

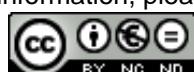


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# **Dissecting the pan-placodal gene regulatory network reveals novel interactions driving sense organ development in vertebrates**

*A thesis submitted in partial fulfilment of the  
requirements of the National University of  
Ireland, Galway for the degree of Doctor of  
Philosophy*

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# Contents

<b>Acknowledgements.....</b>	<b>7</b>
<b>List of abbreviations .....</b>	<b>8</b>
<b>Abstract .....</b>	<b>9</b>
<b>Declaration.....</b>	<b>10</b>
<b>Chapter 1. Introduction.....</b>	<b>11</b>
<b>1.1 Introduction.....</b>	<b>11</b>
<b>1.2 Cranial placodes .....</b>	<b>11</b>
1.2.1 Evolutionary origin of cranial placodes .....	11
1.2.2 Cranial placodes give rise to multiple cell types .....	12
1.2.3 Cranial placodes arise from a common domain .....	15
<b>1.3 Induction of the PPE .....</b>	<b>16</b>
1.3.1 Signalling events required for PPE induction .....	16
1.3.2 Competence factors .....	17
1.3.3 Six genes.....	18
1.3.4 Eya genes .....	19
<b>1.4 Six1 and Eya1 in Development and Disease.....</b>	<b>20</b>
1.4.1 Six1 and Eya1 in development.....	20
1.4.1.1 Six1 and Eya1 in neuronal differentiation.....	21
1.4.1.2 Six1 and Eya1 in proliferating neuronal progenitors.....	22
1.4.2 Six1 and Eya1 in disease.....	23
<b>1.5 Aims of this study.....</b>	<b>23</b>
<b>Chapter 2. Materials and Methods .....</b>	<b>25</b>
<b>2.1 Animal housing and egg laying .....</b>	<b>25</b>
<b>2.2 Embryo manipulation.....</b>	<b>25</b>
2.2.1 Microscopy .....	25
2.2.2 In vitro fertilisation .....	25
2.2.3 Microinjection of mRNA or morpholino antisense oligonucleotides (MO) .....	26
2.2.4 DAPT treatment.....	26
2.2.5 Explanting pre-placodal ectoderm and treatment with cycloheximide/dexamethasone.....	27
2.2.6 Trizol RNA Isolation (placodal explants).....	27
2.2.7 Fixation of embryos .....	28
2.2.8 X-gal staining .....	28
<b>2.3 General molecular techniques .....</b>	<b>28</b>
2.3.1 cDNA synthesis.....	28
2.3.2 PCR.....	28
2.3.3 Colony PCR.....	28
2.3.4 Quantitative PCR .....	29
2.3.5 Restriction digestion .....	29
2.3.6 DNA purification .....	29
2.3.7 RNA purification.....	29
2.3.8 Gel electrophoresis .....	29
2.3.9 Agarose gel DNA extraction .....	30

2.3.10 Subcloning and gene synthesis .....	30
2.3.11 Ligation .....	30
2.3.12 Bacterial transformation .....	30
2.3.13 Midiprep .....	31
2.3.14 Capped mRNA synthesis for microinjection .....	31
2.3.15 RNA probe synthesis for in-situ-hybridisation .....	31
<b>2.4 Wholemount <i>in-situ</i>-hybridisation and immunohistochemistry.....</b>	<b>32</b>
2.4.1 Wholemount in-situ-hybridisation.....	32
2.4.2 Immunostaining on whole mounts .....	33
2.4.3 Vibratome sectioning.....	34
<b>2.5 Bioinformatic analysis of RNA-Seq data.....</b>	<b>34</b>
2.5.1 Initial quality assessment and filtering.....	34
2.5.2 Align reads to genome .....	34
2.5.3 Transcript assembly and differential expression.....	35
2.5.4 Estimating variance between biological replicates .....	35
2.5.5 Generating a scatterplot to visualise FPKM distribution.....	35
2.5.6 Annotating transcript models .....	35
2.5.7 Gene enrichment analysis .....	36
2.5.8 Finding co-differentially expressed genes.....	36
2.5.8.1 Addressing genomic redundancy and tetraploidy in <i>X. laevis</i> .....	36
2.5.8.2 Excluding DEX-induced genes .....	36
2.5.8.3 Identifying common differentially expressed genes .....	37
2.5.9 Gene set enrichment analysis and gene ontology.....	37
2.5.10 Phylogenetic analysis of Hes8 to determine orthology .....	38
<b>Chapter 3. Revealing direct targets of Six1 and Eya1 in the PPE .....</b>	<b>39</b>
<b>3.1 Introduction .....</b>	<b>39</b>
<b>3.2 Conditional overexpression of <i>Six1</i> and <i>Eya1</i> allows only direct targets to be transcribed in the presence of a protein synthesis inhibitor .....</b>	<b>41</b>
3.2.1 GR fusion proteins allow conditional overexpression in the presence of dexamethasone .....	41
3.2.2 Cycloheximide and protein synthesis inhibition.....	41
<b>3.3 The pre-placodal transcriptome dissected .....</b>	<b>42</b>
<b>3.4 An RNA-Seq screen to reveal the direct targets of Six1 and Eya1 in the PPE.....</b>	<b>44</b>
3.4.1 Assessment of sequencing data and rationale for analysis of merged treatment groups .....	44
3.4.2 Direct targets of Six1 and Eya1.....	45
3.4.3 Many direct targets are co-regulated by Six1 and Eya1 .....	46
<b>3.5 Characterisation of RNA-Seq data .....</b>	<b>50</b>
3.5.1 Gene Ontology and gene set enrichment analysis of individual and merged data sets .....	50
<b>3.6 Selection of candidates for in-depth characterisation .....</b>	<b>55</b>
<b>Chapter 4. Characterisation and verification of direct targets of Six1 and Eya1 .</b>	<b>61</b>
<b>4.1 Initial characterisation of selected genes.....</b>	<b>61</b>
4.1.1 Annotation and verification of gene classification.....	61
4.1.2 Hes8 broadly clusters with other vertebrate Hes5 genes .....	61

<b>4.2 Spatio-temporal characterisation of selected target genes.....</b>	<b>64</b>
4.2.1 Genes not expressed in placodes.....	64
4.2.2 Genes expressed broadly in cranial ectoderm including PPE.....	67
4.2.3 Placodal genes expressed in proliferating progenitors of PPE .....	69
4.2.4 Placodal genes expressed in differentiating cells .....	71
4.2.5 General observations on expression of candidate genes .....	75
4.2.6 Selection of core sub-set of genes for functional analysis .....	76
<b>4.3 Verification of selected genes as direct target of Six1 and Eya1 .....</b>	<b>76</b>
4.3.1 qPCR confirms that overexpression of either Six1 or Eya1 is sufficient to up-regulate target genes .....	76
<b>Chapter 5. Analysis of the role Six1 and Eya1 play in regulating their direct targets.....</b>	<b>79</b>
<b>    5.1 An RNA-Seq experiment to reveal effects of Six1 and Eya1 loss of function in the PPE .....</b>	<b>79</b>
5.1.1 Initial analysis of RNA-Seq data .....	79
5.1.2 Very few targets are significantly affected after morpholino knockdown of Six1/Eya1 in RNA-Seq data .....	80
5.1.3 Gene set enrichment for MO data .....	81
<b>    5.2 Six1 and Eya1 regulate direct targets in a complex fashion .....</b>	<b>84</b>
5.2.1 Six1 and Eya1 are necessary for the expression of direct targets .....	86
5.2.2 Six1-GR or Eya1-GR overexpression promotes both repression and activation of targets .....	92
<b>Chapter 6. Six1 and Eya1 regulate neurogenesis by activating Hes5/8 in a Notch-independent way .....</b>	<b>98</b>
<b>    6.1 Notch signalling in neurogenesis .....</b>	<b>98</b>
<b>    6.2 Six1/Eya1 are not required for <i>Hes5/8</i> activation via Notch signalling ..</b>	<b>99</b>
6.2.1 Hes5/8 is a direct effector of Notch signalling .....	99
6.2.2 Six1/Eya1 are not required for inhibition of neuronal differentiation via lateral inhibition.....	102
<b>    6.3 Six1/Eya1 repress neuronal differentiation in a Notch-independent fashion .....</b>	<b>102</b>
6.3.1 Six1 represses neuronal differentiation by activating Hes5/8 in a Notch-independent fashion .....	102
<b>Chapter 7. Discussion .....</b>	<b>105</b>
<b>    7.1 Introduction .....</b>	<b>105</b>
<b>    7.2 A screen to reveal direct targets of Six1 and Eya1 .....</b>	<b>105</b>
7.2.1 Activation of direct targets using CHX and DEX.....	106
7.2.2 Technical issues with analysis of RNA-Seq data .....	107
7.2.3 Technical issues with developed programs .....	109
<b>    7.3 Data quality.....</b>	<b>111</b>
7.3.1 Appraisal of target genes revealed by RNA-Seq screen .....	111
7.3.2 Six1 and Eya1 co-regulate genes in the PPE.....	112
7.3.3 Merged analysis strongly supports individual analysis .....	113
7.3.4 Candidates selected for further characterisation .....	114
7.3.5 Interpretation and analysis of morpholino data .....	115
7.3.5.1 Indirect effects of MOs.....	115

7.3.5.2 Difficulty in interpreting MO phenotype.....	116
7.3.5.3 Indirect activation of p53.....	116
<b>7.4 Dual role of Six1/Eya1 in neuronal differentiation and proliferation .....</b>	<b>118</b>
7.4.1 Six1/Eya1 and neuronal differentiation .....	119
7.4.2 Six1/Eya1 and proliferation .....	120
7.4.3 Six1/Eya1 play a dual role in promoting both neuronal differentiation and proliferation in a dose-dependent fashion .....	121
7.4.4 Six1/Eya1 inhibit neuronal differentiation by directly activating Hes5/8 ..	123
<b>7.5 Towards a gene regulatory network .....</b>	<b>123</b>
7.5.1 General features of GRNs .....	124
7.5.1.1 Autoregulation.....	125
7.5.1.2 Feed-forward loops.....	127
7.5.1.3 Double-negative gates .....	129
7.5.2 A neuronal differentiation cascade .....	130
7.5.3 A model of Six1/Eya1-driven gene regulation in the PPE .....	131
<b>7.6 Conclusion .....</b>	<b>134</b>
7.6.1 Future work.....	134
7.6.2 Summary .....	135
<b>References .....</b>	<b>136</b>
<b>Appendix A. Solutions .....</b>	<b>149</b>
A.1 General solutions.....	149
A.2 Stock solutions for <i>in-situ</i> -hybridisation and RNA synthesis .....	150
<b>Appendix B. Primer list .....</b>	<b>154</b>
B.1 Primer sequences and PCR conditions .....	154
<b>Appendix C. Script.....</b>	<b>156</b>
C.1 General bioinformatic pipeline .....	156
C.2 Pearson's correlation.....	158
C.3 Merger .....	159
C.4 Scatterplot .....	160
C.5 Transcripts .....	162
C.6 DE genes.....	164
C.7 Annotator .....	165
C.8 Enrichment .....	167
C.9 Co-reg .....	169
C.10 Sets.....	175
C.11 qPCR plots .....	178
<b>Appendix D. Placodal transcriptome .....</b>	<b>179</b>
D.1 Placodal transcriptome .....	179
<b>Appendix E. Individual analysis.....</b>	<b>190</b>
E.1 Six1 <sub>i</sub> : Up-regulated genes.....	190
E.2 Six1 <sub>i</sub> : Down-regulated genes.....	198
E.3 Eya1 <sub>i</sub> : Up-regulated genes .....	204
E.4 Eya1 <sub>i</sub> : Down-regulated genes .....	215

E.5 Six1+Eya1 <sub>i</sub> : Up-regulated genes .....	220
E.6 Six1+Eya1 <sub>i</sub> : Down-regulated genes .....	237
<b>Appendix F. Merged analysis .....</b>	<b>247</b>
F.1 Six1 <sub>m</sub> : Up-regulated genes.....	247
F.2 Six1 <sub>m</sub> : Down-regulated genes.....	251
F.3 Eya1 <sub>m</sub> : Up-regulated genes .....	252
F.4 Eya1 <sub>m</sub> : Down-regulated genes .....	255
F.5 Six1+Eya1 <sub>m</sub> : Up-regulated genes .....	256
F.6 Six1+Eya1 <sub>m</sub> : Down-regulated genes .....	260
<b>Appendix G. Hes phylogeny.....</b>	<b>261</b>
G.1 Hes alignment.....	261
G.2 Hes ML phylogeny .....	265
<b>Appendix H. <i>In-situ</i> sense controls.....</b>	<b>266</b>
H.1 <i>In-situ</i> sense controls for newly described genes .....	266
<b>Appendix I. Morpholino RNA-Seq data.....</b>	<b>267</b>
I.1 Six1-MO: Down-regulated genes .....	267
I.2 Six1-MO: Up-regulated genes.....	274
I.3 Eya1-MO: Down-regulated genes .....	279
I.4 Eya1-MO: Up-regulated genes .....	290
I.5 Six1-MO+Eya1-MO: Down-regulated genes.....	295
I.6 Six1-MO+Eya1-MO: Up-regulated genes .....	299
<b>Appendix J. Raw embryo counts for MO statistical analysis .....</b>	<b>302</b>

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## List of abbreviations

<b>BO</b>	Branchio-Otic
<b>BOR</b>	Branchio-Oto-Renal
<b>CHX</b>	Cycloheximide
<b>DEX</b>	Dexamethasone
<b>E</b>	Enrichment score
<b>Eya1<sub>m</sub></b>	Analysis after merging replicates from Eya1 <sub>i</sub> and Six1+Eya1 <sub>i</sub> datasets
<b>Eya<sub>i</sub></b>	Analysis after individual overexpression of <i>Eya1</i> -GR
<b>FC</b>	Log <sub>2</sub> Fold Change
<b>FPKM</b>	Fragments Per Kilobase of exon per Million fragments mapped
<b>GO</b>	Gene Ontology
<b>GR</b>	Glucocorticoid Receptor
<b>GRN</b>	Gene Regulatory Network
<b>MO</b>	Morpholino antisense oligonucleotides
<b>Notch-ICD</b>	Notch Intracellular Domain
<b>PPE</b>	Pan-Placodal Ectoderm
<b>Six1+Eya1<sub>i</sub></b>	Analysis after overexpression of both <i>Six1</i> -GR and <i>Eya1</i> -GR
<b>Six1+Eya1<sub>m</sub></b>	Analysis after merging replicates from Six1 <sub>i</sub> , Eya1 <sub>i</sub> and Six1+Eya1 <sub>i</sub> datasets
<b>Six1<sub>i</sub></b>	Analysis after individual overexpression of <i>Six1</i> -GR
<b>Six1<sub>m</sub></b>	Analysis after merging replicates from Six1 <sub>i</sub> and Six1+Eya1 <sub>i</sub> datasets
<b>Su(H)-DBM</b>	Suppressor of Hairless DNA Binding Mutant

## Abstract

The genes encoding the transcription factor Six1, and its co-factor Eya1 together mark the embryonic territory - the pan-placodal ectoderm (PPE) - that gives rise to most cranial sense organs in vertebrates. Mutation of these genes gives rise to severe craniofacial defects, and they are implicated in the progression of a range of cancers. However, how Six1 and Eya1 regulate such processes is unclear. This study reveals the direct targets of Six1 and Eya1 in the PPE, and further verifies and characterises a subset of these to elucidate important, and novel, regulatory interactions underlying sense organ formation. The direct targets revealed in this study are involved in both the proliferation (including *Hes5/8*, *Hes9*, *Sox2* and *Sox3*) and differentiation (including *Ngn1*, *Atoh1*, *Pou4f1.2*, *Isl2*, *Gfi1a* and *Tlx1*) of neuronal progenitors, and this study provides compelling evidence that Six1 and Eya1 directly affect both processes. Additionally, this study demonstrates that overexpression of *Six1* and *Eya1* inhibits neuronal differentiation in a Notch-independent way, by directly activating *Hes5/8*, suggesting a likely mechanism through which Six1 and Eya1 promote proliferation in neuronal progenitors. Finally, this study proposes a model of gene regulation in the PPE, whereby Six1 and Eya1 synergistically regulate a complex gene regulatory network underlying the development of sense organs in vertebrates.

## Declaration

This thesis has not been submitted in whole, or in part, to this or any other University for any other degree and is, except where otherwise stated, the original work of the author.

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Signed \_\_\_\_\_

Nick Riddiford, February 2016

# Chapter 1. Introduction

## 1.1 Introduction

A complex and specialised head is one of the key features that distinguishes vertebrates from other chordates. Indeed, a number of novel cranial structures evolved in the vertebrate lineage - including the paired sense organs and craniofacial skeleton - that together comprise the vertebrate “new head” (Gans and Northcutt, 1983; Northcutt, 2005). Surprisingly, many of these evolutionary novel structures arise from just two embryonic tissues, the neural crest and cranial placodes. The neural crest comprises a unique population of multipotent cells that migrate throughout the embryo to generate a wide array of cell types, contributing to the peripheral nervous system, craniofacial cartilage and smooth muscle, and has been the focus of an intense research programme since it was first discovered (Baker and Bronner-Fraser, 1997; Endo et al., 2002; Knecht and Bronner-Fraser, 2002; Mayor and Aybar, 2001; Mayor et al., 1998; Sauka-Spengler et al., 2007). Like the neural crest, cranial placodes also give rise to populations of migratory cells, and differentiate into a range of cell types including sensory neurons, glia and secretory cells, and make important contributions to many cranial sensory organs and ganglia. However, the mechanisms underlying cranial placode development have remained largely unexplored until recently, where the discovery of placode-specific expression of many transcription factors supported suggestions that all placodes share a common developmental origin has driven a productive new field of research (Baker and Bronner-Fraser, 2001; Brugmann and Moody, 2005; Schlosser, 2006; 2005; Schlosser and Ahrens, 2004; Streit, 2008; 2004). Although progress has been made to identify the inductive signals required for the establishment of a pre-placodal territory (Grocott et al., 2012), and for subsequent segregation of individual placodes (reviewed in Schlosser, 2006), little is known about the signalling events that promote and maintain placodal fate.

## 1.2 Cranial placodes

### 1.2.1 Evolutionary origin of cranial placodes

Cranial placodes are a series of non-neural ectodermal thickenings that, together with the neural crest, give rise to many of the evolutionary novelties of the vertebrate head. This “new head” is proposed to have evolved on the adoption of an increasingly active and predatory lifestyle during the evolution of vertebrates from filter-feeding ancestors (Gans and Northcutt, 1983; Northcutt, 2005). Evolutionary novel tissues and structures, such as the cranial placodes

and their derivatives, often arise from the redeployment of developmental genes in new contexts, the so-called “tinkering” of evolution (Jacob, 1977). In this vein, small changes in gene regulation over evolutionary time, perhaps the addition or deletion of a *cis*-regulatory element, may lead to the gain or loss of an upstream regulator or downstream target and facilitate the re-wiring of regulatory networks, driving the evolution of novel structures. In light of recent advances in developmental genetics, the gene regulatory networks (GRNs) that underlie both cranial placodes and neural crest are beginning to be explored (Grocott et al., 2012; Meulemans and Bronner-Fraser, 2004; Neves et al., 2013; Simoes-Costa et al., 2014), however there is currently little known about the regulatory events that underlie placode development. In order to unravel the sort of regulatory changes that must have taken place during the evolution of sense organs, a model of gene regulation must first be postulated for both a modern vertebrate and in other extant chordates, to enable the reconstruction of the condition present in vertebrate ancestors.

As the fossil record of early stem vertebrates is patchy, inferences of the evolutionary history of placodes must be made by comparison with other extant taxa. Historically, morphological similarities positioned cephalochordates (*amphioxus*) as the closest living relatives of vertebrates, with urochordates (tunicates) considered more distantly related, however, recent and strongly supported molecular phylogenies now place tunicates as the sister group to vertebrates (Bourlat et al., 2006; Delsuc et al., 2006). Thus tunicates, despite in many ways being poor models for inferring characteristics of the last tunicate-vertebrate ancestor, share some developmental characteristics with vertebrates and, as such, may provide a suitable reference for examining the evolution of placodes via re-wiring of a GRN. Indeed, tunicates utilise many of the same transcription factors as vertebrates to specify several tissues required for the establishment of placodes (see section 1.3), and later form ectodermal thickenings that give rise to several of the same cell types found in vertebrate placodes (reviewed in Schlosser, 2015). The ur-vertebrate then, probably possessed such protoplacodal regions; however only in vertebrates did placodes proper (comprised of clusters of proliferating progenitors that give rise to the complex sense organs) originate from these domains.

### *1.2.2 Cranial placodes give rise to multiple cell types*

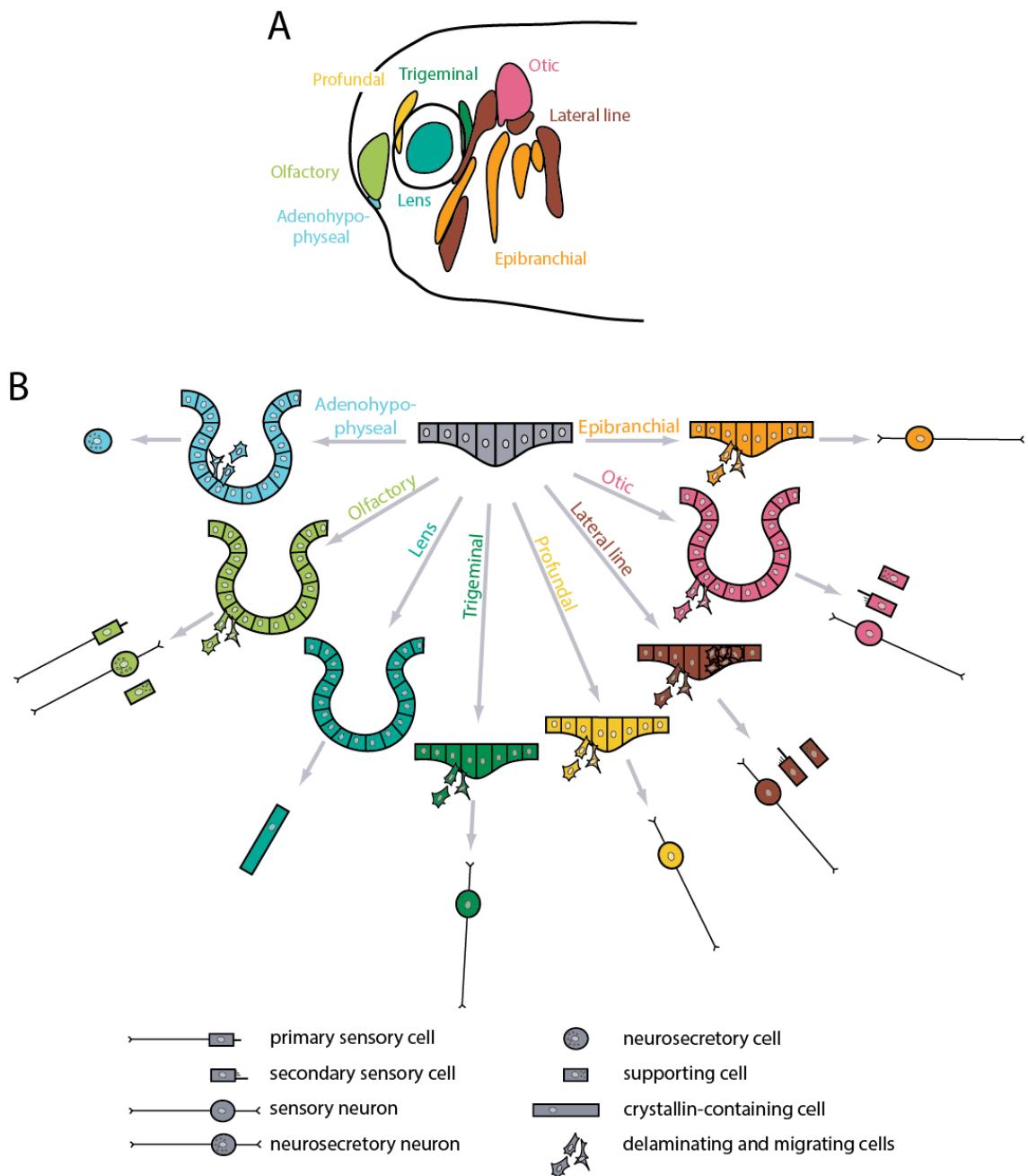
Cranial placodes arise from a common precursor region in the embryonic ectoderm - the pre-placodal ectoderm (PPE) - and contribute to many ganglia and paired sense organs of the vertebrate head (McCabe and Bronner-Fraser, 2009; Schlosser, 2006; 2005; Streit, 2007; 2004).

Seven distinct placodes can be distinguished: The adenohypophyseal, olfactory, lens, otic, lateral line, epibranchial, and profundal/trigeminal placodes (fig. 1.1). In light of extensive recent review (Baker and Bronner-Fraser, 2001; Grocott et al., 2012; Schlosser, 2010; 2006), this introduction will only provide a brief overview of each placode type and its derivatives, before considering likely factors involved in placode induction and specification.

The un-paired adenohypophyseal placode is located in the anterior midline and gives rise to the adenohypophysis - the rostral lobe of the pituitary gland - and generates a variety of endocrine cell types that release hormones important for the regulation of various body functions such as gonadotropes and somatotropes (reviewed in Dubois et al., 1997). The olfactory placode produces chemosensory neurons of the olfactory epithelium and the vomeronasal organ, as well as gonadotropin-releasing hormone (GnRH)-secreting cells that influence reproductive behaviour. These neuronal cells delaminate from the olfactory epithelium and migrate along the olfactory nerve to inhabit regions of the forebrain (reviewed in L. Crews and Hunter, 1993). Together with the adenohypophyseal placode, the lens placode is the only placode that does not give rise to neurons, rather developing into cells that accumulate crystalline to form the lens of the eye (reviewed in Piatigorsky, 1981). The profundal and trigeminal placodes generate migratory cells which delaminate from the placode and, together with contributions from the neural crest, form the sensory neurons of the craniofacial ganglia that mediate temperature, touch and pain sensation in the head (e.g. see Schlosser, 2006). Lateral line placodes give rise to mechanosensory cells and as well as the neurons innervating them, which detect water movements and electric fields in many aquatic vertebrates. Mechanosensory cells are also produced by the otic placode, which gives rise to the entire inner ear (comprising mechanosensory hair cells, their support cells and secretory cells) (reviewed in Torres and Giráldez, 1998). Finally, viscerosensory neurons also delaminate from the epibranchial placodes and innervate cranial sense organs associated with the digestive tract, for example taste buds in the mouth cavity (reviewed in Northcutt, 2004).

Despite the diversity of cell types generated by different placodes, there are several striking similarities between the various placodes. As well as the high levels of proliferation apparent in placodal cells, they all undergo cell shape changes or movements during their development. For example, while all placodes form focal thickenings, several (adenohypophyseal, olfactory, lens, and otic placodes) also invaginate, and the development of all except the lens placode involves delamination and migration of sensory or neuronal precursor cells. Finally, with the exception of the lens and adenohypophyseal placodes, all are

neurogenic, including neurons among their derivatives (fig. 1.1B). It is now clear that these shared, underlying characteristics of placodes, is a consequence of their development from a common developmental precursor (reviewed in Schlosser, 2006; Streit, 2004).

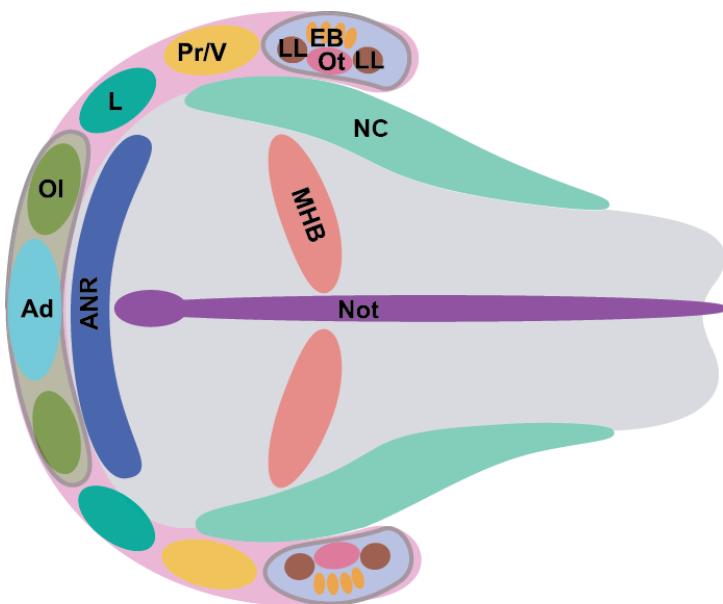


**Fig. 1.1** Vertebrate cranial placodes. **(A)** Schematic of tail bud stage *Xenopus* embryo showing segregated placodes (after Schlosser, 2010). **(B)** Developmental fates and derivative cell types arising from different cranial placodes (after Schlosser, 2010).

### *1.2.3 Cranial placodes arise from a common domain*

Whether or not placodes arise from a common developmental precursor, or are essentially unrelated structures that form in distinct regions of the head, has been a central question in placode research (e.g. Baker and Bronner-Fraser, 2001; Streit, 2004). Fate maps provide compelling evidence that all placodes originate from a crescent-shaped ectodermal domain established at the border of the neural plate/neural crest and future epidermis (fig. 1.2) (Bhattacharyya and Bronner-Fraser, 2004; Pieper et al., 2011; Streit, 2002; H. Xu et al., 2008).

In neural plate stage embryos, the regions from which individual placodes originate overlap extensively within the PPE, and segregate later in development. However, a more hotly contested question is whether PPE is biased towards a placodal fate (e.g. Schlosser, 2006; 2005; Schlosser and Ahrens, 2004; Streit, 2004). Classically, the observation that placodes develop from a common ectodermal thickening was taken as evidence to support a placodal bias. However, such morphological characteristics do not appear to be consistent between taxa, with some thickenings not apparent in the chick, for example (Schlosser, 2006; Schlosser and Northcutt, 2000). A more recent and convincing argument for the existence of a generic placodal bias in the PPE has been suggested by the identification of transcription factors expressed within the PPE whose expression is maintained in some or all cranial placodes (Brugmann et al., 2004; Schlosser and Ahrens, 2004, reviewed in Baker and Bronner-Fraser, 2001; Brugmann and Moody, 2005; Schlosser, 2006; 2005; Schlosser and Ahrens, 2004; Streit, 2004). Genes such as *Dlx*, *GATA4*, and *FoxI*, for example, are expressed in broad ectodermal domains that overlap the PPE, and are promising candidates for factors that establish and demarcate the PPE. Several other gene families (such as *Pax*, *Sox*, *Fox*, *Irx*, *Pitx*, and *Thx*) are expressed in more restricted sub-regions of the PPE, and may thus be involved in its segregation into individual placodal territories, or possibly confer a bias towards specific placodal fate (reviewed in Schlosser, 2010). However, only genes of the Six1/2 and Six4/5 subfamilies, as well as the Eya family, are confined to the PPE at neural plate stages, and are subsequently maintained in all cranial placodes, making them excellent candidates for placodal markers that confer a general placodal bias on the PPE and regulate placode development. These, along with several other factors that appear to promote placodal competence, are discussed below.



**Fig. 1.2** All placodes arise from a pan-placodal primordium. Schematic of a neural plate stage *Xenopus* embryo showing position of placodes (coloured ovals) contained within a common pan-placodal ectoderm (PPE; pink horseshoe-shaped domain). The anterior placodal domain (green overlay) comprises the prospective adenohypophyseal (blue) and olfactory (green) placodes. The posterior placodal domain (blue overlay) includes the prospective otic (pink), lateral line (brown), and epibranchial placodes (orange). The lens (blue-green) and profundal/trigeminal (yellow) placodes border the anterior and posterior regions of the PPE, respectively. To provide morphological landmarks, the anterior neural ridge (ANR; dark blue), midbrain-hindbrain boundary (MHB; salmon pink), neural crest (NC; mint green) and notochord (Not; purple) are labelled on top of the grey neural plate. Abbreviations: Ad, Adenohypophyseal placode; EB, Epibranchial placodes; L, Lens placode; LL, Lateral line placodes; OI, Olfactory placode; Ot, Otic placode; Pr/V, Profundal/trigeminal placodes; ANR, anterior neural ridge; MHB, midbrain-hindbrain boundary; Not,

### 1.3 Induction of the PPE

#### 1.3.1 Signalling events required for PPE induction

Induction of the PPE occurs concomitantly with the origin of other ectodermally derived domains such as the epidermis, neural plate, and neural crest (reviewed in Grocott et al., 2012; Saint-Jeannet and Moody, 2014; Schlosser, 2010). Establishment of such tissues begins during early gastrulation, where the ectoderm becomes segregated into future neural and non-neural domains by signals including inhibitors of bone morphogenetic proteins (BMPs), inhibitors of wingless/integrated proteins (Wnts), and fibroblast growth factors (FGFs). Spatiotemporal combinations of these signals establish differences in the expression of transcription factors that confer identity to neural and non-neural ectoderm. In the non-neural ectoderm, BMP signals from the neural plate border induce the expression of *Dlx*, *GATA* and *FoxI* genes, whereas in the neural territory, BMP represses genes such as *Sox2*, *Sox3* and *Zic*. At the intersection of these neural and non-neural competence territories a neural plate border region is established, comprising precursors of neural, neural crest and PPE lineages. During

gastrulation these expression domains become resolved into complementary non-neural (*Dlx*, *GATA*, *FoxI*) and neural (*Zic*) territories, with the competence to form PPE restricted to the former (non-neural), and neural crest restricted to the latter (neural) (Pieper et al., 2012).

While BMP inhibition is required for neural competence, alone it is not sufficient to induce the expression of neural markers. Rather, BMP inhibition combined with ectopic FGF signals at blastula stage is sufficient to induce neural fate (Delaune et al., 2005), while similar treatment of non-neural ectoderm (previously exposed to high BMP levels) at late gastrula stage in *Xenopus* has been shown to induce ectopic expression of *Six1* (Ahrens and Schlosser, 2005). Although BMP inhibition and FGF signals appear to promote the expression of PPE marker genes (such as *Six1*), additional inhibition of the Wnt system is required for proper formation of PPE. Here, antagonising Wnt signalling causes expansion of the PPE (Brugmann et al., 2004; Litsiou et al., 2005), whereas its activation inhibits PPE gene expression.

Thus, induction of the PPE proceeds in a complicated and multistep fashion, requiring multiple input signals from underlying mesoderm and the neural plate. Here, a Wnt signal from lateral and posterior endomesoderm appears to restrict the posterior PPE territory but expand the neural crest domain, in addition to subdividing the PPE into future anterior (Wnt signal inhibited; prospective adenohypophyseal and olfactory) and posterior (Wnt signal active; prospective otic, lateral line, and epibranchial) placodal domains (Brugmann et al., 2004; Litsiou et al., 2005; Schlosser and Ahrens, 2004, reviewed in Grocott et al., 2012; Saint-Jeannet and Moody, 2014; Schlosser, 2010). In addition, mesoderm directly underlying future PPE expresses high levels of FGF as well as BMP and Wnt antagonists, shielding the PPE from adopting a neural fate, and promoting the expression of PPE marker genes (such as *Six* and *Eya* genes) (reviewed in Bailey and Streit, 2005; Schlosser, 2010; 2006).

### 1.3.2 Competence factors

Whether or not the effect of BMPs, FGFs and Wnts on the expression of *Six1/Eya1* is direct or indirect is unknown. While a *Six1* enhancer that can drive its expression in the PPE does not contain direct binding sites for direct effectors of BMP, FGF or Wnt pathways (Sato et al., 2012; 2010), it does contain a binding site for *Dlx5*, which is regulated by BMP signalling (Litsiou et al., 2005; Monsoro-Burq et al., 2005; Streit, 2007). *Dlx5* is expressed in prospective epidermis and the PPE at neural plate stages and is able to activate the expression of both *Six1* and *Eya1*, and its knockdown is able to reduce PPE gene expression (McLaren et al.,

2003), making it a prime candidate (along with closely related *Dlx3*) for an important upstream regulator of Six1, and mediator of PPE induction (Pieper et al., 2012; Sato et al., 2010). Several additional markers of non-neural ectoderm may also play a role in regulation of Six1 and inducing PPE formation. Both FoxI and GATA2/3 are regulated by BMP and double knockdown of both genes attenuates *Six1* and *Eya1* expression in the PPE (H. J. Kwon et al., 2010). In addition, the expression of these genes appears to be required for the activation of PPE genes in response to BMP, FGF and Wnt signals (H. J. Kwon et al., 2010; Pieper et al., 2012). Taken together, the expression of *Dlx*, *FoxI* and *GATA2/3* genes in the non-neural ectoderm, their capacity to respond to endogenous signals in the non-neural ectoderm (BMP, FGF and Wnt) makes them promising candidates for factors that define the non-neural vs. neural competence territories, and mediate PPE induction.

### 1.3.3 Six genes

Several members of the Six gene family, including Six1/2 and Six4/5, are expressed in a horseshoe-shaped domain coincident with the PPE at neural plate stages, and are maintained in all placodes (with the exception of the lens in *Xenopus*) later in development, making them prime candidates for factors promoting placodal fate (Ghanbari et al., 2001; Kawakami et al., 2000; Pandur and Moody, 2000). Six genes encode homeobox transcription factors, containing both a highly conserved DNA binding homeodomain and a Six domain located toward the N-terminal region of the protein, which allows various protein-protein interactions (Brugmann et al., 2004; Kawakami et al., 2000; Kobayashi et al., 2001; Pignoni et al., 1997). Depending on the co-factor, Six genes can act as either transcriptional activators (e.g. Six-Eya) or repressors (e.g. Six-Dach or Six-Groucho) (Brugmann et al., 2004; Silver et al., 2003). While several members of the Six1/2 and Six4/5 subfamilies show widespread expression in the PPE (Ghanbari et al., 2001; Ikeda et al., 2007; Kobayashi et al., 2001; Laclef et al., 2003; X. Li et al., 2003; Pandur and Moody, 2000; Schlosser and Ahrens, 2004; Zheng et al., 2003; Zou et al., 2004), and may play important roles in placode development (Hoskins et al., 2007; Klesert et al., 2000), given the relative paucity of information regarding other Six family members, only Six1 will be addressed here.

*Six1* is the vertebrate homologue of the *Drosophila* gene *sine oculis* (*so*), which is a core member of the network of genes (*Six-Eya-Dach-Pax*) directing proliferation and cell movements during compound eye development (Cheyette et al., 1994), roles that are conserved in vertebrates (Bonini et al., 1997; Kawakami et al., 2000). Knockout of *Six1* in vertebrates is associated with the severe disruption of the development of multiple placodal

derivatives (Bricaud and Collazo, 2011; Ikeda et al., 2007; Laclef et al., 2003; Zheng et al., 2003; Zou et al., 2004), and causes branchio-otic (BO) and branchio-oto-renal (BOR) syndromes, which are characterised by craniofacial defects and hearing loss (BO) and additional malformations of the kidney and urinary tract (BOR) (Abdelhak et al., 1997; Hoskins et al., 2007; S. Kumar et al., 1997; Ruf et al., 2004). In addition, Six1 is required for the regulation of both proliferation of neural progenitors and subsequently their differentiation in *Xenopus* (Ahmed et al., 2012a; 2012b; Schlosser et al., 2008).

Six1 is also required for the formation of the pre-placodal ectoderm; its overexpression represses neural plate (*FoxD3*) and epidermis (*Keratin*) markers while activating other genes expressed in the PPE (such as *Sox11* and *Eya1*), suggesting that Six1 functions to promote a placodal fate at the expense of epidermis and neural plate. Consistent with the requirement of Six1 for PPE formation, morpholino (MO)-mediated knockdown of *Six1* reduced the expression domains of the PPE markers, and expanded those of neural plate and epidermis markers. Taken together, these results demonstrate that Six1 is required for the expression of PPE genes, and acts to promote general placodal properties (Brugmann et al., 2004).

#### 1.3.4 Eya genes

Genes belonging to the Eya family are vertebrate homologues of the *Drosophila* gene *eyes absent* (*eya*) which, together with *so*, forms the core of the *Six-Eya-Dach-Pax* network required for normal eye development in *Drosophila* (Bonini et al., 1997; Cheyette et al., 1994; Donner and Maas, 2004; J. P. Kumar, 2009). Misexpression of eya is sufficient to induce ectopic eyes in *Drosophila* (Bonini et al., 1997; R. Chen et al., 1997; Pignoni et al., 1997). Like *Six1*, vertebrate *Eya1* plays a crucial role in placode development. It is initially expressed throughout the PPE, and maintained in multiple placodal derivatives (Abdelhak et al., 1997; David et al., 2001; P. X. Xu et al., 1997). In mice, *Eya1* plays an important role in ear, kidney and thymus development (P. X. Xu et al., 1999; 2002), and mutation of *Eya1* in humans leads to BO and BOR syndromes (S. Kumar et al., 1998).

*Eya1* encodes a transcriptional co-activator that requires Six1 for its nuclear translocation, and for its ability to positively regulate gene expression (Y. Zhang et al., 2004). However, Eya1 is unusual in that, in addition to having transactivation potential when coupled with a DNA-binding partner, it also demonstrates tyrosine phosphatase activity (Rayapureddi et al., 2003; Tootle et al., 2003). Such phosphatase activity may modulate the transcriptional

consequence of binding with additional, non-Six co-factors, such as Dach. Dach proteins are able to directly bind DNA, and act as transcriptional co-repressors or co-activators in a context-dependent fashion (X. Li et al., 2003). When coupled with Six1 (Six-Dach), Dach acts as a strong repressor of transcription, however, in complex with Six1 and Eya1 (Six-Eya-Dach), the phosphatase activity of Eya1 appears to convert it into a transcriptional activator (X. Li et al., 2003).

The co-expression in the PPE and direct interaction of Six1 and Eya1 makes them ideal candidates for factors that promote generic placodal properties. Indeed, both Six1 and Eya1 are essential for the expression of other PPE marker genes at neural fold stages (Brugmann et al., 2004; Christophorou et al., 2009), and are additionally required for subsequent segregation and morphogenesis of individual placodes (Ahmed et al., 2012b; 2012a; Laclef et al., 2003; Schlosser et al., 2008; P. X. Xu et al., 1999; Zheng et al., 2003; Zou et al., 2004). However, while overexpression of *Six1/Eya1* can induce expression of other PPE genes, in isolation, it cannot act to confer PPE status on naïve ectoderm (Christophorou et al., 2009). Instead, as discussed in section 1.3.1, Six1/Eya1 act to drive a region of non-neural ectoderm towards a PPE fate against a backdrop of placodal competence, as established by the combined action of signalling events from nearby tissues.

## 1.4 Six1 and Eya1 in Development and Disease

### 1.4.1 *Six1 and Eya1 in development*

Apart from their involvement in sense organ development, Six1 and Eya1 also have important functions in the development of other tissues such as muscle, kidney, heart and skeleton, where they regulate many processes including cell fate specification and maintenance (reviewed in Tadjudide and Hegde, 2013; W. Wu et al., 2015). While Six1 and Eya1 are involved in multiple stages of development, their mode of action is conserved, and in various systems they often act synergistically to maintain cells in a proliferative progenitor state as well as initiating differentiation programmes (P. X. Xu, 2013). Indeed, both Six1 and Eya1 have recently been identified as important regulators of both proliferation and neuronal differentiation in cranial placodes (Ahmed et al., 2012b; 2012a; Schlosser et al., 2008; Zou et al., 2004). Although these two facets of their function are ostensibly paradoxical (considering that a single cell must choose either to proliferate or differentiate), Six1 and Eya1 appear to play a dual role in regulating the balance between proliferating progenitors and differentiating neurons in populations of cells in a dose-dependent fashion (Schlosser et al., 2008).

Considering this dual role, the function of Six1 and Eya1 in progenitors and differentiating neurons will be discussed separately.

#### 1.4.1.1 *Six1* and *Eya1* in neuronal differentiation

In *Drosophila*, *so* (*Six1*) and *eya* (*Eya1*) are often co-expressed with various neuronal differentiation markers and directly activate the neuronal determination (or proneural) gene *atonal*, suggestive of an ancient role in promoting neuronal differentiation (Tanaka-Matakatsu and Du, 2008; T. Zhang et al., 2006). Such a role for Six1 and Eya1 in neuronal differentiation is widely conserved throughout metazoa (Ahmed et al., 2012b; Gehring, 2005; Maier et al., 2014; Schlosser et al., 2008; Zou et al., 2004), and loss-of-function or knockdown of *Six1* or *Eya1* in vertebrates results in strong defects in neuronal differentiation affecting multiple placodal derivatives (Bricaud and Collazo, 2011; Ikeda et al., 2007; Laclef et al., 2003; X. Li et al., 2003; Schlosser et al., 2008; P. X. Xu et al., 1999; Zheng et al., 2003; Zou et al., 2004). It is suggested that together, Six1 and Eya1 mediate neuronal differentiation by activating (directly and/or indirectly) proneural (*Ngn1*, *Ngn2* and *Atoh1*) and neuronal differentiation (*NeuroD1*) genes and, congruent with this, mutants or morphants of *Six1* or *Eya1* show reduced or completely abated expression of such genes in many placodes (Ahmed et al., 2012b; 2012a; Bosman et al., 2009; Bricaud and Collazo, 2011; Ikeda et al., 2007; Schlosser et al., 2008; Zou et al., 2004), indicating that both Six1 and Eya1 are essential for sensory neuron differentiation and maintenance in placodes.

In addition to activating proneural genes, Six1 and Eya1 also appear to directly regulate several *SoxB1* genes (*Sox2* and *Sox3*; Schlosser et al., 2008). *SoxB1* genes are known to play important roles in biasing cells towards neuronal development and are required for several steps of neuronal differentiation in the central nervous system (Cavallaro et al., 2008; Kishi et al., 2000; Miyagi et al., 2009; Mizuseki et al., 1998; Schlosser et al., 2008; Taranova et al., 2006; Zhao et al., 2004). While the precise mechanism of how *SoxB1* genes act to promote neuronal differentiation is unclear, it seems likely they operate in a highly context-dependent fashion, in part mediated by co-factor binding (Miyagi et al., 2009). Indeed, it has recently been demonstrated that Sox2 synergises with both Six1 and Eya1 to co-ordinate neuronal development by directly activating transcription of *Atoh1* in hair cells (Ahmed et al., 2012b; 2012a).

#### 1.4.1.2 Six1 and Eya1 in proliferating neuronal progenitors

Apart from their role in neuronal differentiation, Six1 and Eya1 also promote proliferating neuronal progenitors, possibly through several different mechanisms. On the one hand, Six1 and Eya1 may affect progenitors via *SoxB1* genes. While *SoxB1* genes act under some circumstances to promote neuronal differentiation (see section 1.4.1.1), they are more typically known for their ability to confer stem cell-like properties, and maintain progenitor cells in a proliferative and undifferentiated state (Chew and Gallo, 2009; Miyagi et al., 2009; Wegner, 1999; Wegner and Stolt, 2005). Indeed, in placodes, *SoxB1* genes are primarily expressed in proliferating progenitors, but are also required for neuronal differentiation and maintenance of placode-derived neurons (Kishi et al., 2000; Schlosser et al., 2008). Although it is currently not clear how *SoxB1* genes act to promote both proliferation and neuronal differentiation, one possibility is that, like their up-stream regulators Six1 and Eya1, they act in a dose-dependent fashion, activating proneural genes (*Ngn1* and *Atoh1*) at low doses, and promoting proliferation at high doses (Dabdoub et al., 2008; Schlosser et al., 2008; Taranova et al., 2006).

As well as initiating a cascade of neuronal differentiation, activation of proneural genes by Six1 and Eya1 (possibly through, or in concert with, *SoxB1* genes) additionally represses neuronal differentiation in adjacent cells via lateral inhibition (Chitnis, 1995; Ma et al., 1996). Here, proneural genes such as *Ngn1* activate Notch ligands (e.g. Delta), inducing Notch signalling in adjacent cells which, through activation of effectors of Notch signalling (such as Hes1 and Hes5), inhibits neuronal differentiation (Cau et al., 2000; Kageyama and Ohtsuka, 1999; Kageyama et al., 2008; Ohtsuka, 1999). Conversely, since Hes5 has also been shown to be a direct target of Sox2 (Engelen et al., 2011), the direct activation of such Notch effectors by *SoxB1* genes would also potentially allow Six1 and Eya1 to block differentiation and promote proliferation cell-autonomously.

Additionally, Six1/Eya1 may maintain progenitors in a proliferative state by directly activating cell cycle control genes to promote cell cycle progression and proliferation (such as *CyclinA1*, *CyclinD1*, and *c-Myc*) (Coletta et al., 2004; X. Li et al., 2003; Z. Li et al., 2013; Yu et al., 2006). Such genes regulate cell cycle checkpoint functions, and maintain cells in an undifferentiated state by preventing cell cycle exit. Interestingly, CyclinD1 is also a direct transcriptional target of Sox2, which suggests a significant degree of overlap between direct targets of Six1/Eya1 and Sox2, further indicating a combined role in promoting both proliferation and neuronal differentiation (Ahmed et al., 2012b; 2012a; Y. Chen et al., 2008).

#### *1.4.2 Six1 and Eya1 in disease*

The ability of Six1 and Eya1 to maintain and expand populations of progenitor cells was first revealed by the observation that *Six1* expression is up-regulated in mammary carcinomas, and correlates strongly with breast cancer metastasis (Ford et al., 1998). *Six1* overexpression can induce proliferation in breast cancer, and stimulate tumorigenesis by directly activating CyclinA1, CyclinD1, c-Myc and Ezrin (Coletta et al., 2004; Ford et al., 1998; X. Li et al., 2003; Z. Li et al., 2013; Yu et al., 2006). Interestingly, Eya1 has also been shown to directly interact with CyclinD1 in a Six1-independent fashion in breast cancers, where it acts to promote proliferation as well as inhibit apoptosis (K. Wu et al., 2013), suggesting that Six1 and Eya1 may influence cancer formation and growth via different mechanisms. Since the early discoveries linking Six1 to cancer, both Six1 and Eya1 have been implicated in multiple “hallmarks of cancer” (such as sustained proliferation, invasion and metastasis), and the Six1-Eya1 complex is considered a therapeutically valuable target in cancer treatment (Blevins et al., 2014).

In addition to being implicated in cancer formation, dysregulation of *Six1* and *Eya1* genes is also associated with multiple craniofacial disorders. In humans, mutation of *Six1* or *Eya1* causes abnormalities in several organ systems leading to Branchio-oto-renal (BOR) syndrome (Abdelhak et al., 1997; Hoskins et al., 2007; Ruf et al., 2004), of which the most prominent symptoms are hearing loss (in 93% of affected individuals) (Heimler et al., 1986), urinary tract malformations and chronic renal failure (Ruf et al., 2004). Some *Eya1* mutations in humans are also associated with the formation of congenital cataracts (Azuma et al., 2000). Mice homozygous for *Eya1* null alleles mice, display full loss of ears and kidneys (P. X. Xu et al., 1999). In mice, cranial ganglia reductions or total absence are also observed in mutants of either Six1 or Eya1, and in *Xenopus* their MO-mediated knockdown impairs neuronal differentiation in all neurogenic placodes (Schlosser et al., 2008; P. X. Xu et al., 1999; Zheng et al., 2003; Zou et al., 2004).

#### **1.5 Aims of this study**

Given the important roles Six1 and Eya1 play in multiple stages of development, such as progenitor state maintenance, initiation of differentiation programmes and cell cycle control (reviewed in P. X. Xu, 2013), and the broad range of developmental disorders arising from the disruption of their normal function, it is surprising so little is known about their mode of action. Particularly lacking is an understanding of their direct transcriptional targets in such processes. While several of their direct targets (*CyclinA1*, *CyclinD1*, *c-Myc* and *Ezrin*) are

implicated in cancer metastasis and are known to regulate cell proliferation, adhesion, survival and motility (Coletta et al., 2004; Ford et al., 1998; X. Li et al., 2003; Yu et al., 2006), little is known about how Six1 and Eya1 influence their expression in cranial placodes. Studies demonstrating that Six1-Eya1 directly activate *Gdnf* in muscle precursors and kidney, also suggest mechanisms through which they might influence neuron survival (X. Li et al., 2003), and *Sk12a2* has also been identified as a direct target in dorsal root ganglia (Ando et al., 2005). However, the role these genes may play in placode development is currently unclear. Currently, only two known direct targets of the Six1-Eya1 complex in the PPE and placodes have been identified - *Sox3* and *Atoh1* - both of which are known to play important roles in sense organ development (Ahmed et al., 2012b; 2012a; Schlosser et al., 2008).

Accordingly, this study aims to reveal the direct transcriptional targets of both Six1 and Eya1 in the PPE, using *Xenopus laevis* as a model system. This will be achieved using a combined approach of hormone-induced overexpression of Six1 and Eya1 individually, as well as Six1 and Eya1 together, in the presence of a protein synthesis inhibitor. Such a technique has recently been used to great effect in the reliable activation of direct, but not indirect, targets of transcription factors in *Xenopus* (Kolm and Sive, 1995; Logan et al., 2005; Pozzoli et al., 2001; Schlosser et al., 2008; Seo et al., 2007). Once target lists are compiled, a small number of candidates deemed of high-interest for placode development will be selected for further verification and functional characterisation. Based on these findings, a gene regulatory network underlying sense organ development will be proposed, and network topology probed using gain- and loss-of-function experiments. Characterisation of such a network will have wide-reaching impacts not only on our basic understanding of how Six1 and Eya1 drive sense organ development, but also in potentially revealing interesting insights into factors involved in diseases stemming from their dysregulation, such as cancer and various craniofacial defects. Finally, the construction of such a network in a model vertebrate will provide a foundation for further work to explore the sort of re-wiring that might have taken place in the vertebrate ancestor that drove the evolutionary innovations of the vertebrate “new head”.

## Chapter 2. Materials and Methods

### 2.1 Animal housing and egg laying

The African clawed frog *Xenopus laevis* was used as a model organism throughout this study. Frogs were purchased from Nasco (Fort Atkinson, WI, USA). All animal experiments were performed in full accordance with Irish and European legislation, were approved by the NUI Galway Animal Care Research Ethics Committee (ACREC, 003/10) and were covered under the animal license (Cruelty to Animals Act, 1876) B100/4291 to G. Schlosser. Egg laying was induced by injection of human Chorionic Gonadotropin (hCG) into the dorsal lymph sac of female frogs (Sive et al., 2000). Each female was primed with 50 U (50 µl 1x hCG) at least five days prior to egg laying and then injected with 800 U (800 µl 1x hCG) approximately 12 hours prior to egg laying. Females were housed individually overnight in tank water at 17°C immediately before egg laying.

### 2.2 Embryo manipulation

#### 2.2.1 Microscopy

All embryo observation was performed using an Olympus SZX7 stereomicroscope. GFP screening and wholomount photography were performed using an Olympus SZX16 with an attached DP71 camera with fluorescent illumination capabilities. High-magnification photographs of sections were captured with an Olympus BX51 compound microscope and processed with the Olympus CellD software package, and later adjusted for light balance in Adobe Illustrator 6.

#### 2.2.2 *In vitro* fertilisation

On the day of fertilisation, females were transferred to 1x MBSH (appendix A.1) at room temperature (RT) and eggs were collected regularly (approximately once an hour) into a large petri dish. Males were anaesthetised in 0.1% MS 222 (Tricaine methanesulfonate) in tap water (pH 7.4) for at least 30 minutes and the testes were subsequently removed and stored in 1x MBSH at 4°C (for up to two weeks) and the cadaver frozen. Small pieces of testis were macerated with forceps and mixed with the eggs, and sperm were then activated by the addition of 0.1x MBS. Jelly coats were removed once the eggs were fertilised (as indicated by upward-facing pigmented animal poles; usually after around 15-20 minutes at RT) by cysteine treatment (4% in dH<sub>2</sub>O; pH 8) for approximately 10 minutes, and cysteine fully removed with

several rinses in 0.1x MBS. Embryos were incubated on a cold plate at 14°C (first cleavage occurs at 2.5 hours).

### 2.2.3 Microinjection of mRNA or morpholino antisense oligonucleotides (MO)

*Six1*-GR (500 pg), *Eya1*-GR, (500 pg), *Six1*-EnR (100 pg; Brugmann et al., 2004), Notch Intracellular Domain (*Notch-ICD*; 500 pg; Wettstein et al., 1997) and Suppressor of Hairless (Su(H)) DNA binding mutant (*Su(H)-DBM*; 500 pg; Wettstein et al., 1997) mRNAs and *Six1*-MO1+2 (*Six1*-MO1: 5'-GGAAGGCAGCATAGACATGGCTCAG-3'; *Six1*-MO2: 5'-CGCACACGCAAACACATAACACGGG-3'; Brugmann et al., 2004), *Eya1*-MO1+2 (*Eya1*-MO1: 5'-TACTATGTGGACTGGTTAGATCCTG-3'; *Eya1*-MO2: 5'-ATATTGTTCTGTCAGTGGCAAGTC-3'; Schlosser et al., 2008) and *Eya1*-mismatch-MO (*Eya1*-mmMO; mismatches shown in lower case: 5'-ATtITaGTTCTGaCAGTGGgAAcTC-3') morpholinos (MOs) were freshly prepared before each injection. While relatively high amounts of MO (20 ng and 16 ng of *Eya1*- or *Six1*-MO, respectively) were injected for RNA-Seq experiments, lower amounts were used in follow-up experiments (*Eya1*-MO: 2 ng; *Six1*-MO: 2 ng). *lacZ* (250 pg) or *myc-GFP* (125 pg) mRNAs were co-injected to mark the injected side. Agar dishes (4% agar with wells for eggs) were used for injection. Microelectrodes (10-15 µm tip) used for injection were prepared from glass capillaries (Narishige; cat no.: GD-1) on a microneedle puller (Narishige; cat no.: PN-31) using the following settings: (Heat 98.1; sub magnet 22.0; main magnet 94.1). Microelectrodes were attached to a microinjector (Narishige; cat no.: IM-300), and calibrated by injecting DEPC-H<sub>2</sub>O into a drop of mineral oil on a micro ruler and adjusting the injection diameter to 0.2-0.22 mm for a 5 nl injection volume. Solutions for injection were inspired into the microelectrode from a Parafilm covered glass slide. Approximately 100-200 embryos were transferred to an agar dish containing 5% Ficoll (in 0.1x MBS), and all liquid removed prior to injection of 5 nl per embryo. Depending on the experiment, either one or both blastomeres of a two-cell stage embryo were injected. After injection, the agar dish was flooded with 5% Ficoll and embryos transferred to a labelled petri dish containing 5% Ficoll and cultured on a cold plate at 14°C overnight. Embryos were transferred to 0.1x MBS the following morning, and staged according to Nieuwkoop and Faber, 1967.

### 2.2.4 DAPT treatment

Embryos were treated with N-[N-(3,5-Difluorophenacetyl)-L-alanyl]-S-phenylglycine t-butyl ester (DAPT) from stage 11 on. A stock solution of 10mM DAPT in DMSO was diluted

1:100 into 0.1x MBS for treatments. Control embryos were treated with 1:100 DMSO diluted into 0.1x MBS.

#### *2.2.5 Explanting pre-placodal ectoderm and treatment with cycloheximide/dexamethasone*

24 well cell culture dishes were pre-incubated with 0.5x MBS, 1% Bovine Serum Antibody (BSA), which was replaced by 2 ml 0.5x MBS immediately before explants were added. Vitelline membranes were removed from neural plate stage embryos using forceps in a plasticine dish filled with transplantation solution, and explants of the posterior pre-placodal ectoderm were cut using flame-sharpened tungsten needles, with care taken to remove mesodermal cells. Explants were transferred to a cell culture well containing 0.5x MBS + cycloheximide (CHX; dilution 1:2000; final concentration 10 µg/ml) and incubated at 25°C for 30 minutes. Half of these explants were then transferred to 0.5x MBS + CHX + dexamethasone (DEX; dilution 1:500; final concentration 10 µM) and incubated at 25°C for 2.5 hours. As a reference, sibling embryos kept at 25°C for 3 hours were at stage 20. Explants from the RNA-Seq and qPCR experiments were performed under the same conditions, however fewer explants were taken per biological replicate in the qPCR experiment (approximately 20 per treatment group, compared to approximately 50 in the RNA-Seq experiment).

#### *2.2.6 Trizol RNA Isolation (placodal explants)*

After removing as much liquid as possible, tissue was homogenised in 400 µl Trizol by pipetting until fully lysed. The sample was then centrifuged at 4°C, 10,000 x G for 10 minutes. Approximately 400 µl from the supernatant was recovered and transferred to a new Eppendorf tube. 100 µl chloroform was added (under hood) and the tube inverted vigorously before centrifugation at 4°C, 10,000 x G for 10 minutes. The aqueous phase was carefully removed by tilting the tube to 45° and pipetting off the clear aqueous phase, taking care not to include the thick white protein layer, and approximately 220 µl was recovered into a new Eppendorf tube. 1:1 Isopropanol was added before vortexing and incubated at 20°C for at least 30 minutes. RNA was pelleted by centrifugation at RT, 11,000 x G for 7 minutes and washed twice after removal of the supernatant with 400 µl ice-cold 70% ethanol. The clean RNA pellet was completely dried by first manually removing any remaining EtOH with a small pipette, and then allowing it to dry at RT for 10 minutes with the lid open. Pelleted RNA was dissolved in 15 µl Tris elution buffer (TE; 10 mM pH 8.0; pre-heated to 65°C) for 5 minutes and stored at -20°C. RNA was quality assayed using a BioAnalyser 2100. All RNAs sent for sequencing had an RNA Integrity Number (RIN) > 7.0 with one exception (*Eya1*-

MO biological replicate 3; RIN = 5.30). The latter, however, yielded 82 M high quality RNA-Seq reads and thus was included in the analysis.

### *2.2.7 Fixation of embryos*

Embryos injected with *myc-GFP* were sorted under a fluorescent stereomicroscope prior to fixation. Embryos were fixed in 4% w/v paraformaldehyde (PFA) in 0.1 M PB for 30-45 minutes at RT and rinsed in 0.1 M PB twice for 10 minutes. Unless subjected to X-gal staining (see section 2.2.8), embryos were then rinsed in 50% EtOH for 10 minutes before being stored at 4°C in 70% EtOH.

### *2.2.8 X-gal staining*

Embryos were fixed as above and rinsed in 0.1 M PB twice for 10 minutes. Before dehydration in EtOH, lacZ staining was revealed by treatment with X-gal staining solution in the dark at 37°C until blue staining is apparent. Embryos were then rinsed in 0.1 M PB, 50% EtOH and stored in 70% EtOH as described in section 2.2.7.

## **2.3 General molecular techniques**

### *2.3.1 cDNA synthesis*

Placodal explants and total RNAs were obtained as described (see sections 2.2.5 and 2.2.6). cDNA was synthesised using the QuantiTect Reverse Transcription Kit (Qiagen; cat no.: 205311) according to the manufacturer's instructions, using 500 ng total RNA in a 20 µl reaction.

### *2.3.2 PCR*

PCR was performed using the Phusion® High-Fidelity PCR Kit (New England BioLabs; cat no.: E0553S) according to the manufacturer's instructions. Each 20 µl reaction contained: 4 µl 5X Phusion HF buffer; 0.4 µl 10 mM dNTPs; 1 µl 10 µM forward primer; 1 µl 10 µM reverse Primer; variable template DNA; 0.2 µl MgCl<sub>2</sub>; 0.2 µl Phusion DNA Polymerase; nuclease-free H<sub>2</sub>O to 20 µl. Typical cycling conditions as used in a G-Storm GS1 Thermal Cycler were 98°C for 30 seconds; 30 cycles of 98°C for 5-10 seconds, 45°C-72°C for 30 seconds, 72°C for 30 seconds; 72°C for 1-2 minutes (appendix B).

### *2.3.3 Colony PCR*

Colony PCR was used to amplify plasmid DNA from several bacterial colonies in order to

determine presence and orientation of insert prior to midiprep. A PCR master mix was prepared as described above (see section 2.3.2) in 10 µl reactions, omitting DNA. A single colony was picked with a sterile 0.1 µl pipette tip and added to an individual PCR tube to provide a plasmid DNA template.

#### *2.3.4 Quantitative PCR*

qPCR was performed using TaqMan® reagents (Applied Biosystems; cat no.: 4352042) according to the manufacturer's instructions, using 2 µl cDNA (5 ng/µl), 8 µl Master mix (final concentration 900 nM forward primer; 900 nM reverse primer; 300 nM probe). The following cycling conditions were used in a StepOnePlus™ Real-Time PCR System (cat no.: 4376600): 95°C for 10 minutes; 40 cycles of 95°C for 15 seconds, 55°C for 1 minute. Primers and probes (with added 5' FAM, 3' TQ2) were designed using Primer3 (version 2.3.4; Rozen and Skaletsky, 2000) to quantify gene expression. Relative gene expression was calculated after normalisation using *Smn2* as a reference (Dhorne-Pollet et al., 2013). qPCR was performed in triplicate and the entire experiment was repeated three times from independently prepared RNA. Relative Quantification (RQ) values and log<sub>2</sub> fold change (FC) were averaged across biological replicates.

#### *2.3.5 Restriction digestion*

Restriction digestion was carried out in 50 µl reactions (10 µl plasmid (at 1 µg/µl); 10 units restriction enzyme; 5 µl restriction buffer; adjusted to 50 µl with dH<sub>2</sub>O) and incubated at 37°C for several hours or overnight (or according to manufacturer's instructions; Fisher).

#### *2.3.6 DNA purification*

DNA was purified using the QIAquick PCR purification Kit according to the manufacturer's instructions (Qiagen; cat no.: 28104). DNA was eluted in TE buffer preheated to 65°C.

#### *2.3.7 RNA purification*

RNA was purified using the RNeasy Mini Kit according to the manufacturer's instructions (Qiagen; cat no.: 74106). RNA was eluted in RNase-free H<sub>2</sub>O preheated to 65°C.

#### *2.3.8 Gel electrophoresis*

1.2% Agarose gels were made in 1x TAE buffer with 2 µl SybrSafe (10000x; Invitrogen) added at approximately 50°C. Gels were allowed to cool in-chamber for around 30 minutes before being flooded with 1x TAE and loaded with DNA (1 µl loading buffer + 10 µl DNA; 3 µl

DNA ladder). Gels were run at 90 V for approximately 45 minutes (or until lower blue bands approached the end of the gel).

### 2.3.9 Agarose gel DNA extraction

≤ 1% Agarose gels were prepared as detailed above (see section 2.3.8). Bands were separated under UV illumination and DNA purified using the Promega Wizard® SV Gel and PCR Clean up Kit according to the manufacturer's instructions (Promega; cat no.: A9282). DNA was eluted in TE buffer.

### 2.3.10 Subcloning and gene synthesis

The full coding region of *Hes5/8*, *Crem*, *Atoh1*, *Tbx15*, *FosB* and *Is/2* based on transcript models from RNA-Seq data (GenBank: KT722743; KT722744; KT722747; KT722746; KT722745; KT722748) and flanked by suitable restriction sites (see below), was synthesised by Genscript into the cloning vector pUC57. Subsequently it was sub-cloned into the expression vector pCS2+ after restriction digestion with enzymes cutting at flanking restriction sites (3', 5') as follows: *Hes5/8* and *Crem*: ClaI/EcoRI; *Atoh1*: XbaI/XbaI *Tbx15* and *FosB*: BamHI/EcoRI; *Is/2*: EcoRI/StuI. Primers with added ClaI and EcoRI sites (to the forward and reverse primers, respectively) were designed (appendix B) to amplify the entire coding region of *Tbx6*, which was then subcloned into pCS2+ between the ClaI/EcoRI sites. *Znf214*, *Mab21l2b* and *Pou3f2b* were ordered (pCMV-SPORT6, Fisher Scientific; Clone IDS: 5512398, 5515985 and 4203106). *Hes9* (pCR4-TOPO) was ordered from Source Bioscience (Clone accession: BC169570) and was subcloned into pCS2+ in the EcoRI site.

### 2.3.11 Ligation

DNA was ligated using T4 DNA ligase according to the manufacturer's instructions (Thermo; cat no.: EL0014), using approximately 5x more insert than plasmid (e.g. 10 ng insert with 50 ng plasmid in a 10 µl reaction).

### 2.3.12 Bacterial transformation

2.5% Luria Broth (LB) was prepared in dH<sub>2</sub>O in 100 ml Erlenmeyer flasks, sealed with aluminium foil and autoclaved. Agar plates were prepared from freshly autoclaved 3.7% LB agar in dH<sub>2</sub>O, and supplemented with an appropriate antibiotic (e.g. ampicillin 100 µg/ml) when cold enough to touch. Plates were then poured and allowed to cool at RT and stored upside down at 4°C. XL1-Blue competent cells (Agilent; cat no.: 200249) were thawed on ice and transformed according to manufacturer's instructions. Bacterial cells were concentrated

following recovery by centrifugation at 4,000 rpm for 3 minutes and 50 µl of the bottom 10% of resulting cell suspension was plated onto 37°C pre-warmed LB agar plates using a flame-sterilised tool. Plates were sealed and bred upside down at 37°C overnight.

### 2.3.13 Midiprep

Discrete colonies were picked using a sterile pipette held between flame-sterilised forceps into Erlenmeyer flasks containing 100 ml LB supplemented with the appropriate antibiotic (e.g. ampicillin 100 µg/ml). Bacteria were grown at 37°C for 12-16 hours shaking at 250 rpm. Plasmids were extracted using the NucleoBond® Xtra Midi Kit according to the manufacturer's instructions (Macherey-Nagel; cat no.: 740410.10). Pelleted DNA was dissolved in 100 µl TE buffer, preheated to 65°C, and transferred to labelled Eppendorf tubes.

### 2.3.14 Capped mRNA synthesis for microinjection

mRNA was synthesised from 1 µg of cleaned and linearised plasmids using the Ambion mMessage mMachine Kit according to manufacturer's instructions (Ambion; cat no.: AM1340) and purified as detailed above (see section 2.3.7). Synthesised mRNAs were adjusted to a final concentration of 500 ng/µl with RNase-free H<sub>2</sub>O, aliquotted and stored at -20°C.

### 2.3.15 RNA probe synthesis for *in-situ-hybridisation*

Antisense oligonucleotide probes were synthesised from 1 µg of cleaned, linearised plasmids using the DIG RNA Labelling Kit according to manufacturer's instructions (Roche; cat no.: 11175025910). Synthesised probes were purified as detailed above (see section 2.3.7) and adjusted to a final concentration of 100 ng/µl with hybridisation buffer and stored at -20°C. Wholemount *in-situ-hybridisation* was carried out as described (see section 2.4.1) using digoxigenin-labelled antisense probes for *Six1*, *N-tub*, *Sox2*, *Sox3*, *Ripply3*, *Hes2*, *Sim1*, *Lhx5*, *Sox21*, *Emx1.2*, *Tlx1*, *Hes5/8*, *Hes9*, *Gfi1a*, *Tbx15*, *Ngn1*, *Pou4f1.2*, *Isl2*, *Znf214*, *Mab21l2b*, *Pou3f2b*, *Tbx6*, *FosB*, *Crem* and *Atoh1*. Primers were designed with promoter sites added (forward, T7; reverse, SP6) for *Hes5/8*, *Hes9*, *Gfi1a*, *Tbx15*, *Ngn1*, *Pou4f1.2* and *Isl2* and were used to amplify a ~800bp fragment from plasmid DNA which was then used as a template for probe synthesis using T7 RNA polymerase to make an antisense probe (table 2.1).

**Table 2.1** Vector and probe synthesis information for constructs used

<b>Gene</b>	<b>Vector</b>	<b>Linearise</b>	<b>Probe</b>	<b>Source</b>
<i>Hes2</i>	pBS-CMV	EcoRI	T7	Solter, 2006
<i>Lhx5</i>	pGE-MT	NotI/NdeI	T7	Bachy et al., 2001
<i>Pou4f1.2</i>	pCS2+MT	PCR from pCS2+MT	T7	Hutcheson and Vetter, 2001
<i>Tlx1</i>	Bluescript SK+	Xhol	T3	Patterson and Krieg, 1999
<i>Sim1</i>	pCS108	Sall	T7	Martin et al., 2007
<i>Ripply3</i>	pCDG1	NotI	T7	Janesick et al., 2012
<i>Gbx2</i>	BLUESCRIPT SK+	SmaI	T7	Bubnoff et al., 1996
<i>Sox21</i>	pGEM	BamHI	T7	Cunningham et al., 2008
<i>Gfi1a</i>	pCS111	PCR from pCS111	T7	Ciau-Uitz et al., 2010
<i>Emx1.2</i>	pCMV-SPORT6	EcoRI	T7	Green and Vetter, 2011
<i>Sox2</i>	pCS2+	EcoRI	T7	De Robertis et al., 1997
<i>Sox3</i>	pBK-CMV	EcoRI	T7	Penzel et al., 1997
<i>Six1</i>	pBSII SK	NotI	T7	Pandur and Moody, 2000
<i>N-Tub</i>	pCS2+	BamHI	T3	Oschwald et al., 1991
<i>Ngn1</i>	pCS2+	PCR from pCS2+	T7	Ma et al., 1998
<i>Tbx6</i>	pCS2+	BamHI	T7	PCR
<i>Hes5/8</i>	pCS2+	PCR from pCS2+	T7	Accession: KT722743
<i>Crem</i>	pCS2+	BamHI	T7	Accession: KT722744
<i>FosB</i>	pCS2+	BamHI	T7	Accession: KT722745
<i>Tbx15</i>	pCS2+	PCR from pCS2+	T7	Accession: KT722746
<i>Atoh1</i>	pCS2+	NotI	SP6	Accession: KT722747
<i>Isl2</i>	pCS2+	PCR from pCS2+	T7	Accession: KT722748
<i>Mab21l2b</i>	pCMV-SPORT6	Sall	T7	Clone ID: 5515985
<i>Pou3f2b</i>	pCMV-SPORT6	Sall	T7	Clone ID: 4203106
<i>Znf214</i>	pCMV-SPORT6	Sall	T7	Clone ID: 5512398
<i>Hes9</i>	pCS2+	PCR from pCS2+	T7	Accession: BC169570

## 2.4 Wholemount *in-situ*-hybridisation and immunohistochemistry

### 2.4.1 Wholemount *in-situ*-hybridisation

*In-situ*-hybridisation was conducted in RNase-free conditions, in autoclaved glass vials, with all solutions prior to the RNase treatment step on day two either DEPC treated and autoclaved, or prepared with DEPC-H<sub>2</sub>O. Fixed embryos were rehydrated in 50% EtOH and 30% EtOH for 5 minutes each, and washed three times for 5 minutes in Ptw (1x PBS + 0.1% Tween-20). Embryos were treated with Proteinase K (PK) for 8 minutes (20 µl 25 mg/ml PK in 50 ml Ptw) and rinsed twice in 0.1 M TEA (last step in 4 ml) for 5 minutes each (appendix A.2). 0.5 ml TEA/acetic anhydride (250 µl acetic anhydride in 10 ml 0.1 M TEA) was added, and vials were rocked for 5 minutes before another 0.5 ml TEA/acetic anhydride was added and incubated for 5 minutes. This was followed by washing twice in Ptw for 5 minutes each. Embryos were re-fixed in MEMFA for 20 minutes and washed five times in Ptw for 5 minutes each. Embryos were transferred to screw capped tubes during the final Ptw wash and 1 ml Ptw added. 250 µl hybridisation buffer was added and the entire solution removed after the embryos settled before a further 250 µl hybridisation buffer was added and embryos were incubated at 60°C for 10 minutes. Hybridisation buffer was again removed and replaced with 250 µl fresh hybridisation buffer and embryos were incubated at 60°C for 6 hours. This was replaced with 250 µl of digoxigenin-labelled RNA probes diluted 1:100 in hybridisation buffer

(to a final concentration of 1 ng/μl) and returned to 60°C.

After overnight incubation, the probe was saved and replaced with 200 μl hybridisation buffer and embryos incubated at 60°C for 10 minutes before washing with 2x SSC three times for 20 minutes at 60°C (for this and subsequent washes 1 ml solution per vial was used). 2x SSC was replaced with freshly prepared 2x SSC + RNase A (10 μl [2 mg/ml]) + 0.1 μl RNase T1) and incubated for 30 minutes at 37°C. Embryos were then washed at RT in 2x SSC for 10 minutes, twice in 0.2x SSC at 60°C for 30 minutes each and twice in MAB for 10 minutes total. Embryos were then pre-incubated for 1 hour in MAB + 2% BBR and then 1 hour in MAB + 2% BBR + 20% HIGS. Embryos were then incubated overnight in MAB + 2% BBR + 20% HIGS with 1:1000 AP-coupled, anti-digoxigenin antibody (Roche) in the dark at 4°C.

Antibody incubation solution was removed, and embryos washed in MAB five times for 1 hour each before washing twice in freshly prepared AP buffer for 5 minutes each. AP buffer was replaced by AP buffer + NBT (4.5 μl [100 mg/ml] in 10 ml AP buffer) + BCIP (35 μl [50 mg/ml] in 10 ml AP buffer) and embryos were incubated at RT in the dark until staining was apparent, at which point the reaction was stopped by washing in MAB three times for 5 minutes each. Embryos were re-fixed overnight (2.5 ml Formaldehyde; 0.5 ml glacial acetic acid; 7.0 ml dH<sub>2</sub>O).

Background staining was removed by washing in 100% EtOH three times for 30 minutes each, and 70% EtOH, 50% EtOH for 5 minutes each before embryos were bleached in bleaching solution at RT until pigmentation was fully removed. Embryos were then washed in 1x SSC and 50% EtOH for 5 minutes each and stored at 4°C in 70% EtOH.

#### *2.4.2 Immunostaining on whole mounts*

Embryos were rehydrated in 50% EtOH and PB for 10 minutes each and washed twice in PBS for 1 hour each before the addition of primary antibody (rabbit anti-lacZ, 1:1000: Cappel; cat no.: 55976; mouse anti-p53, 1:200: antibody X77 from Abcam; cat no.: ab16465) in normal goat serum with 5% DMSO for two days. Primary antibody was then removed by washing in PBS five times for 1 hour each before incubation overnight in the dark in secondary antibody (lacZ: 1:1000 Alexa594-conjugated anti-rabbit antibody; Molecular Probes; cat no.: A31631. p53: 1:1000 Alexa488-conjugated anti-mouse antibody; Molecular Probes; cat no.: A-21121). Secondary antibody was then removed by washing in PBS five times for 1 hour each. Embryos were analysed and imaged in PBS, before long-term storage in 70% glycerol with

0.2% sodium azide at 4°C.

### *2.4.3 Vibratome sectioning*

Post *in-situ*-hybridisation, selected embryos were rehydrated in 50% and 30% EtOH and finally PB for 30 minutes each. Embryos were carefully and thoroughly dried with paper tissues before being embedded in semi-cooled 4% Agar in phosphate buffer (PB). Agar was allowed to harden before being trimmed to allow the correct orientation of embryos on the mounting platform of a Leica VT1000S vibratome. 50 µm sections were cut and mounted on slides (Fisher Superfrost™ Plus; cat no.: 12-550-15) and dried overnight before sealing with transparent nail polish.

## **2.5 Bioinformatic analysis of RNA-Seq data**

Unless otherwise noted, scripts referred to between single quotation marks were developed in-house. Scripts ending with the extension ‘.pl’ were written in Perl, and those with ‘.r’ written in R (Breiman, 2001). All scripts mentioned, as well as the general bioinformatic pipeline, are detailed in appendix C.

### *2.5.1 Initial quality assessment and filtering*

Fastqc (<http://www.bioinformatics.babraham.ac.uk/projects/fastqc/>) was run on all read pairs to visually inspect read quality. Trimmomatic (Bolger et al., 2014) was run on read pairs for each biological replicate to quality-trim forward and reverse reads simultaneously, preserving read pairings. Standard Illumina format (-phred33) was specified, and primer/adapter sequences were removed. Leading and trailing low quality bases (below quality score 3) were removed, and the reads were scanned with a 4-base wide sliding window, trimming when the average quality per base dropped below 15. Additionally, the first 12 bases from the start of each read were removed, as were reads shorter than 36 bases after trimming.

### *2.5.2 Align reads to genome*

Bowtie2 (version 2.2.5; Langmead and Salzberg, 2012) was used to generate a genome index from a draft version of the *Xenopus laevis* genome (version XI7; N50 = 19.3kb; <http://www.xenbase.org/other/static/ftpDatafiles.jsp>). Tophat2 (2.0.13; D. Kim et al., 2013), a splice junction mapper for RNA-Seq, was used to align reads to the genome reference using the default settings but specifying a mate-inner-distance of 250 (estimated empirically using Stampy (Lunter and Goodson, 2011)), before analysing mapping results to identify splice

junctions between exons. Mapping statistics were generated using Samtools (version 1.2; H. Li et al., 2009).

### *2.5.3 Transcript assembly and differential expression*

Cufflinks (version 2.1.1; Roberts et al., 2012) was used to assemble transcripts and estimate their abundance using default settings, following default ‘classic-fpkm’ library normalisation. Cuffdiff (version 2.1.1; Roberts et al., 2012) was used to estimate differential expression between the CHX treatment and the CHX+DEX treatment, specifying biological replicates for each treatment group, using the default geometric library normalisation.

### *2.5.4 Estimating variance between biological replicates*

‘pearsons.pl’, was written to provide  $\log_2$  transformed FPKM (Fragments Per Kilobase of exon per Million fragments mapped) values for two biological replicates in a given condition. To normalise the data, FPKM values of 0 and  $> 5000$  were removed. These values were then used to estimate the Pearson’s correlation of replicates in R.

### *2.5.5 Generating a scatterplot to visualise FPKM distribution*

‘merger.pl’ was developed to curate Cuffdiff output for each condition, attaching directional information. ‘scatterplot.grid.r’ was used to extract FPKM values and information on direction of differential expression and generate a scatterplot of CHX FPKM vs. CHX+DEX FPKM values.

### *2.5.6 Annotating transcript models*

‘transcripts.pl’ was used to build transcripts from the genome using output from Cufflinks. Another script ‘de.genes.pl’ was then used to annotate transcripts with differential expression information. These annotated transcripts were then BLASTed against a *Xenopus* mRNA database (*X. laevis*: <ftp://ftp.xenbase.org/pub/Genomics/Sequences/xlaevisMRNA.fasta>; *X. tropicalis*: <ftp://ftp.xenbase.org/pub/Genomics/Sequences/xtropMRNA.fasta>) using blastn with an e-value cut-off of 1E-5. For each query, only the hit with the lowest e-value was considered. These were then related to sequence information and prepared for downstream analysis using ‘annotator.pl’. During further characterisation, several transcripts initially identified as ‘uncharacterised’ or ‘hypothetical’ during annotation were manually re-annotated using blastn for presentation in final tables (tables 3.2 and 3.3). In such cases, the original annotations are maintained in appendices E, F and I.

### *2.5.7 Gene enrichment analysis*

‘enrichment.pl’ was used to estimate the extent to which the top 10% of expressed transcripts (ranked by FC) in one condition were enriched in the top 10% of expressed transcripts in another condition. The output is given as values and co-ordinates to construct a 2 x 2 contingency table for each pairwise comparison, from which enrichment was determined using Fisher’s exact test in R.

### *2.5.8 Finding co-differentially expressed genes*

‘co-reg.pl’ was used to find transcripts that were differentially regulated in at least two conditions. This program performed two major functions that will be detailed below: (1) to pool identically annotated transcripts in an attempt to try to overcome the problem of mapping reads to a draft genome of a tetraploid and, (2) to identify co-differentially expressed transcripts among treatment groups in the absence of reliable statistics (i.e. when specifying two replicates in Cuffdiff).

#### *2.5.8.1 Addressing genomic redundancy and tetraploidy in X. laevis*

In an attempt to address several major problems of performing an RNA-Seq experiment in *X. laevis*, ‘co-reg.pl’ first extracts and combines FPKM values for transcripts that return identical annotations. By default, Bowtie2 will assign reads to a single transcript in the genome, with reads mapping identically to more than one location randomly assigned to one of these regions. Thus, both artificial redundancy in the assembly, and genuine redundancy in the tetraploid genome, will result in populations of reads derived from a single transcript to be distributed among a number of transcript models. In addition, long (100 bp) paired-end reads often overlap in sequencing, which at least partially negates their ability to link together exons and thus, resolve full-length transcript models. In these cases, several transcript models are returned for each transcript and are annotated identically. By summing FPKM values for identically annotated transcripts, and calculating the FC from these pooled values, this program aims to more accurately report differential expression dynamics, at the cost of not being able to distinguish pseudoalleles.

#### *2.5.8.2 Excluding DEX-induced genes*

In order to exclude transcripts activated by DEX, rather than as a result of the overexpression of *Six1*, *Eya1* or *Six1+Eya1*, transcripts differentially expressed in individual experimental conditions (injected with either *Six1-GR*, *Eya1-GR* or *Six1-GR+Eya1-GR*; treatment with CHX alone vs. CHX + DEX) were compared to those differentially expressed in the control

(un-injected; treatment with CHX alone vs. CHX + DEX). Transcripts showing less than a two-fold difference between the control and experimental FC values were excluded from further analysis.

#### *2.5.8.3 Identifying common differentially expressed genes*

‘co-reg.pl’ also acts to determine differential expression where no reliable statistic can be produced (on account of using two experimentally treated biological replicates). Transcripts were considered differentially expressed if (1) the (summed) FPKM for that transcript was  $> 1$  in the DEX treatment group and (2) the transcript was at least two-fold up-regulated/down-regulated in DEX treated groups. Furthermore, in order to prioritise transcripts activated by several conditions for further analysis, only those that met the above criteria, as well as being at least two-fold up-regulated in at least two out of three experimental conditions (individual injection of *Six1* [*Six1<sub>i</sub>*], *Eya1* [*Eya1<sub>i</sub>*] or *Six1+Eya1* [*Six1+Eya1<sub>i</sub>*]) were considered.

As a second approach to finding genes that showed similar differential expression between treatment groups, RNA-Seq data were merged to add statistical power to the analysis. In one analysis, all six experimental conditions were considered as biological replicates to focus on genes with similar differential expression profiles across all conditions (comprising the *Six1+Eya1<sub>m</sub>* dataset). In another analysis, all treatment groups involving *Six1* overexpression (i.e. injection of *Six1* alone or *Six1+Eya1*; *Six1<sub>m</sub>*) or *Eya1* overexpression (*Eya1*, *Six1+Eya1*; *Eya1<sub>m</sub>*) conditions were treated as replicates in order to look for genes preferentially activated by *Six1* or *Eya1*. In these instances, transcripts were considered differentially expressed if they met the above criteria as well as passing Cuffdiff’s default statistical test (where reliable q-values were available), using a pooled dispersion estimation model.

#### *2.5.9 Gene set enrichment analysis and gene ontology*

‘sets.pl’ was used to find discrete gene sets using output from ‘co-reg.pl’. These sets were then converted to their human orthologs by blasting against the Human Uniprot database with blastx with an e-value cut-off of 1E-2, and functionally annotated using the online tools ‘PantherDB’ (Mi et al., 2012; <http://pantherdb.org>) and ‘DAVID’ (Huang:2009be; Huang et al., 2009a; <https://david.ncifcrf.gov>), specifying an un-injected, CHX treated, placodal transcriptome as a background set. The enrichment score (E) for each group is reported as the geometric mean of the EASE scores (a modified Fisher’s exact) that are associated with the enriched annotation terms belonging to that group (Huang et al., 2007).

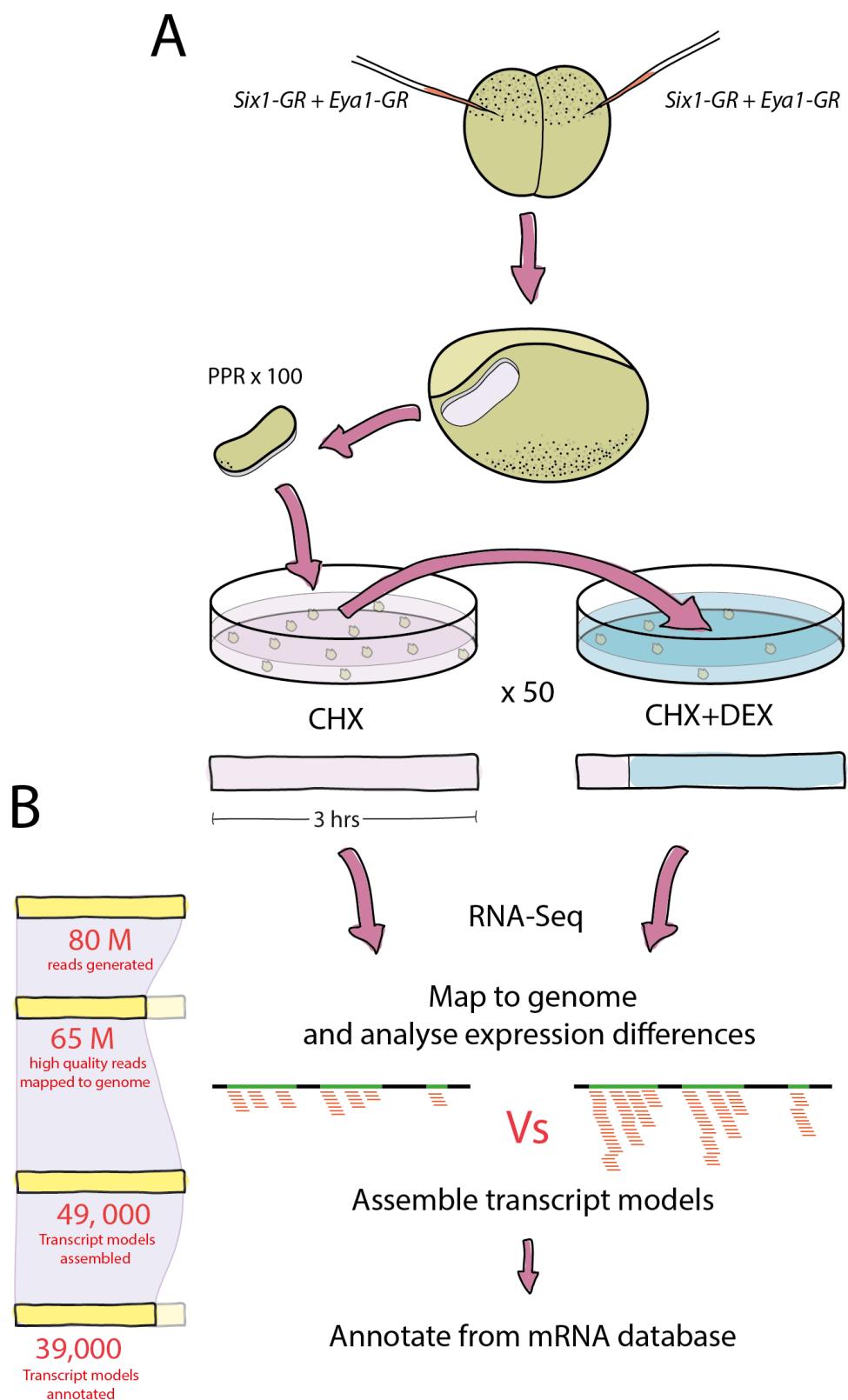
### *2.5.10 Phylogenetic analysis of Hes8 to determine orthology*

Amino acid residues of vertebrate Hes genes (see appendix G.1 for sequence accession numbers) were aligned across the conserved basic helix-loop-helix (bHLH) and Orange domains. Positions with alignment gaps in a majority of the sequences were removed and the regions concatenated, resulting in an alignment (fig. 4.1) of 83 residues shown here as a 50% consensus with invariant sites underlined and concatenated regions separated by dashes 5' - RK-KPJVEKRRRARINXSLXQLKXLLXXXXXXSXRXSKLEKADILEMTVXXL-  
GXXCLXEXXXFLSXXXGXBXXXXXXLLXHL- 3'. Phylogenetic relationships of the genes were estimated via Bayesian inference (Ronquist and Huelsenbeck, 2003) using the WAG (Whelan and Goldman, 2001) amino acid substitution model including gamma among-site rate variation. Analyses were run for 1,100,000 generations, with samples saved every 200 generations, and the first 50,000 generations eliminated as 'burn-in' prior to forming a consensus tree using the sumt command. In addition to the Bayesian approach, a Maximum Likelihood (ML) tree was also generated using RaxML (Stamatakis et al., 2005) to verify topology, and is shown in appendix G.2.

## Chapter 3. Revealing direct targets of Six1 and Eya1 in the PPE

### 3.1 Introduction

Six1 and Eya1 are known to play central roles in the development of placodes from the pre-placodal ectoderm (PPE) but little is currently known about the target genes that mediate these effects (Brugmann et al., 2004). To identify such targets, *Xenopus laevis* embryos were injected with *Six1*-GR and *Eya1*-GR either alone or in combination in three different treatment groups: *Six1*-GR alone, *Eya1*-GR alone and *Six1*-GR +*Eya1*-GR co-injected. PPE was then explanted from neural plate stage embryos and, to ensure only direct targets were activated, explants were treated with the protein synthesis inhibitor cycloheximide (CHX; see section 2.2.5). Explants were split into two directly comparable groups; one treated with dexamethasone (DEX) in the presence of CHX, to conditionally overexpress fusion proteins and thus induce transcription of direct targets only, and the other treated only with CHX, to serve as a direct control for DEX-induced overexpression (fig. 3.1A). An RNA-Seq screen was then conducted on RNA extracted from explants, using two biological replicates for each treatment group, and differential gene expression was analysed between explants treated with CHX alone against those treated with both CHX and DEX. RNA-Seq was also performed on explants taken from un-injected embryos treated with CHX or CHX+DEX to control for the effect of DEX treatment by itself and on un-injected and un-treated embryos, in order to fully characterise the transcriptome of the PPE.



**Fig. 3.1** Experimental pipeline and overview of bioinformatic analysis. **(A)** Both blastomeres of two-cell stage embryos were injected with *Six1-GR*, *Eya1-GR* or *Six1-GR* and *Eya1-GR* and explants were cut from pre-placodal ectoderm. Explants were incubated in CHX for 30 minutes before being split into two groups; 50% were kept in CHX for 2.5 hours and 50% were transferred to CHX+DEX for 2.5 hours. RNA was extracted from both treatment groups and submitted to RNA-Sequencing. **(B)** On average, 80 million reads were generated in sequencing for each treatment group, and 65 million quality trimmed reads were successfully mapped to the *Xenopus* genome. An average of 49,000 transcript models were assembled, of which 80% (39,000) were successfully annotated against a *Xenopus* mRNA database, and differential expression analysis was then performed on annotated transcripts using CHX treated explants as a control for those treated with CHX+DEX.

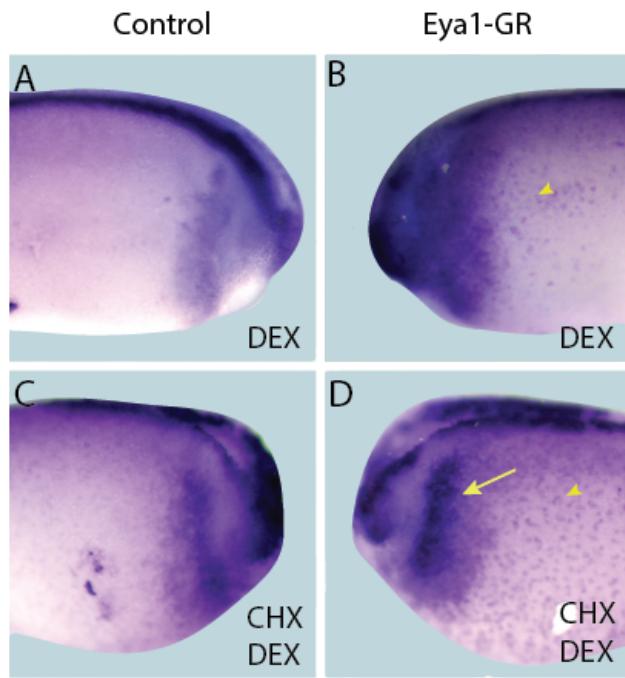
### **3.2 Conditional overexpression of *Six1* and *Eya1* allows only direct targets to be transcribed in the presence of a protein synthesis inhibitor**

#### *3.2.1 GR fusion proteins allow conditional overexpression in the presence of dexamethasone*

Six1 and Eya1 proteins fused to the human glucocorticoid receptor (GR) hormone-binding domain were used to allow conditional activation in neural plate stages. In the absence of DEX, GR-fusion proteins are sequestered in the cytoplasm where they remain transcriptionally inactive (Kolm and Sive, 1995). However, the addition of DEX rapidly activates GR-fusion transcription factors, allowing them to translocate to the nucleus and regulate target gene expression. In order to reproduce previous findings and validate this system for conditional overexpression, the expression of *Sox3*, a predicted target of Eya1 (Schlosser et al., 2008), was analysed after injection of *Eya1*-GR and DEX activation at neural plate stage (fig. 3.2A and B). Consistent with published results, *Eya1*-GR injection and treatment with DEX promoted extensive ectopic expression of *Sox3*.

#### *3.2.2 Cycloheximide and protein synthesis inhibition*

In order to reveal only direct targets of Six1, Eya1 and Six1+Eya1, DEX-induced overexpression was combined with CHX treatment, which blocks translation, and thus the induction of non-direct targets. In combination with conditional activation of fusion proteins, this system efficiently identifies direct target genes (Kolm and Sive, 1995; Logan et al., 2005; Pozzoli et al., 2001; Seo et al., 2007). To reproduce the finding that *Sox3* is a direct target gene of Eya1 (Schlosser et al., 2008), *Sox3* expression was analysed after injection of *Eya1*-GR and, in combination with CHX, treated with DEX at neural plate stage (fig. 3.2C and D). Phenotypes observed were similar to those observed for embryos only treated with DEX (fig. 3.2B), with Eya1 maintaining the ability to ectopically induce *Sox3* during CHX treatment, corroborating *Sox3* as a direct target of Eya1. Also consistent with previous findings was that the ectopic expression of *Sox3* was stronger and broader in CHX treated embryos, suggesting that there are indirect inhibitory effects on *Sox3* that are alleviated when protein synthesis is blocked.

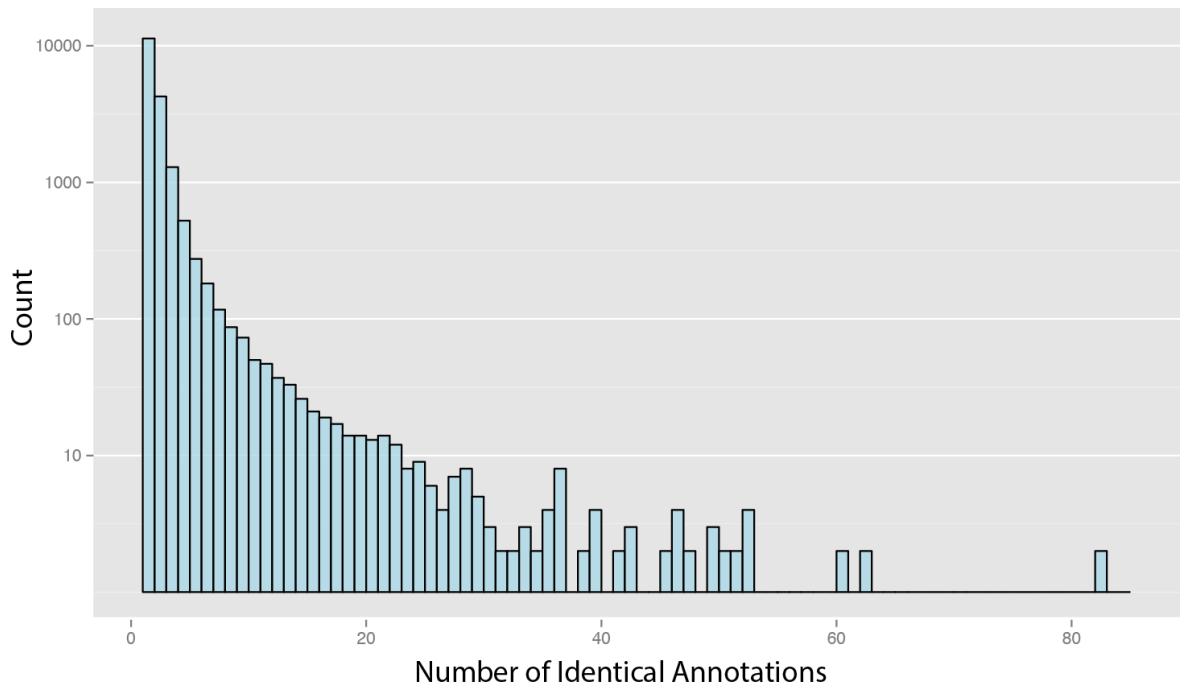


**Fig. 3.2** Effects of overexpression of *Eya1-GR* on *Sox3* expression in early tail bud (stage 22) embryos. Control (**A** and **C**) and injected (**B** and **D**) sides of tail bud stage embryos after unilateral injection of *Eya1-GR* and DEX activation in the absence (**A** and **B**) or presence (**C** and **D**) of CHX at stage 16-18. Arrows mark increased or ectopic *Sox3* expression in the placodal or non-neural ectoderm (arrowheads).

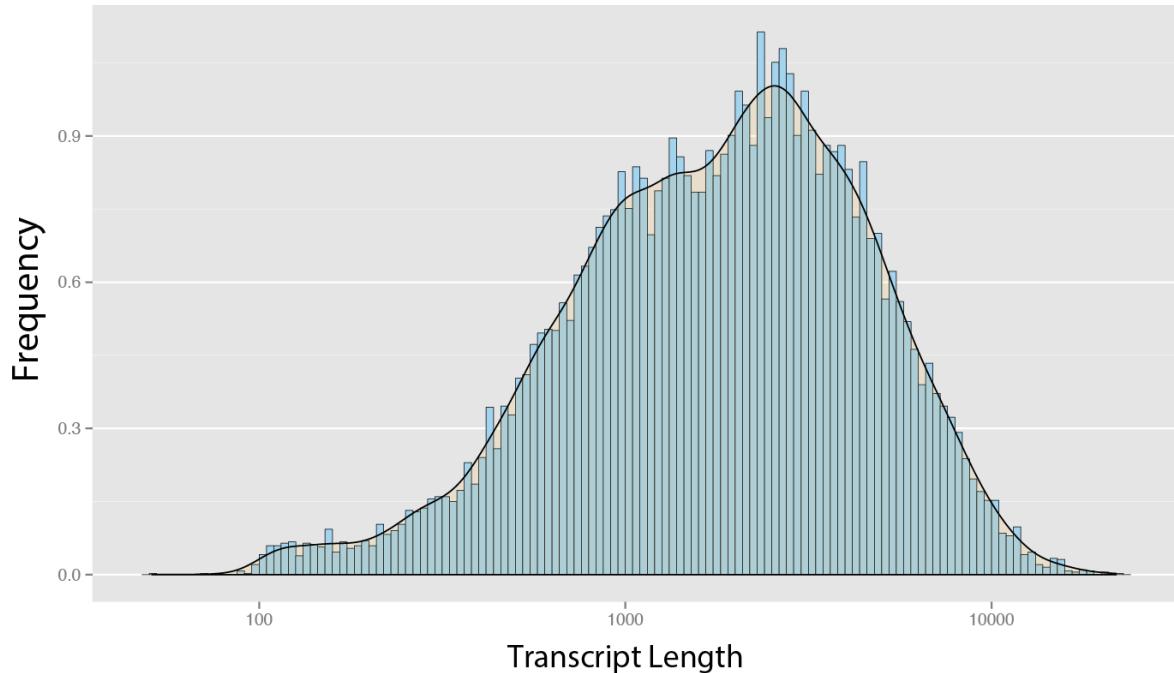
### 3.3 The pre-placodal transcriptome dissected

RNA was extracted from PPE explants cut from un-injected embryos and characterised using RNA-Seq (fig. 3.1A). On average, this generated 80 million paired-end reads, reduced to 72 million reads after quality trimming. 65 million reads were then successfully paired and mapped to a draft version of the *Xenopus laevis* genome (version XI7). Initially, 49,000 transcript models were assembled and annotated against a *Xenopus* blast database, with 80% of transcript models successfully annotated. However, as a large number of these annotations were redundant (see section 2.5.8.1 for further details), with 72% of annotated transcript models sharing the same annotation with at least one other, and 41% with at least four others (likely due to the combined effects of pseudotetraploidy and incompleteness of the genome assembly; fig. 3.3), and 5,039 (25%) of these expressed at levels insufficient for consideration in analysis ( $\text{FPKM} < 1$ ), the number of expressed, unique transcript models was reduced to 15,794, with an average length of 2,813bp and GC content of 43% (fig. 3.4). Gene set enrichment analysis was then conducted on the 1,000 most highly expressed transcripts (appendix D) to explore general characteristics of the PPE. Most notably, RNA processing/splicing was very highly enriched in the PPE data set (enrichment score [E]: 43), suggesting that RNA-binding proteins and mRNA splicing mechanisms may play an

important role in placodal development. Translation elongation and ribosomal proteins were also highly enriched (E: 32), perhaps reflecting the high rate of protein turnover in the rapidly changing PPE (McCabe et al., 2004). Other enriched terms include ubiquitination (E: 10.7), DEAD-box (E: 5), neurogenesis (E: 2.1) and cell cycle (E: 2). Other than neurogenesis and cell cycle, few of the terms were typically associated with placodal development, possibly due to the high number of housekeeping genes found in the most highly expressed genes.



**Fig. 3.3** Distribution of number of non-unique annotations for assembled transcript models.



**Fig. 3.4** Distribution of read lengths of uniquely annotated transcript models from the placodal transcriptome. Blue bars show the read length frequency, and the orange overlay shows the smoothed kernel density estimation, which represents the average value over a specified binwidth.

### 3.4 An RNA-Seq screen to reveal the direct targets of Six1 and Eya1 in the PPE

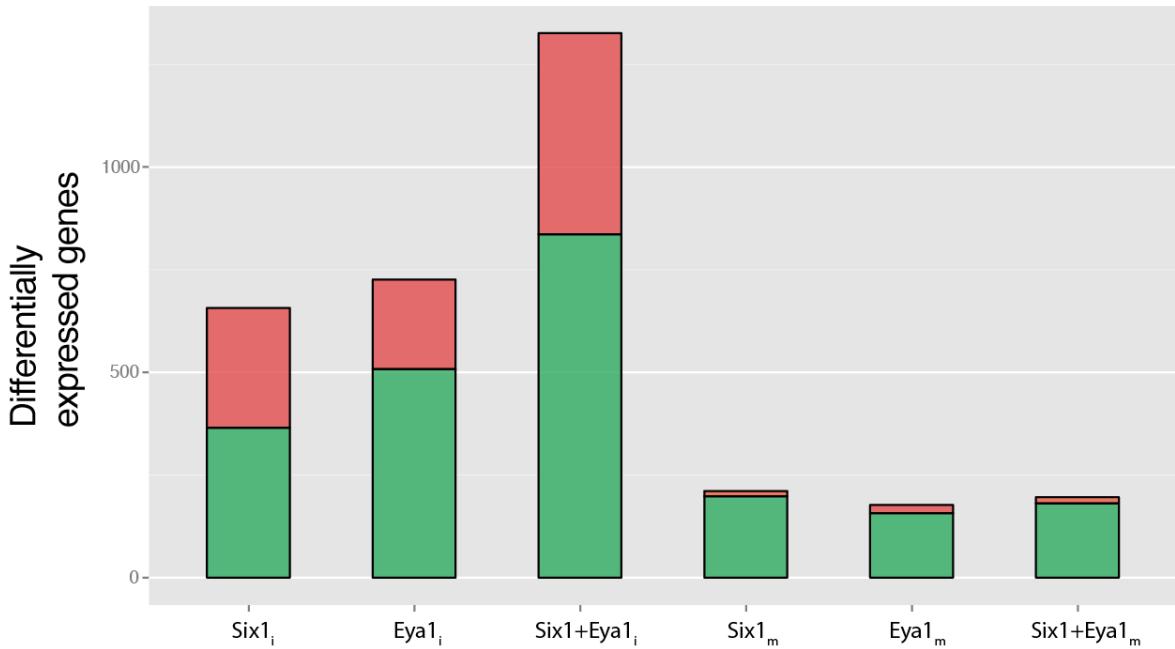
#### 3.4.1 Assessment of sequencing data and rationale for analysis of merged treatment groups

As a preliminary estimate of how well correlated biological replicates were within treatment groups, a Pearson's correlation was calculated for each condition. Biological replicates showed high correlation ( $> 0.9$ ) in each pairwise comparison, indicating the similarity of expression profiles between independently treated samples. Although replicates appeared to show little variability, differential expression analysis conducted for each treatment group revealed few significantly affected ( $q < 0.05$ ) genes, probably reflective of using only two biological replicates. In such cases, and particularly with treated samples (where both the control [CHX] and experimental [CHX+DEX] conditions were chemically treated) from dissected tissue, minor variations between dissected area, injection volume, treatment potency and concentration would introduce variation in expression profiles of target genes. In two biological replicates, there is insufficient data available to resolve this variation and the statistics returned will underestimate the number of significantly affected genes. Therefore, FC values were prioritised over statistical significance in order to rank differentially expressed genes in analysis of individual treatment conditions. As a means to augment these initial analyses with statistical information, treatment conditions were then merged together in groups of either six (all treatment groups) or four (all treatment groups involving injection of *Six1*; all treatment groups involving injection of *Eya1*) biological replicates (see sections 2.5.8.3

and 3.4.3) and re-analysed in order to produce more accurate statistics against which the initial analysis could be compared.

### 3.4.2 Direct targets of Six1 and Eya1

To identify direct targets of Six1 and Eya1, Six1-GR and Eya1-GR fusion proteins were overexpressed either alone or in combination in PPE. In combination with a protein synthesis inhibitor (CHX), nuclear translocation of Six1 and Eya1 was induced by adding DEX for 2.5h, and gene expression was analysed using RNA-Seq. To preclude the inclusion of genes affected by DEX treatment alone, explants taken from un-injected embryos were treated as above (CHX vs. CHX+DEX) and included in sequencing. Direct targets of Six1, Eya1 or Six1+Eya1 were determined by comparing *Six1*-GR, *Eya1*-GR or *Six1*-GR+*Eya1*-GR injected embryos treated with CHX (as controls) against CHX+DEX treated samples. Resultant data sets from these individual treatment groups will henceforth be referred to as Six1<sub>i</sub>, Eya1<sub>i</sub> and Six1+Eya1<sub>i</sub>. After removing genes differentially expressed in the control CHX+DEX treatment group (and thus affected by DEX alone; see section 2.5.8.2), 365 genes that satisfied criteria for differential expression (see section 2.5.8.3) were up-regulated in Six1<sub>i</sub>, 508 in Eya1<sub>i</sub> and 836 in Six1+Eya1<sub>i</sub>, whereas 292 were down-regulated in Six1<sub>i</sub>, 218 in Eya1<sub>i</sub> and 490 in Six1+Eya1<sub>i</sub> (fig. 3.5; appendix E). To check for data set quality, both known (*Slc12a2* (Ando et al., 2005); *CyclinA1* (Coletta et al., 2004); *CyclinD1* (Z. Li et al., 2013); *c-Myc* (X. Li et al., 2003); *Ezrin* (Yu et al., 2006); *Gdnf* (X. Li et al., 2003); *Sox3* (Schlosser et al., 2008)) and predicted (*Atoh1* (T. Zhang et al., 2006); *Sox2* (Schlosser et al., 2008); *Sall1* (Chai, 2006); *MyoD1* (Liu et al., 2013)) targets of Six1 were searched for in the pre-placodal transcriptome as well as in Six1<sub>i</sub> and Six1+Eya1<sub>i</sub> data sets. With the exception of *c-Myc*, all genes were present in the transcriptome, and most were found in either Six1<sub>i</sub> (*CyclinD1* [ccndx] FC: 7.48; *Slc12a2*, FC: -2.75; *CyclinA1*, FC: -3.68; *Sox2*, FC: 1.2; *MyoD*, FC: 3.4) or Six1+Eya1<sub>i</sub> (*Sox3*, FC: 0.9; *Atoh1*, FC: 1.4; *Sall1*, FC: 1) data sets, indicating their fidelity.

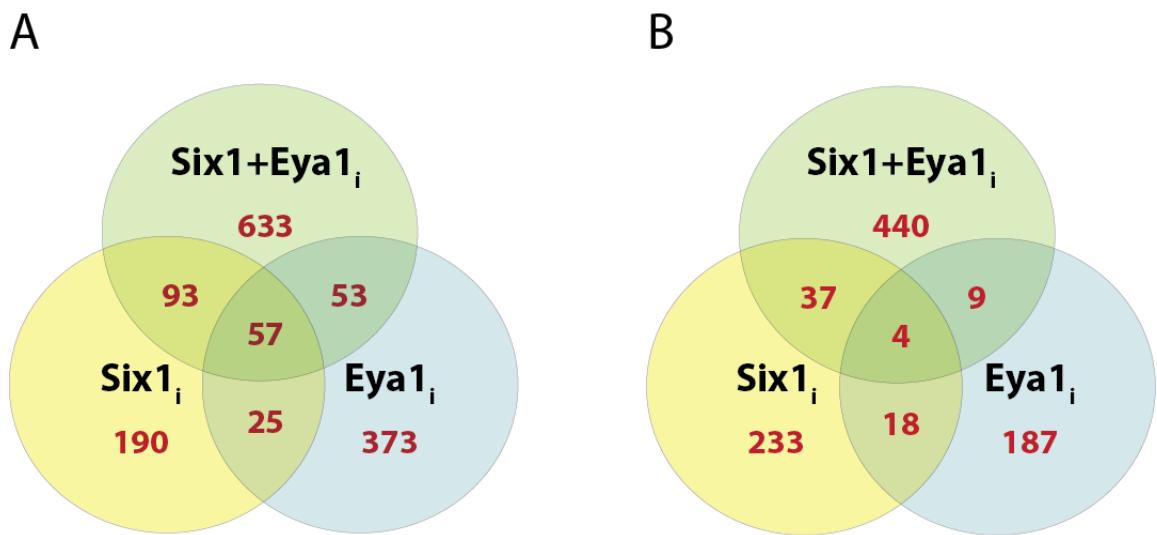


**Fig. 3.5** Number of genes differentially expressed in each condition. Green bars show the number of up-regulated genes per condition and red bars show down-regulated genes. Condition names succeeded by a subscript 'i' represent individual treatments (injected with either *Six1*, *Eya1* or *Six1+Eya1*), whereas those succeeded by a subscript 'm' show merged treatments. Refer to text in sections 3.4.2 and 3.4.3 for further details. 365 genes were up-regulated in *Six1<sub>i</sub>*, 508 in *Eya1<sub>i</sub>* and 836 in *Six1+Eya1<sub>i</sub>* whereas 292 genes were down-regulated in *Six1<sub>i</sub>*, 218 in *Eya1<sub>i</sub>* and 490 in *Six1+Eya1<sub>i</sub>*. When combined and filtered for statistical significance ( $q < 0.05$ ), substantially fewer differentially expressed genes were reported: 149 genes were up-regulated in *Six1<sub>m</sub>* (*Six1* alone or co-injected with *Eya1*), 112 after *Eya1<sub>m</sub>* overexpression (*Eya1* alone or co-injected with *Six1*) and 183 in *Six1+Eya1<sub>m</sub>* (*Six1*, *Eya1*, *Six1+Eya1*), with even fewer down-regulated genes found with 11 in *Six1<sub>m</sub>*, 13 in *Eya1<sub>m</sub>* and 14 in *Six1+Eya1<sub>m</sub>*.

### 3.4.3 Many direct targets are co-regulated by *Six1* and *Eya1*

*Six1* and *Eya1* are known to operate both independently (Blevins et al., 2014; Brugmann et al., 2004; P. X. Xu et al., 2002) and synergistically (Ahmed et al., 2012a; Brugmann et al., 2004; Christophorou et al., 2009; X. Li et al., 2003; Ruf et al., 2004) to regulate transcription. To gain further insight into the extent to which target genes are co-regulated by *Six1* and *Eya1*, genes responding similarly between treatment groups were examined. Of the up-regulated genes, 57 were found in all treatment groups, and 93 shared only between *Six1<sub>i</sub>* and *Six1+Eya1<sub>i</sub>*, 53 between *Eya1<sub>i</sub>* and *Six1+Eya1<sub>i</sub>* and 25 between *Six1<sub>i</sub>* and *Eya1<sub>i</sub>* (fig. 3.6A). While this supports previous findings that *Six1* and *Eya1* act co-operatively in regulating target genes, there are also many genes that appear to be affected exclusively in individual treatment groups, with 190 up-regulated in *Six1<sub>i</sub>*, 373 in *Eya1<sub>i</sub>* and 633 in *Six1+Eya1<sub>i</sub>*. In addition to acting as a transcriptional enhancer, *Six1* also acts as a repressor in a context and cofactor dependent fashion (Brugmann et al., 2004; X. Li et al., 2003). This is perhaps reflected in the proportion of genes down-regulated exclusively in *Six1<sub>i</sub>* (233) compared to those up-regulated (190; fig. 3.6): Alone, *Six1* represses more genes than it activates, possibly due to its interaction with

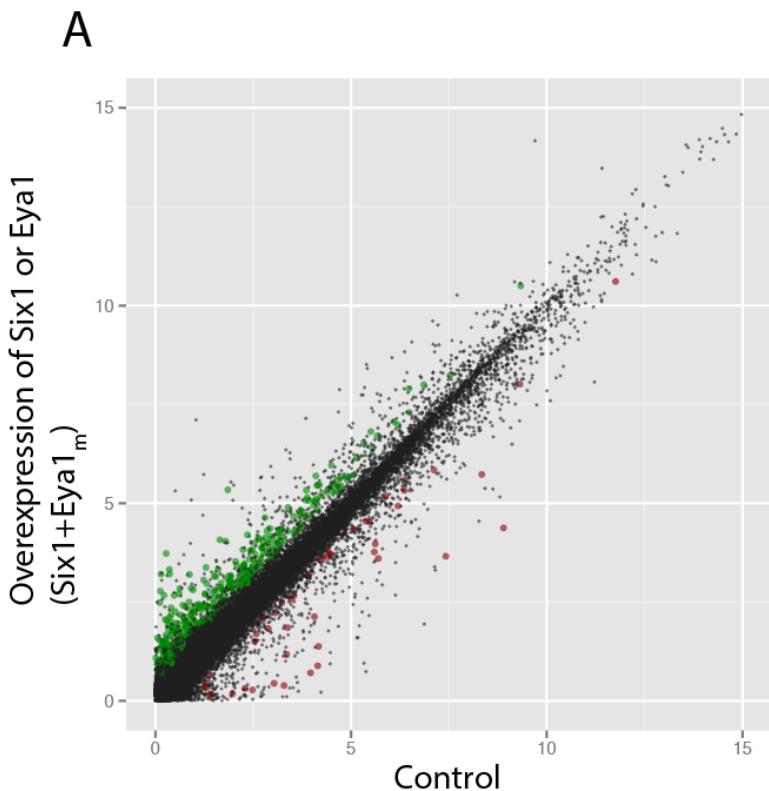
non-Eya binding partners, whereas very few genes appear to be down-regulated co-operatively (4 in all treatment groups; 37 between Six1<sub>i</sub> and Six1+Eya1<sub>i</sub>; 9 between Eya1<sub>i</sub> and Six1+Eya1<sub>i</sub>; 18 between Six1<sub>i</sub> and Eya1<sub>i</sub>), as shown in figure 3.6B, consistent with Eya1 binding having a positive effect on transcription.

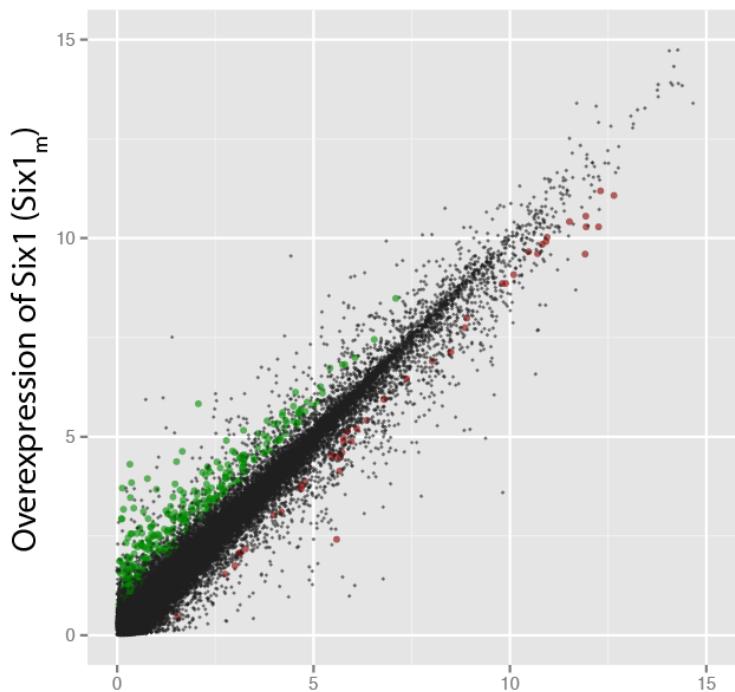
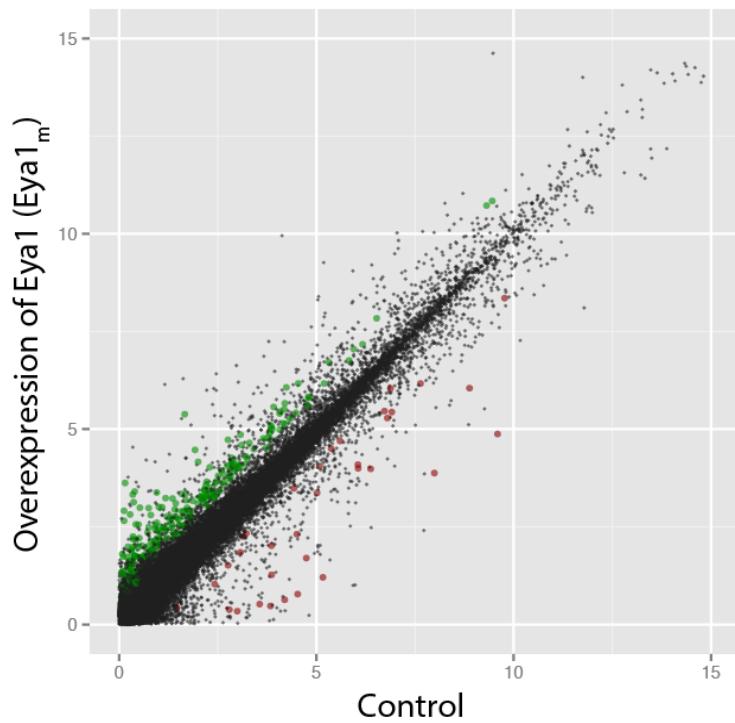


**Fig. 3.6** Genes differentially regulated after overexpression of *Six1* alone (*Six1*; yellow), *Eya1* alone (*Eya1*; blue) or *Six1* and *Eya1* combined (*Six1+Eya1*; green). Venn diagrams show numbers of genes (red) unique for each treatment group or shared between them. **(A)** Number of genes up-regulated and **(B)** down-regulated after injection with *Six1*, *Eya1* or *Six1+Eya1*.

To further examine whether direct targets of any one treatment were similarly affected in other treatment groups, pairwise comparisons were made and tested for significance. Direct targets of Six1, Eya1 and Six+Eya1 are highly enriched for direct targets of the other experimental treatment groups ( $p < 0.0001$ ; Fisher's exact test). When compared to the control group, no enrichment was found in any of the treatment groups Six1<sub>i</sub> ( $p = 0.16$ ), Eya1<sub>i</sub> ( $p = 0.12$ ) or Six1+Eya1<sub>i</sub> ( $p = 0.12$ ).

In an attempt to scrutinise this enrichment, and to add more statistical weight to the analyses by combining replicates, data were merged together and re-analysed (see section 2.5.8.3). Initially, all treatment groups were merged ( $\text{Six1}_i$ ,  $\text{Eya1}_i$ ,  $\text{Six1+Eya1}_i$ ; henceforth  $\text{Six1+Eya1}_m$ ) in order to look for genes regulated by either Six1 or Eya1. Next, data sets were merged in which either Six1 ( $\text{Six1}_i$ ,  $\text{Six1+Eya1}_i$ ; henceforth  $\text{Six1}_m$ ), or Eya1 ( $\text{Eya1}_i$ ,  $\text{Six1+Eya1}_i$ ; henceforth  $\text{Eya1}_m$ ) were overexpressed to discover genes individually regulated by Six1 and Eya1, respectively. In all cases, more genes were significantly affected ( $q < 0.05$ ) in the merged analyses compared to the individual analyses, with 183 significantly up-regulated genes found in  $\text{Six1+Eya1}_m$ , 149 in  $\text{Six1}_m$  and 112 in  $\text{Eya1}_m$  (appendix F). In addition, another known direct target of Six1, *Gdnf* (Z. Li et al., 2013; not found to be differentially expressed in individual conditions), was identified as one of the most highly significantly differentially expressed genes in the  $\text{Six1}_m$  data set ( $\text{Six1}_m$  FC: 5.8, q-value: 0.009), confirming the utility of re-analysis. Strikingly, substantially fewer genes were negatively regulated in merged groups, with 14 significantly down-regulated genes found in  $\text{Six1+Eya1}_m$ , 11 in  $\text{Six1}_m$  and 13 in  $\text{Eya1}_m$  (figs. 3.5 and 3.7), re-enforcing the notion that Six1 and Eya1 together act as transcriptional activators. As in the individual analyses, differentially expressed genes under merged experimental conditions were not found enriched in DEX treated controls ( $\text{Six1}_m$ ,  $p = 0.72$ ;  $\text{Eya1}_m$ ,  $p = 0.25$ ;  $\text{Six1+Eya1}_m$ ,  $p = 0.5$ ).



**B****C**

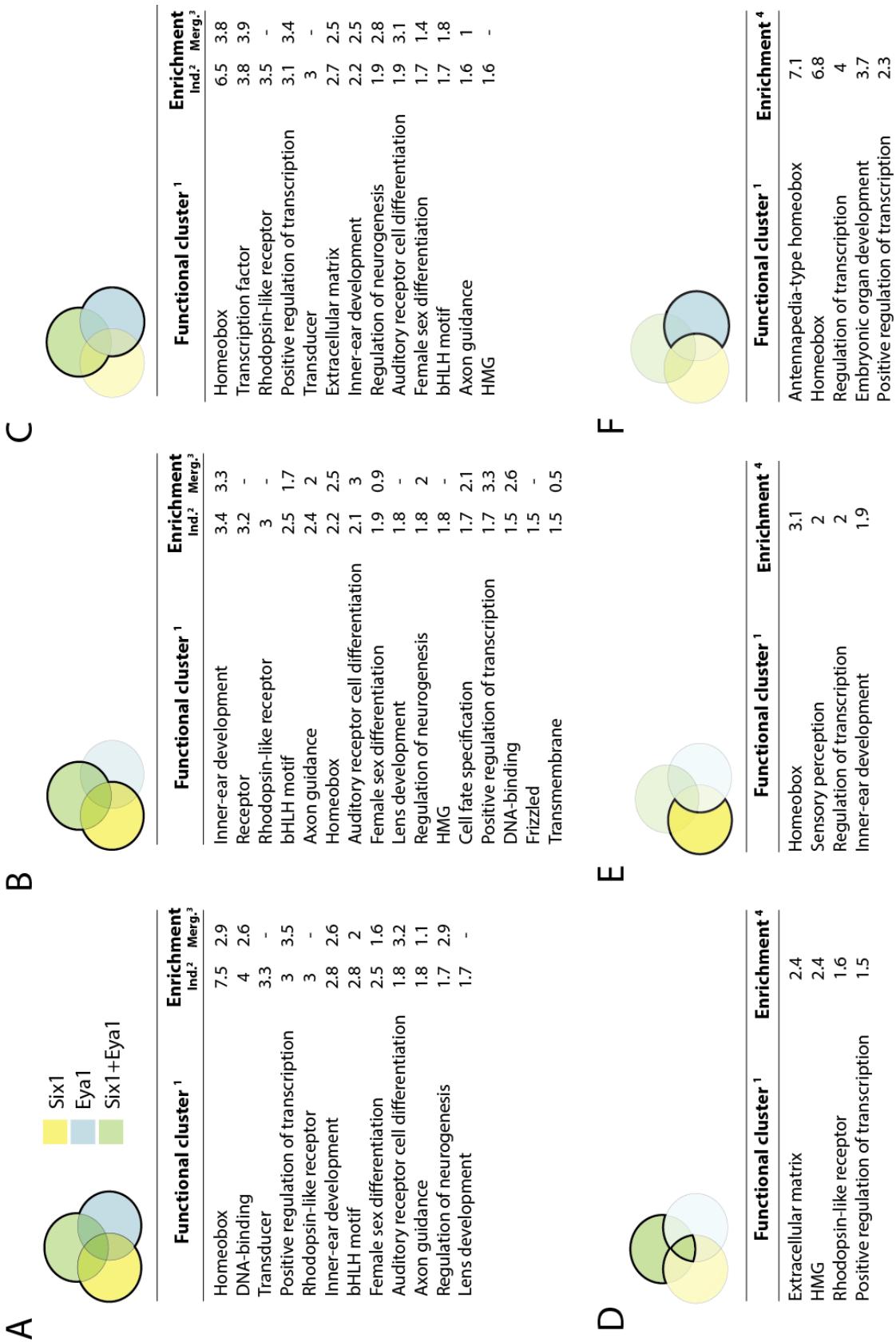
**Fig. 3.7** Pre-placodal transcriptome data set reveals hundreds of genes up-regulated in cranial placodes. Plots show  $\log_2$  transformed (FPKM +1) values after overexpression of (A) *Six1* or *Eya1* (combination of all treatment groups; *Six1+Eya1<sub>m</sub>*); (B) *Six1* (overexpression of *Six1* alone or in combination with *Six1+Eya1*; *Six1<sub>m</sub>*) and (C) *Eya1* (overexpression of *Eya1* alone or in combination with *Six1+Eya1*; *Eya1<sub>m</sub>*). Green dots represent significantly ( $q < 0.05$ ) up-regulated genes and red dots show significantly down-regulated genes.

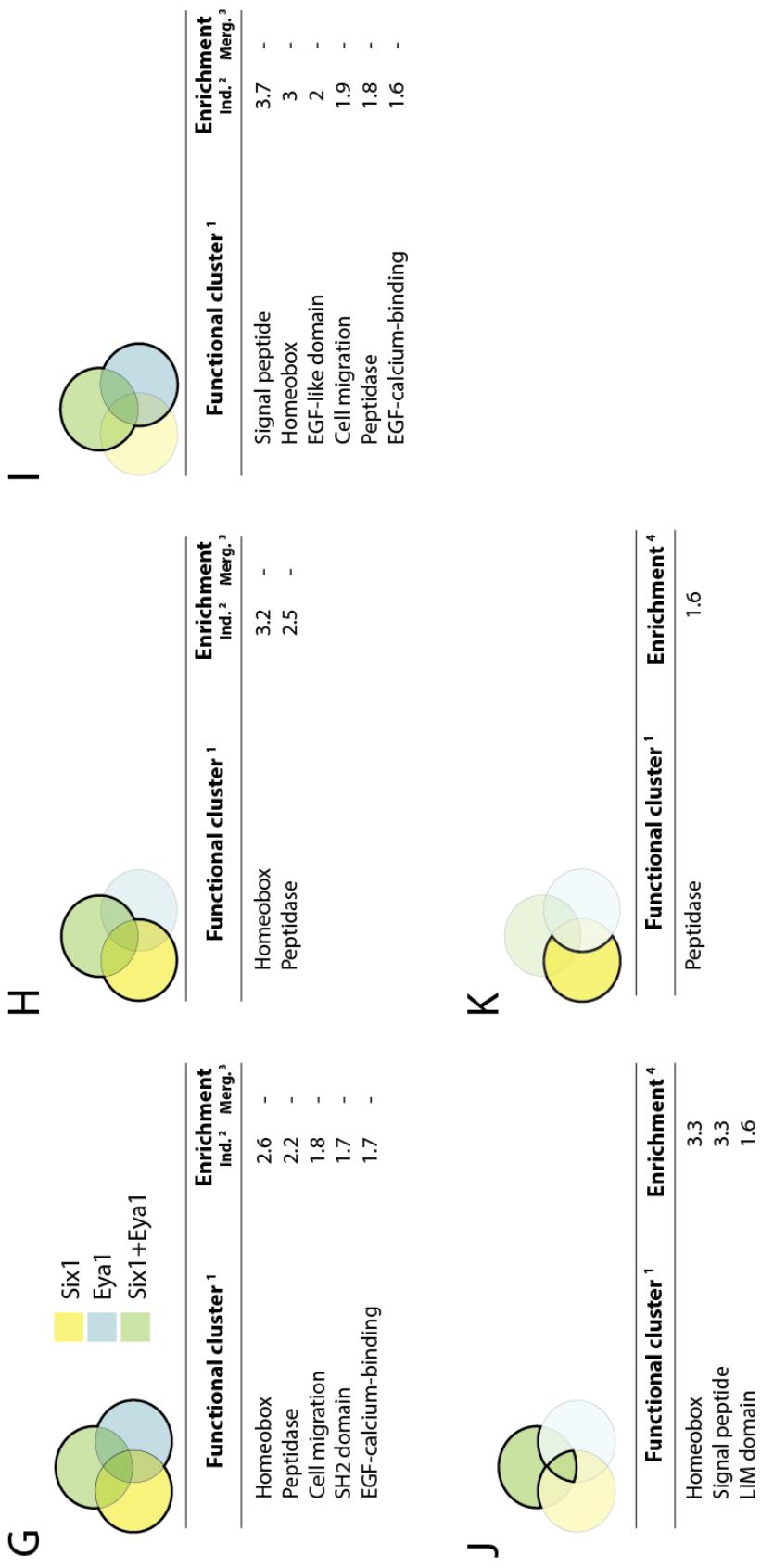
### 3.5 Characterisation of RNA-Seq data

#### 3.5.1 Gene Ontology and gene set enrichment analysis of individual and merged data sets

Gene set enrichment analysis was conducted on differentially expressed genes returned from both individual and merged analyses using DAVID (Huang et al., 2009a; 2009b) and the results are shown in figure 3.8. Overall, terms enriched in the individual data were highly consistent with those enriched in the merged data set, indicating that the categories of genes revealed by the analysis of individual data were strongly supported by the statistical significance of the merged data. Genes directly up-regulated by Six1, Eya1 and Six1+Eya1 show high enrichment for similar biological processes (inner-ear development, auditory receptor cell differentiation, neurogenesis and axon guidance) and molecular functions (DNA-binding and positive regulation of transcription) consistent with their synergistic role in sensory development (Grocott et al., 2012; Schlosser, 2010) and neurogenesis (Maier et al., 2014; Schlosser and Northcutt, 2000).

In order to drill-down on processes driven by different combinations of treatments, several different gene sets were considered. Initially, gene set enrichment analysis was performed on the combined data sets of individual treatment conditions (i.e. Six1<sub>i</sub>, Eya1<sub>i</sub>, Six1+Eya1<sub>i</sub>) to give a general overview of processes driven by either *Six1* or *Eya1* overexpression. Highly enriched terms in the up-regulated data (fig. 3.8A) included homeobox (E: 7.5), DNA-binding (E: 4), inner-ear development (E: 2.8), auditory receptor (E: 1.8), regulation of neurogenesis (E: 1.7) and lens development (E: 1.7), all of which are typical for placode development, and are known to be regulated by Six1 and Eya1 (sensu Schlosser, 2010). Next, sets of genes were considered that excluded either Eya1-induced (fig. 3.8B) or Six1-induced (fig. 3.8C) genes from analysis, in order to reveal processes driven primarily by Six1 or Eya1, respectively. In both cases, and consistent with the data analysed together, enriched terms were typical for placode development and regulation by Six1 and Eya1. Gene set enrichment analyses on the merged datasets (Six1<sub>m</sub>, Eya1<sub>m</sub>, Six1+Eya1<sub>m</sub>) revealed similar profiles (fig. 3.8A-C).





**Fig. 3.8** Gene Set Enrichment Analysis using DAVID, showing clusters of functional terms for different treatment groups. In each case, treatment groups considered are highlighted and outlined in bold in the accompanying Venn diagram. Yellow colouring indicates Six1 treatment; blue shows Eya1 and green Six1+Eya1. Enrichment scores  $\geq 1.5$  are reported for individual conditions (Ind.) and, where available,  $\geq 0.5$  for merged conditions (Merg.). **(A-F)** Genes up-regulated in the various conditions; **(G-K)** genes down-regulated in the various conditions. **(A, G)** All treatment groups included in analysis; **(B, H)** Six1 overexpression only; **(C, I)** Eya1 overexpression only; **(D, J)** genes differentially expressed exclusively after combined overexpression of Six1 and Eya1; **(E, K)** exclusively after Six1 overexpression; **(F)** exclusively after Eya1 overexpression.

<sup>1</sup> Functional cluster of terms that show enrichment

<sup>2</sup> Enrichment for individual conditions; Six1<sub>i</sub>, Eya1<sub>i</sub> and Six1+Eya1<sub>i</sub>

<sup>3</sup> Enrichment for merged conditions; Six1<sub>m</sub>, Eya1<sub>m</sub> and Six1+Eya1<sub>m</sub>

<sup>4</sup> Enrichment for individual conditions only

While these analyses provide insights into processes driven by Six and Eya1 individually and those co-regulated by Six1 and Eya1, they do not allow the determination of which genes are specifically up-regulated by Six1 but not Eya1 or vice versa. For example, the set of genes directly activated in the presence of Six1, but not Eya1, could only be delimited by removing all subsets of Six1<sub>i</sub> that intersect with Eya1<sub>i</sub> or Six1+Eya1<sub>i</sub>. Therefore, to investigate processes driven by Six1 or Eya1 alone, or those that require joint expression of both *Six1* and *Eya1* (target genes for which both Six1 and Eya1 are limiting factors), gene enrichment analysis was also conducted on discrete subsets as depicted in fig. 3.8 D-F). Six1 appears to drive processes associated with placodal development both independent of (sensory perception E: 2; inner ear development, E: 1.9), and in partnership with, Eya1 (positive regulation of transcription, E: 1.5). Particularly interesting however, was the extreme enrichment of homeobox genes (E: 6.8, and specifically those belonging to the Antennapedia-type class; E: 7.1) in the Eya1-specific subset of up-regulated genes (fig. 3.8F). Indeed, of the top 20 genes in this set (when ranked by FC), ten are homeobox genes (table 3.1), suggesting that Eya1 acts independently of Six1 in regulating homeobox, in particular Hox, gene expression. This implies an exciting, and previously un-identified, role for Eya1 in regulating Hox gene expression.

Down-regulated genes were also considered for each of these gene sets. Interestingly, similar to the up-regulated data, homeobox genes appear to be strongly enriched in most data sets, but particularly in the Six1-treatment set (fig. 3.8H; E: 3.2). Several other terms that are down-regulated include peptidases in the Six1-treatment set (E: 2.5), cell migration in the Eya1-treatment set (fig. 3.8I; E: 1.9) and signal peptides in the Six1+Eya1-treatment set (fig. 3.8J; E: 3.3), which may indicate interesting and un-explored repressive modes of action for Six1 and Eya1 in developing placodes.

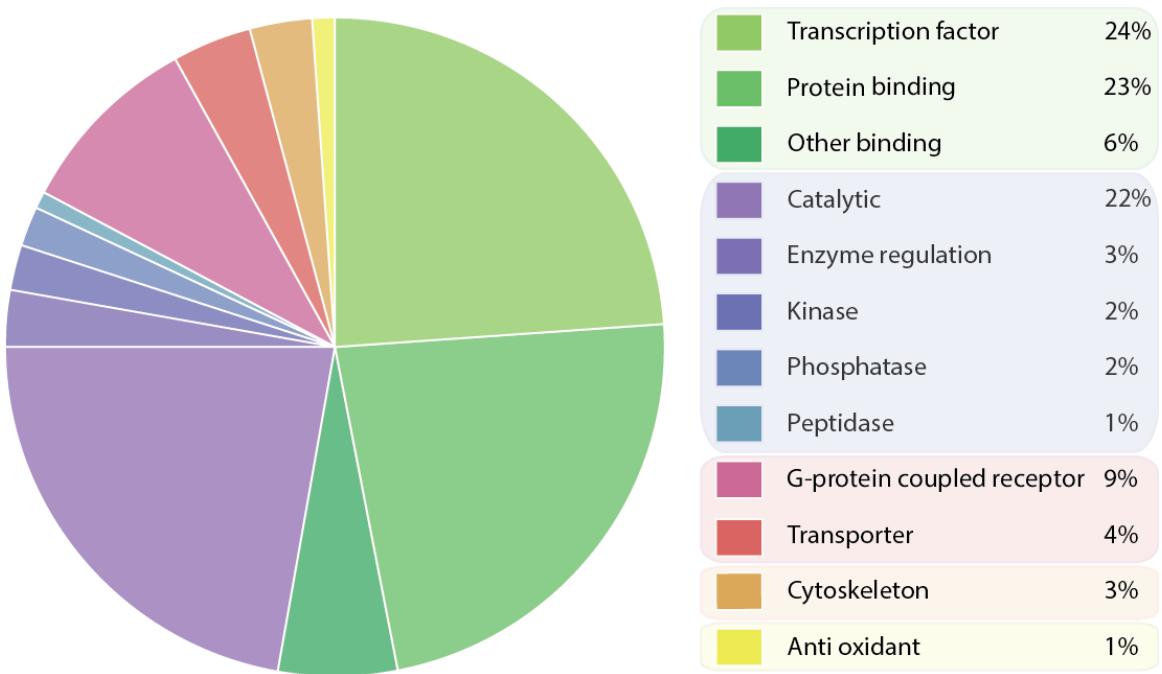
**Table 3.1** The top 20 genes (ranked by FC) up-regulated exclusively after injection of *Eya1*-GR and treatment with CHX+DEX

Gene	Accession	Eya1 FC <sup>a</sup>
X.laevis mRNA for P2Y8 nucleotide receptor	X99953.1	12
* Xenopus laevis homeobox D11 (hoxd11) mRNA	NM_001096497.1	10.4
Xenopus laevis cDNA clone IMAGE:6950606	BC093562.1	10.1
* Xenopus laevis mRNA for XIHbox1 protein (PR I type)	X12499.1	7
* Xenopus laevis homeobox C10 (hoxc10) mRNA	NM_001090479.1	6.4
* Xenopus tropicalis homeobox A11 (hoxa11) mRNA	XM_004915395.1	6
* Xenopus laevis homeobox A9 (hoxa9) mRNA	NM_001097795.1	5.5
Xenopus tropicalis cDNA clone IMAGE:7017143	BC089232.1	5.5
Xenopus laevis cDNA clone MGC:160241 IMAGE:8526708 complete cds	BC129574.1	5
* Xenopus tropicalis homeobox A10 (hoxa10) mRNA	XM_002933394.2	4.8
Xenopus ( <i>Silurana</i> ) tropicalis aldolase A fructose-bisphosphate (aldoa) mRNA	NM_001005643.1	4.5
* Xenopus tropicalis homeobox protein Hox-C11a-like (LOC100487406) mRNA	XM_002936646.2	4.4
Xenopus laevis hypothetical protein LOC100049089 mRNA	BC099046.1	4.4
* Xenopus laevis Hoxc10 mRNA complete cds	AY167741.1	4.2
* Xenopus laevis homeobox D9 (hoxd9) mRNA	NM_001174043.1	4.2
Xenopus tropicalis TYRO protein tyrosine kinase-binding protein-like (LOC100493958)	XM_002940624.2	3.9
Xenopus laevis mRNA for XFGF-20 complete cds	AB012615.1	3.9
Xenopus laevis creatine kinase mitochondrial 1B (ckmt1b-b)	NM_001086060.1	3.8
* Xenopus tropicalis homeobox protein Hox-C9-like (LOC101733557) mRNA	XM_004911933.1	3.7
Xenopus laevis carbonic anhydrase IV (ca4) mRNA	NM_001086319.1	3.6

\* Marks homeobox genes

<sup>a</sup>Log<sub>2</sub> Fold change values for Eya1

Finally, to provide a general summary of gene ontology, differentially expressed genes in the up-regulated merged data set Six1+Eya1<sub>m</sub> were analysed using Panther (Mi et al., 2012) to show the relative abundance of genes grouped by molecular function (fig. 3.9). Transcription factors and protein binding together accounted for the largest fraction of up-regulated genes (53% in total), followed by enzymes (30%) and transporter molecules (13%). That such a high proportion of up-regulated genes were involved in binding (and particularly that 24% were transcription factors), as opposed to other functions (such as housekeeping etc.) suggests the developmental importance of the genes up-regulated by Six1 and Eya1.



**Fig. 3.9** Enrichment of molecular function terms after overexpression of *Six1* and *Eya1*. The area of the pie represents the total number of functional terms contained in the analysis, with each slice representing the percentage of genes against this total. Molecular functions shown can be broadly divided into five categories: Green slices are related to binding functions (53%); purple/blue represents enzyme activity (30%); pink/red shows transmembrane proteins (13%); orange cytoskeleton (3%) and yellow anti-oxidant (1%). Significantly differentially expressed genes from the merged data set (*Six1+Eya1<sub>m</sub>*; appendix F.5) were used for this analysis.

### 3.6 Selection of candidates for in-depth characterisation

In order to construct a *Six1*/*Eya1*-activated gene regulatory network comprising an interacting set of regulatory genes, only up-regulated transcription factors (or co-factors) were considered for further characterisation. As the analysis using individual treatment groups yielded substantially more candidates compared to the merged data (the latter applying a more stringent selection process), target genes were selected from the former data set. In an attempt to minimise the number of false positives selected, and to focus on genes co-regulated by *Six1* and *Eya1*, genes were selected if they showed at least a two-fold up-regulation in at least two of the three treatment groups (table 3.2). This condition was met by 228 genes from which 29 transcription factors and one co-factor (*Ripply3*) were selected for further characterisation. In addition, several other genes were selected even though they didn't meet these criteria: *Gbx2* (*Six1<sub>i</sub>* FC: 0.9; *Eya1<sub>i</sub>* FC: 0.6; *Six1+Eya1<sub>i</sub>* FC: 1.7) was selected owing to its well characterised role in posterior placode development (Steventon et al., 2012), as well as *Ngn1* (*Six1<sub>i</sub>* FC: 0.8; *Eya1<sub>i</sub>* FC: 0.9; *Six1+Eya1<sub>i</sub>* FC: 0.8) which is known to be co-expressed with *Six1* and *Eya1* in the PPE (Schlosser and Northcutt, 2000). Despite falling short of the stringent selection

criteria, *Sox3* (*Six1<sub>i</sub>* FC: 0.5; *Eya1<sub>i</sub>* FC: 0.4 *Six1+Eya1<sub>i</sub>* FC: 0.9) was also considered for characterisation, as it has previously been suggested to be a direct target of *Six1/Eya1* in PPE (Schlosser et al., 2008). Table 3.3 shows the final list of 33 genes selected for further analysis.

**Table 3.2** Genes with at least two-fold up-regulation in at least two out of three individual treatment groups (*Six1<sub>i</sub>*; *Eya1<sub>i</sub>*; *Six1+Eya1<sub>i</sub>*)

	<b>Gene<sup>a</sup></b>	<b>Accession</b>	<b>Six1<sup>b</sup></b>	<b>Eya1<sup>c</sup></b>	<b>Six1+Eya1<sup>d</sup></b>
	X. laevis Ras-related associated with diabetes (rrad)	NM_001092750.1	8	4.6	4.2
	Chromosome unknown open reading frame	XM_002938866.2	6.2	-	7.9
	cDNA clone IMAGE:7022272	BC094950.1	5.6	5.1	7.5
	X. laevis cyclin Dx (ccndx)	NP_001087887.1	7.5	-	5.2
	Calcitonin gene-related peptide-like	XM_002941675.2	7	-	3
	Tripartite motif containing 63, E3 ubiquitin protein ligase (trim63)	NM_001093214.1	5.3	3.6	6.3
	ATP-sensitive inward rectifier potassium channel 11-like	XM_004916278.1	5.1	-	6.1
	Leucine rich repeat containing 52 (lrrc52)	XM_002933773.2	6.1	-	2.8
*	SIX homeobox 2 (six2)	NM_001100275.1	5	3.5	5.9
	Potassium voltage-gated channel shaker-related subfamily 2 (kcna2)	XM_004910736.1	5.1	-	4.9
	Butyrophilin subfamily 2 member A1 (btn2a1)	NM_001094508.1	-	1.2	4.9
*	X. laevis for Xsox17-alpha protein	AJ001730.1	3.6	2.6	4.8
	Glutathione peroxidase 2 (gpox2)	NM_001256315.1	-	2.5	4.8
*	X. laevis Myoblast determination protein 1 homolog A	BC041190.1	3.5	2.7	4.7
	X. laevis ectodysplasin A receptor (edar)	NM_001087047.1	2.8	2.5	4.7
	Calcium/calmodulin-dependent protein kinase kinase 2beta (camkk2)	XM_002937701.2	4.4	2.6	3.5
	Uncharacterized (LOC101734405)	XM_004918247.1	4.4	0.8	3.5
	Glutathione peroxidase 2 (gpox2)	NM_001256315.1	3.4	2.1	4.4
	Cytochrome P450, family 2, subfamily D, polypeptide 6 (cyp2d6)	NM_001093574.1	1.1	-	4.4
	Cytochrome P450 family 26 subfamily B polypeptide 1 (cyp26b1)	NM_001079187.2	3.3	4	4.3
	Ectodysplasin A receptor (edar)	NM_001087047.1	4.3	-	3.3
	Troponin I type 1 (skeletal, slow)	BC061268	1.8	-	4.3
	Protein phosphatase 2 regulatory subunit B'beta (ppp2r5b)	NM_001100279.1	2.4	1.4	4.2
	72 kDa inositol polyphosphate 5-phosphatase-like (LOC101734556)	XM_004916572.1	-	4.2	1.3
	Chemokine (C-X-C motif) receptor 7 (cxcr7)	NM_001030434.1	3	2.8	4.1
	C-X-C motif chemokine 10-like	XM_002940578.2	1.9	4	3.5
	X-linked inhibitor of apoptosis (xiap)	NM_001030412.1	4	3.1	2.3
	X. laevis hedgehog-interacting protein	BC046952.1	-	2.7	4
	Espin (espn) transcript variant X3	XM_004916193.1	-	2.1	4
	B-cell CLL/lymphoma 11B (zinc finger protein) (bcl11b)	XM_004917116.1	-	1.9	4
*	X. laevis xSox17 alpha 2	AB052691.1	1.7	1.4	4
	X. laevis RDC1 like protein	BC098974.1	3.6	2.1	3.9
	X. laevis uncharacterized (LOC496300)	NM_001095458.1	1.4	3.9	1.1
	X. laevis for thimet oligopeptidase	BC070748.1	3.8	-	2
	X. laevis for frizzled 4 protein (fz4 gene)	AJ251750.1	1.3	0.6	3.8
	Espin (espn) transcript variant X1	XM_002933856.2	3.1	2.7	3.7
	Paired box 1 (pax1) transcript variant X1	JQ929179.1	-	3	3.7
	Calcyphosine (caps)	NM_001097320.1	-	1.4	3.7
*	Hairy and enhancer of split 8 (hes8)	XM_002933849.2	2.8	1.7	3.6
	Echinoderm microtubule-associated protein-like 1-like	XM_004917169.1	-	3.6	2.8
	Leucine rich adaptor protein 1-like (lurap1l)	XM_002940127.2	3.6	1.4	2.4
	Potassium voltage-gated channel subfamily F member 1 (kcnf1)	NM_001102926.1	3.6	-	2.1
	Sine oculis binding protein homolog (Drosophila)	BC154687.1	2.7	1.6	3.4
	Anoctamin 2 (ano2)	XM_002932297.2	2.2	1.3	3.4
	RNA-directed DNA polymerase homolog	XM_004916122.1	3.4	-	2.1
	Rho GTPase activating protein 9 (arhgap9), transcript variant X2	XM_012957829	1.8	1.2	3.4
	Kinesin family member 3C (kif3c) transcript variant X1	XM_004914940.1	1.4	0.8	3.4
	Poly (ADP-ribose) polymerase 14-like (LOC101731378)	XM_004920062.1	3.1	-	3.3
	Uncharacterized (LOC101734952)	XM_004916172.1	2.5	-	3.3
	X. laevis natriuretic peptide C (nppc)	NM_001112924.1	2.1	-	3.3
	Uncharacterized (LOC101733225)	XM_004919937.1	2.5	3.2	1.6
*	Growth factor independent 1 transcription repressor (gfi1)	XM_002933803.2	1.8	1.8	3.2
	Protocadherin-11 X-linked-like (LOC100493938)	XM_004916890.1	-	3.2	1.4
	XLU76636 X. laevis calbindin D28k	BC170542.1	2.2	-	3.1
	Calcium channel voltage-dependent beta 4 subunit (cacnb4)	NM_001142151.1	3.1	-	1.9
	F-box protein 32 (fbxo32) transcript variant X1	XM_002941397.2	1.8	-	3.1
*	X. laevis POU class 3 homeobox 2 (pou3f2-b)	NM_001096751.1	3	2.3	2.9
	cDNA clone TEgg026p17	CR761997.2	3	2.6	-
	Mucin-2-like (LOC100494747)	XM_002936043.2	3	2	1.7
	X. laevis transforming growth factor beta-induced (tgfb1)	NM_001095238.1	1.3	-	3
*	X. laevis mab-21-like 2 (mab21l2-b)	NM_001096770.1	-	2.8	2.9
	Integrin beta 6 (itgb6)	NM_001097306.1	2.3	0.6	2.8
	X. laevis uncharacterized protein (MGC68450)	NM_001089841.1	2.2	-	2.8

Gene <sup>a</sup>	Accession	Six1 <sup>b</sup>	Eya1 <sup>c</sup>	Six1+Eya1 <sup>d</sup>
X. laevis U3 snRNA	X07318.1	1	2.8	1.1
Potassium voltage-gated channel I-sk-related (kcne1)	XM_004912135.1	2.2	1.5	2.7
Alpha-kinase 2 (alpk2)	XM_004910401.1	1.1	2.2	2.7
Olfactory receptor 5G3-like (LOC100492086)	XM_002942220.1	1.9	-	2.7
X. laevis arginyl amino peptidase (amino peptidase B) b (rnpep-b)	NM_001092079.1	-	2.7	1.7
X. laevis neuregulin alpha-1	AF076618.1	1.4	0.8	2.7
* T-cell leukemia homeobox 1 (tlx1) transcript variant 1	XM_002936768.2	2.6	2.3	2.6
* Xenopus laevis empty spiracles homeobox 1, gene 2 (emx1.2)	NM_001093430.1	2.6	1.4	2.1
* X. laevis SRY-box containing protein (Sox1)	EF672727.1	-	2.6	2.1
* X. laevis empty spiracles homeobox 1 gene 2 (emx1.2)	NM_001093430.1	2.6	1.9	1.1
X. laevis p21-activated kinase (PAK1)	AF169794.1	1.4	1.8	2.6
KIAA0895 protein (kiaa0895)	NM_001114073.1	1.6	2.6	-
Copine II (cpne2) transcript variant X1	XM_004913481.1	1	1.2	2.6
X. laevis hemoglobin, gamma A (hbga)	NM_001096347	1.2	-	2.6
Finished cDNA clone TNeu143f19	CR760056.2	2.2	2.5	-
X. laevis uncharacterized (LOC100036933)	NM_001097704.1	1.5	1.5	2.5
G protein-coupled receptor 153 (gpr153)	NM_001128052.1	2.5	1.1	1.5
Monocyte to macrophage differentiation-associated (mmd)	XM_004918560.1	-	1.2	2.5
Homeobox B8 (hoxb8) transcript variant X1	XM_002938021.2	1.1	2.5	-
Chromosome unknown open reading frame C2orf80	XM_002937119.2	1.4	2.1	2.4
Transmembrane protein 2-like (LOC100491930)	XM_002932255.2	2.4	1.9	1.3
* Single-minded homolog 1 (sim1) transcript variant X2	XM_004914545.1	-	1.4	2.4
PX domain containing 1 (pxdc1)	NM_001130262.1	1.4	-	2.4
Kinase insert domain receptor (kdr)	XM_002934669.2	1.9	0.9	2.3
Uncharacterized (LOC100490228)	XM_002942932.2	1.8	-	2.3
Alpha-2 3-sialyltransferase ST3Gal V (st3gal5)	FN550108.1	1.8	-	2.3
Beta-1 3-galactosyltransferase 2-like (LOC101732799)	XM_004918863.1	1.6	2.3	-
X. laevis uncharacterized protein (MGC64538)	NM_001086337.1	-	1.6	2.3
* X. laevis SIX homeobox 1 (six1)	AF279254.1	1.4	1.2	2.3
Transmembrane channel-like protein 7-like (LOC100493700)	XM_002932222.2	1.4	0.9	2.3
X. laevis Cep63	FJ464988.1	-	1.4	2.3
Aldehyde dehydrogenase 1 family member L2 (aldh1l2)	XM_002938070.2	0.9	1.3	2.3
X. laevis degr03	DQ096846.1	2.1	2.2	2
Pancreatic lipase-related protein 2 (pnliprp2)	NM_001089647.1	2.1	0.7	2.2
Poly (ADP-ribose) polymerase 14-like (LOC100485144)	XM_002943546.2	2	2.2	1.2
X. laevis chromogranin A (parathyroid secretory protein 1) (chga)	NM_001094724.1	1.6	1.4	2.2
Integrin beta 4 (itgb4) transcript variant X1	XM_002939974.2	1.4	-	2.2
Serine/threonine kinase 32A (stk32a)	XM_002936707.2	1.3	2.2	-
WD repeat domain 27 (wdr27)	XM_002931515.2	1.2	2.2	1.1
X. laevis nephrin (NPHS1)	AY902238.1	-	2.2	1.1
Deoxyribonuclease gamma-like (LOC100497175)	XM_002938386.2	1.8	2.1	2
X. laevis gamma-glutamyl hydrolase (ggh)	NM_001092691.1	2.1	1.3	2
Putative N-acetyltransferase 16-like (LOC100490742)	XM_002943189.1	2.1	1	1.7
Cytochrome P450 family 2 subfamily C polypeptide 18 (cyp2c18)	NM_001091776.1	2.1	1.4	1.6
Neuropeptide Y receptor Y2 (npy2r)	XM_004911153.1	2.1	-	1.6
X. laevis claudin 3 (cldn3)	NM_001005709.1	2.1	1.3	1.5
X. laevis dehydrogenase/reductase (SDR family) member 11 (dhrs11)	NM_001094963.1	-	1.5	2.1
Frizzled family receptor 4 (fzd4)	XM_002936543.2	1.4	0.7	2.1
X. laevis p21 GTPase-associated kinase 1 (PAK1)	AF000239.1	1.2	-	2.1
X. laevis fibroblast growth factor 3 (fgf3)	NM_001008153.1	2	1.2	2
Atlastin GTPase 1 (atl1)	NM_001078754.1	1.8	2	1.7
* T-box 15 (tbx15)	XM_002940981.2	2	1	1.8
Mannosyl-glycoprotein beta-1 4-N-acetylglicosaminyltransferase	NM_001091975.1	2	-	1.8
X. laevis CD81 antigen (target of anti proliferative antibody 1)	BC041217.1	1.7	1.1	2
Avidin-like (LOC100487365)	XM_002939983.2	2	1.6	-
Pyruvate dehydrogenase phosphatase catalytic subunit 1 (pdp1)	NM_001094221.1	1.5	2	1
Fibroblast growth factor 19 (fgf19)	NM_001142825.1	-	2	1.5
X. laevis uncharacterized protein (MGC83079)	NM_001091250.1	2	1.5	-
Uncharacterized (LOC101732195)	XM_004912378.1	2	-	1.5
Ceramide kinase-like (cerkl)	XM_002932015.2	1.4	1.3	2
X. laevis 7-transmembrane receptor frizzled-1	AF231711.1	1.4	1	2
* F-box protein 41 (fbxo41)	NM_001079043.1	1.3	0.6	2
Opsin-3-like	XM_002932623.2	1	1.2	2
* X. laevis xRipply3 for xRipply3 protein	AB455086.1	0.9	1.1	2
X. laevis transmembrane protein 56 (tmem56-b)	NM_001086447.1	-	1.1	2
FH2 domain-containing protein 1-like (LOC100496216)	XM_002934907.2	1.9	0.9	1.9
X. laevis similar to calsequestrin 2 (cardiac muscle)	BC097545.1	1.8	1.5	1.9
ArfGAP with SH3 domain ankyrin repeat and PH domain 3 (asap3)	XM_002939360.2	1.7	-	1.9
X. laevis Kazal-type serine peptidase inhibitor domain 1 (kazald1)	NM_001092073.1	1.6	1.1	1.9
* Early growth response 3 (egr3)	XM_002932703.2	1.6	0.8	1.9
X. laevis neurotrophin 3 (ntf3)	NM_001092740.1	1.4	1.5	1.9
Transmembrane proteaseserine 13 (tmpRSS13)	XM_002932904.2	1.5	1.1	1.9
Metalloprotease TIK1-like (LOC100491951)	XM_002936336.2	1.1	1.4	1.9

	Gene <sup>a</sup>	Accession	Six1 <sup>b</sup>	Eya1 <sup>c</sup>	Six1+Eya1 <sup>d</sup>
*	POU class 4 homeobox 1 (pou4f1.2)	NM_001097307.1	1.3	1	1.9
*	SRY (sex determining region Y)-box 2 (sox2)	NM_213704.3	1.1	1.3	1.9
	Uncharacterized (LOC101734664)	XM_004910525.1	1.2	0.6	1.9
	X. laevis alcohol dehydrogenase iron containing1 (adhfe1)	NM_001127802.1	-	1.9	1.2
	X. laevis CD81 protein (cd81-a)	NM_001086613.1	0.7	1.1	1.9
	X. laevis COMM domain containing 3 (commd3)	NM_001095386.1	1.1	1.9	0.6
	Four and a half LIM domains 2 (fhl2)	NM_001126761.1	-	1.1	1.9
	Monocyte to macrophage differentiation-associated (mmd)	XM_002937811.2	1.7	1.1	1.8
	Activin beta B subunit	S61773.1	-	1.7	1.8
	X. laevis ribosomal protein S2e	BC130122.1	-	1.8	1.7
*	X. laevis hairy and enhancer of split 9, gene 1 (hes9.1-b)	NP_001089097.1	1.8	1.5	1.6
	Ornithine decarboxylase antizyme 2 (oaz2), transcript variant 2	NP_001106583.2	1.8	-	1.5
	X. laevis ectonucleoside triphosphate diphosphohydrolase 1 (entpd1)	NM_001092268.1	1.8	0.6	1.3
	X. laevis arginase 3	U08408.1	-	1.3	1.8
	Tocopherol (alpha) transfer protein (ttpa)	NM_001008184.1	-	1.7	1.6
*	X. laevis for enhancer of split related 9 (esr9 gene)	AJ009282.1	1.7	1.6	-
	G protein-coupled receptor 56 (gpr56)	XM_002931653.2	1.7	-	1.6
*	ISL LIM homeobox 2 (isl2)	NM_001166041.1	1.5	-	1.7
*	X. laevis Tbx6 (Tbx6)	DQ55794.1	1.4	1.7	1
	X. laevis DIRAS family GTP-binding RAS-like 3 (diras3)	NM_001095243.1	0.8	1.7	1.4
*	Protein fosB-like transcript variant X2	XM_004916957.1	-	1.7	1.4
	X. laevis regulator of cell cycle (rgcc)	NM_001093976.1	1.3	1.1	1.7
*	X. laevis Hes2	BC084134.1	1.7	0.9	1.3
	X. laevis p21 GTPase-associated kinase 1	BC081113.1	1.3	0.8	1.7
	Family with sequence similarity 198 member A (fam198a)	XM_002937853.2	1.7	0.7	1.3
	X. laevis uncharacterized (LOC100036989)	NM_001097746.1	-	1.3	1.7
	X. laevis cDNA clone IMAGE:6947552	BC093552.1	1.3	1.7	-
	X. laevis ATPaseNa+/K+ transporting beta 1 polypeptide (atp1b1)	NM_001086759.1	1.2	1	1.7
	B-cell CLL/lymphoma 10 (bcl10)	NM_001015777.2	1.7	-	1.2
	Progesterin and adiponectin receptor family member IX (paqr9)	XM_004914351.1	1.7	-	1.2
	X. laevis tetraspanin repeat domain 39B (ttc39b)	NM_001094701.1	1.1	-	1.7
	Phospholipase C delta 3 (plcd3)	XM_002935518.2	1.1	1.5	1.6
	X. laevis kiaa0930	NM_001086221.1	1.5	1	1.6
	X. laevis complement factor I (cfi-a)	NM_001085952.1	1.4	1.2	1.6
	Uncharacterized (LOC100494710)	XM_002939048.2	1.4	1.6	-
	Proline rich 15 (prrr15)	XM_002933381.2	1.6	-	1.3
	X. laevis adenomatosis polyposis coli down-regulated 1 (apcdd1)	NM_001094109.1	1.2	1	1.6
	X. laevis ras homolog family member V (rhov)	NM_001128659.1	1.2	0.8	1.6
	X. laevis keratin 17 (krt17)	NM_001094941.1	-	1.2	1.6
	Membrane metallo-endopeptidase-like 1 (mmel1)	NM_001127095.1	0.9	1.1	1.6
	X. laevis calcitonin receptor-like (calcr1)	NM_001086737.1	1.1	0.8	1.6
	Putative methyltransferase KIAA1456 homolog	XM_002934674.2	1.1	-	1.6
	X. laevis family with sequence similarity 101 member B (fam101b)	NM_001093870.1	1.5	0.8	1.5
	Uncharacterized (LOC100486093) transcript variant X2	XM_002939117.2	1.5	-	1.5
	IdnK gluconokinase homolog (E. coli) (idnk)	NM_001126592.1	1.4	0.9	1.5
	X. laevis cDNA clone IMAGE:8332229	BC155363.1	1.5	0.9	1.4
	X. laevis prostaglandin reductase 2 (ptgr2)	NM_001079334.1	1.4	1.5	-
*	cAMP responsive element modulator (crem)	XM_002935162.2	-	1.4	1.5
	ChaC cation transport regulator homolog 1 (chac1)	XM_002939546.2	1.2	1.3	1.5
	X. laevis apelin (apln-a)	NM_001097924.1	0.9	1.3	1.5
*	X. laevis zinc finger protein 214 (znf214)	NM_001097042.1	1.2	0.8	1.5
*	SRY (sex determining region Y)-box 1 (sox1)	NM_001080996.1	0.6	1.5	1.2
	X. laevis cdc25Ba	AB363840.1	1.2	-	1.5
*	Xenopus laevis SRY (sex determining region Y)-box 21 (sox21)	NM_001172213.1	1.2	0.6	1.5
*	Atonal homolog 1 (Drosophila) (atoh1)	XM_004911085.1	0.9	1.1	1.5
	Suppressor of cytokine signalling 2 (soc2)	NM_001095760.1	-	1.1	1.5
	X. laevis similar to calsequestrin 2 (cardiac muscle)	BC041283.1	1.1	-	1.5
	Piwi-like RNA-mediated gene silencing 2 (piwil2)	NM_001112999.1	1.1	-	1.5
	X. laevis fast troponin T (TNNT3)	AY114144.1	-	1.1	1.5
	X. laevis similar to envoplakin	BC045116.1	1.4	1.4	-
*	X. laevis ets-2a proto-oncogene	BC133183.1	1.3	1	1.4
	X. laevis uncharacterized protein (MGC81120)	NM_001091225.1	1.4	0.9	1.3
	X. laevis cDNA clone IMAGE:5085355	BC073731.1	1.3	-	1.4
	X. laevis mal T-cell differentiation protein (mal)	NM_001086577.1	-	1.2	1.4
	RAS-like family 11 member B (rasl11b)	NM_001015774.1	-	1.2	1.4
	Ras homolog family member V (rhov)	NM_001095566.1	1.4	1	1.2
	X. laevis clone IMAGE:4684003	BC042305.1	1.4	-	1.2
*	V-maf musculoaponeurotic fibrosarcoma oncogene A (mafa)	NM_001032304.1	1.4	0.9	1.1
	Zinc finger and BTB domain containing 20 (zbtb20)	XM_002939649.2	1.4	-	1.1
*	Single-minded homolog 1 (Drosophila) (sim1) transcript variant X3	XM_004914546.1	1.1	1.3	1.2
	X. laevis tetraspanin 1 (tspan1)	NM_001095473.1	1.2	0.7	1.3
	X. laevis lipaseendothelial (lipg)	NM_001090061.1	1.2	1.3	0.6
	Samd9l protein (samd9l)	XM_002943522.2	-	1.2	1.3

<b>Gene<sup>a</sup></b>	<b>Accession</b>	<b>Six1<sup>b</sup></b>	<b>Eya1<sup>c</sup></b>	<b>Six1+Eya1<sup>d</sup></b>
Flocculation protein FLO11-like (LOC100490389)	XM_002942555.2	1.2	-	1.3
FERM domain containing 4A (frmd4a)	XM_002935243.2	1.1	0.6	1.3
X. laevis TGF-beta2 for transforming growth factor-beta2	X51817.1	1.3	-	1.1
Transmembrane serine protease 9	BC087611.1	1.1	1.1	1.2
X. laevis Dickkopf-1 (Xdkk-1)	AF030434.1	1	1.2	1.1
Uncharacterized (LOC101730819)	XM_004915204.1	0.9	1.1	1.2
X. laevis ectoderm neural cortex related-3 (Engr-3)	AY216793.1	1.1	0.8	1.2
Tumor necrosis factor receptor superfamily member 21 (tnfrsf21)	NM_001079136.1	1.1	0.8	1.2
Finished cDNA clone TNeu008g03	CR761907.2	1.2	1.1	0.7
c-Jun-amino-terminal kinase-interacting protein 4-like	XM_002939963.2	1.1	-	1.2
X. laevis Dnaj (Hsp40) homolog subfamily C member 27 (dnajc27-b)	NM_001095422.1	1.1	0.8	1.1
* X. laevis LIM class homeodomain protein	BC084744.1	1.1	0.7	1.1
Xenopus laevis alpha-2-macroglobulin-like 1 (a2m1)	NM_001135077.1	1.1	-	1.1

\* Indicates transcription factors with at least two-fold change in at least two treatment groups selected for further analysis

<sup>a</sup>Genes are ranked by FC value, using the highest FC in each of the three treatments groups. Genes included must have FC  $\geq 1$  in at least two out of the three treatment groups as well as showing at least a two-fold difference in FC to the un-injected control (not shown). Corresponding values  $\geq 0.5$  are shown for all treatments

<sup>b</sup> Log<sub>2</sub> Fold change values after *Six1* overexpression

<sup>c</sup> Log<sub>2</sub> Fold change values after *Eya1* overexpression

<sup>d</sup> Log<sub>2</sub> Fold change values after *Six1+Eya1* overexpression

**Table 3.3** Transcription factors and co-factors selected for characterisation by *in-situ*-hybridisation ranked by FC value in individual treatment

Gene	Short	Accession	Individual			Merged	
			Six1 <sup>a</sup>	Eya1 <sup>b</sup>	Six1+Eya1 <sup>c</sup>	Six1 <sup>d</sup>	Eya1 <sup>e</sup>
SIX homeobox 2 (six2)	Six2	NM_001100275.1	5	3.5	5.9	5.4*	5*
X. laevis for Xsox17-alpha protein	Sox17	AJ001730.1	3.6	2.6	4.8	4.6*	3.3*
X. laevis Myoblast determination protein 1 homolog A	MyoD1	BC041190.1	3.5	2.7	4.7	4.1*	4.2*
Hairy and enhancer of split 8 (hes8)	Hes8/5	XM_002933849.2	2.8	1.7	3.6	3.2*	3.2*
Growth factor-independent 1 transcription repressor (grf1)	Grf1a	XM_002933803.2	1.8	1.8	3.2	4*	4*
X. laevis POU class 3 homeobox 2 (pou3f2-b)	Pou3f2b	NM_001096751.1	3	2.3	2.9	3*	2.6*
X. laevis mab-21-like 2 (mab21l2-b)	Mab21l2b	NM_001096770.1	-	2.8	2.9	-	2.7*
T-cell leukemia homeobox 1 (tlx1) transcript variant 1	Tlx1	XM_002936798.2	2.6	2.3	2.6	2.5*	2.5*
X. laevis empty spiracles homeobox 1 gene 2 (emx1.2)	Emx1.2	NM_001093430.1	2.6	1.9	1.1	2.3*	2*
X. laevis SRY-box containing protein (Sox1)	Sox1	EF672727.1	-	2.6	2.1	-	-
Single-minded homolog 1 (sim1) transcript variant X2	Sim1	XM_004914545.1	-	1.4	2.4	-	-
X. laevis SIX homeobox 1 (six1)	Six1	AF279254.1	1.4	1.2	2.3	1.9*	1.6*
F-box protein 41 (fbxo41)	Fbxo41	NM_001079043.1	1.3	0.6	2	-	-
T-box 15 (tbx15)	Tbx15	XN_002940981.2	2	1	1.8	1.9*	1.7*
X. laevis xRipply3 for xRipply3 protein	Ripply3	AB455086.1	0.9	1.1	2	1.6*	1.4*
Early growth response 3 (egr3)	Egr3	XM_002932703.2	1.6	0.8	1.9	2.2*	1.9*
SRY (sex determining region Y)-box 2 (sox2)	Sox2	NM_213704.3	1.1	1.3	1.9	1.6*	1.5*
POU class 4 homeobox 1 (pou4f1.2)	Pou4f1.2	NM_001097307.1	1.3	1	1.9	1.7*	1.6*
X. laevis for enhancer of split related 9 (esr9 gene)	Es9.1a	AJ009282.1	1.7	1.6	-	-	-
ISL-LIM homeobox 2 (isl2)	Isl2	NM_001166041.1	1.5	-	1.7	1.8*	1.3*
X. laevis Tbx6 (Tbx6)	Tbx6	DQ355794.1	1.4	1.7	1	-	-
Protein fosB-like transcript variant X2	FosB	XM_004916957.1	-	1.7	1.4	-	1.5
X. laevis Hes2	Hes2	BC084134.1	1.7	0.9	1.3	-	1.2*
XLHOX7A Xenopus laevis homeobox protein Xgbx-2	Gbx2	U04867.1	0.9	0.6	1.7	1.3*	-
cAMP responsive element modulator (crem)	Crem	XN_002935162.2	-	1.4	1.5	-	1.4*
X. laevis zinc finger protein 214 (znf214)	Znf214	NM_001097042.1	1.2	0.8	1.5	6.1*	5.9*
Xenopus laevis SRY (sex determining region Y)-box 21 (sox21)	Sox21	NM_001172213.1	1.2	0.6	1.5	1.4*	1.2*
Atonal homolog 1 (Drosophila) (atoh1)	Atoh1	XN_004911085.1	0.9	1.1	1.5	1	1*
X. laevis ets-2a proto-oncogene	Ets2a	BC133183.1	1.3	1	1.4	1.3*	1.2*
V-maf musculoaponeurotic fibrosarcoma oncogene homolog A (mafA)	MafA	NM_001032304.1	1.4	0.9	1.1	1.9*	-
X. laevis LIM class homeodomain protein	Lhx5	BC084744.1	1.1	-	1.1	-	-
Xenopus (Silurana) tropicalis neurogenin 1 (neurog1)	Ngn1	NM_001123423.1	0.8	0.9	0.8	0.8	0.8*
Xenopus laevis SOX3 protein	Sox3	BC072222.1	0.5	-	0.9	0.7	0.6

<sup>a</sup> Log<sub>2</sub> fold change values after Six1 overexpression (Six1<sub>0</sub>)<sup>b</sup> Log<sub>2</sub> fold change values after Eya1 overexpression (Eya1<sub>0</sub>)<sup>c</sup> Log<sub>2</sub> fold change values after Six1+Eya1 overexpression (Six1+Eya1<sub>0</sub>)<sup>d</sup> Log<sub>2</sub> fold change values after overexpression of Six1 or Six1+Eya1 (Six1<sub>m</sub>)<sup>e</sup> Log<sub>2</sub> fold change values after overexpression of Eya1 or Six1+Eya1 (Eya1<sub>m</sub>)<sup>f</sup> Log<sub>2</sub> fold change values after overexpression of Six1 or Eya1 or Six1+Eya1 (Six1+Eya1<sub>m</sub>)  
\* Denotes statistically supported data (q < 0.05)

# Chapter 4. Characterisation and verification of direct targets of Six1 and Eya1

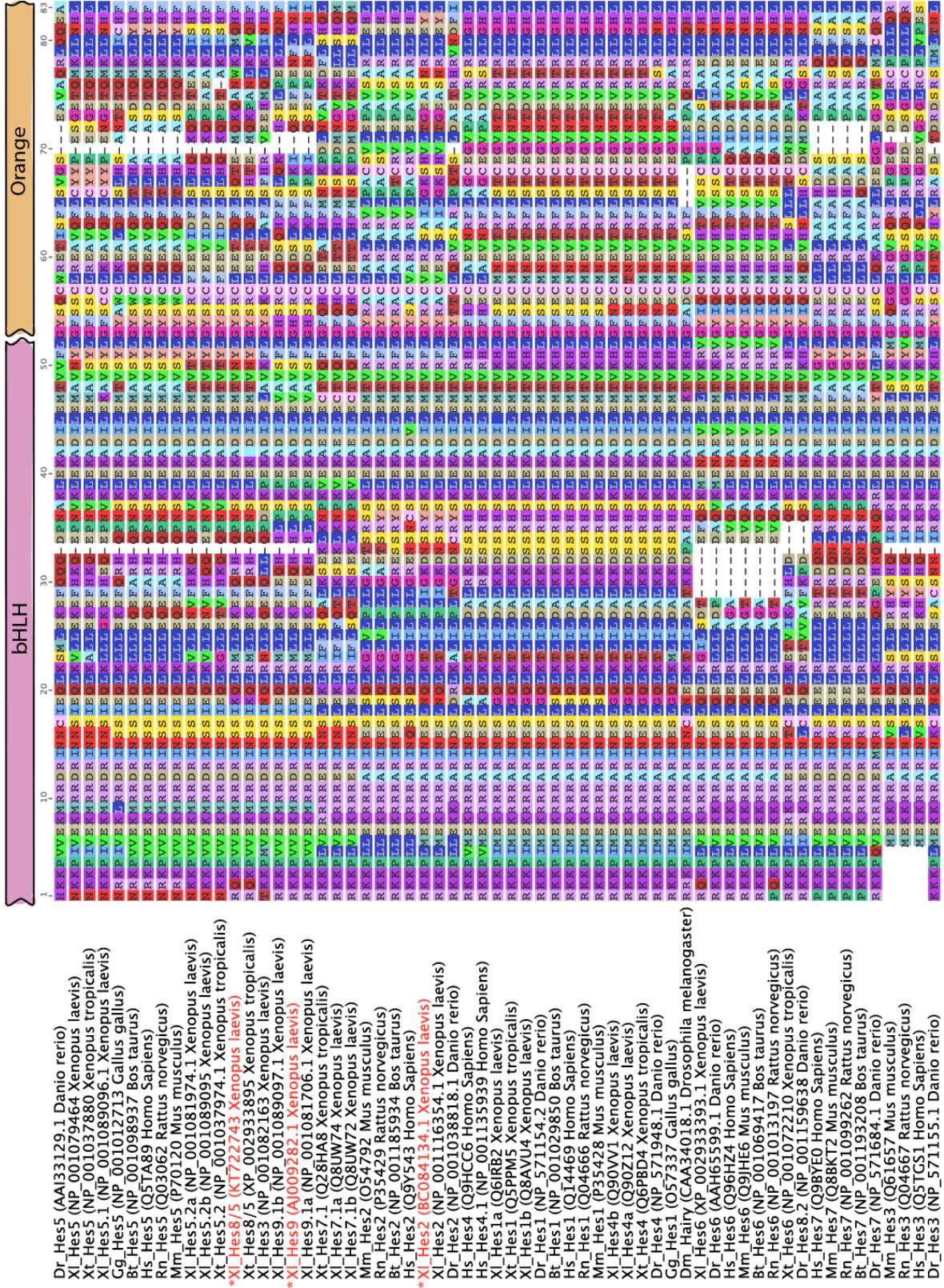
## 4.1 Initial characterisation of selected genes

### 4.1.1 Annotation and verification of gene classification

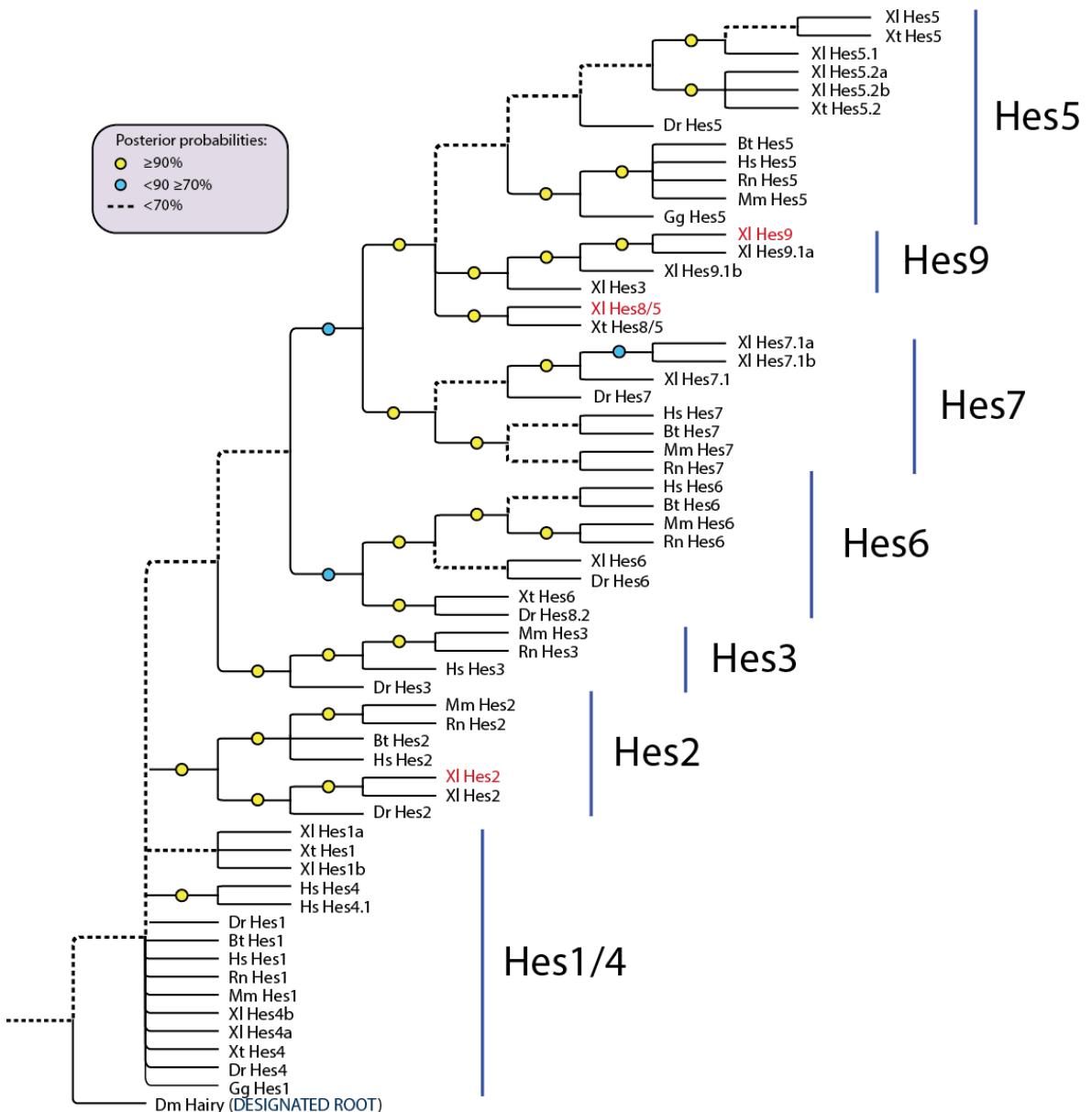
Initially, all genes were annotated with 5' and 3' UTRs and where possible, full-length coding regions (CDS). The CDS for each gene was then used as a BLAST search against the NCBI non-redundant nucleotide database to verify its initial classification. With the exception of *Hes8*, all genes returned hits consistent with their initial annotation. Each gene was then used as a query to the online *Xenopus* resource Xenbase (<http://www.xenbase.org>) in order to survey previously published expression data. Genes with expression patterns published for embryonic stages 15 - 28 that show no placodal expression (*Ets2a* (Salanga et al., 2010); *Mafa* (Coolen et al., 2005); *MyoD1* (Hopwood et al., 1989); *Sox1* (Nitta et al., 2006); *Sox17* (Hudson et al., 1997)) were removed from consideration.

