

Contents

Glossary	xii
Acronyms	xiv
1 Introduction and Literature Review	1
1.1 Cancer Research in the Post-Genomic Era	1
1.1.1 Cancer is a Global Health Issue	2
1.1.1.1 The Genetics and Molecular Biology of Cancers	3
1.1.2 The Genomics Revolution in Cancer Research	3
1.1.2.1 High-Throughput Technologies	4
1.1.2.2 Bioinformatics and Genomic Data	5
1.1.3 Genomics Projects	5
1.1.3.1 The Cancer Genome Project	6
1.1.3.2 The Cancer Genome Atlas Project	6
1.1.4 Genomic Cancer Medicine	8
1.1.4.1 Cancer Genes and Driver Mutations	8
1.1.4.2 Precision Cancer Medicine	9
1.1.4.3 Molecular Diagnostics and Pan-Cancer Medicine	9
1.1.4.4 Targeted Therapeutics and Pharmacogenomics	10
1.1.5 Systems and Network Biology	11
1.2 Synthetic Lethal Cancer Medicine	12
1.2.1 Synthetic Lethal Genetic Interactions	12
1.2.2 Synthetic Lethal Concepts in Genetics	14
1.2.3 Synthetic Lethality in Model Systems	14
1.2.3.1 Synthetic Lethal Pathways and Networks	15
1.2.3.2 Evolution of Synthetic Lethality	15
1.2.4 Synthetic Lethality in Cancer	16
1.2.5 Clinical Impact of Synthetic Lethality in Cancer	18
1.2.6 High-throughput Screening for Synthetic Lethality	19
1.2.6.1 Synthetic Lethal Screens	21
1.2.7 Computational Prediction of Synthetic Lethality	22
1.2.7.1 Bioinformatics Approaches to Genetic Interactions	22
1.2.7.2 Comparative Genomics	24
1.2.7.3 Analysis and Modelling of Protein Data	26
1.2.7.4 Differential Gene Expression	28
1.2.7.5 Data Mining and Machine Learning	29

1.2.7.6	Mutual Exclusivity and Bimodality	31
1.2.7.7	Rationale for Further Development	33
1.3	E-cadherin as a Synthetic Lethal Target	33
1.3.1	The <i>CDH1</i> gene and its Biological Functions	33
1.3.1.1	Cytoskeleton	34
1.3.1.2	Extracellular and Tumour Micro-environment	34
1.3.1.3	Cell-Cell Adhesion and Signalling	34
1.3.2	<i>CDH1</i> as a Tumour (and Invasion) Suppressor	35
1.3.2.1	Breast Cancers and Invasion	35
1.3.3	Hereditary Diffuse Gastric (and Lobular Breast) Cancer	35
1.3.4	Cell Line Models of <i>CDH1</i> Null Mutations	37
1.4	Summary and Research Direction of Thesis	37
1.4.1	Thesis Aims	39
2	Methods and Resources	40
2.1	Bioinformatics Resources for Genomics Research	40
2.1.1	Public Data and Software Packages	40
2.1.1.1	Cancer Genome Atlas Data	41
2.1.1.2	Reactome and Annotation Data	42
2.2	Data Handling	42
2.2.1	Normalisation	42
2.2.2	Sample Triage	43
2.2.3	Metagenes and the Singular Value Decomposition	43
2.2.4	Candidate Triage and Integration with Screen Data	45
2.3	Techniques	46
2.3.1	Statistical Procedures and Tests	46
2.3.2	Gene Set Over-representation Analysis	47
2.3.3	Clustering	47
2.3.4	Heatmap	47
2.3.5	Modelling and Simulations	48
2.3.5.1	Receiver Operating Characteristic Curves	49
2.3.6	Resampling Analysis	49
2.4	Pathway Structure Methods	50
2.4.1	Network and Graph Analysis	50
2.4.2	Sourcing Graph Structure Data	51
2.4.3	Constructing Pathway Subgraphs	51
2.4.4	Network Analysis Metrics	52
2.5	Implementation	53
2.5.1	Computational Resources and Linux Utilities	53
2.5.2	R Language and Packages	54
2.5.3	High Performance and Parallel Computing	57
3	Methods Developed During Thesis	59
3.1	A Synthetic Lethal Detection Methodology	59
3.2	Synthetic Lethal Simulation and Modelling	61
3.2.1	A Model of Synthetic Lethality in Expression Data	62

3.2.2	Simulation Procedure	66
3.3	Detecting Simulated Synthetic Lethal Partners	69
3.3.1	Binomial Simulation of Synthetic Lethality	69
3.3.2	Multivariate Normal Simulation of Synthetic Lethality	71
3.3.2.1	Multivariate Normal Simulation with Correlated Genes	73
3.3.2.2	Specificity with Query-Correlated Pathways	81
3.4	Graph Structure Methods	83
3.4.1	Upstream and Downstream Gene Detection	83
3.4.1.1	Permutation Analysis for Statistical Significance	84
3.4.1.2	Hierarchy Based on Biological Context	84
3.4.2	Simulating Gene Expression from Graph Structures	85
3.5	Customised Functions and Packages Developed	90
3.5.1	Synthetic Lethal Interaction Prediction Tool	90
3.5.2	Data Visualisation	91
3.5.3	Extensions to the iGraph Package	93
3.5.3.1	Sampling Simulated Data from Graph Structures	93
3.5.3.2	Plotting Directed Graph Structures	93
3.5.3.3	Computing Information Centrality	94
3.5.3.4	Testing Pathway Structure with Permutation Testing	94
3.5.3.5	Metapackage to Install iGraph Functions	95
4	Synthetic Lethal Analysis of Gene Expression Data	96
4.1	Synthetic Lethal Genes in Breast Cancer	97
4.1.1	Synthetic Lethal Pathways in Breast Cancer	98
4.1.2	Expression Profiles of Synthetic Lethal Partners	100
4.1.2.1	Subgroup Pathway Analysis	103
4.2	Comparing Synthetic Lethal Gene Candidates	105
4.2.1	Primary siRNA Screen Candidates	105
4.2.2	Comparison with Correlation	105
4.2.3	Comparison with Primary Screen Viability	108
4.2.4	Comparison with Secondary siRNA Screen Validation	110
4.2.5	Comparison to Primary Screen at Pathway Level	111
4.2.5.1	Resampling Genes for Pathway Enrichment	113
4.2.6	Integrating Synthetic Lethal Pathways and Screens	118
4.3	Synthetic Lethal Pathway Metagenes	119
4.4	Replication in Stomach Cancer	121
4.5	Discussion	122
4.5.1	Strengths of the SLIPT Methodology	122
4.5.2	Synthetic Lethal Pathways for E-cadherin	123
4.5.3	Replication and Validation	125
4.5.3.1	Integration with siRNA Screening	125
4.5.3.2	Replication across Tissues	126
4.6	Summary	126

5	Synthetic Lethal Pathway Structure	128
5.1	Synthetic Lethal Genes in Reactome Pathways	128
5.1.1	The PI3K/AKT Pathway	129
5.1.2	The Extracellular Matrix	131
5.1.3	G Protein Coupled Receptors	134
5.1.4	Gene Regulation and Translation	134
5.2	Network Analysis of Synthetic Lethal Genes	136
5.2.1	Gene Connectivity and Vertex Degree	137
5.2.2	Gene Importance and Information Centrality	138
5.2.3	Gene Importance and PageRank Centrality	139
5.3	Upstream or Downstream Synthetic Lethality	141
5.3.1	Measuring Structure of Candidates within PI3K	141
5.3.2	Resampling for Synthetic Lethal Pathway Structure	143
5.4	Discussion	145
5.5	Summary	147
6	Simulation and Modelling of Synthetic Lethal Pathways	149
6.1	Synthetic Lethal Detection Methods	150
6.1.1	Performance of SLIPT and χ^2 across Quantiles	151
6.1.1.1	Correlated Query Genes affects Specificity	154
6.1.2	Alternative Synthetic Lethal Detection Strategies	156
6.1.2.1	Correlation for Synthetic Lethal Detection	157
6.1.2.2	Testing for Bimodality with BiSEp	158
6.2	Simulations with Graph Structures	159
6.2.1	Performance over Graph Structures	160
6.2.1.1	Simple Graph Structures	160
6.2.1.2	Constructed Graph Structures	163
6.2.2	Performance with Inhibitions	165
6.2.3	Synthetic Lethality across Graph Structures	171
6.2.4	Performance within a Simulated Human Genome	174
6.3	Simulations in More Complex Graph Structures	179
6.3.1	Simulations over Pathway-based Graphs	180
6.3.2	Pathway Structures in a Simulated Human Genome	182
6.4	Discussion	185
6.4.1	Simulation Procedure	185
6.4.2	Comparing Methods with Simulated Data	186
6.4.3	Design and Performance of SLIPT	187
6.4.4	Simulations from Graph Structures	189
6.5	Summary	190
7	Discussion	192
7.1	Synthetic Lethality and <i>CDH1</i> Biology	192
7.1.1	Established Functions of <i>CDH1</i>	193
7.1.2	The Molecular Role of <i>CDH1</i> in Cancer	193
7.2	Significance	194
7.2.1	Synthetic Lethality in the Genomic Era	194