### 4.1.2 Hes8 broadly clusters with other vertebrate Hes5 genes

*Hes8* was annotated against a predicted *X. tropicalis* gene that, when further scrutinised, appeared to bear close sequence similarity to other vertebrate *Hes5* orthologs. In order to correctly classify *Hes8*, amino acid sequences of vertebrate *Hes* genes (see appendix G.1 for raw sequences and accession numbers) were aligned across the conserved basic helix-loop-helix (bHLH) and Orange domains (fig. 4.1), and analysed using both Bayesian and Maximum Likelihood (ML) methods. The Bayesian tree clearly resolves most *Hes* subfamilies, showing strong support (posterior probability  $\geq 90\%$ ) for *Hes2*, *Hes3*, *Hes7* and *Hes9*, and good support (posterior probability  $\geq 70\%$ ) for *Hes6*, as shown in figure 4.2. *Hes1* and *Hes4* remain unresolved as a polytomy. The *Hes5/Hes9* subfamily is strongly supported as a group, consistent with a previous report that the *Hes9* gene family arose either from a *Hes5* duplication or a divergent *Hes5* pseudoallele (Y. Li et al., 2003). While *Hes8* clusters within the *Hes5/Hes9* clade, it is outside the *Hes9* clade and thus it would seem appropriate to consider it a *Hes5/8* gene, and it will henceforth be referred to as such. As well as successfully resolving the classification of *Xenopus Hes* genes, this phylogeny represents the most complete survey of *Hes* genes in vertebrates to date, and would additionally recommend the reclassification of the *X. laevis* gene *Hes3* (NP\_001082163), which clusters with strong support within the *Hes9* clade, as a *Hes9* gene as well the *D. rerio* gene *Hes8.2* (NP\_001159638) as a *Hes6* gene. The ML tree resolves gene families almost identically to the Bayesian analysis, showing agreement among all subfamilies, and can be found in appendix G.2.



**Fig. 4.1** Alignment of Vertebrate Hes genes. bHLH and Orange (indicated by pink and orange bars at the top of alignment) domains were concatenated resulting in an alignment of 83 amino acids across 61 species. Sequence names in red indicate those derived from RNA-Seq data (Hes2, Hes8/5 and Hes9). The Drosophila gene hairy was specified as the outgroup in subsequent phylogenetic analysis. Raw sequences used in alignment and their accessions can be found in appendix G.1.



**Fig. 4.2** Bayesian inference analysis of Vertebrate *Hes* genes based on a concatenated alignment of 83 amino acid residues encompassing the conserved bHLH and Orange domains (fig. 4.1). Nodal support shown as posterior probabilities where  $\geq 70$  (solid lines); support below 70% shown as dashed lines. *Hes* genes derived from RNA-Seq data shown in red. Accessions are found in appendix G.1. Abbreviations; Dr *Danio rerio*, XI *Xenopus laevis*, Xt *Xenopus tropicalis*, Gg *Gallus gallus*, Bt *Bos taurus*, Hs *Homo sapiens*, Rn *Rattus norvegicus*, Mm *Mus musculus*, Dm *Drosophila melanogaster* (outgroup).

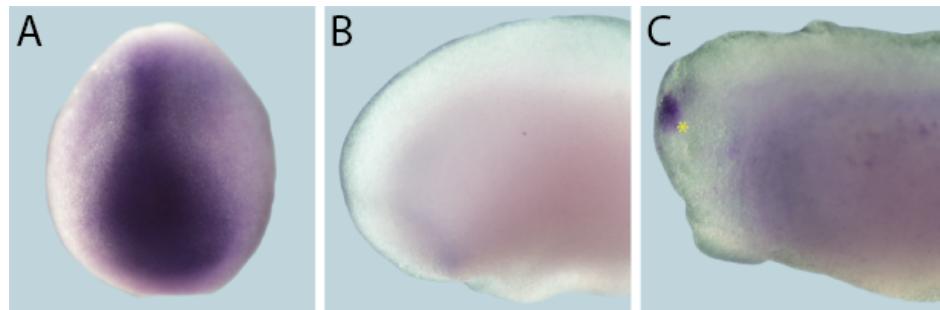
## 4.2 Spatio-temporal characterisation of selected target genes

To elucidate the spatio-temporal dynamics of target gene expression, *in-situ*-hybridisation was performed on an informative range of embryonic stages (neural plate, stages 14-18; early tail bud, stages 21-24; mid/late tail bud, stages 28-32) for each gene. Genes previously undescribed in *Xenopus* (*Crem*, *FosB*, *Hes5/8*, *Isl2*, *Tbx15*, *Znf214*) were fully characterised along with those for which expression has been described only for a few stages (*Atoh1*, *Emx1.2*, *Gfi1a*, *Hes2*, *Hes9*, *Lhx5*, *Mab21l2b*, *Pou3f2b*, *Pou4f1.2*, *Ripply3*, *Sim1*, *Sox21*, *Tbx6*, *Tlx1*) in figures 4.3-4.22. Genes with extensively characterised expression patterns (*Ngn1* (Nieber et al., 2009); *Six1* (Pandur and Moody, 2000); *Six2* (Ghanbari et al., 2001); *Sox2* (Mizuseki et al., 1998); *Sox3* (Penzel et al., 1997)) are not shown here. Several genes (*Egr3*, *Fbxo41* and *Gbx2*) were also omitted from further characterisation owing to technical difficulties: Neither *Egr3* nor *Fbxo41* could successfully be amplified from cDNA, and the *in-situ* probe synthesised for *Gbx2* did not hybridise. Expression patterns for each gene are detailed below, broadly grouped by their expression domains in the following categories: (1) Genes not expressed in placodes or placode-derived structures; (2) genes expressed broadly in cranial ectoderm including PPE; (3) genes expressed in proliferating placodal progenitors of PPE; (4) genes expressed in differentiating placodal cells. Sections were taken in cases where expression was not fully resolved from analysis of whole-mounts, and are shown accompanying whole-mount images. Sense controls are provided for genes not previously characterised and can be found in appendix H.

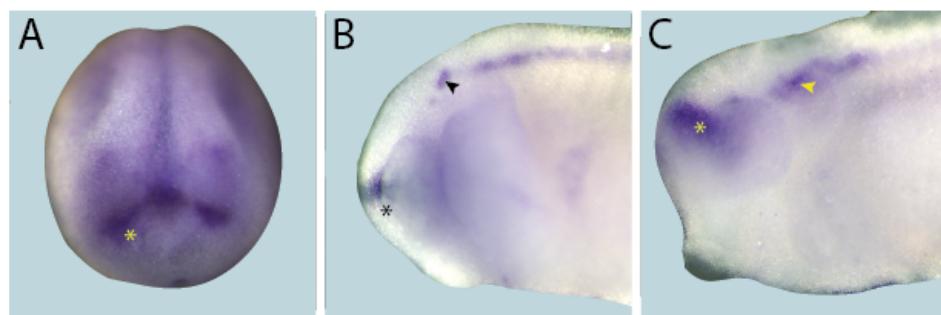
### 4.2.1 Genes not expressed in placodes

A first group of genes is composed of genes that are not expressed in placodes or any placodal derivatives. Among the genes analysed, *Emx1.2* (fig. 4.3), *Lhx5* (fig. 4.4), *Pou3f2b* (fig. 4.5), *Tbx15* (fig. 4.6), *Sim1* (fig. 4.7) and *Tbx6* (fig. 4.8) are included in this category, and are expressed in either the adjacent neural plate (*Emx1.2*), neural folds (*Lhx5*, *Pou3f2b*, *Tbx15*) or broadly throughout dorsal tissues (*Sim1*, *Tbx6*) at neural plate stages. At tail bud stages, both *Emx1.2* and the Lim-homeodomain gene *Lhx5* are expressed in the dorsal forebrain and telencephalon (Bachy et al., 2001; Green and Vetter, 2011; Pannese et al., 1998; fig. 4.3C and fig. 4.4C, respectively), whereas *Pou3f2b* (Baltzinger et al., 1996; fig. 4.5C) is expressed in all levels of the brain (forebrain, midbrain, hindbrain and spinal cord). Previously undescribed, *Tbx15* is expressed in a subdomain of the anterolateral neural folds at neural plate stages (fig. 4.6A), and in some neural crest streams, as well as prominently in the somites at tail bud stages (fig. 4.6B and C). *Sim1* appears to be confined to the pronephros (fig. 4.7C) at tail bud stages, however, published expression patterns clearly indicate additional expression domains in the head, pharyngeal arches and somites (Martin et al., 2007). *Tbx6* is prominently expressed in

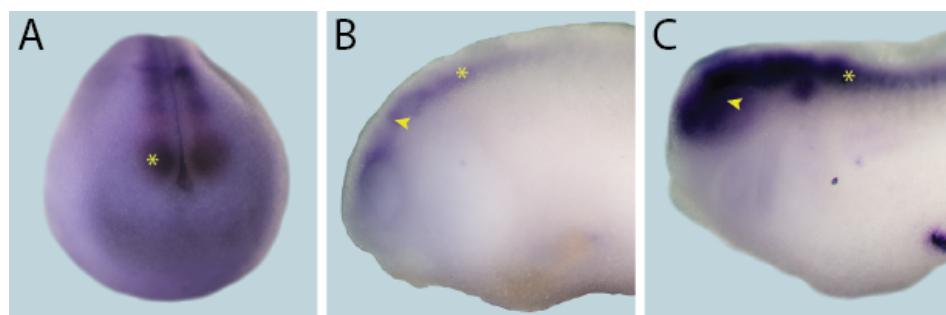
the posterior lateral plate mesoderm at neural plate stages (fig. 4.8A), and in the developing tail bud at later stages (data not shown). Due to the broad and neighbouring expression domains exhibited by these genes, it is likely that they were included in the PPE screen as a result of contamination from surrounding or underlying tissues.



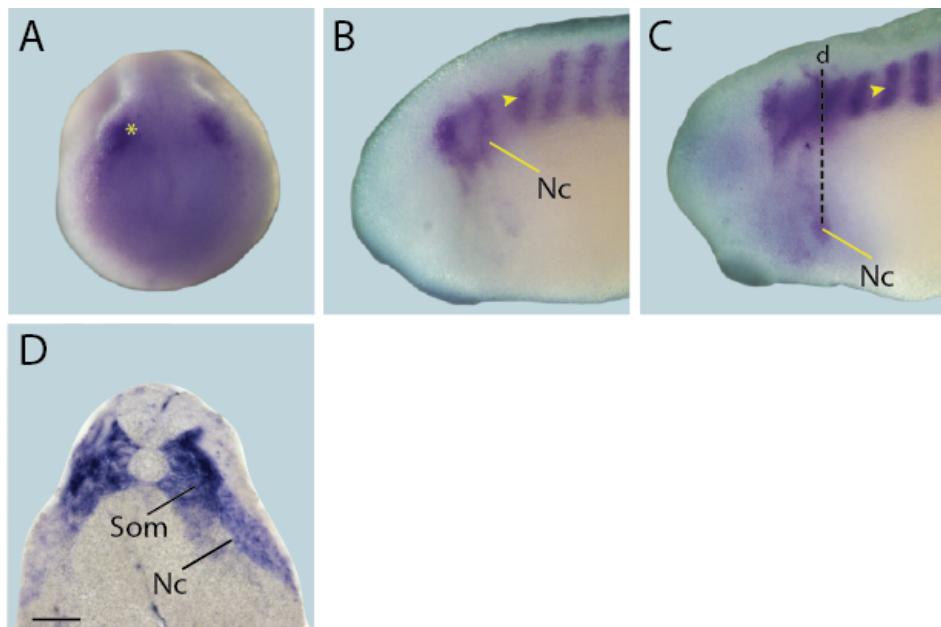
**Fig. 4.3** Expression of *Emx1.2* in whole-mount *Xenopus* embryos. *Emx1.2* is expressed broadly in the neural plate at neural plate stages (A), and becomes restricted to the forebrain (indicated by an asterisk) in late tail bud stages (C).



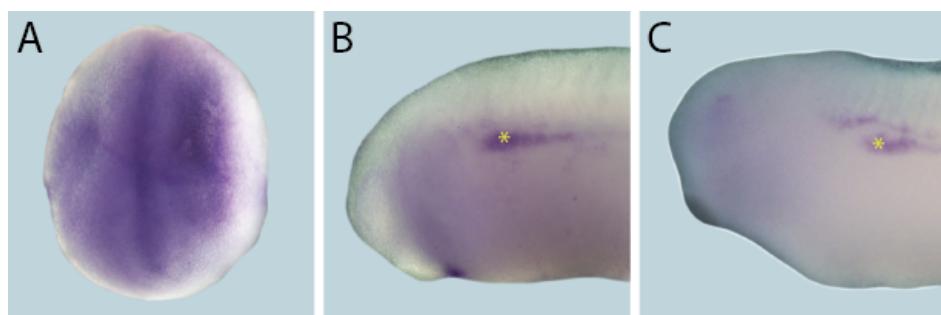
**Fig. 4.4** Expression of *Lhx5* in whole-mount *Xenopus* embryos. *Lhx5* is expressed in the forebrain at all developmental stages (A-C). At early and late tail bud stages *Lhx5* is also expressed in the hindbrain and spinal cord. The forebrain and hindbrain are marked by an asterisk and arrowhead respectively.



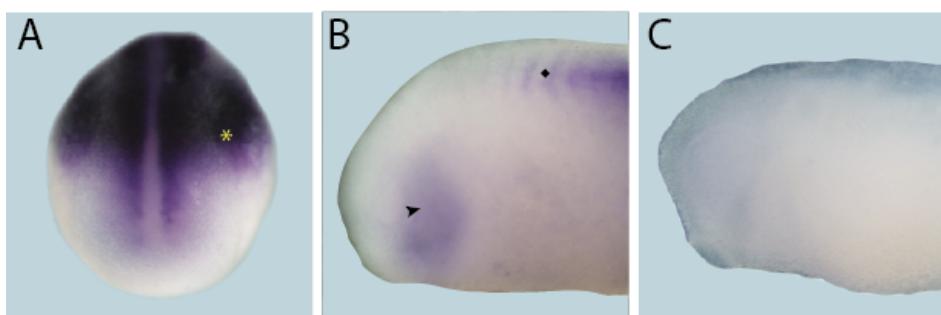
**Fig. 4.5** Expression of *Pou3f2b* in whole-mount *Xenopus* embryos. During neural plate stages (A), *Pou3f2b* is expressed in the neural plate and developing neural tube (shown by an asterisk). Expression in the brain (arrowhead) and spinal cord (asterisk) is maintained during early and late tail bud stages (B and C).



**Fig. 4.6** Expression of *Tbx15* in whole-mount *Xenopus* embryos and sections. During neural plate stages (**A**), *Tbx15* is expressed in a restricted domain of the anterolateral neural folds. At tail bud stages (**B**) expression is prominent in somites and migrating neural crest cells of the hyoid and first branchial neural crest streams (Nc). Both of these expression domains are maintained into late tail bud stages (**C** and **D**). **D** shows section at level indicated in **C** (dotted line). Neural fold expression is marked by an asterisk, and an arrowhead shows somites (Som). Bar in **D**: 100 µm.



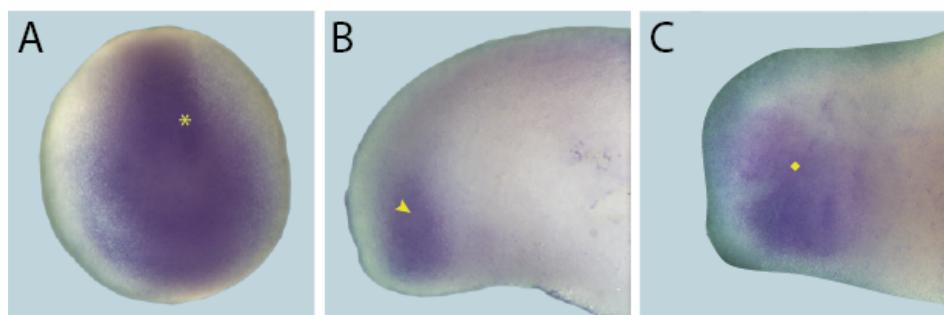
**Fig. 4.7** Expression of *Sim1* in whole-mount *Xenopus* embryos. *Sim1* is expressed in the presumptive pronephros in both early and late tail bud stages (**B** and **C**), as indicated by an asterisk.



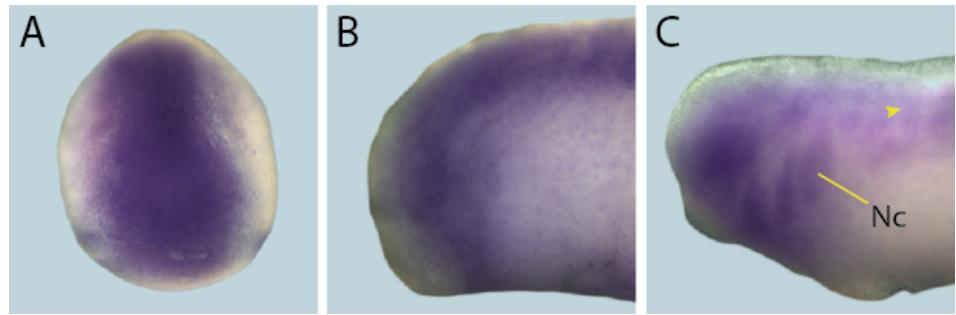
**Fig. 4.8** Expression of *Tbx6* in whole-mount *Xenopus* embryos. Throughout all developmental stages (**A-C**) *Tbx6* is expressed strongly in the posterior paraxial and lateral plate mesoderm (indicated by an asterisk) with weaker expression in the pharyngeal arches (arrowhead). Subsequently, it is expressed in somites, as indicated by a diamond.

#### 4.2.2 Genes expressed broadly in cranial ectoderm including PPE

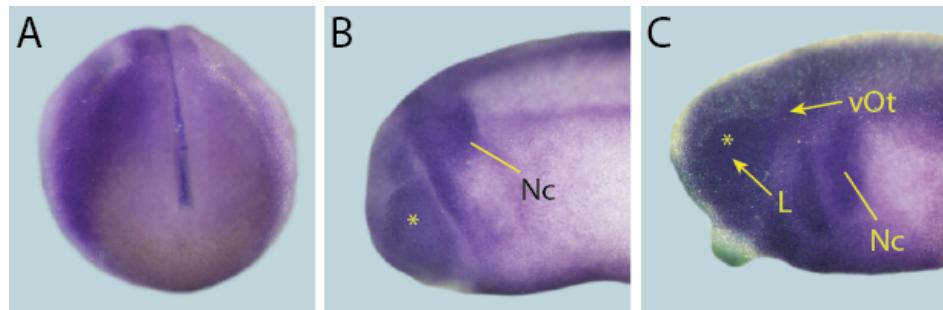
A second group of genes includes those expressed broadly across the cranial ectoderm and at least partially overlap with the PPE at neural plate stages. Included in this group are *Crem* (fig. 4.9), *FosB* (fig. 4.10), *Znf214* (fig. 4.11), *Ripphyl3* (fig. 4.12) and *Gbx2* (Bubnoff et al., 1996; Steventon et al., 2012; data not shown). The basic-leucine zipper (bZIP) transcription factor *Crem* is a member of the CREB (cAMP response element-binding protein) family and, like the immediate early gene *FosB*, promotes neurogenesis in the adult hippocampus (Fujioka, 2004; Jagasia et al., 2009; Yutsudo et al., 2013). Both genes are initially expressed broadly across the cranial mesoderm and ectoderm during neural plate stages. At tail bud stages, *Crem* maintains such broad expression in the pharyngeal arches (fig. 4.9C), whereas *FosB* is additionally expressed in the somites and migrating neural crest stream (fig. 4.10C). *Znf214* (*Znf420*), a zinc finger transcription factor of unknown function is also expressed broadly throughout the ectoderm at neural plate stages. This broad expression is largely confined to the cranial ectoderm at tail bud stages (fig. 4.11B and C) but there is clear concentration in the otic vesicle and lens, as well as in the migrating neural crest stream and retina. *Ripphyl3* is a recently described member of the Ripphey/Bowline family, and is a Groucho-associated co-repressor that interacts with T-box proteins to regulate regional boundaries of gene expression (Janesick et al., 2012). *Ripphyl3* is expressed broadly in the posterior placodal region at neural plate stages (fig. 4.12A), and is implicated as a key factor in positioning and demarcating the boundaries of the PPE (Janesick et al., 2012). However, expression is not maintained in placodes at tail bud stages, where the gene is expressed in the posterior cranial ectoderm (fig. 4.12C). The expression of *Gbx2* has been extensively detailed elsewhere (Bubnoff et al., 1996; B. Li et al., 2009; Simeone, 2000; Steventon et al., 2012), and was not re-analysed here. In published accounts, *Gbx2* is reported to be expressed in the posterior cranial ectoderm, where it is required for early placode specification and later for initiating otic placode-specific gene expression (Steventon et al., 2012).



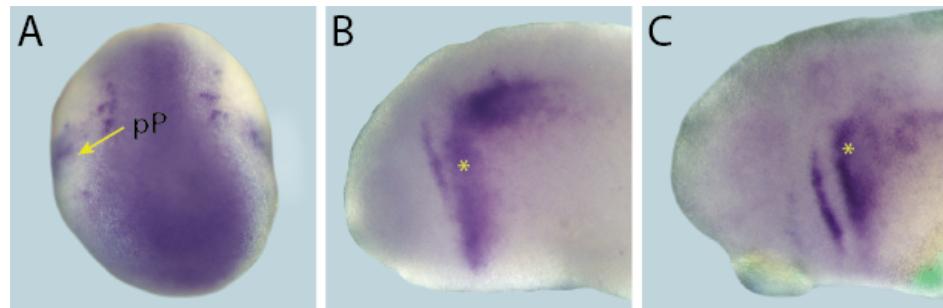
**Fig. 4.9** Expression of *Crem* in whole-mount *Xenopus* embryos. *Crem* is initially expressed broadly in paraxial mesoderm (asterisk) at neural plate stages (A), and in pharyngeal arches at tail bud stages (B and C; arrowhead and diamond, respectively).



**Fig. 4.10** Expression of *FosB* in whole-mount *Xenopus* embryos. *FosB* is expressed in a broad pattern across the cranial ectoderm and trunk mesoderm at both neural fold and early tail bud stages (**A** and **B**). At late tail bud stages (**C**) expression is maintained in cranial ectoderm as well as becoming apparent in the migrating neural crest cells (Nc) as well as weakly in the somites (as indicated by an arrowhead).



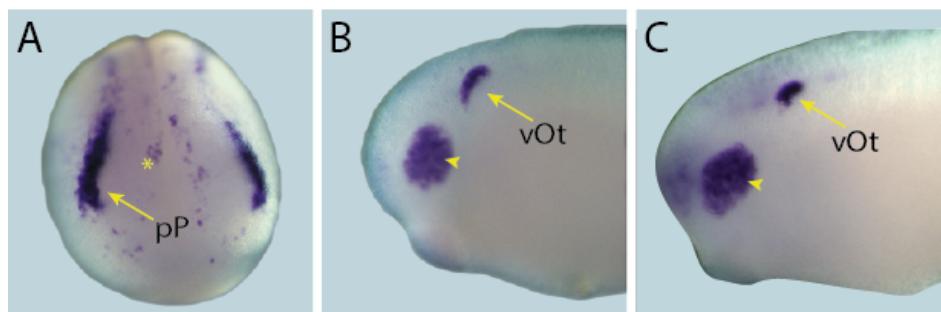
**Fig. 4.11** Expression of *Znf214* in whole-mount *Xenopus* embryos. *Znf214* is expressed broadly across the ectoderm at all developmental stages (**A-C**). At early (**B**) and late (**C**) tail bud stages there is expression in the migrating neural crest cells (Nc) as well as in the retina, and in late tail bud stages *Znf214* is expressed in the otic vesicle and lens placode. The retina is indicated by an asterisk. Abbreviations: L: lens placode; vOt: otic vesicle.



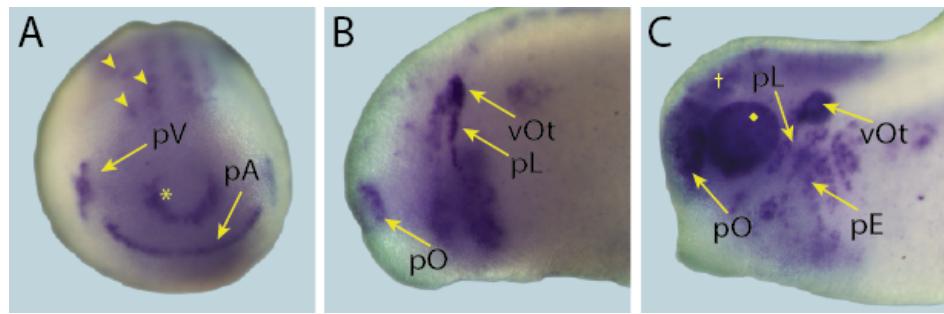
**Fig. 4.12** Expression of *Ripply3* in whole-mount *Xenopus* embryos. *Ripply3* is expressed broadly in the posterior placodal region at neural fold stages (**A**). At both early and late tail bud stages (**B** and **C**) expression is confined to posterior cranial ectoderm (shown by an asterisk). Yellow arrows mark placodal expression. Abbreviations: pP: posterior placodal region.

#### 4.2.3 Placodal genes expressed in proliferating progenitors of PPE

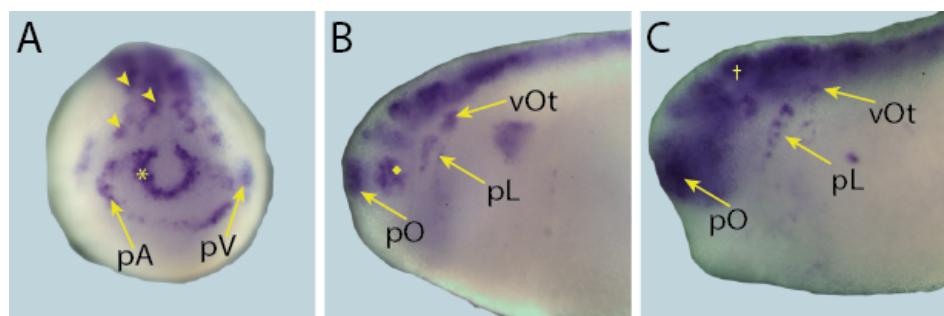
A third group of genes comprises those expressed in the proliferating progenitors of the PPE, and includes *Hes2* (fig. 4.13), *Hes5/8* (fig. 4.14), *Hes9* (fig. 4.15), *Mab21l2b* (fig. 4.16), *Six1* (Pandur and Moody, 2000; data not shown), *Six2* (Ghanbari et al., 2001; data not shown), *Sox2* ((Mizuseki et al., 1998); data not shown), *Sox3* (Penzel et al., 1997; data not shown) and *Sox21* (Cunningham et al., 2008; fig. 4.17). Genes in this group are largely expressed in progenitors of neurogenic placodes, with the exception of *Mab21l2b*, which is lens specific (Kennedy et al., 2004). *Hes* genes are basic helix-loop-helix (bHLH) transcriptional repressors, and are the vertebrate homologs of the *Drosophila* genes *hairy* and *Enhancer of split* genes (Petrovic et al., 2014). *Hes* genes typically inhibit neuronal differentiation and maintain neural progenitor cells via Notch signalling (Kageyama et al., 2014). *Hes2* is predominantly expressed in groups of cells in the posterior placodal region that give rise to sensory organs, and at later stages its expression is confined to the otic placode and retinal precursor cells (Solter, 2006; fig. 4.13C). *Hes5/8* and *Hes9* are dynamically expressed throughout embryogenesis, first in the anterior neural tube, primary neurons and anterior placodal region at neural plate stages (Lamar and Kintner, 2005; Takada et al., 2005; figs. 4.14A and 4.15A). At later stages, expression is restricted to a number of placodes (olfactory, lateral line, otic) and the developing brain (figs. 4.14C and 4.15C).



**Fig. 4.13** Expression of *Hes2* in whole-mount *Xenopus* embryos. During neural plate stages (**A**), *Hes2* is expressed very strongly in a broad region corresponding to the posterior placodal domain including the prospective otic and lateral line placodes, as well as weakly in a scattered subset of neuroectodermal cells (as indicated by an asterisk). Expression is later restricted to the otic vesicle and a new expression domain becomes established in the developing retina at early and late tail bud stages (**B** and **C**). Yellow arrows mark placodal expression, and an arrowhead indicates the developing retina. Abbreviations: pP: posterior placodal region; vOt: otic vesicle.



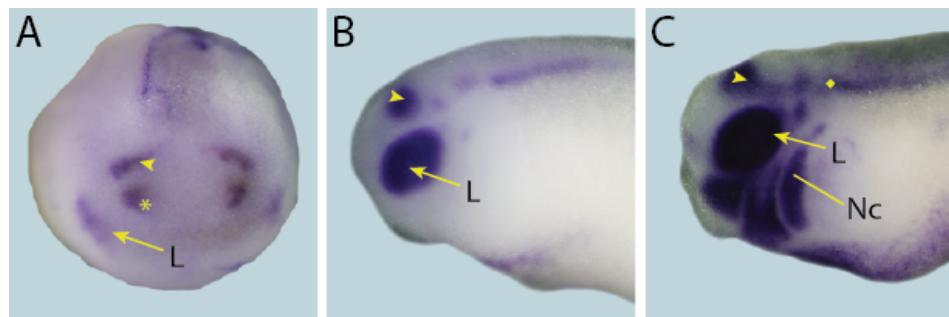
**Fig. 4.14** Expression of *Hes5/8* in whole-mount *Xenopus* embryos. During neural plate stages (**A**), *Hes5/8* is expressed in the developing profundal and trigeminal placodes as well as in the anterior placodal region, the anterior neural plate (asterisk) and the primary neurons (motor neurons, intermediary neurons and sensory neurons) of the posterior neural plate (arrowheads). In early tail bud stages (**B**) trigeminal expression is lost and expression is found in the otic vesicle, as well as lateral line and olfactory placodes. Expression is maintained in all three regions throughout late tail bud stages (**C**) and initiated in epibranchial placodes as well as prominent expression in the retina (diamond) and broadly throughout the brain (cross). Yellow arrows mark placodal expression. Abbreviations: pA: anterior placodal region; pE: epibranchial placode; pL: lateral line placodes; pO: olfactory placode; pV: trigeminal placode; vOt: otic vesicle.



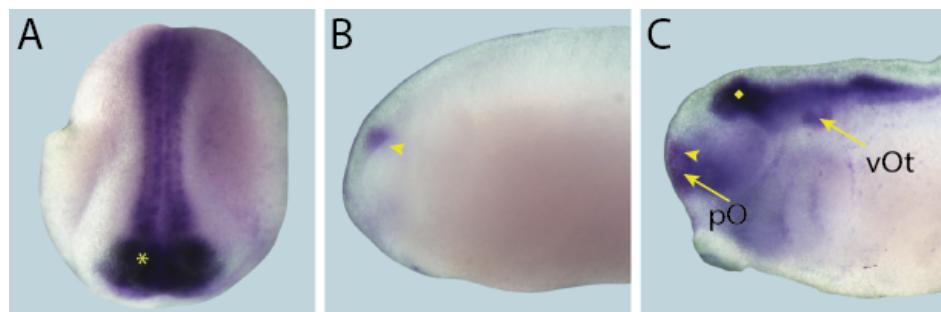
**Fig. 4.15** Expression of *Hes9* in whole-mount *Xenopus* embryos. During neural plate stages (**A**), *Hes9* is expressed in the developing profundal and trigeminal placodes as well as in the anterior placodal region, the anterior neural plate (asterisk) and the primary neurons (motor neurons, intermediary neurons and sensory neurons) of the posterior neural plate (arrowheads). In early tail bud stages (**B**) trigeminal expression is lost and the early anterior placodal expression is apparent in the olfactory placodes, as well as in the otic and lateral line placodes and retina (diamond). In late tail bud stages (**C**) *Hes9* is expressed broadly thought the brain (cross), and is maintained in the lateral line and olfactory placodes as well as the otic vesicle. Yellow arrows mark placodal expression. Abbreviations: pA: anterior placodal region; pL: lateral line placodes; pO: olfactory placode; pV: trigeminal placode; vOt: otic vesicle.

*Mab21l2b* is a vertebrate ortholog of the *Caenorhabditis elegans* *mab21* gene, a developmental regulator of male tail development (Chow and Lang, 2001), and is expressed in proliferating lens progenitors and in the mid and hind brain (Kennedy et al., 2004; fig. 4.16A-C). *Six1/Six2* are members of the Six homeobox family of transcription factors, and are vertebrate homologues of the *Drosophila sine oculis* gene, required for development of the *Drosophila* visual system (Wawersik and Maas, 2000). Both genes are expressed similarly throughout early development (Ghanbari et al., 2001), in a crescent surrounding the anterior neural plate, which marks the PPE, and their expression is maintained in all cranial placodes (Schlosser, 2010). *Sox2/Sox3* genes are members of the Sry-related HMG box-containing gene family (Sox), and are classified as *SoxB1* genes which typically function as transcriptional activators and are expressed in similar domains (Bylund et al., 2003; Kishi et al., 2000). *Sox21*

belongs to the closely related *SoxB2* family and, unlike *SoxB1* family members, promotes differentiation of neural cells by counteracting the activity of *SoxB1* proteins (Sandberg et al., 2005). Both *Sox2* and *Sox3* are expressed in most progenitor cells of the developing nervous system at neural plate stages (Mizuseki et al., 1998; Penzel et al., 1997), as well as in the PPE, where they act to lock in a proliferative progenitor state and block differentiation (Schlosser et al., 2008). *Sox21* expression is generally confined to *SoxB1<sup>+</sup>* cells, and thus overlaps significantly with the expression of *Sox2* and *Sox3* in the developing CNS (Sandberg et al., 2005; fig. 4.17A), which persists into late tail bud stages, as well as in the presumptive forebrain, olfactory placode, and otic vesicle (Cunningham et al., 2008; fig. 4.17C).



**Fig. 4.16** Expression of *Mab21l2b* in whole-mount *Xenopus* embryos. During neural plate stages (A), *Mab21l2b* is expressed in the prospective lens placode, as well as in the eye field (prospective retina) of the forebrain (asterisk) and the prospective midbrain (arrowhead). At early tail bud stages expression in the lens and midbrain is maintained and its expression becomes apparent in the hindbrain (diamond) (B). In late tail bud stages (C) *Mab21l2b* is additionally prominently expressed in migrating neural crest cells (Nc). Yellow arrows mark placodal expression. Abbreviations: L: lens.

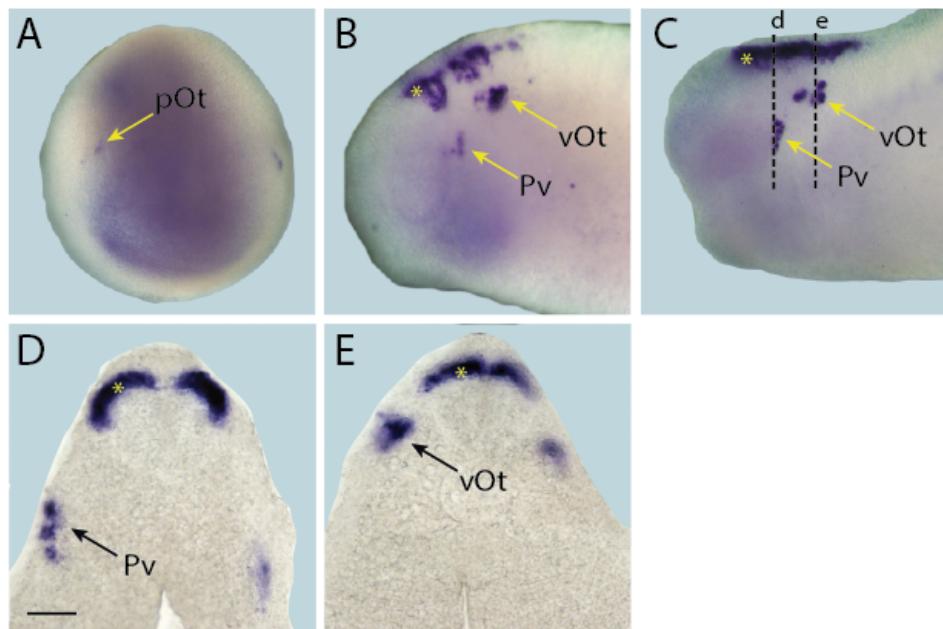


**Fig. 4.17** Expression of *Sox21* in whole-mount *Xenopus* embryos. During neural plate stages *Sox21* is expressed broadly throughout the anterior neural plate (asterisk) (A). This expression becomes confined to the forebrain at early neural plate stages (B), and is maintained into late tail bud stages, in which prominent expression also appears in the forebrain (arrowhead) (C). In late tail bud stages, *Sox21* is also expressed in the olfactory placode, otic vesicle and hindbrain (diamond). Yellow arrows mark placodal. Abbreviations: pO: olfactory placode; vOt: otic vesicle.

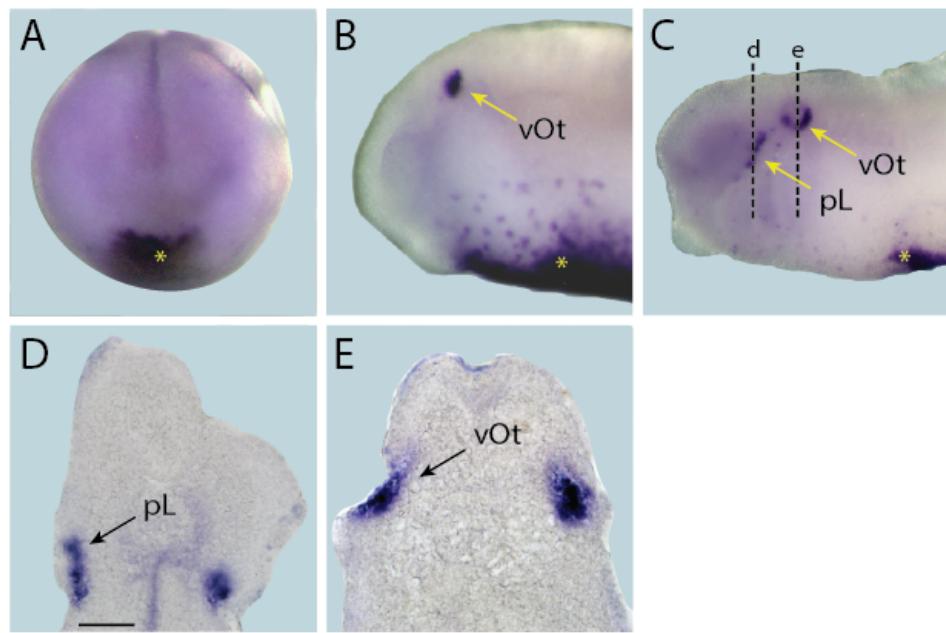
#### 4.2.4 Placodal genes expressed in differentiating cells

The final group of genes includes those expressed in differentiating cells and comprises *Atoh1* (fig. 4.18), *Ngn1* (Nieber et al., 2009; data not shown), *Gfi1a* (fig. 4.19), *Isl2* (fig. 4.20), *Pou4f1.2* (fig. 4.21) and *Tlx1* (fig. 4.22). *Atoh1* is a pro-neural bHLH transcription factor involved in neuronal differentiation and the development of mechanoreceptors. Along with two closely

related bHLH genes, *Ngn1* and *NeuroD1*, *Atoh1* drives neurosensory development in the ear, where it is both necessary and sufficient to induce auditory hair cell differentiation (Bermingham et al., 1999). Very low levels of *Atoh1* expression were detected at neural plate stages in the prospective otic vesicle (P. Kim et al., 1997; fig. 4.18A). Following neural tube closure, *Atoh1* expression becomes prominent in the otic vesicle as well as in the developing trigeminal ganglion and the developing hindbrain (fig. 4.18B-E). At neural plate stages *Ngn1* is transiently expressed in a subset of neurogenic placodes (e.g. profundal and trigeminal placodes), and in cranial ganglia in tail bud stages (Nieber et al., 2009). Both *Ngn1* and *Atoh1* are expressed in proliferating neuronal progenitors, and initiate differentiation by activating pro-neural factors. One such factor is *Gfi1a*, which is crucial for hair cell differentiation, and relies on *Atoh1* for its sustained expression (Chonko et al., 2013). As well as being prominently expressed in haematopoietic stem cells (fig. 4.19B-E), *Gfi1a* expression is confined to the otic vesicle and cranial ganglia in tail bud stages, where it is required for the differentiation and survival of the mechanosensory hair cells of the inner ear, as well as for the survival of cochlear ganglion neurons (Phelan et al., 2010; fig. 4.19C-E).

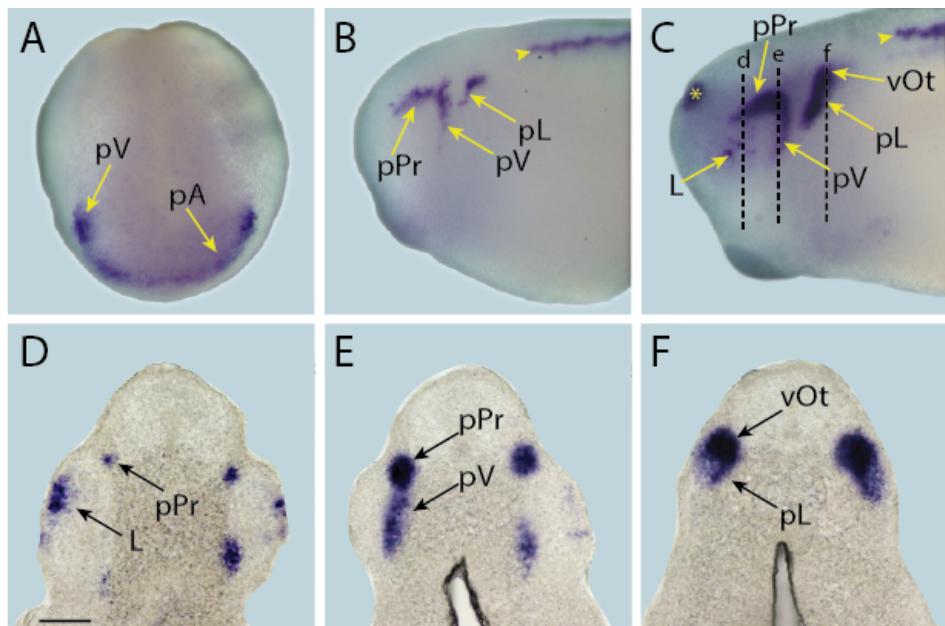


**Fig. 4.18** Expression of *Atoh1* in whole-mount *Xenopus* embryos and sections. During neural plate stages (**A**), *Atoh1* is initially expressed at very low levels in presumptive otic placodes. This expression becomes more pronounced at early tail bud stages (**B**) concomitant with the initiation of expression in trigeminal ganglia and strong expression in the hindbrain (asterisk). Expression becomes more pronounced in all three regions at late tail bud stages (**C-E**). Yellow and black arrows mark placodal expression. Bar in **D**: 100 µm (for **D** and **E**). Abbreviations: pOt: presumptive otic placode; Pv: trigeminal placode; vOt: otic vesicle.

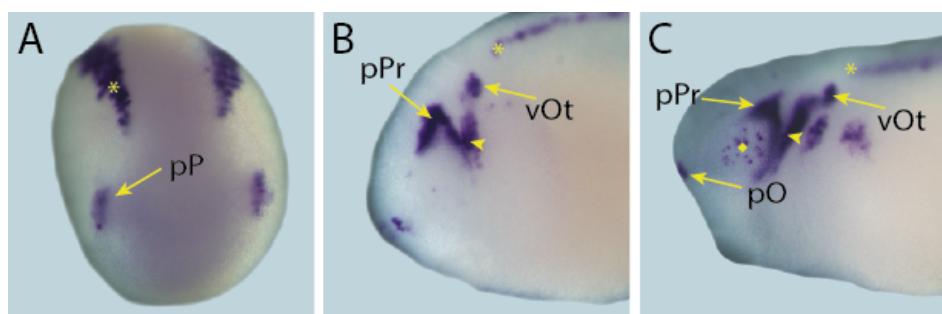


**Fig. 4.19** Expression of *Gfi1a* in whole-mount *Xenopus* embryos and sections. During neural plate stages (**A**), *Gfi1a* is expressed at high levels in haematopoietic cells (asterisk). At early tail bud stages (**B**) this expression becomes more pronounced and diffuse, and expression is also initiated in the otic vesicle. At late tail bud stages (**C-E**) *Gfi1a* is expressed in lateral line placodes as well as otic vesicles as the haematopoietic expression begins to subside. Yellow and black arrows mark placodal expression. Bar in **D**: 100 µm (for **D** and **E**). Abbreviations: pL: lateral line placodes; vOt: otic vesicle.

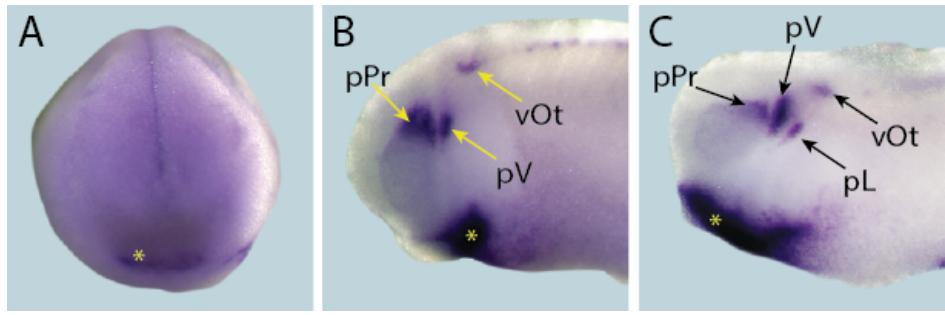
*Isl2* is a LIM domain transcription factor of the LIM-homeodomain subfamily and acts to direct neuron specification with its close homolog *Isl1*, which is important for neuron survival and differentiation (Pfaff et al., 1996). While not fully characterised in *Xenopus*, published accounts of its expression are extremely similar to *Isl1*, and it is expressed in both the trigeminal placode and anterior placodal region at neural plate stages (Bardine et al., 2009; fig. 4.20A), and in several placodes (lateral line trigeminal/profundal) and cranial ganglia at tail bud stages (fig. 4.20C). Its expression in later stages of development is similar to that of both the class-IV POU family member *Pou4f1.2* (*Brn3d*; Hutcheson and Vetter, 2001; fig. 4.21C) and homeodomain gene and sensory ganglia marker, *Tlx1* (*Hox11*; Hutcheson and Vetter, 2001; Patterson and Krieg, 1999; fig. 4.22C), which both show prominent expression in the cranial sensory ganglia. *Isl1* (and so *Isl2*) and *Pou4f1* genes are often co-expressed, and function synergistically in regulating neuronal differentiation and survival, and both are essential factors for sensory neurogenesis and function downstream of the neuronal differential gene *NeuroD1* (Deng et al., 2014).



**Fig. 4.20** Expression of *Isl2* in whole-mount *Xenopus* embryos and sections. During neural plate stages *Isl2* is expressed in the trigeminal placode (**A**) and in the anterior placodal region along the anterior edge of the neural plate. At early tail bud stages *Isl2* expression is maintained in the profundal and trigeminal placodes/ganglia as well as in otic and lateral line placodes/ganglia (e.g the anterodorsal lateral line placode shown here) and primary neurons in the spinal cord (arrowhead) (**B**). Expression is maintained in placodes/cranial ganglia at late tail bud stages and becomes apparent in the forebrain (asterisk) and lens (**C-F**). Yellow and black arrows mark placodal expression. Bar in **D**: 100 µm (for **D-F**). Abbreviations: pA: anterior placodal region pL: lateral line placodes; pPr: profundal placode; L: lens; pV: trigeminal placode; vOt: otic vesicle.



**Fig. 4.21** Expression of *Pou4f1.2* in whole-mount *Xenopus* embryos. During neural plate stages (**A**), *Pou4f1.2* is expressed in the profundal and trigeminal placodes as well as in a stripe of primary sensory neurons (asterisk). In early tail bud stages expression in the profundal/trigeminal placodes/ganglia and primary neurons are both maintained, and expression in the otic and lateral line placodes is upregulated (**B**). Expression is maintained in all domains into late tail bud stages as well as in the retina (diamond) and cranial ganglia (arrowhead) (**C**). Yellow arrows mark placodal expression. Abbreviations: pO: olfactory placode; vOt: otic vesicle; pP: posterior placodes; pV: trigeminal placode.



**Fig. 4.22** Expression of *Tlx1* in whole-mount *Xenopus* embryos. *Tlx1* is expressed in the presumptive ventral visceral arches (asterisk) in neural plate stages (**A**). This is maintained into early and late tail bud stages (**B** and **C**), which also exhibit prominent expression in the profundal/trigeminal placodes/ganglia and otic vesicle. Arrows mark placodal expression. Abbreviations: pL: lateral line placodes; vOt: otic vesicle; pPr: profundal placode; pV: trigeminal placode.

#### 4.2.5 General observations on expression of candidate genes

While almost all of the 25 genes characterised via *in-situ*-hybridisation were expressed in either placodes, or placode derived structures, six were not. Most of these non-placodal genes were expressed either in the adjacent neural plate and neural folds (*Emx1.2*, *Lhx5*, *Pou3f2b*, *Tbx15*) or throughout dorsal tissues (*Sim1*, *Tbx6*) at neural plate stages. Due to these broad or neighbouring expression patterns, it is likely that these genes were included in the screen as a result of contamination from surrounding or underlying tissues (where *Six1*, and possibly *Eya1* are expressed at low levels (Ahrens and Schlosser, 2005)), and thus (with the exception of *Tbx15*) were not considered as potential constituents of a placodal gene regulatory network. Due to its diffuse cranial expression (specifically that underlying the lateral line placodes), *Tbx15* was erroneously included for further analysis before sections were taken, which later revealed expression to be non-placodal (fig. 4.6).

Of the genes expressed broadly across the cranial ectoderm and at least partially overlapping with the PPE, both *Ripply3* and *Gbx2* are known to be involved in placode development (Janesick et al., 2012; B. Li et al., 2009; Steventon et al., 2012), whereas neurogenic factors *Crem*, *FosB* as well as *Znf214* show widespread expression at neural plate stages. Of the remaining candidates expressed in placodes, genes were categorised as being expressed in proliferating progenitors or differentiating cells. While these categories are non-exclusive, considering that a gene may play an early role in proliferation and a later role in differentiation, genes were categorised into such discrete groups in an attempt to represent their predominant expression profile.

The Hes genes are canonical effectors of Notch signalling, and thus are known to promote proliferation by inhibiting neurogenesis. In placodes, *Hes5/8* and *Hes9* show extremely dynamic expression, for example, both genes are expressed in profundal and

trigeminal placodes in neural plate stages but not in early or late tail bud stages (figs. 4.14 and 4.15), suggesting that these genes are transiently expressed in placodes. The final group of genes includes those expressed in differentiating cells. While several of these are expressed in early placodal domains (e.g. at neural plate stages: *Isl2* is expressed in trigeminal placodes and anterior placodal region; *Pou4f1.2* in the posterior placodal region) all of them are prominently expressed in placodally-derived cranial sensory ganglia and are essential for the establishment of one or more sensory organs (Bermingham et al., 1999; Chonko et al., 2013; Deng et al., 2014; Ma et al., 1998; Qian, 2002).

#### 4.2.6 Selection of core sub-set of genes for functional analysis

After the examination of the expression of each gene, a reduced sub-set of genes was selected for further verification and functional analyses. In order to reduce the number of genes considered, and to maximise the chance of revealing cross-regulatory interactions, only transcription factors showing expression in posterior placodes (i.e. those derived from the posterior placodal area; the lateral line, otic and epibranchial placodes) were selected. Since *Six1* and *Six2* show very similar expression (Ghanbari et al., 2001), and *Six1* is already known to positively autoregulate its own expression (Sato et al., 2012), neither *Six1* nor *Six2* were considered for further analysis. *Tbx15* was included before sections were taken, which later revealed its non-placodal expression (fig. 4.6D). This resulted in a set of 11 genes (*Atoh1*, *Gfi1a*, *Hes5/8*, *Hes9*, *Isl2*, *Ngn1*, *Pou4f1.2*, *Sox2*, *Sox3*, *Tlx1* and *Tbx15*) being selected for verification and functional analysis.

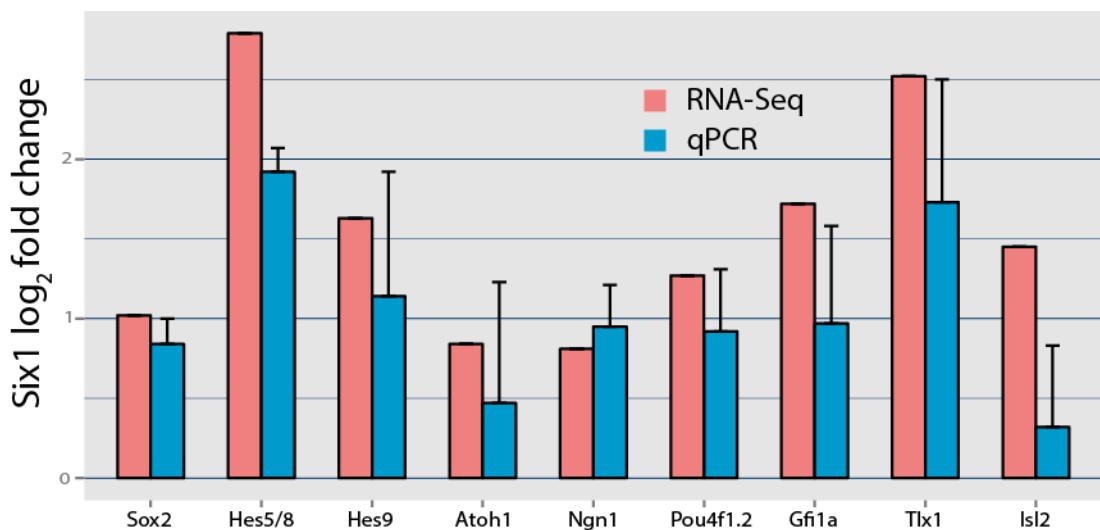
### 4.3 Verification of selected genes as direct target of Six1 and Eya1

To verify that these genes were indeed direct targets of Six1 and Eya1, qPCR was performed on each target after the overexpression of Six1-GR or Eya1-GR in placodal explants (see section 2.2.5). As before, *Six1*-GR or *Eya1*-GR injected embryos treated with CHX were compared against CHX+DEX treated samples. Given that *Sox2* and *Sox3* belong to the same highly conserved Sox subgroup SoxB1 (C. Zhang et al., 2004) and are highly similar at the sequence level (sharing 90% amino acid identity (Kishi et al., 2000)) and are co-expressed in the PPE (Schlosser et al., 2008), *Sox2* was taken to be broadly representative of both genes in the qPCR analysis, and *Sox3* was not assayed.

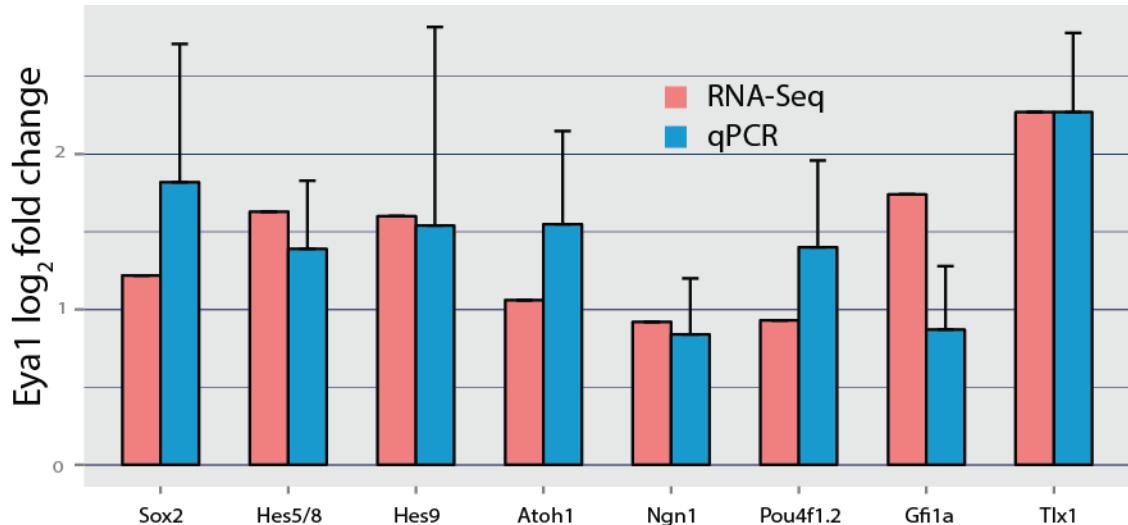
#### 4.3.1 qPCR confirms that overexpression of either Six1 or Eya1 is sufficient to up-regulate target genes

All of the selected targets expressed in placodes show up-regulation after overexpression of *Six1* or *Eya1* (shown as compared against the *Six1<sub>i</sub>* and *Eya1<sub>i</sub>* data sets; figs. 4.23 and 4.24),

providing independent confirmation of their status as direct targets of Six1 and Eya1 in the PPE. As discussed previously (see sections 4.2.1 and 4.2.6), *Tbx15* was included for analysis via qPCR despite not being expressed in placodes, and was not up-regulated in the qPCR data (Six1 FC: -0.01; Eya1 FC: -0.3, data not shown). This can be interpreted in two ways, either that (1) *Tbx15* was a false positive in the RNA-Seq data, and is actually not a direct target of Six1/Eya1, which is reflected in the qPCR data or, (2) the comparatively fewer placodal explants used for each replicate of the qPCR experiment (see section 2.2.5) included RNA from a narrower range of tissue types and developmental stages. If this were the case, while *Tbx15* may still be a direct target of Six1/Eya1 (although in non-placodal tissue), it may not have been sufficiently well represented in the qPCR data for up-regulation to be apparent. Given that *Tbx15* was relatively highly up-regulated in all individual (Six1<sub>i</sub> FC: 2; Eya1<sub>i</sub> FC: 1; Six1+Eya1<sub>i</sub> FC: 1.8), independent, experimental conditions, and that these results were bolstered by statistical significance ( $q < 0.05$ ) in all merged data sets (Six1<sub>m</sub> FC: 1.9; Eya1<sub>m</sub> FC: 1.7; Six1+Eya1<sub>m</sub> FC: 1.7), the latter scenario seems more likely. The same rationalisation may also explain why *Isl2* was not affected after Eya1 overexpression in the qPCR data (FC: -0.4), but showed mild up-regulation in the RNA-Seq data set (FC: 0.3), as well explaining the mild variation (reflected by the standard deviation [SD]) seen in several targets. *Hes9* shows particularly high variation (Six1 SD: 0.78; Eya1 SD: 1.28), possibly reflective of the transiency of its expression in the PPE: An ephemerally expressed gene may show quite different responses to the same treatment at slightly different stages or in explanted regions. With fewer explants, this variation would become more pronounced between replicates (see section 4.2.3; fig. 4.15A).



**Fig. 4.23** qPCR after *Six1* overexpression verifies the majority of genes as direct targets of Six1. Log<sub>2</sub> fold change values were calculated from qPCR data obtained after overexpression of *Six1-GR* in placodal explants and are shown next to fold change values obtained from the RNA-Seq data. In all cases shown, qPCR values broadly corroborate those from the RNA-Seq data - showing up-regulation of target genes after *Six1* overexpression. Vertical error bars show the standard deviation of the mean of biological triplicates.



**Fig. 4.24** qPCR after *Eya1* overexpression verifies the majority of genes as direct targets of *Eya1*. Log<sub>2</sub>fold change values were calculated from qPCR data obtained after overexpression of *Eya1-GR* in placodal explants and are shown next to fold change values obtained from the RNA-Seq data. In all cases shown, qPCR values broadly corroborate those from the RNA-Seq data - showing up-regulation of target genes after *Eya1* overexpression. Vertical error bars show the standard deviation of the mean of biological triplicates.

A further observation of the qPCR data is that while targets generally showed higher up-regulation after *Six1* (as opposed to *Eya1*) overexpression in the RNA-Seq data, the opposite was true for the qPCR data. This can be explained by the observation that targets showed consistently lower FC values in the second biological replicate for the *Six1* experiment compared to the other replicates, possibly reflective of the inherent variability introduced by the experimental set-up (as discussed in section 3.4.1). While this has the effect of further increasing variation between replicates and reducing the average FC across replicates, the overall picture remains clear: Overexpression of either *Six1* or *Eya1* was sufficient to up-regulate all placodally expressed target genes (thus excluding *Tbx15*) consistent with findings from the RNA-Seq data, providing additional and compelling evidence that they are direct targets of *Six1* and *Eya1* in the PPE.

# Chapter 5. Analysis of the role Six1 and Eya1 play in regulating their direct targets

## 5.1 An RNA-Seq experiment to reveal effects of Six1 and Eya1 loss of function in the PPE

In addition to screening for direct target genes of Six1 and Eya1 in the PPE, a further RNA-Seq experiment was conducted to reveal genes affected by either *Six1* or *Eya1* knockdown. Morpholino antisense oligonucleotides (MOs) against *Six1* (Brugmann et al., 2004) and *Eya1* (Schlosser et al., 2008) were unilaterally injected into two-cell stage embryos, and explants taken at neural plate stage as previously described (see section 2.2.5). RNA was extracted from explants using three biological replicates, sequenced using RNA-Seq, and data analysed using the untreated and un-injected explants (which constitutes the PPE transcriptome; see section 3.3) as a control.

### 5.1.1 Initial analysis of RNA-Seq data

To investigate whether Six1 and Eya1 are necessary for the expression of selected targets, gene expression was analysed using RNA-Seq after injection of *Six1*-MO (16 ng) or *Eya1*-MO (20 ng) and placodal dissection as previously described (see section 2.2.5). Affected genes were determined by comparing morpholino-injected embryos to un-injected control embryos. To ensure knockdown efficiency, sibling embryos to those from which explants were taken were analysed via *in-situ*-hybridisation for the expression of the neuronal marker *N-tubulin* (*tubb2b*; Oschwald et al., 1991), which has been demonstrated to be consistently reduced in placodes after injection of *Six1*-MO or *Eya1*-MO (Schlosser et al., 2008). *N-tubulin* was down-regulated in placodes in all assayed replicates, indicating the efficacy of *Six1*-MO (*N-tubulin* reduced after *Six1*-MO injection in 94% [replicate 1, n = 17]; 69% [replicate 2, n = 29]; 75% [replicate 3, n = 24] of embryos) and *Eya1*-MO (93% [replicate 1, n = 27]; 78% [replicate 2, n = 9]; data for replicate 3 unavailable). As for the overexpression screen, the Pearson's correlation between biological replicates within treatment groups was calculated as an initial estimate of data set quality. Replicates showed relatively poor correlation (i.e. *Six1*-MO average correlation: 0.81; *Eya1*-MO average correlation: 0.79), indicating variability in expression profiles within each treatment group. As discussed previously (see section 3.4.1), this may be a methodological artefact, where minor differences between MO concentration and volume, as well as the dissected area, could introduce variation between replicates. However, as three biological replicates were collected for each treatment condition, statistics were available to attempt to resolve this variation, and data were analysed using the same differential expression

criteria as defined previously (see section 2.5.8.3), and filtered for statistical significance ( $q < 0.05$ ). Initial analysis revealed 387 genes significantly down-regulated after *Six1*-MO injection, 597 after *Eya1*-MO injection, 283 up-regulated in *Six1*-MO and 244 in *Eya1*-MO (appendix I.1 - I.4). That 42% of significantly affected genes after *Six1* knockdown were up-regulated (as opposed to 29% after *Eya1* knockdown) again suggests that *Six1* may have a more prominent role as a repressor than *Eya1*.

To further estimate MO efficacy, the lists of significantly down-regulated genes were examined for the presence of *N-tubulin* and neuronal differentiation gene *NeuroD1*, which has been shown to act downstream of *Six1/Eya1* (Ahmed et al., 2012b; Schlosser et al., 2008). *NeuroD1* was found to be highly significantly down-regulated ( $FC \geq 1$ ) in both treatment groups (*Six1*-MO FC: -1.72;  $q = 0.007$ ; *Eya1*-MO FC: -1.79;  $q = 0.03$ ), whereas *N-tubulin* was found to be significantly down-regulated only in *Eya1*-MO, albeit at FC levels insufficient to pass the stringent criteria for differential expression (*Eya1*-MO FC: -0.18;  $q$ -value: 0.003). However, another related neuron marker *Tuj1* (*tubb3*; Ahmed et al., 2012b) was highly significantly down-regulated after *Eya1* knockdown (FC: -3.4;  $q = 0.003$ ), and down-regulated after *Six1* knockdown (FC: -0.9;  $q = 0.16$ ), corroborating previous findings showing that knockdown of either *Six1* or *Eya1* reduces the number of mature neurons (Schlosser et al., 2008).

### 5.1.2 Very few targets are significantly affected after morpholino knockdown of *Six1/Eya1* in RNA-Seq data

Significantly affected genes were further scrutinised for the presence of known (*Six1*-MO: *Ezrin* FC: -0.8; *c-Myc* FC: 0.84. *Eya1*-MO: *CyclinA1* FC: -2.53; *Ezrin* FC: -1.09) or putative (*Six1*-MO: *Ripply3* FC: -2.88; *Crem* FC: -2.07; *Znf214* FC: 1.13. *Eya1*-MO: *Ripply3* FC: -3.38; *Crem* FC: -1.64) direct targets in either data set. Very few known or predicted targets were found, with the vast majority of direct targets identified in this study failing to meet both FC ( $FC \geq 1$ ) and  $q$ -value threshold ( $q < 0.05$ ) (table 5.1). There are several good explanations for why this might be. Firstly, as indicated by the relatively weak correlation between replicates, there simply may have been too much variation between many affected genes for them to have been identified as significantly differentially expressed ( $q > 0.05$ ), which would underestimate the number of genes affected. Secondly, as several of the putative direct targets of *Six1* and *Eya1* are expressed at neural plate stages at relatively low levels (*Atoh1*, *Gfi1a*, *Tlx1*), expression values for these genes may be so low as to exclude them from consideration (FPKM < 1). In this case, genes would be identified in the overexpression screen but not after *Six1/Eya1* knockdown. Thirdly, as this screen captures the response of both direct and indirect targets after either *Six1* or *Eya1* knockdown, multiple and complex indirect

interactions, which may exist downstream of Six1 or Eya1, may obfuscate the interpretation of direct interactions. In this scenario, a gene directly activated by Six1 may also be indirectly repressed, and thus up-regulated after MO-mediated knockdown of *Six1*. As a potential illustration of this effect, while the neuronal differentiation gene *Ngn1* acts downstream of Six1/Eya1 (Ahmed et al., 2012b), knockdown of *Six1* results in a reduction of *Ngn1* expression in placodes in 45% of embryos injected and expansion in 36% (Schlosser et al., 2008; table 5.3). While this phenomenon can be observed via *in-situ*-hybridisation, it would not be evident in RNA-Seq analysis, resulting in low FC values (as a result of pooling explants) and high q-values (as a result of variation between replicates). Additional to this is the difficulty of interpreting knockdown phenotypes, whereby an expression domain can be reduced in area but with more intense expression, or expanded but weakened. While these caveats should be considered during further characterisation, taken together, they serve only to increase the number of false negatives identified, and hence create an overly conservative estimate of the number of genes affected by *Six1* or *Eya1* knockdown.

**Table 5.1** Target gene expression values after injection of *Six1*-MO or *Eya1*-MO

Gene	Control FPKM <sup>a</sup>	<i>Six1</i> -MO FPKM <sup>b</sup>	<i>Six1</i> -MO FC <sup>c</sup>	<i>Six1</i> -MO q-value	Control FPKM <sup>d</sup>	<i>Eya1</i> -MO FPKM <sup>e</sup>	<i>Eya1</i> -MO FC <sup>f</sup>	<i>Eya1</i> -MO q-value
<i>Atoh1</i>	0.54	0.44	-0.31	0.88	0.52	0.39	-0.44	0.86
<i>Gfi1a</i>	1.41	3.20	1.18	0.31	1.37	4.22	1.62	0.09
<i>Hes5/8</i>	4.34	3.61	-0.27	0.57	3.76	2.18	-0.79	0.52
<i>Hes9</i> *	-	-	-	-	-	-	-	-
<i>Isl2</i>	1.37	1.77	0.37	0.46	1.50	1.31	-0.21	0.82
<i>Ngn1</i>	11.36	7.98	-0.51	0.46	11.23	5.29	-1.09	0.15
<i>Pou4f1.2</i>	7.51	6.36	-0.24	0.77	7.21	2.28	-1.66	0.06
<i>Sox2</i>	25.82	20.64	-0.33	0.49	24.68	20.55	-0.27	0.73
<i>Sox3</i>	16.30	23.41	0.52	0.33	21.39	30.35	0.5	0.51
<i>Tlx1</i>	0.50	0.03	-4.02	0.43	0.48	0.01	-5.98	0.56

\* *Hes9* was not identified in either *Six1*-MO or *Eya1*-MO data sets

<sup>a</sup> Expression levels (FPKM) in pre-placodal explants in un-injected embryos

<sup>b</sup> Expression levels (FPKM) in pre-placodal explants embryos injected with *Six1*-MO

<sup>c</sup> Log<sub>2</sub> Fold change values after *Six1*-MO injection

<sup>d</sup> Expression levels (FPKM) in pre-placodal explants in un-injected embryos

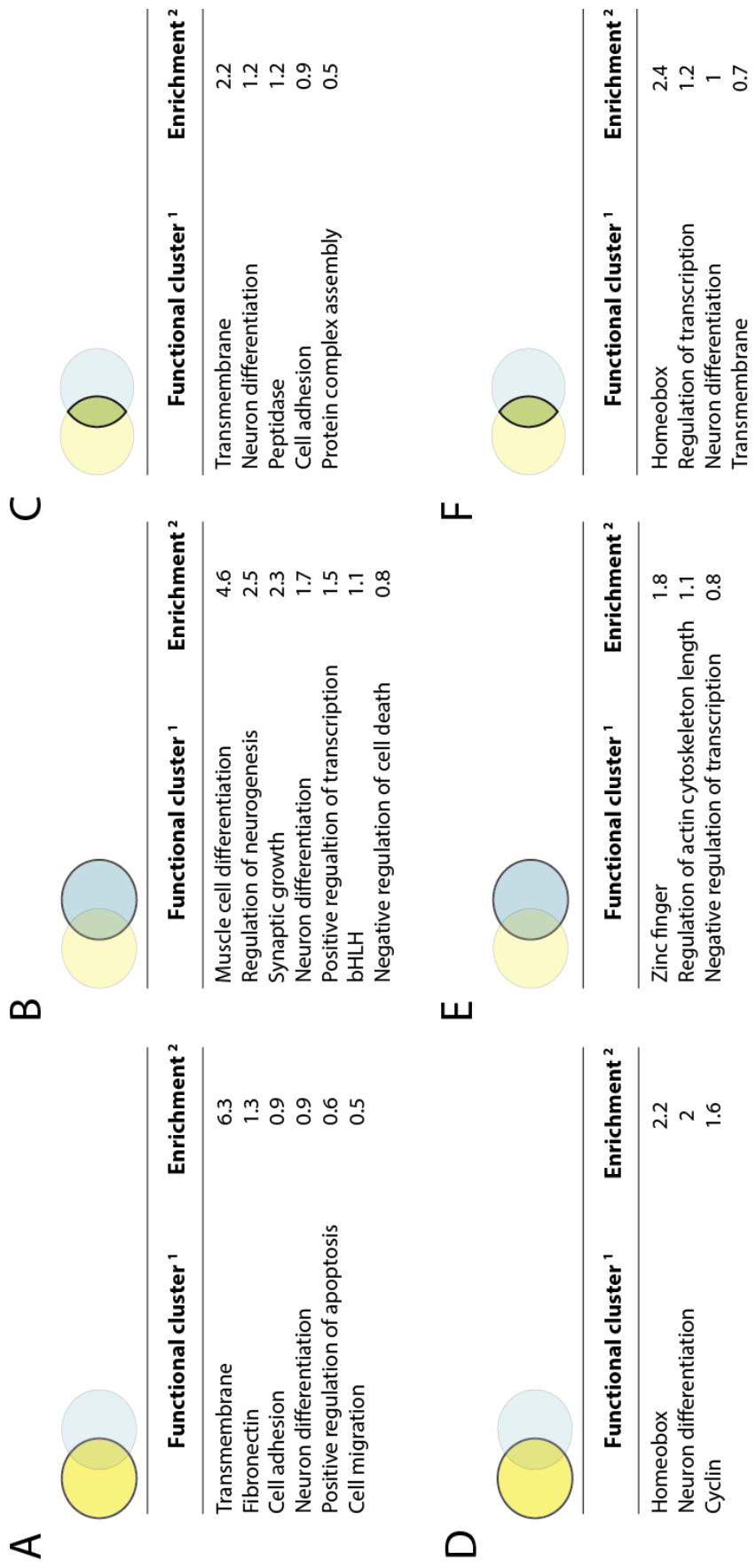
<sup>e</sup> Expression levels (FPKM) in pre-placodal explants embryos injected with *Eya1*-MO

<sup>f</sup> Log<sub>2</sub> Fold change values after *Eya1*-MO injection

### 5.1.3 Gene set enrichment for MO data

As Six1 and Eya1 are known to act synergistically to regulate transcription (Ahmed et al., 2012a; Brugmann et al., 2004; Christophorou et al., 2009; X. Li et al., 2003; Ruf et al., 2004), data were re-analysed in order to find genes similarly affected after *Six1* knockdown as well as *Eya1* knockdown. Here, a gene was considered to be statistically affected in both treatment groups provided that as well as showing statistical significance (q-value < 0.05) in both groups, at least one condition showed at least a two-fold change in expression levels, and the remaining condition had a FC value greater than 0.5 in the same direction (i.e. up-regulated or down-regulated in both treatment groups). This analysis revealed 202 significantly down-

regulated and 127 genes significantly up-regulated genes, in both *Six1*-MO and *Eya1*-MO data sets (appendix I.5 and I.6). To further analyse the extent to which genes affected in one condition were also affected in the other condition, a pairwise comparison between treatment groups was made and tested for significance, revealing that genes affected by *Six1* knockdown were highly enriched for those affected by *Eya1* knockdown ( $p < 0.0001$ ; Fisher's exact test). Gene set enrichment analysis was then conducted on significantly differentially expressed genes affected by *Six1* and *Eya1* both individually and synergistically using DAVID (fig. 5.1; Huang et al., 2009b; 2009a). Genes significantly down-regulated in both the *Six1*-MO and *Eya1*-MO data sets were weakly enriched for several biological processes of relevance for placode development including neuron differentiation (E: 1.2), cell adhesion (E: 0.9) and protein complex assembly (E: 0.5), and significantly up-regulated genes were enriched for homeobox (E: 2.4) and regulation of transcription (E: 1.2), for example. Further examination of the individual data sets shows that genes down-regulated after *Six1* knockdown were enriched for processes similar to the combined analysis, including transmembrane (E: 6.3), cell adhesion (E: 0.9) and neuron differentiation (E: 0.9). However, the *Eya1*-MO down-regulated data set was enriched for processes quite distinct from those in the *Six1*-MO data set, including regulation of muscle cell differentiation (E: 4.6), regulation of neurological system (E: 2.5), synaptic growth (E: 2.3), neuron differentiation (E: 1.7). While this data is prone to the same interpretational caveats as before (see section 5.1.2), it is worth noting that knockdown of *Six1* and *Eya1* appears to affect distinct biological processes.



<sup>1</sup>Functional cluster of terms that show enrichment  
<sup>2</sup>Enrichment for individual conditions; Six1-MO, Eya1-MO and Six1-MO and Eya1-MO

**Fig. 5.1** Gene set enrichment analysis using DAVID, showing clusters of functional terms for different treatment groups. In each case, treatment groups considered are highlighted and outlined in bold in the accompanying Venn diagram. Yellow colouring indicates Six1-MO treatment; blue shows Eya1-MO and green both Six1-MO and Eya1-MO. Enrichment scores  $\geq 0.5$  are reported.  
**(A-C)** Gene set enrichment for genes down-regulated in the various conditions; **(D-F)** for genes up-regulated.

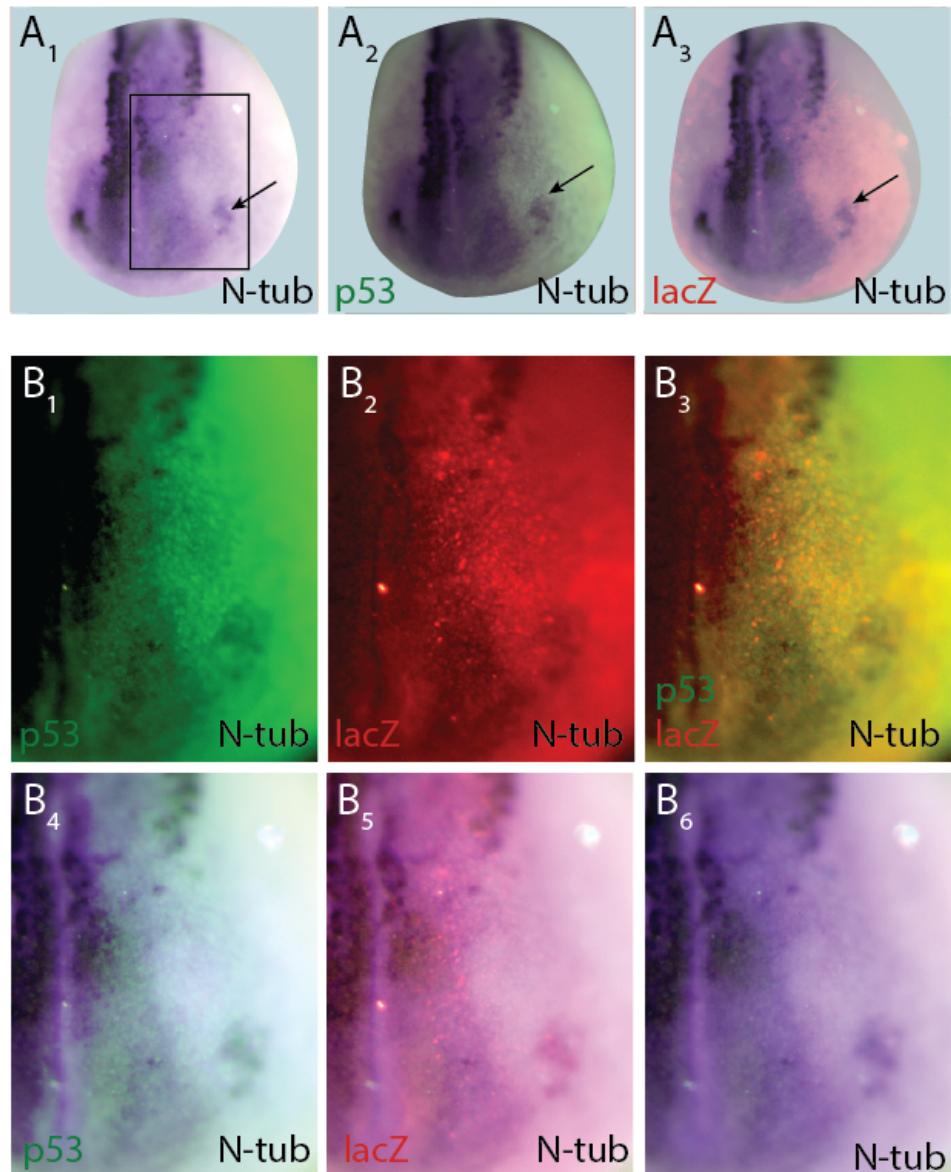
## 5.2 Six1 and Eya1 regulate direct targets in a complex fashion

In addition to the difficulties of interpreting RNA-Seq data derived from MO-injected embryos (see section 5.1.2) a further confounding factor in analysis is the purported tendency of MOs to induce the activation of the tumour suppressor protein p53 (and thus apoptosis), reported to be responsible for off-target effects (Kok et al., 2015; Robu et al., 2007; Schulte-Merker and Stainier, 2014). Such off-target effects may therefore result in gene expression dynamics unrelated to the silencing of the targeted gene. Indeed, *p53* is found significantly up-regulated in both RNA-Seq datasets after injection of relatively high-dose MOs (*Six1*-MO FC: 1; q = 0.006; *Eya1*-MO FC: 1; q = 0.03), potentially representing a significant difficulty in interpreting these results. However, little is known about the mechanism by which MOs activate *p53*, and it seems clear that such off-target effects can be mitigated by conducting appropriate controls such as dosage titration for each MO injected (Rossi et al., 2015), as well as using mismatch MOs to control for toxicity (Eisen and Smith, 2008). Accordingly, as a precaution against *p53*-induced off-target effects, MO dosage titration was conducted (table 5.2) using expression of *N-tubulin* in placodes as a marker for MO efficacy. While high-dose MOs (*Six1*-MO: 16 ng; *Eya1*-MO: 20 ng) resulted in consistent and strong reduction of *N-tubulin* in placodes, *p53* was also found to be activated in the injected area in several cases (fig. 5.2). However, and congruent with other reports (Eisen and Smith, 2008; Rossi et al., 2015), even a ten-fold reduction of MO dose (*Six1*-MO: 2 ng; *Eya1*-MO: 2 ng) was able to consistently reduce *N-tubulin* expression without affecting *p53* (fig. 5.3). To alleviate the possibility of inducing *p53* (and thus off-target effects), such low-dose MO injections were therefore used for all subsequent functional analysis.

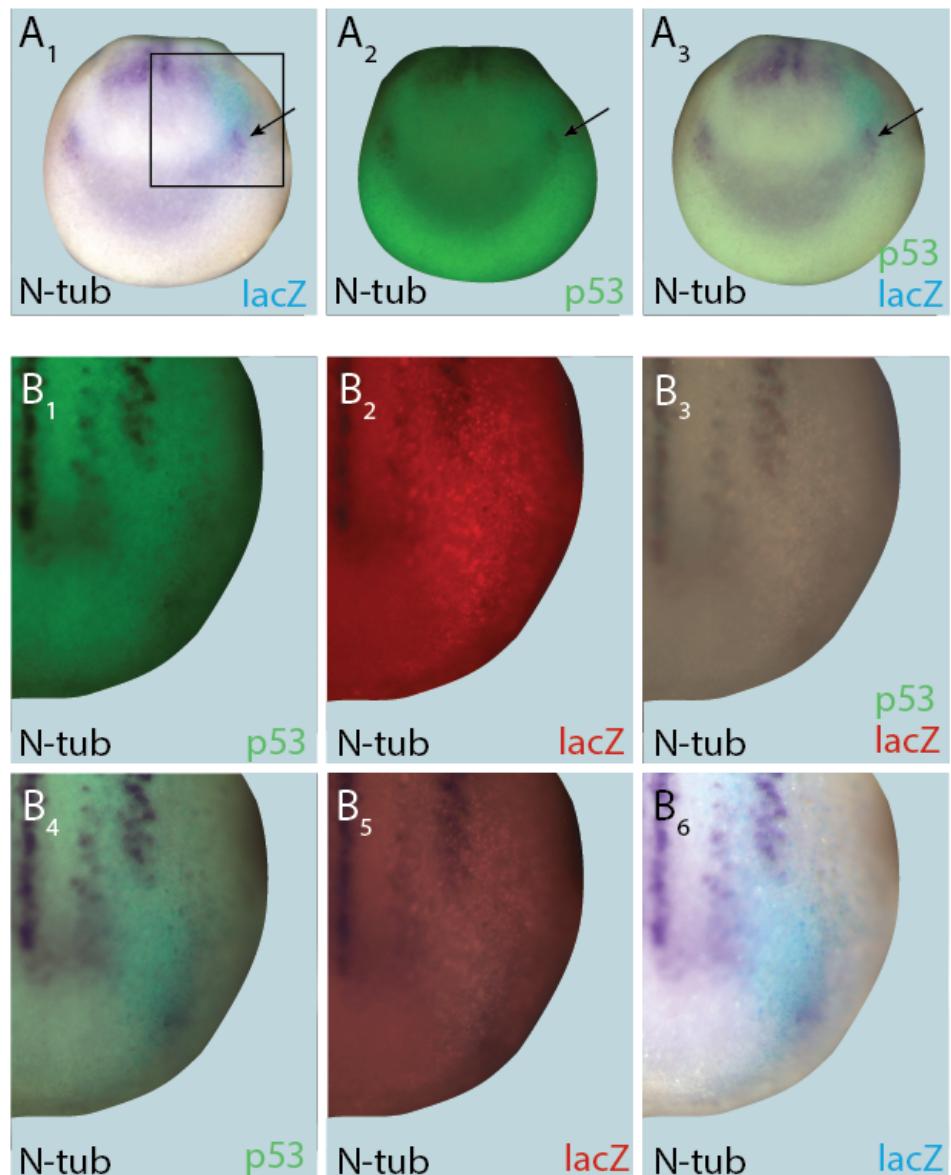
**Table 5.2** Dosage titration of *Six1*-MO and *Eya1*-MO

	<b>Injection</b>	<i>Six1</i> -MO (16 ng)	<i>Six1</i> -MO (8 ng)	<i>Six1</i> -MO (2 ng)	<i>Eya1</i> -MO (20 ng)	<i>Eya1</i> -MO (5 ng)	<i>Eya1</i> -MO (2.5 ng)
	<b>Phenotype</b>	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)
<i>N-tubulin</i>	Reduced	62 (156)	51 (67)	65 (82)	91 (53)	59 (17)	100 (10)

n: number of embryos analysed at both neural plate (stage 14-16) and tail bud (stage 21-26) stage



**Fig. 5.2** Injection of high concentrations of *Eya1*-MO can induce *p53* activation. Panels show expression of *N-tubulin* at neural plate stages after injection of *Eya1*-MO (20 ng), as marked by distribution of lacZ-immunopositive cells. Panels **A<sub>1</sub>-A<sub>3</sub>** show distribution of *N-tubulin* expression in bright field (**A<sub>1</sub>**), superimposed with green fluorescence (p53<sup>+</sup>; **A<sub>2</sub>**) and superimposed with red fluorescence (lacZ<sup>+</sup>; **A<sub>3</sub>**) channels. Arrows in panels **A<sub>1</sub>-A<sub>3</sub>** show reduction of *N-tubulin* in placodes. Panels **B<sub>1</sub>-B<sub>6</sub>** show a magnified region of the embryos shown in panels **A<sub>1</sub>-A<sub>3</sub>**, as indicated by a black box in **A<sub>1</sub>**. **B<sub>1</sub>** and **B<sub>2</sub>** show green (p53<sup>+</sup>; **B<sub>1</sub>**) and red (lacZ<sup>+</sup>; **B<sub>2</sub>**) channels alone and **B<sub>3</sub>** shows the merged image. **B<sub>4</sub>-B<sub>6</sub>** show bright-field images superimposed with green fluorescence (p53<sup>+</sup>; **B<sub>4</sub>**), red fluorescence (lacZ<sup>+</sup>; **B<sub>5</sub>**), or alone (**B<sub>6</sub>**).

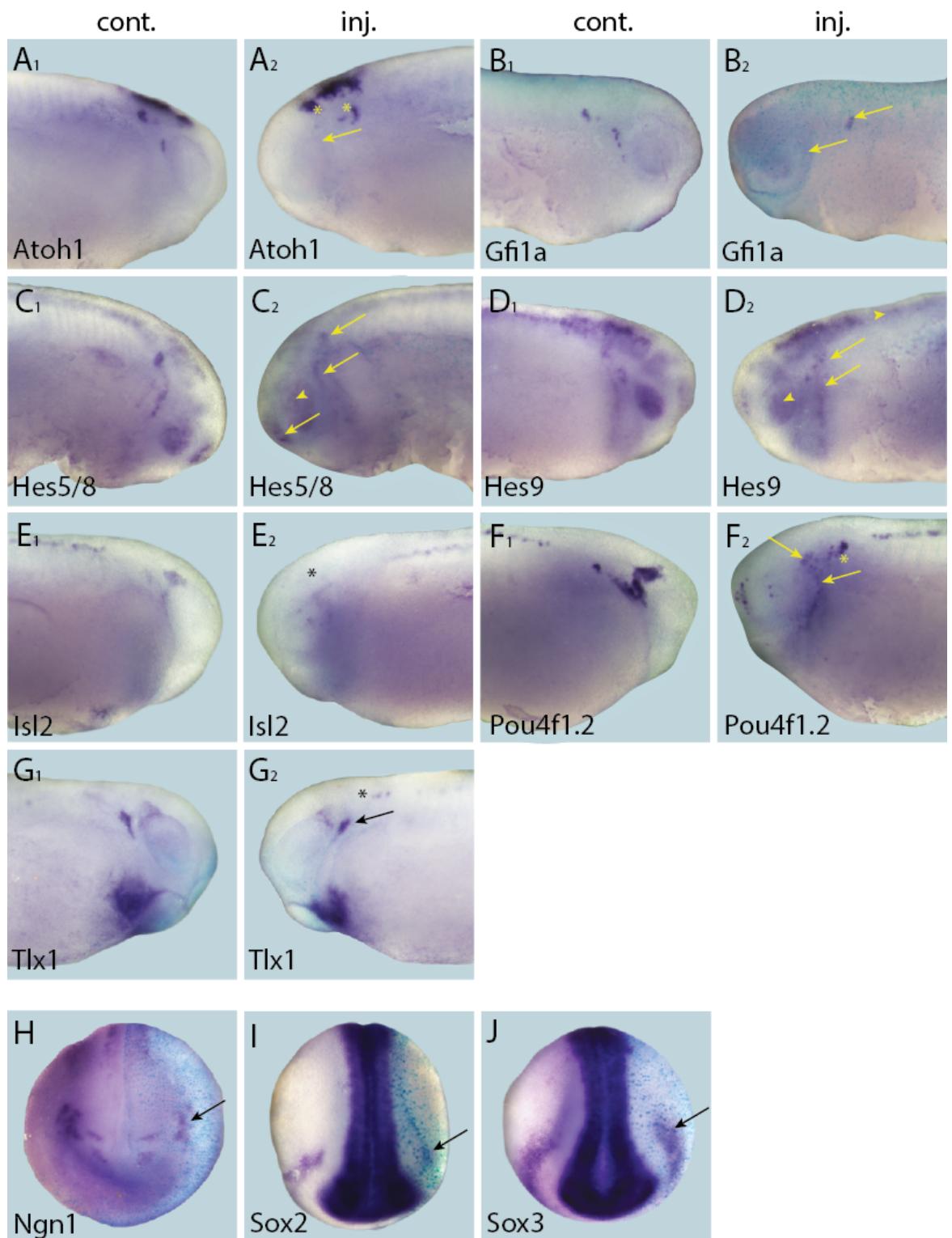


**Fig. 5.3** Injection of low concentrations of *Eya1*-MO does not lead to *p53* activation. Panels show expression of *N-tubulin* at neural plate stages after injection of *Eya1*-MO (4 ng), as marked by distribution of *lacZ*<sup>+</sup> cells (light blue staining in **A**<sub>1</sub>, **A**<sub>3</sub>, **B**<sub>3</sub> and **B**<sub>4</sub>-**B**<sub>6</sub>; red fluorescence in **B**<sub>2</sub>, **B**<sub>5</sub> and **B**<sub>6</sub>). Panels **A**<sub>1</sub>-**A**<sub>3</sub> show bright field whole-embryo distribution of *N-tubulin* expression (**A**<sub>1</sub>), green channel alone (*p53*<sup>+</sup>; **A**<sub>2</sub>) and *N-tubulin* superimposed with green fluorescence (*p53*<sup>+</sup>; **A**<sub>3</sub>). Arrows in panels **A**<sub>1</sub>-**A**<sub>3</sub> show reduction of *N-tubulin* in placodes. Panels **B**<sub>1</sub>-**B**<sub>6</sub> show a magnified region of the embryos shown in panels **A**<sub>1</sub>-**A**<sub>3</sub>, as indicated by a black box in **A**<sub>1</sub>. **B**<sub>1</sub> and **B**<sub>2</sub> show green (*p53*<sup>+</sup>; **B**<sub>1</sub>) and red (*lacZ*<sup>+</sup>; **B**<sub>2</sub>) channels alone and **B**<sub>3</sub> shows the merged image. **B**<sub>4</sub>-**B**<sub>6</sub> show bright-field images superimposed with green fluorescence (*p53*<sup>+</sup>; **B**<sub>4</sub>), red fluorescence (*lacZ*<sup>+</sup>; **B**<sub>5</sub>), or alone (**B**<sub>6</sub>).

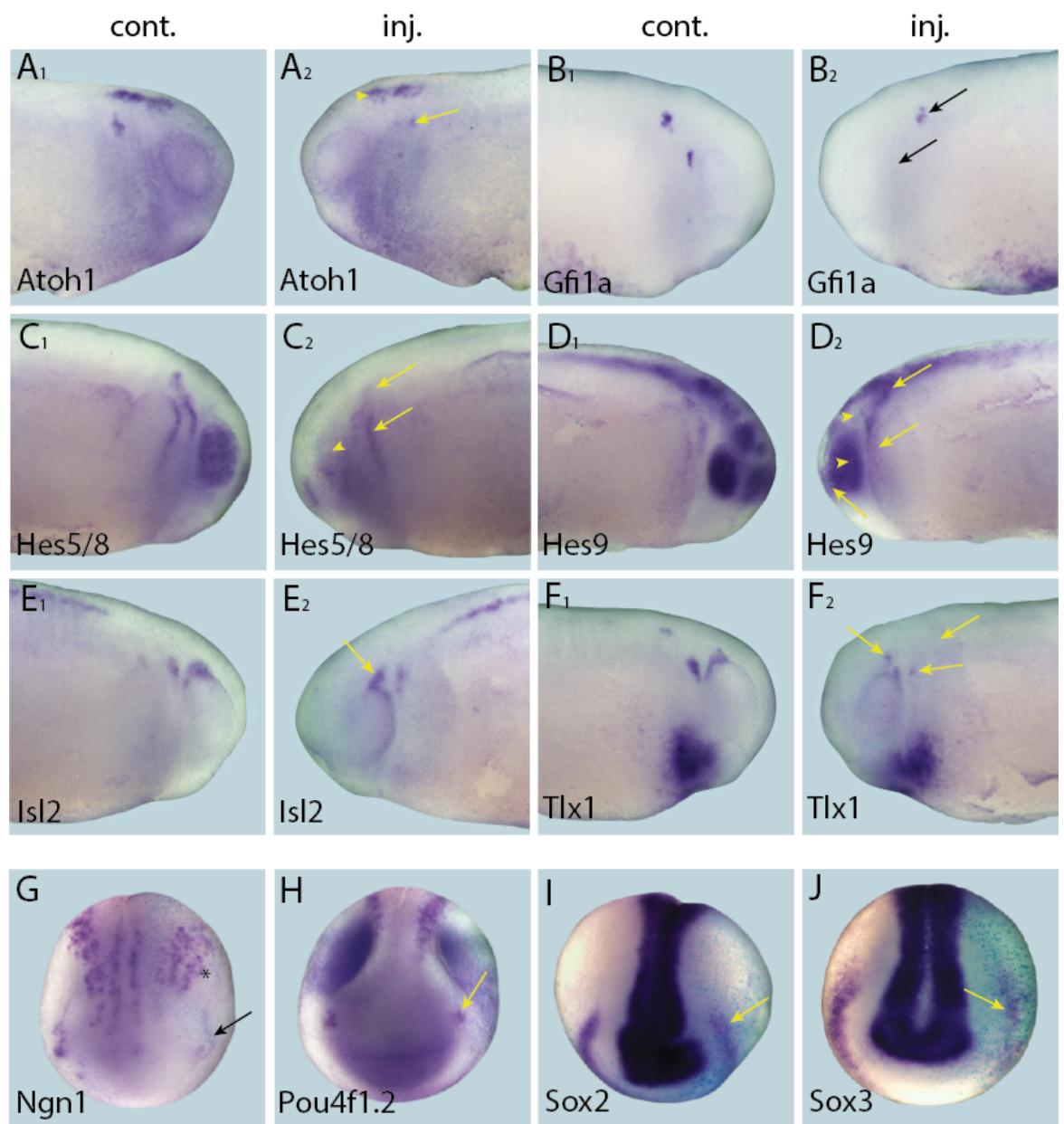
### 5.2.1 *Six1* and *Eya1* are necessary for the expression of direct targets

In order to validate the RNA-Seq results, and to explore whether *Six1* or *Eya1* are necessary for the expression of selected target genes, the expression of each target was analysed by *in-situ*-hybridisation after knockdown of *Six1* or *Eya1*. The efficacy and specificity of both *Six1*-MOs (*Six1*-MO1 and *Six1*-MO2; Brugmann et al., 2004) and both *Eya1*-MOs (*Eya1*-MO1 and *Eya1*-MO2; Schlosser et al., 2008) have been previously reported. While a relatively high concentration of MO (20 ng and 16 ng of *Eya1*-MO and *Six1*-MO, respectively) was injected for RNA-Seq experiments, a lower amount was used in follow-up experiments as a preventative measure against MO-induced *p53* activation (Rossi et al., 2015). Knockdown of

either *Six1* or *Eya1* reduced the expression of all direct target genes in the placodes of at least a subset of embryos, demonstrating that both *Six1* and *Eya1* are required for their expression (figs. 5.4 and 5.5). In all cases, injection of a control-MO (*Eya1*-mmMO) resulted in statistically less frequent effects on all targets analysed, compared to injection of either *Six1*-MO or *Eya1*-MO (table 5.3; appendix J). In addition to reducing target gene expression in placodes, in several instances non-placodal domains were also affected, suggesting that even though *Six1* and *Eya1* are not prominently expressed outside of the PPE at neural plate stage, it is likely that they are expressed at low levels and have a function in other domains (for example this can be seen by changes in expression in *Atoh1* [fig 5.4A and 5.5A] and *Hes9* [fig 5.4D and 5.5D]; arrowheads).



**Fig. 5.4** Effects of *Six1* knockdown on target genes. Tail bud stage (**A - G**) and neural plate stage (**H-J**) embryos after unilateral injection of *Six1*-MO1+2. In each case, *lacZ* was co-injected as a lineage tracer and panels **A<sub>1</sub> - G<sub>1</sub>** show the control (un-injected) side and **A<sub>2</sub> - G<sub>2</sub>** show the injected side. The injected side is positioned to the right in **H - J**, as marked by blue *lacZ* staining. Arrows and arrowheads mark reductions in marker gene expression in placodal and non-placodal derivatives, respectively, and asterisks indicate increased expression.



**Fig. 5.5** Effects of Eya1 knockdown on target genes. Tail bud stage (**A - F**) and neural plate stage (**G - J**) embryos after unilateral injection of *Eya1*-MO1+2. In each case, *lacZ* was co-injected as a lineage tracer and panels **A<sub>1</sub> - F<sub>1</sub>** show the control (un-injected) side and **A<sub>2</sub> - F<sub>2</sub>** show the injected side. The injected side is positioned to the right in **G - J**, as marked by blue *lacZ* staining. Arrows and arrowheads mark reductions in marker gene expression in placodal and non-placodal derivatives, respectively, and asterisks indicate increased expression.

**Table 5.3** Changes in marker gene expression in the placodes after injection of various constructs

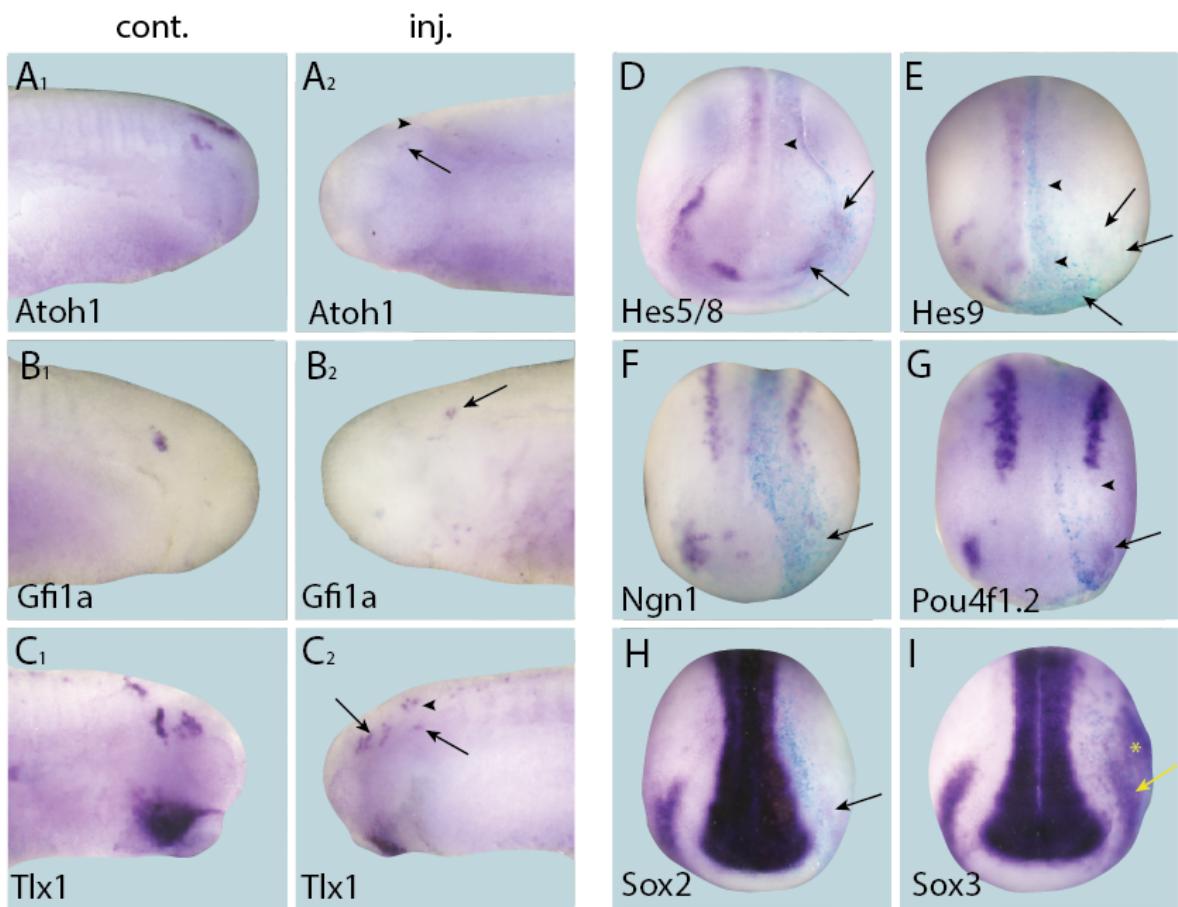
	<b>Injection</b>	<i>Six1-MO</i> <sup>a</sup>	<i>Eya1-MO</i> <sup>a</sup>	<i>Six1-EnR</i>	<i>Six1-GR</i> <sup>b</sup>	<i>Eya1-GR</i> <sup>b</sup>	<i>Eya1-mmMO</i>
	<b>Phenotype</b>	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)
<i>Atoh1</i>	Reduced	77** (26)	90** (20)	94 (18)	26 (19)	42 (12)	10 (21)
	Increased	0 (26)	0 (20)	0 (18)	35 (17)	42 (12)	0 (21)
<i>Gfi1a</i>	Reduced	82* (27)	67* (17)	69 (16)	57 (14)	36 (14)	31 (26)
	Increased	0 (27)	0 (17)	0 (16)	7 (14)	43 (14)	0 (26)
<i>Hes5/8</i>	Reduced	74** (35)	83** (35)	70 (46)	60 (40)	57 (56)	17 (24)
	Increased	0 (35)	0 (35)	24 (46)	15 (40)	29 (56)	0 (24)
<i>Hes9</i>	Reduced	73** (45)	76** (33)	84 (38)	75 (12)	29 (29)	11 (27)
	Increased	0 (45)	0 (33)	8 (38)	0 (12)	0 (29)	0 (27)
<i>Isl2</i>	Reduced	66* (38)	100** (17)	nd	50 (18)	24 (17)	27 (22)
	Increased	6 (38)	0 (17)	nd	31 (16)	41 (17)	0 (22)
<i>Ngn1</i>	Reduced	65** (51)	49* (43)	84 (31)	17 (30)	36 (59)	17 (24)
	Increased	0 (51)	16 (43)	6 (31)	23 (30)	41 (59)	4 (24)
<i>Pou4f1.2</i>	Reduced	67** (48)	63* (30)	71 (35)	47 (15)	81 (37)	16 (19)
	Increased	0 (48)	0 (30)	0 (35)	13 (15)	0 (37)	0 (19)
<i>Sox2</i>	Reduced	74** (19)	78** (18)	87 (30)	90 (21)	48 (33)	6 (16)
	Increased	0 (19)	0 (18)	23 <sup>c</sup> (30)	0 (21)	12 (33)	0 (16)
<i>Sox3</i>	Reduced	68** (25)	54* (26)	39 (31)	49 (25)	40 (23)	9 (22)
	Increased	0 (25)	0 (26)	71 <sup>c</sup> (31)	16 (25)	17 (23)	0 (22)
<i>Tlx1</i>	Reduced	84* (31)	91** (32)	100 (13)	40 (10)	7 (15)	33 (15)
	Increased	6 (31)	0 (32)	0 (13)	40 (10)	73 (15)	0 (15)
<i>N-tubulin</i>	Reduced	65 (82)	100* (10)	88 (41)	53 (17)	40 (15)	43 (21)
	Increased	0 (82)	0 (10)	0 (41)	12 (17)	40 (15)	0 (21)

n: number of embryos analysed at both neural plate (stage 14-16) and tail bud (stage 21-26) stage

nd: not determined

<sup>a</sup> Significant differences (Fisher's exact test; \*: p < 0.05, \*\*: p < 0.001) to *Eya1-mmMO* injections are indicated<sup>b</sup> Dexamethasone treatment from stages 16-18 on<sup>c</sup> Expression ectopic in epidermis

As well as reducing target gene expression, knockdown of *Six1* or *Eya1* occasionally led to an increased expression in placodal and non-placodal domains (expansion of expression domain after injection of *Six1*-MO in *Isl2* [6%] and *Tlx1* [6%] or *Eya1*-MO in *Ngn1* [16%]), indicative of indirect interactions as discussed in section 5.1.2. As a further control for off-target effects associated with MO use, target gene expression was analysed after overexpression of a dominant-negative version of *Six1* (*Six1-EnR*; Brugmann et al., 2004). Expression patterns of all target genes were highly similar to those seen after MO knockdown of either *Six1* or *Eya1*, providing additional support for the reductions in expression being caused by *Six1/Eya1* knockdown as opposed to being an artefact of MO use (fig. 5.6). In addition to reducing the expression of direct targets, injection of *Six1*-MO, *Eya1*-MO or *Six1-EnR* resulted in substantial loss of placodal neurons, as evidenced by the reduction in *N-tubulin* expression in all neurogenic placodes (table 5.3), supporting previous reports of *Six1* and *Eya1* being required for neuronal differentiation in placodes (Ikeda et al., 2007; Schlosser et al., 2008; Zou et al., 2006). Taken together these results demonstrate that loss of *Six1* and *Eya1* function interferes with both progenitor (*Hes5/8*, *Hes9*, *Sox2*, *Sox3*) and differentiation (*Atoh1*, *Tlx1*, *Gfi1a*, *Pou4f1.2*, *Ngn1*, *Isl2*, *N-tubulin*) programmes of neuronal development, and establishes all assayed genes as downstream targets of *Six1* and *Eya1*.

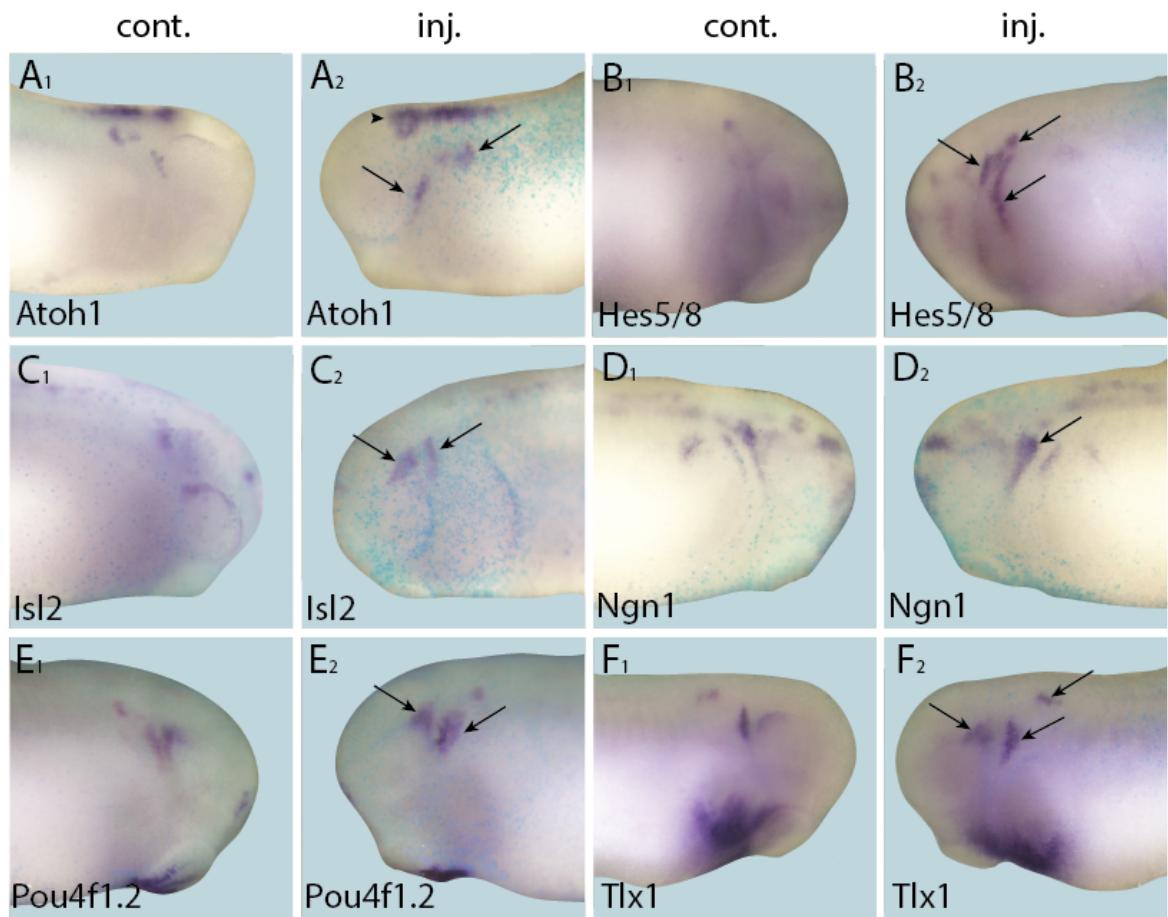


**Fig. 5.6** Down-regulation of *Six1* target genes by *Six1-EnR* injection. Tail bud stage (**A - C**) and neural plate stage (**D - I**) embryos after unilateral injection of *Six1-EnR*. In each case, *lacZ* was co-injected as a lineage tracer and panels **A<sub>1</sub>** - **C<sub>1</sub>** show the control (un-injected) side and **A<sub>2</sub>** - **C<sub>2</sub>** show the injected side. The injected side is positioned to the right in **D - I**, as marked by blue *lacZ* staining. Arrows and arrowheads mark reductions in marker gene expression in placodal and non-placodal derivatives, respectively, and asterisks indicate increased expression.

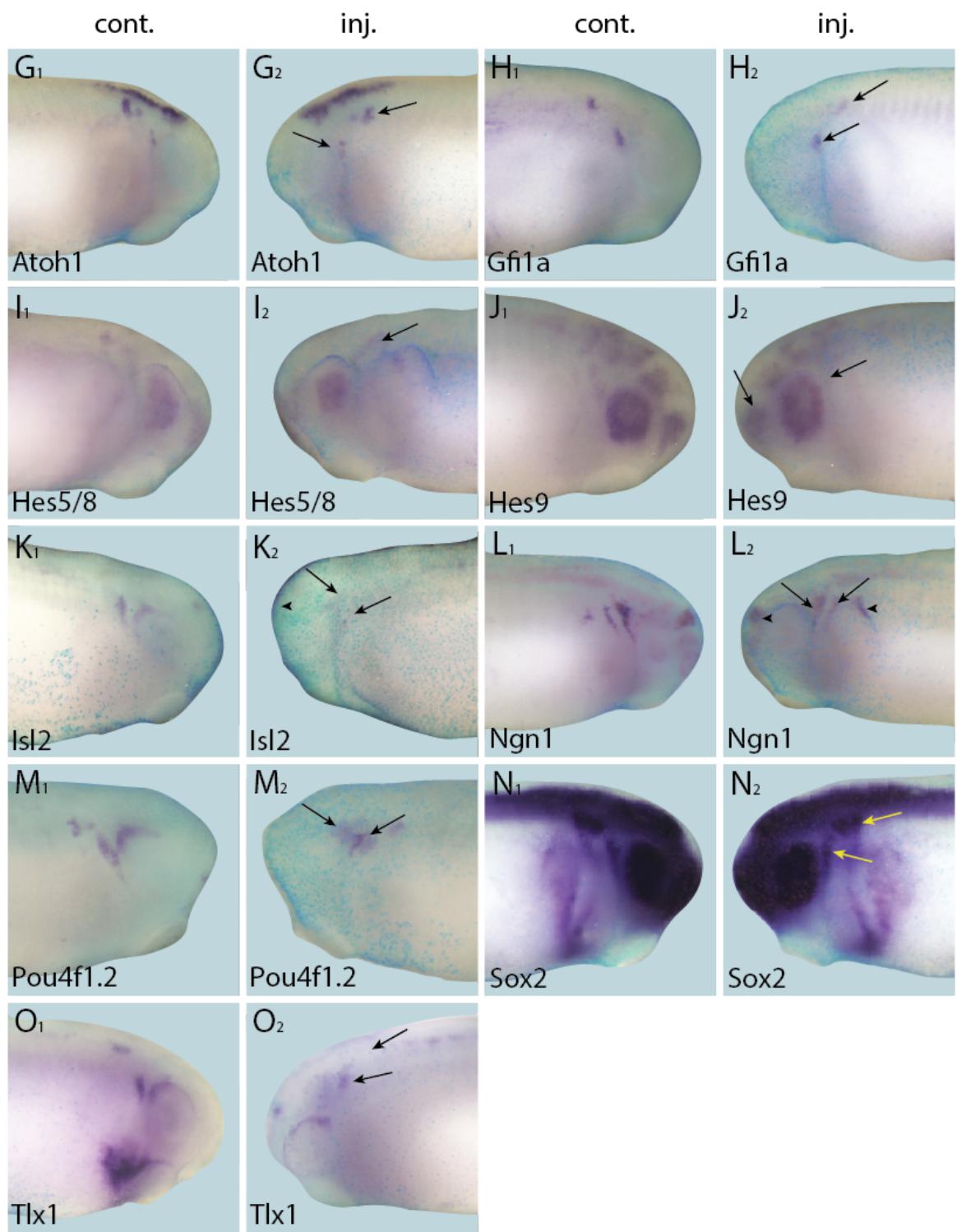
### 5.2.2 Six1-GR or Eya1-GR overexpression promotes both repression and activation of targets

To complement the loss of function studies, and to further examine whether *Six1* and *Eya1* are sufficient to induce the expression of targets, *Six1-GR* (500 pg) and *Eya1-GR* (500 pg) were overexpressed individually and, to ensure that overexpression did not affect early embryogenesis, activated by adding DEX at neural fold stage (stages 16 - 18), after PPE commitment (Ahrens and Schlosser, 2005). Somewhat surprisingly, although injection of *Six1-GR* or *Eya1-GR* resulted in up-regulation of direct targets in a minority of cases (fig. 5.7a and 5.8a; table 5.3), more often, it tended to repress their expression (fig. 5.7b and 5.8b; table 5.3). This was particularly true for genes expressed in proliferating progenitors of placodes at neural plate stage (*Hes5/8*, *Hes9*, *Sox2* and *Sox3*; table 5.3) as opposed to those expressed in differentiating cells (*Atoh1*, *Gf1a*, *Isl2*, *Ngn1*, *Pou4f1.2*, *Tlx1*; table 5.3). For example, overexpression of *Six1* led to down-regulation of *Hes5/8*, *Hes9* and *Sox2* in more than 80% of affected embryos, with neither *Sox2* nor *Hes9* showing any incidence of up-regulation. While overexpression of *Eya1-GR* tended to promote up-regulation more often than after *Six1-GR* overexpression, genes involved in proliferation were also more often down-regulated than

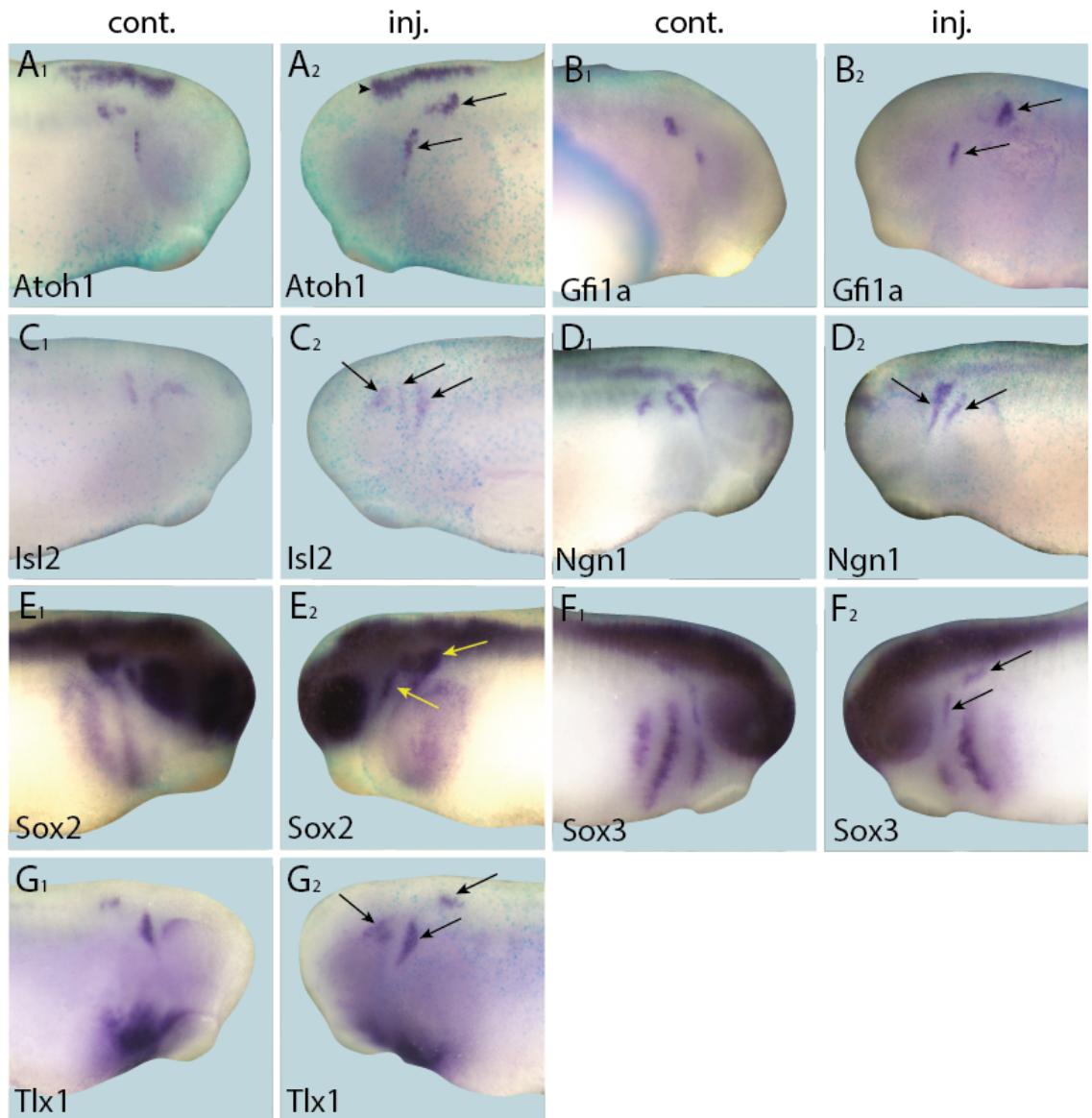
those involved in differentiation, with *Hes5/8*, *Hes9*, *Sox2* and *Sox3* all down-regulated in more than 70% of affected embryos, whereas only *Isl2* and *Tlx1* were consistently up-regulated (in 65% and 92% of embryos, respectively).



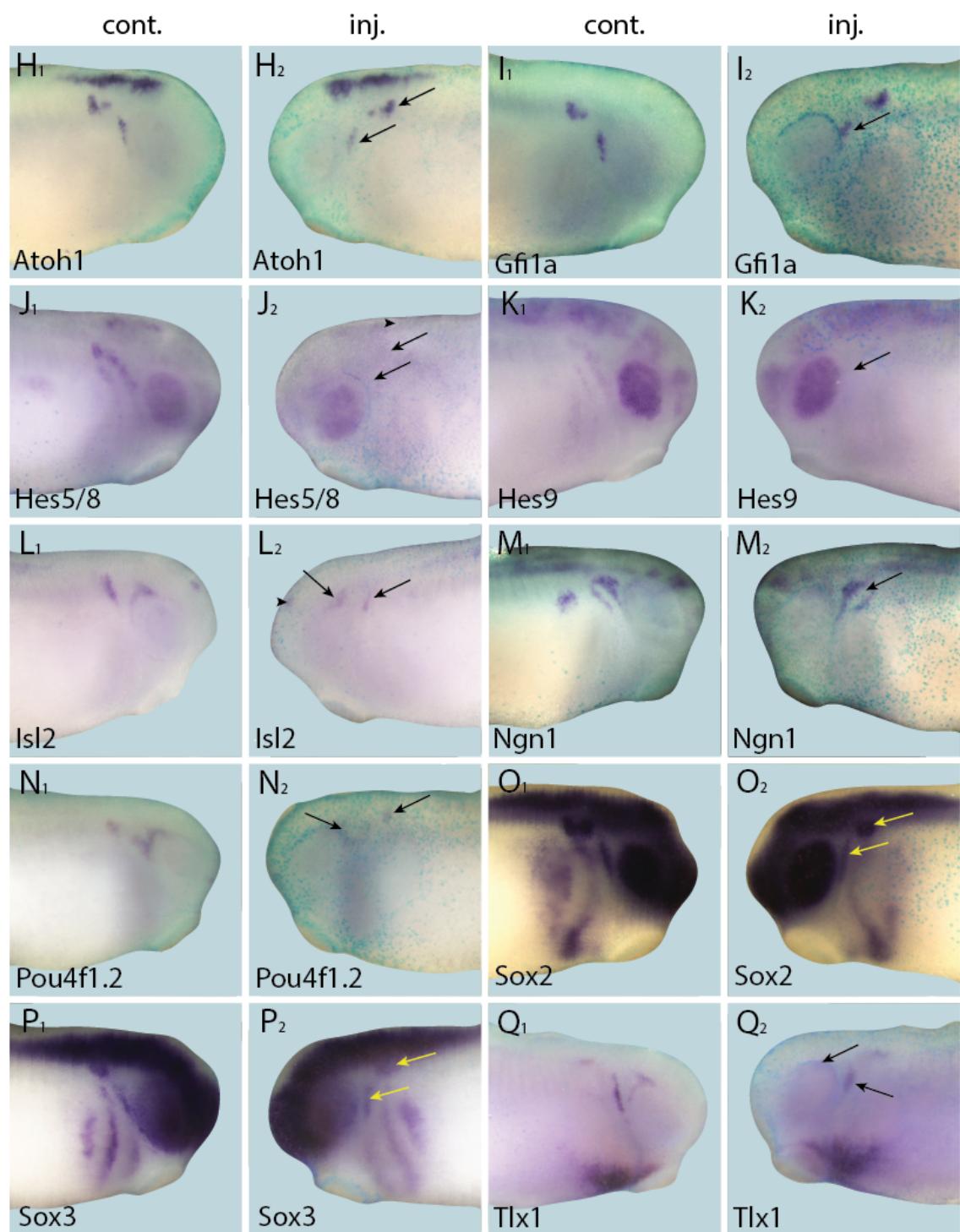
**Fig. 5.7a** Up-regulation of target gene expression domains after overexpression of *Six1-GR*. Tail bud stage embryos (**A** - **F**) after unilateral injection of *Six1-GR* and DEX induction at neural plate stage (16 - 18). In each case, *lacZ* was co-injected as a lineage tracer and panels **A<sub>1</sub>** - **F<sub>1</sub>** show the control (un-injected) side and **A<sub>2</sub>** - **F<sub>2</sub>** show the injected side. Arrows and arrowheads mark expansions in marker gene expression in placodal and non-placodal derivatives, respectively.



**Fig. 5.7b** Down-regulation of target gene expression domains after overexpression of *Six1-GR*. Tail bud stage embryos (**G** - **O**) after unilateral injection of *Six1-GR* and DEX induction at neural plate stage (16 - 18). In each case, *lacZ* was co-injected as a lineage tracer and panels **G<sub>1</sub>** - **O<sub>1</sub>** show the control (un-injected) side and **G<sub>2</sub>** - **O<sub>2</sub>** show the injected side. Arrows and arrowheads mark reductions in marker gene expression in placodal and non-placodal derivatives, respectively.



**Fig. 5.8a** Up-regulation of target gene expression domains after overexpression of *Eya1-GR*. Tail bud stage embryos (**A** - **G**) after unilateral injection of *Eya1-GR* and DEX induction at neural plate stage (16 - 18). In each case, *lacZ* was co-injected as a lineage tracer and panels **A<sub>1</sub>** - **G<sub>1</sub>** show the control (un-injected) side and **A<sub>2</sub>** - **G<sub>2</sub>** show the injected side. Arrows and arrowheads mark expansions in marker gene expression in placodal and non-placodal derivatives, respectively.



**Fig. 5.8b** Down-regulation of target gene expression domains after overexpression of Eya1. Tail bud stage embryos (**H - Q**) after unilateral injection of *Eya1*-GR and DEX induction at neural plate stage (16 - 18). In each case, *lacZ* was co-injected as a lineage tracer and panels **H<sub>1</sub>** - **Q<sub>1</sub>** show the control (un-injected) side and **H<sub>2</sub>** - **Q<sub>2</sub>** show the injected side. Arrows and arrowheads mark reductions in marker gene expression in placodal and non-placodal derivatives, respectively.

Such conflicting phenotypes are likely explained by a number of factors. One explanation is that Six1 and Eya1 serve a dual role in promoting proliferation and differentiation in a dose-dependent fashion. As has been previously shown (Schlosser et al., 2008), Six1/Eya1 promote proliferation at high doses but, below a concentration threshold, drive a differentiation programme. In this scenario, the concentration injected in this experiment may have been sufficiently low to slightly favour differentiation over proliferation, and thus reduce the observed frequency of up-regulation in genes associated with proliferation, compared to those involved in differentiation. Indeed, using the same mRNA concentrations, Schlosser *et al.* report repression of the proliferation marker *Sox2* in approximately 65% of embryos affected but expansion of the expression domain of the neuronal differentiation gene *NeuroD1* in approximately 70% of cases (Schlosser et al., 2008). While *NeuroD1* was not analysed here, several other neurogenic genes show similar frequencies of up-regulation (particularly after *Eya1*-GR overexpression: *Isl2* up-regulated in 65% and *Tlx1* in 92%, of embryos), although interestingly the neuronal gene *Pou4f1.2* was predominantly down-regulated after either *Six1*-GR or *Eya1*-GR overexpression. While these results seemingly contradict those reported in chapter 4 - which show that all direct targets analysed here were up-regulated after *Six1*-GR/*Eya1*-GR overexpression - DEX induction in this experiment was not preceded by CHX treatment, and thus translation of indirect targets was not blocked (unlike in both the RNA-Seq and qPCR experiments). The different phenotypes observed here are therefore suggestive of indirect effects that act to counteract direct effects observed in the RNA-Seq results.

Considering that, for a given cell, the balance between proliferation and differentiation are opposing forces, and that Six1 and Eya1 are known to promote both under different circumstances (Ahmed et al., 2012b; Ikeda et al., 2007; Z. Li et al., 2013; Schlosser et al., 2008), it is perhaps unsurprising that phenotypes consistent with both states are observed after *Six1*/*Eya1* overexpression. As several of the direct targets of Six1/Eya1 revealed in this study are implicated in Notch signalling (*Hes5/8*, *Hes9*, *Atoh1* and *Ngn1*), which directly prevents cell fate commitment via lateral inhibition, the interaction between Six1/Eya1 and the Notch system in the PPE presents itself as an obvious and immediate mechanism by which Six1/Eya1 may exert this dual role.

# Chapter 6. Six1 and Eya1 regulate neurogenesis by activating Hes5/8 in a Notch-independent way

## 6.1 Notch signalling in neurogenesis

During the development of the nervous system, a wide array of neurons differentiate from multi-potent progenitors. Generation of such cell types is mediated by direct cell-cell interactions through Notch signalling, whereby early-differentiating cells signal to neighbouring cells to prevent them from differentiating via lateral inhibition (reviewed in Chitnis, 1995; Muskavitch, 1994). During primary neurogenesis in *Xenopus*, nascent neuroblasts express the membrane-bound ligand Delta (Chitnis et al., 1995), which binds to and activates the Notch receptor on immediately neighbouring cells, inducing the release of the intracellular domain (ICD) of Notch. Notch-ICD, in association with Suppressor of Hairless (*Su(H)*), activates target genes such as *Hes1* and *Hes5* (Ohtsuka, 1999) which inhibit the expression of proneural bHLH genes including *Ngn1*, *NeuroD1* and *Atoh1*, thus preserving the cell as a proliferating progenitor. Notch signalling therefore is an important regulator of proliferative cell maintenance and the commitment to neural fate; indeed, dominant-negative interference with Notch function - as induced by overexpression of a DNA-binding mutant version of *Su(H)* (*Su(H)-DBM*) - results in the activation of a neurogenic programme at the expense of proliferating progenitor maintenance and, conversely, activation of Notch signalling by overexpression of the intrinsically active *Notch-ICD* inhibits neurogenesis (Chitnis, 1995; Muskavitch, 1994).

The bHLH genes *Hes1* and *Hes5* encode transcriptional repressors that are essential effectors of Notch signalling (Ohtsuka, 1999). Therefore, that *Xenopus Hes5/8* is a direct target of Six1/Eya1 in the PPE presents an interesting link between Six1/Eya1 expression and the Notch system. Several previous studies have already reported connections between Six1/Eya1 and the Notch pathway, namely that Six1 interacts with the Notch ligand Jag1 (Bosman et al., 2009) and that Six1 represses Hes1 and Hes5 and activates the proneural genes *Ngn1* and *NeuroD1* (Ikeda et al., 2007) as well as Notch itself (Ikeda et al., 2010). However, whether Six1 acts independent of, or in concert with Notch to activate *Hes5/8* is unexplored.

## 6.2 Six1/Eya1 are not required for *Hes5/8* activation via Notch signalling

### 6.2.1 *Hes5/8* is a direct effector of Notch signalling

In order to examine whether *Xenopus Hes5/8* is a primary target of Notch signalling, consistent with other *Hes5* genes (Ohtsuka, 1999), its expression was analysed after overexpression of *Notch-ICD*. As expected for an effector of Notch, activation of Notch signalling resulted in a reduction of neuronal differentiation in the placodes, as indicated by a reduction in both *Ngn1* and *N-tubulin* (96% and 87%, respectively; fig 6.1A and B), as well as inducing ectopic expression of *Hes5/8* throughout both the neural plate and epidermis (fig. 6.1C) in 100% of embryos analysed (table 6.1).

The opposite was true when Notch signalling was blocked by overexpression of *Su(H)-DBM*, which resulted in an expansion in the expression domains of *Ngn1* and *N-tubulin* (81% and 46%, respectively; fig 6.1D and E) and reduction of placodal *Hes5/8* expression (95%; fig 6.1F). Additionally, and typical of a *Hes5* gene, *Xenopus Hes5/8* acts to repress neuronal differentiation, reducing the expression of both *Ngn1* and *N-tubulin* when overexpressed (83% and 91%, respectively; fig 6.1G and H). Taken together, these results indicate that *Hes5/8* is a primary effector of Notch signalling, and acts to repress neuronal differentiation in a fashion consistent with other *Hes5* genes (Ohtsuka, 1999).

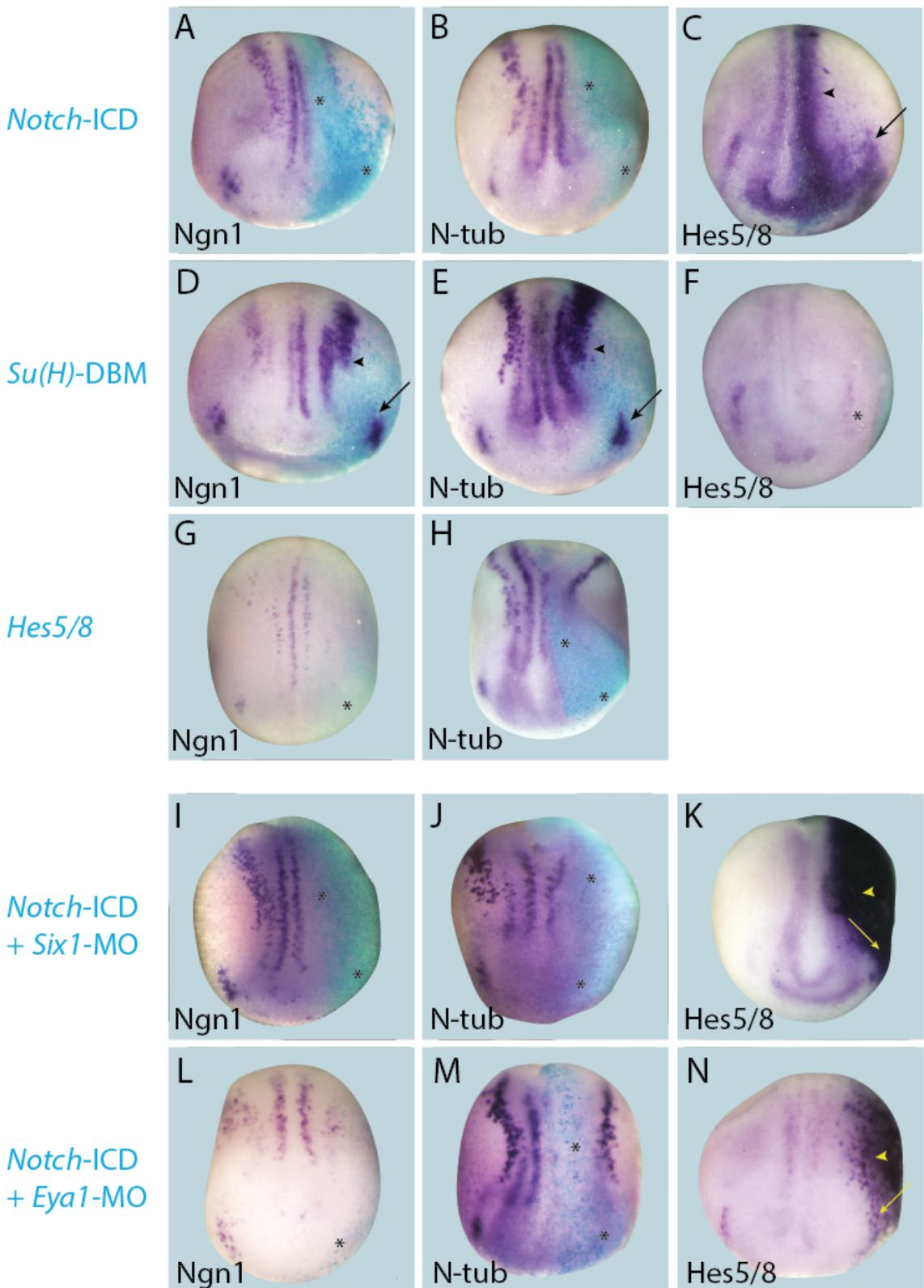
**Table 6.1** Changes in marker gene expression in the placodes after injection of various constructs

	<b>Injection</b>	<b>Notch-ICD</b>	<b>Notch-ICD + Six1</b>	<b>Notch-ICD + Eya1</b>	<b>Notch-ICD + Six1-MO</b>	<b>Notch-ICD + Eya1-MO</b>	<b>Su(h)-DBM</b>	<b>Su(h)-DBM + Six1</b>	<b>Su(h)-DBM + Eya1</b>	<b>Six1 + DAPT</b>	<b>Eya1</b>	<b>Hes5/8</b>
<b>Phenotype</b>	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	
	Reduced (33)	0 (41)	0 (36)	0 (38)	0 (17)	95 (19)	73 (22)	77 (31)	nd (31)	nd (nd)	nd (nd)	nd
<i>Hes5/8</i>	Increased 100 <sup>a</sup> (33)	100 <sup>a</sup> (41)	100 <sup>a</sup> (36)	74 <sup>a</sup> (38)	100 <sup>a</sup> (17)	0 (19)	50 <sup>a</sup> (22)	42 <sup>a</sup> (31)	nd (nd)	nd (nd)	nd (nd)	nd
<b>Phenotype</b>	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	
	Reduced (25)	96 (18)	83 (19)	79 (15)	87 (13)	70 (27)	0 (23)	21 (24)	81 (16)	50 (36)	83 (18)	
<i>Ngn1</i>	Increased 0 (25)	0 (18)	0 (19)	7 (15)	0 (13)	81 (27)	57 (23)	67 (24)	0 (16)	5 (36)	14 (18)	0
<b>Phenotype</b>	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	% (n)	
	Reduced (39)	87 (14)	100 (14)	96 (23)	100 (18)	93 (14)	0 (28)	48 (23)	26 (26)	81 (9)	45 (29)	91 (35)
<i>N-tubulin</i>	Increased 0 (39)	0 (14)	0 (23)	0 (23)	0 (18)	0 (14)	46 (28)	43 (23)	57 (23)	0 (26)	0 (9)	0 (29)

n: number of embryos analysed at both neural plate (stage 14-16) and tail bud (stage 21-26) stage

nd: not determined

<sup>a</sup> Denotes ectopic expression in the epidermis



**Fig. 6.1** *Hes5/8* is a direct effector of Notch. Overexpression of *Notch-ICD* reduces expression of *Ngn1* and *N-tubulin* (**A** and **B**) but broadens the expression domain of *Hes5/8* (**C**) in both the placodes and neural tube. The opposite is true for overexpression of *Su(H)-DBM*, which increases the expression of *Ngn1* and *N-tubulin* (**D** and **E**) and reduces *Hes5/8* expression (**F**). *Hes5/8* represses neuronal differentiation, as can be seen by reductions in *Ngn1* and *N-tubulin* expression after its overexpression (**G** and **H**). When *Notch-ICD* is overexpressed in conjunction with *Six1-MO*, *Ngn1* and *N-tubulin* expression are both reduced (**I** and **J**), whereas *Hes5/8* is expressed ectopically (**K**). Similar results are observed when *Notch-ICD* is co-expressed with *Eya1-MO*: *Ngn1* and *N-tubulin* are both down-regulated (**L** and **M**), and *Hes5/8* expression is increased and ectopic throughout the epidermis (**N**). Together, these results indicate that neither *Six1* nor *Eya1* are necessary for repression of neuronal differentiation via Notch signalling. Arrows and arrowheads mark an increase in marker gene expression in placodal and non-placodal domains, respectively, and asterisks mark a reduction.

### *6.2.2 Six1/Eya1 are not required for inhibition of neuronal differentiation via lateral inhibition*

Although Six1/Eya1 directly activate several genes implicated in Notch signalling (*Atoh1*, *Hes5/8* and *Ngn1*), and *Hes5/8* is a direct Notch effector (see section 6.2.1), whether Six1/Eya1 regulate such genes indirectly through the Notch system (as well as directly) is unclear. If this were the case, Six1/Eya1 would be required for Notch-mediated activation of *Hes5/8* and consequently block neuronal differentiation via lateral inhibition. To examine this, the expression of *Hes5/8* was analysed after either *Six1* or *Eya1* MO-mediated knockdown combined with overexpression of *Notch-ICD*. This co-injection led to a reduction in neuronal markers *Ngn1* and *N-tubulin* in a high proportion of injected embryos after both *Six1*-MO+*Notch-ICD* (87% and 100%, respectively; fig 6.1I and J; table 6.1) and *Eya1*-MO+*Notch-ICD* (70% and 93%, respectively; fig 6.1L and M) and, correspondingly, resulted in increased and ectopic expression of *Hes5/8* throughout the epidermis after both *Six1*-MO (74%; fig 6.1K) and *Eya1*-MO (100%; fig 6.1N) co-injections with *Notch-ICD*. These phenotypes are similar to those observed after injection of *Notch-ICD* alone (fig. 6.1A - C), suggesting that neither Six1 nor Eya1 are required either upstream of, or in conjunction with, Notch signalling to block neuronal differentiation.

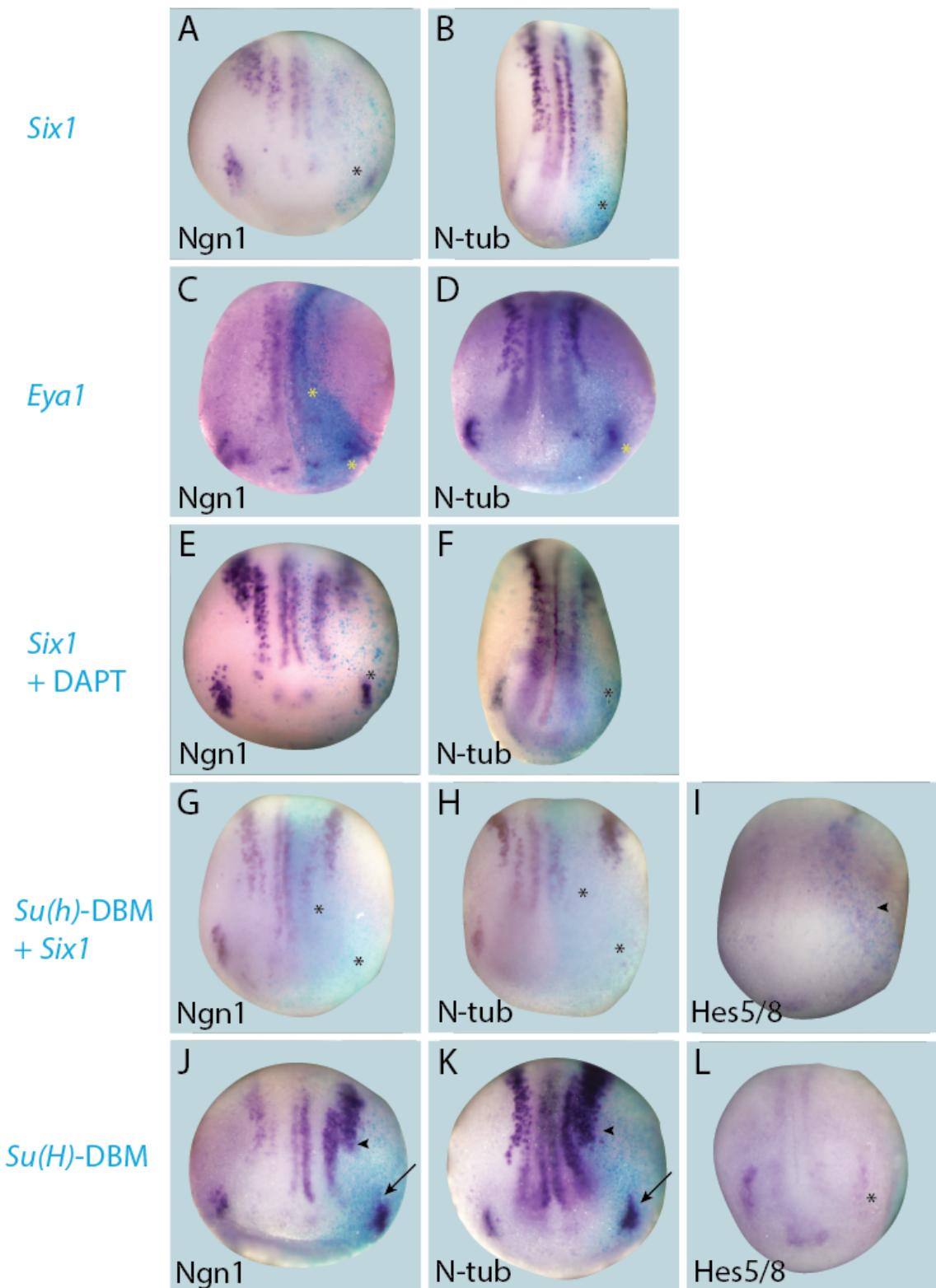
## **6.3 Six1/Eya1 repress neuronal differentiation in a Notch-independent fashion**

### *6.3.1 Six1 represses neuronal differentiation by activating Hes5/8 in a Notch-independent fashion*

Although Six1/Eya1 appear to be non-essential factors in Notch-mediated repression of neuronal differentiation, it remains possible that, conversely, their effects on neuronal differentiation are Notch-dependent. While this study has shown that Six1 and Eya1 directly activate *Ngn1* and promote neuronal differentiation (as shown by up-regulation of *N-tubulin*), previous studies indicate that overexpression of *Six1* and *Eya1* leads initially mostly to an expansion of proliferating progenitors and concomitant reduction of *Ngn1* and *N-tubulin* expression at neural plate stages (with ectopic neuronal differentiation being observed only later (Schlosser et al., 2008)). This was confirmed by injection of *Six1* and *Eya1* individually, and analysis of the expression of *Ngn1* and *N-tubulin* (table 6.1), with both *Six1* (*Ngn1* and *N-tubulin* expression reduced in 81% and 89% of embryos, respectively; fig 6.2A and B) and *Eya1* (50% and 45% of embryos, respectively; fig 6.2C and D) repressing neuronal differentiation.

To examine whether this propensity to initially repress neuronal markers is Notch-dependent, *Six1* was overexpressed before the Notch pathway was chemically antagonised prior to neural plate stage using DAPT (see section 2.2.4; Dovey et al., 2001; Geling et al.,

2002). While DAPT affects both hemispheres of a treated embryo, its ability to stimulate neuronal differentiation due to abrogation of Notch activity is apparent in the increased expression levels of both *Ngn1* and *N-tubulin* following DAPT treatment, compared to untreated embryos (compare expression in the un-injected side of embryos in fig 6.2A and B with fig 6.2E and F, respectively). In contrast, overexpression of *Six1* combined with DAPT treatment resulted in a reduction of expression in both *Ngn1* and *N-tubulin* in the majority of embryos (81% and 81%, respectively; fig 6.2E and F), which strongly suggests that Six1 represses neuronal differentiation in a Notch-independent way, presumably at least partly through direct activation of *Hes5/8*. While DAPT antagonises Notch signalling by preventing the proteolytic cleavage of Notch and the release of Notch-ICD, Su(H)-DBM interferes with DNA-binding of the Notch-ICD binding partner and transcriptional activator Su(H). Therefore, to further corroborate the ability of Six1 to repress neuronal differentiation independent of Notch signalling, this experiment was repeated by overexpressing *Su(H)-DBM* in combination with either *Six1* or *Eya1*. In both cases, co-injection with *Su(H)-DBM* resulted in a loss of differentiating neurons - as evidenced by a reduction in *Ngn1* (*Six1*: 35%; fig 6.2G and table 6.1; *Eya1*: 21%; table 6.1) and *N-tubulin* (*Six1*: 48%; fig 6.2H and table 6.1; *Eya1*: 26%; table 6.1). Again the effects were weaker after *Eya1* compared to *Six1* overexpression. The reverse was true for the expression of *Hes5/8*, which was broadened and ectopic after both *Six1+Su(H)-DBM* (50%; fig 6.2I and table 6.1) and *Eya1+Su(H)-DBM* (42%; table 6.1) overexpression. Taken together, these results independently demonstrate that Six1, and to a lesser extent Eya1, repress neuronal differentiation, and activate *Hes5/8*, in a Notch-independent way.



**Fig. 6.2** *Six1* represses neuronal differentiation in a Notch-independent way. Overexpression of either *Six1* (**A** and **B**) or *Eya1* (**C** and **D**) represses neurogenesis, indicated by a reduction in the expression of *Ngn1* (**A** and **C**) and *N-tubulin* (**B** and **D**). *Six1* can repress neuronal differentiation in a Notch-independent way, reducing the expression of both *Ngn1* (**E** and **G**) and *N-tubulin* (**F** and **H**) and an increase in *Hes5/8* expression (**I**) after treatment with DAPT (**E** and **F**) or co-injection with *Su(h)-DBM* (**G - I**; compare to injection of *Su(h)-DBM* alone [**J - L**]). The ability of DAPT to increase the number of differentiated neurons (due to abrogation of the Notch system) can be seen by comparing the expression of *Ngn1* and *N-tubulin* in the un-injected (left-facing) side of un-treated embryos (**A** and **B**) with the un-injected (left-facing) side of DAPT-treated embryos (**E** and **F**). Asterisks mark a reduction in marker gene expression.

# Chapter 7. Discussion

## 7.1 Introduction

This study has uncovered a gene regulatory network (GRN) underlying sense organ development in vertebrates. The expression of the transcription factor *Six1* and co-factor *Eya1* is initially restricted to the pan-placodal ectoderm (PPE), where together they act to drive lineage decisions towards a placodal fate, making them likely candidates for placode-initiating factors (for review see Schlosser, 2010; 2006). As a first step in exploring the placodal GRN activated by Six1 and Eya1, an RNA-Seq experiment was conducted to reveal their direct targets in the PPE. Genes revealed as direct targets of Six1 and Eya1 were highly enriched for GO terms associated with neurogenesis and placode development, and many appeared to be co-activated by Six1 and Eya1. Ten transcription factors were verified as direct targets using qPCR, and were subsequently demonstrated to require Six1/Eya1 function for their expression. Furthermore, this study has revealed a novel mechanism through which Six1/Eya1 may influence lineage decisions, via direct interaction with genes implicated in lateral inhibition and Notch signalling, such as *Ngn1*, *Atoh1* and *Hes5/8*.

## 7.2 A screen to reveal direct targets of Six1 and Eya1

Conducting an RNA-Seq screen yields a vast amount of raw data. As well as carefully controlling for potential sources of experimental variation, the use of appropriate analytical and statistical methods is paramount in order to ensure accurate interpretation. The screen presented in this study aimed to reveal direct transcriptional targets of Six1 and Eya1 in the PPE by using a hormone-inducible (DEX) overexpression assay in the presence of a protein synthesis inhibitor (CHX). *Six1*-GR and *Eya1*-GR were injected both individually (Six1<sub>i</sub>, Eya1<sub>i</sub>) and in combination (Six1+Eya1<sub>i</sub>), comprising three distinct treatment groups. To ensure that each treatment group had a directly comparable control, PPE was explanted from injected embryos, and split into two groups; one (control) group of explants was treated with CHX only, and the other (experimental) group treated with both CHX and DEX (fig. 3.1). In this manner, major sources of experimental variation including embryo age and genotype, as well as chemical (CHX) potency, were controlled for within individual biological replicates of each treatment groups. However, variation was apparent between replicates, and the low number of replicates used was insufficient to resolve such variation, resulting in few genes considered as significantly affected in analysis using Cuffdiff. As a means to mitigate this apparent inter-replicate variation, a bioinformatic pipeline was developed to prioritise the

selection of genes based on stringent criteria, for further characterisation in the absence of reliable statistics. In addition to this approach, individual treatment conditions were merged and re-analysed to increase the analytical power by using either four (*Six1<sub>m</sub>*, *Eya1<sub>m</sub>*) or six replicates (*Six1+Eya1<sub>m</sub>*). Although both individual and merged analyses yielded similar results (fig. 3.8; table 3.3) they introduce different interpretational caveats. These are discussed in-depth below, along with potential sources of the observed inter-replicate variation.

### *7.2.1 Activation of direct targets using CHX and DEX*

After injection of GR-fused *Six1* and *Eya1*, this study revealed direct transcriptional targets of *Six1* and *Eya1* in the PPE in a two-step process. First, to ensure that only direct targets were transcriptionally activated, protein synthesis was inhibited by CHX treatment. Using the same concentration as was used in this study, this method has been consistently shown to be highly efficient in blocking translation (Logan et al., 2005; Pozzoli et al., 2001). Second, to activate hormone-inducible fusion constructs by promoting their nuclear translocation, DEX was added (in combination with CHX) for 2.5 hours to allow the accumulation of transcripts of direct target genes. This system of DEX-induced conditional overexpression in the presence of CHX has been widely used to reliably activate direct, but not indirect, targets of transcription factors in *Xenopus* (Kolm and Sive, 1995; Logan et al., 2005; Pozzoli et al., 2001; Schlosser et al., 2008; Seo et al., 2007), zebrafish (Ramel and Lekven, 2004) and plants (Bargmann et al., 2013; Levesque et al., 2006). Although DEX treatment alone is known to induce expression of some genes - due to DEX-dependent nuclear translocation of endogenous glucocorticoid receptor (GR) (Reddy et al., 2009; Wang et al., 2004) - these effects were controlled for by comparing differential gene expression between un-injected CHX-treated against un-injected CHX+DEX-treated embryos. Genes found to be differentially affected by DEX treatment alone were excluded from consideration as direct targets of *Six1* or *Eya1*.

Despite this system's ability to activate direct target genes, it is unclear how the efficacy of CHX and DEX might change with dose. Although both were used at concentrations previously shown to be effective (Kolm and Sive, 1995; Schlosser et al., 2008; Seo et al., 2007), it seems likely that to some extent, variations in the concentration of both compounds (as well as their potency, with differences potentially resulting from storage conditions and duration) would affect their ability to block protein synthesis or activate GR-fused genes, respectively. If this were the case, such differences (along with the inherent variation in mRNA injection volume and concentration) would amplify variation in expression profiles between biological replicates. While some amount of variation between replicates observed in this screen (as evidenced by low number of significantly affected genes

assigned by Cuffdiff) may be ascribed to differences in chemical treatment, a greater source is potentially that generated during the PPE dissection experiments. That variation between replicates cannot have resulted purely from chemical treatment is evident in the MO RNA-Seq data set, where un-treated MO-injected embryos showed relatively weak correlations in expression profiles between biological replicates (*Six1*-MO average correlation: 0.81; *Eya1*-MO average correlation: 0.79). Despite this variability, three biological replicates were able to uncover statistically significant expression differences between a considerable number of genes. Taken together, this suggests that while the experimental treatment of embryos prior to RNA extraction (i.e. microinjection, chemical treatment and PPE dissection) does introduce inter-replicate variation, using additional biological replicates enables at least some of this to be resolved. For this reason, data obtained from the individual overexpression analysis (*Six1<sub>i</sub>*, *Eya1<sub>i</sub>* and *Six1+Eya1<sub>i</sub>*) was merged (*Six1<sub>m</sub>*, *Eya1<sub>m</sub>* and *Six1+Eya1<sub>m</sub>*) and re-analysed to provide more power in statistical testing. This second approach of merging replicates also facilitated the identification of genes synergistically regulated by *Six1* and *Eya1*. For example, in merging replicates from *Six1<sub>i</sub>* and *Six1+Eya1<sub>i</sub>* datasets (*Six1<sub>m</sub>*), genes co-regulated by *Six1* and *Eya1* are revealed. In this case, however, while such analysis provides insight into genes co-regulated by *Six1* and *Eya1*, only those whose expression is limited by *Six1* expression levels (but not *Eya1*) will be revealed as significantly different (see section 7.3.2 for further discussion).

### 7.2.2 Technical issues with analysis of RNA-Seq data

In order to capture the expression differences of genes expressed in the developing placodes, RNA was extracted from dissected PPE explants in biological duplicates (for the overexpression screen) and triplicates (for the MO experiment), and sequenced using RNA-Seq. For all experiments (except for one replicate in the *Eya1*-MO experiment - see section 2.2.5), high-quality RNA (RIN > 7) was sent for sequencing, and yielded high-quality paired-end RNA-Seq reads, of which a large proportion (on average 80%) were mapped to a draft version of the *Xenopus laevis* genome (Xl7; for a summary see fig. 3.1). However, perhaps owing to their length (on average > 85 bp), a very large proportion of paired-end reads overlapped in sequencing. This issue likely stems from using a cDNA fragment library size too small to prevent long paired-end reads from overlapping, and is a relatively common observation in RNA-Seq experiments using long sequencing reads (Magoč and Salzberg, 2011). While this is not a problem *per se* in mapping to an annotated reference genome (where each read mapping to an annotated gene contributes to the total FPKM value for that gene), this presents difficulties when mapping reads to an un-annotated genome. The most relevant of these is that when paired-end reads overlap in sequencing they at least partially lose their ability to link exons together, and therefore assemble full-length transcripts. In this scenario,

for a given gene (assuming that the full-length transcript is contiguously represented on a single genomic scaffold), several distinct transcript models are reported, and are annotated and assigned statistical significance independently. As discussed in more detail below (see section 7.2.3 or 2.5.8 for a technical description), the annotation pipeline developed for this study attempted to mitigate this by pooling expression values for transcript models returning identical annotations.

In addition to the limited ability of overlapping reads to fully resolve full-length transcripts when mapped to an un-annotated genome, the incompleteness and redundancy of the *Xenopus* genome further confounds analysis. Examination of the PPE transcriptome in this study revealed that 72% of transcript models assembled by Cufflinks were annotated identically to at least one other, 41% to at least four others (fig. 3.3) and several to over 100 other transcript models. While much of this could be indicative of either redundancy (where a gene is represented in the genome several times) or incompleteness (where a gene is fragmented over several scaffolds, thus returning several transcript models), or both, the relative contribution of these different scenarios is currently unclear. To ease further discussion, two discrete sources of redundancy will be distinguished: Artificial redundancy (as an artefact of a draft genome) and genuine redundancy (where multiple paralogs of a gene exist in the genome, arising from gene duplication). While incompleteness can be accounted for by pooling expression values for duplicated annotations, the genuine genomic redundancy as a result of tetraploidy in *Xenopus laevis* is more troublesome.

The *X. laevis* genome underwent tetraploidisation approximately 40 million years ago, when two ancestral species hybridised to give rise to a descendent species carrying the complete nuclear genome of both ancestors (Evans et al., 2004; Uno et al., 2013). However, cell division occurs in *X. laevis* in a diploid fashion (whereby each chromosome aligns with only one partner), and it is hence referred to as a pseudotetraploid (Evans and T. Kwon, 2015). While many of these duplicated genes (pseudoalleles or alloalleles) are assumed to have been lost through mutation owing to relaxed selection on the redundant copy (recently estimated at between 50-75% (Hellsten et al., 2007)), pseudotetraploidy is maintained in many genes. A recent study surveyed 2,218 genes duplicated in *X. laevis* but existing as singletons in the closely related (diploid) *X. tropicalis*, and demonstrated accelerated evolution of *X. laevis* paralogs, with a significant fraction (estimated at about one third to one half) showing quite divergent expression patterns and dynamics in development (Hellsten et al., 2007). Interestingly, these paralogs were found to have diverged symmetrically (approximately 1 - 2 amino acids per peptide), with both copies equally distantly related to their *X. tropicalis* orthologous counterpart, highly suggestive of evolution under purifying selection (Hellsten et

al., 2007; M. K. Hughes and A. L. Hughes, 1993). Thus, it is clear that many of these duplicated genes have been maintained in *X. laevis*, and their sequence similarity poses a further challenge when mapping RNA-Seq reads.

When using default mapping parameters in Bowtie2, as were used in this study, reads that map to more than one gene are randomly assigned to one of the two genes. Thus, given the close sequence similarity of duplicated genes in *X. laevis*, it is reasonable to assume that reads mapping to a gene present twice in the genome will be equally distributed between the two. Although ideally such pseudoalleles would be distinguished in analysis and differential expression reported for each paralog, this study did not attempt to make such a distinction. Instead it attempted to sum expression values across duplicated genes, and annotate and report differential expression as if they were singletons. Thus, in situations where reads map identically to multiple genomic loci which, as discussed above, may arise owing to both artificial redundancy as a consequence of mapping to a draft assembly, as well as genuine genomic redundancy as a result of tetraploidy in *X. laevis*, the bioinformatic pipeline developed in this study pools expression values for transcript models returning identical annotations, as discussed below.

### 7.2.3 Technical issues with developed programs

Performing an RNA-Seq experiment in an organism lacking a complete, annotated genome, presents several problems. Perhaps the most important of these from an interpretational point of view are the issues of gene annotation, and resolution of artificial genomic redundancy (as discussed above). In-house scripts were developed for this study in an attempt to mediate these issues, and the mechanics of several of these are discussed in detail below.

Accurately annotating the transcript models returned by Cufflinks is a crucial step in any RNA-Seq experiment; a poor initial annotation may result in significant difficulties in data interpretation, and lead to a high false positive/negative discovery rate. The script ‘annotator.pl’ was developed to annotate the longest splice-variant for each transcript model against a *Xenopus* mRNA database. Annotation strategies are only as good as the database they derive annotations from, and for this reason a database containing mRNA sequences from both *X. laevis* and *X. tropicalis* was used (in total 81,902 sequences). To enable efficient annotation of each transcript, a high e-value threshold was specified in blastn searches, and the sequence with the highest score was used to annotate each transcript model. While using such a large (combined) database maximises the chance of a given transcript model being annotated, the reliability of the annotations is varied, with some being derived from empirical data (and thus of high quality) and others from EST libraries using generic clone ids. In the

latter case, while such a generic annotation may share 100% sequence identity to a transcript model query (and thus be returned during in-house annotation), it is wholly uninformative for this study, and serves only to increase the false negative rate. While this is confirmed by a manual inspection of the genes reported in table 3.2, where several genes of high interest for this study (such as *Six1* and *CyclinD1*) were originally annotated in an uninformative fashion (as ‘uncharacterized protein MGC130961’ and ‘cDNA clone MGC: 83328’, respectively), genes found to be highly differentially expressed between treatment groups were generally examined manually, thereby reducing the number of false negatives produced by such uninformative annotation.

Another problem in mapping to a draft genome, and particularly that of a pseudotetraploid, is that of redundancy (both artificial and genuine). As discussed above, reads derived from a single transcript may be mapped to multiple positions in the genome, either as a consequence of genuine redundancy (multiple pseudoalleles) and/or artificial redundancy in the genomic assembly. In such instances, the resulting FC values (and accompanying statistics) generated by Cuffdiff will be unrepresentative of the transcript from which the reads were derived, and thus confound interpretation. In an attempt to address this issue, ‘co-reg.pl’ was developed to sum raw expression values for transcript models sharing the exact same annotation, and output a single annotation with a new FC value calculated from these pooled expression values. To ease the following discussion, transcript models sharing the same annotation (and thus treated as representing the same gene in this study) will be referred to as being ‘twins’, and the final summed annotation reported by ‘co-reg.pl’ will henceforth be termed the ‘flattened’ annotation. This program therefore treats all potential sources of redundancy (i.e. incompleteness due to fragmentation of scaffolds, genuine redundancy due to pseudotetraploidy and artificial redundancy as an artefact of genome assembly) as equal, and while this may enable more accurate interpretation of FC values, it sacrifices the ability to differentiate between pseudoalleles (unless they happen to be annotated separately in the *X. laevis* mRNA database).

A separate issue becomes relevant when using this approach to analyse data resulting from merged replicates. Whereas in the individual analyses FC values were used as a principal gauge of differential expression, the merged data combined replicates to add power to the statistical analysis performed by Cuffdiff, and thus used both q-values and FC values to determine significant differential expression. However, as each transcript model is associated with a q-value, when pooling expression data from twins a single q-value must be selected for the resulting flattened annotation.

In order to mitigate the impact of selecting an individual q-value to represent each flattened annotation (itself potentially derived from many transcript models) only q-values associated with transcript models expressed at FPKM > 1 in the CHX+DEX treatment group were considered, and for each flattened annotation, the lowest (most highly significant) q-value was selected.

Finally, to remove genes activated by DEX-treatment alone, FC values for CHX+DEX-treated, but un-injected (control), embryos was calculated as above. For experimental conditions, only transcripts showing a FC value at least twice that of this control were considered.

### 7.3 Data quality

#### 7.3.1 Appraisal of target genes revealed by RNA-Seq screen

As an initial check of data set quality, both known and predicted direct targets of Six1 were searched for in the genes differentially affected by *Six1/Eya1* overexpression. Of the ten predicted and known targets found expressed in the PPE (i.e. those present in the transcriptome), nine were differentially expressed after either *Six1*, or *Six1+Eya1* overexpression, confirming the high quality of this data (see section 3.4.2). To explore the processes Six1 and Eya1 may drive in the PPE, the targets revealed in this screen were examined to assess the extent to which they are implicated in known functions of Six1 and Eya1 in both the maintenance of progenitors and their differentiation into neurons (Bricaud and Collazo, 2011; Brugmann et al., 2004; Schlosser et al., 2008; P. X. Xu et al., 2002; Zou et al., 2006). Figure 3.8 shows the results of a gene set enrichment analysis, where human orthologs of differentially expressed genes were used as input to DAVID, and functional terms enriched in relation to a background set (comprising the PPE transcriptome) are clustered based on their similarity. Of the functional terms enriched in genes up-regulated after *Six1/Eya1* overexpression, many relate to neurogenesis (axon guidance, regulation of neurogenesis), or are associated with sensory development (inner-ear development, sensory perception, auditory receptor cell differentiation), consistent with known roles of Six1 and Eya1 in such processes. While Six1 and Eya1 have important functions outside of PPE development, such as skeletal muscle differentiation and the development of kidneys (Heanue et al., 1999; P. X. Xu et al., 2002), the absence of such terms from the gene set enrichment analysis provides further confidence that these targets are PPE-specific. Interestingly, an as-yet unexplored link between Eya1 and Hox genes is suggested by the high enrichment of Homeobox genes, particularly those belonging to the Antennapedia-type class, in the Eya1-

specific up-regulated dataset (fig. 3.8F; table 3.1), providing an exciting opportunity for follow-up research.

### 7.3.2 *Six1* and *Eya1* co-regulate genes in the PPE

While transcription factors regulate gene expression by directly binding target DNA, the influence they have on transcription is often mediated by the co-factors they synergise with. For example, the *Drosophila* transcription factor and homologue of vertebrate *Six* genes, *sine oculis* (*so*), is known to synergise with both *eyes absent* (*eya*; homologue of vertebrate *Eya* genes), and *dachshund* (*dac*; homologue of vertebrate *Dach* genes) (R. Chen et al., 1997). In vertebrates, although both Eya and Dach act as co-factors for Six1, their binding promotes different regulatory outcomes; while a Six-Eya interaction results in activation of downstream targets, a Six-Dach complex strongly represses transcription (X. Li et al., 2003). The observation that Six1 acts as a transcriptional activator when synergised with Eya1 is widely supported (Ahmed et al., 2012a; Brugmann et al., 2004; Christophorou et al., 2009; Ruf et al., 2004; P. X. Xu et al., 2002), along with Six1 acting to repress transcription when in complex with non-Eya binding partners such as Dach or Groucho (Brugmann et al., 2004).

In order to examine the degree to which the RNA-Seq experiment supported such co-regulation, datasets were examined for genes differentially affected in multiple treatment groups. Co-regulated genes were considered to be those differentially expressed in two or more treatment groups in the individual analysis and, consistent with Six1-Eya1 synergism having a positive effect on transcription, many more co-regulated genes were up-regulated (228; fig. 3.6A) as opposed to down-regulated (68; fig. 3.6B). Indeed, further examination revealed a highly significant enrichment ( $p < 0.0001$ ; Fisher's exact test) of targets between all experimental treatment groups, supporting a synergistic regulation of direct targets. However, many genes were also found to be exclusively regulated by either Six1 or Eya1, suggesting that they also regulate target genes independently of each other. Interestingly, when considering genes exclusively regulated, while *Eya1* overexpression tended to promote transcription more often than repressing it, the opposite was true for *Six1* overexpression (fig. 3.5), suggesting an additional role for Six1 as a transcriptional repressor. As Six1 is known to function as a repressor in a context and co-factor dependent fashion (Brugmann et al., 2004; X. Li et al., 2003), this finding suggests that Six1 is able to interact with non-Eya1 binding partners in this screen.

To scrutinise these findings, individual datasets ( $\text{Six1}_i$ ,  $\text{Eya1}_i$  and  $\text{Six1}+\text{Eya1}_i$ ) were merged together and re-analysed in order to find genes co-regulated by Six1 and Eya1 either using four ( $\text{Six1}_m$ ,  $\text{Eya1}_m$ ), or all six ( $\text{Six1}+\text{Eya1}_m$ ) replicates. In the individual analyses, genes

were considered as differentially expressed if they were at least two-fold up-regulated in at least two treatment groups (see section 2.5.8), however genes requiring high levels of both Six1 and Eya1 for activation (where both Six1 and Eya1 are limiting factors in a direct target's activation) would only be found in the Six1+Eya1<sub>i</sub> group. Merging data therefore provides an opportunity to examine the apparent enrichment of differentially expressed genes between treatment groups, and thus the extent to which Six1 and Eya1 co-regulate their direct targets in the PPE. While merging all replicates together (Six1+Eya1<sub>m</sub>) reveals co-regulated genes (where both Six1 and Eya1 are limiting factors in direct targets' expression), Six1<sub>m</sub> and Eya1<sub>m</sub> reveal targets whose expression levels are limited by Six1 or Eya1, respectively. For example in the former case (Six1<sub>m</sub>), genes identified as significantly differentially expressed must respond similarly to both individual overexpression of *Six1*, and co-expression of both *Six1* and *Eya1*, suggesting that they are either exclusively regulated by Six1, or that the addition of Eya1 does not quantitatively affect their expression. This re-analysis found more genes significantly affected ( $q < 0.05$ ) than the individual analysis (appendix F), and as for the individual analysis, all experimental data sets were highly significantly enriched for genes activated by the other conditions. Similar to the individual analysis, high numbers of genes were up-regulated in these merged data sets, whereas very few were down-regulated (figs. 3.5 and 3.7), consistent with Six1 and Eya1 synergism promoting activation of target genes.

### 7.3.3 Merged analysis strongly supports individual analysis

In addition to allowing further identification of co-regulated genes, merging replicates together in re-analysis increased statistical power, and enabled the comparison of differentially expressed genes identified by the individual analysis ( $FC \geq 1$ ) to statistically differentially expressed genes ( $q < 0.05$ ;  $FC \geq 1$ ) found by the merged analysis. While far fewer genes were significantly differentially expressed (and substantially fewer genes were negatively regulated) in the merged analysis compared to the individual analysis (figs. 3.5 and 3.7), many of the most highly differentially expressed genes were common to both, suggesting a broad agreement between results from both analytical strategies (compare appendices E and F, and see table 3.3 showing support for both analyses). That fewer genes were statistically significantly affected in this re-analysis is likely explained by a number of factors. Firstly, as the merged analysis makes use of Cuffdiff's statistical testing (on account of having more replicates) the threshold for differential expression is more stringent, and the observed discrepancy between genes affected could simply reflect this. If this were the case, while the results obtained from both analyses strongly support each other (table 3.3 and fig. 3.8), the number of false positives reported by the individual analysis may be higher than for the merged analysis.

Additionally, while the merged analysis provides statistical support on account of combining replicates, there are some instances where it would be unable to identify a gene as differentially affected. For example, if a target was repressed by Six1 individually, but activated by Six1+Eya1, while it might be identified as being up-regulated in Six1+Eya1<sub>m</sub> and/or Eya1<sub>m</sub> datasets, it would not be identified as being down-regulated in Six1<sub>m</sub>. While this may contribute to the paucity of genes found to be significantly down-regulated in the merged datasets, that in all cases the overwhelming proportion of genes reported is up-regulated strongly corroborates previous reports of Six1 and Eya1 acting synergistically to positively regulate transcription, as discussed above. Taken together, the results of this re-analysis - using merged replicates in order to increase statistical power - broadly bolsters the analysis of individual treatment groups, and provides a further means to identify false positives.

#### 7.3.4 Candidates selected for further characterisation

In order to limit functional studies to genes potentially involved in a placodal GRN, only up-regulated transcription factors (or co-factors) were selected for characterisation. From the 33 genes that satisfied these criteria in at least two of the treatment groups, three (*Fbxo41*, *Egr3* and *Gbx2*) were omitted from analysis due to technical difficulties (see section 4.2), as well as a further five (*Ets2a* (Salanga et al., 2010); *Mafa* (Coolen et al., 2005); *MyoD1* (Hopwood et al., 1989); *Sox1* (Nitta et al., 2006); *Sox17* (Hudson et al., 1997)) showing no expression in placodes in published accounts. Several other genes with extensively detailed expression in placodes were not repeated in this study (*Ngn1* (Niebler et al., 2009); *Six1* (Pandur and Moody, 2000); *Six2* (Ghanbari et al., 2001); *Sox2* (Mizuseki et al., 1998); *Sox3* (Penzel et al., 1997)). Thus, 20 were fully characterised across several informative developmental stages via *in-situ*-hybridisation, and broadly clustered in four groups: (1) Genes not expressed in placodes or placode-derived structures (figs. 4.3 - 4.8); (2) genes expressed broadly in cranial ectoderm including PPE (figs. 4.9 - 4.12); (3) genes expressed in proliferating placodal progenitors of PPE (figs. 4.13 - 4.17); (4) genes expressed in differentiating placodal cells (figs. 4.18 - 4.22). From this characterisation, a number of genes were found not to be expressed in placodes (*Emx1.2*, *Lhx5*, *Pou3f2b*, *Tbx15*, *Sim1* and *Tbx6*; figs 4.3 - 4.8), however the majority of these showed expression in either the adjacent neural plate and neural folds or throughout dorsal tissues at neural plate stages. Thus, it is likely that these genes were included in the screen as a result of contamination from surrounding or underlying tissues. However, of the 31 genes selected for further characterisation (discounting *Fbxo41*, *Egr3* but not *Gbx2* according to its known role in placode development (Steventon et al., 2012)), 65% were expressed in placodes (as found either in this study or in published accounts), and 80% of these were supported by both the individual and merged analysis (table 3.3). Furthermore,

of the ten genes selected for further functional analysis, all were confirmed as direct targets of either Six1 or Eya1 via qPCR (*Isl2* was verified as a direct target of Six1 but not Eya1 both in the RNA-Seq data and qPCR). Taken together, these results suggest a low rate of false discovery in the bioinformatic analysis.

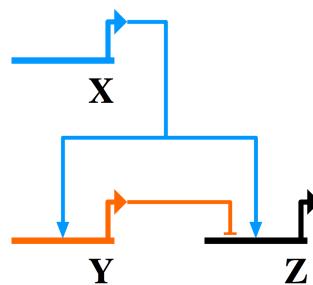
### 7.3.5 Interpretation and analysis of morpholino data

In order to assess whether Six1 and Eya1 were required for the expression of the ten genes verified as their direct targets, a second RNA-Seq experiment was conducted after MO-mediated knockdown of either *Six1* or *Eya1*. None of these genes was found to be significantly differentially expressed in either *Six1*-MO or *Eya1*-MO dataset (table 5.1). In addition, the neuronal marker *N-tubulin* - previously shown to be down-regulated after knockdown of either *Six1* or *Eya1* (Schlosser et al., 2008) - was also absent from both data sets. However, several other neuronal markers known to be downstream of Six1/Eya1 (*NeuroD1*, *Tuj1*; Ahmed et al., 2012b) were significantly down-regulated in either one or both data sets, and genes affected by *Six1*-MO were significantly enriched ( $p < 0.0001$ ; Fisher's exact test) for those affected by *Eya1*-MO. As discussed in section 5.1.2, a combination of factors likely explains the absence of direct targets confirmed in this study from MO data sets, but perhaps the most salient for this discussion are (1) the complex regulatory interactions existing between downstream targets; (2) the difficulty in interpreting knockdown phenotypes; (3) the complexity introduced by *p53* induction as a result of indirect effects of MOs. Each of these will be discussed below.

#### 7.3.5.1 Indirect effects of MOs

That gene knockdown affects both direct and indirect targets immediately presents conceptual interpretational difficulties. Figure 7.1 shows a simple scenario that would lead to such obfuscation, whereby the protein product of gene *X* directly activates two genes *Y* and *Z*, but indirectly represses *Z*. Here, knockdown of gene *X* would reduce the expression of genes *Y* and *Z* (by removing the activating signal from gene *X*) while simultaneously promoting the expression of gene *Z* (by alleviating repression from *Y*). In this scenario, the expression dynamics of gene *Z* after knockdown of *X* would entirely depend on external influences such as the relative expression levels of each gene and the existence of direct and indirect interactions with other factors, and could therefore vary in both cellular space and developmental time. As has previously been shown, while the neuronal differentiation gene *Ngn1* acts downstream of Six1/Eya1 (Ahmed et al., 2012b), knockdown of *Six1* results in a reduction of *Ngn1* expression in placodes in 45% of embryos injected and expansion in 36%, with similar results reported in the same study for both *Sox2* and *Sox3* (Schlosser et al., 2008).

Context-dependent effects of MO action may also explain the observed expression dynamics of *Atoh1* expression, which is reduced in the trigeminal placodes, but simultaneously increased in the otic vesicle after *Six1*-MO mediated knockdown (fig. 5.3A). While to some extent this phenomenon can be observed by comparing individual embryos via *in-situ*-hybridisation, detecting such effects in an RNA-Seq experiment where data from many embryos are pooled would be significantly more difficult. Such variability in the response of gene Z (and thus its expression level) would not be apparent, and would simply be reported as an average of both effects.



**Fig. 7.1** Direct and indirect effects of MO-mediated knockdown. The protein product of gene X directly activates genes Y and Z, but indirectly represses gene Z (through activating gene Y). Even in a simple system such as this, the effect of MO-mediated knockdown of gene X may produce seemingly paradoxical results, for example by indirectly promoting the expression of gene Z (by alleviating the repression from Y).