7.2.2	Clinical Interventions based on Synthetic Lethality	196
7.3	Future Directions	197
7.4	Conclusions	199
	Bibliography	201
A	Sample Quality	225
A.1	Sample Correlation	225
A.2	Replicate Samples in TCGA Breast Cancer Data	227
B	Software Used for Thesis	231
C	Mutation Analysis in Breast Cancer	240
C.1	Synthetic Lethal Genes and Pathways	240
C.2	Synthetic Lethal Expression Profiles	243
C.3	Comparison to Primary Screen	246
C.3.1	Resampling Analysis	248
C.4	Compare SLIPT genes	250
D	Metagene Analysis	252
D.1	Pathway Signature Expression	252
D.2	Somatic Mutation	261
D.3	Synthetic Lethal Reactome Metagenes	262
D.4	Expression of Somatic Mutations	264
E	Intrinsic Subtyping	267
F	Stomach Expression Analysis	269
F.1	Synthetic Lethal Genes and Pathways	269
F.2	Comparison to Primary Screen	273
F.2.1	Resampling Analysis	275
F.3	Metagene Analysis	277
G	Synthetic Lethal Genes in Pathways	278
H	Network Analysis for Mutation SLIPT	285
I	Pathway Structure for Mutation SLIPT	288
J	Performance of SLIPT and χ^2	290
J.1	Correlated Query Genes affects Specificity	296
K	Simulations on Graph Structures	302
K.0.1	Simulations from Inhibiting Graph Structures	303
K.1	Simulation across Graph Structures	306
K.2	Simulations from Complex Graph Structures	310
K.2.1	Simulations from Complex Inhibiting Graphs	313
K.3	Simulations from Pathway Graph Structures	319

List of Figures

1.1	Synthetic genetic interactions	13
1.2	Synthetic lethality in cancer	17
2.1	Read count density	44
2.2	Read count sample mean	44
3.1	Framework for synthetic lethal prediction	60
3.2	Synthetic lethal prediction adapted for mutation	61
3.3	A model of synthetic lethal gene expression	63
3.4	Modelling synthetic lethal gene expression	64
3.5	Synthetic lethality with multiple genes	65
3.6	Simulating gene function	67
3.7	Simulating synthetic lethal gene function	67
3.8	Simulating synthetic lethal gene expression	68
3.9	Performance of binomial simulations	70
3.10	Comparison of statistical performance	70
3.11	Performance of multivariate normal simulations	72
3.12	Simulating expression with correlated gene blocks	74
3.13	Simulating expression with correlated gene blocks	75
3.14	Synthetic lethal prediction across simulations	77
3.15	Performance with correlations	78
3.16	Comparison of statistical performance with correlation structure	79
3.17	Performance with query correlations	80
3.18	Statistical evaluation of directional criteria	81
3.19	Performance of directional criteria	82
3.20	Simulated graph structures	86
3.21	Simulating expression from a graph structure	87
3.22	Simulating expression from graph structure with inhibitions	88
3.23	Demonstration of violin plots with custom features	92
3.24	Demonstration of annotated heatmap	92
3.25	Simulating graph structures	94
4.1	Synthetic lethal expression profiles of analysed samples	101
4.2	Comparison of SLIPT with siRNA	106
4.3	Comparison of SLIPT and siRNA genes with correlation	106
4.4	Comparison of SLIPT and siRNA genes with correlation	108
4.5	Comparison of SLIPT and siRNA genes with screen viability	109

4.6	Comparison of SLIPT genes with siRNA screen viability	109
4.7	Resampled intersection of SLIPT and siRNA candidate genes	114
5.1	Synthetic lethality in the PI3K cascade	130
5.2	Synthetic lethality in Elastic Fibre Formation	132
5.3	Synthetic lethality in Fibrin Clot Formation	133
5.4	Synthetic lethality in the GPCRs	135
5.5	Synthetic lethality and vertex degree	137
5.6	Synthetic lethality and centrality	139
5.7	Synthetic lethality and PageRank	140
5.8	Structure of synthetic lethality resampling in PI3K	142
6.1	Performance of χ^2 and SLIPT across quantiles	152
6.2	Performance of χ^2 and SLIPT across quantiles with more genes	153
6.3	Performance of χ^2 and SLIPT across quantiles with query correlation	154
6.4	Performance of χ^2 and SLIPT across quantiles with query correlation and more genes	155
6.5	Performance of negative correlation and SLIPT	158
6.6	Simple graph structures	161
6.7	Performance of simulations on a simple graph	162
6.8	Performance of simulations is similar in simple graphs	163
6.9	Performance of simulations on a pathway	164
6.10	Performance of simulations on a simple graph with inhibition	166
6.11	Performance is higher on a simple inhibiting graph	168
6.12	Performance of simulations on a constructed graph with inhibition	169
6.13	Performance is affected by inhibition in graphs	170
6.14	Detection of synthetic lethality within a graph structure	172
6.15	Performance of simulations including a simple graph	176
6.16	Performance on a simple graph improves with more genes	177
6.17	Performance on an inhibiting graph improves with more genes	178
6.18	Performance of simulations on the PI3K cascade	181
6.19	Performance of simulations including the PI3K cascade	183
6.20	Performance on pathways improves with more genes	184
A.1	Correlation profiles of removed samples	225
A.2	Correlation analysis and sample removal	226
A.3	Replicate excluded samples	227
A.4	Replicate samples with all remaining	228
A.5	Replicate samples with some excluded	229
C.1	Synthetic lethal expression profiles of analysed samples	244
C.2	Comparison of mtSLIPT to short interfering RNA (siRNA)	246
C.3	Compare mtSLIPT and siRNA genes with correlation	250
C.4	Compare mtSLIPT and siRNA genes with correlation	250
C.5	Compare mtSLIPT and siRNA genes with siRNA viability	251
D.1	Pathway metagene expression profiles	254

D.2	Expression profiles for constituent genes of PI3K	256
D.3	Expression profiles for estrogen receptor related genes	257
D.4	Pathway metagene expression profiles	258
D.5	Expression profiles for p53 related genes	259
D.6	Expression profiles for BRCA related genes	260
D.7	Somatic mutation against the PI3K metagene	261
D.8	Somatic mutation against PIK3CA metagene	264
D.9	Somatic mutation against PI3K protein	265
D.10	Somatic mutation against AKT protein	266
F.1	Synthetic lethal expression profiles of stomach samples	271
F.2	Comparison of SLIPT in stomach to siRNA	273
G.1	Synthetic lethality in the PI3K/AKT pathway	278
G.2	Synthetic lethality in the PI3K/AKT pathway in cancer	279
G.3	Synthetic lethality in the Extracellular Matrix	280
G.4	Synthetic lethality in the GPCR Downstream	281
G.5	Synthetic lethality in the Translation Elongation	282
G.6	Synthetic lethality in the Nonsense-mediated Decay	283
G.7	Synthetic lethality in the 3' UTR	284
H.1	Synthetic lethality and vertex degree	285
H.2	Synthetic lethality and centrality	286
H.3	Synthetic lethality and PageRank	286
I.1	Structure of synthetic lethality resampling	288
J.1	Performance of χ^2 and SLIPT across quantiles	290
J.2	Performance of χ^2 and SLIPT across quantiles	292
J.3	Performance of χ^2 and SLIPT across quantiles with more genes	294
J.4	Performance of χ^2 and SLIPT across quantiles with query correlation	296
J.5	Performance of χ^2 and SLIPT across quantiles with query correlation	298
J.6	Performance of χ^2 and SLIPT across quantiles with query correlation and more genes	300
K.1	Performance of simulations on a simple graph	302
K.2	Performance of simulations on an inhibiting graph	303
K.3	Performance of simulations on a constructed graph with inhibition	304
K.4	Performance of simulations on a constructed graph with inhibition	305
K.5	Detection of synthetic lethality within a graph structure	306
K.6	Detection of synthetic lethality within an inhibiting graph	308
K.7	Detection of synthetic lethality within an inhibiting graph	309
K.8	Performance of simulations on a branching graph	310
K.9	Performance of simulations on a complex graph	311
K.10	Performance of simulations on a large graph	312
K.11	Performance of simulations on a branching graph with inhibition	313
K.12	Performance of simulations on a branching graph with inhibition	314

K.13 Performance of simulations on a complex graph with inhibition	315
K.14 Performance of simulations on a complex graph with inhibition	316
K.15 Performance of simulations on a large constructed graph with inhibition	317
K.16 Performance of simulations on a large constructed graph with inhibition	318
K.17 Performance of simulations on the $G_{\alpha i}$ signalling pathway	319
K.18 Performance of simulations including the $G_{\alpha i}$ signalling pathway	320