### 7.3.5.2 Difficulty in interpreting MO phenotype

A further complication in RNA-Seq data analysis after knockdown experiments is the ambiguity of expression domain changes after gene knockdown: In some instances an expression domain may be reduced in area but increased in intensity, whereas expanded but weakened in others. The former scenario is apparent in the expression of *Tlx1* after *Six1* knockdown (fig. 5.3G), where the trigeminal domain is reduced in size but appears more intense, and the latter in the expression of *Hes9* after *Six1* knockdown (fig. 5.3D), where the expression in the lateral line placodes and otic vesicle appears reduced in intensity but more dispersed. While in both cases the overall effect of *Six1* knockdown on the expression of targets such as *Tlx1* and *Hes9* is clear when analysing *in-situ* data, such effects would not be evident in RNA-Seq data.

### 7.3.5.3 Indirect activation of p53

The use of MOs in gene knockdown is widespread in multiple model systems, including *Xenopus*, partly owing to the ease and speed at which a knockdown phenotype can be observed. However, MOs can induce p53-dependent apoptosis (Robu et al., 2007) and off-target changes in gene expression that are cell-type-specific (Gerety and Wilkinson, 2011), both of which can confound analysis. The tumour suppressor p53 is a transcriptional

regulator with crucial roles in genomic stability and apoptosis of damaged cells, and its activation can lead to cell cycle arrest and apoptosis-induced cell death (Vousden and Prives, 2009). Therefore, that p53 is found to be significantly up-regulated after both *Six1*-MO and *Eya1*-MO mediated knockdown (*Six1*-MO FC: 1; q = 0.006; *Eya1*-MO FC: 1; q = 0.03) presents substantial difficulties for interpretation. While p53-induced off-target effects of MO use are certainly problematic, several approaches have been suggested to mitigate such effects: (1) The use of *p53*-specific MOs to reduce its activity (Robu et al., 2007); (2) comparison of morphant phenotype to mutant phenotype to ensure specificity (Kok et al., 2015; Schulte-Merker and Stainier, 2014); (3) the careful control of MO concentration to ensure observed effects are specific (Eisen and Smith, 2008).

While co-injecting target specific MOs with a *p53*-specific MO has been shown to attenuate phenotypes associated with p53 induction in zebrafish (Robu et al., 2007), the mechanism through which p53 is activated is unknown, and complex (Vousden and Prives, 2009). Also, while this method attempts to remedy p53-mediated off-target effects, it does not address those unrelated to p53 induction. The recent advent of genome editing tools such as zinc finger nucleases (ZFNs) transcription activator-like effector nucleases (TALENs), and more recently clustered regularly interspaced short palindromic repeats (CRISPRs), has allowed the rapid generation of programmable site-specific nucleases and targeted gene disruption in a wide range of systems. Early adopters of such tools observed that mutant phenotypes rarely correlate with those found in (MO-injected) morphants. Indeed, one such study has recently reported that in a morphant-mutant comparison, 80% of morphant phenotypes examined were not shared by mutants (Kok et al., 2015). While it is unclear how universal such a finding is, it serves to highlight an astonishing disparity between the two approaches and, if both mutant and morphants are available for a given target, it should be considered recommended practice now to compare the morphant to the mutant phenotype to ensure a correlation (Schulte-Merker and Stainier, 2014). However, and as a further complication, it is also possible that the poor correlation between mutants and morphants could be a result of genetic compensation in the former but not the latter, with the authors concluding that MOs injected at a sufficiently low dose, so as not to induce p53, should be favoured over other systems (Rossi et al., 2015). Such a method for controlling for p53-induced off-target effects was favoured by this study. Figure 5.2 shows the effects of a high dose MO injection (20 ng), as was used in the RNA-Seq experiment, which shows p53 activation corresponding to the injected side, along with reduction of the neuronal marker *N-tubulin*. In an attempt to mediate this, MO titration was performed (table 5.2), and a MO dose 1/10<sup>th</sup> (2 ng) of that initially injected was shown to be effective in reducing *N-tubulin*, but not activating p53 (fig. 5.3); such a low dose was hence used in all future MO experiments.

To further ensure the correct interpretation of MO use, several other precautionary measures were taken. Firstly, a mismatch MO of *Eya1* (*Eya1*-mmMO) was used to control for MO toxicity (Eisen and Smith, 2008), and in all cases, injection of *Eya1*-mmMO resulted in statistically less frequent effects on all targets analysed, compared to injection of either *Six1*-MO or *Eya1*-MO (table 5.3; appendix J). Secondly, to ensure that the knockdown phenotype produced was specific to the gene targeted, injection of a dominant-negative version of *Six1* (*Six1-EnR*; Brugmann et al., 2004) confirmed the ability of low-dose *Six1*-MO to reduce placodal expression of all target genes (as was also the case for *Eya1*-MO; figs. 5.4, 5.5 and 5.6). Although the use of *Six1-EnR* provides an additional line of evidence for the gene-specific action of *Six1*-MO, an important distinction between the two approaches must be made when comparing resulting phenotypes. While *Six1*-MO prevents the translation of endogenous *Six1* mRNA, and as such block Six1's ability to activate or repress transcription, overexpression of *Six1-EnR* converts injected Six1 into a transcriptional repressor. Thus, while *Six1-EnR* overexpression reduces the ability of endogenous Six1 to activate transcription, it has no effect on its ability to repress transcription. Care must be taken therefore in making direct comparisons between MO-mediated knockdown phenotypes and those resulting from dominant-negative overexpression.

#### 7.4 Dual role of Six1/Eya1 in neuronal differentiation and proliferation

Neurogenesis - the process by which progenitor cells become committed to a neural fate - is a tightly regulated process in embryonic and adult development. Normal development requires close co-ordination of the processes required to maintain a balance of proliferating progenitor cells and committed neurons, with deleterious repercussions if such a balance is not maintained: Favouring differentiation over proliferation may hinder the ability to regenerate and compensate for injury, whereas tipping the balance towards proliferation may promote cancer formation. Considering such dramatic consequences for an un-balanced system, little is known about how the decision to keep dividing or to differentiate is regulated in cells.

Six1 and Eya1 have recently been identified as key regulators of both proliferation and differentiation in cranial placodes (Ahmed et al., 2012b; 2012a; Schlosser et al., 2008; Zou et al., 2004). While these two processes are seemingly at odds with one another (given the decision a progenitor must make), Six1/Eya1 appear to play a dual role in regulating the balance between proliferating progenitors and differentiating cells in a dose-dependent fashion (Schlosser et al., 2008). However, while it has been demonstrated that Six1/Eya1 promote progenitors at high doses and differentiation at low doses, precisely how they coordinate such cell fate determination is currently unclear. The direct targets of Six1/Eya1 as

revealed in this study were described as belonging to two groups: Those expressed in proliferating progenitors of PPE and those expressed in differentiating cells of placodes. Accordingly, both facets of their function will be discussed separately below.

#### 7.4.1 *Six1/Eya1* and neuronal differentiation

*Six1* and *Eya1* are ancient metazoan genes involved in neurogenesis. In *Drosophila*, together with *Pax* and *Dach* genes, they form the *Pax-Six-Eya-Dach* network that is essential for compound eye development (Bonini et al., 1997; Cheyette et al., 1994; Donner and Maas, 2004; J. P. Kumar, 2009), and their misexpression is sufficient to induce ectopic eyes (Bonini et al., 1997; R. Chen et al., 1997; Pignoni et al., 1997). The finding that *Drosophila* orthologues of *Six1* and *Eya1* are often co-expressed with various neuronal differentiation markers, and directly activate the proneural gene *ataonal*, suggests an ancient role in promoting neuronal differentiation (Tanaka-Matakatsu and Du, 2008; T. Zhang et al., 2006). Such roles for *Six1/Eya1* in eye development and neurogenesis are widely conserved throughout metazoa (Gehring, 2005), however in chordates, these generic properties of *Six1/Eya1* interaction in promoting neurogenesis appear to have been co-opted for sense organ development (Schlosser, 2014). In vertebrates, mutation or knockdown of *Six1* or *Eya1* results in strong defects in neurogenesis affecting multiple placodal derivatives (Bricaud and Collazo, 2011; Ikeda et al., 2007; Laclef et al., 2003; X. Li et al., 2003; Schlosser et al., 2008; P. X. Xu et al., 1999; Zheng et al., 2003; Zou et al., 2004). For example in mice, *Six1*<sup>-/-</sup> mutants show severe defects in early neurogenesis in the olfactory epithelium (Ikeda et al., 2007), and cessation of inner-ear development (Ahmed et al., 2012a; Zheng et al., 2003). It is suggested that *Six1/Eya1* effect this role in neurogenesis through activation of neuronal determination genes, such as *Ngn1* and *Atoh1* whose expression in *Six1/Eya1* mutants or morphants is reduced or completely absent in many placodes (Bosman et al., 2009; Bricaud and Collazo, 2011; Ikeda et al., 2007; Zou et al., 2004). *Atoh1* is a bHLH pro-neural factor essential for initial hair cell differentiation (Bermingham et al., 1999), and has been demonstrated as a direct target of *Six1/Eya1* in vertebrates (Ahmed et al., 2012a). In addition to driving hair cell development, *Atoh1* is additionally required for the activation of genes involved in differentiation and maintenance of hair cells, such as *Gfi1a* (Wallis et al., 2003a). The closely related bHLH gene *Ngn1* serves a similarly pro-neural function, and (together with *Ngn2*) acts to initiate the neuronal differentiation programme in cranial sensory ganglia by directly activating *NeuroD1* (Seo et al., 2007). Indeed, in *Xenopus*, *Six1/Eya1* are required for *Ngn1* and *NeuroD1* expression in all neurogenic placodes (Schlosser et al., 2008).

This study, in addition to confirming *Atoh1* as a direct target of *Six1/Eya1* in the PPE, has revealed and confirmed several additional neurogenic genes as direct targets

including *Ngn1*, *Pou4f1.2*, *Tlx1*, *Isl2* and *Gf1a*. Interestingly, many of these genes have been demonstrated to positively regulate each others expression in a pro-neural cascade, and *Pou4f1.2*, *Isl2*, *Gf1a* and *Tlx1* are all known to function downstream of *NeuroD1* (Hertzano, 2004; Hutcheson and Vetter, 2001; Lee et al., 1995). Additional functional experiments demonstrated that *Six1/Eya1* overexpression was able to promote up-regulation of each of these targets in a majority of embryos (although this ability to promote up-regulation was stronger after *Eya1* overexpression compared to *Six1*), as well as causing their down-regulation to a lesser extent (table 5.3; figs. 5.7 and 5.8). Considering that these experiments were conducted in the absence of CHX (and therefore allow the activation of both direct and indirect targets of *Six1* and *Eya1*), that the observed down-regulation was considered an artifact of indirect effects. That *Six1* and *Eya1* directly activate such genes (table 3.3), but apparently simultaneously inhibit neuronal differentiation, strongly suggests the existence of complex downstream regulatory landscape that facilitates both processes under different circumstances.

#### 7.4.2 *Six1/Eya1* and proliferation

As well as promoting neuronal differentiation, *Six1* and *Eya1* can also stimulate proliferation, and independently inhibit neurogenesis by inducing apoptosis (Bricaud and Collazo, 2006). Mutation or knockdown of *Six1* or *Eya1* reduces proliferation in placodes, and results in diminished placodal domains (X. Li et al., 2003; Schlosser et al., 2008; Zheng et al., 2003; Zou et al., 2006), while their overexpression promotes maintenance of progenitor state (Kriebel et al., 2007; Schlosser et al., 2008). This role of *Six1/Eya1* in proliferation has garnered substantial interest, particularly after *Six1* was demonstrated to be overexpressed in primary breast cancers and metastatic lesions (Ford et al., 1998). Since these initial observations, both *Six1* and *Eya1* have been implicated in multiple “hallmarks of cancer” (such as sustained proliferation, invasion and metastasis), and the *Six1-Eya1* complex is considered a therapeutically valuable target in cancer treatment (Blevins et al., 2014).

One mechanism through which *Six1/Eya1* may act to maintain progenitors in a proliferative state is by directly activating cell cycle control genes (such as *CyclinA1*, *CyclinD1*, and *c-Myc*) (Coletta et al., 2004; X. Li et al., 2003; Z. Li et al., 2013; Yu et al., 2006). For example one study demonstrated that *Six1* directly activates *CyclinD1* in pancreatic cancer cells, promoting cell cycle progression and proliferation (Z. Li et al., 2013). Consistent with such a result, this study found a *CyclinD1*-like gene highly significantly up-regulated after *Six1* overexpression (*Six1<sub>i</sub>* FC: 7.5 [appendix E.1]; *Six1<sub>m</sub>* FC: 6.2, q = 0.009 [appendix F.1]). However, considering that *CyclinA1* was down-regulated after *Six1* overexpression (*Six1<sub>i</sub>* FC: -3.7 [appendix E.2]), but unaffected in the merged analysis (and so not supported by the

statistical analysis) it seems unlikely that it is directly activated by Six1 in cranial placodes. Consistent with this, a recent study also found the expression of *CyclinA1* unaffected by either *Six1* or *Eya1* overexpression (Schlosser et al., 2008).

Additionally, it has been suggested that Six1/Eya1 may directly activate *SoxB1* genes (*Sox2* and *Sox3*), whose expression is associated with stem cell-like properties and promotes the proliferation of neuronal and sensory progenitor cells (Bosman et al., 2009; Chew and Gallo, 2009; Miyagi et al., 2009; Schlosser et al., 2008). The results from this study strongly support such direct activation, with *Sox2* significantly up-regulated after *Six1/Eya1* overexpression in both RNA-Seq (Six1+Eya1<sub>m</sub> FC: 1.5, q = 0.008 [table 3.3 and appendix F.5]) and qPCR data (figs. 4.25 and 4.26). Although *Sox3* showed weaker up-regulation (Six1+Eya1<sub>m</sub> FC: 0.6, q = 0.18 [table 3.3 and appendix F.5]) it has previously been shown to be ectopically activated by hormone-inducible versions of Six1 and Eya1 in the presence of CHX even in trunk ectoderm (Schlosser et al., 2008), suggesting that it is also a direct target of Six1 and Eya1 (also see fig. 3.2).

In addition to members of the *SoxB1* family, this study has identified several other genes associated with proliferation as direct targets of Six1/Eya1 including *Hes5/8* and *Hes9*. *Hes5* genes are canonical effectors of Notch signaling, where they act to inhibit neuronal differentiation by direct repression of pro-neuronal genes such as *Ngn1*, thus maintaining the cell in a progenitor state (Ohtsuka, 1999). *Hes5* has also been shown to be a direct target of *Sox2* (Engelen et al., 2011), providing an additional mechanism through which *SoxB1* genes may act to block differentiation. Considering the *Hes9* gene identified appears to belong to a *Hes5* derived sub-family (fig. 4.2; possibly resulting from gene duplication or the divergence of a *Hes5* pseudoallele (Y. Li et al., 2003)), it is likely that these two genes serve a similar role in development, although further characterisation is required to determine the extent to which this is true. As was the case for the neuronal differentiation genes, *Six1/Eya1* overexpression in the absence of CHX induced both up- and down-regulation of direct targets (table 5.3; figs. 5.7 and 5.8), suggestive of a context-dependent dual role of Six1 and Eya1 in promoting proliferation and repressing neuronal differentiation.

#### *7.4.3 Six1/Eya1 play a dual role in promoting both neuronal differentiation and proliferation in a dose-dependent fashion*

It is possible that Six1 and Eya1 exert such a dual role by directly activating *SoxB1* genes which, although typically act to maintain cells in a proliferative and undifferentiated stem cell-like state (Chew and Gallo, 2009; Miyagi et al., 2009; Wegner and Stolt, 2005), also bias

lineage choices of progenitor cells towards a neural fate, and are required for subsequent neuronal differentiation (Cavallaro et al., 2008; Hoffmann et al., 2013; Kishi et al., 2000; Mizuseki et al., 1998; Schlosser et al., 2008; Zhao et al., 2004). While it is unclear how exactly *SoxB1* genes function in this seemingly contradictory fashion, one possibility is that they promote different lineage decisions at different concentrations, with high concentrations required for neuronally-biased progenitor maintenance, and low doses facilitating *SoxB1*-dependent neuronal differentiation (Dabdoub et al., 2008; Schlosser, 2010; Taranova et al., 2006). Given these two functions of *SoxB1* genes, it is likely that to at least some extent the ability of Six1/Eya1 to act in both capacities is *SoxB1*-mediated. However, while *SoxB1* genes may act to promote neuronal differentiation in some contexts, unlike Six1/Eya1 they are unable to induce ectopic neurons, suggesting that Six1/Eya1 may promote neuronal differentiation by additional, and *SoxB1*-independent, mechanisms (Schlosser et al., 2008). Like *SoxB1* genes, Six1 and Eya1 have dose-dependent effects in placode development: At high doses they maintain cells in a proliferative state (in part by expanding the expression of *SoxB1* genes), and block the expression of neuronal determination and differentiation genes, whereas at low doses they promote neuronal differentiation (Schlosser et al., 2008). This dosage dependent role of Six1/Eya1 may partially explain the seemingly contradictory expression dynamics of the direct targets identified in this study - where most were both up- and down-regulated - after *Six1/Eya1* overexpression under conditions in which protein synthesis was not blocked allowing the activation of both direct and indirect targets (table 5.3; figs. 5.7 and 5.8). Indeed, considering the concentration of *Six1/Eya1* injected in this study was the same as that previously shown to be sufficient to favour differentiation over proliferation (Schlosser et al., 2008), it is possible that the observed tendency for *Six1/Eya1* overexpression to more frequently induce up-regulation in genes associated with neuronal differentiation (*Atoh1*, *Ngn1*, *Pou4f1.2*, *Tlx1*, *Isl2* and *Gfi1a*), compared to proliferation (*Sox2*, *Sox3*, *Hes5/8* and *Hes9*) may be partly ascribed to the injection dose (table 5.3).

An additional mechanism through which Six1/Eya1 may serve this dual role in placode development concerns the distinct transcriptional outcomes resulting from differential binding of Six1 to co-factors. When bound to Eya1, Six1 acts as a transcriptional activator to promote proliferation, and protect cells from apoptosis (Bricaud and Collazo, 2006). When coupled with non-Eya1 binding partners (e.g. Groucho or Dach) in neuronal lineages, Six1 represses transcription of target genes, blocking the proliferation such cells and inducing programmed cell death, resulting in fewer neurons (Bricaud and Collazo, 2011; 2006). However, while such differential binding dynamics undoubtedly influence placode development (Brugmann et al., 2004), it is unclear how they relate to the described dosage dependent effects. Moreover, *Six1* and *Eya1* are prominently co-expressed throughout the

PPE, and maintained in all neurogenic placodes throughout development (Schlosser and Ahrens, 2004), suggesting that differential cofactor binding of Six1 is unlikely to explain the dual role both genes play in promoting both proliferation and neuronal differentiation.

#### 7.4.4 *Six1/Eya1* inhibit neuronal differentiation by directly activating *Hes5/8*

That Six1/Eya1 directly activate genes other than *Sox2* and *Sox3* that are associated with progenitor maintenance (*Hes5/8* and *Hes9*) suggests that, while their ability to promote proliferation may be influenced by *SoxB1* genes, it is not entirely dependent on them. This study has demonstrated *Xenopus Hes5/8* to be both a direct transcriptional target of Six1/Eya1 in the PPE (figs. 4.23 and 4.24) and a direct effector of Notch signalling (table 6.1 and fig. 6.1), where - typically for a *Hes5* gene (Ohtsuka, 1999) - *Hes5/8* acts to repress neurogenesis (fig. 6.1G and H). By demonstrating that Six1/Eya1 can activate *Hes5/8* directly, and independently from Notch signalling (table 6.1 and fig. 6.2), this study suggests an exciting and novel mechanism through which Six1/Eya1 may act to promote proliferation in the PPE. The basis of this is discussed in detail later (see section 7.5), but can be briefly summarised as follows. At neural plate stage, Six1/Eya1 are co-expressed at high levels throughout the PPE where, through the combinatorial activation of *SoxB1* (*Sox2* and *Sox3*) and *Hes* genes (*Hes5/8* and possibly *Hes9*), they act to promote progenitor maintenance and block the onset of neuronal determination. At later stages, down-regulated Six1/Eya1 are unable to maintain activation of pro-proliferation genes such as Hes and the relief of Hes-mediated repression of pro-neuronal genes (*Atoh1*, *Ngn1*), combined with their direct activation by Six1/Eya1, permits neuronal differentiation. Moreover, at low doses, Six1/Eya1 actively promote neuronal determination and differentiation by directly activating genes downstream of *Ngn1*, including *Pou4f1.2*, *Tlx1*, *Isl2* and *Gfi1a* (after Schlosser et al., 2008).

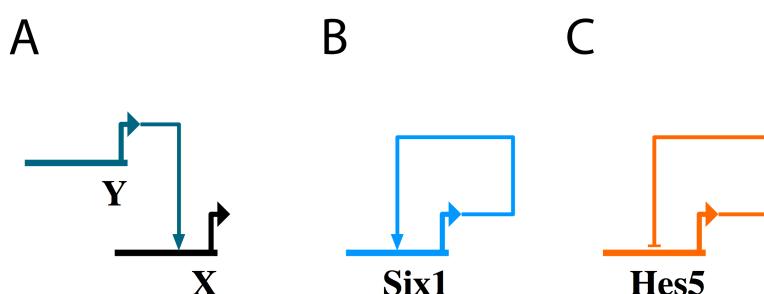
### 7.5 Towards a gene regulatory network

An underlying mechanism in development is the regulation of gene expression, often the product of complex networks of interactions between transcription factors and the genes they regulate (Alon, 2007; Davidson, 2011; Levine and Davidson, 2005). Transcription factors are expressed in a spatio-temporally restricted fashion, and bind *cis*-regulatory elements (which, in combination with other nearby elements, comprise *cis*-regulatory modules) of DNA in the vicinity of target genes to influence their transcription and effect a particular regulatory state. In general, individual *cis*-regulatory modules can be described as driving a specific pattern of gene expression in space and time, and can be combinatorially actuated to produce complex patterns of gene expression (Gray et al., 1994; Levine and Davidson, 2005). Development, then, proceeds by the iterative (and often ephemeral) installation of particular transcriptional

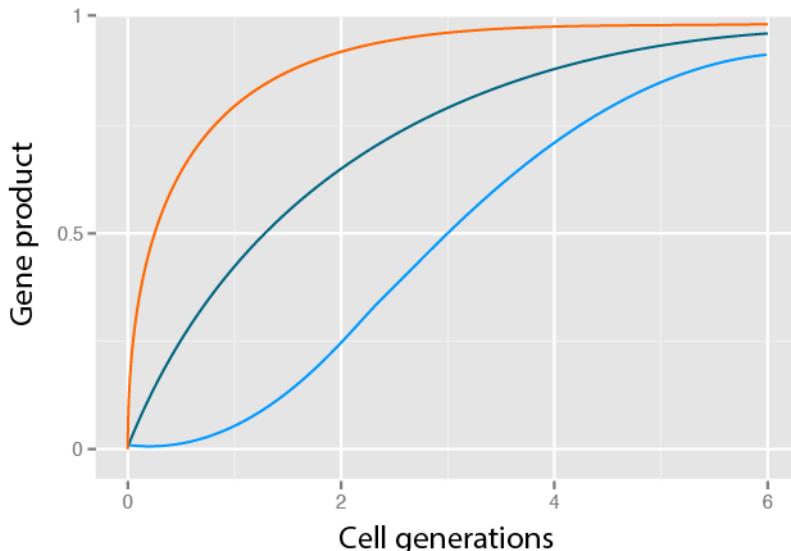
regulatory states in embryonic space. Such states result from the expression of a specific set of regulatory genes (transcription factors) that drive particular developmental programmes. Gene regulatory network models (GRNs) attempt to formalise the manner in which cellular states are specified by such interactions by consolidating functional experimental data with theoretical models of gene interaction and transcriptional dynamics (Bolouri and Davidson, 2003; Peter et al., 2012).

### 7.5.1 General features of GRNs

While GRN complexity can be staggering (Peter and Davidson, 2011; 2010), gene networks are composed of multiple sets of recurrent regulatory patterns, sub-circuits, that are responsible for accomplishing a given regulatory task (Alon, 2007; Davidson, 2011; Milo et al., 2002; Shen-Orr et al., 2002). Thus, the sub-circuits that comprise a given GRN indicate its operational significance, and therefore a passing familiarity with them can aid prediction of network interactions, and inform experimental design. Considering such network features have been reviewed thoroughly (e.g. Alon, 2007; Davidson, 2011), this discussion will only highlight some of the most salient sub-circuits in light of constructing a Six1/Eya1-driven placodal GRN. As a reference for understanding GRN dynamics, it is fruitful to consider first a simple two-gene regulatory interaction, where gene X is activated by transcription factor Y (fig. 7.2A). While the following discussion will treat all sub-circuits presented here as existing in an isolated state, with no external interactants, all such sub-circuits require initial activation, and are subject to finite cellular resources.



**Fig. 7.2** Simple regulation and autoregulation. In simple regulation (**A**), an upstream factor Y activates the expression of gene X. In positive autoregulation (**B**; PAR), a transcription factor (e.g. Six1) activates its own transcription. In negative autoregulation (**C**; NAR), a transcription factor (e.g. Hes5) represses its own transcription.



**Fig. 7.3** Transcriptional dynamics resulting from simple regulation and autoregulation. NAR (orange line) under a strong promoter speeds up the response time required to reach a given steady-state of expression relative to the time taken for a gene under simple regulation (blue-green line) to attain the same level. PAR (blue line) slows the response time (adapted from Alon, 2007).

In simple regulation, on initiation of transcription, the concentration of X increases and reaches a steady state - equal to the ratio between production and degradation (as a result of both active degradation and dilution due to cell growth) (fig. 7.3; Alon, 2007). X concentration declines exponentially if the activating signal is removed. Transcription under simple regulation tends to be slow (Rosenfeld et al., 2002), largely to prevent undesirably high production rate. Autoregulation, however, results in different response times generally by utilising weak or strong promoters: Whereas strong promoters are associated with genes that negatively autoregulate (and thus stem their own production), the opposite is true for genes that positively autoregulate, or are produced under simple regulation (Rosenfeld et al., 2002).

#### 7.5.1.1 Autoregulation

Autoregulation is the simplest network interaction, involving only a single transcription factor. Autoregulation is a remarkably common feature of GRNs - two thirds of transcription factors autoregulate in *E. coli* transcriptional networks (Shen-Orr et al., 2002) - and has been associated with several developmental properties. First, it has been suggested that autoregulation may be positively correlated with the developmental importance of that transcription factor. Thus, regulators that control large numbers of genes will be autoregulated, as their expression levels must be tightly controlled. In addition, the ability of autoregulating genes to mediate their own expression levels may play a role in maintaining cell fate and differentiation, thereby constituting the epigenetic memory of a differentiation state (S. T. Crews and Pearson, 2009).

### 7.5.1.1.1 Positive autoregulation

In positive autoregulation (PAR), a transcription factor activates its own promoter, and enhances its own rate of production (fig. 7.2B). Considering Six1 is known to positively autoregulate its own transcription (Sato et al., 2012) - which this study corroborates (table 3.3) - it can be considered here as an example of PAR. Even for such a simple interaction, the transcriptional dynamics resulting from PAR warrant further consideration. On initial activation, protein levels of X are low, and X production is correspondingly slow. Only when levels approach the concentration threshold required to activate its own promoter does production increase, and thus a steady-state is subject to a delayed response time and achieved in a sigmoidal curve (fig. 7.3). In addition to slowing response time, PAR also tends to increase cell-cell variation in protein levels. Such variation is inherent between cells, as a result of intrinsic fluctuations in protein production and degradation rates (Kærn et al., 2005). Thus, while such a delayed response may facilitate many developmental processes (for example when a protein produced at an early stage of development is used at a later stage), PAR canonically functions to broaden the range of protein distribution between cells, and in extreme cases produces bimodal distributions - generating a differentiation-like division of cells into two populations (Alon, 2007; Becskei et al., 2001; Fang et al., 2008; Karmakar and Bose, 2007; Maeda and Sano, 2006). As it has been indicated that *Six1* expression in the otic vesicle is dependent on the presence of functional Six1 protein (Grifone et al., 2005), autoregulation may play an important role in placode development.

### 7.5.1.1.2 Negative autoregulation

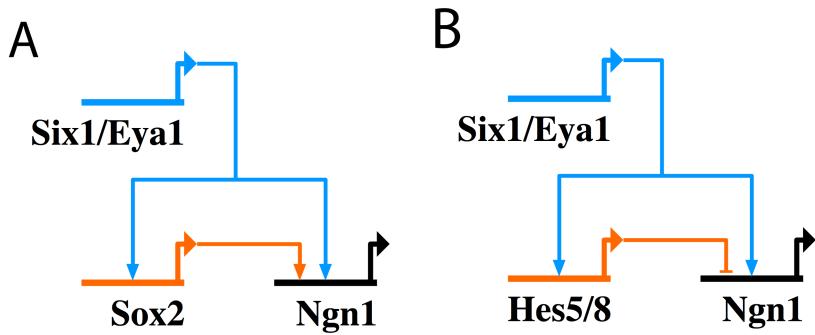
Conversely, in negative autoregulation (NAR; for example Hes5 (Cooper et al., 2000; Fior and Henrique, 2005)), a transcription factor represses transcription of its own gene (fig. 7.2C). NAR is demonstrated by approximately half of all negative regulators in *E. coli* (Thieffry et al., 1998), and like PAR, displays characteristic transcriptional dynamics. Initially, NAR reduces the response time by using a strong promoter to obtain a rapid initial rise in the concentration of protein X. When this concentration approaches a threshold sufficient to repress its own promoter, the production rate of X decreases, and thus X levels are locked into a steady-state marginally below the threshold for repression (fig. 7.3). In addition to speeding up response time, NAR provides robustness to cellular noise: High levels of X repress its own transcription, whereas low levels increase production. Typically this results in a narrower distribution of protein level compared to that found in simple regulation (Becskei and Serrano, 2000).

### *7.5.1.2 Feed-forward loops*

A second group of network interactions involve a feed-forward loop (FFL). Here, a transcription factor X regulates a second transcription factor Y, and X and Y both regulate gene Z. Considering that each of these interactions can be either positive (activation) or negative (repression), there are eight possible configurations of FFL. These structural arrangements result in either coherent (i.e. the sign of the direct [X - Z] and indirect [X - Y - Z] path from X to Z is the same) or incoherent (i.e. the sign of the two paths is different) feed-forward following X activation (Mangan et al., 2003). Furthermore, regulation of gene Z can require a combinatorial input from both X and Y (AND-gate), or individual input from either X or Y (OR-gate). Thus, each of the eight FFL types can occur with either of these *cis*-regulatory input functions (Buchler et al., 2003; Yuh et al., 1998). Like autoregulatory motifs, FFLs are abundantly common in diverse transcriptional networks (Mangan et al., 2003; Milo et al., 2002; Neves et al., 2013; Raft and Groves, 2014; Shen-Orr et al., 2002), however two appear far more regularly than others (the coherent type-1 FFL and the incoherent type-1 FFL), and only these will be considered by this discussion (however, see Mangan and Alon, 2003 for a thorough review of all FFLs).

#### *7.5.1.2.1 Coherent feed-forward loops*

In the coherent type-1 FFL (for simplicity this will henceforth be referred to as ‘coherent FFL’) both X and Y are transcriptional activators (fig. 7.4A). Assuming that the Z promoter has an AND input function (requiring both X and Y for activation) this coherent FFL results in a delayed activation of Z. Initially, Y is activated by X, however Z activation depends on the concentrations of both X and Y to be above the activation threshold for the Z promoter, and thus its activation is subject to delay. However, on removal of the X-activating signal, Z concentration rapidly diminishes (due to its requirement of both X and Y). Thus, this coherent FFL is known as being ‘sign-sensitive’ - an activating ‘ON’ signal results in delayed Z activation, whereas an ‘OFF’ signal experiences no delay. Coherent FFLs of this type therefore only express Z under persistent signals, and act to filter out ephemeral X activation (Alon, 2007). Alternatively, if the Z promoter responds to an OR input function, the reverse is true: There is no delay in Z production after activation, but reduction is delayed on removal of X-activating signal, allowing continued production of Z with a transient loss of input signal (Mangan and Alon, 2003).



**Fig. 7.4** Feed-forward loops. In the coherent type-1 FFL shown here (**A**), the Six1/Eya1 complex ('X') directly activates both Sox2 ('Y') and Ngn1 ('Z'), and Sox2 also activates Ngn1. In the incoherent type-1 FFL (**B**), while the Six1/Eya1 complex ('X') directly activates both Hes5/8 ('Y') and Ngn1 ('Z'), Hes5/8 also represses Ngn1. In both cases, production of Ngn1 can either require a combinatorial input from both X and Y (AND-gate), or individual input from either X or Y (OR-gate). Solid arrows show direct activation; solid barred lines indicate direct repression.

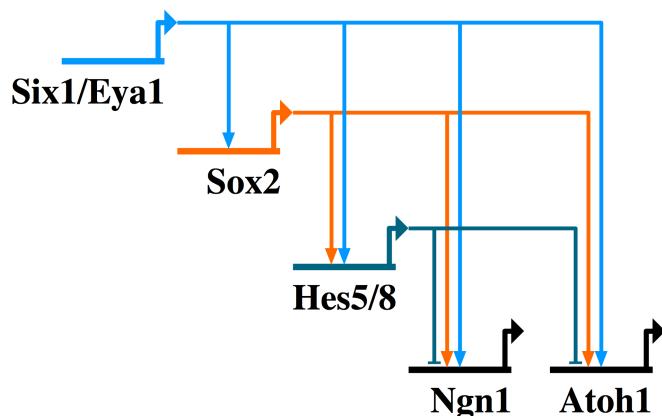
Among the direct Six1/Eya1 targets revealed in this study are *Sox2*, *Ngn1* and *Atoh1*. Considering that *Sox2* has been shown to directly bind the promoter of both *Ngn1* (Cimadomore et al., 2011) and *Atoh1* (Neves et al., 2013), these novel interactions provide insight into the mechanisms through which Six1/Eya1 drive the PPE programme. Assuming the ability of *Sox2* to directly activate *Ngn1* is conserved in the placodes, these findings permit the construction of a coherent FFL consisting of Six1/Eya1, *Sox2* and *Ngn1* (fig. 7.4A). Interestingly, this three-gene interaction alone is potentially able to explain some of the paradoxical results this study has revealed: Provided that the *Ngn1* promoter responds to a AND input function, activation of *Ngn1* would be subject to delay (see above). Initial production of *Sox2* would be rapid (and promote progenitor maintenance) but would eventually cross the threshold required to activate *Ngn1* in conjunction with Six1/Eya1 (and promote neuronal differentiation) at later stages. Although this study found that Six1/Eya1 directly activate *Ngn1* in the absence of protein synthesis (combined treatment with CHX+DEX), it cannot rule out that *Sox2* may be required for co-activation of *Ngn1*. A likely alternative to a dual requirement of Six1/Eya1 and *Sox2* for *Ngn1* activation (AND-gate) is that while Six1 acts as an essential factor for the expression of *Ngn1*, synergism with Eya1 and/or *Sox2* amplifies its ability to drive the expression of *Ngn1* (Ahmed et al., 2012b; 2012a).

#### 7.5.1.2.2 Incoherent feed-forward loops

The defining characteristic of incoherent FFLs is the paradoxical effect of X-activation on levels of Z. In the incoherent type-1 FFL (henceforth ‘incoherent FFL’), X activates both Y and Z, but Z is additionally repressed by Y (fig. 7.4B), therefore X both directly activates (X - Z), and indirectly (X - Y - Z) represses production of Z. Such a conformation results in a rapid initial production of Z (as activated by X) followed by a decrease in Z production (on

X-activated Y reaching the repression threshold for the Z promoter), creating a pulse-like dynamic of Z production. In addition, like NAR, regulation under incoherent FFLs results in an accelerated response time of Z, compared to a corresponding simple regulation system. However, while NAR necessitates that X be a transcription factor, such an incoherent FFL can speed-up production of any gene (Alon, 2007).

This study has demonstrated that Six1/Eya1 regulate Hes5/8 and Ngn1 in an incoherent FFL (fig. 7.4B) whereby Six1/Eya1 directly activates Hes5/8 and Ngn1 (figs. 4.25 and 4.26), and Hes5/8 represses Ngn1 (fig. 6.1). While this study presents no evidence that Hes5/8 directly represses Ngn1, it demonstrates that Hes5/8 functions as a canonical *Hes5* gene, and would thus strongly predict a direct repression of neuronal differentiation genes consistent with other reports (Ohtsuka, 1999). Interestingly, a similar mechanism has been proposed for hair cell development - where Sox2 positively regulates both Ngn1 and a bHLH factor (possibly a Hes gene), which in turn represses *Ngn1* (Raft and Groves, 2014) (a similar mechanism also regulates Atoh1 (Neves et al., 2013)) - suggesting substantial redundancy between the Sox2 (and/or Sox3)-induced regulation of pro-neural genes and their regulation by Six1/Eya1 as revealed in this study (fig. 7.5). Further work is required to determine in what circumstances Six1/Eya1 act upstream of, or synergistically with, Sox2.

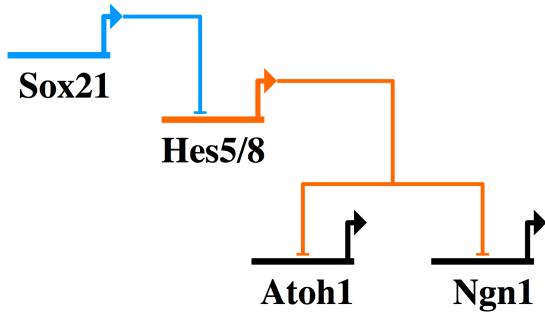


**Fig. 7.5** Combined Six1/Eya1/Sox2 network. Both the Six1/Eya1 complex and Sox2 regulate multiple players involved in cell fate commitment. Solid arrows show direct activation; solid barred lines indicate direct repression.

#### 7.5.1.3 Double-negative gates

Another important sub-circuit in development is the double-negative gate. Here, two repressors (X and Y) are wired in tandem such that X represses Y, and Y represses downstream regulatory genes  $Z_1 - Z_n$  (fig. 7.6). Thus, effector genes  $Z_1 - Z_n$  are only expressed in cells where X is active (domain X), and are shut down in all other cells (domain Y).

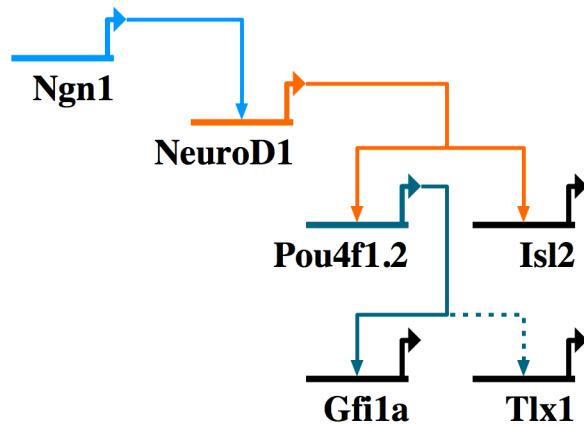
Assuming that Y is a broadly expressed (or global) inhibitor, this sub-circuit serves to install a particular regulatory state in domain X and inhibit that state in all other cells (Davidson and Levine, 2008). Recently, it has been demonstrated that *Sox21* promotes neuronal differentiation by directly repressing *Hes5* (Matsuda et al., 2012), and in light of this it is interesting that *Sox21* (as well as *Hes5/8*) was also found to be significantly differentially affected in the RNA-Seq screen in this study (table 3.3). Considering the restricted expression of *Sox21* in placodes (olfactory and otic vesicle at late tail bud stage; fig. 4.17), and that its expression appears to coincide with that of *Hes5/8* in these placodes (fig. 4.14), it seems unlikely that this serves a global role in directing neuronal differentiation in placodes. However, without first establishing whether *Sox21* and *Hes5/8* are co-expressed (or expressed in neighbouring cells), that *Sox21* plays some role in directing neuronal differentiation in placodes can't be dismissed (fig. 7.6).



**Fig. 7.6** Double-negative gate. Two repressors (here *Sox21* and *Hes5/8*) are wired in tandem such that *Sox21* represses transcription of *Hes5/8*, and *Hes5/8* represses downstream genes, shown here as a neuronal differentiation battery composed of *Atoh1* and *Ngn1*. In double-negative gates, downstream genes *Atoh1* and *Ngn1* are only expressed in cells where *Sox21* is active. Solid barred lines indicate direct repression.

### 7.5.2 A neuronal differentiation cascade

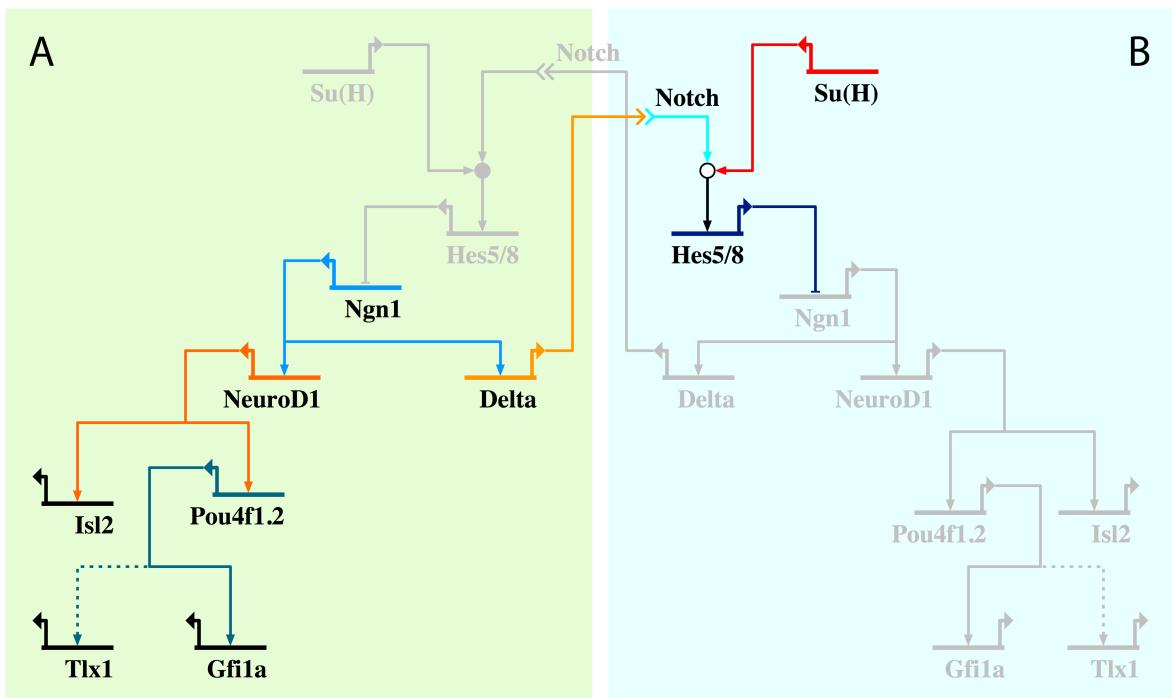
The bHLH gene *Ngn1* is a key regulator of neurogenesis, required for initial commitment to a neurogenic fate (Ma et al., 1998; 1996) and thus normal sense organ development (Ma et al., 2000). Expression of *Ngn1* locks in neuronal differentiation by directly activating *NeuroD1* which in turn activates a battery of neuronal differentiation genes (Seo et al., 2007) including *Pou4f1.2* (Hutcheson and Vetter, 2001) and *Isl1* (Lee et al., 1995). *Pou4f1.2* (*Brn3d*; considered a functional homologue of *Brn3b* and *Brn3c* (Hutcheson and Vetter, 2001)) furthermore activates *Gfi1a* directly (Hertzano, 2004) and *Tlx1* indirectly (Hutcheson and Vetter, 2001). Thus, neurogenesis proceeds in a step-wise cascade as shown in fig 7.7. As well as locking the cell in a neuronal differentiation state by initiation of this cascade, *Ngn1* also directly activates the Notch ligand *Delta*, which inhibits neighbouring cells from committing to a similar state (and thus keeping them in a progenitor state) via lateral inhibition (fig. 7.8; Ma et al., 1996).



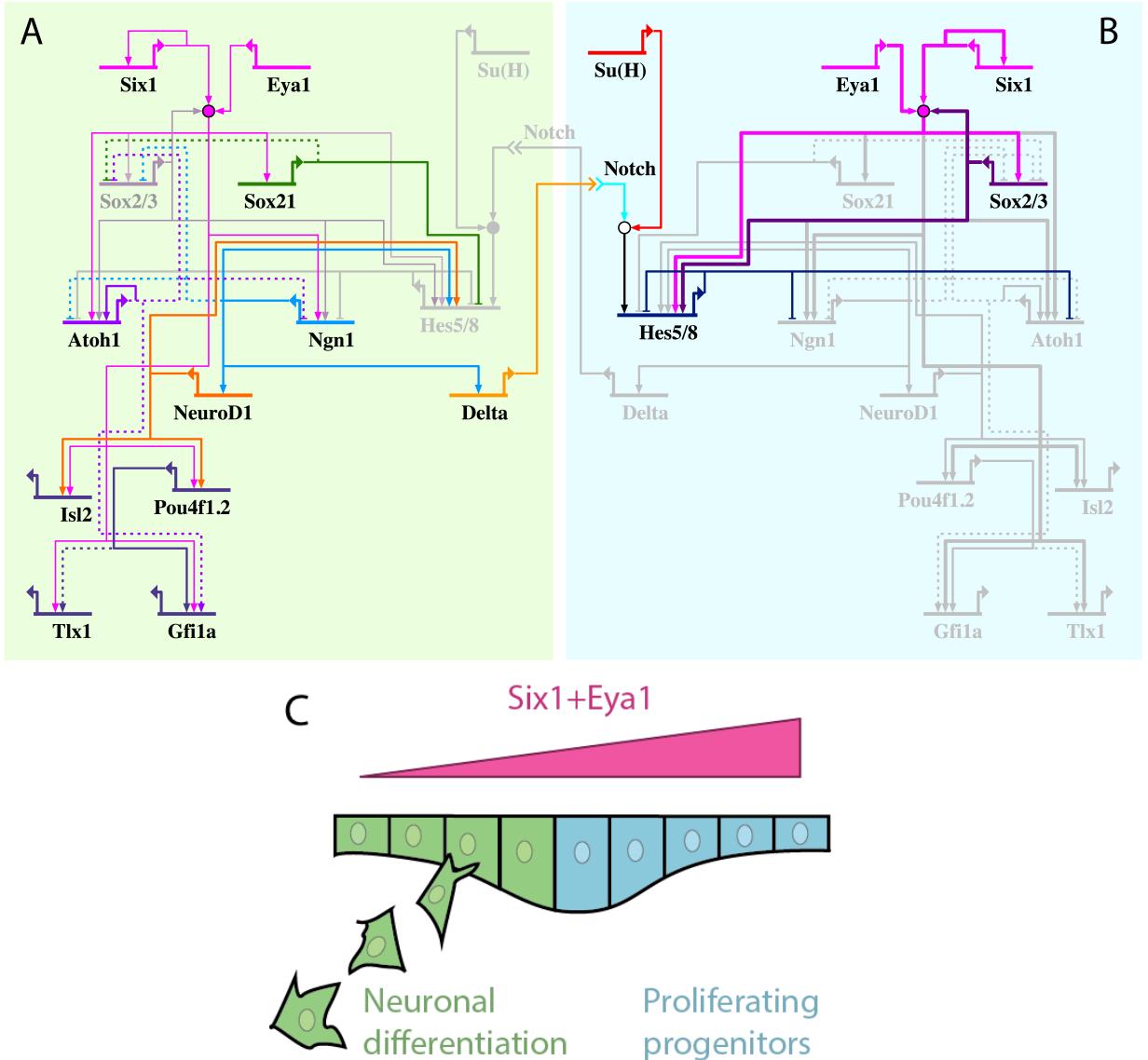
**Fig. 7.7** Neuronal differentiation proceeds in a cascade of gene regulation. Arrowheads indicate up-regulation. Arrows indicate direct (solid line) and indirect (dotted line) activation.

### 7.5.3 A model of Six1/Eya1-driven gene regulation in the PPE

Consistent with previous reports that Six1/Eya1 act not only to activate *Ngn1* and *NeuroD1*, but are additionally required to synergise with *Ngn1* and *NeuroD1* to promote the expression of neuronal differentiation genes (Ahmed et al., 2012b), this study has demonstrated that Six1/Eya1 directly activate several neuronal differentiation genes downstream of *NeuroD1* (*Pou4f1.2*, *Isl2*, *Gfi1a* and *Tlx1*). Six1/Eya1 therefore interact at multiple steps of this cascade to mediate neuronal differentiation. When this neuronal differentiation cascade is considered in context with the other sub-circuits previously discussed, and alongside Notch signalling, a complete model of Six1/Eya1-directed gene regulation in the PPE can be considered (fig. 7.9A and B). Here, Six1/Eya1 drive two distinct but inextricable programmes of neuronal differentiation and progenitor maintenance, at low and high doses, respectively (fig. 7.9C), to orchestrate placode formation and the development of sense organs. At low doses, Six1/Eya1 promote neuronal differentiation by directly activating pro-neural genes such as *Ngn1* and *Atoh1*, but their low expression is unable to activate pro-proliferation genes such as *Sox2* and *Hes5/8* above a certain threshold required for maintenance of a progenitor state. Conversely, at high doses, Six1/Eya1 strongly activates (and probably complexes with) *Sox2* to promote proliferation, at least partially by directly activating *Hes5/8*, which represses neuronal differentiation. Feedback circuitry with the Notch system prevents cells immediately surrounding differentiating neurons from adopting a neural fate, and they are locked in a progenitor state via *Hes5/8* repression of *Ngn1*.



**Fig. 7.8** Lateral inhibition between two cells maintains the balance between proliferation and neuronal differentiation. When Notch signalling is turned off (**A**; differentiating cell), neuronal differentiation is allowed to proceed, and *Ngn1* initiates a cascade of pro-neuronal gene activation, as well as directly activating *Delta*. In neighbouring cells (**B**; proliferating cell), *Delta* interacts with Notch to activate Notch signalling, and inhibits neuronal differentiation via *Hes5/8* activation, which in turn represses pro-neuronal genes such as *Ngn1* (genes repressed shown as greyed out), keeping the cell in a progenitor state. Arrows indicate direct (solid line) and indirect (dotted line) activation; barred lines show direct (solid line) and indirect (dotted line) repression.



**Fig 7.9** Final GRN model for Six1/Eya1-activated gene regulation in the PPE. At low doses (**A**; thin pink lines) Six1/Eya1 act to promote neuronal differentiation by direct activation of pro-neuronal genes (*Ngn1*, *Atoh1*), but are unable to activate pro-proliferation genes such as *Sox2* and *Hes5/8* above a certain threshold required for maintenance of a progenitor state. At high doses of Six1/Eya1 (**B**; thick pink lines), *Sox2* is strongly activated (thick violet lines), and acts in concert with Six1/Eya1 to promote proliferation by directly activating *Hes5/8*, which represses neuronal differentiation. Feedback circuitry with the Notch system prevents cells immediately surrounding differentiating neurons from adopting a neural fate, and they are locked in a progenitor state via *Hes5/8* repression of *Ngn1*. Arrows indicate direct (solid line) and indirect (dotted line) activation; barred lines show direct (solid line) and indirect (dotted line) repression. Thin lines (e.g. those stemming from Six1/Eya1 and *Sox2* in **A**) indicate a low level of expression, whereas thick lines (e.g. Six1/Eya1 and *Sox2* in **B**) show high levels of expression. At low doses, *Sox2* is activated, but at correspondingly low levels (show as faded violet in **A**). Greyed out genes represent those inactive in current cell (e.g. *Hes5/8* in **A**; neuronal differentiation genes in **B**). Evidence for interactions: Six1 positively autoregulates (Sato et al., 2012); Six1/Eya1 directly activate *Sox2*, *Sox3*, *Sox21*, *Ngn1*, *Atoh1*, *Isl2*, *Pou4f1.2*, *Tlx1* and *Gfi1a* (this study); *Sox2* synergises with Six1/Eya1 (Ahmed et al., 2012a; 2012b); *Sox2* directly activates *Hes5* (Engelen et al., 2011), *Atoh1* (Ahmed et al., 2012a) and *Ngn1* (Cimad amore et al., 2011). *Sox21* directly represses *Hes5* (Matsuda et al., 2012) and indirectly inhibits *Sox2* (Ferletta et al., 2011); *Atoh1* and *Ngn1* indirectly repress each other (Gowan et al., 2001); *Ngn1* indirectly represses *Sox2* (Evsen et al., 2013); *Ngn1* directly activates *NeuroD1* and *Hes5* (Seo et al., 2007); *Atoh1* positively autoregulates (Helms et al., 2000); *Atoh1* indirectly represses *Sox2* (Neves et al., 2012) and activates *Gfi1* (Wallis et al., 2003); *Hes5* negatively autoregulates (Cooper et al., 2000); *Hes5* directly represses *Atoh1* and *Ngn1* (Neves et al., 2013; Ohtsuka, 1999; Raft and Groves, 2014); *NeuroD1* directly activates *Hes5* (Seo et al., 2007), *Pou4f1.2* (Hutcheson and Vetter, 2001) and *Isl1* (Lee et al., 1995); *Pou4f1.2* directly activates *Gfi1* (Hertzano, 2004) and indirectly activates *Tlx1* (Hutcheson and Vetter, 2001). **C** Shows a schematic representation of the dose-dependent effects of Six1 and Eya1 on proliferation and differentiation.

## 7.6 Conclusion

### 7.6.1 Future work

By contextualising the results generated by this present study with published and empirically demonstrated interactions between target genes, a model of Six1/Eya1-driven PPE specification and placode development was constructed (fig. 7.9). Such a GRN allows the formulation of testable hypotheses regarding neuronal differentiation and proliferation in placodes, several of which will be considered here. First, an immediately obvious line of enquiry concerns the dose-dependent role of Six1/Eya1 in promoting neuronal differentiation and proliferation. As the direct targets of Six1 and Eya1 revealed in this study are considered as being either pro-progenitor maintenance (*Sox2*, *Sox3*, *Hes5/8* and *Hes9*) or pro-neuronal differentiation (*Atoh1*, *Ngn1*, *Pou4f1.2*, *Isl2*, *Tlx1* and *Gfi1a*) it would be interesting to consider the effect of low and high dose of Six1/Eya1 on their expression. One would expect that, in light of similar approaches (e.g. Schlosser et al., 2008) and considering the hierarchy of regulatory events in neurogenesis (e.g. fig. 7.7), low doses of Six1/Eya1 would promote the expression of pro-neural genes, but not those promoting proliferation. However, to further probe this dual role of Six1 and Eya1, it must first be explained how exactly such dose-dependent effects are implemented. A likely scenario is that Six1 has different binding affinities for the enhancers of different target genes, although this requires further investigation. Additionally, it would be interesting to investigate, via gain- and loss-of-function experiments, whether Six1/Eya1 promote differentiation at low doses in a Hes5/8-dependent way (i.e. does Hes5/8 need to be down-regulated to allow the progression of neuronal differentiation?), and conversely whether Hes5/8 is required to inhibit Ngn1.

Another productive area of investigation pertains to the role *SoxB1* genes play in placode development. As highlighted previously (see sections 7.5.1.2.1 and 7.5.1.2.2), *SoxB1* genes appear to share several direct targets with Six1/Eya1, and have been shown to work synergistically with Six1/Eya1 in both facets of their dual function (Ahmed et al., 2012b; 2012a). One possibility is that Six1 acts as a critical activating factor in promoting pro-neural genes such as *Atoh1* and *Ngn1*, and can complex with Eya1 and Sox2 to strengthen this activation. Considering that a co-requirement of Sox2 in activation of Six1/Eya1 direct targets as revealed in this study cannot be excluded, further gain and loss-of-function experiments should be conducted to reveal the extent of Sox2's involvement. For example, does MO-mediated knockdown of *Sox2* (and/or *Sox3*) combined with overexpression of Six1/Eya1 reduce the extent to which *Ngn1* or *Hes5/8* are expressed ectopically? Furthermore, considering the role Sox21 plays in antagonising *SoxB1* genes to promote neuronal differentiation (Chew and Gallo, 2009; Sandberg et al., 2005), it would also be

fruitful to explore its function in placode development. Interestingly, it has recently been shown that Sox21 is required for neuron formation in the neural plate but, similar to *SoxB1* genes, appears to play a dual role: High levels prevent neuron formation and promote proliferation (by directly repressing *Ngn2*), whereas low doses are required for cell viability and differentiation of neurons (Whittington et al., 2015). Whether high levels of Six1/Eya1 are able to convert Sox21 from a pro-differentiation to a pro-proliferation factor in placodes is an exciting, and open, question.

Finally, while Six1/Eya1 appear to drive neuronal differentiation at low doses by interacting with multiple tiers of the neuronal differentiation cascade (fig. 7.9), whether they require co-factors to achieve this is unclear. The expression of *Ngn1* is necessary for the commitment of ectodermal cells to a neural fate in the otic vesicle (Ma et al., 2000; 1998), and directly activates *NeuroD1* to promote neuronal differentiation (Seo et al., 2007). However, *Ngn1* alone, or *Ngn1* synergising with either Six1 or Eya1 (*Ngn1-Six1* or *Ngn1-Eya1*) is unable to induce the expression of *NeuroD1*, indicating that both Six1 and Eya1 are required to form a complex with *Ngn1* to mediate neuronal differentiation and maturation (Ahmed et al., 2012b). Furthermore, Six1 or Eya1 can complex with *NeuroD1* to induce neuronal differentiation. Taken together, this raises the possibility that Six1 and Eya1 may activate several of the downstream neuronal differentiation genes (e.g. *Pou4f1.2*, *Isl2*, *Tlx1* and *Atoh1*) in collaboration with *Ngn1* and/or *NeuroD1*.

### 7.6.2 Summary

This study has revealed and verified ten direct transcriptional targets of Six1 and Eya1 in the pan-placodal ectoderm. These genes appear to play important roles in both progenitor maintenance and neuronal differentiation programmes, and form the basis of a Six1/Eya1-activated gene regulatory network for placode development, as shown in figure 7.9. Considering the important roles Six1 and Eya1 play in regulating sense organ development, as well as their implication in several cancers and craniofacial defects, the network revealed in this study is predicted to have wide-reaching impacts across these fields. In addition, considering that the pan-placodal ectoderm, as well as the individual placode-derived structures, are considered to be evolutionary novelties of the vertebrate “new head”, such a network provides the foundations for further work to explicate how evolutionary novelty can be generated by re-wiring gene regulatory networks.

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# Appendix A. Solutions

## A.1 General solutions

### 10 x MBS salts

51.3 g NaCl  
0.75 g KCl  
2 g MgSO<sub>4</sub> x 7 H<sub>2</sub>O  
23.8 g HEPES  
2.0 g NaHCO<sub>3</sub>  
pH adjusted to 7.6 with NaOH pellets  
Adjusted to 1 litre with dH<sub>2</sub>O and autoclaved

### 1 x MBSH

100 ml 10 x MBS salts  
7 ml 0.1 M CaCl<sub>2</sub>  
4 ml 5 M NaCl  
Adjusted to 1 litre with dH<sub>2</sub>O

### 1 x MBS

100 ml 10 x MBS salts  
7 ml 0.1 M CaCl<sub>2</sub>  
Adjusted to 1 litre with dH<sub>2</sub>O

### Cysteine

4 g Cysteine  
Adjusted to 100 ml with dH<sub>2</sub>O  
Adjusted pH to 8.0 with NaOH pellets

### 100 x Triple antibiotic (3A)

250 mg Gentamycine  
4 g Penicillin  
4 g Streptomycin  
100 ml dH<sub>2</sub>O  
Dispensed into 1 ml aliquots  
Stored at -20°C

### Transplantation solution

98 ml 1 x MBSH  
2 ml 0.1 M CaCl<sub>2</sub>  
1 ml 100 x 3A

### 50 x TAE

242 g Tris  
Dissolved in 800 ml dH<sub>2</sub>O  
57.1 ml Glacial acetic acid  
18.6 g EDTA  
Adjusted pH to 7.6-7.8 with glacial acetic acid  
Adjusted to 1 litre with dH<sub>2</sub>O and stored at room temperature (RT)

## A.2 Stock solutions for *in-situ*-hybridisation and RNA synthesis

### Alkaline phosphate (AP) buffer

5 ml 1M Tris (pH 9.5)  
2.5 ml 1M MgCl<sub>2</sub>  
1 ml 5M NaCl  
50 µl Tween-20  
Adjusted to 50 ml with dH<sub>2</sub>O  
50 ml AP buffer + 500 µl levamisole (24 mg/ml)

**Anti-digoxigenin antibody, AP-coupled** (Roche 11093274910): Store at 4°C

**BCIP, 50 mg/ml** (Roche 11383221001): Store at -20°C

### Bleaching solution

8.30 ml dH<sub>2</sub>O  
0.5 ml 20 x SSC  
0.70 ml H<sub>2</sub>O<sub>2</sub> (30%)  
0.5 ml formamide

### Boehringer Blocking Reagent (BBR), 10%

10 g Boehringer Blocking Reagent (Roche 11096176)  
Adjusted to 100 ml with 1 x MAB  
Stirred and heated to dissolve  
Autoclaved and made into 50ml aliquots  
Stored at -20°C

### DEPC-H<sub>2</sub>O

Added 1 ml DEPC (Diethylpyrocarbonate) to 1 litre dH<sub>2</sub>O  
Mixed thoroughly and left to evaporate under fume hood overnight (at least 1 hour)  
Autoclaved to destroy DEPC and stored at RT

### EDTA, 0.5M

73.05 g EDTA (Ethylenediaminetetraacetate)  
or  
93.05 g Disodium EDTA\*2 H<sub>2</sub>O  
Dissolved in 400 ml dH<sub>2</sub>O and adjusted pH to roughly pH 8.0 with NaOH pellets  
(will only go into solution at pH > 7)  
Adjusted to 500 ml with dH<sub>2</sub>O  
Added 0.5 ml DEPC  
Mixed thoroughly and left to evaporate under fume hood overnight (at least 1 hour)  
Autoclaved and stored at RT

### EGTA, 0.2M

7.61g EGTA  
Dissolved in 70 ml dH<sub>2</sub>O and adjusted pH to roughly pH 8.0 with NaOH pellets (will  
only go into solution at pH > 7)  
Adjusted to 100 ml with dH<sub>2</sub>O  
Added 0.1 ml DEPC  
Mixed thoroughly and left to evaporate under fume hood overnight (at least 1 hour)  
Autoclaved and stored at RT

### HIGS (heat inactivated goat serum)

Normal goat serum  
Heated for 30 minutes at 60°C

Stored at 4°C

#### **Hybridisation buffer**

Prepared in 50 ml falcon tubes  
25 ml Formamide (Roche 11814320001)  
12.5 ml 20 x SSC  
50 mg Torula yeast RNA (Roche 10109223001)  
5 mg Heparin  
1ml 50 x Denhart's solution (prepared with DEPC-H<sub>2</sub>O)  
50 µl Tween 20  
50 mg CHAPS  
1 ml 0.5M EDTA  
Adjusted to 50 ml with DEPC-H<sub>2</sub>O  
Vortexed until precipitate went into solution  
Stored at -20°C

#### **Levamisole, 100 x (24mg/ml)**

1.2 g Levamisole  
50 ml dH<sub>2</sub>O  
Dispensed into 500 µl aliquots  
Stored at -20°C

#### **MAB, 10x**

58 g Maleic acid  
43.8 g NaCl  
Dissolved in 400 ml dH<sub>2</sub>O and adjusted pH to 7.5 with NaOH pellets (roughly 39.5 g)  
Adjusted to 500 ml with dH<sub>2</sub>O  
Autoclaved and stored at 4°C

#### **MEMFA**

79 ml DEPC-H<sub>2</sub>O  
10 ml 1 M MOPS  
1 ml 200 mM EGTA  
100 µl 1M MgSO<sub>4</sub>  
10 ml Formaldehyde solution

#### **MgCl<sub>2</sub>, 1M**

20.33 g MgCl<sub>2</sub>\*6 H<sub>2</sub>O  
Adjusted to 100 ml with dH<sub>2</sub>O  
Autoclaved and stored at 4°C

#### **MgSO<sub>4</sub>, 1M**

12.04 g MgSO<sub>4</sub>  
Adjusted to 100 ml with dH<sub>2</sub>O  
Added 0.1 ml DEPC  
Mixed thoroughly and left to evaporate under fume hood overnight (at least 1 hour)  
Autoclaved and stored at RT

#### **MOPS, 1M**

104.65g MOPS  
10.25 g NaOAc (sodium acetate)  
50 ml 0.5 M EDTA  
Dissolved in 300 ml dH<sub>2</sub>O and adjusted pH to 7.0 with NaOH pellets  
Adjusted to 500 ml with DEPC-H<sub>2</sub>O and stored at 4°C

**NaCl, 5M**

146.1 g NaCl  
Adjusted to 500 ml with DEPC-H<sub>2</sub>O  
Autoclaved and stored at RT

**NaOAc (sodium acetate), 3M**

123.04 g NaOAc  
Dissolved in 400 ml dH<sub>2</sub>O and adjusted pH to 5.0 with glacial acetic acid  
Adjusted to 500 ml with dH<sub>2</sub>O  
Added 0.5 ml DEPC  
Mixed thoroughly and left to evaporate under fume hood overnight (at least 1 hour)  
Autoclaved and stored at RT

**NBT, 100 mg/ml** (Roche 11383213001): store at -20°C

**NH<sub>4</sub>OAc, 10M**

77g NH<sub>4</sub>OAc (ammonium acetate)  
Dissolved in 25ml DEPC-H<sub>2</sub>O  
Adjusted to 100 ml with DEPC-H<sub>2</sub>O

**Phosphate buffer (PB), 0.1M**

2.63g NaH<sub>2</sub>PO<sub>4</sub>\*H<sub>2</sub>O  
21.65g Na<sub>2</sub>HPO<sub>4</sub>\*7H<sub>2</sub>O  
Dissolved in 800 ml dH<sub>2</sub>O and adjusted pH to 7.4  
Adjusted to 1 litre with dH<sub>2</sub>O  
Autoclaved and stored at RT

**Phosphate buffered saline (PBS), 10x**

80 g NaCl  
2 g KCl  
14.4 g Na<sub>2</sub>HPO<sub>4</sub> (or 26.4 g Na<sub>2</sub>HPO<sub>4</sub>\*7 H<sub>2</sub>O)  
2.4 g KH<sub>2</sub>PO<sub>4</sub>  
Dissolved in 800 ml dH<sub>2</sub>O and adjusted pH to 7.4  
Adjusted to 1 litre with dH<sub>2</sub>O  
Add 1 ml DEPC  
Mixed thoroughly and left to evaporate under fume hood overnight (at least 1 hour)  
Autoclaved and stored at RT

**Paraformaldehyde (PFA), 4%**

20 g PFA  
500 ml 0.1M PB (pH 7.4)  
Heated to approximately 60°C, stirring constantly  
Filtered and dispensed in 15-50 ml aliquots  
Stored at -20°C

**Potassium Ferricyanide, 1M (red)**

3.3g Potassium Ferricyanide  
10 ml dH<sub>2</sub>O  
Stored at 4°C

**Potassium Ferrocyanide, 1M (yellow)**

4.2g Potassium Ferrocyanide  
10 ml dH<sub>2</sub>O  
Stored at 4 °C

**Proteinase K, 25mg/ml**

25 mg Proteinase K (Roche 03115836001)  
1 ml DEPC-H<sub>2</sub>O  
Dispensed into 20 µl aliquots  
Stored at -20°C

**RNA digoxigenin labeling kit:** SP6/T7 (Roche 11175025910): store at -20°C

**RNase A, 2mg/ml**

25 mg RNaseA (Roche 10109142001)  
12.5 ml dH<sub>2</sub>O  
Dispensed into 100 µl aliquots  
Stored at -20°C

**RNase T1** (Roche 10109193001): store at 4°C

**SSC, 20 x**

87.65 g NaCl  
44.1g Nacitrate\*2 H<sub>2</sub>O (trisodium citrate)  
Dissolved in 400 ml dH<sub>2</sub>O and adjusted pH to 7.0  
Adjusted to 500 ml with dH<sub>2</sub>O  
Added 0.5 ml DEPC  
Mixed thoroughly and left to evaporate under fume hood overnight (at least 1 hour)  
Autoclaved and stored at RT

**TEA, 1M**

66.7 ml 100% TEA (triethanolamine) solution (7.5M)  
433.3 ml DEPC-H<sub>2</sub>O  
Store at 4°C

**Tris, 1M**

60.55 g Tris base  
Dissolved in 400 ml dH<sub>2</sub>O and adjusted pH to 9.5  
Adjusted to 500 ml with dH<sub>2</sub>O  
Autoclaved and stored at RT

**X-Gal, 50 x (50 mg/ml)**

0.5 g X-Gal  
10 ml Dimethylformamide  
Dispensed into 100 µl aliquots  
Stored at -20°C

**X-Gal staining solution**

5 ml 0.1M PB  
250 µl 1M Potassium Ferricyanide  
250 µl 1M Potassium Ferrocyanide  
10 µl MgCl<sub>2</sub>  
100 µl X-Gal (50 x in DMF)

# Appendix B. Primer list

## B.1 Primer sequences and PCR conditions

**Table B.1** Primer sequences with modifications

Primer	Sequence	Modifications	Annealing temp
XI.Tbx6_F.ClaI	<b>ATCGATAATGTA</b> CTGACCCCCAGCAG	5' <b>ClaI</b> site	65°C
XI.Tbx6_R.EcoRI	<b>GAATT</b> C GCTGTACCTGCCAATCAATAGT	5' <b>EcoRI</b> site	65°C
XI-Sox2-qPCR-F	TGATGCAAGAGCAGCTTGGAA		55°C
XI-Sox2-qPCR-R	GCGTTGTGCGCTTGAG		55°C
XI-Sox2-qPCR-P	ACCCCCAACACCCG	5' FAM 3'TQ2	55°C
XI-Hes5-qPCR-F	TAAGAAAAGCCGGTGGTGGAG		55°C
XI-Hes5-qPCR-R	CCAGCAACATTGGAGCTG		55°C
XI-Hes5-qPCR-P	AGACGGGACC GGATAAACAGCAGCAT	5' FAM 3'TQ2	55°C
XI-Is12-qPCR-F	GCAGCAGCAACTGAGTGAC		55°C
XI-Is12-qPCR-R	GCTGTCATGCCGAATTGGAC		55°C
XI-Is12-qPCR-P	CCTGCAAGGCTTGACAGGAACACCAA	5' FAM 3'TQ2	55°C
XI-Hes9-qPCR-F	GCTGACATCCTGGAGGTG		55°C
XI-Hes9-qPCR-R	TGGGCTGGATGATTGTGCATA		55°C
XI-Hes9-qPCR-P	AGCTTCTGCAGCAGCTAATGGCTCCA	5' FAM 3'TQ2	55°C
XI-Tbx15-qPCR-F	TGACCGCCTATCAGAACAG		55°C
XI-Tbx15-qPCR-R	TTCCTCCCAGAGTCTCGGAA		55°C
XI-Tbx15-qPCR-P	ACTCGGGCTGAAGATTGACAGGAACCCGT	5' FAM 3'TQ2	55°C
XI-Ngn1-qPCR-F	GAGGACTCCTCAGCATGCA		55°C
XI-Ngn1-qPCR-R	CCTGGCACTTCTCAGGAGTT		55°C
XI-Ngn1-qPCR-P	TCCCTCTCCTCTGGACACATGACATCCC	5' FAM 3'TQ2	55°C
XI-Gfi1-qPCR-F	CCACACCACATGGACTCGAA		55°C
XI-Gfi1-qPCR-R	TTGCTCTAAGCTGACCGCAT		55°C
XI-Gfi1-qPCR-P	ACGAAGGTCTCACAGTGGTACAAGGCCA	5' FAM 3'TQ2	55°C
XI-Smn2-qPCR-F	TGGGCAGTAGCTGATCTCC		55°C
XI-Smn2-qPCR-R	GAAGATTCCATTGCCCTTGC		55°C
XI-Smn2-qPCR-P	AGCGGATATCACACTGGCTACTACCTGGG	5' FAM 3'TQ2	55°C
XI-Atoh1-qPCR-F	AAGACAGCAAGACATGCCA		55°C
XI-Atoh1-qPCR-R	TGGTCTCGTCTGAATCGCTG		55°C
XI-Atoh1-qPCR-P	TCATCGGAGTGACGGCGAATTCTCCC	5' FAM 3'TQ2	55°C
XI-Pou4f1.2-qPCR-F	TTCTGTTTAACAACCCACCACTC		55°C
XI-Pou4f1.2-qPCR-R	CTAATAGATCGCTGCTAAGCCTTG		55°C
XI-Pou4f1.2-qPCR-P	TCACCATCATCACCACCACCGCCT	5' FAM 3'TQ2	55°C
XI-Tlx1-qPCR-F	GGTTGTATTGTCAGCAGCGCCTAC		55°C
XI-Tlx1-qPCR-R	TCATGTTGAAGAGCCTGTGAGAGC		55°C
XI-Tlx1-qPCR-P	ACGGGCAATTACAGCAACAGCGGCA	5' FAM 3'TQ2	55°C
Hes5-SP6-F	<b>TAGCAATTAGGTGACACTATAGAA</b> GGCTTCTACCCACGTTGC	5' <b>SP6</b> promoter	55°C
Hes5-T7-R	<b>GATCATAATACGACTCACTATAGGG</b> TTACCAAGGCCTCCAGATTA	5' <b>T7</b> promoter	55°C
Gfi1a-SP6-F	<b>TAGCAATTAGGTGACACTATAGAA</b> GCCTGCCCCTCTGGATG	5' <b>SP6</b> promoter	55°C
Gfi1a-T7-R	<b>GATCATAATACGACTCACTATAGGG</b> GGTATGGTCTGGTATCCGAA	5' <b>T7</b> promoter	55°C
Hes9-SP6-F	<b>TAGCAATTAGGTGACACTATAGAA</b> ATGGCTCCATACTGCTAGC	5' <b>SP6</b> promoter	55°C
Hes9-T7-R	<b>GATCATAATACGACTCACTATAGGG</b> ACCAGGGCTCCAGAGAACT	5' <b>T7</b> promoter	55°C
Tbx15-SP6-F	<b>TAGCAATTAGGTGACACTATAGAA</b> GTTCCACGAAATCGGACTG	5' <b>SP6</b> promoter	55°C
Tbx15-T7-R	<b>GATCATAATACGACTCACTATAGGG</b> ACAGCTGATTCTCCCTGCAG	5' <b>T7</b> promoter	55°C

Primer	Sequence	Modifications	Annealing temp
Ngn1-SP6-F	<b>TAGCAATTTAGGTGACACTATAGAA</b> GACTATGACACCTGCAGCCA	5' <b>SP6</b> promoter	55°C
Ngn1-T7-R	<b>GATCATAATACGACTCACTATAGGG</b> GCTGGAAGTGCAGACTGAGA	5' <b>T7</b> promoter	55°C
Pou4f1.2-SP6-F	<b>TAGCAATTTAGGTGACACTATAGAA</b> ATCAGAACGCCATTGGAGAGC	5' <b>SP6</b> promoter	55°C
Pou4f1.2-T7-R	<b>GATCATAATACGACTCACTATAGGG</b> TCCCTTGCTCTCTGC	5' <b>T7</b> promoter	55°C
Isl2-SP6-F	<b>TAGCAATTTAGGTGACACTATAGAA</b> AGTGGATGTCATCTGCCCT	5' <b>SP6</b> promoter	55°C
Isl2-T7-R	<b>GATCATAATACGACTCACTATAGGG</b> GCTGTCATGCCGAATTGGAC	5' <b>T7</b> promoter	55°C
pCS2+-SP6-F	TGGCGTTCATTGACGTA		55°C
pCS2+-T7-R	AAGCAATAGCATCACAAATTCA		55°C

# Appendix C. Script

## C.1 General bioinformatic pipeline

```
#####
### Bioinformatic Pipeline #####
#####

# 2.5.1 Run fastqc to visually inspect all sequencing results
# Run as perl script:

#!/usr/bin/perl
use warnings;
use strict;
use feature qw(say);

open (FILES, "ls *.txt |");

while (<FILES>) {
    chomp;
    my ($file) = (split)[0];
    say "Parsing $file...";
    system ("fastq_quality_trimmer -t 30 -l 75 -i $file -o $file.fastq");
}

# 2.5.1 Run Trimmomatic to quality filter reads:

java -classpath /path/to/Trimmomatic/trimmomatic-0.25.jar
org.usadellab.trimmomatic.TrimmomaticPE
-threads 12 \
-phred33 \
<pe_1> <pe_2> <paired_output_1> <unpaired_output_1> <paired_output_2>
<unpaired_output_2> \
ILLUMINACLIP:<filter_set> \
LEADING:3 TRAILING:3 SLIDINGWINDOW:4:15 HEADCROP:12 MINLEN:36

# 2.5.2 Build bowtie index

bowtie2-build <genome.fasta>

# 2.5.2 Tophat2 on each set of paired reads

tophat2 -p 12 -r 250 -N 3 -o <output_dir> <bowtie-index> <pe_1> <pe_2>
cd <output_dir>
samtools flagstat accepted_hits.bam > stats.txt
mv accepted_hits.bam <condition.rep.bam>

# 2.5.2 Assemble transcripts and abundance estimation with Cufflinks

cufflinks -p 12 -b <bowtie-index> \
-o <output_dir> <condition.rep.bam>

# 2.5.2 Merge assemblies using cuffmerge

ls -1 <path/to/cufflinks_output_condition_1/transcripts.gtf> \
<condition_n/transcripts.gtf> > assemblies.txt
cuffmerge assemblies.txt

# 2.5.3 Differential expression with Cuffdiff

cd <cufflinks_output_dir> \
cuffdiff -p 12 \
-o <cufflinks_output_dir/diff_out> \
-b <bowtie-index/.fasta> \
-L <L1,L2> <merged.gtf> <L1_rep1.bam>,<L1_rep2.bam> <L2_rep1.bam>,<L2_rep2.bam>

# 2.5.4 Estimating variance between biological replicates
# run pearsons.pl
# Run ops package in R:

library(ops)
fpkm<-read.table("output/from/pearsons.pl")
fpkm1<-$fpkm[1]
fpkm2<-$fpkm[2]
x=cbind(fpkm1,fpkm2)
p=findP(x)$maxIQR
```

```

y=x^p

# 2.5.5 Generating a scatterplot to visualise FPKM distribution
# run merger.pl
# run scatterplot.grid.R

# 2.5.6 Annotating transcript models
# run transcripts.pl
# run de.genes.pl

# 2.5.6 Annotating transcript models
# BLAST output ('de_transcripts_.fa') against Xenopus mRNA database:

blastn -db <path_to_Xenopus_DB> \
-query <de_transcripts_.fa> \
-num_threads 12 \
-evalue '0.00001' \
-perc_identity 80 \
-outfmt "6 qseqid pident sseqid" \
-task blastn \
-max_target_seqs 1 | sort -u -k1,1 > <output.txt>

# 2.5.6 Annotating transcript models
# run annotator.pl

# 2.5.7 Gene Enrichment Analysis
# run enrichment.pl

# perform chi squared test using output from 'enrichment.pl' ( e.g. use
# http://www.socscistatistics.com/tests/chisquare/)

# 2.5.8 Finding co-differentially expressed genes
# run co-reg.pl

# 2.5.9 Gene ontology analysis on discrete gene sets
# run sets.pl

# Blast output (e.g. 'setA') against Human Uniprot DB

blastx -db <path_to_Human_UniDB> \
-query <setA.fa> \
-num_threads 12 \
-evalue '0.001' \
-outfmt "6 qseqid pident sseqid" \
-max_target_seqs 1 | sort -u -k1,1 > <output.txt>

```

## C.2 Pearson's correlation

```
#####
### Pearson's ###
#####

#!/usr/bin/perl
use warnings;
use strict;
use feature qw(say);
use Data::Dumper;
$Data::Dumper::Sortkeys = 1;

# This script will read in fpkm values from cuffdiff output file
'genes.read_group_tracking' and open a .csv file containing:

# FPKM rep 1    FPKM rep 2
# This can then be used as input for a standard Pearson's correlation (e.g. in R)

# Run as perl pearsons.pl

# Will open output in excel

open my $in, '<',
"/Users/Nick/Documents/Bioinformatics/Data/eya1/diff_out/genes.read_group_tracking" or die
"usage perl pearsons.pl\n";

my %data;
while (<$in>){
    chomp;
    next unless /^XLOC/;
    my @split = split(/\t/);
    $data{$split[0]}{$split[1]}{$split[2]} = $split[6];
}

open my $out, '>', 'output.csv';
print $out "fpkm1,fpkm2\n";

my (%rep_count, $log_trans_fpkm, %filter, $fpkm);

for my $xloc (sort keys %data){
    for my $condition (keys $data{$xloc}){
        for my $replicate (sort keys $data{$xloc}{$condition}){
            $fpkm = $data{$xloc}{$condition}{$replicate};
            next if $fpkm == 0;
            $rep_count{$xloc}{$condition}++;
            $filter{$xloc}{$condition}{$replicate} = $fpkm if $replicate == 0 and
$rep_count{$xloc}{$condition} == 1 and $condition eq 'Echx';
            $filter{$xloc}{$condition}{$replicate} = $fpkm if $replicate == 1 and
$rep_count{$xloc}{$condition} == 2 and $condition eq 'Echx';
        }

        delete $filter{$xloc} if $fpkm == 0 or $fpkm > 5000;
    }
}

# Print out log2 fpkm to excel
for my $id (sort keys %filter){
    for my $condition (keys $filter{$id}){
        for my $replicate (sort keys $filter{$id}{$condition}){
            my $fpkm = log2($filter{$id}{$condition}{$replicate});
            print $out "$fpkm," if $replicate == 0 and $condition eq 'Cchxdex';
            print $out "$fpkm" if $replicate == 1 and $condition eq 'Cchxdex';
        }

        print $out "\n";
    }
}

sub log2 {
    my $n = shift;
    $n = $n + 1;

    return log($n)/log(2);
}
system("open output.csv");

```

### C.3 Merger

```
#####
### Merger ####
#####

#!/usr/bin/perl
use warnings;
use strict;
use Data::Dumper;
$Data::Dumper::Sortkeys = 1;
use autodie;

# This script will annotate 'gene_exp.diff' information on the direction of change
# To be used as input for scatterplot
# Run as perl merger.pl

open my $in, '<', 'gene_exp.diff';

my (%data, $sig);

while (<$in>){
    chomp;
    next unless /^XLOC/;
    my @split = split(/\t/);
    $data{$split[0]} = $split[13];
}

open my $fpkm, '<', 'new.txt';
open my $out, '>', 'merged.txt';

print $gene_names "gene\tcontrol\texpt\n";

while (<$fpkm>){
    chomp;
    my @split = split(/\t/);
    my ($con, $exp) = ($split[9], $split[13]);
    if ($data{$split[0]}) {
        if ($data{$split[0]} eq 'yes' and $con < $exp){
            print $out "$_\t$data{$split[0]}\tup\n";
        }
        elsif ($data{$split[0]} eq 'yes' and $con > $exp){
            print $out "$_\t$data{$split[0]}\tdown\n";
        }
        elsif ($data{$split[0]} eq 'no'){
            print $out "$_\t$data{$split[0]}\tno\n";
        }
    }
}
```

## C.4 Scatterplot

```
#####
### Scatterplot.grid.R #####
#####

# Either copy and paste entire script into R, or run script from R:

# $ R > source("scatterplot.grid.R")

# Load packages
library(reshape)
library(ggplot2)
require(gridExtra)

# Specify output path
pdf(file="/Users/Nick/Desktop/grid_FPKM_scatter.pdf", width=10, height=6)

merged<-read.table("/Users/Nick/Documents/Bioinformatics/Data/Merged_reps/Plots/merged.txt",header=FALSE, row.names=1)

sse<-read.table("/Users/Nick/Documents/Bioinformatics/Data/Six1+Six1-Eya1/Plots/merged.txt",header=FALSE, row.names=1)

ese<-read.table("/Users/Nick/Documents/Bioinformatics/Data/Eya1+Six1-Eya1/Plots/merged.txt",header=FALSE, row.names=1)

# Six1-Eya1 data
con<-log2((merged[,9])+1)
exp<-log2((merged[,13])+1)
sig<-merged[,17]
dir<-merged[,18]

# Six1 data
sse_con<-log2((sse[,9])+1)
sse_exp<-log2((sse[,13])+1)
sse_sig<-sse[,17]
sse_dir<-sse[,18]

# Eya1 data
ese_con<-log2((ese[,9])+1)
ese_exp<-log2((ese[,13])+1)
ese_sig<-ese[,17]
ese_dir<-ese[,18]

# Set plot variables for each condition
ese_plot<-ggplot(ese, aes(ese_con, ese_exp , colour=ese_dir, size=ese_sig) ) +
  xlim(0, 15) +
  ylim(0, 15) +
  xlab("Control log(2) (1+FPKM)") +
  ylab("Eya1+Six1-Eya1 overexpression log(2) (1+FPKM)") +
  # Shades for density of points
  geom_point(alpha = 0.6) +
  # point size
  scale_size_manual(values=c(I(1), I(1.8))) +
  scale_colour_manual(values=c('no'="grey12", 'down'="red4", 'up'="green4"))

sse_plot<-ggplot(sse, aes(sse_con, sse_exp , colour=sse_dir, size=sse_sig) ) +
  xlim(0, 15) +
  ylim(0, 15) +
  xlab("Control log(2) (1+FPKM)") +
  ylab("Six1+Six1-Eya1 overexpression log(2) (1+FPKM)") +
  # Shades for density of points
  geom_point(alpha = 0.6) +
  # point size
  scale_size_manual(values=c(I(1), I(1.8))) +
  scale_colour_manual(values=c('no'="grey12", 'down'="red4", 'up'="green4"))

merged_plot<-ggplot(merged, aes(con, exp , colour=dir, size=sig) ) +
  xlim(0, 15) +
  ylim(0, 15) +
  xlab("Control log(2) (1+FPKM)") +
  ylab("Eya1+Six1+Six1-Eya1 overexpression log(2) (1+FPKM)") +
  # Shades for density of points
  geom_point(alpha = 0.6) +
  # point size
  scale_size_manual(values=c(I(1), I(1.8))) +
  scale_colour_manual(values=c('no'="grey12", 'down'="red4", 'up'="green4"))
```

```
# Define legends
g_legend<-function(a.gplot){
  tmp <- ggplot_gtable(ggplot_build(a.gplot))
  leg <- which(sapply(tmp$grobs, function(x) x$name) == "guide-box")
  legend <- tmp$grobs[[leg]]
  return(legend)}

mylegend<-g_legend(merged_plot)

# Arrange scatterplots in a grid
grid.arrange(arrangeGrob(merged_plot + theme(legend.position="none"),
  sse_plot + theme(legend.position="none"),
  ese_plot + theme(legend.position="none"), nrow=1))

dev.off()
```

## C.5 Transcripts

```
#####
### Transcripts ###
#####

#!/usr/bin/perl
use warnings;
use strict;
use Data::Dumper;
$Data::Dumper::Sortkeys = 1;

# This script will read in exonic positions from cufflinks output file 'merged.gtf'
# and relate then to genomic positions. The program then prints out

# a) All transcript variants for a given gene
# b) All exons in each gene
# c) The longest transcript variant for all assembled genes (numbered by transcript
# variant)

# Run as perl transcripts.pl in the same directory as 'merged.gtf'

open my $merged, '<', 'merged.gtf' or die "usage perl transcripts.pl\nIs 'merged.gtf' in
the current directory?\n";

my (%gene, $scaf, $start, $stop, $xloc, $tcons, $exon, $length, $info);

while(<$merged>){
    my @split = split('\t');
    ($scaf) = ">$split[0]";
    ($start) = $split[3];
    ($stop) = $split[4];
    ($info) = $split[8];
    ($xloc, $tcons, $exon) = $info =~ /gene_id "(.+?)"; transcript_id "(.+?)"; exon_number
"(\d+)";/g;
    $gene{$xloc}{$tcons}{$exon} = [$start, $stop, $scaf];
}

open my $genome, '<', '/Laevis_7/Xl7.fa' or die $!;
my (%seq, $head);

while (<$genome>) {
    chomp;
    $head = $_ if /^>/;
    $seq{$head} = $_ if /^[A-Z]/;
}

open my $transcript_variant, '>', 'transcript_variants.txt' or die $!;
open my $exons, '>', 'transcripts_by_exon.txt' or die $!;
open my $transcripts, '>', 'transcripts.txt' or die $!;

my ($exon_seq, %transcript, %seq_length, $exon_length);
my %most_exons;
my $splice_var;

for my $xloc (sort keys %gene){
    my $seen = 0;
    for my $tcons (sort keys $gene{$xloc}){
        $seen++;
        $splice_var = "$xloc.$seen";
        my $exon_count = 0;
        for my $exon (sort { $a <=> $b } keys $gene{$xloc}{$tcons}){
            my ($start, $stop, $scaf) = @{$gene{$xloc}{$tcons}{$exon}};
            $exon_count++;
            $start = ($start - 1);
            $exon_length = $stop - $start;
            $exon_seq = substr $seq{$scaf}, $start, $exon_length;
            print $exons ">$xloc.$seen\_$exon\n$exon_seq\n";
            $transcript{$tcons} .= $exon_seq;
            if ( (not exists $most_exons{$xloc})
                or
                ($most_exons{$xloc}[0] < $exon_count) ){
                $most_exons{$xloc} = [$exon_count, $splice_var,
                $transcript{$tcons}];
            }
        }
    }
}
```

```
        print $transcript_variant ">$xloc.$seen\n$transcript{$tcons}\n";
    }

for my $gene (sort keys %most_exons){
    my ($exon_count, $splice_var, $sequence) = @{$most_exons{$gene}};
    print $transcripts ">$splice_var\n$sequence\n";
}
```

## C.6 DE genes

```
#####
### De.genes ###
#####

#!/usr/bin/perl
use strict;
use warnings;
use Data::Dumper;
$Data::Dumper::Sortkeys = 1;

# This script will attach differential expression info from the cufdiff output file
'gene_exp.diff' to transcripts assembled in 'transcripts.pl'

# Run as 'perl de.genes.pl'

open my $in, '<', 'diff_out/gene_exp.diff' or die "usage perl de.genes.pl\nIs 'diff_out'
in the current directory?\n";

my %diff_out;

while (<$in>){
    chomp;
    next if /test_id/;
    my @split = split(/\t/);
    my $xloc = $split[0];
    my $scaffold = $split[3];
    my ($control_val, $expt_val, $change_val) = @split[7,8,9];
    $diff_out{$xloc} = [$scaffold, $control_val, $expt_val, $change_val];
}

open my $hits, '<', 'transcripts.txt' or die "'transcripts.txt' must be in the
current directory\n";

my (%genes, $xloc);

while (<$hits>) {
    chomp;
    ($xloc) = /(XL0C_\d+)/ if /^>/;
    $genes{$xloc} = $_ if /^[A-Z]/;
}

open my $de_genes_print, '>', 'de_transcripts_new.txt' or die "$!\\n";
foreach my $key (sort keys %diff_out){
    my ($scaffold, $control_val, $expt_val, $change_val) = @{$diff_out{$key}};
    print $de_genes_print
">$key\_$control_val:$expt_val,_Change:$change_val\\n$genes{$key}\\n" if exists
$genes{$xloc};
}
```

## C.7 Annotator

```
#####
### Annotator ####
#####

#!/usr/bin/perl
use strict;
use warnings;
use Data::Dumper;
$Data::Dumper::Sortkeys = 1;

# This script takes blast results as input and annotates all transcripts with best
# hit and user specified condition name
# It also counts the number of genes that return no hit. Output as two .txt files
# which can be blasted against a different database (or with a lower E-value) and added to
# the blast output.

# Run as perl annotator.pl <Xenopus blast output> <condition> <de_genes.txt>

if ($#ARGV != 2){
    usage();
    exit
}

my $input1 = $ARGV[0];

open my $blast, '<', $input1 or die $!;

my $condition = $ARGV[1];

# edit.input will trim input, removing genes that don't hit the database

my $editedblast = "$condition.$input1";

open my $unknowns_removed, '>', $editedblast or die "Can't write to $editedblast: $!";

# Split output from blast and outout edited input (with non-hits removed)
my $blast_count = 0;
my (%blasthits, @test, @blastID);

while (<$blast>) {
    chomp;
    $blast_count++;
    my @genematch = ( $_ =~ /(XLOC_\d+)/g );
    push @blastID, @genematch;
    print $unknowns_removed "$condition\t$_\n";
}

push @{$blasthits{$blastID[$_]}} , [ $blastID[$_] ] for 0 .. $#blastID;

# read in de_genes
my $de_seqs = $ARGV[2];

open my $de_genes, '<', $de_seqs or die "Can't open '$de_seqs'";

my $gene_count = 0;
my (@gene, @head, @seq);

while (<$de_genes>) {
    chomp;
    my @match = ( $_ =~ /(XLOC_\d+)/g );
    push @gene, @match;
    push @head, $_ if /^>/;
    push @seq, $_ if /^[A-Z]/;
    $gene_count++ if />/;
}
print "$gene_count queries\n";
print "$blast_count hits in $input1 \n";
my %genes;

push @{$genes{ $gene[$_] }} , [ $head[$_], $seq[$_] ] for 0 .. $#gene;

my $nohit = 'unknown_genes.txt';

open my $unknown_print, '>', $nohit or die "Can't write to $nohit: $!";
```

```

# Output to 'unknown_genes.txt' all the de_genes that return no blast hit
my $i = 0;
for my $key (sort keys %genes) {
    unless (exists $blasthits{$key}) {
        $i++;
        for my $part (@ { $genes{$key} } ) {
            my ($head, $seq) = @$part;
            print $unknown_print "$head\n$seq\n";
        }
    }
}

print "$i genes that return no blast hit\n";

sub usage {
    print "\n*****ANNOTATOR*****\n\n";
    print "Usage: annotator.pl <Xenopus blast output> <condition> <de_genes.txt>\n\n";
    print "Takes BLAST results as input and finds all genes that return no hit (not
present\n";
    print "in the blast output but present in the de_genes).\n\n";
    print "Outputs: 'unknown_genes.txt' to BLAST against a different database\n";
    print "Outputs: 'edit.<input>.txt' to be used as input for 'co-reg.pl'\n\n";
    print "Nick Riddiford\n";
    print "August 2013\n\n";
}

```

## C.8 Enrichment

```
#####
### Enrichment ####
#####

#!/usr/bin/perl
use warnings;
use strict;
use Data::Dumper;
$Data::Dumper::Sortkeys = 1;
use feature 'say';
use POSIX;

# This script will read in differentially expressed genes (output from 'blast.pl')
# for two conditions and output the top 10% for each list, as well as input data for a
# contingency table

# Run as perl enrichment.pl <blast.file1> <blast.file2>

if ($#ARGV < 0) {
    die "usage enrichment.pl <blast.file1> <blast.file2>";
    exit;
}

my ($g1, $g2);
my $p = 10;
my ($r1, $r2);
my (%top1, %top2);
my $threshold = 0.5;
my (%duplicates, %gene_list, %change);
my $fileno;
my $all_genes;
blast_extractor($_) foreach @ARGV;
sub blast_extractor {
    my $file = shift;
    open my $output, '<', $file or die "Can't read file '$file' [$!]\n";
    $fileno++;
    my @_change;
    while (<$output>) {
        chomp;
        next if /^$/;
        my ($xloc) = /(XLOC_\d+)/;
        my ($change) = /Change:(-?\d+\.\d+|-?inf|-?\d+)/;
        my ($container) = /XLOC_\d+_(_+:+)_Change/g;
        my ($con_val, $expt_val) = split('/: ', $container);
        my @split = split(/\t/);
        my ($condition, $percent_id, $gene) = (@split[0], $split[2], $split[3]);
        $gene =~ s/_/ /g;
        $gene =~ s/_//g;
        my @small_split = split(/\|/, $gene);
        my $small_gene = $small_split[4];
        my ($count) = $duplicates{$small_gene}++;
        $gene_list{$small_gene}{$count} = [$xloc, $con_val, $expt_val, $change];
    }
}

my (%filtered, %expt_count, %pos_count, %neg_count, %con_sum, %expt_sum,
%genes_by_count, %all, @xlocs);
my $change_sum;
my $gene;

foreach $gene (sort keys %gene_list) {
    foreach my $count (sort keys $gene_list{$gene}) {
        my ($xloc, $con_val, $expt_val, $change_val) =
 @{$gene_list{$gene}{$count}};
        $count];
    }
}

# Remove the occurrences of 'inf' log(2) values by adding 0.001 to
# each raw value of 0
$con_val = ($con_val + 0.001) if $con_val == 0;
$expt_val = ($expt_val + 0.001) if $expt_val == 0;
# Foreach gene/condition sum up all the raw values
$con_sum{$gene} += $con_val;
$expt_sum{$gene} += $expt_val;
$con_val = $con_sum{$gene};
$expt_val = $expt_sum{$gene};
my $raw_val = $expt_val/$con_val;
```

```

$change_sum = eval sprintf('%.6f', (log($raw_val)/ log(2)));
$genes_by_count{$gene} = [$xloc, $con_val, $expt_val, $change_sum,
    delete $genes_by_count{$gene} if abs $change_sum < $threshold or
$genes_by_count{$gene}[2] < 1;
}

}

for my $g (sort keys %genes_by_count) {
    my ($x, $c, $e, $fc, $co) = @{$genes_by_count{$g}};
    push @change, [$g, $fc];
}

my @top;
my $top10;
my $counter = 0;

for (sort { $b->[1] <=> $a->[1] } @change){
    $counter++;
    $top10 = int(10/100 * (#change));
    push @top, @_ [0] if $counter < $top10;
}

$r1 = $top10 if $fileno == 1;
$r2 = $top10 if $fileno == 2;
$g1 = $#top if $fileno == 1;
$g2 = $#top if $fileno == 2;
say "$top10 genes in top $p% of $file";
say "$counter genes in $file";
$all_genes += $counter;
$all_genes -= $top10;
$top10 = ();
if ($fileno == 1) {$top1{$\_} = 1 foreach @top}
if ($fileno == 2) {$top2{$\_} = 2 foreach @top}

@change=();

delete $genes_by_count{$_} for keys %genes_by_count;
delete $gene_list{$_} for keys %gene_list;
}

my $overlap = 0;
my (@overlap, %overlap);

foreach my $gene (keys %top1){
    if (exists $top2{$gene}){
        $overlap++;
        push @overlap, $gene;
        print "$gene\n";
        $overlap{$gene} = 1;
    }
}

say "\n'a' -> Genes in top $p% for condition1 & condition2 = $overlap";
my $not_overlap1 = ceil($r1 - $overlap);
my $not_overlap2 = ceil($r2 - $overlap);
my $non_overlap = ceil(($g1 + $g2)/2);
say "'b' -> Genes in the top $p% for condition2, but not in the top $p% for
condition1 = $not_overlap1";
say "'c' -> Genes in the top $p% for condition1, but not in the top $p% for
condition2 = $not_overlap2";
say "'d' -> Genes in neither the top $p% for condion1 nor condition2 = $all_genes ";

print "-----\n";
print "|      | |\n";
print "| overlap | $not_overlap2 |\n";
print "|      | |\n";
print "-----\n";
print "|      | |\n";
print "|$not_overlap1 | $all_genes |\n";
print "|      | |\n";
print "-----\n";

```

## C.9 Co-reg

```
#####
#### Co-reg ####
#####

#!/usr/bin/perl
use warnings;
use strict;
use Data::Dumper;
$Data::Dumper::Sortkeys = 1;
use feature qw(say);
use autodie;

# Run `perl co-reg.pl` for usage statementnt

# This program will take N blast hit results and output those genes which are found
# to be overlapping in $n conditions.

# Run as follows: `perl co-reg.pl control.txt six.txt six-eya.txt eya.txt` where
# blast output has the following, tab delimited format:
# gene XLOC_control.fpkm:expt.fpkm,_Change:FC_val %id blast hit

# Always run 'control' condition first

if ($#ARGV < 0) {
    usage();
    exit; }

# set a flag for 'silent run' which will not read in transcriptomes or open .csv
# file behind script
my $flag1;
if ($ARGV[0] eq '-s'){
    $flag1 = shift @ARGV;
}
else {$flag1 = 1; }

my (%duplicates, %gene_list, %change, %seen, %seen_all);

print "Opening blast ouput foreach condition...\n";

blast_extractor($_) foreach @ARGV;# To extract data from blast output

# Select for up or down regulation
print "Enter direction: ";
chomp(my $direction = <STDIN>);

# Select the condition being run to read in corresponding transcriptome
my $seqs_to_print;
print "Enter sequences to print (six1 = 0 eya1 = 1 six1-eya1 = 2): ";
chomp($seqs_to_print = <STDIN>);

my (%filtered, %expt_count, %pos_count, %neg_count, %con_sum, %expt_sum,%genes_by_count,
%all, @xlocs);

my $change_sum;
my $threshold = 1;
my $n = 2;

say "Filtering genes for DE genes...";

my $gene;

foreach $gene (sort keys %gene_list) {
    foreach my $con (sort keys $gene_list{$gene}) {
        $expt_count{$gene}++ unless $con eq 'control';
        foreach my $count (sort keys $gene_list{$gene}{$con} ) {
            my ($xloc, $con_val, $expt_val, $change_val) =
                @{$gene_list{$gene}{$con}}{$count};
            # Remove the occurrences of 'inf' log(2) values by adding 0.001 to each
            # raw value of 0
            $con_val = ($con_val + 0.001) if $con_val == 0;
            $expt_val = ($expt_val + 0.001) if $expt_val == 0;
            # Foreach gene/condition sum up all the raw values
            $con_sum{$gene}{$con} += $con_val;
            $expt_sum{$gene}{$con} += $expt_val;
            $con_val = $con_sum{$gene}{$con};
            $expt_val = $expt_sum{$gene}{$con};
```

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my $raw_val = $expt_val/$con_val;
$change_sum = eval sprintf('%.6f', (log($raw_val)/ log(2)));
# Make a new hash. As the loop cycles though sorted change values, the
# last value will be the highest count (and thus the correct log(2)
# value for that gene/condition). Use this in the next loop to pull out
# only those genes that are all positive or negative...
$genes_by_count{$gene}{$con} = [$xloc, $con_val, $expt_val,$change_sum,
$count];
$all{$gene}{$con} = [$xloc, $con_val, $expt_val, $change_sum, $count];
}

# Only genes with log(2) > 0.5 allowed through
# Only genes with chx+dex FPKM > 1 allowed through
# In upreg run mode, consider genes with chx+dex FPKM > 1 as expressed
# In downreg run mode, consider genes with chx FPKM > 1 as expressed

my $num = 2 if $direction eq 'up';
$num = 1 if $direction eq 'down';
delete $genes_by_count{$gene}{$con} if abs $change_sum < 0.5 or
$genes_by_count{$gene}{$con}[$num] < 1;

# Treat control in the same way
delete $all{$gene}{$con} if $all{$gene}{$con}[$num] < 1 and $con eq 'control';
$pos_count{$gene}++ if $change_sum > 0 and $con ne 'control';
$neg_count{$gene}++ if $change_sum < 0 and $con ne 'control';
}

}

# Experimental conditions only
# Only look for genes that are up or down reg in all conditions

for my $gene (sort keys %genes_by_count) {
    if ( defined $expt_count{$gene} and ( ($pos_count{$gene} or $neg_count{$gene})
    == $expt_count{$gene}) ) {
        next if exists $neg_count{$gene} and $direction eq 'up';
        next if exists $pos_count{$gene} and $direction eq 'down';
        foreach my $con ( sort keys $genes_by_count{$gene} ) {
            next if $con eq 'control';
            $seen{$gene}++ if @{$genes_by_count{$gene}{$con}}[3] > 1;
            foreach my $count ( @{$genes_by_count{$gene}{$con}}[4] ){
                my ($xloc, $con_val, $expt_val, $change_val, $count) =
                @{$genes_by_count{$gene}{$con}};
                # only real duplicates are printed
                $change_val, $count];
                $count = '' unless $count > 0;
                $filtered{$gene}{$con} = [$xloc, $con_val, $expt_val,
                # Put the smallest change value for each gene in new hash,
                as long as it's > 1
                # This means that if while all genes log(2) > 0.5 are
                considered, we need at least 1 of the conditions to have
                log(2) > 1
                if (( (not exists $change{$gene}) || (abs
                $change{$gene} < abs $change_val) ) ) && (abs $change_val
                >= $threshold) ) {
                    }
                }
            }
        }
    }
    $change{$gene} = $change_val;
}

my %control_filt;

for my $gene (sort keys %all) {
    next unless exists $change{$gene};
    foreach my $con (sort keys $all{$gene}) {
        next unless $con eq 'control';
        foreach my $count ( @{$all{$gene}{$con}}[4] ) {
            my ($xloc, $con_val, $expt_val, $change_val, $count) =
            @{$all{$gene}{$con}};
            $count = '' unless $count > 0;
            $control_filt{$gene} = [$con_val, $expt_val, $change_val, $count];
        }
    }
}

```

```

        }

    }

my $overlapping_count = 0;
my $control_seen = 0;
my %control_ratio;
print "Calculating control ratios for DE genes..\n\n";

fraction(%change);

my $sequences;

my (%six_seqs, %eya_seqs, %sixeya1_seqs, $xloc);

# Skip reading in transcriptomes if in silent mode
unless ($flag1 eq '-s'){

open $sequences, '>',
"/Users/nickriddiford/Desktop/$seqs_to_print.$direction.sequences.txt" or die $!;
if ($seqs_to_print == 0){
    print "Opening transcriptome for six1...\n";
    open my $S_transcriptome, '<', '/six1/transcripts.txt' or die $!;
    while (<$S_transcriptome>){
        chomp;
        ($xloc) = />(XLOC_\d+)/./ if /^[^>]$/;
        $six_seqs{$xloc} = $_ if /[^\>]/;
    }
}

if ($seqs_to_print == 1){
    print "Opening transcriptome for eya1...\n";
    open my $E_transcriptome, '<','/eya1/transcripts.txt' or die $!;
    while (<$E_transcriptome>){
        chomp;
        ($xloc) = />(XLOC_\d+)/./ if /^[^>]$/;
        $eya_seqs{$xloc} = $_ if /[^\>]/;
    }
}

if ($seqs_to_print == 2){
    print "Opening transcriptome for six1-eya1...\n";
    open my $SE_transcriptome, '<','/six1-eya1/transcripts.txt' or die $!;
    while (<$SE_transcriptome>){
        chomp;
        ($xloc) = />(XLOC_\d+)/./ if /^[^>]$/;
        $sixeya1_seqs{$xloc} = $_ if /[^\>]/;
    }
}

my $overlapping_genes = 'overlapping_genes.csv';
open my $overlap_print, '>', $overlapping_genes or die "Can't write to
$overlapping_genes: $!";

print $overlap_print "Rank, Gene, Six1 CHX, Six1 CHX+DEX, Log(2), Eya1 CHX, Eya1
CHX+DEX, Log(2), Six1-Eya1 CHX, Six1-Eya1 CHX+DEX, Log(2), Control CHX, Control
CHX+DEX, Log(2)\n";

my ($Sx, $Sc, $Se, $Sch, $Sco, $Ex, $Ec, $Ee, $Ech, $Eco, $SEx, $SEc, $SEe, $SEch,
$SEco);

say "Printing ranked DE gene list to 'overlapping_genes.csv'";
say "Printind DE sequences to '/Users/nickriddiford/Desktop/sequences.txt'\n";

# Output 'out.txt' as input for 'sets.pl'
open my $out, '>', "/Users/nickriddiford/Desktop/$seqs_to_print.$direction.out.txt";

```

```

# Sort first by rank, and second by FC
for my $gene (sort {
    abs $control_ratio{$a}[0] <=> abs $control_ratio{$b}[0]
    or
    abs $control_ratio{$b}[1] <=> abs $control_ratio{$a}[1] }
keys %control_ratio) {

    my ($con_val, $expt_val, $change_val, $count) = @{$control_filt{$gene}} if
exists $control_filt{$gene};
    ($con_val, $expt_val, $change_val, $count) = (0,0,0, '') if not exists
$control_filt{$gene};
    print $overlap_print "$control_ratio{$gene}[0],$gene,";
    print $out "$control_ratio{$gene}[0]\t$gene\t";
    for my $condition (sort { $filtered{$gene}{$a} <=> $filtered{$gene}{$b} })
keys %{$filtered{$gene}} ) {
        $count++ if $count;
        ($Sx, $Sc, $Se, $Sch, $Sco) = @{$filtered{$gene}{'six1'}} if exists
$filtered{$gene}{'six1'};
        ($Ex, $Ec, $Ee, $Ech, $Eco) = @{$filtered{$gene}{'eya1'}} if exists
$filtered{$gene}{'eya1'};
        ($SEx, $SEc, $SEe, $SEch, $SEco) = @{$filtered{$gene}{'six1-eya1'}} if
exists $filtered{$gene}{'six1-eya1'};
    }

    # Input for 'sets.pl'
    print $out "$Sx\t" if exists $filtered{$gene}{'six1'};
    print $out "$Ex\t" if exists $filtered{$gene}{'eya1'};
    print $out "$SEx\t" if exists $filtered{$gene}{'six1-eya1'};
    exists $filtered{$gene}{'six1'} ? print $overlap_print "$Sc,$Se,$Sch," :
print $overlap_print "-,-,-";
    exists $filtered{$gene}{'eya1'} ? print $overlap_print "$Ec,$Ee,$Ech," :
print $overlap_print "-,-,-";
    exists $filtered{$gene}{'six1-eya1'} ? print $overlap_print
"$SEc,$SEe,$SEch," : print $overlap_print "-,-,-";
    print $overlap_print "$con_val,$expt_val,$change_val\n";

    # Input for 'sets.pl'
    exists $filtered{$gene}{'six1'} ? print $out "$Sc\t$Se\t$Sch\t" : print
$out "-\t-\t-\t";
    exists $filtered{$gene}{'eya1'} ? print $out "$Ec\t$Ee\t$Ech\t" : print
$out "-\t-\t-\t";
    exists $filtered{$gene}{'six1-eya1'} ? print $out "$SEc\t$SEe\t$SEch\t" :
print $out "-\t-\t-\t";
    print $out "$con_val\t$expt_val\t$change_val\n";

unless ($flag1 eq '-s'){
    sequence($Sx, $Sc, $Se, $Sch, $Sco, 'six1') if $seqs_to_print == 0;
    sequence($Ex, $Ec, $Ee, $Ech, $Eco, 'eya1') if $seqs_to_print == 1;
    sequence($SEx, $SEc, $SEe, $SEch, $SEco, 'six1-eya1')if $seqs_to_print == 2;
}

say "$overlapping_count genes $direction-regulated in $n conditions";
say "$control_seen genes were removed with Expt.log(2)/Cont.log(2) > 0.5";

unless ($flag1 eq '-s'){
my $sleep = 2;
    while($sleep--){
        sleep(1);
    }
}

system("open overlapping_genes.csv");
}

### subs ###

# Attach sequences

unless ($flag1 eq '-s'){
    sub sequence {
        my (%seqs, $xloc);
        my ($xlocs, $c, $e, $ch, $gene, $count, $condition) = @_;
        $gene =~ s/^ //g;
}

```

```

        print $sequences ">$xlocs $gene $condition \n$six_seqs{$xlocs}\n" if
        $seqs_to_print == 0;
        print $sequences ">$xlocs $gene $condition \n$veya_seqs{$xlocs}\n" if
        $seqs_to_print == 1;
        print $sequences ">$xlocs $gene $condition \n$sixeya1_seqs{$xlocs}\n" if
        $seqs_to_print == 2;
    }
}

# 'fraction' takes genes in %change and calculates the ratio between the highest
# expt. condition and the control value and filters out genes with a value > 0.5.

# Outputs %control_ratio to sort genes by this ratio.

sub fraction {
    for my $gene ( sort keys %change){
        next unless $seen{$gene} >= $n;
        my ($con_val, $expt_val, $change_val, $count) = @{$control_filt{$gene}} if
        exists $control_filt{$gene};
        ($con_val, $expt_val, $change_val, $count) = (0,0,0.001, '') if not exists
        $control_filt{$gene};

        # Ranks the control genes -ve reg. as higher than absent
        if ($direction eq 'up'){
            $change_val = 0 if $change_val < 0;
        }
        else {
            $change_val = 0 if $change_val > 0;
        }
        my $fraction = eval abs sprintf('.3f', ($change_val/$change{$gene}));
        if (eval sprintf('1f', $fraction) > 0.5){
            $control_seen++;
            next;
        }
        else {
            $control_ratio{$gene} = [$fraction, $change{$gene}];
            $overlapping_count++;
        }
    }
}

# Extracts information from blast output foreach @ARGV and outputs %gene_list
sub blast_extractor {
    my $file = shift;
    open my $output, '<', $file or die "Can't read file '$file' [$!]\n";
    while (<$output>) {
        chomp;
        next if /^$/;
        my ($xloc) = /(XLOC_\d+)/;
        my ($change) = /Change:(-\?\d+\.\d+|-?inf|-?\d+)/;
        my ($container) = /XLOC_\d+_(_+:+),_Change/g;
        my ($con_val, $expt_val) = split(/:/, $container);
        my @split = split(/\t/);
        my ($condition, $percent_id, $gene) = (@split[0], $split[2], $split[3]);
        $gene =~ s/_/ /g;
        $gene =~ s/_//g;
        my @small_split = split(/\|/, $gene);
        my $small_gene = $small_split[4];
        my ($count) = $duplicates{$small_gene}{$condition}++;
        $gene_list{$small_gene}{$condition}{$count} = [$xloc, $con_val, $expt_val,
$change];
    }
    return \%gene_list;
}

sub usage {
    print "\n****CO-REG****\n\n";
    print "Usage: co-reg.pl <control gene list> <de gene list condition 1> .. <de
gene list condition N>\n";
    print "This program takes N blast hit results and searches for overlapping
genes.\n";
    print "Only conditions with a log(2) fold change value > $threshold are
considered.\n\n";
    print "All control values are parsed, and are used to calculate the ratio

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between control change value/experimental change value.\n";
print "All control values are parsed, and are used to calculate the ratio
between control change value/experimental change value.\n";
print "Output in .csv will contain genes sorted 1st on rank and 2nd on FC\n\n";}
```

## C.10 Sets

```
#####
### Sets ####
#####

#!/usr/bin/perl
use warnings;
use strict;
use Data::Dumper;
$Data::Dumper::Sortkeys = 1;

# This script divides blast input into discrete sets.
# The order input is given dictates the naming of the output:
# if run as perl sets.pl list1 list2 list3
# output will be as follows:
# SetA = list1 not list2 not list3
# SetB = list1 and list2 not list3
# SetC = list1 and list3 not list2
# SetD = list1 and list2 and list3

unless ($#ARGV == 2) {
    usage();
    exit; }

# used to specify which sequence to print (and which column to sort on)
print "Enter primary set: ";
chomp(my $set = <STDIN>);

my (%c1_vals, %c2_vals, %c3_vals);

parse($_) foreach @ARGV;

my $count = 0;
my $c;
my ($c1, $c2, $c3);
my (%c1_data, %c2_data, %c3_data);
my $c1_c2_count = 0;
my $c1_c3_count = 0;
my $cond1 = 0;
my $total = 0;
my $all = 0;
my (%six_seqs, %eya_seqs, %sixeya1_seqs, $xloc);

open my $S_transcriptome, '<',
'/Users/nickriddiford/Desktop/Data/Genes/six1/transcripts.txt' or die $!;

while (<$S_transcriptome>){
    chomp;
    ($xloc) = />(XLOC_\d+)\./ if /^>/;
    $six_seqs{$xloc} = $_ if /[>]/;
}

open my $E_transcriptome, '<',
'/Users/nickriddiford/Desktop/Data/Genes/eya1/transcripts.txt' or die $!;

while (<$E_transcriptome>){
    chomp;
    ($xloc) = />(XLOC_\d+)\./ if /^>/;
    $eya_seqs{$xloc} = $_ if /[>]/;
}

open my $SE_transcriptome, '<',
'/Users/nickriddiford/Desktop/Data/Genes/six1-eya1/transcripts.txt' or die $!;

while (<$SE_transcriptome>){
    chomp;
    ($xloc) = />(XLOC_\d+)\./ if /^>/;
    $sixeya1_seqs{$xloc} = $_ if /[>]/;
}

open my $set_A_seqs, '>', "$c1\set_A_seqs.txt" or die $!;
open my $set_B_seqs, '>', "$c1\set_B_seqs.txt" or die $!;
open my $set_C_seqs, '>', "$c1\set_C_seqs.txt" or die $!;
open my $set_D_seqs, '>', "$c1\set_D_seqs.txt" or die $!;
```

```

my $switch;
$switch = 5 if $set eq 'six';
$switch = 8 if $set eq 'eya';
$switch = 11 if $set eq 'six-eya';

open my $space_A, '>', "$c1\_\_set_A.csv" or die $!;
open my $space_B, '>', "$c1\_\_set_B.csv" or die $!;
open my $space_C, '>', "$c1\_\_set_C.csv" or die $!;
open my $space_D, '>', "$c1\_\_set_D.csv" or die $!;

print $space_A "Rank, Gene, Six1 CHX, Six1 CHX+DEX, Log(2), Eya1 CHX, Eya1
CHX+DEX, Log(2), Six1-Eya1 CHX, Six1-Eya1 CHX+DEX, Log(2), Control CHX, Control
CHX+DEX, Log(2)\n";

print $space_B "Rank, Gene, Six1 CHX, Six1 CHX+DEX, Log(2), Eya1 CHX, Eya1
CHX+DEX, Log(2), Six1-Eya1 CHX, Six1-Eya1 CHX+DEX, Log(2), Control CHX, Control
CHX+DEX, Log(2)\n";

print $space_C "Rank, Gene, Six1 CHX, Six1 CHX+DEX, Log(2), Eya1 CHX, Eya1
CHX+DEX, Log(2), Six1-Eya1 CHX, Six1-Eya1 CHX+DEX, Log(2), Control CHX, Control
CHX+DEX, Log(2)\n";

print $space_D "Rank, Gene, Six1 CHX, Six1 CHX+DEX, Log(2), Eya1 CHX, Eya1
CHX+DEX, Log(2), Six1-Eya1 CHX, Six1-Eya1 CHX+DEX, Log(2), Control CHX, Control
CHX+DEX, Log(2)\n";

# sort the lists first by control rank, then by FC and print out as in
# 'overlapping.pl'
for my $gene (sort { abs @{$c1_data{$a}}[0] <=gt abs @{$c1_data{$b}}[0]
                      or abs @{$c1_data{$b}}[$switch] <=gt abs @{$c1_data{$a}}[$switch]}
                      keys %c1_data)
{
    # second sort: six1 = 5
    # eya1 = 8

    # six-eya = 11      my ($rank, $gene, $xloc, $sc, $scd, $sch, $ec, $ecd,
    $ech, $sec, $secd, $sech,
    $cc, $ccd, $cch) = @{$c1_data{$gene}};
    $total++;
    if (not $c2_vals{$gene} and not $c3_vals{$gene}) {
        $cond1++;
        print $space_A
        "$rank, $gene, $sc, $scd, $sch, $ec, $ecd, $ech, $sec, $secd, $sech, $cc, $ccd, $cch\n";
        print $set_A_seqs ">$xloc\_$gene\_$c1\n$six_seqs{$xloc}\n" if $set eq 'six';
        print $set_A_seqs ">$xloc\_$gene\_$c1\n$eya_seqs{$xloc}\n" if $set eq 'eya';
        print $set_A_seqs ">$xloc\_$gene\_$c1\n$sixeya1_seqs{$xloc}\n" if $set eq
        'six-eya';
    }
    if ($c2_vals{$gene} and not $c3_vals{$gene}) {
        $c1_c2_count++;
        print $space_B
        "$rank, $gene, $sc, $scd, $sch, $ec, $ecd, $ech, $sec, $secd, $sech, $cc, $ccd, $cch\n";
        print $set_B_seqs ">$xloc\_$gene\_$c1\n$six_seqs{$xloc}\n" if $set eq
        'six';
        print $set_B_seqs ">$xloc\_$gene\_$c1\n$eya_seqs{$xloc}\n" if $set eq
        'eya';
        print $set_B_seqs ">$xloc\_$gene\_$c1\n$sixeya1_seqs{$xloc}\n" if $set eq
        'six-eya';
    }
    if ($c3_vals{$gene} and not $c2_vals{$gene}) {
        $c1_c3_count++;
        print $space_C
        "$rank, $gene, $sc, $scd, $sch, $ec, $ecd, $ech, $sec, $secd, $sech, $cc, $ccd, $cch\n";
        print $set_C_seqs ">$xloc\_$gene\_$c1\n$six_seqs{$xloc}\n" if $set eq
        'six';
        print $set_C_seqs ">$xloc\_$gene\_$c1\n$eya_seqs{$xloc}\n" if $set eq
        'eya';
        print $set_C_seqs ">$xloc\_$gene\_$c1\n$sixeya1_seqs{$xloc}\n" if $set eq
        'six-eya';
    }
    if ($c2_vals{$gene} and $c3_vals{$gene}) {
        $all++;
        print $space_D
        "$rank, $gene, $sc, $scd, $sch, $ec, $ecd, $ech, $sec, $secd, $sech, $cc, $ccd, $cch\n";
        print $set_D_seqs ">$xloc\_$gene\_$c1\n$six_seqs{$xloc}\n" if $set eq
        'six';
        print $set_D_seqs ">$xloc\_$gene\_$c1\n$eya_seqs{$xloc}\n" if $set eq
        'eya';
        print $set_D_seqs ">$xloc\_$gene\_$c1\n$sixeya1_seqs{$xloc}\n" if $set eq
    }
}

```

```

'six-eya';
}

# print numbers for each set
print "$c1 total = $total\n";
print "$c1 = $cond1\n";
print "$c1 + $c2 = $c1_c2_count\n";
print "$c1 + $c3 = $c1_c3_count\n";
print "$c1+$c2+$c3 = $all\n";
my $check = ($cond1 + $c1_c2_count + $c1_c3_count + $all);
print "check = $check\n";

# read in each file (output from 'co-reg.pl'). $ARGV[0] is set as the 'primary'
# set (ie that which intersecting lists are found for)
# read in each file (output from 'co-reg.pl'). $ARGV[0] is set as the 'primary'
# set (ie that which intersecting lists are found for)

sub parse {
$count++;
my $file = shift;
($c = $file) =~ s/\. [^.]+//;
open my $list, '<', $file or die "Can't read file '$file' [$!]\n";
while(<$list>) {
    chomp;
    if ($count == 1){
        my @split = split(/\t/);
        $c1_vals{$split[1]}++;
        $c1_data{$split[1]} = [@split];
        $c1 = $c;

    }

    if ($count == 2){
        my @split = split(/\t/);
        $c2_vals{$split[1]}++;
        $c2_data{$split[1]} = [@split];
        $c2 = $c;

    }

    if ($count == 3){
        my @split = split(/\t/);
        $c3_vals{$split[1]}++;
        $c3_data{$split[1]} = [@split];
        $c3 = $c;

    }
}

sub usage {
print "Usage: set.pl <gene list1> <gene list2> <gene list3>\n";
print "Calculates intersections between different sets\n";
}

```

## C.11 qPCR plots

```
#####
### qPCR.plot.R ####
#####

library(ggplot2)
library(gridExtra)

# This script will take in qPCR dat arranged as follows:
#   Gene   FC      expt    se
# e.g. Sox2    1.02    RNA-Seq 0
# and produce two barplots one for six1 and one for eya1 # Copy and past into an active R
session, or run as:

# > source("qPCR.plot.R")

dodge <- position_dodge(width = 0.6)

# Six1
six1 <- read.table("Six1_qPCR.txt", header=TRUE)

# Manual re-order
six1$Gene <- factor(six1$Gene,
c("Sox2","Hes5","Hes9","Atoh1","Ngn1","Pou4f1","Gfi1a","Tlx1","Isl2"))

s<-ggplot(data=six1, aes(x=Gene, y=FC, fill=expt, group=expt)) +
  xlab("\nGene") +
  ylab("log2 Fold Change\n") +
  geom_bar(colour="black", stat="identity", position=dodge, width=0.6) +
  theme(
    legend.position="none", # remove legend
    panel.grid.major.x = element_blank(), # remove x grids
    panel.grid.minor.x = element_blank(),
    panel.grid.major.y = element_line( size = .25, color = "#003366"),
    panel.grid.minor.y = element_line(size = .1, colour = "#003366"),
    # panel.background = element_rect(fill = "transparent",colour = NA), #
    transparent background and plotplot.background = element_rect(fill = "transparent",colour =
NA) )+

  geom_errorbar(aes(ymax = FC + se, ymin = FC, group=expt), position = dodge,
width = 0.25) +
  scale_fill_manual(values=c("lightcoral", "deepskyblue3"))

# Eya1
eya1 <- read.table("Eya1_qPCR.txt", header=TRUE)
eya1$Gene <- factor(eya1$Gene
,c("Sox2","Hes5","Hes9","Atoh1","Ngn1","Pou4f1","Gfi1a","Tlx1"))

e<-ggplot(data=eya1, aes(x=Gene, y=FC, fill=expt, group=expt)) +
  xlab("\nGene") +
  ylab("log2 Fold Change\n") +
  geom_bar(colour="black", stat="identity", position=dodge, width=0.6) +
  theme(
    legend.position="none", # remove legend
    panel.grid.major.x = element_blank(), # remove x grids
    panel.grid.minor.x = element_blank(),
    panel.grid.major.y = element_line( size = .25, color = "#003366"),
    panel.grid.minor.y = element_line(size = .1, colour = "#003366"),
    panel.background = element_rect(fill = "transparent",colour = NA), #
    transparent background and plotplot.background = element_rect(fill = "transparent",colour =
NA) )+

  geom_errorbar(aes(ymax = FC + se, ymin = FC, group=expt), position = dodge,
width = 0.25) +
  scale_fill_manual(values=c("lightcoral", "seagreen3"))

# Merge
grid.arrange(s, e, ncol=2)
```

# Appendix D. Placodal transcriptome

## D.1 Placodal transcriptome

**Table D.1** The top 1000 genes in the placodal transcriptome

Gene	Accession	FPKM
PREDICTED: Xenopus (Silurana) tropicalis inverted formin-2-like (LOC101733927)	XM_004919623.1	4960072.75
Xenopus tropicalis cDNA clone IMAGE:6991249	BC158454.1	582492.88
Xenopus tropicalis cDNA clone IMAGE:6992595	BC082728.1	148349.73
Xenopus laevis lactamase, beta 2 (lactb2)	NM_001094943.1	48580.58
Xenopus laevis, clone IMAGE:4884491	BC041318.1	46888.45
Xenopus laevis cDNA clone IMAGE:7200336	BC130151.1	19136.87
Xenopus laevis hypothetical protein LOC100049126 (cDNA clone MGC:161039 IMAGE:7299620)	BC133776.1	16475.8
Xenopus laevis rexp44 mRNA, complete sequence	DOQ96861.1	15632
Xenopus laevis cortactin (cttn)	NM_001092402.1	14507.47
Xenopus laevis uncharacterized LOC100158288 (LOC100158288)	NM_001127745.1	13556.38
XELUB1 Xenopus laevis ubiquitin mRNA	M11512.1	11460.32
Xenopus tropicalis cDNA clone IMAGE:6983060	BC156029.1	9353.43
Xenopus laevis similar to tissue specific transplantation antigen P35B (cDNA clone IMAGE:3399217), partial cds	BC108857.1	8885.36
Xenopus tropicalis finished cDNA, clone TNeu023c10	CT030610.1	7247.24
X. laevis mRNA for 42Sp48 protein	X56699.1	6511.4
Xenopus laevis thyroid hormone receptor associated protein 3 (thrap3)	NM_001103187.1	6287.39
Xenopus laevis small ubiquitin-like modifier 3 (sumo3)	NM_001086290.1	5560.28
Xenopus tropicalis finished cDNA, clone TNeu057o19	CR760167.2	5559.86
Xenopus laevis four and a half LIM domains 3 (fhl3)	NM_001092256.1	5362.83
X.laevis posterior (Xpo) gene	X58487.1	5326.76
Xenopus laevis cDNA clone MGC:98823 IMAGE:3401284	BC084658.1	4784.96
Xenopus laevis cold inducible RNA-binding protein (cDNA clone MGC:52663 IMAGE:4681597)	BC041204.1	4767.19
Xenopus laevis uncharacterized protein MGC78908 (MGC78908)	NM_001091061.1	4467.43
Xenopus laevis uncharacterized LOC100049124 (LOC100049124)	NM_001097957.1	4366.26
Xenopus laevis mRNA for cytokeratin type II (XenCK5(5/6))	X14427.1	4268.43
Xenopus laevis hypothetical protein MGC64297 (cDNA clone IMAGE:5570778)	BC043947.1	4200.66
Xenopus laevis tetraspanin 31 (tspan31-b)	NM_001086181.1	4139.75
Xenopus laevis keratin (krt-a)	NM_001087819.1	3986
Xenopus laevis holo cytochrome c synthase (hccs-b)	NM_001093680.1	3724
PREDICTED: Xenopus (Silurana) tropicalis filamin B, beta (flnb), transcript variant X2	XM_004914170.1	3353
PREDICTED: Xenopus (Silurana) tropicalis ubiquitin B (ubb), transcript variant X2	XM_004911681.1	3216.73
PREDICTED: Xenopus (Silurana) tropicalis tubulin alpha-1A chain-like (LOC100490289), transcript variant X2	XM_004917637.1	2880.36
AF231347 Xenopus laevis adenine nucleotide translocase (Ant1) mRNA; nuclear gene for mitochondrial product	AF231347.1	2832.78
Xenopus laevis gastrula stage epidermal type I cytokeratin (cDNA clone MGC:68905 IMAGE:4889094)	BC055996.1	2831.8
Xenopus tropicalis cDNA clone IMAGE:7547330	BC166316.1	2800.8
Xenopus laevis retrotransposon-like element, partial sequence	AF057166.1	2571.05
Xenopus laevis epid21 mRNA, complete sequence	DOQ96899.1	2463.46
Fragment of Xenopus laevis mRNA for ribosomal protein S19	V01443.1	2437.21
Xenopus laevis Ferritin heavy chain, oocyte isoform (cDNA clone MGC:64558 IMAGE:6880106)	BC056858.1	2424.81
XELB1AA X.borealis B1 protein mRNA	M63663.1	2408.68
Xenopus laevis nuclelease sensitive element binding protein 1 (cDNA clone MGC:52606 IMAGE:5570493)	BC042217.1	2395.6
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100498107 (LOC100498107)	XM_002936780.2	2376.46
Xenopus laevis keratin 8 (krt8)	NM_001087056.1	2356.22
Xenopus laevis hypothetical protein MGC64320 (cDNA clone IMAGE:4683880)	BC040972.1	2243.09
PREDICTED: Xenopus (Silurana) tropicalis epiplakin-like (LOC101733226)	XM_004919968.1	2215.4
XELRGT Figure 1. (c) Base sequence of the terminator and flanking DNA	M28530.1	2186.05
Xenopus laevis actin, beta (actb)	NM_001088953.1	2167.58
Xenopus laevis cDNA clone IMAGE:5156129	BC083028.1	2150.83
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733888 (LOC101733888)	XM_004911452.1	2143.35
Xenopus laevis XEEL mRNA for embryonic epidermal lectin	AB105372.1	2110.7
Xenopus laevis eukaryotic translation initiation factor 5 (eif5)	NM_001086842.1	2097.94
Xenopus laevis Nup98 protein (cDNA clone IMAGE:5536418), partial cds	BC077959.1	2086.85
Xenopus laevis uncharacterized protein MGC53311 (MGC53311)	NM_001085987.1	2058.32
Xenopus laevis proline-serine-threonine phosphatase interacting protein 2 (pstPIP2)	NM_001093301.1	2036.98
Xenopus laevis Heat shock cognate protein 70 (cDNA clone MGC:52655 IMAGE:4681532)	BC041201.1	1986.92
Xenopus laevis cold-inducible RNA binding protein 2 (cDNA clone MGC:64461 IMAGE:6880852)	BC054250.1	1960.75
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100495124 (LOC100495124)	XM_002935579.2	1788.39
X.laevis mRNA for nucleolin	X72957.1	1754.01
Xenopus laevis keratin 12 (krt12)	NM_001088963.1	1712.09
Xenopus laevis actin, gamma 1 (actg1-a)	NM_001096383.1	1704.34
Xenopus laevis MGC78980 protein (MGC78980)	NM_001091921.1	1691.82
Xenopus laevis eukaryotic translation elongation factor 1 alpha, somatic form (cDNA clone MGC:52631 IMAGE:4681345)	BC041196.1	1653.21
Xenopus tropicalis cDNA clone IMAGE:6982245	BC158446.1	1604.58
Xenopus laevis tumor protein, translationally-controlled 1 (tp1)	NM_001086678.1	1564.26
Xenopus laevis retrotransposon-like element 10A1 gag-like protein mRNA, partial cds; and 3' LTR, complete sequence	AF145966.1	1515.83
Xenopus laevis cDNA clone MGC:196526 IMAGE:9041433	BC169799.1	1496.12
Xenopus laevis uncharacterized LOC100158370 (LOC100158370)	NM_001127815.1	1481.3
Xenopus laevis otogelin (otog)	NM_001089059.1	1458.54
Xenopus laevis uncoupling protein 2 (mitochondrial, proton carrier) (ucp2), nuclear gene encoding mitochondrial protein	NM_001086754.1	1446.64
Xenopus laevis eukaryotic translation initiation factor 2, subunit 1 alpha, 35kDa (eif2s1)	NM_001087283.1	1383.76
Xenopus laevis hypothetical protein MGC68423 (cDNA clone MGC:68423 IMAGE:6863193)	BC055957.1	1377.11
Xenopus laevis mRNA for putative XIRG protein	AJ278067.1	1360.33
Xenopus laevis calreticulin (calr)	NM_001086627.1	1358.93
Xenopus laevis eukaryotic translation elongation factor 1 alpha, somatic form (cDNA clone MGC:53288 IMAGE:5570176)	BC043843.1	1332.88
Xenopus laevis heat shock 70kDa protein 8 (hspa8)	NM_001086163.1	1322.7
Xenopus laevis transmembrane protein TA-2-like (cDNA clone MGC:196571 IMAGE:9041478)	BC169844.1	1282.35
Xenopus laevis Xpo protein (cDNA clone IMAGE:6863526)	BC064682.1	1258.04
Xenopus laevis ribosomal protein S2 (rps2)	NM_001087413.1	1225.29
Xenopus laevis marginal coil Xmc (Xmc) mRNA	AF394111.1	1184.28
Xenopus (Silurana) tropicalis Dnaj (Hsp40) homolog, subfamily C, member 28 (dnajc28)	NM_001015977.1	1172.44
XELRPS2A X.laevis ribosomal protein S22, 40S subunit mRNA	M34706.1	1165.18
Xenopus laevis MGC80249 protein (MGC80249)	NM_001092031.1	1150.28
Xenopus laevis interleukin enhancer binding factor 2 (ilf2)	NM_001087106.1	1137.11

Gene	Accession	FPKM
Xenopus tropicalis finished cDNA, clone TNeu115i10	CT025176.2	1103.42
Xenopus laevis SET nuclear oncogene (set-b)	NM_001086440.1	1098.24
Xenopus laevis tRNA selenocysteine associated protein (secp43)	NM_001087119.1	1081.62
Xenopus laevis mitogen-activated protein kinase kinase 7 (map2k7)	NM_001087648.1	1064.78
Xenopus laevis mediator complex subunit 21 (med21)	NM_001092469.2	1050.54
Xenopus laevis high-mobility group nucleosome binding domain 1 (cDNA clone MGC:64449 IMAGE:6880568)	BC054972.1	1022.67
XELXLO1 Xenopus laevis mRNA for pleiotrophic factor-beta1	D42059.1	1022.47
Xenopus laevis TAF10 RNA polymerase II, TATA box binding protein (TBP)-associated factor, 30kDa (taf10)	NM_001097045.1	1012.4
Xenopus laevis cDNA clone IMAGE:7009319, **** WARNING: chimeric clone ****	BC086463.1	1005.89
Xenopus laevis ADP-ribosylation factor 1 (arf1)	NM_001086588.1	980.57
Xenopus (Silurana) tropicalis ribosomal protein L23a (rpl23a)	NM_001005109.2	969.34
PREDICTED: Xenopus (Silurana) tropicalis filamin C, gamma (flnc), transcript variant 1	XM_002931838.2	953.78
Xenopus laevis xPRMT1 mRNA for protein arginine methyltransferase 1	AB085173.1	931.73
Xenopus laevis coagulation factor 2 (thrombin) receptor-like 1 (f2rl1)	NM_001095470.1	923.95
PREDICTED: Xenopus (Silurana) tropicalis serine peptidase inhibitor, Kunitz type 1 (spint1), transcript variant X1	XM_002939545.2	921.59
Xenopus laevis splicing factor 3b, subunit 4, 49kDa (sf3b4)	NM_001086631.1	912.34
Xenopus laevis calpain 9 (capn9)	NM_001092528.1	903.17
Xenopus laevis serine/arginine-rich splicing factor 2 (srsf2)	NM_001087274.1	893.84
Xenopus laevis ribosomal protein L34 (cDNA clone MGC:64481 IMAGE:6881480)	BC053809.1	887.48
PREDICTED: Xenopus (Silurana) tropicalis AHNAK nucleoprotein (ahnak), transcript variant X2	XM_004912909.1	876.02
Xenopus laevis ribosomal protein L13a (rpl13a)	NM_001086661.1	871.17
Xenopus laevis cDNA clone IMAGE:4957472	BC130094.1	869.92
Xenopus laevis ribosomal protein S14 (cDNA clone MGC:53662 IMAGE:4887979)	BC041512.1	865.95
Xenopus (Silurana) tropicalis ribosomal protein L37a (rpl37a)	NM_001005137.1	862.33
Xenopus laevis ribosomal protein S21 (rps21)	NM_001093455.1	856.29
Xenopus laevis hypothetical protein LOC100049101 (cDNA clone IMAGE:4970197), partial cds	BC133742.1	843.4
Xenopus laevis ribosomal protein L10 (rpl10)	NM_001087197.1	843.19
Xenopus laevis tubulin, alpha 1 (cDNA clone MGC:52630 IMAGE:4681338)	BC041195.1	836.05
Xenopus laevis NADH dehydrogenase (ubiquinone) 1 alpha subcomplex, 4, 9kDa (ndufa4-a), nuclear gene encoding mitochondrial protein	NM_001171709.1	825.45
PREDICTED: Xenopus (Silurana) tropicalis Fc fragment of IgG binding protein (fcgbp)	XM_002940534.2	824.96
Xenopus laevis protein phosphatase 1, catalytic subunit, alpha isozyme (ppp1ca)	NM_001086753.1	821.87
Xenopus laevis XIrg protein-like (cDNA clone MGC:196449 IMAGE:9041356)	BC169722.1	820.57
Xenopus (Silurana) tropicalis actin, gamma 1 (actg1)	NM_204001.1	818.41
Xenopus laevis thymosin beta 4 peptide (cDNA clone IMAGE:7009418)	BC077646.1	812.67
Xenopus laevis nucleophosmin (nucleolar phosphoprotein B23, numatrin) (npm1)	NM_001086038.1	811.79
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 850-like (LOC101732212)	XM_004919126.1	811.08
XELABPEF Xenopus laevis poly(A)-binding protein (ABP-EF) mRNA	M27072.1	807.15
Xenopus laevis high mobility group HMG-17 (hmg-17)	NM_001088261.1	794.75
Xenopus laevis ribosomal protein S23 (rps23)	NM_001093433.1	793.54
Xenopus laevis keratin (krt-b)	NM_001087241.1	792.52
X.laevis mRNA for nucleolin	X72957.1	772.77
Xenopus laevis ribosomal protein L11 (rpl11)	NM_001093818.1	772.4
Xenopus laevis ribosomal protein L10a (rpl10a)	NM_001086736.1	769.66
PREDICTED: Xenopus (Silurana) tropicalis HECT, UBA and WWE domain containing 1, E3 ubiquitin protein ligase (huwe1)	XM_004914192.1	767.57
Xenopus laevis high mobility group nucleosomal binding domain 2 (hmgn2)	NM_001086309.1	764.25
Xenopus laevis tubulin, alpha 1a (tuba1a-b)	NM_001086587.1	755.65
Xenopus laevis lrr peptidase 2, peroxisomal (lornp2)	NM_001096479.1	753.37
Xenopus laevis heat shock protein 90kDa alpha (cytosolic), class B member 1 (hsp90ab1)	NM_001093155.1	751.29
Xenopus laevis metastasis suppressor 1, gene 2 (mtss1.2)	NM_001093916.1	744.81
Xenopus laevis ribosomal protein L3 (rpl3)	NM_001086872.1	739.09
Xenopus laevis putative transmembrane protein TA-2 mRNA	AF310008.1	737.17
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733854 (LOC101733854)	XM_004912652.1	732.35
Xenopus laevis clone S10-45-H5 mRNA sequence	AF549891.1	730.09
Xenopus laevis UPF0688 protein C1orf174 homolog (MGC115548)	NM_001096207.1	725.97
Xenopus laevis chgr07 mRNA, complete sequence	DCQ96903.1	723.2
Xenopus laevis ribosomal protein L36a (rpl36a)	NM_001093853.1	710.44
Xenopus laevis ribosomal protein S12 (rps12-a)	NM_001087125.1	701.39
Xenopus laevis MGC82136 protein (MGC82136)	NM_001092903.1	698.14
Xenopus laevis family with sequence similarity 135, member B (fam135b)	NM_001094366.1	695.15
Xenopus laevis X-epilectin mRNA	AY689185.1	694.65
Xenopus laevis MGC82416 protein (MGC82416)	NM_00109323.1	694.62
Xenopus laevis angiopoietin 4 (angpt4)	NM_001130950.1	694.4
Xenopus laevis uncharacterized protein MGC114621 (MGC114621)	NM_001095972.1	678.13
Xenopus laevis clone S10-56-G6 mRNA sequence	AF549908.1	675.09
Xenopus laevis lectin, galactoside-binding, soluble, 3 (lgals3)	NM_001086174.1	665.77
Xenopus laevis mRNA for DUF87	AB004793.1	665.76
TPA exp: Xenopus laevis glutamine synthetase (gso3) mRNA	BK000049.1	650.23
Xenopus laevis progestin and adipoQ receptor family member VI (paqr6-a)	NM_001093937.1	647.09
Xenopus laevis uncharacterized protein MGC68448 (MGC68448)	NM_001089645.1	646.5
Xenopus laevis ribosomal protein S19 (rps19)	NM_001087327.1	644.47
Xenopus laevis histone H2A.ZI2 (cDNA clone MGC:53404 IMAGE:5571643)	BC044011.1	642.6
Xenopus laevis ribosomal protein S27a (rps27a)	NM_001092596.1	642.52
XELRPS8B X.laevis ribosomal protein S8 (rps8B) mRNA	M21486.1	641.9
Xenopus laevis uncharacterized LOC100189560 (LOC100189560)	NM_001135063.1	641.88
Xenopus laevis mRNA for P11F3	AB072001.1	636.32
Xenopus laevis heterogeneous nuclear ribonucleoprotein D-like (hnrnndl)	NM_001086140.1	636.19
Xenopus laevis ribosomal protein S18 (rps18)	NM_001091278.1	631.34
Xenopus laevis signal sequence receptor beta subunit mRNA	AF346565.1	625.67
Xenopus laevis ribonucleotide reductase subunit M1 (RRM1) mRNA, partial cds	AF168794.1	624.96
Xenopus laevis eukaryotic translation elongation factor 2, gene 1 (eef2.1)	NM_001087187.1	618.67
X.laevis mRNA for variant histone H2A.ZI1	X98535.1	618.62
Xenopus tropicalis cDNA clone IMAGE:6991509	BC158455.1	616.86
Xenopus laevis hypothetical protein MGC64263 (cDNA clone MGC:181800 IMAGE:7391560)	BC152715.1	616.08
Xenopus laevis claudin 4 (cldn4)	NM_001096114.1	591.99
Xenopus laevis arginine methyltransferase 1b mRNA	AY330768.1	591.71
Xenopus laevis ribosomal protein L35 (rpl35)	NM_001096138.1	591.31
Xenopus laevis MGC78885 protein (cDNA clone MGC:154294 IMAGE:8070732)	BC123118.1	589.31
Xenopus laevis ribosomal protein S3A (rps3a-a)	NM_001086856.1	588.79
Xenopus laevis CWF19-like 2, cell cycle control (cwf19l2)	NM_001096193.1	584.91
Xenopus tropicalis finished cDNA, clone TEgg07Bb22	CR761304.2	582.68
PREDICTED: Xenopus (Silurana) tropicalis IgGFc-binding protein-like (LOC100492852)	XM_002940533.2	582.58
Xenopus laevis uncharacterized protein MGC81434 (MGC81434)	NM_001091189.1	578.6

Gene	Accession	FPKM
PREDICTED: Xenopus (Silurana) tropicalis protein S100-G-like (LOC100494945)	XM_002942678.2	574.14
Xenopus laevis ribosomal protein L7 (rpl7)	NM_001096628.1	569.95
Xenopus laevis eukaryotic translation initiation factor 5A (eif5a)	NM_001087067.1	569.61
Xenopus laevis hypothetical protein LOC398551 (cDNA clone MGC:64328 IMAGE:6876388)	BC054189.1	565.1
Xenopus tropicalis finished cDNA, clone TGas030a18	CR855708.2	561.72
Xenopus laevis ribosomal protein L7a (rpl7a)	NM_001092017.1	561.08
Xenopus laevis zinc finger and BTB domain containing 17 (zbtb17)	NM_001096528.1	560.82
Xenopus laevis nuclear RNA helicase (ddx39)	NM_001087264.1	559.15
Xenopus laevis COP9 signalosome subunit 6 (cops6)	NM_001091150.1	557.99
Xenopus laevis hypothetical protein MGC64236 (cDNA clone IMAGE:5542101), partial cds	BC046648.1	555.78
Xenopus laevis serine/arginine-rich splicing factor 3 (srsf3)	NM_001086991.1	553.93
Xenopus laevis MGC80994 protein (MGC80994)	NM_001092419.1	549.82
PREDICTED: Xenopus (Silurana) tropicalis ubiquitin B (ubb), transcript variant X3	XM_004911682.1	547.73
Xenopus laevis isocitrate dehydrogenase 1 (NADP+), soluble (idh1)	NM_001094553.1	543.15
Xenopus laevis cDNA clone IMAGE:7211162	BC106480.1	543.01
Xenopus laevis signal sequence receptor, gamma (translocon-associated protein gamma) (ssr3-a)	NM_001086847.1	542.12
Xenopus laevis cDNA clone MGC:85498 (IMAGE:6940048)	BC084859.1	539.68
Xenopus laevis ribosomal protein L4 (cDNA clone MGC:130828 IMAGE:7981421)	BC106315.1	538.23
Xenopus laevis solute carrier family 22 (organic cation/ergothioneine transporter), member 4 (slc22a4)	NM_001094580.1	538.05
Xenopus laevis hypothetical protein LOC443682 (cDNA clone IMAGE:5570626), partial cds	BC073668.1	537.51
Xenopus laevis ribosomal protein SA (rpsa)	NM_001087261.1	534.38
Xenopus laevis ribosomal protein S9 (rps9)	NM_001092340.1	531.44
Xenopus laevis calreticulin (calr)	NM_001086627.1	529.17
Xenopus laevis uncharacterized LOC100137635 (LOC100137635)	NM_001114785.1	529.14
Xenopus tropicalis finished cDNA, clone TEgg074l07	CT030370.1	524.18
Xenopus laevis signal sequence receptor, beta (translocon-associated protein beta) (ssr2)	NM_001086212.1	522.3
Xenopus laevis beta-Tubulin at 56D (cDNA clone MGC:64554 IMAGE:6879809)	BC054297.1	520.73
X.laevis mRNA for PP2A 65 kDa regulatory subunit alpha-isotype	X80696.1	517.14
Xenopus laevis ribosomal protein L14 (rpl14)	NM_001091255.1	516.53
XELTFIDS Xenopus laevis oocyte mRNA for transcription factor TFIID subunit	D50054.1	516.23
Xenopus laevis cico01 mRNA, complete sequence	DQ096916.1	514.44
Xenopus laevis tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, theta polypeptide (ywhaq)	NM_001086903.1	512.14
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100491360 (LOC100491360), transcript variant X1	XM_004917936.1	511.62
XELRPS8A X.laevis ribosomal protein S8 (rps8A) mRNA	M21485.1	510.73
XELXGS Xenopus laevis mRNA for glutamine synthetase	D50062.1	506.13
Xenopus laevis Nuclelease-sensitive element-binding protein 1 (cDNA clone MGC:52597 IMAGE:4681490)	BC041191.1	503.7
Xenopus laevis ribosomal protein L19 (rpl19-a)	NM_001086798.1	500.12
Xenopus laevis ribosomal protein S16 (cDNA clone MGC:130741 IMAGE:7973798)	BC106273.1	498.12
Xenopus laevis annexin A4 (anxa4)	NM_001090012.1	495.03
Xenopus laevis archain 1 (arcn1)	NM_001087214.1	493.27
Xenopus tropicalis novel protein similar to prothymosin, alpha (cDNA clone MGC:184634 IMAGE:7673450)	BC157756.1	491.73
Xenopus laevis 40S ribosomal protein S4 (rps4)	NM_001091472.1	491.19
Xenopus laevis heterogeneous nuclear ribonucleoprotein A1 (hnrmrp1)	NM_001087220.1	490.76
Xenopus laevis mRNA for TAF-Ibeta2	AB022692.1	490.69
Xenopus laevis Xesp-1 mRNA for embryonic serine protease-1	AB038496.1	489.65
Xenopus laevis uncharacterized protein MGC114789 (MGC114789)	NM_001095990.1	486.93
Xenopus laevis cDNA clone IMAGE:6876276, containing frame-shift errors	BC053781.1	484.9
Xenopus laevis cDNA clone MGC:82601 (IMAGE:4971138)	BC077493.1	483.28
Xenopus laevis cDNA clone MGC:85435 (IMAGE:6935251)	BC078558.1	477.93
XELEF1ALA X.laevis elongation factor-1 alpha-chain (EF-1-alpha) mRNA	M25504.1	477.92
Xenopus laevis uroplakin 1B (upk1b)	NM_001093265.1	474.46
Xenopus (Silurana) tropicalis KH domain containing, RNA binding, signal transduction associated 1 (khdrbs1)	NM_001017045.2	470.32
Xenopus (Silurana) tropicalis ribosomal protein S26 (rps26)	NM_001005121.2	470.13
Xenopus laevis ribosomal protein, large, P1 (rplp1)	NM_001090161.1	466.54
Xenopus laevis proline-rich Vg1 mRNA-binding protein (cDNA clone IMAGE:6864417), partial cds	BC063270.1	466.44
Xenopus laevis ribosomal protein S9 (rps9)	NM_001092340.1	464.31
Xenopus tropicalis cDNA clone IMAGE:7030532, containing frame-shift errors	BC082738.1	460.85
Xenopus laevis histone cluster 2, H3d (hist2h3d)	NM_001092605.1	458.88
Xenopus laevis fused in sarcoma (fus)	NM_001086914.1	451.98
PREDICTED: Xenopus (Silurana) tropicalis vitellogenin-A2-like (LOC100494633)	XM_004913911.1	447.36
Xenopus laevis ribosomal protein L13 (rpl13)	NM_001093003.1	442.42
Xenopus laevis nucleolar protein NO38 (no38)	NM_001086291.1	441.29
Xenopus laevis ribosomal protein L27a (cDNA clone MGC:196755 IMAGE:9041663)	BC170028.1	440.56
Xenopus (Silurana) tropicalis ribosomal protein L26 (rpl26)	NM_001005104.2	438.13
Xenopus laevis cDNA clone 12F11	AJ009287.1	434.01
Xenopus laevis cDNA clone IMAGE:6934659	BC099261.1	429.42
40S ribosomal small subunit protein S10 [Xenopus laevis, splenocytes, 538 nt]	S57432.1	428.89
Xenopus laevis cDNA clone IMAGE:3400494	BC080436.1	428.43
Xenopus laevis 3-mercaptopyruvate sulfurtransferase (mpst)	NM_001097073.1	427.68
Xenopus laevis heterogeneous nuclear ribonucleoprotein A/B (hnrmrpab)	NM_001087020.1	426.27
Xenopus laevis ribosomal protein L15 (rpl15)	NM_001087297.1	425.7
Xenopus laevis uncharacterized LOC100037021 (LOC100037021)	NM_001097761.1	425.62
Xenopus laevis ribosomal protein L8 mRNA	U00920.1	422.8
Xenopus (Silurana) tropicalis ribosomal protein S27.1 (rps27.1)	NM_0011171651.1	422.57
Xenopus laevis cDNA clone IMAGE:3398708	BC108862.1	421.95
Xenopus laevis ribosomal protein L34 (rpl34)	NM_001093842.1	421.55
Xenopus laevis cDNA clone MGC:81816 (IMAGE:7007960)	BC075124.1	420.86
Xenopus laevis ribosomal protein L6 (rpl6)	NM_001092921.1	420.11
Xenopus laevis ribosomal protein L8 (rpl8)	NM_001086996.1	419
Xenopus laevis hypothetical protein MGC80045 (cDNA clone MGC:80045 IMAGE:4032724)	BC070571.1	418.54
Xenopus laevis guanine nucleotide binding protein, beta 2, related sequence 1 (gnb2l1)	NM_001086664.1	417.94
Xenopus laevis ribosomal protein L5 (rpl5-b)	NM_001085968.1	416.83
Xenopus laevis hypothetical protein LOC100126627 (cDNA clone IMAGE:8533789), partial cds	BC153770.1	414.43
Xenopus laevis histone H3.3 (MGCS2708)	NM_001085906.1	413.72
Xenopus laevis hypothetical protein LOC398440 (cDNA clone IMAGE:4889483), partial cds	BC044967.1	413.4
PREDICTED: Xenopus (Silurana) tropicalis transcription elongation factor B (SII), polypeptide 3 (110kDa, elongin A) (tceb3)	XM_002933844.2	410.08
PREDICTED: Xenopus (Silurana) tropicalis 60S ribosomal protein L32-like (LOC100493057)	XM_002936246.2	408.44
Xenopus laevis ribosomal protein L15 (cDNA clone MGC:81851 IMAGE:7008333)	BC075126.1	407.35
Xenopus tropicalis finished cDNA, clone TNeu079d01	CT030536.1	406.29
Xenopus laevis heterogeneous nuclear ribonucleoprotein A2/B1 (hnrmrpab2b1)	NM_001086921.1	405.86
Xenopus laevis hypothetical LOC495285 (cDNA clone IMAGE:4970860), partial cds	BC084641.1	405.28
XLU20977 Xenopus laevis cellular nucleic acid binding protein (CNBP) mRNA	U20977.1	403.91

Gene	Accession	FPKM
Xenopus laevis ribosomal protein S13 (rps13)	NM_001086882.1	403.44
Xenopus laevis far upstream element (FUSE) binding protein 3 (fubp3)	NM_001089921.1	402.75
X.laevis mRNA for ribosomal protein L22 homologue	X94243.1	402.19
Xenopus (Silurana) tropicalis uncharacterized protein MGC75872 (MGC75872)	NM_203836.1	400.57
Xenopus laevis CD2-associated protein (cd2ap)	NM_001092963.1	400.51
Xenopus laevis tumor-associated calcium signal transducer 1 mRNA	AY166594.1	399.16
Xenopus laevis zinc finger, CCCH-type with G patch domain (zgpat)	NM_001095068.1	396.33
Xenopus laevis ribosomal protein L21 (rpl21)	NM_001086258.1	395.21
Xenopus laevis cDNA clone IMAGE:5072983, **** WARNING: chimeric clone ****	BC072757.1	394.53
Xenopus laevis hatching gland-like Xhel protein (LOC398485)	NM_001088989.1	393.98
Xenopus (Silurana) tropicalis tyrosine 3-monooxygenase/triptophan 5-monooxygenase activation protein, theta polypeptide (ywhaq)	NM_001030399.1	392.26
Xenopus laevis HBS1-like (hbs1l)	NM_001092382.1	391.37
PREDICTED: Xenopus (Silurana) tropicalis IQ motif containing GTPase activating protein 1 (iqgap1)	XM_002932266.2	391.02
Xenopus laevis XISOX-2 (Sox-2) mRNA	AF005476.1	390.83
Xenopus laevis death inducer-obliterator 1 (dido1)	NM_001094588.1	389.29
Xenopus laevis heavy-chain binding protein BiP (cDNA clone MGC:79065 IMAGE:4957353)	BC077757.1	389.24
Xenopus laevis ribosomal protein S17 (rps17)	NM_001092464.1	388.61
Xenopus laevis ribonucleoprotein A1a (hnrrnpa1)	NM_001087813.1	385.96
Xenopus laevis ribosomal protein S15 (rps15)	NM_001087364.1	385.89
PREDICTED: Xenopus (Silurana) tropicalis retrotransposon-like protein 1-like (LOC101730840)	XM_004914237.1	384.46
Xenopus laevis mRNA similar to ribosomal protein L13 (cDNA clone MGC:53222 IMAGE:5543356)	BC041531.1	383.69
Xenopus laevis GID complex subunit 8 homolog (S. cerevisiae) (gid8)	NM_001096765.1	382.85
PREDICTED: Xenopus (Silurana) tropicalis proline-rich coiled-coil 2B (prrc2b)	XM_002936190.2	379.6
Xenopus laevis Xran mRNA for ran GTP-binding protein	AB030945.1	377.32
Xenopus tropicalis finished cDNA, clone TNeu086n11	CU075727.1	377.18
Xenopus laevis WBP2 N-terminal like (wbp2nl)	NM_001094568.1	376.76
Xenopus (Silurana) tropicalis disrupted in renal carcinoma 2 (dirc2)	NM_001016971.3	376.19
Xenopus (Silurana) tropicalis eukaryotic translation elongation factor 2, gene 1 (eef2.1)	NM_203924.1	373.12
Xenopus laevis ribosomal protein L24 (rpl24)	NM_001093723.1	372.2
Xenopus laevis uncharacterized protein MGC80262 (MGC80262)	NM_001091737.1	371.92
Xenopus laevis small nuclear ribonucleoparticle-associated protein (snRNP) mRNA, clone Sm51	BC041275.1	370.78
Xenopus laevis cDNA clone MGC:83385 IMAGE:7008012	BC074212.1	370.24
Xenopus laevis ribosomal protein L12 (rpl12)	NM_001086653.1	370.22
Xenopus laevis ribosomal protein L28 (rpl28-a)	NM_001099866.1	367.76
Xenopus laevis RAD51 homolog (S. cerevisiae) (rad51-a)	NM_001087767.1	363.83
Xenopus tropicalis novel protein similar to human angiopoietin-like ANGPTL (cDNA clone MGC:185766 IMAGE:7546975)	BC158370.1	363.33
Xenopus laevis MGC80767 protein (MGC80767)	NM_001092324.1	363.23
Xenopus laevis ribosomal protein, large, P2 (rlp2)	NM_001096852.1	362.87
Xenopus laevis churchill domain containing 1 (churc1)	NM_001085739.1	360.09
Xenopus laevis ribosomal protein S11 (cDNA clone MGC:64491 IMAGE:6872649)	BC053813.1	356.25
Xenopus laevis XNop56 protein (cDNA clone IMAGE:5085468), partial cds	BC097676.1	353.52
X. laevis ODC mRNA for ornithine decarboxylase (EC 4.1.1.17)	X56316.1	353.49
Xenopus laevis uncharacterized protein MGC116435 (MGC116435)	NM_001096142.1	353
Xenopus laevis MGC78845 protein (MGC78845)	NM_001093149.1	352.93
Xenopus laevis mRNA for ribosomal protein L14b	X06223.1	352.1
xnf7-O=nuclear factor 7 [Xenopus laevis, oocytes, 2343 nt]	S80988.1	351.51
Xenopus (Silurana) tropicalis splicing factor proline/glutamine-rich (sfpq)	NM_001127955.1	351.45
Xenopus laevis Cysteine and histidine-rich protein 1A (cDNA clone IMAGE:5513813), partial cds	BC073380.1	350.98
Xenopus laevis coiled-coil domain containing 63 (ccdc63)	NM_001090143.1	350.94
XELRNPA3B Xenopus laevis ribonucleoprotein mRNA	L02957.1	349.99
Xenopus laevis uncharacterized LOC446289 (LOC446289)	NM_001092996.1	347.77
Xenopus laevis Sec61 alpha 1 subunit (sec61a1)	NM_001086775.1	343.83
PREDICTED: Xenopus (Silurana) tropicalis embryonic protein UVS.2-like (LOC101734297), transcript variant X1	XM_004913368.1	343.51
Xenopus laevis CD63 antigen-like (LOC398555)	NM_001089077.1	341.85
Xenopus laevis H3 histone, family 3B (H3.3B) (h3f3b)	NM_001091579.1	341.57
Xenopus laevis hypothetical LOC398633 (cDNA clone IMAGE:7983501)	BC110715.1	341.15
Xenopus laevis calpain 5 (capn5)	NM_001087339.1	340.6
Xenopus laevis hypothetical protein LOC446245 (cDNA clone IMAGE:5440250), partial cds	BC078043.1	338.93
Xenopus laevis heterogeneous nuclear ribonucleoprotein K (hnrrpk)	NM_001087094.1	338.65
Xenopus laevis high mobility group box 3 (hmgb3)	NM_001087116.1	336.55
Xenopus laevis high mobility group nucleosomal binding domain 3 (cDNA clone IMAGE:4681438)	BC041718.1	333.8
Xenopus tropicalis MGC69457 protein (cDNA clone MGC:135204 IMAGE:7529780)	BC158916.1	333.12
Xenopus laevis soc001 mRNA, complete sequence	DQ096857.1	333.08
Xenopus laevis StAR-related lipid transfer (START) domain containing 10 (stard10)	NM_001094732.1	331.72
Xenopus laevis ATPase, Na <sup>+</sup> /K <sup>+</sup> transporting, beta 2 polypeptide (cDNA clone MGC:82363 IMAGE:4203104)	BC082868.1	331.4
Xenopus laevis ribosomal protein S5 (rps5)	NM_001086331.1	330.82
Xenopus laevis keratin 18 (krt18-b)	NM_001087341.1	330.19
Xenopus laevis epithelial cell adhesion molecule (epcam-a)	NM_001093506.1	330.05
Xenopus laevis cDNA clone IMAGE:5570937, partial cds	BC168523.1	330.03
Xenopus laevis mRNA for ribosomal protein L32	X55030.1	327.74
Xenopus laevis small nuclear ribonucleoprotein polypeptide F (snrpf)	NM_001087432.1	327.11
Xenopus laevis cnef01 mRNA, complete sequence	DQ096911.1	326.64
Xenopus laevis ribosomal protein S3A (rps3a-b)	NM_001110740.1	326.3
Xenopus tropicalis finished cDNA, clone TNeu059k22	CT025295.2	325.16
Xenopus laevis, Similar to splicing factor, arginine-serine-rich 5, clone IMAGE:5570132	BC044284.1	325.09
Xenopus laevis translocation associated membrane protein 1 (tram1)	NM_001093156.1	323.78
Xenopus laevis DECD-box RNA helicase (DDX39) mRNA	AY298950.1	323.47
PREDICTED: Xenopus (Silurana) tropicalis INO80 homolog (ino80)	XM_004917309.1	323.4
Xenopus laevis ribosomal protein, large, P0 (rlp0)	NM_001086665.1	323.35
Xenopus laevis heterogeneous nuclear ribonucleoprotein H3 (2H9) (hnrrph3)	NM_001094504.1	322.6
XELRNPA2B Xenopus laevis ribonucleoprotein mRNA	L02955.1	322.13
Xenopus laevis U2 small nuclear RNA auxiliary factor 2 (u2af2)	NM_001087126.1	320.51
Xenopus laevis ribosomal protein S12 (rps12-b)	NM_001086433.1	320.36
Xenopus laevis Rho GDP dissociation inhibitor (GDI) gamma (arhgdi)	NM_001094028.1	319.77
PREDICTED: Xenopus (Silurana) tropicalis thymosin beta-15A homolog (LOC100488215), transcript variant X1	XM_002940401.2	319.05
Xenopus laevis mRNA for ribosomal protein L14a	X06222.1	318.17
Xenopus laevis heterogeneous nuclear ribonucleoprotein D-like (hnrrpd1-b)	NM_001091139.1	317.27
Xenopus laevis ribosomal protein S10 (rps10)	NM_001087259.1	315.74
Xenopus laevis cathepsin V (ctsv)	NM_001094020.1	315.07
Xenopus tropicalis cDNA clone IMAGE:7030014, containing frame-shift errors	BC080160.1	314.51
Xenopus laevis ribosomal protein S4, X-linked (rps4x)	NM_001097003.1	309.81
Xenopus laevis tubulin, alpha 3c (tuba3c)	NM_001086054.1	308.76

Gene	Accession	FPKM
Xenopus laevis acidic (leucine-rich) nuclear phosphoprotein 32 family, member C (anp32c)	NM_001096478.1	306.53
Xenopus (Silurana) tropicalis ribosomal protein L6 (rpl6)	NM_001142035.1	306.02
Xenopus laevis histone cluster 1, H2bj (hist1h2bj)	NM_001093284.1	304.5
Xenopus laevis stem cell leukemia protein SCL (cDNA clone MGC:79923 IMAGE:4175038)	BC072130.1	303.06
Xenopus laevis uncharacterized protein MGC131332 (MGC131332)	NM_001096476.1	302.14
Xenopus laevis high mobility group B3b protein (cDNA clone MGC:154337 IMAGE:7974582)	BC123147.1	301.48
Xenopus laevis hypothetical protein MGC64286 (cDNA clone MGC:64286 IMAGE:6874131)	BC053768.1	298.13
Xenopus laevis ribosomal protein L27a (rpl27a)	NM_001086720.1	295.7
Xenopus laevis ribosomal protein L9 (rpl9)	NM_001087300.1	294.3
Xenopus laevis galectin IX mRNA, partial cds	JN975639.1	293.84
Xenopus tropicalis finished cDNA, clone TGas076k16	CR761730.2	291.59
Xenopus laevis epco02 mRNA, complete sequence	DQ096943.1	291.36
PREDICTED: Xenopus (Silurana) tropicalis ribosomal protein S17 (rps17), transcript variant X1	XM_002938873.2	291.05
Xenopus laevis cDNA clone IMAGE:6938161	BC099319.1	290.35
Xenopus laevis uncharacterized protein MGC64293 (MGC64293)	NM_001089194.1	290.25
Xenopus laevis ribosomal protein L5 (rpl5-a)	NM_001085908.1	289.35
Xenopus laevis uncharacterized LOC495835 (LOC495835)	NM_001095192.1	288.07
Xenopus laevis NF-protocadherin (NFPC) mRNA	AF043643.1	287.98
Xenopus (Silurana) tropicalis eukaryotic translation initiation factor 5 (eif5)	NM_001008208.1	287.24
Xenopus laevis hypothetical protein MGC52708 (cDNA clone MGC:52708 IMAGE:4683607)	BC041218.1	285.48
Xenopus laevis glutamine-fructose-6-phosphate transaminase 2 (gfpt2)	NM_001087014.1	284.48
Xenopus laevis MGC80163 protein (MGC80163)	NM_001092006.1	284.26
Xenopus laevis small integral membrane protein 7 (smim7)	NM_001091931.1	282.26
Xenopus laevis solute carrier family 25 (mitochondrial carrier; phosphate carrier), member 3 (slc25a3)	NM_001086726.1	282.13
Xenopus laevis MGC82859 protein (cDNA clone MGC:82859 IMAGE:5511530)	BC073568.1	281.2
XLU10108 Xenopus laevis Na <sup>+</sup> -K <sup>+</sup> -ATPase alpha 1 subunit mRNA	U10108.1	280.95
Xenopus laevis transgelin 2 (tagln2)	NM_001087314.1	280
Xenopus laevis ribosomal protein L28 (rpl28-b)	NM_001086269.1	279.87
Xenopus (Silurana) tropicalis cathepsin Z (ctsz)	NM_001112956.1	279.67
Xenopus laevis Probable ATP-dependent RNA helicase DDX5-like (MGC53795)	NM_001086234.1	279.13
Xenopus laevis hypothetical protein LOC446267 (cDNA clone IMAGE:5542041), partial cds	BC079725.1	278.11
XLU07156 Xenopus laevis dsRNA-binding protein 4F.2 mRNA, partial cds	U07156.1	277.3
Xenopus (Silurana) tropicalis helicase, lymphoid-specific (hells)	NM_001142221.2	277.24
Xenopus laevis orga04 mRNA, complete sequence	DQ096925.1	276.75
Xenopus laevis ubiquitin A-52 residue ribosomal protein fusion product 1 (uba52)	NM_001091987.1	276.64
Xenopus laevis eukaryotic translation initiation factor XeIF-4AIII mRNA	AF020432.1	276.4
Xenopus laevis Sec61 beta subunit (sec61b)	NM_001086687.1	274.25
Xenopus laevis stromal interaction molecule 1 (stim1)	NM_001097037.1	273.13
Xenopus laevis DnaJ (Hsp40) homolog, subfamily A, member 1 (dnaja1)	NM_001086896.1	272.66
Xenopus laevis elongation factor 1 homolog (elf1)	NM_001091854.1	272.4
Xenopus laevis DEAD (Asp-Glu-Ala-Asp) box polypeptide 19B (ddx19b)	NM_001087163.1	272.12
Xenopus laevis pendrin-like anion exchanger mRNA	AF544021.1	270.39
PREDICTED: Xenopus (Silurana) tropicalis EF-hand calcium binding domain 12 (efcab12), transcript variant X2	XM_004914125.1	268.71
Xenopus laevis anterior gradient 2 homolog (agr2)	NM_001086251.1	268.42
Xenopus laevis coatomer protein complex, subunit beta 2 (beta prime) (copb2)	NM_001086752.1	268.12
Xenopus laevis NSA2 ribosome biogenesis homolog (nsa2)	NM_001174021.1	267.68
Xenopus (Silurana) tropicalis high density lipoprotein binding protein (viginil) (hdlbp)	NM_001006853.1	266.87
XLU76752 Xenopus laevis putative secreted protein XAG (XAG) mRNA	U76752.1	266.38
Xenopus laevis peptidylprolyl isomerase D (cyclophilin D) (ppid)	NM_001094385.1	265.79
Xenopus laevis NHP2 non-histone chromosome protein 2-like 1 (nhp2l1-a)	NM_001094930.1	265.71
Xenopus laevis mRNA for Na/K-ATPase beta-2 subunit (atp1B2 gene)	AJ293961.1	263.38
Xenopus laevis uroplakin III (cDNA clone MGC:80399 IMAGE:5154763)	BC073184.1	261.84
PREDICTED: Xenopus (Silurana) tropicalis NXPE family member 2-like (LOC100487011)	XM_002940452.2	261.41
Xenopus laevis claudin 7 (cldn7)	NM_001086240.1	261.19
Xenopus laevis uncharacterized LOC100049136 (LOC100049136)	NM_001097965.1	260.51
Xenopus laevis CYCLOPhilin (18.4 kD) (cyp-7) (cyp-7)	NM_001086791.1	258.89
PREDICTED: Xenopus (Silurana) tropicalis transmembrane 9 superfamily member 3 (tm9sf3)	XM_002935964.2	258.73
Xenopus laevis U2 small nuclear RNA auxiliary factor 1 (u2af1)	NM_001096391.1	257.75
Xenopus laevis p68 RNA helicase mRNA	AF218580.1	257.51
Xenopus laevis high mobility group box 2 (hmgb2)	NM_001085918.1	256.27
Xenopus laevis ribosomal protein L30 (rpl30)	NM_001087152.1	256
Xenopus laevis cDNA clone IMAGE:6871346	BC088971.1	255.28
XELRPS6X Xenopus laevis ribosomal protein S6 mRNA and flanking regions	L19996.1	255.07
Xenopus laevis fructose-1,6-bisphosphatase 1 (fbp1)	NM_001087059.1	255.05
Xenopus laevis prostaglandin-endoperoxide synthase 2 (prostaglandin G/H synthase and cyclooxygenase) (ptgs2)	NM_001093477.1	254.99
Xenopus laevis ribosomal protein L27 (rpl27)	NM_001086954.1	254.95
Xenopus laevis epithelial membrane protein 2 (emp2)	NM_001086112.1	254.88
Xenopus (Silurana) tropicalis nucleosome assembly protein 1-like 1 (nap1l1), transcript variant 2	NM_001015984.3	253.68
Xenopus laevis uncharacterized LOC100127268 (LOC100127268)	NM_001112845.1	252.21
Xenopus laevis ribosomal protein L38 (rpl38)	NM_001093847.1	252.08
Xenopus laevis zinc finger protein 182 (znf182)	NM_001096703.1	251.95
Xenopus laevis claudin4L1 (cDNA clone MGC:81414 IMAGE:6635231)	BC068813.1	251.85
Xenopus laevis ferritin, heavy polypeptide 1 (fth1-b)	NM_001086111.1	251.31
Xenopus laevis phosphatidylinositol transfer protein, beta (pitpnB)	NM_001086805.1	250.95
Xenopus laevis uncharacterized protein MGC53997 (MGC53997)	NM_001086046.1	247.75
Xenopus laevis cldn4L2 mRNA for Claudin4L2	AB072909.1	246.4
X.laevis mRNA for ribosomal protein S1a	X57322.1	245.21
Xenopus laevis ATPase, H <sup>+</sup> transporting, lysosomal 16kDa, V0 subunit c (atp6v0c)	NM_001089206.1	245.2
Xenopus laevis upstream transcription factor 2, c-fos interacting (usf2-b)	NM_001095231.1	244.2
Xenopus laevis RNA-binding protein 8A-B (rbm8a-b)	NM_001086436.1	244.19
Xenopus laevis ras homolog family member C (rhoc)	NM_001086170.1	243.72
Xenopus laevis uncharacterized LOC100158267 (LOC100158267)	NM_001127726.1	243.68
PREDICTED: Xenopus (Silurana) tropicalis peroxisome proliferator-activated receptor alpha-like (LOC100497346)	XM_002940738.2	243.57
Xenopus laevis ubiquitin C (ubc)	NM_001087396.1	243.28
PREDICTED: Xenopus (Silurana) tropicalis mucin-5B-like (LOC100488209)	XM_002938895.2	241.18
Xenopus laevis clone S10-51-H5 mRNA sequence	AF549994.1	240.83
Xenopus laevis activating transcription factor 4 (atf4-a)	NM_001089743.1	240.79
Xenopus laevis TAF-Ibeta2 (cDNA clone MGC:79899 IMAGE:6634045)	BC072127.1	240.44
Xenopus laevis ribosomal protein L23a (rpl23a)	NM_001093830.1	239.03
Xenopus laevis MGC81939 protein (MGC81939)	NM_001092608.1	238.68
PREDICTED: Xenopus (Silurana) tropicalis parvin, alpha (parva), partial mRNA	XM_004913335.1	238.11
Xenopus laevis hypothetical protein LOC100127277 (cDNA clone MGC:196106 IMAGE:9041013)	BC169379.1	237.55

Gene	Accession	FPKM
Xenopus laevis MGC85550 protein (cDNA clone MGC:85550 IMAGE:6881408)	BC078605.1	236.54
Xenopus laevis pyruvate dehydrogenase E1 alpha 1 (cDNA clone MGC:132282 IMAGE:3399753)	BC106670.1	236.24
Xenopus laevis anterior gradient 1 (ag1-b)	NM_001088200.1	233.85
Xenopus (Silurana) tropicalis tubulin, alpha 1a (tuba1a)	NM_203798.1	233.54
Xenopus laevis small nuclear ribonucleoprotein D2 polypeptide 16.5kDa (snrpd2)	NM_001174027.1	232.69
Xenopus laevis cDNA clone IMAGE:4173903	BC083045.1	232.45
Xenopus tropicalis finished cDNA, clone TGas068j09	CT025399.2	232.41
PREDICTED: Xenopus (Silurana) tropicalis cingulin-like (LOC100488374), transcript variant X1	XM_004917136.1	232.28
Xenopus (Silurana) tropicalis clone IMAGE:7794105 kinetochore component (KNL1) mRNA	JF804775.1	232.07
Xenopus laevis hypothetical protein LOC100137623 (cDNA clone IMAGE:5440018), partial cds	BC142545.1	231.97
Xenopus laevis hypothetical protein LOC443682 (cDNA clone IMAGE:5542496)	BC041544.1	231.46
Xenopus laevis ribosomal protein L23 (rpl23)	NM_001092452.1	230.81
XLU86699 Xenopus laevis oocyte lectin mRNA	U86699.1	230.08
Xenopus laevis ribosomal protein L4-b (cDNA clone MGC:64318 IMAGE:6876082)	BC054956.1	230.06
Xenopus tropicalis finished cDNA, clone TTpA078d11	CU025077.1	229.41
Xenopus laevis ribosomal protein S15a (rps15a)	NM_001096225.1	228.96
Xenopus laevis RNA-binding protein Y14 (rbm8)	NM_001090403.1	228.38
Xenopus tropicalis finished cDNA, clone TGas028j07	CT025482.2	228.37
Xenopus laevis 67kD laminin receptor precursor	AY730625.1	228.25
Xenopus laevis, clone IMAGE:4683795	BC044062.1	228.19
Xenopus laevis YTH domain containing 1 (ythdc1)	NM_001094824.1	227.64
Xenopus laevis proteasome (prosome, macropain) 26S subunit, ATPase, 4 (psmc4)	NM_001089831.1	226.85
Xenopus laevis STT3A, subunit of the oligosaccharyltransferase complex (catalytic) (stt3a)	NM_001090517.1	226.77
Xenopus laevis serine/threonine kinase 38 like (stk38l)	NM_001091259.1	226.7
Xenopus tropicalis myelin expression factor 2 (cDNA clone MGC:184667 IMAGE:7671690)	BC161028.1	226.05
Xenopus laevis cement gland-specific protein CGS (cDNA clone MGC:160454 IMAGE:8642735)	BC128956.1	225.71
Xenopus laevis, clone IMAGE:5542876	BC046379.1	225.21
Xenopus laevis CD81 protein (cd81-a)	NM_001086613.1	225.21
PREDICTED: Xenopus (Silurana) tropicalis zinc finger BED domain-containing protein 6-like (LOC101730718), transcript variant X1	XM_004919553.1	224.91
Xenopus laevis family with sequence similarity 3, member A (fam3a)	NM_001087002.1	223.79
PREDICTED: Xenopus (Silurana) tropicalis tripartite motif containing 29 (trim29), transcript variant X2	XM_004916119.1	223.78
Xenopus laevis hypothetical protein LOC414678 (cDNA clone MGC:83086 IMAGE:6316990)	BC071149.1	221.28
Xenopus laevis protein phosphatase methylesterase 1 (ppme1)	NM_001093413.1	220.93
Xenopus laevis solute carrier family 2 (facilitated glucose transporter), member 12 (slc2a12)	NM_001096426.1	220.63
Xenopus laevis keratin 18 (krt18-a)	NM_001088985.1	220.61
Xenopus laevis similar to procollagen-proline, 2-oxoglutarate 4-dioxygenase (proline 4-hydroxylase)	BC046736.1	219.44
Xenopus laevis zinc finger and BTB domain containing 8B (zbtb8b)	NM_001087386.1	219.29
Xenopus laevis uncharacterized protein MGC115605 (MGC115605)	NM_001096076.1	218.44
Xenopus laevis death-associated protein kinase 2 (dapk2)	NM_001095452.1	216.69
Xenopus laevis translation initiation factor eIF4A II (cDNA clone IMAGE:4887970), partial cds	BC045237.1	216.68
Xenopus laevis MGC82844 protein (MGC82844)	NM_001092466.1	216.42
Xenopus laevis high mobility group nucleosomal binding domain 3 (hmgn3-b)	NM_001088702.1	216.33
Xenopus (Silurana) tropicalis coiled-coil domain containing 81 (ccdc81)	NM_001126915.1	216.24
TPA inf: Xenopus laevis RTN1.1-C (RTN1) mRNA	BK004007.1	216.17
Xenopus laevis Int-6 protein mRNA	AF162775.1	215.6
Xenopus laevis cDNA clone MGC:83396 IMAGE:7008271	BC075193.1	214.65
Xenopus laevis uncharacterized LOC100101301 (LOC100101301)	NM_001099887.1	214.44
Xenopus laevis cDNA clone IMAGE:6876600	BC054330.1	213.53
Xenopus laevis mago-nashi homolog, proliferation-associated (magoh)	NM_001086255.1	213.19
Xenopus laevis glycosylphosphatidylinositol anchor attachment 1 (gpaa1)	NM_001089943.1	213.17
Xenopus (Silurana) tropicalis ribosomal protein L37 (rpl37)	NM_001113853.1	212.9
Xenopus laevis cDNA clone IMAGE:4965486	BC130061.1	212.61
Xenopus laevis CCAAT box transcription factor p122 subunit (CBTF122) mRNA	AY008299.1	212.46
Xenopus (Silurana) tropicalis ER membrane protein complex subunit 7 (emc7)	NM_001079099.1	211.83
Xenopus laevis Sin3A-associated protein, 18kDa (sap18)	NM_001095804.1	211.38
Xenopus laevis DnaJ (Hsp40) homolog, subfamily A, member 2 (dnaja2)	NM_001087156.1	211.38
Xenopus laevis calnexin (canx-b)	NM_001085946.1	210.66
Xenopus laevis annexin A7 (anxa7)	NM_001094206.1	210.52
Xenopus laevis uncharacterized protein MGC68822 (MGC68822)	NM_001090083.1	210.38
Xenopus tropicalis finished cDNA, clone TEgg006b24	CT025277.2	209.9
Xenopus laevis huntingtin interacting protein K (hypk)	NM_001093721.1	209.4
Xenopus laevis MGC78899 protein (cDNA clone IMAGE:5543041)	BC041552.1	208.56
PREDICTED: Xenopus (Silurana) tropicalis dynein, cytoplasmic 2, heavy chain 1 (dync2h1)	XM_002934933.2	207.71
PREDICTED: Xenopus (Silurana) tropicalis serine-rich adhesin for platelets-like (LOC100485162)	XM_002934057.2	207.39
Xenopus laevis hypothetical LOC495462 (cDNA clone IMAGE:4724777)	BC057725.1	207.1
Xenopus laevis Xepsin mRNA for epidermis specific serine protease	AB018694.1	207.05
Xenopus laevis signal sequence receptor, alpha (translocon-associated protein alpha) (ssr1)	NM_001086452.1	206.57
Xenopus laevis solute carrier family 35 (UDP-N-acetylglucosamine (UDP-GlcNAc) transporter), member A3, gene 2 (slc35a3.2)	NM_001086268.1	206.23
Xenopus laevis splicing factor 3b, subunit 5, 10kDa (sf3b5)	NM_001171762.1	205.96
Xenopus laevis ATP synthase, H+ transporting, mitochondrial F1 complex, beta polypeptide (atp5b)	NM_001086657.1	204.96
X.laevis XFG 5-2 mRNA for zinc finger protein	X70678.1	204.79
PREDICTED: Xenopus (Silurana) tropicalis NXPE family member 2-like (LOC100495388)	XM_002941335.2	204.78
PREDICTED: Xenopus (Silurana) tropicalis sodium-dependent proline transporter-like (LOC100490052)	XM_002942712.2	204.59
Xenopus laevis profilin 1 (pfn1)	NM_001096625.1	203.55
Xenopus tropicalis finished cDNA, clone TNeu075k21	CR761923.2	202.63
Xenopus laevis SEC13 homolog (sec13)	NM_001086973.1	202.57
X.laevis mRNA for mRNA for NM23/nucleoside diphosphate kinase, subunit A1	X97900.1	201.44
Xenopus laevis beclin 1, autophagy related (beclin1)	NM_001092282.1	200.66
Xenopus laevis keratin 19 (krt19)	NM_001091523.1	200.1
Xenopus laevis kaptin (actin binding protein) (kptn)	NM_001091722.1	199.68
Xenopus laevis solute carrier family 16, member 3 (monocarboxylic acid transporter 4) (slc16a3-b)	NM_001092618.1	199.54
X.laevis mRNA for rat translocon-associated protein delta homolog	X90584.1	199.1
Xenopus laevis telomeric repeat binding factor 1 mRNA	AF525882.2	198.87
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100498593 (LOC100498593), transcript variant X2	XM_004915063.1	198.57
Xenopus (Silurana) tropicalis stress-associated endoplasmic reticulum protein 1 (serp1)	NM_203754.1	197.72
Xenopus laevis mRNA similar to envelopakin (cDNA clone IMAGE:5571064)	BC045116.1	197.02
Xenopus laevis transformer 2 beta homolog (tra2b)	NM_001088933.1	196.4
Xenopus laevis oligosaccharyltransferase complex subunit (non-catalytic) (ostc-a)	NM_001092333.1	196.05
Xenopus laevis cDNA clone IMAGE:6877235	BC082926.1	195.71
Xenopus laevis uncharacterized protein MGC68479 (MGC68479)	NM_001090016.1	194.78
Xenopus laevis zinc finger protein 207 (znf207)	NM_001086855.1	194.44
Xenopus laevis basic leucine zipper and W2 domains 1 (bzw1)	NM_001086822.1	194

Gene	Accession	FPKM
Xenopus laevis ribosomal protein S28 pseudogene 9 (rps28p9)	NM_001093885.1	192.9
Xenopus laevis F1B (cDNA clone IMAGE:5543421), partial cds	BC045093.1	192.81
Xenopus laevis eukaryotic translation initiation factor 3, subunit D (eif3d)	NM_001087118.1	192.62
Xenopus (Silurana) tropicalis tyrosine 3-monoxygenase/tryptophan 5-monoxygenase activation protein, epsilon polypeptide (ywhae)	NM_001008155.1	192.46
Xenopus laevis liver RNA helicase II/Gu mRNA	AF302423.2	192.06
Xenopus laevis similar to RAN binding protein 7 (cDNA clone MGC:52556 IMAGE:5571758)	BC046568.1	191.53
Xenopus (Silurana) tropicalis eukaryotic translation initiation factor 4B (eif4b)	NM_001005064.1	191.48
Xenopus laevis cDNA clone 23G1-2	AJ009297.1	190.43
Xenopus laevis uncharacterized protein MGC83900 (MGC83900)	NM_001091294.1	190.09
Xenopus laevis uncharacterized protein MGC68910 (MGC68910)	NM_001086379.1	188.82
XELCADH Xenopus laevis (clone: XTCAD-1) cadherin gene	L29057.1	188.59
Xenopus laevis acid phosphatase 5, tartrate resistant (acp5)	NM_001091758.1	187.92
Xenopus laevis guanine nucleotide binding protein (G protein), beta polypeptide 3 (gnb3-b)	NM_001086983.1	187.35
Xenopus (Silurana) tropicalis serine/arginine-rich splicing factor 5 (srsf5)	NM_001004783.1	187.3
PREDICTED: Xenopus (Silurana) tropicalis L-2-hydroxyglutarate dehydrogenase (l2hgdh), transcript variant 2	XM_002938125.2	186.67
Xenopus laevis 5'-3' exoribonuclease 2 (xrn2)	NM_001097942.1	186.29
Xenopus laevis ATPase, H <sup>+</sup> transporting, lysosomal 13kDa, V1 subunit G3 (atp6v1g3)	NM_001094939.1	185.93
Xenopus laevis Peptidyl-prolyl cis-trans isomerase FKBP1A-like (fkbp1a)	NM_001085913.1	184.37
Xenopus laevis RAB15, member RAS oncogene family (rab15)	NM_001090236.1	183.98
Xenopus laevis stress-associated endoplasmic reticulum protein 1 (serp1)	NM_001171777.1	182.92
Xenopus laevis calpain, small subunit 1 (capns1)	NM_001094441.1	182.89
Xenopus laevis ribosomal protein S29 (rps29)	NM_001171730.1	182.84
Xenopus laevis myosin 10 mRNA	AY616034.1	182.68
Xenopus (Silurana) tropicalis RAN binding protein 1 (ranbp1)	NM_203689.1	182.64
Xenopus laevis MGC80700 protein (MGC80700)	NM_001092290.1	182.18
Xenopus laevis cytohesin 1 (cyth1)	NM_001097054.1	182.07
Xenopus laevis guanine nucleotide binding protein (G protein), gamma 5 (gng5-b)	NM_001092487.1	181.91
Xenopus laevis eukaryotic translation initiation factor 2, subunit 2 beta, 38kDa (eif2s2)	NM_001096819.1	181.6
Xenopus laevis NOP10 ribonucleoprotein (nop10)	NM_001171790.1	180.75
PREDICTED: Xenopus (Silurana) tropicalis nuclear pore complex protein Nup160-like (LOC100498194), transcript variant X1	XM_004915406.1	180.68
Xenopus laevis XGB mRNA for GTP-binding protein	AB250408.1	180.53
Xenopus laevis NO27 mRNA	AB018189.1	180.45
Xenopus tropicalis hypothetical protein LOC779580 (cDNA clone IMAGE:7859548), partial cds	BC123995.1	180.1
XELA Xenopus laevis mRNA for ornithine decarboxylase antizyme	D32141.1	179.77
Xenopus laevis Translocon-associated protein subunit alpha-like (ssr1)	NM_001090515.1	179.63
Xenopus laevis protein disulfide isomerase family A, member 3 (pdia3)	NM_001086582.1	179.48
Xenopus laevis eukaryotic translation initiation factor 3, subunit M (eif3m)	NM_001086592.1	179.43
Xenopus tropicalis inositol (1,4,5) trisphosphate receptor type 2 (cDNA clone IMAGE:7685633), partial cds	BC168572.1	178.71
Xenopus tropicalis desmoplakin (cDNA clone IMAGE:7692386), partial cds	BC121309.1	177.43
Xenopus laevis hypothetical protein LOC446950 (cDNA clone IMAGE:7012159), partial cds	BC081273.1	176.75
Xenopus laevis Rho GTPase Cdc42 mRNA	AF275252.1	176.56
Xenopus laevis eukaryotic translation initiation factor 1A, X-linked (eif1ax)	NM_001092602.1	176.08
Xenopus laevis basigin (Ok blood group) (bsg)	NM_001096135.1	176.05
Xenopus laevis hypothetical protein LOC100174804 (cDNA clone IMAGE:4032912), partial cds	BC167499.1	175.84
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 638 (znf638)	XM_002936276.2	175.4
Xenopus laevis similar to nucleolar protein NOP5/NOP58 (cDNA clone IMAGE:7978699), partial cds	BC106291.1	175.16
X.laevis XLCL1 mRNA	Z14253.1	174.83
Xenopus laevis uncharacterized protein MGC83511 (MGC83511)	NM_001091637.1	174.71
Xenopus laevis capping protein (actin filament) muscle Z-line, beta (capzb)	NM_001086647.1	174.12
Xenopus tropicalis finished cDNA, clone TNeu057c05	CR848298.2	174.02
Xenopus laevis Hsp90beta mRNA	AY785160.1	173.93
Xenopus laevis SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily e, member 1	BC041216.1	173.85
Xenopus laevis smg-9 homolog, nonsense mediated mRNA decay factor (smg9)	NM_001097020.1	173.71
Xenopus laevis myelin expression factor 2 (myef2)	NM_001096673.1	173.46
Xenopus laevis transcription elongation factor B (SII), polypeptide 2 (18kDa, elongin B) (tceb2)	NM_001086945.1	173.21
Xenopus laevis degenerative spermatocyte homolog 3, lipid desaturase (degs3)	NM_001087376.1	172.85
Xenopus laevis signal recognition particle 19kDa (srp19)	NM_001094752.1	172.8
Xenopus laevis cytoskeleton-associated protein 4 (ckap4)	NM_001095897.1	171.81
Xenopus laevis enhancer of rudimentary homolog (erh-b)	NM_001171729.1	171.74
Xenopus laevis uncharacterized LOC100037078 (LOC100037078)	NM_001097802.1	171.73
Xenopus laevis TAR DNA binding protein (tardbp)	NM_001087168.1	170.97
Xenopus laevis uncharacterized protein MGC115332 (MGC115332)	NM_001095924.1	170.78
Xenopus laevis guanine nucleotide binding protein-like 2 (nucleolar) (gnl2)	NM_001087044.1	170.54
Xenopus laevis trk-fused gene (tfg-b)	NM_001086952.1	169.49
Xenopus laevis tubulin, alpha 6 (tuba6)	NM_001087320.1	169.44
Xenopus laevis cDNA clone MGC:160189 IMAGE:8460550	BC129528.1	169.41
Xenopus laevis epo mRNA for erythropoietin receptor homologue	AB189477.1	169.02
Xenopus laevis citrate synthase (cs), nuclear gene encoding mitochondrial protein	NM_001086725.1	168.69
Xenopus laevis uncharacterized protein MGC81333 (MGC81333)	NM_001091069.1	167.6
Xenopus laevis synaptotagmin 1 (syt1)	NM_001094138.1	167.29
XLU23535 Xenopus laevis epithelial sodium channel, alpha subunit (alphaxENaC) mRNA	U23535.1	166.84
Xenopus laevis endothelin converting enzyme-1 (ECE-1) mRNA	EF127992.1	166.5
Xenopus laevis prothymosin, alpha (ptma-b)	NM_001094011.1	165.4
Xenopus laevis outer dense fiber of sperm tails 2 (odf2)	NM_001095273.1	165.1
Xenopus laevis eukaryotic translation initiation factor 3, subunit I (eif3i)	NM_001094195.1	164.94
Xenopus (Silurana) tropicalis ataxin 2-like (atxn2l)	NM_001142213.1	164.92
Xenopus laevis dolichyl-diphospholigosaccharide--protein glycosyltransferase subunit (non-catalytic) (ddost)	NM_001092387.1	164.91
Xenopus laevis cDNA clone 17G2	AJ009293.1	164.15
XLU19253 Xenopus laevis/gilli complement component C3 mRNA, partial cds	U19253.1	163.52
Xenopus laevis uncharacterized protein MGC68696 (MGC68696)	NM_001089779.1	163.29
Xenopus laevis ornithine decarboxylase-2 (cDNA clone MGC:52527 IMAGE:5542547)	BC047954.1	162.87
Xenopus laevis xpo mRNA, complete sequence	DOQ96856.1	162.71
Xenopus laevis sulfotransferase family, cytosolic, 2B, member 1 (sult2b1)	NM_001097677.1	162.48
Xenopus laevis tyrosine 3-monoxygenase/tryptophan 5-monoxygenase activation protein, epsilon polypeptide (ywhae)	NM_001087236.1	162.48
Xenopus laevis transmembrane 9 superfamily member 2 (tm9sf2)	NM_001095114.1	161.72
Xenopus laevis hypothetical protein LOC100137647 (cDNA clone IMAGE:8328787), partial cds	BC157450.1	161.44
Xenopus laevis DEAD (Asp-Glu-Ala-Asp) box polypeptide 3, X-linked (ddx3x)	NM_001086814.1	161.07
Xenopus (Silurana) tropicalis metadherin (mtdh)	NM_203833.1	160.8
Xenopus laevis RNA helicase II/Gu (cDNA clone MGC:130693 IMAGE:7975823)	BC108448.1	160.77
Xenopus laevis solute carrier family 18 (vesicular monoamine), member 1 (slc18a1)	NM_001085945.1	160.36
Xenopus laevis MGC80186 protein (MGC80186)	NM_001092014.1	160.34
Xenopus laevis ubiquinol-cytochrome c reductase, complex III subunit VII, 9.5kDa (uqcrc1), nuclear gene encoding mitochondrial protein	NM_001171758.1	160.34

Gene	Accession	FPKM
Xenopus laevis mRNA for TAF-Ibeta1	AB022691.1	160.23
Xenopus laevis acidic (leucine-rich) nuclear phosphoprotein 32 family, member B (anp32b)	NM_001085964.1	159.93
Xenopus laevis signal peptidase complex subunit 2 homolog (spcs2)	NM_001098688.1	159.74
Xenopus laevis PERP, TP53 apoptosis effector (perp)	NM_001092762.1	159.73
Xenopus laevis cDNA clone MGC:30731 IMAGE:7983334	BC108459.1	159.65
Xenopus tropicalis finished cDNA, clone TNeu031k16	CR926181.2	159.23
Xenopus laevis G patch domain and ankyrin repeats 1 (gpank1)	NM_001097131.1	158.84
Xenopus laevis hypothetical LOC494716 (cDNA clone IMAGE:4898644), partial cds	BC082657.1	158.79
Xenopus laevis tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, beta polypeptide (ywhab-a)	NM_001172053.1	158.73
Xenopus laevis Eukaryotic translation initiation factor 3 subunit E-A (cDNA clone MGC:160178 IMAGE:8737627)	BC128927.1	158.56
Xenopus laevis ring finger protein 20, E3 ubiquitin protein ligase (rnf20)	NM_001112861.1	158.34
Xenopus laevis SET and MYND domain containing 1 (smyd1)	NM_001091994.1	158.33
Xenopus laevis epid41 mRNA, complete sequence	DQ096906.1	158.28
Xenopus laevis Baf57 mRNA	AF487782.1	158.23
Xenopus laevis elongation factor 1 beta (cDNA clone MGC:130878 IMAGE:7210653)	BC106340.1	157.65
Xenopus laevis eukaryotic translation initiation factor 2, subunit 3 gamma, 52kDa (eif2s3)	NM_001087096.1	157.35
Xenopus laevis nestin (nes)	NM_001087857.1	157.28
Xenopus laevis, Similar to S-100 related protein, clone 42C, clone IMAGE:4682756	BC041295.1	157.19
Xenopus (Silurana) tropicalis transformer 2 beta homolog (tra2b)	NM_001006877.1	156.84
Xenopus laevis hypothetical protein LOC100127337 (cDNA clone IMAGE:7980696), partial cds	BC155957.1	156.61
Xenopus laevis basic transcription factor 3 (btf3)	NM_001094887.1	156.56
Xenopus laevis MGC82181 protein (MGC82181)	NM_001091894.1	156.19
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730819 (LOC101730819)	XM_004915204.1	155.77
Xenopus laevis epithelial splicing regulatory protein 1 (esrp1)	NM_001086055.1	155.61
Xenopus laevis socrin (sri)	NM_001097890.1	155.16
Xenopus tropicalis finished cDNA, clone TNeu066d19	CR760077.2	154.95
Xenopus laevis tubulin, beta 4A class IVa (tubb4a)	NM_001086203.1	154.81
Xenopus tropicalis finished cDNA, clone TGas113h12	CR761753.2	154.47
Xenopus laevis ribophorin II (rpn2)	NM_001086192.1	154.41
Xenopus laevis brain protein 44-like (brp44l-b)	NM_001171779.1	153.82
Xenopus laevis mRNA for nucleoporin CAN (CAN gene)	AJ243889.1	153.78
Xenopus laevis dynein, light chain, LC8-type 1 (dynll1-a)	NM_001171695.1	153.35
XELIIANX1 X.laevis annexin II type 1 mRNA	M60768.1	152.8
Xenopus laevis hypothetical protein LOC443623 (cDNA clone IMAGE:5156152), partial cds	BC073076.1	152.63
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730873 (LOC101730873)	XM_004911776.1	152.41
Xenopus laevis cDNA clone IMAGE:5515566, partial cds	BC073639.1	152.38
Xenopus (Silurana) tropicalis transmembrane emp24 domain trafficking protein 2 (tmmed2)	NM_001045776.1	152.35
Xenopus laevis alpha-tubulin (cDNA clone MGC:114676 IMAGE:5572530)	BC097546.1	151.96
Xenopus laevis caveolin-3 (Cav-3) mRNA	AF455044.1	151.8
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733049 (LOC101733049)	XM_004916041.1	151.34
Xenopus laevis MGC80429 protein (MGC80429)	NM_001092090.1	151.19
Xenopus laevis MGC82380 protein (cDNA clone MGC:154259 IMAGE:8077010)	BC122514.1	150.93
Xenopus (Silurana) tropicalis eukaryotic translation initiation factor 3, subunit F (eif3f)	NM_001016190.2	150.84
Xenopus laevis ADP-ribosylation factor-like 1 (arl1)	NM_001092343.1	150.49
Xenopus laevis chromosome 20 open reading frame 24 (c20orf24)	NM_001093217.1	150.46
Xenopus (Silurana) tropicalis basic transcription factor 3 (btf3)	NM_001016697.2	150.14
Xenopus laevis transmembrane tight junction protein claudin (cla)	NM_001088604.1	149.94
Xenopus laevis similar to deoxyribonuclease I-like 3 (cDNA clone IMAGE:5570084), partial cds	BC043839.1	149.93
Xenopus laevis uncharacterized LOC100158313 (LOC100158313)	NM_001127765.1	149.04
Xenopus laevis hypothetical protein LOC100049142 (cDNA clone IMAGE:6867315), partial cds	BC134825.1	148.96
Xenopus (Silurana) tropicalis Rtf1, Paf1/RNA polymerase II complex component, homolog (rtf1)	NM_001005068.1	148.92
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734416 (LOC101734416), transcript variant X1	XM_004912947.1	148.78
Xenopus laevis metastasis associated 1 (mta1)	NM_001094428.1	148.76
Xenopus laevis uncharacterized protein MGC52856 (MGC52856)	NM_001085890.1	148.75
Xenopus laevis casein kinase 2, beta polypeptide (csnk2b)	NM_001090657.1	148.56
Xenopus laevis uncharacterized LOC446286 (LOC446286)	NM_001092994.1	148.36
Xenopus laevis scellin (scel)	NM_001092354.1	148.18
Xenopus laevis brsc02 mRNA, complete sequence	DQ096983.1	148.01
Xenopus (Silurana) tropicalis SERPINE1 mRNA binding protein 1 (serbp1), transcript variant 1	NM_001001227.2	147.14
Xenopus laevis ornithine decarboxylase antizyme 1 (oaz1)	NM_001086576.1	146.15
Xenopus laevis cDNA clone IMAGE:3745226	BC100243.1	145.84
Xenopus laevis transmembrane emp24 protein transport domain containing 9 (tmmed9)	NM_001094622.1	145.81
Xenopus laevis MGC84169 protein (alref-b)	NM_001092746.1	145.48
Xenopus laevis MGC80893 protein (MGC80893)	NM_001093575.1	144.9
Xenopus laevis mRNA for calpastatin (calp3 gene)	AJ271209.2	144.77
Xenopus laevis solute carrier family 30 (zinc transporter), member 8 (slc30a8)	NM_001095453.1	144.64
Xenopus laevis basic transcription factor 3-like 4 (btf3l4)	NM_001096139.1	144.41
Xenopus laevis MKI67 FHA domain-interacting nucleolar phosphoprotein-like (cDNA clone IMAGE:6878823)	BC054288.1	144.35
Xenopus laevis uncharacterized protein MGC81054 (MGC81054)	NM_001091253.1	144.27
Xenopus laevis epco01 mRNA, complete sequence	DQ096854.1	144.07
PREDICTED: Xenopus (Silurana) tropicalis carcinoembryonic antigen-related cell adhesion molecule 8-like (LOC100493418), partial mRNA	XM_002942459.2	143.78
Xenopus laevis splicing factor 3B, 14 kDa subunit (sf3b14)	NM_001086471.1	143.72
Xenopus laevis uncharacterized LOC495270 (LOC495270)	NM_001094944.1	143.12
Xenopus laevis epid18 mRNA, complete sequence	DO096907.1	142.6
Xenopus laevis mitochondrial pyruvate carrier 2 (mpc2), nuclear gene encoding mitochondrial protein	NM_001086062.1	142.56
Xenopus laevis ribosomal protein L7-like 1 (rlp7l1)	NM_001095013.1	142.22
Xenopus laevis G protein pathway suppressor 2 (gps2)	NM_001086717.1	141.95
Xenopus tropicalis cDNA clone IMAGE:7719339	BC121654.1	141.87
PREDICTED: Xenopus (Silurana) tropicalis periplakin (ppl)	XM_002941048.2	141.85
Xenopus laevis KDEL (Lys-Asp-Glu-Leu) endoplasmic reticulum protein retention receptor 1 (kdelr1)	NM_001087169.1	141.8
Xenopus laevis ribosomal protein L31 (rlp31)	NM_001091241.1	141.7
Xenopus laevis coatomer protein complex, subunit epsilon (cope)	NM_001091858.1	141.22
Xenopus laevis malate dehydrogenase 1, NAD (soluble) (mdh1)	NM_001089866.1	140.92
Xenopus laevis transcription elongation factor B (SII), polypeptide 1 (15kDa, elongin C) (tcbe1)	NM_001086659.1	140.88
Xenopus laevis hypothetical protein LOC446963 (cDNA clone IMAGE:6872282), partial cds	BC078469.1	140.84
Xenopus laevis U1 70K (cDNA clone MGC:130688 IMAGE:7976400)	BC108447.1	140.71
Xenopus (Silurana) tropicalis h/ACA ribonucleoprotein complex subunit 1 (gar1)	NM_001011252.1	140.09
Xenopus laevis XGATA-2 protein (cDNA clone MGC:131004 IMAGE:7978680)	BC108544.1	140.06
Xenopus laevis ATPase, H+ transporting, lysosomal 13kDa, V1 subunit G1 (atp6v1g1)	NM_001086967.1	140.04
Xenopus laevis TAF13 RNA polymerase II, TATA box binding protein (TBP)-associated factor, 18kDa (taf13)	NM_001095765.1	140.02
XELTM4ISO Xenopus laevis non-muscle tropomyosin (TM-4) mRNA	L35238.1	139.91
Xenopus laevis Dnaj (Hsp40) homolog, subfamily A, member 2 (dnaja2)	NM_001087156.1	139.7

Gene	Accession	FPKM
Xenopus laevis cDNA clone MGC:154541 IMAGE:8320906	BC124896.1	139.54
Xenopus laevis glycerol-3-phosphate dehydrogenase 1 (soluble) (gpd1)	NM_001090509.1	139.43
Xenopus laevis dynamin 1-like (dnm1)	NM_001086714.1	139.38
Xenopus laevis uncharacterized protein MGC68811 (MGC68811)	NM_001090098.1	138.95
Xenopus laevis ER membrane protein complex subunit 6 (emc6)	NM_001114827.1	138.88
Xenopus laevis ribosomal protein S27 (rps27)	NM_001087225.1	138.15
Xenopus laevis lamin B1 (lmbn1)	NM_001086584.1	137.99
Xenopus laevis GRAM domain containing 1C (gramd1c)	NM_001091428.1	137.73
Xenopus laevis NHP2 non-histone chromosome protein 2-like 1 (nhp2l1-b)	NM_001087255.1	137.59
Xenopus laevis serine peptidase inhibitor, Kunitz type, 2 (spint2)	NM_001096173.1	137.45
Xenopus laevis small nuclear ribonucleoprotein D1 polypeptide 16kDa (snrp1)	NM_001091853.1	137.32
Xenopus laevis zinc finger protein 326 (znf326)	NM_001097844.1	137.19
Xenopus laevis vesicle-associated membrane protein-associated protein B/C (LOC495400)	NM_001090509.1	137.17
Xenopus laevis SPRY domain containing 3 (spry3)	NM_001091822.1	136.26
Xenopus laevis cDNA clone MGC:180066 IMAGE:6632424	BC160763.1	136.22
Xenopus laevis small ubiquitin-like modifier 2 (sumo2-a)	NM_001086616.1	136.03
Xenopus laevis GA binding protein transcription factor, beta subunit 2 (gabpb2)	NM_001093038.1	135.78
Xenopus laevis small nuclear ribonucleoprotein polypeptide A (snrpa)	NM_001086620.1	135.37
Xenopus laevis deoxyribonuclease gamma mRNA	AF059612.1	135.29
Xenopus laevis histocompatibility (minor) 13 (hmc13)	NM_001087405.1	135.25
Xenopus laevis prostaglandin E synthase 3 (cytosolic) (ptges3)	NM_001087139.1	135.15
Xenopus laevis serrate RNA effector molecule homolog (srrt-b)	NM_001089063.1	135.11
Xenopus laevis neurexophilin and PC-esterase domain family, member 2 (nxpe2)	NM_001095145.1	135.03
Xenopus laevis calpain 1, (mu/l) large subunit (capn1)	NM_001093584.1	134.64
PREDICTED: Xenopus (Silurana) tropicalis chromosome X open reading frame 58 (cxorf58)	XM_002935189.2	134.62
Xenopus (Silurana) tropicalis ADP-ribosylation factor 5 (arf5)	NM_203687.1	134.55
Xenopus laevis coactivator-associated arginine methyltransferase 1 (carm1)	NM_001094676.1	134.39
Xenopus laevis UDP-glucose dehydrogenase (cDNA clone MGC:196075 IMAGE:9040982)	BC169348.1	134.09
Xenopus laevis eukaryotic translation initiation factor 3, subunit B (eif3b)	NM_001096535.1	133.88
Xenopus tropicalis transmembrane emp24 domain containing 1 (cDNA clone MGC:146643 IMAGE:7660000)	BC123004.1	133.35
Xenopus laevis selenoprotein T (selt)	NM_001086037.1	133.09
Xenopus laevis cell division cycle 34 (cdc34)	NM_001086142.1	132.82
Xenopus laevis guanine nucleotide binding protein-like 3 (nucleolar) (gnl3)	NM_001087179.1	132.74
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100488736 (LOC100488736), transcript variant X2	XM_004916677.1	131.81
Xenopus laevis cDNA clone IMAGE:3401392, containing frame-shift errors	BC070522.1	131.75
Xenopus laevis spermatid perinuclear RNA binding protein (strbp)	NM_001093702.1	131.42
Xenopus laevis Ferritin light chain, oocyte isoform (MGC68606)	NM_001086458.1	131.29
Xenopus laevis anocatinin 5 (ano5)	NM_001093341.1	131.08
PREDICTED: Xenopus (Silurana) tropicalis protein kinase C-like 2-like (LOC101733770)	XM_004911451.1	130.81
Xenopus laevis RAB10, member RAS oncogene family (rab10)	NM_001089799.1	130.49
Xenopus laevis scaffold attachment factor A (cDNA clone MGC:79920 IMAGE:6866280)	BC084742.1	130.21
Xenopus laevis H3 histone, family 3A (h3f3a)	NM_001098432.1	130.04
Xenopus laevis ribosomal protein L37 (rpl37)	NM_001092508.1	130.03
Xenopus laevis solute carrier family 3 (activators of dibasic and neutral amino acid transport), member 2 (slc3a2)	NM_001086777.1	129.99
Xenopus laevis ankyrin repeat and BTB (POZ) domain containing 2 (abtb2)	NM_001094801.1	129.81
Xenopus laevis mRNA for hnRNP-E2 protein	AJ243591.1	129.81
Xenopus laevis Sp1 transcription factor (sp1)	NM_001091419.1	129.53
Xenopus laevis MGC80472 protein (MGC80472)	NM_001092221.1	129.41
Xenopus laevis solute carrier family 39 (zinc transporter), member 7 (slc39a7)	NM_001127819.1	129.28
Xenopus laevis pescadillo ribosomal biogenesis factor 1 (pes1)	NM_001087088.1	129.05
Xenopus laevis villin-like (vill)	NM_001086147.1	128.97
Xenopus laevis alpha(E)-catenin (cDNA clone IMAGE:6631200), partial cds	BC110961.1	128.95
Xenopus laevis cDNA clone MGC:82639 IMAGE:5049047	BC073031.1	128.84
Xenopus laevis hypothetical protein LOC100036868 (cDNA clone IMAGE:8464182), partial cds	BC129563.1	128.77
Xenopus laevis proteasome (prosome, macropain) subunit, beta type, 1 (psmb1)	NM_001086966.1	128.73
Xenopus laevis zinc finger protein 622 (znf622-b)	NM_001087081.1	128.7
Xenopus laevis nuclear casein kinase and cyclin-dependent kinase substrate 1 (nucks1)	NM_001091242.1	128.6
Xenopus laevis GDP dissociation inhibitor 2 (cDNA clone MGC:82555 IMAGE:4968535)	BC078017.1	128.42
Xenopus (Silurana) tropicalis prostaglandin E synthase 3 (cytosolic) (ptges3)	NM_001007104.1	128.33
Xenopus laevis coatomer protein complex, subunit gamma 1 (copg1)	NM_001090106.1	128.19
Xenopus laevis Rab acceptor 1 (prenylated) (rabac1-b)	NM_001094533.1	128.11
Xenopus laevis eukaryotic translation initiation factor 3, subunit L (eif3l)	NM_001086876.1	128.04
Xenopus laevis coatomer protein complex, subunit alpha (copa)	NM_001093019.1	127.41
Xenopus laevis ribosomal protein S8 (rps8)	NM_001087374.1	127.23
Xenopus tropicalis finished cDNA, clone TNeu055d12	CR942696.2	127.1
Xenopus laevis splicing factor 3b, subunit 2, 145kDa (sf3b2)	NM_001095946.1	126.89
Xenopus laevis 14-3-3 protein zeta (cDNA clone MGC:64423 IMAGE:6879629)	BC056850.1	126.51
Xenopus laevis RAB6A, member RAS oncogene family (rab6a)	NM_001087037.1	126.51
Xenopus laevis junction plakoglobin (jup-a)	NM_001089007.1	126.45
Xenopus (Silurana) tropicalis solute carrier family 16, member 3 (monocarboxylic acid transporter 4) (slc16a3)	NM_001007911.1	126.4
Xenopus laevis MGC80314 protein (MGC80314)	NM_001092053.1	125.65
Xenopus laevis solute carrier family 16, member 12 (monocarboxylic acid transporter 12) (slc16a12)	NM_001092655.1	125.48
Xenopus laevis xnf7 protein (cDNA clone MGC:181714 IMAGE:8821190)	BC155882.1	125.34
Xenopus laevis hypothetical protein LOC733296 (cDNA clone IMAGE:6323284), partial cds	BC099011.1	125.13
Xenopus tropicalis finished cDNA, clone TTpA012b11	CR760877.2	125.08
Xenopus laevis cytochrome c oxidase subunit IIb (cox7b), nuclear gene encoding mitochondrial protein	NM_001093985.1	124.9
Xenopus laevis hypothetical protein LOC431876 (cDNA clone IMAGE:4175265), partial cds	BC160685.1	124.81
Xenopus (Silurana) tropicalis small nuclear ribonucleoprotein 70kDa (U1) (snrnp70)	NM_001008125.2	124.78
Xenopus laevis pre-mRNA processing factor 3 (prpf3)	NM_001091804.1	124.7
Xenopus laevis ATP-binding cassette, sub-family F (GCN20), member 2 (abcf2), nuclear gene encoding mitochondrial protein	NM_001086598.1	124.35
PREDICTED: Xenopus (Silurana) tropicalis AT rich interactive domain 1A (SWI-like) (arid1a), transcript variant 1	XM_002934639.2	124.31
Xenopus laevis uncharacterized LOC100037062 (LOC100037062)	NM_001097788.1	123.67
Xenopus laevis beta-catenin protein (cDNA clone MGC:132060 IMAGE:4755420)	BC108764.1	123.66
Xenopus laevis proteasome (prosome, macropain) subunit, beta type, 7 (psmb7)	NM_001094066.1	123.63
Xenopus laevis uncharacterized protein MGC80175 (MGC80175)	NM_001091684.1	123.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734517 (LOC101734517)	XM_004910341.1	122.98
Xenopus tropicalis finished cDNA, clone TNeu107m13	CU075760.1	122.52
Xenopus tropicalis finished cDNA, clone TtbA040d21	CU025123.1	122
XLU15444 Xenopus laevis beta 5 tubulin mRNA	U15444.1	121.75
Xenopus laevis hypothetical protein LOC443583 (cDNA clone IMAGE:4970100), partial cds	BC072805.1	121.39
Xenopus laevis ribosomal L24 domain containing 1 (rsl24d1)	NM_001085972.1	121.38
Xenopus (Silurana) tropicalis WD repeat and SOCS box containing 1 (wsb1)	NM_204056.1	120.95

Gene	Accession	FPKM
Xenopus laevis p54nrb mRNA for 54 kD nuclear RNA-binding protein proteasome subunit XC3 [Xenopus laevis, 890 nt]	AB238228.1	120.94
Xenopus laevis hypothetical protein LOC100037232 (cDNA clone IMAGE:8548789), partial cds	S51111.1	120.83
Xenopus laevis ras homolog family member T1 (rhot1)	BC129782.1	120.71
Xenopus laevis dsRNA and RNA-DNA hybrid binding protein dsRBP-ZFa mRNA	NM_001127846.1	120.54
Xenopus laevis MGC80203 protein (MGC80203)	AF005083.1	120.49
Xenopus laevis Aly/REF export factor (alyref)	NM_001092020.1	120.29
Xenopus laevis eukaryotic translation elongation factor 1 epsilon 1 (eef1e1)	NM_001094972.1	120.2
Xenopus laevis casein kinase 2, alpha 1 polypeptide (csnk2a1)	NM_001090655.1	120.14
Xenopus laevis glutamine-fructose-6-phosphate transaminase 1 (gfpt1)	NM_001091267.1	119.73
Xenopus laevis G protein alpha subunit 14 mRNA	AF059182.1	119.72
Xenopus laevis transmembrane emp24 domain trafficking protein 2 (tmed2)	NM_001087150.1	119.71
Xenopus laevis Riddle 3 (rdd3)	NM_001135062.1	119.65
XLU58683 Xenopus laevis nuclear orphan receptor xGCNF mRNA	U58683.1	119.57
Xenopus laevis carbonyl reductase 1 (cbr1)	NM_001096272.1	119.54
Xenopus (Silurana) tropicalis cytochrome c oxidase subunit Va (cox5a), nuclear gene encoding mitochondrial protein	NM_001017049.3	119.49
Xenopus tropicalis bagpipe homeobox homolog-related (cDNA clone IMAGE:7879321), partial cds	BC125785.1	119.42
Xenopus laevis uncharacterized LOC100158314 (LOC100158314)	NM_001127766.1	119.37
Xenopus laevis signal recognition particle 72kDa (srp72)	NM_001092495.1	118.95
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 4 (anion exchanger), member 1, adaptor protein (slc4a1ap)	XM_002938421.2	118.89
Xenopus laevis hypothetical protein LOC100049130 (cDNA clone IMAGE:6867328), partial cds	BC133780.1	118.62
Xenopus (Silurana) tropicalis polypyrimidine tract binding protein 1 (ptbp1)	NM_001011140.1	118.45
Xenopus laevis cDNA clone MGC:79935 IMAGE:6865551	BC076717.1	118.43
Xenopus laevis Long-chain-fatty-acid-CoA ligase 1-like (MGC53832)	NM_001086196.1	118.27
Xenopus (Silurana) tropicalis envelopakin (evpl)	NM_001134819.1	118.26
Xenopus laevis splicing factor, arginine/serine-rich 6 (bsf2)	NM_001086679.1	118.1
Xenopus laevis heat shock factor binding protein 1 (hsbp1-a)	NM_001112810.1	117.96
PREDICTED: Xenopus (Silurana) tropicalis proline arginine rich coiled coil 1 (rprc1), transcript variant X1	XM_002936218.2	117.92
Xenopus laevis serine/arginine-rich splicing factor 4 (srsf4)	NM_001091912.1	117.87
Xenopus XA-1 mRNA, anterior and ectodermic-specific	X53821.1	117.67
Xenopus laevis uncharacterized LOC495046 (LOC495046)	NM_001094749.1	117.62
PREDICTED: Xenopus (Silurana) tropicalis fer-1-like protein 4-like (LOC101734097)	XM_004918505.1	117.58
Xenopus laevis poly(A) binding protein, cytoplasmic 1 (pabpc1-b)	NM_001091600.1	117.56
Xenopus laevis KIAA1109 (kiaa1109)	NM_001092862.1	117.25
Xenopus laevis eukaryotic translation initiation factor 3, subunit J (eif3j)	NM_001091733.1	117.23
Xenopus laevis oocyte RNA helicase II/Gu mRNA	AF302422.1	117.2
Xenopus laevis uncharacterized protein MGC130708 (MGC130708)	NM_001096817.1	117.1
Xenopus laevis epithelial sodium channel alpha subunit (cDNA clone MGC:179940 IMAGE:7011875)	BC161708.1	117.01
Xenopus laevis cnso01 mRNA, complete sequence	DQ096896.1	117.01
Xenopus laevis comp43 mRNA, complete sequence	DQ096849.1	116.84
Xenopus (Silurana) tropicalis ribosomal protein L29 (rpl29)	NM_001171676.1	116.73
Xenopus laevis LIM domain containing 2 (limd2-a)	NM_001092306.1	116.7
Xenopus laevis sorting nexin 18 (snx18-a)	NM_001094806.1	116.64
Xenopus laevis small nuclear ribonucleoprotein polypeptide G (snrpg)	NM_001171650.1	116.62
Xenopus laevis protein LLP homolog (llph)	NM_001086052.1	116.62
Xenopus (Silurana) tropicalis cytochrome P450, family 26, subfamily B, polypeptide 1 (cyp26b1)	NM_001079187.2	116.58
Xenopus laevis uncharacterized protein MGC83105 (MGC83105)	NM_001091275.1	116.49
PREDICTED: Xenopus (Silurana) tropicalis ribosomal protein S6 kinase alpha-3-like (LOC100496161)	XM_002936044.2	116.11
Xenopus laevis dynein, light chain, LC8-type 1 (dynll1-b)	NM_001092156.1	116.09
Xenopus (Silurana) tropicalis far upstream element (FUSE) binding protein 3 (fubp3)	NM_001097365.1	115.9
Xenopus laevis calcium/calmodulin-dependent protein kinase II gamma L subunit mRNA	AF233632.1	115.68
Xenopus laevis high density lipoprotein binding protein (viginin) (cDNA clone MGC:52662 IMAGE:4681587)	BC044314.1	115.54
Xenopus laevis voltage-dependent anion channel 3 (vdac3)	NM_001091887.1	114.69
Xenopus laevis transmembrane emp24 protein transport domain containing 7 (tmed7)	NM_001093965.1	114.69
Xenopus laevis uncharacterized LOC100049121 (LOC100049121)	NM_001097954.1	114.66
Xenopus laevis integrin beta-3 subunit (cDNA clone MGC:80757 IMAGE:5512565)	BC073343.1	114.39
Xenopus laevis T-complex protein 1 subunit alpha-like (MGC53348)	NM_001086097.1	113.99
Xenopus tropicalis transcription factor AP-2 alpha (activating enhancer binding protein 2 alpha)	BC135698.1	113.87
Xenopus laevis small nuclear ribonucleoprotein 27kDa (U4/U6.U5) (snrnp27)	NM_001092711.1	113.73
Xenopus laevis RNA binding motif protein 23 (rbm23)	NM_001092339.1	113.66
Xenopus laevis protein disulfide isomerase family A, member 6 (pdia6-b)	NM_001086836.1	113.65
Xenopus laevis uncharacterized LOC495465 (LOC495465)	NM_001095117.1	113.11
Xenopus laevis uncharacterized protein MGC85015 (MGC85015)	NM_001095857.1	112.64
Xenopus laevis fatty acid Coenzyme A ligase (cDNA clone MGC:132122 IMAGE:5085511)	BC108787.1	112.52
PREDICTED: Xenopus (Silurana) tropicalis CUB domain containing protein 1 (cdcp1)	XM_002938305.1	112.46
Xenopus laevis MGC81971 protein (cDNA clone IMAGE:5542873)	BC043899.1	112.36
Xenopus laevis uncharacterized LOC398959 (LOC398959)	NM_001090029.1	112.17
Xenopus laevis protein disulfide isomerase family A, member 6 (pdia6-a)	NM_001093174.1	112.01
Xenopus laevis mRNA for Id3 protein	AJ292558.1	111.92
Xenopus laevis cDNA clone IMAGE:6950416	BC094193.1	111.84
Xenopus laevis golgi membrane protein 1 (golm1)	NM_001094405.1	111.83
Xenopus laevis hypothetical protein MGC130860 (cDNA clone MGC:130860 IMAGE:7205580)	BC106331.1	111.73
Xenopus laevis cDNA clone IMAGE:3379429, partial cds	BC089277.1	111.72
Xenopus laevis elongation factor 1-alpha O (cDNA clone MGC:86322 IMAGE:7009531)	BC079786.1	111.58
Xenopus laevis, Similar to Ewing sarcoma breakpoint region 1, clone IMAGE:5543106	BC043904.1	111.58
PREDICTED: Xenopus (Silurana) tropicalis desmoglein 2 (dsg2)	XM_002934131.2	111.45
Xenopus laevis histone deacetylase 1 (hdac1-b)	NM_001085927.1	111.35
Xenopus laevis enhancer of rudimentary homolog (erh-a)	NM_001090354.1	111
Xenopus laevis uncharacterized LOC100037113 (LOC100037113)	NM_001097823.1	110.89
Xenopus (Silurana) tropicalis cystic fibrosis transmembrane conductance regulator (ATP-binding cassette sub-family C, member 7) (cfr)	XM_002934956.2	110.81
Xenopus laevis hypothetical protein LOC446231 (cDNA clone MGC:114833 IMAGE:4970962)	BC097603.1	110.71
Xenopus laevis TEA domain family member 4 (tead4)	NM_001094370.1	110.64
Xenopus laevis transmembrane protein 258 (tmem258)	NM_001092261.1	110.58
Xenopus laevis Ras association (RalGDS/AF-6) domain family member 2 (rassf2)	NM_001096747.1	110.44
Xenopus laevis RuvB-like protein 2 (rvbl2)	NM_001086931.1	110.18
Xenopus (Silurana) tropicalis UTP3, small subunit (SSU) processome component, homolog (utp3)	NM_001126577.1	109.96
Xenopus laevis heat shock 70kDa protein 5 (glucose-regulated protein, 78kDa) (hspa5)	NM_001086595.1	109.93
Xenopus laevis histidine ammonia-lyase, gene 1 (hal.1)	NM_001093175.1	109.6
Xenopus (Silurana) tropicalis DnaJ (Hsp40) homolog, subfamily A, member 2 (dnaja2)	NM_001004807.1	109.31
Xenopus laevis catenin (cDNA clone MGC:154290 IMAGE:8071429)	BC128668.1	109.25
Xenopus laevis jun B proto-oncogene (junb)	NM_001097035.1	109.08
Xenopus tropicalis MGC89417 protein (cDNA clone MGC:89417 IMAGE:6990058)	BC075526.1	109.01

Gene	Accession	FPKM
Xenopus tropicalis finished cDNA, clone TNeu115i02	CR848278.2	108.72
Xenopus laevis guanine nucleotide binding protein (G protein), beta polypeptide 3 (gnb3)	NM_001087217.1	108.69
Xenopus laevis cDNA clone MGC:80932 IMAGE:5516092	BC077950.1	108.48
PREDICTED: Xenopus (Silurana) tropicalis eukaryotic translation initiation factor 4 gamma, 3 (eif4g3), transcript variant X4	XM_004914400.1	108.48
Xenopus laevis LIM domain and actin binding 1 (lima1)	NM_001094929.1	108.48
Xenopus laevis cofilin 1 (non-muscle) (cfl1-a)	NM_001086102.1	108.42
Xenopus laevis cDNA clone IMAGE:6326815	BC097767.1	108.35
Xenopus laevis casein kinase 1, alpha 1 (csnk1a1)	NM_001087077.1	108.29
Xenopus laevis chaperonin containing TCP1, subunit 5 (epsilon) b (cct5b)	NM_001097075.1	107.99
Xenopus laevis retinol saturase (all-trans-retinol 13,14-reductase) (retsat)	NM_001122880.1	107.7
XELDG42A X.laevis D42 protein mRNA, and 5' flank	M22249.1	107.68
Xenopus tropicalis finished cDNA, clone TNeu07715	CR761948.2	107.66
Xenopus (Silurana) tropicalis NDRG family member 2 (ndrg2)	NM_001007897.1	107.53
Xenopus laevis hypothetical protein LOC398627 (cDNA clone IMAGE:6880461), partial cds	BC054305.1	107.41
PREDICTED: Xenopus (Silurana) tropicalis synaptotagmin binding, cytoplasmic RNA interacting protein (syncrip), transcript variant X7	XM_004914532.1	107.1
Xenopus laevis protein phosphatase 2, catalytic subunit, alpha isozyme (ppp2ca)	NM_001086884.1	106.73
Xenopus laevis bx24 mRNA	AF319877.1	106.64
Xenopus laevis cDNA clone IMAGE:6634171	BC098965.1	106.3
Xenopus laevis peptidylprolyl isomerase B (cyclophilin B) (ppib)	NM_001087036.1	106.27
Xenopus tropicalis finished cDNA, clone TGas135103	CR848092.2	106.22
Xenopus laevis retinal pigment epithelium-specific protein 65kDa (rpe65)	NM_001086800.1	106.05
PREDICTED: Xenopus (Silurana) sosondowah ankyrin repeat domain family member D (sowahd)	XM_004916878.1	105.86
Xenopus laevis jumonji C domain containing histone demethylase 1 homolog D (jhdmd1)	NM_001092110.1	105.61
Xenopus laevis cDNA clone IMAGE:6948748	BC094119.1	105.58
Xenopus laevis phosphoribosyl pyrophosphate synthetase 2 (prps2)	NM_001087186.1	105.54
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 850-like (LOC100493303), transcript variant X1	XM_002942342.2	105.28
Xenopus laevis cDNA clone MGC:68554 IMAGE:4031347	BC060335.1	105.18
Xenopus laevis nectin-2beta (LOC100505446)	NM_001197282.1	105.05
Xenopus laevis thioredoxin reductase 3 (txnrd3)	NM_001094191.1	104.89
Xenopus laevis small ubiquitin-like modifier 2 (sumo2-b)	NM_001092126.1	104.88
Xenopus laevis proline-rich Vg1 mRNA-binding protein mRNA	AY028920.1	104.85
Xenopus laevis poly(A) binding protein, nuclear 1 (pabpn1-a)	NM_001088588.1	104.74
X.laevis gene for thyroid hormone receptor beta (promoter region)	Z30971.1	104.59
Xenopus laevis cDNA clone MGC:82763 IMAGE:5156829	BC078036.1	104.54
Xenopus laevis nucleolar phosphoprotein (cDNA clone MGC:80737 IMAGE:5512141)	BC073328.1	104.53
Xenopus laevis MGC84462 protein (MGC84462)	NM_001092815.1	104.16
Xenopus laevis lipoprotein stimulated lipoprotein receptor (lsr)	NM_001092196.1	104
Xenopus laevis coagulation factor 9 (f9)	NM_001098678.1	103.95
Xenopus laevis hypothetical protein LOC398627 (cDNA clone MGC:130680 IMAGE:7979747)	BC108445.1	103.88
Xenopus laevis MGC80562 protein (MGC80562)	NM_001092240.1	103.88
Xenopus laevis proteasome subunit Y (LOC397716)	NM_001087748.1	103.61
Xenopus laevis mRNA for putative SURF6 protein (surf6 gene)	AJ276843.1	103.59
Xenopus laevis glioma tumor suppressor candidate region gene 2 (gltscr2)	NM_001096553.1	103.55
Xenopus laevis heterogeneous nuclear ribonucleoprotein H1 (H) (hnrrph1-b)	NM_001093454.1	103.54
Xenopus laevis proteasome (prosome, macropain) subunit, beta type 4 (psmb4)	NM_001087421.1	103.42
Xenopus laevis hypothetical protein LOC398643 (cDNA clone IMAGE:6879166), partial cds	BC054293.1	103.22
Xenopus laevis calponin 2 (cDNA clone MGC:53326 IMAGE:5570733)	BC046257.1	103.09
Xenopus laevis proteasome maturation protein (pomp)	NM_001091284.1	102.91
Xenopus laevis fatty acid 2-hydroxylase (fa2h)	NM_001089238.1	102.86
Xenopus laevis chaperon containing TCP1, subunit 8 (theta) (cct8)	NM_001087244.1	102.47
Xenopus laevis yeast RPD3 homologue (cDNA clone MGC:83956 IMAGE:6862376)	BC081136.1	102.45
Xenopus laevis carbonic anhydrase 2 (ca2)	NM_001086611.1	102.14
Xenopus laevis UMP-CMP kinase (cDNA clone IMAGE:4684066), partial cds	BC045275.1	101.7
Xenopus laevis golgi reassembly stacking protein 2, 55kDa (gorasp2)	NM_001087050.1	101.7
Xenopus laevis uncharacterized protein MGC154858 (MGC154858)	NM_001097031.1	101.68
Xenopus laevis heterogeneous nuclear ribonucleoprotein C (C1/C2) (hnrcpc)	NM_001086787.1	101.64
Xenopus laevis junction plakoglobin (jup-b)	NM_001090582.1	101.44
XELUVS2 X.laevis UVS.2 protein gene, 3' end	M27162.1	101.43
Xenopus (Silurana) tropicalis protein phosphatase 1, regulatory subunit 16A (ppp1r16a)	NM_001078885.1	101.4
elongation factor 1 gamma [type 1] [Xenopus laevis=South African clawed frogs, oocytes, 1441 nt]	S69724.1	101.31
Xenopus laevis Siah-interacting protein (sip)	NM_001086745.1	101.22
Xenopus (Silurana) tropicalis spectrin, alpha, non-erythrocytic 1 (sptan1)	NM_001097205.1	101.1
Xenopus laevis eukaryotic translation initiation factor 4 gamma, 2 (eif4g2)	NM_001112853.1	100.96
Xenopus laevis high mobility group protein-1 (cDNA clone MGC:80952 IMAGE:5516344)	BC073449.1	100.92
Xenopus laevis uncharacterized protein MGC114665 (MGC114665)	NM_001095981.1	100.63
Xenopus laevis suppressor of Ty 4 homolog 1 (supt4h1)	NM_001095737.1	100.41
Xenopus laevis RAB11A, member RAS oncogene family (rab11a)	NM_001094296.1	100.3
Xenopus laevis Sp7 transcription factor (sp7)	NM_001099905.1	100.28
Xenopus laevis hypothetical protein MGC116483 (cDNA clone MGC:116483 IMAGE:6935947)	BC099291.1	100.23
Xenopus laevis ATPase, Na+-K+ transporting, alpha 1 polypeptide (atp1a1-b)	NM_001089111.1	99.94
Xenopus laevis aldolase B (cDNA clone MGC:53389 IMAGE:5571492)	BC044676.1	99.89
Xenopus laevis uncharacterized protein MGC68480 (MGC68480)	NM_001090033.1	99.89
Xenopus tropicalis cDNA clone IMAGE:7025807, partial cds	BC077652.1	99.81
Xenopus laevis TIMP metallopeptidase inhibitor 3 (timp3)	NM_001085595.1	99.66
Xenopus laevis EF-hand domain family, member D1 (efhd1)	NM_001092304.1	99.3
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100489973 (LOC100489973), transcript variant X1	XM_002937695.2	99.25
PREDICTED: Xenopus (Silurana) tropicalis uro-adherence factor A-like (LOC100486894), transcript variant X1	XM_002937232.2	98.9
Xenopus laevis heterogeneous nuclear ribonucleoprotein A0 (hnrrna0)	NM_001088945.1	98.88
Xenopus laevis arginase, liver (arg1)	NM_001086948.1	98.83
Xenopus laevis protein phosphatase 4, catalytic subunit (ppp4c)	NM_001091769.1	98.65
PREDICTED: Xenopus (Silurana) tropicalis ring-box 1, E3 ubiquitin protein ligase (rbx1), transcript variant X1	XM_002934726.2	98.64
Xenopus laevis nuclear apoptosis inducing factor 1 (naif1)	NM_001096902.1	98.29
Xenopus tropicalis split hand/foot malformation (ectrodactyly) type 1 (cDNA clone MGC:147791 IMAGE:7531611)	BC167420.1	97.97
Xenopus laevis serine/arginine-rich splicing factor 9 (srsf9)	NM_001094931.1	97.86
Xenopus laevis retinol dehydrogenase 7 (rdh7)	NM_001085720.1	97.85
Xenopus laevis translocase of outer mitochondrial membrane 20 homolog (tomm20), nuclear gene encoding mitochondrial protein	NM_001086854.1	97.8
Xenopus laevis synovial sarcoma translocation gene on chromosome 18-like 1 (ss18l1)	NM_001093632.1	97.78
Xenopus laevis nonmuscle myosin II heavy chain A mRNA	AF055895.1	97.68
Xenopus laevis GTPase activating protein (SH3 domain) binding protein 1 (g3bp1)	NM_001087229.1	97.59

## Appendix E. Individual analysis

### E.1 Six1; Up-regulated genes

**Table E.1** Genes with at least two-fold up-regulation after injection of Six1-GR and treatment with CHX + DEX

Gene	Accession	Six1 CHX <sup>a</sup>	Six1 CHX+DEX <sup>b</sup>	FC Six1 CHX+DEX <sup>b</sup>	FC Six1 CHX <sup>a</sup>	Control CHX <sup>a</sup>	Control CHX+DEX <sup>b</sup>	FC Control CHX+DEX <sup>b</sup>	Rank <sup>c</sup>
PREDICTED: Xenopus (Silurana) tropicalis tyrosine-protein phosphatase non-receptor type substrate 1-like (LOC101733531) mRNA	XM_004918397.1	0.01	31.04	14.93	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine-protein kinase N2-like (LOC101734962) mRNA	XM_004912328.1	0.01	3.35	11.71	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis RNA polymerase II elongation factor ELU2-like (LOC100492932) transcript variant X1 mRNA	XM_002932261.2	0.01	1.53	10.58	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine-protein kinase N2-like (LOC101734328) mRNA	XM_004910385.1	0.01	1.51	10.56	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TNel116d14	CR759989.2	0.01	1.29	10.33	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100485669 (LOC100485669) mRNA	XM_002932396.2	0.01	5.84	9.58	0	0	0	0	0
Xenopus laevis cyclin D (cncd1) mRNA	NP_001087887.1	0.13	22.8	7.48	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis calcitonin gene-related peptide-like (LOC100487527) mRNA	XM_002941675.2	0.03	3.27	6.93	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C14orf166B (LOC100490780) mRNA	XM_002938866.2	0.02	1.32	6.11	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis leucine rich repeat containing 52 (lrc52) mRNA	XM_002933773.2	0.03	1.35	6.01	0	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE7022272	BC094950.1	0.12	5	5.51	0	0	0	0	0
Xenopus laevis uncharacterized LOC10036338 (LOC10036338) mRNA	NM_001097708.1	0.14	5.33	5.26	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis ATP-sensitive inward rectifier potassium channel 11-like (LOC100492679) mRNA	XM_004916278.1	0.04	1.24	5.06	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis voltage-gated channel shaker-related subfamily member 2 (kcnal2) mRNA	XM_004910736.1	0.04	1.21	5.02	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis calcium/calmodulin-dependent protein kinase kinase kinase 2 (camkk2) transcript variant X2 mRNA	XM_004910513.1	0.08	2.48	4.99	0	0	0	0	0
Xenopus (Silurana) tropicalis SK/homeobox 2 (skx2) mRNA	NM_001100275.1	0.55	16.44	4.92	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC10036338 (LOC10036338) mRNA	XM_004918247.1	0.06	1.16	4.37	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis inward rectifier potassium channel 11-like (camkk2) transcript variant X1 mRNA	XM_002937701.2	0.08	1.57	4.32	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis calmodulin-dependent protein kinase kinase kinase 2 (camkk2) transcript variant X2 mRNA	XM_002934546.2	0.19	3.11	4.06	0	0	0	0	0
Xenopus (Silurana) tropicalis SK/homeobox 2 (skx2) mRNA	XM_004914258.1	0.12	1.8	3.95	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734405 (LOC101734405) mRNA	NM_001030412.1	0.35	5.15	3.91	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis calmodulin-dependent protein kinase kinase kinase 2 (camkk2) transcript variant X2 mRNA	XM_002937701.1	0.15	2.18	3.89	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis mastemid-like domain containing 1 (mamid1) mRNA	XM_004913441.1	0.1	1.27	3.71	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis intestinal mucin-like protein-like (LOC101730508) mRNA	H2266327.1	0.18	2.29	3.69	0	0	0	0	0
Xenopus (Silurana) tropicalis potassium inwardly-rectifying channel subfamily J member 8 (kcnj8) mRNA	NM_001102926.1	0.17	2.01	3.6	0	0	0	0	0
Xenopus (Silurana) tropicalis finished cDNA clone TIB05016	CL025142.1	0.16	1.89	3.58	0	0	0	0	0
Xenopus laevis mRNA for Xoso17-alpha protein	A001730.1	0.83	9.79	3.56	0	0	0	0	0
Xenopus laevis RDC1 like protein mRNA (cDNA clone MGIC114801 IMAGE4632706) complete cds	BC098974.1	0.57	6.53	3.53	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis RNA-directed DNA polymerase homolog (LOC101730974) partial mRNA	XM_004911612.1	0.19	1.94	3.39	0	0	0	0	0
Xenopus (Silurana) tropicalis glutathione peroxidase 2 (gastrointestinal) (gpx2) mRNA	NM_001256315.1	1.18	11.69	3.32	3.02	2.57	-0.24	0	0
Xenopus laevis cytochrome P450 family 26 subfamily B polypeptide 1 (Cyp26b1) mRNA partial cds	EU816560.1	0.12	1.09	3.26	0	0	0	0	0
Xenopus (Silurana) tropicalis cytochrome P450 family 26 subfamily B polypeptide 1 (Cyp26b1) mRNA	NM_001079187.2	0.18	1.63	3.23	3.19	1.81	-0.82	0	0
PREDICTED: Xenopus (Silurana) tropicalis poly(A)-ribose polymerase 14-like (LOC101731378) mRNA	XM_004920062.1	0.16	1.32	3.08	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis espin (espn) transcript variant X1 mRNA	NM_002933895.2	0.69	5.66	3.06	1.8	1.75	-0.05	0	0
Xenopus (Silurana) tropicalis calcium channel voltage-dependent beta 4 subunit (cacnb4) mRNA	NM_001142151.1	0.13	1.08	3.05	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TEG9026p17	CR761997.2	0.2	1.56	2.99	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC100494747) mRNA	XM_002936043.2	0.32	2.44	2.95	0	0	0	0	0
Xenopus (Silurana) tropicalis chemokine (C-X-C motif) receptor 7 (cxcr7) mRNA	NM_001030434.1	0.45	3.32	2.91	0	0	0	0	0
Xenopus laevis solute carrier family 43 member 1 (slc43a1) mRNA	NM_001094328.1	1.15	8.28	2.86	2.77	1.88	-0.57	0	0
Xenopus laevis gelatinase B (Mmp-9) mRNA complete cds	AF072455.1	0.66	4.7	2.85	0	0	0	0	0
Xenopus laevis ectodysplasin A receptor (edan) mRNA	NM_001087047.1	0.4	2.65	2.76	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC100497756) mRNA	NM_002940161.2	0.29	1.82	2.67	0	0	0	0	0

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC Six1 <sup>+</sup>	FC Six1 <sup>-</sup>	Control CHX	Control CHX+DEX	FC Control CHX	FC Control CHX+DEX	Rank <sup>a</sup>
Xenopus tropicalis sine ocellis binding protein homolog (Drosophila) mRNA (cDNA clone MGC:172604 IMAGE:7636239) complete cds	BC154687.1	0.58	3.53	2.63	0	0	0	0	0	0
Xenopus (Silurana) tropicalis protease serine 27 (prss27) mRNA	NM_001011195.1	0.25	1.42	2.55	0	0	0	0	0	0
Xenopus laevis mRNA for papillary renal cell carcinoma protein (lprcc gene)	AJ318531.1	229.62	1338.74	2.55	39.47	36.08	-0.13	0	0	0
Xenopus laevis empty sites homeobox 1 gene 2 (emx1.2) mRNA	NM_001093430.1	0.4	2.31	2.53	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis protein kinase C delta type-like (LOC101734135) partial mRNA	XM_004920514.1	0.34	1.94	2.52	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733225 (LOC101733225) mRNA	XM_004919937.1	0.22	1.18	2.46	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734952 (LOC101734952) mRNA	XM_004916172.1	0.24	1.29	2.44	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101732555 (LOC101732555) mRNA	XM_004913816.1	0.21	0	2.39	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 2-like (LOC100491930) mRNA	XM_002832254.2	0.91	4.61	2.36	11.58	6.57	-0.82	0	0	0
Xenopus laevis f23 protein mRNA (cDNA clone MGC:196642 IMAGE:9041549) complete cds	BC169915.1	0.3	1.51	2.33	3.2	3.13	-0.04	0	0	0
Xenopus tropicalis finished cDNA clone TNeu076f16	CR848409.2	0.73	3.6	2.32	0	0	0	0	0	0
Xenopus laevis actin alpha 3 smooth muscle aorta (acta2) mRNA	NM_001091337.1	0.33	1.55	2.24	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TNeu143f9	CR760056.2	0.32	1.44	2.2	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis myomesin 1 (myom1) mRNA	NM_002838907.2	0.26	1.17	2.19	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis voltage-gated channel 1sk-related family member 1 (kcne1) transcript variant X2 mRNA	XM_004912135.1	1.01	4.46	2.16	0	0	0	0	0	0
Xenopus laevis uncharacterized protein MGCG6450 (MGCG6450) mRNA	NM_001089841.1	0.37	1.61	2.16	0	0	0	0	0	0
Xenopus (Silurana) tropicalis myomesin 1 (myom1) mRNA	NM_001129936.1	0.44	1.91	2.15	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis anocamtin 2 (ano2) mRNA	NM_002932297.2	0.31	1.35	2.13	0	0	0	0	0	0
Xenopus laevis natruretic peptide C (nppc) mRNA	NM_001093118.1	0.5	2.12	2.1	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis metallothionein-like (LOC100488960) mRNA	NM_002940036.2	0.31	1.32	2.1	0	0	0	0	0	0
Xenopus laevis Rab11B member RAS oncogene family gene (rab11b.2) mRNA	NM_001095383.1	0.51	2.17	2.09	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tripartite motif-containing protein 7-like (LOC100491025) mRNA	NM_002943573.2	0.3	1.28	2.09	0	0	0	0	0	0
Xenopus laevis janus kinase and microtubule interacting protein 1 (jakmip1) mRNA	NM_001091623.1	0.28	1.16	2.08	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) nuclear receptor corepressor mRNA complete cds	AF495886.1	77.6	323.28	2.06	62.12	49.96	-0.32	0	0	0
PREDICTED: Xenopus (Silurana) pancreatic cancer Y receptor Y2 (inpy2n) mRNA	XM_004911153.1	0.61	2.5	2.06	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) calcitonin receptor (calc) mRNA	NM_002834599.2	0.26	1.08	2.05	0	0	0	0	0	0
Xenopus (Silurana) pancreatic lipase-related protein 2 (plnlp2) mRNA	NM_001078963.1	1.52	6.27	2.05	3.07	3	-0.04	0	0	0
Xenopus laevis gamma glutamyl hydrolase (conjugase folylpolygamma glutamyl hydrolase) (ggth) mRNA	NM_001092691.1	1.96	7.95	2.03	4.24	4.22	-0.01	0	0	0
Xenopus laevis integrin beta-3 subunit mRNA (cDNA clone MGCG80757) (MGEGE5512565) complete cds	BC073343.1	19.29	77.73	2.02	17.36	15.68	-0.15	0	0	0
Xenopus (Silurana) tropicalis eva-1 homolog A (eva1a) mRNA	NM_001102800.2	2.36	8.81	1.91	2.69	2.63	-0.04	0	0	0
Xenopus laevis serine palmitoyltransferase long chain base subunit 2 (splc2) mRNA	NM_001092294.1	57.38	209.58	1.87	43.75	38.3	-0.2	0	0	0
Xenopus (Silurana) tropicalis calcitonin receptor (calc) mRNA	CR855512.2	54.43	188.85	1.8	36.81	15.78	-1.23	0	0	0
Xenopus (Silurana) sema domain immunoglobulin domain (lg) short basic domain secreted (semaphorin) 3E (sema3e)	NM_002935256.2	0.64	2.19	1.78	1.2	1.06	-0.18	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100490228 (LOC100490228) mRNA	NM_002942932.2	0.41	1.38	1.75	12.15	6.67	-0.87	0	0	0
Xenopus (Silurana) tropicalis ST3Gal IV (st3gal4) mRNA	FN1550108.1	5.36	17.83	1.74	5.83	4.91	-0.25	0	0	0
Xenopus laevis hyaluronan synthase related sequence protein (has-rs) mRNA	NM_001093680.1	1552.09	5059.89	1.71	2219.06	2209.29	-0.01	0	0	0
PREDICTED: Xenopus (Silurana) macrophage differentiation-associated (mmnd) transcript variant X1 mRNA	NM_002937811.2	1.04	3.28	1.67	2.81	2.39	-0.23	0	0	0
Xenopus tropicalis finished cDNA clone THd029C02	CR855543.2	2.72	7.97	1.56	40.19	6.65	-2.6	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis ArfGAP with SH3 domain ankyrin repeat and PH domain 3 (asap3) mRNA	NM_001107474.1	1.34	3.88	1.54	1.85	2.21	-0.07	0	0	0
Xenopus laevis hyaluronan synthase related sequence protein (has-rs) mRNA	NM_002932703.2	1.19	3.38	1.51	1.83	1.26	-0.54	0	0	0
PREDICTED: Xenopus (Silurana) nuclear gene encoding mitochondrial protein mRNA	NM_001114489.1	1.36	3.82	1.5	15.65	15.21	-0.05	0	0	0
Xenopus laevis pyruvate dehydrogenase catalytic subunit 1 (pdhp1) mRNA	NM_001094221.1	160.75	448.4	1.48	48.37	41.62	-0.22	0	0	0
PREDICTED: Xenopus (Silurana) nuclear gene encoding mitochondrial protein mRNA	BC074452.1	9.05	25.11	1.48	12.2	11.1	-0.14	0	0	0
Xenopus laevis dopa decarboxylase (aromatic L-amino acid decarboxylase) (ddc) mRNA	NM_001096636.1	7.42	20.55	1.48	11.34	9.3	-0.29	0	0	0
PREDICTED: Xenopus (Silurana) early growth response 3 (egfr3) mRNA	NM_002937352.2	0.42	1.11	1.43	6.2	5.44	-0.19	0	0	0
Xenopus (Silurana) nuclear gene containing 3 (litch3) mRNA	NM_001090008.1	1.79	4.78	1.43	1.57	1.56	-0.02	0	0	0
Xenopus laevis cyclic nucleotide-binding protein with multiple splicing (rbpms) mRNA	AF076618.1	15.35	40.15	1.39	33.39	25.96	-0.37	0	0	0

Gene	Accession	Six1 CHX	Six1 CHX+DXP	FC Six1 <sup>1</sup>	FC Control CHX	Control CHX	Control CHX-DEX	FC Control CHX	Rank <sup>7</sup>
PREDICTED: Xenopus (Silurana) tropicalis synaptic vesicle glycoprotein 2C (sv2c) mRNA	XM_002934827.2	4.64	12.11	1.39	5.15	3.71	-0.48	0	
Xlaevis XFD2 mRNA for fork head protein	XJ4315.1	10.11	26.34	1.39	14.77	13.66	-0.12	0	
PREDICTED: Xenopus (Silurana) tropicalis thiamine transporter 1-like (LOC100489793) mRNA	XM_002935783.2	1.64	4.27	1.38	3.02	1.69	-0.85	0	
Xenopus laevis clone IMAGE:684003 mRNA	BC042305.1	8.14	21.16	1.38	7.39	6.5	-0.19	0	
Xenopus laevis poly(A/P-ribose) glycohydrolase (parl) mRNA	NM_001096133.1	823.44	2127.02	1.37	51.87	45.79	-0.18	0	
Xenopus laevis adenosine deaminase (ada) mRNA	NM_001092271.1	10.25	26.33	1.37	27.42	24.12	-0.19	0	
PREDICTED: Xenopus (Silurana) tropiclin (CDNA clone IMAGE:5571064)	BC045116.1	467197.2	1178098.76	1.34	713849.63	67869.31	-3.4	0	
Xenopus laevis mRNA similar to envelopin (CDNA clone IMAGE:3206.2)	XM_002933206.2	10.88	27.31	1.33	12.93	9.41	-0.46	0	
Xenopus laevis uncharacterized LOC496300 (LOC496300) mRNA	NM_001095458.1	3.98	9.98	1.33	10.26	7.72	-0.42	0	
Xenopus laevis Nlo mRNA complete cds	AY177202.1	1.25	3.12	1.32	2.18	1.71	-0.36	0	
Xenopus (Silurana) tropicalis ras homolog family member V (rhov) mRNA	NM_001128659.1	16.26	40.5	1.32	11.9	11.47	-0.06	0	
PREDICTED: Xenopus (Silurana) tropicalis frizzled family receptor 4 (frz4) mRNA	XM_002936543.2	19.89	49.45	1.32	20.82	14.32	-0.54	0	
PREDICTED: Xenopus (Silurana) tropicalis transmembrane channel-like protein 7-like (LOC100493700) mRNA	XM_002932222.2	2.36	5.86	1.32	4.07	2.12	-0.95	0	
Xenopus laevis CUE domain containing 2 (cuedc2-b) mRNA	NM_001094007.1	0.57	1.41	1.31	1.14	1.05	-0.12	0	
PREDICTED: Xenopus (Silurana) tropicalis mannose receptor C type 1 (mrc1) mRNA	XM_002939002.2	0.86	2.12	1.31	1.72	1.09	-0.67	0	
Xenopus (Silurana) tropicalis tenascin R (tnr) mRNA	NM_00113815.1	1.63	4	1.3	3.1	2.18	-0.51	0	
Xenopus (Silurana) tropicalis POU class A-homeobox 1 (pou4f1.2) mRNA	NM_001097307.1	5.06	12.13	1.27	10.09	7.19	-0.49	0	
Xenopus laevis ets-2a proto-oncogene mRNA (CDNA clone MGCI60372) [IMAGE:8544815] complete cds	BC133183.1	8.1	19.18	1.25	11.41	8.58	-0.42	0	
Xenopus laevis mRNA for frizzled 4 protein (frz4 gene)	AJ251750.1	7.72	18.24	1.25	9.31	7.66	-0.29	0	
Xenopus laevis transmembrane protein 117 (tmem117) mRNA	NM_001096969.1	0.93	2.14	1.22	1.51	1.02	-0.58	0	
Xenopus laevis uncharacterized LOC503678 (LOC503678) mRNA	NM_001095567.1	77.36	178.56	1.21	16.09	13.06	-0.31	0	
Xenopus laevis zinc finger protein 21.4 (znf21.4) mRNA	NM_001097042.1	121.42	276.51	1.19	161.35	146.92	-0.14	0	
PREDICTED: Xenopus (Silurana) tropicalis flocculation protein FLO11-like (LOC100490389) transcript variant X1 mRNA	XM_002942555.2	2.86	6.42	1.17	1.75	1.6	-0.14	0	
Xenopus laevis RELT-like 2 (retl2) mRNA	NM_001094085.1	127.69	286.74	1.17	42.22	36.16	-0.23	0	
Xenopus laevis cdc25Ba mRNA for cdc25B phosphatase complete cds	AB363840.1	7.83	17.53	1.17	7.43	6.94	-0.1	0	
Xenopus laevis keratin 16 (focal non-epidermolytic palmaroplantar keratoderma) (krt16) mRNA	NM_001093439.1	3.1	6.86	1.15	3.16	2.24	-0.5	0	
PREDICTED: Xenopus (Silurana) tropicalis hematopoietic cell signal transducer (hst) partial mRNA	XM_004916621.1	0.84	1.83	1.14	1.44	1.15	-0.33	0	
PREDICTED: Xenopus (Silurana) tropiclin (CDNA clone IMAGE:39359) transcript variant X1 mRNA	XM_0049139359.1	3.64	7.92	1.13	4.59	2.62	-0.81	0	
Xenopus laevis adenomatous polyposis coli down-regulated 1 (apcd1) mRNA	NM_001094109.1	4.8	10.39	1.12	6.58	5.29	-0.32	0	
Xenopus (Silurana) tropicalis forkhead box C1 (foxc1) mRNA	NM_001097863.1	0.6	1.29	1.12	2.83	2.83	-0.01	0	
Xenopus laevis homeobox transcription factor Six3 (Ski3) mRNA complete cds	AF183357.1	4.65	10.05	1.12	8.05	6.17	-0.39	0	
Xenopus laevis ER membrane protein complex subunit 8 (emc8) mRNA	NM_001094734.1	13.1	28.16	1.11	26.67	23.82	-0.17	0	
Xenopus laevis cDNA clone IMAGE:6317850	BC155359.1	9.12	19.58	1.11	21.2	16.45	-0.37	0	
Xenopus laevis uncharacterized LOC503674 (LOC503674) mRNA	NM_001095631.1	14.21	30.48	1.11	15.18	11.67	-0.39	0	
Xenopus laevis forkhead box 14 gene 1 (fox4.1) mRNA	NM_001097651.1	16.76	35.52	1.09	31.6	31.31	-0.02	0	
Xenopus (Silurana) tropicalis uncharacterized LOC100127867 (LOC100127867) mRNA	NM_001113158.1	3.3	6.93	1.07	108.94	75.13	-0.54	0	
Xenopus laevis LM class homeodomain protein mRNA (cDNA clone MGC79961) [IMAGE:6865745] complete cds	BC084744.1	1.63	3.39	1.06	3.31	2.6	-0.35	0	
Xenopus (Silurana) tropicalis O-linked N-acetylglucosamine (GlcNAc) transferase (egt) mRNA	NM_001097223.1	0.77	1.6	1.06	1.06	1.17	-0.06	0	
Xenopus (Silurana) tropicalis leucine zipper putative tumor suppressor 2 (litz2) mRNA	NM_001096720.1	4.61	9.54	1.05	4.64	4.1	-0.18	0	
PREDICTED: Xenopus (Silurana) tropicalis CD36 molecule epsilon associated protein (cd36ap) transcript variant 2 mRNA	XM_002932605.2	2.52	5.18	1.04	1.51	1.51	-0.01	0	
Xenopus laevis polymerase (RNA) II (DNA directed) polypeptide L 7.6kDa gene 1 (pol2L1) mRNA	NM_001112227.1	71.4	146.25	1.04	51.33	45.46	-0.18	0	
PREDICTED: Xenopus (Silurana) tropicalis tripartite motif-containing protein 7-like (LOC100490338) mRNA	CLO24921.1	0.32	1.28	2	0	0	-0.01	0	
PREDICTED: Xenopus (Silurana) tropicalis poly(A)-linked N-acetylglucosaminyltransferase (mgat4b) mRNA	XM_002943546.2	0.45	1.79	2	0	0	0.01	0	
Xenopus (Silurana) tropicalis mannosyl (alpha-1-3-glycoprotein beta-1-N-acetylglucosaminyltransferase isozyme B (mgat4b) mRNA	NM_001102914.1	0.52	1.99	1.95	0	0	0.01	0	
PREDICTED: Xenopus (Silurana) tropicalis avdin-like (LOC100487365) mRNA	XM_002939983.2	0.54	2.05	1.93	0	0	0.01	0	
PREDICTED: Xenopus (Silurana) tropicalis complement C3-like mRNA	NM_004919091.1	0.45	1.6	1.85	0	0	0.01	0	

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC Six1 <sup>1</sup> CHX+DEX	Control CHX	Control CHX-DEX	FC Control CHX-DEX	Control CHX	Rank <sup>7</sup>
PREDICTED: Xenopus (Silurana) tropicalis c-X-C motif chemokine 10-like (LOC100490785) mRNA.	XM_002940578.2	0.51	1.8	1.85	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis polycystic kidney disease 1-like 3 (pkd1l3) mRNA	XW..004914252.1	0.36	1.26	1.84	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis kinase insert domain receptor (a type III receptor tyrosine kinase) (kdr) mRNA	XW..002934669.2	1.14	4.06	1.84	0	0	0	0	0.01
Xenopus laevis uncharacterized LOC10037217 (LOC100037217) mRNA	NM..001097906.1	0.31	1.08	1.84	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis olfactory receptor 563-like (LOC100492086) mRNA	XW..002942220.1	1	3.5	1.83	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis UDP-GalbetaGlcNAc beta 1-4-galactosyltransferase polypeptide 2 (b4galt2)	XW..002931472.2	0.57	2	1.81	0	0	0	0	0.01
Xenopus laevis cytochrome P450 family 3 subfamily A polypeptide 4 (cyp3a4) mRNA	NM..001097988.1	0.3	1.02	1.8	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis deoxyribonuclease gamma-like (LOC100497175) transcript variant X1 mRNA	XW..002938386.2	1.37	4.69	1.79	0	0	0	0	0.01
Xenopus laevis similar to caliskeptin 2 (cardiac muscle) mRNA (cDNA clone MGC:114674 IMAGE:5161 32) complete cds	BC097545.1	0.37	1.26	1.79	0	0	0	0	0.01
Xenopus laevis uncharacterized protein MGCB2209 (MGCB2209) mRNA	NM..001091541.1	0.42	1.41	1.76	0	0	0	0	0.01
Xenopus laevis ectonucleotide triphosphate diphosphohydrolase 1 (entpd1) mRNA	NM..001092268.1	0.47	1.56	1.75	0	0	0	0	0.01
Xenopus laevis MGCB0947 protein (MGCB0947) mRNA	NM..001092397.1	0.62	2.07	1.75	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone Thd4609	CU025205.1	0.37	1.22	1.75	0	0	0	0	0.01
Xenopus laevis shisa-2 mRNA for Shisa2 complete cds	AB242597.1	1.44	4.81	1.74	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis F-box protein 32 (fbxo32) transcript variant X1 mRNA	XW..002941397.2	0.46	1.52	1.74	0	0	0	0	0.01
Xlaevis mRNA for interphotoreceptor retinoid-binding protein	XW5473.1	3.58	11.87	1.74	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis membrane protein 74 (tnem74) transcript variant X2 mRNA	XW..004915071.1	0.33	1.09	1.73	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TEG013309	CT025230.1	2.69	8.87	1.73	0	0	0	0	0.01
Xenopus laevis cDNA clone MGCB0675 IMAGE:5511 09 complete cds	BC073294.1	0.7	2.21	1.68	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100493356 (LOC100493256) transcript variant X1 mRNA	XW..002931666.2	1.09	3.46	1.67	0	0	0	0	0.01
Xenopus laevis mRNA for xox1 (Alpha1) partial cds	XW051691.1	0.37	1.15	1.67	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis death domain containing 1 (dthd1) mRNA	XW..002938654.2	0.62	1.96	1.67	0	0	0	0	0.01
Xenopus laevis collagen type II alpha 1 (col2a1) mRNA	NM..001087789.1	1.23	3.84	1.66	0	0	0	0	0.01
Xenopus (Silurana) tropicalis leucine rich repeat containing 39 (lrrc39) mRNA	NM..001130264.1	3.56	10.94	1.62	0	0	0	0	0.01
Xenopus (Silurana) tropicalis sorting nexin 16 (snx16) mRNA	NM..001017302.3	0.6	1.84	1.62	0	0	0	0	0.01
Xenopus (Silurana) tropicalis B-Cell CLLymphoma 10 (hc10) mRNA	NM..001015777.2	0.51	1.55	1.62	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TEG019K04	CF7613320.2	0.43	1.29	1.59	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis nicotinamide N-methyltransferase-like (LOC100491286) transcript variant X4 mRNA	XW..004916098.1	0.56	1.65	1.58	0	0	0	0	0.01
Xenopus laevis Kazal-type serine peptidase inhibitor domain 1 (kazald1) mRNA	NM..001092073.1	0.47	1.37	1.57	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis beta-1,3-galactosyltransferase 1 (LOC101732799) transcript variant X1 mRNA	XW..004918863.1	0.59	1.72	1.56	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis tripartite motif-containing protein 14-like (LOC100493056) mRNA	XW..002935873.2	0.37	1.08	1.56	0	0	0	0	0.01
Xenopus (Silurana) tropicalis KIAA0895 protein (KIAA0895) mRNA	NM..001114073.1	0.5	1.42	1.52	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis zinc finger CCHC domain containing 7 (zcch7) mRNA	XW..002935880.2	0.61	1.73	1.51	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis unknown open reading frame human C2orf80 (LOC100486093) transcript variant X2 mRNA	XW..004918907.1	1.08	3.01	1.5	0	0	0	0	0.01
Xenopus laevis uncharacterized LOC100336933 (LOC100336933) mRNA	NM..001097704.1	0.86	2.4	1.48	0	0	0	0	0.01
Xenopus laevis calcium binding and coiled-coil domain 1 (calcooc1-b) mRNA	NM..001093489.1	0.65	1.78	1.46	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis homeobox A7 (hoxa7) mRNA	XW..004915298.1	0.79	2.16	1.46	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis microtubule associated serine/threonine kinase family member 4 (mas4) mRNA	XW..002934204.2	0.76	2.05	1.45	0	0	0	0	0.01
Xenopus laevis MGCB0823 protein (MGCB0823) mRNA	XW..002937064.2	0.43	1.16	1.44	0	0	0	0	0.01
Xenopus tropicalis hypothetical protein LOC594926 mRNA (cDNA clone IMAGE:7024052) partial cds	BC092555.1	0.85	2.26	1.42	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis CDP-diacylglycerol synthase (phosphatidate cytidyltransferase) 1 (cds1) mRNA	XW..002937878.2	0.43	1.14	1.41	0	0	0	0	0.01
Xenopus (Silurana) tropicalis family with sequence similarity 155 member B (snf15b) mRNA	NM..001028631.1	0.46	1.22	1.41	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C2orf80 (LOC100489057) mRNA	XW..002937119.2	1.16	3.06	1.4	0	0	0	0	0.01
Xenopus laevis uncharacterized protein MGCG4484 (MGCG4484) mRNA	NM..001097066.1	0.39	1.03	1.4	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis kinase family member 3C (kif3c) transcript variant X1 mRNA	XW..002942110.2	0.58	1.51	1.39	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis zinc finger and BTB domain containing 20 (zbtf20) mRNA	XW..002935849.2	0.79	2.05	1.39	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis cernamide kinase-like (cerkl) mRNA	XW..002932015.2	0.56	1.45	1.38	0	0	0	0	0.01
Xenopus laevis Tbxf6 (Tbx6) mRNA complete cds	D0355794.1	3.94	10.14	1.37	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis putative nuclease HARB1-like (LOC100491442) mRNA	XW..002933801.1	0.69	1.77	1.37	0	0	0	0	0.01
Xenopus (Silurana) tropicalis $\gamma$ -maf musculoaponeurotic fibrosarcoma oncogene homolog A (mafafa) mRNA	NM..001032304.1	1.07	2.75	1.37	0	0	0	0	0

Gene	Accession	Six1 CHX	Six1 CHX+Dex	FC Six1 CHX+Dex	Control CHX	Control CHX-Dex	FC Control CHX- Control CHX-Dex	Rank <sup>a</sup>
PREDICTED: Xenopus (Silurana) tropicalis oocyte zinc finger protein XICOF6-like [LOC100486602] mRNA	XM_004915718.1	0.42	1.07	1.36	0	0	0	0.01
Xenopus laevis cDNA clone 21 (G4-1)	AJ009295.1	2.28	5.8	1.36	0	0	0	0.01
Xenopus laevis TGF-beta family member lefty-A mRNA complete cds	AF283562.1	0.41	1.03	1.35	0	0	0	0.01
Xenopus (Silurana) tropicalis adenosine monophosphate deaminase 3 (ampd3) mRNA	NM_001030516.1	0.47	1.18	1.35	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis sush1 von Willebrand factor type A EGF and pentraxin domain containing 1 (sven1) mRNA	XM_002942621.2	1.21	3.05	1.35	0	0	0	0.01
Xenopus tropicalis finished cDNA clone Thd04035f23	C0025180.1	1.16	2.93	1.35	0	0	0	0.01
Xenopus (Silurana) tropicalis RNA binding motif protein 34 (rbm34) mRNA	NM_001004890.1	0.48	1.2	1.34	0	0	0	0.01
Xenopus (Silurana) tropicalis PX domain containing 1 (pxdc1) mRNA	NM_001130262.1	0.72	1.81	1.34	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis adenosine monophosphate deaminase 1 (ampd1) mRNA	XM_002935775.2	0.5	1.26	1.34	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis novel protein similar to PHD finger protein 6 [LOC733724] mRNA	XM_002935750.2	0.81	2.02	1.34	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis metallopeptidase with thrombospondin type 1 motif 12 (adams12) partial mRNA	XM_002942444.2	0.52	1.3	1.33	0	0	0	0.01
Xenopus (Silurana) tropicalis drhk glucuronidase homolog (E. coli) (idnk) mRNA	NM_001126592.1	0.88	2.2	1.33	0	0	0	0.01
Xenopus laevis uncharacterized protein MGC81120 (MGC81120) mRNA	NM_001091225.1	1.05	2.6	1.31	0	0	0	0.01
Xenopus (Silurana) tropicalis SWAD family member 3 (smad3) mRNA	NM_001008436.1	0.52	1.28	1.31	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100494710 (LOC100494710) mRNA	XM_002939048.2	0.64	1.56	1.31	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis bone morphogenetic protein 5 (bmp5) transcript variant X1 mRNA	XM_002934847.2	0.79	1.95	1.31	0	0	0	0.01
Xenopus laevis cDNA clone IMAGE6952399 partial cds	BC074332.1	0.54	1.31	1.3	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis glyican 3 (gpc3) mRNA	XM_002932619.2	0.43	1.04	1.28	0	0	0	0.01
Xenopus (Silurana) tropicalis LIM class homeodomain protein (lim5/Lhx5) mRNA complete cds	NM_001079043.1	0.93	2.25	1.28	0	0	0	0.01
Xenopus laevis hypothetical protein LOC443526 mRNA (cDNA clone IMAGE4885284) partial cds	BC072948.1	1.78	4.24	1.26	0	0	0	0.01
Xenopus laevis cDNA clone IMAGE6952398 partial cds	BC093552.1	0.49	1.17	1.26	0	0	0	0.01
Xenopus (Silurana) tropicalis transforming growth factor beta-induced 68kDa (tgfb1) mRNA	NM_001102875.1	0.54	1.27	1.25	0	0	0	0.01
Xenopus laevis transforming growth factor beta-linked recessive (tp2) mRNA	NM_001095238.1	0.62	1.46	1.25	0	0	0	0.01
YELLIM5R: Xenopus laevis LIM class homeodomain protein (lim5/Lhx5) mRNA	L42546.1	3.33	7.87	1.25	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine kinase 32A (stks2a) mRNA	XM_002936707.2	0.6	1.39	1.23	0	0	0	0.01
Xenopus laevis matrix-remodeling associated 7 (mxm7) mRNA	NM_001094445.1	0.72	1.68	1.22	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100486501 (LOC100486501) mRNA	XM_002942085.2	1.53	3.51	1.21	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis pigmentosa 2 (X-linked recessive) (rp2) mRNA	XM_002935977.2	2.54	5.85	1.21	0	0	0	0.01
XENLIM5R: Xenopus (Silurana) tropicalis containing 9 (comm9) mRNA	NM_001114206.1	0.46	1.06	1.21	0	0	0	0.01
Xenopus (Silurana) tropicalis melanoma inhibitory activity (mia) mRNA	NM_001142244.1	0.45	1.02	1.2	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis PEST proteolytic signal containing nuclear protein (pcnp) transcript variant X1 mRNA	XM_002936816.2	1.26	2.87	1.19	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100486511 (LOC100486511) mRNA	XM_002942816.2	0.5	1.13	1.19	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TEG093313	CP942784.1	0.55	1.25	1.18	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis CAMP responsive element binding protein 5 (creb5) transcript variant X1 mRNA	XM_002933873.2	0.49	1.08	1.17	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor 126 (gr126) mRNA	XM_002933873.2	0.49	1.08	1.17	0	0	0	0.01
Xenopus laevis allograft phosphoribosyltransferase (qprt) mRNA	XG06592.1	0.52	1.14	1.15	0	0	0	0.01
Xenopus laevis hypothetical protein MGC132374 mRNA (CDNA clone MGC:132374 IMAGE3396440) complete cds	BG106693.1	1.55	3.43	1.15	0	0	0	0.01
Xenopus tropicalis N-deacetylase/N-acetyltransferase (heparan glucosaminyl) 3 (ndts3) mRNA	NM_001120704.1	1.14	2.51	1.15	0	0	0	0.01
Xenopus laevis ALG11 alpha-1,2-mannosyltransferase (alg11) mRNA	NM_001122888.1	1.3	2.86	1.15	0	0	0	0.01
Xenopus laevis quinolinate phosphoribosyltransferase (qprt) mRNA	NM_001093324.1	0.52	1.14	1.15	0	0	0	0.01
Xenopus laevis X1Hbox2 mRNA (homeobox-containing p52 clone)	XM_002938828.2	0.65	1.4	1.13	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734664 (LOC101734664) mRNA	XM_004910525.1	1.17	2.53	1.12	0	0	0	0.01
Xenopus (Silurana) tropicalis solute carrier family 13 (sodium-dependent citrate transporter) member 5 (slc13a5)	XM_004911730.1	0.5	1.07	1.12	0	0	0	0.01
Xenopus tropicalis lin-28 homolog mRNA (cDNA clone MGC:97527 IMAGE538262) complete cds	BC09084.1	10.08	21.77	1.12	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis collagen alpha-1(VII) chain-like (LOC101732997) mRNA	XM_004913190.1	2.17	4.68	1.12	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC10049750 (LOC10049750) mRNA	NM_001098690.1	0.51	1.09	1.11	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis canavanine 3 (scd3) mRNA	XM_002938828.2	0.52	1.11	1.11	0	0	0	0.01
Xenopus tropicalis 28S ribosomal RNA (rRNA) clone MGC:121354 (IMAGE7605119) complete cds	BC135512.1	0.9	1.91	1.1	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis C delta 3 (plcd3) mRNA	XM_002935518.2	0.83	1.77	1.1	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis 6-phosphofructo-2-kinase/phosphatase-like (LOC100488759) mRNA	XM_004919631.1	1.42	3.01	1.09	0	0	0	0.01
Xenopus laevis Williams Beuren syndrome chromosome region 27 (wbscr27) mRNA	NM_001091059.1	0.54	1.14	1.09	0	0	0	0.01

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC Six1 <sup>a</sup>	Control CHX	Control CHX+DEX	FC Control CHX	Rank <sup>b</sup>
Xenopus laevis solute carrier family 11 (proton-coupled divalent metal ion transporters) member 1 (slc11a1) mRNA	NM_001059453.9.1	0.7	1.47	1.08	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis homeobox B8 (bxhd8) transcript variant X1 mRNA	XW..002938021.2	1.01	2.11	1.07	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis alpha-kinase 2 (alpk2) mRNA	XW..004910401.1	0.59	1.23	1.07	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis single-minded homolog 1 (Drosophila) (sim1) transcript variant X3 mRNA	XW..004914546.1	0.61	1.27	1.07	0	0	0	0.01
Xenopus laevis COMM domain containing 3 (comm3) mRNA	NM..001059586.1	0.68	1.41	1.07	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis brain-specific angiogenesis inhibitor 3 (bai3) transcript variant X1 mRNA	XW..002936521.2	0.97	2	1.06	0	0	0	0.01
Xenopus (Silurana) tropicalis piwi-like RNA-mediated gene silencing 2 (piwil2) mRNA	NM..001112999.1	0.74	1.54	1.06	0	0	0	0.01
Xenopus laevis tetraopeptide repeat domain 39B (ttc39b) mRNA	NM..001094701.1	0.94	1.94	1.06	0	0	0	0.01
Xenopus laevis lapl02 mRNA complete sequence	DQ096843.1	112.29	231.36	1.05	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis PylA/PGLa A-like (LOC100488375) transcript variant 2 mRNA	XW..002941627.2	0.52	1.07	1.04	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis putative methyltransferase KIAA1456 homolog (LOC100488302) transcript variant X1 mRNA	XW..002934674.2	0.85	1.74	1.04	0	0	0	0.01
Xenopus (Silurana) tropicalis uncharacterized LOC100145323 (LOC100145323) mRNA	NM..001126798.1	0.62	1.26	1.04	0	0	0	0.01
Xenopus laevis twisted gastrulation homolog 2 (twg2) mRNA	NM..001095289.1	0.52	1.06	1.04	0	0	0	0.01
Xenopus laevis embryonic serine protease-1 mRNA (cDNA clone IMAGE:6864235) partial cds	BC082670.1	11.5	23.45	1.03	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis arachidonate 5-lipoxygenase (alox5) transcript variant X2 mRNA	XW..004915925.1	0.73	1.48	1.03	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis metalloprotease ITIM1-like (LOC00491951) mRNA	XW..002936336.2	1.54	3.13	1.03	0	0	0	0.01
Xenopus (Silurana) tropicalis protein expressed (pdit) mRNA	XW..002931984.2	1	2.02	1.03	0	0	0	0.01
Xenopus laevis similar to casqueinut 2 (cardiac muscle) mRNA (cDNA clone IMAGE:4681496) partial cds	BC041283.1	0.87	1.77	1.03	0	0	0	0.01
Xenopus laevis clone IMAGE:681494 mRNA	BC044027.1	1.38	2.76	1.01	0	0	0	0.01
Xenopus (Silurana) tropicalis mitochondrial calcium uniporter (mcu) nuclear gene encoding mitochondrial protein mRNA	NM..001130227.1	0.91	1.81	1.01	0	0	0	0.01
Xenopus laevis cDNA clone IMAGE:6957925	BC..30190.1	4.84	9.68	1.01	0	0	0	0.01
Xenopus laevis fibroblast growth factor 7 (fgf7) mRNA	NM..001096168.1	1.27	2.54	1.01	0	0	0	0.01
Xenopus laevis lipase endothelial (lipg) mRNA	NM..001090061.1	0.79	1.72	1.12	1.38	1.38	0.01	0.01
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor 56 (gpr56) mRNA	XW..002931653.2	0.47	1.48	1.68	1.79	1.79	0.01	0.01
Xenopus laevis similar to casqueinut 2 (cardiac muscle) mRNA (cDNA clone IMAGE:4681496) partial cds	NM..001094724.1	2.8	7.98	1.52	3.64	3.66	0.01	0.01
Xenopus laevis chromosome A (parathyroid calbindin D28k) mRNA	NM..001092314.1	0.76	1.89	1.33	1.16	1.17	0.01	0.01
Xenopus laevis cryptochrome 2b mRNA complete cds	AY049035.1	1.58	10.36	2.73	2.39	2.43	0.03	0.01
Xenopus laevis acyl-CoA synthetase medium-chain family member 3 (acsmt3) mRNA	NM..001092901.1	9.05	19.98	1.15	4.42	4.51	0.03	0.03
Xenopus laevis family with sequence similarity 101 member B (fam101b) mRNA	NM..001093870.1	7.46	19.75	1.41	9.23	9.48	0.04	0.03
XLJ7636 Xenopus laevis calbindin D28k mRNA complete cds	U76636.1	0.94	4.04	2.11	1.3	1.4	0.11	0.06
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100491360 (LOC100491360) transcript variant X1 mRNA	NM..004917936.1	5.38	14.76	1.46	29.58	313.13	0.09	0.06
PREDICTED: Xenopus (Silurana) tropicalis extended synaptotagmin-1-like (LOC100493855) mRNA	XW..004914206.1	1.7	6.35	1.91	1.84	1.99	0.12	0.07
XELPDFAA XIaevis platelet-derived growth factor A 4-domain bicarbonate transporter member 4 (slc4a4)	NM..004919133.1	1.04	2.82	1.44	1.54	1.63	0.09	0.07
XENOPUS laevis family with sequence similarity 101 member B (fam101b) mRNA	M23228.1	3.91	10.18	1.39	5.15	5.46	0.09	0.07
XENOPUS laevis ATPase Na+/K+-transporting beta 1 polypeptide (atp1b1) mRNA	NM..001086759.1	57.26	131.35	1.2	28.11	29.57	0.08	0.07
Xenopus laevis TGF-beta 2 mRNA (for transforming growth factor-beta 2)	X51817.1	0.68	1.6	1.25	1.52	1.6	0.08	0.07
Xenopus laevis mRNA for enhancer of split related 9 (esr9 gene) clone 8c9	AJ009282.1	1.19	3.66	1.63	1.63	1.75	0.11	0.07
Xenopus laevis prostaglandin reductase 2 (pgtr2) mRNA	NM..001095485.1	2.27	5.97	1.4	8.42	8.97	0.1	0.07
Xenopus laevis 7-transmembrane receptor-like protein (7tmprss13) mRNA	AF231711.1	1.4	3.53	1.34	1.39	1.48	0.1	0.07
Xenopus laevis Ras-related associated with diabetes (rad1) mRNA	NM..001092750.1	0.02	2.99	8	1.03	1.68	0.72	0.09
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 13 (tmprss13) mRNA	NM..002932904.2	1.19	3.33	1.5	2	2.2	0.14	0.09
Xenopus laevis mRNA similar to erythrocyte protein band 4.1-like 1 mRNA (cDNA clone IMAGE:4889018) partial cds	BC044964.1	1.25	3.48	1.49	6.96	7.63	0.14	0.09
PREDICTED: Xenopus (Silurana) tropicalis SH3-domain binding protein 1 (sh3bp1) transcript variant X1 mRNA	NM..002935188.2	1.1	3.18	1.54	1.14	1.26	0.15	0.1
Xenopus laevis CASP2 and RIPK1 domain-containing adaptor with death gene 2 (cadd2) mRNA	NM..001091948.1	1.67	4.4	1.41	1.9	2.09	0.15	0.1
Xenopus laevis serine/threonine kinase 4 (stk4) mRNA	NM..001089968.1	0.71	1.65	1.23	1.24	1.37	0.14	0.12
PREDICTED: Xenopus (Silurana) tropicalis FH2 domain-containing protein 1-like (LOC100496216) mRNA	NM..002934907.2	0.54	1.94	1.86	1.28	1.49	0.22	0.12
Xenopus (Silurana) tropicalis finished cDNA clone TNel21R01	CP926202.2	16.97	34.6	1.03	12.92	14.07	0.13	0.12
Xenopus (Silurana) tropicalis sex determining region Y-box 2 (sox2) mRNA	NM..213704.3	35.87	72.35	1.02	49.46	53.9	0.13	0.13
Xenopus laevis solute carrier family 22 (organic anion transporter) member 7 (slc22a7) mRNA	NM..001092498.1	3.66	13.67	1.91	18.04	21.34	0.25	0.13
Xenopus laevis complement factor I (cf-i) mRNA	NM..001085952.1	1.74	4.35	1.33	1.3	1.49	0.17	0.13
Xenopus laevis cone-rod homeobox (crx-b) mRNA	NM..001088447.1	4.03	8.53	1.09	8.52	9.41	0.15	0.14

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC Six1 <sup>a</sup>	Control CHX	Control CHX+DEX	FC Control CHX	Rank <sup>b</sup>
PREDICTED; Xenopus (Silurana) tropicalis CTTNBP2 N-terminal like (cttnbp2nl) transcript variant X1 mRNA	XM_002932128.2	1.06	2.43	1.21	1.5	1.7	0.18	0.15
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC100487960 (LOC100487960) transcript variant X1 mRNA	XM_002939939.2	6.53	13.84	1.09	2.59	2.89	0.16	0.15
PREDICTED; Xenopus (Silurana) tropicalis integrin beta 4 (itgb4) transcript variant X1 mRNA	XM_002939974.2	0.77	1.94	1.34	1.66	1.9	0.2	0.15
Xenopus laevis POU class 3 homeobox 2 (pou3f2-b) mRNA	NM_001096751.1	0.44	3.47	3	1.01	1.38	0.45	0.15
Xenopus (Silurana) tropicalis G-protein-coupled receptor 153 (grp153) mRNA	NM_001128052.1	1.29	7.07	2.46	1.32	1.71	0.38	0.16
Xenopus laevis uncharacterized protein MGCI16527 (MGCI16527) mRNA	NM_001096165.1	2.55	5.3	1.06	2.41	2.7	0.17	0.16
PREDICTED; Xenopus (Silurana) tropicalis T-cell leukemia homeobox 1 (ttx1) transcript variant 1 mRNA	XM_002936768.2	1.01	5.73	2.52	1.12	1.47	0.4	0.16
Xenopus laevis cardiac myosin heavy chain-alpha (MHC-alpha) mRNA complete cds	AY913767.1	1.99	5.1	1.37	4.01	4.66	0.22	0.17
Xenopus laevis CD81 antigen (target of anti-proliferative antibody 1) mRNA (cDNA clone MGCI52702 IMAGE:4682721) complete cds	BC041217.1	25.98	82.18	1.67	34.23	41.3	0.28	0.17
Xenopus laevis regulator of cell cycle (rgcc) RNA	NM_001093976.1	30.73	72.87	1.25	32.68	37.69	0.21	0.17
Xenopus laevis cDNA clone IMAGE:405515 partial cds	BC072262.1	1.85	6.07	1.73	1.38	1.69	0.3	0.17
Xenopus laevis uncharacterized protein MGCI83079 (MGCI83079) mRNA	NP_001084719.1	9.68	38.06	1.98	17.66	22.59	0.36	0.18
Xenopus laevis uncharacterized protein MGCI30961 (MGCI30961) mRNA	NM_001096456.1	7.64	19.78	1.38	7.5	8.91	0.25	0.19
Xenopus laevis tspanparin 1 (tspan1) mRNA	NM_001095473.1	17.7	39.82	1.17	20.69	23.95	0.22	0.19
PREDICTED; Xenopus (Silurana) tropicalis family with sequence similarity 198 member A (fam198a) mRNA	XM_002937953.2	1.88	5.9	1.66	2.49	3.1	0.32	0.2
PREDICTED; Xenopus (Silurana) tropicalis progestin and adiponQ receptor family member IX (pacq9) mRNA	XM_004914351.1	1.99	6.15	1.63	1.47	1.83	0.32	0.2
Xenopus laevis cDNA clone IMAGE:085355 partial cds	BC073731.1	5.3	12.21	1.21	3.75	4.44	0.25	0.21
Xenopus laevis cytochrome P450 family 2 subfamily C polypeptide 18 (cytP2c18) mRNA	NM_001091776.1	96.74	421.33	2.13	34.82	47.17	0.44	0.21
Xenopus laevis Myoblast determination protein 1 (homolog A) mRNA (cDNA clone MGCI52596 IMAGE:488525) complete cds	BC041190.1	3.56	38.7	3.45	3.86	6.38	0.73	0.22
Xenopus (Silurana) tropicalis uncharacterized LOC100158501 (LOC100158501) mRNA	NM_001127941.1	1.29	3.17	1.31	1.04	1.26	0.29	0.22
Xenopus laevis finished cDNA clone TNed010893	CR761907.2	1.65	3.59	1.13	1.7	2.03	0.26	0.23
PREDICTED; Xenopus (Silurana) tropicalis CD48 antigen-like (LOC101731872) mRNA	XM_004919314.1	2.02	4.9	1.28	3.69	4.5	0.29	0.23
Xenopus tropicalis finished cDNA clone Tga091104	CR855728.2	18.98	39.66	1.07	3.09	3.65	0.24	0.23
Xenopus laevis MGCI80983 protein (MGCI80983) mRNA	NM_001092413.1	0.61	1.49	1.32	2.56	3.15	0.3	0.23
Xenopus tropicalis finished cDNA clone Tga006k01	CR848587.2	0.72	1.6	1.17	2.84	2.27	0.24	0.22
Xenopus laevis neurotrophin 3 (ntf3) mRNA	NM_0010922740.1	1.13	2.87	1.35	1.33	1.66	0.32	0.23
Xenopus (Silurana) tropicalis uncharacterized LOC549508 (LOC549508) mRNA	NM_001016754.2	106.61	319.8	1.59	51.06	66.83	0.39	0.25
Xenopus laevis cDNA clone IMAGE:7009602	BC084260.1	6.83	23.76	1.8	4.3	5.84	0.45	0.25
PREDICTED; Xenopus (Silurana) tropicalis hairy and enhancer of split 8 (hes8) mRNA	XM_002933849.2	1.67	11.43	2.79	1	1.64	0.72	0.26
Xenopus laevis mRNA for thimer oligopeptide complete cds	A053094.1	3.7	48.93	3.73	3.38	6.77	1.01	0.27
Xenopus laevis p21 GTPase-associated kinase 1 mRNA (cDNA clone MGCI548287) complete cds	BC081113.1	5.42	12.47	1.21	7.71	9.74	0.34	0.28
Xenopus (Silurana) tropicalis CD48 antigen-like (LOC10173522) transcript variant X2 mRNA	NM_004910924.1	1.43	3.45	1.28	2.61	3.35	0.37	0.29
Xenopus laevis mitochondrial ribosomal protein S2 (mrs2) nuclear gene encoding mitochondrial protein mRNA	NM_001091834.1	1.42	3	1.09	4.61	5.7	0.31	0.29
Xenopus laevis cDNA clone MGCI130954 IMAGE:7973902 complete cds	BC106382.1	2	7.97	2	1.9	2.86	0.6	0.3
Xenopus laevis cDNA clone IMAGE:8332229	BC155363.1	6.15	16.75	1.45	8.37	11.34	0.44	0.31
Xenopus laevis p21 GTPase-associated kinase 1 (PAK1) mRNA complete cds	AF169794.1	0.82	1.82	1.17	4.11	5.25	0.36	0.31
PREDICTED; Xenopus (Silurana) tropicalis zinc-finger protein XSPR2 mRNA (cDNA clone MGCI5154993) partial cds	BC072763.1	7.32	18.38	1.33	7.2	9.82	0.32	0.34
Xenopus laevis uncharacterized LOC495489 (LOC495489) mRNA	NM_001095132.1	1.46	3.35	1.21	1.2	1.57	0.39	0.32
Xenopus laevis zinc-finger protein S21 (znf521) mRNA	NM_001091128.1	4.4	10.22	1.22	4.49	5.87	0.39	0.36
PREDICTED; Xenopus (Silurana) tropicalis proline rich 15 (prn15) mRNA	NM_002933388.1	1.47	4.42	1.59	4.43	5.05	0.52	0.33
Xenopus tropicalis finished cDNA clone TNed052k18	CR76018.2	0.51	2.9	2.53	0.83	1.5	0.86	0.34
PREDICTED; Xenopus (Silurana) tropicalis myb-related transcription factor partner of profilin-like (LOC101733690)	NM_004917223.1	7.58	17.54	1.22	9	12.09	0.43	0.36
Xenopus laevis SRY (sex determining region Y)-box 21 (sox21) mRNA	CU075546.1	0.21	1.05	2.35	1.03	1.84	0.84	0.36
Xenopus (Silurana) tropicalis Tcf10/100h07	NM_001006681.9	0.18	3.37	4.25	0.39	1.13	1.53	0.36
Xenopus laevis alkyne repeat 2 (asrb2) mRNA	NM_001093794.1	0.72	1.76	1.29	0.74	1.02	0.48	0.37
PREDICTED; Xenopus (Silurana) tropicalis CD48 antigen-like (LOC10173236) mRNA	NM_004919320.1	0.94	1.89	1.02	1.32	1.72	0.38	0.37
Xenopus laevis uncharacterized LOC495222 (LOC495222) mRNA	NM_001094904.1	1.03	2.26	1.15	1.99	2.67	0.43	0.37
PREDICTED; Xenopus (Silurana) tropicalis hepatic and glial cell adhesion molecule (hepcam) mRNA	NM_002937519.2	1.53	3.12	1.03	1.46	1.91	0.39	0.38
Xenopus laevis degf03 mRNA complete sequence	DQ096846.1	1	4.21	2.08	0.58	1.01	0.81	0.39

Gene	Accession	Six1 CHX	Six1 CHX+DEX <sup>2</sup>	FC Six1 <sup>1</sup>	Control CHX	Control CHX+DEX <sup>3</sup>	FC Control CHX	Rank <sup>7</sup>
Xenopus (Silurana) tropicalis SL LIM homeobox 2 (sl2) mRNA	NM_001166041.1	5.76	15.63	1.45	5.65	8.33	0.57	0.39
Xenopus (Silurana) tropicalis integrin beta 6 (itgb6) mRNA	NM_001097306.1	0.38	1.83	2.27	0.67	1.24	0.91	0.4
PREDICTED: Xenopus (Silurana) tropicalis putative N-acetyltransferase 16-like (LOC100490742) mRNA	NM_002943189.1	0.8	3.27	2.05	0.82	1.48	0.86	0.42
PREDICTED: Xenopus (Silurana) tropicalis T-box 15 (tbx15) mRNA	NM_002940981.2	0.87	3.3	1.93	0.79	1.39	0.81	0.42
PREDICTED: Xenopus (Silurana) tropicalis WD repeat domain 27 (wdr27) mRNA	NM_002931515.2	4.35	9.95	1.2	1.26	1.78	0.51	0.42
Xenopus laevis KIAA0930 (kaad930) mRNA	NM_001086221.1	1.96	5.48	1.49	3.28	5.06	0.63	0.43
Xenopus (Silurana) tropicalis protein phosphatase 2 regulatory subunit B' beta (pp2cb5b) mRNA	NM_001100279.1	0.37	1.85	2.36	0.51	1.02	1.01	0.43
Xenopus (Silurana) tropicalis cytochrome P450 family 2 subfamily C polypeptide 18 (cytP2c18) mRNA	NM_001112980.1	2.15	9.09	2.08	2.38	4.46	0.91	0.44
Xenopus laevis claudin 3 (cldn3) mRNA	NM_001093931.1	0.89	3.6	2.03	1.06	1.96	0.9	0.45
PREDICTED: Xenopus (Silurana) tropicalis growth factor independent 1 transcription repressor (gf1) mRNA	NM_002933803.2	1.39	4.54	1.72	0.78	1.32	0.76	0.45
Xenopus laevis cDNA clone MGCG-181613 IMAGE8737127 complete cds	BC154955.1	12.76	45.6	1.84	12.92	23.07	0.84	0.46
Xenopus laevis glutathione S-transferase alpha 4 (gstaa4) mRNA	NM_001096027.1	0.95	1.97	1.06	1.08	1.51	0.49	0.47
Xenopus laevis Hes 2 mRNA (cDNA clone IMAGE7011946) partial cds	BC084134.1	7.41	23.73	1.68	6.02	10.31	0.78	0.47
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101732195 (LOC101732195) mRNA	NM_004912378.1	0.56	2.08	1.92	0.98	1.87	0.95	0.5
Xenopus tropicalis finished cDNA clone TIP008c06	CP926432.2	0.61	1.61	1.42	0.75	1.24	0.74	0.52
Xenopus (Silurana) tropicalis atlastin GTPase 1 (atl1) mRNA	NM_001078754.1	0.62	2.02	1.72	0.66	1.22	0.9	0.53
Xenopus (Silurana) tropicalis tumor necrosis factor receptor superfamily member 21 (tnfrsf21) mRNA	NM_001079136.1	4.39	9.14	1.07	2.95	4.39	0.58	0.55

<sup>1</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1 and CHX treatment

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1 and CHX+DEX treatment

<sup>3</sup> Log<sub>2</sub> Fold change values for Six1

<sup>4</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup> Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup> Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Six1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## E.2 Six1; Down-regulated genes

**Table E.2** Genes with at least two-fold down-regulation after injection of Six1-GR and treatment with CHX + DEX

Gene	Accession	Six1 CHX <sup>a</sup>	Six1 CHX+DEX <sup>b</sup>	FC Six <sup>c</sup>	Control CHX <sup>d</sup>	Control CHX+DEX <sup>e</sup>	FC Control CHX+DEX <sup>f</sup>	Rank <sup>g</sup>
Xenopus laevis histone cluster 2 H2ab (hist2h2ab) mRNA	NM_001092630.1	1.45	0.01	-9.5	0	0	0	0
Xenopus tropicalis finished cDNA clone Theu12e04	CR855425.2	19.93	0.66	-4.87	5.7	9.45	0.73	0
PREDICTED: Xenopus (Silurana) tropicalis SVOP-like (svop) mRNA	XM_002940328.2	3.98	0.26	-3.98	1.91	2.26	0.25	0
Xenopus laevis adenosine deaminase domain containing 1 (testis-specific) (adad1) mRNA	NM_001096430.1	1.16	0.08	-3.97	0	0	0	0
XELUVS2 Xlaevis UVS-2 protein gene 3' end	M27162.1	539.15	38.91	-3.8	273.92	419.74	0.62	0
Xenopus laevis cyclin A1 mRNA (cDNA clone NIGC-132235 IMAGE:5048077) complete cds	BC11150.1	1.16	0.1	-3.68	0	0	0	0
Xenopus laevis WW and C2 domain containing 1 mRNA (IMAGE:557647) partial cds	BC04290.1	157.94	14.69	-3.43	25.38	43.13	0.77	0
PREDICTED: Xenopus (Silurana) tropicalis VP530 domain-containing receptor SorsCS1-like (LOC101733791) partial mRNA	XM_004920598.1	22.71	2.12	-3.43	0	0	0	0
Xenopus laevis ferritin heavy chain mRNA complete cds	AF538970.1	1.88	0.18	-3.41	0	0	0	0
Xenopus laevis mRNA for PYL1 (prepro-PYL1) precursor from skin	X01824.1	744.08	82.87	-3.17	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis neurogenin locus notch homolog protein 1-like (LOC100488695) mRNA	XM_002942601.2	1.35	0.16	-3.16	0	0	0	0
Xenopus laevis Ly-6/uPAR-related protein-1 (LURP-1) mRNA complete cds	AF364819.1	1.84	0.22	-3.13	0	0	0	0
Xenopus (Silurana) tropicalis myosin heavy chain 2 skeletal muscle adult (myh2) mRNA	NM_001096804.1	1.14	0.15	-3	0	0	0	0
Xenopus laevis high-mobility group nucleosome binding domain 1 mRNA (cDNA clone MGC:64449 IMAGE:6880568) complete cds	BC054972.1	8.33	1.17	-2.84	333.6	363.56	0.13	0
XELC3 X.laevis complement component 3 (C3) mRNA 3' end	M21790.1	1.12	0.17	-2.8	0	0	0	0
Xenopus laevis F-box and leucine-rich repeat protein 5 (fbx5) mRNA	NM_001087279.1	3204.58	467.36	-2.78	305.78	326.96	0.1	0
Xenopus tropicalis cDNA clone IMAGE:7690587	BC161097.1	1.03	0.16	-2.71	0	0	0	0
Xenopus laevis mitogen-activated protein kinase 7 (mapk7) mRNA	NM_001087648.1	3382.5	537.07	-2.66	274.19	382.01	0.48	0
PREDICTED: Xenopus (Silurana) tropicalis roundabout axon guidance receptor homolog 2 (Drosophila) (robo2) transcript variant X2 mRNA	XM_004912079.1	1.51	0.27	-2.54	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis 26-like (LOC100493683) mRNA	XM_002941439.2	2.87	0.52	-2.47	1.26	1.61	0.36	0
PREDICTED: Xenopus (Silurana) tropicalis collagen type XVII alpha 1 (col16a1) transcript variant X2 mRNA	XM_004912079.1	1.19	0.22	-2.46	0	0	0	0
Xenopus (Silurana) tropicalis vomeronasal type-2 receptor homolog 2 (mboat2) mRNA	NM_001102685.1	2.56	0.48	-2.44	1.11	1.24	0.17	0
Xenopus (Silurana) tropicalis membrane bound O-acetyltransferase domain containing 2 (mboat2) mRNA	NM_001032331.1	1.07	0.21	-2.34	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis N-myristoyl transferase (nmtf) mRNA	XM_004919395.1	1.29	0.26	-2.34	0	0	0	0
Xenopus laevis enolase-phosphatase 1 (enol1) mRNA	NM_001095889.1	1.91	3.06	-2.28	1.56	1.8	0.22	0
PREDICTED: Xenopus (Silurana) tropicalis glutamate receptor ionotropic N-methyl-D-aspartate 3B (grin3b) mRNA	XM_002937382.2	1.42	0.3	-2.25	0	0	0	0
Xenopus (Silurana) tropicalis protein phosphatase 1 regulatory subunit 12B (pp12b) mRNA	NM_001079292.1	62.12	13.24	-2.24	0	0	0	0
Xenopus laevis mRNA for hatching enzyme complete cds	D89632.1	82.66	17.86	-2.22	72.54	101.29	0.49	0
Xenopus (Silurana) tropicalis ElMO/CED-12 domain containing 1 (elmod1) mRNA	NM_001078847.1	1.04	0.24	-2.18	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100485064 (LOC100485064) transcript variant X2 mRNA	XM_004918508.1	105.65	243.19	-2.12	0	0	0	0
Xenopus laevis transporter associated with antigen processing 2 (TAP2) mRNA Tap2-1 allele complete cds	AY204554.1	5.04	1.17	-2.11	2.19	4.15	0.93	0
PREDICTED: Xenopus (Silurana) tropicalis phosphopantethenylysteine decarboxylase-like (LOC100495698) transcript variant X2 mRNA	XM_004915988.1	489.05	114.31	-2.1	17.86	20.16	0.18	0
PREDICTED: Xenopus (Silurana) tropicalis tyrosine receptor (tyr1) transcript variant X2 mRNA	XM_004917103.1	1.65	0.4	-2.07	0	0	0	0
Xenopus laevis uncharacterized protein MGCI31332 (MGCI31332) mRNA	NM_001096476.1	313.69	753.75	-2.06	206.4	230.3	0.16	0
Xenopus tropicalis protein mRNA (cDNA clone MGCI39417 IMAGE:6990058) complete cds	BC075526.1	5.89	1.43	-2.05	5.56	7.94	0.52	0
Xenopus tropicalis cDNA clone IMAGE:7692177	BC121991.1	1.16	0.29	-2.03	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00496845 (LOC100496845) mRNA	XM_002935176.2	651.37	160.86	-2.02	449.83	2036.85	2.18	0
PREDICTED: Xenopus (Silurana) tropicalis mediator of RNA polymerase II transcription subunit 24-like (LOC100491557) transcript variant X1 mRNA	XM_002937856.2	1.18	0.3	-2.02	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis anchor protein 6 (akap6) transcript variant X2 mRNA	XM_004917275.1	1.84	0.47	-1.99	0	0	0	0
Xenopus laevis ecto-nucleoside triphosphate diphosphohydrolase 7 mRNA partial cds	DQ117608.1	190.66	48.39	-1.98	33.3	34.81	0.07	0
Xenopus (Silurana) tropicalis ras homolog family member H (rhoh) mRNA	NM_001126963.1	2.12	0.35	-1.96	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00490207 (LOC100490207) mRNA	NM_001094695.1	292.48	77.56	-1.92	79.36	85.32	0.11	0
Xenopus laevis laminin beta 2 (laminin S) (lamb2) mRNA	BC129550.1	201.04	53.62	-1.91	51.44	169.75	1.73	0
Xenopus tropicalis finished cDNA clone TGA098017	CR942563.2	302.6	81.12	-1.9	44.24	50.01	0.18	0
Xenopus (Silurana) tropicalis phd finger protein 19 (phf19) mRNA	NM_001078978.1	1.76	0.47	-1.9	0	0	0	0

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC	Six1 CHX	Control CHX	FC	Control CHX+DEX	FC	Control CHX+DEX	Rank <sup>a</sup>
Xenopus laevis cDNA clone IMAGE:115069 [IMAGE:3404671] complete cds	BC100222.1	195:83	529:82	-1.89	323:66	782:18	1.28	0			
Xenopus laevis malic enzyme 3 NADP(+)-dependent mitochondrial (m3) nuclear gene encoding mitochondrial protein mRNA	NM_001095050.1	50:78	13:81	-1.88	12.89	19.21	0.58	0			
PREDICTED: Xenopus (Silurana) tropicalis tenasin XB (tnxb) mRNA	XM_002941267.2	1.36	0.37	-1.88	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis polymerase 4-like (LOC100991505) mRNA	XM_002957967.2	1.17	0.33	-1.84	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 58A-like (LOC100487634) transcript variant X1 mRNA	XM_004919038.1	22:23	62:71	-1.83	45.53	60.18	0.41	0			
PREDICTED: Xenopus (Silurana) tropicalis poly (ADP-ribose) polymerase family member 4 (parp4) mRNA	XM_002957966.2	3.03	0.87	-1.81	0	0	0	0			
Xenopus laevis (Silurana) tropicalis zinc finger protein 58A-like (LOC100487634) transcript variant X2 mRNA	CR761732.2	1.17	0.34	-1.79	0	0	0	0			
Xenopus laevis ubiquitin-conjugating enzyme E2K (ube2k) mRNA	NM_001096821.1	133:44	39:27	-1.77	34.98	35.34	0.02	0			
Xenopus (Silurana) tropicalis novel protein containing an inosine-uridine preferring nucleoside hydrolase domain (LOC734042) mRNA	NM_001045726.1	1.13	0.34	-1.75	0	0	0	0			
Xenopus laevis homeobox protein Otx2 mRNA complete cds	AF081352.1	1.37	0.42	-1.74	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis stialic acid binding Ig-like lectin 1 stiabodhesin (siglec1) mRNA	XM_002938400.2	1.7	0.52	-1.74	0	0	0	0			
Xenopus laevis circadian rhythmicity protein CLOCK mRNA complete cds	AF203107.1	369:09	112	-1.73	1.72	3.54	1.05	0			
Xenopus laevis cDNA clone IMAGE:7211103	BC106476.1	1.8	0.55	-1.72	1.79	4.84	1.45	0			
Xenopus tropicalis cDNA clone IMAGE:7017207	BC087776.1	1.57	0.49	-1.71	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis retina and anterior neural fold homeobox (rax) mRNA	XM_002936669.2	1.25	0.39	-1.7	1.01	1.89	0.91	0			
PREDICTED: Xenopus (Silurana) tropicalis neurofascin (nfasc) transcript variant X8 mRNA	XM_004910702.1	1.12	0.35	-1.7	0	0	0	0			
Xenopus laevis cDNA clone IMAGE:6635274	BC108828.1	7.98	2.98	-1.69	0	0	0	0			
Xenopus laevis cDNA clone IMAGE:7211103	NM_001092943.1	1.09	0.35	-1.67	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis clone IMAGE:7017207	NM_001096745.1	1113:763	353:57	-1.66	81.94	124:24	0.61	0			
PREDICTED: Xenopus (Silurana) tropicalis synaptotagmin 4 (syt4) mRNA	NM_001091620.1	1.32	0.42	-1.65	0	0	0	0			
Xenopus laevis pyruvate dehydrogenase E1 alpha 1 mRNA (cdna clone MGC:132282 [IMAGE:3399753]) complete cds	NM_001171514.1	1.29	0.41	-1.65	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis myosin VIIA (myo7a) transcript variant X2 mRNA	BC106670.1	271:327	868:78	-1.65	126.66	162.61	0.37	0			
PREDICTED: Xenopus (Silurana) tropicalis fibroblast growth factor 9 (glia-activating factor) (fgf9) mRNA	XM_00491226.1	4.19	1.35	-1.64	0	0	0	0			
Xenopus laevis coiled-coil domain containing 3 (ccdc3) mRNA	NM_002938575.2	1.08	0.35	-1.63	0	0	0	0			
Xenopus laevis Rho GTPase activating protein 30 (argap30) mRNA	NM_001093552.1	1.37	0.45	-1.62	0	0	0	0			
Xenopus laevis ankyrin repeat domain 1 (cardiac muscle) (ankrd1) mRNA	NM_001096171	3.19	1.04	-1.62	0	0	0	0			
Xenopus laevis uncharacterized LOC496105 (LOC496105) mRNA	NM_001095360.1	1.22	0.41	-1.61	0	0	0	0			
Xenopus laevis RNA binding motif protein 23 (rbm23) mRNA	NM_001092339.1	199:85	665:92	-1.58	158.55	238.74	0.71	0			
Xenopus laevis hypothetical protein LOC100127287 mRNA (cdna clone IMAGE:88222377) partial cds	BC1155883.1	61:64	20:7	-1.58	16.31	18.27	0.17	0			
Xenopus laevis neuronal differentiation 1 (neurod1-b) mRNA	NM_001092127.1	263:51	89:04	-1.57	30.95	33.76	0.13	0			
Xenopus laevis family with sequence similarity 98 member B (fam98b) mRNA	NM_001093330.1	154:31	52:2	-1.57	122.52	282.09	1.21	0			
PREDICTED: Xenopus (Silurana) tropicalis BR serine/threonine kinase 1 (brsk1) transcript variant X2 mRNA	XM_004916304.1	1.56	0.53	-1.56	0	0	0	0			
Xenopus laevis cDNA clone IMAGE:5156129	BC083028.1	1026:026	352:06	-1.55	792.53	1191.92	0.59	0			
PREDICTED: Xenopus (Silurana) tropicalis embryonic protein USV2-like (LOC100996122) transcript variant X1 mRNA	XM_002957302.2	5.09	1.79	-1.52	4.08	5.46	0.43	0			
Xenopus laevis dimethylarginine dimethylaminohydrolase 2 (ddah2) mRNA	NM_001094280.1	2.96	1.04	-1.52	0	0	0	0			
Xenopus laevis recombinase activating gene 1 (rag1) mRNA	JF439312.1	1.1	0.39	-1.51	0	0	0	0			
Xenopus laevis mitochondrial ribosomal protein-like (LOC101735189) transcript variant X2 mRNA	NM_001172083.1	1.48	0.53	-1.5	0	0	0	0			
Xenopus laevis mitochondrial fission factor 1 (mff1) nuclear gene encoding mitochondrial protein mRNA	XM_004915893.1	443:39	158:35	-1.49	18.84	70.63	1.91	0			
Xenopus laevis ventral pancreatic precursor 1 short isoform (vpbp1) mRNA complete cds alternatively spliced	NM_001093456.1	356:04	127:34	-1.49	20.27	264.63	0.39	0			
Xenopus laevis recombination activating gene 1 a (rag1) mRNA	JF439312.1	1.29	0.47	-1.48	0	0	0	0			
Xenopus laevis chromosome 21 open reading frame 2 (c21orf2) nuclear gene encoding mitochondrial protein mRNA	NM_001094049.1	86:58	31:18	-1.48	0	0	0	0			
Xenopus laevis cDNA clone IMAGE:6948748	BC094119.1	506:41	182:49	-1.48	83.09	95.07	0.2	0			
Xenopus tropicalis finished cDNA clone Teg018h13	CR761350.2	1.02	0.38	-1.46	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734416 transcript variant X1 mRNA	NM_004912947.1	4.42	1.63	-1.45	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis interleukin-17 receptor C-like (LOC100486154) mRNA	NM_002901672.2	1.26	0.46	-1.48	0	0	0	0			
Xenopus laevis cDNA clone IMAGE:5570266 partial cds	BC081240.1	129:79	48:12	-1.44	0	0	0	0			
Xenopus laevis cDNA clone IMAGE:6939268	NM_001096615.1	1.35	0.5	-1.43	10.31	11.96	0.22	0			
Xenopus laevis zinc finger protein MGCI14819 (MGCI14819) mRNA	NM_001096464.1	1.97	0.74	-1.43	3.56	3.74	0.08	0			
Xenopus laevis FZ2 family zinc finger 2 (fzef2) mRNA	NM_001095791.1	2.04	0.77	-1.43	3.19	3.28	0.04	0			
PREDICTED: Xenopus (Silurana) tropicalis ankyrin G (ankyrin G) transcript variant X2 mRNA	NM_004920571.1	38:7	14.53	-1.42	0	0	0	0			
PREDICTED: Xenopus (Silurana) tropicalis ankyrin G (ankyrin G) transcript variant X3 mRNA	NM_001096138.1										
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731138 (LOC101731138) mRNA	NM_004920571.1										

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC Six1 CHX	FC Six1 CHX	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Rank <sup>a</sup>
Xenopus tropicalis finished cDNA clone TGasd076k16	CR761730_2	185.06	694.84	-1.42	97.55	142.27	0.55	0	
PREDICTED: Xenopus (Silurana) tropicalis translocated promoter region nuclear basket protein (tpn) transcript variant X2 mRNA	XM_0010913764.1	1.22	0.46	-1.41	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis proteasomal ATPase-associated factor 1-like (LOC100491421) mRNA	XM_002943314.2	1.01	0.39	-1.39	0	0	0	0	0
Xenopus laevis Leucine-rich repeat-containing protein 42 mRNA (cDNA clone IMAGE:68633997) partial cds	BC078092.1	132.28	509.27	-1.38	160.5	215.05	0.43	0	0
Xenopus laevis cDNA clone IMAGE:37245844	BC097849.1	3.16	1.22	-1.38	0	0	0	0	0
Xenopus laevis uncharacterized protein MGCB8546 (MGCB8546) mRNA	NM_001090093.1	1.05	0.41	-1.37	0	0	0	0	0
Xenopus laevis insect-derived growth factor-B-like protein mRNA complete cds	AY986979.1	138.71	541.37	-1.37	166.5	203.7	0.3	0	0
PREDICTED: Xenopus (Silurana) tropicalis embryonic protein USV-2-like (LOC101734297) transcript variant X3 mRNA	XM_0010913370.1	9.72	3.81	-1.36	0	0	0	0	0
Xenopus laevis uncharacterized LOC100505442 (LOC100505442) mRNA	NM_001197278.1	393.29	154.06	-1.36	37.12	42.62	0.2	0	0
Xenopus laevis hypothetical protein LOC73266 mRNA (cDNA clone IMAGE:6324856) partial cds	BC09782.1	7.98	3.13	-1.35	0	0	0	0	0
Xenopus laevis hypothetical protein LOC43691 mRNA (cDNA clone IMAGE:5082323) partial cds	BC07379.1	243.14	95.82	-1.35	46.67	47.64	0.03	0	0
Xenopus laevis carbonic anhydrase XIII (ca13) mRNA	NM_001093512.1	1.2	0.48	-1.34	0	0	0	0	0
Xenopus (Silurana) tropicalis hydroxysteroid dehydrogenase 3 beta- and steroid delta-isomerase 7 (hsd3b7) mRNA	NM_001127029.1	1.61	0.64	-1.34	0	0	0	0	0
Xenopus laevis uncharacterized LOC100505442 (LOC100505442) mRNA	DQ646916.1	1.93	0.77	-1.34	0	0	0	0	0
Xenopus laevis solute carrier family 44 member 1 (slc4a1) mRNA	NM_001091778.1	862.85	344	-1.33	181.1	193.92	0.1	0	0
Xenopus laevis carbonic anhydrase XII (ca12) mRNA	XM_002943984.2	2.16	0.87	-1.33	0	0	0	0	0