List of Tables

1.1	Methods for predicting genetic interactions	23
1.2	Methods for predicting synthetic lethality in cancer	23
1.3	Methods used by Wu <i>et al.</i> (2014)	25
2.1	Excluded samples by batch and clinical characteristics.	43
2.2	Computers used during thesis	53
2.3	Linux utilities and applications used during thesis	54
2.4	R installations used during thesis	55
2.5	R Packages used during thesis	55
2.6	R packages developed during thesis	57
4.1	Candidate synthetic lethal gene partners of <i>CDH1</i> from SLIPT	98
4.2	Pathways for <i>CDH1</i> partners from SLIPT	99
4.3	Pathways for clusters of <i>CDH1</i> partners from SLIPT	104
4.4	ANOVA for synthetic lethality and correlation with <i>CDH1</i>	107
4.5	Comparison of Synthetic Lethal Interaction Prediction Tool (SLIPT) genes against secondary siRNA screen	111
4.6	Pathways for <i>CDH1</i> partners from SLIPT and siRNA	112
4.7	Pathways for <i>CDH1</i> partners from SLIPT	115
4.8	Pathways for <i>CDH1</i> partners from SLIPT and siRNA primary screen .	116
4.9	Examples of candidate metagenes synthetic lethal for <i>CDH1</i> from SLIPT	120
5.1	ANOVA for synthetic lethality and vertex degree	138
5.2	ANOVA for synthetic lethality and information centrality	139
5.3	ANOVA for synthetic lethality and PageRank centrality	141
5.4	Resampling for pathway structure of synthetic lethal detection methods	144
B.1	Complete list of R packages used during this thesis	231
C.1	Candidate synthetic lethal gene partners of <i>CDH1</i> from mtSLIPT . . .	241
C.2	Pathways for <i>CDH1</i> partners from mtSLIPT	242
C.3	Pathways for clusters of <i>CDH1</i> partners from mtSLIPT	245
C.4	Pathways for <i>CDH1</i> partners from mtSLIPT and siRNA	247
C.5	Pathways for <i>CDH1</i> partners from mtSLIPT	248
C.6	Pathways for <i>CDH1</i> partners from mtSLIPT and siRNA primary screen	249
D.1	Candidate synthetic lethal metagenes against <i>CDH1</i> from mtSLIPT . .	263

E.1	Comparison of intrinsic subtypes	267
F.1	Synthetic lethal gene partners of <i>CDH1</i> from SLIPT in stomach cancer	269
F.2	Pathways for <i>CDH1</i> partners from SLIPT in stomach cancer	270
F.3	Pathways for clusters of <i>CDH1</i> partners in stomach SLIPT	272
F.4	Pathways for <i>CDH1</i> partners from SLIPT and siRNA	274
F.5	Pathways for <i>CDH1</i> partners from SLIPT in stomach cancer	275
F.6	Pathways for <i>CDH1</i> partners from SLIPT in stomach and siRNA	276
F.7	Synthetic lethal metagenes against <i>CDH1</i> in stomach cancer	277
H.1	ANOVA for synthetic lethality and vertex degree	287
H.2	ANOVA for synthetic lethality and information centrality	287
H.3	ANOVA for synthetic lethality and PageRank centrality	287
I.1	Resampling for pathway structure of synthetic lethal detection methods	289

Glossary

bioinformatics	Statistical or computational approaches to biological data or research tools.
centrality	A network metric which identifies important vertices .
E-cadherin	Epithelial cadherin (calcium-dependent adhesion), a cell-adhesion protein encoded by <i>CDH1</i> .
edge or link	A relationship connecting a pair of elements of a graph structure or network, may be weighted or directional.
essential	A gene which is required to be functional or expressed for a cell or organism to be viable, grow or develop.
gene expression	A measure of the relative expression of each gene from the mRNA extracted from (pooled) cells.
graph or network	A mathematical structure modelling or depicting the relationships between elements.
hub	A central or highly connected component of a network.
information centrality	A network centrality metric which uses the impact of removing a vertex or node on connections in the network.
metagene	A consistent signal of expression for a collection of genes such as a biological pathway, derived from singular value decomposition.
mutation	A change in DNA sequence that disrupts gene function.

non-oncogene addiction	The dependence of a cancer cell on functioning non-mutant genes.
oncogene	A gene that potentially causes cancer, typically by over-expression or mutant gene variants.
oncogene addiction	The dependence of a cancer cell on a specific oncogenic pathway.
PageRank centrality	A network centrality metric which uses eigenvectors with a scaling factor (Brin and Page, 1998).
scale-free	A property of a network which has a power law vertex degree distribution, that is several highly connected hub genes and many with very few connections.
shortest path	A path with the fewest possible edges which connects two particular vertices .
synthetic lethal	Genetic interactions where inactivation of multiple genes is inviable (or deleterious) which are viable if inactivated separately.
tumour suppressor	A gene potentially causes cancer, typically by disruption of functions which protect the cell from cancer.
vertex degree	A network metric of connectivity of vertices which uses the number of edges connected to each vertex or node .
vertex or node	An element of a graph structure or network.

Acronyms

AMP	Adenosine Monophosphate.
AMPK	AMP -activated Protein Kinase.
ANOVA	Analysis of Variance.
BioPAX	Biological Pathway Exchange.
BMP	Bone Morphogenic Protein.
CXCR	Chemokine Receptor.
EMT	Epithelial-Mesenchymal Transition.
GPCR	G Crotein Coupled Receptor.
JAK	Janus Kinase.
mtSLIPT	Synthetic Lethal Interaction Prediction Tool (against mutation).
NMD	Nonsense-Mediated Decay.
PDE	Phosphodiesterase.
PI3K	Phosphoinositide 3-kinase.
PIP ₂	Phosphatidylinositol-(4,5)-bisphosphate.
PIP ₃	Phosphatidylinositol-(3,4,5)-trisphosphate.
RGS	G-protein Signalling.
RHO	Ras Homolog Family.
RNA	Ribonucleic Acid.
siRNA	Short Interfering RNA.
SLIPT	Synthetic Lethal Interaction Prediction Tool.
TCGA	The Cancer Genome Atlas (genomics project).
TGF β	Transforming Growth Factor β .

UTR Untranslated Region (of mRNA).

WNT Wingless-Related Integration Site.

Chapter 5

Synthetic Lethal Pathway Structure

Having identified key pathways implicated in [synthetic lethal](#) genetic interactions with *CDH1* (in Chapter 4), these were investigated for the [synthetic lethal](#) genes within them and their relationships to [pathway](#) structure in Reactome pathways. This chapter will focus on the [pathway](#) structure of biological pathways detected across analyses in Chapter 4. Specifically, investigations were performed to determine whether [synthetic lethal](#) candidates, detected by [SLIPT](#) or [siRNA](#), exhibited differences with respect to metrics of [pathway](#) structure of network connectivity and importance (as described in Sections 2.4.4 and 3.5.3). The relationships between [synthetic lethal](#) candidates, detected by either approach, were also examined to determine whether [SLIPT](#) candidate genes were upstream or downstream [siRNA](#) candidate genes. These directional relationships were tested by resampling (as described in Sections 3.4.1 and 3.4.1.1) and comparisons to the pathway hierarchical score based on biological context (as derived in Section 3.4.1.2). Together these investigations into structural relationships demonstrate how a combination of network biology and statistical techniques can be performed with genes identified by a [bioinformatics](#) analysis.

5.1 Synthetic Lethal Genes in Reactome Pathways

The [graph](#) structure for Reactome pathways was obtained from Pathway Commons via [Biological Pathway eXchange \(BioPAX\)](#) (as described in Section 2.4.2). The pathways describe the (directional) relationships between biomolecules, including genes that encode proteins in biological pathways. These relationships include cell signalling (e.g., kinase phosphorylation cascades), gene regulation (e.g., transcription factors, chromatin modifiers, [RNA](#) binding proteins), and metabolism (e.g., the product of an enzyme being the substrate of another). Together these relationships describe the

known functional pathways in a human cell with a reasonable resolution, from a curated database supported by publications documenting pathway relationships.

Pathway structures from the Reactome network (as described in Section 2.4.3) were used to derive the [graph](#) structure of each biological pathway. The [synthetic lethal](#) candidate genes for notable pathways discussed in Chapter 4, including candidate [synthetic lethal](#) pathways of *CDH1*, were examined to show the [SLIPT](#) and [siRNA](#) candidates within these pathways. The [synthetic lethal](#) genes considered here are those candidates detected by [SLIPT](#) (as described in Section 3.1) in [The Cancer Genome Atlas \(TCGA\)](#) breast cancer [expression](#) and [mutation](#) data ([Koboldt *et al.*, 2012](#)) in comparison to the candidate gene partners from the [siRNA](#) screening in breast cell lines ([Telford *et al.*, 2015](#)).

5.1.1 The PI3K/AKT Pathway

The [phosphoinositide 3-kinase \(PI3K\)](#) cascade signalling pathway is important in cancer because it is involved in mediating signals between the [G protein coupled receptors](#) and regulation of protein translation have both been strongly implicated to be [synthetic lethal](#) pathways with loss of *CDH1* function (in Chapter 4). These pathways have are all subject to dysregulation in cancer ([Courtney *et al.*, 2010](#); [Dorsam and Gutkind, 2007](#); [Gao and Roux, 2015](#)). Thus the PI3K cascade will be examined along with the most supported [synthetic lethal](#) pathways (as identified in Chapter 4). It also exhibited a relationship with *CDH1* mutations in [metagene](#) analyses (in Appendix D).

The [phosphoinositide 3-kinase \(PI3K\)](#) pathway is well characterised and has an established direction of signal transduction from extracellular stimuli (and membrane bound receptors) to the inner mechanisms of the cell, namely, the regulation of protein translation. The production of proteins is necessary for the growth of the cell so it is reasonable to suggest that these processes may be subject to (non-[oncogene](#)) addiction in some cancer cells which rely upon them for sustained protein production and cell growth. This is also supported by the [oncogenes](#) *PIK3CA* and *AKT1* being involved with the PI3K cascade and related PI3K/AKT pathway which may be subject to [oncogene addiction](#) when these proto-oncogenes are activated.

The [PI3K](#) cascade was not supported across [SLIPT](#) in [TCGA](#) breast [expression](#) data and the [siRNA](#) primary screen by over-representation (in Section 4.2.5) or resampling (in Section 4.2.5.1) but genes were detectable by either approach (as shown in Figure 5.1). While few genes were identified by both approaches, they include genes that are highly connected in the PI3K cascade and are hubs to information transmission

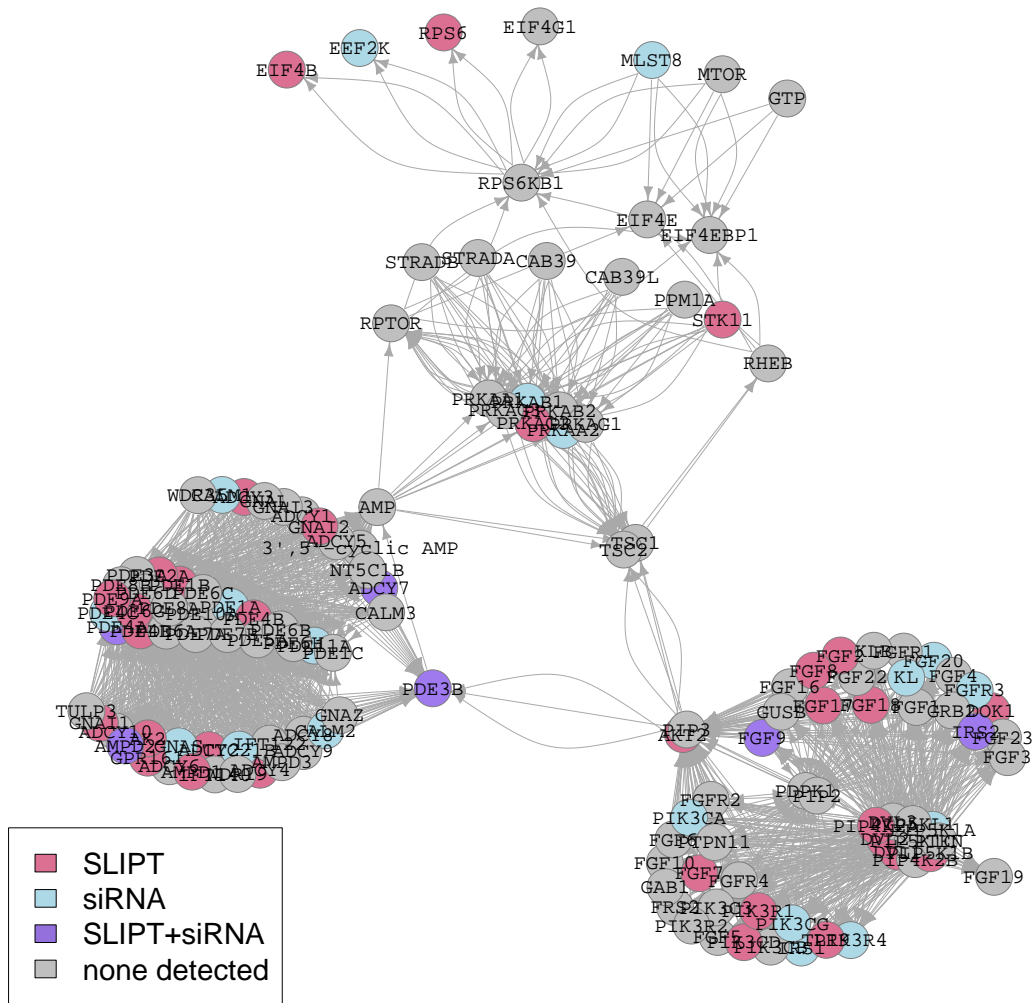


Figure 5.1: **Synthetic lethality in the PI3K cascade.** The Reactome [PI3K](#) Cascade pathway with [synthetic lethal](#) candidates coloured as shown in the legend.

such as *FGF9*, *PDE3B*, and *PDE4A*. The key upstream genes *PIK3CA* and *PIK3CG* were detected by [siRNA](#) whereas the downstream *PIK3R1* and *AKT2* genes were detected by [SLIPT](#). Gene detected by either method were also prevalent in the [PI3K](#), [phosphodiesterase \(PDE\)](#), and [AMP-activated protein kinase \(AMPK\)](#) modules, in addition to the downstream translation factors and ribosomal genes (*EIF4B*, *EEF2K*, and *RPS6*). Together these suggest that there may be further structure between the [SLIPT](#) and [siRNA](#) candidate partners of *CDH1* in pathways as illustrated by [PI3K](#). As such, [pathway](#) structure will be investigated to detect differences in the upstream and

downstream gene candidates of those detected by either method. Pathway structure may account for the disparity between SLIPT and siRNA genes, even in pathways such as PI3K where they did not significantly intersect. For instance, SLIPT gene partners may be downstream of siRNA candidates rather than replicating them directly.

This disparity between SLIPT and siRNA gene candidate synthetic lethal partners of CDH1 (i.e., a high number of genes detected by either approach with few detected by both) was replicated in the related PI3K/AKT pathway and the “PI3K/AKT in cancer” pathway (shown in Appendix Figures G.1 and G.2). Many synthetic lethal candidates were at the upstream core of these pathway networks and the downstream extremities. It is particularly notable that the many genes important in cell signalling and gene regulation were detected by either synthetic lethal detection approach. These include *AKT1*, *AKT2*, and *AKT3*, the Calmodulin signalling genes *CALM1* and *CAMK4*, and the forkhead family transcription factors *FOXO1* (a tumour suppressor) and *FOXO4* (an inhibitor of EMT).

5.1.2 The Extracellular Matrix

The extracellular pathways “elastic fibre formation” and “fibrin clot formation” (shown in Figures 5.2 and 5.3 respectively) were both supported across analyses (in Chapter 4). These pathways were identified by both SLIPT (for TCGA breast cancer) and siRNA gene candidates as they had significant over-representation and resampling.

Particularly for elastic fibres (Figure 5.2), the vast majority of genes were detected by either approach in addition to a significant proportion of genes detected by both approaches (as determined in Section 4.2.5). The genes detected by both approaches also appeared to have a non-random distribution in the network, with *TFGB1*, *ITGB8*, and *MFAP2* exhibiting high connectivity, and having a central role in their respective pathway modules. In addition to a structural role in the extracellular matrix and connective tissue (including the tumour microenvironment), these proteins including Furin, transforming growth factor β (TGF β), and the bone morphogenic proteins (BMPs), are also involved in responses to endocrine signals and interact with the cellular receptors for signalling pathways. Therefore it is plausible that *CDH1* deficient tumours will be subject to non-oncogene addiction to the extracellular environment and growth signals arising from this pathway. The pathway structure also indicated that the genes detected by siRNA (or by both approaches) may be downstream of those detected by SLIPT, in addition to whether connectivity or centrality is higher for synthetic lethal candidates than other genes in the pathway.