Xenopus (Silurana) tropicalis hydroxysteroid dehydrogenase 3 beta- and steroid delta-isomerase 7 (hsd3b7) mRNA	XM_0010916850.1	1.74	0.7	-1.33	0	0	0	0	0
Xenopus laevis receptor tyrosine kinase Erbb4 mRNA complete cds	D42060.1	65.54	26.27	-1.32	9.82	14.46	0.56	0	0
XENXO1A: Xenopus laevis mRNA for pleiotropic factor-beta2 complete cds	NM_001091174.1	231.4	92.77	-1.32	26.71	39.63	0.57	0	0
Xenopus laevis potassium channel tetramerization domain containing 14 (kctd14) mRNA	DQ096919.1	574.33	231.05	-1.32	114.82	190.48	0.74	0	0
Xenopus laevis comp37 mRNA complete sequence	AY160969.1	1.68	0.68	-1.32	0	0	0	0	0
Xenopus tropicalis homeodomain transcription factor (Hex) mRNA complete cds	BC159126.1	34.06	13.76	-1.31	0	0	0	0	0
Xenopus tropicalis cDNA clone MG_C172468 (MG_C172468) mRNA	NM_001094923.1	6.44	2.62	-1.31	2.11	2.76	0.39	0	0
Xenopus laevis transmembrane protein 82 (tnem82) mRNA	NM_002941330.2	2.36	0.96	-1.31	1.01	1.32	0.39	0	0
PREDICTED: Xenopus (Silurana) tropicalis tripartite motif-containing protein 7-like (LOC100494464) mRNA	AF168794.1	1091.56	445.39	-1.3	200.28	431.25	1.11	0	0
Xenopus laevis ribonucleotide reductase subunit M1 (RRM1) mRNA partial cds	XM_002939415.2	3.37	1.39	-1.29	3.48	3.76	0.12	0	0
PREDICTED: Xenopus (Silurana) tropicalis BA1-associated protein 3 (baip3) mRNA	XM_002931434.2	1.14	0.47	-1.29	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor 52 (gpr52) mRNA	XM_002938125.2	95.86	39.53	-1.29	128.6	159.14	0.31	0	0
PREDICTED: Xenopus (Silurana) tropicalis L-2-hydroxyglutarate dehydrogenase (l2hgdh) transcript variant 2 mRNA	XM_002941571.1	179.77	739.46	-1.28	39.62	103.78	1.39	0	0
PREDICTED: Xenopus (Silurana) tropicalis heavy polypeptide-like (LOC101732029) mRNA	BC084339.1	6.53	2.7	-1.28	1.31	1.62	0.3	0	0
Xenopus laevis comp37 mRNA containing frame-shift errors	EF010533.1	1.05	0.44	-1.28	0	0	0	0	0
Xenopus laevis glucose-dependent insulinotropic polypeptide mRNA complete cds	BC087617.1	423.03	176.48	-1.27	11.19	13.9	0.32	0	0
Xenopus laevis cDNA clone IMAGE:5514602	NM_001091902.1	22.42	9.39	-1.26	2.52	2.73	0.12	0	0
Xenopus laevis MGCB2394 protein (MGCB2394) mRNA	NM_001091302.1	2.94	1.24	-1.26	1.92	2.19	0.2	0	0
PREDICTED: Xenopus (Silurana) tropicalis gamma-1-phosphatidylinositol 4,5-bisphosphate phosphodiesterase eta-2-like (LOC100487353) mRNA	XM_002936582.2	2.47	1.04	-1.25	0	0	0	0	0
Xenopus laevis enhancer of split related protein-7 mRNA (cDNA clone IMAGE:5959168) partial cds	BC07298.1	1.2	0.51	-1.25	0	0	0	0	0
Xenopus (Silurana) tropicalis cholinergic receptor nicotinic alpha 4 (neuronal) (chnra4) mRNA	NM_001113843.1	30.98	13.07	-1.25	18.5	21.72	0.24	0	0
PREDICTED: Xenopus (Silurana) tropicalis 1-phosphatidylinositol 4,5-bisphosphate phosphodiesterase activating protein (gap) transcript variant X2 mRNA	XM_001091436.1	2.8	1.18	-1.25	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis c-X-C motif chemokine 10-like (LOC100490227) mRNA	DQ096849.1	483.79	204.33	-1.25	69.01	75.74	0.14	0	0
Xenopus laevis uncharacterized LOC100487353 mRNA	NM_001127039.1	1.1	0.47	-1.24	0	0	0	0	0
Xenopus laevis phosphodiesterase 8B (pde8b) transcript variant X1 mRNA	XM_00290874.2	1.41	0.61	-1.24	0	0	0	0	0
Xenopus laevis multi-homolog 1 colon cancer nonpolyposis type 2 (mh1) mRNA	NM_001098707.1	1.02	0.44	-1.23	0	0	0	0	0
Xenopus (Silurana) tropicalis oxy steroid binding protein-like 3 (osbp3) mRNA	NM_001256919.1	42.69	18.25	-1.23	11.78	15.19	0.37	0	0
PREDICTED: Xenopus (Silurana) tropicalis c-X-C motif chemokine 10-like (LOC100490227) mRNA	XM_002940575.1	2.91	1.25	-1.23	1.21	1.69	0.49	0	0
Xenopus laevis uncharacterized LOC100487353 mRNA	NM_001086446.1	1.76	0.75	-1.23	0	0	0	0	0
Xenopus laevis ectropic viral integration site 1 (evi1) mRNA complete cds	DQ088677.1	1.23	0.53	-1.23	0	0	0	0	0
Xenopus laevis histone H1 histone family member X (11x) mRNA	NM_001086796.1	99.58	42.69	-1.23	23.76	31.87	0.31	0	0
Xenopus (Silurana) tropicalis arhgap33 mRNA	NM_001097300.1	248.92	107.09	-1.22	11.1	11.33	0.03	0	0
PREDICTED: Xenopus (Silurana) tropicalis GTPase activating protein 33 (arhgap33) mRNA	CR762223.2	1.16	0.5	-1.22	0	0	0	0	0
Xenopus laevis histone H1 histone family member X (11x) mRNA	NM_001094932.2	97.08	41.93	-1.22	0	0	0	0	0
Xenopus laevis lactamase beta 2 (lacbt2) mRNA	NM_001094943.1	231.8653	100.25568	-1.21	175.34433	413960.9	1.24	0	0

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC	Six1 CHX	Control CHX	FC	Control CHX+DEX	FC	Control CHX	Rank <sup>a</sup>
Xenopus laevis cathepsin S (ctts-a) mRNA	NM_001087353.1	1.16	0.5	-1.21	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100486850 (LOC100486850) transcript variant X2 mRNA	XM_0041915362.1	92.36	40.13	-1.21	0	0	0	0	0	0	0
Xenopus laevis death inducer-blitzter 1 (dido1) mRNA	NM_001094588.1	1047.17	455.08	-1.21	238.74	281.96	0.25	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein containing 8 (zswim8) mRNA	XM_002935767.2	57.88	25.29	-1.2	15.53	18.18	0.23	0	0	0	0
Xenopus laevis hypothetical protein LOC100037229 mRNA (DNA clone [MAGE:8541663]) partial cds	BC129775.1	2.1	0.93	-1.19	0	0	0	0	0	0	0
Xenopus laevis MGc80465 protein (MGc80465) mRNA	NM_001092219.1	210.05	92.47	-1.19	35.43	36.14	0.03	0	0	0	0
Xenopus laevis uncharacterized LOC00101301 (LOC100101301) mRNA	NM_001099887.1	1822.93	804.94	-1.19	365.08	436.07	0.26	0	0	0	0
Xenopus laevis G patch domain and ankyrin repeats 1 (bpank1) mRNA	NM_001097131.1	1377.97	605.92	-1.19	194.14	259.78	0.43	0	0	0	0
Xenopus laevis SEC13 homolog (sec13) mRNA	NM_001086973.1	4014.16	1773.07	-1.18	198.79	804.95	2.02	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100488736 (LOC100488736) transcript variant X2 mRNA	XM_004916677.1	179.58	79.32	-1.18	37.79	42.25	0.24	0	0	0	0
Xenopus laevis cDNA clone MGc79935 (MAGE:8865551) complete cds	BC076771.1	791.12	349.55	-1.18	246.09	269.65	0.14	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis rho GTPase-activating protein 44-like (LOC100497222) mRNA	XM_002937019.2	1.23	0.55	-1.18	0	0	0	0	0	0	0
Xenopus laevis uncharacterized protein MGc81054 (MGc81054) mRNA	NM_001091253.1	1582.3	702.7	-1.18	276.15	312.57	0.18	0	0	0	0
Xenopus laevis seizure related 6 homolog-like 2 (sez6l2) mRNA	NM_001093958.1	1.58	0.7	-1.18	1.4	1.52	0.13	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00494305 (LOC00494305) partial mRNA	XM_002942838.2	11	4.9	-1.17	0	0	0	0	0	0	0
Xenopus laevis synaptic vesicle 2-related protein mRNA complete cds	DQ167576.1	2.35	1.06	-1.16	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis spermidine synthase (smr) transcript variant X1 mRNA	XM_002939409.2	2.05	0.92	-1.16	0	0	0	0	0	0	0
XLA001049 Xenopus laevis retinal homeobox 2A (R2A) mRNA complete cds	AF001049.1	1.53	0.69	-1.16	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) mucin-22-like (LOC101734541) mRNA	XM_004911425.1	1.07	0.48	-1.16	0	0	0	0	0	0	0
Xenopus laevis synovial sarcoma translocation gene on chromosome 18-like 1 (s18l1) mRNA	NM_001093632.1	171.12	77.12	-1.16	48.5	61.68	0.35	0	0	0	0
Xenopus laevis X-ray repair complementing defective repair in Chinese hamster cells 1 (xrccl1) mRNA	NM_001087242.1	30.28	137.19	-1.15	48	49.68	0.05	0	0	0	0
Xenopus laevis neuronal leucine-rich repeat protein 6 (NLRR-6) mRNA complete cds	DG315790.1	1.73	0.79	-1.14	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis piggybac transposable element-derived protein 4-like (LOC100488733) transcript variant X1 mRNA	XM_002939902.2	105.62	483.24	-1.13	22.54	35.95	0.68	0	0	0	0
Xenopus laevis cDNA clone MAGE:69464628	BC081201.1	1.06	0.49	-1.13	1.03	1.17	0.19	0	0	0	0
Xenopus laevis uncharacterized LOC1095835 (LOC1095835) mRNA	NM_001095192.1	630.05	289.98	-1.12	565.5	818.25	0.54	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis sodium channel voltage-gated type III alpha subunit (scn3a) mRNA	XM_002939271.2	1.04	0.48	-1.12	0	0	0	0	0	0	0
Xenopus laevis cDNA clone MGc130909 (MAGE:7320281) complete cds	BC106360.1	1.43	0.66	-1.12	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis polyplasmic polyadenylation element binding protein 2 (pepb2) transcript variant X2 mRNA	XM_004911230.1	297.24	137.22	-1.12	77.25	79.18	0.04	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis proline-serine-threonine phosphatase interacting protein 2 (pstPIP2) mRNA	XM_002936514.2	2.41	1.12	-1.12	3.76	5.18	0.47	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis apoptosis-associated speck-like protein containing a CARD-like (loc101732196) partial mRNA	XM_004920313.1	1.44	0.67	-1.11	0	0	0	0	0	0	0
XELPDS Xenopus laevis mRNA for prostaglandin D synthase complete cds	D83712.1	1.18	0.56	-1.09	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis trichohyalin-like (LOC100495334) mRNA	XM_00294276.1	2.79	1.3	-1.11	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis oligodendrocyte lineage transcription factor 2 (olig2) transcript variant X1 mRNA	XM_004912344.1	551.24	257.59	-1.1	137.87	138.42	0.01	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homolog (linos8) mRNA	XM_004917309.1	7.91	3.7	-1.1	8.48	9.19	0.12	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis painmodulatory transferase ZDHHC2-like (LOC101732299) mRNA	XM_004920314.7	1.78	0.87	-1.1	304.79	397.58	0.39	0	0	0	0
XELPDS Xenopus laevis mRNA for prostaglandin D synthase complete cds	D83712.1	1.18	0.56	-1.09	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis trichohyalin-like (LOC101731437) mRNA	XM_004913803.1	103.03	48.58	-1.09	29.97	34.97	0.23	0	0	0	0
Xenopus laevis HESX homeobox 1 (hesx1-b) mRNA	NM_001162570.1	1.22	0.58	-1.08	1.07	1.39	0.39	0	0	0	0
PREDICTED: Xenopus (Silurana) COUP transcription factor 1 mRNA (MGc196736 (MAGE:9041644)) complete cds	HM535699.1	1.46	0.56	-1.07	9.43	10.4	0.15	0	0	0	0
PREDICTED: Xenopus (Silurana) tau-like protein 1 (xtp1) mRNA complete cds alternatively spliced	BC170009.1	1.36	6.5	-1.0	1.33	1.42	0.1	0	0	0	0
PREDICTED: Xenopus (Silurana) painmodulatory transferase ZDHHC2-like (LOC101732299) mRNA	AY032847.1	1.24	0.61	-1.02	27.86	29.39	0.08	0	0	0	0
Xenopus (Silurana) tropicalis cyclin-dependent kinase inhibitor 2C (p18 inhibits CDK4) (cdkn2c) mRNA	NM_001123460.1	1.76	0.87	-1.04	1.21	1.29	0.09	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis retinol dehydrogenase 7 (rdh7) mRNA	NM_001085720.1	333.05	172.76	-1.04	112.23	119.24	0.09	0	0	0	0
Xenopus laevis ATP-binding cassette sub-family A member 3 (ABCA3) mRNA partial cds	HM535699.1	1.46	0.72	-1.03	7.87	8	0.03	0	0	0	0
PREDICTED: Xenopus (Silurana) COUP transcription factor 1 mRNA (DNA clone MGc196736 (MAGE:9041644)) complete cds	BC170009.1	1.4	0.56	-1.03	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tau-like protein 1 (xtp1) mRNA complete cds alternatively spliced	AY032847.1	1.24	0.61	-1.02	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) GTPase-activating protein rap1gap mRNA	NM_001094661.1	77.03	38.24	-1.04	1.21	1.29	0.09	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis carbohydrate (N-acetylgalactosamine 4-sulfate 6-O)-sulfotransferase 15 (chst15) mRNA	XM_002937806.2	1.34	0.67	-1.02	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) COUP transcription factor 1 mRNA (DNA clone MGc196736 (MAGE:9041644)) complete cds	XM_004916474.1	316.56	157.33	-1.01	51.84	62.74	0.28	0	0	0	0
PREDICTED: Xenopus (Silurana) COUP transcription factor 1 mRNA (DNA clone MGc196736 (MAGE:9041644)) complete cds alternatively spliced	CR942341.2	1.15	0.58	-1.01	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis protein kinase C delta type-like (LOC101734683) mRNA	NM_001092991.1	158.48	251.6	-2.66	262.85	260.12	-0.02	0.01	0.01	0.01	0.02
Xenopus laevis synaptotagmin-like 4 (sytl4) mRNA	NM_001096993.1	112.32	24.95	-2.18	5.2	5.05	-0.05	0.02	0.02	0.02	0.02

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC	Six1 CHX	Control CHX	FC	Control CHX+DEX	FC	Control CHX+DEX	Rank <sup>a</sup>
Xenopus laevis cytochrome b561 (cyb561) mRNA	NM_001095578.1	708.13	233.32	-1.61	166.87	163.36	-0.04	0.02			
Xenopus laevis glutaminyl-peptide cyclotransferase (qpcr) mRNA	NM_001094123.1	246.55	38.33	-2.69	8.63	8.32	-0.06	0.02			
Xenopus laevis homeodomain protein (phox2a) mRNA complete cds alternatively spliced	AY371496.1	90.33	34.25	-1.4	22.48	22.02	-0.03	0.03			
Xenopus laevis sphingosine-1-phosphate receptor 1 (s1pr1) mRNA	NM_001092761.1	1.37	0.45	-1.63	1.3	1.26	-0.04	0.03			
Xenopus laevis pyruvate dehydrogenase phosphatase catalytic subunit 2 (pdhc2) mRNA	NM_001096881.1	11.78	5.12	-1.21	1.39	1.37	-0.03	0.03			
Xenopus laevis HIV-1 Tat interactive protein 2 (3k0ba) (mifatip2) mRNA	NM_001096049.1	1881.99	2378.05	-2.99	48.48	45.67	-0.09	0.03			
Xenopus laevis MGC84737 protein (MGC84737) mRNA	NM_001094305.1	266.81	745.58	-1.85	174.63	168.33	-0.06	0.03			
Xenopus laevis uncharacterized LOC495021 (LOC495021) mRNA	NM_001094727.1	116.91	45.49	-1.37	37.75	36.59	-0.05	0.04			
Xenopus laevis family with sequence similarity 175 member B (fam17b5) mRNA	NM_001094420.1	97.85	41.44	-1.24	10.03	9.72	-0.05	0.04			
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733467 (LOC101733467) mRNA	XM_004917412.1	2.01	0.49	-2.05	2.93	2.76	-0.09	0.05			
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00495801 (LOC00495801) transcript variant X2 mRNA	XM_004919213.1	902.81	360.11	-1.33	55.08	52.69	-0.07	0.05			
Xenopus laevis membrane bound O-acyltransferase domain containing 1 (mboat1) mRNA	NM_001097001.1	228.94	51.84	-2.15	28.84	26.74	-0.11	0.06			
Xenopus (Silurana) tropicalis numb (numb) mRNA	NM_001097359.1	139.74	37.05	-1.92	17.42	16.23	-0.11	0.06			
Xenopus laevis retrotransposon-like element 10A1 gag-like protein mRNA partial cds, and 3' UTR complete sequence	AF45966.1	387.67	906.13	-2.1	739.44	683.24	-0.12	0.06			
Xenopus laevis splicing factor 1 (sf1) mRNA	NM_001087311.1	2.29	0.56	-2.05	49.68	45.91	-0.12	0.06			
Xenopus laevis dolichol kinase (dolk) mRNA	NM_001092539.1	404.14	166.69	-1.28	116.62	110.86	-0.08	0.06			
Xenopus laevis elongator acetyltransferase complex subunit 4 (elp4) mRNA	NM_001094894.1	186.55	41.4	-2.18	21.45	19.61	-0.13	0.06			
PREDICTED: Xenopus (Silurana) tropicalis TNF receptor-associated factor 2-like (LOC101734662) mRNA	NM_001094894.1	1.97	0.76	-1.38	1.3	1.23	-0.09	0.06			
Xenopus laevis cDNA clone IMAGE:904.1493	BC169859.1	118.92	41.92	-1.51	26.84	25.09	-0.1	0.07			
PREDICTED: Xenopus (Silurana) tropicalis multiple C2 domains transmembrane 2 (mctp2) transcript variant X1 mRNA	NM_002933312.2	1.8	0.77	-1.23	2.49	2.34	-0.09	0.07			
Xenopus laevis density protein 95 mRNA (CDNA clone IMAGE:7661545) partial cds	BC123007.1	1.75	0.81	-1.12	2.14	2.03	-0.08	0.07			
Xenopus laevis HoxD 3 protein mRNA (CDNA clone MGC:86545; IMAGE:3395571) complete cds	BC080434.1	1.22	0.52	-1.24	1.02	0.96	-0.1	0.08			
Xenopus (Silurana) tropicalis autophagy related 7 (atg7) mRNA	NM_001129922.1	113.49	413.72	-1.46	228.56	211.12	-0.12	0.08			
Xenopus laevis chromosome 4 open reading frame 22 (cdorf22) mRNA	NM_001095253.1	4.51	1.98	-1.21	33.18	30.96	-0.1	0.09			
Xenopus laevis retrochisin 1 (rs1) mRNA	NM_001096951.1	82.42	40.88	-1.02	2.21	2.05	-0.11	0.09			
PREDICTED: Xenopus (Silurana) tropicalis ALK tyrosine kinase receptor-like (LOC100495071) mRNA	NM_002938422.2	5.05	1.92	-1.4	38.74	36.28	-0.1	0.1			
Xenopus laevis ankyrin repeat domain 37 (ankrd37) mRNA	NM_001098697.1	4.25	1.47	-1.54	3.31	2.99	-0.15	0.1			
Xenopus laevis aryl-hydrocarbon receptor nuclear translocator 2 (arnt2) mRNA	NM_001087071.1	34.04	11.06	-1.63	12.01	10.73	-0.17	0.1			
PREDICTED: Xenopus (Silurana) tropicalis vomerine type-2 receptor 26-like (LOC100498040) mRNA	NM_002940523.2	3.02	1.14	-1.42	4.37	3.96	-0.15	0.1			
Xenopus laevis uncharacterized protein MGCI15605 (MGCI15605) mRNA	NM_001096076.1	158.78	647.88	-1.3	395.47	357.44	-0.15	0.12			
Xenopus laevis GID complex subunit 1 (rs1) mRNA	NM_001096765.1	290.58	750.29	-1.96	20.52	17.49	-0.24	0.12			
Xenopus (Silurana) tropicalis WW and C2 domain containing 2 (wwc2) mRNA	NM_001102742.1	557.48	154.36	-1.86	108.62	92.99	-0.23	0.13			
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 12 (sodium/potassium/chloride transporters) member 2 (slc12a2)	NM_0010941047.1	106.43	15.91	-2.75	20.24	16.04	-0.34	0.13			
XLRGAGP3-X:leuvs oocyte 35-s rRNA pseudogene clone px031	K01368.1	10.93	3.33	-1.72	1.55	1.34	-0.22	0.13			
Xenopus laevis KIAA1524 (kiaa1524) mRNA	NM_001094671.1	103.64	47.21	-1.14	17.28	15.65	-0.15	0.13			
Xenopus laevis dickkopf WNT signaling pathway inhibitor 3 (dkk3) mRNA	NM_001127781.1	1.43	0.68	-1.09	1.99	1.8	-0.15	0.14			
PREDICTED: Xenopus (Silurana) tropicalis sphingosine-1-phosphate receptor 3 (s1pr3) mRNA	NM_001102702.2	1.04	0.34	-1.63	1.02	0.88	-0.22	0.14			
Xenopus tropicalis finished DNA clone Tga12117	CJ075630.1	3.97	1.79	-1.16	8.72	7.8	-0.17	0.14			
Xenopus laevis glucosamine (N-acetyl)-6-sulfatase (gns) mRNA	NM_001086792.1	33.37	10.22	-1.71	46.76	39.54	-0.25	0.15			
PREDICTED: Xenopus (Silurana) tropicalis platelet-derived growth factor beta polypeptide (pdgb) transcript variant X1 mRNA	NM_002933750.2	1.23	0.48	-1.37	1.03	0.9	-0.21	0.15			
Xenopus tropicalis dickkopf WNT signaling pathway inhibitor 3 (dkk3) mRNA	CR71867.2	56.87	91.13	-2.65	1.68	1.27	-0.41	0.16			
Xenopus (Silurana) hypothetical protein MGCI185039 (MGCI185039) complete cds	BC161088.1	3.1	1.43	-1.12	4.24	3.75	-0.18	0.16			
Xenopus laevis YTH domain family member 1 (ythdf1) mRNA	NM_001090010.1	1.77	0.67	-1.43	16.04	13.7	-0.23	0.16			
PREDICTED: Xenopus (Silurana) tropicalis membrane-associated ring finger (C3HC4) 4 E3 ubiquitin-protein ligase (march4) mRNA	NM_002940050.2	43.78	11.47	-1.94	52.56	42.01	-0.33	0.17			
Xenopus laevis CCAAT/enhancer binding protein (CEBP) beta (cebpb-b) mRNA	27.38	13.21	-1.06	21.59	19.06	-0.19	0.18				
Xenopus tropicalis hypothetical protein MGCI68423 (MGCI68423) mRNA (DNA clone MGCI68423) complete cds	BC055957.1	4149.06	1553.18	-1.42	1222.4	1032.41	-0.25	0.18			
Xenopus laevis pancreatic lipase (plip) mRNA	NM_001097678.1	1.96	0.83	-1.26	104.45	89.71	-0.22	0.18			
PREDICTED: Xenopus (Silurana) tropicalis probable phospholipid-transporting ATPase 11c-like (LOC10498300) transcript variant X4 mRNA	NM_004916950.1	1.89	0.72	-1.41	1.54	1.29	-0.26	0.19			
Xenopus laevis repx44 mRNA complete sequence	DQ095861.1	66738.8	25094.2	-1.42	43685.14	36364.38	-0.27	0.19			
PREDICTED: Xenopus (Silurana) tropicalis 2-epi-5-epi-vallolone synthase-like (LOC100492806) transcript variant X1 mRNA	NM_002940475.2	39.94	17.14	-1.23	24.73	21.04	-0.24	0.2			

Gene	Accession	Six1 CHX <sup>1</sup>	Six1 CHX+DEX <sup>2</sup>	FC Six1 CHX	FC Six1 CHX+DEX <sup>3</sup>	Control CHX	Control CHX+DEX <sup>4</sup>	FC Control CHX+DEX <sup>5</sup>	Rank <sup>6</sup>
Xenopus laevis SYST1 Golgi-localized integral membrane protein homolog (sy61) mRNA	NM_001085953.1	2122.45	810.88	-1.4	48.47	40.25	-0.27	0.2	
Xenopus (Silurana) tropicalis SH3 and cysteine rich domain 3 (soc3) mRNA	NM_L001007506.1	263.57	40.01	-2.72	64.2	44.1	-0.55	0.2	
Xenopus laevis homeobox protein mRNA (cDNA clone MGC:5161842) complete cds	BC079970.1	2.56	1.09	-1.24	2.15	1.81	-0.25	0.2	
Xenopus (Silurana) tropicalis ataxin 2-like (atxnd2) mRNA	NM_001142213.1	552.18	175.49	-1.66	260.74	206.44	-0.34	0.21	
Xenopus laevis transporter associated with antigen processing 1 (TAP1) mRNA	AY204551.1	19.5	8.31	-1.24	30	24.93	-0.27	0.22	
Xenopus laevis uncharacterized LOC100031777 (LOC100031777) mRNA	NM_001097871.1	42.72	21.23	-1.01	21.21	18.16	-0.23	0.23	
XLUA9509 Xenopus laevis DNA polymerase gamma mRNA complete cds	U49509.1	136.67	64.15	-1.1	33.87	28.51	-0.25	0.23	
Xenopus laevis interferon regulatory factor 1 (irf1) mRNA	NM_001089781.1	34.94	16.56	-1.08	8.85	6.26	-0.26	0.24	
PREDICTED: Xenopus (Silurana) tropicalis IL-2-inducible T-cell kinase (itk) mRNA	NM_002939610.2	94.19	26.58	-1.83	30.34	22.29	-0.45	0.25	
PREDICTED: Xenopus (Silurana) tropicalis spectrin repeat containing nuclear envelope family member 3 (synex3) transcript variant X2 mRNA	NM_004917083.1	2.09	0.98	-1.1	1.67	1.38	-0.28	0.25	
XLU61839 Xenopus laevis thyroid hormone down-regulated protein (gene 18) mRNA complete cds	U41839.1	7.12	2.94	-1.28	13.65	10.9	-0.33	0.26	
Xenopus laevis XRIP1 mRNA for receptor-interacting protein 1 complete cds	AB206440.1	6.09	1.78	-1.78	2.15	1.56	-0.47	0.27	
Xenopus laevis nestin (nes) mRNA	NM_001087857.1	3144.75	985.77	-1.68	441.75	321.19	-0.46	0.28	
Xenopus laevis hypothetical protein LOC46267 mRNA (cDNA clone IMAGE:5542041) partial cds	BC079725.1	979.4	388.15	-1.34	275.84	212.6	-0.38	0.29	
Xenopus laevis chemokine (C-X-C motif) receptor 4 (cxcr4-a) mRNA	NM_001137581.1	29.38	12.77	-1.21	27.24	21.5	-0.35	0.29	
Xenopus laevis sphingomyelin phosphodiesterase 3 neutral membrane (neutral sphingomyelinase II) (smnpd3) mRNA	NM_001096194.1	9.37	4.55	-1.05	8.23	6.7	-0.3	0.29	
Xenopus tropicalis finished cDNA clone TTpaG77b029	CU075890.1	855.21	362.86	-1.24	18.21	14.22	-0.36	0.29	
Xenopus tropicalis finished cDNA clone TEG114K13	CT485687.1	1.4	0.45	-1.66	1.06	0.75	-0.5	0.31	
Xenopus laevis autoproteolytic 4D cysteine peptidase (ag4d) mRNA	NM_001093948.1	291.9	117.85	-1.31	33.28	25.08	-0.41	0.32	
PREDICTED: Xenopus (Silurana) tropicalis tensin X-like (LOC101733293) mRNA	NM_004914933.1	24.2	11.19	-1.12	33.06	25.92	-0.36	0.32	
Xenopus laevis hypothetical protein LOC442706 mRNA (cDNA clone IMAGE:6946515) partial cds	BC126044.1	136.14	40.78	-1.74	5.18	3.51	-0.57	0.33	
Xenopus tropicalis uncharacterized LOC495461 (LOC495461) mRNA	NM_001128621.1	662.67	163.44	-2.02	67.57	42.88	-0.66	0.33	
Xenopus tropicalis cDNA clone IMAGE:78066304	BC166285.1	479.98	196.49	-1.29	16.85	12.55	-0.43	0.33	
PREDICTED: Xenopus (Silurana) tropicalis mitogen-activated protein kinase kinase 4 (map3k4) transcript variant X1 mRNA	NM_002936262.2	51.42	23.34	-1.14	21.7	16.69	-0.38	0.34	
PREDICTED: Xenopus (Silurana) tropicalis me3 homolog A/C (me3a) partial mRNA	NM_002933115.2	32.63	12.57	-1.38	19.58	14.18	-0.47	0.34	
Xenopus laevis suppressor of cytokine signaling 1 (soc1-sa) mRNA	NM_001092026.1	7.28	2.62	-1.48	8.91	6.24	-0.52	0.35	
Xenopus laevis cDNA clone NMGC98823 IMAGE:3401284 complete cds	BC084658.1	12149.48	5603.31	-1.12	13751.58	10505.88	-0.39	0.35	
Xenopus laevis hypothetical protein LOC398632 mRNA (cDNA clone IMAGE:4965716) partial cds	BC056077.1	1.54	0.61	-1.35	1.2	0.86	-0.49	0.37	
Xenopus tropicalis finished cDNA clone TGas131n17	CR762128.2	38.5	17.58	-1.14	31.48	23.36	-0.44	0.38	
PREDICTED: Xenopus (Silurana) tropicalis acyl-CoA dehydrogenase family member 11 (acad11) transcript variant 1 mRNA	NM_00293772.7	1.49	0.66	-1.18	2.94	2.13	-0.47	0.4	
Xenopus (Silurana) tropicalis G protein-coupled receptor 83 (gpr83) mRNA	NM_001079037.1	175.22	67.05	-1.39	96.96	66.06	-0.56	0.4	
PREDICTED: Xenopus (Silurana) tropicalis zinc finger CCCH domain-containing protein 3-like (LOC100495750) mRNA	NM_004917436.1	91.33	42.66	-1.1	29.17	21.45	-0.45	0.41	
Xenopus tropicalis finished cDNA clone Theud30109	CR942608.2	1.21	0.71	-1.37	12.08	8.17	-0.57	0.42	
Xenopus (Silurana) tropicalis cytokine inducible SH2-containing protein (cish) mRNA	NM_00113689.1	51.36	21.87	-1.24	63.48	43.84	-0.54	0.44	
Xenopus laevis suppressor of cytokine signaling 3 (soc3) mRNA	NM_00112112.1	228.53	70.49	-1.7	252.68	150.06	-0.76	0.45	
Xenopus laevis mRNA for eph receptor tyrosine kinase (epn84b) partial	AJ236868.1	8.21	3.36	-1.3	8.78	5.87	-0.59	0.46	
Xenopus tropicalis finished cDNA clone TGas50hn04	CR761765.2	29.64	13.07	-1.19	14.38	9.74	-0.57	0.48	
PREDICTED: Xenopus (Silurana) tropicalis Ig-motif and Sec7 domain 2 (qsec2) transcript variant X3 mRNA	XM_004916746.1	14.57	6.31	-1.21	6.71	4.5	-0.58	0.48	
Xenopus tropicalis finished cDNA clone TEG086m16	CT025204.2	1.45	0.47	-1.62	1.71	1	-0.78	0.49	
Xenopus (Silurana) tropicalis hepsin (hepn) mRNA	NM_001126815.1	1.86	0.87	-1.11	2.84	1.91	-0.58	0.52	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730751 mRNA	NM_004917395.1	2.8	1.2	-1.23	2.97	1.9	-0.65	0.54	
Xenopus laevis uncharacterized LOC100462907 (LOC100462907) mRNA	NM_001190395.1	8.76	4.2	-1.07	14.47	9.72	-0.58	0.54	

<sup>1</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1 and CHX treatment

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1 and CHX+DEX treatment

<sup>3</sup> Log<sub>2</sub> Fold change values for Six1

<sup>4</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup> Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup> Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Six1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

### E.3 Eya1; Up-regulated genes

**Table E.3** Genes with at least two-fold up-regulation after injection of Eya1-GR and treatment with CHX + DEX

Gene	Accession	Eya1 CHX <sup>a</sup>	Eya1 CHX+DEX <sup>b</sup>	FC Eya1 <sup>c</sup>	FC Control CHX <sup>a</sup>	FC Control CHX+DEX <sup>b</sup>	Rank <sup>d</sup>
Xlaevis mRNA for P2Y8 nucleotide receptor	X99533.1	0.01	4.03	11.98	0	0	0
Xenopus laevis homeobox D11 (hoxd11) mRNA	NM_001096497.1	0.01	1.34	10.38	0	0	0
Xenopus laevis cDNA clone IMAGE:6950606	BC0933562.1	0.01	1.06	10.04	0	0	0
Xenopus laevis mRNA for XlHbox1 protein (Prl type)	X12499.1	0.03	2.54	6.91	0	0	0
Xenopus laevis homeobox C10 (hoxc10) mRNA	NM_001090479.1	0.03	2.27	6.31	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox A11 (hoxa11) mRNA	XM_004915395.1	0.04	2.14	5.99	0	0	0
Xenopus laevis homeobox A9 (hoxa9) mRNA	NM_001097795.1	0.05	2.23	5.5	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7022272	BC094950.1	0.07	2.83	5.43	0	0	0
Xenopus laevis cDNA clone IMAGE:7017143	BC094957.1	0.11	3.47	5.1	0	0	0
Xenopus laevis cDNA clone MGCI:160241 (IMAGE:8526708 complete cds	BC129574.1	0.04	1.05	4.98	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox A10 (hoxa10) mRNA	XM_002933394.2	0.19	4.73	4.71	0	0	0
Xenopus (Silurana) tropicalis adduct A fructose-bisphosphate aldolase mRNA	NM_001095643.1	0.06	1.19	4.41	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox protein Hox-C11a-like (LOC100487406) mRNA	XM_002936646.2	0.37	7.49	4.37	0	0	0
Xenopus laevis hypothetical protein LOC10049089 mRNA (cDNA clone MGCI:115644 (IMAGE:3749571) complete cds	BC090046.1	7.47	153.26	4.36	30.94	9.11	-1.77
PREDICTED: Xenopus (Silurana) tropicalis polyphosphate 5'-phosphatase-like (LOC101734556) mRNA	XM_004916572.1	0.08	1.36	4.16	0	0	0
Xenopus laevis Hoxc10 mRNA complete cds	AY16741.1	0.15	2.58	4.13	0	0	0
Xenopus laevis homeobox D9 (hoxd9) mRNA	NM_001740431.1	0.06	1.03	4.12	0	0	0
Xenopus (Silurana) tropicalis cytochrome P450 family 26 subfamily B polypeptide 1 (cytP450B1) mRNA	NM_001079187.2	0.24	3.81	4	3.19	1.81	-0.82
PREDICTED: Xenopus (Silurana) tropicalis c-X-C motif chemokine 10-like (LOC100490785) mRNA	XM_002940578.2	0.28	4.19	3.93	0	0	0
Xenopus laevis uncharacterized LOC496300 (LOC496300) mRNA	NM_001095458.1	7.47	153.26	4.36	30.94	9.11	-1.77
PREDICTED: Xenopus (Silurana) tropicalis polyphosphate 5'-phosphatase-like (LOC101734556) mRNA	XM_004916572.1	0.08	1.36	4.16	0	0	0
Xenopus laevis Hoxc10 mRNA complete cds	AB012615.1	0.15	2.58	4.13	0	0	0
Xenopus laevis homeobox D9 (hoxd9) mRNA	NM_001086050.1	0.11	1.44	3.81	0	0	0
Xenopus (Silurana) tropicalis creatine kinase mitochondrial 1B (ckmt1b-b) nuclear gene encoding mitochondrial protein mRNA	NM_001091933.1	0.12	1.62	3.77	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis Hox-C5-like (LOC101733557) mRNA	XM_002935582.2	0.43	5.19	3.62	0	0	0
Xenopus laevis uncharacterized LOC10036338 (LOC10036338) mRNA	NM_001097708.1	0.16	1.92	3.59	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tyrosine kinase-binding protein-like (LOC100493958) mRNA	XM_0029406242	0.1	1.34	3.85	0	0	0
Xenopus laevis mRNA for XI GF-20 complete cds	AB012615.1	0.11	1.44	3.81	0	0	0
Xenopus (Silurana) tropicalis c-X-C motif chemokine 10-like (LOC100490785) mRNA	NM_001086050.1	0.12	1.62	3.77	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tachykinin-like peptide (MGCI:32043) mRNA	XM_002935582.2	0.43	5.19	3.62	0	0	0
Xenopus laevis uncharacterized LOC10036338 (LOC10036338) mRNA	NM_001097708.1	0.16	1.92	3.59	0	0	0
Xenopus laevis carbonic anhydrase IV (ca4) mRNA	NM_001086319.1	94.76	109.868	3.54	145.35	118.44	-0.3
Xenopus (Silurana) tropicalis 5'X homeobox 2 (si52) mRNA	NM_001100275.1	0.61	6.75	3.49	0	0	0
Xenopus laevis homeobox D8 (hoxd8) mRNA	NM_001089872.1	0.14	1.48	3.42	0	0	0
Xenopus tropicalis cDNA clone IMAGE:796027 partial cds	BC122054.1	0.78	8.11	3.4	0	0	0
Xenopus laevis uncharacterized LOC495161 (LOC495161) mRNA	NM_001094854.1	0.14	1.19	3.19	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tachykinin-like peptide (MGCI:32043) mRNA	XM_004916890.1	0.15	2.03	3.37	0	0	0
Xenopus laevis gamma-aminobutyric acid receptor subunit rho-3 (Gabr3) mRNA partial cds	HQ437681.1	0.11	1.09	3.33	0	0	0
Xenopus laevis XGrp2 mRNA for glutamate receptor subunit rho-2 complete cds	AB290863.1	0.11	1.08	3.32	0	0	0
Xenopus tropicalis hypothetical protein MGCI:146277 mRNA (cDNA clone MGCI:146277 (IMAGE:7690007) complete cds	BC123945.1	0.17	1.52	3.22	0	0	0
Xenopus laevis uncharacterized LOC495161 (LOC495161) mRNA	NM_001030412.1	0.19	1.49	3.05	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis protocadherin-1 X-linked-like (LOC100493938) transcript variant X2 mRNA	XM_002934245.2	0.68	5.48	3.02	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis paired box 1 (pax1) transcript variant X1 mRNA	XM_002934245.2	0.14	1.11	3	0	0	0
Xenopus tropicalis finished cDNA clone Ga3313e15	C705282.2	0.15	1.15	2.99	0	0	0
Xenopus laevis hypothetical protein LOC446860 mRNA (cDNA clone MGCI:7203732) partial cds	BC082220.1	0.19	1.46	2.99	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis extracellular calcium-sensing receptor-like (LOC101734639) partial mRNA	XM_004919948.1	3.62	26.66	2.89	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tryosin-3-like (LOC10173092) mRNA	NM_004914194.1	0.18	1.32	2.88	0	0	0

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX	FC Eva1 <sup>a</sup>	Control CHX	Control CHX+DEX	FC Control CHX	Control CHX+DEX	FC Control CHX	Rank <sup>b</sup>
Xenopus (Silurana) tropicalis leucine rich repeat containing 8 family member D (lrrc8d) mRNA	NM_001126681.1	0.18	1.28	2.86	0	0	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7758182	BG154876.1	0.22	1.51	2.83	0	0	0	0	0	0
Xenopus laevis branched chain amino-acid transaminase 2 mitochondrial (lbcat2) nuclear gene encoding mitochondrial protein mRNA	NM_001112883.1	0.16	1.11	2.83	0	0	0	0	0	0
Xenopus (Silurana) tropicalis chemokine (C-X-C motif) receptor 7 (cxcr7) mRNA	NM_001030434.1	0.15	1.03	2.8	0	0	0	0	0	0
Xenopus laevis CD93 molecule (cd93) mRNA	NM_001095151.1	0.25	1.69	2.76	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox D13 (hoxd13) mRNA	NM_002935677.2	0.27	1.76	2.76	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox D3 (hoxd3) transcript variant X3 mRNA	NM_004917730.1	0.29	1.86	2.72	0	0	0	0	0	0
Xenopus laevis homeobox B7 (hoxb7-a) mRNA	NM_001127798.1	0.93	6.05	2.71	0	0	0	0	0	0
Xenopus laevis homeobox B8 (hoxb7-a) mRNA	NM_001085641.1	0.28	1.78	2.69	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone (Ga0003a01)	CR942341.2	0.72	4.55	2.68	0	0	0	0	0	0
Xenopus (Silurana) tropicalis sushi domain containing 1 (susd1) mRNA	NM_203891.1	0.23	1.41	2.66	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis espin (espn) transcript variant X1 mRNA	NM_002933856.2	0.25	1.57	2.66	1.8	1.75	-0.05	0	0	0
Xenopus laevis hedgehog-interacting protein mRNA complete cds	AY328923.1	0.35	2.12	2.64	0	0	0	0	0	0
Xenopus laevis oviduct protein p20 mRNA complete cds	AY563621.1	0.31	1.9	2.64	0	0	0	0	0	0
Xenopus laevis arginyl aminopeptidase (aminopeptidase B) b (mpepb) mRNA	NM_001092079.1	0.3	1.85	2.64	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis poly (ADP-ribose) polymerase 4-like (LOC100491505) mRNA	NM_002937967.2	0.68	4.2	2.63	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis estradiol 17-beta-dehydrogenase 1-like (LOC100489679) mRNA	NM_002935902.2	0.18	1.06	2.61	0	0	0	0	0	0
Xenopus laevis SRY-box containing protein (Sox1) mRNA complete cds	EF67227.1	0.18	1.08	2.6	0	0	0	0	0	0
Xenopus laevis mRNA for olfactory receptor class II (xb177 gene)	AJ250754.1	109.2	656.03	2.59	120.19	32.5	-1.89	0	0	0
Xenopus laevis pyridine nucleotide-disulfophosphate oxidoreductase domain 2 (pyroxd2) mRNA	NM_001095120.1	0.94	5.54	2.57	0	0	0	0	0	0
Xenopus (Silurana) tropicalis RNA895 protein (kiaa0895) mRNA	NM_001114073.1	0.32	1.86	2.56	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical calcium/calmodulin-dependent protein kinase kinase 2 beta (camkk2) transcript variant X1 mRNA	NM_002937701.2	0.24	1.38	2.56	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone (Tegg026p17)	CR761997.2	0.35	2.01	2.55	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical integrin alpha 8 (itga8) mRNA	NM_002939028.2	0.21	1.19	2.55	0	0	0	0	0	0
Xenopus laevis mRNA for Xoxx17-alpha protein	AJ001730.1	1.65	9.56	2.54	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical myosin heavy chain 7 cardiac muscle beta (myh7) mRNA	NM_002939015.2	0.19	1.06	2.53	0	0	0	0	0	0
Xenopus (Silurana) tropicalis solute carrier family 16 member 7 (monocarboxylic acid transporter 2) (slc16a7) mRNA	NM_001112921.1	0.19	1.03	2.51	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone (TeggA03p05)	CU075479.1	0.83	4.68	2.5	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical homeobox B8 (hoxb8) transcript variant X1 mRNA	NM_002938021.2	0.71	3.97	2.49	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical polycystic kidney and hepatic disease 1 (autosomal recessive)-like 1 (pkhd1l1) mRNA	NM_002942314.2	0.24	1.34	2.49	0	0	0	0	0	0
Xenopus laevis family with sequence similarity 46 member A (fam46a) mRNA	NM_001127822.1	0.89	4.86	2.47	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:807580.1	BC123196.1	0.77	4.2	2.46	0	0	0	0	0	0
Xenopus laevis hypothetical LOC495273 mRNA (cDNA clone IMAGE:7018338) partial cds	BC084403.1	0.47	2.57	2.46	0	0	0	0	0	0
XLAFO1596 Xenopus laevis homeobox protein pXhox8 (khox8) mRNA complete cds	AF001596.1	0.85	4.62	2.46	0	0	0	0	0	0
Xenopus laevis ectodysplasin A receptor (edan) mRNA	NM_001087047.1	0.83	4.5	2.46	0	0	0	0	0	0
Xenopus (Silurana) tropical methionine sulfoxide reductase A gene 2 (msra2) mRNA	NM_001114258.1	0.19	1.04	2.45	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TNel143f19	CR760056.2	0.59	3.12	2.41	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone (Ga0371e17)	BC263377.2	0.31	1.62	2.41	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical dimethylamine monooxygenase [N-oxide-forming] 2-like (LOC100489107) mRNA	NM_00293744.2	0.35	1.82	2.4	0	0	0	0	0	0
Xenopus laevis Hoxd13 protein mRNA (cDNA clone MGCS36545 IMAGET3399571) complete cds	BC080334.1	0.75	3.95	2.4	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical mitogen-activated protein kinase 3 (mapkap3) mRNA	NM_002939594.2	18.79	97.31	2.38	1.68	1.54	-0.13	0	0	0
Xenopus (Silurana) tropicalis mitogen-activated protein kinase 3 (mapkap3) mRNA	NM_001079394.1	1.13	5.83	2.37	0	0	0	0	0	0
Xenopus laevis orga04 mRNA complete sequence	DQ096925.1	3422.97	17656.8	335.36	183.58	-0.87	0	0	0	0
Xenopus (Silurana) tropical pre-mRNA processing factor 31 (ppf31) mRNA	NM_213694.1	0.34	1.73	2.36	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical embryonic protein LUV5.2-like (LOC100488711) mRNA	NM_00491998.1	0.6	2.99	2.34	0	0	0	0	0	0
Xenopus laevis ectodysplasin A receptor (edan) mRNA	NM_00491038.1	1.24	6.08	2.31	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical mitogen-activated protein kinase 3 (mapkap3) mRNA	NM_004911935.1	1.62	7.93	2.3	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical homeobox C6 (hoxc6) transcript variant X7 mRNA	NM_002936227.2	0.27	1.27	2.29	0	0	0	0	0	0
Xenopus (Silurana) tropical transcript variant X2 mRNA	NM_203994.1	0.29	1.38	2.29	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropical contactin-2-like (LOC100487049) mRNA	BC124896.1	141.25	67.28	2.26	193.25	125.6	-0.63	0	0	0
Xenopus laevis cDNA clone MGCS15451 IMAGET3320906 complete cds	AF072455.1	1.18	5.53	2.23	0	0	0	0	0	0

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX	FC Eva1 CHX	FC Control CHX	FC Control CHX+DEX	FC Control CHX	Rank*
PREDICTED: Xenopus (Silurana) tropicalis beta-1,3-galactosyltransferase 2-like (LOC101732799) transcript variant X1 mRNA	XM_004918863.1	0.34	1.55	2.23	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis multidrug and toxin extrusion protein 2-like (LOC100489463) mRNA	XM_002933835.2	0.62	2.87	2.23	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis poly (ADP-ribose) polymerase family member 4 (parp4) mRNA	XM_002937966.2	0.27	1.25	2.22	0	0	0	0
Xenopus laevis cat eye syndrome chromosomal region candidate 5 (cecr5) mRNA	NM_001097147.1	2.81	12.91	2.21	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine kinase 32A (sk32a) mRNA	XM_002936707.2	0.23	1.03	2.2	0	0	0	0
Xenopus laevis fin and gill-specific type-1 keratin (fgk) mRNA	NM_001246316.1	0.24	1.08	2.18	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis alpha-1 kinase 2 (alk2) mRNA	NM_004910401.1	0.46	2.04	2.17	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis mucin 2-like (LOC100494822) mRNA	XM_002938567.2	0.38	1.69	2.16	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734503 (LOC101734503) mRNA	XM_004919251.1	0.29	1.28	2.15	0	0	0	0
Xenopus laevis SRY (sex determining region Y-box 17 beta gene 2 (sox17b.2) mRNA	NM_001098715.1	0.39	1.72	2.15	0	0	0	0
Xenopus laevis nephrin (NPHS1) mRNA complete cds	AY902238.1	0.51	2.21	2.14	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis poly (ADP-ribose) polymerase 14-like (LOC100485144) mRNA	XM_002943546.2	0.52	2.25	2.13	0	0	0	0
Xenopus laevis hypothetical protein LOC414703 mRNA (cDNA clone IMAGE663551)2 partial cds	BG068953.1	1.94	8.45	2.13	7.99	5.93	-0.43	0
Xenopus laevis uncharacterized LOC495373 (LOC495373) mRNA	NM_001095035.1	0.24	1.03	2.12	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC495373 (LOC495373) mRNA	XM_002939392.2	0.29	1.25	2.12	0	0	0	0
Xenopus laevis SRY (sex determining region Y-box 17 beta gene 2 (sox17b.2) mRNA	NM_001098715.2	0.72	3.05	2.1	0	0	0	0
Xenopus laevis nephrin (NPHS1) mRNA complete cds	DQ096885.1	0.91	3.88	2.1	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis leucine rich repeat containing 15 (lrrc15) mRNA	NM_004916193.1	1.13	4.77	2.09	0	0	0	0
Xenopus laevis ceg121 mRNA complete sequence	NM_001256315.1	1.04	4.4	2.09	3.02	2.57	-0.24	0
PREDICTED: Xenopus (Silurana) tropicalis espin (espn) transcript variant X3 mRNA	XM_00534121	0.56	2.32	2.06	0	0	0	0
Xenopus tropicalis (XCT1) mRNA complete cds	NM_002938386.2	1.6	6.61	2.06	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis deoxyribonucleic acid gamma-like (LOC10049175) transcript variant X1 mRNA	XM_002933384.2	0.34	1.39	2.05	0	0	0	0
Xenopus laevis elongation factor 1 delta mRNA (cDNA clone MGCI80004 IMAGE6317228) complete cds	BC072139.1	35.84	146.44	2.04	35.53	32.98	-0.11	0
Xenopus laevis RDC1 like protein mRNA (cDNA clone MGCI114801 IMAGE6332706) complete cds	BG098974.1	0.43	1.74	2.04	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C2orf80 (LOC100489057) mRNA	XM_0029371192	0.91	3.66	2.03	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transcription factor EC (tfec) transcript variant X1 mRNA	XM_002934967.2	0.49	1.96	2.02	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis elongation factor 1 delta mRNA (cDNA clone MGCI80004 IMAGE6317228) complete cds	XM_002937441.2	0.42	1.68	2.02	0	0	0	0
Xenopus laevis cDNA clone IMAGE5440130 mRNA	XM_002935272.2	14.88	59.93	2.02	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis histone acetyltransferase 2 (fecf2) transcript variant X1 mRNA	BC094441.1	0.57	2.23	1.98	1.44	1.42	-0.03	0
PREDICTED: Xenopus (Silurana) tropicalis histone acetyltransferase 2 (fecf2) transcript variant X2 mRNA	XM_00293761.2	1	3.9	1.98	3.69	3.65	-0.02	0
PREDICTED: Xenopus (Silurana) tropicalis histone acetyltransferase 2 (fecf2) transcript variant X3 mRNA	XM_002938846.2	42.84	167.35	1.97	26.37	15.92	-0.73	0
Xenopus laevis calbindin D28k mRNA (cDNA clone MGCI197288 IMAGE:9093927) complete cds	BC170541.1	2.45	9.4	1.95	4.23	3.75	-0.18	0
Xenopus laevis pyruvate dehydrogenase phosphatase catalytic subunit 1 (pdph1) nuclear gene encoding mitochondrial protein mRNA	NM_001094221.1	95.14	360.44	1.93	48.37	41.62	-0.22	0
PREDICTED: Xenopus (Silurana) tropicalis histone acetyltransferase 2 (fecf2) transcript variant X1 mRNA	XM_002932255.2	1.02	3.72	1.87	11.58	6.57	-0.82	0
Xenopus laevis MYST histone acetyltransferase 2 (myst2) mRNA	NM_001096764.1	241.6	834.22	1.79	9.54	7.71	-0.31	0
Xenopus laevis cyclin-dependent kinase 14 (cdk14) mRNA	NM_001092883.1	1.34	4.6	1.78	1.89	1.69	-0.17	0
Xenopus laevis transmembrane emp24 transport domain containing 4 (tm4sf4) mRNA	NM_001092877.1	1106.73	3701.63	1.75	37.01	35.33	-0.07	0
Xenopus laevis ribosomal protein S2e mRNA (cDNA clone MGCI160651 IMAGE:63181725) complete cds	BC130122.1	0.52	1.71	1.74	5.59	4.71	-0.25	0
active beta 1 subunit (Xenopus laevis mRNA 2716 nt)	BC170087.1	3.55	11.63	1.72	4.04	3.42	-0.25	0
Xenopus (Silurana) tropicalis 5T6 beta-galactosidase mRNA (cDNA clone MGCI96814 IMAGE:9041722) complete cds	NM_0011114199.1	7.98	24.22	1.61	5.11	4.12	-0.32	0
PREDICTED: Xenopus (Silurana) tropicalis caspase-3 (casp3) mRNA	XM_004915237.1	680.5	2218.36	1.71	12.28	6.73	-0.87	0
PREDICTED: Xenopus (Silurana) tropicalis cyclin-dependent kinase 14 (cdk14) mRNA	XM_002942837.2	0.55	1.78	1.7	3.09	1.91	-0.7	0
Xenopus laevis interferon regulatory factor 9 (irf9) mRNA	NM_001091377.1	824.57	2617.25	1.67	4.67	3.88	-0.27	0
PREDICTED: Xenopus (Silurana) tropicalis NPF family member 1-like (LOC101731316) partial mRNA	XM_004920026.1	2.73	8.49	1.64	10.68	10.45	-0.04	0
activation beta 1 subunit (Xenopus laevis mRNA 2716 nt)	S61773.1	0.97	2.98	1.63	1.85	1.35	-0.46	0
Xenopus (Silurana) tropicalis 5T6 beta-galactosidase mRNA (cDNA clone MGCI96814 IMAGE:9041722) complete cds	NM_0011114199.1	7.98	24.22	1.61	5.11	4.12	-0.32	0
PREDICTED: Xenopus (Silurana) tropicalis caspase-3 (casp3) mRNA	XM_001011068.1	1.71	5.03	1.57	2.79	2.75	-0.79	0
XL062288 Xenopus laevis Wnt-2b (Xwnt-2b) mRNA complete cds	U62288.1	1.84	5.33	1.54	2.35	1.32	-0.84	0
Xenopus (Silurana) tropicalis family with sequence similarity 78 member A (fam78a) mRNA	NM_001127018.1	0.98	2.79	1.52	2.01	1.87	-0.11	0
Xenopus (Silurana) tropicalis hypothetical LOC492883 mRNA (cDNA clone IMAGE:66863036) partial cds	BC108836.1	0.47	1.33	1.52	4.87	1.78	-1.45	0
Xenopus laevis homeobox C6 (hoxc6) mRNA	NM_001087546.1	1.72	4.83	1.5	1.63	1.19	-0.46	0
PREDICTED: Xenopus (Silurana) tropicalis transcript variant X1 mRNA	NM_004915536.1	2.94	8.25	1.49	1.9	1.37	-0.48	0

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX	FC Eva1 <sup>a</sup>	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Rank <sup>b</sup>
Xenopus (Silurana) tropicalis methylsterol monooxygenase 1 (msmo1) mRNA	NM_0010179341.1	0.96	2.62	1.47	1.86	1.81	-0.05	0
Xenopus laevis cDNA clone IMAGE:6886279	BC099021.1	0.47	1.25	1.44	3.79	2.17	-0.81	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox C12 (hoxc12) mRNA	XM_002936647.2	4.88	12.98	1.42	4.64	3.59	-0.38	0
PREDICTED: Xenopus (Silurana) tropicalis cAMP uncharacterized transcript variant X1 mRNA	XM_002935162.2	6.6	17.11	1.38	9.03	7.83	-0.21	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100498064 (LOC100498064) mRNA	XM_004915062.1	137.92	357.43	1.38	24.18	18.77	-0.37	0
Xenopus (Silurana) tropicalis uncharacterized LOC100498064 (LOC100498064) mRNA	NM_001126985.1	0.64	1.66	1.38	2.59	1.83	-0.51	0
Xenopus laevis mRNA similar to envelopin (cDNA clone IMAGE:5571064)	XN_002942992.2	0.7	1.75	1.34	713849.63	67869.31	-3.4	0
PREDICTED: Xenopus (Silurana) tropicalis tripartite motif-containing protein 7-like (LOC100495288) mRNA	XM_002938821.2	1.42	3.56	1.33	1.32	1.23	-0.81	0
PREDICTED: Xenopus (Silurana) tropicalis one cut domain family member 2-like (LOC100491290) mRNA	XM_00110257.1	2.07	5.19	1.33	1.24	1.04	-0.25	0
Xenopus (Silurana) tropicalis distal-less homeobox 1 (dlx1) mRNA	NM_0010197001.1	96.79	241.7	1.33	28.84	26.74	-0.11	0
Xenopus laevis membrane bound O-acetyltransferase domain containing 1 (mbat1) mRNA	CT030471.1	1.55	3.85	1.32	4.99	4.68	-0.1	0
Xenopus laevis mRNA for ZIC-related 2 complete cds	AB014461.1	2.01	4.9	1.29	3.44	3.27	-0.08	0
Xenopus laevis XI-c-ets-2b protein mRNA (cDNA clone MGCS8006 IMAGE:6631306) complete cds	BC077264.1	19.72	48	1.29	21.13	18.01	-0.24	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100490339 (LOC100490339) mRNA	XM_002942884.2	1.89	4.59	1.29	1.76	1.13	-0.65	0
Xenopus laevis putative transmembrane protein TA-2 mRNA complete cds	AF310008.1	677.97	1637.22	1.28	1548.14	1093.89	-0.51	0
Xenopus laevis finished cDNA clone Tgas056c05	NM_0010192691.1	3.44	8.23	1.27	4.24	4.22	-0.01	0
Xenopus laevis mRNA for ZIC-related 2 complete cds	NM_0010193055.1	30.36	72.75	1.27	12.68	11.68	-0.12	0
Xenopus laevis XI-c-ets-2b protein mRNA (cDNA clone MGCS8006 IMAGE:6631306) complete cds	BC099044.1	1.82	4.27	1.24	1.92	1.37	-0.5	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100490339 (LOC100490339) mRNA	GU775438.1	0.79	1.83	1.23	1.34	1.22	-0.15	0
Xenopus laevis runt-related transcription factor 3 (Runx3) mRNA complete cds	NM_001172167.1	4.08	9.48	1.22	7.77	4.13	-0.92	0
Xenopus laevis CCAAT/enhancer binding protein (CEBP) beta (cebpb-al) mRNA	DQ096879.1	1193.82	2739.84	1.2	1245.62	1237.76	-0.01	0
Xenopus laevis mito mRNA complete sequence	XN_002932117.2	1.64	3.75	1.2	1.38	1.36	-0.02	0
PREDICTED: Xenopus (Silurana) tropicalis Mov10 Moloney leukemia virus 10 homolog (mouse) (mov10) mRNA	AF168465.1	2.95	6.73	1.2	4.43	2.8	-0.67	0
Xenopus laevis gamma-glutamyl hydrolase (conjugase folylpolygamma glutamyl hydrolase) (gg) mRNA	NM_002943522.2	0.76	1.72	1.19	8.84	8.87	-0.01	0
Xenopus laevis glyoxalase 1 (glo1-b) mRNA	NM_001129915.1	30.51	69.02	1.18	13.42	10.87	-0.31	0
Xenopus laevis cDNA clone IMAGE:3747628	XM_002932622.2	0.94	2.05	1.14	1.33	1.23	-0.12	0
Xenopus laevis runt-related transcription factor 3 (Runx3) mRNA complete cds	AB197251.1	1.8	3.94	1.14	5.17	4.75	-0.13	0
Xenopus laevis mito mRNA complete sequence	NM_0010192381.1	6.93	15.56	1.17	8.6	8.47	-0.03	0
PREDICTED: Xenopus (Silurana) tropicalis Mov10 Moloney leukemia virus 10 homolog (mouse) (mov10) mRNA	BC043732.1	0.86	1.91	1.17	1.6	1.37	-0.24	0
Xenopus laevis hyaluronan synthase 2 (HAS2) mRNA complete cds	NM_002934799.1	0.92	2.02	1.15	1.91	1.25	-0.62	0
PREDICTED: Xenopus (Silurana) partial mRNA (sand9) protein (sand9) partial mRNA	AB458410.1	221.25	480.68	1.12	1013.22	580.92	-0.81	0
PREDICTED: Xenopus (Silurana) nuclear receptor interacting protein 1 (nrip1) mRNA	NM_0010192473.1	1.16	2.52	1.12	1.96	1.9	-0.05	0
Xenopus laevis Kruppel-like factor 2 (lung) mRNA (cDNA clone MGCS52512 IMAGE:5570193) complete cds	AB197251.1	5.86	12.79	1.13	1.78	1.15	-0.64	0
PREDICTED: Xenopus (Silurana) Kruppel-like factor 2 (lung) mRNA up regulated post tail amputation-2 complete sequence	NM_0010192381.1	1.72	3.73	1.13	4.25	2.42	-0.82	0
Xenopus laevis non-coding RNA up regulated post tail amputation-2 complete sequence	AF030344.1	4.12	8.97	1.13	8.9	6.35	-0.49	0
PREDICTED: Xenopus (Silurana) tropicops opsin-3-like (LOC100488877) mRNA	AB458410.1	221.25	480.68	1.12	1013.22	580.92	-0.81	0
Xenopus laevis mRNA for xMAP complete cds	NM_0010195566.1	21.17	45.72	1.12	29.62	24.66	-0.27	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein adipocyte associated 1 (tpa1) mRNA	BC110723.1	54.54	117.44	1.11	84.18	78.18	-0.1	0
Xenopus laevis Dickkopf-1 (Xdkk-1) mRNA complete cds	NM_001130334.1	55.56	119.37	1.11	75.33	38.66	-0.97	0
Xenopus laevis non-coding RNA up regulated post tail amputation-2 complete sequence	AB458410.1	0.9	1.93	1.11	1.09	1.05	-0.06	0
Xenopus laevis saccharopin dehydrogenase (putative) (scpdh) mRNA	NM_0010192473.1	2.38	5.1	1.11	2.24	1.36	-0.72	0
Xenopus laevis ras homolog family member V (rhov) mRNA	NM_0010195566.1	0.81	1.73	1.11	21.10	1.44	-0.31	0
PREDICTED: Xenopus (Silurana) tropicalis X-box binding protein 1 mRNA (cdna clone MGCI30816 IMAGE:7975357) complete cds	BC110723.1	98.98	211.07	1.1	29.16	14.76	-0.99	0
Xenopus (Silurana) tropicalis transmembrane protein epitelial specific (elf3) mRNA	NM_0010198435.2	3.63	7.64	1.08	7.16	6.14	-0.23	0
PREDICTED: Xenopus (Silurana) tropicalis transcription factor GitnU-like (LOC100496329) mRNA	XM_002934032.2	1.65	3.45	1.07	1.83	1.2	-0.61	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730819 (LOC101730819) mRNA	XM_0010198697.1	1.18	2.45	1.07	2.81	2.39	-0.23	0
PREDICTED: Xenopus (Silurana) tropicalis transcription factor epitelial specific (elf3) mRNA	NM_0010193439.1	2.18	4.53	1.06	3.16	2.24	-0.15	0
Xenopus (Silurana) tropicalis Kruppel-like factor 16 (Xdkk-16) mRNA	BC157491.1	109.39	227.03	1.06	172.6	80.9	-1.1	0
Xenopus laevis zinc finger protein 706 gene 2 (znt706-2-b) mRNA	X58487.1	14539.42	30040.73	1.05	4351.26	2955.11	-0.56	0
Xlaevis posterior (Xpo) gene	NM_0010192619.1	1.27	2.62	1.05	4.43	3.09	-0.53	0

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX	FC Eva1 CHX	FC Control CHX	FC Control CHX+DEX	Rank <sup>a</sup>
PREDICTED: Xenopus (Silurana) tropicalis interleukin-8-like (LOC100498389) mRNA	XM_002942531.1	15.33	31.55	1.05	22.66	14.96	-0.6
XELHX03: Xenopus laevis mRNA for Xhox3 protein complete cds	D 0455.1	2.2	4.46	1.03	2.77	1.65	-0.75
PREDICTED: Xenopus (Silurana) tropicalis cAMP responsive element modulator (crem) transcript variant X3 mRNA	XM_004915473.1	4.38	8.9	1.03	5.98	5.2	-0.21
Xenopus laevis cDNA clone 1/G2	AL009293.1	9.26	187.96	1.03	177.16	143.34	-0.31
Xenopus laevis BMI1 polycomb ring finger oncogene (bm1-1b) mRNA	NM_001094538.1	5.6	11.34	1.02	14.39	10.38	-0.48
Xenopus (Silurana) tropicalis homeobox A5 (xoxa5) mRNA	NM_001014051.1	3.72	7.52	1.02	1.39	1.25	-0.15
Xenopus (Silurana) tropicalis suppressor of cytokine signaling 2 (soc2) mRNA	NM_001127426.1	2.77	5.6	1.02	5.22	2.32	-1.18
Xenopus laevis prepro-PcLa mRNA (cDNA clone MGCI_97244 IMAGE:0093902) complete cds	BG170517.1	3.36	6.78	1.02	2.22	1.52	-0.55
Xenopus laevis hyaluronan synthase related sequence protein (has-s) mRNA	NM_001088359.1	1.58	3.17	1.02	1.9	1.54	-0.31
Xenopus (Silurana) tropicalis uncharacterized LOC100158544 (LOC100158544) mRNA	NM_001127978.2	3.37	6.79	1.02	3.01	2.79	-0.12
Xenopus laevis eukaryotic translation initiation factor 2B subunit 2 beta 39kDa (elf2b2) mRNA	NM_001086799.1	1	2.01	1.01	2.02	1.97	-0.04
PREDICTED: Xenopus (Silurana) tropicalis ALR tyrosine kinase receptor-like (LOC100495071) mRNA	XM_002938422.2	2.01	4.04	1.01	2.53	2.3	-0.14
Xenopus laevis transmembrane protein 56 (tmem56-b) mRNA	NM_001086647.1	58.93	118.12	1.01	32.63	29	-0.17
PREDICTED: Xenopus (Silurana) tropicalis interferon-induced very large GTPase 1-like (LOC1004920501) mRNA	XM_002939852.1	0.33	1.32	2	0	0	0.01
Xenopus (Silurana) tropicalis uncharacterized LOC100153584 (LOC100153584) mRNA	NM_001114050.1	0.47	1.84	1.99	0	0	0.01
Xenopus tropicalis finished cDNA clone TEG9750/20	CR926268.2	0.98	3.81	1.97	0	0	0.01
Xenopus laevis PNK-FBP1 precursor mRNA complete cds	DQ141336.1	0.75	2.88	1.95	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis chromosome-associated kinesin KIF4-like (LOC100494653) mRNA	XM_004916831.1	0.3	1.13	1.94	0	0	0.01
Xenopus (Silurana) tropicalis fibroblast growth factor 19 (fgf19) mRNA	NM_001142825.1	0.32	1.19	1.94	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC100494747) mRNA	XM_002936043.2	0.35	1.32	1.93	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100494754 (LOC100494754) mRNA	NM_001093542.2	0.27	1.01	1.92	0	0	0.01
Xenopus (Silurana) tropicalis ALR tyrosine kinase receptor-like (LOC100496761) mRNA	XM_002942891.2	1.07	4.03	1.92	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100491693 (LOC100491693) transcript variant X1 mRNA	NM_001097161.1	0.33	1.21	1.9	0	0	0.01
Xenopus laevis mRNA for cardiac troponin C (complete cds)	AB0303080.1	0.67	2.44	1.88	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis membrane protein 238-like (LOC100487502) mRNA	XM_0029349715.2	0.33	1.18	1.88	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100493190 (LOC100493190) mRNA	NM_001093629.2	0.56	2.04	1.87	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis glucose-regulated protein-like (LOC100492570) mRNA	XM_002941644.2	0.37	1.32	1.85	0	0	0.01
Xenopus (Silurana) tropicalis general transcription factor IIA 19/37Da (gtf2a1) mRNA	NM_001016980.2	1.32	4.72	1.84	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis phosphatase orphan 1 (phospho1) transcript variant X2 mRNA	NM_00104918610.1	0.33	1.16	1.83	0	0	0.01
Xenopus laevis COMM domain containing 3 (comm3) mRNA	NM_001095386.1	0.93	3.29	1.83	0	0	0.01
Xenopus tropicalis finished cDNA clone TEG9133d14	CR848487.1	0.39	1.36	1.82	0	0	0.01
Xenopus laevis alcohol dehydrogenase ion containing 1 (adhfe1) nuclear gene encoding mitochondrial protein mRNA	NM_001127802.1	0.56	1.96	1.82	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis mediator of RNA polymerase II transcription subunit 24-like (LOC100491557)	XM_002937856.2	1.24	4.36	1.82	0	0	0.01
Xenopus laevis empty spiracles homeobox (gene 2 lemx1.2) mRNA	NM_001093430.1	0.43	1.49	1.81	0	0	0.01
Xenopus laevis hypothetical protein LOC10036810 mRNA (cDNA clone IMAGE:8070472) partial cds	BG128671.1	0.52	1.8	1.8	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis insulin receptor substrate 2-like (LOC100498409) transcript variant X2 mRNA	NM_0010911986.1	0.46	1.58	1.79	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor family C group 5 member C (gpr5c) transcript variant X4 mRNA	XM_004916328.1	0.61	2.04	1.77	0	0	0.01
Xenopus laevis xPR-1 mRNA (cDNA clone IMAGE:8317959) partial cds	BG124878.1	0.38	1.29	1.77	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C15orf61 (LOC100488454)	NM_0010912591.1	0.32	1.07	1.77	0	0	0.01
Xenopus laevis p21ING1c variant 1 (ing1) mRNA complete cds alternatively spliced	AY601664.1	0.39	1.29	1.74	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis galactosidase beta 1-like 2 (glb12) mRNA	XM_002937982.2	0.86	2.84	1.74	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis nicotinamide N-methyltransferase-like (LOC100491286) transcript variant X4 mRNA	NM_001091194.1	0.49	1.62	1.73	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis zinc finger CCHC domain-containing protein 3-like (LOC100491757) mRNA	NM_001091195.1	0.35	1.16	1.73	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis histamine H3 receptor-like (LOC101735223) mRNA	NM_001096910.1	1.21	3.97	1.72	0	0	0.01
Xenopus laevis fucosyltransferase 10 (alpha 1,3)fucosyltransferase (fut10) mRNA	NM_001091194.1	0.34	1.1	1.71	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis protein fos-like (LOC100490871) transcript variant X2 mRNA	NM_001091195.1	2.77	8.93	1.69	0	0	0.01
Xenopus laevis haplotype i MH class I antigen mRNA complete cds	AF185286.1	0.65	2.08	1.69	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis phospholipid scramblase 2-like (LOC100489273) mRNA	NM_0010938406.2	0.75	2.41	1.69	0	0	0.01
Xenopus (Silurana) tropicalis transfer protein (tppa) mRNA	NM_001090818.1	0.5	1.58	1.68	0	0	0.01
Xenopus laevis 71 kDa protein mRNA (cDNA clone MGCI_96627 IMAGE:9041534) complete cds	BG169900.1	1.25	3.98	1.68	0	0	0.01

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX	FC Eva1 CHX	FC Control CHX	FC Control CHX+DEX	FC Control CHX	Rank <sup>a</sup>
Xenopus tropicalis cDNA clone IMAGE:7004781	BC096007.1	0.44	1.39	1.68	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis butyrophilin subfamily 3 member A3-like (LOC100494525) transcript variant X3 mRNA	XM_004919164.1	0.48	1.51	1.68	0	0	0	0.01
Xenopus laevis trypophan hydroxylase 1 (tph1) mRNA	NM_001087454.1	0.57	1.8	1.67	0	0	0	0.01
Xenopus laevis finished cDNA clone TNed066cd19	CR760077.2	0.55	1.73	1.67	0	0	0	0.01
Xenopus laevis Thbx6 (Thbx6) mRNA complete cds	DQ355794.1	4.13	12.94	1.65	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis non-SMC condensin II complex subunit D3 (incapd3) mRNA	XM_002937980.2	0.49	1.52	1.64	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis angiogeninogen serpin peptidase inhibitor clade A, member 8 (agt) mRNA	NM_001096277.1	0.4	1.24	1.63	0	0	0	0.01
Xenopus (Silurana) tropicalis serum amyloid P component (LOC496665) mRNA.	NM_001011229.2	0.47	1.45	1.63	0	0	0	0.01
Xenopus (Silurana) tropicalis calcium and integrin binding family member 3 (cb3) mRNA	NM_001011260.1	0.34	1.04	1.63	0	0	0	0.01
Xenopus (Silurana) collagen triple helix repeat containing 1 mRNA (cDNA clone IMAGE:7003453) partial cds	BC089073.1	0.39	1.18	1.63	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis 5'-protein-coupled receptor 135 (grp135) mRNA	NM_002933733.2	0.34	1.02	1.62	0	0	0	0.01
Xenopus laevis cDNA clone IMAGE:7552.1 partial cds	BC093552.1	0.39	1.19	1.62	0	0	0	0.01
Xenopus tropicalis hypothetical protein LOC10058502 mRNA (cDNA clone MGCI81221) IMAGE:8914481) complete cds	BC16114.1	0.45	1.36	1.6	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis vomeronasal type-2 receptor 26-like (LOC100487815) mRNA	XM_002945022.2	407.36	1228.41	1.6	0	0	0	0.01
Xenopus tropicalis sine oculis binding protein (Drosophila) mRNA (cDNA clone MGCI722604 IMAGE:7656239) complete cds	BC154687.1	0.49	1.45	1.59	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis undifferentiated LOC100498210 (LOC100498210) mRNA	NM_001173597.1	0.72	2.16	1.59	0	0	0	0.01
Xenopus laevis protein tyrosine phosphatase non-receptor type 6 (ptpn6) mRNA	NM_00129390482.2	0.92	2.73	1.57	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis undifferentiated LOC100494710 (LOC100494710) mRNA	NM_001086337.1	0.35	1.02	1.57	0	0	0	0.01
Xenopus laevis uncharacterized protein MGCG64538 (MGCG64538) mRNA	NM_001086337.1	0.97	2.85	1.56	0	0	0	0.01
Xenopus laevis homeobox 89 (hoxb9) mRNA	BC082918.1	0.5	1.47	1.56	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis avdin-like (LOC100487365) mRNA	NM_002939983.2	0.59	1.7	1.54	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis excitatory amino acid transporter 5-like (LOC100496215) mRNA	NM_002934804.2	0.46	1.33	1.54	0	0	0	0.01
Xenopus laevis transporter superfamily 5 (cd40) mRNA	NM_001145216.1	0.43	1.22	1.53	0	0	0	0.01
Xenopus laevis transporter associated with antigen processing (TAP1) mRNA TAP1-1 allele partial cds	AY204553.1	0.9	2.58	1.52	0	0	0	0.01
Xenopus laevis mRNA for xsurvivin B complete cds	AB197248.1	0.36	1.03	1.52	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis serine peptidase inhibitor Kazal type 2 (acrosin-trypsin inhibitor) (spink2)	NM_002940600.2	0.57	1.62	1.51	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis phosphoprotein Peptidase C delta 3 (picd3) mRNA	NM_002933518.2	1.76	4.97	1.5	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropocalis neurotachin (nbea) transcript variant X2 mRNA	NM_004912032.1	0.49	1.35	1.48	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis protein kinase C delta type-I-like (LOC101730464) mRNA	NM_004912333.1	0.62	1.7	1.48	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis pre-B-cell leukemia homeobox 3 (pbx3) transcript variant X1 mRNA	NM_002937875.2	0.51	1.39	1.47	0	0	0	0.01
Xenopus laevis Zic-4 mRNA complete cds	AB259564.1	0.53	1.42	1.45	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropocalis potassium voltage-gated channel 1sk-related family member 1 (kcnel1) transcript variant X2 mRNA	NM_004912135.1	0.6	1.63	1.44	0	0	0	0.01
Xenopus laevis troponin T type 1 skeletal slow (tnnt1) mRNA	NM_001092738.1	0.56	1.51	1.44	0	0	0	0.01
Xenopus laevis hemopoitically expressed homeobox mRNA (cDNA clone MGCG69042 IMAGE:4965399) complete cds	NM_002935535.2	0.75	1.97	1.41	0	0	0	0.01
Xenopus laevis uncharacterized LOC10036933 (LOC10036933) mRNA	NM_001097704.1	1.17	3.14	1.43	0	0	0	0.01
Xenopus laevis wnt9a (wnt9a) mRNA partial cds	GQ150813.1	0.81	2.15	1.42	0	0	0	0.01
Xenopus laevis similar to calsequitin 2 (cardiac muscle) mRNA (cDNA clone MGCI114674 IMAGE:5516132) complete cds	BC097545.1	0.4	1.04	1.41	0	0	0	0.01
Xenopus laevis dehydrogenase/reductase (Sdh family) member 11 (dhhs11) mRNA	NM_001094963.1	1.39	3.67	1.41	0	0	0	0.01
Xenopus tropicalis solute carrier family 4 anion exchanger member 1 (erythrocyte membrane protein band 3 Diego blood group) (slcda1)	NM_002935535.2	0.75	1.97	1.41	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis metalloprotease (IK1-like (LOC100491951) mRNA	NM_002936336.2	1.52	3.98	1.4	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropocalis claudin 16 (cldn16) mRNA	NM_002934041.2	0.68	2.15	1.4	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropocalis single-minded homolog 1 (Drosophila) (sim1) transcript variant X2 mRNA	NM_002934145.1	0.72	1.89	1.39	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropocalis myo-related transcription factor partner of profilin-like (LOC101732135) mRNA	NM_004911411.1	0.41	1.06	1.39	0	0	0	0.01
Xenopus laevis mRNA for <i>sox17</i> alpha2 complete cds	AB052691.1	0.88	2.28	1.38	0	0	0	0.01
XELFOX36 Xenopus laevis (clone 36.12) mRNA complete cds	MW24752.1	0.97	2.53	1.38	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis KIAA1239 ortholog (kiaa1239) transcript variant X1 mRNA	NM_001142088.1	0.55	1.36	1.38	0	0	0	0.01
Xenopus (Silurana) tropocalis chloride channel voltage-sensitive 3 (clcn3) mRNA	NM_001091700.1	0.46	1.17	1.37	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis myo-1H (myo1h) mRNA	NM_002932063.2	0.88	2.26	1.37	0	0	0	0.01
Xenopus (Silurana) tropicalis sulfotransferase family cytosolic 2B member 1 (sulf2b1) mRNA	NM_001007921.1	0.88	2.26	1.37	0	0	0	0.01

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX	FC Eva1 CHX	FC Control CHX	FC Control CHX+DEX	FC Control CHX	Rank <sup>a</sup>
Xenopus laevis SOUL/heme-binding protein mRNA complete cds	D0004682.1	0.66	1.68	1.36	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis alpha 2-macroglobulin-like (LOC100488238) transcript variant X2 mRNA	XM_004920161.1	0.53	1.35	1.36	0	0	0	0.01
Xenopus tropicalis hypothetical protein LOC594926 mRNA (cDNA clone IMAGE7024052) partial cds	BC092555.1	0.46	1.16	1.34	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis serine protease 48-like (LOC100493951) mRNA	XM_002939691.2	0.98	2.46	1.34	0	0	0	0.01
Xenopus laevis cDNA clone (MAGEf6)5274	BC108482.1	5.33	13.4	1.34	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis PDZ domain containing 2 (pdz2) mRNA	XN_002936677.2	0.57	1.44	1.33	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis homeobox A6 (hoxa6) mRNA	XM_002933395.2	1.97	4.93	1.33	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TIB05016	CU025142.1	0.53	1.31	1.32	0	0	0	0.01
Xenopus laevis hypothetical protein LOC10037233 mRNA (cDNA clone IMAGE8549225) partial cds	BC129784.1	0.51	1.24	1.31	0	0	0	0.01
Xenopus laevis partial mRNA for cAMP-dependent protein kinase catalytic subunit beta	AJ413219.1	0.75	1.83	1.3	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis SH3-domain GRB2-like 3 (sh3g3) mRNA	XM_002940099.2	0.65	1.59	1.3	0	0	0	0.01
Xenopus laevis uncharacterized LOC494801 (LOC494801) mRNA	NM_001094633.1	0.44	1.05	1.29	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TIBdA036f09	CU025205.1	0.51	1.22	1.29	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TNeu070h09	CR848334.2	0.69	1.63	1.26	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis cernamide kinase-like (cerkl) mRNA	XM_002932015.2	1.07	2.54	1.26	0	0	0	0.01
Xenopus tropicalis protein phosphatase 1 regulatory inhibitor subunit 3B mRNA (cDNA clone MGCG135308 IMAGE7541173) complete cds	BC167311.1	0.58	1.37	1.25	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis LOC398438 mRNA (cDNA clone IMAGE5524088) partial cds	BC045073.1	2.75	6.53	1.25	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis anoctamin 2 (ano2) mRNA	XM_002932297.2	0.75	1.17	1.25	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis homolog subfamily B member 5 (dnajb5) transcript variant X8 mRNA	NM_001097746.1	0.62	1.46	1.24	0	0	0	0.01
Xenopus laevis uncharacterized LOC10036689 (LOC10036689) mRNA	NM_001097746.1	0.55	1.29	1.23	0	0	0	0.01
PREDICTED: Xenopus (Silurana) protein phosphatase 1 (Drosophila) (sim 1) transcript variant X3 mRNA	XN_004914546.1	0.46	1.07	1.22	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis single-minded homolog 1 (Drosophila) (sim 1) transcript variant L2 (alldh12) mRNA	XM_002938070.2	0.55	1.26	1.22	0	0	0	0.01
Xenopus laevis kelch-like family member 17 (khl17) mRNA	NM_001095757.1	1.05	2.41	1.22	0	0	0	0.01
Xenopus laevis arylsulfatase family member K (arks) mRNA	NM_001096834.1	1.03	2.38	1.22	0	0	0	0.01
Xenopus laevis CCHZ zinc finger protein C3H-finger protein 1 mRNA (cDNA clone MGCG197081 IMAGE9093736) complete cds	BC170354.1	0.5	1.12	1.19	0	0	0	0.01
Xenopus laevis keratin 17 (krt17) mRNA	NM_001094941.1	0.51	1.16	1.19	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TGA010h16	CT027968.1	0.73	1.65	1.19	0	0	0	0.01
Xenopus (Silurana) tropicalis solute carrier family 25 (mitochondrial carrier; citrate transporter) member 1 (slc25a1)	NM_204060.2	41.84	94.58	1.18	0	0	0	0.01
Xenopus laevis BRCA1 associated protein (brap) mRNA	NM_001089891.1	0.68	1.54	1.18	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TEG086m16	CT025204.2	0.6	1.34	1.18	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TNeu137j02	CR942725.1	0.93	2.09	1.18	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis homeobox A7 (hoxa7) mRNA	NM_004915398.1	0.78	1.76	1.17	0	0	0	0.01
Xenopus laevis Ly-6/αPF-receptor-related protein-1 (LURP-1) mRNA complete cds	AF364819.1	0.8	1.78	1.17	0	0	0	0.01
Xenopus laevis uncharacterized LOC495206 (LOC495206) mRNA	NM_001094893.1	3.36	7.51	1.16	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TIBdA016e20	CU025163.1	1.75	3.9	1.16	0	0	0	0.01
Xenopus (Silurana) tropicalis WRE11 meiotic recombination 11 homolog A (mre11a) mRNA	NM_001114047.1	2	4.42	1.15	0	0	0	0.01
Xenopus (Silurana) tropicalis methyltransferase like 2/IA (mettl2/ia) mRNA	NM_001097392.1	0.57	1.26	1.15	0	0	0	0.01
Xenopus laevis protease serine 2 (trypsin 2) (prss2) mRNA	NM_001095641.1	0.66	1.45	1.15	0	0	0	0.01
Xenopus laevis B-cell receptor-associated protein 29 (bcrap29) mRNA	NM_001093099.1	0.52	1.14	1.15	0	0	0	0.01
Xenopus laevis poly (ADP-ribose) polymerase family member 9 (parp9) mRNA	NM_001091415.1	0.57	1.25	1.15	0	0	0	0.01
Xenopus laevis uncharacterized LOC495513 (LOC495513) mRNA	NM_001095148.1	1.2	2.65	1.15	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis platelet derived growth factor C (pdgfC) mRNA	NM_002932874.2	1.01	2.21	1.14	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TEG035o03	CR926335.2	0.88	1.92	1.14	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 16 member 14 (monocarboxylic acid transporter 14) (slc16a14)	NM_002944218.2	0.66	1.44	1.13	0	0	0	0.01
Xenopus (Silurana) tropicalis butyrophilin subfamily 2 member A1 (btrn2a1) mRNA	NM_001132016.1	0.88	1.91	1.12	0	0	0	0.01
Xenopus laevis chemokine (C-X-C motif) ligand 12 (cxcl12) transcript variant 2 mRNA	NM_001091631.1	0.6	1.28	1.11	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis platelet derived growth factor C (pdgfC) mRNA	NM_002934746.2	1.84	1.8	1.11	0	0	0	0.01
Xenopus laevis transporter associated with antigen processing 1 (TAP1) mRNA	AY204552.1	0.62	1.32	1.1	0	0	0	0.01
Xenopus (Silurana) tropicalis solute carrier family 2 (tfl2) mRNA	NM_001126761.1	0.51	1.07	1.1	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis proton antiporter 5 (parp5) mRNA	NM_002931691.2	0.63	1.32	1.09	0	0	0	0.01
Xenopus laevis hypothetical protein LOC79088 mRNA (cDNA clone IMAGE8328337) partial cds	BC123337.1	0.62	1.32	1.09	0	0	0	0.01

Gene	Accession	Eya1 CHX	Eya1 CHX+DEX	FC Eya1 CHX	FC Control CHX	FC Control CHX+DEX	FC Control CHX	Rank <sup>a</sup>
Xenopus laevis KazaI-type serine peptidase inhibitor domain 1 (kazal1) mRNA	NM_001092073.1	0.79	1.66	1.09	0	0	0	0.01
Xenopus laevis hypothetical protein LOC779026 mRNA (cDNA clone MAGE:3397679) partial cds	BC087548.1	14.82	31.32	1.09	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101733395 (LOC101733395) mRNA	XM_004916468.1	0.83	1.74	1.08	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TNeu016sh19	CR848302.2	1.82	3.82	1.08	0	0	0	0.01
Xenopus (Silurana) tropicalis zinc finger protein 71 (znf71) mRNA	NM_001167599.1	0.9	1.87	1.07	0	0	0	0.01
Xenopus laevis cDNA clone TNeu016sh19	BC160698.1	0.88	1.81	1.06	0	0	0	0.01
Xenopus (Silurana) tropicalis membrane metallo-endopeptidase-like 1 (mmel1) mRNA	NM_001127095.1	1.4	2.89	1.05	0	0	0	0.01
Xenopus tropicalis cDNA clone IMAGE:4964804 partial cds	BC135913.1	1.29	2.66	1.05	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C18orf42 (LOC100488626) mRNA	XM_002938057.2	0.56	1.15	1.05	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis adenylyl cyclase 1 (brain) (adcy1) mRNA	NM_002933415.2	0.66	1.34	1.04	0	0	0	0.01
Xenopus laevis neutral cholesterol ester hydrolase 1 (nch1) mRNA	NM_001099890.1	0.81	1.64	1.03	0	0	0	0.01
Xenopus laevis cDNA clone IMAGE:506637 partial cds	BC073275.1	0.52	1.05	1.03	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis copine II (cpne2) transcript variant X2 mRNA	XM_004913481.1	0.71	1.43	1.03	0	0	0	0.01
Xenopus laevis cDNA clone IMAGE:7205a04	NM_001094724.1	0.75	1.52	1.02	0	0	0	0.01
Xenopus (Silurana) tropicalis adenosylhomocysteinase-like 2 (ahcy2) mRNA	NM_001016409.2	0.57	1.15	1.02	0	0	0	0.01
PREDICTED; Xenopus (Silurana) tropicalis C-type natriuretic peptide 1-like (LOC100495918) mRNA	NM_002937526.1	0.72	1.45	1.02	0	0	0	0.01
Xenopus laevis family with sequence similarity 13 member A (fam13a) mRNA	NM_001099879.1	0.82	1.64	1.01	0	0	0	0.01
Xenopus laevis lipase endothelial (lipg) mRNA	NM_001090061.1	1.56	3.64	1.23	1.38	1.38	0.01	0.01
Xenopus laevis chromogranin A (parathyroid secretory protein 1) (chga) mRNA	NM_001094724.1	2.39	6	1.33	3.64	3.66	0.01	0.01
Xenopus laevis PhyHd mRNA for phytanoyl-CoA dioxygenase like protein complete cds	AB759708.1	2.59	11.34	2.14	37.71	38.07	0.02	0.01
PREDICTED; Xenopus (Silurana) tropicalis monoacylglycerol lipase ABHD12-like (LOC10495642) transcript variant X2 mRNA	NM_001097166.1	0.86	2.61	1.61	1.58	1.59	0.02	0.01
Xenopus laevis copper chaperone for superoxide dismutase (ccs) mRNA	NM_001093342.2	196.09	632.44	1.69	145.99	147.38	0.02	0.01
PREDICTED; Xenopus (Silurana) tropicalis integrin alpha 6 (itga6) mRNA	NM_002934619.2	1.54	3.32	1.12	1.53	1.54	0.01	0.01
Xenopus (Silurana) tropicalis PAF-related orphan receptor A (fora) mRNA	NM_001079195.1	0.44	1.92	2.15	1.49	1.52	0.03	0.02
Xenopus laevis uncharacterized protein MGCG14631 (MGCG14631) mRNA	NM_001096178.1	593.81	2248.48	1.93	23.45	23.88	0.03	0.02
PREDICTED; Xenopus (Silurana) tropicalis cytoplasmic polyadenylation element binding protein 2 (cpeb2) transcript variant X2 mRNA	NM_004912301.1	80.17	221.63	1.47	77.75	79.18	0.04	0.03
Xenopus tropicalis cDNA clone IMAGE:6983060	BC156029.1	10407.3	21758.6	1.07	9823.63	10007.8	0.03	0.03
Xenopus laevis cDNA clone IMAGE:6946680	BC081202.1	0.38	2.92	2.97	1.45	1.53	0.09	0.03
PREDICTED; Xenopus (Silurana) tropicalis NOP2/Sun domain family member 3 (nsun3) transcript variant X2 mRNA	NM_004912074.1	36.17	90.53	1.33	47.75	48.7	0.04	0.03
Xenopus laevis Cbl proto-oncogene E3 ubiquitin protein ligase B (cblb-a) mRNA	NM_001091959.1	34.15	294.44	3.11	23.68	25.4	0.11	0.04
Xenopus laevis cep02 mRNA complete sequence	DQ096881.1	16.8	98.74	2.56	23.03	24.81	0.11	0.05
Xenopus laevis ALX homeobox 1 (alk1) mRNA	NM_001085647.1	2.21	4.53	1.04	2.6	2.69	0.05	0.05
Xenopus laevis CD81 protein (cd81-a) mRNA	NM_001086613.1	270.71	578.42	1.1	110.37	114.58	0.06	0.05
PREDICTED; Xenopus (Silurana) tropicalis protein-glutamine gamma-glutamyltransferase E-like (LOC100486230) mRNA	NM_002935530.2	9.2	19.4	1.08	14.31	14.86	0.06	0.05
PREDICTED; Xenopus (Silurana) tropicalis NEIL-like 1 (nelli) mRNA	AB099282.1	0.47	1.4	1.6	1.63	1.75	0.11	0.07
Xenopus laevis hypothetical protein LOC10127285 mRNA (cDNA clone MAGE:8734320) partial cds	NM_001097261.1	1.32	3.09	1.23	1.08	1.14	0.09	0.07
Xenopus laevis ribosomal protein L35a mRNA (cDNA clone MGCG64295 (MAGE:6874721)) complete cds	BC155879.1	0.6	1.82	1.61	1.2	1.27	0.1	0.06
PREDICTED; Xenopus (Silurana) tropicalis prostaglandin reductase 2 (pgdr2) mRNA	BC053377.1	2.24	6.17	1.47	7.01	7.43	0.09	0.06
Xenopus laevis mRNA for enhancer of split related 9 (esr9 gene) clone 8C9	NM_001095485.1	1.86	5.08	1.46	8.42	8.97	0.07	0.07
Xenopus (Silurana) tropicalis hepcidin antimicrobial peptide (hamp2) mRNA	AJ009282.1	0.47	1.4	1.6	1.63	1.75	0.11	0.06
Xenopus laevis MgCG84338 protein (MGC84338) mRNA	NM_001092786.1	0.56	1.22	1.13	1.15	1.22	0.09	0.08
PREDICTED; Xenopus (Silurana) tropicalis colo-epithelial domain containing 117 (cdce17) transcript variant X1 mRNA	NM_002933702.2	3.07	6.47	1.08	4.01	4.25	0.09	0.08
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC495099 (LOC495099) mRNA	XM_004910896.1	0.82	1.84	1.17	1.31	1.39	0.1	0.08
Xenopus laevis mRNA for uncharacterized LOC495099 (LOC495099) mRNA	NM_001094799.1	1.28	2.93	1.2	2.74	2.93	0.1	0.09
Xenopus laevis mal T-cell differentiation protein (mal) mRNA	NM_001086577.1	6.72	15.14	1.18	10.98	11.74	0.1	0.09
PREDICTED; Xenopus (Silurana) tropicalis hepcidin antimicrobial peptide (hamp2) mRNA	NM_002942678.2	11593.1	2338.54	1.02	653.87	693.24	0.09	0.09
PREDICTED; Xenopus (Silurana) tropicalis cytochrome P450 26-like (LOC100487674) transcript variant X2 mRNA	NM_002935590.2	1.29	3.25	1.35	2.31	2.31	0.12	0.09
PREDICTED; Xenopus (Silurana) tropicalis characterized LOC495099 (LOC495099) mRNA	XM_004917782.1	0.88	1.78	1.02	4.36	4.66	0.1	0.1
Xenopus laevis transmembrane protein 163 (tmem163) mRNA	NM_00109281.1	840.03	1779.43	1.09	24.49	26.34	0.11	0.1
PREDICTED; Xenopus (Silurana) tropicalis ubiquitin protein ligase E3D (ube3d) mRNA	NM_002938455.2	0.41	1.54	1.93	137.19	156.81	0.2	0.1
Xenopus laevis microtubule-associated protein 1 light chain 3 alpha (maplc3a) mRNA	NM_001086397.1	12.87	77.81	2.6	10.56	12.66	0.27	0.11

Gene	Accession	Eya1 CHX	Eya1 CHX+DEX	FC Eya1 <sup>a</sup>	Control CHX	Control CHX+DEX	FC Control CHX	Rank <sup>b</sup>
Xenopus (Silurana) tropicalis SRY (sex determining region Y)-box 2 (sox2) mRNA	NM_213704.3	35.45	82.55	1.22	49.46	53.9	0.13	0.11
PREDICTED; Xenopus (Silurana) tropicalis coiled-coil domain containing 175 (ccdc175) mRNA	XW_004917212.1	1.33	2.76	1.06	1.17	1.27	0.12	0.11
Xenopus (Silurana) tropicalis follistatin (fst) mRNA	NM_001008056.1	2.67	18.04	2.76	1.51	1.87	0.31	0.12
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101731410 (LOC101731410) mRNA	XW_004918259.1	3.4	6.8	1.01	2.45	2.66	0.13	0.13
Xenopus laevis cDNA clone (MAGF551)1680 partial cds	BC097628.1	4.55	9.12	1.01	5.49	5.98	0.13	0.13
PREDICTED; Xenopus (Silurana) tropicalis transmembrane protease serine 13 (tmprss13) mRNA	XW_0029329042	2.39	5	1.07	2	2.2	0.14	0.13
Xenopus (Silurana) tropicalis monothiophosphate-4-phosphatase type I (107Da (inpp4a)) mRNA	NM_001079252.1	3.35	8.44	1.34	5.3	5.96	0.17	0.13
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101733775 (LOC101733775) partial mRNA	XW_004912842.1	0.37	1.03	1.49	2.37	2.71	0.2	0.13
Xenopus laevis mb-2-like 2 (mb2l2.2b) mRNA	NM_001096770.1	0.6	3.95	2.74	1.19	1.53	0.37	0.14
Xenopus (Silurana) tropicalis cholinergic receptor nicotinic alpha 4 (neuronal) (chnaa4) mRNA	NM_001113843.1	17.79	57.5	1.7	18.5	21.72	0.24	0.14
PREDICTED; Xenopus (Silurana) tropicalis small G protein signalling modulator 2 (sgsm2) mRNA	XW_002935292.2	0.53	1.67	1.67	1.94	2.28	0.24	0.14
Xenopus laevis complement factor I (cfi-a) mRNA	NM_001085952.1	2.75	6.16	1.17	1.33	1.49	0.17	0.15
Xenopus laevis solute carrier family 27 (fatty acid transporter) member 6 (slc27a6) mRNA	NM_001096974.1	1.13	4.15	1.88	2.63	3.19	0.28	0.15
PREDICTED; Xenopus (Silurana) tropicalis phospholipase A2 group IV (pla2g4h) mRNA	XW_002932839.2	1.45	3.07	1.09	1.89	2.12	0.17	0.15
Xenopus laevis mb-2-like 2 (mb2l2.2b) mRNA	AB435403.1	1.62	4.39	1.45	2.82	3.28	0.22	0.16
Xenopus laevis Ras-related associated with diabetes (rad1) mRNA	NM_001092750.1	0.11	2.42	4.57	1.03	1.68	0.72	0.16
Xenopus laevis uncharacterized protein MGCI16527 (MGCI16527) mRNA	NM_001096165.1	2.83	5.7	1.01	2.41	2.7	0.17	0.15
Xenopus laevis hypothetical protein LOC445837 mRNA (CDNA clone IMAGE6859328) partial cds	BC076873.1	2.95	6.33	1.11	4.77	5.4	0.19	0.17
Xenopus laevis matrix metalloproteinase 1 (interstitial collagenase) (mmpl1) mRNA	NM_001087049.1	0.47	1.54	1.72	1.2	1.46	0.3	0.17
Xenopus laevis HECT and RLD domain containing E3 ubiquitin protein ligase family member 6 (herc6) mRNA	NM_001092286.1	0.84	2.34	1.5	2.67	3.2	0.27	0.18
PREDICTED; Xenopus (Silurana) 1-t cell leukemia virus 1 (tlx1) transcript variant 1 mRNA	XW_002936768.2	1.38	6.6	2.27	1.12	1.47	0.18	0.18
PREDICTED; Xenopus (Silurana) hypothetical protein LOC445837 mRNA (CDNA clone IMAGE6859328) partial cds	XW_002941495.2	2.28	5.62	1.31	3.91	4.6	0.24	0.19
Xenopus (Silurana) matrix metalloproteinase 1 (interstitial collagenase) (mmpl1) mRNA	NM_001079177.1	0.45	1.82	2.04	0.94	1.23	0.39	0.19
Xenopus laevis mb-2-like 2 (mb2l2.2b) mRNA	NM_001113813.1	1.96	4.73	1.28	1.38	1.63	0.24	0.19
Xenopus laevis regulator of cell cycle (rgcc) mRNA	NM_001093976.1	29.1	61.6	1.09	32.68	37.69	0.21	0.19
Xenopus laevis regulator of cell cycle (rgcc) mRNA	NM_001094263.1	7.96	49.61	2.65	44.18	63.01	0.52	0.2
Xenopus laevis POU class 3 homeobox 2 (pou3f2-b) mRNA	NM_001096751.1	0.52	2.52	2.3	1.01	1.38	0.45	0.2
Xenopus laevis clone IMAGE5571419 mRNA	BC047264.1	28.43	59.82	1.08	24.44	28.28	0.22	0.2
Xenopus laevis cDNA clone IMAGE4405515 partial cds	BC072262.1	0.8	2.19	1.47	1.38	1.69	0.3	0.2
PREDICTED; Xenopus (Silurana) spermatozoal transposon silencer (mael) mRNA	XW_004914247.1	0.57	1.61	1.52	1.28	1.58	0.31	0.21
Xenopus laevis Cep63 mRNA complete cds	NM_001135063.1	47.828	1030.46	1.11	455.15	531.81	0.23	0.21
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC100495928 (LOC100495928) transcript variant X3 mRNA	XW_004911592.1	6.02	12.75	1.09	5.19	6.08	0.23	0.22
Xenopus laevis uncharacterized LOC100158325 (LOC100158325) mRNA	NM_001127773.1	0.75	2.26	1.6	1.24	1.57	0.34	0.22
Xenopus laevis neurotrophin 3 (ntf3) mRNA	NM_001092740.1	1.64	4.54	1.48	1.33	1.66	0.32	0.22
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101733058 (LOC101733058) mRNA	BC070556.1	18.27	74.6	2.08	23.73	32.5	0.46	0.22
Xenopus laevis uncharacterized LOC100189660 (LOC100189560) mRNA	NM_00109385082	13.05	32.18	1.31	5.21	6.35	0.29	0.22
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC100495928 (LOC100495928) transcript variant X3 mRNA	NM_001096456.1	10.29	22.49	1.13	7.5	8.91	0.25	0.22
Xenopus laevis clone IMAGE5571419 mRNA	NM_00129333042	1.06	2.71	1.36	2.72	3.37	0.31	0.23
Xenopus laevis Cyb561 protein mRNA (cDNA clone MGCI79653)IMAGE6637204) complete cds	NM_004913736.1	0.99	2.05	1.06	1.1	1.31	0.24	0.23
PREDICTED; Xenopus (Silurana) tropicalis nucleoporin (07Rda) (nup107) transcript variant X1 mRNA	FJ464988.1	10.6	26.6	1.33	3.05	3.78	0.31	0.24
Xenopus laevis regulator of cell cycle (rgcc) mRNA	NM_001092276.1	1.81	4.11	1.19	2.12	2.57	0.28	0.24
PREDICTED; Xenopus (Silurana) tropicalis Wdr repeat domain 27 (wdr27) mRNA	NM_0029315152	6.04	26.57	2.14	1.26	1.78	0.51	0.24
Xenopus laevis finished cDNA clone TN008903	CR761907.2	1.75	3.63	1.06	1.7	2.03	0.26	0.24
Xenopus laevis uncharacterized protein MGCI3079 (MGCI3079) mRNA	NP_001084719.1	11.41	31	1.45	17.66	22.59	0.36	0.25
Xenopus laevis cDNA clone IMAGE4405515 partial cds	XW_004919220.1	0.61	1.42	1.23	1.1	1.36	0.31	0.25
PREDICTED; Xenopus (Silurana) tropicalis eucube rich repeat containing 72 (lrc72) mRNA	BC157718.1	77.59	157.46	1.03	177.19	210.94	0.26	0.25
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC100492765 (LOC100492765) transcript variant X2 mRNA	XW_0049131.1	2	4.23	1.09	2.25	2.71	0.27	0.25
Xenopus laevis A virus cellular homolog (LOC101732881) mRNA	NM_001011473.1	0.43	1.9	2.15	5.74	8.31	0.54	0.25
PREDICTED; Xenopus (Silurana) tropicalis brain-specific angiogenesis inhibitor 2 (baii2) transcript variant X2 mRNA	XW_004911241.1	0.97	3.04	1.66	2.29	3.05	0.42	0.25
Xenopus laevis Cep63 mRNA complete cds	XW_002933082.2	1.02	2.42	1.25	0.9	1.11	0.32	0.25
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101730954 (LOC101730954) mRNA	XW_004917398.1	1.41	3.43	1.29	0.88	1.1	0.33	0.26

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX	FC Eva1 <sup>a</sup>	Control CHX	Control CHX+DEX	FC Control	Rank <sup>b</sup>
Xenopus laevis CD81 antigen (target of antiproliferative antibody 1) mRNA (cDNA clone MGC:52702 IMAGE:4682721) complete cds	BC041217_1	12.96	27.09	1.07	34.23	41.3	0.28	0.26
PREDICTED: Xenopus (Silurana) tropicalis protein fosB-like (LOC100490871) transcript variant X1 mRNA	NM_001079375_1	2.22	6.46	1.55	7.81	10.35	0.41	0.27
Xenopus (Silurana) tropicalis adrenoceptor alpha 2A (adra2a) mRNA	CR926183_2	1.27	2.74	1.11	1.63	2	0.3	0.27
Xenopus laevis tRNA clone TNeu1 25p14	BC041190_1	0.23	1.22	2.41	15.15	23.93	0.66	0.28
Xenopus laevis Myoblast determination protein 1 homolog A mRNA (cDNA clone MGC:52596 IMAGE:488525) complete cds	NM_001092918_1	1.6	9.79	2.63	3.86	6.38	0.73	0.28
Xenopus laevis nipsnap homolog 3A (nipsnap-a-a) mRNA	NM_001096304_1	0.61	1.47	1.29	0.95	1.22	0.37	0.29
PREDICTED: Xenopus (Silurana) tropicalis dynein cytoplasmic 2 heavy chain 1 (dynch2h1) mRNA	NM_001092256_1	147.2	354.81	1.27	62.59	81.01	0.38	0.3
Xenopus laevis RIB43A domain with coiled-coils 5 (ribc1) mRNA	NM_001092256_1	36.42	87.44	1.27	15.71	20.55	0.39	0.31
Xenopus laevis four and a half LIM domains 5 (fh13) mRNA	NM_001092256_1	128.61	339.33	1.4	132.19	179.14	0.44	0.32
Xenopus tropicalis finished cDNA clone TIPa078d11	CU025077_1	46.9	105.43	1.17	60.92	78.52	0.37	0.32
Xenopus laevis sulfotransfase family cytosolic 1A phenol-prefering member 1 (sult1a1) mRNA	NM_001094083_1	0.26	1.04	2.03	1.05	1.63	0.65	0.32
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor 153 (grp153) mRNA	NM_002934681_2	0.82	1.92	1.24	0.78	1.03	0.41	0.33
Xenopus (Silurana) tropicalis RAS-like family 11 member B (rasl11b) mRNA	NM_001015774_1	2.89	6.22	1.11	5.38	6.94	0.37	0.34
Xenopus tropicalis finished cDNA clone TEg0033a21	CT030593_1	0.52	1.03	1.01	0.97	1.23	0.34	0.34
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100493999 (LOC100493999) transcript variant X2 mRNA	NM_004491590_1	2.55	5.36	1.08	3.16	4.06	0.37	0.34
Xenopus (Silurana) tropicalis G protein-coupled receptor 153 (grp153) mRNA	NM_001128052_1	2.29	4.8	1.07	1.32	1.71	0.38	0.35
PREDICTED: Xenopus (Silurana) tropicalis fast troponin T (TNNT3) mRNA complete cds	AY114144_1	0.95	1.89	1.01	0.81	1.04	0.37	0.37
PREDICTED: Xenopus (Silurana) tropicalis matrix metalloproteinase-18-like (LOC100491214) mRNA	NM_002934930_2	0.51	1.49	1.57	1.4	2.09	0.59	0.38
Xenopus laevis degf03 mRNA complete sequence	DQ968466_1	0.29	1.26	2.14	0.58	1.01	0.81	0.38
Xenopus laevis B-cell transcription gene 5 (btg5) mRNA	NM_001095985_1	12.88	31.79	1.31	18.25	25.64	0.5	0.38
Xenopus laevis comp37 mRNA complete sequence	DQ09619_1	152.7	583.23	1.94	114.82	190.48	0.74	0.38
PREDICTED: Xenopus (Silurana) tropicalis cytokine receptor common subunit beta-like (LOC101733452) mRNA	NM_004913882_1	0.8	2.03	1.36	3.23	4.61	0.52	0.39
Xenopus laevis RELT-like 1b (relt1b) mRNA	NM_001086425_1	1.38	3.39	1.31	1.08	1.53	0.5	0.39
PREDICTED: Xenopus (Silurana) tropicalis TRIM25-like (LOC100496318) mRNA	NM_002937032_2	1.38	2.95	1.1	1.71	2.3	0.44	0.4
PREDICTED: Xenopus (Silurana) tropicalis domain containing 25 mRNA (cDNA clone MGC:154423 IMAGE:875585) complete cds	BC123194_1	1.22	3.96	1.71	0.64	1.03	0.69	0.41
Xenopus laevis heart and neural crest derivatives expressed 2 mRNA (cDNA clone MGC:154423 IMAGE:875585)	NM_001091231_1	3.29	6.92	1.98	2.23	3.01	0.44	0.41
PREDICTED: Xenopus (Silurana) tropicalis ring finger protein 223 (rf223) mRNA	NM_004491652_1	1.05	2.22	1.09	1.71	2.32	0.44	0.41
Xenopus laevis RELT-like 1a (relt1a) mRNA	NM_001091231_1	20.59	57.07	1.48	40.32	61.62	0.62	0.42
PREDICTED: Xenopus (Silurana) tropicalis E3 ubiquitin ligase TRIM15-like (LOC100496318) mRNA	NM_002938641_2	1.05	3.13	1.58	1.24	1.96	0.67	0.43
Xenopus laevis sex determining region Y-box 1 (sox1) mRNA	NM_001086956_1	1.03	2.79	1.45	1.94	3	0.63	0.44
Xenopus (Silurana) tropicalis acyl-CoA oxidase 3 pristanoyl (aco3) mRNA	NM_001015864_1	0.74	1.95	1.4	0.84	1.28	0.61	0.44
PREDICTED: Xenopus (Silurana) tropicalis growth factor independent 1 transcription repressor (gf1) mRNA	NM_002933803_2	0.61	2.01	1.74	0.78	1.32	0.76	0.44
Xenopus laevis RELT-like 1b (relt1b) mRNA	NM_002933849_2	0.94	2.88	1.63	1	1.64	0.72	0.44
PREDICTED: Xenopus (Silurana) tropicalis protein convertase subtilisin/kexin type 9 (pcsk9) mRNA	NM_001095144_1	2.2	4.7	1.1	1.75	2.44	0.49	0.45
PREDICTED: Xenopus (Silurana) tropicalis deasines autosomal dominant 5 (daas5) mRNA	NM_002933392_1	75.701	2110.18	1.48	151.22	239.69	0.67	0.45
Xenopus (Silurana) tropicalis atlastin GTPase 1 (atl1) mRNA	NM_001078754_1	1.11	4.4	2	0.66	1.22	0.9	0.45
Xenopus laevis GRAM domain containing 3 (gram3) mRNA	NM_001095481_1	1.94	3.93	1.02	2.61	3.6	0.47	0.46
PREDICTED: Xenopus (Silurana) tropicalis atonal homolog 1 (atoh1) mRNA	NM_002933849_2	2.29	4.76	1.06	2.26	3.19	0.5	0.47
Xenopus laevis U3 snRNA	X07318_1	3.18	21.45	2.76	1.62	4.03	1.32	0.48
Xenopus laevis xrRIPly3 mRNA for xrRIPly3 protein complete cds	AB45086_1	24.68	49.63	1.01	6.33	8.93	0.5	0.5
Xenopus laevis p21-activated kinase (PAK1) mRNA complete cds	AF00239_1	23.38	79.25	1.77	5.75	10.51	0.88	0.5
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100496891 (LOC100496891) mRNA	NM_001094085_1	1.5	4	1.42	1.05	1.7	0.71	0.5
Xenopus laevis JNK1/MAFK8-associated membrane protein (jamp) mRNA	NM_001094100_1	1.65	3.73	1.18	1.88	2.82	0.59	0.5
Xenopus laevis heat shock protein (hsp) mRNA	NM_0078115_1	72.35	208.23	1.53	36.55	62.19	0.77	0.51
Xenopus laevis PDZ domain containing ring finger 3 (pdzrn3) mRNA	NM_001097041_1	176.66	448.87	1.35	183.25	295.42	0.69	0.52
Xenopus laevis PSMC3 interacting protein (psmc3ip) mRNA	NM_001094071_1	130.77	332.4	1.35	65.9	106.5	0.7	0.52
Xenopus (Silurana) tropicalis salivary homolog 1 (sav1) mRNA	NM_001094931_1	0.87	1.86	1.11	1.3	1.92	0.58	0.52
Xenopus tropicalis 60S ribosomal export protein NMD3 mRNA (cDNA clone MGC:83630 IMAGE:5084801) complete cds	B064238_1	1.39	3.49	1.34	2.27	3.66	0.7	0.52
XELUV52 Xlaevis UV5.2 protein gene 3' end	M27162_1	169.35	380.83	1.17	273.92	419.74	0.62	0.53
Xenopus laevis DIRAS family GTP-binding RAS-like 3 draf3 mRNA	NM_001095243_1	1.09	3.35	1.62	0.96	1.75	0.87	0.54

Gene	Accession	Eya1 CHX	Eya1 CHX+DEX	FC Eya1 <sup>1</sup>	Control CHX	Control CHX+DEX	FC Control	Rank <sup>6</sup>
Xenopus laevis cDNA clone MGC:130954 IMAGE:7973902 complete cds	BC106382.1	2.37	5.11	1.12	1.9	2.86	0.6	0.54
Xenopus tropicalis CDC91 cell division cycle 91-like 1 (S. cerevisiae) mRNA (cDNA clone MGC:197523 IMAGE:9040054) complete cds	BC170796.1	0.78	2.27	1.55	2.21	3.93	0.84	0.54
Xenopus (Silurana) tropicalis early growth response 2 (Krox-20 homolog) (egf2/mRNA)	NM_001100255.1	1.54	4.86	1.67	1.21	2.25	0.91	0.55

<sup>1</sup>Expression levels (FPKM) in pre-placodal explants after injection of Eya1 and CHX treatment

<sup>2</sup>Expression levels (FPKM) in pre-placodal explants after injection of Eya1 and CHX+DEX treatment

<sup>3</sup>Log<sub>2</sub> Fold change values for Eya1

<sup>4</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup>Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup>Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## E.4 Eya1: Down-regulated genes

**Table E.4** Genes with at least two-fold down-regulation after injection of Eya1-GR and treatment with CHX + DEX

Gene	Accession	Eya1 CHX	Eya1 CHX+DEX <sup>2</sup>	FC Eya1 <sup>3</sup>	Control CHX	Control CHX+DEX <sup>4</sup>	FC Control CHX+DEX <sup>5</sup>	Rank <sup>6</sup>
Xenopus laevis Similar to myosin light polypeptide 9 regulatory clone IMAGE:5570659 mRNA	BC042231.1	28.17	0.01	-14.79	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101735120 (LOC101735120) mRNA	XM_004919916.1	20.54	0.01	-14.33	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis granzyme A-like (LOC100496550) transcript variant X2 mRNA	XM_004910357.1	17.77	0.01	-14.12	0	0	0	0
Xenopus (Silurana) tropicalis chromosome 66 (cl9orf66) mRNA	NM_001097193.1	5.47	0.01	-12.42	0	0	0	0
XELNKF7AA-X.leius xnf7 protein mRNA complete cds	M63705.1	1.91	0.01	-10.9	0	0	0	0
Xenopus laevis lactamase beta 2 (lactzb) mRNA	NM_001094943.1	94877.61	60.3	-10.62	175344.33	413960.9	1.24	0
PREDICTED: Xenopus (Silurana) tropicalis extensin-like (LOC101731394) mRNA	XM_004918752.1	1.47	0.01	-10.52	0	0	0	0
XELNUC1B-X.leius mucin B1 consensus repeat mRNA	M83821.1	1.44	0.01	-10.49	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis serum deprivation-response protein-like (LOC100495531) mRNA	XM_002937440.2	1.06	0.01	-10.04	0	0	0	0
Medicago truncatula Glucan 1,3-beta-glucosidase (MTR_Bg091310) mRNA complete cds	NM_003630099.1	11.03	0.38	-4.89	0	0	0	0
Xenopus laevis MAX dimerization protein 3 (maxd3) mRNA	NM_001096719.1	313.61	16.55	-4.25	4.05	5.56	0.46	0
Xenopus laevis causequestrin 1 (fast-twitch skeletal muscle) (casq1) nuclear gene encoding mitochondrial protein mRNA	NM_001086758.1	1.49	0.1	-4	0	0	0	0
Xenopus laevis uncharacterized LOC100127280 (LOC100127280) mRNA	NM_001112852.1	1.63	0.11	-3.96	0	0	0	0
Xenopus laevis mRNA repeat sequence Xstar	AB039922.1	146.95	9.94	-3.89	4.78	16.12	1.76	0
PREDICTED: Xenopus (Silurana) tropicalis sodium- and chloride-dependent creatine transporter 1-like (LOC100489909)	NM_002935493.2	1.28	0.1	-3.71	0	0	0	0
Xenopus tropicalis finished DNA clone Tegg36.d01	CR761262.2	7.27	0.67	-3.46	3.93	5.02	0.36	0
Xenopus laevis NK3 transcription factor related locus 1 (Drosophila) mRNA (cDNA clone MGIC:97890 IMAGE:6317935) complete cds	BC085075.2	3.65	0.36	-3.37	1.04	1.06	0.04	0
PREDICTED: Xenopus (Silurana) tropicalis sterile alpha motif domain containing 12 (samd12) mRNA	XM_002940572.2	1.08	0.11	-3.34	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis 2-aminoethanethiol (cysteamine) dioxygenase (ado) mRNA	NM_002939541.2	4.23	0.47	-3.2	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis gamma-crystallin 1-like (LOC100493812) mRNA	NM_002934791.2	1.36	0.16	-3.13	0	0	0	0
Xenopus tropicalis finished DNA clone The0242k05	CB760075.2	14.11	1.66	-3.1	0	0	0	0
Xenopus laevis cDNA clone IMAGE:5514602	BC087617.1	1305.27	166.66	-2.97	11.19	13.9	0.32	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101732389 (LOC101732389) mRNA	NM_004918291.1	1.07	0.16	-2.78	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 4b (gladb) mRNA	XM_002942676.2	2.21	0.33	-2.76	0	0	0	0
Xenopus laevis cDNA clone IMAGE:6946466 containing frame-shift errors	BC110925.1	1.8	0.28	-2.71	0	0	0	0
X.laevis XFO-1 mRNA for zinc finger protein	X70676.1	881.13	14.22	-2.64	15.78	23.84	0.6	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733743 (LOC101733743) mRNA	NM_004915875.1	2.43	0.4	-2.62	0	0	0	0
Xenopus laevis gap junction protein alpha 4 b (gap4b) mRNA	NM_001171972.1	72.49	12.11	-2.59	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 585A-like (LOC00487634) transcript variant X1 mRNA	NM_0029419038.1	469.57	85	-2.47	45.53	60.18	0.41	0
PREDICTED: Xenopus (Silurana) tropicalis elastin microfibril interfacer 3 (elmin3) mRNA	NM_002932856.2	2.65	0.5	-2.41	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731841 (LOC101731841) mRNA	NM_004914855.1	1.37	0.26	-2.4	0	0	0	0
Xenopus laevis cDNA clone IMAGE:4032905 partial cds	BC060385.1	1.96	0.38	-2.37	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tripartite motif-containing protein 7-like (LOC100494464) mRNA	NM_002941303.2	3.85	0.75	-2.37	1.01	1.32	0.39	0
Xenopus laevis hypothetical protein LOC46932 mRNA (cDNA clone IMAGE:7766060) partial cds	BC108446.1	1.18	0.23	-2.36	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis elastin microfibril interfacer 3 (elmin3) mRNA	NM_004913662.1	1.12	0.25	-2.22	0	0	0	0
Xenopus (Silurana) tropicalis lectin galactoside-binding soluble 12 (lgals12) transcript variant X2 mRNA	NM_001049578.1	5.23	1.14	-2.21	0	0	0	0
Xenopus laevis cDNA clone IMAGE:720336	BC130151.1	31672.08	7106.13	-2.16	4716.89	486957	0.05	0
Xenopus (Silurana) tropicalis E74-like factor 1 (ets domain transcription factor) (elf1) mRNA	NM_001079386.1	2.45	0.55	-2.16	0	0	0	0
Xenopus laevis similar to co-stimulatory complex subunit zeta 1 mRNA (cDNA clone IMAGE:5569945) partial cds	BC047988.1	595.61	135.29	-2.14	1738	19.45	0.13	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00495491 (LOC100495491) transcript variant 1 mRNA	NM_002941655.2	1.2	0.28	-2.1	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis vomer nasal type-2 receptor 26-like (LOC100493683) mRNA	NM_002935452.2	9.1	2.17	-2.08	1.26	1.61	0.36	0
PREDICTED: Xenopus (Silurana) tropicalis putative all-trans-retinol 13,14-reductase-like (LOC100488563) transcript variant X1 mRNA	NM_001033947.1	4.3	1.07	-2.02	0	0	0	0
Xenopus (Silurana) tropicalis neutrophil cytosolic factor 2 (ncf2) mRNA	BC059203.1	1.62	0.41	-2	1.14	2.05	0.86	0
Xenopus tropicalis cone arrestin mRNA (cDNA clone MGIC:15048 IMAGE:6553716) complete cds	BC160462.1	45.33	11.4	-2	9.01	15.05	0.75	0
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC101730562) mRNA	NM_004914259.1	1.01	0.26	-2	0	0	0	0

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX <sup>2</sup>	FC Eva1 <sup>1</sup>	Control CHX	Control CHX+DEX <sup>2</sup>	FC Control CHX	Rank <sup>2</sup>
PREDICTED: Xenopus (Silurana) tropicalis tenascin XB (tnxb) mRNA	XM_002941267.2	1.02	0.26	-1.99	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis purmilio homolog 2 (pum2) mRNA	XM_002934483.2	1.52	0.4	-1.96	0	0	0	0
Xenopus laevis tripartite motif containing 2 (trin2) mRNA	NM_001092023.1	13.55	3.5	-1.96	1.87	2.82	0.6	0
PREDICTED: Xenopus (Silurana) tropicalis latrophilin 1 (lphn1) transcript variant X6 mRNA	XM_004918881.1	1.38	0.37	-1.94	0	0	0	0
Xenopus laevis uncharacterized protein MGCB1517 (MGCB1517) mRNA	NM_001091649.1	1.53	0.41	-1.92	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100488736 (LOC100488736) transcript variant X2 mRNA	XM_004916677.1	73.06	19.67	-1.9	35.79	42.25	0.24	0
Xenopus tropicalis finished cDNA clone Thieu124c03	CR760142.2	4.71	1.29	-1.88	5.68	7.54	0.41	0
PREDICTED: Xenopus (Silurana) tropicalis olfactory receptor 5M10-like (LOC101731270) partial mRNA	NM_001095116.1	1.68	0.46	-1.88	0	0	0	0
Xenopus laevis uncharacterized LOC495464 (LOC495464) mRNA	U58834.1	1.2	0.33	-1.87	0	0	0	0
XLUS834. Xenopus laevis mothers against DPP (XWad) mRNA complete cds	XM_00491828.1	10.01	2.84	-1.83	8.6	12.14	0.5	0
PREDICTED: Xenopus (Silurana) tropicalis thymocyte selection associated family member 2 (themis2) mRNA	XM_004912318.1	1.21	0.35	-1.82	0	0	0	0
Xenopus laevis delta/notch-like GF repeat containing b mRNA (cDNA clone IMAGE:6956228) partial cds	BC094199.1	8.78	2.56	-1.78	1.44	2.07	0.52	0
PREDICTED: Xenopus (Silurana) tropicalis ADAMTS-like 1 (adams1) mRNA	NM_004910864.1	6.74	1.99	-1.77	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tenasin 1 (tns1) mRNA	XM_004918228.1	1.18	0.35	-1.76	0	0	0	0
Xenopus tropicalis finished cDNA clone TEB133f09	CT025230.1	3.3	0.98	-1.76	0	0	0	0
Xenopus laevis hypothetical protein LOC100073003 mRNA (cDNA clone IMAGE:6633659) partial cds	BC130091.1	2.27	0.68	-1.74	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis ADAM metallopeptidase with thrombospondin type 1 motif 3 (adams3) mRNA	XM_002940352.2	1.97	0.6	-1.74	0	0	0	0
Xenopus laevis repetitive 17/23 element fragment C17	X00077.1	396.5	119.46	-1.74	97.57	125.81	0.37	0
Xenopus (Silurana) tropicalis matrix metallopeptidase 13 (collagenase 3) (mmpl3) mRNA	NM_001032335.1	1.2	0.37	-1.71	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis granulocyte colony-stimulating factor receptor-like (LOC100498116) mRNA	XM_002938741.2	3.19	0.99	-1.7	0	0	0	0
Xenopus laevis mindbomb E3 ubiquitin protein ligase 2 (mib2) mRNA	NM_001127864.1	1.23	0.39	-1.67	0	0	0	0
Xenopus (Silurana) tropicalis ring finger protein 182 (rnf182) mRNA	XM_002940635.2	1.03	0.33	-1.66	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis sarcoglycan epsilon (sgce) mRNA	AJ011652.1	3.91	1.28	-1.64	0	0	0	0
Xenopus laevis mRNA for brain factor 2				-1.62	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis PEST proteolytic signal containing nuclear protein (pcnp) transcript variant X1 mRNA	XM_002936816.2	1.36	0.45	-1.6	0	0	0	0
Xenopus (Silurana) tropicalis zinc finger protein 182 (zfp182) mRNA	XM_00293217.2	1.06	0.36	-1.57	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis mannosidase alpha class 2B member 2 (man2b2) mRNA	NM_001113044.1	1.68	0.57	-1.56	0	0	0	0
Xenopus laevis zinc finger protein Fez mRNA (complete cds)	AF195021.1	1.05	0.37	-1.53	0	0	0	0
Xenopus laevis MGCB4396 protein (MGCB4396) mRNA	NM_001094447.1	1.24	0.43	-1.53	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis traf2, and NCK-interacting protein kinase-like (LOC101735220) transcript variant X1 mRNA	XM_004915408.1	8.13	2.85	-1.52	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7619279 partial cds	BC161331.1	1.52	0.55	-1.48	0	0	0	0
Xenopus tropicalis finished cDNA clone TEBg07-d17	CR855513.2	2.64	0.96	-1.47	1.13	2.24	0.99	0
Xenopus laevis solute carrier family 25 (mitochondrial iron transporter) member 28 (slc25a28-b)	NM_001094037.1	2.96	1.07	-1.47	0	0	0	0
Xenopus laevis XEBF-3 protein (xebf-3) mRNA (complete cds)	AF040994.3	6.25	2.27	-1.47	2.23	2.61	0.23	0
Xenopus tropicalis finished cDNA clone TEBg06hb10	CU075091.1	1.14	0.42	-1.47	0	0	0	0
Xenopus laevis MGCB3624 protein mRNA (cDNA clone MGCB3624) IMAGE:5084399 complete cds	BC073118.1	680.23	247.48	-1.46	406.5	996.49	1.3	0
PREDICTED: Xenopus (Silurana) tropicalis integrin alpha 9 (itga9) transcript variant X2 mRNA	NM_004915354.1	1.14	0.42	-1.46	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis ephrin A5b-like (LOC101730271) mRNA	XM_004921023.1	7.75	2.86	-1.45	12.7	13.93	0.14	0
Xenopus laevis Wiskott-Aldrich syndrome (was) mRNA	NM_001097930.1	1.2	0.45	-1.45	0	0	0	0
Xenopus laevis hypothetical LOC96307 mRNA (cDNA clone IMAGE:6868091) partial cds	BC088825.1	3.66	1.36	-1.43	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis 6-phosphofructo-2-kinase/fructose 2,6-bisphosphatase-like (LOC100488795) transcript variant X3 mRNA	NM_004919631.1	55.84	20.87	-1.43	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7790739	BC161549.1	1.18	0.45	-1.42	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731402 (LOC101731402) mRNA	XM_004914240.1	2.82	1.07	-1.41	0	0	0	0
Xenopus laevis HESX homeobox 1 (hesx1-b) mRNA	NM_001162570.1	1.54	0.59	-1.39	1.07	1.39	0.39	0
PREDICTED: Xenopus (Silurana) tropicalis collagen type XI alpha 1 (col11a1) transcript variant X2 mRNA	NM_004918818.1	1.44	0.56	-1.39	0	0	0	0
Xenopus laevis mitogen-activated protein kinase (mapk) 7 mRNA (partial)	NM_001087648.1	779.79	300.64	-1.38	274.19	382.01	0.48	0
Xenopus tropicalis sphingomyelin phosphodiesterase (smpd2) mRNA	NM_001126737.1	2.89	1.12	-1.38	0	0	0	0
Xenopus laevis prostaglandin F2 receptor 3 (subtype EP3) (pig3) mRNA	NM_001098711.1	3.04	1.19	-1.37	0	0	0	0
Xenopus laevis MGCB3480 protein (MGCB3480) mRNA	NM_001093688.1	7181.29	2801.7	-1.36	1879.71	4171.42	1.16	0
Xenopus laevis chromosome 2 open reading frame 40 (C2orf40) mRNA	NM_001085712.1	1.07	0.42	-1.36	0	0	0	0
Xenopus (Silurana) tropicalis cytochrome P450 family 7 subfamily B poly(omega peptide 1 (cyp7b1) mRNA	NM_001127039.1	1.74	0.68	-1.36	0	0	0	0

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX <sup>2</sup>	FC Eva1 CHX+DEX <sup>2</sup>	Control CHX	Control CHX+DEX <sup>2</sup>	FC Control CHX	Rank <sup>4</sup>
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized [LOC100486501] (LOC100486501) mRNA	XM_002942085.2	15.03	5.92	-1.35	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:5936686	BC168616..1	18.27	7.2	-1.35	1.18	2.75	1.23	0
Xenopus laevis cDNA clone IMAGE:6323682	BC099050..1	1.57	0.63	-1.34	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized [LOC101730913] (LOC101730913) mRNA	XM_004916814..1	1.26	0.51	-1.33	0	0	0	0
Xenopus laevis myelin PI-ribosyl membrane protein DM gamma1 mRNA complete cds	Af329364..1	138.03	55.18	-1.33	1.43	2.11	0.57	0
PREDICTED: Xenopus (Silurana) tropicalis mitogen-activated protein kinase MLT-like (LOC100495797) transcript variant X1 mRNA	XM_002934606..2	617.43	247.89	-1.32	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis myelin protein zero-like 1 (mpz1) transcript variant X5 mRNA	XM_004912271..1	325.63	131.56	-1.31	49.31	57.29	0.22	0
PREDICTED: Xenopus (Silurana) tropicalis formin-like 3 (fmln3) transcript variant X5 mRNA	XM_004911841..1	6.54	2.65	-1.31	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis sole carrier family 35 member Ed (slc35e4) transcript variant X2 mRNA	XM_004910461..1	1.39	0.57	-1.3	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis CDG2 effector protein (Rho GTPase binding) 1 (cdg2ep1) transcript variant X2 mRNA	XM_004913388..1	1.52	0.63	-1.28	0	0	0	0
Xenopus laevis SH2 domain containing 3C (sh2d3c) mRNA	NM_001086327..1	1.78	0.74	-1.28	1.3	2.12	0.71	0
PREDICTED: Xenopus (Silurana) tropicalis oocyte zinc finger protein XIQCF22-like (LOC100491191) mRNA	XM_004919970..1	1.45	0.61	-1.27	0	0	0	0
Xenopus laevis epor mRNA for erythropoietin receptor homolog complete cds	AB189477..1	952.82	397.21	-1.27	159.6	198.2	0.32	0
Xenopus laevis enolase-phosphatase 1 (enopl1) mRNA	NM_001095889..1	9.52	4.07	-1.23	1.56	1.8	0.22	0
Xenopus laevis CUE domain containing 1 (cuecd1) mRNA	NM_001086219..1	1.39	0.6	-1.22	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zinc finger CCCH domain-containing protein 3-like (LOC101735217) mRNA	XM_004913331..1	5.07	2.19	-1.22	0	0	0	0
Xenopus laevis zona pellucida C glycoprotein mRNA (cDNA clone MGCS3138 IMAGE:6318867) complete cds	BC072326..1	1.36	0.59	-1.21	0	0	0	0
Xenopus laevis uncharacterized LOC100049773 (LOC100049773) mRNA	NM_001098708..1	1.09	0.48	-1.21	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7808293	BC157693..1	1.35	0.59	-1.21	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis protein FAM19A-like (LOC100485411) partial mRNA	XM_002944313..2	1.17	0.51	-1.2	0	0	0	0
Xenopus tropicalis finished DNA clone Nieu40415	CR848351..2	7.81	3.43	-1.19	0	0	0	0
Xenopus laevis epithelial sodium channel alpha 1 subunit mRNA (cDNA clone MGCS196631 IMAGE:9041538) complete cds	BC169904..1	361.3	15.91	-1.19	6	8.02	0.42	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized [LOC100488691] (LOC100488691) mRNA	XM_004919929..1	19	8.39	-1.18	0	0	0	0
Xenopus laevis coiled-coil domain containing 6 (ccdc6) mRNA	NM_001086144..1	221.11	97.99	-1.18	26.03	27.34	0.08	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100498475 (LOC100498475) mRNA	XM_002932828..1	2.2	1	-1.15	1.22	2.15	0.82	0
PREDICTED: Xenopus (Silurana) tropicalis ATP/GTP binding protein-like 3 (pgb3) mRNA	XM_004921447..1	8.41	3.81	-1.15	2.94	3.56	0.28	0
Xenopus tropicalis finished DNA clone Tgas17k07	CR848082..2	1.09	0.5	-1.14	0	0	0	0
Xenopus laevis DEXH (Asp-Glu-X-His) box poly peptide 5.8 (dhx5) mRNA	NM_001092446..1	1.36	0.62	-1.13	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis glycerol-3-phosphate acyltransferase 3-like (LOC100485787) mRNA	XM_002939586..2	2.77	1.28	-1.12	0	0	0	0
Xenopus (Silurana) tropicalis DnaI (Hsp40) homolog subfamily C member 28 (dnajc28) mRNA	NM_001015977..1	3208.1	1480.26	-1.12	288.13	396.24	0.46	0
Xenopus laevis forkhead box H1 gene 2 (foxh1.2) mRNA	NM_001172075..1	2	0.93	-1.12	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis retrotransposon-derived protein PEG10-like (LOC101734357) mRNA	XM_004911607..1	259.18	120.8	-1.11	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:59292505	BC082728..1	14709.84	6887.53	-1.1	3931.48	4809.24	0.3	0
PREDICTED: Xenopus (Silurana) tropicalis araf domain containing kinase 2 (adck2) mRNA complete cds	XM_002941614..2	1.22	0.58	-1.08	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis slingshot protein phosphatase 2 (ssh2) transcript variant X4 mRNA	XM_004911701..1	3.32	1.57	-1.08	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis element-derived protein 4-like (LOC100487842) mRNA	XM_004911445..1	4.85	2.31	-1.08	2.89	3.12	0.11	0
Xenopus tropicalis genomic-screened homeobox 2 (ghs2) mRNA complete cds	DQ195531..1	1.75	0.84	-1.08	0	0	0	0
Xenopus laevis uncharacterized protein MGCS8797 (MGCS8797) mRNA	NM_001090141..1	3.35	1.6	-1.07	2.71	3.04	0.17	0
Xenopus laevis sole carrier family 35 member F2 (slc35f2) mRNA	NM_001094969..1	56.86	27.19	-1.07	33.18	37.47	0.18	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731321 (LOC101731321) partial mRNA	XM_004920844..1	1.45	0.69	-1.07	2.28	2.93	0.36	0
Xenopus (Silurana) tropicalis nuclear factor of activated T-cells cytoplasmic calcineurin-dependent 1 (nfatc1) mRNA	NM_001079272..1	1.01	0.49	-1.07	0	0	0	0
Xenopus laevis fsh beta mRNA for follicle-stimulating hormone beta-subunit complete cds	A9175888..1	996.71	477.14	-1.07	2.21	9.93	2.17	0
Xenopus laevis cyclin A1 mRNA (cDNA clone MGCS132235 IMAGE:5048077) complete cds	BC111510..1	1.04	0.5	-1.07	0	0	0	0
Xenopus laevis FEZ family zinc finger 2 (fezf2) mRNA	NM_00109644..1	1.05	0.51	-1.06	0	0	0	0
Xenopus laevis sole carrier family 3 mRNA complete cds	DC327671..1	1.6	0.77	-1.06	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis bicarbonate transporter member 10 (slc4a10) transcript variant X2 mRNA	NM_001093916..1	831.19	404.12	-1.05	82.21	100.36	0.29	0
Xenopus laevis metallothionein 1 gene 2 (mtt2) mRNA	XM_004917699..1	1.63	0.79	-1.04	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis carrier family 4 sodium bicarbonate transporter member 10 (slc4a10) transcript variant X2 mRNA	BC074494..1	2.66	1.31	-1.03	0	0	0	0
Xenopus laevis trafficking protein particle complex 9 (trapc9) mRNA	NM_001089879..1	41.49	20.39	-1.03	13.62	19.06	0.49	0
Xenopus (Silurana) tropicalis lanthionotic synthetase component C-like 1 (bacterial) (lancl1) mRNA	NM_001016046..3	3.37	1.66	-1.03	0	0	0	0

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX <sup>2</sup>	FC Eva1 <sup>1</sup>	Control CHX	Control CHX+DEX <sup>2</sup>	FC Control CHX	Rank <sup>4</sup>
Xenopus laevis hypothetical LOC495397 mRNA (cDNA clone IMAGE:6876609) partial cds	BC084868.1	24.78	12.21	-1.03	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis protein FAM107B-like (LOC101730771) transcript variant X1 mRNA	NM_004916718.1	1.18	0.59	-1.02	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7680666 partial cds	BC168625.1	1.4	0.7	-1.02	0	0	0	0
Xenopus tropicalis MPN domain containing (mpn) mRNA	NM_00102976.1	1.45	0.72	-1.02	0	0	0	0
Xenopus laevis hypothetical protein 1 (LOC33298 mRNA (cDNA clone IMAGE:6934329) partial cds	BC099260.1	1.86	0.93	-1.02	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis Ras protein-specific guanine nucleotide-releasing factor 1 (rasrf1) mRNA	NM_002932963.2	1.29	0.64	-1.01	1.44	1.51	0.07	0
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C12orf55 (LOC101735126) mRNA	NM_004913179.1	13.2	6.58	-1.01	0	0	0	0
Xenopus laevis uncharacterized LOC496105 (LOC496105) mRNA	NM_001095360.1	1.76	0.88	-1.01	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis phosphopanthotheyone/steine decarboxylase-like (LOC100495698) transcript variant X2 mRNA	NM_00491598.1	65.41	32.64	-1.01	17.86	20.16	0.18	0
Xenopus laevis GRB2 associated regulator of MAPK1-like (garem) mRNA	NM_001094088.1	1.76	0.88	-1.01	1.86	2.12	0.19	0
Xenopus laevis C/EBP delta-2 mRNA for CCAAAT-enhancer binding protein delta complete cds	AB107221.1	27.16	0.01	-14.73	57.97	54.62	-0.09	0.01
Xenopus laevis transmembrane protein 55B (tmem55b) mRNA	NM_001094660.1	48.12	19.92	-1.28	25.26	25.01	-0.02	0.02
XLRGAOP3_X.laevis oocyte Ss RNA pseudogene clone pxL031	KO1368.1	792.69	4.56	-7.45	1.55	1.34	-0.22	0.03
PREDICTED: Xenopus (Silurana) tropicalis peroxisomal biogenesis factor 6 (pexf6) partial mRNA	NM_002943464.2	43.48	5.92	-2.88	4.39	4.13	-0.09	0.04
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 510 (znf510) mRNA	NM_002941361.2	6.04	1.88	-1.69	4.84	4.64	-0.07	0.04
Xenopus laevis thromboxane A2 receptor (tbx2r) mRNA	NM_001093555.1	3153.44	364.94	-3.12	2.54	2.33	-0.13	0.05
PREDICTED: Xenopus (Silurana) tropicalis receptor-type tyrosine-protein phosphatase eta-like (LOC101732323) mRNA	NM_004914831.1	3.03	0.84	-1.87	12.52	11.77	-0.09	0.05
Xenopus laevis solute carrier family 25 (carnitine/acylcarnitine translocase) member 20 gene 2 (slc25a20_2) mRNA	NM_001095111.1	1903.43	21.99	-6.44	31.81	25.12	-0.35	0.06
PREDICTED: Xenopus (Silurana) tropicalis vomeronasal type-2 receptor 26-like (LOC100487329) partial mRNA	NM_002943234.2	1786.19	57.94	-4.95	33.81	27.6	-0.3	0.06
Xenopus laevis hypothetical protein MG58493 mRNA (cDNA clone MG:CS3264) (IMAGE:6637488) complete cds	BC068970.1	2640.1	976.84	-1.44	329.36	309.45	-0.09	0.07
Xenopus laevis protein phosphatase 2A activator regulatory subunit 4 (ppp4r) mRNA	NM_001095603.1	407.22	143.36	-1.51	93.35	86.44	-0.12	0.08
PREDICTED: Xenopus (Silurana) tropicalis putative nuclelease HARB1-like (LOC100495422) mRNA	NM_002938286.2	105.5	4.06	-1.38	12.62	11.75	-0.11	0.08
Xenopus laevis splicing factor 1 (sf1) mRNA	NM_001086731.1	1.97	0.7	-1.5	49.68	45.91	-0.12	0.08
Xenopus tropicalis postsynaptic density protein 95 mRNA (cDNA clone IMAGE:7661545) partial cds	BC123007.1	3.37	1.69	-1.01	2.14	2.03	-0.08	0.08
PREDICTED: Xenopus (Silurana) tropicalis WW family member 3 (wwc3) mRNA	NM_002932795.2	121.79	49.19	-1.31	3.7	3.44	-0.11	0.08
Xenopus laevis empty spliceosomal homeobox 2 (enx2) mRNA	NM_001092583.1	600.74	300.32	-1.01	149.38	140.98	-0.09	0.09
Xenopus laevis MGCB82906 protein (MGCB82906) mRNA	NM_001092491.1	32.71	15.36	-1.1	43.16	40.45	-0.1	0.09
Xenopus laevis dyrinin 1-like (dhnml) mRNA	NM_001086714.1	321.36	150.54	-1.1	28.42	26.6	-0.1	0.09
Xenopus laevis MGCB84462 protein (MGCB84462) mRNA	NM_001092815.1	122.93	58.45	-1.08	82.7	77.28	-0.1	0.1
Xenopus laevis Similar to RIKEN cDNA 181036f124 gene clone IMAGE:5542252 mRNA	BC043892.1	2.15	0.01	-1.07	9.51	4.64	-1.04	0.14
PREDICTED: Xenopus (Silurana) tropicalis aryl-hydrocarbon receptor nuclear translocator 2 (arnt2) mRNA	NM_001087070.1	1.22	0.4	-1.63	12.01	10.73	-0.17	0.1
Xenopus laevis maleolin (malec-a) mRNA	NM_001091743.1	4749.94	1268.79	-1.91	143.12	125.01	-0.2	0.11
Xenopus (Silurana) tropicalis zinc-finger protein 160 (zfp160) mRNA	NM_001016517.2	46.71	17.02	-1.46	7.24	6.46	-0.17	0.12
XELBLAA X borealis B1 protein mRNA complete cds	M63663.1	39327.31	9126.19	-2.11	4051.44	3420.12	-0.25	0.12
Xenopus laevis uncharacterized LOC100158321 (LOC100158321) mRNA	NM_001127770.1	22.54	3.54	-2.68	2.73	2.17	-0.34	0.13
Xenopus (Silurana) tropicalis ZFP2 zinc finger protein (zf2) mRNA	NM_00107894.6	4.14	1.81	-1.2	12.08	10.83	-0.16	0.14
Xenopus laevis hypothetical LOC495219 mRNA (cDNA clone IMAGE:4174149) partial cds	BC084630.1	2.24	0.97	-1.21	1.27	1.13	-0.18	0.15
Xenopus laevis RAB15 member RAS oncogene (rab15) mRNA	NM_00108923.6	306.74	133.91	-1.2	114.66	100.5	-0.2	0.16
Xenopus laevis GID complex subunit 8 (homolog (S. cerevisiae) (gid8)) mRNA	NM_001096765.1	2351.78	869.87	-1.44	20.52	17.49	-0.24	0.17
PREDICTED: Xenopus (Silurana) tropicalis platelet-derived growth factor beta polypeptide (pdgb) transcript variant X1 mRNA	NM_002933750.2	1.1	0.46	-1.25	1.03	0.9	-0.21	0.17
PREDICTED: Xenopus (Silurana) tropicalis dynamin heavy chain 5 (axonemal-like (LOC100488102) mRNA	NM_004915670.1	70.06	28.85	-1.29	9.62	8.32	-0.21	0.17
PREDICTED: Xenopus (Silurana) tropicalis protein phosphatase 1 regulatory subunit 9B (ppp19b) mRNA	NM_002935559.2	3.59	1.67	-1.11	2.58	2.27	-0.19	0.17
Xenopus tropicalis finished DNA clone Tgg13819	CT025274.2	11.56	5.68	-1.03	3.79	3.34	-0.19	0.18
Xenopus laevis Similar to hypothetical protein FLJ3687 clone IMAGE:5569934 mRNA	BC03838.1	367.03	86.87	-2.08	12.02	9.23	-0.39	0.19
PREDICTED: Xenopus (Silurana) tropicalis alysulfatase A (arsa) mRNA	NM_00293829.1	1.64	0.41	-2.01	1.34	1.01	-0.41	0.21
PREDICTED: Xenopus (Silurana) tropicalis chloride channel voltage-sensitive 2 (clcn2) transcript variant X1 mRNA	NM_002935462.2	35.63	13.51	-1.4	4.08	3.33	-0.3	0.22
Xenopus laevis histamine N-methyltransferase (hmtc-a) mRNA	NM_00109508.1	1.45	0.47	-1.65	6.38	5.01	-0.35	0.22
PREDICTED: Xenopus (Silurana) tropicalis regulatory subunit 12 (med12) mRNA	NM_002934903.2	67.63	17.01	-2	18.81	13.77	-0.45	0.23
PREDICTED: Xenopus (Silurana) tropicalis plectin B3 (pknb3) mRNA	NM_002935494.2	142.87	68.21	-1.07	106.77	90.18	-0.25	0.23
Xenopus laevis Rsk-2 mRNA (cDNA clone MGCB82193) (IMAGE:3401402) complete cds	BC080017.1	260.43	126.85	-1.04	67.44	56.67	-0.26	0.25
Xenopus tropicalis finished DNA clone Tgg0954d21	CR942489.2	47.02	21.7	-1.12	1.13	0.93	-0.28	0.25

Gene	Accession	Eya1 CHX	Eya1 CHX+DEX <sup>2</sup>	FC Eya1 <sup>3</sup>	Control CHX	Control CHX+DEX	FC Control <sup>4</sup>	Rank <sup>5</sup>
PREDICTED: Xenopus (Silurana) tropicalis ALX homeobox 4 (aka4) transcript variant X2 mRNA	XM_004913349.1	4.23	1.24	-1.78	1.07	0.78	-0.46	0.26
PREDICTED: Xenopus (Silurana) tropicalis Fraser syndrome 1 (fas1) mRNA	XM_002935451.2	68.62	19.21	-1.84	7.54	5.39	-0.49	0.27
Xenopus laevis cd42 binding protein kinase beta (DMPK-like) (cd42bpb) mRNA	NM_001171955.2	53.73	25.1	-1.1	14.08	11.43	-0.31	0.28
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101732750 (LOC101732750) mRNA	XM_004914348.1	32.06	13.94	-1.21	8.11	6.42	-0.34	0.29
PREDICTED: Xenopus (Silurana) tropicalis transmembrane domain containing 14 (krtd14) mRNA	XM_002934654.2	17.64	3.81	-2.22	2.73	1.77	-0.64	0.29
PREDICTED: Xenopus (Silurana) tropicalis sodium channel protein gene 1 (scn1) mRNA	XM_002935077.2	1.68	0.68	-1.31	2.24	1.73	-0.38	0.29
PREDICTED: Xenopus (Silurana) tropicalis cadherin EGF LAG seven-pass G-type receptor 3 (cefs3) mRNA	AB727591.1	1.1	0.51	-1.13	1.02	0.8	-0.36	0.32
PREDICTED: Xenopus (Silurana) tropicalis urtensin I related peptide precursor complete cds	XM_004914172.1	12.64	5.9	-1.1	4.73	3.69	-0.36	0.33
Xenopus laevis SH3 and multiple ankyrin repeat domains 2 (shank2) mRNA	NM_00109539.1	4.08	1.94	-1.08	2.06	1.58	-0.39	0.36
PREDICTED: Xenopus (Silurana) tropicalis cystatin-A-like (LOC101734653) transcript variant X1 mRNA	XM_004912278.1	1.9	0.8	-1.26	1.46	1.06	-0.47	0.37
Xenopus laevis cDNA clone IMAGE7012129	BC080836.1	54.17	18.6	-1.55	28.26	18.6	-0.61	0.4
Xenopus laevis cholinergic receptor nicotinic alpha 1 (muscle) gene 2 (chma1.2) mRNA	NM_00109240.1	1.28	0.54	-1.25	1.51	1.07	-0.5	0.4
PREDICTED: Xenopus (Silurana) tropicalis argonaute RISC catalytic component 4 (ago4) mRNA	NM_002936214.2	2.24	0.92	-1.29	1.97	1.38	-0.53	0.41
Xenopus laevis afadin mRNA complete cds	GU290317.1	1106.77	382.21	-1.54	122.43	76.55	-0.68	0.45
PREDICTED: Xenopus (Silurana) tropicalis STE20/SPS1-related proline-alanine-rich protein kinase-like (LOC100488948) mRNA	XM_002936751.2	1.88	0.76	-1.32	1.07	0.71	-0.6	0.46
PREDICTED: Xenopus (Silurana) tropicalis CASP8 and FADD-like apoptosis regulator (cifar) transcript variant X2 mRNA	XM_004917781.1	1.78	0.81	-1.16	1.3	0.9	-0.33	0.46
Xenopus (Silurana) tropicalis SH3 and cysteine rich domain 3 (sac3) mRNA	NM_001007506.1	130.42	58.85	-1.15	64.2	44.1	-0.55	0.48
Xenopus laevis peroxisome proliferator activated receptor gamma mRNA (cDNA clone MGC:58675 IMAGE:4032975) complete cds	BC050474.1	161.15	7.7	-1.07	19.06	13.07	-0.55	0.51
Xenopus laevis extended synaptotagmin-like protein 2 (esyt2-a) mRNA	NM_001095791.1	119.19	32.83	-1.87	41.75	20.89	-1	0.54

<sup>1</sup> Expression levels (FPKM) in pre-placodal explants after injection of Eya1 and CHX treatment

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Eya1 and CHX+DEX treatment

<sup>3</sup> Log<sub>2</sub> Fold change values for Eya1

<sup>4</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup> Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup> Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## E.5 Six1+Eya1; Up-regulated genes

**Table E.5** Genes with at least two-fold up-regulation after injection of Six1+GR+Eya1-GR and treatment with CHX + DEX

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DEX <sup>2</sup>	FC Six1+Eya1 <sup>3</sup>	FC Control CHX	FC Control CHX+DEX <sup>4</sup>	Rank <sup>5</sup>
PREDICTED: Xenopus (Silurana) tropicalis parvalbumin thymic CPV3-like (LOC100485867) transcript variant X1 mRNA	XM_002931951.1	0.01	9.49	12.22	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zona pellucida-like domain-containing protein 1-like (LOC101732051) mRNA	XM_004916514.1	0.01	4.72	11.21	0	0	0
Xenopus laevis cDNA clone IMAGE:688110 partial cds	BC054974.1	0.01	2.11	11.04	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 255B (tmem255b) transcript variant X2 mRNA	XM_004911801.1	0.01	3.91	10.94	0	0	0
Xenopus laevis cDNA clone MGCI:310958 (IMAGE:794381) complete cds	BC106383.1	0.01	1.59	10.63	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734650 (LOC101734650) mRNA	XM_004913366.1	0.01	1.33	10.38	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis floculation protein FL011-like (LOC101732963) mRNA	XM_004919443.1	0.01	1.28	10.32	0	0	0
Xenopus laevis parvalbumin gene 1 (pvab1.1) mRNA	NM_001112856.1	0.01	1.21	10.24	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis oocyte zinc finger protein XlCOF8.4-like (LOC100496403) transcript variant X1 mRNA	XM_002924241.0	0.01	1.15	10.17	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis traf2 and NCK-interacting protein kinase-like (LOC101730922) mRNA	XM_004912405.1	0.01	1.14	10.15	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis pyruvate dehydrogenase kinase isozyme 1 (pdik1) mRNA	XM_0029234618.2	0.01	1.03	10	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 1 (tgzf1) mRNA	XM_004918651.1	0.01	1.01	9.97	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis integrin alpha 7 (itga7) transcript variant X2 mRNA	XM_002936351.1	0.01	1.45	9.51	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis clain 2 (cln2) mRNA	XM_004918679.1	0.01	2.02	9.39	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis leucine-rich repeat-containing protein 3B-like (LOC100492739) t	NM_001093369.1	5.27	2153.19	8.68	0	0	0
Xenopus laevis storkhead box 2 (stox2) mRNA	BC127415.1	0.02	3.18	8.04	0	0	0
Xenopus laevis cDNA clone IMAGE:514689 partial cds	XM_002938866.2	0.02	2.85	7.88	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C14orf166B (LOC100490780) mRNA	XM_0029236334.2	0.02	3.67	7.83	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis cartilage intermediate layer protein 2 (clip2) mRNA	XM_002943394.2	0.03	4.1	7.53	0	0	0
Xenopus tropicalis cDNA clone IMAGE:100488329 mRNA	BC094950.1	0.06	9.13	7.5	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis stereocilin (strc) mRNA	XM_00293262.2	0.03	3.93	7.36	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis neurofilament medium polypeptide-like (LOC100497735) mRNA	XM_002933777.2	0.02	1.8	6.96	0	0	0
Xenopus laevis uncharacterized LOC10036938 (LOC10036938) mRNA	NM_001097708.1	0.15	10.85	6.27	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis ATP-sensitive inward rectifier potassium channel 11-like (LOC100492679) mRNA	XM_004919278.1	0.05	2.78	6.06	0	0	0
Xenopus laevis similar to myosin heavy polypeptide 4 skeletal muscle mRNA (cDNA clone IMAGE:4681347) partial cds	NM_001102275.1	0.59	33.82	5.86	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7022272	BC041716.1	0.03	1.29	5.82	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis neurofilament medium polypeptide-like (LOC100497735) mRNA	BC070615.1	0.13	5.5	5.52	0	0	0
Xenopus laevis finished cDNA clone Tga00518 (Tga00518)	CR761455.2	0.12	4.7	5.37	0	0	0
Xenopus laevis fibrinogen-like 1 (fgf1) mRNA	NM_001092837.1	0.05	1.7	5.23	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis NFkB inhibitor interacting Ras-like 2 (inkras2) transcript variant X3 mRNA	XM_004913708.1	0.07	2.49	5.21	0	0	0
Xenopus laevis cyclin Dx (cdnx) mRNA	NP_001083887.1	0.46	16.55	5.19	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis dimethylamine monooxygenase [N-oxide-forming] 2-like (LOC100492603)	XM_002934166.2	0.16	5.58	5.19	0	0	0
Xenopus laevis sorting nexin 31 (snx31) mRNA	XM_002935561.2	0.04	1.07	5.11	0	0	0
Xenopus (Silurana) tropicalis plasin 1 (pls1) mRNA	NM_001132361.1	0.04	1.26	5.07	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis voltage-gated channel shaker-related subfamily member 2 (kcn2a) mRNA	NM_00491036.1	0.08	2.3	4.89	0	0	0
Xenopus laevis butyrophilin subfamily 1 member A1 (bnf2a1) mRNA	NM_00113216.1	0.07	1.8	4.87	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100494619 (LOC100494619) mRNA	XM_002940483.2	0.09	2.4	4.8	0	0	0
Xenopus tropicalis finished cDNA clone ThdA039p05	CU075479.1	1.15	31.22	4.78	0	0	0
Xenopus laevis mRNA for Xoxx17alpha protein	AJ001730.1	0.64	17.33	4.78	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis STAMP domain-containing protein 2-like (LOC100497585) mRNA	XM_002935263.2	0.07	1.87	4.75	0	0	0
Xenopus laevis ectodysplasin A receptor (edar) mRNA	NM_001083471.1	0.35	8.7	4.67	0	0	0
Xenopus (Silurana) tropicalis enoyl-CoA delta isomerase 1 (ecdl) nuclear gene encoding mitochondrial protein mRNA	NM_001012129.1	0.05	1.22	4.67	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731096 (LOC101731096) mRNA	NM_004911510.1	4.63	110.38	4.58	60.22	8.76	-2.79
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100491937 (LOC100491937) mRNA	NM_002933961.2	0.09	1.89	4.55	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis potassium voltage-gated channel shaker-related subfamily beta member 2 (kcnab2)	NM_004916188.1	0.05	1.06	4.48	0	0	0

Gene	Accession	Six1+Eva1 CHK	Six1+Eva1 CHK+DXR <sup>a</sup>	FC Six1+Eva1 CHK	FC Six1+Eva1 CHK+DXR <sup>a</sup>	Control CHK	Control CHK+DXR <sup>a</sup>	FC Control CHK	FC Control CHK+DXR <sup>a</sup>	Rank <sup>b</sup>
Xenopus laevis cDNA clone IMAGE:5.51.4733; partial cds	BC161680_1	0.11	2.25	4.44	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis ornithine decarboxylase-like (LOC100498433) mRNA	XM_002940019_2	0.07	1.48	4.42	0	0	0	0	0	0
Xenopus (Silurana) tropicalis myosin heavy chain 4 skeletal muscle (myh4) mRNA	NM_001001244_1	0.38	7.56	4.34	0	0	0	0	0	0
Xenopus (Silurana) tropicalis glutaminase peroxidase 2 (gpx2) mRNA	NM_001253151_1	1.46	29.46	4.34	3.02	2.57	-0.24	0	0	0
Xenopus (Silurana) tropicalis uncharacterized LOC100145323 (LOC100145323) mRNA	NM_001125798_1	0.09	1.8	4.34	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis thrombospondin type-1 domain-containing protein 4-like (LOC101733981)	XM_004919735_1	0.06	1.03	4.3	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis solute carrier family 4 sodium bicarbonate cotransporter member 4 (slc4a4)	XM_004919134_1	0.18	3.46	4.3	0	0	0	0	0	0
Xenopus (Silurana) tropicalis cytochrome B450 family 26 subfamily 8A member 1 (cyb26d1) mRNA	NM_001079187_2	0.53	10.15	4.28	3.19	1.81	-0.82	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis interleukin-8-like (LOC100498334) mRNA	XM_002942530_2	0.06	1.13	4.27	0	0	0	0	0	0
Xenopus laevis mRNA for xSox17alpha2 complete cds	NM_001092397_1	0.21	3.85	4.21	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis metal-like (LOC100485014) mRNA	XM_002935804_2	0.13	2.24	4.18	0	0	0	0	0	0
Xenopus (Silurana) tropicalis chemokine (C-X-C motif) receptor 7 (cxcr7) mRNA	NM_001030434_1	0.43	7.01	4.03	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis ankyrin repeat and SOCS box-containing 2 (asb2) transcript variant X3 mRNA	XM_004917073_1	0.09	1.39	4.03	0	0	0	0	0	0
Xenopus laevis apelin (apelin) mRNA	NM_001097924_1	0.08	1.15	4.01	0	0	0	0	0	0
Xenopus laevis mRNA for xSox17alpha2 complete cds	AB052691_1	0.65	10.06	3.96	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis espin (espn) transcript variant X3 mRNA	XM_004916193_1	0.49	7.42	3.94	0	0	0	0	0	0
Xenopus laevis MGCB1526 protein (MGCB1526) mRNA	NM_001093266_1	0.14	2.03	3.93	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis B-cell CLL/symphoma 11B (zinc finger protein) (bcl11b) transcript variant X2 mRNA	NM_001093267_1	0.15	2.24	3.93	0	0	0	0	0	0
Xenopus laevis hedgehog-interacting protein mRNA complete cds	AY328923_1	0.23	3.37	3.91	0	0	0	0	0	0
Xenopus laevis sodium solute transporter Vho-2 (Vto) mRNA complete cds; alternatively spliced	AY260729_1	0.13	1.89	3.9	0	0	0	0	0	0
Xenopus tropicalis tyrosine kinase 1 (tk1) mRNA clone IMAGE:8944/08) partial cds	BC161731_1	0.14	2.02	3.9	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis domain-containing protein Spartan-like (LOC100485137) mRNA	XM_002942357_2	0.21	3.03	3.89	7.3	6.73	-0.12	0	0	0
Xenopus laevis RDC1 like protein mRNA (cDNA clone MGCB114801 IMAGE:4632706) complete cds	BC098974_1	0.56	7.89	3.84	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis baculoviral IAP repeat-containing protein 1-like (LOC100490991) mRNA	XM_002934832_2	0.16	2.14	3.76	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TIP1008n14	CR848248_2	0.09	1.1	3.74	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis glutamate receptor ionotropic N-methyl D-aspartate 2C (grin2c)	XM_004919478_1	0.11	3.71	3.71	408.64	3.71	9.31	7.66	-0.29	0
Xenopus laevis mRNA for frizzled 4 protein (fzd4 gene)	AJ251750_1	31.3	2.69	3.66	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis proline-rich transmembrane protein 3 (prt3) transcript variant X1 mRNA	XM_00491103_1	0.11	1.34	3.71	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis tyrosine-protein kinase transforming protein Fps-like (LOC100489191) mRNA	XM_002932240_2	0.09	1.08	3.68	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis espin (espn) transcript variant X1 mRNA	XM_002933856_2	0.15	1.87	3.67	0	0	0	0	0	0
Xenopus laevis kelch-like family member 30 (khh30) mRNA	NM_00109724_1	0.19	2.3	3.67	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis SLT and NTRK-like family member 5 (sltr5) transcript variant X1 mRNA	XM_002940106_2	0.22	2.69	3.66	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis tyrosine-protein phosphatase non-receptor type substrate 1-like (LOC101733531) mRNA	XM_004913397_1	5.48	68.31	3.65	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis paired box 1 (pax1) transcript variant X1 mRNA	XM_002932947_2	0.22	2.63	3.63	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis calcineurin homoeostasis modulator protein 2-like (LOC101735003) partial mRNA	XM_004920484_1	0.1	1.19	3.62	0	0	0	0	0	0
Xenopus (Silurana) tropicalis synaptic Ras GTPase activating protein 1 (syngap1) mRNA	NM_001134816_1	0.11	1.3	3.58	0	0	0	0	0	0
Xenopus (Silurana) tropicalis glyoxalase 1 (glu1) mRNA	NM_001030374_1	0.1	1.08	3.57	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis G-protein-coupled receptor 84 (gpr84) mRNA	NM_001078867_1	0.27	3.09	3.53	0	0	0	0	0	0
Xenopus laevis pancreatic lipase-related protein 1 (plipnp1) mRNA	NM_001094231_1	25.12	284.23	3.51	66.79	66.87	0.01	0	0	0
Xenopus laevis uncharacterized protein MGCB2269 (MGCB2269) mRNA	NM_001091363_1	0.14	1.58	3.5	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone Tga071e17	CR926377_2	0.3	3.37	3.5	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis membrane-associated ring finger (C3HC4) 4/3 ubiquitin protein ligase (march4) mRNA	XM_002940052_2	3.47	38.78	3.49	52.56	42.01	-0.33	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis carinoembryonic antigen-related cell adhesion molecule 3-like (LOC101734405) mRNA	XM_002932442_2	0.23	2.51	3.48	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101734405 (LOC101734405) mRNA	XM_00491247_1	0.45	4.97	3.48	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis endothelin 2 (edn2) mRNA	XM_00293354_2	0.15	1.59	3.46	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis calmodulin-dependent protein kinase kinase 2 beta (cankk2)	XM_00491631_1	4.05	44.01	3.45	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis calcium/calmodulin-dependent cell adhesion molecule 3-like (canakk2)	XM_00293701_2	0.62	6.69	3.45	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis transcriptional coactivator p300 (p300)	XM_004913389_1	0.21	2.18	3.44	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis phosphotyrosine protein kinase 10-like (LOC100490785) mRNA	XM_002940578_2	0.59	6.34	3.44	0	0	0	0	0	0
Xenopus laevis myosin V (Myo5) mRNA	JX494698_1	0.12	1.27	3.43	2.27	1.95	-0.23	0	0	0
Xenopus tropicalis sine oculis binding protein homolog (Drosophila) mRNA (cDNA clone MGC:172604 IMAGE:7656239)	BC154687_1	0.62	6.44	3.4	0	0	0	0	0	0

Gene	Accession	Six1+Eya1 CHK	Six1+Eya1 CHK+DXR <sup>a</sup>	FC Six1+Eya1 CHK	FC Six1+Eya1 CHK+DXR <sup>a</sup>	Control CHK	Control CHK+DXR <sup>a</sup>	FC Control CHK	FC Control CHK+DXR <sup>a</sup>	Rank <sup>b</sup>
PREDICTED; Xenopus (Silurana) tropicalis kinesin family member 3C (kif3c) transcript variant X1 mRNA	XM_002942110.2	0.28	2.87	3.39	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis andocatin 2 (aho2) mRNA	XM_002932297.2	0.17	1.64	3.33	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis sense/threonine-protein kinase N2-like (LOC100492906) mRNA	XM_002940991.2	0.15	1.47	3.32	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis tankyrase-1-like (LOC100498424) mRNA	XM_00293393.2	0.16	1.49	3.3	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TNeu089c16	CR760308.2	3.42	33.23	3.29	0	0	0	0	0	0
Xenopus laevis natriuretic peptide C (nppc) mRNA	NM_001093118.1	0.37	3.45	3.24	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101734952 (LOC101734952) mRNA	XM_004916172.1	0.13	1.2	3.23	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone Tlp0409c08	CR926447.1	0.23	2.04	3.21	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis poly (ADP-ribose) polymerase 14-like (LOC101731378) mRNA	XM_004920062.1	0.19	1.73	3.21	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE7394277	BC106376.1	0.18	1.62	3.21	0	0	0	0	0	0
XERGAOP3.X.laevis oocyte 5s rRNA pseudogene clone pXo3.1	K01368.1	0.94	8.65	3.21	1.55	1.34	-0.22	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis regulatory factor X-associated protein (rkcap) mRNA	XM_002939581.2	23.54	215.9	3.2	1.78	1.66	-0.1	0	0	0
Xenopus (Silurana) tropicalis APC membrane recruitment protein 2 (amer2) mRNA	NM_001275500.1	0.12	1.09	3.19	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101735004 (LOC101735004) mRNA	XM_004913048.1	97.28	884.32	3.19	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis tyrosine kinase (ibtk)	XM_00491523.1	32.4	294.1	3.19	65.26	30.08	-1.12	0	0	0
Xenopus laevis cDNA clone IMAGE5049275 containing frame-shift errors	BC077506.1	0.19	1.68	3.19	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis espin (espn) transcript variant X2 mRNA	XM_002946653.2	0.14	1.4	3.13	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis membrane-associated-like (LOC100492698) mRNA	XM_004911404.1	0.2	1.67	3.09	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C14orf164 (LOC101735053) mRNA	BC161435.1	0.14	1.11	3.07	0	0	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE8850885	XM_002944797.2	0.54	4.33	3.03	0	0	0	0	0	0
Xenopus (Silurana) tropicalis F-box protein 32 (fbx32) transcript variant X1 mRNA	NM_001129441.1	0.21	1.67	3.02	0	0	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE7609381	BC135338.1	0.17	1.33	3.01	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101734178 (LOC101734178) mRNA	XM_00491385.1	0.15	1.18	2.99	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C14orf164 (LOC101735053) mRNA	XM_004911404.1	0.2	1.67	3.09	0	0	0	0	0	0
Xenopus laevis matrix metalloproteinase 8 (neutrophil collagenase) (mmp8) mRNA	BC161435.1	0.14	1.11	3.07	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis transforming growth factor beta-induced 68kDa (tgfb1) mRNA	XM_001095238.1	0.27	2.1	2.98	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis outer dense fiber of sperm tails 3-like 2 (odf3l2) mRNA	XM_002939767.2	0.65	5.01	2.97	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) calcitonin gene-related peptide-like (LOC100487527) mRNA	XM_002941675.2	0.22	1.63	2.95	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis furin (paired basic amino acid cleaving enzyme) (furin) transcript variant X1 mRNA	XM_002932341.2	0.16	1.16	2.93	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) transforming growth factor beta-induced 68kDa (tgfb1) mRNA	XM_001095034.1	0.17	1.29	2.99	0	0	0	0	0	0
Xenopus laevis transforming growth factor beta-induced 68kDa (tgfb1) mRNA	NM_00110956.1	0.26	2.1	2.98	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis receptor-type tyrosine-protein phosphatase eta-like (LOC101733351) mRNA	XM_00491833.1	0.42	3.01	2.86	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis microtubule-associated protein-like 1-like (LOC100495318) mRNA	XM_002936583.2	0.32	2.18	2.8	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) leucine rich repeat containing S2 (lrfc52) mRNA	XM_00293373.2	0.72	4.9	2.79	0	0	0	0	0	0
Xenopus laevis uncharacterized protein MGc68450 (MGc68450) mRNA	NM_00108841.1	0.46	3.04	2.76	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis cubilin (intrinsic factor-cobalamin receptor) (cubn) mRNA	XM_002939005.2	0.17	1.09	2.75	0	0	0	0	0	0
Xenopus (Silurana) tropicalis potassium inwardly-rectifying channel subfamily J member 8 (kcnj8) mRNA	NM_00110855.1	0.31	2.05	2.74	0	0	0	0	0	0
Xenopus tropicalis cDNA clone Gas018h01	CR835799.2	0.46	2.91	2.67	0	0	0	0	0	0
Xenopus laevis neuregulin alpha-1 mRNA complete cds	XM_004910401.1	0.57	3.61	2.67	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis potassium voltage-gated channel sk-related family member 1 (kcnel1)	NM_001112949.1	0.46	2.85	2.65	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis open reading frame human C1orf233 (LOC101733145) mRNA	NM_001096768.1	24.12	148.35	2.63	22.76	21.11	-0.11	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis oyster binding protein-like 1A (osbp1a) mRNA	XM_004916491.1	0.19	1.12	2.6	0	0	0	0	0	0
Xenopus laevis zinc finger protein 16 (znf16) mRNA	AF076618.1	18.71	120.5	2.69	33.39	25.96	-0.37	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC779603 (LOC779603) transcript variant X1 mRNA	XM_004912135.1	0.81	5.14	2.69	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis open reading frame human C1orf233 (LOC101733145) mRNA	XM_00294220.1	0.54	3.39	2.67	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis finished cDNA clone (Gas058c12	XM_002934726.1	0.26	1.03	2.71	0	0	0	0	0	0
Xenopus (Silurana) tropicalis alpha-1 (osbp1a) mRNA	NM_004912026.1	0.26	1.56	2.6	0	0	0	0	0	0
Xenopus laevis zinc finger protein 16 (znf16) mRNA	NM_002934799.1	1.56	9.28	2.59	1.91	1.25	-0.62	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis copine II (cpne2) transcript variant X1 mRNA	BC106693.1	0.58	3.42	2.57	0	0	0	0	0	0
Xenopus laevis hypothetical protein MGc132374 mRNA clone MGc132374 (IMAGE3396440) complete cds	XM_004914057.1	1.44	8.45	2.56	0	0	0	0	0	0

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DEX*	FC Six1+Eya1 CHX+DEX*	Control CHX	Control CHX+DEX	FC Control CHX+DEX Control*	Rank*
PREDICTED; Xenopus (Silurana) tropicalis GRB2-associated binding protein 1 (gab1) transcript variant 1 mRNA	XM_002940078.2	0.21	1.2	2.56	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis shisa homolog 2 (Xenopus laevis) (shisa2) mRNA	XM_002940554.2	1.45	8.48	2.56	0	0	0	0
Xenopus (Silurana) tropicalis SH3 domain containing 19 (sh3d19) transcript variant X2 mRNA	XM_00491139.1	0.42	2.41	2.54	0	0	0	0
Xenopus (Silurana) tropicalis 5'-nucleotidase domain containing 3 (nt5dc3) mRNA	NM_00102844.1	0.43	2.45	2.54	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis nucleotide-binding protein 1 (LOC100488874) transcript variant X1 mRNA	XM_00293153.2	0.18	1.04	2.54	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis 3'-nucleotidase domain-containing 1 LOC100488874 (LOC100488874) transcript variant X1 mRNA	XM_004916310.1	0.44	2.55	2.54	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis lym domain-binding protein 3-like (LOC101736564) transcript variant X1 mRNA	XM_002935866.2	0.31	1.75	2.53	0	0	0	0
Xenopus tropicalis finished cDNA clone TNeu121p10	CR8-48378.2	1.21	6.9	2.52	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis 3-O-sulfotransferase 3A1-like (LOC100488739) mRNA	XM_004913335.1	304.26	1734.55	2.52	18.36	17.05	-0.11	0
PREDICTED; Xenopus (Silurana) tropicalis immunoglobulin superfamily member 10 (igsf10) mRNA	XM_0029241106.2	0.26	1.47	2.51	0	0	0	0
Xenopus laevis cDNA clone IMAGE7764223	BC106227.1	0.22	1.2	2.49	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis histone H1B-like (LOC101734349) mRNA	XM_00491999.1	54.13	302.41	2.49	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis monocyte to macrophage differentiation-associated (mmnd) transcript variant X2 mRNA	XM_004918560.1	1.4	7.72	2.47	2.24	1.36	-0.72	0
Xenopus (Silurana) tropicalis transforming growth factor beta-induced 68kDa (tgfb1) mRNA	NM_001073861.1	0.28	1.54	2.46	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis protein kinase C delta type-like (LOC101731320) mRNA	XM_004913086.1	0.98	5.31	2.45	0	0	0	0
Xenopus laevis transient receptor potential cation channel subfamily M member 8b (TRPM8b) mRNA complete cds	FJ948759.1	0.29	1.58	2.45	0	0	0	0
Xenopus (Silurana) tropicalis endothelial cell adhesion molecule (esam) mRNA	NM_001142053.1	0.38	2.03	2.45	0	0	0	0
Xenopus laevis uncharacterized LOC100336523 (LOC100365933) mRNA	NM_00107704.1	0.66	3.53	2.43	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C2orf80 (LOC100489057) mRNA	XM_00293719.2	0.79	4.08	2.39	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis single-minded homolog 1 (Drosophila) (sim1) transcript variant X2 mRNA	XM_00491545.1	0.3	1.51	2.38	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101731841 (LOC101731841) mRNA	XM_004914855.1	2.72	13.87	2.36	0	0	0	0
Xenopus laevis forkhead box D2 (foxD2) mRNA	NM_001094052.1	0.82	4.17	2.36	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC100491349 (LOC100491349) mRNA	XM_004919358.1	0.29	1.44	2.35	0	0	0	0
Xenopus (Silurana) tropicalis endothelin receptor B subtype 2 (ednrB2) mRNA	NM_001079208.1	0.25	1.24	2.33	0	0	0	0
Xenopus tropicalis finished cDNA clone TTb05016	CU025142.1	0.38	1.91	2.33	0	0	0	0
Xenopus (Silurana) tropicalis PY domain containing 1 (pxdc1) mRNA	NM_00113262.1	0.39	1.92	2.33	0	0	0	0
Xenopus tropicalis TRAF and TNF receptor associated protein mRNA (cDNA clone MGCI:184699) IMAGE7661931 complete cds	BC166146.1	0.38	1.87	2.32	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis integrin beta 4 (itgb4) transcript variant X3 mRNA	XM_004918457.1	0.3	1.43	2.3	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis aldehyde dehydrogenase 1 family member L2 (alhd1L2) mRNA	XM_002938070.2	1.08	5.25	2.3	0	0	0	0
Xenopus laevis hypothetical protein LOC432141 mRNA (cDNA clone IMAGE4407523) partial cds	BC072094.1	0.22	1.05	2.29	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis UPF3 regulator of nonsense transcripts homolog A (yeast) (upf3a)	XM_00491795.1	0.35	1.66	2.29	0	0	0	0
Xenopus (Silurana) tropicalis X-linked inhibitor of apoptosis (xiap) mRNA	NM_001030412.1	0.54	2.61	2.28	0	0	0	0
Xenopus (Silurana) tropicalis chloride intracellular channel 5 (clc5) nuclear gene encoding mitochondrial protein mRNA	NM_001007907.1	0.23	1.08	2.28	0	0	0	0
Xenopus laevis hypothetical protein LOC1017335 mRNA (cDNA clone IMAGE582075) partial cds	BC146639.1	0.35	1.66	2.28	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis heatocyclic carcinomatous-associated protein TD26-like (LOC101732836) mRNA	NM_004913895.1	0.43	2.04	2.27	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis UPF3 regulator of nonsense transcripts homolog A (yeast) (upf3a)	XM_00491795.1	0.31	1.47	2.27	0	0	0	0
Xenopus (Silurana) tropicalis 5'-linked inhibitor of apoptosis (xiap) mRNA	NM_002939382.2	4.48	21.39	2.26	2.22	2.08	-0.1	0
Xenopus (Silurana) tropicalis folate receptor 4 (delta) homolog (fol4) mRNA	NM_001174032.1	0.39	1.86	2.26	0	0	0	0
Xenopus laevis GID complex subunit 8 homolog (S. cerevisiae) (gid8) mRNA	NM_001095765.1	57.59	273.28	2.25	20.52	17.49	-0.24	0
PREDICTED; Xenopus (Silurana) tropicalis differentiation LOC100490228 (LOC100490228) mRNA	NM_002949332.2	0.65	3.07	2.25	12.15	6.67	-0.87	0
PREDICTED; Xenopus (Silurana) tropicalis fibillin-3-like (LOC100488202) transcript variant X1 mRNA	NM_002937657.2	1.59	7.48	2.25	0	0	0	0
Xenopus laevis hypothetical protein LOC733211 mRNA (cDNA clone IMAGE5949404) partial cds	BC094125.1	0.23	1.08	2.25	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis serine/threonine-protein kinase N1-like (LOC101734642) mRNA	NM_00491842.1	0.53	2.49	2.24	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis sphingomyelin cycle protein kinase AB (epha8) partial mRNA	XM_002943929.2	0.26	1.21	2.23	0	0	0	0
Xenopus (Silurana) tropicalis mRNA for alpha 2,3-sialyltransferase ST3Gal V (st3gal5 gene)	FN500108.1	6.69	31.19	2.23	5.83	4.91	-0.25	0
PREDICTED; Xenopus (Silurana) tropicalis HMP-like protein 1-like (LOC100488066) mRNA	NM_002946092.0	0.37	1.72	2.22	0	0	0	0
Xenopus laevis uncharacterized protein MGCG538 (MGCG538) mRNA	NM_001086337.1	0.39	1.79	2.22	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis membrane channel-like protein 7-like (LOC100493700) mRNA	XM_002932222.2	3.04	14.09	2.22	4.07	2.12	-0.95	0
Xenopus (Silurana) tropicalis SREBF chaperone (scap) mRNA	NM_001126690.1	3.09	14.28	2.22	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis kinase insert domain receptor tyrosine kinase (kdr) mRNA	NM_002934669.2	1.07	4.94	2.22	0	0	0	0
Xenopus laevis morphogen mRNA	BC160760.1	0.4	1.84	2.21	0	0	0	0

Gene	Accession	Six1+Eya1 CHK	Six1+Eya1 CHK+DEx <sup>a</sup>	FC Six1+Eya1 CHK	Six1+Eya1 CHK+DEx <sup>a</sup>	FC Six1+Eya1 CHK	Control CHK	Control CHK+DEx <sup>a</sup>	FC Control CHK	Rank <sup>b</sup>
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101730377 (LOC101730377) mRNA	XM_004917426.1	0.46	2.11	2.2	0	0	0	0	0	0
Xenopus laevis cDNA clone MGCE2592 IMAGE4970942 complete cds	BC073007.1	0.29	1.3	2.19	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis regulator of G-protein signaling 3-like (LOC100486204) transcript variant X2 mRNA	XM_004915922.1	0.68	3.08	2.19	0	0	0	0	0	0
Xenopus (Silurana) tropicalis pancreatic lipase-related protein 2 (pliprp2) mRNA	NM_001073963.1	8.55	38.56	2.18	3.07	3	-0.04	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC101734213 (LOC101734213) mRNA	XM_004914869.1	0.67	3.01	2.17	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis protein 2-like (LOC101733103) partial mRNA	XM_004920933.1	0.38	1.71	2.17	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis sense/threonine-protein kinase N1-like (LOC101731840) mRNA	XM_004914268.1	0.34	1.5	2.17	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis uncharacterized LOC100489072 (LOC100489072) mRNA	XM_002924175.2	0.88	3.91	2.16	0	0	0	0	0	0
Xenopus (Silurana) tropicalis midline 1 (Opitz/BBB syndrome) (mid1) mRNA	NM_001006875.1	0.35	1.54	2.16	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis midline 10-like (LOC100496232) mRNA	XM_002923526.2	0.25	1.1	2.15	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis SKI family transcriptional corepressor 2 (skor2) mRNA	XM_002930516.2	0.33	1.43	2.14	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TIB#07948	CU075394.1	0.46	1.96	2.11	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis homolog mitochondrial-like (LOC101734069) transcript variant X1 mRNA	XM_004915745.1	1.98	8.48	2.1	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis frizzled family receptor 4 (frz4) mRNA	XM_002923543.2	37.22	158.62	2.1	20.82	14.32	-0.54	0	0	0
Xenopus (Silurana) tropicalis potassium voltage-gated channel subfamily E member 1 (kcnf1) mRNA	NM_001102926.1	0.44	1.85	2.08	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis RNA-directed DNA polymerase homolog (LOC101730374) partial mRNA	XM_004916122.1	0.34	1.41	2.07	0	0	0	0	0	0
Xenopus laevis tandem C2 domains nuclear (tc2n) mRNA	NM_001095408.1	0.49	2.03	2.07	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis transferrin receptor (p90 CD71) (tfrc) mRNA	XM_002923405.2	0.44	1.82	2.06	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis waprin-Ph1-like (LOC100495457) mRNA	XM_002923513.2	0.7	2.89	2.06	0	0	0	0	0	0
Xenopus laevis dehydrogenase/reductase (SDR family) member 11 (dhns11) mRNA	NM_001094963.1	0.97	4.01	2.05	0	0	0	0	0	0
Xenopus laevis synbun (syntaxin-interacting) (sybu) mRNA	NM_001093953.1	0.28	1.12	2.05	0	0	0	0	0	0
Xenopus laevis uncharacterized LOC100126648 (LOC100126648) mRNA	NM_001110749.1	0.31	1.27	2.04	1.48	1.45	-0.03	0	0	0
Xenopus (Silurana) tropicalis G protein-coupled receptor 176 (gpr176) mRNA	NM_001108264.1	0.26	1.05	2.03	0	0	0	0	0	0
Xenopus laevis secretogranin II (sgc2) mRNA	NM_001088120.1	0.42	1.7	2.03	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis repeat containing protein (bp) mRNA	XM_004915636.1	2.71	11.04	2.15	1.23	-0.82	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis tetrasoricopeptid repeat and ankyrin repeat containing (trknk1) mRNA	XM_002923422.2	0.86	3.5	2.03	0	0	0	0	0	0
Xenopus (Silurana) tropicalis albumin promoter (albumin D-box) binding protein (dbp) mRNA	XM_002923278.2	0.39	1.56	2.03	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis glial cell-line-derived neurotrophic factor-like (LOC100495890) mRNA	XM_004915965.1	0.56	2.24	2.02	0	0	0	0	0	0