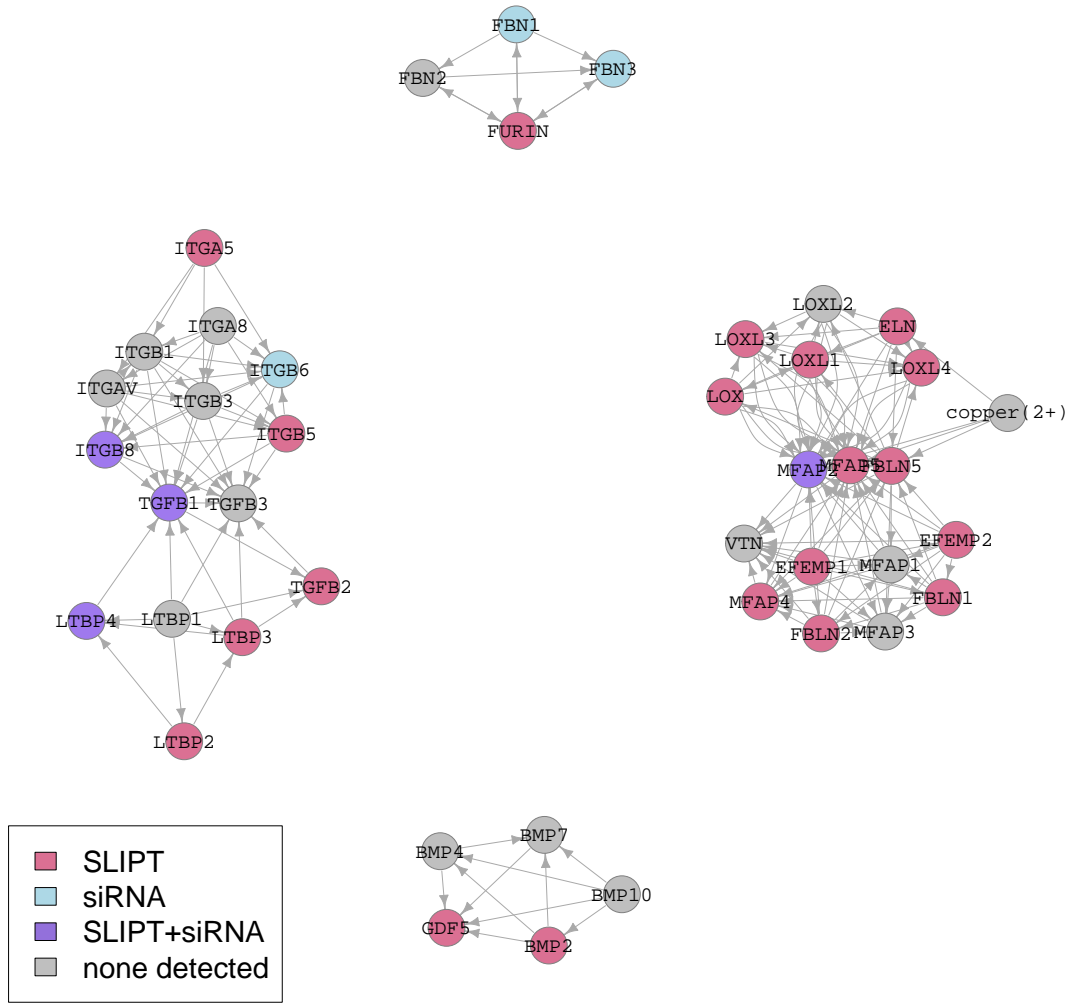


Figure 5.2: **Synthetic lethality in Elastic Fibre Formation.** The Reactome Elastic Fibre Formation pathway with **synthetic lethal** candidates coloured as shown in the legend.

Genes detected as **synthetic lethal** partners of *CDH1* by **SLIPT** or **siRNA** screening were also common in the Fibrin clot formation pathway (shown in Figure 5.3). This is consistent with the established pleiotropic role of *CDH1* in regulating fibrin clotting. It is also notable that the genes detected by either method appear to be highly connected such as *C1QBP*, *KNG1*, *F8*, *F10*, *F12*, *F13A*, and *PROC* (including many of the coagulation factors). **Synthetic lethal** candidates also include *SERPINE2* and *PRCP*, which only affect downstream genes, in addition to *PROCR* and *VWF*, which are only affected by upstream genes.

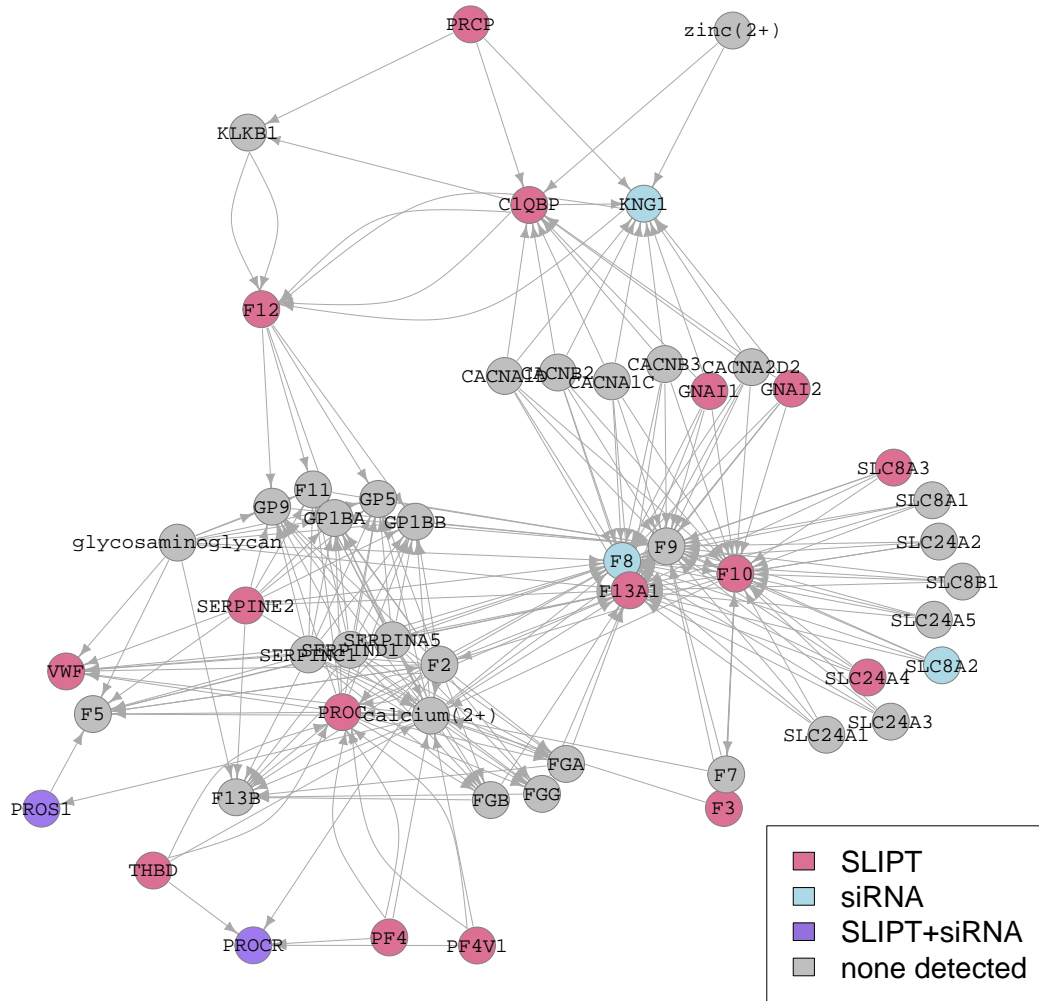


Figure 5.3: **Synthetic lethality in Fibrin Clot Formation.** The Reactome Fibrin Clot Formation pathway with **synthetic lethal** candidates coloured as shown in the legend.

Many of these genes are involved in the larger Extracellular Matrix pathway (shown in Appendix Figure G.3), including many of the **synthetic lethal** candidates discussed for elastic fibres. The number of **SLIPT** candidate genes outnumbers those identified by **siRNA**, as expected from an isolated cell model. However, the endocrine response genes (e.g., *TGFB1* and *LTBP4*) which are potentially artifacts of the cell line growth process were replicated with **SLIPT** analysis in patient tumours (TCGA breast cancer data). There is also additional support for **synthetic lethal** genes (e.g., *ITGB2*, *MFAP2*, and *SPARC*) being highly connected networks hubs of the pathway. The complexity of

the extracellular matrix pathway lends credence to the need for formal network analysis approaches to interpret the [pathway](#) structure of [synthetic lethal](#) candidates. Furthermore statistical approaches are needed to determine whether structural relationships are unlikely to be observed between [synthetic lethal](#) candidates by chance

5.1.3 G Protein Coupled Receptors

[G protein coupled receptor](#) (GPCR) pathways are highly complex (as shown in [Figure 5.4](#) and [Appendix Figure G.4](#)). Many of genes in these pathways were [synthetic lethal](#) candidates, detected by either [SLIPT](#) or [siRNA](#) screening, including genes frequently detected with both approaches, consistent with these pathways being supported by prior analyses (in [Sections 4.2.5](#) and [4.2.5.1](#)). [Synthetic lethal](#) candidates include the [PDE](#) and [Calmodulin](#) genes (as discussed in [Section 5.1.3](#)) in addition to others such as the regulators of [G-protein signalling](#) ([RGS](#)), [chemokine receptors](#) ([CXCR](#)), [Janus kinase](#) ([JAK](#)), and the [Ras homolog family](#) ([RHO](#)) genes. These are important regulatory signalling pathways necessary for cellular growth and cancer proliferation. Thus the [GPCR](#) pathways (and downstream [PI3K/AKT](#) signals) are a potentially actionable vulnerability against *CDH1* deficient cancers, particularly since many existing drug targets exist among these signalling pathways, some of which have been experimentally validated ([Telford et al., 2015](#)). While a statistically significant number of genes in GPCR pathways was detected by both approaches (in [Sections 4.2.5](#) and [4.2.5.1](#)), the complexity of [GPCR](#) networks (containing hundreds of genes) further support the needs for a rational network-based approach to the relationships between [SLIPT](#) and experimental candidates.