XENXSH Xenopus laevis morphogen (sonic hedgehog) mRNA complete cds	EF672722.1	0.58	2.33	2.02	0	0	0	0	0	0
Xenopus (Silurana) laevis transmembrane protein 56 (tmem56-1b) mRNA	L39213.1	0.36	1.42	2.01	0	0	0	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis calcium channel voltage-dependent gamma subunit 7 (cacng7) mRNA	NM_001072981.1	211.99	831.51	2.01	0	0	0	0	0	0
Xenopus (Silurana) laevis PR domain containing 16 (prd16) transcript variant X1 mRNA	XM_002923492.2	0.6	2.4	2.01	0	0	0	0	0	0
Xenopus laevis hypothetical LOC495219 mRNA (cDNA clone IMAGE4174149) partial cds	BC084630.1	0.37	1.45	1.99	1.27	1.13	-0.18	0	0	0
Xenopus laevis CCR4-NOT transcription complex 4 (cntr4) mRNA	NM_0010743.1	20.47	81.01	1.99	18.81	15.97	-0.24	0	0	0
Xenopus laevis gamma-glutamyl hydrolase conjugase polygamma-glutamylutamyl hydrolase (ggth) mRNA	NM_00109691.1	3.95	15.57	1.99	4.24	4.22	-0.01	0	0	0
PREDICTED; Xenopus (Silurana) laevis opsin-3-like (LOC100488877) mRNA	XM_002923523.2	0.79	3.03	1.95	1.33	1.23	-0.12	0	0	0
Xenopus laevis transmembrane protein 26 (tmem56-1) mRNA	NM_00108447.1	70.16	268.07	1.94	32.63	29	-0.17	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis monocyte to macrophage differentiation-associated (mmnd) transcript variant X1 mRNA	XM_002924161.2	1.92	7.14	1.9	1.27	1.13	-0.18	0	0	0
Xenopus laevis SH3 domain ankyrin repeat and PH domain 3 (asap3) mRNA	NM_001087303.2	0.5	1.84	1.89	1.83	1.26	-0.54	0	0	0
PREDICTED; Xenopus (Silurana) laevis early growth response 3 (egfr3) mRNA	XM_002923703.2	1.21	4.32	1.85	1.9	1.36	-0.49	0	0	0
Xenopus (Silurana) laevis homeobox protein Xpf4 mRNA partial cds	NM_001097307.1	8.95	32.14	1.85	10.09	7.19	-0.49	0	0	0
activin beta A subunit (Xenopus laevis mRNA) 2716 nt]	S61773.1	1.68	5.85	1.8	1.85	1.35	-0.46	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis phospholipid scramblase 1-like (LOC101731311) partial mRNA	NM_004918998.1	76.58	266.43	1.8	31.63	27.22	-0.22	0	0	0
Xenopus (Silurana) laevis ArfGAP with SH3 domain ankyrin repeat and PH domain 3 (putative) (ub3) mRNA	XM_0029237811.2	4.86	16.8	1.8	2.81	2.39	-0.23	0	0	0
XLHOX7A Xenopus laevis homeobox protein Xpf4-2 mRNA partial cds	U04867.1	11.35	499.72	1.78	56.21	51.98	-0.12	0	0	0
Xenopus laevis hypoxia-inducible factor 1 alpha (hif-1α) mRNA	NM_00109257.1	349.83	1143.15	1.71	432.51	148.4	-1.55	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis sorting nexin 24 (snx24) mRNA	NM_001016125.3	0.98	3.19	1.71	1.33	1.32	-0.02	0	0	0
Xenopus laevis ribosomal protein S2e mRNA (cdNA clone MGCI60657) IMAGE6318125 complete cds	BC130122.1	0.6	1.95	1.7	5.59	4.71	-0.25	0	0	0
PREDICTED; Xenopus (Silurana) tropicalis alpha-tectorin-like (LOC100490936) rRNA	NM_00293023.2	1.43	4.57	1.69	3.52	3.08	-0.2	0	0	0
Xenopus tropicalis finished cDNA clone TNed065ii3	CU025050.1	4.07	12.98	1.68	1.86	1.55	-0.27	0	0	0

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DIX*	FC	Control CHX	Control CHX+DIX*	FC	Control CHX	Control CHX+DIX*	Rank*
PREDICTED: Xenopus (Silurana) tropicalis tenascin-X-like (LOC101733293) mRNA	XM_004914832.1	199.98	629.22	1.66	33.06	25.92	-0.36	0		
Xenopus laevis finished cDNA clone Tegg107p05	CR942552.2	5.94	18.51	1.65	11.03	9.94	-0.15	0		
PREDICTED: Xenopus (Silurana) tropicalis potassium voltage-gated channel subfamily KQT member 1-like (LOC100492908) mRNA	XM_0029241385.2	3.37	10.36	1.63	2.55	2.03	-0.34	0		
Xenopus (Silurana) tropicalis low density lipoprotein receptor class A domain containing 3 (ldlr3) mRNA	NM_001102860.1	25.31	77.28	1.62	9.81	7.84	-0.33	0		
PREDICTED: Xenopus (Silurana) tropicalis tetrastricopeptide repeat protein 21B-like (LOC100485767) transcript variant X2 mRNA	XM_00491518.1	0.89	2.69	1.61	15.77	10.73	-0.56	0		
Xenopus (Silurana) tropicalis ral guanine nucleotide dissociation stimulator (ragds) mRNA	NM_001113823.1	12.17	36.78	1.6	10.53	9.4	-0.17	0		
Xenopus (Silurana) tropicalis stanniocalcin 2 mRNA (cDNA clone MGC:145691 IMAGE:7706668) complete cds	BC121338.1	1.84	5.5	1.59	1.9	1.61	-0.25	0		
Xenopus laevis B-cell CLL/lymphoma 6 (bcf6) mRNA	NM_00109569.1	3.66	10.93	1.59	7.22	4.62	-0.65	0		
Xenopus laevis uncharacterized LOC503674 (LOC101730579) mRNA	NM_001095563.1	11.49	34.34	1.58	15.18	11.67	-0.39	0		
PREDICTED: Xenopus (Silurana) tropicalis protein NLRC5-like (LOC101730579) mRNA	XM_00491480.1	4.44	13.08	1.57	6.26	6.08	-0.05	0		
Xenopus laevis adenomatous polyposis coli down-regulated 1 (apcd1) mRNA	NM_001094109.1	4.83	14.02	1.54	6.58	5.29	-0.32	0		
Xenopus laevis EFR3 homolog A (efr3a) mRNA	NM_001094422.1	42.14	121.72	1.54	27.24	19.12	-0.52	0		
PREDICTED: Xenopus (Silurana) tropicalis membrane-bound isoform-like (LOC100496613) mRNA	XM_002933991.2	26.13	73.14	1.49	22.9	18.22	-0.34	0		
Xenopus (Silurana) tropicalis l-fnsg O-fucosylpeptide 3-beta-N-acetylglucosaminyltransferase (lfng) mRNA	NM_00109707.1	6.69	18.67	1.49	5.68	5.16	-0.14	0		
Xenopus (Silurana) tropicalis sema domain transmembrane domain (Tm) and cytoplasmic domain (Tm) (semaphorin) 6D (sema6d)	NM_001126680.1	3.63	10.06	1.48	2.13	2.12	-0.01	0		
Xenopus (Silurana) tropicalis claudin 3 (cldn3) mRNA	NM_001005709.1	0.5	1.38	1.48	458.05	383.04	-0.26	0		
Xenopus (Silurana) tropicalis family with sequence similarity 78 member A (fam78a) mRNA	NM_001127108.1	4.32	14.8	2.01	1.87	1.87	-0.11	0		
Xenopus laevis cDNA clone IMAGE:6317767 partial cds	BC071098.1	2.04	5.61	1.47	3.37	2.87	-0.24	0		
Xenopus laevis tripartite motif containing 36 (trim36) mRNA	NM_001091117.1	8.8	24.2	1.47	5.82	5.59	-0.06	0		
Xenopus laevis zinc finger protein 214 (znf214) mRNA	NM_001097042.1	148.18	404.64	1.45	161.35	146.92	-0.14	0		
PREDICTED: Xenopus (Silurana) tropicalis claudin 3 (cldn3) mRNA	XM_002936508.2	35.21	95.77	1.45	54.03	22.03	-1.3	0		
Xenopus laevis met proto-oncogene (hepatocyte growth factor receptor) (met) mRNA	NM_001085560.1	2.33	6.26	1.44	1.69	1.38	-0.29	0		
Xenopus laevis cdc25 Ba mRNA for cdc25B phosphatase complete cds	AB363840.1	8.82	23.74	1.43	7.43	6.94	-0.1	0		
Xenopus laevis cDNA clone IMAGE:374762.8	BC099044.1	1.52	4.07	1.43	1.92	1.37	-0.5	0		
Xenopus (Silurana) tropicalis suppressor of cytokine signaling 2 (soc2) mRNA	NM_00112426.1	1.69	4.54	1.43	5.22	2.32	-1.18	0		
PREDICTED: Xenopus (Silurana) tropicalis myosin VB (myo5b) mRNA	XM_00492050.1	1.46	3.88	1.42	1.24	1.22	-0.03	0		
PREDICTED: Xenopus (Silurana) tropicalis probable phospholipid-translocating ATPase 11C-like (LOC100495324) partial mRNA	XM_004916950.1	2.69	7.13	1.41	1.54	1.29	-0.26	0		
Xenopus (Silurana) tropicalis ST6 alpha-N-acetyl-neuraminyl-2-3-beta-galactosyl-I-3-N-acetylgalactosaminide alpha	NM_001078839.1	36.89	97.91	1.41	6	3.78	-0.67	0		
PREDICTED: Xenopus (Silurana) tropicalis cAMP responsive element modulator (crem) transcript variant XI mRNA	XM_002935162.2	10.55	27.99	1.41	9.03	7.83	-0.21	0		
PREDICTED: Xenopus (Silurana) tropicalis nuclear receptor coactivator 6 (ncoa6) transcript variant X2 mRNA	XM_004918529.1	30.43	80.61	1.41	8.37	7.31	-0.2	0		
PREDICTED: Xenopus (Silurana) tropicalis Frasier syndrome 1 (fras1) mRNA	XM_00293451.2	17.32	45.81	1.41	7.54	5.39	-0.49	0		
Xenopus laevis cDNA clone IMAGE:69917249	BC158454.1	758798.78	2002747.58	1.41	5219726.2	477202	-0.13	0		
PREDICTED: Xenopus (Silurana) tropicalis tapasin-related protein-like (LOC100488748) transcript variant X2 mRNA	NM_004910522.1	1	2.6	1.4	3.32	2.83	-0.24	0		
Xenopus laevis fucosyltransferase 4 (alpfa1) fucosyltransferase myelin-specific (fu4) mRNA	NM_001172165.1	1.26	3.26	1.38	2.9	1.53	-0.93	0		
PREDICTED: Xenopus (Silurana) tropicalis Kruppel-like factor 1 (klf15) transcript variant X1 mRNA	XM_00293233.2	3.71	9.6	1.38	4.88	4.08	-0.26	0		
Xenopus laevis ets-2a proto-oncogene mRNA (cDNA clone MGCI:60372 IMAGE:854815) complete cds	BC13183.1	9.13	23.42	1.36	11.41	8.58	-0.42	0		
Xenopus laevis hypothetical protein LOC100127276 mRNA (cDNA clone IMAGE:4030363) partial cds	BC153555.1	5.21	13.32	1.36	5.8	3.03	-0.94	0		
PREDICTED: Xenopus (Silurana) tropicalis Sulf-1 gene family member 6 (slrif6) mRNA	NM_004911827.1	4.06	10.4	1.36	6.56	4.23	-0.64	0		
Xenopus laevis nuclear receptor subfamily 2 group F member 6 (nr2f6) mRNA	NM_001087019.1	4.68	11.93	1.36	1.97	1.93	-0.04	0		
PREDICTED: Xenopus (Silurana) tropicalis ADP-ribosylation factor guanine nucleotide-exchange factor 1 (breffelin A-inhibited) (arfgef1)	NM_001094828.1	1.57	3.97	1.35	1.34	1.25	-0.11	0		
PREDICTED: Xenopus (Silurana) tropicalis stromal antigen 1 (stag1) mRNA	NM_004914622.1	4.06	9.99	1.3	3.92	3.87	-0.03	0		
Xenopus laevis biphenyl hydrolase-like serine hydrolase (bpnl) mRNA	NM_001096325.1	0.61	1.49	1.3	2.3	1.25	-0.89	0		
Xenopus laevis nudE nuclear distribution E homolog (A_nudilans)-like 1 (nde1-a) mRNA	NM_001134806.1	45.33	110.97	1.3	1.88	1.12	-0.75	0		
PREDICTED: Xenopus (Silurana) tropicalis camtine O-acylonyltransferase (cot) mRNA	NM_001130376.1	46.05	112.71	1.3	2.62	21.37	-0.32	0		
Xenopus (Silurana) tropicalis G protein-coupled receptor 161 (grpr161) mRNA	NM_00293322.2	23.22	56.8	1.3	31.52	28.15	-0.17	0		
PREDICTED: Xenopus (Silurana) tropicalis ADP-ribosylation factor guanine nucleotide-exchange factor 1 (breffelin A-inhibited) (arfgef1)	NM_001094828.1	1.87	4.56	1.29	32.85	21.45	-0.62	0		
Xenopus laevis clone S1-046-G6 mRNA sequence	AF549892.1	17.26	42.1	1.29	28.19	20.93	-0.43	0		
Xenopus laevis nudE nuclear distribution E homolog (A_nudilans)-like 1 (nde1-a) mRNA	NM_001092863.1	3.27	7.88	1.28	3.21	2.83	-0.19	0		
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 2-like (LOC100491930) mRNA	NM_00293255.2	6.11	14.68	1.27	11.58	6.57	-0.82	0		
Xenopus laevis clone TNeu129d01	CU075707.1	25.01	60.1	1.27	30.12	27.71	-0.13	0		
Xenopus laevis cdc42 effector protein (Rho GTPase binding) 3 (cdc42ep3) mRNA	NM_001095138.1	4.13	9.92	1.27	3.79	2.64	-0.53	0		

Gene	Accession	Six1+Eya1 CHK	Six1+Eya1 CHK+DIX*	FC	Control CHK	Control CHK+DIX*	FC	Control CHK	Control CHK+DIX*	Rank*
PREDICTED: Xenopus (Silurana) tropicalis SKI family transcriptional corepressor 1 (skor1) mRNA	XM_002933958.2	2.05	4.88	1.26	3.31	3.07	-0.11	0	0	
Xenopus laevis histidine ammonia-lyase gene 1 (hal.1) mRNA	NM_001093175.1	51.14	121.9	1.26	72.6	67.29	-0.11	0	0	
Xenopus laevis mab-21-like 1 (mab21l1) mRNA	NM_001091996.1	1.66	3.94	1.25	2.38	2.18	-0.13	0	0	
PREDICTED: Xenopus (Silurana) tropicalis sorting nexin-20-like (LOC104959820) mRNA	XM_00293877.2	3.03	7.19	1.25	15.54	10.22	-0.61	0	0	
PREDICTED: Xenopus (Silurana) tropicalis gastrula zinc finger protein XIC Gf57 -like (LOC101733825) transcript variant X3 mRNA	XM_004915381.1	43.59	103.45	1.25	5.84	2.66	-1.14	0	0	
PREDICTED: Xenopus (Silurana) tropicalis Sand91 protein (sand91) partial mRNA	XM_002945522.2	0.6	1.41	1.25	8.84	8.8	-0.01	0	0	
PREDICTED: Xenopus (Silurana) tropicalis alpha-2D adrenergic receptor-like (LOC00493940) mRNA	XM_004912868.1	0.72	1.7	1.24	1.69	1.43	-0.24	0	0	
Xenopus laevis cDNA clone (Hsp40) homolog subfamily 1 member 18 (dnajc18) mRNA	NM_001102878.1	8.62	20.35	1.24	6.23	6.13	-0.03	0	0	
Xenopus laevis cDNA clone MGCI:181855 IMAGE:83319770 complete cds	BC167562.1	3.18	7.47	1.24	3.54	3.22	-0.14	0	0	
PREDICTED: Xenopus (Silurana) tropicalis c-protein-coupled receptor 37 (endothelin receptor type B-like) (grp37) mRNA	XM_002944831.2	1.17	2.74	1.23	1.78	1.52	-0.23	0	0	
Xenopus (Silurana) tropicalis distal-less homeobox 1 (dlx1) mRNA	NM_00110257.1	0.78	1.8	1.23	1.24	1.04	-0.25	0	0	
Xenopus laevis hypothetical protein MGCI:114768 mRNA clone MGCI:114768 IMAGE:3400761 complete cds	BC100184.1	26.85	62.62	1.23	35.89	30.76	-0.23	0	0	
PREDICTED: Xenopus (Silurana) tropicalis histone-lysine N-methyltransferase p7r-set7-like (LOC101731509) partial mRNA	XM_004920537.1	4.3	9.99	1.22	4.07	2.8	-0.55	0	0	
Xenopus laevis uncharacterized protein MGCI:15231 (MGCI:15231) mRNA	NM_00109191.1	21.85	50.76	1.22	27.55	23.76	-0.22	0	0	
Xenopus tropicalis finished cDNA clone TNed053012	CR855435.2	12.43	28.86	1.22	20.71	15.32	-0.44	0	0	
PREDICTED: Xenopus (Silurana) tropicalis floculation protein FLO11-like (LOC100490389) transcript variant XI mRNA	XM_002945552.2	2.82	6.52	1.22	1.75	1.6	-0.14	0	0	
Xenopus (Silurana) tropicalis leucine zipper putative tumor suppressor 2 (lzs2) mRNA	NM_00106720.1	6.22	14.37	1.21	4.64	4.1	-0.18	0	0	
Xenopus laevis sizzled (szz1) mRNA	NM_00108521.1	5.24	12.11	1.21	4.4	2.3	-0.94	0	0	
PREDICTED: Xenopus (Silurana) tropicalis protein 1-like (LOC100498628) transcript variant XI mRNA	XM_002934358.2	2.04	4.69	1.21	2.03	1.78	-0.19	0	0	
Xenopus laevis Etk4 ETS-domain protein (SRF accessory protein 1) (elk4-b) mRNA	NM_001092639.1	2.27	5.23	1.21	1.16	1.08	-0.11	0	0	
Xenopus laevis cDNA clone MGCI:160982 IMAGE:8320056 complete cds	BC146638.1	14.53	33.26	1.2	14.23	12.87	-0.15	0	0	
Xenopus laevis popeye domain-containing 3 mRNA (cDNA clone MGCI:136149 IMAGE:78223917) complete cds	BC160588.1	1.82	4.14	1.2	1.45	1.44	-0.01	0	0	
PREDICTED: Xenopus (Silurana) tropicalis CLE1 RNA export mediator/homolog (yeast) (gle1) transcript variant XI mRNA	XM_002935501.2	33	74.01	1.17	40.04	36.8	-0.13	0	0	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100497755 (LOC100497755) mRNA	XM_004917587.1	52.6	117.83	1.17	71.77	44.21	-0.7	0	0	
PREDICTED: Xenopus (Silurana) tropicalis SLA family member 5-like (LOC101731813) mRNA	XM_004919313.1	17.12	38.34	1.17	8.78	1.1	-3.01	0	0	
Xenopus (Silurana) tropicalis zinc and ring finger 3 (zrf3) mRNA	NM_001073996.1	2.64	5.89	1.17	3.01	1.98	-0.61	0	0	
PREDICTED: Xenopus (Silurana) tropicalis protein MGCI:6693 mRNA (cDNA clone MGCI:6693 IMAGE:7488) complete cds	BC068970.1	144.83	323.85	1.17	329.36	309.45	-0.09	0	0	
PREDICTED: Xenopus (Silurana) tropicalis C-jun amino-terminal kinase-interacting protein 4-like (LOC100493724) mRNA	XM_00293963.2	4.22	9.4	1.16	3.3	2.86	-0.21	0	0	
PREDICTED: Xenopus (Silurana) calbindin D28k mRNA (cDNA clone MGCI:197268 IMAGE:903927) complete cds	BC170541.1	4.66	10.34	1.16	4.23	3.75	-0.18	0	0	
PREDICTED: Xenopus (Silurana) tropicalis apoptosis peptidase activating factor 1 (lapaf1) mRNA	XM_00293256.2	4.48	9.93	1.16	6.78	3.76	-0.86	0	0	
Xenopus laevis clone IMAGE:46844003 mRNA	BC042305.1	23.83	52.82	1.15	7.39	6.5	-0.19	0	0	
PREDICTED: Xenopus (Silurana) tropicalis sphingosine-1-phosphate phosphatase 1-like (LOC101730648) mRNA	XM_004917200.1	7.49	16.57	1.15	9.18	7.42	-0.31	0	0	
PREDICTED: Xenopus (Silurana) cortical neural cortex related-3 (encr-3) mRNA complete cds	XM_002937195.2	66.82	146.52	1.14	47.7	39.09	-0.29	0	0	
PREDICTED: Xenopus (Silurana) calmodulin D28k mRNA (cDNA clone MGCI:197268 IMAGE:903927) complete cds	XM_002939112.2	14.43	31.63	1.14	45.29	29.11	-0.64	0	0	
Xenopus laevis calmodulin D28k mRNA (cDNA clone MGCI:197268 IMAGE:903927) complete cds	NM_001091365.1	7.93	17.29	1.13	1.37	1.26	-0.13	0	0	
Xenopus laevis clone IMAGE:46844003 mRNA	NM_001091430.1	9.4	20.41	1.12	5.57	5.46	-0.03	0	0	
Xenopus (Silurana) tropicalis neprin (hpn) mRNA	NM_001126697.1	2.11	4.53	1.11	4.9	3.43	-0.52	0	0	
Xenopus laevis ectoderm neural cortex related-3 (encr-3) mRNA complete cds	AY216793.1	47.45	102.62	1.12	55.04	45.63	-0.28	0	0	
PREDICTED: Xenopus (Silurana) tropicalis mastermind-like 1 (Drosophila) transcript variant XI mRNA	XM_002944798.2	28.26	60.37	1.11	18	16.88	-0.1	0	0	
Xenopus laevis calbindin D28k mRNA (cDNA clone MGCI:197268 IMAGE:903927) complete cds	BC080836.1	14.25	30.76	1.12	28.26	18.6	-0.61	0	0	
Xenopus (Silurana) tropicalis uncharacterized LOC100145209 (LOC100145209) mRNA	NM_001126697.1	10.46	22.47	1.11	7.19	6.1	-0.24	0	0	
PREDICTED: Xenopus (Silurana) tropicalis pleckstrin homology domain containing family G (with RhoGef domain) member 1 (plekhhg1)	NM_001128515.1	21.97	47.17	1.11	11.9	11.47	-0.06	0	0	
PREDICTED: Xenopus (Silurana) tropicalis rho GTPase-activating protein 1 (rho1) mRNA	XM_002941204.1	232.64	502.96	1.12	29.16	14.76	-0.99	0	0	
Xenopus laevis Dickkopf-1 (Xdkk-1) mRNA complete cds	AF030434.1	14.75	31.33	1.09	8.9	6.35	-0.49	0	0	
PREDICTED: Xenopus (Silurana) tropicalis rho guanine nucleotide exchange factor (GEF) 3 (argef3) transcript variant XI mRNA	XM_002940292.2	14.72	31.25	1.09	21.32	16.03	-0.42	0	0	
PREDICTED: Xenopus (Silurana) tropicalis pleckstrin homology domain containing family O member 1 (plekhhg1)	XM_002939867.2	7.69	16.3	1.09	8.27	6.17	-0.43	0	0	
Xenopus laevis Rsk-2 mRNA (cDNA clone MGCI:82193 IMAGE:3401402) complete cds	BC080017.1	170.6	360.7	1.09	67.44	56.67	-0.26	0	0	
PREDICTED: Xenopus (Silurana) tropicalis pleckstrin homology domain containing family O member 1 (plekhhg1)	XM_004917337.1	3.87	8.15	1.08	3.3	2.76	-0.26	0	0	
PREDICTED: Xenopus (Silurana) tropicalis prickle homolog 3 (Drosophila) transcript variant XI mRNA	NM_001093583.1	5.22	10.96	1.08	7.2	4.29	-0.75	0	0	
Xenopus laevis von Willebrand factor A domain-containing protein 2 (MGCI:80919) mRNA	NM_001093583.1	8.56	17.76	1.06	8.93	7.58	-0.24	0	0	

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DXR <sup>a</sup>	FC Six1+Eva1 CHX+DXR <sup>a</sup>	Control CHX	Control CHX+DXR <sup>a</sup>	FC Control CHX+DXR <sup>a</sup>	Rank <sup>b</sup>
PREDICTED: Xenopus (Silurana) tropicalis extracellular calcium-sensing receptor-like (LOC100495404) partial mRNA	XM_002944348.2	1.17	2.42	1.06	2.45	2.21	-0.15	0
PREDICTED: Xenopus (Silurana) tropicalis POU domain class 3 transcription factor 3-B-like (LOC100497507) mRNA	NM_004911792.1	1.6	3.31	1.05	6.93	2.73	-1.35	0
Xenopus laevis arrestin beta 2 (arrb2) mRNA	NM_001092112.1	86.97	178.95	1.05	56.09	48.74	-0.21	0
Xenopus laevis sperm-specific transmembrane protein (sns) mRNA	NM_001258422.1	2.65	5.43	1.04	4.55	3.11	-0.56	0
Xenopus laevis uncharacterized LOC733172 (LOC733172) mRNA	NM_001095603.1	2.86	5.85	1.04	2.5	2	-0.33	0
Xenopus laevis LIM class homeodomain protein mRNA (cDNA clone MGCI9961 IMAGE:6865745) complete cds	BC084744.1	2.37	4.85	1.04	3.31	2.6	-0.35	0
Xenopus laevis mRNA for fibronectin leucine-rich transmembrane protein 3 (flt3 gene)	AJ605776.6	6.3	12.9	1.04	7.76	6.48	-0.27	0
Xenopus laevis ADP-ribosylation factor-like 5A (arf5) mRNA	NM_001095436.1	9.43	19.3	1.04	24.55	19.91	-0.31	0
Xenopus laevis hypothetical protein LOC432287 mRNA (cDNA clone IMAGE:4220281) partial cds	BC072256.1	1.61	3.28	1.03	1.09	1.04	-0.07	0
PREDICTED: Xenopus (Silurana) tropicalis rho guanine nucleotide exchange factor (GEF) 38 (arhge138) mRNA	XM_002934776.2	23.98	48.78	1.03	23.77	13.22	-0.85	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100491806 (LOC100491806) mRNA	XM_002942171.2	7.43	15.09	1.03	8.19	4.31	-0.93	0
Xenopus laevis Similar to hypothetical protein FLJ20297 (clone IMAGE:4681271) mRNA	BC044113.1	1.36	2.75	1.02	1.81	1.12	-0.7	0
PREDICTED: Xenopus (Silurana) tropicalis vitamin D (1,25-dihydroxyvitamin D3) receptor (vdrb) mRNA	XM_002935672.2	3.18	6.45	1.02	6.36	2.82	-1.18	0
Xenopus laevis hypothetical protein LOC100037005 mRNA (cDNA clone IMAGE:4958929) partial cds	BC130095.1	21.24	43.01	1.02	9.95	8.8	-0.18	0
Xenopus laevis huntingtin interacting protein 1 related (hip1r) mRNA	NM_001093146.1	7.1	14.35	1.02	15.23	12.47	-0.29	0
Xenopus laevis uncharacterized LOC49630 (LOC49630) mRNA	NM_001095458.1	12.17	24.49	1.01	10.26	7.72	-0.42	0
Xenopus (Silurana) tropicalis abhydrolase domain containing 2 (abhd2) mRNA	NM_001142063.1	6.67	13.36	1.01	6.71	4.71	-0.52	0
PREDICTED: Xenopus (Silurana) tropicales extended synaptotagmin-like protein 1 (esyt1) mRNA	NM_002934533.2	86.99	174.19	1.01	46.94	32.28	-0.55	0
PREDICTED: Xenopus (Silurana) tropicalis lysophosphatidic acid receptor 6-like (LOC100497020) mRNA	NM_004916183.1	1.18	4.68	2	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis ceramide kinase-like (cerk) mRNA	NM_002933015.2	0.4	1.58	1.99	0	0	0.01	0
Xenopus laevis pituitary lactotropin-releasing peptide (lrcp1) mRNA complete cds	AF187871.1	2.38	9.4	1.99	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis arylalkylamine N-acetyltransferase receptor 52D1-like (LOC100487277) mRNA	NM_002943661.1	0.26	1.03	1.99	0	0	0.01	0
XLU1861:Xenopus taeni gene 19 mRNA partial cds	U41861.1	0.3	1.17	1.99	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis LY6/plAUR domain containing 6B (lypd6b) mRNA	NM_002935292.2	7.87	30.99	1.98	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis fibronectin type III domain-containing protein 7-like (LOC1004932589) mRNA	NM_002932171.2	0.43	1.67	1.98	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis leucine rich repeat containing 7 (lrrc7) mRNA	NM_0029419772.0	0.3	1.17	1.97	0	0	0.01	0
Xenopus laevis leucine rich repeat containing 10 (lrrc10) mRNA	NM_001094706.1	0.62	2.38	1.97	0	0	0.01	0
PREDICTED: Xenopus (Silurana) BEN domain containing 7 (bend7) mRNA	NM_00293251.2	0.32	1.24	1.96	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicales DEAD (Asp-Glu-Ala-Asp) box polypeptide 31 (ddx31) mRNA	NM_00293195.2	2.4	9.24	1.95	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicales deoxyribonucleoprotein gamma-like (LOC100497175) transcript variant X1 mRNA	NM_002933836.2	1.34	5.12	1.95	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis olfactory receptor 56B1-like (LOC100497911) mRNA	NM_002943370.2	0.35	1.34	1.95	0	0	0.01	0
Xenopus laevis NK6 transcription factor related locus 2 mRNA (CDNA clone MGC:154471 IMAGE:8317795) complete cds	BC123229.1	0.56	2.13	1.95	0	0	0.01	0
Xenopus laevis SH3 domain containing ring finger 3 (sh3rf3) mRNA	NM_001096464.1	0.52	1.98	1.94	0	0	0.01	0
Xenopus tropicalis finished cDNA clone Theu10816	CT025340.2	1.3	4.92	1.93	0	0	0.01	0
Xenopus (Silurana) tropicalis F-box protein 41 (fibx41) mRNA	NM_001079043.1	0.94	3.55	1.93	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis versican (vcan) transcript variant X1 mRNA	NM_002935704.2	0.52	1.96	1.92	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis metalloproteinase-19 (mmp19) mRNA	NM_002935949.2	0.34	1.27	1.92	0	0	0.01	0
Xenopus tropicalis hypothetical protein LOC10049336 (cDNA clone IMAGE:7672916) partial cds	BC158227.1	0.31	1.16	1.91	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicales elastin-like (LOC1013709293) mRNA	NM_004913983.1	0.31	1.14	1.91	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis zinc finger 1 (zf1) mRNA	NM_002939425.2	0.8	2.96	1.9	0	0	0.01	0
Xenopus laevis fast skeletal troponin C beta mRNA (cDNA clone MGCI5923 IMAGE:4889693) complete cds	BC041495.1	0.38	1.42	1.9	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis SCAN domain-containing protein 3-like (LOC100489631) mRNA	NM_00293613.1	10.59	39.13	1.89	0	0	0.01	0
Xenopus (Silurana) tropicalis cathepsin K (ctsk) mRNA	NM_001078967.1	0.73	2.67	1.89	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 38 member 2 (slc38a2) mRNA	NM_001014056.1	0.64	2.33	1.89	0	0	0.01	0
Xenopus (Silurana) tropicalis zinc finger protein 1 (zf1) mRNA	NM_001126761.1	0.51	1.88	1.88	0	0	0.01	0
Xenopus tropicalis finished cDNA clone TEG077e15	CR761269.2	0.42	1.51	1.88	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis transfrin receptor 1-like (LOC101734813) mRNA	NM_004914949.1	1.1	4.02	1.88	0	0	0.01	0
Xenopus (Silurana) tropicalis phosphoprotein enriched in astrocytes 15 (pea15) mRNA	NM_001126828.1	0.49	1.8	1.88	0	0	0.01	0
PREDICTED: Xenopus (Silurana) tropicalis NCK-interacting protein kinase-like (LOC101732883) mRNA	NM_004913352.1	0.34	1.22	1.88	0	0	0.01	0
Xenopus (Silurana) tropicalis ABC1 member 5 (abc15) mRNA	NM_001142127.1	1.09	3.99	1.88	0	0	0.01	0
Xenopus tropicalis zinc finger protein 64 homolog mRNA (cDNA clone IMAGE:7577708) partial cds	BC159085.1	1.3	4.74	1.87	0	0	0.01	0

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DXR <sup>a</sup>	FC Six1+Eva1 CHX+DXR <sup>a</sup>	Control CHX	Control CHX+DXR <sup>a</sup>	FC Control CHX+DXR <sup>a</sup>	Rank <sup>b</sup>
Xenopus (Silurana) tropicalis uncharacterized LOC100170569 (LOC100170569) mRNA	NM_001130346.2	1.86	6.71	1.86	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis metalloprotease TIK1-like (LOC100491951) mRNA	XM_002936336.2	2.64	9.52	1.86	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis insulinoma-associated protein 2 (insm2) mRNA	XM_002935349.2	0.41	1.46	1.85	0	0	0	0.01
Xenopus laevis Kazal-type serine peptidase inhibitor domain 1 (kazld1) mRNA	NM_001092073.1	0.81	2.9	1.85	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis proteocadherin Fat 2-like (LOC100495486) mRNA	XM_002940209.2	0.32	1.15	1.84	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 26-like (LOC101733991) mRNA	XM_004916528.1	0.49	1.75	1.84	0	0	0	0.01
Xenopus tropicalis finished cDNA clone Thiel124c02	CT030609.1	4	14.28	1.84	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733069 (LOC101733069) mRNA	XM_004917478.1	5.14	18.33	1.84	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734664 (LOC101734664) mRNA	XM_004910525.1	0.79	2.79	1.83	0	0	0	0.01
Xenopus (Silurana) tropicalis calcium channel voltage-dependent beta 4 subunit (cacnb4) mRNA	NM_001142151.1	0.5	1.75	1.83	0	0	0	0.01
Xenopus laevis similar to calquestrin 2 (cardiac muscle) mRNA (cDNA clone MGC:114674 IMAGE:5516132) complete cds	BC097545.1	0.38	1.32	1.83	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TGA502z12	CT030353.1	1.13	3.97	1.82	0	0	0	0.01
Xenopus laevis cytochrome c450 family A polypeptide 4 (cyt450a4) mRNA	NM_001109781.1	0.95	3.32	1.82	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis bradykinin receptor B2 (bkdrb2) transcript variant X2 mRNA	XM_004917087.1	0.44	1.53	1.82	0	0	0	0.01
Xenopus tropicalis Transferin receptor (p90 Cd71) mRNA (cDNA clone IMAGE:7540105) partial cds	BC167509.1	0.33	1.15	1.81	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis LanC antibiotic synthetase component C-like 3 (bacterial) (lanC3) mRNA	XM_002937661.2	0.44	1.51	1.81	0	0	0	0.01
Xenopus laevis (Silurana) tropicalis uncharacterized LOC100485552 (LOC100485552) transcript variant X2 mRNA	XM_00491642.1	0.38	1.3	1.81	0	0	0	0.01
Xenopus tropicalis retinoid X receptor beta mRNA (cDNA clone IMAGE:7686927) complete cds	BC167577.1	0.4	1.39	1.81	0	0	0	0.01
Xenopus laevis P2Y nucleotide receptor mRNA complete cds	AF43254.1	0.72	2.5	1.81	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis hephaestin-like 1 (heph1) mRNA	NM_0012930721.2	0.84	2.87	1.79	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis rho GTPase-activating protein 20-like (LOC100486629) mRNA	XM_002936793.2	0.32	1.09	1.79	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis transforming growth factor beta receptor III (tgfb3) transcript variant X2 mRNA	XM_004913724.1	1.12	3.82	1.78	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis neurobeachin (nbea) transcript variant X2 mRNA	XM_004912032.1	0.75	2.54	1.78	0	0	0	0.01
Xenopus laevis mRNA for brain factor 2	AJ011652.1	1.49	5.02	1.76	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis hypermethylated in cancer 1 (hc1) transcript variant X2 mRNA	XM_00491637.1	0.59	2	1.76	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis retrotransposon-derived protein PEG10-like (LOC101734357) mRNA	XM_00491607.1	107.29	361.09	1.76	0	0	0	0.01
Xenopus laevis uncharacterized LOC100505431 (LOC100505431) mRNA	NM_001197268.1	0.7	2.35	1.75	0	0	0	0.01
Xenopus laevis uncharacterized LOC100127334 (LOC100127334) mRNA	NM_001112888.1	0.38	1.25	1.75	0	0	0	0.01
Xenopus (Silurana) tropicalis mannosyl (alpha1-3)-beta coprotein beta-1-N-acetylglucosaminyltransferase isozyme B (mgat4b)	NM_001102914.1	0.69	2.29	1.75	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis eyes absent homolog 4 (Drosophila) transcript variant X2 mRNA	XM_004914950.1	0.78	2.6	1.74	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis oxytocer binding protein-like 10 (osbp10) mRNA	XM_002941816.2	1.32	4.38	1.74	0	0	0	0.01
Xenopus laevis serine/threonine-protein kinase SRPK1-like (MGC115587) mRNA	NM_001095585.1	0.63	2.08	1.73	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis transcription factor COE3-like (LOC101734223) mRNA	XM_004914891.1	0.35	1.16	1.73	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropical calcium channel voltage-dependent N type alpha 1b subunit (cacna1b) partial mRNA	XM_002942747.2	0.77	2.54	1.73	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TGA66K03	CT030428.1	1.79	5.9	1.73	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropical AtP-binding cassette sub-family D (Abcd2) transcript variant X1 mRNA	XM_00491502.1	0.9	2.96	1.73	0	0	0	0.01
Xenopus laevis iKAROS family zinc finger 3 (Ikaros) mRNA	NM_0011092483.1	0.33	1.06	1.73	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropical CD8 antigen-like (LOC100490323) transcript variant X1 mRNA	XM_002938993.2	1.68	5.5	1.72	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropical tubulin monoglycolase TLL3-like (LOC101730880) mRNA	XM_004913340.1	0.66	2.14	1.72	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropical calcium channel voltage-dependent N type alpha 1b subunit (cacna1b) partial mRNA	XM_004913082.1	2.84	9.33	1.72	0	0	0	0.01
Xenopus (Silurana) tropicalis mucosa associated lymphoid tissue lymphoma translocation protein 1-like (LOC100497102)	XM_00293571.2	0.66	2.15	1.71	0	0	0	0.01
Xenopus (Silurana) tropical protease serine 23 (prss23) mRNA	NM_001113120.1	2.87	9.35	1.71	0	0	0	0.01
Xenopus laevis cDNA clone IMAGE:6946930	BC076679.1	1.96	6.38	1.71	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TGA106F21	BC092116.1	0.65	2.09	1.7	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropical ankyrin repeat and SOCS box containing 2 (asb2) transcript variant X1 mRNA	CR85737.2	0.56	1.81	1.69	0	0	0	0.01
Xenopus tropicalis CDNA clone TBA07209	CU075384.1	0.87	2.8	1.69	0	0	0	0.01
Xenopus (Silurana) tropical kinesin family member 7 (kif7) mRNA	NM_001102886.1	0.83	2.64	1.68	0	0	0	0.01
Xenopus (Silurana) tropical Cdc42 guanine nucleotide exchange factor (GEF) 9 (arfgef9) mRNA	NM_001092056.2	0.8	2.55	1.67	0	0	0	0.01
Xenopus laevis arginyl aminopeptidase (aminopeptidase B) (rnpep-b) mRNA	NM_001092079.1	0.52	1.65	1.67	0	0	0	0.01
Xenopus (Silurana) tropicalis jumonji domain containing 7 (jmjd7) mRNA	NM_00101279.1	0.57	1.78	1.67	0	0	0	0.01

Gene	Accession	Six1+Eya1 CHK	Six1+Eya1 CHK+DXR <sup>a</sup>	FC Six1+Eya1 CHK	FC Six1+Eya1 CHK+DXR <sup>a</sup>	Control CHK	Control CHK+DXR <sup>a</sup>	FC Control CHK	FC Control CHK+DXR <sup>a</sup>	Rank <sup>b</sup>
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC100490280) mRNA	XM_0029541317.2	0.54	1.71	1.67	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis cysteine-rich secretory protein LCL1 domain containing 2 (crisp1d2) mRNA	NM_001032328.1	0.46	1.42	1.66	0	0	0	0	0	0.01
Xenopus laevis protein kinase repeat domain 39B (ttc39b) mRNA	NM_001094701.1	0.55	1.71	1.66	0	0	0	0	0	0.01
Xenopus laevis kinase AWP-activated gamma 3 non-catalytic subunit b (pkag3-3-b) mRNA	NM_00108963.1	0.47	1.47	1.66	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis tandem C2 domains nuclear (ttc2n) mRNA	XM_002933204.2	0.68	2.12	1.66	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis uncharacterized LOC10045450 (LOC100491247) mRNA	NM_001126904.1	0.88	2.75	1.65	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis putative N-acetyltransferase 16-like (LOC100491247) mRNA	XM_002943191.2	0.55	1.68	1.64	0	0	0	0	0	0.01
Xenopus laevis uncharacterized protein MGCB8735 (MGCB8735) mRNA	NM_001090211.1	0.64	1.98	1.63	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis putative protein female sterile-like (LOC100488372) mRNA	XM_002940610.2	2.39	7.39	1.63	0	0	0	0	0	0.01
Xenopus laevis uncharacterized LOC100036989 (LOC100036989) mRNA	NM_001097746.1	0.36	1.12	1.63	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis uncharacterized LOC100145136 (LOC100145136) mRNA	NM_001126643.1	0.6	1.86	1.63	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC100494747) mRNA	XM_00293603.2	1.12	3.45	1.63	0	0	0	0	0	0.01
Xenopus laevis unc-45 homolog B (uncd5b) mRNA	NM_00109403.1	0.85	2.61	1.63	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis synaptic vesicle glycoprotein 2B (sv2b) transcript variant X1 mRNA	XM_002933209.2	1.74	5.33	1.63	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis calcium/calmodulin-dependent protein kinase (CaM kinase) II delta (camk2d) mRNA	NM_001113692.1	0.5	1.52	1.62	0	0	0	0	0	0.01
Xenopus laevis uncharacterized LOC100037217 (LOC100037217) mRNA	NM_001097906.1	0.37	1.13	1.62	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis (Drosophila) (pum2) mRNA	XM_002934483.2	1.04	3.16	1.61	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis sulfomucin core protein 2-like (LOC101732584) mRNA	XM_00491548.1	0.98	2.97	1.61	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis forkhead box P2 (foxp2) mRNA	NM_0011010252.1	1.67	5.06	1.61	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis ras-related protein Rab-39B-like (LOC1000494491) mRNA	XM_00293466.2	1.44	4.33	1.6	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis C-terminal like (ctnbp2l) transcript variant X2 mRNA	NM_004910727.1	1.9	5.72	1.6	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis growth factor beta receptor III (tgfb3) transcript variant X3 mRNA	XM_004913725.1	0.38	1.14	1.59	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis beta-site APP-cleaving enzyme 1 (bacel1) mRNA	NM_001079017.1	0.46	1.36	1.58	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis transfer protein (tppa) mRNA	NM_001008184.1	1.49	4.42	1.58	0	0	0	0	0	0.01
Xenopus laevis hypothetical protein LOC100037229 mRNA (cDNA clone IMAGE:8541663) partial cds	BC129775.1	1	2.97	1.58	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis C-terminal like (LOC100495987) mRNA	XM_002937799.2	0.45	1.31	1.57	0	0	0	0	0	0.01
Xenopus tropicalis cDNA clone IMAGE:7682896	BC168014.1	0.37	1.08	1.57	0	0	0	0	0	0.01
Xenopus laevis keratin 17 (krt17) mRNA	NM_00109941.1	0.73	2.13	1.56	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis transcript variant X1 mRNA	XM_002936727.2	1.04	3.04	1.55	0	0	0	0	0	0.01
XELMMNNN Xenopus laevis low molecular weight endopeptidase-like 1 (lmmel1) mRNA	M86654.1	0.35	1.03	1.55	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis membrane metallo-endopeptidase-like 1 (lmmel1) mRNA	NM_001127095.1	1.38	4	1.55	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis C delta type-like (LOC101732924) mRNA	XM_004917384.1	0.6	1.73	1.55	0	0	0	0	0	0.01
Xenopus laevis uncharacterized LOC100036876 (LOC100036876) mRNA	NM_001097657.1	0.54	1.57	1.55	0	0	0	0	0	0.01
Xenopus tropicalis finished DNA clone TEG07462.19	CR942787.2	0.41	1.17	1.54	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC10004891674 (LOC10004891674) mRNA	XM_002934643.2	0.4	1.16	1.54	0	0	0	0	0	0.01
Xenopus laevis ret proto-oncogene mRNA (cDNA clone IMAGE:165904) (IMAGE:9093558) complete cds	BC170177.1	0.64	1.85	1.54	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine-protein kinase N2-like (LOC100487885) mRNA	XM_002937894.1	0.39	1.13	1.54	0	0	0	0	0	0.01
Xenopus tropicalis cDNA clone IMAGE:7644543	BC128642.1	0.57	1.62	1.53	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis APP-cleaving enzyme 1 (bacel1) mRNA	XM_002932950.2	0.5	1.42	1.53	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis metallopeptidase with thrombospondin type 1 motif 7 (adams7)	XM_002934674.2	0.76	2.19	1.53	0	0	0	0	0	0.01
Xenopus tropicalis finished DNA clone Theul04c17	BC168062.1	0.7	2.01	1.52	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis phospholipase C beta 2 (plcb2) mRNA	XM_00293518.2	0.74	2.1	1.52	0	0	0	0	0	0.01
Xenopus laevis Wilms tumor 1 mRNA (cDNA clone MGC:197240) (IMAGE:9093898) complete cds	BC170513.1	0.82	2.33	1.52	0	0	0	0	0	0.01
Xenopus tropicalis novel helix-loop-helix DNA binding domain protein mRNA (cDNA clone MGC:185767 IMAGE:7551946)	BC161271.1	0.9	2.54	1.51	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis mitochondrial protein mRNA	XM_004919937.1	1.28	3.64	1.51	0	0	0	0	0	0.01
Xenopus tropicalis finished DNA clone LOC101733225 (LOC101733225) mRNA	CU075784.1	0.52	1.47	1.51	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis neuropeptide Y receptor Y2 (npyp2) mRNA	XM_004917294.1	1.25	3.54	1.51	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis casein kinase I (fast-twitch skeletal muscle) (casq1) nuclear gene encoding mitochondrial protein mRNA	XM_004915133.1	0.84	2.38	1.51	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis glucose-6-phosphate isomerase mRNA	NM_001086758.1	0.51	1.43	1.51	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis DNA dedicator of cytokinesis 10 (dock10) mRNA	NM_001126592.1	0.57	1.6	1.5	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis cDNA clone IMAGE:7644543	XM_002937096.2	0.4	1.11	1.49	0	0	0	0	0	0.01

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DXR <sup>a</sup>	FC Six1+Eva1 CHX	FC Six1+Eva1 CHX+DXR <sup>a</sup>	Control CHX	Control CHX+DXR <sup>a</sup>	FC Control CHX	FC Control CHX+DXR <sup>a</sup>	Rank <sup>b</sup>
Xenopus (Silurana) tropicalis cofilin 2 (non-muscle) (Ctf2) mRNA	NM_001011156.2	0.38	1.04	1.49	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis protein C13orf12 homolog (LOC100488697) mRNA	XW_002942813.2	0.67	1.86	1.49	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis synaptotagmin VI (Syt6) mRNA	XM_002935776.2	9.95	27.83	1.49	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis X (M14 family) member 1 (cpxm1) transcript variant XI mRNA	XM_002935688.2	1.56	4.34	1.49	0	0	0	0	0	0.01
Xenopus laevis progesterone receptor mRNA (cDNA clone MGC:79872 (MAGEF6;317815)) complete cds	BC068635.2	0.42	1.17	1.49	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis X (M14 family) member 1 (cpxm1) transcript variant XI mRNA	NM_203640.1	1.69	4.68	1.48	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis glycine amidinotransferase (L-arginine-glycine amidinotransferase) (gatm)	NM_001142825.1	0.45	1.24	1.47	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis fibroblast growth factor 19 (fgf19) mRNA	NM_001011117.2	0.92	2.5	1.46	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis calmodulin-like (calm14) mRNA	NM_004916711.1	0.86	2.35	1.45	0	0	0	0	0	0.01
XELSTAGE: Xenopus laevis mRNA sequence	LO4278.1	1.34	3.62	1.45	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100486093 (LOC100486093) transcript variant X2 mRNA	NM_004918907.1	0.61	1.65	1.44	0	0	0	0	0	0.01
Xenopus tropicalis finished DNA clone TTPaG6g09	CU075826.1	1.04	2.79	1.44	0	0	0	0	0	0.01
Xenopus laevis hypothetical protein LOC100037130 mRNA (cDNA clone IMAGE:8527261) partial cds	BC133180.1	0.49	1.32	1.44	0	0	0	0	0	0.01
Xenopus tropicalis finished DNA clone TTPaG41k24	CU075800.1	0.83	2.22	1.44	0	0	0	0	0	0.01
Xenopus laevis reticulin 1A (RTN1-A) mRNA complete cds	AY484582.1	0.57	1.53	1.44	0	0	0	0	0	0.01
Xenopus laevis similar to calsequestrin 2 (cardiac muscle) mRNA (cDNA clone IMAGE:4581496) partial cds	BC041283.1	0.54	1.45	1.43	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis cartilage-intermediate layer protein nucleotide pyrophosphorylase (clip) mRNA	NM_002938356.2	0.59	1.57	1.42	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis radiation-inducible immediate-early gene EX-1-like (LOC100497513)	NM_004910807.1	27.24	72.51	1.42	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis E3 ubiquitin-protein ligase TRIM41-like (LOC100487181) mRNA	NM_004915623.1	0.84	2.21	1.41	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis piwi-like RNA-mediated gene silencing 2 (piwi2) mRNA	NM_001112999.1	0.95	2.52	1.41	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis mucin 2-like (LOC101373230) partial mRNA	NM_00492631.1	0.43	1.14	1.41	0	0	0	0	0	0.01
Xenopus laevis X-ATPase beta-m subunit (ATP6B4) mRNA complete cds	DQ413025.1	0.89	2.37	1.41	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis kinase-like protein KIFC3-like (LOC100485082) transcript variant X1 mRNA	NM_0029245102.2	0.89	2.35	1.41	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100495639 (LOC100495639) mRNA	NM_002935483.2	52.37	138.89	1.41	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis zinc finger DHHC-type containing 22 (Zdhhc22) mRNA	NM_00293862.2	0.52	1.36	1.41	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101735158 (LOC101735158) mRNA	NM_004923018.1	0.47	1.25	1.41	0	0	0	0	0	0.01
Xenopus laevis beta-m subunit (ATP6B4) mRNA complete cds	NM_001142082.1	1.23	3.23	1.4	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis kinase-like protein KIFC3-like (LOC100485082) transcript variant X2 mRNA	BC081275.1	6.89	18.01	1.39	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100495639 (LOC100495639) mRNA	NM_001087398.1	0.63	1.64	1.38	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis protein kinase AMP-activated alpha 2 catalytic subunit (prkaa2) mRNA	NM_004921541.1	2.28	5.91	1.38	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100493102 (LOC100493102) transcript variant X2 mRNA	NM_004919158.1	2.21	5.69	1.37	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis MGC:108117 protein (MGC:108117) mRNA	NM_001015694.1	4.27	10.95	1.37	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis scavenger receptor class F member 2-like (plb2ga) mRNA	NM_002923202.2	1.48	3.79	1.36	0	0	0	0	0	0.01
Xenopus laevis phospholipase A2 group IVA (cytosolic calcium-dependent) (pla2ga) mRNA	NM_004910523.1	0.69	1.74	1.35	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis ERH receptor A7 (leph7) transcript variant X3 mRNA	NM_0011012041.1	0.44	1.11	1.35	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis zinc finger DNA binding protein Gli-1 mRNA partial cds	U57154.1	0.78	1.97	1.35	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis Ly-9-like (LOC100486657) mRNA	NM_002924086.2	8.56	21.57	1.34	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733060 (LOC101733060) partial mRNA	NM_004920141.1	0.61	1.51	1.33	0	0	0	0	0	0.01
Xenopus tropicalis Sam69 protein mRNA (cDNA clone IMAGE:6997843) partial cds	BC091702.1	7.86	19.6	1.32	0	0	0	0	0	0.01
Xenopus tropicalis finished DNA clone TEG049b19	CT027912.1	0.54	1.33	1.32	0	0	0	0	0	0.01
Xenopus laevis uncharacterized LOC495139 (LOC495139) mRNA	NM_001094834.1	0.46	1.14	1.32	0	0	0	0	0	0.01
Xenopus laevis transcription factor 21 (Tcf21) mRNA complete cds	AY660871.1	2.53	6.28	1.32	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis glial cell-line-derived neurotrophic factor-like (LOC100495890) transcript variant X2 mRNA	NM_00491964.1	0.47	1.17	1.32	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis unc-5 homolog C (C. elegans) (unc5c) transcript variant Y2 mRNA	NM_004919103.1	2.02	5.01	1.32	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis protocadherin-11 X-linked-like (LOC10048398) transcript variant X2 mRNA	NM_00491890.1	0.71	1.76	1.31	0	0	0	0	0	0.01
Xenopus tropicalis finished DNA clone TBA06611	CU075734.1	0.44	1.09	1.31	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis mediator complex subunit 11 (med11) mRNA	NM_204048.1	0.45	1.1	1.31	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100488173 (LOC100488173) mRNA	NM_002940932.2	0.45	1.09	1.3	0	0	0	0	0	0.01
Xenopus laevis ectonucleoside triphosphate diphosphohydrolase 1 (entpd1) mRNA	NM_001092268.1	1.3	3.19	1.3	0	0	0	0	0	0.01
Xenopus laevis major histocompatibility complex class II DR alpha (h2-ea) mRNA	NM_001096778.1	1.02	2.5	1.3	0	0	0	0	0	0.01

Gene	Accession	Six1+Eva1 CHK	Six1+Eva1 CHK+DXR <sup>a</sup>	FC Six1+Eva1 CHK	FC Six1+Eva1 CHK+DXR <sup>a</sup>	Control CHK	Control CHK+DXR <sup>a</sup>	FC Control CHK	FC Control CHK+DXR <sup>a</sup>	Rank <sup>b</sup>
Xenopus (Silurana) tropicalis p21 protein [Cdc42/Rac]-activated kinase 7 (pdk7) mRNA	NM_001126638.1	0.52	1.26	1.3	0	0	0	0	0	0.01
Xenopus laevis ES98b mRNA for enhancer of split related protein 9b complete cds	AB211547.1	1.33	3.26	1.3	0	0	0	0	0	0.01
Xenopus tropicalis hypothetical protein LOC733725 mRNA [cDNA clone MGC:180542 IMAGE:7866825] complete cds	BC154856.1	1.88	4.55	1.28	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis mitofusin 1 interacting protein 1 (mfp1) transcript variant X1 mRNA	XM_002934662.2	0.45	1.08	1.28	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis growth differentiation factor 11 (gdf11) mRNA	XM_00293453.2	0.56	1.35	1.28	0	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone Tgg05b24	CT025277.2	398.32	961.91	1.28	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis ovomucoid-like (LOC101734374) (LOC101734374) mRNA	XM_002939809.2	1.66	4	1.27	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734374 (LOC101734374) mRNA	XM_004914707.1	0.77	1.85	9.33	0	0	0	0	0	0.01
Xenopus laevis similar to booby sox homolog (Drosophila) mRNA [cDNA clone IMAGE:5543330] partial cds	BC043817.1	3.91	9.33	1.26	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis mRNA for enhancer of split related protein 9b (es98b) mRNA	NM_001129762.1	1.04	2.47	1.26	0	0	0	0	0	0.01
Xenopus laevis Nr2B2 protein (nr2f2) mRNA	NM_001094481.1	2.43	5.79	1.26	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis leucine rich repeat containing 17 (lrrc17) mRNA	XM_004913023.1	0.62	1.47	1.26	0	0	0	0	0	0.01
Xenopus laevis hypothetical LOC495343 mRNA [cDNA clone IMAGE:69465904] partial cds	BC084805.1	0.95	2.26	1.25	0	0	0	0	0	0.01
Xenopus laevis similar to booby sox homolog (Drosophila) mRNA [cDNA clone IMAGE:5543330] partial cds	BC166311.1	0.59	1.4	1.25	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis transient receptor potential cation channel subfamily A member 1 (trpa1) mRNA	XM_004915572.1	1.04	2.46	1.25	0	0	0	0	0	0.01
Xenopus laevis Nr2B2 protein (nr2f2) mRNA	BC135560.1	0.57	1.33	1.25	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis unknown open reading frame human C14orf37 (LOC100497155)	CR926181.2	309.74	730.3	1.24	0	0	0	0	0	0.01
Xenopus laevis uncharacterized protein MG81120 (MG81120) mRNA	XM_00491219.1	0.52	1.21	1.24	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis diacylglycerol kinase eta (dgkhn) transcript variant X3 mRNA	NM_00109125.1	0.74	1.73	1.23	0	0	0	0	0	0.01
Xenopus laevis MG81672 protein mRNA [cDNA clone MG81672 IMAGE:586451.3] complete cds	XM_00491835.1	0.61	1.42	1.23	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis ERFR3 homolog B (S. cerevisiae) (erf3b) mRNA	BC071761.1	1.66	3.87	1.23	0	0	0	0	0	0.01
Xenopus laevis hypothetical protein LOC100049770 mRNA [cDNA clone IMAGE:8076078] partial cds	CU075356.1	1.12	2.61	1.23	0	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TTBAQ20h11	XM_00491495.1	1.44	3.35	1.23	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730802 (LOC101730802) mRNA	CR85706.2	0.74	1.71	1.23	0	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone Tga1070m05	NM_001096294.1	1.33	3.07	1.22	0	0	0	0	0	0.01
Xenopus laevis N-acetylneuraminate pyruvate lyase (dihydrodipicolinate synthase) (npl-a) mRNA	BC099050.1	1.92	4.44	1.22	0	0	0	0	0	0.01
Xenopus laevis cDNA clone IMAGE:63235682	XM_004919785.1	0.54	1.25	1.22	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis extracellular calcium-sensing receptor-like (LOC100496505) mRNA	CR94285.2	98.75	266.1	1.2	0	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone Tgg098905	BC088916.1	0.86	1.95	1.2	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis transmembrane protein 163 (tmem163) mRNA	NM_00112984.1	1.15	2.58	1.18	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis GIPC PDZ domain containing family member 3 (gipc3) mRNA	NM_001109302.1	3.1	6.98	1.18	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis protein phosphatase 1 regulatory subunit 16B (pp1r16b) mRNA	NM_00113280.1	0.52	1.16	1.18	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis ATP-binding cassette sub-family B (MDR/TAP) member 9 (abcb9)	NM_004910518.1	0.99	2.22	1.17	0	0	0	0	0	0.01
Xenopus laevis collagen type II alpha 1 mRNA [cDNA clone MG812599 IMAGE:4930289] complete cds	BC049862.1	1.14	2.55	1.17	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis poly (ADP-ribose) polymerase 14-like (LOC100481441) mRNA	NM_00294546.2	1.25	2.8	1.17	0	0	0	0	0	0.01
Xenopus laevis mitofusin 1 (mfn1) nuclear gene encoding mitochondrial protein mRNA	NM_001094981.1	1.28	2.86	1.17	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis V-set and transmembrane domain containing 2B (vtm2b) mRNA	NM_001161389.1	0.97	2.17	1.16	0	0	0	0	0	0.01
Xenopus laevis MD51 and EV1 complex locus (necom-b) mRNA	NM_001094933.1	1.88	4.19	1.16	0	0	0	0	0	0.01
Xenopus laevis sema domain immunoglobulin domain (lg) short basic domain secreted (sema3c) mRNA	NM_001094281.1	0.72	1.59	1.16	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100494257 (LOC100494257) mRNA	NM_00293249.2	1.72	3.8	1.15	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis maltooligosidase-glucosidase (mgam) mRNA	NM_00491703.1	0.71	1.57	1.15	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis ryanodine receptor 1 (skeletal) (ryr1) transcript variant X2 mRNA	BC118722.1	0.52	2.69	1.15	0	0	0	0	0	0.01
Xenopus laevis MD51 and EV1 complex locus (necom-b) mRNA	NM_00293462.2	0.48	1.05	1.15	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis corin serine peptidase (corin) transcript variant X1 mRNA	NM_001094308.1	1.69	3.72	1.14	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis transcript receptor potential cation channel subfamily C member 6 (trpc6) mRNA	NM_00293570.2	0.91	2	1.14	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC101734194) mRNA	NM_004915927.1	0.86	1.88	1.14	0	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis FAM 3D-like (LOC100496780) transcript variant X2 mRNA	NM_004912158.1	0.81	1.77	1.14	0	0	0	0	0	0.01
Xenopus (Silurana) tropicalis ADAM metallopeptidase with thrombospondin type 1 motif 9 (adams9) mRNA	NM_001007896.1	2.72	5.94	1.14	0	0	0	0	0	0.01

Gene	Accession	Six1+Eva1 CHK	Six1+Eva1 CHX+DXR <sup>a</sup>	FC Six1+Eva1 CHX	FC Six1+Eva1 CHX+DXR <sup>a</sup>	Control CHK	Control CHX+DXR	FC Control CHK	Rank <sup>b</sup>
Xenopus (Silurana) tropicalis iroquois homeobox 5 (irx5) mRNA	NM_001171927.1	0.93	2.03	1.14	0	0	0	0	0.01
Xenopus laevis mRNA for transcription factor (foxo2 gene)	AJ344435.1	3.74	8.18	1.13	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis transmembrane gamma-carboxyglutamic acid protein 4-like (LOC100486092) mRNA	XM_002938713.2	0.53	1.15	1.13	0	0	0	0	0.01
Xenopus laevis MGC80350 protein (MGC80350) mRNA	NM_001092066.1	0.69	1.49	1.13	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TTPA07205	CR855825.2	1.72	3.73	1.12	0	0	0	0	0.01
Xenopus laevis alcohol dehydrogenase iron containing 1 (adhfe1) nuclear gene encoding mitochondrial protein mRNA	NM_001127802.1	0.76	1.65	1.12	0	0	0	0	0.01
Xenopus (Silurana) tropicalis zinc finger MYND-type containing 8 (zmynd8) transcript variant 1 mRNA	NM_001006790.1	87.33	189.49	1.12	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone Theud03708	CT025170.2	0.58	1.25	1.12	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis single-minded homolog 1 (Drosophila) (sim1) transcript variant X3 mRNA	XM_001491546.1	0.59	1.28	1.12	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis myosin XVA (myo15a) mRNA	NM_002938562.2	0.84	1.8	1.11	0	0	0	0	0.01
Xenopus (Silurana) tropicalis wingless-type MMTV integration site family member 7B (wnt7b) mRNA	NM_001126633.1	1.4	3.01	1.11	0	0	0	0	0.01
Xenopus laevis Rad17 (rad17) mRNA complete cds	AY169965.1	0.71	1.52	1.11	0	0	0	0	0.01
Xenopus laevis villin 1 (vill1) mRNA	NM_001087034.1	0.67	1.44	1.11	0	0	0	0	0.01
Xenopus (Silurana) tropicalis zinc finger protein 572 (znf572) mRNA	NM_001095926.1	1.61	3.46	1.11	0	0	0	0	0.01
Xenopus laevis zinc finger protein 572 (znf572) mRNA	NM_001015777.2	0.71	1.53	1.11	0	0	0	0	0.01
Xenopus (Silurana) tropicalis protease serine 29 (prss29) mRNA	NM_001001228.1	1.87	4	1.1	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis zinc finger BTB/POU domain containing 20 (zbfb20) mRNA	XM_002938649.2	2.48	5.28	1.1	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TGA07315	CR762215.2	0.8	1.71	1.1	0	0	0	0	0.01
Xenopus laevis clone IMAGE:4681494 mRNA	BC044072.1	1.9	4.04	1.1	0	0	0	0	0.01
Xenopus laevis Nk6 transcription factor related locus 1 (nkx6-1) mRNA complete cds	EF460789.1	0.69	1.47	1.1	0	0	0	0	0.01
Xenopus laevis empty chain 1 gene 2 (emx1.2) mRNA	NM_00109430.1	0.96	2.03	1.09	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis zinc finger CCHC domain-containing protein 3-like (LOC101735217) mRNA	XM_001491331.1	4.18	8.87	1.09	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C18orf42 (LOC100488626) mRNA	NM_002938057.2	0.53	1.11	1.08	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis failed axon connections homolog (Drosophila) (fac) mRNA	XM_00293867.2	0.87	1.82	1.08	0	0	0	0	0.01
Xenopus laevis sidekick cell adhesion molecule 2 (sdk2) mRNA	NM_001097930.1	0.59	1.24	1.07	0	0	0	0	0.01
Xenopus laevis acyl-CoA oxidase 2 branched chain (acox2) mRNA	NM_00109064.1	0.95	1.98	1.07	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis transmembrane and tetrapeptide repeat containing 1 (tmrct1)	XM_002942552.1	0.82	1.72	1.07	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731582 (LOC101731582) mRNA	XM_00491821.1	2.32	4.85	1.07	0	0	0	0	0.01
Xenopus laevis small glutamine-rich tetrapeptide repeat (TPR) containing beta (gfb) mRNA	NM_00109693.1	1.75	3.65	1.07	0	0	0	0	0.01
Xenopus laevis clone 1668	AJ009292.13	1.79	3.73	1.06	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis glutathione S-transferase 3-like (LOC100496158) transcript variant X1 mRNA	XM_002934854.2	0.89	1.85	1.06	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis dixi-like (LOC101734507) partial mRNA	XM_004920311.1	3.84	7.98	1.06	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101735163 (LOC101735163) mRNA	XM_004913712.1	2.62	5.44	1.06	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis kinase C delta type-like (LOC10497253) mRNA	XM_002933331.2	1.35	2.79	1.05	0	0	0	0	0.01
Xenopus tropicalis hypothetical protein LOC10045805 mRNA (cdna clone IMAGE:8962257) partial cds	BC161793.1	0.85	1.76	1.06	0	0	0	0	0.01
XLRNPA: Xenopus laevis ribonucleoprotein mRNA complete cds	L15430.1	0.55	1.14	1.06	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis short stature homeobox (shox) transcript variant X2 mRNA	NM_001491817.1	1.14	2.36	1.06	0	0	0	0	0.01
Xenopus laevis nephrin (NPHS1) mRNA complete cds	AY902238.1	0.55	1.14	1.05	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis threonine-protein kinase N2-like (LOC101733271) mRNA	XM_004913766.1	1.17	2.41	1.05	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731630 (LOC101731630) mRNA	NM_001079290.1	21.83	44.63	1.05	0	0	0	0	0.01
Xenopus (Silurana) tropicalis kinase C delta type-like (LOC101731809) mRNA	NM_00491377.1	0.61	1.23	1.03	0	0	0	0	0.01
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 5-1-like (LOC100337651) mRNA	NM_001127295.1	1.31	2.67	1.03	0	0	0	0	0.01
Xenopus laevis clone IMAGE:5079328	NM_004919054.1	1.01	2.05	1.03	0	0	0	0	0.01
Xenopus laevis cDNA clone TPA65d19	BC085001.1	2.44	4.95	1.03	0	0	0	0	0.01
Xenopus tropicalis finished cDNA clone TPA65d19	CU075897.1	2.05	4.12	1.02	0	0	0	0	0.01
Xenopus laevis uncharacterized LOC100127341 mRNA	NM_00112894.1	0.58	1.16	1.01	0	0	0	0	0.01
Xenopus tropicalis cDNA clone IMAGE:7790739	BC161549.1	0.94	1.87	1.01	0	0	0	0	0.01

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DIX*	FC Six1+Eya1 CHX	FC Six1+Eya1 CHX+DIX*	Control CHX	Control CHX+DIX*	FC Control CHX+DIX*	Rank*
Xenopus (Silurana) tropicalis v-maf musculoaponeurotic fibrosarcoma oncogene homolog A (mafA) mRNA	NM_001032304.1	1.46	2.91	1.01	0	0	0	0.01	
PREDICTED: Xenopus (Silurana) tropicalis bc12 antagonist of cell death-like (LOC100487418) mRNA	XM_002940225.2	5.44	298.33	5.78	11.45	11.56	0.02	0.01	
Xenopus laevis chromogranin A (parathyroid secretory protein 1) (chga) mRNA	NM_001094724.1	5.12	22.96	2.17	3.64	3.66	0.01	0.01	
PREDICTED: Xenopus (Silurana) tropicales G protein-coupled receptor 56 (Gpr56) mRNA	XM_002931653.2	0.6	1.76	1.56	1.79	1.79	0.01	0.01	
Xenopus tropicalis leucine zipper transcription factor RNA (cDNA clone MGC:346870) (IMAGE:7800970) complete cds	BC121507.1	0.78	2.82	1.86	1.03	1.04	0.01	0.01	
Xenopus laevis uncharacterized LOC100036823 (LOC100036823) mRNA	NM_001097113.1	0.29	2.12	2.9	1.89	1.92	0.03	0.01	
Xenopus (Silurana) tropicalis thyroid adenoma associated (thada) mRNA	NM_001091149.1	3.83	7.8	1.03	6.35	6.4	0.02	0.02	
PREDICTED: Xenopus (Silurana) tropicalis thyrocalcitonin receptor type 1 mRNA	XM_002937199.2	1.02	2.98	1.55	1.45	1.48	0.03	0.02	
Xenopus laevis sorting nexin 15 (snx15) mRNA	NM_001097066.1	2.91	5.91	1.03	2.33	2.36	0.02	0.02	
Xenopus laevis mRNA decapping enzyme 1 mRNA (cDNA clone IMAGE:5074458) partial cds	BC073062.1	641.11	2968.47	2.22	65.02	67.36	0.06	0.03	
Xenopus laevis family with sequence similarity 101 member B (fam101b) mRNA	NM_001098707.1	8.81	23.36	1.41	9.23	9.48	0.04	0.03	
Xenopus laevis CD81 protein (cd81-a) mRNA	NM_00108613.1	192.94	699.12	1.86	110.37	114.58	0.06	0.03	
PREDICTED: Xenopus (Silurana) tropicalis thyrocalcitonin receptor type 1 mRNA	XM_002932394.2	12.07	121.38	3.34	18.84	20.2	0.11	0.03	
PREDICTED: Xenopus (Silurana) tropicalis KIAA1332 (kiaa1332) transcript variant X2 mRNA	XM_004910792.1	0.45	1.12	1.32	1.31	1.35	0.05	0.04	
Xenopus (Silurana) tropicalis MGC89395 protein (MGC89395) mRNA	NM_001088113.1	1.64	3.77	1.21	2.13	2.19	0.05	0.04	
Xenopus laevis CD81 protein (cd81-b) mRNA complete cds	U76636.1	1.1	8.82	3.01	1.3	1.4	0.11	0.04	
Xenopus laevis CD81 protein (cd81-b) mRNA complete cds	NM_001136171.1	2.85	7.33	1.37	2.61	2.7	0.05	0.04	
PREDICTED: Xenopus (Silurana) tropicalis thyrocalcitonin receptor type 1 mRNA	XM_002932378.2	1.39	3.49	1.33	1.76	1.82	0.06	0.04	
Xenopus laevis CD81 protein (cd81-b) mRNA	NM_00109434.1	155.5	342.15	1.14	82.05	84.62	0.05	0.04	
Xenopus laevis CD81 protein (cd81-b) mRNA	AF05476.1	159.78	5107.36	1.73	812.21	855.19	0.08	0.05	
Xenopus (Silurana) tropicalis cablinbindin D28K mRNA complete cds	NM_001127155.1	1.74	4.3	1.32	1.63	1.69	0.05	0.04	
Xenopus laevis ATPase Na+/K+-transporting ATPase I (atp1b1) mRNA	NM_001086759.1	58.44	180.75	1.63	28.11	29.57	0.08	0.05	
Xenopus laevis 7-transmembrane receptor frizzled-1 mRNA complete cds	AF231711.1	1.45	5.43	1.91	1.39	1.48	0.1	0.05	
Xenopus laevis goliath-related E3 ubiquitin ligase 1 mRNA complete cds	DQ911466.1	1.42	3.2	1.19	1.76	1.84	0.06	0.05	
Xenopus tropicalis finished DNA clone TGA072h19	CT025400.2	1.9	6.95	1.88	1.58	1.7	0.12	0.06	
PREDICTED: Xenopus (Silurana) tropicalis inositol polyphosphate-4KDa gene 2 (inpp5a2) mRNA	NM_00293507.2	2.92	6.87	1.24	3.18	3.35	0.08	0.06	
Xenopus laevis ATPase Na+/K+-transporting ATPase I (atp1b1) mRNA	NM_213704.3	44.76	166.72	1.9	49.46	53.9	0.13	0.07	
Xenopus tropicalis 7-transmembrane receptor frizzled-1 mRNA	CT032929.1	0.55	2.95	2.43	1.17	1.31	0.16	0.07	
Xenopus laevis sema domain immunoglobulin domain (Ig) transmembrane domain (TM) and short cytoplasmic domain (sema4b)	NM_001095593.1	1.33	3.84	1.54	1.37	1.47	0.11	0.07	
Xenopus laevis nail T-cell differentiation protein (mal) mRNA	NM_001085577.1	6.65	16.92	1.35	10.98	11.74	0.1	0.08	
PREDICTED: Xenopus (Silurana) tropicalis membrane protease serine 13 (tmprss13) mRNA	XM_002932904.2	1.96	7.06	1.85	2	2.2	0.14	0.08	
Xenopus laevis sex-determining region Y-box 2 (sox2) mRNA	X51817.1	1.3	2.73	1.08	1.52	1.6	0.08	0.08	
PREDICTED: Xenopus (Silurana) tropicalis transforming growth factor-beta2 mRNA	Q0292559.1	30.98	74.97	1.28	29.94	32.31	0.11	0.09	
Xenopus (Silurana) tropicalis protocadherin 10 (pcdh10) mRNA	BC135795.1	117.29	12.78	1.02	2.31	2.44	0.08	0.08	
XLRP7CA-X laevis repeat element from bestrophin mRNA	XM_002945982.2	2.26	47.65	1.08	1.67	1.77	0.09	0.09	
PREDICTED: Xenopus (Silurana) tropicalis Xlaevis repeat element from bestrophin mRNA	M36867.1	0.67	24.65	5.2	5.3	7.17	0.44	0.09	
PREDICTED: Xenopus (Silurana) tropicalis Zfp485 mRNA	XM_00293201.2	3.68	8.1	1.14	3.7	3.95	0.1	0.09	
Xenopus laevis sex-determining region Y-box 2 mRNA complete cds	AB007035.1	0.71	1.58	1.18	1.25	1.35	0.11	0.1	
Xenopus tropicalis hypothetical protein LOC349562 mRNA (cDNA clone MGCI349562) complete cds	NM_004910503.1	1.54	5.2	1.76	1.28	1.43	0.17	0.1	
PREDICTED: Xenopus (Silurana) tropicalis protocadherin 14-like (pcdh14) mRNA	CR926200.2	16.49	39.46	1.26	12.92	14.07	0.13	0.1	
PREDICTED: Xenopus (Silurana) tropicalis ribonuclease 14-like (rnase14) mRNA	NM_001102975.1	4.19	8.89	1.09	2.89	3.11	0.11	0.11	
Xenopus tropicalis finished DNA clone TGA072h19	CT030363.1	1.51	6.88	2.19	1.66	1.9	0.2	0.09	
Xenopus tropicalis finished DNA clone The10515	CR760241.2	831.32	6224.09	2.91	52.87	63.81	0.28	0.1	
Xenopus laevis mRNA for FGFR receptor 3 complete cds	NM_001130271.1	2.08	5.38	1.38	2.32	2.55	0.15	0.11	
PREDICTED: Xenopus (Silurana) tropicalis putative leucine-rich repeat-containing protein DBB (LOC101734904) mRNA	NM_001094248.1	2.34	8.52	1.87	0.98	1.12	0.2	0.11	
Xenopus tropicalis finished DNA clone Theu21k01	NM_001089592.1	3.7	10.98	1.58	1.33	1.49	0.17	0.11	
Xenopus laevis paired-like homeobox 2 mRNA (cDNA clone MGCI30961) complete cds	BC124891.1	46.71	104.03	1.16	24.29	26.49	0.13	0.11	
Xenopus laevis anion channel activated chloride channel (an101) mRNA	NM_001096456.1	6.71	30.88	2.21	7.5	8.91	0.25	0.12	

Gene	Accession	Six1+Eya1 CHK	Six1+Eya1 CHK+DXR <sup>a</sup>	FC Six1+Eya1 <sup>b</sup>	FC Six1+Eya1 <sup>b</sup>	Control CHK	Control CHK+DXR <sup>a</sup>	FC Control CHK+DXR <sup>a</sup>	Rank <sup>c</sup>
Xenopus laevis MGCB5342 protein mRNA (cDNA clone MGCB5342 IMAGE:6876390) complete cds	BC081224.1	0.69	2.49	1.87	1.05	5.96	6.71	0.22	0.12
X. laevis mRNA for epithelial sodium channel gamma 2 subunit	Y12001.1	33.48	95.99	1.52	1.01	2.35	2.54	0.18	0.12
Xenopus (Silurana) tropicalis family with sequence similarity 21.4 member A (fam214a) mRNA	NM_001015702.1	2.52	5.07	1.01	1.44	1.6	1.16	0.12	0.12
Xenopus tropicalis Shox2 mRNA complete cds	DQ675518.1	1.38	3.57	1.38	1.25	1	1.1	0.15	0.12
PREDICTED: Xenopus (Silurana) tropicalis paraneoplastic antigen Ma2 homolog (LOC101733383) mRNA	XM_00495951.1	20.37	48.24	2.97	1.87	1.28	1.49	0.22	0.12
PREDICTED: Xenopus (Silurana) tropicalis Fh2 domain-containing protein 1-like (LOC100496216) mRNA	XM_002934907.2	0.82	1.94	1.44	1.84	2.07	1.18	0.18	0.12
PREDICTED: Xenopus (Silurana) tropicalis mitochondrial fission factor (mff) transcript variant X9 mRNA	XM_001941282.1	0.72	2.23	1.01	1.63	1.77	1.01	0.13	0.13
PREDICTED: Xenopus (Silurana) tropicalis Kruppel-like factor 3 (basic) (kif3) mRNA	XM_002936648.2	1.11	5.49	2.89	1.19	1.53	1.37	0.13	0.13
Xenopus laevis mab-2-like 2 (mab212b) mRNA	NM_001095701.2	0.75	1.19	1.61	32.68	37.69	0.21	0.13	
Xenopus laevis regulator of cell cycle (rgcc) mRNA	NM_001093976.1	23.49	71.51	5.59	1.6	1.88	2.17	0.21	
PREDICTED: Xenopus (Silurana) tropicalis kelch-like family member 35 (kh35) mRNA	XM_00294162.2	1.86	38.59	2.26	3.05	3.78	3.31	0.14	
Xenopus laevis Cep33 mRNA complete cds	FJ469888.1	8.1	38.75	1.13	2.65	2.95	0.16	0.14	
PREDICTED: Xenopus (Silurana) tropicalis sorting nexin 19 (snx19) mRNA	XM_002938108.2	10.92	23.75	1.95	34.23	41.3	0.28	0.14	
Xenopus laevis cDNA clone CD81 antigen (target of antiproliferative antibody 1) mRNA (cDNA clone MGCB52702 IMAGE:4682721) complete cds	BC041217.1	36.76	141.59	1.95	4.42	13.91	22.22	0.68	0.16
PREDICTED: Xenopus (Silurana) tropicalis uveal autoantigen with coiled-coil domains and ankyrin repeats-like (LOC101731603) mRNA	XM_004913328.1	4.09	10.99	1.43	5.29	6.08	0.21	0.15	
Xenopus laevis uncharacterized protein MGCB516527 (MGCB116527) mRNA	NM_001096165.1	3.36	7.44	1.16	2.41	2.7	0.17	0.15	
Xenopus laevis RNA binding motif protein 24 (rbm24-a) mRNA	NM_001097526.1	2.55	8.58	1.76	2.6	3.1	0.26	0.15	
Xenopus laevis cDNA clone IMAGE:6637905 containing frame-shift errors	BC076852.1	532.99	202.83	1.95	44.57	54.41	0.29	0.15	
Xenopus laevis uncharacterized protein MGCB6508 (MGCB6508) mRNA	NM_00108814.1	5.52	117.67	4.42	13.91	20.16	0.18	0.15	
PREDICTED: Xenopus (Silurana) tropicalis phosphopantethoylase decarboxylase-like (LOC100495698)	XM_004915998.1	34.63	76.2	1.14	17.86	22.9	0.68	0.16	
Xenopus laevis Myoblast determination protein 1 homolog A mRNA (cDNA clone MGCB52596 IMAGE:4885255) complete cds	BC041190.1	2.96	74.94	4.67	3.86	6.38	0.73	0.16	
Xenopus laevis RNA binding motif protein 24 (rbm24-b) mRNA	NM_00109751.1	0.67	4.87	2.89	1.01	1.38	0.45	0.16	
PREDICTED: Xenopus (Silurana) tropical T-cell leukemia homeobox 1 (ttx1) transcript variant 1 mRNA	XM_002936768.2	1.84	10.49	2.52	1.12	1.47	0.4	0.16	
PREDICTED: Xenopus (Silurana) tropicalis putative nuclelease HARB1-like (LOC100488339) mRNA	XM_002937555.2	0.48	1.25	1.4	1.65	1.92	0.23	0.16	
Xenopus tropicalis finished DNA clone TEg0372ad6	CT030667.1	0.08	1.33	4.21	22.9	36.69	0.18	0.17	
Xenopus laevis coagulation factor 5 (f5) mRNA	NM_00109678.1	51.72	220.87	2.1	11.52	14.6	0.35	0.17	
Xenopus (Silurana) tropical meningioma (disrupted in balanced translocation) 1 gene 1 (mn1.1) mRNA	NM_00110202.1	19.05	38.78	1.03	16.21	18.2	0.17	0.17	
PREDICTED: Xenopus (Silurana) tropical Shc1 (Src homology 2-domain containing) transforming protein 2 (shc2) mRNA	XM_002939766.2	1.28	2.95	1.21	0.97	1.12	0.2	0.17	
Xenopus laevis H2A histone family member Y2 (hzay2) mRNA	NM_001087412.1	321.3	646.37	1.01	10.52	11.8	0.17	0.17	
PREDICTED: Xenopus (Silurana) tropicalis flocculation protein F1 (lo11-like) transcript variant X2 mRNA	XM_002939280.2	16.64	45.37	1.43	7.12	8.38	0.24	0.17	
XLU13183: Xenopus laevis (Xwnt-4) mRNA complete cds	U13183.1	1.76	4.21	1.26	1.45	1.68	0.21	0.17	
Xenopus (Silurana) tropical adenylate cyclase activating polypeptide 1 (pituitary) (adcycl1) mRNA	NM_001096781.1	61.13	1.9	1.33	1.66	1.32	0.17	0.17	
PREDICTED: Xenopus (Silurana) tropical Shc1 (Src homology 2-domain containing) transforming protein 2 (shc2) mRNA	AF169794.1	1.02	4.34	2.1	4.11	5.25	0.36	0.17	
Xenopus laevis Ras-related associated with diabetes (rad) mRNA	NM_001092750.1	0.92	16.61	4.19	1.03	1.68	0.72	0.17	
PREDICTED: Xenopus (Silurana) tropicalis flocculation protein F1 (lo11-like) transcript variant X2 mRNA	XM_00491263.1	9.83	26.35	1.43	3.7	4.4	0.25	0.18	
Xenopus laevis tetraspain 1 (tspan1) mRNA	NM_001095473.1	19.72	45.37	1.21	20.69	23.95	0.22	0.18	
Xenopus (Silurana) tropical neurotrophin 3 (ntf3) mRNA	NM_001102740.1	1.65	1.36	4.46	0.66	1.14	0.18	0.18	
Xenopus laevis p21 GTPase-associated kinase 1 (PAK1) mRNA complete cds	NM_0011173404.1	32.14	71.7	1.16	27.43	31.78	0.22	0.19	
Xenopus laevis Ras-related associated with diabetes (rad) mRNA	BC073731.1	5.71	14.27	1.33	3.75	4.44	0.25	0.19	
PREDICTED: Xenopus (Silurana) tropicalis wingless-type MMTV integration site family member 4 (wnt4) mRNA	NM_004913802.1	1.26	2.76	1.14	1.12	1.29	0.22	0.19	
Xenopus laevis tetraspain 1 (tspan1) mRNA	NM_001095472.2	7.4	1.95	5.19	6.7	0.37	0.19	0.17	
Xenopus (Silurana) tropical adenylate cyclase activating polypeptide 1 (pituitary) (adcycl1) mRNA	BC072262.1	4.22	12	1.51	1.38	1.69	0.3	0.2	
Xenopus laevis xox-1 protein (xsox-1) mRNA	AY766060.1	22.37	50.41	1.18	10.21	11.98	0.24	0.2	
Xenopus laevis yemein2 (yem2) mRNA complete cds	NM_001084866.1	5.61	12.6	1.17	5.49	6.44	0.24	0.2	
PREDICTED: Xenopus (Silurana) tropical chimerin 1 (chn1) mRNA	XM_00293849.2	1.99	23.28	3.56	1	1.64	0.72	0.21	
Xenopus (Silurana) tropical wingless-type MMTV integration site family member 4 (wnt4) mRNA	NM_00125086.1	1.01	2.52	1.33	1.26	1.51	0.27	0.21	
PREDICTED: Xenopus (Silurana) tropical leucine rich adaptor protein 1-like (lrap1) mRNA	AF048994.3	0.63	1.38	1.13	2.23	2.61	0.23	0.21	
Xenopus laevis Xbp-3 protein (Xbp1) mRNA complete cds	BC072262.1	5.59	17.71	1.67	7.71	9.74	0.34	0.21	
Xenopus laevis p21 GTPase-associated kinase 1 mRNA (cDNA clone MGCB83445 IMAGE:5084287) complete cds	BC081113.1	0.57	4.61	3.04	0.77	1.19	0.63	0.21	
Xenopus (Silurana) tropical calnexin (canx) mRNA	BC106345.1	0.63	1.67	1.42	3.18	3.97	0.32	0.23	
PREDICTED: Xenopus (Silurana) tropical transcription repressor (grf1) mRNA	NM_001005668.1	0.73	6.52	3.18	0.78	1.32	0.76	0.24	
Xenopus (Silurana) tropical calcineurin B (calnB) mRNA	NM_002933803.2								

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DX*	FC Six1+Eva1 CHX+DX*	Six1+Eva1 CHX	FC Six1+Eva1 CHX	Control CHX+DX	FC Control CHX+DX	Rank*
PREDICTED: Xenopus (Silurana) tropicalis HemK methyltransferase family member 1 (hemk1) mRNA	XM_004510733.1	4.06	8.45	1.07	3.6	4.29	0.26	0.24	
Xenopus (Silurana) tropicalis protein phosphatase 2 regulatory subunit B' beta (pp275b) mRNA	NM_001100279.1	0.12	2.08	4.14	0.51	1.02	1.01	0.25	
Xenopus laevis fast tropomin T (TNNNT3) mRNA complete cds	AY114144.1	0.38	1.06	4.48	0.81	1.04	0.37	0.25	
Xenopus laevis natriuretic peptide receptor A/guanylyl cyclase A (atrionatriuretic peptide receptor A) (npr1) mRNA	NM_00109234.1	1.05	2.31	1.15	1.12	1.36	0.29	0.25	
Xenopus laevis corinfilin homolog B (crnf1-b) mRNA	NM_00109858.1	0.94	3.92	2.07	1.06	1.51	0.52	0.25	
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 198 member A (fam198a) mRNA	NM_001293785.2	3.24	7.82	2.28	2.49	3.1	0.32	0.26	
Xenopus laevis R3Hypiy3 mRNA for R3Hypiy3 protein complete cds	AB55086.1	12.56	49.31	1.98	6.33	8.93	0.5	0.26	
Xenopus laevis zinc finger RAN-binding domain containing 1 (ranbb1-b) mRNA	NM_001091229.1	9.59	21.1	1.14	27.85	33.96	0.29	0.26	
Xenopus laevis Rab19 member RAS oncogene family (rab19) mRNA	NM_00109665.1	83.5	478.75	2.52	161.98	252.93	0.65	0.26	
Xenopus laevis uncharacterized protein MGCG8724 (MGCG8724) mRNA	NM_001089949.1	3.4	8.97	1.41	1.76	2.27	0.37	0.26	
Xenopus (Silurana) tropicalis G protein-coupled receptor 153 (grp153) mRNA	NM_001128052.1	5.63	15.08	1.43	1.32	1.71	0.38	0.27	
Xenopus (Silurana) tropicalis member B (rasl1b) mRNA	NM_001015774.1	5.03	13.13	1.39	5.38	6.94	0.37	0.27	
Xenopus laevis 5'-nucleotidase domain containing 4 (n5dd4) mRNA	NM_001094603.1	6557	14579.89	1.16	1849.38	2307.69	0.32	0.28	
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 35 member E4 (slc35e4) transcript variant X2 mRNA	NM_004910496.1	0.6	1.83	1.62	0.75	1.03	0.46	0.29	
Xenopus tropicalis finished cDNA clone Theud85b13	CR760373.2	58.13	158.05	1.45	47.52	62.92	0.41	0.29	
PREDICTED: Xenopus (Silurana) tropicalis progestin and adiponQ receptor family member IX (paro9) mRNA	XM_004910351.1	4.16	8.97	1.11	1.47	1.83	0.32	0.29	
Xenopus tropicalis uncharacterized LOC00492708 (LOC00492708) mRNA	XM_00294289.2	16.17	48.77	1.6	13.09	18.07	0.47	0.3	
Xenopus laevis cDNA clone IMAGE7009602	BC084260.1	8.26	23.11	1.49	4.3	5.84	0.45	0.3	
PREDICTED: Xenopus (Silurana) tropicalis FH2 domain containing 1 (fhdc1) mRNA	XM_002933493.2	2.78	6.53	1.24	2.87	3.72	0.38	0.31	
Xenopus laevis cDNA clone MGCG130954 (MGCG130954) mRNA	BC106382.1	2.71	10.51	1.96	1.9	2.86	0.6	0.31	
Xenopus laevis uncharacterized protein MGCG8265 (MGCG8265) mRNA	NM_001095998.1	4.61	9.29	1.02	5.18	6.47	0.32	0.29	
XL21860 Xenopus laevis type I serine/threonine kinase receptor (X-Trk1) mRNA complete cds	U21860.1	2.23	4.52	1.03	2.08	2.61	0.33	0.33	
PREDICTED: Xenopus (Silurana) tropicalis Tgg-79/3902 complete cds	XN_002931673.2	3.01	6.19	1.04	3.37	4.25	0.34	0.33	
Xenopus laevis cDNA clone MGCG130953 (MGCG130953) mRNA	CR761262.2	1.58	3.35	1.09	3.93	5.02	0.36	0.33	
Xenopus laevis uncharacterized protein MGCG883 (MGCG883) mRNA	BC155263.1	6.96	17.76	8.37	11.34	14.44	0.32	0.33	
PREDICTED: Xenopus (Silurana) tropicalis integrin beta 1 (itgb6) mRNA	XM_002941653.2	8.26	40.54	2.3	6.44	10.98	0.76	0.33	
Xenopus (Silurana) tropicalis protein-coupled receptor 63 (grp63) mRNA	NM_001097306.1	0.37	2.37	2.72	0.67	1.24	0.91	0.34	
Xenopus tropicalis finished cDNA clone TGas076K6	XM_002934868.2	1.27	2.74	1.12	0.8	1.03	0.38	0.34	
Xenopus tropicalis lysophosphatidic acid receptor 3 (lpar3) mRNA	CR761730.2	238.91	729.1	1.61	97.55	142.27	0.55	0.55	
Xenopus laevis cDNA clone IMAGE8332229	NM_00109374.1	0.43	1.54	0.67	1.04	1.63	0.34	0.33	
PREDICTED: Xenopus (Silurana) tropicalis radiation resistance associated (rrasra) transcript variant 1 mRNA	AF00239.1	1.66	9.76	2.56	5.75	10.51	0.88	0.34	
Xenopus (Silurana) tropicalis uncharacterized LOC01070544 (LOC01070544) mRNA	NM_001130321.1	9.14	18.6	1.03	6.04	7.68	0.35	0.34	
Xenopus (Silurana) LIM homeobox 2 (lsl2) mRNA	NM_001165041.1	5.26	16.33	1.64	5.65	8.33	0.57	0.35	
PREDICTED: Xenopus (Silurana) atonal homolog 1 (drosophila) (atoh1) mRNA	NM_00491085.1	8.54	22.6	1.41	2.26	3.19	0.5	0.36	
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 46 member A (fam46a) mRNA	NM_001092306.1	6.46	16.71	1.38	4.17	5.83	0.49	0.36	
Xenopus laevis LIM domain containing 2 (limd2-a) mRNA	NM_001092306.1	122.67	808.85	2.73	212.32	416.49	0.98	0.36	
PREDICTED: Xenopus (Silurana) tropicalis ribosomal protein S19 (rps19) transcript variant X4 mRNA	NM_004910834.1	1.26	2.87	1.2	1.05	1.42	0.44	0.37	
Xenopus (Silurana) calyphosine (span31) mRNA	NM_001097320.1	0.82	10.49	3.7	0.88	2.32	0.39	0.39	
PREDICTED: Xenopus (Silurana) tetraspain 31 (span31-b) mRNA	NM_004920603.1	0.71	2.62	1.9	0.78	1.29	0.73	0.39	
Xenopus laevis mex3b mRNA for Mex-3 homolog b complete cds	AB499003.2	6.23	16.91	1.45	3.24	4.76	0.56	0.39	
Xenopus laevis mRNA for xPL1 complete cds	AB127963.1	14.79	60.12	2.03	0.58	1.01	0.8	0.4	
PREDICTED: Xenopus (Silurana) tropicalis proliffe rich 15 (pr15) mRNA	NM_00293381.2	6.86	16.85	1.3	1.43	2.05	0.52	0.4	
Xenopus laevis F-box protein 46 (fbx46) mRNA	NM_001098714.1	18.06	38.58	1.1	15.57	21.08	0.44	0.4	
Xenopus (Silurana) LIM and senescent cell antigen-like domains 1 (lims1) mRNA	NM_001142098.1	1.64	4.25	1.38	1.16	1.71	0.57	0.41	
Xenopus laevis tetraspain 31 (span31-b) mRNA	NM_001086181.1	3238.41	7035.72	1.12	1437.06	1970.58	0.46	0.41	
Xenopus laevis kiaa0930 (kiaa0930) mRNA	NM_001086221.1	3.7	10.67	1.54	3.28	5.06	0.63	0.41	
PREDICTED: Xenopus (Silurana) tropicalis tanabin-like (LOC02098609) mRNA	NM_002945032	3.08	6.3	1.04	1.75	2.34	0.43	0.41	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC01730439 (LOC01730439) mRNA	NM_004910303.1	1.49	3.02	1.03	1.28	1.71	0.42	0.41	
Xenopus laevis deg05 mRNA complete sequence	DQ096846.1	1.14	4.37	1.95	0.58	1.01	0.81	0.42	
Xenopus (Silurana) kinesin family member 5A (kif5a) mRNA	NM_0011010745.1	1.56	4.45	1.52	1.27	1.97	0.64	0.42	
Xenopus tropicalis finished cDNA clone Theuds2k8	CR760018.2	1.23	5	2.04	0.83	1.5	0.86	0.42	

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DEX <sup>2</sup>	FC Six1+Eya1 CHX+DEX <sup>2</sup>	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Rank <sup>6</sup>
Xenopus laevis RNA binding motif protein 24 (rbm24-b) mRNA	NM_001086109.1	2.51	5.43	1.12	1.84	2.54	0.47	0.42
PREDICTED: Xenopus (Silurana) tropicalis ankyrin repeat and MYND domain containing 2 (ankrm72) transcript variant X1 mRNA	XM_002933303.2	22.13	49.02	1.15	5.57	7.78	0.49	0.42
Xenopus laevis calcitonin receptor-like (calcrl) mRNA	NM_001086737.1	0.94	2.82	1.59	0.68	1.09	0.7	0.44
PREDICTED: Xenopus (Silurana) tropicalis poliovirus receptor-related protein 1 (pvr1) mRNA	XM_002940925.2	0.86	4.31	2.33	2.38	4.84	1.03	0.45
Xenopus laevis serine dehydratase-like (sds1) mRNA	NM_001086679.1	0.43	1.23	1.53	0.97	1.55	0.68	0.45
Xenopus laevis carbohydrate (N-acetylglucosamine 6-O) sulfotransferase 7 (chst7) mRNA	NM_0010966351.1	2.81	6.95	1.31	4.36	6.52	0.59	0.45
PREDICTED: Xenopus (Silurana) tropicalis putative UDP-GlcNAc-betaGal beta-1,3-N-acetylglicosaminyltransferase LOC02377-like	XM_002935226.2	1.43	5.69	2	2566.55	4825.17	0.92	0.46
PREDICTED: Xenopus (Silurana) tropicalis T-box 15 (tbx15) mRNA	XM_002940981.2	2.57	8.73	1.77	0.79	1.39	0.81	0.46
Xenopus laevis aminoacyl-ribosidic ester synthase 2 (alas2) nuclear gene encoding mitochondrial protein mRNA	NM_001094030.1	0.88	2.2	1.33	1.75	2.67	0.61	0.46
Xenopus laevis cDNA clone IMAGE:5511657	BC083033.1	0.23	2.31	3.39	0.46	1.36	1.57	0.47
Xenopus (Silurana) tropicalis ectodysplasin A receptor (edar) mRNA	NM_001006819.1	0.15	1.33	3.24	0.39	1.13	1.53	0.48
PREDICTED: Xenopus (Silurana) tropicalis WD repeat domain 27 (wir27) mRNA	XM_002933151.2	11.09	23.06	1.06	1.26	1.78	0.51	0.48
Xenopus (Silurana) tropicalis tumor necrosis factor superfamily member 21 (tnfrsf21) mRNA	NM_001079136.1	4.51	10	1.16	2.95	4.39	0.58	0.5
PREDICTED: Xenopus (Silurana) tropicalis etn homologous factor (ethf) mRNA	XM_002937289.2	1.09	3.52	1.71	1.41	2.54	0.86	0.51
Xenopus laevis DBB1 and CUL4 associated factor 17 (dcif17) mRNA	NM_001096802.1	15.03	41.27	1.46	4.31	7.17	0.74	0.51
XLU08408 Xenopus laevis arginase 3 mRNA complete cds	U08408.1	1.16	3.85	1.74	1.06	1.96	0.89	0.51
Xenopus tropicalis cDNA clone IMAGE:7593686	BC168516.1	6.74	35.56	2.41	1.18	2.75	1.23	0.52
Xenopus tropicalis cDNA clone IMAGE:7648616	BC136038.1	3.55	9.61	1.44	0.95	1.58	0.74	0.52
PREDICTED: Xenopus (Silurana) tropicalis chaotic cation transport regulator homolog 1 (E. coli) (chaet1) mRNA	XM_00293546.2	8.59	24.08	1.49	6.52	11.18	0.78	0.53
Xenopus laevis mRNA for thimet oligopeptidase complete cds	AB030904.1	0.34	1.27	1.91	3.38	6.77	1.01	0.53
PREDICTED: Xenopus (Silurana) tropicalis putative N-acetylmuramylpeptidase 16-like (LOC10090742) mRNA	XM_002941383.1	0.79	2.4	1.62	0.82	1.48	0.86	0.53
Xenopus laevis MGC79035 protein (MGC79035) mRNA	NM_001093448.1	1.69	8.11	2.27	0.66	1.54	1.24	0.55
Xenopus laevis Dnat (Hsp40) homolog subfamily C member 27 (dnac27-b) mRNA	NM_001095422.1	2.2	4.7	1.1	1.35	2.04	0.6	0.55
Xenopus (Silurana) tropicalis SRV (sex-determining region Y)-box 1 (sox1) mRNA	NM_001080996.1	1.1	2.42	1.15	1.94	3	0.63	0.55
Xenopus (Silurana) tropicalis atlastin GTPase 1 (atl1) mRNA	NM_0010738754.1	0.96	2.97	1.64	0.66	1.22	0.9	0.55

<sup>1</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1+Eya1 and CHX treatment

<sup>2</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1+Eya1 and CHX+DEX treatment

<sup>3</sup>Log<sub>2</sub> Fold change values for Six1+Eya1

<sup>4</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup>Expression levels (FPKM) in un-injected pre-placodal explants after injection of Six1+Eya1 and CHX+DEX treatment

<sup>6</sup>Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup>Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Six1+Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## E.6 Six1+Eya1; Down-regulated genes

**Table E.6** Genes with at least two-fold down-regulation after injection of Six1-GR+Eya1-GR and treatment with CHX + DEX

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DEX <sup>a</sup>	FC Six1+Eya1 <sup>b</sup>	Control CHX	Control CHX+DEX <sup>c</sup>	FC Control CHX+DEX <sup>c</sup>	Rank <sup>d</sup>
Xenopus laevis cardiac troponin T mRNA complete cds alternatively spliced	AF467919.1	19.33	0.01	-14.24	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox protein zampogna-like (LOC100485176) mRNA	XM_002934132.2	4.06	0.01	-11.99	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis desmoglein 1 (dsy4) mRNA	AY362041.1	3.46	0.01	-11.76	0	0	0	0
Xenopus (Silurana) manganese superoxide dismutase mRNA complete cds	NM_001011234.1	2.92	0.01	-11.51	0	0	0	0
Xenopus (Silurana) tropicalis lipoic acid synthetase (las) nuclear gene encoding mitochondrial protein mRNA	NM_001079059.1	2.75	0.01	-11.43	0	0	0	0
Xenopus laevis hepatocyte nuclear factor 4 alpha (hnf4a) mRNA	NM_001086601.1	2.69	0.01	-11.39	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis protein phosphatase EF-hand calcium binding domain 1 (pepf1) mRNA	XM_002936047.2	2.63	0.01	-11.36	0	0	0	0
Xenopus tropicalis finished DNA clone Thel1.15h01	CR760212.2	2.08	0.01	-11.03	1.31	1.46	0.16	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100494519 (LOC100494519) transcript variant X1 mRNA	XM_002942124.2	1.72	0.01	-10.75	0	0	0	0
Xenopus (Silurana) tropicalis RAB35A member RAS oncogene family (rab33a) mRNA	NM_001079059.1	1.67	0.01	-10.71	0	0	0	0
Xenopus tropicalis finished cDNA clone TEG143019	CR848438.2	1.56	0.01	-10.61	0	0	0	0
Xenopus laevis ankyrin repeat and SOCS box containing 5 (asrb5) mRNA	NM_001093879.1	1.56	0.01	-10.61	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis septin 3 (sep13) transcript variant X1 mRNA	XM_002934722.2	1.5	0.01	-10.55	0	0	0	0
Xenopus tropicalis finished cDNA clone Tcas128C14	CR942568.2	1.44	0.01	-10.49	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis myosin H (myo1) mRNA	XM_002932063.2	1.29	0.01	-10.34	0	0	0	0
Xenopus laevis unchartered protein MGc8345 (MGc8345) mRNA	NM_001090093.1	1.23	0.01	-10.26	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100488459 (LOC100488459) mRNA	XM_002934414.2	1.07	0.01	-10.07	0	0	0	0
Xenopus laevis unchartered LOC493561 (LOC493561) mRNA	NM_001095024.1	39.83	0.55	-6.2	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis sterol 26-hydroxylase mitochondrial-like (LOC100496109) mRNA	XM_002933939.2	1.51	0.04	-5.55	0	0	0	0
Xenopus laevis chromosome 10 open reading frame 11 (cl.00r11) mRNA	NM_001096630.1	9.76	0.24	-5.39	0	0	0	0
Xenopus laevis transient receptor potential cation channel subfamily M member 8 (TRPM8) mRNA complete cds	FJ608757.1	26.97	0.89	-4.94	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis unchartered LOC100496109 (LOC100496109) mRNA	XM_002935750.2	13.14	0.52	-4.68	0	0	0	0
Xenopus laevis unchartered LOC493564 (LOC493564) mRNA	XM_002936544.2	5.38	0.29	-4.25	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 135 (tmem135) transcript variant X1 mRNA	NM_001092444.1	242.09	15.15	-4	16.26	18.82	0.22	0
Xenopus laevis chimerin 2 (chr2) mRNA	FJ6784	29.86	4.26	-46.09	63.53	0.47	0	0
Xenopus (Silurana) tropicalis synapsin 2 (cps2) mRNA	NM_001012579.1	6.9	0.44	-3.99	0	0	0	0
Xenopus (Silurana) tropicalis homeobox C9 (hexc9) mRNA	NM_001012756.2	6.9	0.44	-3.99	0	0	0	0
Xenopus tropicalis tumor necrosis factor receptor superfamily member 6 (fas) mRNA complete cds	EF555573.1	1.82	0.12	-3.99	0	0	0	0
Xenopus laevis mRNA for PVL (prepro-PGLa) precursor from skin	X01824.1	1011.64	64.25	-3.98	0	0	0	0
Xenopus laevis solute carrier family 28 (sodium-coupled nucleoside transporter) member 2 (slc28a2) mRNA	NM_001093313.1	1.36	0.1	-3.81	0	0	0	0
Xenopus laevis cDNA clone IMAGE:55206734	BC080026.1	1.16	0.09	-3.73	0	0	0	0
Xenopus laevis chimerin 1 (chr1) mRNA	NM_00113.1307.1	1.23	0.1	-3.71	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis myosin IIb (myo2b) mRNA	XM_002934652.1	1.04	0.09	-3.69	0	0	0	0
Xenopus laevis phosphodiesterase D family member 6 (pde6) nuclear gene encoding mitochondrial protein mRNA	NM_001016875.3	3.46	0.27	-3.69	0	0	0	0
XL63818-Xenopus laevis RING finger protein mRNA complete cds	U63818.1	263.32	20.8	-3.67	11.42	18.78	0.72	0
Xenopus tropicalis partial mRNA for double-stranded RNA activated protein kinase 3 (pk3 gene)	AVR850092.1	3.4	0.28	-3.64	0	0	0	0
Xenopus laevis lactate dehydrogenase B (ldhb-b) mRNA	NM_001087581.1	1.45	0.13	-3.58	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis integrin beta 8 (itgb8) mRNA	XM_002933298.2	2.26	0.19	-3.58	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis dimethylalanine monooxygenase [N-oxide-forming] 2-like (LOC100489107) mRNA	XM_002934145.2	11.89	1.01	-3.56	0	0	0	0
Xenopus laevis bowline2 mRNA for Bowline2 partial cds	A9300353.1	2.86	0.25	-3.54	0	0	0	0
Xenopus (Silurana) tropicalis kinesin family member 18A (kif18a) mRNA	NM_001016245.3	1.68	0.15	-3.49	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis Pr domain containing 13 (prdm13) mRNA	XM_002932160.2	1.02	0.1	-3.48	0	0	0	0
Xenopus laevis transmembrane protease serine 15 (tpnss15) mRNA	NM_001095642.1	1.55	0.15	-3.45	1.3	1.76	0.44	0
Xenopus laevis CR3 short transcript variant (CR3) mRNA complete cds alternatively spliced	AY796188.1	1.24	0.12	-3.44	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis hemacentrin-1 (hmcn1) mRNA	XM_00293766.2	1.55	0.15	-3.44	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 108 (tmem108) transcript variant X2 mRNA	XM_004915366.1	1.28	0.13	-3.4	0	0	0	0
Xenopus (Silurana) tropicalis chloride channel voltage-sensitive 3 (clcn3) mRNA	NM_001142088.1	1.01	0.1	-3.24	0	0	0	0

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DEX	FC Six1+Eva1 CHX	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Control CHX	FC Control CHX	Rank <sup>a</sup>
PREDICTED: Xenopus (Silurana) tropicalis coordinator of PRMT5 and differentiation stimulator-like (LOC100490247) mRNA	XM_002932712.2	1.44	0.16	-3.25	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TTbA49J122	CU025137.1	13.84	1.5	-3.22	0	0	0	0	0	0
Xenopus laevis checkpoint with forkhead and ring finger domains E3 ubiquitin protein ligase (chfr) mRNA	NM_001095771.1	3939322	426.57	-3.21	256.13	323.57	0.34	0	0	0
Xenopus (Silurana) tropicalis CD40 receptor superfamily member 5 (cd40) mRNA	NM_004917368.1	2.39	0.28	-3.12	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tumor necrosis factor (llegnd) superfamily member 8 (tnfsf8) mRNA	XM_002935385.2	206.06	25.08	-3.11	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tumor necrosis factor 5 (dok5) mRNA	AF253504.1	1.7	0.21	-3.04	0	0	0	0	0	0
Xenopus (Silurana) tropicalis homeodomain protein dbx mRNA complete cds	NM_00126696.1	3.35	0.43	-3.04	0	0	0	0	0	0
Xenopus (Silurana) tropicalis beta-1,4-N-acetyl-beta-D-galactosaminyl transferase 1 (b4galnt1) mRNA	NM_001095129.1	24.03	3.12	-2.98	0	0	0	0	0	0
Xenopus laevis N-myc (and STA1) interactor (nmi) mRNA	NM_001126775.1	100.93	13.25	-2.95	0	0	0	0	0	0
Xenopus (Silurana) tropicalis family with sequence similarity_192 member A (fam192a) mRNA	BC161185.1	1.37	0.21	-2.76	0	0	0	0	0	0
Xenopus tropicalis hypothetical protein LOC100145515 mRNA (cDNA clone IMAGE:7864048) partial cds	XM_002934142.2	2.63	0.4	-2.75	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis vascular cell adhesion molecule 1 (vcam1) mRNA	XM_002938074.2	2.67	0.4	-2.75	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis nuclear transcription factor Y beta (nfyb) transcript variant X3 mRNA	NM_001130384.1	1.34	0.21	-2.74	0	0	0	0	0	0
Xenopus (Silurana) tropicalis uncharacterized LOC10170623 (LOC100170623) mRNA	CR848409.2	13.02	2.05	-2.68	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone Theta76f616	BC154996.1	8.07	1.3	-2.64	2.19	2.39	0.14	0	0	0
Xenopus laevis cDNA clone IMAGE:8322189 complete cds	BC123277.1	1.71	0.29	-2.61	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis membrane-associated ring finger (C3HC4) 5 (march5) mRNA	XM_00293611.2	1.46	0.25	-2.59	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zinc finger 2 (fezf2) mRNA	NM_001095644.1	5.78	0.98	-2.57	3.56	3.74	0.08	0	0	0
Xenopus tropicalis finished DNA clone Theta76f616	CR842619.2	3.62	0.63	-2.53	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:8322189	NM_0014431.1	1.54	0.27	-2.52	0	0	0	0	0	0
Xenopus laevis homeobox protein mRNA (cDNA clone MGCI54555 IMAGE:7864048) partial cds	NM_001092862.1	711.19	126.2	-2.5	81.65	103.99	0.35	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis adenomatous polyposis coli (apc) transcript variant XI mRNA	NM_00123478.1	1.39	0.26	-2.46	0	0	0	0	0	0
Xenopus laevis FEZ family zinc finger 2 (fezf2) mRNA	NM_001091468.1	3.45	0.64	-2.44	0	0	0	0	0	0
Xenopus tropicalis finished DNA clone Theta76f616	U28370.1	1.64	0.31	-2.44	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis protein phosphatase Mg <sup>2+</sup> /Mn <sup>2+</sup> -dependent 1L (ppml1) transcript variant X1 mRNA	NM_0014431.1	1.54	0.27	-2.44	0	0	0	0	0	0
Xenopus laevis KIAA1109 (kiaa1109) mRNA	NM_001092862.1	711.19	126.2	-2.43	81.65	103.99	0.35	0	0	0
Xenopus (Silurana) tropicalis membrane-associated ring finger (C3HC4) 5 (march5) mRNA	NM_00123478.1	1.39	0.26	-2.44	0	0	0	0	0	0
Xenopus laevis tubulin alpha 1a (tuba1a-a) mRNA	NM_001091468.1	3.45	0.64	-2.44	0	0	0	0	0	0
XL28370: Xenopus laevis homeobox protein (XANF2) mRNA complete cds	U28370.1	1.64	0.31	-2.44	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis anchor protein 7 isoforms: alpha and beta-like (LOC10096426) transcript variant X2 mRNA	NM_001204059.1	1.57	0.3	-2.44	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:5514602	XM_002936206.2	1006.2	188.02	-2.43	111.19	133.9	0.32	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis FERM and PDZ domain containing 4 (frmpd4) mRNA	XM_002933807.2	1.15	0.22	-2.41	0	0	0	0	0	0
Xenopus laevis yak-1 mRNA for adult keratin YAK-1 complete cds	AB045600.1	1.16	0.22	-2.41	0	0	0	0	0	0
Xenopus (Silurana) tropicalis complement component 5 (c5) mRNA	NM_001204059.1	132.72	25.38	-2.39	2.25	3.21	0.52	0	0	0
Xenopus tropicalis finished DNA clone TTbA455m12	CU075365.1	4.7	0.91	-2.39	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC46919 mRNA (cDNA clone IMAGE:6873809) partial cds	BC087617.1	1.51	0.29	-2.39	0	0	0	0	0	0
Xenopus laevis family with sequence similarity_22 member B (fam22b) mRNA	NM_001094184.1	523.94	101.25	-2.38	47.69	107.48	1.18	0	0	0
Xenopus laevis cDNA clone MGCI31018 (IMAGE:7979650) complete cds	BC108553.1	122.53	23.9	-2.36	38.03	46.4	0.29	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis myotubularin related protein 11 (mtmr11) mRNA	XM_002935174.2	1.52	0.3	-2.35	0	0	0	0	0	0
Xenopus laevis tyrosine phosphatase receptor type N polypeptide 2 (ptprn2) mRNA	AY937405.1	1.25	0.25	-2.33	0	0	0	0	0	0
Xenopus laevis egg envelope glycoprotein (ZP1) mRNA partial cds	NM_001128639.1	1.48	0.31	-2.28	0	0	0	0	0	0
Xenopus (Silurana) tropicalis leucine rich repeat neuronal 4 (lrn4) mRNA	BC078490.1	1.51	0.29	-2.31	0	0	0	0	0	0
Xenopus laevis phosphodiesterase 5a (GMP-AMP) nucleotide AMP-activated protein kinase regulatory subunit 1 mRNA	NM_001094184.1	523.94	101.25	-2.31	5.54	6.46	0.23	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis myotubularin related protein 11 (mtmr11) mRNA	BC108553.1	122.53	23.9	-2.31	5.57	1.23	0.07	0	0	0
Xenopus laevis hypothetical protein LOC100137614 mRNA (cDNA clone IMAGE:6998652) complete cds	BC074469.1	710.87	145.95	-2.29	88.57	104.43	0.19	0	0	0
Xenopus (Silurana) tropicalis par-6 partitioning defective 6 homolog beta (par6b) mRNA	NM_001128639.1	1.48	0.31	-2.28	0	0	0	0	0	0
Xenopus (Silurana) tropicalis 5-hydroxytryptamine (serotonin) receptor 5A G protein-coupled (hrt5a) mRNA	XM_002932497.2	1.91	0.32	-2.26	0	0	0	0	0	0
Xenopus laevis phosphodiesterase 5a (GMP-AMP) nucleotide AMP-activated protein kinase regulatory subunit 1 mRNA	NM_001094802.1	75.02	15.24	-2.25	1.31	1.49	0.19	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transcript variant X4 mRNA	NM_0010947340.1	2.79	0.57	-2.24	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC100137614 mRNA (cDNA clone IMAGE:6998652) complete cds	BC07805.1	1.1	0.24	-2.24	0	0	0	0	0	0
Xenopus (Silurana) tropicalis transcription factor Rfx4-like (LOC101732419) mRNA	NM_004913128.1	1.37	0.3	-2.22	0	0	0	0	0	0
Xenopus laevis retina and anterior neural fold homeobox (rax-b) mRNA	NM_001098220.1	2.17	0.47	-2.22	2.42	3.76	0.64	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis chromaffin granule amine transporter like (LOC100495897) transcript variant X1 mRNA	NM_004912333.1	1.39	0.31	-2.18	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor family C group 5 member C (gprc5c) transcript variant X4 mRNA	NM_004916328.1	1.23	0.27	-2.24	0	0	0	0	0	0
Xenopus tropicalis ubiquitin specific peptidase 13 (sopeptidase T-3) mRNA (cDNA clone MGCI108005 IMAGE:6998652) complete cds	BC07805.1	1.1	0.24	-2.24	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis par-6 partitioning defective 6 homolog beta (par6b) mRNA	NM_004913128.1	1.37	0.3	-2.22	0	0	0	0	0	0
Xenopus laevis retina and anterior neural fold homeobox (rax-b) mRNA	NM_001098220.1	2.17	0.47	-2.22	2.42	3.76	0.64	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis kinase C delta type-like (LOC101730464) mRNA	NM_004912333.1	1.39	0.31	-2.18	0	0	0	0	0	0
Xenopus laevis tudor domain containing 5 (tdrd5) mRNA	NM_001097130.1	8.66	1.94	-2.17	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:5154485 (IMAGE:8318484) complete cds	BC12327.1	2.29	0.53	-2.12	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:15156571 (partial cds)	BC073207.1	4.91	1.15	-2.11	2.71	2.94	0.12	0	0	0

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DEX	FC Six1+Eya1 CHX	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Control CHX	Control CHX+DEX	FC Control CHX	Rank <sup>a</sup>
Xenopus (Silurana) tropicalis KIAA0146 (Kiaa0146) mRNA	NM_001142908.1	107.53	25.11	-2.1	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis potassium voltage-gated channel subfamily H (eag-related) member 7 (kcnh7) mRNA	XM_002933260.2	4.03	0.94	-2.1	0	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:4202370 containing frame-shift errors	BC061937.1	1838.6	430.02	-2.1	55.12	260.73	2.25	0	0	0	0
PREDICTED: Xenopus (Silurana) K+ transporting ATPase interacting 2 (nkain2) mRNA	XM_002936008.2	1.02	0.24	-2.1	0	0	0	0	0	0	0
Xenopus laevis mRNA for imo4 protein (Imo4 gene)	A1511277.1	52.85	12.4	-2.1	15.5	18.11	0.23	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis lymphoidlastic leukemia derived sequence 1 (yl1) transcript variant X2 mRNA	XM_004918902.1	1.3	0.31	-2.09	0	0	0	0	0	0	0
Xenopus laevis mRNA for imo4 protein (Imo4 gene)	XM_004916041.1	457.08	108.21	-2.08	3.16	8.14	1.37	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733049 (LOC101733049) mRNA	AB016717.1	1.14	0.27	-2.08	0	0	0	0	0	0	0
Xenopus laevis cDNA clone for aldehyde dehydrogenase class 1 complete cds	BC085199.1	958.98	232.3	-2.05	31.78	46.91	0.57	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis integrin alpha 2 (CD49B) alpha 2 subunit of VLA-2 receptor (Itga2) mRNA	XM_002933043.2	2.21	0.54	-2.05	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis peroxisome proliferator-activated receptor alpha-like (LOC100497346) mRNA	XM_002930738.2	1021.26	249.37	-2.04	73.79	81.5	0.15	0	0	0	0
Xenopus tropicalis finished cDNA clone TGas107e13	CR848617.1	88.03	21.68	-2.03	0	0	0	0	0	0	0
Xenopus laevis jifaya (jy) mRNA	NM_001195095.1	2.67	0.66	-2.02	1.51	1.54	0.04	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis c-X-C motif chemokine 10-like (LOC100490277) mRNA	XM_002940775.1	6.43	1.59	-2.02	1.21	1.69	0.49	0	0	0	0
Xenopus laevis uncharacterized protein MGCG4526 (MGCG4526) mRNA	NM_001086455.1	1.16	0.29	-2.02	0	0	0	0	0	0	0
Xenopus laevis SEC13 homolog (Sec13) mRNA	NM_001086973.1	7163.9	1782.78	-2.01	198.79	804.95	2.02	0	0	0	0
Xenopus (Silurana) tropicalis glutathione synthetase (Gss) mRNA	NM_001008044.1	1.03	0.26	-2.01	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis phosphopantethenylycysteine synthetase (ppcs) mRNA	NM_001079164.1	1.14	0.29	-2	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis CtpA caspaseolytic peptidase B homolog (ctpB) mRNA	XM_002940744.2	75.63	19.15	-1.99	8.43	10.12	0.27	0	0	0	0
Xenopus (Silurana) tropicalis cartilage acidic protein 1 (CRTAC-1) mRNA complete cds	DO2551712.1	1.96	0.5	-1.98	0	0	0	0	0	0	0
Xenopus laevis solute carrier family 12 (potassium/chloride transporter) member 9 (slc12a9) mRNA	NM_001086782.1	41.26	10.62	-1.96	4.81	6.75	0.5	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis phosphotransferase-like protein (LARGE1-like) (LOC100495555) partial mRNA	XM_002943048.2	1.08	0.29	-1.94	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis SET and MYND domain containing 3 (syndy3) transcript variant X2 mRNA	XM_004914626.1	1.04	0.28	-1.93	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis substrate 1 (polk1s1) transcript variant 1 mRNA	XM_002939634.2	2.12	0.56	-1.93	0	0	0	0	0	0	0
Xenopus laevis dipeptidase E mRNA (cDNA clone IMAGE:7542807) complete cds	BC167317.1	1.15	0.31	-1.92	0	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.91	4.04	8.07	1	0	0	0	0
Xenopus laevis transporter associated with antigen processing 1 (TAP1) mRNA TAP1 allele partial cds	AY204553.1	1.37	0.37	-1.9	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis glycosyltransferase-like protein LOC446960 mRNA (cDNA clone IMAGE:7203732) partial cds	BC082220.1	1.66	0.46	-1.87	0	0	0	0	0	0	0
Xenopus laevis tumor protein 053 inducible protein 11 (tp53i11) mRNA	NM_001097917.1	91.43	25.47	-1.85	12.96	16.28	0.33	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis polypeptide kinase 1.35325 (IMAGE:135325) transcript variant 1 mRNA	XM_004914728.1	1.11	0.32	-1.84	0	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC167317.1	43.35	11.62	-1.83	4.04	8.07	1	0	0	0	0
Xenopus laevis transporter associated with antigen processing 1 (TAP1) mRNA TAP1 allele partial cds	BC1417650.1	43.35	11.62	-1.83	4.04	8.07	1	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis SET and MYND domain containing 3 (syndy3) transcript variant X2 mRNA	XM_002943048.2	1.08	0.29	-1.82	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis substrate 1 (polk1s1) transcript variant 1 mRNA	XM_002939634.2	2.12	0.56	-1.82	0	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.81	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC446960 mRNA (cDNA clone IMAGE:7203732) partial cds	BC082220.1	1.66	0.46	-1.81	0	0	0	0	0	0	0
Xenopus laevis tumor protein 053 inducible protein 11 (tp53i11) mRNA	NM_001097917.1	91.43	25.47	-1.85	12.96	16.28	0.33	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis chain-like (LOC100495918) mRNA	XM_004914728.1	1.11	0.32	-1.84	0	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.83	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.82	4.04	8.07	1	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis chain-like (LOC100495918) transcript variant X1 mRNA	XM_004914728.1	1.11	0.32	-1.82	0	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC446960 mRNA (cDNA clone IMAGE:7203732) partial cds	BC082220.1	1.66	0.46	-1.81	0	0	0	0	0	0	0
Xenopus laevis tumor protein 053 inducible protein 11 (tp53i11) mRNA	NM_001097917.1	91.43	25.47	-1.85	12.96	16.28	0.33	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis chain-like (LOC100495918) mRNA	XM_004914728.1	1.11	0.32	-1.84	0	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.83	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.82	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.81	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.80	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.79	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.78	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.77	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.76	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.75	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.74	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.73	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.72	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.71	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.70	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.69	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.68	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.67	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.66	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.65	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.64	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.63	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.62	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.61	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.60	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.59	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.58	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.57	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.56	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.55	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.54	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.53	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.52	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.51	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.50	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.49	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.48	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11.62	-1.47	4.04	8.07	1	0	0	0	0
Xenopus laevis hypothetical protein LOC10009766 mRNA (cDNA clone IMAGE:8024638) partial cds	BC1417650.1	43.35	11								

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DEX	FC Six1+Eva1 CHX	FC Six1+Eva1 CHX+DEX	Control CHX	Control CHX+DEX	FC Control CHX	FC Control CHX+DEX	Rank <sup>a</sup>
Xenopus laevis partial mRNA for lysyl oxidase-like 3 (foxl3 gene)	AM040262.2	65.19	20.35	-1.68	0	0	0	0	0	0
Xenopus laevis Sh3 domain and leucine-rich repeats 1 (sh3rlc1) mRNA	NM_001095875.1	10.23	3.2	-1.68	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis cytoplasmic polyadenylation element binding protein 2 (cpeb2) transcript variant X2 mRNA	XM_004911230.1	278.13	87.43	-1.67	77.25	79.18	0.04	0	0	0
Xenopus laevis crystallin alpha A mRNA (cdna clone MGC:19657 / IMAGE:909632) complete cds	BC170250.1	3.63	1.15	-1.67	0	0	0	0	0	0
Xenopus laevis uncharacterized protein MGC8961 (MGC68961) mRNA	NM_001086495.1	2.62	0.83	-1.66	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TTPA048d11	CU75859.1	1.23	0.39	-1.66	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734067 (LOC101734067) mRNA	XM_004920793.1	3.55	1.14	-1.64	0	0	0	0	0	0
Xenopus laevis transcription factor (saf) mRNA safa-A allele complete cds	AY508933.1	9.65	3.12	-1.64	2.91	3.66	0.34	0	0	0
Xenopus laevis peroxidase 2 (POX2) mRNA complete cds	AY056992.2	4.57	1.48	-1.63	0	0	0	0	0	0
Xenopus tropicalis finished cDNA clone TTPA022g13	CR926448.2	932.18	302.24	-1.63	6.65	8.12	0.29	0	0	0
Xenopus laevis nuclear apoptosis inducing factor 1 (naf1) mRNA	NM_001096902.1	100.63	32.83	-1.62	18.68	50.64	1.44	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis regulator of cell cycle RGCC-like (LOC100495028) mRNA	XM_0029939201.2	1.51	0.5	-1.62	0	0	0	0	0	0
Xenopus (Silurana) tropicalis syntaxin 2 (stx2) mRNA	NM_001079176.1	1.39	0.46	-1.62	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis poly (ADP-ribose) polymerase 12-like (LOC100490338) mRNA	XM_002942857.2	1.52	0.5	-1.62	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis wingless-type MMTV integration site family member 8B (wn8b) transcript variant X1 mRNA	XM_002936348.2	1.24	0.41	-1.62	0	0	0	0	0	0
Xenopus (Silurana) tropicalis gameteigenin binding protein 2 (gbnp2) mRNA	NM_001006808.1	1.12	0.37	-1.61	0	0	0	0	0	0
Xenopus (Silurana) tropicalis adenosinomycinesteinase-like 2 (alynl2) mRNA	NM_001016409.2	2.12	0.7	-1.61	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis upstream binding transcription factor RNA polymerase I (ubrif) transcript variant X1 mRNA	XM_002935336.2	147.71	48.79	-1.6	26.15	31.52	0.27	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101732516 (LOC101732516) mRNA	XM_004913167.1	1.3	0.43	-1.6	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC43671 mRNA (cDNA clone IMAGE:5512708) partial cds	BG073592.1	2.42	0.81	-1.59	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis upf-Gal1betaGalNac beta 1,3-galactosidase transferase poly peptide 4 (bg3gal4) mRNA	XW_002938685.2	1.36	0.46	-1.59	0	0	0	0	0	0
Xenopus laevis phosphoribosylglycamide synthetase (gart)	NM_001099882.1	546.33	183.96	-1.58	26.53	27.77	0.07	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis poly (ADP-ribose) polymerase family member 4 (parp4) mRNA	XM_002937966.2	8.78	2.99	-1.56	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis brain-specific angiogenesis inhibitor 2 (baii2) transcript variant X2 mRNA	XM_004919220.1	2.09	0.72	-1.56	1.1	1.36	0.31	0	0	0
Xenopus laevis Ras association (RalGDS/AF-6) domain family (N-terminal) member (N-terminal) member 7 (rasrf7) mRNA	NM_001093161.1	1.85	0.64	-1.55	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis interferon gamma receptor 2-like (LOC100482598) transcript variant X2 mRNA	XW_004912129.1	9.04	3.1	-1.55	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis embryonic protein Uvs2-like (LOC100496122) transcript variant X1 mRNA	XM_002937302.2	5.75	1.98	-1.55	4.08	5.46	0.43	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis chromosome X open reading frame 58 (xorf58) mRNA	XM_00293589.2	125.8	43.25	-1.55	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC73228 mRNA (cDNA clone IMAGE:5512944)	BC123324.1	2.47	0.85	-1.55	4.85	5.37	0.15	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis terminal uridylyl transferase 1 U6 snRNA-specific (utrl) mRNA	XM_002941456.2	1.19	0.42	-1.54	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00487518 (LOC100487518) mRNA	XW_002938497.2	1.03	0.36	-1.53	1.42	1.7	0.26	0	0	0
Xenopus laevis transcription adaptor 2a (tad2a) mRNA	NM_001094008.1	3.65	1.27	-1.53	3.35	3.89	0.22	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis NXP6 family member 4-like (LOC101734002) mRNA	XM_004920234.1	1.01	0.36	-1.51	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis extracellular calcium-sensing receptor-like (LOC100492511) mRNA	XM_002940813.2	23.5	8.29	-1.51	7.63	16.85	1.15	0	0	0
Xenopus laevis cDNA clone IMAGE:6559127	BC084402.1	1594.78	562.92	-1.51	11.065	27.959	1.34	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis terminal uridylyl transferase 1B histone chaperone (asfb) mRNA	NM_001086841.1	53.49	18.89	-1.51	11.05	17.18	0.64	0	0	0
Xenopus laevis anti-silencing function 1B (asfb) mRNA	NM_001102967.1	1.42	0.51	-1.5	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis LIM-only protein LMO-2 mRNA	AF374473.1	4.09	1.46	-1.5	1.19	2.3	0.95	0	0	0
Xenopus laevis XHR1-A mRNA for HES-related 1A' complete cds	AB071433.1	4.99	1.79	-1.49	3.27	6.08	0.9	0	0	0
Xenopus tropicalis cDNA clone IMAGE:65978032	BC084737.1	2.42	0.88	-1.48	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis matrix metalloproteinase-23-like (LOC100488482) transcript variant X2 mRNA	XM_004914394.1	2.7	0.98	-1.47	1.53	1.85	0.28	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis serine/arginine repetitive matrix 3 (srm3) mRNA	XM_002933830.2	597.43	217.23	-1.46	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis tetrapsin 12 (tspan12) transcript variant X2 mRNA	XM_004912984.1	8.31	3.03	-1.46	1.09	1.35	0.31	0	0	0
Xenopus laevis cDNA clone IMAGE:5511680 partial cds	BC097628.1	196.58	71.95	-1.46	5.49	5.98	0.13	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis thyrotropin subunit beta-like (LOC100496349) transcript variant X2 mRNA	XM_004910716.1	1.34	0.5	-1.45	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis unc-5 homolog D (cel-1) mRNA	XM_002932956.2	1.03	0.38	-1.45	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis E1A binding protein p300 (ep300) transcript variant X2 mRNA	XM_004913798.1	538.7	198.29	-1.45	41.79	48.56	0.22	0	0	0
Xenopus laevis tau-like protein-1 (ktb) mRNA complete cds alternatively spliced	AY032847.1	3.91	1.44	-1.45	1.33	1.42	0.1	0	0	0
Xenopus (Silurana) tropicalis uncharacterized LOC10045501 mRNA	NM_001126945.1	1.3	0.48	-1.44	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis peroxidase 4-like (LOC100491505) mRNA	XM_002937967.2	15.71	5.82	-1.44	0	0	0	0	0	0

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DEX	FC Six1+Eva1 CHX*	Control CHX	Control CHX+DEX	FC Control CHX+DEX*	Control CHX*	Control CHX+DEX	FC Control CHX*	Rank <sup>c</sup>
Xenopus laevis chromosome 11 open reading frame 65 (c11orf65) mRNA	NM_001093524.1	9.96	3.7	-1.44	4.37	5.2	0.26	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734353 (LOC101734353) mRNA	XM_004918777.1	31.28	11.62	-1.43	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100493999 (LOC100493999) transcript variant X2 mRNA	NM_004915901.1	1.16	0.43	-1.43	3.16	4.06	0.37	0	0	0	0
Xenopus laevis hypothetical protein LOC10137710 mRNA (DNA clone IMAGE:4030892) partial cds	BC157465.1	1.56	0.58	-1.43	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis GS homeobox 1 (gsex1) mRNA	NM_001095789.1	2.83	1.05	-1.43	0	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:3401855	BC084599.1	84.76	31.56	-1.43	3201.32	158456.62	2.31	0	0	0	0
Xenopus laevis allograft inflammatory factor 1-like (aif1) mRNA	NM_001092679.1	1.03	0.39	-1.43	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis caspase recruitment domain family member 14 (card14) transcript variant X1 mRNA	XM_002937309.2	1.05	0.4	-1.42	0	0	0	0	0	0	0
Xenopus tropicalis finished DNA clone TEG003m08	CT025197.2	62.73	23.48	-1.42	15.43	16.28	0.08	0	0	0	0
Xenopus laevis cDNA clone IMAGE:6348748	BC094199.1	232.19	87.2	-1.42	83.09	95.07	0.2	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis TBC1 (tre2/USP6-BU22 cdc16) domain family member 1 (tbc1d1) transcript variant X2 mRNA	NM_004910931.1	6.26	2.35	-1.42	1.23	2.18	0.83	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis Zic family member 5 (zic5) mRNA	XM_002937082.2	1.97	0.74	-1.42	0	0	0	0	0	0	0
Xenopus laevis polymerase (RNA) III (DNA-directed) polymerase A1.5KDa (polr2a) mRNA	NM_001086833.1	2.38	0.9	-1.41	1.3	1.19	0.55	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis actin binding LIM protein family member 2 (ablim2) mRNA	XM_002938262.2	3.46	1.31	-1.41	1.21	1.49	0.31	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis SH2 domain containing 6 (sh2d6) transcript variant X1 mRNA	NM_004912569.1	1.12	0.43	-1.4	0	0	0	0	0	0	0
Xenopus tropicalis genomic-screened homeobox X (gsh2) mRNA complete cds	DQ1995531.1	2.63	1	-1.4	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis embryonic protein UV5-2-like (LOC100495416) mRNA	XM_002934087.2	10.12	3.86	-1.4	5.51	7.06	0.36	0	0	0	0
Xenopus laevis sulfatase 2 (sulf2) mRNA	NM_001094945.1	9.79	3.74	-1.4	2.98	5.12	0.79	0	0	0	0
Xenopus laevis RNA binding motif protein 23 (rbm23) mRNA	NM_001092339.1	1334.51	509.16	-1.4	156.55	258.74	0.71	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis brain-specific angiogenesis inhibitor 1-associated protein 2-like (LOC100485322) mRNA	XM_002935002.2	2.14	0.82	-1.39	0	0	0	0	0	0	0
Xenopus laevis cathepsin S (ctss-a) mRNA	NM_00108353.1	1.57	0.6	-1.39	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis solute carrier family 35 member E3 (slc35e3) mRNA	NM_001078809.1	1.61	0.62	-1.39	0	0	0	0	0	0	0
Xenopus tropicalis finished DNA clone Theu29g02	CT03608.1	14.74	5.66	-1.39	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis olfactory receptor 51-like (LOC100496897) mRNA	NM_004914310.1	2.47	0.96	-1.38	1.68	1.96	0.23	0	0	0	0
Xenopus laevis uncharacterized protein MGCI31201 (MGCI31201) mRNA	NM_001096283.1	1.3	0.5	-1.38	0	0	0	0	0	0	0
Xenopus laevis recombination activating gene 1a (rag1) mRNA	NM_001172983.1	1.1	0.43	-1.38	0	0	0	0	0	0	0
XLU86699 Xenopus laevis oocyte lectin mRNA complete cds	U86699.1	437.96	169.25	-1.38	39.36	62.57	0.78	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis LINE-1 type transposase domain-containing protein 1-like (LOC100493625) mRNA	XM_0029342620.2	5.15	2	-1.37	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis Sp4 transcription factor (sp4) mRNA	NM_00293296.2	4.99	1.94	-1.37	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis orthodontic protein 1 (otx1) mRNA	NM_203385.1	6.52	2.53	-1.37	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis MGCG9309 protein (MGCG9309) mRNA	NM_001004837.1	133.73	51.87	-1.37	21.25	23.47	0.15	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis low density lipoprotein receptor class A domain containing 2 (ldlrad2) mRNA	XM_002932622.2	1.25	0.49	-1.37	0	0	0	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7741920	BC122065.1	1.52	0.59	-1.37	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis neutral cholesterol ester hydrolase 1 (nche1) mRNA	NM_001098980.1	1.03	0.4	-1.37	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis MGCG82053 protein (MGCG82053) mRNA	NM_001092892.1	1.17	0.46	-1.36	0	0	0	0	0	0	0
Xenopus laevis nutrient peptide receptor type C mRNA complete cds	AF231035.1	2.2	0.87	-1.35	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis retina and anterior neural fold homeobox (rax) mRNA	NM_00293669.2	1.04	0.41	-1.35	0	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:8541893 containing frame-shift errors	BC129668.1	56.49	22.3	-1.35	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis metalloproteinase 3 (bpap3) mRNA	NM_002938415.2	4.61	1.82	-1.34	3.48	3.76	0.12	0	0	0	0
Xenopus (Silurana) tropicalis protocadherin 20 (pcdh20) mRNA	NM_001100230.1	1.28	0.51	-1.34	1.12	1.28	0.2	0	0	0	0
Xenopus laevis RCC1 domain containing 1 (rccl1) mRNA	NM_00109773.1	2.18	0.87	-1.33	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis v-myc myeloblastosis viral oncogene homolog (myb) transcript variant X2 mRNA	NM_004914575.1	26.02	10.4	-1.33	16.47	22.75	0.47	0	0	0	0
Xenopus laevis SPRY domain containing 3 (spry3) mRNA	NM_001091822.1	440.13	176.22	-1.33	207.82	262.29	0.34	0	0	0	0
Xenopus laevis gamma-aminobutyric acid (GABA) A receptor alpha 2 (gabra2) mRNA	NM_001093962.1	2.84	1.14	-1.32	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis kinase family member 1A (kif1a) mRNA	NM_002933334.2	2.2	0.89	-1.31	0	0	0	0	0	0	0
Xenopus laevis cell division cycle associated 9 (cdca9) mRNA	NM_001092946.1	1.86	0.76	-1.31	0	0	0	0	0	0	0
XLA0156 Xenopus laevis homeobox protein Xhox8 (xhox8) mRNA complete cds	AF001596.1	1.53	0.62	-1.31	0	0	0	0	0	0	0
Xenopus tropicalis finished DNA clone Theu89k08	CR848306.2	5.17	2.1	-1.31	3.94	5.66	0.53	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis SaT1 transcript variant X2 mRNA	NM_004917760.1	1.97	0.81	-1.3	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis metalloproteinase-18-like (LOC100493214) mRNA	NM_002934930.2	1.09	0.45	-1.3	1.4	2.09	0.59	0	0	0	0
Xenopus laevis LIM domain only 4 gene 2 (limo4.2-b) mRNA	NM_001086236.1	15.73	6.46	-1.29	4.94	5.26	0.1	0	0	0	0

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DEX	FC Six1+Eva1 CHX+DEX	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Control CHX	FC Control CHX	Rank <sup>c</sup>
PREDICTED : Xenopus (Silurana) tropicalis uncharacterized LOC101734544 (LOC101734544) mRNA	XM_004919663.1	1.72	0.71	-1.29	0	0	0	0	0	0
Xenopus laevis transcription elongation factor A (SII) 3 (tce43-b) mRNA	NM_001088343.1	1.08	0.45	-1.28	0	0	0	0	0	0
Xenopus (Silurana) tropicalis CUB domain containing protein 2 (cdq2) mRNA	NM_001102795.1	1.74	0.72	-1.28	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis cyclin-dependent kinase 10 (LOC100492493) mRNA	XM_002934072.2	8.76	3.64	-1.27	0	0	0	0	0	0
Xenopus laevis dihydroprymidine-nase-like 4 (dphys4) mRNA	NM_001086033.1	133.99	55.7	-1.27	37.24	47.16	0.35	0	0	0
Xenopus tropicalis finished cDNA clone TTB4Q40h11	CU025138.1	1.03	0.43	-1.27	0	0	0	0	0	0
Xenopus laevis 3'-spastic acid methyltransferase 1 (trdm1) mRNA	NM_001086204.1	3.54	1.48	-1.27	6.82	6.99	0.04	0	0	0
Xenopus laevis mRNA for Requiem protein partial cds clone reed201	AB021738.1	58.02	24.18	-1.27	44.26	60.37	0.45	0	0	0
Xenopus laevis matrix-remodelling associated 7 (mxra7) mRNA	NM_001094445.1	1.31	0.55	-1.27	0	0	0	0	0	0
Xenopus (Silurana) tropicalis ankyrin repeat domain 13B (ankrd13b) mRNA	NM_001102966.1	1.22	0.51	-1.27	0	0	0	0	0	0
Xenopus (Silurana) tropicalis VENT homeobox 3 gene 1 (ventbx3.1) mRNA	NM_001114266.1	1.17	0.49	-1.26	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis ATPas class I type 88 member 4 (atp8b4) mRNA	XM_002956572.2	62.02	25.98	-1.26	14.52	15.72	0.12	0	0	0
Xenopus laevis component of oligomeric gof complex 6 (cog6-b) mRNA	NM_001096804.1	1.22	0.51	-1.26	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis serine/threonine-protein kinase N1-like (LOC100487973) mRNA	XM_002942439.2	2.45	1.04	-1.24	0	0	0	0	0	0
Xenopus laevis Rh family B glycoprotein (rhbg-b) mRNA	NM_001093583.1	4.25	1.81	-1.24	1.16	11.07	3.26	0	0	0
Xenopus tropicalis finished cDNA clone TEG134-C23	CR761104.2	202.85	86.12	-1.24	67.44	72.59	0.11	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis uncharacterized LOC00497422 (LOC100497422) mRNA	XM_002934048.2	1.84	0.79	-1.24	0	0	0	0	0	0
Xenopus laevis zeta-chain (TCR) associated protein kinase 70kDa (zap70) mRNA	NM_001093536.1	17.71	7.54	-1.24	1.44	2.39	0.74	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis tormin 2 (fmm2) mRNA	XM_002935817.2	3.48	1.49	-1.23	0	0	0	0	0	0
Xenopus laevis metastasis suppressor 1 gene 2 (mirs1.2) mRNA	NM_001093916.1	759.5	326.49	-1.22	82.21	100.36	0.29	0	0	0
Xenopus laevis sulfotransferase family cytosolic 1C member 2 (sulftc2) mRNA	NM_001094956.1	8.91	3.84	-1.22	2.52	5.41	1.1	0	0	0
Xenopus laevis nucleolar protein 12 (nol12) mRNA	NM_001096931.1	435.05	187.31	-1.22	96.87	46.06	2.25	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis ligand of Numb protein X-2-like (LOC100494793) transcript variant X4 mRNA	XM_004916836.1	1.55	0.67	-1.22	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis progressive rod-cone degeneration (prcd) mRNA	XM_004918442.1	1.78	0.77	-1.21	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis ubiquitin carboxyl-terminal hydrolase CYLD-like (LOC100489307) mRNA	XM_002943432.2	3.13	1.36	-1.21	2.95	3.1	0.08	0	0	0
PREDICTED : Xenopus (Silurana) uncharacterized LOC01730712 (LOC101730712) mRNA	XM_004911430.1	2.88	1.25	-1.21	0	0	0	0	0	0
Xenopus laevis RAD51 homolog (S. cerevisiae) (rad51-a) mRNA	NM_001087767.1	8893.73	3805.78	-1.21	57.81	716.77	0.32	0	0	0
Xenopus laevis cone arrestin mRNA (CDNA clone MGC-15048) IMAGE:6953716 complete cds	BC094203.1	1.06	0.47	-1.21	1.14	2.05	0.86	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis uncharacterized LOC00492003 (LOC100492003) mRNA	XM_002956027.1	1.16	0.51	-1.2	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis similar to D-dopachrome tautomerase clone (LOC100217317) mRNA	XM_002939964.2	5.16	2.25	-1.2	0	0	0	0	0	0
Xenopus laevis colony stimulating factor-1 mRNA complete cds	XK418294.1	1.79	0.78	-1.2	0	0	0	0	0	0
Xenopus laevis PDZ domain containing ring finger 3 (pofrn3) mRNA	NM_001097041.1	527.57	230.55	-1.2	183.25	295.42	0.69	0	0	0
Xenopus (Silurana) tropicalis malonyl CoA:ACP acyltransferase (mcat) nuclear gene encoding mitochondrial protein mRNA	NM_001004788.1	1.22	0.54	-1.2	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis bicoid/C homolog 1 (biccl1) mRNA	XM_002937107.2	1.57	0.69	-1.2	0	0	0	0	0	0
Xenopus laevis uncharacterized LOC100217317 (LOC100217317) mRNA	NM_001142789.1	619.64	271.64	-1.19	144.84	196.2	0.44	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis aldehyde dehydrogenase 1 family member A3 (alhd1a3) transcript variant X1 mRNA	XM_002941452.2	1.55	0.68	-1.19	0	0	0	0	0	0
Xenopus laevis pmp1 transcript variant X1 mRNA	NM_002931991.2	35.75	15.77	-1.19	9.02	9.23	0.04	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis acyltransferase slow (pirp2) mRNA	NM_001086347.1	1.49	0.66	-1.18	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis bim1 mRNA	XM_002932750.2	1.24	0.55	-1.18	0	0	0	0	0	0
Xenopus laevis Similar to D-dopachrome tautomerase clone IMAGE:4930459 mRNA	BC043871.1	1.75	0.78	-1.18	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis prostate transmembrane protein androgen induced 1 (pmipa1) transcript variant X1 mRNA	NM_002902577.2	5.53	2.45	-1.18	3.48	3.87	0.16	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis ubiquitin specific peptidase 31 (usp31) mRNA	XM_00293122.1	1.64	0.73	-1.18	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis peripherin 2 (retinal degeneration slow) (prp12) mRNA	XM_002938740.2	1.07	0.48	-1.18	0	0	0	0	0	0
Xenopus laevis Similar to D-dopachrome tautomerase clone IMAGE:6952313 partial cds	NM_001103003.1	1.17	0.52	-1.18	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis scopardin-like (LOC100497129) mRNA	BC157297.1	2.76	1.23	-1.17	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC33133 mRNA (cDNA clone IMAGE:6952313)	BC074438.1	1.82	0.81	-1.17	0	0	0	0	0	0
Xenopus laevis peripherin 2 (retinal degeneration slow) (prp12) mRNA	DQ09656.1	1.18	0.53	-1.17	0	0	0	0	0	0
PREDICTED : Xenopus (Silurana) tropicalis finger protein 1-like (LOC100497960) mRNA	NM_001096814.1	381.95	170.16	-1.17	107.09	107.06	-0.01	0	0	0
Xenopus (Silurana) tropicalis uncharacterized LOC100215092 (LOC100125092) mRNA	BC1301833.1	1.13	0.51	-1.17	0	0	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7535733	NM_002931574.2	561.24	250.25	-1.17	148.1	175.9	0.25	0	0	0
Xenopus laevis lipocalin mRNA (cDNA clone IMAGE:6946540) complete cds	NM_001079403.1	1.69	0.76	-1.17	0	0	0	0	0	0

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DEX	FC Six1+Eya1 CHX	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Control CHX	FC Control CHX	Rank <sup>a</sup>
PREDICTED: Xenopus (Silurana) tropicalis Arp5 actin-related protein 5 homolog (actr5) transcript variant X2 mRNA	XM_004918537..1	1.56	0.7	-1.17	0	0	0	0	0	0
Xenopus laevis double stranded RNA activated protein kinase 1 mRNA (cDNA clone MGIC:179851 (IMAGE:4969041) complete cds	BC157421..1	1.13	0.51	-1.17	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine-protein kinase N1-like (LOC100486934) mRNA	XM_002945400..2	4.7	2.11	-1.16	2.46	2.93	0.26	0	0	0
Xenopus laevis ORAI calcium release-activated calcium channel subunit 1 (ora1) mRNA	NM_001090975..1	810..7	363..51	-1.16	141..53	205..07	0.54	0	0	0
Xenopus (Silurana) tropicalis mediator complex subunit 8 (med8) mRNA	NM_001016614..3	12..3	5..53	-1.16	3..04	3..9	0..37	0	0	0
Xenopus (Silurana) tropicalis T-box protein 1 mRNA	NM_00126670..1	1.74	0.79	-1.15	0	0	0	0	0	0
Xenopus (Silurana) tropicalis member 5 (sicd4as) mRNA	NM_001092825..1	16..94	7..66	-1..15	8..9	14..69	0..73	0	0	0
Xenopus laevis clone p50007..1	AF50007..1	2..11	0..96	-1..15	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis stimulated by retinoic acid 13 (stra13) transcript variant X1 mRNA	XM_002942767..2	4.39	1.99	-1.15	0	0	0	0	0	0
X. laevis AE-III mRNA for peptidyl hydroxyacyl-Nc-lyase	X62771..1	2.35	1.07	-1.15	0	0	0	0	0	0
Xenopus tropicalis finished DNA clone Tgg129d15	CR848505..2	2.18	0.99	-1.14	0	0	0	0	0	0
Xenopus laevis nuclear domain-10 protein NDp2 mRNA (cDNA clone MGIC:32052 (IMAGE:6641847) complete cds	BC108760..1	1.01	0.46	-1.14	0	0	0	0	0	0
Xenopus laevis RNA helicase II/Gu mRNA (cDNA clone IMAGE:5047862) partial cds	BC079823..1	2.78	1.27	-1.14	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis regulatory factor X4 (influences HLA class II expression) (rfx4) mRNA	XM_002943913..2	3..31	1..51	-1..14	1..28	1..45	0..19	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis vacuolar protein sorting 37 homolog D (S. cerevisiae) (vps37d) mRNA	XM_004911761..1	3..43	1..57	-1..13	0	0	0	0	0	0
Xenopus laevis CCR1 (CCR1) mRNA complete cds	AY902778..1	1585..3	724..95	-1..13	195..12	283..3	0..54	0	0	0
Xenopus laevis cDNA clone IMAGE:7767654 partial cds	BC122467..1	1..63	0..75	-1..13	0	0	0	0	0	0
Xenopus laevis MTG16b (MTG16) mRNA MTG16-A allele complete cds alternatively spliced	AY714078..1	1..47	0..68	-1..13	0	0	0	0	0	0
Xenopus XA-1 mRNA anterior and ectodermis-specific	X53821..1	144..79	66..59	-1..13	135..4	208..3	0..63	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis interferleukin 16 (il16) mRNA	XM_002932267..2	2..92	1..34	-1..12	1..74	2..55	0..56	0	0	0
Xenopus (Silurana) tropicalis bone morphogenic protein receptor type 1A (bmpr1a) mRNA	XM_002937571..2	1..54	0..71	-1..12	0	0	0	0	0	0
Xenopus laevis neuronal cell adhesion molecule (ncam) mRNA	NM_001092018..1	1..24	0..58	-1..11	0	0	0	0	0	0
Xenopus laevis clone IMAGE:4683763 mRNA	BC045254..1	2..24	1..04	-1..11	4..13	4..77	0..21	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis syncoilin intermediate filament protein (sync) transcript variant X1 mRNA	XM_002940022..2	2	0..93	-1..11	0	0	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7615867 containing frame-shift errors	BC133731..1	6..25	2..91	-1..11	37..87	55..31	0..55	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis arachitin 10 (anop10) mRNA	XM_002937850..2	1..87	0..87	-1..11	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis spermatogenesis associated serine-rich 2-like (spats2l) mRNA	XM_002937711..2	4..77	2..23	-1..11	5..03	5..04	0..01	0	0	0
Xenopus laevis clone IMAGE:7767653 mRNA	NM_00101193..2	4..36	2..04	-1..1	3..13	4..14	0..41	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis prostaglandin reductase 1 gene 1 (pigf1..1) mRNA	XM_004918765..1	1..06	0..5	-1..1	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis syncoilin intermediate filament protein 3-like (LOC101732678) mRNA	XM_004917684..1	1..63	0..77	-1..1	3..05	3..24	0..09	0	0	0
Xenopus tropicalis homeobox 2 (hmx2) transcript variant X2 mRNA	NM_001095105..1	6..87	3..22	-1..1	7..57	15..53	1..04	0	0	0
Xenopus tropicalis hypothetical protein LOC779596 mRNA (cDNA clone IMAGE:7679615) partial cds	BC125718..1	6..25	2..94	-1..1	1..09	1..67	0..62	0	0	0
Xenopus (Silurana) tropicalis Friend leukemia virus integration 1 (flv1) mRNA	NM_001102719..1	1..03	0..48	-1..09	0	0	0	0	0	0
Xenopus laevis family with sequence similarity 69 member B (fam69b) mRNA	NM_001097225..1	1..28	0..6	-1..09	0	0	0	0	0	0
Xenopus laevis cDNA clone IMAGE:5570266 partial cds	BC073666..1	39..31	18..51	-1..09	22..37	35..46	0..67	0	0	0
Xenopus laevis Bar homeobox protein XBH4 mRNA partial cds	AF283692..1	1..94	0..91	-1..09	0	0	0	0	0	0
Xenopus tropicalis clone IMAGE:7529854	BC167287..1	20..44	9..64	-1..09	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis Friend leukemia virus integration 1 (flv1) mRNA	XM_002934933..2	265..61	125..26	-1..09	62..59	8..01	0..38	0	0	0
Xenopus laevis hypothetical protein LOC733197 mRNA (cDNA clone IMAGE:6643031) complete cds	BC093582..1	1..24	0..59	-1..09	1..15	1..12	0..07	0	0	0
XENCAR Xenopus laevis cone arrestin mRNA complete cds	L40463..1	1..81	0..86	-1..08	1..34	1..88	0..5	0	0	0
Xenopus tropicalis finished DNA clone Teg037g14	CR760426..2	1..32	0..63	-1..08	0	0	0	0	0	0
Xenopus tropicalis finished DNA clone Tgas140f03	CR762155..2	1410..93	667..94	-1..08	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zonepellucida sperm-binding protein 4-like (LOC100494768) mRNA	XM_002939873..2	5..58	2..65	-1..08	2..28	5..15	1..18	0	0	0
Xenopus laevis T-box protein mRNA (cDNA clone MGIC:83452 (IMAGE:6643031) complete cds	B070708..1	1..47	0..7	-1..08	0	0	0	0	0	0
XENCAR Xenopus laevis cone arrestin mRNA complete cds	L40463..1	1..81	0..86	-1..08	1..34	1..88	0..5	0	0	0
Xenopus tropicalis finished DNA clone Teg037g22	CT027886..1	1..04	0..5	-1..08	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis leucine rich repeat containing 41 (lrr41) mRNA	XM_002931474..2	1..34	0..64	-1..08	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis erm ERM-like protein (ermn) mRNA	XM_00293283..2	168..4	80..26	-1..07	0	0	0	0	0	0
Xenopus laevis wee1 homolog mRNA (cDNA clone MGIC:196573 (IMAGE:59414486) complete cds	BC169846..1	1..97	0..94	-1..07	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis methylmalonic aciduria (cobalamin deficiency) cb1a type (mmal) mRNA	XM_002938933..2	29..8	14..26	-1..07	2..24	2..62	0..23	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis vomeronasal type-2 receptor 26-like (LOC100498282) mRNA	XM_004911543..1	2..67	1..28	-1..07	0	0	0	0	0	0

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DEX	FC Six1+Eva1 CHX	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Control CHX	Control CHX+DEX	FC Control CHX	Rank <sup>a</sup>
Xenopus laevis retron dehydrogenase 7 (rdh7) mRNA	NM_001085720.1	257.52	123.45	-1.07	112.23	119.24	0.09	0	0	0	0
Xenopus laevis blood island-enriched kruppel-like factor mRNA (cDNA clone MGC:130763 IMAGE:7980258) complete cds	BC1.10713.1	61.08	29.35	-1.06	0	0	0	0	0	0	0
Xenopus laevis angiogenin receptor-like 1b (agribil) mRNA complete cds	DO473442.1	18.11	8.7	-1.06	6.62	6.82	0.05	0	0	0	0
Xenopus laevis galectin 1X mRNA partial cds	JN975639.1	2522.2	1213.88	-1.06	262.02	508.63	0.96	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis filaggrin-like (LOC10097400) mRNA	XM_004914931.1	40.94	19.77	-1.06	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis dacshund homolog 2 (dash2) mRNA	NM_00110045.1	1.04	0.5	-1.06	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis N-deacetylase/N-sulfotransferase (heparan glucosaminyl) 3 (ndst3) mRNA	NM_001127047.1	1.44	0.7	-1.05	0	0	0	0	0	0	0
Xenopus laevis mRNA for thyaccine 1	Y14446.1	1.44	0.7	-1.05	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis embryonic protein IUVS-2-like (LOC101734297) transcript variant X3 mRNA	XM_004913370.1	11.04	5.35	-1.05	0	0	0	0	0	0	0
Xenopus laevis hypothetical protein LOC100127287 mRNA (cDNA clone IMAGE:8822377) partial cds	BC155883.1	309.23	149.86	-1.05	16.31	18.27	0.17	0	0	0	0
Xenopus laevis clone IMAGE:4885140 mRNA	BC041321.1	2.66	1.29	-1.05	1.09	1.98	0.87	0	0	0	0
Xenopus laevis palmelphlin (palmed) mRNA	NM_001087189.1	1.79	0.87	-1.05	0	0	0	0	0	0	0
Xenopus laevis uncharacterized protein MGC115316 (MGC115316) mRNA	NM_001095891.1	1.23	0.6	-1.04	0	0	0	0	0	0	0
Xenopus laevis neurotisin (nts) mRNA	NM_001092681.1	2.03	1	-1.04	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis ankyrin repeat domain 6 (ankrd6) mRNA	NM_00112901.1	1.17	0.58	-1.04	0	0	0	0	0	0	0
Xenopus tropicalis finished DNA clone TGA80807	CR942563.2	878.29	430.79	-1.03	44.24	50.01	0.18	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7715753	BC1.69136.1	4.36	2.14	-1.03	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis cysteine-rich motor neuron 1 protein-like (LOC101731600) mRNA	NM_004916186.1	2.77	1.36	-1.03	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis melanin-concentrating hormone receptor 2 (mchr2) mRNA	XM_002932159.2	1.76	0.87	-1.03	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis GTP-binding protein like (X0674) (MGC147600) mRNA	NM_001079409.1	3.57	1.76	-1.03	1.31	1.38	0.08	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis patatin-like phospholipase domain-containing protein 1-like (LOC100497390) mRNA	XM_002938433.2	5.11	2.52	-1.03	5.64	5.72	0.02	0	0	0	0
Xenopus laevis PRKR interacting protein 1 (I1.11 inducible) (prkrp1) mRNA	NM_001092454.1	246.86	121.79	-1.02	16.56	18.39	0.16	0	0	0	0
Xenopus (Silurana) tropicalis hyaluronan synthase 2 (has2) mRNA	NM_001127085.1	2.17	1.07	-1.02	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis myobdiumenum cofactor synthase 1 (mcos1) mRNA	XM_002902944.2	40.48	19.99	-1.02	11.37	16.14	0.51	0	0	0	0
Xenopus laevis homeobox B7 (hoxd7-b) mRNA	NM_001096049.1	2.6	1.29	-1.02	0	0	0	0	0	0	0
Xenopus laevis hypothetical LOC958321 mRNA (cDNA clone MGC:90030 IMAGE:5514059) complete cds	RC086469.1	1.36	0.68	-1.02	0	0	0	0	0	0	0
Xenopus tropicalis finished DNA clone TGA13724	CT025404.2	129.99	64.54	-1.02	28.17	29.17	0.06	0	0	0	0
Xenopus (Silurana) tropicalis synaptosomal-associated protein 91kDa (snap91) mRNA	NM_001130277.1	1.64	0.82	-1.01	2.05	2.23	0.13	0	0	0	0
Xenopus laevis transmembrane protein 82 (tmem82) mRNA	NM_001094923.1	1.76	0.88	-1.01	2.11	2.76	0.39	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00498222 (LOC100498222) mRNA	XM_002939879.2	1.67	0.83	-1.01	0	0	0	0	0	0	0
XELTGF85 Xlewis transforming growth factor-beta (TGF-beta 5) mRNA complete cds	J05180.1	1.3	0.65	-1.01	1.06	1.85	0.81	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC01730969 (LOC101730969) mRNA	XM_004913153.1	19.4	9.7	-1.01	1.92	2.18	0.19	0	0	0	0
Xenopus laevis hypothetical protein LOC33224 mRNA (cDNA clone IMAGE:6946334) partial cds	BC094170.1	2.62	1.31	-1.01	0	0	0	0	0	0	0
Xenopus laevis TRAF3 interacting protein 1 (trafip1) mRNA	NM_001094677.1	27.76	6.81	-1.01	5.92	5.87	-0.02	0.01	0.01	0	0
Xenopus laevis hypothetical protein LOC33214 mRNA (cDNA clone IMAGE:6957873) partial cds	BC094137.1	498.43	174.21	-1.02	30.73	30.72	-0.02	0.01	0.01	0	0
Xenopus laevis small nuclear RNA activating complex polypeptide 5 (snap5c-b) mRNA	NM_001171809.1	290.38	142.95	-1.03	45.11	44.77	-0.02	0.01	0.01	0	0
PREDICTED: Xenopus (Silurana) tropicalis NPX E family member 1-like (LOC101731316) partial mRNA	XM_00293275.2	2.88	1.4	-1.05	7.41	7.33	-0.02	0.02	0.02	0	0
Xenopus laevis paired domain transcription factor variant A (Pax-6) mRNA Pax-6 allele complete cds	AF154555.1	9.92	2.78	-1.04	9.72	9.49	-0.04	0.02	0.02	0	0
Xenopus laevis myobdiumenum cofactor synthase 1 (mcos1) mRNA	NM_001092732.1	1549.14	750.98	-1.05	28.28	28.87	-0.03	0.02	0.02	0	0
Xenopus laevis cDNA clone IMAGE:3402714 partial cds	AY648303.1	8.92	1.78	-2.34	7.38	7.13	-0.05	0.03	0.03	0	0
Xenopus laevis matrix metalloproteinase 7 (MMP-7) mRNA complete cds	AY573380.1	114.83	47.6	-1.28	47.07	46.14	-0.03	0.03	0.03	0	0
Xenopus laevis uncharacterized LOC95021 (LOC95021) mRNA	NM_001094727.1	193.55	51.83	-1.91	37.75	36.59	-0.05	0.03	0.03	0	0
Xenopus laevis sphingosine-1-phosphate receptor 1 (s1pr1) mRNA	NM_001092761.1	1.23	0.42	-1.56	1.3	1.26	-0.04	0.03	0.03	0	0
Xenopus laevis doublesex and mab-3 related transcription factor 5 mRNA complete cds	DQ23558.1	4.79	2.08	-1.08	3.89	3.82	-0.03	0.03	0.03	0	0
Xenopus tropicalis cDNA clone IMAGE:7659670 containing frame-shift errors	BC123956.1	1.02	0.44	-1.24	5.74	5.6	-0.04	0.03	0.03	0	0
Xenopus laevis elongation factor 1 delta mRNA (cDNA clone IMAGE:80004 IMAGE:6317228) complete cds	BC072139.1	676.31	61.24	-3.47	35.53	32.98	-0.11	0.04	0.04	0	0
Xenopus laevis troponin C complete cds	A803080.1	3.66	0.73	-2.34	1.03	0.97	-0.09	0.04	0.04	0	0
Xenopus laevis chromosome 4 open reading frame 22 (corf22) mRNA	NM_001095253.1	1.44	0.23	-2.67	2.21	2.05	-0.11	0.04	0.04	0	0

Gene	Accession	Six1+Eva1 CHX	Six1+Eva1 CHX+DEX	FC	Control CHX	Control CHX+DEX	FC	Control CHX	Control CHX+DEX	FC	Rank <sup>c</sup>
Xenopus laevis MGCB2906 protein (MGCB2906) mRNA	NM_001092491.1	3333.03	708.23	-2.24	43.16	40.45	-0.1	0.05	-0.05	0.05	0.05
Xenopus laevis cardiolipin synthase 1 (crls1) mRNA	NM_001096593.1	115	57.29	-1.01	5.2	5.05	-0.05	0.05	-0.05	0.05	0.05
Xenopus laevis cDNA clone MGCC179753 (MAGE:3396437 complete cds)	BC167483.1	162.85	3.54	-5.53	69.63	58.47	-0.26	0.05	-0.26	0.05	0.05
Xenopus laevis egg cortical granule lectin mRNA (cDNA clone MG:196814 IMAGE:9041722) complete cds	BC167487.1	18.45	0.67	-4.79	4.04	3.42	-0.25	0.06	-0.25	0.06	0.06
Xenopus laevis SPB-a mRNA complete cds	EF164945.1	1.3	0.43	-1.61	1.28	1.2	-0.1	0.06	-0.1	0.06	0.06
Xenopus laevis hypothetical protein LOC100127319 mRNA (cDNA clone IMAGE:8319360) partial cds	BC155933.1	585.47	185.25	-1.67	341.9	320.07	-0.1	0.06	-0.07	0.07	0.07
Xenopus laevis cDNA clone NM_001098070.1	BC099326.1	1.28	0.61	-1.07	1.41	1.34	-0.07	0.07	-0.09	0.07	0.07
Xenopus (Silurana) tropicales serine hydrolase-like (sehl) mRNA	NM_001096049.1	2.18	0.93	-1.24	2.18	2.05	-0.09	0.08	-0.09	0.08	0.08
Xenopus laevis HIV-1 Tat interactive protein 2 (30 kDa) (htatip2) mRNA	NM_001096049.1	36108.37	15638.76	-1.21	48.48	45.67	-0.09	0.08	-0.09	0.08	0.08
Xenopus laevis cDNA clone IMAGE:4173903	BC083045.1	103.41	34.57	-1.59	228.56	211.12	-0.12	0.08	-0.12	0.08	0.08
Xenopus laevis cytochrome P450 family 2 subfamily E polypeptide 1 (cytP2e1) mRNA	NM_001091741.1	26.87	13.27	-1.02	17.38	16.43	-0.09	0.08	-0.09	0.08	0.08
Xenopus laevis uncharacterized protein MGCB8331 (MGCB8331) mRNA	NM_001090920.1	465.34	171.95	-1.44	92.52	84.44	-0.14	0.1	-0.14	0.1	0.1
PREDICTED: Xenopus (Silurana) tropicales zinc finger protein 850-like (LOC101732212) mRNA	NM_004919126.1	2237.78	1102.38	-1.03	42.35	39.52	-0.1	0.1	-0.09	0.1	0.1
Xenopus laevis adducin 1 (alpha) (add1) mRNA	NM_001087641.1	1609.19	57.73	-4.81	35.49	25.21	-0.5	0.11	-0.5	0.11	0.11
PREDICTED: Xenopus (Silurana) tropicales UDP-N-acetylglucosamine/UDP-glucose/GDP-mannose transporter-like (LOC100497699)	NM_002938972.2	19.46	2.04	-3.26	8.62	6.8	-0.35	0.11	-0.35	0.11	0.11
Xenopus laevis ankyrin repeat domain 37 (ankrd37) mRNA	NM_001098697.1	1.97	0.75	-1.4	3.31	2.99	-0.15	0.11	-0.15	0.11	0.11
Xenopus laevis gamma-glytamyltransferase 1 b (ggt1-b) mRNA	NM_001092395.1	188.73	60.29	-1.65	24.3	21.4	-0.19	0.12	-0.19	0.12	0.12
Xenopus laevis KIAA1143 (kiaa1143) mRNA	NM_001094698.1	78.97	28.75	-1.46	10.3	9.2	-0.17	0.12	-0.17	0.12	0.12
PREDICTED: Xenopus (Silurana) tropicales platelet-derived growth factor beta polypeptide (pdgfb) transcript variant X1 mRNA	NM_002933750.2	1.13	0.37	-1.65	1.03	0.9	-0.21	0.13	-0.21	0.13	0.13
Xenopus laevis adducin 1 (alpha) (add1) mRNA	NM_002938972.2	30.61	4.81	-2.68	5.36	4.21	-0.36	0.14	-0.36	0.14	0.14
PREDICTED: Xenopus (Silurana) tropicales associated protein 4 (gemin4) transcript variant X2 mRNA	NM_004911679.1	1591.24	753.14	-1.08	517.89	469.41	-0.15	0.14	-0.15	0.14	0.14
Xenopus laevis cDNA clone IMAGE:3398.08	BC108862.1	78.58	37.37	-1.08	17.28	15.65	-0.15	0.14	-0.15	0.14	0.14
Xenopus laevis KIAA11524 (kiaa1524) mRNA	NM_001094671.1	76.17	36.28	-1.08	15.87	14.36	-0.15	0.14	-0.15	0.14	0.14
Xenopus laevis WD repeat domain 5 (wdrp5-b) mRNA	NM_00109454.1	327.29	96.07	-1.77	20.59	17.41	-0.25	0.14	-0.25	0.14	0.14
Xenopus laevis CD38 molecule (cd38) mRNA	NM_001097679.1	4.54	2.23	-1.03	2.64	2.39	-0.15	0.14	-0.15	0.14	0.14
Xenopus laevis POU class 3 homeobox 1 (pou3f1-b) mRNA	RC129575.1	14.66	7.27	-1.02	15.79	14.34	-0.14	0.14	-0.14	0.14	0.14
Xenopus laevis hypothetical protein LOC100066874 mRNA (DNA clone IMAGE:9526743) partial cds	NM_002931687.2	8.37	2.75	-1.62	1.02	0.87	-0.23	0.14	-0.23	0.14	0.14
PREDICTED: Xenopus (Silurana) tropicales metastasis suppressor 1-like (mstsl1) transcript variant X1 mRNA	BC133196.1	9.94	4.69	-1.09	6.07	5.44	-0.16	0.15	-0.16	0.15	0.15
Xenopus laevis hypothetical protein LOC100071325 mRNA (DNA clone IMAGE:5570551) partial cds	NM_002935465.2	1.15	0.56	-1.04	2.68	2.41	-0.16	0.16	-0.16	0.16	0.16
Xenopus laevis nesos5 mRNA complete sequence	DQ098855.1	3.9	1.42	-1.46	7.69	7.67	-0.22	0.16	-0.22	0.16	0.16
PREDICTED: Xenopus (Silurana) tropicales chloride channel voltage-sensitive 2 (clcn2) transcript variant X1 mRNA	XM_002935464.2	90.82	24.04	-1.92	4.08	3.33	-0.3	0.16	-0.3	0.16	0.16
PREDICTED: Xenopus (Silurana) tropicales mitogen-activated protein kinase kinase 5 (map3k5) mRNA	XM_002935759.2	2606.82	1041.21	-1.33	203.37	172.96	-0.22	0.16	-0.22	0.16	0.16
Xenopus laevis hypothetical protein LOC100037135 mRNA (DNA clone IMAGE:5570551) partial cds	NM_0010110739.1	28.96	13.45	-1.11	2.2	1.94	-0.19	0.17	-0.19	0.17	0.17
PREDICTED: Xenopus (Silurana) tropicales solute carrier family 26 (sulfate transporter) member 1 (slc26a1) mRNA	XM_004914445.1	1.4	0.52	-1.44	1.47	1.22	-0.27	0.19	-0.27	0.19	0.19
Xenopus laevis mso05 mRNA complete sequence	BC106201.1	16.7	7.3	-1.2	9.77	8.35	-0.23	0.2	-0.23	0.2	0.2
PREDICTED: Xenopus (Silurana) tropicales uncharacterized LOC00495124 (LOC100495124) transcript variant X1 mRNA	DQ096861.1	70488.69	2726.49	-1.38	43665.14	36364.38	-0.27	0.2	-0.27	0.2	0.2
Xenopus laevis tRNA for Yom protein	AF174488.1	2.85	1.22	-1.23	1.55	1.3	-0.26	0.22	-0.26	0.22	0.22
Xenopus laevis tRNA for Xom protein	NM_001095781.1	39.65	19.49	-1.03	28.54	24.44	-0.23	0.22	-0.23	0.22	0.22
Xenopus laevis AdP-ribosyltransferase 1 (art1) mRNA	BC079790.1	2.64	1.21	-1.13	2.15	1.81	-0.25	0.22	-0.25	0.22	0.22
PREDICTED: Xenopus (Silurana) tropicales methylbind-like splicing regulator 1 (mbrn1) transcript variant X2 mRNA	CR942489.2	2.21	0.92	-1.27	1.13	0.93	-0.28	0.22	-0.28	0.22	0.22
Xenopus laevis hypothetical protein LOC10003895 mRNA (cDNA clone Tgg95cd21)	CJ075333.1	1.17	0.38	-1.64	5.96	5.95	-0.38	0.23	-0.38	0.23	0.23
Xenopus laevis rep44 mRNA complete sequence	X8454.1	58.51	20.95	-1.49	22.4	17.63	-0.35	0.24	-0.35	0.24	0.24
Xenopus laevis axia telangiectasia mutated (atm) mRNA partial cds	NM_001090563.1	43.64	15.09	-1.54	18.03	13.95	-0.38	0.25	-0.38	0.25	0.25
Xenopus laevis uncharacterized protein MGCB4886 (MGCB4886) mRNA	DQ096925.1	1707.39	149.71	-3.52	335.56	183.58	-0.87	0.25	-0.87	0.25	0.25
Xenopus laevis homeobox protein mRNA (cDNA clone MGCB79389 IMAGE:5161842) complete cds	XM_002939921.2	2.09	0.77	-1.45	29.71	23.12	-0.37	0.25	-0.37	0.25	0.25
Xenopus tropicalis finished DNA clone Ttg95cd1605	AF168464.1	5.6	1.91	-1.56	8.27	6.27	-0.4	0.26	-0.4	0.26	0.26
Xenopus laevis forhead protein lens 1 (lens1) mRNA complete cds	AB005655.1	7.9	2.97	-1.42	3.72	2.87	-0.38	0.27	-0.38	0.27	0.27
Xenopus laevis uncharacterized protein MGCB4886 (MGCB4886) mRNA	NM_001096633.1	2.49	0.98	-1.35	1.74	1.36	-0.37	0.27	-0.37	0.27	0.27
PREDICTED: Xenopus (Silurana) tropicales ALX homeobox 4 (alk4) transcript variant X2 mRNA	XM_004913349.1	2.11	0.67	-1.66	0.78	0.78	-0.46	0.28	-0.46	0.28	0.28
Xenopus laevis uncharacterized LOC100101308 (LOC100101308) mRNA	NM_001095892.1	6.98	3.1	-1.18	1.5	1.5	-0.33	0.28	-0.33	0.28	0.28

Gene	Accession	Six1+Eya1 CHX	Six1+Eya1 CHX+DEX <sup>1</sup>	FC Six1+Eya1 <sup>1</sup>	Control CHX	Control CHX+DEX <sup>1</sup>	FC Control CHX+DEX <sup>1</sup>	Rank <sup>7</sup>
Xenopus laevis VENT homeobox 1 gene 1 (ventnx1.1-b) mRNA	NM_001172121.1	3.25	0.89	-1.89	1.36	0.95	-0.53	0.28
Xenopus laevis hypothetical protein LOC43706 mRNA (cDNA clone IMAGE:6946515) partial cds	BC126044.1	148.62	38.29	-1.96	5.18	3.51	-0.57	0.29
Xenopus tropicalis stimulated by retinoic acid gene 6 homolog mRNA (cDNA clone IMAGE:7621800) partial cds	BC135711.1	2.97	1.35	-1.15	2.27	1.8	-0.34	0.3
PREDICTED: Xenopus (Silurana) tropicalis protein tyrosine kinase 6 (ptk6) mRNA	XM_002942383.2	1.58	0.72	-1.14	2.81	2.23	-0.34	0.3
X laevis mRNA for Xvent-1 protein	X92851.1	8.29	2.87	-1.53	2.96	2.14	-0.48	0.31
Xenopus laevis cholesterol 25-hydroxylase mRNA (cDNA clone IMAGE:8460553) partial cds	BC128953.1	62.87	31.39	-1.01	18.37	14.76	-0.32	0.32
Xenopus tropicalis finished cDNA clone Toas120721	CT485737.1	16.38	7.08	-1.22	6.56	5.03	-0.39	0.32
Xenopus laevis solute carrier family 22 (organic cation/ergothioneine transporter) member 4 (slc22a4) mRNA	NM_001094580.1	1338.92	623.75	-1.11	480.06	376.13	-0.36	0.32
PREDICTED: Xenopus (Silurana) tropicalis diencephalon/mesencephalon homeobox protein 1A-like (LOC100493907) transcript variant 1 mRNA	XM_002931403.2	26.62	6.47	-2.05	25.34	16.03	-0.67	0.33
Xenopus (Silurana) tropicalis snail family zinc finger 1 (snail1) mRNA	NM_203936.1	103.39	46.81	-1.15	73.63	56.58	-0.39	0.34
PREDICTED: Xenopus (Silurana) tropicalis polycystic kidney disease protein 1-like 3-like (LOC101733273) mRNA	NM_004915060.1	220.56	93.85	-1.24	63.93	47.74	-0.43	0.35
Xenopus laevis jun B proto-oncogene (junB) mRNA	NM_001097035.1	1069.01	356	-1.59	383.85	264.74	-0.55	0.35
Xenopus (Silurana) tropicalis steroid 5-alpha-reductase alpha polypeptide 2 (3-oxo-5-alpha-steroid delta 4-dehydrogenase alpha 2) (sr5a2)	NM_001017113.2	2.78	1.35	-1.05	2.92	2.28	-0.36	0.35
Xenopus laevis proline-serine-threonine phosphatase interacting protein 2 (pstip2) mRNA	NM_001093301.1	2219	1022.23	-1.12	222.25	170.85	-0.39	0.35
Xenopus laevis uncharacterized protein NGC68811 (NGC68811) mRNA	NM_00109098.1	931.16	313.5	-1.58	197.57	135.42	-0.55	0.35
Xenopus laevis cDNA clone MGC:98823 (IMAGE:3401284 complete cds	BC084658.1	7827.64	3612.11	-1.12	1375.58	1050.88	-0.39	0.35
Xenopus laevis transmembrane protein 55A (cnmem55a) mRNA	NM_001095826.1	1.44	0.67	-1.13	1.88	1.44	-0.4	0.35
Xenopus laevis ephrin-A4 (efha4) mRNA	NM_001127882.1	68.27	32.6	-1.07	63.18	48.78	-0.38	0.35
Xenopus (Silurana) tropicalis SH3 and cysteine rich domain 3 (stac3) mRNA	NM_001007506.1	151.02	53.54	-1.5	64.2	44.1	-0.55	0.37
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100498593 (LOC100498593) transcript variant X2 mRNA	XM_004915063.1	492.77	182.71	-1.44	37.61	25.86	-0.55	0.38
Xenopus laevis cDNA clone MGC:154541 (IMAGE:8320906 complete cds	BC124896.1	805.33	259.64	-1.64	192.5	125.6	-0.63	0.39
Xenopus tropicalis finished cDNA clone Theu01319	CR760197.2	487.72	193.86	-1.34	1.56	1.08	-0.54	0.41
Xenopus tropicalis finished cDNA clone Toas14948	CT025385.2	8.69	3.17	-1.46	8.37	5.56	-0.6	0.41
PREDICTED: Xenopus (Silurana) tropicalis CASP8 and FADD-like apoptosis regulator (cflar) transcript variant X2 mRNA	XM_004917781.1	1.94	0.83	-1.23	1.3	0.9	-0.53	0.43
XLU3528 Xenopus laevis homeobox protein (Vox-1) mRNA complete cds	US3528.1	76.07	30.11	-1.34	44.85	29.37	-0.61	0.45
Xenopus laevis zinc finger protein 2 (zfzf706.2-b)	NM_001092619.1	1.1	0.5	-1.16	4.43	3.09	-0.53	0.46
Xenopus (Silurana) tropicalis epithelial membrane protein 2 (emp2) mRNA	NM_001030386.1	1888.2	825.3	-1.2	76.86	52.86	-0.55	0.46
Xenopus laevis EGFR-like-domain multiple 6 (egfr6) mRNA	NM_001086685.1	5.18	2.57	-1.02	3.72	2.7	-0.47	0.46
Xenopus laevis ATPase Na+/K+ transporting alpha 3 polypeptide (atp1a3) mRNA	NM_001086971.1	3.83	0.57	-2.77	1.39	0.57	-1.3	0.47
Xenopus laevis axin 2 (conductin axil) (axin2) mRNA	NM_001094372.1	14.27	6.48	-1.14	11.17	7.71	-0.54	0.47
PREDICTED: Xenopus (Silurana) tropicalis carbohydrate sulfotransferase 10-like (LOC100497654) transcript variant X1 mRNA	XM_002938346.2	208.54	73.39	-1.51	26.37	15.92	-0.73	0.49
Xenopus tropicalis finished cDNA clone Theu03909	CR942608.2	18.44	8.26	-1.16	12.08	8.17	-0.57	0.49
X laevis mRNA for transcription factor VENT	X98849.1	32.59	13.19	-1.31	11.42	7.28	-0.66	0.5
Xenopus laevis cDNA clone IMAGE:5158422 *** WARNING: chimeric clone ***	BC157480.1	3.82	1.52	-1.34	1.93	1.22	-0.68	0.51
Xenopus laevis Ras homolog enriched in brain like 1 (nebl1) mRNA	NM_001094758.1	154.15	74.92	-1.05	119.74	83.26	-0.53	0.51
Xenopus (Silurana) tropicalis inositol hexaphosphate kinase 1 (ip6k1) mRNA	NM_001079117.1	251.62	104.33	-1.27	4.8	3.06	-0.66	0.52

<sup>1</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1+Eya1 and CHX treatment

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1+Eya1 and CHX+DEX treatment

<sup>3</sup> Log<sub>2</sub> fold change values for Six1+Eya1

<sup>4</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup> Log<sub>2</sub> fold change values for un-injected control

<sup>7</sup> Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Six1+Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## Appendix F. Merged analysis