5.1.4 Gene Regulation and Translation

While very few [synthetic lethal](#) genes were detected in translational pathways in an experimental screen against *CDH1* ([Telford et al., 2015](#)), these were highly over-represented in translational elongation (as shown in [Appendix Figure G.5](#)). These [SLIPT](#) genes include many ribosomal proteins and the regulatory “elongation factors” which may be subject to responses in the upstream signalling pathways. This observation further indicates that [pathway](#) structure may be used to identify relationships between [synthetic lethal](#) candidates detected by [SLIPT](#) and [siRNA](#). The computational approach with [SLIPT](#) may exhibit the ability to detect downstream genes in the core translational processes, which experimental screening did not identify. The experimental screening may similarly detect upstream regulatory genes less sensitive

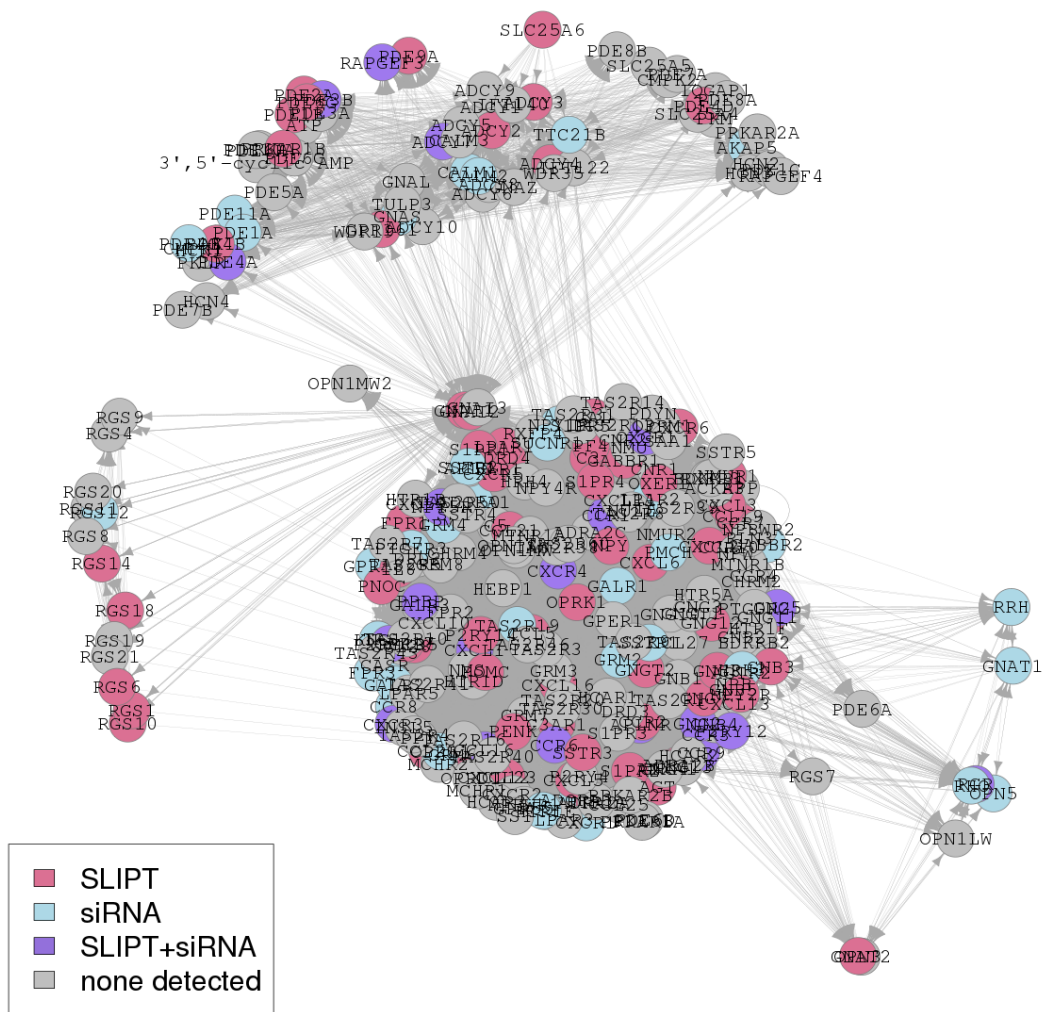


Figure 5.4: **Synthetic lethality in the GPCRs.** The Reactome $G_{\alpha i}$ pathway with synthetic lethal candidates, coloured as shown in the legend.

to inactivation, that is, genes that are less likely to be indiscriminately lethal to both genotypes at high doses of inactivation.

Many of these **SLIPT** candidate genes are also among the **nonsense-mediated decay (NMD)** pathway (shown in Appendix Figure G.6) or **3' untranslated region (UTR)** mediated translational regulation (shown in Appendix Figure G.7). While genes in these pathways were also supported by experimental screening with **siRNA**, there were differences in which genes were detected within the **pathway** structures. In particular, *UPF1* was detected in the **siRNA** screen and is the focal downstream gene for the entire **NMD** pathway showing that (in this case) **siRNA** genes are downstream effectors of those detected by **SLIPT**. **3' UTR** mediated translational regulation has a similar structure with two modules connected solely by *RPL13A*, giving an example of **SLIPT** candidate genes with high connectivity, although there were many ribosomal proteins detected by **SLIPT**. However, the detection of *EIF3K*, a regulatory elongation factor (not **essential** to ribosomal function) was replicated across **SLIPT** and **siRNA** screening, while the majority of the elongation factors were not detected by either approach. Regulatory genes, being more amenable to experimental validation, also support further investigation into **pathway** structure. The **SLIPT** candidates may support experimental candidates in biological pathways by detecting downstream genes, which may not be detectable by experimental screening with high dose inhibitors. This difference between the approaches may explain the greater number of **SLIPT** candidate partners of *CDH1* than those experimentally identified.

5.2 Network Analysis of Synthetic Lethal Genes

Genes detected as **synthetic lethal** partners of *CDH1* with the **SLIPT** computational approach and the **siRNA** screen (Telford *et al.*, 2015) were compared across network metrics in the example of $G_{\alpha i}$ signalling, a **GPCR** pathway. This pathway was used to demonstrate deeper network analysis approaches to **synthetic lethal** candidates within complex pathways it was supported across analyses (in Chapter 4), with significant over-representation in both **SLIPT** and **siRNA** screening, and the genes differed considerably between **synthetic lethal** detection methods (shown in Appendix Figures 5.4). These network metrics were used to measure whether the network properties differed between groups of genes detected by either or both approaches. These analyses serve to test both whether **synthetic lethal** gene candidates had higher connectivity or importance in a network and whether either detection approach is biased towards genes with different network properties.

5.2.1 Gene Connectivity and Vertex Degree

Vertex degree (the number of connections) for each gene is a fundamental property of a network. The vast majority of genes had a relatively modest number of connections, each with only a few genes in the $G_{\alpha i}$ pathway (shown in Figure 5.5) having pathway relationships with a high number of genes, consistent with the [scale-free](#) property of biological networks ([Barabási and Oltvai, 2004](#)). The number of connections was similar between gene groups (by [synthetic lethal](#) detection). Genes detected by [siRNA](#) included those with the fewest connections, despite there being fewer genes that were detected by either approach. There was no statistically significant effect of either computational or experimental [synthetic lethal](#) detection method on [vertex](#) degree, as determined by [analysis of variance \(ANOVA\)](#) (shown by Table 5.1).

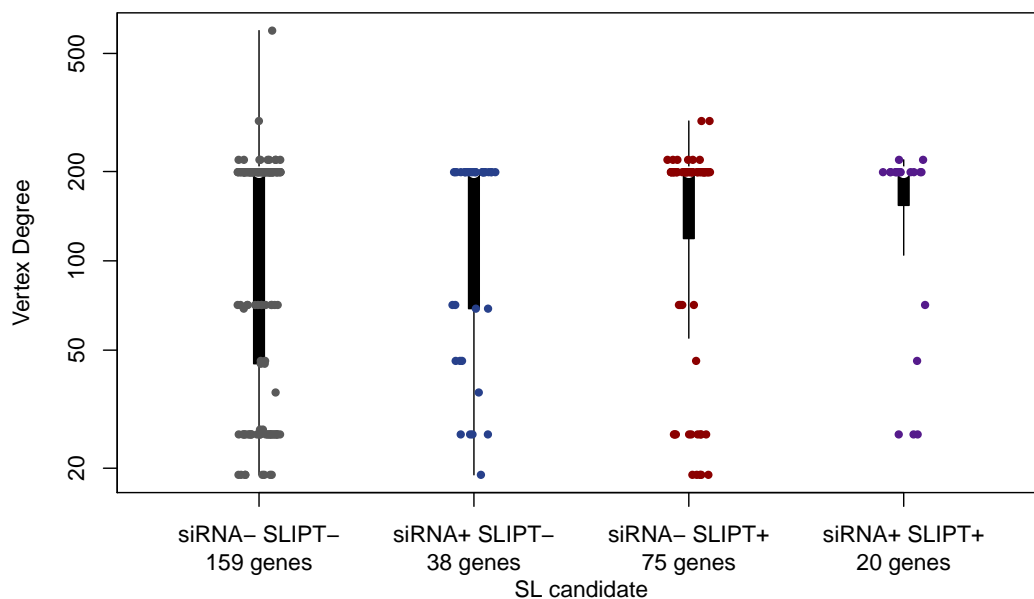


Figure 5.5: **Synthetic lethality and vertex degree.** The number of connected genes ([vertex degree](#)) was compared (on a log-scale) across genes detected by [SLIPT](#) and [siRNA](#) screening in the Reactome $G_{\alpha i}$ cascade pathway. There were no differences in [vertex](#) degree between the groups (shown in Table 5.1), although genes detected by [siRNA](#) included those with the fewest connections.