### F.1 Six1<sub>m</sub>: Up-regulated genes

**Table F.1** Significantly differentially expressed genes with at least two-fold up-regulation after injection of Six1-GR or Six1-GR+Eya1-GR and treatment with CHX + DEX

Gene	Accession	Sik1 CHX <sup>1</sup>	Sik1 CHX+DEX <sup>2</sup>	FC Six1 <sup>3</sup>	q-value	Control CHX <sup>4</sup>	Control CHX+DEX <sup>5</sup>	FC Control CHX+DEX <sup>6</sup>	Rank <sup>7</sup>
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731096 (LOC101731096) mRNA	XM_004916510.1	0.02	54.22	11.4	0.023	61.07	9.12	-2.7	0
PREDICTED: Xenopus (Silurana) tropicalis parvalbumin thymic CPV3-like (LOC100485867) transcript variant X1 mRNA	XM_002931951.1	0.03	5.03	7.4	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zona pellucida-like domain-containing protein 1-like (LOC101732051) mRNA	XM_004916514.1	0.03	2.36	6.3	0.009	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE7022272	BC094950.1	0.09	6.66	6.2	0.009	0	0	0	0
Xenopus laevis MGc83328 protein mRNA cDNA clone MGc83328 (IMAGE:6864428) complete cds	EC082426.1	0.26	1.88	6.2	0.009	0	0	0	0
Xenopus laevis uncharacterized LOC100363538 (LOC100363538) mRNA	NM_001109778.1	0.13	8.02	5.9	0.009	0	0	0	0
Xenopus (Silurana) tropicalis SIX homeobox 2 (six2) mRNA	NM_0029100275.1	0.57	23.41	5.4	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis potassium voltage-gated channel shaker-related subfamily member 2 (kcn2a) mRNA	XN_004910736.1	0.08	1.92	4.6	0.009	0	0	0	0
Xenopus (Silurana) tropicalis ectodysplasin A receptor (edar) mRNA	NM_001006819.1	0.18	4.03	4.5	0.009	0.4	1.13	1.5	0.33
Xenopus laevis mRNA for Xsox17/alpha protein	AU001730.1	0.54	11.26	4.4	0.009	0	0	0	0
Xenopus (Silurana) tropicalis glutathione peroxidase 2 (gastrointestinal) (gpx2) mRNA	NM_001256315.1	0.7	14.61	4.4	0.017	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis calcium/calmodulin-dependent protein kinase kinase 2 beta (camkk2) transcript variant X2 mRNA	XM_004910513.1	0.14	2.68	4.3	0.009	0	0	0	0
Xenopus laevis Myoblast determination protein 1 homolog A mRNA (cdNA clone MGc52596 (IMAGE:4885255) complete cds	BC041190.1	3.21	55.9	4.1	0.009	3.87	6.38	0.7	0.17
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 4 sodium bicarbonate cotransporter member 4 (slc4a4) transcript variant X3	XN_0049119134.1	0.15	2.06	3.8	0.039	0	0	0	0
Xenopus laevis RDC1 like protein mRNA (cdNA clone MGc114801 (IMAGE:632706)) complete cds	BC098974.1	0.6	7.77	3.7	0.009	0	0	0	0
Xenopus laevis mRNA for Xsox17/alpha2 complete cds	AB02691.1	0.5	5.53	3.5	0.009	0	0	0	0
Xenopus (Silurana) tropicalis uncharacterized protein MGc82269 (MGc82269) mRNA	NM_001091363.1	0.1	1.11	3.5	0.009	0	0	0	0
Xenopus (Silurana) tropicalis chemokine (C-X-C motif) receptor 7 (cxcr7) mRNA	CX_001030434.1	0.43	4.92	3.5	0.009	0	0	0	0
Xenopus (Silurana) tropicalis P450 family 26 subfamily B polypeptide 1 (cytP450b1) mRNA	NM_0010379187.2	0.29	2.61	3.2	0.009	3.54	1.95	-0.9	0
Xenopus laevis hedgehog-interacting protein mRNA complete cds	AY328923.1	0.23	2.05	3.2	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis hairy and enhancer of split 8 (hes8) mRNA	XN_002933849.2	1.64	15.4	3.2	0.009	1.01	1.64	0.7	0.22
Xenopus tropicalis sine oculis binding protein (Drosophila) mRNA (cdNA clone MGc172604 (IMAGE:7656239) complete cds	BC154687.1	0.6	5.01	3.1	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis espin (espn) transcript variant X1 mRNA	XN_002933856.2	1.02	8.56	3.1	0.017	1.81	1.76	0	0
Xenopus (Silurana) tropicalis cytochrome P450 family 26 subfamily B polypeptide 1 (cytP450b2) mRNA	NM_001030412.1	0.45	3.68	3	0.009	0	0	0	0
Xenopus laevis hedgehog-interacting protein mRNA complete cds	NM_001100279.1	0.17	1.39	3	0.009	0.52	1.02	1	0.33
PREDICTED: Xenopus (Silurana) tropicalis hairy and enhancer of split 8 (hes8) mRNA	XN_002933849.2	1.64	15.4	3.2	0.009	1.01	1.64	0.7	0.22
Xenopus tropicalis sine oculis binding protein (Drosophila) mRNA (cdNA clone MGc172604 (IMAGE:7656239) complete cds	BC154687.1	0.6	5.01	3.1	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis espin (espn) transcript variant X1 mRNA	XN_002933856.2	1.02	8.56	3.1	0.017	1.81	1.76	0	0
Xenopus (Silurana) tropicalis X-linked inhibitor of apoptosis (xiap) mRNA	NM_0010379187.2	0.53	3.68	3	0.009	0	0	0	0
Xenopus (Silurana) tropicalis protein phosphatase 2 regulatory subunit B' beta (pp2B5b) mRNA	XN_002933849.2	0.25	1.71	2.8	0.009	0	0	0	0
Xenopus laevis gelatinase B (Mmp-9) mRNA complete cds	AF072455.1	0.89	6.86	2.9	0.009	0	0	0	0
Xenopus laevis POU class 3 homeobox 2 (pou3f2) mRNA	NM_001096715.1	0.53	4.01	2.9	0.009	1.02	1.39	0.4	0.14
Xenopus laevis potassium inwardly-rectifying channel subfamily J member 8 (Kcnj8) mRNA complete cds	HQ266327.1	0.19	1.39	2.9	0.039	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis $\lambda$ -C motif chemokine 10-like (LOC100490785) mRNA	XN_002940578.2	0.53	3.8	2.8	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis paired box 1 (pax1) transcript variant X1 mRNA	XN_002939437.2	0.25	1.71	2.8	0.009	0	0	0	0
Xenopus tropicalis finished cDNA clone TNeu143f19	CR760056.2	0.55	3.85	2.8	0.009	1.03	1.1	0.1	0.04
XLJ76536 Xenopus laevis calbindin D28k mRNA complete cds	UJ66336.1	0.9	5.98	2.7	0.009	1.58	1.59	0	0
Xenopus laevis transforming growth factor beta-induced 68kDa (tgfb1) mRNA	NM_001095238.1	0.23	1.39	2.6	0.023	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis octamin 2 (ano2) mRNA	XN_002932297.2	0.27	1.49	2.5	0.029	0	0	0	0
Xenopus laevis uncharacterized protein MGc68450 (MGc68450) mRNA	NM_001089841.1	0.43	2.3	2.4	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis T-cell leukemia homeobox 1 (ttx1) transcript variant 1 mRNA	XN_002936768.2	1.59	8.16	2.4	0.009	1.12	1.47	0.4	0.17
PREDICTED: Xenopus (Silurana) tropicalis integrin beta 4 (itgb4) transcript variant X3 mRNA	XN_004918457.1	0.37	1.94	2.4	0.017	0	0	0	0
Xenopus laevis cDNA clone IMAGE7394277	BC106376.1	0.23	1.18	2.4	0.044	0	0	0	0
Xenopus laevis mRNA for frizzled 4 protein (fz4 gene)	AJ251750.1	43.34	213.37	2.3	0.009	14.14	10.8	-0.4	0
Xenopus laevis shisa-2 mRNA for Shisa2 complete cds	AB24297.1	1.29	6.19	2.3	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis growth factor-independent 1 transcription repressor (gf1) mRNA	XN_002933803.2	1.06	5.4	2.3	0.009	0.79	1.33	0.8	0.35

Gene	Accession	Six1 CHX	Six1 CHX+DEX <sup>1</sup>	FC Six1 CHX+DEX <sup>1</sup>	q-value	Control CHX	Control CHX+DEX <sup>1</sup>	FC Control CHX+DEX <sup>1</sup>	Rank <sup>2</sup>
Xenopus tropicalis finished cDNA clone TNeu052k18	CR760018.2	0.81	3.87	2.3	0.009	0.84	1.5	0.8	0.35
Xenopus (Silurana) tropicalis integrin beta 6 (itgb6) mRNA	NM_001097306.1	0.53	2.37	2.2	0.009	0.67	1.25	0.9	0.41
PREDICTED: Xenopus (Silurana) tropicalis monocyte to macrophage differentiation-associated (mmnd) transcript variant X1 mRNA	XM_009378112	1.11	4.8	2.1	0.009	3.32	2.75	-0.3	0
Xenopus laevis chromogranin A (parathyroid secretory protein 1) (chga) mRNA	NM_001094724.1	4.17	16.32	2	0.009	3.65	3.66	0	0
Xenopus laevis claudin 3 (cd3) mRNA	NM_001093931.1	1.32	5.39	2	0.009	459.11	385	-0.3	0
Xenopus laevis gamma-glutamyl hydrolase conjugase folylpolygammaglutamyl hydrolase (ggm) mRNA	NM_001092691.1	2.92	11.51	2	0.009	4.24	4.22	0	0
Xenopus laevis cDNA clone MGCI:30954 (IMAGE:973902) complete cds	BC106382.1	2.21	8.61	2	0.009	1.9	2.87	0.6	0.3
PREDICTED: Xenopus (Silurana) tropicalis vesicle glycoprotein 2C (sv2c) mRNA	NM_002934827.2	4.76	18.55	2	0.017	0	0	0	0
Xenopus tropicalis finished cDNA clone TIBa05016	CU25142.1	0.41	1.63	2	0.029	0	0	0	0
Xenopus laevis deg03 mRNA complete sequence	DQ096846.1	1	3.92	2	0.039	0	0	0	0
Xenopus (Silurana) tropicalis mRNA for alpha 2-3-sialyltransferase ST3Gal V (st3gal5 gene)	PN550108.1	5.43	20.55	1.9	0.009	5.83	4.91	-0.2	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734405 (LOC101734405) mRNA	XM_004918247.1	0.98	3.56	1.9	0.009	0	0	0	0
Xenopus (Silurana) tropicalis G protein-coupled receptor 153 (gpr153) mRNA	NM_001128052.1	2.41	8.88	1.9	0.009	1.46	1.97	0.4	0.21
PREDICTED: Xenopus (Silurana) tropicalis T-box 15 (tbx15) mRNA	NM_002940981.2	1.47	5.66	1.9	0.009	1.39	1.91	0.5	0.26
Xenopus laevis uncharacterized LOC10036533 (LOC10036533) mRNA	NM_001097704.1	0.71	2.56	1.9	0.023	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane channel-like protein 7-like (LOC100493700) mRNA	NM_002932222.2	3.02	10.32	1.8	0.009	4.08	2.12	-0.9	0
PREDICTED: Xenopus (Silurana) tropicalis deoxyribonuclease gamma-like (LOC100497751) transcript variant X1 mRNA	NM_002938385.2	5.51	25.29	1.8	0.009	3.7	2.3	-0.7	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 13 (tmprs13) mRNA	NM_002932904.2	1.35	4.62	1.8	0.009	2.01	2.2	0.1	0.06
Xenopus laevis uncharacterized protein MGCI:30961 (MGCI:30961) mRNA	NM_001096456.1	6.76	24.27	1.8	0.009	7.5	8.91	0.2	0.11
PREDICTED: Xenopus (Silurana) tropicalis putative N-acetyltransferase 16-like (LOC100490742) mRNA	NM_002943182.1	0.69	2.4	1.8	0.009	0.83	1.49	0.8	0.44
Xenopus (Silurana) tropicalis cytochrome c-450 family 2 subfamily C polypeptide 18 (cyp2c18) mRNA	NM_001129801	2.4	8.33	1.8	0.009	2.38	4.47	0.9	0.5
Xenopus tropicalis hypothetical protein LOC1004765 mRNA (CDNA clone IMAGE:894708) partial cds	BC16733.1	0.49	1.54	1.7	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis aldehyde dehydrogenase 1 family member L2 (alhd1l2) mRNA	NM_002938076.2	1.05	3.4	1.7	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized protein MGCI:30961 (MGCI:30961) mRNA	NM_002936543.2	23.43	74.02	1.7	0.009	36.06	26.56	-0.4	0
PREDICTED: Xenopus (Silurana) tropicalis putative N-acetyltransferase 16-like (LOC100490742) mRNA	NM_002932703.2	1.17	3.75	1.7	0.009	1.91	1.36	-0.5	0
Xenopus laevis DNA clone IMAGE:7009602	EC09426.1	6.99	23.28	1.7	0.009	4.3	5.84	0.4	0.24
Xenopus (Silurana) tropicalis putative UDP-GlcNAcbetaGal beta-1,3-N-acetylglucosaminyltransferase LOC402377-like (LOC100487553)	NM_002935226.2	1.08	3.39	1.7	0.049	2567.99	4826.33	0.9	0.53
Xenopus (Silurana) tropicalis LOC1004765 mRNA (CDNA clone IMAGE:894708) partial cds	NM_001097307.1	6.16	19.11	1.6	0.009	10.1	7.2	-0.5	0
PREDICTED: Xenopus (Silurana) tropicalis aldehyde dehydrogenase 1 family member L2 (alhd1l2) mRNA	NM_001093118.1	0.92	2.83	1.6	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis frizzled family receptor 4 (frz4) mRNA	AF23711.1	1.38	4.27	1.6	0.009	1.39	1.48	0.1	0.06
PREDICTED: Xenopus (Silurana) tropicalis early growth response 3 (egfr) mRNA	NM_002932703.2	1.17	3.75	1.7	0.009	4.3	5.84	0.4	0.24
Xenopus laevis DNA clone IMAGE:7009602	EC09426.1	6.99	23.28	1.7	0.009	4.3	5.84	0.4	0.24
Xenopus (Silurana) POU class 4-homeobox 1 (pou4f1.2) mRNA	NM_001093118.1	0.92	2.83	1.6	0.009	0	0	0	0
Xenopus laevis nutrient peptide C (nppc) mRNA	NM_001093118.1	0.92	2.83	1.6	0.009	0	0	0	0
Xenopus laevis 7-transmembrane receptor 1 mRNA complete cds	BC072262.1	3.05	9.45	1.6	0.009	1.39	1.7	0.3	0.19
Xenopus laevis cDNA clone IMAGE:440551.5 partial cds	AB45086.1	11.03	32.84	1.6	0.009	6.33	8.94	0.5	0.31
Xenopus (Silurana) tropicalis 3 mRNA for xRippl3 protein complete cds	NM_001166041.1	5.19	15.41	1.6	0.009	5.65	8.34	0.6	0.37
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor 56 (gpr56) mRNA	NM_002931653.2	0.54	1.6	1.6	0.017	1.79	1.8	0	0
Xenopus laevis p21 GTPase-associated kinase 1 mRNA	BC081113.1	3.89	11.67	1.6	0.023	7.72	9.74	0.3	0.19
PREDICTED: Xenopus (Silurana) tropicalis kinase insert domain receptor (a type III receptor tyrosine kinase) (kdr) mRNA	NM_002934669.2	0.93	2.83	1.6	0.029	0	0	0	0
Xenopus laevis neurotrophin 3 (ntf3) mRNA	NM_001092740.1	1.34	4.18	1.6	0.039	1.34	1.67	0.3	0.19
PREDICTED: Xenopus (Silurana) tropicalis SH3 domain ankyrin repeat and PH domain 3 (asap3) mRNA	NM_00293393662	0.41	1.28	1.6	0.044	1.83	1.27	-0.5	0
Xenopus laevis p21 GTPase-associated kinase 1 mRNA	CT025400.2	2.23	6.19	1.5	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis sex determining region Y-box 2 (sox2) mRNA	NM_213704.3	39.86	116.36	1.5	0.009	49.47	53.9	0.1	0.07
Xenopus laevis regulator of cell cycle (rgcc) mRNA	NM_001093976.1	23.88	65.38	1.5	0.009	32.69	37.69	0.2	0.13
Xenopus laevis complement factor I (cf-i) mRNA	NM_001085952.1	2.77	7.86	1.5	0.023	1.33	1.5	0.2	0.13
Xenopus laevis Mgcb0983 protein (MGC80983) mRNA	NM_001092413.1	1.22	3.33	1.4	0.009	11.66	10.57	-0.1	0
Xenopus laevis tripartite motif containing 36 (trm36) mRNA	NM_001091117.1	5.23	14.11	1.4	0.009	5.83	5.6	-0.1	0
Xenopus laevis family with sequence similarity 101 member B (fam101b) mRNA	NM_001093870.1	8.49	22.36	1.4	0.009	9.24	9.48	0	0
Xenopus laevis SRY (sex determining region Y)-box 2 (sox2) mRNA	NM_001172213.1	7.83	21.11	1.4	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis CT1-N-terminal like (cttnbp2nl) transcript variant X1 mRNA	NM_00293282.2	1.48	3.99	1.4	0.009	1.51	1.71	0.2	0.14
Xenopus laevis RNA binding motif protein 24 (rbm24-a) mRNA	NM_001087526.1	2.43	6.31	1.4	0.009	2.61	3.11	0.3	0.21
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 198 member A (fam198a) mRNA	NM_002937853.2	2.6	6.71	1.4	0.009	2.49	3.11	0.3	0.21
Xenopus laevis cDNA clone IMAGE:8332229	BC155363.1	6.32	16.79	1.4	0.009	8.37	11.35	0.4	0.29
Xenopus (Silurana) tropicalis NMN23 family member 7 (nmn7) mRNA	NM_203572.1	17.48	45.42	1.4	0.009	14.29	18.28	0.4	0.29

Gene	Accession	Six1 CHX	Six1 CHX+DEX	FC Six1 q-value	FC Six1 q-value	Control CHX	Control CHX+DEX	FC Control	Rank <sup>a</sup>	
XLHOX7A	Xenopus laevis homeobox protein Xgpb-2 mRNA partial cds	U04867_1	10.68	28.6	1.4	0.049	11.26	101.7	-0.1	0
	Xenopus laevis pituitary adenylate cyclase-activating peptide (PACAP) mRNA complete cds	AF-18787_7_1	2	4.99	1.3	0.009	0	0	0	0
	Xenopus laevis LFNG O-fucosylpeptide 3-beta-N-acetylglucosaminyltransferase (lfng) mRNA	NM_001097707_1	4.12	9.92	1.3	0.009	5.68	5.16	-0.1	0
	Xenopus laevis (Silurana) tropicalis ras homolog member V (rho) mRNA	NM_001128659_1	15.93	39.42	1.3	0.009	21.1	193.5	-0.1	0
	Xenopus laevis cdc25B mRNA for cdc25B phosphatase complete cds	AB363840_1	8.31	20.41	1.3	0.009	7.44	6.95	-0.1	0
	Xenopus laevis undifferentiated 1 (apccdd1) mRNA	NM_001094563_1	13.14	32.48	1.3	0.009	15.19	116.7	-0.4	0
	Xenopus laevis adenomatous polyposis coli down-regulated 1 (apccdd1) mRNA	NM_001094199_1	4.99	12.61	1.3	0.009	6.58	5.29	-0.3	0
	Xenopus laevis ets-2a proto-oncogene mRNA (CdNA clone MGCI_60372 IMAGE:8544815) complete cds	BC1331_83_1	8.04	19.73	1.3	0.009	11.41	8.59	-0.4	0
PREDICTED:	Xenopus (Silurana) tropicalis uveal autoantigen with coiled-coil domains and ankyrin repeats-like (LOC101731603) mRNA	XK_004918328_1	3.81	9.21	1.3	0.009	5.29	6.08	0.2	0.15
PREDICTED:	Xenopus (Silurana) tropicalis family with sequence similarity 46 member A (fam46a) mRNA	XK_002938458_2	4.22	10.25	1.3	0.009	4.17	5.84	0.5	0.38
	Xenopus laevis cDNA clone IMAGE:5085355 partial cds	BC073731_1	5.37	12.92	1.3	0.023	3.75	4.44	0.2	0.15
	Xenopus (Silurana) tropicalis uveal autoantigen with coiled-coil domains and ankyrin repeats-like (LOC101731603) mRNA	NM_001102800_2	3.07	7.06	1.2	0.009	2.7	2.64	0	0
	Xenopus laevis cDNA clone IMAGE:631776_7 partial cds	BC071098_1	1.9	4.3	1.2	0.009	3.38	2.88	-0.2	0
	Xenopus (Silurana) tropicalis uveal autoantigen with coiled-coil domains and ankyrin repeats-like (LOC101731603) mRNA	NM_001113823_1	12.05	27.96	1.2	0.009	10.54	9.41	-0.2	0
	Xenopus laevis zinc finger protein 21.4 (znf21.4) mRNA	NM_001097402_1	169.67	376.92	1.2	0.009	152.98	127.99	-0.3	0
	Xenopus laevis CD81 protein (cd81-a) mRNA	NM_001086613_1	235.41	543.6	1.2	0.009	110.38	114.58	0.1	0.08
	Xenopus laevis tetraspanin 1 (tspan1) mRNA	NM_001095473_1	18.56	41.66	1.2	0.009	20.7	23.96	0.2	0.17
PREDICTED:	Xenopus (Silurana) tropicalis progestin and adipooQ receptor family member IX (pacq9) mRNA	NM_004914357_1	3.66	8.21	1.2	0.009	1.47	1.83	0.3	0.25
	Xenopus laevis mex3b mRNA for Mex-3 homolog b complete cds	AB499903_2	5.33	12.13	1.2	0.009	3.24	4.76	0.6	0.5
PREDICTED:	Xenopus (Silurana) tropicalis flocculation protein FL01-1-like (LOC100490389) transcript variant X1 mRNA	XK_002942555_2	2.8	6.33	1.2	0.017	1.76	1.6	-0.1	0
	Xenopus laevis pyruvate dehydrogenase phosphatase catalytic subunit 1 (pdph1) nuclear gene encoding mitochondrial protein mRNA	NM_001094221_1	221.69	516.82	1.2	0.017	48.01	41.03	-0.2	0
	Xenopus tropicalis finished cDNA clone TNeu0651_3	CU025050_1	3.82	8.82	1.2	0.029	1.86	1.55	-0.3	0
PREDICTED:	Xenopus (Silurana) tropicalis ATP-binding cassette sub-family D (ALD) member 2 (abcd2) transcript variant X1 mRNA	XK_004913002_1	0.91	2.08	1.2	0.035	0	0	0	0
PREDICTED:	Xenopus (Silurana) tropicalis hypermethylated in cancer 1 (hc1) transcript variant X2 mRNA	XK_004911637_1	0.76	1.7	1.2	0.044	0	0	0	0
	Xenopus laevis ectoderm neural cortex related 3 (Engr-3) mRNA complete cds	AY216793_1	43.03	89.5	1.1	0.009	55.05	45.63	-0.3	0
	Xenopus (Silurana) tropicalis v-HAT nucleophosmin oncogene homolog A (nufafa) mRNA	NM_001093204_1	1.26	2.75	1.1	0.009	0	0	0	0
	Xenopus laevis histidine ammonia-lyase gene (hal1) mRNA	NM_001093175_1	52.33	111.32	1.1	0.009	72.61	67.3	-0.1	0
PREDICTED:	Xenopus (Silurana) tropicalis Kruppel-like factor 15 (klf15) transcript variant X1 mRNA	NM_002938233_2	3.98	8.75	1.1	0.009	4.88	4.09	-0.3	0
	Xenopus laevis nudeE nuclear distribution E homolog (A_nudl1-a) mRNA	NM_001092863_1	3.07	6.44	1.1	0.009	3.21	2.84	-0.2	0
	Xenopus laevis nudE nuclear distribution E homolog (A_nudl1-a) mRNA	NM_001102878_1	6.64	14.34	1.1	0.009	6.24	6.13	0	0
	Xenopus (Silurana) tropicalis v-HAT nucleophosmin oncogene homolog A (nufafa) mRNA	NM_002939963_2	3.92	8.44	1.1	0.009	3.3	2.86	-0.2	0
	Xenopus laevis sex-determining region Y-box 2 mRNA complete cds	XM_002934358_2	1.82	3.78	1.1	0.009	2.04	1.79	-0.2	0
PREDICTED:	Xenopus (Silurana) tropicalis Nhs-like protein 1-like (LOC100493724) transcript variant X1 mRNA	AF59892_1	17.47	36.76	1.1	0.009	28.2	20.93	-0.4	0
	Xenopus laevis clone S10-46-56 mRNA sequence	AF005476_1	1650.04	3532.83	1.1	0.009	812.21	855.19	0.1	0.09
PREDICTED:	Xenopus (Silurana) tropicalis transcript variant X1 mRNA	NM_001093593_1	1.49	3.1	1.1	0.009	1.37	1.47	0.1	0.09
	Xenopus (Silurana) tropicalis immunoglobulin domain (Ig) transmembrane domain (TM) and short cytoplasmic domain (semaphorin) 4B (sema4b)	NM_001093593_1	4.85	10.6	1.1	0.009	2.06	2.16	0.1	0.09
	Xenopus (Silurana) tropicalis leucine zipper putative tumor suppressor 2 (litz2) mRNA	GQ292559_1	27.65	57.63	1.1	0.009	29.94	32.31	0.1	0.09
	Xenopus laevis sex-determining region Y-box 2 mRNA complete cds	NM_001095138_1	3.21	7.03	1.1	0.023	3.8	2.64	-0.5	0
PREDICTED:	Xenopus (Silurana) tropicalis Rho GTPase binding 3 (cdc42ep3) mRNA	XM_002932552_2	5.47	11.65	1.1	0.029	11.58	6.58	-0.3	0
	Xenopus laevis cDNA clone IMAGE:1776_1	BC099044_1	1.49	3.22	1.1	0.039	1.93	1.37	-0.5	0
PREDICTED:	Xenopus (Silurana) tropicalis Dickkopf-1 (Dkk1) mRNA complete cds	XM_002934454_2	7.67	15.02	1	0.009	8.05	7.02	-0.2	0
	Xenopus (Silurana) tropicalis leucine zipper putative tumor suppressor 2 (litz2) mRNA	CG075707_1	24.98	50.96	1	0.009	30.12	27.72	-0.1	0
PREDICTED:	Xenopus (Silurana) tropicalis sodium/hydrogen exchanger 2-like (LOC101732511) mRNA	NM_004911791_1	5.48	10.78	1	0.009	6.55	5.69	-0.2	0
	Xenopus laevis hypothetical protein LOC4436524 mRNA (CDNA clone IMAGE:5155631) partial cds	BC073512_1	16.76	33.64	1	0.009	12.91	10.53	-0.3	0
PREDICTED:	Xenopus (Silurana) tropicalis transmembrane protein 125 (tmem125) transcript variant X1 mRNA	NM_004913959_1	3.6	7.3	1	0.009	4.59	2.63	-0.8	0
	Xenopus laevis Dickkopf-1 (Dkk1) mRNA complete cds	AF030434_1	10.78	22.04	1	0.009	8.9	6.35	-0.5	0
PREDICTED:	Xenopus (Silurana) tropicalis leucine zipper putative tumor suppressor 2 (litz2) mRNA	NM_001095383_1	7.13	13.79	1	0.009	8.94	7.59	-0.2	0
	Xenopus laevis cDNA clone IMAGE:1776_1	NM_001096276_1	4.9	10.09	1	0.009	4.5	5.11	0.2	0.2
PREDICTED:	Xenopus (Silurana) tropicalis transmembrane protein 125 (tmem125) transcript variant X1 mRNA	NM_0011173404_1	24.01	46.73	1	0.009	27.44	31.78	0.2	0.2
	Xenopus laevis Dickkopf-1 (Dkk1) mRNA complete cds	AY913767_1	2.04	4.22	1	0.009	4.01	4.67	0.2	0.2
PREDICTED:	Xenopus (Silurana) tropicalis leucine zipper putative tumor suppressor 2 (litz2) mRNA	NM_001098714_1	17.14	33.53	1	0.009	15.58	21.08	0.4	0.4

Gene	Accession	Six1 CHX	Six1 CHX+DEX <sup>2</sup>	FC Six1 <sup>1</sup>	q-value	Control CHX	Control CHX+DEX <sup>3</sup>	FC Control	Rank <sup>7</sup>
PREDICTED: Xenopus (Silurana) tropicalis cAMP responsive element modulator (crem) transcript variant X2 mRNA	XM_004915472.1	4.63	9.24	1	0.023	0	0	0	0
Xenopus laevis peroxiredoxin 5 (prdx5) nuclear gene encoding mitochondrial protein mRNA	NW_001092111.1	18.82	36.59	1	0.029	15.94	18.58	0.2	0.2
PREDICTED: Xenopus (Silurana) tropicalis nuclear gene encoding mitochondrial protein mRNA	XM_004911827.1	4.37	8.94	1	0.035	6.57	4.24	-0.6	0
Xlaevis XFD2 mRNA for fork head protein	XJ4315.1	11.24	22.8	1	0.035	14.77	13.67	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730819 (LOC101730819) mRNA	XM_004915204.1	138.22	256.17	1	0.039	253.37	186.49	-0.4	0

<sup>1</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1 or Six1+Eya1 and CHX treatment  
<sup>2</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1 or Six1+Eya1 and CHX+DEX treatment  
<sup>3</sup>Log<sub>2</sub> Fold change values for Six1 or Six1+Eya1  
<sup>4</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment  
<sup>5</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment  
<sup>6</sup>Log<sub>2</sub> Fold change values for un-injected control  
<sup>7</sup>Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Six1 or Six1+Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## F.2 Six1<sub>m</sub>: Down-regulated genes

**Table F.2.** Significantly differentially expressed genes with at least two-fold down-regulation after injection of Six1-GR or Six1-GR+Eya1-GR and treatment with CHX + DEX

Gene	Accession	Six1 CHX <sup>1</sup>	Six1 CHX+DEX <sup>2</sup>	FC Six1 <sup>3</sup>	q-value	Control CHX <sup>4</sup>	Control CHX+DEX <sup>5</sup>	FC Control CHX+DEX <sup>6</sup>	Rank <sup>7</sup>
Xenopus laevis FEZ family zinc finger 2 (fez2) mRNA	NM_001056464.1	3.68	0.88	-2.1	0.023	3.56	3.74	0.1	0
Xenopus laevis respx44 mRNA complete sequence	DQ96861.1	68206.75	25945.22	-1.4	0.009	43840.89	36446.9	-0.3	0.21
PREDICTER; Xenopus (Silurana) tropicalis diencephalon/mesencephalon homeobox protein 1-A-like (LOC100493907) transcript variant 1	XM_002931403.2	18.47	7.18	-1.4	0.009	25.34	16.03	-0.7	0.5
Xenopus laevis paired domain transcription factor variant A (Fax-6) mRNA Pax-6-2 allele complete cds	AF154535.1	7.73	3.18	-1.3	0.009	0	0	0	0
XLU41839 Xenopus laevis thyroid hormone down-regulated protein (gene 18) mRNA complete cds	U41839.1	7.71	3.23	-1.3	0.049	13.65	10.9	-0.3	0.23
XLU43938 Xenopus laevis homeodomain protein (Xb-1b) mRNA complete cds	U43938.1	61.43	28.47	-1.1	0.009	0	0	0	0
Xenopus laevis chemokine (C-X-C motif) receptor 4 (cxcr4-a) mRNA	NM_001137581.1	24.76	11.88	-1.1	0.009	27.25	21.51	-0.3	0.27
Xenopus laevis cDNA clone MGEG3401284 complete cds	BC084658.1	1000277	4606.81	-1.1	0.009	13751.59	10505.89	-0.4	0.36
Xenopus laevis uncharacterized LOC100462907 (LOC100462907) mRNA	NM_001190395.1	9.07	4.44	-1	0.009	8.92	8.52	-0.1	0.1
Xenopus laevis cholesterol 25-hydroxylase mRNA (cDNA clone IMAGE8460553) partial cds	BC128955.1	48.72	2465	-1	0.009	18.38	14.76	-0.3	0.3
Xenopus (Silurana) tropicalis snail family zinc finger 1 (snail) mRNA	NM_2039396.1	80.64	41.55	-1	0.009	73.64	56.58	-0.4	0.4

<sup>1</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1 or Six1+Eya1 and CHX treatment

<sup>2</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1 or Six1+Eya1 and CHX+DEX treatment

<sup>3</sup>Log<sub>2</sub> Fold change values for Six1 or Six1+Eya1

<sup>4</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup>Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup>Rank = FC ( $\log_2$ ) Control/FC ( $\log_2$ ) Six1 or Six1+Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

### F.3 Eya1<sub>m</sub>: Up-regulated genes

**Table F.3** Significantly differentially expressed genes with at least two-fold up-regulation after injection of Eya1-GR or Six1-GR+Eya1-GR and treatment with CHX + DEX

Gene	Accession	Eya1 CHX <sup>a</sup>	Eya1 CHX+DEX <sup>b</sup>	FC	q-value	Control CHX <sup>c</sup>	Control CHX+DEX <sup>c</sup>	FC	Rank <sup>d</sup>
Xenopus tropicalis cDNA clone IMAGE:7022272	BC094950.1	0.1	6.52	6	0.016	0	0	0	0
Xenopus (Silurana) tropicalis 5'K homeobox 2 (skx2) mRNA	NM_001100275..1	0.6	18.62	5	0.009	0	0	0	0
Xenopus laevis uncharacterized LOC10036338 (LOC10036938) mRNA	NM_001097708..1	0.22	6.05	4.8	0.009	0	0	0	0
Xenopus laevis MGc8.3328 protein mRNA (cDNA clone MGC:83328 [IMAGE:6864428]) complete cds	BC024261..1	0.3	7.77	4.7	0.009	0	0	0	0
Xenopus laevis Myoblast determination protein 1 homolog A mRNA (cDNA clone MGC:52596 [IMAGE:4885255]) complete cds	BC041190..1	2.18	40.77	4.2	0.009	3.87	6.38	0.7	0.17
PREDICTED: Xenopus (Silurana) tropicalis ATP-sensitive inward rectifier potassium channel 1-like (LOC100492679) mRNA	XM_004916278..1	0.09	1.51	4.1	0.043	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox A1 (hoxa10) mRNA	NM_002933394..2	0.17	2.57	3.9	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 4 sodium bicarbonate cotransporter member 4 (slc4a4) transcript variant X3 mRNA	XM_004919134..1	0.16	2.32	3.9	0.016	0	0	0	0
Xenopus (Silurana) tropicalis glutathione peroxidase 2 (gastrintestinal) (gpx2) mRNA	NM_001256315..1	1.01	14.17	3.8	0.009	0	0	0	0
Xenopus (Silurana) tropicalis chemokine (C-X-C motif) receptor 7 (cxcr7) mRNA	NM_001030434..1	0.29	3.78	3.7	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis potassium voltage-gated channel shaker-related subfamily member 2 (kcnas2) mRNA	XM_004910736..1	0.12	1.45	3.6	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis $\times$ C-X-C motif chemokine 10-like (LOC100490785) mRNA	XM_002940578..2	0.42	4.9	3.5	0.009	0	0	0	0
Xenopus laevis hedgehog-interacting protein mRNA complete cds	AY328923..1	0.25	2.74	3.5	0.009	0	0	0	0
Xenopus laevis mRNA for Xox1/1 alpha protein	AJ001730..1	0.97	9.73	3.3	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis paired box 1 (pax1) transcript variant X1 mRNA	XM_002939437..2	0.18	3.3	0.009	0	0	0	0	0
Xenopus (Silurana) tropicalis ectodysplasin A receptor (edan) mRNA	NM_00106819..1	0.36	3.51	3.3	0.009	0.4	1.13	1.5	0.45
Xenopus laevis ectodysplasin A receptor (edan) mRNA	NM_001087047..1	0.27	2.66	3.3	0.023	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis hairy and enhancer of split 8 (hes8) mRNA	XM_0029333849..2	1.34	12.31	3.2	0.009	1.01	1.64	0.7	0.22
Xenopus (Silurana) tropicalis cytochrome b5 subfamily B polypeptide 1 (cyt26b1) mRNA	NM_001079187..2	0.42	3.62	3.1	0.009	3.54	1.95	-0.9	0
PREDICTED: Xenopus (Silurana) tropicalis espin (espn) transcript variant X1 mRNA	XM_002933556..2	0.22	1.83	3.1	0.034	1.81	1.76	0	0
Xenopus (Silurana) tropicalis A receptor (edan) mRNA	AB052691..1	0.75	5.99	3	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis hairy and enhancer of split 8 (hes8) mRNA	XM_002936646..2	0.49	4.04	3	0.048	0	0	0	0
Xenopus (Silurana) tropicalis sine oculis binding protein homolog (Drosophila) mRNA (cDNA clone MGC:172604 [IMAGE:7656239]) complete cds	BC154687..1	0.64	4.74	2.9	0.009	0	0	0	0
Xenopus (Silurana) tropicalis sine oculis binding protein homolog (Drosophila) mRNA (cDNA clone MGC:172604 [IMAGE:7656239]) complete cds	CR76056..2	0.69	4.66	2.8	0.009	1.03	1.1	0.1	0.04
Xenopus laevis mRNA for Xox1/1 alpha protein	NM_001093318..1	0.26	1.85	2.8	0.043	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis espin (espn) transcript variant X3 mRNA	XM_004916193..1	1.14	7.03	2.6	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox 2 (poj372-b) mRNA	NM_001096751..1	0.5	3.06	2.6	0.009	1.02	1.39	0.4	0.15
PREDICTED: Xenopus (Silurana) tropicalis growth factor independent 1 transcription repressor (gf1) mRNA	NM_002938303..2	0.65	4.05	2.6	0.009	0.79	1.33	0.8	0.31
Xenopus laevis cDNA clone IMAGE:7394277	BC106376..1	0.17	1.04	2.6	0.029	0	0	0	0
Xenopus laevis natruretic peptide C (nppc) mRNA	NM_001093312..1	0.26	1.85	2.8	0.043	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox 2 (poj372-b) mRNA	AF072455..1	1.28	7.32	2.5	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox 1 (titx1) transcript variant 2 mRNA	XM_002936768..2	1.57	8.36	2.4	0.009	0	0	0	0
Xenopus laevis uncharacterized protein (MGc648450 [MGc648450]) mRNA	NM_001059841..1	0.32	1.58	2.3	0.009	0	0	0	0
Xenopus laevis arginyl aminopeptidase (aminopeptidase B) b (mpep-b) mRNA	NM_001092079..1	0.31	1.4	2.2	0.023	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis potassium voltage-gated channel 1sk-related family member 1 (kcnel1) transcript variant X2 mRNA	NM_004912135..1	0.66	3	2.2	0.038	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis calmodulin-dependent protein kinase kinase 2 beta (camkk2) transcript variant X1 mRNA	NM_002937701..2	0.61	2.7	2.1	0.009	0	0	0	0
XLU76636 Xenopus laevis calbindin D28k mRNA complete cds	U76636..1	1.19	4.62	2	0.009	1.58	1.59	0.9	0.47
PREDICTED: Xenopus (Silurana) tropicalis aldehyde dehydrogenase 1 family member 12 (aldh12) mRNA	XM_002938070..2	0.78	3.03	2	0.009	0	0	0	0
Xenopus laevis chromogranin A (parathyroid secretory protein 1) (chga) mRNA	NM_001094724..1	3.93	15.29	2	0.009	3.65	3.66	0	0
PREDICTED: Xenopus (Silurana) tropicalis deoxyribonuclease gamma-like (LOC100497175) transcript variant X1 mRNA	NM_002938386..2	1.66	6.7	2	0.023	3.7	2.33	-0.7	0
PREDICTED: Xenopus (Silurana) laevis calbindin D28k mRNA complete cds	EF672727..1	0.35	1.38	2	0.043	0	0	0	0
Xenopus (Silurana) laevis calcitonin gene-related peptide (Sox1) mRNA	NM_001078754..1	0.98	3.77	1.9	0.009	0.66	1.23	0.9	0.47
Xenopus (Silurana) tropicalis hypothetical protein LOC100145765 mRNA (cDNA clone IMAGE:8944708) partial cds	BC16733..1	0.32	1.23	1.9	0.023	0	0	0	0
Xenopus tropicalis finished cDNA clone TNef052k18	CR76001..2	0.83	3.15	1.9	0.043	0.84	1.5	0.8	0.42
Xenopus laevis claudin 3 (cldn3) mRNA	NM_001093931..1	1.43	4.85	1.8	0.009	459.11	385	-0.3	0
PREDICTED: Xenopus (Silurana) tropicalis monocyte to macrophage differentiation-associated (mmd) transcript variant X1 mRNA	NM_002937811..2	0.47	5.25	1.8	0.029	3.32	2.75	-0.3	0
Xenopus laevis uncharacterized LOC10036333 (LOC10036933) mRNA	NM_001097704..1	0.91	3.15	1.8	0.029	0	0	0	0

Gene	Accession	Eva1 CHX	Eva1 CHX+DEX	FC Eva1 <sup>a</sup>	q-value	Control CHX	Control CHX+DEX <sup>b</sup>	FC Control CHX+DEX <sup>b</sup>	Rank <sup>c</sup>
Xenopus laevis pituitary adenylate cyclase-activating peptide (PACAP) mRNA complete cds	AF1878771	1.49	5.09	1.8	0.038	0	0	0	0
Xenopus laevis gamma-glutamyl hydrolase conjugate folylpolygamma glutamyl (hydrolyse) (ggf) mRNA	NM_001092691..1	3.66	11.6	1.7	0.009	4.24	4.22	0	0
Xenopus laevis mRNA for frizzled 4 protein (fz4 gene)	AJ251750..1	70.58	222.52	1.7	0.009	14.14	10.8	-0.4	0
Xenopus laevis SRY sex determining region Y-box 2 (sox2) mRNA	NM_213704..3	39.66	120.79	1.6	0.009	49.47	53.9	0.1	0.06
Xenopus laevis uncharacterized protein MGCI30961 (MGCI30961) mRNA	NM_00109656..1	7.58	23.12	1.6	0.009	7.5	8.91	0.2	0.12
Xenopus laevis neurotrophin 3 (ntf3) mRNA	NM_0010922740..1	1.6	4.97	1.6	0.009	1.34	1.67	0.3	0.19
Xenopus (Silurana) tropicalis G protein-coupled receptor 153 (grp153) mRNA	NM_00128052..1	2.27	7.04	1.6	0.009	1.46	1.97	0.4	0.25
PREDICTED: Xenopus (Silurana) tropicalis metallopeptidase trypsin-like (LOC100491951) mRNA	XM_002936336..2	1.89	5.73	1.6	0.016	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis DIX domain containing 1 (dixdc1) mRNA	XM_002932938..2	2.96	9.13	1.6	0.016	2.23	2.09	-0.1	0
Xenopus laevis cDNA clone MGCI30954 (MGCI30954) mRNA	BC106382..1	2.39	7.11	1.6	0.016	1.9	2.87	0.6	0.37
PREDICTED: Xenopus (Silurana) tropicalis ceramide kinase-like (cerkl) mRNA	XM_002932015..2	0.35	1.08	1.6	0.038	0	0	0	0
Xenopus laevis homeobox C8 (hoxc8) mRNA	NM_001127798..1	1.84	5.47	1.6	0.038	0	0	0	0
Xenopus (Silurana) tropicalis POU class A-homeobox 1 (pou4f1) mRNA	NM_001097307..1	6.77	19.73	1.5	0.009	10.1	7.2	-0.5	0
Xenopus laevis shisa-2 mRNA for Shisa2 complete cds	AB242597..1	1.76	5.02	1.5	0.009	0	0	0	0
Xenopus (Silurana) tropicalis mRNA for alpha 2-3-sialyltransferase ST3Gal V (st3gal5 gene)	FN550108..1	5.33	15	1.5	0.009	5.83	4.91	-0.2	0
activin beta B subunit (Xenopus laevis mRNA) [2716 nt]	S61773..1	1.52	4.38	1.5	0.009	1.86	1.35	-0.5	0
Xenopus laevis 7-transmembrane receptor frizzled-1 mRNA complete cds	AF231711..1	1.5	4.11	1.5	0.009	1.48	1.48	0.1	0.07
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 13 (tmprss13) mRNA	XM_002932904..2	1.99	5.68	1.5	0.009	2.01	2.2	0.1	0.07
PREDICTED: Xenopus (Silurana) tropicalis opsin-3-like (LOC104888771) mRNA	XM_002932623..2	0.87	2.48	1.5	0.016	1.34	1.23	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis monocyte to macrophage differentiation-associated (mmnd) transcript variant X2 mRNA	XM_004918560..1	2.09	6.09	1.5	0.034	2.24	1.37	-0.7	0
Xenopus tropicalis finished cDNA clone (Ga072h19)	CT02540..2	1.88	4.88	1.4	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis protein fbx-like (LOC104909871) transcript variant X2 mRNA	XM_004916957..1	3.17	8.6	1.4	0.009	0	0	0	0.07
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734405 (LOC101734405) mRNA	XM_004918247..1	1.31	3.46	1.4	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane channel-like protein 7-like (LOC100493700) mRNA	XM_002932222..2	3.53	9.47	1.4	0.009	4.08	2.12	-0.9	0.07
Xenopus laevis X15OX-2 (Sox2-2) mRNA complete cds	AF005476..1	1380.73	3642.54	1.4	0.009	812.21	355.19	0.1	0.07
Xenopus laevis complete factor 1 (clf-a) mRNA	NM_001085952..1	3.2	8.53	1.4	0.009	1.33	1.5	0.2	0.14
Xenopus laevis regulator of cell cycle (rcg) RNA	NM_001093976..1	23.27	59.66	1.4	0.009	32.69	37.69	0.2	0.14
Xenopus (Silurana) tropicalis family member 7 (nmf7) mRNA	NM_203572..1	15.81	41.26	1.4	0.009	14.29	18.28	0.4	0.29
PREDICTED: Xenopus (Silurana) tropicalis T-box 15 (tbx15) mRNA	XM_002940981..2	2.19	5.73	1.4	0.009	1.39	1.91	0.5	0.36
Xenopus laevis xRIPPLY3 mRNA for xRIPPLY3 protein complete cds	AB45506..1	16.01	42.82	1.4	0.009	6.33	8.94	0.5	0.36
PREDICTED: Xenopus (Silurana) tropicalis hypermethylated in cancer 1 (hic1) transcript variant X2 mRNA	XM_004911637..1	0.55	1.41	1.4	0.016	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis frizzled family receptor 4 (frz4) mRNA	XM_002936543..2	29.08	72.12	1.3	0.009	36.06	26.56	-0.4	0
PREDICTED: Xenopus (Silurana) tropicalis cAMP responsive element modulator (crem) transcript variant X2 mRNA	XM_004915472..1	5.29	13.44	1.3	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis ATP-binding cassette sub-family D (ALD) member 2 (abcd2) transcript variant X1 mRNA	XM_004913002..1	0.84	2.08	1.3	0.009	1.9	0.37	-0.5	0
Xenopus laevis cDNA clone IMAGE3.747628	BC09904..1	1.68	4.07	1.3	0.009	1.93	1.37	-0.5	0
Xenopus laevis tripartite motif-containing 36 (trm36) mRNA	NM_001092413..1	5.13	12.36	1.3	0.009	5.83	5.6	-0.1	0
Xenopus (Silurana) tropicalis RAS-like family 11 member B (ras11b) mRNA	NM_001091117..1	3.62	8.75	1.3	0.009	5.38	6.94	0.4	0.31
Xenopus laevis B-cell CLL/lymphoma 6 (bcl6) mRNA	NM_001095069..1	3.63	8.75	1.3	0.016	7.22	4.63	-0.6	0
PREDICTED: Xenopus (Silurana) tropicalis early growth response 3 (egfr3) mRNA	NM_002937032..2	1.29	3.26	1.3	0.034	1.91	1.36	-0.5	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox C12 (hoxc12) mRNA	XM_002936647..2	4.52	10.62	1.2	0.009	4.65	3.59	-0.4	0
Xenopus laevis MGC80983 protein (MGC80983) mRNA	NM_001092413..1	1.3	3.09	1.2	0.009	11.66	10.57	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730819 (LOC101730819) mRNA	XM_004915204..1	68.29	161.1	1.2	0.009	253.37	186.49	-0.4	0
Xenopus laevis B-cell CLL/lymphoma nucleotide dissociation stimulator (balb) mRNA	NM_00113823..1	11.37	25.31	1.2	0.009	10.54	9.41	-0.2	0
PREDICTED: Xenopus (Silurana) tropicalis LLOC503674 (LLOC503674) mRNA	NM_001095563..1	11.62	26.19	1.2	0.009	15.19	11.67	-0.4	0
Xenopus laevis ets-2a proto-oncogene mRNA (cdna clone MGCI60372) (MAGE8544815) complete cds	BC133183..1	6.83	15.65	1.2	0.009	11.41	8.59	-0.4	0
Xenopus laevis adenomatous polyposis coli down-regulated 1 (apcdcl1) mRNA	NM_001094109..1	5.05	11.69	1.2	0.009	6.58	5.29	-0.3	0
Xenopus laevis RNA binding motif protein 2 (rbm24-al) mRNA	NM_00108526..1	2.49	5.79	1.2	0.009	2.61	3.1	0.3	0.25
PREDICTED: Xenopus (Silurana) tropicalis CTTNBP2 N-terminal like (cttnbp2nl) transcript variant X2 mRNA	NM_004910727..1	1.66	3.77	1.2	0.023	0	0	0	0
Xenopus laevis mal T-cell differentiation protein (mal) mRNA	NM_00108657..1	6.6	15.66	1.2	0.029	10.99	11.74	0.1	0.08
Xenopus laevis SRY (sex determining region Y-box 21 (sox21) mRNA	NM_001172213..1	9.68	21.48	1.1	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis CA/MH responsive element modulator (crem) transcript variant X3 mRNA	NM_004915473..1	3.57	7.65	1.1	0.009	0	0	0	0

Gene	Accession	Eya1 CHX	Eya1 CHX+DEX <sup>2</sup>	FC Eya1 <sup>1</sup>	q-value	Control CHX	Control CHX+DEX <sup>3</sup>	FC Control	Rank <sup>4</sup>
PREDICTED: Xenopus (Silurana) tropicalis E74-like factor 3 (ets domain transcription factor epithelial-specific) (lef3) mRNA	XM_002932989.2	27.12	56.63	1.1	0.009	75.33	38.66	-1	0
Xenopus (Silurana) tropicalis ras homolog family member V (rhov) mRNA	NM_001128659.1	13.04	28.7	1.1	0.009	21.1	19.35	-0.1	0
Xenopus laevis LFNG O-fucosylpeptide 3-beta-N-acetylglucosaminyltransferase (lfng) mRNA	NM_001097707.1	5.32	11.56	1.1	0.009	5.68	5.16	-0.1	0
Xenopus laevis family with sequence similarity 101 member B (fam101b) mRNA	NM_001093870.1	8.52	18.05	1.1	0.009	9.24	9.48	0	0
Xenopus laevis Dickkopf-1 (Xdkk-1) mRNA complete cds	AF030434.1	8.87	19.3	1.1	0.023	8.9	6.35	-0.5	0
PREDICTED: Xenopus (Silurana) tropicalis uveal autotagigen with coiled-coil domains and ankyrin repeats-like (LOC101731603) mRNA	XM_004918328.1	4.05	8.65	1.1	0.023	5.29	6.08	0.2	0.18
Xenopus laevis cdc25Ba mRNA for cdc25B phosphatase complete cds	AB363840.1	8.17	16.26	1	0.009	7.44	6.95	-0.1	0
Xenopus laevis paired box transcription factor (Pax1) mRNA complete cds	JQ929179.1	6.02	12.04	1	0.009	7.15	6.55	-0.1	0
Xenopus laevis ectoderm neural cortex related-3 (Encl-3) mRNA complete cds	AY216793.1	42.61	82.56	1	0.009	55.05	45.63	-0.3	0
Xenopus (Silurana) tropicalis DnaJ (Hsp40) homolog subfamily C member 18 (dnajc18) mRNA	NM_001102878.1	7.4	15.01	1	0.009	6.24	6.13	0	0
Xenopus laevis XI-1-ets-2b protein mRNA (cDNA clone MGC:66631306) complete cds	BC077264.1	16.86	34.6	1	0.009	21.13	18.01	-0.2	0
Xenopus laevis sex-determining region Y-box 2 mRNA complete cds	GO292559.1	26.95	54.65	1	0.009	29.94	32.31	0.1	
Xenopus laevis tetraspanin 1 (span1) mRNA	NM_001095473.1	15.82	30.74	1	0.009	20.7	23.96	0.2	
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 198 member A (fam198a) mRNA	XM_002937853.2	2.92	5.92	1	0.016	2.49	3.11	0.3	
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 46 member A (fam46a) mRNA	XM_002938458.2	5.28	10.62	1	0.034	4.17	5.84	0.5	

<sup>1</sup> Expression levels (FPKM) in pre-placodal explants after injection of Eya1 or Six1+Eya1 and CHX treatment

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Eya1 or Six1+Eya1 and CHX+DEX treatment

<sup>3</sup> Log<sub>2</sub> Fold change values for Eya1 or Six1+Eya1

<sup>4</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup> Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup> Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Eya1 or Six1+Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

#### F.4 Eya1<sub>m</sub>: Down-regulated genes

**Table F.4** Significantly differentially expressed genes with at least two-fold down-regulation after injection of Eya1-GR or Six1-GR+Eya1-GR and treatment with CHX + DEX

Gene	Accession	Eya1 CHX <sup>1</sup>	Eya1 CHX+DEX <sup>2</sup>	FC Eya1 <sup>3</sup>	q-value Eya1 <sup>3</sup>	Control CHX <sup>4</sup>	Control CHX+DEX <sup>5</sup>	FC Control	Rank <sup>6</sup>
Xenopus laevis Similar to myosin light polypeptide 9 regulatory clone IMAGE:5570659 mRNA	BC042231.1	14.85	0.02	-95	0.038	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101735120 (LOC101735120) mRNA	XM_004919916.1	13.41	0.02	-9.4	0.009	0	0	0	0
XELRGAOP3 x.laevis socyce 5srRNA pseudogene clone pXe631	K01368.1	419.49	5.34	-6.3	0.009	1.56	1.34	-0.2	0.93
Medicago truncatula Glucan 1-3-beta-glucosidase (MTR_8g091310) mRNA complete cds	XM_003630009.1	5.94	0.31	-4.3	0.009	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis vomeronasal type-2 receptor 26-like (LOC100487329) partial mRNA	XM_002943234.2	963.48	190.84	-2.3	0.009	33.81	27.61	-0.3	0.13
Xenopus tropicalis finished cDNA clone TNel07616	CR84409.2	6.06	1.21	-2.3	0.048	0	0	0	0
Xenopus laevis FEZ family zinc finger 2 (fezf2) mRNA	NM_001096464.1	3.06	0.68	-2.2	0.043	3.56	3.74	0.1	0
Xenopus laevis DMR14 (Dmrt4) mRNA complete cds	AY1648303.1	5.63	1.79	-1.7	0.034	7.38	7.14	0	0
XELB1AA_X.borealis B1 protein mRNA complete cds	M63663.1	24155.96	8217.38	-1.6	0.009	4051.45	3420.13	-0.2	0.12
PREDICTED: Xenopus (Silurana) tropicalis diencephalon/mesencephalon homeobox protein 1-A-like (LOC100493907) transcript variant 1	XM_002931403.2	19.06	7.11	-1.4	0.009	25.34	16.03	-0.7	0.5
Xenopus laevis repetitive 1723 element fragment C17	X00077.1	234.66	103.84	-1.2	0.009	97.58	125.81	0.4	0
PREDICTED: Xenopus (Silurana) tropicalis elastin microfibril interfacer 3 (emilin3) mRNA	XM_002932865.2	1.8	0.84	-1.1	0.009	1.38	1.92	0.5	0
X.laevis mRNA for Xom protein	X98454.1	34.65	16.95	-1	0.009	21.45	16.65	-0.4	0.4

<sup>1</sup>Expression levels (FPKM) in pre-placodal explants after injection of Eya1 or Six1+Eya1 and CHX treatment

<sup>2</sup>Expression levels (FPKM) in pre-placodal explants after injection of Eya1 or Six1+Eya1 and CHX+DEX treatment

<sup>3</sup>Log<sub>2</sub> Fold change values for Eya1 or Six1+Eya1

<sup>4</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup>Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup>Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Eya1 or Six1+Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## F.5 Six1+Eya1<sub>m</sub>: Up-regulated genes

**Table F.5** Significantly differentially expressed genes with at least two-fold up-regulation after injection of Six1-GR or Eya1-GR or Six1-GR+Eya1-GR and treatment with CHX + DEX

Gene	Accession	Six1-Eya1		Six1-Eya1		Six1-Eya1		Control		FC CHX+DEX*		Rank*
		CHX	CHX+DEX	CHX	CHX+DEX	Eva 1 <sup>a</sup>	FC Six1-Eya1	q-value	CHX	CHX+DEX*	Control	
Xenopus laevis cytochrome b reductase 1 (cyb1) mRNA	NM_001092967.1	0.02	2.96	7.2	0.033	15.09	18.42	0.3	0.04			
PREDICTED: Xenopus (Silurana) tropicalis parvalbumin thymic CPV-3-like (LOC100485867) transcript variant X1 mRNA	XM_002931951.1	0.03	3.27	6.8	0.008	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis glutathione peroxidase 2 (gastrintestinal) (gp-x2) mRNA	NM_001256315.1	0.12	8.24	6.1	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis zona pellucida-like domain-containing protein 1-like (LOC101732051) mRNA	XM_004916514.1	0.02	1.26	6	0.008	0	0	0	0	0	0	0
Xenopus tropicalis cDNA clone IMAGE:7022_272	BC094950.1	0.1	5.87	5.9	0.008	0	0	0	0	0	0	0
Xenopus laevis MGC83328 protein mRNA (cDNA clone MGCI03328 IMAGE:684428) complete cds	BC082426.1	0.02	12.27	5.8	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family, 26 member 7 (sc26a7) mRNA	XM_002934422.2	0.02	1.13	5.8	0.004	0	0	0	0	0	0	0
Xenopus laevis zinc finger protein 214 (znf214) mRNA	NM_001097042.1	0.09	4.61	5.7	0.008	152.98	127.99	-0.3	-0.4	0	0	0
Xenopus tropicalis finished cDNA clone Thel107m13	CU075760.1	0.05	2.04	5.4	0.008	243.21	186.6	-0.4	0	0	0	0
Xenopus laevis uncharacterized LOC100036938 (LOC100036938) mRNA	NM_001097708.1	0.18	5.38	4.9	0.008	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis SIX homeobox 2 (six2) mRNA	NM_001100275.1	0.58	17.27	4.9	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734405 (LOC101734405) mRNA	XM_004918247.1	0.08	1.91	4.6	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis growth factor independent 1 transcription repressor (gf1) mRNA	NM_001253803.2	0.2	3.18	4	0.008	0.79	1.33	0.8	0.2	0	0	0
Xenopus laevis Myoblast determination protein 1 homolog mRNA (cDNA clone MGCI52596 IMAGE:4985255) complete cds	BC041190.1	2.62	39.49	3.9	0.008	3.87	6.38	0.7	0.18	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis potassium voltage-gated channel shaker-related subfamily member 2 (kcna2) mRNA	XM_004910736.1	0.11	1.56	3.8	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 4 sodium bicarbonate cotransporter member 4 (slc4a4) transcript variant X3 mRNA	XM_004919134.1	0.14	1.76	3.7	0.008	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis ectodysplasin A receptor (edar) mRNA	NM_001006819.1	0.24	3.11	3.7	0.008	0.4	1.13	1.5	0.41	0	0	0
Xenopus laevis hypothetical protein LOC443654 mRNA (cDNA clone IMAGE:5155631) partial cds	BC073512.1	0.65	8.15	3.6	0.008	12.91	10.53	-0.3	0	0	0	0
Xenopus laevis paired box transcription factor (Pax1) mRNA complete cds	JQ29179.1	0.33	4.13	3.6	0.008	7.15	6.55	-0.1	0	0	0	0
Xenopus laevis mRNA for Xsox7/alpha protein	AJ001730.1	0.55	6.2	3.5	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis putative UDP-GlcNAc beta-1,3-N-acetylglucosaminyltransferase LOC402377-like (LOC100487553)	XM_002932262.2	0.15	1.68	3.5	0.008	256.79	482.63	0.9	0.26	0	0	0
Xenopus (Silurana) tropicalis cytochrome P450 family 26 subfamily B polypeptide 1 (cyt26b1) mRNA	NM_00109187.2	0.26	2.68	3.4	0.008	3.54	1.95	-0.9	0	0	0	0
Xenopus (Silurana) tropicalis chemokine (C-X-C motif) receptor 7 (cxcr7) mRNA	NM_001030434.1	0.33	3.52	3.4	0.008	0	0	0	0	0	0	0
Xenopus laevis RDC1 like protein mRNA (cDNA clone MGCI114801 IMAGE:46432706) complete cds	BC098974.1	0.53	5.57	3.4	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis calcium/calmodulin-dependent protein kinase kinase 2 beta (camkk2) transcript variant X1 mRNA	NM_002937701.2	0.16	1.62	3.3	0.008	0	0	0	0	0	0	0
Xenopus (Silurana) tropicalis G protein-coupled receptor 153 (gpr153) mRNA	NM_001128052.1	0.47	4.65	3.3	0.008	1.46	1.97	0.4	0.12	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis leucine rich adaptor protein 1-like (lrap1) mRNA	XM_002940127.2	0.23	2.05	3.2	0.008	0	0	0	0	0	0	0
Xenopus laevis hedgehog-interacting protein mRNA complete cds	AY328923.1	0.21	1.88	3.2	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis c-X-C motif chemokine 10-like (LOC100490785) mRNA	XM_002940578.2	0.43	3.8	3.1	0.008	0	0	0	0	0	0	0
Xenopus laevis sulfatotransferase family cytosolic 2B member 1 (slt2b1) mRNA	NM_001097677.1	0.35	2.95	3.1	0.008	197.68	216.54	0.1	0.03	0	0	0
Xenopus laevis reticulin 1-C2 (Rtn1-C2) mRNA complete cds	AF498369.1	0.2	1.57	3	0.008	18.97	18.01	-0.1	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis hairy and enhancer of split 8 (hes8) mRNA	XM_002933849.2	1.31	10.75	3	0.008	1.01	1.64	0.7	0.23	0	0	0
Xenopus laevis mRNA for XSox7/Talpah2 complete cds	BC153370.1	2.15	15.86	2.9	0.008	454.26	191.26	-1.2	0	0	0	0
Xenopus laevis gelatinase B (MMP-9) mRNA complete cds	AB05269.1	0.63	4.37	2.8	0.008	0	0	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis paired box 1 (pax1) transcript variant X1 mRNA	AF072455.1	0.9	6.23	2.8	0.008	0	0	0	0	0	0	0
Xenopus laevis reticulin 1-C2 (Rtn1-C2) mRNA complete cds	XM_002933856.2	1.29	8.99	2.8	0.008	1.81	1.76	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis X-linked inhibitor of apoptosis (xiap) mRNA	NM_001030412.1	0.37	2.57	2.8	0.008	0	0	0	0	0	0	0
Xenopus laevis eva-1 homolog A (eva1a) mRNA	NM_001102800.2	0.38	2.68	2.8	0.008	2.7	2.64	0	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis paired box 1 (pax1) transcript variant X1 mRNA	NM_002939437.2	0.22	1.46	2.7	0.008	0	0	0	0	0	0	0
Xenopus laevis natriuretic peptide C (nppc) mRNA	NM_001093118.1	0.27	1.71	2.7	0.008	0	0	0	0	0	0	0
Xenopus laevis POU class 3 homeobox 2 (pou3f2-b) mRNA	NM_001096751.1	0.45	2.94	2.7	0.008	1.02	1.39	0.4	0.15	0	0	0
Xenopus tropicalis sine oocells binding protein (Drosophila) mRNA (cDNA clone MGCI03329) complete cds	BC154687.1	0.76	4.74	2.6	0.008	0	0	0	0	0	0	0
Xenopus laevis eva-1 homolog A (eva1a) mRNA	CR760562.2	0.57	3.53	2.6	0.008	1.03	1.1	0.1	0.04	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis inward-rectifying channel subfamily J member 1 (Kcnj8) mRNA complete cds	HQ260327.1	0.17	1.02	2.6	0.025	0	0	0	0	0	0	0
Xenopus laevis von Willebrand factor A domain-containing protein 2 (MGC80919) mRNA	NM_001093583.1	0.72	4.21	2.5	0.008	8.94	7.59	-0.2	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 2-like (LOC100491930) mRNA	XM_002932252.2	0.83	4.67	2.5	0.008	11.58	6.58	-0.8	0	0	0	0

Gene	Accession	Six1-Eya1 CHX	Six1-Eya1 GFX+DX <sup>2</sup>	FCSix1- Eya1 <sup>1</sup>	q-value	Control CHX	Control CHX+DX <sup>2</sup>	FC Control CHX+DX <sup>2</sup>	Rank <sup>7</sup>
PREDICTED: Xenopus (Silurana) tropicalis T-cell leukemia homeobox / (tlx1) transcript variant 1 mRNA	XM_002936768.2	1.36	7.34	2.4	0.008	1.12	1.47	0.4	0.17
Xenopus laevis cDNA clone MAGE6635346	BC168524.1	0.48	2.35	2.3	0.008	88.44	76.06	-0.2	0
PREDICTED: Xenopus (Silurana) tropicalis aldehyde dehydrogenase 1 family member L2 / (aldh1l2) mRNA	XM_002938070.2	0.32	1.6	2.3	0.008	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis Skt-related family member 1 (skrt1) mRNA	XM_002932958.2	0.21	1.02	2.3	0.008	3.32	3.08	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis potassium voltage-gated channel Skt-related family member 1 (kcnel1) transcript variant X2 mRNA	XM_004912351.1	0.69	3.08	2.2	0.008	0	0	0	0
Xenopus laevis SCL/TAL1 interrupting locus (stil) mRNA	NM_001091352.1	1.5	6.81	2.2	0.008	13.12	10.5	-0.3	0
Xenopus laevis shisa-2 mRNA for Shisa-2 complete cds	AB242597.1	0.89	3.99	2.2	0.008	0	0	0	0
Xenopus laevis uncharacterized protein LOC495015 (LOC495015) mRNA	NM_001094721.1	0.27	1.28	2.2	0.008	21.32	19.21	-0.2	0
Xenopus laevis uncharacterized protein MGCC68450 (MGCC68450) mRNA	NM_001089841.1	0.33	1.42	2.1	0.008	0	0	0	0
Xenopus laevis pituitary adenylate cyclase-activating peptide (PACAP) mRNA complete cds	AF187877.1	0.69	3.03	2.1	0.008	0	0	0	0
Xenopus laevis ankyrin repeat and BTB/POZ domain containing 2 (abtb2) mRNA	NM_001094801.1	0.7	3.01	2.1	0.008	155.02	152.05	0	0
Xenopus laevis uncharacterized protein MGCS0223 (MGCS0223) mRNA	NM_001091687.1	1.85	7.96	2.1	0.008	11.83	12.78	0.1	0.05
Xenopus tropicalis finished cDNA clone Theud52k18	CR760018.2	0.71	3.06	2.1	0.008	0.84	1.5	0.8	0.38
Xenopus laevis transforming growth factor beta-induced 68kDa (tgfb1) mRNA	NM_001095238.1	0.25	1.04	2.1	0.004	0	0	0	0
Xenopus laevis glial cell line-derived neurotrophic factor (GDNF) mRNA complete cds	DQ77994.1	0.87	3.48	2	0.008	21.14	21.26	0	0
Xenopus laevis arginyl aminopeptidase (aminopeptidase B) b (rnpepb) mRNA	NM_001092079.1	0.29	1.19	2	0.008	0	0	0	0
Xenopus laevis claudin 3 (cldn3) mRNA	NM_001093931.1	0.96	3.75	2	0.008	459.11	385	-0.3	0
Xenopus laevis hypothetical LOC495621 mRNA (cDNA clone MGCS81644 IMAGE6664239) complete cds	BC084933.1	1.92	7.84	2	0.008	14.55	10.89	-0.4	0
XL76636: Xenopus (Silurana) tropicalis desoxyribonuclease gamma-like (LOC100497175) transcript variant X1 mRNA	U76636.1	1.09	4.39	2	0.008	1.58	1.59	0	0
PREDICTED: Xenopus (Silurana) tropicalis calbindin D28k mRNA complete cds	NM_002938386.2	0.9	3.63	2	0.008	3.7	2.33	-0.7	0
Xenopus laevis Grb2/SH3 adapter Grb2 mRNA (cDNA clone MGCS81797 IMAGE7007774) complete cds	BC074118.1	0.89	3.52	2	0.008	84.34	87.79	0.1	0.05
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 13 (tmprss13) mRNA	NM_001093930.2	0.98	3.81	2	0.008	2.01	2.2	0.1	0.05
Xenopus laevis cardia myosin heavy chain-alpha (MHC-alpha) mRNA complete cds	AY913767.1	0.33	1.28	2	0.008	4.01	4.67	0.2	0.1
Xenopus (Silurana) tropicalis atlasin GTPase 1 (atl1) mRNA	NM_001078754.1	0.62	2.51	2	0.008	0.66	1.23	0.9	0.45
Xenopus (Silurana) tropicalis cytochrome P450 mRNA complete sequence	NM_001112980.1	1.42	5.56	2	0.008	2.38	4.47	0.9	0.45
Xenopus laevis degl3 mRNA complete sequence	DQ09686.1	0.72	2.81	2	0.014	0	0	0	0
Xenopus laevis SCY1-like 3 (scy3) mRNA	NM_001091462.1	0.28	1.09	2	0.029	74.46	89.43	0.3	0.15
Xenopus laevis carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2 (chs2) mRNA	NM_001127775.1	1.43	5.48	1.9	0.008	21.03	18.54	-0.2	0
PREDICTED: Xenopus (Silurana) tropicalis early growth response 3 (egf3) mRNA	XM_002932703.2	0.59	2.17	1.9	0.008	1.91	1.36	-0.5	0
Xenopus laevis uncharacterized LOC495396 (LOC495396) mRNA	NM_001095055.1	1.16	4.45	1.9	0.008	88.4	74.24	-0.3	0
Xenopus laevis caspase 1 apoptosis-related cysteine peptidase (casp1-a) mRNA	NM_001087754.1	0.75	2.71	1.9	0.014	5.42	5.59	0	0
PREDICTED: Xenopus (Silurana) tropicalis anocatin 2 (ano2) mRNA	XM_002932297.2	0.37	1.42	1.9	0.033	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis monocyte to macrophage differentiation-associated (mmd) transcript variant X1 mRNA	XM_002937811.2	1.25	4.33	1.8	0.008	3.32	2.75	-0.3	0
Xenopus laevis v-maf musculoaponeurotic fibrosarcoma oncogene homolog A (mafA) mRNA	NM_00101032304.1	0.53	1.83	1.8	0.008	0	0	0	0
Xenopus laevis chromogranin A (parathyroid secretory protein 1) (chga) mRNA	NM_001094724.1	3.73	13.18	1.8	0.008	3.65	3.66	0	0
PREDICTED: Xenopus (Silurana) tropicalis homeobox protein Hox-C13-like (LOC100487255) mRNA	XM_002936645.2	0.79	2.8	1.8	0.008	3.52	3.33	-0.1	0
Xenopus laevis zinc finger domain containing 1 (gata1) mRNA	NM_001087134.1	0.62	2.13	1.8	0.008	48.48	51.84	0.1	0.06
Xenopus laevis pyruvate phosphatase catalytic subunit 1 (pdpl1) nuclear gene encoding mitochondrial protein mRNA	NM_001094221.1	15.51	55.79	1.8	0.02	48.01	41.03	-0.2	0
Xenopus tropicalis finished cDNA clone TBAA05016	CU025142.1	0.35	1.23	1.8	0.029	0	0	0	0
Xenopus laevis similar to RNA binding motif protein 14 mRNA (cdNA clone IMAGE5542213) partial cds	BC046684.1	1.27	4.14	1.7	0.008	60.94	56.52	-0.1	0
Xenopus laevis uncharacterized LOC100036933 (LOC100036933) mRNA	NM_001097704.1	0.81	2.59	1.7	0.008	0	0	0	0
Xenopus laevis gamma-glutamyl hydrolase (conjugate poly(gamma-glutamyl hydrolase) (ggf) mRNA	NM_001092691.1	3.15	10.55	1.7	0.008	4.24	4.22	0	0
Xenopus (Silurana) tropicalis leucine zipper putative tumor suppressor 2 (lzs2) mRNA	NM_001006720.1	1.87	6.03	1.7	0.008	2.06	2.16	0.1	0.06
Xenopus laevis empty splices homeobox 2 (emx2) mRNA	BC106382.1	2.14	7.02	1.7	0.008	1.9	2.87	0.6	0.35
Xenopus laevis similar to RNA binding motif protein 14 (emx1) mRNA	NM_001093430.1	0.52	1.73	1.7	0.044	0	0	0	0
Xenopus laevis uncharacterized LOC10172165.1	NM_001092691.1	0.41	1.22	1.6	0.008	2.91	1.54	-0.9	0
Xenopus (Silurana) tropicalis mRNA for alpha-1 (I) fucosyltransferase 5 (fut5 gene)	FN530108.1	5.1	15.37	1.6	0.008	5.83	4.91	-0.2	0
Xenopus laevis empty splices homeobox 2 (emx2) mRNA	BC153804.1	2.17	6.38	1.6	0.008	127.86	115.55	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis ATP-binding cassette sub-family D (ALD) member 2 (abcd2) transcript variant X1 mRNA	NM_004913002.1	0.37	1.09	1.6	0.008	0	0	0	0
Xenopus laevis MGC80983 protein (MGC80983) mRNA	NM_001092413.1	0.65	1.93	1.6	0.008	11.66	10.57	-0.1	0

Gene	Accession	Six1-Eya1 CHX	Six1-Eya1 GFX+DEX <sup>2</sup>	FCSix1- Eya1 <sup>1</sup>	q-value	Control CHX	Control CHX-DEX <sup>2</sup>	FC Control CHX-DEX <sup>2</sup>	Rank <sup>7</sup>
Xenopus laevis CD81 protein (cd81-1) mRNA	NM_001086613.1	1.27	3.92	16	0.008	110.38	114.58	0.1	0.06
Xenopus laevis uncharacterized protein MGCI30961 (MGCI30961) mRNA	NM_001096456.1	6.65	19.59	16	0.008	7.5	8.91	0.2	0.12
Xenopus (Silurana) tropicalis family with sequence similarity 107 member A (fam107a) mRNA	NM_001128182.1	0.67	2.1	1.6	0.008	42.99	53.05	0.3	0.19
Xenopus laevis neurotrophin 3 (ntf3) mRNA	NM_001092740.1	1.41	4.17	1.6	0.008	1.34	1.67	0.3	0.19
Xenopus laevis cDNA clone IMAGE:4405515 partial cds	BC072262.1	2.23	6.92	1.6	0.008	1.39	1.7	0.3	0.19
PREDICTED: Xenopus (Silurana) tropicalis T-box 15 (tbx15) mRNA	XM_002940981.2	1.38	4.31	1.6	0.008	1.39	1.91	0.5	0.31
PREDICTED: Xenopus (Silurana) tropicalis flocculation protein FLO1-like (LOC100490389) transcript variant X1 mRNA	XM_0029425552.0	0.51	1.53	1.6	0.014	1.76	1.6	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis synaptic vesicle glycoprotein 2c (sv2c) mRNA	XM_0029348272.0	4.8	14.96	1.6	0.029	0	0	0	0
Xenopus laevis CD38 mRNA for CD38 complete cds	AB19489.1	0.9	2.81	1.6	0.04	43.44	58.54	0.4	0.25
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730819 (LOC101730819) mRNA	XM_004915204.1	15.11	41.42	1.5	0.008	253.37	186.49	-0.4	0
Xenopus (Silurana) tropicalis POU class 4 homeobox 1 (pot4f1.2) mRNA	NM_001097307.1	5.53	15.18	1.5	0.008	10.1	7.2	-0.5	0
Xenopus (Silurana) cDNA clone IMAGE:5770937 partial cds	BC168523.1	2.6	7.57	1.5	0.008	253.84	247.76	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis leucine-rich repeats and calponin homology (CH) domain containing 3 (lrich3) mRNA	XM_002937352.2	1.55	4.29	1.5	0.008	0	0	0	0
Xenopus laevis mRNA for frizzled 4 protein (fz4 gene)	AJ251750.1	9.35	26.46	1.5	0.008	14.14	10.8	-0.4	0
Xenopus laevis paired box transcription factor (Pax9) mRNA complete cds	JQ929180.1	0.79	2.21	1.5	0.014	3.73	3.71	0	0
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 9 subfamily A (NHES cation proton antiporter 5) member 5 (slc9a5) mRNA	XM_002931691.2	0.87	2.44	1.5	0.002	1.41	1.09	-0.4	0
Xenopus tropicalis monocyte to macrophage differentiation-associated mRNA (CDNA clone IMAGE:7650877) partial cds	BC136008.1	1.78	4.92	1.5	0.025	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis metalloprotease Tlk1-like (LOC100491951) mRNA	XM_002936336.2	1.67	4.65	1.5	0.037	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis metalloprotease Tlk1-like (LOC100493700) mRNA	XM_002932222.2	2.26	6.06	1.4	0.008	4.08	2.12	-0.9	0
Xenopus laevis Dickkopf-1 (Xdkk-1) mRNA complete cds	AF030434.1	4.27	11.07	1.4	0.008	8.9	6.35	-0.5	0
PREDICTED: Xenopus (Silurana) tropicalis frizzled family receptor 4 (fz4d) mRNA	XM_002936543.2	24.58	63.12	1.4	0.008	36.06	26.56	-0.4	0
activin beta B subunit (Xenopus laevis mRNA:2716 nt)	SG1773.1	1.11	2.89	1.4	0.008	1.86	1.35	-0.5	0
Xenopus (Silurana) tropicalis SRY (sex determining region Y)-box 2 (sox2) mRNA	NM_213704.3	38.14	104.02	1.4	0.008	49.47	53.9	0.1	0.07
Xenopus laevis 7-transmembrane receptor frizzled-1 mRNA complete cds	AF231711.1	1.42	3.79	1.4	0.008	1.39	1.48	0.1	0.07
Xenopus laevis complement factor 1 (cf1a) mRNA	NM_001085952.1	2.77	7.25	1.4	0.008	1.33	1.5	0.2	0.14
Xenopus laevis Sial-interacting protein (sip) mRNA	NM_001086745.1	2.76	7.27	1.4	0.008	73.89	84.03	0.2	0.14
Xenopus (Silurana) tropicalis NMN/NMN23 family member 1 (nmne7) mRNA	NM_203572.1	16.68	43	1.4	0.008	14.29	18.28	0.4	0.29
Xenopus laevis F1.3 (LOC779037) mRNA	NM_001096707.1	8.54	22.31	1.4	0.008	103.56	142.84	0.5	0.36
Xenopus (Silurana) LIM homeobox 2 (fsl2) mRNA	NM_001166041.1	3.85	10.1	1.4	0.008	5.65	8.34	0.6	0.43
PREDICTED: Xenopus (Silurana) tropicalis kinase insert domain receptor (a type III receptor tyrosine kinase) (krk) mRNA	NM_0012934669.2	0.6	1.59	1.4	0.014	0	0	0	0
TPA-inf. Xenopus laevis RTNL1-C (RTNL1) mRNA complete cds	BK004907.1	0.39	1.02	1.4	0.004	269.94	187.34	-0.5	0
Xenopus tropicalis finished cDNA clone Theul12901	CU075/70.1	13.31	32.71	1.3	0.008	30.12	27.72	-0.1	0
Xenopus tropicalis finished cDNA clone Tga5072h19	CT025/00.2	2.05	5.11	1.3	0.008	0	0	0	0
Xenopus (Silurana) tropicalis rat guanine nucleotide dissociation stimulator (rlgds) mRNA	NM_001113823.1	7.44	18.68	1.3	0.008	10.54	9.41	-0.2	0
PREDICTED: Xenopus (Silurana) ATPase H+-transporting lysosomal 42kDa VI subunit C2 (atp6v1c2) mRNA	AB45086.1	2.73	6.85	1.3	0.008	66.35	55.48	-0.3	0
PREDICTED: Xenopus (Silurana) tropicalis uveal autoantigen with coiled-coil domains and ankyrin repeats-like (LOC101731603) mRNA	NM_001091975.1	2.61	6.26	1.3	0.008	5.29	6.08	0.2	0.15
Xenopus laevis regulator of cell cycle (gcc) mRNA	NM_001093976.1	23.67	59.42	1.3	0.008	32.69	37.69	0.2	0.15
Xenopus laevis MGC81121 protein (MGC81121) mRNA	NM_001091866.1	2.65	6.48	1.3	0.008	20.55	25.64	0.3	0.23
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 198 member A (fam198a) mRNA	NM_002937853.2	2.4	5.84	1.3	0.008	2.49	3.11	0.3	0.23
Xenopus laevis Rplpy3 mRNA for Rplpy3 protein complete cds	AB45086.1	13.76	34.42	1.3	0.008	6.33	8.94	0.5	0.38
Xenopus laevis mannose/(alpha-1-3)-glycoprotein beta-1,4-N-acetylglucosaminyltransferase isozyme B (mgat4b) mRNA	NM_001172213.1	1.16	2.8	1.3	0.014	7.71	8.11	0.1	0.08
PREDICTED: Xenopus (Silurana) tropicalis CTNBP2 N-terminal like (ctnnb2nl) transcript variant X2 mRNA	NM_001091294.1	5.74	14.28	1.3	0.002	210.56	286.82	0.4	0.31
Xenopus laevis homeobox C8 (hoxc8) mRNA	NM_001127798.1	1.64	4.09	1.3	0.004	0	0	0	0
Xenopus laevis family with sequence similarity 101 member B (fam101b) mRNA	NM_001093870.1	8.37	19.28	1.2	0.008	9.24	9.48	0	0
Xenopus laevis tripartite motif containing 35 (trm35) mRNA	NM_001091117.1	5	11.38	1.2	0.008	5.83	5.6	-0.1	0
Xenopus laevis SRY sex determining region Y-box 21 (sox21) mRNA	NM_001172213.1	7.98	17.96	1.2	0.008	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis CTNBP2 N-terminal like (ctnnb2nl) transcript variant X2 mRNA	NM_00491072.1	1.5	3.37	1.2	0.008	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis Ig5Fc-binding protein-like (LOC100492852) mRNA	NM_002940533.2	3.85	8.74	1.2	0.008	502.53	351.49	-0.5	0
Xenopus laevis adenomatous polyposis coli down-regulated 1 (apcd1) mRNA	BC099044.1	1.57	3.51	1.2	0.008	1.93	1.37	-0.5	0
Xenopus laevis adenosine triphosphate-binding cassette subfamily A member 1 (apca1) mRNA	NM_001094109.1	5.07	11.54	1.2	0.008	6.58	5.29	-0.3	0
Xenopus laevis ats-2a proto-oncogene mRNA (cDNA clone MGCI60372 IMAGE:85244815) complete cds	BC133183.1	6.96	16.18	1.2	0.008	11.41	8.59	-0.4	0

Gene	Accession	Six1-Eya1 CHX	Six1-Eya1 CHX+DEX <sup>2</sup>	FC Six1- Eya1 <sup>1</sup>	q-value	Control CHX	Control CHX+DEX	FC Control CHX+DEX	Rank <sup>7</sup>
Xenopus laevis uncharacterized LOC503674 (LOC503674) mRNA	NM_001095563.1	12.11	27.13	1.2	0.008	15.19	11.67	-0.4	0
Xenopus (Silurana) tropicalis ras homolog family member V (rhvov) mRNA	NM_001128659.1	14.06	32.57	1.2	0.008	21.1	19.35	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis protein fos-B-like (LOC100490871) transcript variant X2 mRNA	XM_004916957.1	3.42	7.9	1.2	0.008	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis cAMP responsive element modulator (crem) transcript variant X2 mRNA	XM_004915472.1	4.73	10.64	1.2	0.008	0	0	0	0
Xenopus laevis E-box protein 46 (fbxo46) mRNA	NM_00109814.1	7.85	18.34	1.2	0.008	15.58	21.08	0.4	0.33
Xenopus laevis cDNA clone IMAGE8332229	BC155363.1	6.34	14.87	1.2	0.008	8.37	11.35	0.4	0.33
Xenopus laevis nestin (nest) mRNA	NM_001087857.1	3.88	8.71	1.2	0.02	438.87	324.97	-0.4	0
PREDICTED: Xenopus (Silurana) tropicalis vasohibin 2 (vash2) mRNA	XM_002934687.2	1.02	2.28	1.2	0.002	2.74	3.08	0.2	0.17
PREDICTED: Xenopus (Silurana) transcript variant X2 mRNA	XM_004911637.1	0.67	1.42	1.1	0.008	0	0	0	0
Xenopus laevis LFNc O-fucosylpeptide 3-beta-N-acetylglucosaminyltransferase (lfng) mRNA	NM_001097707.1	3.91	8.11	1.1	0.008	5.68	5.16	-0.1	0
Xenopus laevis cdk2Ba mRNA for cdk2B phosphatase complete cds	AB363840.1	8.11	16.81	1.1	0.008	7.44	6.95	-0.1	0
PREDICTED: Xenopus (Silurana) transcript domain containing family G (with RhoGef domain) member 1 (plekhh1) mRNA	NM_002934454.2	5.76	12.21	1.1	0.008	8.05	7.02	-0.2	0
Xenopus laevis transcr. factor Ap-2 beta (activating enhancer binding protein 2 beta) (tfap2b) mRNA	NM_001094232.1	21.33	44.25	1.1	0.008	50.79	37.03	-0.5	0
Xenopus laevis NSOX-2 (Sox-2) mRNA complete cds	AF005476.1	149a.4	3115.21	1.1	0.008	812.21	855.19	0.1	0.09
PREDICTED: Xenopus (Silurana) tropicalis progestin and adiponQ receptor family member IX (paqr9) mRNA	NM_004914351.1	1.89	4.03	1.1	0.008	1.47	1.83	0.3	0.27
Xenopus laevis RNA binding motif protein 24 (rbm24-a) mRNA	NM_001087526.1	2.4	5.31	1.1	0.008	2.61	3.11	0.3	0.27
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 46 member A (fam46a) mRNA	XM_002938458.2	4.46	9.48	1.1	0.008	4.17	5.84	0.5	0.45
PREDICTED: Xenopus (Silurana) protein-coupled receptor 56 (grp56) mRNA	XM_002931653.2	0.63	1.37	1.1	0.014	1.79	1.8	0	0
Xenopus laevis cDNA clone MGCI79935 IMAGE686551 complete cds	BC076717.1	4.09	8.59	1.1	0.014	246.09	269.65	0.1	0.09
PREDICTED: Xenopus (Silurana) tropicalis tripartite motif-containing protein 7-like (LOC100490838) mRNA	NM_004922015.1	1.87	4.05	1.1	0.02	0	0	0	0
Xenopus laevis cDNA clone IMAGE5083355 partial cds	BC073731.1	4.23	8.9	1.1	0.02	3.75	4.44	0.2	0.18
Xenopus (Silurana) tropicalis Drai (Hpd40) Homolog subfamily C member 18 (dhajc18) mRNA	NM_001102878.1	6.59	13.2	1	0.008	6.24	6.13	0	0
Xenopus laevis X1-c-ets-2b protein mRNA (CDNA clone MGCI80006 IMAGE6631306) complete cds	BC077264.1	15.97	31.91	1	0.008	21.13	18.01	-0.2	0
Xenopus laevis forkhead box 14 gene 1 (foxi4.1) mRNA	NM_001097651.1	4.84	9.48	1	0.008	31.16	31.31	0	0
Xenopus laevis E-box protein 6 (fbxo6) mRNA	NM_001095085.1	7.77	15.27	1	0.008	6.66	5.84	-0.2	0
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 850-like (LOC101733241) mRNA	NM_002919000.1	100.67	202.63	1	0.008	84.94	70.39	-0.3	0
PREDICTED: Xenopus (Silurana) tropicalis neural precursor cell expressed developmentally down-regulated 9 (nedd9) mRNA	XM_002932666.2	4.5	9.2	1	0.008	17.39	14.67	-0.2	0
Xenopus laevis neural cortex related-3 (Enrc-3) mRNA complete cds	AY216793.1	41.19	80.94	1	0.008	55.05	45.63	-0.3	0
X.laevis XFD2 mRNA for fork head protein	X74315.1	9.45	18.54	1	0.008	14.77	13.67	-0.1	0
PREDICTED: Xenopus (Silurana) tropicalis sodium/hydrogen exchanger 2-like (LOC101732511) mRNA	NM_004911791.1	4.64	9.21	1	0.008	6.55	5.69	-0.2	0
Xenopus laevis tetraspanin 1 (tspn1) mRNA	NM_001095473.1	16.27	33.47	1	0.008	20.7	23.96	0.2	0.2
Xenopus laevis peroxiredoxin 5 (prdx5) nuclear gene encoding mitochondrial protein mRNA	NM_001092111.1	12.11	24.61	1	0.008	15.94	18.58	0.2	0.2
Xenopus laevis malT-cell differentiation protein (mal) mRNA	NM_001086577.1	6.25	12.83	1	0.014	10.99	11.74	0.1	0.1
Xenopus laevis dual specificity phosphatase 18 (dusp18) nuclear gene encoding mitochondrial protein mRNA	NM_001096276.1	3.35	6.74	1	0.025	4.5	5.11	0.2	0.2
Xenopus laevis cDNA clone 2164-1	AJ009295.1	2.98	6.14	1	0.029	0	0	0	0

<sup>1</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1 or Eya1 or Six1+Eya1 and CHX treatment

<sup>2</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1 or Eya1 or Six1+Eya1 and CHX+DEX treatment

<sup>3</sup>Log<sub>2</sub> Fold change values for Six1 or Eya1 or Six1+Eya1

<sup>4</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup>Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup>Log<sub>2</sub> Fold change values for un-injected control  
<sup>7</sup>Log<sub>2</sub> FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Six1 or Eya1 or Six1+Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## F.6 Six1+Eya1<sub>m</sub>: Down-regulated genes

**Table F.6** Significantly differentially expressed genes with at least two-fold down-regulation after injection of Six1-GR or Eya1-GR or Six1-GR+Eya1-GR and treatment with CHX+DEX

Gene	Accession	Six1-Eya1 CHX <sup>1</sup>	Six1-Eya1 CHX+DEX <sup>2</sup>	FC Six1- Eya1 <sup>3</sup>	q-value CHX <sup>4</sup>	Control CHX <sup>5</sup>	Control CHX+DEX <sup>6</sup>	FC Control	Rank <sup>7</sup>
Xenopus laevis Similar to myosin light polypeptide 9 regulatory clone IMAGE:5570659 mRNA	BC042231.1	9.67	0.02	-8.9	0.029	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101735120 (LOC101735120) mRNA	XM_004919916.1	8.77	0.02	-8.8	0.014	0	0	0	0
XELRG-AOP3 xlaevis oocyte 5s rRNA clone pXl031	K01368.1	189.71	0.88	-7.8	0.008	1.56	1.34	-0.2	0.03
Medicago truncatula Glucan 1,3-beta-glucosidase (MTR_Bg091310) mRNA complete cds	XM_003630009.1	3.92	0.26	-3.9	0.008	0	0	0	0
PREDICTED: Xenopus (Silurana) tropicalis vomer nasal type-2 receptor 2s-like (LOC100487329) partial mRNA	XM_002945234.2	356.12	23.72	-3.9	0.008	33.81	27.61	-0.3	0.08
XELB1AA X borealis B1 protein mRNA complete cds	N63663.1	322.95	52.01	-2.6	0.008	4051.45	3420.13	-0.2	0.08
Xenopus laevis FEZ family zinc finger 2 (fezf2) mRNA	NM_001096464.1	2.62	0.72	-1.9	0.014	3.56	3.74	0.1	0
Xenopus laevis Similar to RIKEN cDNA 2610301D06 gene clone IMAGE:5572543 mRNA	BC042289.1	7.62	2.44	-1.6	0.008	56.31	70.87	0.3	0
Xenopus laevis vesicle-associated membrane protein 1 (synaptobrevin 1) (vamp1) mRNA	NM_001094764.1	1.6	0.58	-1.5	0.008	49.86	61.84	0.3	0
Xenopus laevis Dmrt4 (Dmrt4) mRNA complete cds	AY648303.1	4.84	1.79	-1.4	0.029	7.38	7.14	0	0
Xenopus laevis repetitive 173 element fragment C17	XN00077.1	133.6	68.58	-1.2	0.008	97.58	125.81	0.4	0
Xenopus laevis uncharacterized LOC_00462907 (LOC100462907) mRNA	NM_001190395.1	10.59	5.27	-1	0.008	8.92	8.52	-0.1	0.1
PREDICTED: Xenopus (Silurana) tropicalis dynen axonemal heavy chain 6 (dnah6) mRNA	XM_002935453.2	10.04	4.95	-1	0.044	7.12	6.61	-0.1	0.1
Xenopus tropicalis cDNA clone IMAGE:6991249	BC158454.1	1245.89	624.22	-1	0.044	521972.63	477202.01	-0.1	0.1

<sup>1</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1 or Eya1 or Six1+Eya1 and CHX treatment

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1 or Eya1 or Six1+Eya1 and CHX+DEX treatment

<sup>3</sup> Log<sub>2</sub> Fold change values for Six1 or Eya1 or Six1+Eya1

<sup>4</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX treatment

<sup>5</sup> Expression levels (FPKM) in un-injected pre-placodal explants after CHX+DEX treatment

<sup>6</sup> Log<sub>2</sub> Fold change values for un-injected control

<sup>7</sup> Rank = FC (log<sub>2</sub>) Control/FC (log<sub>2</sub>) Six1 or Eya1 or Six1+Eya1. Instances where a gene is either not expressed in the un-injected control or has a negative rank (i.e. the opposite direction of change compared to the experimental condition) are equally ranked as 0.

## Appendix G. Hes phylogeny

## G.1 Hes alignment

**Table G.1** Full coding regions for 61 *Hes* genes

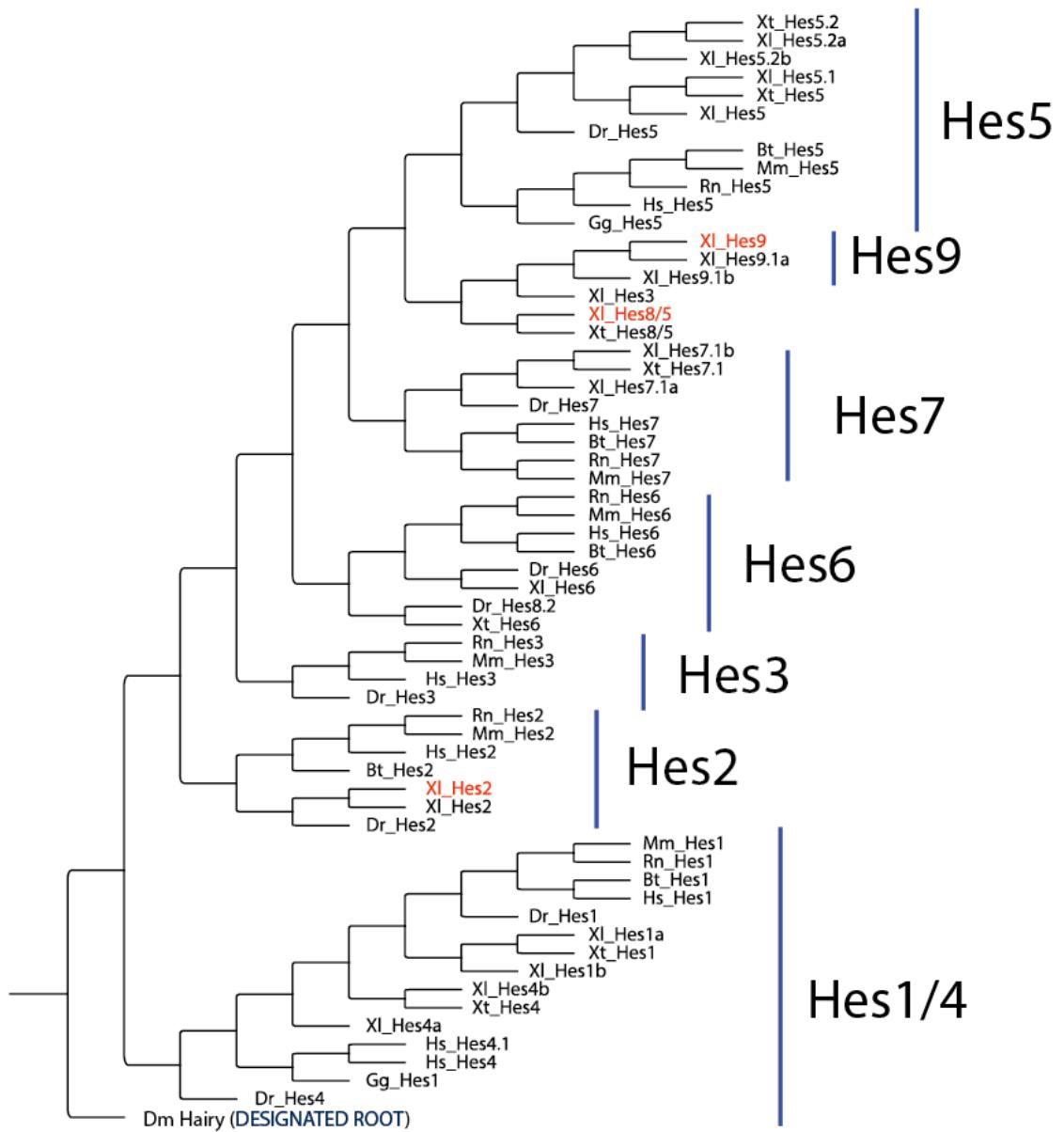
Gene	Accession	Species	Sequence
Gg_Hes5	NP_001012713	<i>Gallus gallus</i>	MAPSALSILETPKEKNRLRKPIVEKLRDRINSSIEQLKLLKEFQRHQPNSKLEKADILEMTVSYLKYSRAFAASAKSLQOODYCEGYAWCLKEALQFLSANTETQMKLICHFQRSQAMPKD
Hs_Hes1	Q14469	<i>Homo Sapiens</i>	SGSPSASTSTHOAPSAKOTPVKPCSNLWRPW
Hs_Hes2	Q9Y543	<i>Homo Sapiens</i>	MPADIMEKNSSPVAAATPASVNTTPDKPTASEHRKSSKPMIEKRRARINESISQLKLTDALKDSSRRHSKLEKADEILEMTVKHLRNLQRAQMTAALSTDPSVIGKYRAGFSCECMNEVTRFLS
Hs_Hes3	Q5TGS1	<i>Homo Sapiens</i>	TCEGVNTEVTRLLGHIANCMTQINAMYTPGQPHPAQAPPPLVPIPPGAAPPGGGPQHQAPAPPVLPREGYSACVALARVLPCACRVLEPAVSARLLEHLWRRAAA
Hs_Hes4	Q9HCC6	<i>Homo Sapiens</i>	TSNSGTSGVNAVSPSSGSPSLTADSMMWRPWNS
Hs_Hes4.1	NP_001135939	<i>Homo Sapiens</i>	MGLPRRAGDAELRKSLLEKRRARINQSLQSLKGKLLPLGRENSNCSKLEKADEILEMTVRFQELPASSWPTAAPLPCDSYREGYSACVALARVLPCACRVLEPAVSARLLEHLWRRAAA
Hs_Hes5	Q5TA89	<i>Homo Sapiens</i>	TLDGGRAGDSSGSPAPAPASAPEAASPVPSPSPCGPGLWRPW
Hs_Hes6	Q96HZ4	<i>Homo Sapiens</i>	MEKKRARINIVSLEQQLSLLEKHSHQIRKRLKLEKADEILSVKYMRSLQSLQGLWPVPRGAEQPSGFRSCLCPGYSQLRRGDEVSGLRCPLVPESAAGSTMDSAGLQEAPALFRPCTPAV
Hs_Hes7	Q9BYE0	<i>Homo Sapiens</i>	WAAPAAGGPRSPRPPLLIPESLPGSSAVPPQPAASSRCAESPGLGIRWWRPWSPGDNN
Mm_Hes1	P35428	<i>Mus musculus</i>	MAADTPGKPSASPMAGAPASAASRTDPKPSAAEHRKSRGPRVGRATGGREGRGTQPVDPQSSKPVMERARRARINESLAQKTLTILDALKKESRHSKLEKADEILEMTVRHLRSLLRRQVQTA
Mm_Hes2	Q54792	<i>Mus musculus</i>	ALSDAPAVLGKYRAGFHECLAEVNRFLAGCEGVADWRSRLLGHLACLRQLGPSRSPRASLSPAAEAAPAEVYAGRPLLPSLQGPFPPLAPPLPGLTTRALPAAPRAGPQGPQGPWRPWLR
Mm_Hes3	Q61657	<i>Mus musculus</i>	MAPSTVAELSPKEKNRLRKPVVEKMRDRINSSIEQLKLLQEFAHQPNSKLEKADEILEMAVSYLHKSKAFVAAGPKSLHQDYSEGWSCLQEAVQFLTLHAASDTQMKLLYHFQRPPA
Mm_Hes6	Q9JHE6	<i>Mus musculus</i>	APAAPAKEPKAPGAAPPALSAKATAAAAAAHOPACGLWRPW
Mm_Hes7	Q8BK72	<i>Mus musculus</i>	MAPPAAPGDRVGRGREDEGWTRGDRKARPKLVKEKRRARINESIQLRLLLAGAEVQAKLENALIELTYVRVQGVLRGRAREQLOAEASERAFAAGYIQCIMHEVHTFVSTCQAIATVAA
Rn_Hes1	Q04666	<i>Rattus norvegicus</i>	ELLNLLESMPUREGSSFDQLLGDALAGPRAPIGRSGWPGAGPSIPSPGQGDLCSDLLEAPEAKSQAEGPDLYPAALGSLTTAQIARSVWRPW
Rn_Hes2	P35429	<i>Rattus norvegicus</i>	MNTRDRAENRDFGPKMLKPLVVEKRRDRINRSLLEFLRLLERTRDQNRLRNPKLEKADEILEAVYLRERSRVEPPGVPRSPVQDAEALASCYLSGFRECLLRLAAFAHDASPAARAQLFSALHGYL
Rn_Hes3	Q04667	<i>Rattus norvegicus</i>	RPKPPRKPVDPRPPAPRSPPLDAPALPQMLHQRPPSPRCAWSPSLCSFRAGDSGAPAPLTGLLPPPQQPHQDGAPAKLPPPAFWRPW
Rn_Hes5	Q03062	<i>Rattus norvegicus</i>	MPADIMEKNSSPVAAATPASVNTTPDKPTASEHRKSSKPMIEKRRARINESISQLKLTDALKDSSRRHSKLEKADEILEMTVKHLRNLQRAQMTAALSTDPSVIGKYRAGFSCECMNEVTRFLS
Rn_Hes6	NP_001013197	<i>Rattus norvegicus</i>	TCEGVNTEVTRLLGHIANCMTQINAMYTPGQAHPALQAPPSSGSPGQHAPAPPVLPVPGGAAPPGSACPKLGSQAGEAAKVFGGFQVWPAPDQFAHSGPVPV
Rn_Hes2	Q04667	<i>Rattus norvegicus</i>	YTNSNGTSVGPNAVSPSSGSSLTDSMMWRPWNS
Rn_Hes3	Q03062	<i>Rattus norvegicus</i>	MRLPRVVEDAAELRKNLKPLLEKRRARINESIQLKGIVLVLPLGAETSRSSKLEKADEILEMTVRFQEQPATLYSSAAPGPNLSDYLEGYRACLARLVPACSYLEPAVSARLLEHLRQRTVSDDS
Rn_Hes5	Q04666	<i>Rattus norvegicus</i>	PSLTPPAPAPSPVPPGSSGLWRPW
Rn_Hes6	Q04667	<i>Rattus norvegicus</i>	MEKKRARINIVSLEQRLSLLERHYSHQIRKRLKLEKADEILESVKYMRSLQNSLQGLWPVPSGVYDPSGFQGGLRGVSQRLRPGEDSGLRCPLLLLQRREGSTDSANPQATSVLNPCLPAINAPS
Rn_Hes7	Q04666	<i>Rattus norvegicus</i>	RAAGGSHSPQSPPLPQGLLESSTDWAPHPSNCQAESTRPGFRVWRPW
Rn_Hes2	P35429	<i>Rattus norvegicus</i>	MAPSTVAEVMSLSPKEKNRLRKPVVEKMRDRINSSIEQLKLLQEFAHRHOPNSKLEKADEILEMAVSYLHKSKAFAAAGPKSLHQDYSEGWSCLQEAVQFLTLHAASDTQMKLLYHFQRPPA
Rn_Hes3	Q04667	<i>Rattus norvegicus</i>	PAAPAAKEPPAPGAAPPARSSAKAAAAAVTSRQAPACGLWRPW
Rn_Hes5	Q04666	<i>Rattus norvegicus</i>	MAPSOQASPDSDRAGQEDDRWEARGDRKARPKLVKEKRRARINESIQLRLLLAGTEVQAKLENALIELTYVRVQGALRGRAREQLOAEASERAFAAGYIQCIMHEVHTFVSTCQAIATVAA
Rn_Hes6	Q04667	<i>Rattus norvegicus</i>	LNHHLESMPUREGSSFDQLLGDASLPGGSGRSSWPGGSPESLSSPPGPGDCLCSDSLLEIPEAEELNRV AEGDLVSTSLGSLTAAARRAQSIVWRPW
Rn_Hes7	Q04666	<i>Rattus norvegicus</i>	MVTTERAENRDFGPKMLKPLVVEKRRDRINRSLLEFLRLLERTRDQNRLRNPKLEKADEILEAVYLRERSRVEPPGVPRSPGQDAEALAKSPLPPYQDGAQKPLPPPPPYRQDGAQKPLPPAFWRPW
Rn_Hes1	Q04666	<i>Rattus norvegicus</i>	RPKPPRPEAVDGPLPAPRPPPLDTPDKPTASEHRKSSKPMIEKRRARINESISQLKLTDALKDSSRRHSKLEKADEILEMTVKHLRNLQRAQMTAALSTDPSVIGKYRAGFSCECMNEVTRFLS
Rn_Hes2	P35429	<i>Rattus norvegicus</i>	TCEGVNTEVTRLLGHIANCMTQINAMYTPGQAHPALQAPPSSGSPGQHAPAPPVLPVPGGAAPPGSACPKLGSQAGEAAKVFGGFQVWPAPDQFAHSGPVPV
Rn_Hes3	Q04667	<i>Rattus norvegicus</i>	MRLPRGYGDAELRKSLKPLLEKRRARINESIQLKGIVLVLPLGAETSRSYNSKLEKADEILEMTVRFLEQEPASVCSTEAPGSDLSYLEGYRACLARLVPACSYLEPAVSARLLEHLRQRTVSDDP
Rn_Hes5	Q04666	<i>Rattus norvegicus</i>	PSLTPASAPASPPVPPSSGLWRPW
Rn_Hes6	NP_001013197	<i>Rattus norvegicus</i>	MEKKRARINIVSLEQRLSLLERHYSHQIRKRLKLEKADEILESVKYMRSLQNSLQGLWLPSGVYDPSGFQGGLRGVSQRLRPGEDSGLRCPLLLLQRREGSTDSANPQATSVLNPCLPAINAPS
Rn_Hes7	Q04666	<i>Rattus norvegicus</i>	PAGGSQSQSPQSPFPPLGGLLLESSTGILAPPASNCQAENPRPGRFVWRPW
Rn_Hes2	Q04667	<i>Rattus norvegicus</i>	MAPSTVAEVMSLSPKEKNRLRKPVVEKMRDRINSSIEQLKLLQEFAHRHOPNSKLEKADEILEMAVSYLHKSKAFAAAGPKSLHQDYSEGWSCLQEAVQFLTLHAASDTQMKLLYHFQRPPA
Rn_Hes3	Q04667	<i>Rattus norvegicus</i>	PAAPVKEPTPGAAQPARSTKAAASVSTSRSACGIWRPW
Rn_Hes5	Q04666	<i>Rattus norvegicus</i>	MRTVGKHGGTARCQGLRVDGPAGGTGQGLSRFQARKLVEKRRARINESLQEIRLLLAGTEVQAKLENAEVLETVRYOGALGRAREQLOAEASERFAAGYIQCIMHEVHTFVST
Rn_Hes6	NP_001013197	<i>Rattus norvegicus</i>	CQAIATVSAELNHLESMPLREGSSFDLLGDSLAGLPGGSSPESLSSPPGPWDILCSDEEIPAELNRPVAPGPDLVPTSLGILTARRAQSWRPW



<b>Gene</b>	<b>Accession</b>	<b>Species</b>	<b>Sequence</b>
Xt_Hes4	Q6PBD4	<i>Xenopus tropicalis</i>	MAPDSMEKPTASPIAGAPASSAQTPDKPKSAEHRKSXPIMERRRARINESLQLQKTLILDALKKDSRRHSKLEKADILEMTVKHLRLNLRQVQMTAALTADPSVLGKYRAGFNECMNEVTREL STCEGVNTVERTRLLGLHSCLQLQIVAMNYQQPPSSQPLHVQLPSSTPAVPMPCKVNPAAISPCKVPAETSPGPVILPNPAYTSPGGFQLPVATDQFAFLIPNPAVTFSPGGFQSPVQGLIT TFGHKMPHMAQAVSPLGGSTGADSAYW/RPW
Xt_Hes5	NP_001037880	<i>Xenopus tropicalis</i>	MAPTSISVDTFSCKERNKLNKLKPIVEKMRRDRINNSIEQLKALLEKEFHKKQEPNPKLEKADILEMAVSYLOQQKSOSPNLAKLEQDYKQGFSSCLREAVQFLCYYPSEGETQMKLKHLOAQPKLSV APLTYPSVSDSKQAAIASPNPSKIVRPW
Xt_Hes5.2	NP_001037974.1	<i>Xenopus tropicalis</i>	MAPSTDPLDQKMTTPKENKLKPVVEKMRDRINSSIEQLKGILLETVFHKKQOPNVKLEKADILEMTVTYLHQQTOLIKSEPHNNNDIQMDYKDGYSRCFEEVIDFLSHQKQPETAKLISHFHSK ATASSISSFPRCQSOKTANGTGSSLW/RPW
Xt_Hes6	NP_001072210	<i>Xenopus tropicalis</i>	MSGAGRSQKLACSAKEERKLRLKPILERKRERINTCLEQLKETVKAFFHDQSKLEKADILEMTVRHLQNIQKSSTGEPSQGSVDAQQRFSTGYIQCMLHELSLLTCDWMDPALGARLLNHLL KSLPRPEGRATAFLQDYEGDTGRTMSPSJSDCEAEQTSPLHSDSAQGKTQCSLLRSQMLWRPW
Xt_Hes7.1	Q28HA8	<i>Xenopus tropicalis</i>	MKGASEYRPMEAHRKLLKPLVERRRERRINNSLEKLRIFLQALKSEKLNPKVKEAELLECTVQFLQOSKLVPDGDVGNGKYQSGFQHCLETALHFMNNSKPDINVATKDFLSHQLSSYKPPAE AWSPTDTPKPTPSIGYQDASPHLSSNTIVSPITKLVDGQFSPQITFQIWRPW
Xt_Hes8/5	XP_002933895	<i>Xenopus tropicalis</i>	MTSAHITQDCSMENPKSKTSLRQIRKPVKEWMRRDRINNSIKQLRMILLEKEFQRHQPNSKLEKADILEMTVNLYLKEHQLQMINAVAFAARKSPFQDYNQGYSRCLEETLQFLSHTEMQKPANLKLV QHFNRTVTPADNNLPGQAPSQPKQPSNTAAIWRPW
* XI_Hes2	BC084134.1	<i>Xenopus laevis</i>	MAPNVALADSMHNQYQPKPGKRNRQEASELRKTLPLMEKRRARINESLNQQLKTLPLIGDONSRYSKLEKADILEMTVRLRDIPPVQAQNQADRYKEGYRACVERLSAILGKSHVLTGEASNRL LEYLQRSPESLCCSSDCNHPPKPQRPRVLOVSPRTSQFGSPLNQOPSSHRPAPCPQQLNSIIWRPW

\* Indicates sequences derived from RNA-Seq data

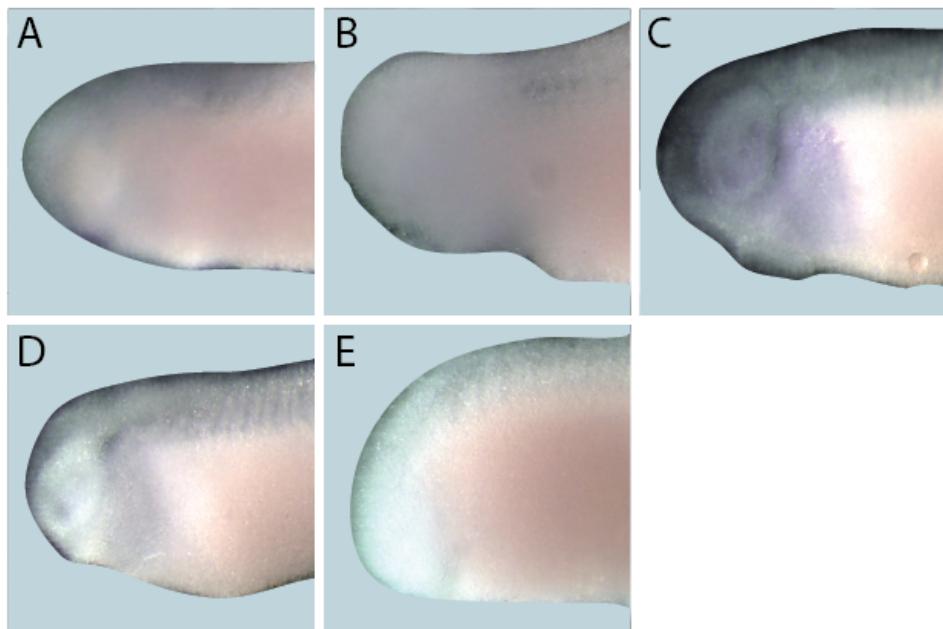
## G.2 Hes ML phylogeny



**Fig. G.2** Maximum likelihood tree of Vertebrate Hes genes based on a concatenated alignment of 83 amino acid residues encompassing the conserved bHLH and Orange domains (fig. 4.1). Hes genes derived from RNA-Seq data shown in red. Accessions are found in Appendix A7. Abbreviations; Dr *Danio rerio*, XI *Xenopus laevis*, Xt *Xenopus tropicalis*, Gg *Gallus gallus*, Bt *Bos taurus*, Hs *Homo sapiens*, Rn *Rattus norvegicus*, Mm *Mus musculus*, Dm *Drosophila melanogaster* (outgroup).

## Appendix H. *In-situ* sense controls

### H.1 *In-situ* sense controls for newly described genes



**Fig H.1** Sense controls for newly described genes at tail bud stages (stages 26 - 32). **(A)** *Crem*; **(B)** *FosB*; **(C)** *Znf214*; **(D)** *Tbx15*; **(E)** *Isl2*.

# Appendix I. Morpholino RNA-Seq data

## I.1 Six1-MO: Down-regulated genes

**Table I.1** Genes significantly down-regulated after injection of Six1-MO

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO FC <sup>3</sup>	q-value
Xenopus laevis tetraspanin 31 (tspan31b) mRNA	NM_001086181.1	4238.61	0.01	-22.02	0.002
Xenopus laevis mitogen-activated protein kinase kinase kinase 7 (map2k7) mRNA	NM_001087648.1	1098.13	0.01	-20.07	0.002
Xenopus laevis shg07 mRNA complete sequence	DD096903.1	379.32	0.01	-18.53	0.002
Xenopus laevis tubulin folding cofactor E-like (tbc1) mRNA	NM_001096263.1	28.39	0.01	-14.79	0.002
Xenopus (Silurana) tropicalis RAS protein activator-like 1 (GAP1 like) (rasal1) mRNA	NM_001080848.1	10.85	0.01	-13.4	0.002
PREDICTED: Xenopus (Silurana) tropicalis phosphodiesterase A2 inhibitor and Ly6/PLAUR domain-containing protein-like (LOC101730430) mRNA	XM_004919780.1	5.47	0.01	-12.42	0.002
Xenopus laevis caspase-7 mRNA (CDNA clone MGc82818 IMAGE5506240) complete cds	BC078049.1	5.38	0.01	-12.39	0.002
XLU62711 Xenopus laevis zefillin mRNA complete cds	U63711.1	4.68	0.01	-12.19	0.002
Xenopus laevis G-protein type G-alpha-i-3 partial cds	X56090.1	2.98	0.01	-11.54	0.002
Xenopus laevis mRNA for alpha-subunit of G-protein type G-alpha-i-3 partial cds	NM_001087145.1	1.91	0.01	-10.9	0.002
Xenopus laevis N-methyltransferase hmt1-b) mRNA	NM_001095993.1	1.77	0.01	-10.78	0.002
Xenopus laevis histamine N-methyltransferase hmt1-b) mRNA	XM_004917598.1	1.61	0.01	-10.65	0.002
PREDICTED: Xenopus (Silurana) transcript variant X1 mRNA	NM_001137612.1	2.56	0.01	-10.32	0.002
Xenopus (Silurana) tropicalis reelin (rein) mRNA	XM_004918188.1	1.29	0.01	-9.33	0.002
PREDICTED: Xenopus (Silurana) tropicalis otoferlin (otof) mRNA	BC136162.1	1.19	0.01	-9.22	0.002
Xenopus laevis patch domain and ankyrin repeats 1 (gapnk1) mRNA	NM_001097131.1	150.16	0.27	-9.12	0.002
Xenopus laevis holoxyochrome synthase (hcs-b) mRNA	NM_001093680.1	3838.02	15.59	-7.94	0.002
Xenopus laevis four and a half LIM domains 3 (fh3) mRNA	NM_001092256.1	5526.3	63.28	-6.45	0.002
Xenopus laevis mucin-2-like (LOC101734541) mRNA	XM_004911425.1	1.85	0.03	-6.12	0.002
PREDICTED: Xenopus (Silurana) tropicalis serine protease 48-like (LOC104939351) mRNA	XM_002939691.2	6.1	0.11	-5.89	0.002
Xenopus laevis GID complex subunit 8 homolog (S. cerevisiae) (gid8) mRNA	NM_001096765.1	388.53	9.75	-5.32	0.002
PREDICTED: Xenopus (Silurana) tropicalis mitochondrial fission factor (mff) transcript variant X1 mRNA	XM_002933362.2	114.93	3.04	-5.24	0.002
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100493999 transcript variant X2 mRNA	XM_004915901.1	8.42	0.39	-4.44	0.002
Xenopus laevis integrin beta-3 subunit mRNA (CDNA clone MGc80757 IMAGE5512265) complete cds	BC073343.1	119.91	5.94	-4.34	0.002
Xenopus tropicalis finished cDNA clone Thle05417	CR843402	35.63	2	-4.16	0.002
XLU21933 Xenopus laevis high mobility group protein-H (HMG-1) mRNA complete cds	U21933.1	22.96	1.34	-4.11	0.002
Xenopus laevis SERPINE1 mRNA binding protein 1 (serbp1) mRNA	NM_001086826.1	43.97	2.73	-4.01	0.002
Xenopus laevis hypothetical LOC496090 mRNA (CDNA clone IMAGE7205280) partial cds	BC087515.1	51.07	3.21	-3.99	0.002
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC100494222) mRNA	NM_002938567.2	3.47	0.23	-3.92	0.002
Xenopus (Silurana) tropicalis Mgcs89248 protein (MGc89248) mRNA	NM_001004956.1	1.04	0.07	-3.9	0.002
Xenopus laevis DNA clone IMAGE6948275 **** chimeric clone ***	BC095925.1	4.09	0.36	-3.53	0.002
Xenopus laevis uncharacterized protein MGc69077 (MGc69077) mRNA	NM_001090073.1	23.31	2.07	-3.5	0.002
XE1XL01 Xenopus laevis mRNA for pleiotrophic factor-beta 1 complete cds	D20259.1	1058.69	99.43	-3.41	0.002
Xenopus laevis ion peptidase 2 peroxisomal (lmp2) mRNA	NM_001096479.1	779.96	75.25	-3.37	0.002
Xenopus laevis cysteine and histidine-rich protein 1A mRNA (CDNA clone IMAGE5513813) partial cds	BC073380.1	347.25	36.26	-3.26	0.002
Xenopus laevis DNA clone MGc154541 IMAGE8320906 complete cds	BC124896.1	154.01	17.96	-3.1	0.002
Xenopus (Silurana) tropicalis sema domain immunoglobulin domain (lg) short basic domain secreted (semaphorin) 3B (sema3b) mRNA	NM_001127436.1	1.03	0.13	-3	0.002
Xenopus laevis PKS06-binding protein mRNA complete cds	AF232672.1	7.09	0.91	-2.97	0.002
Xenopus laevis characterized protein MGc81434 (MGc81434) mRNA	NM_001091189.1	539.69	69.59	-2.96	0.002
PREDICTED: Xenopus (Silurana) tropicalis homeobox protein Dixa-like (LOC100494171) mRNA	NM_002935712.2	15.94	2.09	-2.94	0.002
Xenopus laevis characterized LOC101733743 (LOC101733743) mRNA	XM_004915875.1	6.27	0.83	-2.92	0.002
Xenopus laevis xRippl3 protein complete cds	AE455086.1	11.8	1.61	-2.88	0.002
Xenopus laevis DNA clone MGc115069 IMAGE3304671 complete cds	BC100222.1	94.87	13.05	-2.86	0.002
Xenopus laevis retinol dehydrogenase 7 gene 2 (rdh7) mRNA	NM_001094615.1	3.45	0.49	-2.83	0.002
Xenopus laevis DNA clone IMAGE8642610	BC153809.1	5.69	0.85	-2.74	0.002
Xenopus laevis DNA clone MGc114987 IMAGE5084306 complete cds	BC098991.1	35.12	5.33	-2.72	0.002
PREDICTED: Xenopus (Silurana) transcript variant 2 mRNA	NM_002938125.2	163.56	25.98	-2.65	0.002

Gene	Accession	Control <sup>1</sup>	Sik1 <sup>-/-</sup> MO <sup>2</sup>	Sik1 <sup>-/-</sup> MO <sup>2</sup> FC <sup>3</sup>	q-value
PREDICTED: Xenopus (Silurana) tropicalis toll-like receptor 2-like (LOC100485358) mRNA	XM_002943050.2	1.52	0.25	-2.65	0.002
Xenopus laevis cdc01 mRNA complete sequence	D0096916..1	554.4	89.1	-2.64	0.002
Xenopus laevis hypothetical protein LOC733412 mRNA (cDNA clone IMAGE8070819) partial cds	BC124835..1	38.91	6.45	-2.59	0.002
Xenopus laevis connelin gene 1 (cnf1..1-a) mRNA	NM_001011231..1	46.41	8.32	-2.48	0.002
PREDICTED: Xenopus (Silurana) tropicales uncharacterized LOC101730819 (LOC101730819) 91 mRNA	NM_004915204..1	47.93	8.69	-2.46	0.002
PREDICTED: Xenopus (Silurana) tropicalis NXPE family member 2-like (LOC100495388) mRNA	NM_002941335..2	199.84	36.22	-2.46	0.002
PREDICTED: Xenopus (Silurana) tropicales brain-specific angiogenesis inhibitor 1 (bai1) mRNA	XN_002934468..2	11.29	2.07	-2.45	0.002
Xenopus laevis (Silurana) tropicalis transcription factor epithelial-specific (elf3) mRNA	XN_002932989..2	9.48	1.74	-2.45	0.002
PREDICTED: Xenopus (Silurana) tropicalis E74-like factor 3 (ets domain transcription factor epithelial-specific) (elf3) mRNA	NM_001092839..1	30.49	5.59	-2.45	0.002
Xenopus laevis (Silurana) tropicalis 3 (ets domain transcription factor epithelial-specific) (elf3) mRNA	NM_004912208..1	5.58	1.03	-2.44	0.002
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 2-like (LOC101731874) transcript variant X3 mRNA	CB855646..2	4.9	0.92	-2.42	0.002
Xenopus tropicalis finished cDNA clone Tgas015e21	BC045104..1	56.89	10.81	-2.4	0.002
Xenopus tropicalis finished cDNA clone TTPA07n02	CB848229..2	2.54	0.49	-2.38	0.002
XLU2535 Xenopus laevis epithelial sodium channel alpha subunit (alphaENaC) mRNA complete cds	U23535..1	180.91	35.54	-2.35	0.002
Xenopus (Silurana) tropicalis chromosome 12 open reading frame (c12orf4) mRNA	NM_001007513..1	11.32	2.27	-2.32	0.002
PREDICTED: Xenopus (Silurana) tropicales synaptotagmin-13-like (LOC100490466) mRNA	XM_002944648..2	4.5	0.91	-2.31	0.002
Xenopus laevis (Silurana) tropicalis vasopressin V2 receptor-like (LOC100487051) transcript variant X2 mRNA	XN_004916721..1	1.65	0.34	-2.29	0.002
Xenopus laevis cDNA clone IMAGE958024 partial cds	BC074386..1	27.75	5.74	-2.28	0.002
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 6 (neurotransmitter transporter dopamine) member 3 (slc6a3)	XM_004915332..1	9.5	1.98	-2.27	0.002
Xenopus laevis ribosomal protein S6 kinase 90kDa polypeptide 6 (ros6ka6) mRNA	NM_001091806..1	9.86	2.05	-2.27	0.002
PREDICTED: Xenopus (Silurana) tropicalis mesothelin (msln) mRNA	NM_004918064..1	8.41	1.77	-2.25	0.002
Xenopus laevis uncharacterized LOC100127277 (LOC100127277) mRNA	NM_001112850..1	225.09	47.43	-2.25	0.002
PREDICTED: Xenopus (Silurana) tropicalis FAM26E-like (LOC101730841) mRNA	XN_004914563..1	2.56	0.57	-2.19	0.002
Xenopus laevis C80266 protein (MC80266) mRNA	NM_001092037..1	2.53	0.57	-2.17	0.002
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100487313 (LOC100487313) mRNA	XN_002939177..2	15.11	3.35	-2.17	0.002
Xenopus laevis ION peptidase N-terminal domain and ring finger 3 (lonrf3) mRNA	XN_002934542..2	5.25	1.2	-2.14	0.002
PREDICTED: Xenopus (Silurana) tropicales LON peptidase N-terminal domain (IMAGE8822377) partial cds	BC155885..1	4.11	1.17	-2.08	0.002
Xenopus laevis uncharacterized protein MGC83713 (MGC83713) mRNA	NM_001091448..1	163.64	38.9	-2.07	0.002
PREDICTED: Xenopus (Silurana) tropicalis AMP responsive element modulator (crem) transcript variant X2 mRNA	XM_004915472..1	4.67	1.11	-2.07	0.002
Xenopus laevis uncharacterized LOC100487159 (LOC100487159) transcript variant X3 mRNA	XN_004918159..1	97.77	23.4	-2.06	0.002
Xenopus laevis DNA clone IMAGE7207942	BC157742..1	12.87	3.12	-2.05	0.002
Xenopus laevis SH3 domain binding glutamatic acid-rich protein like 2 (sh3bgrl2-b) mRNA	NM_001096649..1	19.36	4.7	-2.04	0.002
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100489259 (LOC100489259) mRNA	XN_002935863..2	6.05	1.52	-2	0.002
Xenopus laevis glycosidase 1 (glc1..-a) mRNA	NM_001094108..1	102.07	25.47	-2	0.002
Xenopus laevis uncharacterized LOC100158303 (LOC100158303) mRNA	NM_001127759..1	18.37	4.63	-1.99	0.002
PREDICTED: Xenopus (Silurana) tropicalis alpha-1-N-acetylglucosaminyltransferase-like (LOC100493555) mRNA	XN_002938120..2	17.08	4.34	-1.98	0.002
Xenopus laevis DNA clone IMAGE3398708	BC108862..1	103.26	26.78	-1.95	0.002
Xenopus laevis glycerol-3-phosphate dehydrogenase 2 (mitochondrial) (gpd2) nuclear gene encoding mitochondrial protein mRNA	NM_001092540..1	2.02	0.53	-1.95	0.002
Xenopus laevis ferritin light chain oocyte isoform (MGC68606) mRNA	NM_001086458..1	123.51	32.94	-1.91	0.002
Xenopus laevis uncharacterized LOC100497516 (LOC100497516) mRNA	XN_002944045..1	12.27	3.27	-1.91	0.002
Xenopus laevis suppressor of cytokine signaling 3 (soc3) mRNA	NM_001094112..1	37.87	10.24	-1.89	0.002
Xenopus laevis DNA clone MGC85498 (IMAGE694048) complete cds	BC084859..1	457.82	124.35	-1.88	0.002
Xenopus (Silurana) tropicalis uncharacterized LOC100127867 (LOC100127867) mRNA	NM_001113158..1	2.32	0.64	-1.86	0.002
Xenopus laevis B glycoprotein (rbog-b) mRNA	NM_001093683..1	7.67	2.16	-1.83	0.002
Xenopus (Silurana) tropicalis mannose-6-phosphate isomerase mRNA	NM_001170492..1	30.72	8.76	-1.81	0.002
Xenopus laevis monoglyceride lipase (mgll) mRNA	NM_001094434..1	6.47	1.89	-1.78	0.002
PREDICTED: Xenopus (Silurana) tropicalis myosin VI (myo6) transcript variant X4 mRNA	XM_004914517..1	4.07	1.19	-1.77	0.002
Xenopus (Silurana) tropicalis ribosomal protein L23a (rpL23a) mRNA	NM_001005109..2	40.48	12.03	-1.75	0.002
Xenopus laevis testis derived transcript (3 LIM domains) (tes) mRNA	NM_001087237..1	10.9	3.29	-1.73	0.002
Xenopus laevis spid21 mRNA complete sequence	D00968991..1	2189.8	659.24	-1.73	0.002
Xenopus laevis bigiphe homeobox homolog-related mRNA (cDNA clone IMAGE7879321) partial cds	BC125785..1	44.92	13.53	-1.73	0.002
Xenopus laevis stomatin (EPB72)-like 3 (stom3) mRNA	NM_001096223..1	25.85	7.83	-1.72	0.002
Xenopus laevis potassium voltage-gated channel shaker related subfamily member 4 (Kcnq4) mRNA	NM_001088601..1	3.38	1.05	-1.7	0.002
Xenopus laevis calcium/calmodulin-dependent protein kinase II gamma 1 subunit mRNA complete cds	BC086276..1	54.68	16.82	-1.7	0.002
Xenopus laevis UDP-GlcNAc-beta-1,3-N-acetylgalactosaminyltransferase 5 (b3gnt5-b) mRNA	AF233630..1	12.67	3.91	-1.7	0.002
Xenopus laevis scindrin (scin) mRNA	NM_001092620..1	30.67	9.44	-1.69	0.002
Xenopus laevis uncharacterized protein MGC81349 (MGC81349) mRNA	NM_001091200..1	19.51	6.08	-1.68	0.002

Gene	Accession	Control <sup>1</sup>	Sik1-MO <sup>2</sup>	Sik1-MO FC <sup>3</sup>	q-value
Xenopus laevis putative transmembrane protein TA-2 mRNA complete cds	AF310008.1	479.18	149.5	-1.68	0.002
Xenopus (Silurana) tropicalis cold shock domain containing C2 RNA binding (csdc2) mRNA	NM_001100248.1	5.05	1.63	-1.64	0.002
PREDICTED: Xenopus (Silurana) tropicalis ADAMTS-like 1 (adams1t1) mRNA	XM_004910864.1	2.81	0.92	-1.62	0.002
Xenopus laevis mRNA for aldolase complete cds	AB002267.1	29.82	10	-1.58	0.002
Xenopus tropicalis lin-28 homolog mRNA (cDNA clone MGIC97527 IMAGE5382621) complete cds	BC09084.1	16.15	5.4	-1.58	0.002
Xenopus (Silurana) tropicalis superwillin (svil) mRNA	NM_001097296.1	7.59	2.55	-1.58	0.002
Xenopus (Silurana) tropicalis four and a half LIM domains 3 (fh3) mRNA	NM_001008164.1	20.37	6.86	-1.57	0.002
Xenopus tropicalis finished cDNA clone TEG0172a06	BG04196.1	1717.61	581.25	-1.56	0.002
Xenopus laevis ankyrin repeat domain 24 (membrane-inserted) (mrnp24) mRNA	CR85553.2	3.81	1.3	-1.55	0.002
Xenopus laevis characterized protein MGIC53587 (MGIC53587) mRNA	NM_001085951.1	19.22	6.56	-1.55	0.002
Xenopus laevis brain metalloproteinase 24-like (brp44l-b) mRNA	NM_001093636.1	13.61	4.67	-1.54	0.002
Xenopus laevis synaptotagmin-like 1 (sytl) mRNA	NM_001171779.1	163.09	56.29	-1.53	0.002
PREDICTED: Xenopus (Silurana) tropicalis clustered mitochondria protein homolog (LOC100496889) mRNA	NM_001093193.1	15.92	5.53	-1.53	0.002
Xenopus (Silurana) tropicalis enosin-like (LOC100497841) transcript variant X2 mRNA	XM_002932088.2	3.06	1.07	-1.53	0.002
PREDICTED: Xenopus (Silurana) tropicalis cysteine-rich protein 2-like (LOC101734738) mRNA	XM_004910879.1	9.42	3.31	-1.51	0.002
PREDICTED: Xenopus (Silurana) tropicalis mucin-16-like (LOC100498596) mRNA	XM_002941501.2	17.42	6.18	-1.5	0.002
Xenopus laevis brain protein 44-like (brp44l) mRNA	NM_001088329.1	28.96	10.45	-1.47	0.002
Xenopus laevis caveolin 2 (cav2) mRNA	NM_001088329.1	13.19	4.83	-1.45	0.002
Xenopus laevis semtex2 (XEMD2) mRNA complete cds	AT766060.1	104.49	38.33	-1.45	0.002
Xenopus laevis DNA clone IMAGE6880181	BC05326.1	15.16	5.57	-1.45	0.002
Xenopus laevis calcium/calmodulin-dependent protein kinase II gamma 1 mRNA complete cds	AF233632.1	12.05	4.43	-1.44	0.002
Xenopus tropicalis hypothetical protein LOC548657 mRNA (cDNA clone MGIC895997) complete cds	BC161440.1	19.79	7.33	-1.43	0.002
Xenopus laevis adiponectin receptor 2 adipor2) mRNA	NM_001094102.1	8.34	3.1	-1.43	0.002
Xenopus laevis secreted frizzled-related protein 2 (frzr2) mRNA	NM_0010887194.1	32.5	12.04	-1.43	0.002
Xenopus laevis guanine nucleotide binding protein (G protein) gamma 12 (gng12) mRNA	NM_001095586.1	9.65	3.6	-1.42	0.002
PREDICTED: Xenopus (Silurana) tropicalis serine-rich adhesin for platelets-like (LOC101733625) mRNA	AF231347.1	2942.75	1108.33	-1.41	0.002
Xenopus laevis exp44 mRNA complete sequence	DQ098661.1	16156.88	6135.79	-1.4	0.002
PREDICTED: Xenopus (Silurana) tropicalis protein tyrosine phosphatase receptor type U (ptpru) transcript variant X1 mRNA	XM_002939487.2	9.75	3.72	-1.39	0.002
Xenopus laevis byturate carboxylase gene 1 (oc1) mRNA	NM_001089757.1	62.53	23.88	-1.39	0.002
Xenopus laevis ornithine aminotransferase (oat) gene encoding mitochondrial protein mRNA	NM_001086593.1	14.38	5.55	-1.38	0.002
PREDICTED: Xenopus (Silurana) tropicalis serine-rich adhesin for platelets-like (LOC101733625) mRNA	XM_004912389.1	32.09	12.35	-1.38	0.002
Xenopus (Silurana) tropicalis branched chain amino-acid transaminase 2 mitochondrial (bcat2)	NM_001096739.	32.5	12.67	-1.36	0.002
Xenopus laevis solute carrier family 2 (facilitated glucose transporter) member 12 (slc2a12) mRNA	NM_001096426.1	39.61	15.4	-1.36	0.002
Xenopus laevis myeloid differentiation primary response 88 (myd88-b) mRNA	NM_001095728.1	23.45	9.27	-1.34	0.002
Xenopus laevis solute carrier family 44 member 4 (slc4a4) mRNA	NM_001092233.1	17.66	6.98	-1.34	0.002
Xenopus laevis characterized LOC100127738 (LOC100127738) mRNA	NM_001277737.1	75.24	29.67	-1.34	0.002
Xenopus laevis transmembrane protein 56 (tmem56-b) mRNA	NM_001086447.1	35.59	14.36	-1.31	0.002
Xenopus laevis cDNA clone IMAGE551358 partial cds	BC085045.1	64.49	25.99	-1.31	0.002
Xenopus laevis TSC22 domain family member 2 (tsc22d2) mRNA	NM_001091281.1	14.92	6.07	-1.3	0.002
Xenopus laevis hatching gland-like Xhel protein (LOC398485) mRNA	NM_001088898.1	414.37	169.66	-1.29	0.002
Xenopus (Silurana) tropicalis guanine nucleotide binding protein (G protein) gamma 12 (gng12) mRNA	NM_001102695.1	42.5	17.36	-1.29	0.002
Xenopus laevis characterized LOC100127738 (LOC100127738) mRNA	CR762359.2	5.91	2.44	-1.28	0.002
Xenopus laevis small nuclear RNA activating complex poly peptide 5 19kDa (snap5-b) mRNA	NM_001107180.1	59.76	24.61	-1.28	0.002
Xenopus laevis characterized LOC495461 (LOC495461) mRNA	NM_001128621.1	42.43	17.6	-1.27	0.002
Xenopus laevis mRNA for calcineurin catalytic subunit complete cds	AB037146.1	24.26	10.09	-1.27	0.002
Xenopus laevis hypothetical protein LOC100137163 mRNA	BC142545.1	226.57	95.3	-1.25	0.002
Xenopus (Silurana) tropicalis mannosyl (alpha-1,6)-glycoprotein beta-1-N-acetylglucosaminyltransferase (mgat5) mRNA	NM_001126984.1	12.6	5.29	-1.25	0.002
Xenopus laevis glutathione S-transferase omega 1 (gsto1) mRNA	NM_001095036.1	29.33	12.38	-1.24	0.002
Xenopus laevis ribosomal protein S7 (rps7) mRNA	NM_001091033.1	59.26	25.2	-1.23	0.002
Xenopus laevis sepine palmitoyltransferase small subunit A (spissaa) mRNA	NM_001094202.1	48.88	20.81	-1.23	0.002
Xenopus laevis tnf receptor type I (tnfrsf1a) mRNA	EU0177101.1	37.4	16.22	-1.21	0.002
Xenopus laevis GIPC PDZ domain containing family member 2 (gipc2) mRNA	NM_001088817.1	31.99	13.93	-1.2	0.002
Xenopus (Silurana) tropicalis WNK lysine deficient protein kinase 4 (wnk4) mRNA	XM_002932519.2	3.92	1.71	-1.2	0.002
Xenopus laevis comp42 mRNA complete sequence	DQ096878.1	29.39	12.83	-1.2	0.002
Xenopus laevis XEL mRNA	BC157491.1	40.88	17.8	-1.19	0.002
Xenopus laevis NIPA-like domain containing (nipa1) mRNA	AB105372.1	2762.21	1191.98	-1.18	0.002
PREDICTED: Xenopus (Silurana) tropicalis NIPA-like domain containing (nipa1) mRNA	XM_002933460.2	28.66	12.64	-1.18	0.002
Xenopus laevis sphingosine-1-phosphate phosphatase (sgpp2) mRNA	NM_001097668.1	14.56	6.44	-1.18	0.002

Gene	Accession	Control <sup>1</sup>	Sik1-MO <sup>2</sup>	Sik1-MO FC <sup>3</sup>	q-value
Xenopus laevis hypothetical protein LOC100127337 mRNA (cDNA clone IMAGE7980636) partial cds	BC155957.1	164.19	73.25	-1.16	0.002
Xenopus laevis desmocollin 3 (dscl3) mRNA	NM_001095736.1	20.01	8.96	-1.16	0.002
Xenopus laevis aldehyde dehydrogenase 3 family member B1 (aldh3b1) mRNA	NM_001093704.1	16.77	7.6	-1.14	0.002
Xenopus laevis kepsin mRNA for epidermis specific serine protease complete cds	AB018694.1	265.09	119.91	-1.14	0.002
PREDICTED: Xenopus (Silurana) tropicalis mucin-4-like (LOC100485319) mRNA	XM_002834058.2	15.17	6.88	-1.14	0.002
Xenopus laevis G6L pathogenesis-related 2 (glpr2) mRNA	NM_001095072.1	29.88	13.66	-1.13	0.002
Xenopus (Silurana) tropicalis hyaluronoglycosaminidase 4 (hyal4) mRNA	NM_001097327.1	12.69	5.8	-1.13	0.002
Xenopus tropicalis inositol (1,4,5) trisphosphate receptor type 3 mRNA partial cds	EF507432.1	63.91	29.14	-1.13	0.002
Xenopus laevis lutein-binding protein 1 (cpne1) mRNA	NM_001090183.1	30.98	14.29	-1.12	0.002
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 160 member A1 (fam160a1) transcript variant X2 mRNA	AJ278067.1	3161.52	1464.01	-1.11	0.002
Xenopus laevis synaptotagmin-like 2 (sytl2) mRNA	XN_004911147.1	18.14	8.43	-1.11	0.002
Xenopus laevis characterized LOC100158314 (LOC100158314) mRNA	NM_001094552.1	67.1	31.2	-1.1	0.002
Xenopus laevis lutein-binding protein 1 (cpne1) mRNA	NM_001127766.1	121.97	57.13	-1.09	0.002
Xenopus laevis solute carrier family 38 member 2 (slc38a2) mRNA	NM_001093606.1	25.24	11.9	-1.09	0.002
Xenopus laevis (MGCB4135) protein (MGCB4135) mRNA	NM_001092726.1	26.99	12.7	-1.09	0.002
Xenopus laevis nuclear factor erythroid 2-related factor 2-like (MGCS5355) mRNA	NM_001086053.1	18.63	8.77	-1.09	0.002
Xenopus laevis fatty acid transporter member 2 mRNA (CDNA clone IMAGE4681491) partial cds	BC041766.1	40.46	19.16	-1.08	0.002
Xenopus laevis long-chain-fatty-acid-CoA ligase ACSBG2 mRNA (cDNA clone MGCE132130 (IMAGE637961) complete cds	BC119493.1	59.3	28.26	-1.07	0.002
Xenopus tropicalis finished cDNA clone TPA012b11	NM_001095094.1	72.9	34.65	-1.07	0.002
PREDICTED: Xenopus (Silurana) tropicalis Golgi membrane protein 1-like (LOC100485898) transcript variant X1 mRNA	CR76877.2	128.9	61.95	-1.06	0.002
Xenopus laevis similar to solute carrier family 27 (fatty acid transporter) member 1 mRNA (LOC100494753) mRNA	XM_002837888.2	77.33	37.05	-1.06	0.002
Xenopus laevis tripeptidyl peptidase I (tpp1) mRNA	NM_001091086.1	102.35	49.32	-1.05	0.002
Xenopus laevis tripeptidyl peptidase I (tpp1) mRNA	NM_001085739.1	358.48	174.85	-1.04	0.002
Xenopus laevis ATPase H+ transporting ubiquitous ATPase subunit 1 (atp6uo1a1) mRNA	NM_001086625.1	40.36	19.75	-1.03	0.002
Xenopus laevis cotonester protein complex subunit gamma 1 (cotp1) mRNA	NM_001090106.1	132.9	64.99	-1.03	0.002
Xenopus laevis AMP-regulated phosphoprotein 19 kDa (app19) mRNA	NM_001093165.1	43.96	21.7	-1.02	0.002
Xenopus laevis phosphoprotein 19 kDa (app19) mRNA	XM_002836862.2	30.96	15.31	-1.02	0.002
PREDICTED: Xenopus (Silurana) tropicalis multidrug resistance protein 1-like (LOC100494753) mRNA	XM_002835245.2	42.32	20.99	-1.01	0.002
Xenopus laevis tripeptidyl peptidase I (tpp1) mRNA	AB033718.2	20	9.98	-1	0.002
Xenopus laevis family with sequence similarity 102 member A (fam102a) mRNA	NM_001092210.1	30.92	2.08	-3.9	0.004
Xenopus laevis characterized protein MGCG6899 (MGCG6899) mRNA	NM_001089696.1	40.49	6.77	-2.58	0.004
PREDICTED: Xenopus (Silurana) tropicalis FOS-like antigen 1 (fos1) mRNA	XM_002832164.2	11.44	3.23	-1.83	0.004
Xenopus laevis guanine nucleotide binding protein (G protein) gamma 7 (gng7-a) mRNA	NM_001085727.1	12.55	3.76	-1.74	0.004
Xenopus laevis characterized protein MGCG5210 (MGCG5210) mRNA	NM_001096801.1	3.5	1.08	-1.7	0.004
Xenopus laevis chromogranin A parathyroid secretory protein 1 (chga) mRNA	NM_001094724.1	8.43	2.7	-1.65	0.004
Xenopus laevis glutathione S-transferase theta 1 (gstt1) mRNA	NM_001091734.1	30.16	9.88	-1.61	0.004
PREDICTED: Xenopus (Silurana) tropicalis protein tyrosine phosphatase receptor type U (ptp1u) transcript variant X2 mRNA	XM_002832164.2	4.77	1.67	-1.52	0.004
Xenopus laevis adenylyl kinase 3 (ak3) nuclear gene encoding mitochondrial protein mRNA	NM_004911581.1	11.82	4.33	-1.45	0.004
PREDICTED: Xenopus (Silurana) tropicalis heterogeneous nuclear ribonucleoprotein R (hnrrpr) transcript variant X2 mRNA	NM_004911580.1	10.96	4.23	-1.37	0.004
Xenopus (Silurana) tropicalis intermediate filament protein 1 (ifip1) mRNA	NM_001142051.1	3.66	1.42	-1.36	0.004
Xenopus laevis transmembrane protein 62 (tmem62) mRNA	NM_001096973.1	6.8	2.65	-1.36	0.004
PREDICTED: Xenopus (Silurana) tropicalis ADP-ribosylation factor-like protein 11-like (LOC100490310) mRNA	XM_002835735.2	19.77	7.86	-1.33	0.004
PREDICTED: Xenopus (Silurana) tropicalis TSC2 domain family member 2 (tsc2dd2) transcript variant X2 mRNA	XM_004914464.1	13.86	5.56	-1.32	0.004
Xenopus laevis adenylyl kinase 2 (ak2) mRNA	NM_001095977.1	6.01	2.42	-1.31	0.004
PREDICTED: Xenopus (Silurana) tropicalis mitochondrial protein mRNA	XM_002835108.2	14.73	6.25	-1.24	0.004
Xenopus laevis coctosin-1-like 1 (cot1) mRNA	NM_001087325.1	39.81	17.69	-1.17	0.004
Xenopus laevis transmembrane 9 superfamily protein member 4 (tm9sf4) mRNA	NM_001089674.1	10.51	4.92	-1.1	0.004
Xenopus laevis histidine ammonia-lyase gene 1 (hal1) mRNA	NM_001089550.1	101.78	50.84	-1	0.004
PREDICTED: Xenopus (Silurana) tropicalis SEC22 vesicle trafficking protein homolog C (sec22c) mRNA	NM_001093175.1	113.48	56.69	-1	0.004
Xenopus laevis solute carrier family 18 (vesicular monoamine) member 2 (slc18a2) mRNA	NM_001094278.1	1.69	0.18	-3.28	0.006
Xenopus (Silurana) tropicalis sterile alpha motif domain containing 1 (samd1) mRNA	NM_001086556.9	2.25	0.31	-2.86	0.006
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00497755 (LOC00497755) mRNA	NM_001029351.1	1.79	0.43	-2.06	0.006
Xenopus (Silurana) tropicalis 5-hydroxytryptamine receptor 7-like (LOC01730375) mRNA	XM_004916478.1	1.33	0.34	-1.97	0.006
PREDICTED: Xenopus (Silurana) tropicalis finger FVE domain containing 28 (zfve28) mRNA	NM_001093695.1	33.23	8.57	-1.96	0.006
Xenopus laevis characterized protein LOC10049141 mRNA (cDNA clone IMAGE6326207) partial cds	BC134824.1	4.13	1.55	-1.42	0.006
PREDICTED: Xenopus (Silurana) tropicalis alpha-1-N-acetylglicosaminyltransferase-like (LOC101733902) mRNA	XM_004918245.1	11.88	4.52	-1.39	0.006
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 169 member A (fam169a) mRNA	XM_002834825.2	15.52	6.72	-1.21	0.006

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO FC <sup>3</sup>	q-value
Xenopus laevis SRG protein-like mRNA (cDNA clone MGC:19649) (IMAGE:336) complete cds	BC169722.1	852.14	367.31	-1.21	0.006
PREDICTED: Xenopus (Silurana) tropicalis Rho-related BTB domain containing 2 (rhohtb2) transcript variant X1 mRNA	XM_002932706.2	10.52	4.64	-1.18	0.006
Xenopus laevis neuronal differentiation 1 (neurod1b) mRNA	NM_001092127.1	4.98	1.51	-1.72	0.007
Xenopus laevis male sperm domain containing 1 (mospd2) mRNA	NM_001095457.1	5.39	1.8	-1.59	0.007
Xenopus laevis 5'-phosphoadenosine 5'-phosphosulfate synthase 2 (paps2) mRNA	NM_001089339.1	6.53	2.19	-1.58	0.007
PREDICTED: Xenopus (Silurana) tropicalis cdk5/cdk5/p35 kinase 7 (dock7) mRNA	XM_002939111.2	5.15	1.83	-1.5	0.007
Xenopus laevis stat1 mRNA complete cds	AY101602.1	4.68	1.69	-1.47	0.007
Xenopus laevis spod8 mRNA complete sequence	DQ096907.1	111.4	42.4	-1.39	0.007
PREDICTED: Xenopus (Silurana) tropicalis ATPase class VI type 1A (atp11a) mRNA	XM_002933015.2	5.87	2.32	-1.34	0.007
Xenopus (Silurana) tropicalis homeobox 2 (hmeis2) mRNA	NM_001097218.1	12.32	5.07	-1.28	0.007
Xenopus laevis triplex mRNa complete cds	DS351290.1	56.56	24.5	-1.21	0.007
Xenopus laevis arylacetamide deacetylase-like 4 (adack4) mRNA	NM_001095048.1	18.25	8.69	-1.07	0.007
Xenopus tropicalis hypothetical protein LOC100145465 mRNA (cDNA clone MGC:185063 IMAGE:7690214) complete cds	BC161094.1	18.22	8.84	-1.04	0.007
Xenopus laevis cDNA clone (IMAGE:6871784)	BC099359.1	84.75	41.75	-1.02	0.007
Xenopus laevis peroxiredoxin 6 mRNA (cDNA clone MGC:64522 IMAGE:6877839) complete cds	BC054278.1	6.3	1.5	-2.08	0.009
Xenopus laevis mRNA for Mig30 complete cds	AB055379.1	4.04	0.97	-2.06	0.009
Xenopus laevis hypothetical protein LOC43216 mRNA (cDNA clone IMAGE:5078910) partial cds	BC072375.1	4.55	1.39	-1.72	0.009
Xenopus laevis inhibitor of DNA binding 4 dominant negative helix-loop-helix protein (id4-a) mRNA	NM_001087235.1	13.99	6.31	-1.15	0.009
PREDICTED: Xenopus (Silurana) tropicalis leucine rich repeat containing 31 (lrfc31) mRNA	XM_002931545.2	10.45	4.78	-1.13	0.009
Xenopus laevis (IMAGE:6871784)	NM_001096675.1	15.88	7.32	-1.12	0.009
Xenopus laevis (Silurana) hypothetical protein LOC101733241 mRNA	XM_004919000.1	86.09	40.06	-1.1	0.009
Xenopus laevis metallopeptidase-disintegrin (MDCG) mRNA complete cds	AF052382.1	6.08	3	-1.02	0.009
PREDICTED: Xenopus (Silurana) tropicalis interacting protein 1 (flip1) transcript variant X1 mRNA	XN_002936356.2	1.28	0.13	-3.37	0.010
Xenopus (Silurana) tropicalis interacting protein 1 (flip1) transcript variant X2 mRNA	XM_004914513.1	1.38	0.27	-2.35	0.010
Xenopus (Silurana) tropicalis uncharacterized protein MGC:145260 (MGC:145260) mRNA	NM_001079412.1	1.94	0.54	-1.86	0.010
Xenopus (Silurana) tropicalis carboxypeptidase Z/CpZ mRNA	NM_001130361.1	3.48	1.25	-1.48	0.010
PREDICTED: Xenopus (Silurana) tropicalis mevalonate kinase (mkv) mRNA	XM_002932064.2	2.75	1.07	-1.37	0.010
Xenopus laevis hypothetical LOC398633 mRNA (cDNA clone IMAGE:7983501)	BC110715.1	385.96	164	-1.23	0.010
Xenopus laevis mRNA for fibronectin leucine rich transmembrane protein 3 (flrt3 gene)	AI605776.1	5.73	2.47	-1.22	0.010
Xenopus (Silurana) tropicalis tBC1 domain member 9 (with GRAM domain) (tbc1d9) mRNA	NM_001137618.1	3.21	1.57	-1.04	0.010
PREDICTED: Xenopus (Silurana) tropicalis trimethylcytosine dioxygenase 2 (tet2) mRNA	XN_002934777.2	6.04	3.03	-1	0.010
Xenopus laevis P2Y1 nucleotide receptor mRNA complete cds	AF42354.1	1.28	0.24	-2.46	0.012
Xenopus laevis retinoic acid-inducible gene 3 (raig3) mRNA complete cds	EF456761.1	11.27	4.87	-1.21	0.012
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733661 (LOC101733661) mRNA	XM_004918356.1	10.92	4.88	-1.16	0.012
Xenopus laevis GDP-mannose 4-dehydratase mRNA (cDNA clone MGC:130830 IMAGE:981605) complete cds	BC11472.1	44.23	20.1	-1.14	0.012
Xenopus laevis mRNA complete cds	AY46070.1	7.41	3.45	-1.1	0.012
PREDICTED: Xenopus (Silurana) tropicalis Ral GEF with PH domain and SH3 binding motif 2 (ralgps2) transcript variant X2 mRNA	XM_004914033.1	8.46	3.95	-1.1	0.012
Xenopus (Silurana) tropicalis methylsterol monooxygenase 1 (msmo1) mRNA	NM_001079241.1	2.12	0.51	-2.05	0.013
Xenopus laevis (IMAGE:809) 3 protein mRNA (cDNA clone MGCS86452 IMAGE:7012443) complete cds	BC086298.1	2.52	0.66	-1.94	0.013
Xenopus (Silurana) tropicalis GTP cyclohydrolase 1 (gch1) mRNA	NM_001067788.1	1.59	0.45	-1.82	0.013
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 2-like (LOC101731505) mRNA	NM_004912203.1	11.48	4.26	-1.43	0.013
Xenopus laevis characterized protein MGC:154351 (MGC:154351) mRNA	NM_001096843.1	25.63	11.11	-1.21	0.013
Xenopus laevis MGCS82349 protein (MGCS82349) mRNA	NM_001093641.1	14.65	6.34	-1.21	0.013
PREDICTED: Xenopus (Silurana) tropicalis human immunodeficiency virus type 1 enhancer binding protein 2 (hivep2) transcript variant X2 mRNA	XN_004914605.1	2.1	0.89	-1.25	0.014
Xenopus laevis characterized LOC1001308 (LOC1001308) mRNA	NM_001093025.1	9.76	4.64	-1.07	0.013
Xenopus laevis metastasis suppressor 1 gene (mss1.2) mRNA	NM_004913749.1	5.52	2.77	-1	0.013
PREDICTED: Xenopus (Silurana) tropicalis interferon alpha/beta receptor 1-like (LOC100498552) transcript variant X4 mRNA	NM_001093916.1	688.95	0.38	-10.86	0.015
Xenopus laevis cytochrome b-245 beta polypeptide (cybb) mRNA	BC073564.1	19.39	0.21	-6.56	0.015
PREDICTED: Xenopus (Silurana) tropicalis activating protein 28 (arhgap28) transcript variant X1 mRNA	XM_004912138.1	3.33	1.12	-1.58	0.015
Xenopus laevis (IMAGE:3396339)	BC080170.1	5.21	1.91	-1.45	0.015
PREDICTED: Xenopus (Silurana) tropicalis Rac guanine nucleotide exchange factor (GEF) 18 (arfgef18) transcript variant X1 mRNA	XM_0049109868.1	5.46	2.46	-1.15	0.015
Xenopus laevis secreted protein acidic cysteine-rich (osteonectin) (sparc) mRNA	NM_001086621.1	19.51	9.01	-1.12	0.015
Xenopus laevis capn13 (capn13) mRNA	NM_001086019.1	67.27	31.51	-1.09	0.015
Xenopus laevis characterized protein MGC:131302 (MGC:131302) mRNA	NM_001096306.1	35.65	16.97	-1.07	0.015

Gene	Accession	Control <sup>1</sup>	Sik1 <sup>-/-</sup> MO <sup>2</sup>	Sik1 <sup>-/-</sup> MO FC <sup>3</sup>	q-value
Xenopus tropicalis finished cDNA clone TNel033k10	CF7600833.2	4.15	0.47	-3.17	0.016
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100491360 (LOC100491360) transcript variant X1 mRNA	XM_004917936.1	194.31	24.98	-2.96	0.016
PREDICTED: Xenopus (Silurana) tropicalis homeobox C12 (hoxc12) mRNA	XM_002936647.2	2.91	0.64	-2.19	0.016
Xenopus (Silurana) tropicalis cytokine inducible SH2-containing protein (cish) mRNA	NM_001113689.1	4.39	1.07	-2.04	0.016
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100487159 (LOC100487159) transcript variant X2 mRNA	XM_004918158.1	6.08	2.19	-1.48	0.016
Xenopus laevis signal sequence receptor gamma translocon-associated protein gamma (ss33- $\alpha$ ) mRNA	NW_001086647.1	397.71	143.46	-1.47	0.016
Xenopus tropicalis zinc finger and BTB domain containing 16 (zbfb16) transcript variant X4 mRNA	XN_004916097.1	3.01	1.29	-1.23	0.016
PREDICTED: Xenopus (Silurana) tropicalis zinc finger and BTB domain containing 1 (lmtf1) mRNA	XM_002932282.2	2.6	1.13	-1.21	0.016
Xenopus tropicalis finished cDNA clone TNel06g11	CR926197.2	9.23	4.35	-1.09	0.016
Xenopus (Silurana) tropicalis homolog subfamily C member 18 (dnajc18) mRNA	BC155359.1	21.52	5.85	-1.88	0.018
Xenopus laevis repetitive Y723 element fragment C17	NM_004910287.1	4.39	1.75	-1.33	0.018
Xenopus laevis repetitive Y723 element fragment C17	X00977.1	77.94	31.87	-1.29	0.018
PREDICTED: Xenopus (Silurana) tropicalis fibrocytin-L-like (LOC100487795) mRNA	XM_004911907.1	2.81	1.3	-1.11	0.018
PREDICTED: Xenopus (Silurana) tropicalis interleukin 17 receptor E (il17e) mRNA	XM_004914115.1	3.1	0.22	-3.84	0.019
Xenopus tropicalis finished cDNA clone TtpA008c12	CR942418.4	6.1	2.09	-1.54	0.019
PREDICTED: Xenopus (Silurana) tropicalis fibrocytin-L-like (LOC100486760) mRNA	XM_002942313.2	21.29	8.08	-1.4	0.019
Xenopus (Silurana) tropicalis claudin-1-like (LOC101732192) mRNA	XM_004911749.1	1.79	0.36	-2.33	0.020
Xenopus laevis sodium channel voltage-gated type III beta subunit (scn3b) mRNA	NM_001093207.1	3.44	1.43	-1.27	0.020
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C8orf34 (LOC100492224)	XM_004915169.1	7.58	3.71	-1.03	0.020
Xenopus laevis anchored LOC00037052 (LOC00037052) mRNA	XM_001097782.1	22.82	11.15	-1.03	0.020
Xenopus (Silurana) tropicalis uncharacterized LOC100124990 (LOC100124990) mRNA	NM_001102919.2	1.78	0.6	-1.57	0.021
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100494783 (LOC100494783) partial mRNA	XM_002942126.2	2.39	0.87	-1.47	0.021
Xenopus (Silurana) tropicalis antioxidant 1 copper chaperone atom 1 mRNA	NM_001092119.1	20.39	9.94	-1.04	0.022
Xenopus laevis hypothetical LOC9536403 (MAGE70115) complete cds	BC08413.1	14.32	6.4	-1.22	0.021
PREDICTED: Xenopus (Silurana) tropicalis pyruvate dehydrogenase phosphatase catalytic subunit 2 (pdp2) mRNA	XM_002941994.2	8.23	4.05	-1.03	0.021
Xenopus (Silurana) tropicalis olfactomedin-like 2A (olfm2a) mRNA	NM_001102975.1	1.35	0.33	-2.03	0.022
PREDICTED: Xenopus (Silurana) tropicalis chondroitin sulphatase N-acetylgalactosaminyltransferase 1 (csgalnact1) mRNA	XM_002933993.2	16.27	7.78	-1.07	0.022
Xenopus laevis MGC82544 protein (MGC82544) mRNA	NM_001096118.1	4.17	1.66	-1.33	0.023
Xenopus laevis SCL2-antagonist/killer 1 (bak1) mRNA	XM_004916298.1	5.49	2.2	-1.32	0.023
Xenopus (Silurana) tropicalis BC1.2-associated transcription factor 1 (bcf1) mRNA	NM_001123491.1	6.92	3.29	-1.07	0.023
Xenopus laevis hypothetical protein LOC779043 (mRNA cDNA clone IMAGE6859066) partial cds	BC110935.1	40.04	6.19	-2.69	0.024
Xenopus (Silurana) tropicalis adipocyte plasma membrane associated protein (apnp) transcript variant X2 mRNA	XN_004912925.1	4.66	1.66	-1.5	0.024
PREDICTED: Xenopus (Silurana) tropicalis gastrin zinc finger protein XICGF57.1-like (LOC101733825) transcript variant X3 mRNA	XN_004915538.1	4.8	0.67	-2.86	0.025
PREDICTED: Xenopus (Silurana) tropicalis p21 protein Cdc42/Rac-activated kinase (paka) partial mRNA	XM_002932826.2	1.16	0.34	-1.8	0.025
PREDICTED: Xenopus (Silurana) galactose-3-O-sulfotransferase 4 (gal3st4) mRNA	XN_002934273.2	1.35	0.41	-1.75	0.025
Xenopus laevis galactose-3-O-sulfotransferase 4 (gal3st4) mRNA	NM_001086139.1	2.44	0.75	-1.7	0.025
PREDICTED: Xenopus (Silurana) dorsal inhibitory axon guidance protein (draxin) mRNA	XM_002937523.2	6.87	3.3	-1.06	0.025
Xenopus tropicalis cDNA clone MGCT7659340 complete cds	BC161041.1	2.59	0.52	-2.33	0.026
XLU38225 Xenopus laevis glutamic acid decarboxylase (GAD) mRNA complete cds	U38225.1	2.2	0.87	-1.35	0.026
PREDICTED: Xenopus (Silurana) tropicalis EPH receptor B1 (ephb1) mRNA	XM_002935075.2	2.89	1.19	-1.28	0.026
Xenopus (Silurana) tropicalis zinc finger protein 47-like (LOC100494841) transcript variant X1 mRNA	XN_002931728.2	12.84	5.91	-1.12	0.026
Xenopus (Silurana) LSm7 homolog 6 small nucleolar RNA associated with ribosomal RNA processing	NM_001171662.1	56.71	27.42	-1.05	0.026
Xenopus laevis hypothetical protein MGCS33862 (mRNA cDNA clone IMAGE6643767) complete cds	BC070801.1	9.68	4.82	-1.01	0.026
Xenopus tropicalis hypothetical protein LOC100135268 (mRNA cDNA clone MGCT860392) complete cds	BC157558.1	10.05	5.03	-1	0.026
Xenopus laevis hypothetical protein LOC73334 mRNA (cDNA clone IMAGE6939205) partial cds	XN_0029274.1	1.41	0.18	-2.98	0.028
Xenopus tropicalis cDNA clone IMAGE7684620 containing frame-shift errors	BC122494.1	4.86	1.89	-1.36	0.028
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100498501 (LOC100498501) mRNA	XM_002943984.2	2.85	0.05	-6.14	0.029
PREDICTED: Xenopus (Silurana) tropicalis deoxyribonuclease-1-like (LOC100493098) mRNA	XM_002932463.2	1.07	0.17	-2.68	0.029
Xenopus (Silurana) tropicalis myocardial zona adherens gene (myzap) mRNA	NM_001113953.2	3.26	1.14	-1.52	0.029
PREDICTED: Xenopus (Silurana) tropicalis FERM and PDZ domain containing 1 (frmpd1) mRNA	XM_004910895.1	1.09	0.24	-2.2	0.030
Xenopus laevis cDNA clone IMAGE8073253 partial cds	BC122494.1	2.39	0.61	-1.98	0.030
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein NGC82246 (MGC82246) mRNA	NM_001099192.1	4.54	1.46	-1.64	0.030
PREDICTED: Xenopus (Silurana) tropicalis 5-hydroxytryptamine (serotonin) receptor 7 adenylate cyclase-coupled (ht7) mRNA	XM_002940010.2	1.81	0.07	-4.89	0.031
Xenopus (Silurana) tropicalis interferon regulation factor 8 (irf8) mRNA	NM_001093628.1	4.33	0.66	-2.72	0.031
Xenopus (Silurana) tropicalis solute carrier family 25 member 43 (slc25a43) mRNA	XM_002934539.2	2.09	0.71	-1.56	0.031
Xenopus (Silurana) tropicalis NGF1-A binding protein 2 (EGFR binding protein 2) (nab2) mRNA	NM_00126669.1	16.45	6.85	-1.26	0.031
Xenopus laevis fks506 binding protein 2.13KDa (fkb2) mRNA	NM_001086024.1	32.3	15.14	-1.09	0.032

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO FC <sup>3</sup>	q-value
Xenopus tropicalis major histocompatibility complex class I antigen mRNA clone MGC:88592 IMAGE:89519261 complete cds	BC16748.1	8.68	2.28	-1.93	0.033
Xenopus laevis hypothetical protein LOC43609 mRNA (cDNA clone IMAGE:5048812) partial cds	BC073025.1	7.49	3.02	-1.31	0.033
Xenopus tropicalis finished cDNA clone TEg059J13	CP942514.2	1.3	0.41	-1.68	0.034
PREDICTED: Xenopus (Silurana) tropicalis actin filament associated protein 1 (afap1) transcript variant X1 mRNA	XM_002938272	1.8	0.81	-1.15	0.034
PREDICTED: Xenopus (Silurana) tropicalis theg spermatid protein (theg) transcript variant X1 mRNA	XM_002939761.2	9.7	4.38	-1.15	0.034
PREDICTED: Xenopus (Silurana) tropicalis UFM1-specific ligase 1 (ulf1) mRNA	XM_002934873.2	7.64	3.77	-1.02	0.034
Xenopus laevis UDP-N-acetyl-alpha-D-galactosaminylpeptidase 1 (GalNAc-T1) (galnt1) mRNA	NM_001089941.1	9.18	4.6	-1	0.034
PREDICTED: Xenopus (Silurana) tropicalis sema domain immunoglobulin domain (lg) short basic domain secreted (semaphorin) 3D (sema3d)	XM_002935247.2	1.25	0.36	-1.82	0.035
PREDICTED: Xenopus (Silurana) tropicalis sema domain immunoglobulin domain (lg) short basic domain secreted (semaphorin) 3D (sema3d)	XM_002939668.2	1.71	0.55	-1.65	0.035
Xenopus laevis thymidine kinase 2 (tk2) mRNA	NM_001094950.1	3.16	1.1	-1.53	0.035
Xenopus laevis thrombomodulin 2 (rbtd2) mRNA	EL367938.1	3.73	1.47	-1.35	0.036
Xenopus laevis TMEM16a mRNA complete cds	NM_001135228.1	5.67	2.31	-1.3	0.036
Xenopus laevis phosphatidylserine synthase 2 (ptds2) mRNA	NM_00110208.1	4.44	1.9	-1.23	0.036
Xenopus laevis nuclear receptor subfamily 4 group A member 2 (nr4a2) mRNA	NM_001114824.1	4.84	2.16	-1.17	0.036
Xenopus laevis phosphatidylethanolamine phospholipase D (napeplid) mRNA	CT485686.1	3.72	1.76	-1.08	0.037
Xenopus tropicalis finished cDNA clone TEg083113	XM_004919498.1	13.13	2.27	-2.54	0.038
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733475 (LOC101733475) mRNA	XM_004915314.1	1.09	0.29	-0.38	0.038
Xenopus laevis (Silurana) tropicalis cadherin type 6 K-cadherin fetal kidney (cdh6) transcript variant X2 mRNA	NM_001130940.1	1.55	0.51	-1.63	0.038
Xenopus laevis interleukin 6 signal transducer (gp130 oncostatin M receptor) (il6st) mRNA	NM_001126752.1	1.6	0.6	-1.41	0.038
Xenopus laevis phosphatidylinositol 4,5-bisphosphate 5-kinase 1 (pi4ki1) mRNA	NM_001087052.1	10.66	5.3	-1.01	0.038
Xenopus laevis trafficking protein particle complex 3 (trappc3) mRNA	NM_001079138.1	1.3	0.33	-1.99	0.038
Xenopus laevis tropomyosin ligand of numb protein X2 (inx2) mRNA	BC072179.1	4.35	1.83	-1.25	0.038
Xenopus laevis uncharacterized LOC80311 (IMAGE:5073822) complete cds	BC154684.2	2.25	0.06	-5.29	0.039
Xenopus tropicalis leucine rich repeat and Ig domain containing 1 mRNA (cDNA clone MGC:7652507) complete cds	NM_001097731.1	2.19	0.33	-2.75	0.039
Xenopus laevis fatty acid amide hydrolase gene 1 (fah1) mRNA	NM_001097731.1	2.19	0.33	-2.75	0.039
PREDICTED: Xenopus (Silurana) tropicalis chimerin 2 (chn2) transcript variant X1 mRNA	XN_002933382.2	1.59	0.46	-1.79	0.039
Xenopus laevis minichromosome maintenance complex component 6 (mcn6) mRNA	NM_001088553.1	4.35	1.79	-1.28	0.039
PREDICTED: Xenopus (Silurana) tropicalis chromosomal unknown open reading frame human C14orf137 (LOC100497155)	XN_004917220.1	2.47	0.84	-1.57	0.041
Xenopus (Silurana) tropicalis ATPase class II type 9A (atp9a) mRNA	NM_203901.1	7.02	2.87	-1.29	0.041
Xenopus laevis G517 protein mRNA (cDNA clone MGC:132219) (IMAGE:66863910) complete cds	BC106651.1	25.62	11.83	-1.12	0.042
Xenopus laevis ceroid-lipofuscinosis neuronal 8 (leptin receptor-like protein with mental retardation) (clf8) mRNA	NM_001094043.1	1.2	0.13	-3.28	0.042
Xaevis mRNA for metallothionein	XE9380.1	17.54	6.98	-1.33	0.042
Xenopus laevis uncharacterized LOC498828 (LOC398828) mRNA	NM_001095186.1	10.4	4.6	-1.18	0.042
Xenopus laevis hypothetical protein LOC398637 mRNA (cDNA clone IMAGE:6880240) partial cds	BC054237.1	9.69	3.03	-3.27	0.044
Xenopus laevis (Silurana) tropicalis lipoma HMGC fusion partner (lhfp) mRNA	NM_00127145.1	1.28	0.16	-3.03	0.044
Xenopus laevis galectin X mRNA partial cds	JN975639.1	11.15	2.48	-2.17	0.044
Xenopus laevis ceroid-lipofuscinosis neuronal 6 (zinc finger protein 51) (clf6) mRNA	NM_001093358.1	1.8	0.65	-1.47	0.044
PREDICTED: Xenopus (Silurana) tropicalis epithelial stromal interaction 1 (breast) (epsti1) mRNA	XM_002938048.2	2.2	0.62	-1.84	0.045
PREDICTED: Xenopus (Silurana) tropicalis met proto-oncogene (hepatocyte growth factor receptor) (met) mRNA	XM_002934955.2	1.79	0.69	-1.38	0.045
Xenopus laevis receptor protein tyrosine phosphatase CRYP-alpha mRNA partial cds	AF198450.1	3.8	1.5	-1.34	0.045
Xenopus laevis iroquois homeobox 4 (ix4-4b) mRNA	NM_001161391.1	6.46	3.17	-1.03	0.045
Xenopus laevis ceramide synthase 2 (cers2) mRNA	NM_001085979.1	7.68	3.67	-1.07	0.046
Xenopus laevis paraplegia 21 (autosomal recessive Mast syndrome) (spg21) mRNA	NM_001095297.1	63.84	0.01	-15.96	0.047
Xenopus laevis retinol dehydrogenase 7 (rdh7) mRNA	NM_001085720.1	68.31	3.5	-4.29	0.047
Xenopus laevis high-affinity Na/dicarboxylate cotransporter mRNA complete cds	AY252508.1	3.99	0.39	-3.39	0.047
Xenopus laevis bovine mRNA for bovine protein complete cds	AB105905.1	37.11	4.1	-3.18	0.047
Xenopus laevis protein kinase C beta 1 (ckb1) mRNA	BC072988.1	1.38	0.3	-2.24	0.047
PREDICTED: Xenopus (Silurana) tropicalis cysteine-rich venom protein tigrin-like (LOC100492430) mRNA	XN_002933580.2	1.74	0.51	-1.78	0.047
Xenopus laevis protease-activated receptor 2 (par2) mRNA	NM_001093903.1	5.36	2.56	-1.07	0.047
Xenopus laevis peroxisome proliferator-activated receptor gamma mRNA (cDNA clone MGC:658675) IMAGE:4032975 complete cds	NM_001095490.1	7.11	3.45	-1.05	0.048
Xenopus laevis peroxisome proliferator-activated receptor gamma mRNA (cDNA clone MGC:658675) IMAGE:4032975 complete cds	BC060474.1	4.44	1.42	-1.65	0.049

<sup>1</sup> Expression levels (FPKM) in un-injected pre-placodal explants

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1-MO

<sup>3</sup> Log<sub>2</sub> Fold change values for Six1-MO

## I.2 Six1-MO: Up-regulated genes

**Table I.2** Genes significantly up-regulated after injection of *Six1*-MO

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO FC <sup>3</sup>	q-value
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC100485313) mRNA	XM_009162901	0.01	148.576	20.5	0.002
PREDICTED: Xenopus (Silurana) tropicalis alpha-N-acetylgalactosaminide alpha-2-6-sialyltransferase 2-like (LOC100489217) transcript variant X1 mRNA	XM_002940092.2	0.01	517.63	18.98	0.002
Xenopus laevis hypothetical protein MGCG8493 mRNA (CDNA clone MGCG8493) (mRNA (CDNA clone IMAGE5513111)) partial cds	BC068970.1	0.01	216.93	17.73	0.002
Xenopus laevis hypothetical protein LOC131726 (LOC131726) partial cds	BC091726.1	0.01	19.53	14.25	0.002
Xenopus laevis GTPase activating protein 29 (arntgap29) mRNA	NM_001105265.1	0.01	9.85	13.27	0.002
Xenopus laevis Xpat protein mRNA (cDNA clone MGCG79925) (MAGE5-5155227) complete cds	BC072723.1	0.01	7.07	12.79	0.002
Xenopus laevis CD8alpha receptor mRNA complete cds	HQ116783.1	0.01	4.68	12.19	0.002
PREDICTED: Xenopus (Silurana) tropicalis adenosine receptor A2a-like (LOC10173477) mRNA	XM_004914806.1	0.01	4.38	12.1	0.002
PREDICTED: Xenopus (Silurana) tropicalis containing 2 (enth2) transcript variant X1 mRNA	XM_002940093.2	0.01	3.09	11.59	0.002
XENSTAGE: Xenopus laevis disintegrin and metalloproteinase domain-containing protein 28-like (LOC100498252) mRNA	XM_002932556.1	0.01	2.16	11.07	0.002
Xenopus laevis solute carrier family 28 sodium-coupled nucleoside transporter member 2 (slc28a2) mRNA	LO4278.1	0.01	2.14	1.06	0.002
XENSTAGE: Xenopus laevis keratin filament-binding protein-like (LOC100487003) mRNA	NM_001093333.1	0.01	3.02	10.56	0.002
PREDICTED: Xenopus (Silurana) tropicalis trichorhine keratin non-receptor type substrate 1-like (LOC10173337) mRNA	XM_002937028.2	0.01	1.37	10.41	0.002
Xenopus (Silurana) tropicalis uncharacterized LOC1001730752 (LOC1001730752) mRNA	XM_004917789.1	0.01	1.12	10.12	0.002
PREDICTED: Xenopus (Silurana) tropicalis FXYD domain-containing transport regulator 7 (kyt7) mRNA	XM_002942646.2	0.01	1.09	10.08	0.002
PREDICTED: Xenopus (Silurana) tropicalis DNA-directed RNA polymerase II subunit RPB1-like (LOC101733335) mRNA	XM_004918771.1	0.01	1.08	10.07	0.002
Xenopus laevis cDNA clone MGCG6866291 complete cds	BC082432.1	1.96	1068.47	9.09	0.002
Xenopus tropicalis finished cDNA clone Theud023c10	CT030610.1	7521.61	40682.06	9.08	0.002
Xenopus laevis characterized LOC495337 (LOC495337) mRNA	NM_001093003.1	3.65	1794.42	8.94	0.002
Xenopus (Silurana) tropicalis tyrosine-protein phosphatase non-receptor type substrate 1-like (LOC101733531) mRNA	XM_004918397.1	0.07	24.92	8.69	0.002
Xenopus laevis characterized LOC100145470 (LOC100145470) mRNA	NM_001126922.1	0.06	12.2	7.71	0.002
Xenopus laevis characterized LOC100158288 (LOC100158288) mRNA	NM_001127245.1	13942.08	20413.28	7.94	0.002
Xenopus laevis TAR (HIV-1) RNA binding protein 2 (tarbp2) mRNA	NM_001092051.1	54.7	4845.82	6.47	0.002
PREDICTED: Xenopus (Silurana) tropicalis class I histocompatibility antigen F10 alpha chain-like (LOC100494618) mRNA	XM_002940092.2	0.05	3.83	6.33	0.002
Xenopus laevis cDNA clone IMAGE6866501	BC084311.1	3.28	221.78	6.08	0.002
Xenopus laevis hypothetical LOC494808 mRNA (cDNA clone IMAGE7009702) partial cds	BC082947.1	1.87	101.66	5.77	0.002
Xenopus tropicalis hypothetical protein LOC548740 mRNA (cDNA clone MGCI:147107 IMAGE7729681) complete cds	BC125765.1	1	41.13	5.37	0.002
Xenopus laevis F10x protein 6 (f10x6) mRNA	NM_001090855.1	0.76	30.12	5.31	0.002
Xenopus laevis transmembrane protein 168 (tmem168) mRNA	NM_001087379.1	35.11	123.06	5.13	0.002
Xenopus laevis 5'-nucleotidase domain containing 3 (nt5dc3) mRNA	NM_001092501.1	6.2	200.77	5.02	0.002
Xenopus laevis myelobium cofactor synthesis 1 (mcfs1) mRNA	NM_001092732.1	5.15	162.64	4.98	0.002
Xenopus tropicalis cDNA clone IMAGE754730	BC166316.1	2905.49	79067.79	4.77	0.002
Xenopus laevis nestin (nes) mRNA	NM_001087857.1	147.44	3959.73	4.75	0.002
Xenopus laevis MGCG80661 protein (MGCG80661) mRNA	NM_00109229.1	21.11	550.38	4.7	0.002
Xenopus laevis proteasome (prosome-macropain) 26S subunit ATPase 3 (psmnc3) mRNA	NM_00109302.1	11.41	25	4.36	0.002
Xenopus laevis cDNA clone MGCG131018 (MAGE77379650) complete cds	BC10853.1	14.43	265.89	4.2	0.002
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100505443 (LOC100505443) mRNA	AB194899.1	6.39	107.31	4.07	0.002
Xenopus laevis characterized LOC100496845 (LOC100496845) mRNA	NM_001197229.1	0.24	3.21	3.79	0.002
Xenopus laevis retrotransposon-like element partial sequence	AF057166.1	7.48	86.33	3.53	0.002
Xenopus laevis proteasome (prosome-macropain) 26S subunit ATPase 3 (psmnc3) mRNA	NM_00108214.1	0.33	12251.24	3.5	0.002
Xenopus laevis integrin alpha 2b (platelet glycoprotein lib of lib/lla complex antigen CD41) gene 2 (iga2b2) mRNA	NM_001094754.1	13.26	34.42	3.11	0.002
PREDICTED: Xenopus (Silurana) tropicalis 3-kinase regulatory subunit 5 mRNA (CDNA clone IMAGE4968567) partial cds	BC072301.1	0.29	138.83	3.39	0.002
Xenopus laevis characterized LOC100496845 (LOC100496845) mRNA	NM_001092728.1	25.16	230.37	3.2	0.002
Xenopus laevis squalate dehydrogenase 1 (NADP+)-soluble [idh1] mRNA	NM_00109453.1	54.89	5010.89	3.19	0.002
Xenopus laevis bromodomain containing 3 (bpd3) mRNA	NM_001097126.1	3.76	33.19	3.14	0.002
Xenopus laevis caudal type-homeobox 4 mRNA (CDNA clone MGCG88909) (MAGE4889626) complete cds	AY260729.1	0.34	2.87	3.11	0.002
PREDICTED: Xenopus (Silurana) tropicalis Na+-K+-transporting beta 2 polypeptide (atp1b2) mRNA	NM_001091970.1	1.11	9.26	3.07	0.002
Xenopus laevis alpha 2-macroglobulin-like 1 (a2ml1) mRNA	NM_001135077.1	0.31	2.49	3.04	0.002
Xenopus tropicalis mRNA for beta-1-galactosidase 5T6Gal1 (sgal1) gene	FN996984.1	0.39	40.96	3.02	0.002
Xenopus laevis IMP protein (LOC0203394) mRNA	NM_001096833.1	1.18	9.12	2.95	0.002
		0.18	1.31	2.87	0.002

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO <sup>FC*</sup>	q-value
Xenopus laevis serum/glucocorticoid regulated kinase 1 (sgk1-a) mRNA	NM_00109488256	95.63	690.44	285	0.002	
PREDICTED: Xenopus (Silurana) tropicalis insulin-like growth factor-binding protein complex acid labile subunit-like (LOC100488256) mRNA	XM_004918587.1	0.31	2.18	283	0.002	
PREDICTED: Xenopus (Silurana) tropicalis ectodysplasin A2 receptor (edar2) transcript variant X1 mRNA	XM_002938195.2	0.63	4.44	283	0.002	
Xenopus laevis xCAD2 protein mRNA (MGC:160980   IMAGE:8319613) complete cds	BC146637.1	0.85	5.59	272	0.002	
Xenopus laevis aquaporin 3 (Gill blood group) (aqn3-b) mRNA	NM_001094477.1	3.3	21.46	271	0.002	
Xenopus laevis nucleolar protein 12 (nol 12) mRNA	NM_001096931.1	10.4	65.4	265	0.002	
Xenopus (Silurana) tropicalis solute carrier family 7 (glycoprotein-associated amino acid transporter light chain bo + system) member 9 (slc7a9) mRNA	NM_001092022.1	11.94	73.86	263	0.002	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734826 mRNA	XM_004910729.1	2.8	16.82	259	0.002	
Xenopus laevis hyaluronan synthase related sequence protein (has-19) mRNA	NM_001088339.1	4.39	26.15	257	0.002	
Xenopus laevis phosphatidylinositol 3'-trisphosphate-dependent Rac exchange factor 1 (prex1) mRNA	NM_001090233.1	55.62	316.19	251	0.002	
Xenopus laevis colloid-coil domain containing 149 (ccdc149) mRNA	NM_001091433.1	12.63	72.17	251	0.002	
Xenopus laevis peroxidase 2 (POX2) mRNA complete cds	AY069942.2	2.34	12.95	247	0.002	
PREDICTED: Xenopus (Silurana) tropicalis histone H2B 1.1-like (LOC100495396) transcript variant 1 mRNA	NM_002943267.2	28.07	151.73	243	0.002	
Xenopus (Silurana) tropicalis C-reactive protein-like (LOC100489750) mRNA	XM_002937750.2	0.92	4.82	241	0.002	
PREDICTED: Xenopus (Silurana) tropicalis apoptosis-associated speck-like protein containing a CARD-like (LOC101730604) mRNA	XM_004914290.1	0.43	2.17	234	0.002	
Xenopus laevis phosphatidylglycosidase F (pgif) mRNA	NM_001095344.1	23.51	118.12	233	0.002	
Xenopus laevis Wnt-1/int-1-related mRNA (CDNA clone MAGE:5156763) complete cds	BC082627.1	5.64	27.32	228	0.002	
Xenopus laevis solute carrier family 16 (mannosidase-like acid transporters) member 6 (slc16a6) mRNA	NM_001086838.1	3.47	16.39	224	0.002	
Xenopus laevis tDNA clone MGC:81566 IMAGE:6863092 complete cds	BC082361.1	17.57	80.8	22	0.002	
Xenopus laevis cry1 protein (cry1) mRNA	NM_001095521.1	1.01	4.6	21.9	0.002	
Xenopus laevis retinoschisin 1 (rs1) mRNA	NM_001096951.1	10.47	46.33	21.5	0.002	
Xenopus tropicalis cDNA clone IMAGE:6991249	BC158454.1	595346.95	2596904.78	212	0.002	
Xenopus laevis hypothetical protein LOC443701 mRNA (CDNA clone IMAGE:7008877) partial cds	BC074228.1	0.9	3.92	212	0.002	
Xenopus laevis SPB-a mRNA complete cds	FJ644945.1	0.98	4.13	208	0.002	
Xenopus laevis bone morphogenic protein 7 mRNA (CDNA clone MGC:68434 IMAGE:6862718) complete cds	BC057702.1	1.16	6.39	205	0.002	
Xenopus laevis ancinthor characterized (LOC100037207) (LOC100037207) mRNA	NM_001097896.1	97.4	401.19	204	0.002	
Xenopus (Silurana) tropicalis alpha-3 (latf3) transcript variant X2 mRNA	XM_004915795.1	1.21	4.77	198	0.002	
Xenopus laevis ancinthor characterized protein MGCS1393 (MGCS1393) mRNA	NM_001091196.1	15.26	59.97	197	0.002	
Xenopus laevis hypothetical protein LOC10127319 mRNA (CDNA clone IMAGE:8319350) partial cds	BC155933.1	30.44	114.86	192	0.002	
Xenopus laevis hypothetical LOC495232 mRNA (CDNA clone IMAGE:511906) partial cds	BC084651.1	17.27	63.75	188	0.002	
Xenopus laevis synapsin light chain 2 mRNA (CDNA clone MGC:85772 IMAGE:857060) complete cds	BC088794.1	1.25	4.58	188	0.002	
PREDICTED: Xenopus (Silurana) tropicalis spike homolog 2 (Drosophila) (spire2) transcript variant X1 mRNA	NM_002933684.2	0.79	2.88	187	0.002	
Xenopus laevis ornithine decarboxylase 2 mRNA (CDNA clone MGC:52527 IMAGE:542547) complete cds	BC047954.1	173.45	627.81	186	0.002	
Xenopus laevis orthodontide homolog 5 mRNA (CDNA clone MGC:79897 IMAGE:5156432) complete cds	BC084194.1	2.61	9.47	186	0.002	
Xenopus laevis peripheral myelin protein 22 (pmp22) mRNA	NM_001093533.1	4	14.07	182	0.002	
Xenopus laevis glutathione reductase (grs) mRNA	BC159022.1	3.05	10.53	17.9	0.002	
Xenopus laevis tRNA clone MGC:196526 IMAGE:9041433 complete cds	NM_001095833.1	180.46	620.56	17.8	0.002	
Xenopus laevis 3p -like zinc-finger protein XSPR-2 mRNA complete cds	AY062263.1	4.49	15.35	17.7	0.002	
Xenopus (Silurana) tropicalis cellular retinoic acid binding protein 2 (crabp2) mRNA	NM_001114770.1	1.25	4.21	1.76	0.002	
Xenopus laevis pituitary homeobox gene 1 paired-like homeodomain transcription factor (Pitx1) mRNA complete cds	AF217647.1	5.5	18.43	1.75	0.002	
Xenopus laevis paired-like homeodomain transcription factor 2 mRNA (CDNA clone MGC:7523076) complete cds	NM_001088447.1	8.28	27.18	1.72	0.002	
Xenopus tropicalis p53 inducible protein 11 (tp53i11) mRNA	NM_001097917.1	5.82	17.87	1.62	0.002	
PREDICTED: Xenopus (Silurana) tropicalis nuclear export mediator factor (nemf) transcript variant X1 mRNA	NM_002938127.2	9.35	28.57	1.61	0.002	
Xenopus (Silurana) tropicalis integrin alpha 5 (fibronectin receptor alpha polypeptide) (itga5) mRNA	NM_00109862.1	13.56	40.67	1.59	0.002	
Xenopus laevis aquaporin 35 (Upf-N-acetylglucosamine UDP-GlcNAc) transporter, member A3 gene 1 (slc35a3.1) mRNA	NM_00109261.1	4.96	14.17	1.52	0.002	
Xenopus laevis paired-like homeodomain transcription factor 2 mRNA (CDNA clone MGC:80997 IMAGE:5542798) complete cds	BC073479.1	6.43	18.47	1.52	0.002	
Xenopus laevis RIO kinase 3 (rik3) mRNA	NM_00108923.1	8.28	23.38	1.5	0.002	
Xenopus laevis mRNA for Tlrat complete cds	AB075925.1	69.12	195.84	1.5	0.002	
xCRBP=cellular retinoic acid-binding protein	S74933.1	27.7	78.57	1.5	0.002	
Xenopus laevis synapsin light chain dcdb mRNA complete cds	DQ487758.4	8.72	24.41	1.49	0.002	
PREDICTED: Xenopus (Silurana) tropicalis integrin alpha 5 (fibronectin receptor alpha polypeptide) (itga5) mRNA	NM_001114769.1	1.29	3.58	1.47	0.002	
Xenopus laevis acid phosphatase 1 soluble (acp-1) transcript variant b mRNA	BC073568.1	6.18	17.15	1.47	0.002	
Xenopus laevis synapsin 3 (syn3) protein mRNA (CDNA clone MGC:82859 IMAGE:511530) complete cds	CT025392.2	38.12	103.58	1.44	0.002	
Xenopus tropicalis finished cDNA clone TGA063099	BC082728.1	263.07	706.63	1.43	0.002	
Xenopus laevis tiam1-binding signaling subunit (Tcr22) mRNA complete cds	EF431896.1	2.11	5.66	1.42	0.002	
Xenopus laevis tkrp-like factor 10 (kfr10) mRNA	NM_001095871.1	6.63	17.58	1.41	0.002	

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO <sup>3</sup>	FC <sup>4</sup>	q-value
Xenopus (Silurana) tropicalis tumor protein p53 (tbp53) mRNA	NM_001001903.1	14.73	39.15	1.41	0.002	
Xenopus laevis (MGCB1) 892 protein (MGCB1892) mRNA	NM_001092597.1	6.58	17.11	1.38	0.002	
Xenopus laevis uncharacterized LOC100158370 (LOC100158370) mRNA	NM_001127815.1	1511.73	3889.72	1.36	0.002	
Xenopus laevis receptor-interacting serine-threonine kinase 4 mRNA (cDNA clone MGC:132134 IMAGE:4741010) complete cds	BC108792.1	5.54	13.99	1.34	0.002	
PREDICTED: Xenopus (Silurana) tropicanl kinase B (ubb) transcript variant X2 mRNA	XM_004911681.1	3339.67	8437.52	1.34	0.002	
Xenopus laevis heat shock protein 90kDa alpha (cytosolic) class A member 1 gene 1 (hsps90aa1.1) mRNA	NM_001092129.1	7.8	19.45	1.32	0.002	
Xenopus laevis cytochrome P450 family 26 subfamily 2 protein (cyp26c1) mRNA	NM_001096487.1	14.25	35.09	1.3	0.002	
PREDICTED: Xenopus (Silurana) tropicanl kinase B (ubb) transcript variant X1 mRNA	XM_004920544.1	66.68	163.03	1.29	0.002	
Xenopus laevis pancreatic lipase-related protein 1 (plnlp1) mRNA	NM_001094231.1	151.15	36.92	1.29	0.002	
Xenopus (Silurana) tropicalis methyltransferase like 22 (mettl22) mRNA	NM_001127301.1	0.82	1.96	1.26	0.002	
Xenopus laevis keratin 5 gene 1 b (krk5.1b) mRNA	NM_001086741.1	17.42	41.56	1.25	0.002	
PREDICTED: Xenopus (Silurana) tropicalis transformation/transcription domain-associated protein (trap) transcript variant X2 mRNA	XM_004917975.1	5.22	12.28	1.24	0.002	
Xenopus (Silurana) tropicalis epiregulin (ereg) mRNA	NM_001086754.1	7.55	17.69	1.23	0.002	
Xenopus laevis protein phosphatase 1 catalytic subunit gamma isoform (ppp1cc) mRNA	NM_001087839.1	10.23	24.06	1.23	0.002	
Xenopus laevis (moprbp1) mRNA	NM_001092329.1	16.98	39.53	1.22	0.002	
PREDICTED: Xenopus (Silurana) tropicanl kinase B (ubb) transcript variant X1 mRNA	BC130151.1	1773.67	4128.62	1.22	0.002	
Xenopus laevis CDNA clone IMAGE:7200336	BC087622.1	11.81	1.21	0.002		
Xenopus laevis hypothetical LOC496107 mRNA (cDNA clone MGCG88335 IMAGE:3401690) complete cds	NM_001086720.1	146.30	334.43	1.19	0.002	
Xenopus laevis uncoupling protein 2 (mitochondrial proton carrier) (ucp2) nuclear gene encoding mitochondrial protein mRNA	M6363.1	2133.26	4829.39	1.18	0.002	
XELB1AA XboII restriction endonuclease B1 protein mRNA complete cds	NM_001171732.1	19.51	43.8	1.17	0.002	
Xenopus laevis zinc finger protein 706 gene 2 (znf706.2-a) mRNA	BC070550.1	20.57	45.77	1.15	0.002	
Xenopus laevis CRM1/XPO1 protein mRNA (cDNA clone MGCG77902 IMAGE:6639569) complete cds	NM_00109235086.2	33.91	75.43	1.15	0.002	
PREDICTED: Xenopus (Silurana) tropicanl kinase B (ubb) transcript variant X1 mRNA	D42066.1	111.17	24.26	1.12	0.002	
XELXLO1A Xenopus laevis mRNA for pleiotropic factor-beta2 complete cds	NM_001093576.1	207.4	45.19	1.12	0.002	
Xenopus laevis RIO kinase 3 (yak3) (rik3) mRNA	NM_001092233.1	17.71	38.21	1.11	0.002	
PREDICTED: Xenopus (Silurana) tropicanl kinase 2-like (amot2) transcript variant X2 mRNA	NM_001094747.1	16.41	34.89	1.09	0.002	
Xenopus laevis (moprb6) mRNA	NM_001095726.1	7.85	16.6	1.08	0.002	
Xenopus laevis cyclin A2 (ccna2) mRNA	NM_001096243.1	23.11	48.56	1.07	0.002	
X. laevis ODC mRNA for ornithine decarboxylase (EC 4.1.1.17)	X56316.1	370.56	775.03	1.06	0.002	
Xenopus laevis similar to hyaluronan synthase mRNA (cDNA clone MGCG13076 IMAGE:7978364) complete cds	BC108480.1	114.19	232.35	1.02	0.002	
PREDICTED: Xenopus (Silurana) tropicanl kinase 2-like (amot2) transcript variant X2 mRNA	XM_004916445.1	2.55	5.16	1.02	0.002	
Xenopus laevis G protein-coupled receptor kinase E2O mRNA	NM_002935942.2	9.26	18.53	1	0.002	
PREDICTED: Xenopus (Silurana) tropicanl kinase 17 (amot17) mRNA	NM_001094448.1	0.01	10.35	13.34	0.004	
XENOLX1A Xenopus (Silurana) tropicanl kinase 17 (amot17) mRNA	NM_0029398772.2	0.01	3.42	11.74	0.004	
Xenopus laevis cyclin A1 skeletal muscle mRNA (cDNA clone MGCG2636 IMAGE:4681379) complete cds	BC041197.1	0.56	6.04	3.45	0.004	
Xenopus laevis mRNA for aquaporin-3	AJ131847.1	0.5	5.27	3.42	0.004	
Xenopus (Silurana) tropicalis carbonic anhydrase 6 (ca6) mRNA	NM_001079014.1	3.81	22.7	2.57	0.004	
Xenopus laevis solute carrier family 38 member 5 (slc38a5) mRNA	NM_001092358.1	1.13	4.14	1.88	0.004	
PREDICTED: Xenopus (Silurana) tropicanl kinase 17 (amot17) mRNA	NM_001094738.1	4.45	14.45	1.7	0.004	
Xenopus laevis Nk3 homeobox 1 (nkx3-1-a) mRNA	NM_001086738.1	3.19	8.91	1.48	0.004	
PREDICTED: Xenopus (Silurana) tropicanl kinase 17 (amot17) mRNA	BC108429.1	6.21	16.05	1.37	0.004	
Xenopus laevis posterior fgf gene	X56487.1	537.16	1370.652	1.35	0.004	
Xenopus laevis paired-domain transcription factor Pax3delta isoform mRNA complete cds	AY725268.1	7.73	18.09	1.23	0.004	
XENOLX779 Xenopus laevis xot-79 mRNA 3' end	M60076.1	3.8	8.37	1.14	0.004	
Xenopus laevis solute carrier family 8 (sodium/calcium exchanger) member 3 (slc8a3) mRNA	BC070526.1	12.51	26.65	1.09	0.004	
XELP53AA Xenopus p53 homologue mRNA complete cds	M36362.1	12.65	26.67	1.08	0.004	
Xenopus laevis cydin E2 (ccne2) mRNA	NM_001087058.1	8.53	17.69	1.05	0.004	
Xenopus laevis WWP1 homolog (wwp1) mRNA	NM_001091163.1	0.01	221.41	11.76	0.006	
PREDICTED: Xenopus (Silurana) tropicanl kinase 17 (amot17) mRNA	DQ355794.1	0.57	15.64	4.78	0.006	
Xenopus laevis solute carrier family 8 (sodium/calcium exchanger) member 3 (slc8a3) mRNA	NM_001093062.1	2.69	45.69	4.09	0.006	
Xenopus laevis p53 homologue mRNA complete cds	XM_004917428.1	0.32	3.28	3.4	0.006	
Xenopus laevis mRNA for Zic2 protein complete cds	AB05292.2	2.42	5.74	1.25	0.006	
PREDICTED: Xenopus (Silurana) tropicanl kinase 17 (amot17) transcript variant X1 mRNA	XM_002941364.2	4.19	9.5	1.18	0.006	
Xenopus laevis glucosidase alpha; neutral AB (gabn) mRNA	NM_001097631.1	10.59	21.99	1.06	0.006	
Xenopus laevis tumor-suppressor protein p53 mRNA (cDNA clone MGCG79985 IMAGE:6864775) complete cds	BC084064.1	26.43	54.53	1.04	0.006	
Xenopus laevis hypothetical protein LOC733407 mRNA (cDNA clone IMAGE:7206895) partial cds	BC108604.1	2.01	57.39	4.84	0.007	
Xenopus laevis trimethyllysine hydroxylase epsilon (tmhle) nuclear gene encoding mitochondrial protein mRNA	NM_001135068.1	0.52	2.41	2.22	0.007	

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO <sup>3</sup>	FC <sup>4</sup>	q-value
Xenopus tropicalis finished cDNA clone TEG0155p05	CR761243.2	5.22	15.92	1.61	0.007	
Xenopus laevis amyloid beta (A4) precursor-like protein 1 (aplip1) mRNA	NM_001092950.1	2.75	8.28	1.59	0.007	
Xenopus laevis phosphoinositide 3-kinase regulatory subunit polypeptide 2 (p85 beta) mRNA	NM_001093482.1	0.73	2.17	1.57	0.007	
Xenopus laevis odd-paired-like (opl) mRNA complete cds	AF028805.1	2.32	6.54	1.5	0.007	
X.laevis mRNA for Xom protein	X98454.1	4.32	11.98	1.47	0.007	
PREDICTED: Xenopus (Silurana) tropicalis p53-induced death domain protein (pidd) transcript variant X3 mRNA	XW_004913674.1	2.05	5.07	1.31	0.007	
Xenopus laevis thromboxane A synthase 1 (ptxas1) mRNA	XM_002931500.2	2.3	5.09	1.15	0.007	
X.laevis mRNA for transcription factor V alpha 2 (colva2) mRNA	NM_001096002.1	1.068	23.12	1.12	0.007	
Xenopus laevis family with sequence similarity 115 member A (fam115a) mRNA	NM_001094696.1	51.38	104.86	1.03	0.007	
Xenopus laevis mRNA for Xom protein	AB091393.1	0.01	19.09	14.22	0.009	
Xenopus tropicalis finished cDNA clone TGas11924	CT023457.1	13.45	103.78	2.95	0.009	
Xenopus laevis Ras association domain 8 (RASSF8) mRNA complete cds	EF623991.1	0.71	2.58	1.87	0.009	
Xenopus laevis thiombarane A synthase 1 platelet (ttxas1) mRNA	NM_001096983.1	1.02	3.27	1.68	0.009	
X.laevis mRNA for transcription factor VENT-2	X98849.1	3.1	8.73	1.49	0.009	
Xenopus laevis cDNA clone (ME-E4969048	BC077701.1	1.64	4.19	1.36	0.009	
Xenopus laevis paired-domain transcription factor Pax-3 mRNA complete cds	AY725269.1	5.17	11.19	1.11	0.009	
Xenopus laevis tbx6 mRNA complete cds	BC133187.1	3.3	7.07	1.1	0.009	
Xenopus laevis taurine carrier family 6 (amino acid transporter) member 1 gene 2 (slc6a14.2) mRNA	NM_001093067.1	0.75	6.76	3.19	0.010	
Xenopus laevis DEAD/South RNA helicase (DEADSout) mRNA complete cds	NM_001091185.1	0.53	2.52	2.27	0.010	
Xenopus laevis mRNA for Zic2 protein complete cds	AF190623.1	0.77	2.98	1.96	0.010	
Xenopus laevis (Silurana) tropicalis transformation/transcription domain-associated protein (trtrap) transcript variant X1 mRNA	AB009565.1	1.84	6.01	1.71	0.010	
PREDICTED: Xenopus laevis (Silurana) tropicalis transformation/transcription domain-associated protein (trtrap) transcript variant X1 mRNA	XW_002931947.2	2.78	5.99	1.11	0.010	
Xenopus laevis programmed cell death 6 mRNA (cDNA clone MG:13214 IMAGE:65448231) complete cds	BC110939.2	19.03	39.35	2.05	0.010	
XENRAQBP_X.laevis oocyte 5s rRNA pseudogene clone XRN31	K01368.1	1.79	58.96	5.04	0.012	
Xenopus laevis sarf domain containing kinase 3 (adck3) nuclear gene encoding mitochondrial protein mRNA	NM_001097842.1	3.98	9.16	1.21	0.012	
Xenopus laevis t-box protein TBX20 mRNA complete cds	AF308810.1	2.59	5.21	1.01	0.012	
Xenopus laevis hypothetical protein LOC100495360	BC167561.1	0.01	33.46	15.03	0.013	
PREDICTED: Xenopus laevis (Silurana) tropicalis side-chain cleavage enzyme mitochondrial-like (LOC100495360) partial cds	NM_001093023.1	0.19	2.28	3.6	0.013	
Xenopus laevis actinin alpha 3 (actn3) mRNA	NM_001091063.1	9.58	42.49	2.15	0.013	
Xenopus laevis RAS guanyl-releasing protein 1 (calmodulin and DAG-regulated) (rasgrp1) mRNA	NM_001092094.1	0.01	42.68	15.38	0.014	
Xenopus laevis leucyl-tRNA synthetase 2 mitochondrial (lars2) nuclear gene encoding mitochondrial protein mRNA	AY154394.1	0.51	1.54	1.61	0.014	
Xenopus laevis t-box protein TBX20 mRNA complete cds	XM_002934562.2	11.93	27.73	1.22	0.014	
PREDICTED: Xenopus laevis (Silurana) tropicalis cholesterol side-chain cleavage enzyme mitochondrial-like (LOC100495360) mRNA	NM_001093977.1	135.91	866.67	2.67	0.015	
Xenopus laevis taurine/oxalylacetate aminotransferase 2 (taox2) mRNA	BC070481.1	0.52	2.2	2.1	0.015	
Xenopus laevis thimet oligopeptidase mRNA (cDNA clone MG:683753 IMAGE:66640474) complete cds	XW_004914893.1	15.5	53.15	1.78	0.015	
PREDICTED: Xenopus (Silurana) tropicalis linker PHD RING helicase E3 (ubiquitin protein ligase (shprh) mRNA)	CR855460.2	1.76	4.33	1.3	0.015	
Xenopus tropicalis finished cDNA clone Theu031n03	XM_002934984.2	1.22	31.67	1.01	0.015	
Xenopus laevis cyclin O (ccno) mRNA	NM_001094210.1	15.84	70.53	1.53	0.016	
Xenopus laevis zinc finger CCCH domain containing 10 (zcchc10) mRNA	NM_001093648.1	24.48	0.6	1.37	0.016	
PREDICTED: Xenopus (Silurana) tropicalis neurofascin (nfasc) transcript variant X1 mRNA	XM_002936220.2	0.26	1.86	2.87	0.018	
Xenopus laevis ACT-25 mRNA (cDNA clone MG:68441 IMAGE:6686305) complete cds	BC055964.1	0.01	3.59	1.83	0.018	
Xenopus laevis sulfhydryl oxidase 1 mRNA	XM_002934174.1	0.23	3.19	1.39	0.018	
PREDICTED: Xenopus laevis (Silurana) tropicalis G protein-coupled receptor family C group 5 member C (gprcsc) transcript variant X1 mRNA	X659662.1	3.41	8.1	1.25	0.018	
Xenopus laevis taurine synthetase large subunit	BC072283.1	0.17	4.65	4.81	0.019	
PREDICTED: Xenopus laevis (Silurana) tropicalis beta-1,3-galactosyltransferase 2-like (LOC100485796) partial cds	NM_001093046.1	15.45	349.51	4.5	0.019	
Xenopus laevis myb/SANT-like DNA-binding domain containing 4 with coiled-coil (msantd4) mRNA	NM_001093992.1	0.23	1.56	2.82	0.019	
Xenopus laevis histone H3 NME1 protein (MG:86493) mRNA	NM_001102737.1	0.24	1.4	2.56	0.019	
Xenopus laevis taurine/oxalylacetate aminotransferase 2 (taox2) mRNA	U11731.1	0.51	2.71	2.43	0.019	
X.laevis mRNA for gastrula-specific protein kinase	X99406.1	0.33	1.7	2.4	0.019	
X.laevis mRNA for Zic-related-2 complete cds	AY648303.1	0.53	2.52	2.26	0.019	