Table 5.1: ANOVA for synthetic lethality and vertex degree

	DF	Sum Squares	Mean Squares	F-value	p-value
siRNA	1	21	20.8	0.0030	0.9561
SLIPT	1	16215	16215	2.3722	0.1246
siRNA×SLIPT	1	17	17	0.0025	0.9603

Analysis of variance for **vertex** degree against **synthetic lethal** detection approaches (with an interaction term)

The results for the $G_{\alpha i}$ pathway were very similar when testing **synthetic lethality** against *CDH1* **mutation** (mtSLIPT). In either case, there was no significant evidence that SLIPT or mtSLIPT-specific genes had higher connectivity than those detected by siRNA screening (shown in Appendix Figure H.1 and Appendix Table H.1). Thus **synthetic lethal** detection does not discriminate among genes by their connectivity in this pathway network, nor is either approach constrained to detecting highly connected genes. Both approaches have been demonstrated to detect genes with many and very few connections in the $G_{\alpha i}$ signalling pathway.

5.2.2 Gene Importance and Information Centrality

Information centrality is a measure of the importance of **nodes** in a network in terms of how vital they are to the transmission of information throughout the network. This applies well to biological pathways, particularly gene regulation and cell signalling. The **nodes** with the highest **information centrality** are not necessarily the most connected, as they may also include **nodes** that pass signals between highly connected network hubs. **Information centrality** therefore provides a distinct metric for the connectivity of a gene in a pathway, which has the added benefit of being directly related to the disruption of pathway function were it to be inactivated or removed. **Information centrality** has also been suggested to be indicative of the essentiality of genes or proteins (Kranthi *et al.*, 2013).

Within the $G_{\alpha i}$ pathway (shown in Figure 5.6), the **information centrality** across gene groups detected by either **synthetic lethal** approach did not differ significantly (shown by Table 5.2). Genes detected by SLIPT span the complete range of **PageRank centrality** values for this pathway. These findings were replicated (shown in Appendix Figure H.2 and Appendix Table H.2). Thus neither method was unable to detect **synthetic lethal** genes in the $G_{\alpha i}$ pathway with particular **centrality** constraints but they were also not detecting genes with higher **centrality** than expected by chance.

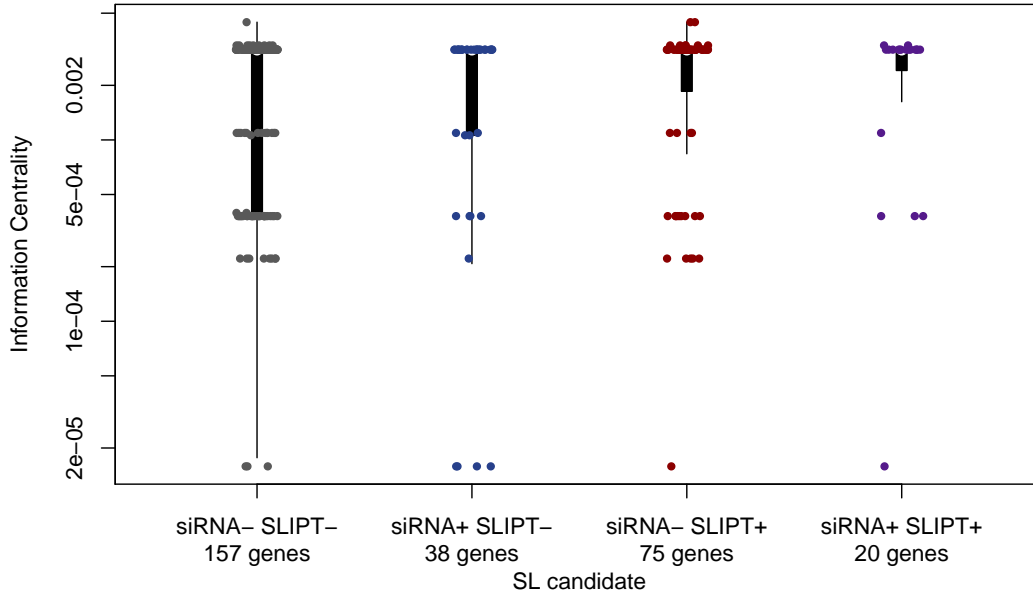


Figure 5.6: **Synthetic lethality and centrality.** The [information centrality](#) was compared (on a log-scale) across genes detected by [SLIPT](#) and [siRNA](#) screening in the Reactome $G_{\alpha i}$ pathway. Genes detected by [SLIPT](#) or [siRNA](#) did not have higher centrality than other genes (shown in Table 5.2). Genes detected by [SLIPT](#) spanned the range of [centrality](#) values.

Table 5.2: [ANOVA](#) for synthetic lethality and information centrality

	DF	Sum Squares	Mean Squares	F-value	p-value
siRNA	1	0.00000000	2.7000×10^{-9}	0.0016	0.96783
SLIPT	1	0.00000548	5.4831×10^{-6}	3.3253	0.06926
siRNA \times SLIPT	1	0.00000002	1.8800×10^{-8}	0.0114	0.91511

Analysis of variance for [information centrality](#) against [synthetic lethal](#) detection approaches (with an interaction term)

5.2.3 Gene Importance and PageRank Centrality

[PageRank centrality](#) is another network analysis procedure to infer a hierarchy of gene importance from a network using connections and structure ([Brin and Page, 1998](#)). In contrast to the [information centrality](#) approach of removing [nodes](#), PageRank uses the eigenvalue properties of the adjacency matrix to rank genes according to the number of connections and paths they are involved in.

This distinction is immediately clear within the $G_{\alpha i}$ pathway (shown in Figure 5.7), which differs considerably from the [information centrality](#) scores (in Figure 5.6). Genes detected by either [synthetic lethal](#) approach did not include those with the highest PageRank centrality. There was a significant association between genes detected by [SLIPT](#) (which had a lower median) with PageRank centrality (shown by Table 5.3). The genes detected by [SLIPT](#) span the range of centrality values of [siRNA](#) showing that both approaches were capable of detecting genes of moderately high centrality (as shown for information centrality) and that the lower centrality of [SLIPT](#) candidates in $G_{\alpha i}$ pathway may be due to [synthetic lethal](#) partners being less critical to the pathway, rather than a limitation of the methodology.

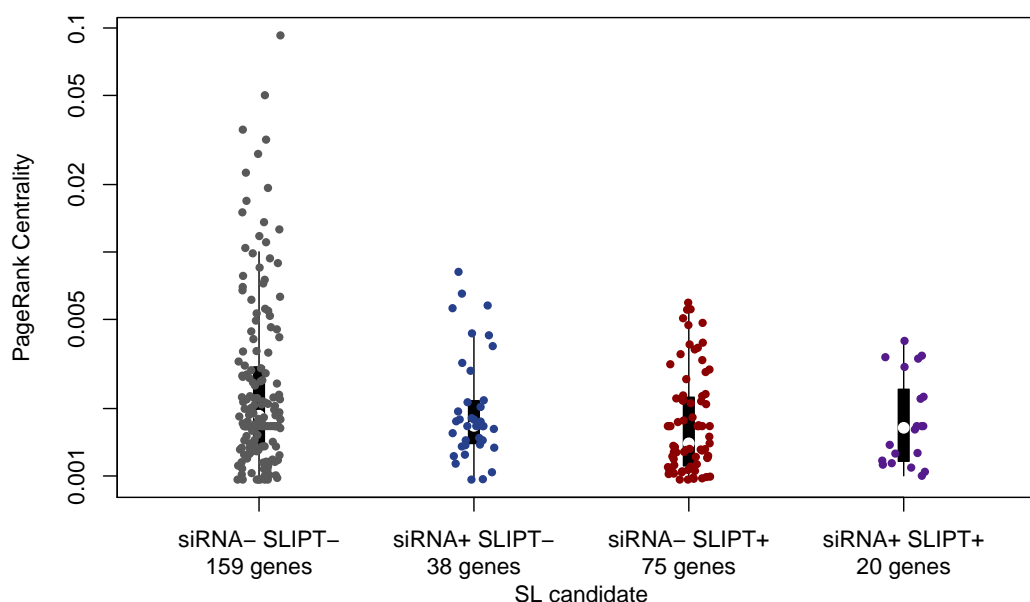


Figure 5.7: **Synthetic lethality and PageRank.** The [PageRank centrality](#) was compared (on a log-scale) across genes detected by [mtSLIPT](#) and [siRNA](#) screening in the Reactome $G_{\alpha i}$ pathway. Genes detected by with either [synthetic lethal](#) detection approach had a more restricted range of [centrality](#) values but only [SLIPT](#) genes had a significant association with centrality (shown in Table 5.3).

There was not a significant association between [siRNA](#) candidates and PageRank centrality. The significant result for [SLIPT](#) was not replicated when testing [synthetic lethality](#) against [CDH1 mutation](#) (shown in Appendix Figure H.3 and Appendix Ta-

ble H.3). However, this may be due to fewer genes being detected by [mtSLIPT](#) and [siRNA](#).