Xenopus (Silurana) tropicalis Zic family member 3 (zic3) mRNA	AB014461.1	2	5.17	1.38	0.019	
Xenopus laevis protein tyrosine kinase (G67) mRNA partial cds	NM_001092895.1	12.06	28.01	1.16	0.019	
Xenopus laevis prostaglandin D2 synthase 21Da (lbrain) (ptgsd-a) mRNA	NM_001083994.1	0.01	1136.15	20.12	0.020	
Xenopus laevis taurine domain containing 1 (ctnd1) mRNA	XM_002935803.2	1.93	4.67	1.28	0.020	
PREDICTED: Xenopus (Silurana) tropicalis glucuronidase beta (glusb) mRNA	NM_203986.1	8.43	17.06	1.02	0.021	
Xenopus laevis polymerase F 39 kDa (polf3) mRNA	NM_00108033.1	6.32	21.2	1.75	0.021	
Xenopus laevis dihydroprymidine-4-dapsylate cyclase 1 (dapsy1) mRNA	NM_001091778.1	1.48	4.65	1.65	0.021	

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO <sup>3</sup>	FC <sup>4</sup>	q-value
Xenopus laevis homeobox B' (hoxb7.6) mRNA	NM_001096491	0.06	1.38	4.91	0.022	
Xenopus (Silurana) tropicalis enjoyl CoA hydrolase domain containing 2 (echdc2) mRNA	NM_001113907.1	0.71	1.56	1.13	0.022	
Xenopus laevis hypothetical protein LOC43704 mRNA (cDNA clone IMAGE6945618) partial cds	BC07422.1	1.52	7.93	2.38	0.023	
Xenopus laevis smad anchor for receptor activation (SARA) mRNA (complete cds)	AF104305.1	0.42	1.12	1.43	0.023	
Xenopus laevis sp1-like zinc-finger protein KSPR-2 mRNA (cDNA clone IMAGE5154993) partial cds	BC072763.1	6.88	14.68	1.09	0.023	
Xenopus laevis n-myc downstream regulated 1 (nrdg1-a) mRNA	NM_001094390.1	0.41	3.88	3.26	0.024	
Xenopus laevis forkhead box H1 gene 2 (foxh1.2) mRNA	NM_001113907.1	0.81	4.08	2.35	0.024	
Xenopus laevis transmembrane protein 60 (tmem60) mRNA	NM_001094207.1	2.64	38.52	3.87	0.026	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101732820 (LOC101732820) mRNA	XM_004918288.1	9.6	50.77	2.4	0.027	
Xenopus laevis heat shock protein mRNA (cDNA clone MGCS3330) (IMAGE5084801) complete cds	BC078115.1	4.59	12.4	1.43	0.027	
Xenopus laevis uncharacterized LOC1000368327 (LOC1000368327) mRNA	NM_001097125.1	105.07	613.23	2.55	0.029	
Xenopus laevis spondin 2 (Rsp2) mRNA complete cds	AY753198.1	0.28	4.6	2.38	0.029	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100485964 (LOC100485964) transcript variant X2 mRNA	XM_004917879.1	1	3.31	1.73	0.029	
Xenopus laevis hypothetical protein MGCS4450 mRNA (cDNA clone IMAGE5073620) partial cds	BC07509.1	2.68	6.51	1.28	0.029	
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor 151 (gpr151) mRNA	XM_002935277.2	0.07	1.43	4.37	0.030	
PREDICTED: Xenopus (Silurana) tropicalis mucin-5c-like (LOC100493765) mRNA	XM_002936037.2	0.14	1.33	3.34	0.030	
Xenopus laevis p7.5-like transmembrane protein fullback mRNA complete cds	AF131890.1	2.29	5.17	1.18	0.030	
Xenopus laevis tropomyosin finished cDNA clone TNel026k19	CR760307.2	2.43	7.27	1.58	0.031	
Xenopus laevis brachyury (T) mRNA (cDNA clone MGCT79815) (IMAGE3402478) complete cds	BC072031.1	0.92	3.64	1.99	0.032	
Xenopus laevis ankyrin anchoring protein MGCS4450 mRNA	NM_001086321.1	2.38	6.08	1.35	0.032	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730720 (LOC101730720) partial mRNA	XM_004920246.1	2.2	12.39	2.5	0.033	
Xenopus laevis p5'-nucleotidase cytosolic IIIA (nt5cA) mRNA	NM_001094563.1	0.93	3.26	1.83	0.033	
Xenopus laevis purinergic receptor P2Y G-protein coupled 10 (p2ry10) mRNA	NM_001094846.1	0.32	3.27	2.87	0.034	
Xenopus laevis DNA ligase III isofrom alpha mRNA (cDNA clone MGCT79845) (IMAGE3409226) complete cds	BC070993.1	5	39.11	2.78	0.034	
Xenopus laevis tropomyosin finished cDNA clone TEG0037/p04	CU075595.1	0.45	1.23	1.46	0.035	
Xenopus laevis uncharacterized protein MGCI15510 (MGCI15510) mRNA	NM_001096122.1	0.08	1.2	3.97	0.036	
PREDICTED: Xenopus (Silurana) tropicalis SET and MYND domain containing 3 (smyd3) transcript variant X2 mRNA	XM_004914626.1	0.32	2.99	3.23	0.036	
PREDICTED: Xenopus (Silurana) tropicalis carcinobionic antigen-related cell adhesion molecule 8-like (LOC100493418) partial mRNA	XM_0029424592	99.48	222.69	1.16	0.036	
Xenopus laevis mRNA for glucokinase	X93494.1	0.28	1.23	2.13	0.038	
Xenopus laevis hypothetical protein LOC14512 mRNA (cDNA clone IMAGE30528) partial cds	BC068857.1	846.67	3065.56	1.86	0.038	
X.laevis mRNA for cpl-1	X84414.1	0.24	5.41	4.52	0.038	
Xenopus laevis Brachyury and Tbx related protein mRNA (cDNA clone IMAGE8328112) partial cds	BC130162.1	0.35	1.42	2.04	0.038	
Xenopus laevis NiNA-related kinase 3 (nek3) mRNA	NM_001091706.1	0.41	1.64	2.04	0.038	
Xenopus laevis sDNA clone IMAGET3397054	BC079827.1	16.33	58.98	1.85	0.038	
Xenopus laevis tordin mRNA (cDNA clone MGCT79971) (IMAGE5161617) complete cds	BC077767.1	0.59	1.91	1.69	0.038	
Xenopus laevis gap junction protein beta 2 (gap2) mRNA	NM_001087009.1	1.42	3.32	1.32	0.039	
Xenopus laevis UPF688 protein C1orf74 homolog (MGCI15548) mRNA	BC077767.1	3.55	157.54	2.15	0.041	
Xenopus laevis hypothetical protein MGCI30860 mRNA (cDNA clone MGCI30860) (IMAGE2705580) complete cds	BC106331.1	93.36	307.66	1.72	0.041	
PREDICTED: Xenopus (Silurana) tropicalis AhNAK nucleoporin (ahnak) transcript variant X2 mRNA	XM_004919209.1	774.41	2752.23	1.83	0.042	
Xenopus laevis heat shock protein 110 (hsp110) mRNA complete cds	DQ48776.1	31.29	95.34	1.61	0.042	
PREDICTED: Xenopus (Silurana) tropicalis protein Wnt-7b-like (LOC100490426) mRNA	XM_002935496.2	3.36	9.47	1.5	0.042	
Xenopus laevis THO complex 5 (tno5c-a) mRNA	NM_001093029.1	19.17	40.06	1.06	0.042	
Xenopus laevis ankyrin repeat and SOCS box containing 1 (absl) mRNA	NM_001092027.1	1.09	3.03	1.47	0.043	
Xenopus laevis adaptin-related protein complex 5 zeta 1 subunit (apz51) mRNA	NM_001092489.1	1.42	3.18	1.16	0.043	
Xenopus laevis acyl-CoA synthetase family member 3 (acsf3) nuclear gene encoding mitochondrial protein mRNA	NM_001092845.1	0.36	1.43	1.99	0.044	
Xenopus laevis zinc finger protein 214 (znf214) mRNA	NM_001097042.1	123.05	269.53	1.13	0.044	
Xenopus laevis INF1 homeobox B (hnfb1b) mRNA	NM_001085811.1	0.34	1.21	1.84	0.045	
Xenopus laevis uncharacterized LOC100127288 (LOC100127288) mRNA	NM_001112845.1	3.60	158.43	2.14	0.045	
Xenopus laevis runt-related transcription factor 1 mRNA (cDNA clone MGCI69003) (IMAGE4963637) complete cds	BC057739.1	0.38	1.08	1.54	0.045	
PREDICTED: Xenopus (Silurana) tropicalis Down syndrome cell adhesion molecule like 1 (dsccml1) transcript variant X1 mRNA	XM_002932905.2	1.54	3.19	1.05	0.045	
Xenopus laevis lysophosphatidic acid acyltransferase-like (MGCI54408) mRNA	NM_001098633.1	0.23	1.25	2.49	0.046	
Xenopus laevis 1 DAN family BMP antagonist (cer1) mRNA	NM_001093918.1	2.22	5.07	1.19	0.046	
Xenopus laevis cerberus 1 DAN family member 1 (zic1) mRNA	NM_001088331.1	3.41	7.2	1.08	0.049	
PREDICTED: Xenopus (Silurana) tropicalis Zic family member 1 (zic1) mRNA	NM_001093918.1	0.32	1.45	2.19	0.049	
PREDICTED: Xenopus (Silurana) tropicalis theme oligopeptide 1 (thop1) mRNA	NM_001093918.1	4.31	1.24	0.049		

<sup>1</sup>Expression levels (FPKM) in un-injected pre-placodal explants

<sup>2</sup>Expression levels (FPKM) in pre-placodal explants after injection of Six1-MO

<sup>3</sup>Log<sub>2</sub> Fold change values for Six1-MO

### I.3 Eya1-MO: Down-regulated genes

**Table I.3** Genes significantly down-regulated after injection of *Eya1*-MO

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	α-value
Xenopus laevis tubulin folding cofactor E-like (tubb3) mRNA	NM_001096263.1	27.24	0.01	-14.73	0.003
Xenopus laevis cDNA clone MGCI14987 (IMAGE5084306) complete cds	BC058991.1	24.37	0.01	-14.57	0.003
PREDICTED: Xenopus (Silurana) tropicalis phospholipase A2 inhibitor and Ly6/PLAUR domain-containing protein-like (LOC101730430) mRNA	XM_001919780.1	5.27	0.01	-12.36	0.003
PREDICTED: Xenopus (Silurana) tropicalis serine protease 48-like (LOC100493951) mRNA	XM_001939691.2	5.88	0.04	-7.23	0.003
PREDICTED: Xenopus (Silurana) tropicalis otoferrin (otof) mRNA	XM_001918188.1	1.24	0.03	-5.9	0.003
Xenopus laevis G1D complex subunit 8 homolog (S. cerevisiae) (g1d8) mRNA	NM_001096765.1	377.05	7.89	-5.58	0.003
PREDICTED: Xenopus (Silurana) tropicalis L-type ion channel protein 2-like (LOC101730242) partial mRNA	XM_001919456.1	2.98	0.12	-4.73	0.003
Xenopus laevis cDNA clone MGCI10012564 mRNA (cDNA clone IMAGE7661075) partial cds	XM_001916360.1	1.27	0.07	-4.35	0.003
Xenopus laevis retinohisin 1 (rs1) mRNA	NM_001096951.1	8.19	0.44	-4.23	0.003
Xenopus tropicalis hypothetical protein LOC100127277 mRNA (cDNA clone MGCI106106 (IMAGE9041013) transcript variant X2 mRNA	BC136162.1	1.15	0.07	-4.09	0.003
XL23535: Xenopus laevis epithelial sodium channel alpha subunit (alphaENaC) mRNA complete cds	U2355.1	152.64	14.38	-3.41	0.003
Xenopus laevis XRPly3 mRNA for XRPly3 protein complete cds	AB45086.1	11.3	1.09	-3.38	0.003
Xenopus laevis subunit beta 3 class III (tubb3) mRNA	NM_001094986.1	6.16	0.6	-3.37	0.003
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain-containing protein 170-like (LOC1004948825) mRNA	XM_001923371.52	9.46	0.96	-3.31	0.003
Xenopus laevis hypothetical protein LOC100127277 mRNA (cDNA clone MGCI106106 (IMAGE9041013) complete cds	BC169379.1	23.75	26.69	-3.15	0.003
Xenopus (Silurana) tropicalis chromosome 17 open reading frame 105 (C17orf105) mRNA	NM_203999.1	5.75	0.67	-3.12	0.003
PREDICTED: Xenopus (Silurana) tropicalis ADAMTS-like 1 (adams1) mRNA	NM_001910864.1	1.81	0.23	-3.02	0.003
Xenopus laevis coiled-coil domain containing 63 (ccdc63) mRNA	NM_001090143.1	350.94	43.6	-3.01	0.003
Xenopus laevis subunit beta 3 class III (tubb3) mRNA	BC044125.1	6.41	0.85	-2.93	0.003
Xenopus laevis MGCA8752 protein (MGCA8752) mRNA	NM_001092839.1	29.13	3.89	-2.91	0.003
Xenopus laevis uncharacterized protein MGCE8948 (MGCE8948) mRNA	NM_001086386.1	8.96	1.2	-2.9	0.003
Xenopus laevis neocentri helix-loop helix 1 (nhhl1-α) mRNA	NM_001094952.1	4.68	0.63	-2.89	0.003
Xenopus (Silurana) tropicalis WD repeat domain 49 (wdr49) mRNA	NM_001272027.1	6.01	0.83	-2.86	0.003
Xenopus laevis uncharacterized protein MGCE9097 (MGCE9097) mRNA	NM_00109073.1	21.64	3.1	-2.8	0.003
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain-containing protein 105-like (LOC100487156) mRNA	NM_00192938674.2	6.78	0.99	-2.78	0.003
Xenopus laevis cDNA clone IMAGE8642610	BC153809.1	5.49	0.82	-2.75	0.003
Xenopus laevis uncharacterized protein MGCS5387 (MGCS5387) mRNA	NM_001085951.1	17.76	2.7	-2.72	0.003
Xenopus laevis corneolin gene 1 (chnl1-1) mRNA	NM_001091231.1	46.21	7.05	-2.71	0.003
Xenopus laevis hypothetical protein LOC100127287 mRNA (cDNA clone IMAGE3822377) partial cds	BC152983.1	2.17	14.12	-2.71	0.003
Xenopus laevis synaptotagmin 1-3-like (LOC100490466) mRNA	NM_001092365.112	5.36	0.82	-2.71	0.003
PREDICTED: Xenopus (Silurana) tropicalis lipoxigenase homology domain 1 (loxdh1) mRNA	XM_00194468.2	4.41	0.68	-2.7	0.003
Xenopus laevis ADP-ribosylarginine hydrolase (adrh1) mRNA	NM_001094795.1	3.35	0.53	-2.68	0.003
PREDICTED: Xenopus (Silurana) tropicalis fibroblast L-like (LOC100486760) mRNA	NM_001094232.1	21.12	3.31	-2.68	0.003
Xenopus laevis uncharacterized LOC496239 (LOC496239) mRNA	NM_00109424.1	16.16	2.52	-2.68	0.003
Xenopus laevis chromosome 9 open reading frame 9 (c9orf9) mRNA	NM_001091855.1	11.67	1.83	-2.68	0.003
PREDICTED: Xenopus (Silurana) tropicalis shaker-related subfamily member 8-like protein 1-like (LOC100494819) transcript variant X1 mRNA	NM_0019293761.2	9.63	1.52	-2.67	0.003
Xenopus laevis caveolin 1 caveole protein 22kDa mRNA (cDNA clone MGCI093805 (LOC100493805) mRNA	XM_001923195.2	5.24	0.84	-2.65	0.003
Xenopus laevis cation channel gamma 7 (gng7-al) mRNA	NM_001086621.1	41.2	6.75	-2.61	0.003
PREDICTED: Xenopus (Silurana) tropicalis circularly permuted Ras protein 1-like (LOC100491292) transcript variant X2 mRNA	NM_001094916298.1	7.74	1.29	-2.59	0.003
Xenopus laevis family B glycoprotein (thbgb) mRNA	NM_001093663.1	7.37	1.24	-2.57	0.003
Xenopus laevis protein phosphatase 1 regulatory subunit 36 (ppp136) mRNA	NM_001095219.1	12.19	2.09	-2.55	0.003
PREDICTED: Xenopus (Silurana) tropicalis voltage-gated channel receptor kinase substrate 8-like protein 1-like (LOC100494819) transcript variant X1 mRNA	NM_0019293820.2	10.14	1.73	-2.55	0.003
Xenopus laevis cation channel gamma 7 (gng7-al) mRNA	BC070672.1	14.2	2.45	-2.54	0.003
Xenopus (Silurana) tropicalis uncharacterized LOC100493805 (LOC100493805) mRNA	NM_001085727.1	12.08	2.1	-2.53	0.003
Xenopus laevis uncharacterized LOC100494171 (LOC100494171) mRNA	NM_001094201.1	11.21	1.95	-2.53	0.003
Xenopus laevis synapsin assembly factor 3 exon 1a (cncna1) mRNA	NM_001096370.1	20.36	3.62	-2.49	0.003
Xenopus (Silurana) tropicalis uncharacterized LOC100158544 (LOC100158544) mRNA	NM_00109127978.2	4.62	0.83	-2.48	0.003
Xenopus laevis potassium voltage-gated channel shaker-related subfamily member 4 (knca4) mRNA	NM_001088606.1	3.3	0.6	-2.47	0.003
PREDICTED: Xenopus (Silurana) tropicalis homeobox protein Dixa-like (LOC100494171) mRNA	NM_0010935712.2	16.07	2.93	-2.46	0.003
Xenopus (Silurana) tropicalis hyaluronoglicosaminidase 4 (hyal4) mRNA	NM_001097327.1	12.17	2.22	-2.46	0.003
Xenopus laevis ASAP (ASAP) mRNA complete cds	EU219608.1	4.47	0.82	-2.45	0.003
Xenopus laevis transcription factor Sox10 mRNA complete cds	AY149116.1	6.63	1.23	-2.43	0.003
Xenopus laevis apoptosis-inducing factor mitochondrial protein mRNA	NM_001097928.1	9.08	1.7	-2.42	0.003

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	c-value
Xenopus laevis uncharacterized protein MGCS2968 (MGCS2968) mRNA	NM_001086175.1	41.68	7.92	-2.4	0.003
Xenopus laevis hypothetical LOC95038 mRNA clone MGCS86403 (IMAGE:7011516) complete cds	BC054133.1	13.61	2.59	-2.39	0.003
Xenopus tropicalis finished cDNA clone TGas058024	CR942040.2	17.56	3.39	-2.38	0.003
Xenopus laevis tetra ricopeptide repeat domain 29 (ttc29) mRNA	NM_001096364.1	8.94	1.72	-2.38	0.003
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain containing 27 (ccdc27) mRNA	XM_002937464.2	6.1	1.18	-2.38	0.003
PREDICTED: Xenopus (Silurana) tropicalis protein FAM154A-like (LOC101731882) transcript variant X1 mRNA	NM_004916066.1	10.05	1.95	-2.37	0.003
Xenopus laevis (MGCS80975) protein (MGCS80975) mRNA	NM_001092408.1	32.52	6.34	-2.36	0.003
Xenopus laevis ATP/GTP binding protein 1 (agtpbp1) mRNA	NM_001093635.1	5.44	1.06	-2.36	0.003
PREDICTED: Xenopus (Silurana) tropicalis tubulin alpha 1b (tuba1b) transcript variant 1 mRNA	XM_002935108.2	14.2	2.76	-2.36	0.003
Xenopus laevis sCAT2 mRNA for calcium transporter 2 complete cds	AB085631.1	20.33	4.07	-2.32	0.003
Xenopus laevis cDNA clone IMAGE:4201936	BC086460.1	16.19	3.24	-2.32	0.003
Xenopus laevis sodium channel nonvoltage-gated 1 gamma (scn1g) mRNA	NM_001096984.1	3.05	0.62	-2.3	0.003
Xenopus laevis ribosome binding protein 1 (rbp1) mRNA	NM_001089623.1	58.86	12	-2.29	0.003
Xenopus laevis TMEV 6s mRNA complete cds	EU367938.1	3.59	0.75	-2.27	0.003
Xenopus laevis tico01 mRNA complete sequence	DQ069916.1	514.45	106.56	-2.27	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100487159 (LOC100487159) transcript variant X2 mRNA	XM_009181581	5.81	1.22	-2.25	0.003
Xenopus laevis (MGES5948275) **** WARNING: chimeric clone ***	XM_002938017.2	6.42	1.37	-2.24	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100487159 (LOC100487159) transcript variant X1 mRNA	NM_001096223.1	24.85	5.34	-2.22	0.003
Xenopus laevis stomatin (EPB72)-like 3 (stom3) mRNA	NM_001094136.1	11.66	2.51	-2.22	0.003
Xenopus laevis TMEV 6s mRNA complete cds	XM_00914518.1	4.48	0.96	-2.22	0.003
Xenopus laevis putative transmembrane BAX inhibitor motif containing 1 (tbabim1) mRNA	BC059525.1	3.93	0.85	-2.21	0.003
PREDICTED: Xenopus (Silurana) tropicalis thymus brain and testes associated (tbata) mRNA	NM_001094993.1	8	1.74	-2.2	0.003
Xenopus laevis glutathione S-transferase omega 1 (gsto1) mRNA	XM_002939725.2	28.8	6.3	-2.19	0.003
Xenopus laevis epoxide hydrolase 2 mRNA complete sequence	NM_001095036.1	24.33	5.22	-2.19	0.003
PREDICTED: Xenopus (Silurana) tropicalis transcript variant 78 (ccdc78) mRNA	DQ068899.1	2463.46	544.49	-2.18	0.003
Xenopus laevis myosin VI (myo6) transcript variant X5 mRNA	XM_00916291.1	5.16	1.15	-2.17	0.003
Xenopus laevis putative transmembrane protein TA-2 mRNA complete cds	AF310008.1	737.17	165.26	-2.16	0.003
Xenopus laevis putative transmembrane protein TA-2 mRNA	NM_001097296.1	7.02	1.57	-2.16	0.003
PREDICTED: Xenopus (Silurana) tropicalis transmembrane BAX inhibitor motif containing 1 (tbabim1) mRNA	XM_0029379872	8.71	1.96	-2.15	0.003
Xenopus laevis glutathione S-transferase omega 1 (gsto1) mRNA	XM_002932210.2	7.57	1.72	-2.14	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731919 (LOC101731919) mRNA	AF343894.1	7.64	1.73	-2.14	0.003
Xenopus laevis putative transmembrane protein TA-2 mRNA	XM_0029331362	6.82	1.55	-2.14	0.003
Xenopus laevis putative transmembrane protein TA-2 mRNA	NM_001096217.1	40.65	9.23	-2.14	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC00497800 (LOC00497800) transcript variant 1 mRNA	NM_001097083.1	7.18	1.65	-2.13	0.003
Xenopus laevis putative transmembrane channel-like 5 (tnnc5) mRNA	BC059045.1	20.07	4.58	-2.13	0.003
Xenopus laevis chisel (CsI) mRNA complete cds	NM_001097405.1	3.95	0.92	-2.11	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100494322 (LOC100494322) mRNA	BC059919.1	290.36	67.23	-2.11	0.003
Xenopus laevis sperm associated antigen 6 (spag6) mRNA	NM_001095293.1	19.25	4.52	-2.09	0.003
Xenopus laevis coiled-coil domain containing 176 (ccdc176) mRNA	NM_001096217.1	22.81	5.36	-2.09	0.003
Xenopus laevis interleukin 1 receptor accessory protein (illtrap) mRNA	AF45044.1	151.8	36.03	-2.08	0.003
Xenopus laevis cDNA clone IMAGE:3749049	NM_001088829.1	12.75	3.05	-2.07	0.003
Xenopus (Silurana) tropicalis sepiotin 5 (sept5) mRNA	NM_001097043.1	7.88	1.88	-2.07	0.003
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain containing 83 (ccdc83) mRNA	XM_002937246.2	18.25	4.27	-2.06	0.003
Xenopus laevis adenylyl kinase 1 (ak1-1b) mRNA	NM_001094214.1	67.12	16.11	-2.06	0.003
PREDICTED: Xenopus (Silurana) tropicalis lymphocyte antigen 6E-like (LOC101732565) mRNA	XM_009198881	46.66	11.29	-2.05	0.003
Xenopus laevis intracellular transport 74 homolog (ittf4) mRNA	NM_00142661.1	27.61	6.65	-2.05	0.003
PREDICTED: Xenopus (Silurana) tropicalis radial spoke head 10 homolog B (Chlamydomonas) (rsph10b) transcript variant X2 mRNA	NM_001095800.1	13.66	3.31	-2.05	0.003
Xenopus laevis testis derived transcript 3 (LIM domains) (tes) mRNA	NM_0010971978.1	8.57	2.07	-2.05	0.003
Xenopus laevis sRF protein kinase 3 (spk3) mRNA	NM_001092877.1	11.12	2.79	-2	0.003
PREDICTED: Xenopus (Silurana) tropicalis centrosomal protein 1 (cep128) transcript variant X1 mRNA	BC044693.1	8.57	2.17	-1.99	0.003
Xenopus laevis sepiotin 3 (tek3) mRNA	NM_001112874.1	10.43	2.7	-1.95	0.003
PREDICTED: Xenopus (Silurana) tropicalis neural cell adhesion molecule L1-like protein-like (LOC101735183) mRNA	NM_001094305.1	10.56	2.74	-1.95	0.003
Xenopus laevis calactin I (annexin II) heavy chain mRNA (CDNA clone MGCS54013 IMAGE:5571501) complete cds	NM_001096459.1	34.58	9.08	-1.93	0.003
Xenopus laevis family with sequence similarity 166 member B (fam166b) mRNA	NM_001092877.2	83.11	22.07	-1.91	0.003
PREDICTED: Xenopus (Silurana) tropicalis centromere protein J-like (LOC100430305) mRNA	NM_002935827.2	42.81	11.46	-1.9	0.003
Xenopus (Silurana) tropicalis myocardial zonula adherens gene (myzpz) mRNA	NM_001113933.2	9.55	2.58	-1.89	0.003

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	c-value
Xenopus laevis cDNA clone IMAGE:6880181	BC058326.1	15.89	4.29	-1.89	0.003
Xenopus tropicalis finished cDNA clone Thel057d16	CR926199.1	18.22	4.96	-1.88	0.003
Xenopus laevis adenylyl kinase 7 mRNA (cDNA clone MGCI-98814 IMAGE:3400314) complete cds	BC089169.1	28.5	7.82	-1.87	0.003
PREDICTED: Xenopus (Silurana) tropicalis tetraacetyl repeat domain 1 8 ttc(18) transcript variant X1 mRNA	XM_002940598.1	14.7	4.05	-1.86	0.003
Xenopus laevis MORN repeat containing 2 (morn2) mRNA	NM_00171791.1	3.51	0.97	-1.86	0.003
Xenopus laevis hypothetical protein LOC100137623 mRNA (cDNA clone IMAGE:5440018) partial cds	BC142545.1	225.15	62.51	-1.85	0.003
PREDICTED: Xenopus (Silurana) tropicalis L-2-hydroxyglutarate dehydrogenase 2(hgdh) transcript variant 2 mRNA	XM_002938125.2	158.96	44.15	-1.85	0.003
Xenopus tropicalis finished cDNA clone Thel057d20	XM_002936375.2	11.79	3.32	-1.83	0.003
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain containing 170 (ccdc170) mRNA	CR926201.2	13.05	3.66	-1.83	0.003
Xenopus (Silurana) tropicalis serine/threonine kinase 33 (stxk33) mRNA	NM_001016979.2	18.61	5.22	-1.83	0.003
PREDICTED: Xenopus (Silurana) tropicalis spermatogenesis associated 18 (spata18) transcript variant X1 mRNA	XM_002932575.2	97.44	27.86	-1.81	0.003
PREDICTED: Xenopus (Silurana) tropicalis WD repeat domain 66 (wdr66) mRNA	XM_002937032.2	18.45	5.25	-1.81	0.003
PREDICTED: Xenopus (Silurana) tropicalis cysteine-rich protein 2-like (LOC101734738) mRNA	XM_002910879.1	17.47	5.04	-1.8	0.003
PREDICTED: Xenopus (Silurana) tropicalis IQ motif containing 1 (iqch) transcript variant X1 mRNA	XM_002932957.2	5.89	1.8	-1.8	0.003
Xenopus laevis adaptor-related protein complex 3 beta 1 subunit (ap3b1) mRNA	NM_001091840.1	17.11	4.94	-1.79	0.003
Xenopus laevis tektin 2 (tekt2) (tekt2) mRNA	NM_001096211.1	65.42	18.9	-1.79	0.003
PREDICTED: Xenopus (Silurana) tropicalis chromosome 6 open reading frame 165 (coorf165) mRNA	XM_002938297.2	9.68	2.8	-1.79	0.003
Xenopus laevis cDNA clone IMAGE:6316572	BC071055.1	11.08	3.23	-1.78	0.003
Xenopus laevis pyruvate dehydrogenase kinase isoform 4 (pdhk4) nuclear gene encoding mitochondrial protein mRNA	NM_001086628.1	12.05	3.54	-1.77	0.003
PREDICTED: Xenopus (Silurana) tropicalis IQ motif containing with AAA domain 1 (iqca1) mRNA	XM_002932002.2	8.95	2.63	-1.77	0.003
Xenopus laevis EF-hand domain C-terminal containing 1 (efhc1) mRNA	NM_001092940.1	42.22	12.4	-1.77	0.003
PREDICTED: Xenopus (Silurana) tropicalis dorsal inhibitory axon guidance protein (draxin) mRNA	XM_002937523.2	6.85	2.03	-1.76	0.003
Xenopus laevis secreted protein acidic cysteine-rich (osteonectin) (sparc) mRNA	NM_001086121.1	18.73	5.53	-1.76	0.003
Xenopus laevis selenocystein (scin) mRNA	NM_001092620.1	10.04	2.99	-1.75	0.003
Xenopus laevis brain protein 44-like (brp44b) mRNA	NM_00171779.1	153.83	45.66	-1.75	0.003
PREDICTED: Xenopus (Silurana) tropicalis neural precursor cell expressed developmentally down-regulated 4 E3 ubiquitin protein ligase (nedd4) transcript variant X1 mRNA	XM_002940252.2	8.89	2.67	-1.74	0.003
Xenopus laevis IgM Fc-domain containing 1 (fcfc1) mRNA	NM_001095561.1	15.05	4.52	-1.74	0.003
PREDICTED: Xenopus (Silurana) ADP ribosylation factor GAP protein (kap3) mRNA	NM_001095719.1	38.8	11.82	-1.72	0.003
Xenopus laevis thymosome 4 open reading frame 47 (coorf47) mRNA	XM_002914651.1	9.05	2.76	-1.72	0.003
Xenopus laevis leucine rich repeat containing 34 (lrrc34) mRNA	NM_001094745.1	23.65	7.22	-1.71	0.003
Xenopus laevis uncharacterized protein MGCS85013 (MGCS85013) mRNA	NM_001095339.1	13.81	4.23	-1.71	0.003
Xenopus (Silurana) tropicalis spermine synthase (sms) mRNA	NM_001096519.1	30.92	9.42	-1.71	0.003
PREDICTED: Xenopus (Silurana) tropicalis adenylate kinase 9 (ak9) mRNA	NM_00258422.1	3.64	1.11	-1.71	0.003
PREDICTED: Xenopus (Silurana) tropicalis cell division cycle 14A (cdc14a) transcript variant X1 mRNA	XM_002940552.2	15.12	4.66	-1.7	0.003
Xenopus (Silurana) tropicalis tubulin alpha 4a (tub4a) mRNA	NM_002934122.2	6.37	1.97	-1.69	0.003
PREDICTED: Xenopus (Silurana) tropicalis V-type proton ATPase subunit S1-like (tnik) mRNA	NM_001114233.1	23.79	7.41	-1.68	0.003
Xenopus laevis T-box 4 (tbx4) mRNA	XM_002932982.1	15.96	5	-1.68	0.003
Xenopus laevis T-box 1 (tbx1-b) mRNA	NM_001096675.1	15.07	4.73	-1.67	0.003
Xenopus laevis kinase 4 (tkt4) mRNA	NM_001095333.1	26.97	8.56	-1.66	0.003
Xenopus laevis hypothetical protein LOC100126627 mRNA (cDNA clone IMAGE:8533789) partial cds	NM_002931550.2	6.23	1.98	-1.66	0.003
PREDICTED: Xenopus (Silurana) tropicalis tyrosine kinase 1 (tkn1) mRNA	NM_001096182.1	16.45	5.19	-1.66	0.003
PREDICTED: Xenopus (Silurana) tropicalis probable S-acetyltransferase Atsg 14255-like (LOC100498452) mRNA	XM_002932057.2	18.76	5.98	-1.65	0.003
Xenopus (Silurana) tropicalis WD repeat domain 63 (wd63) mRNA	NM_00128956.1	12.19	3.89	-1.65	0.003
Xenopus (Silurana) tropicalis synaptotagmin-like 1 (sytl) mRNA	NM_00106772.1	38.93	12.53	-1.64	0.003
PREDICTED: Xenopus (Silurana) tropicalis centrosomal protein 164Da (cep164) transcript variant X2 mRNA	NM_001095072.1	6.48	2.09	-1.64	0.003
Xenopus laevis GLI pathogenesis-related 2 (gplp2) mRNA	BC153770.1	411.59	131.62	-1.64	0.003
PREDICTED: Xenopus (Silurana) tropicalis fermitin family member 1 (fermt1) mRNA	NM_001095331.1	38.86	12.52	-1.63	0.003
Xenopus laevis spid18 mRNA complete sequence	BC080177.1	20.6	6.64	-1.63	0.003
Xenopus laevis finished cDNA clone Thel06g11	DQ06907.1	117.49	38.32	-1.62	0.003
Xenopus laevis ankyrin repeat domain protein 5 mRNA (cDNA clone IMAGE:7391599) partial cds	CR926197.2	8.64	2.84	-1.61	0.003
Xenopus laevis G protein beta 1 (gpb1) mRNA	BC108626.1	12.44	4.09	-1.61	0.003
PREDICTED: Xenopus (Silurana) tropicalis switching-B-cell complex 70kDa subunit (swap70) mRNA	NM_001095344.1	24.73	8.23	-1.59	0.003
Xenopus laevis oncogene partner (fgfr1) mRNA	NM_001095994.1	83.34	27.77	-1.59	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100497279 transcript variant X2 mRNA	NM_001095394.1	28.22	9.53	-1.57	0.003

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>MO<sup>2</sup></sup>	Eya1-MO <sup>FC<sup>3</sup></sup>	c-value
Xenopus laevis isoleucine Dipeptidase 25 domain containing protein (5R/04) (MGCI5481) mRNA	NM_001096967.1	56.04	18.86	-1.57	0.003
Xenopus (Silurana) tropicalis Daz2 interacting zinc finger protein (dzp1) mRNA	NM_001079400.1	18.16	6.12	-1.57	0.003
Xenopus laevis radial spoke head 9 homolog (rsp9) mRNA	NM_001096314.1	16.77	5.71	-1.55	0.003
Xenopus laevis similar to hypothetical protein DKTP2P43AP1735 clone IMAGE:4884518 mRNA	BC042360.1	30.65	10.54	-1.54	0.003
Xenopus (Silurana) tropicalis Ellis van Creveld syndrome (evc) mRNA	NM_001102764.1	5.51	1.9	-1.54	0.003
PREDICTED: Xenopus (Silurana) tropicalis carnitine O-palmitoyltransferase 1 liver isoform-like (LOC100490391) transcript variant X1 mRNA	XM_002942970.2	13.02	4.51	-1.53	0.003
Xenopus laevis exp44 mRNA complete sequence	DQ06861.1	155.0861	538.05	0.003	
PREDICTED: Xenopus (Silurana) tropicalis SEC24 family member B5 (S. cerevisiae) (sec24b) mRNA	XM_0029392862	11.54	3.99	-1.53	0.003
Xenopus laevis polymerase I and transcript release factor (ptrf) mRNA	NM_001087107.1	22.58	7.83	-1.53	0.003
Xenopus laevis similar to solute carrier family 27 (fatty acid transporter) member 2 mRNA (cDNA clone IMAGE:4681491) partial cds	BC041746.1	40.02	13.82	-1.53	0.003
Xenopus (Silurana) tropicalis synaptotagmin-like 1 (sytl1) mRNA	NM_001093193.1	15.3	5.38	-1.51	0.003
Xenopus laevis forkhead box protein mRNA	BC07846.1	26.21	9.23	-1.51	0.003
Xenopus laevis complete sequence	NM_001126544.1	11.55	4.07	-1.51	0.003
PREDICTED: Xenopus (Silurana) tropicalis SEC24 family member B5 (S. cerevisiae) (sec24b) mRNA	AY530633.2	8.82	3.82	-1.5	0.003
Xenopus laevis similar to solute carrier family 27 (fatty acid transporter) member 2 mRNA (cDNA clone IMAGE:4681491) partial cds	XM_002934825.2	12.74	4.54	-1.49	0.003
Xenopus laevis synaptotagmin-like 1 (sytl1) mRNA	BC055994.1	3.61	12.83	-1.49	0.003
Xenopus laevis forhead box protein mRNA	XM_002937598.2	14.26	5.08	-1.49	0.003
Xenopus laevis complete sequence	AY046070.1	7.06	2.55	-1.47	0.003
Xenopus laevis squalin mRNA	NM_001097668.1	17	6.2	-1.46	0.003
Xenopus tropicalis finished cDNA clone Tgas107921	CR835771.2	44.71	16.27	-1.46	0.003
Xenopus laevis outer dense fiber of sperm tails 3 (odf3) mRNA	NM_001085901.1	18.79	6.82	-1.46	0.003
Xenopus laevis tropomyosin family with sequence similarity 169 member A (fam169a) mRNA	NM_001091603.1	18.64	6.79	-1.46	0.003
Xenopus laevis hypothetical protein MGCG8898 mRNA (cDNA clone MGCG8898) (IMAGE:4885361) complete cds	NM_001085976.1	75.09	27.45	-1.45	0.003
Xenopus laevis tropomyosin family with sequence similarity 169 member B (fam169b) mRNA	NM_00127737.1	46.22	16.9	-1.45	0.003
Xenopus laevis complete sequence	BC041195.1	83.65	30.683	-1.45	0.003
PREDICTED: Xenopus (Silurana) tropicalis nuclear receptor subfamily 2 group F member 5-like (LOC100491959) transcript variant X1 mRNA	XM_002938463.2	86.58	31.94	-1.44	0.003
Xenopus laevis WD repeat domain 16 (wdrl6-b) mRNA	NM_0010915704.1	55.97	20.82	-1.43	0.003
Xenopus laevis claudin 1 (cdn1) mRNA	BC09955.1	83.9	31.06	-1.43	0.003
Xenopus laevis claudin 1 (cdn1) mRNA	NM_001127814.1	25.78	9.69	-1.41	0.003
Xenopus laevis uncharacterized LOC100158280 (LOC100158280) mRNA	NM_001094622.29	49.04	3.42	-1.41	0.003
Xenopus laevis squalin 1 mRNA (cDNA clone MGCG8898) (IMAGE:4881338) complete cds	LOC100498229	19.71	19.14	-1.38	0.003
PREDICTED: Xenopus (Silurana) tropicalis Golgi membrane protein 1-like (LOC100485896) transcript variant X1 mRNA	XM_002937888.2	39.76	15.27	-1.38	0.003
Xenopus laevis solute carrier family 39 (zinc transporter) member 9 (slc39a9-a) mRNA	NM_001091036.1	83.13	32.23	-1.37	0.003
Xenopus laevis inositol polyphosphate-5-phosphatase 1 mRNA	NM_001094239.1	32.18	12.5	-1.37	0.003
Xenopus laevis finished cDNA clone Tgas047b06	CR72316.2	37.36	14.58	-1.36	0.003
Xenopus laevis mRNA for PCM-1 complete cds	AB025414.1	56.61	22	-1.36	0.003
Xenopus laevis synaptotagmin-like 2 (sytl2) mRNA	NM_001094552.1	64.84	25.64	-1.34	0.003
Xenopus laevis tubulin beta class I (tubb) mRNA	NM_001087257.1	96.88	38.3	-1.34	0.003
PREDICTED: Xenopus (Silurana) tropicalis SPEC complex locus (spec) mRNA	NM_002939632.2	10.47	4.17	-1.33	0.003
Xenopus laevis mRNA for calcineurin catalytic subunit complete cds	AB037146.1	23.3	9.31	-1.32	0.003
Xenopus laevis metalloendopeptidase 14 (membrane-inserted) (mmp14) mRNA	NM_001091009.1	24.01	9.67	-1.31	0.003
Xenopus laevis hypothetical protein LLOC100158371 mRNA (cDNA clone IMAGE:4406142) partial cds	BC124931.1	16.55	6.9	-1.26	0.003
Xenopus laevis anchored LOC100158288 (LOC100158288) mRNA	NM_001271745.1	13556.4	5681.8	-1.25	0.003
PREDICTED: Xenopus (Silurana) tropicalis NPEP family member 2-like (LOC100487011) mRNA	XM_002940452.2	260.82	109.84	-1.25	0.003
Xenopus laevis Mgcs84135 protein (Mgc84135) mRNA	NM_001092726.1	26.18	11.1	-1.24	0.003
Xenopus laevis formin homology 2 domain containing 1 (fhod1) mRNA	NM_001094812.1	15.77	6.69	-1.24	0.003
Xenopus laevis retinal pigment epithelium-specific protein 65kDa (rp65) mRNA	BC124931.1	156.61	66.89	-1.23	0.003
Xenopus tropicalis novel protein similar to human angiopoietin-like ANGPTL mRNA	NM_001086800.1	106.35	46.75	-1.19	0.003
Xenopus (Silurana) tropicalis CAS1 domain containing 1 (casd1) mRNA	BC158370.1	363.33	160.95	-1.17	0.003
XL15444 Xenopus laevis beta 5 tubulin mRNA complete cds	NM_001142168.1	66.57	29.76	-1.16	0.003
Xenopus laevis ring finger protein 8 E3 ubiquitin protein ligase (rnfl8-a) mRNA	U15444.1	121.76	54.8	-1.15	0.003
Xenopus laevis adenosine kinase (adk) mRNA	NM_001086915.1	34.9	15.78	-1.15	0.003
XELO101 Xenopus laevis mRNA for pleiotrophic factor-beta 1 complete cds	NM_001092888.1	40.48	18.25	-1.15	0.003
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 160 member A1 (fam160a1) transcript variant X2 mRNA	D42059.1	173.23	81.7	-1.08	0.003
Xenopus laevis scc1249 mRNA	NM_001094553.1	17.18	8.2	-1.07	0.003
Xenopus laevis solute carrier family 12 (potassium/chloride transporters) member 9 (scl12a9) mRNA	NM_001096782.1	12.48	6.06	-1.04	0.003

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>FC<sup>2</sup></sup>	Eya1-MO <sup>FC<sup>2</sup></sup>	c-value
Xenopus laevis trecent helix-loop-helix 1 (hhhl-1) mRNA	NM_001088333.1	3.62	0.27	-3.75	0.006
PREDICTED: Xenopus (Silurana) tropicalis EP58-like 2 (ep58l2) transcript variant X1 mRNA	NM_002937505.2	10.75	1.35	-2.99	0.006
PREDICTED: Xenopus (Silurana) tropicalis leucine-rich repeats and death domain containing 1 (ldrd1) mRNA	NM_002934542	3.25	0.51	-2.67	0.006
Xenopus (Silurana) tropicalis chromosome 4 (c12orf4) mRNA	NM_001007513.1	10.9	1.9	-2.53	0.006
PREDICTED: Xenopus (Silurana) tropicalis AT-type 11A (atp11a) mRNA	NM_002933052	5.9	1.09	-2.44	0.006
Xenopus laevis hypothetical protein LOC100049756 mRNA (cDNA clone IMAGE7980299) partial cds	BC141747.1	4.94	0.97	-2.35	0.006
Xenopus laevis hypothetical protein LOC100049752 mRNA (cDNA clone IMAGE7980299) partial cds	BC153758.1	88.05	17.51	-2.33	0.006
Xenopus laevis solute carrier family 34 (sodium phosphate) member 2 (slc34a2-a) mRNA	NM_001091525.1	2.24	0.51	-2.16	0.006
Xenopus laevis matrix metallopeptidase 24 (membrane-inserted) (mmp24) mRNA	NM_001093636.1	12.17	2.75	-2.15	0.006
Xenopus (Silurana) tropicalis EF-hand domain family member B (efhb) mRNA	NM_001201467.1	8.17	1.92	-2.09	0.006
Xenopus laevis ankyrin repeat domain 13A (ankrd13a) mRNA	NM_001094574.1	4.06	1	-2.03	0.006
Xenopus laevis microfibrillar-associated protein 2 (mfap2) mRNA	NM_001086649.1	12.42	3.42	-1.86	0.006
PREDICTED: Xenopus (Silurana) tropicalis leucine-rich repeat-containing protein 45-like (LOC101730385) partial mRNA	NM_002920361.1	8.72	2.6	-1.75	0.006
Xenopus laevis leucine rich repeat containing 18 (lrc18) mRNA	NM_001095397.1	3.77	1.16	-1.71	0.006
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor 156 (gpr156) mRNA	NM_002919779.1	6.82	2.11	-1.7	0.006
Xenopus (Silurana) tropicalis coiled-coil domain containing 147 (ccdc147) mRNA	NM_001113430.1	24.2	7.52	-1.69	0.006
Xenopus (Silurana) tropicalis coiled-coil domain containing 147 (ccdc147) mRNA	NM_0012670.1	12.89	4.09	-1.66	0.006
Xenopus laevis doublecortin domain-containing protein 2-like (MGC52970) mRNA	NM_001086176.1	14.4	4.78	-1.59	0.006
Xenopus (Silurana) tropicalis Tcfex1 domain-containing 1 (ttx1-d1-b) mRNA	NM_00127759.1	18.97	6.35	-1.58	0.006
Xenopus (Silurana) tropicalis mucin-4-like (LOC100492862) mRNA	NM_001197283.1	16.97	6	-1.5	0.006
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 33 member B (fan83b) mRNA	NM_002934613.2	27.64	9.93	-1.48	0.006
Xenopus laevis sperm autoantigenic protein 17 (sap17) mRNA	NM_001920708.1	24.61	8.98	-1.45	0.006
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain-containing 173 (ccdc173) mRNA	AF549903.1	15.39	5.71	-1.43	0.006
Xenopus laevis sterile alpha motif domain-containing protein 12-like (LOC101731956) mRNA	NM_001095209.1	8.63	3.24	-1.41	0.006
Xenopus (Silurana) tropicalis RAB5B member RAS oncogene family (rab5b) mRNA	NM_001005723.1	5.07	1.94	-1.39	0.006
PREDICTED: Xenopus (Silurana) tropicalis mucin-4-like (LOC100492863) mRNA	NM_002934058.2	14.6	5.78	-1.34	0.006
Xenopus (Silurana) tropicalis T-lymphocyte surface antigen Ly-9-like (LOC100492864) mRNA	NM_002943142.2	10.21	4.22	-1.28	0.006
PREDICTED: Xenopus (Silurana) tropicalis intraflagellar transport 88 homolog (Chlamydomonas) (ift88) transcript variant X1 mRNA	NM_002937959.2	21.22	8.89	-1.26	0.006
Xenopus laevis family with sequence similarity 3 member D (fan3d) mRNA	NM_00108382.1	31.67	13.8	-1.2	0.006
Xenopus laevis glucose transporter member 12 (slc2a12) mRNA	NM_001095209.1	38.39	17.18	-1.16	0.006
Xenopus laevis similar to deoxyribonuclease I-like 3 mRNA (cDNA clone IMAGE557084) partial cds	BC043839.1	149.93	69.8	-1.1	0.006
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734576 (LOC101734576) mRNA	NM_002913174.1	3.25	0.13	-4.67	0.008
Xenopus laevis CREB binding protein (crebbp-b) mRNA	NM_001095168.1	4.53	0.86	-2.41	0.008
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 2-like (LOC101731874) transcript variant X3 mRNA	NM_004912208.1	4.81	0.98	-2.29	0.008
Xenopus laevis S10.5-A10 mRNA sequence	NM_001095209.1	3.61	0.82	-2.15	0.008
PREDICTED: Xenopus (Silurana) tropicalis arylsulphatase transporter member 1 (atsp) mRNA	NM_001096426.1	38.39	17.18	-2.06	0.008
Xenopus laevis farnesyldiphosphate synthase (fdps) mRNA	NM_001096328.1	16	3.85	-1.89	0.008
PREDICTED: Xenopus (Silurana) tropicalis leucine-rich repeats and guanylate kinase domain-containing (lrguk) mRNA	NM_001096644.1	10.82	2.93	-1.84	0.008
Xenopus laevis cyclin A1 mRNA (cDNA clone MGC81786 IMAGE886659) complete cds	NM_002931868.2	7.39	2.07	-1.83	0.008
Xenopus laevis secreted protein ethmin mRNA (cDNA clone MGC81786 IMAGE886659) complete cds	BC074115.1	11.07	3.11	-1.83	0.008
Xenopus laevis cdc25 phosphatase (cdc25d) mRNA	NM_002939659.2	12.68	3.58	-1.83	0.008
Xenopus laevis MORN repeat-containing 3 (morn3) mRNA	BC160753.1	6.8	1.99	-1.77	0.008
PREDICTED: Xenopus (Silurana) tropicalis sperm flagellar 2 (spf2) transcript variant X1 mRNA	NM_002939572	5.92	2.07	-1.52	0.008
Xenopus (Silurana) tropicalis uncharacterized LOC101733661 (LOC101733661) mRNA	NM_001094548.1	4.49	1.34	-1.49	0.008
Xenopus laevis transmembrane protein TA-2-like mRNA (cDNA clone MGC196571 IMAGE9041478) complete cds	BC169844.1	8.7	3.1	-1.45	0.008
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain-containing protein 17-like (LOC101733785) mRNA	NM_004910910.1	5.53	2.11	-1.39	0.008
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine-protein kinase MRCK alpha-like (LOC100497398) transcript variant X2 mRNA	NM_004913655.1	10.65	4.69	-1.18	0.008
Xenopus laevis calponin 2 mRNA (cDNA clone MGC53226 IMAGE557073) complete cds	BC046257.1	103.1	47.49	-1.12	0.008
PREDICTED: Xenopus (Silurana) tropicalis histone H2A type 1-like (LOC101733262) transcript variant X1 mRNA	NM_004915788.1	1.56	0.01	-10.16	0.010
Xenopus tropicalis finished cDNA clone The120e6	NM_002935802	1.79	0.17	-3.42	0.010
Xenopus laevis dimethylarginine dimethylaminohydrolase 2 (ddah2) mRNA	CU025070.1	15.8	0.83	-2.4	0.010
Xenopus laevis chromosome 11 open reading frame 65 (c11orf65) mRNA	NM_001094280.1	4.38	0.75	-2.28	0.010
Xenopus laevis chromosome 11 open reading frame 65 (c11orf65) mRNA	NM_001095354.1	3.61	0.75	-2.28	0.010

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	c-value
Xenopus laevis uncharacterized protein MGCT5428 (MGCT5428) mRNA	NM_001968391	6.37	1.48	-2.11	0.010
Xenopus (Silurana) tropicalis armadillo repeat containing 3 (armc3) mRNA	NM_001001246.1	17.57	4.55	-1.95	0.010
Xenopus (Silurana) tropicalis tumor suppressor candidate 5 (tusc5) mRNA	NM_001005033.1	6.92	2.11	-1.72	0.010
Xenopus laevis sepin 8 (sep18-b) mRNA	NM_001091489.1	21.22	6.75	-1.65	0.010
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor 37 (endothelin receptor type B-like) (grp37) mRNA	XM_002941831.2	4.17	1.36	-1.62	0.010
Xenopus (Silurana) tropicalis inhibitor of DNA binding 4 dominant negative helix-loop-helix protein (id4) mRNA	NM_001004859.1	6.41	2.1	-1.61	0.010
Xenopus laevis dihydropyrimidinease-like 3 mRNA (CDNA clone MGCG5258) (IMAGE:4930151) complete cds	BC046636.1	9.23	3.05	-1.6	0.010
PREDICTED: Xenopus (Silurana) tropicalis WD repeat domain 38 (wdr38) mRNA	XM_002941647.2	10.81	3.6	-1.59	0.010
Xenopus laevis hypothetical protein LOC43692 mRNA (CDNA clone IMAGE:5085483) partial cds	BC073734.1	6.04	2.04	-1.57	0.010
Xenopus laevis sperm-tail PG-rich repeat containing 1 (spol1) mRNA	NM_001096224.1	5.24	1.82	-1.53	0.010
PREDICTED: Xenopus (Silurana) tropicalis ortholog KIAA1217 ortholog (kiaa1217) transcript variant X6 mRNA	XM_001091543.91	4.61	1.64	-1.49	0.010
Xenopus laevis histone family member Y2 (h2ay2) mRNA	NM_001087412.1	7.21	2.63	-1.46	0.010
Xenopus laevis UDP-Gal:betaGalNAc beta 1,4-galactosyltransferase polypeptide 4 (b4galta4) mRNA	NM_001092811.1	8.24	3.2	-1.37	0.010
Xenopus laevis thopillin associated tail protein 1-like (tropn1) mRNA	NM_001096022.1	17.44	6.8	-1.36	0.010
Xenopus laevis radial spoke head 1 homolog (rsph1) mRNA	NM_001095320.1	37.27	15.54	-1.26	0.010
Xenopus laevis cref01 mRNA complete sequence	DQ06911.1	32.664	139.09	-1.23	0.010
Xenopus laevis protein disulfide isomerase family A member 4 (pdia4) mRNA	NM_001094982.1	80.24	34.4	-1.22	0.010
Xenopus laevis mRNA for xSox7 protein complete cds	D83649.1	33.82	14.61	-1.21	0.010
Xenopus laevis adenosylhomocysteinase mRNA (CDNA clone MGCG58782) (IMAGE:4201976) complete cds	BC060432.1	43.47	19.07	-1.19	0.010
Xenopus laevis solute carrier family 16 member 12 (monocarboxylic acid transporter 12) (slc6a12) mRNA	NM_001092655.1	71.66	33.82	-1.08	0.010
Xenopus laevis cytochrome P450 family 2 subfamily J polypeptide 2 (cyb2j2) mRNA	NM_00127832.1	1.59	0.26	-2.64	0.011
PREDICTED: Xenopus (Silurana) tropicalis zinc finger B-box domain containing (zbbox) mRNA	NM_002931565.2	8.95	1.85	-2.27	0.011
Xenopus laevis microsomal glutathione S-transferase 1 (most1) mRNA	NM_001091251.1	4.83	1.27	-1.93	0.011
PREDICTED: Xenopus laevis Enolase-like protein ENSP00000345555 homolog mRNA (CDNA clone IMAGE:7202693) partial cds	BC157741.1	8.38	2.31	-1.86	0.011
Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C8orf34 (LOC100492224) transcript variant X1 mRNA	NM_002935332.2	8.11	2.5	-1.7	0.011
Xenopus laevis solute carrier family 16 member 12 (monocarboxylic acid transporter 12) (slc6a12) mRNA	NM_002938880.2	5.28	1.63	-1.7	0.011
Xenopus laevis cytochrome P450 family 2 subfamily J polypeptide 2 (cyb2j2) mRNA	NM_001096041.1	20	6.28	-1.67	0.011
PREDICTED: Xenopus (Silurana) tropicalis cAMP responsive element modulator (crem) transcript variant X1 mRNA	NM_002935162.2	4.36	1.4	-1.64	0.011
Xenopus laevis apoptosis-inducing factor mitochondrial-associated 3 (aimf3) nuclear gene encoding mitochondrial protein mRNA	NM_001087560.1	2.24	0.73	-1.62	0.011
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 385A (znf385a) transcript variant X5 mRNA	NM_004911926.1	3.35	1.15	-1.54	0.011
Xenopus laevis aldo-keto reductase family 1 member C2 (akr1c2-b) mRNA	NM_001086099.1	37.84	13.45	-1.49	0.011
Xenopus laevis family 1 midline 1 (MD1) mRNA complete cds	GU32929.1	5.18	1.98	-1.39	0.011
Xenopus laevis family with sequence similarity 15 (member B) (fam154b) mRNA	NM_001093710.1	25.21	9.64	-1.39	0.011
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100490177 (LOC100490177) mRNA	NM_002931626.2	3.79	1.46	-1.38	0.011
Xenopus laevis adenylate kinase 3 (ak3) nuclear gene encoding mitochondrial protein mRNA	NM_001095997.1	6.04	2.43	-1.32	0.011
Xenopus (Silurana) tropicalis cold shock RNA binding (csdc2) mRNA	NM_0010100248.1	4.47	1.88	-1.25	0.011
Xenopus laevis uncharacterized LOC100158314 (LOC100158314) mRNA	NM_00127766.1	119.37	53.3	-1.16	0.011
Xenopus laevis reelin mRNA (CDNA clone MGCI5453; IMAGE:8320610) complete cds	BC123265.1	78.09	35.84	-1.12	0.011
Xenopus laevis selenine palmitoyltransferase long chain base subunit 1 (spcl1) mRNA	NM_001091494.1	49.08	23.31	-1.07	0.011
Xenopus (Silurana) tropicalis SRY (sex determining region Y-box 10) (sox10) mRNA	NM_00100221.1	3.48	0.81	-2.11	0.013
Xenopus laevis homeobox protein Xhox11.2 mRNA complete cds	AF283693.1	7.26	1.75	-2.06	0.013
Xenopus laevis phosphatidylinositol transfer protein beta (pitpnb2) mRNA	NM_001093495.1	7.79	1.93	-2.02	0.013
PREDICTED: Xenopus (Silurana) tropicalis leucine rich repeat containing 36 (lrrc36) mRNA	NM_002931622.2	16.35	7.14	-1.2	0.013
Xenopus laevis 19S rRNA (MGCG81939) mRNA	NM_001092608.1	19.25	4.88	-1.98	0.013
Xenopus laevis uncharacterized protein MGCG53997 (MGCG53997) mRNA	NM_001095457.1	5.05	1.39	-1.86	0.013
Xenopus laevis solute carrier family 7 (amino acid transporter light chain system) member 7 (slc7a7) mRNA	NM_001092109.1	50.48	17.91	-1.49	0.013
Xenopus laevis 5-hydroxytryptamine (serotonin) receptor 3A ionotropic (htt3a) mRNA	NM_001086077.1	13.48	3.73	-1.34	0.013
PREDICTED: Xenopus (Silurana) tropicalis transient receptor potential cation channel subfamily V member 6-like (LOC100492412) transcript variant X1 mRNA	NM_002931622.2	16.35	7.73	-1.25	0.013
Xenopus laevis nucleophosmin 1 (nucleophosmin) mRNA	BC046493.1	105.24	22.64	-2.22	0.015
Xenopus laevis nucleophosmin 1 (nucleophosmin) mRNA	NM_002939651.1	7.77	1.71	-2.18	0.015
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human Clorf194 (LOC100487688) transcript variant X1 mRNA	NM_001086139.1	2.22	0.52	-2.12	0.015
Xenopus laevis 3-O-sulfotransferase 4 (gal3t4) mRNA	NM_002935587.2	6.74	1.74	-1.96	0.015
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C9orf135 (LOC100485682) mRNA	NM_002933438.2	9.39	2.57	-1.87	0.015

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	c-value
Xenopus laevis testis expressed 26 (tex26) mRNA	NM_001052526.1	13.1	3.75	-1.81	0.015
PREDICTED: Xenopus (Silurana) tropicalis growth factor receptor-bound protein 7 (grb7) transcript variant X2 mRNA	XM_004918677.1	8.04	2.48	-1.7	0.015
Xenopus (Silurana) tropicalis sodium channel and clathrin linker 1 (scnl1) transcript variant 1 mRNA	NM_001199930.1	6.46	2.16	-1.58	0.015
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain containing 17 (ccdc17) mRNA	XM_002931598.2	11.71	3.94	-1.57	0.015
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 116-like (LOC10497315) mRNA	XM_002940124.2	10.52	3.84	-1.46	0.015
Xenopus laevis midosis expressed gene 1 homolog (mieg1) mRNA	NM_001171686.1	39.21	14.69	-1.42	0.015
Xenopus laevis uncharacterized protein MGCI54492 (MGCI54492) mRNA	NM_001096895.1	22.18	8.4	-1.4	0.015
PREDICTED: Xenopus (Silurana) tropicalis dynein assembly factor 1 axoneme-like (LOC101730543) mRNA	XM_004913556.1	10.83	4.43	-1.3	0.015
PREDICTED: Xenopus (Silurana) tropicalis actin filament associated protein 1-like 1 (afap1l1) mRNA	XM_00293106.2	14.56	5.96	-1.29	0.015
PREDICTED: Xenopus (Silurana) tropicalis slit homolog 2 (Drosophila) (slit2) transcript variant XI mRNA	XM_002936715.2	18.85	8.62	-1.13	0.015
Xenopus laevis tubulin tyrosine ligase-like family member 7 (tthal7) mRNA	NM_001092004.1	1.61	0.3	-2.47	0.016
PREDICTED: Xenopus (Silurana) tropicalis Rho GTPase activating protein 2 (arhgap28) transcript variant X1 mRNA	XM_002938054.2	2.59	0.51	-2.37	0.016
PREDICTED: Xenopus (Silurana) tropicalis EPH receptor B1 (lephb1) mRNA	XM_002935075.2	2.87	0.71	-2.03	0.016
Xenopus laevis hypothetical protein LOC133236 mRNA clone IMAGE:4681515 partial cds	BC094440.1	10.09	2.58	-1.97	0.016
PREDICTED: Xenopus (Silurana) tropicalis von Willebrand factor A domain containing 3B (wva3b) mRNA	XM_002931645.2	3.82	1.01	-1.92	0.016
XLU38225: Xenopus laevis glutamic acid decarboxylase (GAD) mRNA; complete cds	U38225.1	2.15	0.62	-1.79	0.016
Xenopus laevis mRNA for fibronectin leucine rich transmembrane protein 3 (flrt3) gene	AL65776.1	5.51	1.82	-1.6	0.016
PREDICTED: Xenopus (Silurana) tropicalis RGPBP1-like (rgrip11) mRNA	XM_002935372.2	4.11	1.57	-1.4	0.016
Xenopus laevis phosphoglucomutase 3 (pgm3) mRNA	NM_001087415.1	23.11	9.2	-1.33	0.016
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100495307 (LOC100495307) mRNA	XM_002933809.2	6.39	2.56	-1.32	0.016
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100495308 (LOC100495308) mRNA	XM_002937332.2	6.29	2.61	-1.27	0.016
Xenopus (Silurana) tropicalis erythrocyte membrane protein band 1-like 4A (ebp4l4a) mRNA	NM_00106909.1	12.76	5.32	-1.26	0.016
Xenopus laevis solute carrier family 37 (glucose-6-phosphate transporter) member 4 (slc37a4) mRNA	NM_001089935.1	26.57	11.27	-1.24	0.016
Xenopus laevis mRNA for Na <sup>+</sup> /H <sup>+</sup> antiporter	Y08635.1	22.95	11.38	-1.01	0.016
PREDICTED: Xenopus (Silurana) tropicalis lysophosphatidic acid receptor 3 (lpqr3) mRNA	XM_002931673.2	3.37	0.45	-2.92	0.018
PREDICTED: Xenopus (Silurana) tropicalis cyclin-dependent kinase-like 5 (cdkl5) transcript variant X2 mRNA	XM_004911758.1	4.18	0.82	-2.36	0.018
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain-containing protein 162-like (LOC100496491) mRNA	XM_002940549.2	2.33	0.51	-2.19	0.018
Xenopus laevis chromosome 1 open reading frame 158 (clf158) mRNA	NM_001095298.1	12.35	3.14	-1.98	0.018
PREDICTED: Xenopus (Silurana) tropicalis putative uncharacterized protein C16orf6 homolog (LOC101734550) partial mRNA	XM_002930946.1	9.66	2.64	-1.87	0.018
Xenopus laevis glutathione S-transferase theta 1 (gstt1) mRNA	NM_001091734.1	25.63	7.91	-1.7	0.018
Xenopus laevis monoglyceride lipase (mgll) mRNA	NM_001094434.1	6.39	2.09	-1.62	0.018
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733381 (LOC101733381) mRNA	XM_004918233.1	8.76	3.02	-1.54	0.018
Xenopus laevis tetratricopeptide repeat domain 26 (ttc26) mRNA	NM_001091757.1	48.92	16.84	-1.54	0.018
PREDICTED: Xenopus (Silurana) tropicalis EF-hand calcium binding domain 2 (efcab2) mRNA	XM_002931483.2	8.21	2.83	-1.54	0.018
Xenopus laevis uncharacterized LOC496226 (LOC496226) mRNA	NM_001095413.1	12.79	5.04	-1.34	0.018
Xenopus laevis RIB4A domain with coiled-coils 2 (ribc2) mRNA	NM_001096247.1	9.56	3.79	-1.34	0.018
PREDICTED: Xenopus (Silurana) tropicalis proprotein convertase subtilisin/kexin type 5-like (LOC100486641) mRNA	XM_002937893.2	3.77	1.56	-1.28	0.018
Xenopus (Silurana) tropicalis tetratricopeptide repeat domain 26 (ttc26) mRNA	NM_001092931.1	19.81	8.36	-1.24	0.018
PREDICTED: Xenopus (Silurana) tropicalis MORN repeat containing 1 (morn1) mRNA	XM_002936560.2	5	0.84	-2.59	0.019
Xenopus laevis Fk506 binding protein 7 (fbp7) mRNA	NM_00142951.1	3.28	0.68	-2.27	0.019
PREDICTED: Xenopus (Silurana) tropicalis WD repeat domain 93 (wd93) mRNA	XM_002933236.2	5.13	1.25	-2.05	0.019
Xenopus laevis hypothetical protein LOC133411 mRNA cDNA clone IMAGE:7210814 partial cds	BC108620.1	5.63	1.53	-1.88	0.019
Xenopus (Silurana) tropicalis WNK lysine deficient protein kinase 4 (wnk4) mRNA	XM_002932519.2	4.77	1.52	-1.66	0.019
PREDICTED: Xenopus (Silurana) tropicalis kinase-like protein KIF9-like (LOC100497230) mRNA	BC090251.1	9.02	4.51	-1	0.019
Xenopus laevis HIG1 hypoxia inducible domain family member 1A (higd1-a) mRNA	NM_001093393.1	48.78	16.91	-1.53	0.019
Xenopus laevis selenocysteotide pyrophosphatase/phosphodiesterase 4 (putative) (lenpp4) mRNA	NM_001090619.1	2.79	0.99	-1.5	0.019
Xenopus (Silurana) tropicalis intrafagellar transport 122 homolog (ift122) mRNA	NM_001113021.1	11.26	4.69	-1.26	0.019
Xenopus laevis uncharacterized LOC100137635 (LOC100137635) mRNA	NM_001114785.1	529.14	227.89	-1.22	0.019
Xenopus laevis hypothetical protein LOC733171 mRNA cDNA clone IMAGE:4057139 partial cds	BC090251.1	9.02	4.51	-1	0.019
Xenopus laevis Janus kinase 1 (a protein tyrosine kinase) (jak1) mRNA	NM_001093393.1	8.2	1.46	-2.49	0.020
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100488736 (LOC100488736) transcript variant X2 mRNA	XM_004919725.1	66.1	15	-2.14	0.020
PREDICTED: Xenopus (Silurana) tropicalis abhydrolase domain-containing protein 15-like (LOC100497109) mRNA	NM_002933650.2	6.93	2.13	-1.77	0.020
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100497755 (LOC100497755) mRNA	XM_004917587.1	23.1	7.21	-1.68	0.020
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100497111 (LOC100497111) mRNA	XM_002937018.2	6.24	2.08	-1.59	0.020
Xenopus laevis secreted frizzled-related protein 2 (frzrp2) mRNA	NM_001087194.1	39.84	13.77	-1.53	0.020
PREDICTED: Xenopus (Silurana) tropicalis resistin-like (LOC101732588) partial mRNA	NM_002930544.1	64.85	23.78	-1.45	0.020

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	c-value
Xenopus (Silurana) tropicalis Mef3 homeobox 2 (mef3.2) mRNA	NM_001097218.1	12.01	4.43	-1.44	0.020
PREDICTED: Xenopus (Silurana) tropicalis echinoderm microtubule associated protein-like 6 (emil6) transcript variant X1 mRNA	XM_002936920.2	14.19	5.37	-1.4	0.020
Xenopus tropicalis finished cDNA clone TNel100n17	CR760194.2	9.48	4.07	-1.22	0.020
PREDICTED: Xenopus (Silurana) tropicalis inositol-trisphosphate 3-kinase B (ipk3b) transcript variant X1 mRNA	XM_002938844.2	10.37	4.54	-1.19	0.020
PREDICTED: Xenopus (Silurana) tropicalis solute carrier family 6 (neurotransmitter transporter dopamine) member 3 (slc6a3) transcript variant X2 mRNA	XM_004915332.1	9.18	4.07	-1.17	0.020
PREDICTED: Xenopus (Silurana) tropicalis brain-specific angiogenesis inhibitor 1 (bai1) mRNA	XM_002934468.2	10.87	2.93	-1.89	0.022
PREDICTED: Xenopus (Silurana) tropicalis phosphotyrosine kinase alpha 2 (liver) (ptk2) transcript variant X2 mRNA	XM_004911663.1	12.21	4.74	-1.37	0.022
Xenopus (Silurana) tropicalis inositol-trisphosphate kinase 3 (ipk3) transcript variant X3 (GalNAc-T3) (galnt3) mRNA	BC088743.1	13	5.43	-1.26	0.022
Xenopus (Silurana) tropicalis meiosis-specific nuclear structural 1 (mnsl) mRNA	NM_00103012.1	13.37	5.81	-1.2	0.022
PREDICTED: Xenopus (Silurana) tropicalis UDP-N-acetyl-alpha-D-galactosaminyltransferase 3 (GalNAc-T3) (galnt3) mRNA	XM_002936748.2	15.23	6.85	-1.15	0.022
Xenopus (Silurana) tropicalis brain-specific angiogenesis inhibitor 1 (bai1) mRNA	NM_001091828.1	41.1	18.68	-1.14	0.022
Xenopus laevis Vip 1 interacting factor homolog B (vip1 b-a) mRNA	CR60752.1	36.22	17.14	-1.08	0.022
Xenopus tropicalis finished cDNA clone Tga5070h121	NM_001091630.1	73.09	36.45	-1	0.022
Xenopus laevis uncharacterized protein MGCT8986 (MGCT8986) mRNA	BC087515.1	13.64	5.95	-2.13	0.023
PREDICTED: Xenopus (Silurana) tropicalis claudin-4-like (LOC101732192) mRNA	XM_004911749.1	3.28	0.96	-1.78	0.023
Xenopus (Silurana) tropicalis uncharacterized LOC100127867 (LOC100127867) mRNA	NM_001113158.1	2.23	0.74	-1.61	0.023
PREDICTED: Xenopus (Silurana) tropicalis tetrastricopeptide repeat domain 24 (ttc24) mRNA	XM_002938944.2	13.66	5.29	-1.37	0.023
Xenopus laevis chromosome 15 open reading frame 26 (c15orf26) mRNA	NM_001092844.1	27.01	10.61	-1.35	0.023
Xenopus (Silurana) tropicalis myoferlin (myf1) mRNA	NM_001128651.1	25.47	10.01	-1.35	0.023
Xenopus laevis intracellular transport 57 homolog (itt57) mRNA	NM_001114796.1	11.62	4.95	-1.23	0.023
Xenopus (Silurana) tropicalis clone IMAGE:5513587 partial cds	BC085045.1	48.07	23.62	-1.03	0.023
Xenopus laevis uncharacterized protein MGCT8908 (MGCT8908) mRNA	NM_001091061.1	4421.68	17.46	-7.99	0.024
Xenopus (Silurana) tropicalis B-cell lymphoma 3 protein homolog (LOC100489600) mRNA	XM_002941035.2	1.5	0.2	-2.96	0.024
PREDICTED: Xenopus (Silurana) tropicalis onlyin-2-like (LOC100497775) mRNA	XM_002931610.2	1.38	0.32	-2.12	0.024
Xenopus tropicalis finished cDNA clone TNel124112	CT025309.2	12.69	3.71	-1.77	0.024
Xenopus laevis sigma protein mRNA (cDNA clone MGCC80311 (IMAGE:5013822) complete cds	BC072179.1	4.19	1.46	-1.52	0.024
PREDICTED: Xenopus (Silurana) tropicalis t-complex-associated-testis-expressed 1 (tte1) mRNA	XM_002942154.2	7.43	2.82	-1.4	0.024
Xenopus (Silurana) tropicalis synaptotagmin 7 (sy7) mRNA	NM_00109366.1	2.69	0.73	-1.89	0.025
PREDICTED: Xenopus (Silurana) tropicalis ATR interactiv domain 1B (swi1-like) (arid1b) transcript variant X8 mRNA	XM_004914635.1	4.05	1.27	-1.67	0.025
Xenopus laevis lobosomal protein S6 kinase 90kDa polypeptide 6 (rps6ka6) mRNA	NM_001091841.1	10.81	3.46	-1.65	0.025
Xenopus (Silurana) tropicalis transmembrane protease serine 4 (tmprss4) mRNA	NM_001045609.1	9.25	3.79	-1.29	0.025
Xenopus laevis matrix metalloproteinase 3 (stromelysin 1 progelatinase) (mmmp3) mRNA	NM_001093594.1	22.68	9.87	-1.2	0.025
Xenopus laevis GIPC PDZ-domain containing family member 2 (gipc2) mRNA	NM_001088817.1	30.54	14.3	-1.1	0.025
PREDICTED: Xenopus (Silurana) tropicalis zinc transporter member 39 (zinc transporter) member 13 (slc39a13) transcript variant X1 mRNA	NM_002942061.2	18.81	9.08	-1.05	0.025
PREDICTED: Xenopus (Silurana) tropicalis myosin VI (myo6) transcript variant X7 mRNA	XM_004914520.1	10.6	5.2	-1.03	0.025
PREDICTED: Xenoura tropicalis unknown open reading frame human C1orf53 (LOC100485377) transcript variant X2 mRNA	NM_001091596.1	87.61	14.46	-2.6	0.026
Xenopus (Silurana) tropicalis HMP-like 1 (hmp1) mRNA	XM_002939668.2	1.64	0.31	-2.42	0.026
PREDICTED: Xenopus (Silurana) tropicalis transient receptor potential cation channel subfamily M member 19 (paqr19) mRNA	NM_00104920605.1	4.12	0.95	-2.11	0.026
Xenopus (Silurana) tropicalis progesterone and adipoo receptor family member IX (paqr9) mRNA	NM_001086571.1	2.8	0.8	-1.82	0.026
Xenopus (Silurana) hypothetical protein LOC548657 mRNA (cDNA clone MGCI86534 (IMAGE:895997) complete cds	BC161440.1	18.82	6.46	-1.54	0.026
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 20 member c (fam20c) mRNA	NM_002933758.2	35.51	13.72	-1.37	0.026
Xenopus laevis suppressor of cytokine signaling 3 (soc3) mRNA	NM_001091112.1	36.91	15.21	-1.28	0.026
Xenopus laevis growth arrest-specific 6 (gas6) mRNA	NM_001089686.1	38.25	17.62	-1.12	0.026
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730819 (LOC101730819) partial mRNA	XM_002938062.2	5.42	1.79	-1.03	0.026
Xenopus (Silurana) tropicalis dehydrogenase 1 (35.5 kD) (Xn278) mRNA (cDNA clone MGCI54458 (IMAGE:8079155) complete cds	BC123216.1	7.76	3.46	-1.17	0.028
Xenopus (Silurana) tropicalis calcium binding domain 7 (lefab7) mRNA	NM_001093685.1	11.99	3.03	-1.99	0.029
PREDICTED: Xenopus (Silurana) tropicalis chromosome 1 open reading frame 222 (clorf222) mRNA	NM_001197222.1	2.12	0.33	-2.68	0.028
Xenopus laevis hypothetical protein LOC100091141 mRNA (cDNA clone IMAGE:5323207) partial cds	BC134824.1	4.83	1.55	-1.64	0.029
Xenopus tropicalis hypothetical protein LOC100158626 mRNA (cDNA clone IMAGE:6786030) partial cds	BC16350.1	9.6	3.71	-1.37	0.029
Xenopus (Silurana) tropicalis aldehyde dehydrogenase 3 family member B1 (aldd13b) mRNA	NM_001093704.1	15.94	6.9	-1.21	0.029
Xenopus (Silurana) tropicalis uncharacterized LOC100487159 (LOC100487159) transcript variant X3 mRNA	XM_002937206.2	87.37	38.98	-1.16	0.030
PREDICTED: Xenopus (Silurana) tropicalis galectin-related protein-like (LOC100492008) mRNA	XM_002940548.2	2.47	0.21	-3.56	0.030
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein FlJ37396-like (LOC100496336) mRNA	XM_002940548.2	1.38	0.21	-2.72	0.030

Gene	Accession	Control	Eya1-MO <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>a</sup>	c-value
PREDICTED: Xenopus laevis rap106 mRNA complete sequence	XM_004915978.1	1.74	0.31	-2.53	0.030	
Xenopus laevis nonboid veinlet-like 2 (rbndl2) mRNA	DQ056844.1	8.42	1.64	-2.36	0.030	
PREDICTED: Xenopus (Silurana) tropicula family with sequence similarity 92 member B (fan92b) transcript variant X2 mRNA	NM_001094890.1	3.08	0.64	-2.27	0.030	
PREDICTED: Xenopus (Silurana) tropicula zinc finger protein 474-like (LOC100498441) transcript variant X1 mRNA	NM_001094913598.1	21.82	5.97	-1.87	0.030	
Xenopus laevis uncharacterized protein MGCI5435.1 (MGCI5435.1) mRNA	NM_001096843.1	11.23	3.99	-1.49	0.030	
Xenopus (Silurana) tropicula kinase D-interacting substrate 220kDa (kidin220) mRNA	NM_001126687.1	19.98	8.21	-1.28	0.030	
PREDICTED: Xenopus (Silurana) tropicula zinc finger protein 474-like (LOC100498441) mRNA	NM_001126687.1	4.25	1.8	-1.24	0.030	
Xenopus (Silurana) tropicula uncharacterized LOC101733938 (LOC101733938) mRNA	NM_001094920692.2	7.05	3.4	-1.05	0.030	
PREDICTED: Xenopus (Silurana) tropicula uncharacterized LOC100496035 (LOC100496035) mRNA	NM_001093421.8.2	34.42	0.01	-15.07	0.031	
PREDICTED: Xenopus (Silurana) tropicula testis expressed 33 (tex33) mRNA	NM_001093421.8.2	3.41	0.48	-2.83	0.031	
Xenopus laevis cDNA clone IMAGE6875640 partial cds	BC054980.1	1.13	0.21	-2.45	0.031	
Xenopus laevis uncharacterized LOC495993 (LOC495993) mRNA	NM_001095260.1	4.64	1.64	-1.5	0.031	
Xenopus laevis lipase member H (lipph-b) mRNA	NM_001094386.1	10.45	3.99	-1.39	0.031	
PREDICTED: Xenopus (Silurana) tropicula cell division cycle 20B (cd20b) transcript variant X2 mRNA	NM_00109410356.1	14.19	5.79	-1.29	0.031	
Xenopus tropicalis cDNA clone IMAGE7679348	BC12831.1	4.96	2.04	-1.28	0.031	
Xenopus laevis inositol-tetrakisphosphate 1-kinase (itpk1) mRNA	NM_001086667.1	7.91	3.28	-1.27	0.031	
PREDICTED: Xenopus (Silurana) tropicula mitochondrial membrane (cyt5b) nuclear gene encoding mitochondrial protein mRNA	NM_001095847.1	15.14	6.6	-1.2	0.031	
Xenopus laevis cytochrome b5 type B (outer mitochondrial membrane) (cytb5) nuclear gene encoding mitochondrial protein mRNA	NM_001086091.1	8.61	3.77	-1.19	0.031	
Xenopus laevis Matrix metalloproteinase-18 mRNA cDNA clone IMAGE6393778 partial cds	BC059337.1	10.33	5.05	-1.03	0.031	
Xenopus laevis purine nucleoside phosphorylase (ppn) mRNA	NM_001086340.1	5.04	1.18	-0.99	0.032	
Xenopus laevis insect-derived growth factor-B-like protein mRNA complete cds	AY986979.1	15.6	3.74	-2.06	0.032	
Xenopus laevis coiled-coil domain containing 104 (ccdc104) mRNA	NM_001087370.1	6.83	2.23	-1.62	0.032	
PREDICTED: Xenopus (Silurana) tropicula primary ciliary dyskinesia protein 1-like (LOC100493735) partial mRNA	NM_00109430322	3.23	1.1	-1.56	0.032	
Xenopus laevis chromosome 2 open reading frame 70 (c2orf70-b) mRNA	NM_001095416.1	12.5	4.35	-1.52	0.032	
PREDICTED: Xenopus (Silurana) tropicula tetratricopeptide repeat domain 8 (ttcb) transcript variant X1 mRNA	NM_0010933198.2	4.64	1.66	-1.49	0.032	
PREDICTED: Xenopus (Silurana) tropicula uncharacterized LOC100492437 (LOC100492437) mRNA	NM_001092947.2	8.52	3.62	-1.23	0.032	
Xenopus laevis spermatogenesis associated 6-like (spata6) mRNA	NM_001096282.1	21.85	9.37	-1.22	0.032	
Xenopus laevis phosphoserine phosphatase (psph) mRNA	NM_001093025.1	9.39	4.12	-1.19	0.032	
Xenopus laevis sti11-1-like (LOC338504) mRNA	NM_001089019.1	97.23	47.82	-1.02	0.032	
PREDICTED: Xenopus (Silurana) tropicula breast cancer anti-estrogen resistance protein 3-like (LOC101731830) mRNA	NM_00109418793.1	1.51	0.44	-0.79	0.033	
Xenopus laevis synemin axonemal light chain 1 (dnal1) mRNA	NM_001094484.1	12.69	4.01	-1.66	0.033	
Xenopus laevis upstream transcription factor 1 (ustf) mRNA	NM_001096002.1	12.77	4.99	-1.36	0.033	
PREDICTED: Xenopus (Silurana) tropicula early activation antigen Cd69-like (LOC101732585) mRNA	NM_001093961.1	9.3	3.81	-1.29	0.033	
Xenopus laevis OP30 domain containing 1 mRNA (cdna clone MCIGC161067) (MAGE77396835) complete cds	BC133793.1	18.76	7.7	-1.29	0.033	
Xenopus laevis solute carrier family 25 (mitochondrial carrier citrate transporter) member 1 b (slc25a1-b) nuclear gene encoding mitochondrial protein mRNA	NM_001098709.1	14.05	6.02	-1.22	0.033	
Xenopus laevis calcium 2mRNA (cdna clone MGCI96805) (MAGE9041773) complete cds	BC170078.1	3.69	0.63	-2.56	0.034	
Xenopus laevis tkttn-2-like (MGCS2862) mRNA	NM_001085998.1	2.41	0.71	-1.77	0.034	
PREDICTED: Xenopus (Silurana) tropicula vasopressin V2 receptor-like (LOC100487051) transcript variant X2 mRNA	NM_001094916721.1	1.62	0.5	-1.71	0.034	
PREDICTED: Xenopus (Silurana) tropicula IQ motif and ubiquitin domain containing (qub) transcript variant X2 mRNA	NM_0010912993.1	13.1	5.53	-1.24	0.034	
Xenopus laevis dynein light chain roadblock-type 2 (dynlb2) mRNA	NM_001097940.1	33.46	15.19	-1.14	0.034	
Xenopus laevis protein cas4c (cas4c) mRNA	NM_001086374.1	20.73	9.69	-1.1	0.034	
PREDICTED: Xenopus (Silurana) tropicula lebercilin-like (LOC100497171) mRNA	AFO53935.2	10.41	4.41	-1.24	0.035	
Xenopus laevis phosphodiesterase 6b (GMP-specific cnc gamma 1) mRNA	NM_0010937222.2	2.82	0.28	-3.34	0.036	
PREDICTED: Xenopus (Silurana) tropicula GABA(A) receptor-associated protein like 1 (gabarpl1-b) mRNA	NM_001011006.1	1.83	0.12	-3.25	0.036	
PREDICTED: Xenopus (Silurana) tropicula neural precursor cell expressed developmentally down-regulated 4 E3 ubiquitin protein ligase (nedd4) transcript variant X2 mRNA	NM_001079044.1	3.98	1.14	-1.81	0.036	
Xenopus laevis catalase gene 1 (cat1) mRNA	NM_00109515647.1	1.95	0.37	-2.42	0.035	
Xenopus laevis protein cas4a (cas4a) mRNA	BC047386.1	1.4	3.96	1.51	0.035	
Xenopus tropicula nitric oxide synthase mRNA complete cds	NM_0010938932.2	1.25	0.06	-4.41	0.035	
PREDICTED: Xenopus (Silurana) tropicula uncharacterized 49 member A mRNA (cdna clone MGCI147023) (IMAGE7822461) complete cds	NM_0010110273.1	1.08	0.12	-2.71	0.036	
Xenopus (Silurana) tropicula lumican (lum) mRNA	NM_001079044.1	3.98	1.14	-1.81	0.036	
PREDICTED: Xenopus (Silurana) tropicula chromosome 20 open reading frame 26 (c20orf26) mRNA	NM_001094912701.1	11.56	3.32	-1.8	0.036	
Xenopus tropicula family with sequence similarity 49 member B mRNA (cdna clone MGCI147023) (IMAGE7822461) complete cds	NM_001095194.1	4	1.24	-1.69	0.036	
Xenopus laevis cDNA clone IMAGE7822461 mRNA	BC121555.1	3.46	1.18	-1.56	0.036	
Xenopus laevis uncharacterized LOC101732297 mRNA	NM_001094912701.1	6.03	2.09	-1.53	0.036	
Xenopus laevis Mdm1 nuclear protein homolog (mdm1) mRNA	NM_001302671	13.6	5.22	-1.38	0.036	
X. laevis mRNA for beta-1-adrenergic receptor receptor	Y09213.1	4.83	2.05	-1.24	0.036	
Xenopus (Silurana) tropicula poly(A) binding protein cytoplasmic 4 (inducible form) (pabpc4) mRNA	NM_001015753.1	15.87	7.06	-1.17	0.036	
Xenopus laevis vesicle-associated membrane protein 2 mRNA (cdna clone MGCG68783) (IMAGE4202036) complete cds	BC060344.1	2.97	0.58	-2.37	0.037	

Gene	Accession	Control	Eya1-MO <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO <sup>FC</sup>	c-value
PREDICTED: Xenopus (Silurana) tropicalis fidgetin-like (LOC100487395) mRNA	XM_004911907.1	2.72	1.15	-1.25	0.037	
Xenopus laevis myeloid differentiation primary response 88 (myd88-b) mRNA	NM_001057861	23.22	10.66	-1.12	0.037	
PREDICTED: Xenopus (Silurana) tropicalis cdk5 transcript variant XI mRNA	XM_0029325332	5.49	2.56	-1.1	0.037	
Xenopus laevis IVD repeat-containing protein 36-like (MGCS3182) mRNA	NM_001086229.1	16.7	8.09	-1.05	0.037	
PREDICTED: Xenopus (Silurana) tropicalis cGMP-dependent protein kinase 1-like (LOC100494278) transcript variant XI mRNA	XM_0029354282	2	0.3	-2.74	0.038	
XL28067: Xenopus laevis neurogenin differentiation factor (neurod) mRNA complete cds	U28067.1	6.77	1.69	-2.01	0.038	
Xenopus laevis neuronal differentiation 1 (neurod1-b) mRNA	NM_001092127.1	4.75	1.25	-1.93	0.038	
Xenopus laevis hypothetical protein LOC797978 mRNA (CDNA clone IMAGE:8321296) partial Cds	BC123275.1	7.71	2.05	-1.91	0.038	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100492104 (LOC100492104) mRNA	XM_002932457.2	4.13	1.66	-1.31	0.038	
Xenopus laevis hypothetical LOC949429 mRNA (CDNA clone MGCI15117 IMAGE:947554) complete cds	BC054118.1	10.6	4.61	-1.2	0.038	
Xenopus laevis neurogenin differentiation factor (neurod) mRNA complete cds	NM_001096479.1	744.79	11.42	-6.03	0.039	
Xenopus laevis ion peptidase 2 peptidomimetic (lon2) mRNA	NM_001096304.1	22.95	4.96	-2.21	0.039	
Xenopus laevis rib43A domain with coiled-coil 1 (ribc1) mRNA	XM_002935592	1.25	0.35	-1.85	0.039	
PREDICTED: Xenopus (Silurana) tropicalis protein phosphatase 1 regulatory subunit 9B (ppp1'9b) mRNA	BC11726.1	7.77	2.18	-1.83	0.039	
Xenopus laevis hypothetical protein LOC100497491 mRNA (CDNA clone IMAGE:7204292) partial Cds	XM_004911446.1	8.07	2.91	-1.47	0.039	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101730828 (LOC101730828) mRNA	BC114211.1	8.67	3.56	-1.29	0.039	
Xenopus laevis cDNA clone IMAGE:7394776	NM_001096665.1	6.76	3.02	-1.17	0.039	
Xenopus laevis Rab19 member RAS oncogene family (rab19) mRNA	XM_004917220.1	2.39	0.63	-1.93	0.040	
PREDICTED: Xenopus (Silurana) tropicalis chromosome unknown open reading frame human C14orf37 (LOC100497155) transcript variant X3 mRNA	XM_002938622.2	3.24	1.07	-1.6	0.040	
Xenopus laevis hypothetical protein LOC100497491 mRNA (CDNA clone IMAGE:7204292) partial Cds	NM_00126955.1	4.71	1.74	-1.44	0.040	
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733849 (LOC101733849) mRNA	NM_001093923.1	34.29	16.1	-1.09	0.040	
Xenopus laevis cDNA clone IMAGE:7394776	NM_001096760.1	2.64	0.25	-3.43	0.041	
Xenopus laevis Rab19 member RAS oncogene family (rab19) mRNA	NM_001099903.1	7.91	3.12	-1.34	0.041	
PREDICTED: Xenopus (Silurana) tropicalis processing of precursor 5'ribonuclease P/MRP subunit (pop5) mRNA	NM_001099903.1	5.21	2.19	-1.25	0.041	
Xenopus laevis protein phosphatase 3 catalytic subunit alpha isoform (pp3ca) mRNA	NM_00126955.1	17.69	7.61	-1.22	0.041	
Xenopus laevis cytochrome b5 domain containing 1 (cyb5d1) mRNA	XM_002937579.2	13.79	6.3	-1.13	0.041	
PREDICTED: Xenopus (Silurana) tropicalis nuclear mitotic apparatus protein 1-like (LOC100496014) transcript variant X1 mRNA	XM_004910911.1	3.49	0.83	-2.08	0.042	
Xenopus laevis mRNA for alpha2 (st3gal2) mRNA	AJ58762.1	4.08	1.06	-1.95	0.042	
Xenopus tropicalis finished cDNA clone Tgas012h22	CR761682.2	11.06	4.22	-1.39	0.042	
Xenopus laevis cDNA clone IMAGE:5512439 partial Cds	BC070863.1	7.79	3.54	-1.14	0.042	
Xenopus laevis thioredoxin (txn) mRNA	NM_001095018.1	19.39	8.84	-1.13	0.042	
Xenopus tropicalis finished cDNA clone Thieu033k10	CR700883.2	4.5	0.63	-2.84	0.043	
PREDICTED: Xenopus (Silurana) tropicalis homothiolate reductase 1 (chpt1) mRNA	XM_002932409.2	2.68	0.56	-2.26	0.043	
Xenopus (Silurana) tropicalis uncharacterized LOC100493999 (LOC100493999) transcript variant X2 mRNA	NM_004915901.1	8.15	1.94	-2.07	0.043	
PREDICTED: Xenopus (Silurana) tropicalis peroxisomal membrane protein 4 (pxmp4) mRNA	XM_002932856.2	6.18	2.21	-1.48	0.043	
Xenopus laevis isletabillin mRNA partial Cds	DQ30728.1	5.88	2.32	-1.34	0.043	
Xenopus laevis intraflagellar transport 52 homolog (ift52) mRNA	NM_001094130.1	12.61	5.22	-1.27	0.043	
Xenopus laevis selenophosphate phosphotransferase 1 (chpt1) mRNA	NM_001096106.1	17.13	7.52	-1.19	0.043	
PREDICTED: Xenopus (Silurana) tropicalis synaptotagmin-like 2 (synt2) mRNA	NM_001007878.2	46.37	20.37	-1.19	0.043	
Xenopus (Silurana) tropicalis uncharacterized LOC100498463 (LOC100498463) transcript variant X2 mRNA	XM_004914061	26.64	12.37	-1.11	0.043	
PREDICTED: Xenopus (Silurana) tropicalis cytosolic 5'-nucleotidase A-like (LOC100495274) mRNA	XM_002933901.2	2.57	0.38	-2.77	0.044	
Xenopus (Silurana) tropicalis clathrin heavy chain	XM_002934054.2	1.28	0.24	-2.42	0.044	
Xenopus (Silurana) tropicalis colled-coil and C2 domain containing 2a (ccdc22a) mRNA	XM_002936643.2	18.17	4.84	-1.93	0.044	
PREDICTED: Xenopus (Silurana) tropicalis glutamate-ammonia ligase (glul) transcript variant X2 mRNA	CT025189.2	46.37	20.37	-1.19	0.044	
Xenopus (Silurana) tropicalis growth arrest-specific 2 like (gas2l2) transcript variant X2 mRNA	XM_004914038.1	6.98	3	-1.22	0.044	
Xenopus (Silurana) tropicalis glutamine synthetase (glu1) transcript variant XI mRNA	NM_0010979184.1	17.65	8.44	-1.06	0.044	
PREDICTED: Xenopus (Silurana) tropicalis synaptogamin 16 (syn16) transcript variant XI mRNA	XM_002939478.2	17.84	8.72	-1.03	0.044	
Xenopus (Silurana) tropicalis uncharacterized LOC100497783 (LOC100497783) partial mRNA	XM_002942126.2	2.78	0.71	-1.97	0.045	
Xenopus (Silurana) tropicalis chromosome 9 open reading frame 117 (cporf117) mRNA	NM_001079306.1	7.04	2.19	-1.68	0.045	
PREDICTED: Xenopus (Silurana) tropicalis growth arrest-specific 2 like 2 (gas2l2) transcript variant X2 mRNA	XM_004911630.1	4.81	0.92	-2.39	0.045	
Xenopus (Silurana) tropicalis finished cDNA clone Tba02710	CR942710.2	5.66	1.46	-1.96	0.045	
Xenopus (Silurana) tropicalis ornithine aminotransferase (oat) nuclear gene encoding mitochondrial protein mRNA	NM_001093221.1	4.24	1.35	-1.65	0.045	
PREDICTED: Xenopus (Silurana) tropicalis 7-alpha-hydroxycholesterol-4-en-3-one 12-alpha-hydroxylase-like (LOC100481658) mRNA	XM_002935221.1	7.22	2.91	-1.21	0.045	
Xenopus (Silurana) tropicalis finished cDNA clone Tegg072a06	CR83553.2	3.72	1.58	-1.23	0.045	
PREDICTED: Xenopus (Silurana) tropicalis microtubule-associated tumor suppressor 1 homolog A-like (LOC100488087) mRNA	XM_002932233.2	3.87	0.46	-3.09	0.046	
Xenopus (Silurana) tropicalis finished cDNA clone Tba027113	CU07555.1	2.16	0.29	-2.91	0.046	
Xenopus laevis premature ovarian failure 1B (poof1) mRNA	NM_001193408.1	16.07	5.84	-1.46	0.046	
PREDICTED: Xenopus (Silurana) tropicalis oxidative stress-induced growth inhibitor 1 (osgin1) mRNA	XM_002935131.2	5.5	2.1	-1.39	0.046	

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	-t-value
PREDICTED: Xenopus (Silurana) tropicalis protein S100-G-like (LOC100494945) mRNA	XM_002942678.2	3/6.78	1/4.93	-1.11	0.046
Xenopus laevis orga4 mRNA complete sequence	DQ056925.1	276.76	131.65	-0.07	0.046
Xenopus (Silurana) tropicalis branched chain amino-acid transaminase 2 mitochondrial (bcat2) nuclear gene encoding mitochondrial protein mRNA	NM_001006739.1	31.56	15	-1.07	0.046
Xenopus laevis keratin 19 (kr19) mRNA	NM_001091523.1	252.38	124.97	-1.01	0.046
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine-protein kinase PIK2-like (LOC100487804) mRNA	XM_002941122.2	1.23	0.21	-2.61	0.047
PREDICTED: Xenopus (Silurana) tropicalis chromosome region candidate 12 (ds2cr12) transcript variant X2 mRNA	XM_002941775.1	3.5	0.83	-2.09	0.047
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine-protein kinase PIK2-like (kr1) transcript variant X4 mRNA	XM_002914441.1	4.15	1.36	-1.61	0.047
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 2-like (LOC101731505) mRNA	NM_001092303.1	11.21	3.91	-1.52	0.047
Xenopus laevis fibrous sheath-interacting protein 1 mRNA (cDNA clone IMAGE8548844) partial cds	BC129783.1	3.53	1.36	-1.37	0.047
Xenopus laevis receptor protein tyrosine phosphatase CRYP-alpha mRNA partial cds	AF198450.1	3.52	1.39	-1.34	0.047
Xenopus laevis HB51-like (hb51) mRNA	NM_001092382.1	391.37	181.23	-1.11	0.047
Xenopus laevis hypothetical LOC443366 mRNA (cDNA clone IMAGE5511185)	BC0733564.1	18.66	0.19	-6.62	0.048
Xenopus laevis impact protein mRNA (cDNA clone IMAGE7010848)	BC084977.1	53.72	8.61	-2.64	0.048
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100489259 (LOC100489259) mRNA	NM_002935863.2	5.69	1.6	-1.83	0.048
Xenopus laevis metalloprotease-disintegrin (MDC11a) mRNA partial cds	AF032384.1	2.22	0.37	-2.61	0.049
PREDICTED: Xenopus (Silurana) tropicalis mucin-2-like (LOC100494822) mRNA	NM_002938567.2	3.73	0.79	-2.25	0.049
PREDICTED: Xenopus (Silurana) tropicalis cadherin-related family member 4 (cdhr4) mRNA	NM_002936486.2	3.92	1.24	-1.67	0.049
Xenopus laevis sNME/NM23 family member 9 (ome9) mRNA	NM_001091578.1	18.15	8	-1.18	0.049
Xenopus laevis putative beta-N-acetylhexosaminidase mRNA partial cds	JN127371.1	17.38	7.75	-1.17	0.049
PREDICTED: Xenopus (Silurana) tropicalis interleukin 17 receptor E (il17re) mRNA	NM_002914115.1	5.02	2.29	-1.13	0.049
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100485911 (LOC100485911) mRNA	NM_002942801.2	3.28	1.2	-1.45	0.049
Xenopus laevis single-stranded DNA binding protein 2 (ssbp2) mRNA	NM_001086878.1	20.12	9.32	-1.11	0.049

<sup>1</sup> Expression levels (FPKM) in un-injected pre-placodal explants

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Eya1-MO

<sup>3</sup> Log<sub>2</sub> Fold change values for Eya1-MO

## I.4 Eya1-MO: Up-regulated genes

**Table I.4** Genes significantly up-regulated after injection of Eya1-MO

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO <sup>3</sup>	q-value
Xenopus laevis Xpat protein mRNA (cDNA clone MGC:39925 IMAGE:55155227) complete cds	BC072773_1	0.01	21.17	14.37	0.003
Xenopus laevis hypothetical protein LOC_33309 mRNA (cDNA clone IMAGE:6939729) partial cds	BC099336_1	0.01	2.41	11.23	0.003
PREDICTED: Xenopus (Silurana) tropicalis NPFY family member 1-like (LOC_01731316) partial mRNA	XM_004920266_1	0.01	2.15	11.07	0.003
Xenopus laevis phosphatidylethanolamine-binding protein 4 (pepb4) mRNA	XM_002932556_1	0.01	1.92	10.9	0.003
Xenopus tropicalis finished cDNA clone TEG019k04	NM_001093828_1	0.01	1.71	10.74	0.003
PREDICTED: Xenopus (Silurana) tropicalis unconventional myosin-XV-like (LOC_0048569) mRNA	CR761320_2	0.01	1.25	10.28	0.003
Xenopus (Silurana) tropicalis homolog subfamily C member 28 (dnajc28) mRNA	NM_00293973_2	0.01	2.24	10.13	0.003
Xenopus tropicalis finished cDNA clone TNelj025c10	NM_001093777_1	1171.46	884808.92	9.56	0.003
Xenopus laevis uncharacterized LOC495410 (LOC01731410) mRNA	CT030610_1	7247.25	4556459.11	9.3	0.003
XELXLOTA Xenopus laevis mRNA for a pleiotrophic factor-beta2 complete cds	NM_001095067_1	0.31	133.07	8.78	0.003
Xenopus laevis ganylate binding protein 2 interferon-inducible (gbp2) mRNA	D42060_1	4.48	191.23	5.42	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC_01733460 (LOC_01733460) mRNA	NM_001097040_1	0.46	13.35	4.86	0.003
Xenopus laevis hypothetical protein LOC_00049126 mRNA (cDNA clone MGC:161039 IMAGE:7299620) complete cds	XM_004913346_1	0.12	2.34	4.4	0.003
PREDICTED: Xenopus (Silurana) tropicalis trafficking protein kinesin binding 2 (trak2) transcript variant X2 mRNA	BC133776_1	16475.81	311078.1	4.24	0.003
Xenopus laevis ganylate binding protein 1 interferon-inducible (gbp1) mRNA	NM_001097797_1	0.09	1.3	3.93	0.003
Xenopus laevis hyaluronan synthase related sequence protein (has-15) mRNA	NM_001088359_1	0.18	11.51	3.72	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC_01732820 (LOC_01732820) mRNA	XM_004913268_1	14.5	46.25	3.49	0.003
PREDICTED: Xenopus (Silurana) tropicalis ectodysplasin A2 receptor (edar2) transcript variant X1 mRNA	NM_002938915_2	0.62	5.67	3.21	0.003
Xenopus tropicalis cDNA clone IMAGE:75473320	BC166316_1	2800.8	24486.51	3.13	0.003
PREDICTED: Xenopus (Silurana) laminin-associated polypeptide 2 isoforms alpha/zeta-like (LOC_01731493) mRNA	NM_004913342_1	4.13	26.9	2.71	0.003
Xenopus laevis serum/glucocorticoid-regulated kinase 1 (sgk1-a) mRNA	NM_001090340_1	88.73	581.69	2.71	0.003
Xenopus laevis SPB-a mRNA complete cds	NM_001094563_1	0.42	6.68	2.67	0.003
Xenopus laevis solute carrier family 16 (monocarboxylic acid transporters) member 6 (slc16a6) mRNA	NM_001086838_1	0.18	19.65	2.63	0.003
Xenopus laevis ornithine decarboxylase 2 mRNA (cDNA clone MGC:55257 IMAGE:552547) complete cds	BC047954_1	162.87	1002	2.62	0.003
Xenopus laevis TAR (HIV-1) RNA binding protein 2 (tarbp2) mRNA	NM_001092105_1	47.15	276.08	2.55	0.003
Xenopus laevis myeloperoxidase peroxidase 2 mRNA (cDNA clone IMAGE:7766107) partial cds	BC108479_1	2.25	13.16	2.55	0.003
Xenopus laevis G-protein signaling 2.24kDa (rgs2) mRNA	FJ644945_1	0.99	5.67	2.52	0.003
Xenopus laevis histone cluster 1-H2bj (hist1h2bj) mRNA	BC155933_1	27.55	144.65	2.39	0.003
Xenopus laevis hypothetical protein MGC:130860 (cDNA clone MGC:130860 IMAGE:7205580) complete cds	NM_00109916_1	0.41	2.13	2.38	0.003
Xenopus laevis histone cluster 1-H2bj (hist1h2bj) mRNA	NM_001093284_1	94.2	474.22	2.33	0.003
PREDICTED: Xenopus (Silurana) tropicalis spike homeobox 2 (Drosophila) (spine2) transcript variant X1 mRNA	BC10633_1	89.71	440.08	2.29	0.003
Xenopus laevis family homeobox 2 (hmnx2) mRNA	NM_00293684_2	0.27	1.15	2.13	0.003
Xenopus laevis G-protein regulator of G-protein signaling 2.24kDa (rgs2) mRNA	NM_001095105_1	6.82	29.64	2.12	0.003
Xenopus laevis saif domain containing kinase 3 (adck3) nuclear gene encoding mitochondrial protein mRNA	NM_001090545_1	7.16	29.59	2.05	0.003
Xenopus laevis CDNA clone IMAGE:7200336	XM_002936941_2	2.6	10.62	2.03	0.003
XENOGAT Xenopus (Silurana) tropicalis prepropeptide cartpt1 mRNA	NM_001097842_1	3.84	15.47	2.01	0.003
Xenopus laevis Xpat protein mRNA for Xam protein	NM_001096261_1	105.78	425.71	2.01	0.003
XELXOCT91 Xenopus laevis xct-91 mRNA complete cds	BC13015_1	18780.85	7496.48	2	0.003
Xenopus laevis CART prepropeptide cartpt1 mRNA	NM_001094096_1	0.77	3.05	1.99	0.003
Xenopus laevis receptor-interacting serine-threonine kinase 4 mRNA (cDNA clone MGC:132134 IMAGE:4741010) complete cds	BC108792_1	5.07	19.98	1.98	0.003
Xenopus laevis mRNA for Xam protein	X98454_1	4.83	19.03	1.98	0.003
Xenopus laevis glutamic pyruvate transaminase (alanine aminotransferase) 2 (gp2) mRNA	NM_001092635_1	1.91	7.45	1.97	0.003
Xenopus laevis solute carrier family 35 (UDP-N-acetylglucosamine (UDP-GlcNAc) transporter) member A3 gene 1 (slc35a3_1) mRNA	NM_001096261_1	4.81	18.51	1.94	0.003
Xenopus laevis BTG3 associated nuclear protein (banp) mRNA	NM_001095794_1	6.63	24.67	1.9	0.003
Xenopus laevis situtary homeobox gene 1 paired-like homeodomain transcription factor (Ptx1) mRNA complete cds	AF217647_1	5.36	20.01	1.9	0.003
Xenopus laevis NK3 homeobox 1 (nkx3-1-a) mRNA	NM_001086738_1	5.78	21.35	1.89	0.003
Xenopus laevis ATPase Na <sup>+</sup> /K <sup>+</sup> -transporting beta 2 polypeptide (atp1b2) mRNA	NM_001092728_1	2.2	8.02	1.87	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC_01730916 (LOC_01730916) mRNA	XM_004917653_1	3.87	13.92	1.85	0.003
XELXOCT91 Xenopus laevis xct-91 mRNA complete cds	M60077_1	16.91	60.26	1.83	0.003
Xenopus laevis krtpp1-like homeobox gene 10 (kif10) mRNA	NM_001095871_1	5.38	18.94	1.82	0.003
Xenopus tropicalis finished cDNA clone TEG017407	CT030570_1	524.18	1816.21	1.79	0.003
Xenopus laevis cone-rod homeobox (cro-1) mRNA	NM_001088447_1	7.7	26.08	1.76	0.003

Gene	Accession	Control <sup>l</sup>	Eya1-MO <sup>2</sup>	Eya1-MO <sup>3</sup>	q-value
Xenopus laevis aquaporin 3 (Gill blood group) (aqp3-b) mRNA	NM_001094477.1	3.17	10.69	1.75	0.003
Xenopus laevis regulator of calcineurin 1 (rcan1) mRNA	NM_001086391.1	8.4	28.03	1.74	0.003
Xenopus laevis acid phosphatase 1 soluble (acp1-a) transcript variant b mRNA	NM_001114769.1	5.94	19.81	1.74	0.003
Xenopus laevis Ito kinase 3 (itoK3) mRNA	NM_001089923.1	7.96	25.95	1.71	0.003
Xenopus laevis thioredoxin interacting protein (txnip) mRNA	NM_001093153.1	4.8	15.75	1.71	0.003
Xenopus laevis upstream binding protein 1 (Lbp-1a) (ubp1) mRNA	NM_001098699.1	22.43	73.05	1.7	0.003
Xenopus tropicalis cDNA clone IMAGE:6991249	BC158454.1	524922.88	1.884478.42	1.69	0.003
YEI1 AA X-borealis B1 protein mRNA complete cds	ME3663.1	2406.5	7672.23	1.67	0.003
PREDICTED: Xenopus (Silurana) tropicalis embryonic protein UV5.2-like (LOC101734297) transcript variant X1 mRNA	NM_004913368.1	335.77	1059.65	1.66	0.003
Xenopus tropicalis finished cDNA clone TGAst063j09	CT0253992	232.42	716.38	1.62	0.003
Xenopus laevis receptor-associated protein of the synapse (raspn) mRNA	NM_001093521	2.83	8.67	1.62	0.003
Xenopus laevis heat shock protein 90kDa alpha (cytosolic) class A member 1 gene 1 (hsp90aa1.1) mRNA	NM_001092129.1	7.49	22.45	1.58	0.003
PREDICTED: Xenopus (Silurana) tropicalis homeobox A4 (hoxc4) mRNA	NM_004915397.1	5.53	16.37	1.57	0.003
Xenopus (Silurana) tropicalis glioma tumor suppressor candidate gene 1 (gtrs1) mRNA	NM_001113827.1	2.66	7.88	1.57	0.003
PREDICTED: Xenopus (Silurana) tropicalis C-reactive protein-like (LOC100489750) mRNA	NM_001085944.1	1.41	4.17	1.57	0.003
Xenopus laevis paired box 6 (pax6-a) mRNA	AB075925.1	3.57	10.55	1.56	0.003
Xenopus laevis mRNA for tailin complete cds	NM_001095344.1	34.05	99.69	1.55	0.003
Xenopus laevis phosphatidylinositol glycan anchor biosynthesis class F (pigf) mRNA	NM_001095344.1	23.7	68.18	1.52	0.003
PREDICTED: Xenopus (Silurana) tropicalis inverted repeat poly (ADP-ribosel) polymerase 14-like (LOC100487753) mRNA	NM_002942598.2	8.8	25.21	1.52	0.003
Xenopus laevis cyclin G1 (cng1) mRNA	NM_001096966.1	10.34	28.34	1.46	0.003
Xenopus laevis solute carrier family 22 organic cation/carnitine transporter member 5 (slc22a5) mRNA	NM_001087429.1	61.49	165.25	1.43	0.003
Xenopus tropicalis arrestin domain containing 3 mRNA cDNA clone MGCI:21258 (IMAGE:7598595) complete cds	BC13514.1	3.99	10.44	1.39	0.003
Xenopus laevis heterogeneous nuclear ribonucleoprotein A0 (hnrpao) mRNA	NM_001087024.1	96.12	247.98	1.37	0.003
PREDICTED: Xenopus (Silurana) tropicalis inverted formin-2-like (LOC101733927) mRNA	NM_004910623.1	4960927.76	12811377.88	1.37	0.003
Xenopus tropicalis cDNA clone IMAGE:7549631	BC158399.1	7.13	18.38	1.37	0.003
XCRAP=cellular retinoic acid-binding protein Xenopus laevis embryos mRNA	ST4933.1	30.09	76.44	1.35	0.003
Xenopus laevis clone S1:105-1-H5 mRNA sequence	AF549994.1	240.84	615.66	1.35	0.003
Xenopus (Silurana) tropicalis cullin 5 (cul5) mRNA	NM_001079368.1	4.48	11.21	1.32	0.003
Xenopus laevis stayopherin alpha 1 mRNA (cdna clone MGCG53789) complete cds	BC04637.3	41.96	103.87	1.31	0.003
X. laevis ODC mRNA for ornithine decarboxylase (Ec 4.1.1.7)	X56316.1	353.5	830.85	1.23	0.003
Xenopus laevis ATPase H+-K+-transporting nongastric alpha polypeptide (atp12a-a) mRNA	NM_001087349.1	10.48	23.94	1.19	0.003
Xenopus tropicalis cDNA clone IMAGE:6992395	BC082272.1	148349.75	338864	1.19	0.003
Xenopus laevis characterized LOC100036942 mRNA	NM_001097712.1	1.87	4.15	1.15	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101734735 (LOC101734735) mRNA	NM_00491602.1	29.8	65.05	1.13	0.003
Xenopus laevis sulfotransferase family cytosolic 2B member (sult2b1) mRNA	NM_001097677.1	180.25	373.77	1.05	0.003
Xenopus laevis mRNA for ornithine-serine-threonine phosphopeptides interacting protein 2 (pstpb2) mRNA	NM_00109301.1	2019.79	4162.68	1.04	0.003
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100497040 (LOC100497040) transcript variant X1 mRNA	NM_002931526.2	0.01	299	1.19	0.006
Xenopus laevis characterized LOC495221 (LOC495221) mRNA	NM_001094903.1	0.77	15.5	4.35	0.006
Xenopus laevis nucleophosphomim/nucleoplasm 2 (pm2) mRNA	NM_001097558.1	2.91	12.8	2.14	0.006
Xenopus laevis DNA clone IMAGE:7974039 partial cds	BC155953.1	1.86	8.13	2.13	0.006
Xenopus laevis paired-like homeodomain transcription factor 2 mRNA (cdna clone MGCG53789) complete cds	BC07347.9.1	6.6	18.65	1.5	0.006
Xenopus laevis mRNA complete cds	AF308810.1	2.18	5.68	1.38	0.006
Xenopus laevis zinc finger protein 166 gene 2 (zfpf6.2-a) mRNA	NM_001171732.1	19.71	49.52	1.33	0.006
Xenopus laevis small nuclear ribonucleoprotein polypeptide A' (snrp1) mRNA	NM_001098394.1	36.81	89.64	1.28	0.006
Xenopus laevis heterogeneous nuclear ribonucleoprotein A0 (hnrpao) mRNA	NM_001088945.1	98.89	226.07	1.19	0.006
Xenopus laevis finished cDNA clone TGA 02107	CR761464.2	0.01	16.97	14.05	0.008
Xenopus laevis characterized LOC100037047 (LOC100037047) mRNA	NM_001097778.1	0.88	4.78	2.45	0.008
Xenopus laevis protease neprin-1 (PN-1) mRNA complete cds	DQ324047.1	0.69	3.62	2.4	0.008
Xenopus laevis amyloid beta (A4) precursor-like protein 1 (ap1j1) mRNA	NM_001095950.1	1.67	8.39	2.33	0.008
Xenopus laevis ankyrin repeat and SOCS box containing 1 (asbl1) mRNA	NM_001096257.1	1.02	4.67	2.21	0.008
Xenopus laevis hypothetical LOC495232 mRNA (cdna clone IMAGE:5511906) partial cds	BC084651.1	0.92	3.58	1.97	0.008
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101731501 (LOC101731501) mRNA	XM_004918616.1	6.14	23.25	1.92	0.008
Xenopus laevis mRNA for Zic-related 2 complete cds	AB014461.1	2.01	6.83	1.77	0.008
PREDICTED: Xenopus (Silurana) tropicalis nuclear export mediator factor (nemf) transcript variant X1 mRNA	NM_002938127.2	9.18	30.43	1.73	0.008
Xenopus laevis odd-paired-like (opl) mRNA complete cds	AF028805.1	2.3	7.5	1.71	0.008
Xenopus laevis ITAM-bearing signaling subunit (Tcr2) mRNA complete cds	EF431896.1	3.11	8.34	1.43	0.008
PREDICTED: Xenopus (Silurana) tropicalis transformation/transcription domain-associated protein (trap) transcript variant X1 mRNA	NM_00293947.2	2.68	6.75	1.33	0.008
PREDICTED: Xenopus (Silurana) tropicalis transformation/transcription domain-associated protein (trap) transcript variant X2 mRNA	NM_004917975.1	5.11	11.23	1.14	0.008

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO <sup>3</sup>	q-value
PREDICTED: Xenopus (Silurana) tropicalis SET and WND domain containing 3 (smyod3) transcript variant X2 mRNA	XM_004946261_1	0.23	2.48	3.44	0.010
Xenopus laevis uncharacterized protein MGC115729 (MGC115729) mRNA	NM_001096099_1	2.27	12.43	2.46	0.010
Xenopus (Silurana) tropicalis sterol carrier protein 2 (scp2) mRNA	NM_001015795_1	1.46	7.46	2.36	0.010
Xenopus laevis xCAD2 protein mRNA (cDNA clone MGC:160980 (IMAGE:8319613)) complete cds	BC1_46637_1	0.82	3.9	2.26	0.010
PREDICTED: Xenopus (Silurana) tropicalis protein Wnt-7b-like (LOC00490426) mRNA	XM_002935496_2	3.98	16.58	2.06	0.010
Xenopus tropicalis finished cDNA clone TEG0377/p04	CU075595_1	0.43	1.71	2	0.010
Xenopus laevis arachidonate 12-lipoxygenase 12R type (lcox12b) mRNA	NM_001095796_1	1.46	5.66	1.96	0.010
Xenopus laevis zinc finger AN1-type domain 2A (zfAn2d2a) mRNA	NM_001093364_1	2.64	9.87	1.9	0.010
Xenopus laevis hypothetical LOC445868 mRNA (cDNA clone MGC:79897 (IMAGE:5156432)) complete cds	BC084194_1	2.54	7.96	1.65	0.010
Xenopus laevis hypothetical LOC445868 mRNA (cDNA clone MGC:7982507)	BC1_08429_1	6.1	15.36	1.33	0.010
Xenopus laevis bone morphogenic protein 6 mRNA (cDNA clone MGC:79851 (IMAGE:6636878)) complete cds	NM_001095169_1	5.84	13.88	1.25	0.010
Xenopus laevis bone morphogenic protein 6 (bipid6-b) mRNA	BC072120_1	12.67	26.54	1.07	0.010
Xenopus laevis protein disulfide isomerase family A member 6 (pidis6-b) mRNA	NM_001098638_1	113.66	235.62	1.05	0.010
Xenopus laevis as -related protein in RAS-DNA mRNA (cDNA clone MGC:131080 (IMAGE:7981700)) complete cds	BC108584_1	42.92	85.85	1	0.010
PREDICTED: Xenopus (Silurana) tropicalis histone N-methyltransferase pre-set7-like (LOC01731509) partial mRNA	XM_004920537_1	19.21	211.91	3.46	0.011
PREDICTED: Xenopus (Silurana) tropicalis histone N-methyltransferase pre-set7-like (LOC01731509) transcript variant X6 mRNA	XM_004912632_1	0.33	1.63	2.32	0.011
Xenopus laevis KAROS family zinc finger 4 (Ecs1) (zfz4) mRNA	BC057702_1	1.55	6.43	2.06	0.011
Xenopus laevis bone morphogenic protein 6 mRNA (cDNA clone MGC:68434 (IMAGE:66862718)) complete cds	BC15359_1	20.31	74.54	1.88	0.011
Xenopus laevis cDNA clone IMage:E5317850	BC157454_1	20	47.45	1.25	0.011
Xenopus laevis MARCKS-like protein mRNA (cDNA clone MGC:179993 (IMAGE:8330180)) complete cds	NM_001127411_1	0.13	1.54	3.64	0.013
Xenopus laevis as -related protein in RAS-DNA mRNA (cDNA clone MGC:131080 (IMAGE:7981700)) partial mRNA	NM_001114813_1	0.74	6.97	3.24	0.013
Xenopus laevis histone cluster 1 (H4d (hist1h4d)) mRNA	NM_001135069_1	2.82	15.1	2.42	0.013
Xenopus laevis mRNA for Zic2 protein complete cds	AB009365_1	1.83	7.31	2	0.013
Xenopus laevis mRNA for transcription factor VENF2	X98849_1	3.46	11.51	1.74	0.013
XEL1HSH1-Xlaevis histone H1-like maternal (B4) protein mRNA complete cds	M36655_1	10.3	31.77	1.63	0.013
Xenopus tropicalis cDNA clone IMAGE:8920859	BC1_6124_1	8.3	24.7	1.57	0.013
Xenopus tropicalis paired-like homeo-domain 2 mRNA (cDNA clone MGCI47762 (IMAGE:7523076)) complete cds	BC1_5902_2	3.87	10.27	1.41	0.013
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein FOG family member 1 (zfpm1) transcript variant X1 mRNA	XM_002936100_2	5.43	14.08	1.38	0.013
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein FOG family member 1 (zfpm1) transcript variant X1 mRNA	XM_004910733_1	13.23	32.02	1.28	0.013
Xenopus laevis high mobility group box 2 (hmgb2) mRNA	NM_001095918_1	256.28	568.25	1.15	0.013
Xenopus laevis crystallin lambda 1 (crl1) mRNA	NM_001097733_1	0.14	2.37	4.08	0.015
Xenopus laevis mRNA for Zic3 protein complete cds	AB00529_2	1.42	4.65	1.71	0.015
Xenopus laevis D-dopachrome autotermerase (ddt) mRNA	NM_001094566_1	1.88	4.75	1.34	0.015
Xenopus laevis PWP1 homolog (fwp1) mRNA	NM_001091163_1	0.01	167.99	17.36	0.016
Xenopus (Silurana) tropicalis uncharacterized LOC100145470 (LOC100145470) mRNA	NM_001126922_1	0.01	1.3	10.34	0.016
PREDICTED: Xenopus (Silurana) tropicalis lipoma HMIGC fusion partner-like 1 (lhp1) transcript variant X1 mRNA	XM_002938786_2	0.27	3.14	3.59	0.016
Xenopus laevis 3-kinase regulatory subunit 1 (MAPK3) mRNA	BC07230_1	0.34	1.96	2.53	0.016
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100485964 (LOC00485964) transcript variant X2 mRNA	XM_004917879_1	0.87	3.56	2.04	0.016
PREDICTED: Xenopus (Silurana) Down syndrome cell adhesion molecule like 1 (dsccml1) transcript variant X2 mRNA	XM_004916065_1	1.55	4.19	1.44	0.016
Xenopus (Silurana) tropicalis cell adhesion molecule like 1 (crabp2) mRNA	NM_001006862_1	30.46	78.24	1.36	0.016
Xenopus laevis Aly/REF export factor (alyref) mRNA	NM_001095845_1	120.26	251.39	1.06	0.016
Xenopus laevis uncharacterized LOC100158389 (LOC100158389) mRNA	NM_001127833_1	0.18	4.48	4.69	0.018
Xenopus laevis XCT-25 mRNA (cDNA clone MGCI68441 (IMAGE:8686305)) complete cds	BC05956_1	0.24	3.18	3.74	0.018
Xenopus laevis GSK-3 binding protein GBP (Gbp) mRNA complete cds	AF062738_1	3.41	7.78	1.19	0.018
Xenopus laevis DNA clone IMAGE:3397054	BC079827_1	14.71	32.07	1.12	0.018
Xenopus laevis pleckstrin homology domain containing family A (phosphoinositide binding specific) member 2 (plekha2) mRNA	AY64830_1	2.07	4.42	1.1	0.018
Xenopus laevis Smn1 mRNA	AB197248_1	0.88	4.48	2.85	0.019
PREDICTED: Xenopus (Silurana) tropicalis metalloionene-like (LOC100495370) mRNA	XM_002937175_2	0.09	16.26	2.4	0.019
Xenopus laevis Runx-related transcription factor 1 mRNA (cDNA clone MGC:69003 (IMAGE:4963637)) complete cds	BC057739_1	0.35	1.43	2.06	0.019
Xenopus laevis baculoviral IAP repeat-containing 5 gene 1 (bir5c1-al) mRNA	NM_001087631_1	2.96	10.28	1.8	0.019
PREDICTED: Xenopus (Silurana) G protein-coupled receptor family C group 5 member C (gprc5c) transcript variant X1 mRNA	XM_002934984_2	1.17	3.54	1.61	0.019
Xenopus tropicalis finished cDNA clone Thel03103	CR85456_2	1.69	4.99	1.56	0.019
Xenopus (Silurana) tropicalis 72 domain family member 3 (tsc22d3) mRNA	NM_001086817_1	4.6	12.44	1.44	0.019
Xenopus laevis protein phosphatase 1 catalytic subunit gamma isozyome (ppp1cc) mRNA	NM_001087839_1	9.73	22.39	1.2	0.019
Xenopus laevis tumor protein p53 inducible protein 11 (tp53i1) mRNA	NM_001097917_1	1.82	24.58	3.76	0.020
Xenopus laevis brachyury (T) mRNA (cDNA clone MGCI78815 (IMAGE:3402478)) complete cds	BC07203_1	0.86	3.37	1.98	0.020
XEL1HSH1-Xlaevis xct-79 mRNA	M60076_1	6.81	15.75	1.21	0.020

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	q-value
Xenopus laevis Eph receptor B3 (ephl3) mRNA	NM_00109526.1	7.54	16.25	1.11	0.020
Xenopus laevis hypothetical protein LOC432278 mRNA (cDNA clone IMAGE:4680796) partial cds	BC072283.1	1.57	11.4	2.86	0.022
Xenopus laevis protein Idas (mida) mRNA	NM_001127804.1	54.51	142.89	1.39	0.022
PREDICTED: Xenopus (Silurana) tropicalis exportin 1 (CRM1 homolog) (xpo1) transcript variant X2 mRNA	XM_004914701.1	12.61	26.13	1.05	0.022
Xenopus (Silurana) tropicalis phosphodiesterase 4D interacting protein (pde4dip) mRNA	NM_001079418.1	3.7	10.17	1.46	0.023
Xenopus laevis cDNA clone (IMAGE:7976113) partial cds	BC107201	84.78	232.31	1.45	0.023
Xenopus (Silurana) tropicalis protein kinase domain containing cytoplasmic (pkdcc2) mRNA	NM_001127171	9.65	23.29	1.27	0.023
Xenopus laevis G-protein coupled 10 (p2y10) mRNA	NM_001098461.1	0.3	4.42	3.9	0.024
Xenopus laevis secreted frizzled-related protein (frpx) mRNA	NM_001105267.1	0.45	2.31	2.36	0.024
PREDICTED: Xenopus (Silurana) tropicalis Crm1 crimped-like (Drosophila) (crimp1) transcript variant X1 mRNA	XM_002932451.2	0.7	2.5	1.85	0.024
PREDICTED: Xenopus (Silurana) tropicalis protein 2 (ck2ap2) transcript variant X2 mRNA	XM_004911970.1	0.36	2.84	3.01	0.025
PREDICTED: Xenopus (Silurana) tropicalis L2-inducible T-cell kinase (tkk) mRNA	XM_002939610.2	9.12	69.89	2.94	0.025
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC01731432 (LOC01731432) mRNA	NM_004919958.1	13.56	73.22	2.43	0.025
Xenopus laevis cDNA clone MGIC160633 IMAGE:5156759 complete cds	BC130110.1	1.79	7.62	2.09	0.025
Xenopus laevis t-nucleotidase domain containing 4 (nt5dc4) mRNA	NM_001094603.1	7.72	19.16	1.31	0.025
Xenopus (Silurana) tropicalis sestin 1 (sesn1) mRNA	NM_00129939.1	3.55	7.99	1.17	0.025
Xenopus laevis t-nucleotidase domain containing 3 (nt5dc3) mRNA	NM_001092501.1	6.44	67.13	3.38	0.026
Xenopus laevis cDNA clone (IMAGE:Es886501)	BC084311.1	7.63	24.22	1.67	0.026
Xenopus laevis mitochondrial ribosomal protein L15 (mrpl15) nuclear gene encoding mitochondrial protein mRNA	NM_001093374.1	5.31	14.77	1.48	0.026
Xenopus laevis gap junction protein beta 2 (gjb2) mRNA	NM_001005691.1	1.59	4.13	1.38	0.026
XL05528 Xenopus laevis homeobox protein (vox-1) mRNA complete cds	U53528.1	1.37	4.07	1.58	0.028
Xenopus laevis mRNA for Taf-1/beta2 complete cds	AB022692.1	11.01	25.71	1.22	0.028
Xenopus laevis apoptosis enhancing nuclelease (an) mRNA	NM_001094575.1	348.85	788.61	1.18	0.028
Xenopus laevis seminin H mRNA (cDNA clone IMAGE:83313 IMAGE:6864113) complete cds	BC082424.1	2.98	6.75	1.18	0.028
X.laevis mRNA for glucokinase	X93494.1	32.02	68.99	1.11	0.029
Xenopus laevis cysteine dioxygenase type 1 (cd1) mRNA	NM_001090037.1	0.24	1.21	2.37	0.030
Xenopus laevis t-nucleotidase uncharacterized LOC100487560 (LOC100487560) mRNA	XM_002937151.2	0.22	2.79	1.16	0.030
Xenopus laevis uncharacterized LOC100037078 (LOC100037078) mRNA	NM_001097802.1	5.59	22.79	2.03	0.031
Xenopus laevis CD44 molecule (Indian blood group) (cd44) mRNA	NM_00109133.1	0.86	3.28	1.94	0.031
Xenopus laevis characterized protein MGIC15323 (MGIC15323) mRNA	NM_001096542.1	6.42	17.94	1.48	0.031
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protein 140 (tmem140) transcript variant 2 mRNA	XM_002941427.2	0.26	1.69	2.75	0.032
Xenopus tropicalis finished cDNA clone Thida4212	CU075474.1	0.48	2.96	2.63	0.032
PREDICTED: Xenopus (Silurana) tropicalis pseudouridine synthase A mitochondrial-like (LOC100490752) transcript variant X3 mRNA	NM_004912623.1	0.68	3	2.15	0.032
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100485383 (LOC100485383) mRNA	NM_002935683.2	0.01	7.64	12.9	0.033
Xenopus laevis quinoid dihydroperoxide reductase (qdrp) mRNA	NM_001096335.1	0.48	11.26	4.56	0.033
Xenopus laevis finger protein 168 E3 ubiquitin protein ligase (rf168) mRNA	NM_001091654.1	3.47	7.72	1.15	0.034
Xenopus laevis WD repeat domain 24 (wd24) mRNA	NM_001087196.1	4.52	10.19	1.17	0.035
Xenopus laevis NKT1 substrate 1 (proline-rich) (akts1) mRNA	NM_001091309.1	8.99	19.65	1.13	0.035
Xenopus laevis HESEX homeobox 1 (hesx1) mRNA	NM_001162570.1	1.25	1.25	2.98	0.037
Xenopus laevis histone cluster 1-h2aj (hist12aj) mRNA	NM_001094479.1	0.48	3.47	2.86	0.037
Xenopus laevis hypothetical protein LOC00137665 mRNA (cDNA clone IMAGE:202803) partial cds	BC157474.1	4.01	14.32	1.84	0.037
Xenopus laevis finger protein 1 (fng1) mRNA	BC074103.1	8.61	38.2	2.15	0.038
PREDICTED: Xenopus (Silurana) tropicalis Rab12 member RAS oncogene family (rab12) mRNA	XM_00293745.2	0.62	2.01	1.71	0.038
Xenopus laevis hypothetical QCA94681 mRNA (cDNA clone IMAGE:58632775) partial cds	BC093542.1	1.02	5.17	2.35	0.039
Xenopus laevis solute carrier family 6 (amino acid transporter) member 14 gene 1 (slc6a14) mRNA	NM_001091185.1	0.47	1.9	2.02	0.042
Xenopus (Silurana) tropicalis carbonic anhydrase 6 (ca6) mRNA	NM_001079014.1	6.37	24.88	1.54	0.041
PREDICTED: Xenopus (Silurana) tropicalis integrin alpha 5 fibronectin receptor alpha polypeptide (itga5) mRNA	NM_002935659.2	1.49	3.73	1.33	0.041
Xenopus laevis stem cell leukemia protein SCL mRNA (cDNA clone MGIC79923 IMAGE:4175038) complete cds	BC072130.1	1.34	3.37	1.33	0.041
Xenopus laevis complete sequence	DQ096858.1	1.47	7.6	2.37	0.042
Xenopus laevis solute carrier family 6 (amino acid transporter) member 14 gene 2 (slc6a14) mRNA	NM_001091309.2	0.47	1.9	2.02	0.042
Xenopus (Silurana) tropicalis uncharacterized LOC00170494 (LOC00170494) mRNA	NM_001091650.1	0.98	3.96	2.15	0.043
Xenopus laevis ring finger protein 2 (sh3rf2) mRNA	NM_004916774.1	1.61	4.34	2.15	0.043
PREDICTED: Xenopus (Silurana) tropicalis cx9c motif-containing protein 4-like (LOC101731920) transcript variant X1 mRNA	NM_0029371766.2	1.09	6.15	2.54	0.044
PREDICTED: Xenopus (Silurana) tropicalis serine/threonine-protein kinase N1-like (LOC100493742) mRNA	NM_001093306.1	2.09	9.46	2.18	0.044
Xenopus laevis histone cluster 1-h2ad (hist12ad) mRNA	NM_001086236.1	4.6	10.35	1.17	0.044

Gene	Accession	Control <sup>1</sup>	Eya1-MO <sup>2</sup>	Eya1-MO FC <sup>3</sup>	q-value
PREDICTED: Xenopus (Silurana) tropicalis PR domain containing 1 with ZNF domain (pfdm1) transcript variant X2 mRNA	XM_004914548..1	6.57	13.69	1.06	0.045
Xenopus laevis trachyury and Tbx related protein mRNA (cDNA clone IMAGE3.28112) partial cds	BC130162..1	0.33	1.59	2.28	0.045
PREDICTED: Xenopus (Silurana) tropicalis synaptoosomal-associated protein 25-like (LOC100491227) transcript variant X1 mRNA	XM_002938586..2	1.49	4.3	1.53	0.045
Xenopus laevis peripheral myelin protein 22 (mpmp22) mRNA	NM_001093533..1	3.9	9.57	1.3	0.045
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC10036715 (LOC10036715) mRNA	XM_002938348..2	2.44	5.03	1.05	0.045
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100127805 (LOC100127805) mRNA	XM_002937441..2	0.79	3.77	2.26	0.046
PREDICTED: Xenopus (Silurana) tropicalis ENTH domain containing 2 (enthid2) transcript variant X1 mRNA	XM_0029240093..2	0.01	3.06	11.58	0.047
Xenopus laevis glutamic-oxaloacetic transaminase 1 soluble (got1) mRNA	NM_001087074..1	6.6	14.05	1.09	0.047
Xenopus laevis peroxisomal biogenesis factor 16 (pexf16) mRNA	NM_001091728..1	1.5	5.15	1.78	0.048
Xenopus laevis uncharacterized protein MGCG68754 (MGCG68754) mRNA	NM_001090147..1	2.17	7.29	1.75	0.048
PREDICTED: Xenopus (Silurana) tropicalis monocarboxylate transporter 9-like (LOC100485780) partial mRNA	XM_002937108..2	6.29	14.64	1.22	0.048
Xenopus laevis cDNA clone IMAGE3401727	BC09348..1	3.74	8.01	1.1	0.048
Xenopus (Silurana) tropicalis thioredoxin reductase 1 (txrd1) mRNA	NM_001256471..1	0.27	1.58	2.58	0.049
Xenopus laevis uncharacterized protein MGCI15496 (MGCI15496) mRNA	NM_001096598..1	8.95	18.58	1.05	0.049
XELTFIIA X.laevis 5S rRNA gene transcription factor (TFIIA) mRNA complete cds	KD2938..1	1.59	5.01	1.66	0.049
PREDICTED: Xenopus (Silurana) tropicalis histone H2B.1-like (LOC100492632) mRNA	XM_004919053..1	5.25	15.52	1.56	0.049

<sup>1</sup> Expression levels (FPKM) in un-injected pre-placodal explants

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Eya1-MO

<sup>3</sup> Log<sub>2</sub> Fold change values for Eya1-MO

## I.5 Six1-MO+Eya1-MO: Down-regulated genes

**Table I.5** Genes significantly down-regulated in both Six1MO and Eya1MO after injection of Six1MO or Eya1MO

Gene	Accession	Control <sup>b</sup>	Six1-MO <sup>c</sup> FC	Six1-MO <sup>c</sup> FC	Control <sup>d</sup>	Eya1-MO <sup>e</sup> FC	Eya1-MO <sup>e</sup> FC	q-value
Xenopus laevis tubulin folding co-factor E-like (tblc) mRNA	NM_001096263.1	28.39	0.01	-14.79	0.002	27.24	0.01	-14.73
Xenopus laevis cDNA clone MGCI:114987 (IMAGE:5084306 complete cds	BC098991.1	35.12	5.33	-2.72	0.002	24.37	0.01	-14.57
PREDICTED: Xenopus (Silurana) tropicalis phospholipase A2 inhibitor and Ly6/PLAUR domain-containing protein-like (LOC:1017330430)	XM_009919780.1	5.47	0.01	-12.42	0.002	5.27	0.01	-12.36
PREDICTED: Xenopus (Silurana) tropicalis histone H2A type 1-like (LOC:101733262) transcript variant X1 mRNA	XM_00991588.1	1.61	0.01	-10.65	0.002	1.56	0.01	-10.16
PREDICTED: Xenopus (Silurana) tropicalis oroferin (otof) mRNA	BC136162.1	1.29	0.01	-9.33	0.002	1.24	0.03	-5.9
Xenopus tropicalis hypothetical protein LOC:00125064 mRNA (cDNA clone IMAGE:7661075) partial cds	NM_001096765.1	1.19	0.01	-9.22	0.002	1.15	0.07	-4.09
Xenopus laevis Gd complex subunit 8 homolog (S. cerevisiae) (gld8) mRNA	XM_009915901.1	8.42	0.39	-4.44	0.002	8.15	1.94	-5.58
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC:00493999 (LOC:100493999) transcript variant X2 mRNA	BC087515.1	51.07	3.21	-3.99	0.002	59.51	13.64	-2.13
PREDICTED: Xenopus (Silurana) tropicalis clone (LOC:100494280) mRNA	XM_009938567.2	3.47	0.23	-3.92	0.002	3.73	0.79	-2.25
Xenopus laevis cDNA clone (LOC:100494282) mRNA	BC099592.1	4.09	0.36	-3.53	0.002	3.93	0.85	-2.21
Xenopus laevis hypothetical protein LOC:00125064 mRNA (cDNA clone IMAGE:7661075) partial cds	NM_001090073.1	23.31	0.07	-3.5	0.002	21.64	3.1	-2.8
XELO101 Xenopus laevis mRNA for pleiotrophic factor-beta1 complete cds	DA2059.1	10586.69	99.43	-3.41	0.002	173.23	81.7	-1.08
Xenopus laevis cysteine and histidine-rich protein 1A mRNA (cDNA clone IMAGE:5513813) partial cds	BC073380.1	347.25	36.26	-3.26	0.002	335.76	199.7	-0.75
PREDICTED: Xenopus (Silurana) tropicalis ADAMTS-like 1 (adams1t1) mRNA	XM_009910864.1	2.81	0.92	-1.62	0.002	1.81	0.23	-3.02
Xenopus laevis uncharacterized protein MGCA81434 mRNA	NM_001091189.1	53.69	69.59	-2.96	0.002	55.77	30.33	-0.86
PREDICTED: Xenopus (Silurana) tropicalis homeobox protein Dikka-like (LOC:100494171) mRNA	XM_009935712.2	15.94	2.09	-2.94	0.002	16.07	2.93	-2.46
Xenopus laevis uncharacterized protein MGCG907 (MGCG9077) mRNA	NM_001092839.1	23.31	5.59	-2.45	0.002	29.13	3.89	-2.91
Xenopus laevis mRNA for pleiotrophic factor-beta1 complete cds	NM_001092839.1	30.49	0.07	-3.41	0.002	173.23	81.7	-1.08
Xenopus laevis histidine-rich protein 1A mRNA (cDNA clone IMAGE:5513813) partial cds	NM_001092839.1	19.22	6.56	-1.55	0.002	17.76	2.7	-2.72
PREDICTED: Xenopus (Silurana) tropicalis 1-adamts1 mRNA	BC155883.1	4.11	17.36	2.08	0.002	2.17	14.12	2.71
Xenopus laevis uncharacterized protein MGCA81434 mRNA	NM_001091231.1	46.41	8.32	-2.48	0.002	46.21	7.05	-2.71
PREDICTED: Xenopus (Silurana) tropicalis 1-adamts1 mRNA	XM_009938125.2	163.56	25.98	-2.45	0.002	158.96	44.15	-1.85
Xenopus laevis uncharacterized protein MGCG84752 mRNA	DQ09616.1	55.64	89.1	-2.64	0.002	514.45	106.56	-2.27
Xenopus laevis uncharacterized protein MGCS3587 (MGCS3587) mRNA	NM_001088606.1	3.38	1.05	-1.7	0.002	3.3	0.6	-2.47
Xenopus laevis uncharacterized protein LOC:00127287 mRNA (cDNA clone IMAGE:38222377) partial cds	NM_001085951.1	12.69	5.8	-1.13	0.002	12.17	2.22	-2.72
Xenopus laevis orofelin gene 1 (orfn1-1-a) mRNA	BC155883.1	4.11	17.36	2.08	0.002	2.17	14.12	2.71
Xenopus laevis uncharacterized protein MGCA81434 mRNA	NM_001091231.1	46.41	8.32	-2.48	0.002	46.21	7.05	-2.71
PREDICTED: Xenopus (Silurana) tropicalis 1-adamts1 mRNA	XM_009938125.2	163.56	25.98	-2.45	0.002	158.96	44.15	-1.85
Xenopus laevis uncharacterized protein MGCG84752 mRNA	DQ09616.1	55.64	89.1	-2.64	0.002	514.45	106.56	-2.27
Xenopus laevis potassium voltage-gated channel shaker related subfamily member 4 (kcnna4) mRNA	NM_001088606.1	3.38	1.05	-1.7	0.002	3.3	0.6	-2.47
Xenopus (Silurana) tropicalis hyaluronoglucosaminidase 4 (hyal4) mRNA	NM_001097397.1	12.69	5.8	-1.13	0.002	12.17	2.22	-2.72
Xenopus (Silurana) tropicalis uncharacterized LOC:01730819 (LOC:101730819) mRNA	NM_009915204.1	47.93	8.69	-2.46	0.002	43.07	12.97	-1.73
PREDICTED: Xenopus (Silurana) tropicalis brain-specific angiogenesis inhibitor 1 (bai1) mRNA	XM_02934468.2	11.29	2.07	-2.45	0.002	10.87	2.93	-1.89
PREDICTED: Xenopus (Silurana) transmembrane protease serine 2-like (LOC:101731874) transcript variant X3 mRNA	XM_009912208.1	5.58	0.03	-2.44	0.002	4.81	0.98	-2.29
PREDICTED: Xenopus (Silurana) transcript variant X2 mRNA	XM_009916721.1	1.65	0.34	-2.29	0.002	1.62	0.5	-1.71
PREDICTED: Xenopus (Silurana) transcript variant X2 receptor-like (LOC:00487051) transcript variant X2 mRNA	BC074386.1	2.775	5.74	-2.28	0.002	1.4	3.96	0.34
Xenopus laevis novel gene 1 (orfn1-1-a) mRNA	NM_009915332.1	9.5	1.98	-2.27	0.002	9.18	4.07	-1.17
Xenopus laevis novel gene 1 (orfn1-1-a) mRNA	NM_001091841.1	9.86	2.05	-2.27	0.002	10.81	3.46	-1.65
Xenopus laevis ribosomal protein S6 kinase 90 kDa polypeptide 6 (psks6) mRNA	NM_001096223.1	25.85	7.83	-1.72	0.002	24.85	5.34	-2.22
Xenopus laevis stomatin (EPB72) like 3 (stom3) mRNA	NM_001095036.1	29.33	12.38	-1.24	0.002	24.33	5.32	-2.19
Xenopus laevis glutathione S-transferase omega 1 (gsto1) mRNA	DQ09616.1	2189.8	659.24	-1.73	0.002	544.49	218.0	-0.02
Xenopus laevis complete sequence	NM_001097291.1	7.59	2.55	-1.58	0.002	7.02	1.57	-0.02
Xenopus (Silurana) tropicalis superillin (svil) mRNA	NM_001097291.1	97.77	23.4	-2.06	0.002	87.37	38.98	-1.16
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC:00487159 (LOC:100487159) transcript variant X3 mRNA	BC158326.1	22.66	-2.04	0.002	363.33	160.95	-1.17	
Xenopus tropicalis novel protein similar to human angiopoietin-1 (angptl) mRNA (cdna clone MGCG7546975)	NM_001097237.1	10.9	3.29	-1.73	0.002	11.12	2.79	-2.002
PREDICTED: Xenopus (Silurana) transcript variant X3 mRNA	XM_029355863.2	6.05	1.52	-2	0.002	5.69	1.6	-1.83
Xenopus laevis uncharacterized LOC:00158303 (LOC:100158303) mRNA	NM_001094434.1	18.37	4.63	-1.99	0.002	18.97	4.36	-1.58
PREDICTED: Xenopus (Silurana) tropicalis alpha-1-N-acetylglucosaminyltransferase-like (LOC:00493555) mRNA	NM_001127299.1	17.08	4.33	-1.98	0.002	28.64	15.24	-0.91
PREDICTED: Xenopus (Silurana) transcript variant X2 mRNA	XM_009914651.1	9.42	3.31	-1.51	0.002	9.05	2.76	-1.72
Xenopus tropicalis novel protein similar to human angiopoietin-1 (angptl) mRNA (cdna clone MGCG7546975)	NM_001092481.1	15.16	5.57	-1.45	0.002	15.89	4.29	-1.89
Xenopus laevis testis derived transcript (3 LIM domains) (tes) mRNA	NM_001091112.1	37.87	10.24	-1.89	0.002	36.91	15.21	-1.28
Xenopus laevis transcript variant X3 (soc3) mRNA	NM_00113158.1	2.32	0.64	-1.86	0.002	2.23	0.74	-1.61
Xenopus laevis uncharacterized LOC:00489259 (LOC:100489259) mRNA	NM_001094043.1	6.47	1.89	-1.78	0.002	6.39	2.09	-1.62
Xenopus laevis uncharacterized LOC:00158303 (LOC:100158303) mRNA	NM_001094434.1	163.09	56.29	-1.53	0.002	153.83	45.66	-0.02
PREDICTED: Xenopus (Silurana) transcript variant X2 mRNA	XM_009914651.1	9.42	3.31	-1.51	0.002	9.05	2.76	-1.72
Xenopus (Silurana) tropicalis cold shock domain containing C2 RNA binding (csc2) mRNA	NM_001092481.1	5.05	1.63	-1.64	0.002	4.47	1.88	-1.25
PREDICTED: Xenopus (Silurana) transcript variant X2 mRNA	XM_009944851.2	7.08	3.76	-0.91	0.002	8.86	2.22	-0.02
Xenopus laevis uncharacterized LOC:00127867 (LOC:100127867) mRNA	BC068645.1	75.99	49.64	-0.61	0.002	8.62	2.92	-1.56
Xenopus laevis uncharacterized LOC:00127867 (LOC:100127867) mRNA	CR05533.2	3.81	1.3	-1.55	0.002	3.72	1.58	-1.23

Gene	Accession	Control <sup>a</sup>	Six1-MO FC	Six1-MO q-value	Control <sup>a</sup>	Six1-MO FC	Six1-MO q-value
Xenopus tropicalis hypothetical protein LOC548657 mRNA (cDNA clone MGC:186334 IMAGE:8959997) complete cds	BC161440.1	19.79	7.33	-1.43	0.002	18.82	6.46
Xenopus laevis synaptotagmin-like 1 (sytl1) mRNA [LOC:100158280] (LOC:100158280) mRNA	NM_001093193.1	15.92	5.53	-1.53	0.002	15.3	5.38
Xenopus laevis anion channel characterized [LOC:100158280] (LOC:100158280) mRNA	NM_001127737.1	75.24	29.67	-1.34	0.002	46.32	16.9
Xenopus laevis calcium/calmodulin-dependent protein kinase I gamma L subunit mRNA complete cds; nuclear gene for mitochondrial product	AF233632.1	12.05	4.43	-1.44	0.002	11.56	4.42
Xenopus laevis adenine nucleotide translocase (Ant1) mRNA complete cds; nuclear gene for mitochondrial product	AF231347.1	2942.75	1108.3	-1.41	0.002	1460.74	676.03
Xenopus laevis pyruvate carboxylase gene 1 (pc1) mRNA	NM_00108977.1	62.53	23.88	-1.39	0.002	61.41	30.84
Xenopus (Silurana) tropicalis Golgi membrane protein 1-like (LOC:100485898) transcript variant X1 mRNA	XM_00297888.2	77.33	37.05	-1.06	0.002	39.76	15.27
Xenopus laevis solute carrier family 2 (facilitated glucose transporter) member 12 (slc2a12) mRNA	NM_001096426.1	39.61	15.4	-1.36	0.002	38.39	17.18
Xenopus (Silurana) tropicalis branched chain amino-acid transaminase 2 mitochondrial (lbcat2)	NM_001067391.1	3.25	12.67	-1.36	0.002	31.56	15
PREDICTED: Xenopus (Silurana) tropicalis mucin-4-like (LOC:10048533.9) mRNA	XM_002934058.2	15.17	6.88	-1.14	0.002	14.6	5.78
Xenopus laevis myeloid differentiation primary response 88 (myd88-3b) mRNA	NM_001095786.1	23.45	9.27	-1.34	0.002	23.22	-1.12
Xenopus laevis synaptotagmin-like 2 (sytl2) mRNA	NM_001094552.1	67.1	31.2	-1.1	0.002	64.84	25.64
Xenopus laevis mRNA for calcineurin catalytic subunit complete cds	AB037146.1	24.26	10.46	-1.27	0.002	23.3	9.31
Xenopus laevis cDNA clone IMAGE:13587 partial cds	BC085045.1	64.49	25.99	-1.31	0.002	48.07	23.62
Xenopus laevis hatching gland-like Xhel protein (LOC398485) mRNA	NM_001088991.1	414.37	169.66	-1.29	0.002	364.23	180.66
Xenopus (Silurana) tropicalis gramicine nucleotide binding protein (G protein) gamma 12 (gng12) mRNA	NM_001026951.1	42.5	17.36	-0.29	0.002	41.12	21.97
Xenopus laevis small nuclear RNA activating complex poly(A)nuclease 5' nuclease (snanc5-b) mRNA	NM_001171809.1	59.76	24.61	-1.28	0.002	75.41	41.15
Xenopus laevis hypothetical protein LOC:10012737 mRNA (cDNA clone IMAGE:7980696) partial cds	BC155957.1	164.19	73.25	-1.16	0.002	156.61	66.89
Xenopus laevis RING finger domain protein Np95 mRNA complete cds	EU177101.1	37.4	16.22	-1.21	0.002	16.16	7.27
Xenopus laevis PDZ domain containing family member 2 (gipz2) mRNA	NM_001088817.1	31.99	13.93	-1.2	0.002	30.54	14.3
Xenopus laevis cDNA clone IMAGE:6634291	BC127414.1	15.26	6.63	-1.2	0.002	15.09	7.54
PREDICTED: Xenopus (Silurana) tropicalis NPA-like domain containing 1 (nipal1) mRNA	XM_002933460.2	28.66	12.64	-1.18	0.002	27.54	-0.71
Xenopus laevis uncharacterized LOC:100158314 (LOC:100158314) mRNA	NM_001127766.1	121.97	57.13	-1.09	0.002	119.37	53.3
Xenopus laevis finished cDNA clone Theul24a018	CR848315.2	19.09	10.19	-0.91	0.002	18.56	8.29
Xenopus laevis RING finger domain protein Np95 mRNA complete cds	NM_001086825.1	40.36	19.75	-1.03	0.002	40.48	18.25
Xenopus laevis cDNA clone IMAGE:11147.1	XM_00109411147.1	18.14	8.43	-1.11	0.002	17.18	8.22
Xenopus laevis mRNA for putative XRG protein	AJ278067.1	3161.52	1464	-1.11	0.002	1360.33	723.8
Xenopus laevis hypothetical LOC:100158314 (LOC:100158314) mRNA	BC083037.1	11.36	5.36	-1.08	0.002	11.29	5.37
Xenopus laevis solute carrier family 16 member 12 (monocarboxylate transporter 12) (scf16a12) mRNA	NM_001277651.1	124.32	71.86	-0.79	0.002	71.66	33.82
Xenopus laevis ATPase Ca++ transporting ubiquitous (atp2a3) mRNA	NM_001095094.1	72.9	34.65	-1.07	0.002	70.51	37.2
Xenopus tropicalis finished cDNA clone TPA012b11	CR760877.2	128.9	61.95	-1.06	0.002	124.76	66.39
Xenopus laevis customer protein complex subunit gamma 1 (cpg1) mRNA	NM_001090106.1	132.9	64.99	-1.03	0.002	128.2	83.31
Xenopus laevis cDNA clone IMAGE:993951 mRNA	XM_002939691.2	6.1	1.1	-0.89	0.002	5.88	0.04
XL202535 Xenopus laevis epithelial sodium channel alpha subunit (alphaENaC) mRNA complete cds	U2355.1	35.54	2.35	-0.02	0.002	152.64	14.38
Xenopus laevis RIPPLY3 mRNA for RIPPLY3 protein complete cds	AB552086.1	11.8	6.61	-2.88	0.002	11.3	1.09
Xenopus laevis DNA clone IMAGE:8642610	BC153809.1	5.69	0.85	-2.74	0.002	5.49	0.82
PREDICTED: Xenopus (Silurana) synaptotagmin-13-like (LOC:100490466) mRNA	XM_002944648.2	4.5	0.91	-2.31	0.002	4.41	0.68
PREDICTED: Xenopus (Silurana) fibrocytin-L-like (LOC:100486760) mRNA	NM_002942313.2	21.29	8.08	-1.4	0.019	21.12	3.31
Xenopus laevis ADP-ribosylation hydrolase (adrph) mRNA	NM_001094795.1	3.38	1.21	-1.49	0.003	3.53	0.53
PREDICTED: Xenopus (Silurana) tropicanth spermatic protein (theq) transcript variant X1 mRNA	XM_002939761.2	9.7	4.38	-1.15	0.003	9.63	1.52
PREDICTED: Xenopus (Silurana) tropicalis circularly permuted Ras protein 1-like (LOC:100491292) transcript variant X2 mRNA	XM_009916298.1	5.49	2.2	-1.32	0.003	7.74	1.29
Xenopus laevis B glycoprotein (rbgb1) mRNA	NM_001093683.1	7.67	2.16	-1.83	0.002	7.37	1.24
PREDICTED: Xenopus (Silurana) homeobox B1 (hoxb1) transcript variant X1 mRNA	XM_002938017.2	6.61	2.44	-1.44	0.003	6.42	1.37
Xenopus laevis putative transmembrane protein TA-2 mRNA complete cds	AF31008.1	479.18	149.5	-1.68	0.002	737.17	165.26
Xenopus laevis tubulin alpha 1b (tuba1b) transcript variant 1 mRNA	XM_002935108.2	14.73	6.25	-1.24	0.004	14.2	2.76
Xenopus laevis ribosome-binding protein 1 (rbp1) mRNA	NM_001089623.1	45.2	23.76	-0.93	0.006	58.86	12
Xenopus laevis TMEM16a mRNA complete cds	EU367938.1	3.73	1.47	-1.35	0.036	3.59	0.75
PREDICTED: Xenopus (Silurana) tropicanth characterized LOC:100487159 (LOC:100487159) transcript variant X2 mRNA	XM_009918158.1	6.08	2.19	-1.48	0.016	5.81	1.22
PREDICTED: Xenopus (Silurana) tropicanth homeobox B1 (hoxb1) transcript variant X1 mRNA	NM_001085722.1	12.55	3.76	-1.74	0.003	12.08	2.1
Xenopus laevis putative transmembrane protein TA-2 mRNA complete cds	BC084133.1	14.32	6.4	-1.16	0.021	13.61	2.59
PREDICTED: Xenopus (Silurana) tropicanth 1 receptor accessory protein (ilrp1) mRNA	XM_002935108.2	14.73	6.25	-1.24	0.004	14.2	2.76
Xenopus laevis sepihin 5 mRNA	NM_00112051.1	3.66	1.42	-1.36	0.003	3.95	0.92
PREDICTED: Xenopus (Silurana) tropicanth 1 receptor accessory protein (ilrp1) mRNA	NM_001097043.1	8.18	3.39	-1.27	0.003	7.88	1.88
PREDICTED: Xenopus (Silurana) tropicanth 1 receptor accessory protein (ilrp1) mRNA	NM_001088829.1	13.19	4.83	-1.45	0.002	12.75	3.05
Xenopus laevis putative transmembrane protein TA-2 mRNA complete cds	NM_001142661.1	12.68	6.51	-0.96	0.003	27.61	6.65
Xenopus laevis putative transmembrane protein TA-2 mRNA complete cds	NM_001113953.2	3.26	1.14	-1.52	0.029	9.55	2.58
Xenopus laevis tropicanth 1 receptor accessory protein (ilrp1) mRNA	BC142545.1	226.57	95.3	-1.25	0.002	225.15	-1.85

Gene	Accession	Control <sup>a</sup>	Six1-MO FC	Six1-MO q-value	Control <sup>a</sup>	Six1-MO FC	Six1-MO q-value
PREDICTED: Xenopus (Silurana) tropicalis coiled-coil domain containing 170 (ccdc170) mRNA	XM_002936375.2	12.05	6.15	<0.97	0.003	11.79	3.32
PREDICTED: Xenopus (Silurana) tropicalis cysteine-rich protein 2-like (LOC101734738) mRNA	XM_004910879.1	17.42	6.18	<1.5	0.002	17.47	<1.8
Xenopus laevis adaptor-related protein 3 beta 1 subunit (ap3b1) mRNA	NM_001091840.1	17.45	8.26	<1.03	0.003	17.11	<1.79
Xenopus laevis cDNA clone IMAGE63165722	BC071055.1	11.41	5.85	<0.97	0.031	11.08	<1.78
Xenopus laevis secreted protein acidic cysteine-rich (osteonectin) (sparc) mRNA	NM_001086121.1	19.51	9.01	<1.12	0.003	18.73	<1.76
Xenopus laevis dorsal inhibitory axon guidance protein (draxin) mRNA	NM_002937523.1	6.87	3.3	<1.06	0.003	6.85	<1.76
Xenopus laevis scinderin (scin) mRNA	NM_001092620.1	10.42	3.23	<1.69	0.002	10.04	<2.99
Xenopus laevis leucine-rich transmembrane protein 3 (lrit3) mRNA	NM_001095561.1	15.91	8.17	<0.96	0.013	15.05	<4.52
Xenopus laevis secreted protein acidic cysteine-rich (osteonectin) (sparc) mRNA	NM_001093131.1	12.57	5.48	<1.2	0.004	14.95	<4.66
Xenopus laevis T-box 1 (tbx1-b) mRNA	NM_001096675.1	15.88	7.32	<1.12	0.003	15.07	<4.7
Xenopus laevis spidil8 mRNA complete sequence	DQ096570.1	111.4	42.4	<1.39	0.007	117.49	<38.32
Xenopus laevis GII pathogenesis-related 2 (gipr2) mRNA	NM_001095072.1	29.88	13.66	<1.13	0.002	30.59	<10.01
Xenopus tropicalis finished cDNA clone Theu06511	CR926197.2	9.23	4.35	<1.09	0.016	8.64	<2.84
Xenopus laevis exp44 mRNA complete sequence	DQ096616.1	16156.88	6135.58	<1.4	0.002	15508.61	<3380.5
Xenopus laevis similar to solute carrier family 27 (fatty acid transporter) member 2 mRNA (cDNA clone IMAGE4681491) partial cds	BC041746.1	40.46	19.16	<1.08	0.002	40.02	<13.82
PREDICTED: Xenopus (Silurana) tropicalis family with sequence similarity 169 member A (fam169a) mRNA	NM_002934825.2	15.52	6.72	<1.21	0.003	12.74	<4.54
Xenopus laevis T-box 1 (tbx1-b) mRNA	AY046070.1	7.41	3.45	<1.1	0.003	7.06	<2.55
Xenopus laevis sphingosine-1-phosphate phosphatase 2 (spp2) mRNA	NM_001097668.1	14.56	6.44	<1.18	0.002	17	<6.2
PREDICTED: Xenopus (Silurana) tropicalis nuclear receptor subfamily 2 group F member 5-like (LOC100491959) transcript variant X1 mRNA	NM_002938463.2	55.49	25.86	<1.1	0.002	86.58	<31.94
Xenopus laevis exp44 mRNA complete sequence	BC098355.1	84.75	41.75	<1.02	0.003	83.9	<31.06
Xenopus laevis similar to solute carrier family 27 (fatty acid transporter) member 2 mRNA (cDNA clone IMAGE4681491) partial cds	NM_001094239.1	34.74	20.19	<0.78	0.012	32.18	<12.5
Xenopus laevis solute carrier family 39 (zinc transporter) member 9 (scs39a9-a) mRNA	NM_001091036.1	58.36	30.2	<0.95	0.002	83.13	<32.23
PREDICTED: Xenopus (Silurana) tropicalis protein phosphatase 1 regulatory subunit 12A-like (LOC100487727) mRNA	NM_002937684.2	11.53	5.91	<0.96	0.015	8.11	<4.27
Xenopus laevis tubulin beta class 1 (tubb1) mRNA	NM_001087257.1	129.91	81.22	<0.68	0.049	96.88	<38.3
PREDICTED: Xenopus (Silurana) tropicalis SPFG complex locus (spgf) mRNA	NM_002933963.2	13.33	8.75	<0.61	0.003	10.47	<4.17
Xenopus laevis clone IMAGE65871841	NM_001091009.1	24.98	12.83	<0.96	0.009	24.01	<9.67
Xenopus laevis solute carrier family 39 (zinc transporter) member 9 (scs39a9-a) mRNA	NM_001092726.1	26.99	12.7	<0.99	0.002	26.18	<11.1
PREDICTED: Xenopus (Silurana) tropicalis protein phosphatase 1 regulatory subunit 12A-like (LOC100487727) mRNA	NM_001086800.1	111	68.07	<0.71	0.003	106.35	<46.75
Xenopus laevis tubulin beta class 1 (tubb1) mRNA	NM_00142168.1	75.73	42.83	<0.82	0.003	66.57	<29.76
PREDICTED: Xenopus (Silurana) tropicalis ADP-ribosylation factor-like protein 11-like (LOC100490310) mRNA	NM_001092888.1	40.96	21.11	<0.96	0.002	37.93	<17.53
Xenopus laevis adenylyl kinase 3 (ak3) nuclear gene encoding mitochondrial protein mRNA	NM_002935735.2	19.77	7.86	<1.33	0.004	19.25	<4.88
Xenopus laevis adenylyl kinase 3 (ak3) nuclear gene encoding mitochondrial protein mRNA	NM_001095977.1	6.01	2.42	<1.31	0.004	6.04	<2.43
PREDICTED: Xenopus (Silurana) tropicalis EPSE-like 2 (eps8l2) transcript variant X1 mRNA	NM_002937505.2	3.43	1.47	<1.23	0.006	10.75	<3.35
Xenopus (Silurana) tropicalis chromosome 12 open reading frame 4 (ct2orf4) mRNA	NM_00107513.1	11.32	2.27	<2.32	0.002	10.9	<1.9
Xenopus (Silurana) tropicalis CAs1 domain containing 1 (casd1) mRNA	NM_00142168.1	5.87	2.32	<1.34	0.007	5.9	<1.09
Xenopus laevis adenosine kinase (adk) mRNA	NM_001092888.1	13.61	6.57	<1.54	0.002	12.17	<2.75
PREDICTED: Xenopus (Silurana) tropicalis membrane-Inserted (mmp24) mRNA	NM_001093573.5	33.23	8.57	<1.96	0.006	23.1	<7.21
Xenopus laevis matrix metalloproteinase 14 (membp24) mRNA	NM_001095977.1	6.01	2.42	<1.31	0.004	6.04	<2.43
PREDICTED: Xenopus (Silurana) tropicalis EPSE-like 2 (eps8l2) transcript variant X1 mRNA	NM_002937505.2	3.43	1.47	<1.23	0.006	10.75	<3.35
Xenopus (Silurana) tropicalis chromosome 12 open reading frame 4 (ct2orf4) mRNA	NM_00107513.1	11.32	2.27	<2.32	0.002	10.9	<1.9
Xenopus (Silurana) tropicalis ATPase class VI type 11A (atp11a) mRNA	NM_002933015.2	5.87	2.32	<1.34	0.007	5.9	<1.09
Xenopus laevis matrix metalloproteinase-24 (membp24) mRNA	NM_001093636.1	13.61	6.57	<1.54	0.002	12.17	<2.75
PREDICTED: Xenopus (Silurana) tropicalis characterized LOC100497755 (LOC100497755) mRNA	NM_001097158.1	3.32	1.31	<1.31	0.004	6.04	<2.43
Xenopus laevis Stat1 mRNA complete cds	AY101602.1	5781.13	302.07	<0.94	0.006	529.14	<227.89
Xenopus (Silurana) tropicalis Meis homeobox 2 (meis2) mRNA	NM_001097218.1	12.32	5.07	<1.28	0.007	4.46	<2.46
Xenopus laevis protein disulfide isomerase family A member 4 (pdia4) mRNA	NM_001094862.1	80.67	43.74	<0.88	0.007	80.24	<34.4
Xenopus laevis thiole intracellular channel 3 (cliC3) mRNA	NM_001092748.1	22.9	9.45	<1.28	0.007	21.58	<7.02
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101733661 (LOC101733661) mRNA	NM_001094836.1	10.92	4.88	<1.16	0.008	8.7	<3.1
Xenopus laevis UDP-GalbetaGlcNAc beta 1-4-galactosyltransferase poly peptide 4 (b4galt4) mRNA	NM_001092811.1	5.03	1.69	<1.47	0.047	1.79	<0.17
Xenopus laevis growth arrest-specific 6 (gas6) mRNA	NM_001089668.1	16.61	7.6	<1.13	0.010	38.25	<17.62
PREDICTED: Xenopus (Silurana) tropicalis transmembrane protease serine 2-like (LOC101731505) mRNA	NM_001094858.1	54.02	29.82	<0.86	0.012	7.87	<3.11
Xenopus laevis sperm domain containing 2 (mospd2) mRNA	NM_0010912203.1	11.48	4.26	<1.43	0.013	11.21	<3.91
PREDICTED: Xenopus (Silurana) tropicalis inhibitor of DNA binding 4 (dominant negative helix-loop-helix protein (id4)-a) mRNA	NM_001095457.1	5.39	1.8	<1.59	0.007	5.05	<1.39
PREDICTED: Xenopus (Silurana) tropicalis arthrinactivating protein 28 (arthrap28) transcript variant X1 mRNA	NM_001087235.1	13.99	6.31	<1.15	0.009	13.48	<5.35
Xenopus laevis UDP-GalbetaGlcNAc beta 1-4-galactosyltransferase poly peptide 4 (b4galt4) mRNA	NM_002938054.2	1.22	0.37	<1.72	0.014	2.59	<0.51
Xenopus laevis protein disulfide isomerase family A member 4 (pdia4) mRNA	NM_001098091.1	95.05	55.47	<0.78	0.014	97.23	<47.82
Xenopus laevis galactose 3-O-sulfotransferase 4 (gal3t4) mRNA	NM_001086139.1	2.44	0.75	<1.7	0.015	2.22	<0.52
PREDICTED: Xenopus (Silurana) tropicalis silk homology 2 (Drosophila) (slit2) transcript variant X1 mRNA	NM_0010926715.2	19.99	10.13	<0.98	0.004	18.85	<8.62
PREDICTED: Xenopus (Silurana) tropicalis rho GTPase activating protein B1 (ephb1) mRNA	NM_002935075.2	2.89	1.19	<1.28	0.016	2.87	<0.71
XL38225: Xenopus laevis glutamic acid decarboxylase (GAD) mRNA complete cds	U38225.1	2.2	0.87	<1.35	0.026	2.15	<0.62
Xenopus laevis mRNA for fibronectin leucine rich transmembrane protein 3 (flrt3) gene	A1605776.1	5.73	2.47	<1.22	0.010	5.51	<1.82

Gene	Accession	Control <sup>1</sup>	Six1-MO FC	q-value	Control <sup>4</sup>	Six1-MO FC	q-value	Eya1-MO FC	q-value
Xenopus tropicalis finished cDNA clone TNeu033k10	CR760083.2	4.15	-3.17	0.016	4.5	0.63	-2.84	0.016	
PREDICTED: Xenopus (Silurana) tropicalis lysophosphatidic acid receptor 3 (lpars3) mRNA	XM_00931673.2	2.69	-0.95	-1.51	0.018	3.37	0.45	-2.92	0.018
Xenopus laevis glutathione S-transferase theta 1 (gstt1) mRNA	NM_001091734.1	30.16	9.88	-1.61	0.004	25.63	7.91	-1.7	0.018
Xenopus laevis cDNA clone IMAGE:3397065	BC088743.1	13.61	6.65	-1.03	0.018	13	5.43	-1.26	0.018
PREDICTED: Xenopus (Silurana) tropicalis interleukin 17 receptor E (il17e) mRNA	NM_0094115.1	3.1	0.22	-3.84	0.019	5.02	2.29	-1.13	0.049
PREDICTED: Xenopus (Silurana) tropicalis WNK lysine deficient protein kinase 4 (wnk4) mRNA	XM_00922519.2	3.92	1.71	-1.2	0.002	4.77	1.52	-1.66	0.019
PREDICTED: Xenopus (Silurana) tropicalis claudin-4-like (LOC101732192) mRNA	XM_00911749.1	1.79	0.36	-2.33	0.020	3.28	0.96	-1.78	0.020
Xenopus laevis secreted frizzled-related protein 2 (frzrp2) mRNA	NM_001087194.1	32.5	12.04	-1.43	0.002	39.84	13.77	-1.53	0.020
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100494783 (LOC100494783) partial mRNA	XM_00942126.2	2.39	0.87	-1.47	0.021	2.78	0.71	-1.97	0.021
PREDICTED: Xenopus (Silurana) tropicalis UDP-N-acetylgalactosamine:polypeptide N-acetylgalactosaminyltransferase 3	XM_00936748.2	15.97	9.04	-0.82	0.035	15.23	6.85	-1.15	0.022
Xenopus laevis uncharacterized protein MGCT8986 (MGCT8986) mRNA	NM_001091630.1	30.18	16.16	-0.9	0.004	73.09	36.45	-1	0.022
Xenopus (Silurana) tropicalis myoferlin (myof) mRNA	NM_001128651.1	10.17	5.39	-0.92	0.042	25.47	10.01	-1.35	0.023
Xenopus laevis sigma protein mRNA (cDNA clone MGCT80311 IMAGE:5073822) complete cds	BC072179.1	4.35	1.83	-1.25	0.024	1.46	0.52	-1.52	0.024
PREDICTED: Xenopus (Silurana) tropicalis myosin VI (myo6) transcript variant X7 mRNA	NM_0094520.1	11.91	6.48	-0.88	0.033	10.6	5.2	-1.03	0.025
Xenopus laevis zearin (ezr) mRNA	NM_001093923.1	21.87	12.64	-0.79	0.026	34.29	16.1	-1.09	0.026
PREDICTED: Xenopus (Silurana) tropicalis Hh1-like 1 (hh1pl1) mRNA	XM_00939668.2	1.71	0.55	-1.65	0.035	1.64	0.31	-2.42	0.026
Xenopus laevis small T-cell differentiation protein (natl) mRNA	NM_001086577.1	4.65	2.77	-0.75	0.038	4.59	0.64	-2.85	0.028
Xenopus laevis hypothetical protein LOC100494141 mRNA (cDNA clone IMAGE:6320207) partial cds	BC134824.1	4.13	1.55	-1.42	0.006	4.83	1.55	-1.64	0.029
Xenopus laevis aldehyde dehydrogenase 3 family member B1 (aldh3b1) mRNA	NM_001093704.1	16.77	7.6	-1.14	0.002	15.94	6.9	-1.21	0.029
Xenopus laevis shamboid veinlet-like 2 (rbhd2) mRNA	NM_001094850.1	3.16	1.1	-1.53	0.030	3.08	0.64	-2.27	0.030
Xenopus laevis activating transcription factor 5 gene 2 (atf5.2) mRNA	NM_001093530.1	3.6	1.28	-1.49	0.024	3.51	1.07	-1.71	0.030
PREDICTED: Xenopus (Silurana) tropicalis zinc finger protein 474-like (LOC100494841) transcript variant X1 mRNA	XM_00931728.2	12.84	5.91	-1.12	0.026	11.23	3.99	-1.49	0.030
Xenopus laevis uncharacterized protein MGCT154351 (MGCT154351) mRNA	NM_001096931	25.63	11.11	-1.21	0.013	19.98	8.21	-1.28	0.030
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC101732938 (LOC101732938) mRNA	XM_009413985.1	7.6	4.93	-0.63	0.026	7.05	3.4	-1.05	0.030
Xenopus laevis putative beta-N-acetylhexosaminidase mRNA partial cds	JN127371.1	17.58	8.92	-0.98	0.030	17.38	7.75	-1.17	0.030
Xenopus laevis phosphoserine phosphatase (psph) mRNA	NM_00109325.1	9.76	4.64	-1.07	0.013	9.39	4.12	-1.19	0.032
Xenopus laevis phosphodiesterase 6/cGMP-specific cone gamma (pcde6h) mRNA	NM_001092763.1	1.13	0.3	-1.91	0.038	1.08	0.12	-3.25	0.035
PREDICTED: Xenopus (Silurana) tropicalis ras and EF-hand domain-containing protein homolog (LOC101732297) mRNA	NM_009413126.1	1.1	0.36	-1.62	0.036	6.03	2.09	-1.53	0.036
Xenopus laevis putative protein MGCT154351 (MGCT154351) mRNA	XM_009411907.1	1.3	-1.11	0.018	2.72	1.15	-1.25	0.037	
Xenopus laevis neuronal differentiation 1 (neurod1-b) mRNA	NM_001092127.1	4.98	1.51	-1.72	0.007	4.75	1.25	-1.93	0.038
Xenopus laevis ion peptidase 2 (peroxisomal (lonp2)) mRNA	NM_001096479.1	779.96	75.25	-3.37	0.002	744.79	11.42	-6.03	0.039
PREDICTED: Xenopus (Silurana) tropicalis chromosomal unknown open reading frame human C14orf37 (LOC10049715)	XM_009417220.1	2.47	0.84	-1.57	0.040	2.39	0.63	-1.93	0.040
Xenopus laevis protein phosphatase 3 catalytic subunit alpha isoform (pp3ca) mRNA	NM_001093903.1	5.36	2.56	-1.07	0.047	5.21	2.19	-1.25	0.041
Xenopus laevis receptor protein tyrosine phosphatase CTP-alpha mRNA partial cds	AF198450.1	3.8	1.5	-1.34	0.045	3.52	1.39	-1.34	0.045
Xenopus laevis hypothetical LOC43666 mRNA (cDNA clone IMAGE:511185)	BC07564.1	19.39	0.21	-6.56	0.015	18.66	0.19	-6.62	0.048
Xenopus laevis impact protein mRNA (cDNA clone IMAGE:7010848)	BC084977.1	10.11	6.75	-0.58	0.002	53.72	8.61	-2.64	0.048

<sup>1</sup> Expression levels (FPKM) in un-injected pre-placodal explants<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1-MO<sup>3</sup> Log<sub>2</sub> Fold change values for Six1-MO<sup>4</sup> Expression levels (FPKM) in un-injected pre-placodal explants<sup>5</sup> Expression levels (FPKM) in pre-placodal explants after injection of Eya1-MO<sup>6</sup> Log<sub>2</sub> Fold change values for Eya1-MO

## I.6 Six1-MO+Eya1-MO: Up-regulated genes

**Table I.6** Genes significantly up-regulated in both Six1-MO and Eya1-MO after injection of Six1-MO or Eya1-MO

Gene	Accession	Control <sup>a</sup>	Six1-MO <sup>b</sup>	Six1-MO <sup>c</sup>	q-value	Control <sup>d</sup>	Eya1-MO <sup>e</sup>	Eya1-MO <sup>f</sup>	q-value
Xenopus laevis hypothetical protein MGc68493 mRNA (cDNA clone MGc83264 [IMAGE63748]) complete cds	BC068970.1	0.01	216.93	11.73	0.002	39.10	81.65	1.06	0.003
PREDICTED: Xenopus (Slurana) tropicalis ENTH domain containing 2 (enth2) transcript variant X1 mRNA	XM_00254093.2	0.01	3.09	11.59	0.002	0.00	3.05	11.58	0.002
Xenopus tropicalis finished cDNA clone Theud23c10	XM_002532556.1	0.01	2.16	11.07	0.002	0.00	1.91	10.9	0.003
Xenopus laevis TAR (HIV-1) RNA binding protein 2 (tarbp2) mRNA	CT03610.1	7521.61	406206.07	9.08	0.002	7247.24	455649.11	9.3	0.002
Xenopus laevis CDNA clone XM_002540931	NM_001092105.1	54.7	4845.82	6.47	0.002	47.14	276.98	2.55	0.003
XELXLOTA Xenopus laevis mRNA for pleiotrophic factor-beta2 complete cds	BC08431.1	3.28	221.78	6.08	0.002	7.63	242.21	1.67	0.002
Xenopus laevis 5'-nucleotidase domain containing 3 (n5dc3) mRNA	D42060.1	11.17	24.26	1.12	0.002	4.47	191.22	5.42	0.002
Xenopus laevis molybdenum cofactor synthesis 1 (mocs1) mRNA	NM_001092732.1	5.15	162.64	4.98	0.002	6.44	67.13	3.38	0.026
Xenopus tropicalis cDNA clone IMAGE757230	BC16631.1	2905.49	79667.79	4.77	0.002	2.89	1.09	6.16	0.002
Xenopus laevis uncharacterized LOC495221 (LOC495221) mRNA	NM_001094903.1	22.84	34.18	0.58	0.002	280.09	244665.50	3.13	0.002
Xenopus laevis tumor protein p53 inducible protein 11 (tp53i11) mRNA	NM_001097917.1	5.82	17.87	1.62	0.002	1.82	245.57	3.76	0.002
Xenopus laevis Phosphoinositide 3-kinase regulatory subunit 5 mRNA (cDNA clone IMAGE4968567) partial cds	BC072301.1	0.29	2.69	3.25	0.002	0.34	1.95	2.53	0.016
Xenopus laevis 5'-nucleotidase domain containing 4 (n5dc4) mRNA	NM_001094603.1	3.83	34.16	3.16	0.002	7.72	191.15	1.31	0.002
Xenopus laevis molybdenum containing 4 (m5dc4) mRNA	NM_001097126.1	3.76	33.19	3.14	0.002	11.68	22.66	0.96	0.008
PREDICTED: Xenopus (Slurana) tropicalis cytoskeleton associated protein 2 (ckap2) transcript variant X2 mRNA	NM_004911970.1	0.31	2.49	3.04	0.002	0.35	2.83	3.01	0.025
Xenopus laevis ATPase Na <sup>+</sup> /K <sup>+</sup> -transporting beta 2 polypeptide (atpb2) mRNA	NM_001092728.1	5.07	40.96	3.02	0.002	2.20	8.02	1.87	0.003
Xenopus laevis serum/glucocorticoid-regulated kinase 1 (sak1-a) mRNA	NM_001090340.1	95.63	690.44	2.85	0.002	88.72	581.69	2.71	0.002
Xenopus laevis s-CAD protein 3 (Gill blood group) mRNA	BC14663.1	0.85	5.59	2.72	0.002	0.81	3.89	2.26	0.010
Xenopus laevis aquaporin 3 (Gill blood group) mRNA	NM_001094477.1	3.3	2.16	2.71	0.002	3.17	106.9	1.75	0.002
Xenopus laevis solute carrier family 16 (monocarboxylic acid transporters) member 6 (slc16a6) mRNA	NM_001086838.1	3.47	16.39	2.24	0.002	16.27	19.65	2.63	0.002
Xenopus laevis ornithine decarboxylase 1 (ODC1) mRNA	BC047954.1	173.45	627.81	1.86	0.002	162.87	1002.00	2.62	0.002
PREDICTED: Xenopus (Slurana) tropicalis C-reactive protein-like (LOC10489750) mRNA	NM_002537750.2	0.92	4.87	2.41	0.002	1.40	4.17	1.57	0.002
Xenopus laevis phosphatidylinositol glycan anchor biosynthesis class F (pigf) mRNA	NM_001095344.1	23.51	118.12	2.33	0.002	23.69	68.18	1.52	0.003
Xenopus laevis aquaporin 3 (CDN3) mRNA	NM_002536842.2	0.79	2.88	1.87	0.002	0.26	1.14	2.13	0.002
Xenopus tropicalis mRNA (GCF160980 IMAGE8319613) complete cds	BC14653.1	0.85	5.59	2.72	0.002	0.81	3.89	2.26	0.010
Xenopus laevis solute carrier family 16 (monocarboxylic acid transporters) member 6 (slc16a6) mRNA	NM_001098638.1	3.3	2.16	2.71	0.002	3.17	106.9	1.75	0.002
Xenopus laevis ornithine decarboxylase 1 (ODC1) mRNA	BC047954.1	173.45	627.81	1.86	0.002	162.87	1002.00	2.62	0.002
PREDICTED: Xenopus (Slurana) tropicalis C-reactive protein-like (LOC10489750) mRNA	NM_002537750.2	0.92	4.87	2.41	0.002	1.40	4.17	1.57	0.002
Xenopus laevis s-CAD protein 3 (Gill blood group) mRNA	BC14663.1	0.85	5.59	2.72	0.002	0.81	3.89	2.26	0.010
Xenopus laevis aquaporin 3 (Gill blood group) mRNA	NM_001094477.1	3.3	2.16	2.71	0.002	3.17	106.9	1.75	0.002
Xenopus laevis solute carrier family 16 (monocarboxylic acid transporters) member 6 (slc16a6) mRNA	NM_001086838.1	3.47	16.39	2.24	0.002	16.27	19.65	2.63	0.002
Xenopus laevis ornithine decarboxylase 1 (ODC1) mRNA	BC047954.1	173.45	627.81	1.86	0.002	162.87	1002.00	2.62	0.002
PREDICTED: Xenopus (Slurana) tropicalis C-reactive protein-like (LOC10489750) mRNA	NM_002537750.2	0.92	4.87	2.41	0.002	1.40	4.17	1.57	0.002
Xenopus laevis s-CAD protein 3 (Gill blood group) mRNA	BC14663.1	0.85	5.59	2.72	0.002	0.81	3.89	2.26	0.010
Xenopus laevis aquaporin 3 (Gill blood group) mRNA	NM_001094477.1	3.3	2.16	2.71	0.002	3.17	106.9	1.75	0.002
Xenopus laevis ornithine decarboxylase 1 (ODC1) mRNA	BC047954.1	173.45	627.81	1.86	0.002	162.87	1002.00	2.62	0.002
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PREDICTED: Xenopus (Slurana) tropicalis C-reactive protein-like (LOC10489750) mRNA	NM_002537750.2	0.92	4.87	2.41	0.002	1.40	4.17	1.57	0.002
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Xenopus laevis ornithine decarboxylase 1 (ODC1) mRNA	BC047954.1	173.45	627.81	1.86	0.002	162.87	1002.00	2.62	0.002
PREDICTED: Xenopus (Slurana) tropicalis C-reactive protein-like (LOC10489750) mRNA	NM_002537750.2	0.92	4.87	2.41	0.002	1.40	4.17	1.57	0.002
Xenopus laevis s-CAD protein 3 (Gill blood group) mRNA	BC14663.1								

Gene	Accession	Control <sup>t</sup>	Six1-MO <sup>c</sup>	Six1-MO <sup>e</sup>	Six1-MO <sup>f</sup>	Control <sup>t</sup>	Eya1-MO <sup>c</sup>	Eya1-MO <sup>e</sup>	Eya1-MO <sup>f</sup>	q-value	q-value
Xenopus laevis tropicalis mediator of RNA polymerase II transcription subunit 1 (med1) mRNA	NM_001097258.1	9.48	22.36	1.24	0.002	26.24	48.19	0.88	0.008		
Xenopus laevis protein phosphatase 1 catalytic subunit gamma isozyme (pp1cc) mRNA	NM_001087839.1	10.23	24.06	1.23	0.002	9.72	22.38	1.2	0.019		
X. laevis ODC mRNA for ornithine decarboxylase (EC 4.1.1.17)	X56316.1	30.56	77.03	1.06	0.002	35.50	83.035	1.23	0.002		
Xenopus laevis kinase muscle (pkm) mRNA	NM_00109872.1	9.51	21.64	1.19	0.002	9.21	20.71	1.17	0.002		
Xenopus tropicalis hypothetical protein LOC549562 mRNA (cDNA clone MGC:121609 IMAGE:7623213) complete cds	BC135795.1	18.06	39.76	1.14	0.002	23.90	37.68	0.66	0.020		
Xenopus laevis sp1 protein mRNA (cDNA clone MGC:155227) complete cds	BC072773.1	0.01	7.07	12.79	0.002	0.00	21.16	14.37	0.003		
Xenopus laevis pyruvate kinase muscle C member 28 (dkajc28) mRNA	NM_001015977.1	121.77	206	0.86	0.003	117.45	88408.92	9.56	0.003		
Xenopus laevis characterized LOC495410 (LOC495410) mRNA	NM_001095067.1	0.75	6.76	3.19	0.003	0.30	133.06	8.78	0.003		
Xenopus laevis sequence protein (hsatrs) mRNA	NM_001088359.1	4.39	26.15	2.57	0.002	4.12	46.25	3.49	0.003		
PREDICTED: Xenopus (Silurana) tropicalis synthase related sequence protein (hsatrs) mRNA	XM_004918268.1	9.6	50.77	2.4	0.003	14.49	138.01	3.25	0.003		
PREDICTED: Xenopus (Silurana) tropicalis ectodysplasin A2 receptor (edazar) transcript variant X1 mRNA	NM_002938915.2	0.63	4.44	2.83	0.002	0.61	5.66	3.21	0.003		
Xenopus laevis 5'-nucleotidase cytosolic-IIIa (nt5c3) mRNA	NM_001094563.1	0.93	3.26	1.83	0.003	0.42	2.67	2.67	0.003		
Xenopus laevis 3'P-B mRNA complete cds	FJ649495.1	0.98	4.13	2.08	0.002	0.99	5.67	2.52	0.003		
Xenopus laevis hypothetical protein LOC100127319 mRNA (cDNA clone IMAGE:8319360) partial cds	BC155933.1	30.44	114.86	1.92	0.002	27.55	144.64	2.39	0.003		
Xenopus laevis hypothetical protein MGCI30860 mRNA (cDNA clone MGCI30860 IMAGE:7205580) complete cds	BC106331.1	93.36	307.66	1.72	0.001	89.70	440.08	2.29	0.003		
Xenopus laevis nuclear gene encoding mitochondrial protein mRNA	NM_001097842.1	3.98	9.16	1.21	0.003	3.83	15.47	2.01	0.003		
X. laevis mRNA for Xom protein	X8454.1	4.32	11.98	1.47	0.003	4.82	19.03	1.98	0.003		
Xenopus laevis pituitary homeobox gene 1 paired-like homeodomain transcription factor (Pttx1) mRNA complete cds	AF217647.1	5.5	18.43	1.75	0.002	5.35	20.00	1.9	0.003		
Xenopus laevis 1k (nk3-1-a) mRNA	NM_001086738.1	3.19	8.91	1.48	0.003	5.78	21.35	1.89	0.003		
Xenopus laevis Kruppel-like factor 10 (klf10) mRNA	NM_001095871.1	6.63	17.58	1.41	0.002	5.38	18.94	1.82	0.003		
Xenopus laevis cone-rod homeobox (crx-b) mRNA	NM_001088447.1	8.28	27.18	1.72	0.002	7.70	26.08	1.76	0.003		
Xenopus laevis acid phosphatase 1 soluble (acp-1) transcript variant b mRNA	NM_00114769.1	6.18	17.15	1.47	0.002	5.94	19.80	1.74	0.003		
Xenopus laevis regulator of calcineurin 1 (rcan1) mRNA	NM_001086391.1	8.69	16.91	0.96	0.003	8.40	28.02	1.74	0.003		
PREDICTED: Xenopus (Silurana) tropicalis carbonic anhydrase A4 (hoxa4) mRNA	XW_004915397.1	5.74	9.17	0.67	0.003	5.52	16.37	1.57	0.003		
Xenopus laevis mRNA for Tia1n complete cds	AB075925.1	69.12	195.84	1.5	0.002	34.04	99.68	1.55	0.003		
Xenopus laevis cone-rod homeobox (crx-b) mRNA	NM_001096966.1	10.68	23.12	1.12	0.003	10.33	28.34	1.46	0.003		
Xenopus laevis transcript variant b mRNA	NM_001087349.1	9.41	13.41	0.51	0.002	10.47	23.94	1.19	0.003		
Xenopus laevis characterized LOC100036942 (LOC100036942) mRNA	NM_001097712.1	2.34	3.32	0.5	0.002	1.87	4.14	1.15	0.003		
Xenopus (Silurana) tropicalis carbonic anhydrase e (ca6) mRNA	NM_001079014.1	3.81	22.7	2.57	0.004	6.37	24.88	1.97	0.004		
Xenopus laevis hypothetical LOC445668 mRNA (cDNA clone IMAGE:7982507)	BC108429.1	6.21	16.05	1.37	0.004	6.10	15.36	1.33	0.004		
Xenopus laevis gravin-like (gl) mRNA complete cds	AF308810.1	2.59	5.21	1.01	0.006	2.18	5.67	1.38	0.006		
Xenopus laevis cyclin G1 (cnrg1) mRNA	NM_001091163.1	0.01	221.41	17.76	0.006	0.00	167.99	17.36	0.006		
Xenopus laevis tumor suppressor protein p53 mRNA (cDNA clone MGCI79985 IMAGE:6864775) complete cds	BC084641.1	26.43	54.53	1.04	0.006	25.70	50.99	0.99	0.033		
Xenopus laevis odd-paired-like (opl) mRNA complete cds	AF28805.1	2.32	6.54	1.5	0.007	2.30	7.49	1.71	0.007		
Xenopus laevis amyloid beta (A4) precursor-like protein 1 (abp1) mRNA	NM_001095950.1	2.75	8.28	1.59	0.007	1.67	8.38	2.33	0.008		
Xenopus laevis snykin repeat and SOCS box containing 1 (abp1) mRNA	NM_001096257.1	1.09	3.03	1.47	0.008	1.01	4.67	2.21	0.008		
Xenopus laevis hypothetical LOC495232 mRNA (cDNA clone IMAGE:55191906) partial cds	BC084651.1	17.27	63.75	1.88	0.002	0.91	3.58	1.97	0.008		
Xenopus laevis mRNA for Zic-related-2 complete cds	AB014461.1	2	5.17	1.38	0.019	1.01	2.00	6.83	0.008		
PREDICTED: Xenopus (Silurana) tropicalis transformation/transcription domain-associated protein (trap)	XM_002931947.2	2.78	5.99	1.11	0.010	2.68	6.75	1.33	0.008		
X. laevis mRNA for transcription factor VENT-2	X98349.1	3.1	8.73	1.49	0.009	3.45	11.50	1.74	0.009		
Xenopus laevis cryptochrome 1 (photolyase-like) mRNA (cDNA clone MGCI79851 IMAGE:6636878) complete cds	BC072120.1	3.05	6.69	1.14	0.009	12.67	26.53	1.07	0.009		
PREDICTED: Xenopus (Silurana) tropicalis SET and WD domain containing 1 (abp1) mRNA	XW_004915396.2	1.09	2.99	3.23	0.036	0.23	4.48	3.44	0.010		
Xenopus tropicalis finished cDNA clone TEG049047-p004	CU075595.1	0.45	1.23	1.46	0.035	0.43	1.65	1.65	0.009		
Xenopus laevis hypothetical protein LOC432279 mRNA (cDNA clone IMAGE:4680368) partial cds	AB005292.2	2.8	6.83	1.29	0.002	2.86	7.92	1.47	0.010		
Xenopus laevis programmed cell death 6 mRNA (cDNA clone MGCI32114 IMAGE:6549231) complete cds	BC110939.1	19.03	39.35	1.05	0.010	1.65	13.72	3.06	0.010		
Xenopus laevis solute carrier family 6 (amino acid transporter) member 14 gene 2 (slc6a14.2) mRNA	NM_001091185.1	0.53	2.52	2.27	0.010	0.47	1.90	2.02	0.012		
Xenopus laevis mRNA for Zic2 protein complete cds	AB00956.1	1.84	6.01	1.71	0.010	1.83	7.30	2	0.010		
Xenopus laevis MARCKS-like protein mRNA (cDNA clone IMAGE:6864261) mRNA	BC157454.1	18.9	36.19	0.94	0.030	20.00	47.45	1.25	0.011		
Xenopus laevis mRNA for Zic3 protein complete cds	AB005292.2	2.42	5.74	1.25	0.006	1.42	4.64	1.71	0.015		
Xenopus laevis programmed cell death 6 mRNA (cDNA clone MGCI32114 IMAGE:6549231) complete cds	XM_004917879.1	1	3.31	1.73	0.029	0.86	3.56	2.04	0.016		
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100145470 (LOC100145470) transcript variant X2 mRNA	BC074103.1	28.23	49.73	0.82	0.016	8.61	38.19	2.15	0.016		
Xenopus laevis mRNA for Zic2 protein complete cds	BC055964.1	0.26	1.86	2.87	0.018	0.24	3.18	3.74	0.018		
Xenopus laevis MARCKS-like protein mRNA (cDNA clone IMAGE:6864261) mRNA	BC079827.1	16.33	58.98	1.85	0.018	14.70	32.07	1.12	0.018		
Xenopus laevis mRNA for Zic3 protein complete cds	BC072283.1	0.17	4.65	4.81	0.019	1.57	11.39	2.86	0.022		
PREDICTED: Xenopus (Silurana) tropicalis uncharacterized LOC100145470 (LOC100145470) transcript variant Y2 mRNA	AY648303.1	0.53	2.52	2.26	0.019	0.51	3.64	2.85	0.019		

Gene	Accession	Control <sup>1</sup>	Six1-MO <sup>2</sup>	Six1-MO <sup>3</sup>	Six1-MO <sup>4</sup>	q-value	Control <sup>4</sup>	Eya1-MO <sup>5</sup>	Eya1-MO <sup>6</sup>	q-value
			F <sub>c</sub>	F <sub>c</sub>	F <sub>c</sub>			F <sub>c</sub>	F <sub>c</sub>	
Xenopus (Silurana) tropicalis Zic family member 3 (zic3) mRNA	NM_001005691.1	2.06	4.83	1.23	0.019	1.59	4.13	1.38	0.019	
Xenopus laevis Runt-related transcription factor 1 mRNA (cDNA clone MGC:69003 IMAGE:4963637) complete cds	BC057739.1	0.38	1.08	1.54	0.045	0.34	1.42	2.06	0.019	
PREDICTED: Xenopus (Silurana) tropicalis G protein-coupled receptor family C group 5 member C (gprc5c)	XW_002934984.2	1.22	3.19	1.39	0.018	1.16	3.54	1.61	0.019	
Xenopus tropicalis finished cDNA clone Thieu03 In03	CR855460.2	1.76	4.33	1.3	0.015	1.69	4.98	1.56	0.019	
XELXOCT79 X.laevis xct-79 mRNA 3' end	M60076.1	3.8	8.37	1.14	0.004	6.80	15.74	1.21	0.020	
Xenopus laevis ePH receptor B3 ephb3 mRNA	NM_001095726.1	7.85	16.6	1.08	0.002	7.54	16.24	1.11	0.020	
Xenopus laevis protein idas (midaes) mRNA	NM_001127804.1	52.7	105.03	0.99	0.002	54.50	142.88	1.39	0.022	
Xenopus laevis purinergic receptor P2Y G-protein coupled 10 (p2ry10) mRNA	NM_001094846.1	0.32	2.27	2.87	0.024	0.30	4.42	3.9	0.024	
Xenopus (Silurana) tropicalis sestrin 1 (sesn1) mRNA	NM_001129939.1	3.17	5.87	0.89	0.034	3.54	7.99	1.17	0.025	
Xenopus laevis mucolipin 1 (mcoln1) mRNA	NM_001092218.1	17.74	27.05	0.61	0.014	12.85	26.91	1.07	0.025	
Xenopus laevis gap junction protein beta 2 26kDa (gapj2) mRNA	NM_001087009.1	1.42	3.53	1.32	0.028	1.36	4.07	1.58	0.028	
Xenopus laevis mRNA for Taf-1beta2 complete cds	AB022692.1	359	727.52	1.02	0.004	348.84	788.61	1.18	0.028	
X.laevis mRNA for glucokinase	X93494.1	0.28	1.23	2.13	0.030	0.23	1.20	2.37	0.030	
Xenopus laevis brachyury (T) mRNA (cDNA clone MGC:78815 IMAGE:3402478) complete cds	BC072031.1	0.92	3.64	1.99	0.032	0.86	3.37	1.98	0.020	
Xenopus laevis ring finger protein 168 E3 ubiquitin protein ligase (rnf168) mRNA	NM_001091654.1	3.71	7.92	1.09	0.014	3.47	7.71	1.15	0.034	
Xenopus (Silurana) tropicalis uncharacterized LOC100170494 (LOC100170494) mRNA	NM_001130274.1	1.61	2.67	0.73	0.037	0.66	3.96	2.58	0.037	
Xenopus laevis brachyury and Tbx related protein mRNA (cDNA clone IMAGE:3328112) partial cds	BC130162.1	0.35	1.42	2.04	0.038	0.33	1.59	2.28	0.045	
Xenopus laevis peroxisomal biogenesis factor 16 (dex1) mRNA	NM_001091728.1	1.48	4.65	1.65	0.021	1.50	5.14	1.78	0.048	

<sup>1</sup> Expression levels (FPKM) in un-injected pre-placodal explants

<sup>2</sup> Expression levels (FPKM) in pre-placodal explants after injection of Six1-MO

<sup>3</sup> Log<sub>2</sub> Fold change values for Six1-MO

<sup>4</sup> Expression levels (FPKM) in un-injected pre-placodal explants

<sup>5</sup> Expression levels (FPKM) in pre-placodal explants after injection of Eya1-MO

<sup>6</sup> Log<sub>2</sub> Fold change values for Eya1-MO

## Appendix J. Raw embryo counts for MO statistical analysis

**Table J.1** Number of genes affected after either *Six1*-MO or *Eya1*-MO injection compared to *Eya1*-mmMO

	<i>Six1</i> -MO			<i>Eya1</i> -MO			<i>Eya1</i> -mmMO		
	<i>Knocked-down</i>	<i>Un-affected</i>	<i>p-value</i>	<i>Knocked-down</i>	<i>Un-affected</i>	<i>p-value</i>	<i>Knocked-down</i>	<i>Un-affected</i>	<i>p-value</i>
<i>Atoh1</i>	20	6	***	18	2	**	2	19	
<i>Gfi1</i>	14	3	0.0016	18	9	0.0135	8	18	
<i>Hes5</i>	26	9	**	29	6	**	4	20	
<i>Hes9</i>	33	12	**	25	8	**	3	24	
<i>Is2</i>	25	13	0.0068	17	0	**	6	16	
<i>Ngn1</i>	33	18	***	21	22	0.0099	4	20	
<i>Pou4f1.2</i>	32	19	0.0009	16	11	0.0056	3	16	
<i>Sox2</i>	14	5	**	14	4	**	1	15	
<i>Sox3</i>	17	8	**	14	12	0.0017	2	20	
<i>Tlx1</i>	26	5	0.0017	29	3	**	5	10	
<i>N-tub</i>	53	29	0.0835	10	0	0.0041	9	12	

\*\* Denotes a p value < 0.0001