Table 5.3: [ANOVA](#) for synthetic lethality and PageRank centrality

	DF	Sum Squares	Mean Squares	F-value	p-value
siRNA	1	0.0001059	1.0589×10^{-4}	2.1021	0.14818
SLIPT	1	0.0002881	2.8808×10^{-4}	5.7188	0.01743
siRNA×SLIPT	1	0.0000477	4.7704×10^{-5}	0.9470	0.33131

Analysis of variance for [PageRank centrality](#) against [synthetic lethal](#) detection approaches (with an interaction term)

5.3 Upstream or Downstream Synthetic Lethality

This approach does not ascertain whether [SLIPT](#) and [siRNA](#) candidate partners of *CDH1* are upstream or downstream of one and other within a pathway such as the [PI3K](#) cascade. The hierarchical approach is designed to detect differences in pathway location between gene groups. An alternative [pathway](#) structure method has been devised to use [network](#) structures to identify directional relationships between individual [SLIPT](#) and [siRNA](#) genes. This [pathway](#) structure methodology will be applied (as described in Section 3.4.1) to detect the direction of [shortest paths](#) between [SLIPT](#) and [siRNA](#) gene candidates. This will be used to demonstrate the methodology on the [PI3K](#) pathway, to develop a statistical test for [pathway](#) structure between [SLIPT](#) and [siRNA](#) gene candidate using resampling (as described in Section 3.4.1.1), and to apply this test for [pathway](#) structure among [synthetic lethal](#) gene candidates to the pathways identified in Chapter 4 and discussed in Section 5.1.

5.3.1 Measuring Structure of Candidates within PI3K

Shortest paths in a pathway network were used to devise a strategy to detect [pathway](#) structure between [SLIPT](#) and [siRNA](#) gene candidate partners of *CDH1* (as described in Section 3.4.1). Thus we can determine whether individual [SLIPT](#) genes have upstream or downstream [siRNA](#) candidates (scored as “up” or “down” events respectively). This procedure enables the detection of directional relationships between [SLIPT](#) and [siRNA](#) gene candidates (in contrast to the hierarchical approach).

The total number of gene candidate pairs in either direction can be compared within a pathway network to assess the overall directional relationships in a pathway. This directionality is detectable by the difference between the number of [SLIPT](#) candidate

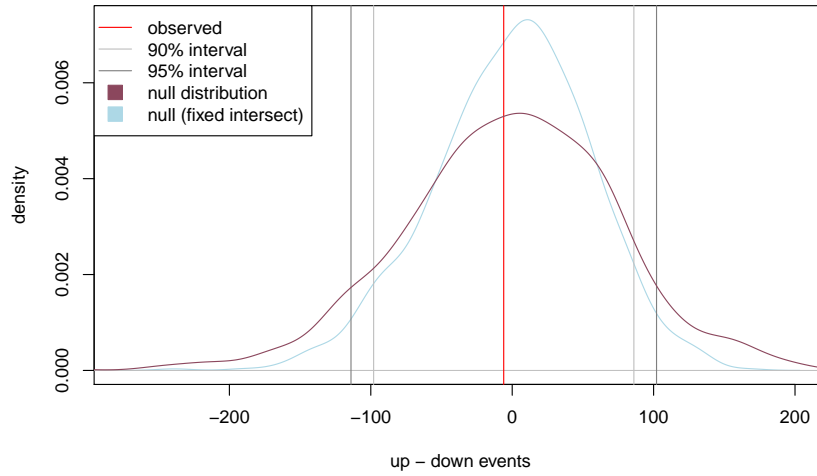


Figure 5.8: **Structure of synthetic lethality resampling in PI3K.** A null distribution with 10,000 iterations of the number of siRNA genes upstream or downstream of SLIPT genes (depicted as the difference of these) in the PI3K pathway. To assess significance, the observed events (with shortest paths) were compared to the 90% and 95% intervals for the null distribution (shown in violet). Genes detected by both methods were fixed to the same number as observed for the alternative null distribution (shown in blue), although the observed number of events (red) was not significant in either case. In both cases, these genes detected by both approaches were included in computing the number of shortest paths (in either direction) between SLIPT and siRNA genes.

genes with upstream and downstream siRNA gene partners. However, this measure alone is not sufficient to determine whether there is evidence of pathway structure between SLIPT and siRNA gene candidate partners of *CDH1* in a pathway network. Nevertheless, it does serve to measure the magnitude (and direction) of the consensus of directional relationships (upstream and downstream) between SLIPT and siRNA gene candidate partners. This measure of pathway structure can be used for testing for statistical significance of pathway structure by resampling, using a permutation procedure to test whether these relationships are detectable among randomly selected gene groups rather than the detected SLIPT and siRNA gene candidate partners (as described in Sections 2.3.6 and 3.4.1.1).

This resampling procedure was performed for the PI3K network to generate a null distribution for the difference in the number of “up events” and “down events” for this pathway (as shown in Figure 5.1). Resampling yields a distribution to detect whether

genes detected by **SLIPT** had significantly more upstream or downstream **siRNA** candidates. While there was modest indication that **siRNA** genes were downstream of **SLIPT** candidate genes, resampling for the **PI3K** pathway (as shown in Figure 5.8) did not detect a significant number of **siRNA** genes upstream or downstream.

In contrast, when testing **synthetic lethality** against *CDH1* mutation (**mtSLIPT**) there was modest indication that **siRNA** genes were upstream of **SLIPT** candidate genes. However, resampling (as shown in Appendix Figure I.1) was also unable to detect a significant number of **siRNA** genes upstream or downstream of **mtSLIPT** candidates. Neither fixing the number of genes detected by both approaches (as shown by the blue line in Figure 5.8 and Appendix Figure I.1) nor excluding these jointly detected genes altered the findings of this approach. These genes were included in the analysis because they can disproportionately count towards **siRNA** genes being upstream (or downstream) of **SLIPT** genes as they may still have different proportions of gene detected by either approach upstream (or downstream) of them. Furthermore, expanding the range of **shortest paths** to consider **links** in related pathways (using the “metapathways” constructed in Section 2.4.3) also had little effect on the null distribution generated, despite increasing the computational demands of the procedure.

5.3.2 Resampling for Synthetic Lethal Pathway Structure

The permutation procedure (as described in Section 3.4.1.1) that was performed in Section 5.3.1 for the **PI3K** cascade was also applied to other pathways identified in Chapter 4 and discussed in Section 5.1. These include extracellular matrix (with constituent elastic fibre and fibrin pathways), cell signalling (by **PI3K/AKT** and **GPCRs**), and translational pathways (with **NMD** and **3'UTR** regulation). The resampling results across these pathways (as shown in Table 5.4) had limited support for association between **pathway** structure and detection of **synthetic lethal** genes, with the majority of these being non-significant as shown for **PI3K** (in Appendix Figure I.1). However, the distribution for these pathways will differ depending on their structure, the number of genes they consist of, and the proportion of **synthetic lethal** candidates among them (including a higher frequency of genes detected by both methods for the pathways identified in Section 4.2.5.1). This resampling is an appropriate procedure to use to detect structural relationships across pathways as it does not assume an underlying test statistic distribution.

Pathway structure was supported for the **NMD** pathway (which is consistent with **siRNA** being downstream in Appendix Figure G.6). However, this observation rests

upon a single gene and was not replicated when testing [synthetic lethality](#) ([mtSLIPT](#)) against [CDH1 mutation](#) (as shown in Appendix Table I.1) nor was it supported by the related 3'UTR regulation and translational elongation pathways.

Table 5.4: Resampling for [pathway](#) structure of [synthetic lethal](#) detection methods

Pathway	Graph		States		Observed				Permutation p-value	
	Nodes	Edges	SLIPT	siRNA	Up	Down	Up-Down	Up/Down	Up-Down	Down-Up
PI3K Cascade	138	1495	38	25	122	128	-6	0.953	0.5326	0.4606
PI3K/AKT Signalling in Cancer	275	12882	98	44	779	679	100	1.147	0.3255	0.6734
G_{αi} Signalling	292	22003	95	58	836	1546	-710	0.541	0.9971	0.0029
GPCR downstream	1270	142071	312	160	9755	9261	494	1.053	0.3692	0.6305
Elastic fibre formation	42	175	24	7	1	2	-1	0.500	0.5461	0.3865
Extracellular matrix	299	3677	127	29	547	455	92	1.202	0.3351	0.6636
Formation of Fibrin	52	243	18	5	12	17	-5	0.706	0.6198	0.3564
Nonsense-Mediated Decay	103	102	74	2	0	74	-74	0	1.0000	< 0.0001
3' -UTR-mediated translational regulation	107	2860	77	1	0	0	0		0.4902	0.5027
Eukaryotic Translation Elongation	92	3746	76	0	0	0	0		0.4943	0.4933

Pathways in the Reactome network tested for structural relationships between [SLIPT](#) and [siRNA](#) genes by resampling. The raw p-value (computed without adjusting for multiple comparisons over pathways) is given for the difference in upstream and downstream paths from [SLIPT](#) to [siRNA](#) gene candidate partners of *CDH1* with significant pathways highlighted in bold. Sampling was performed only in the target pathway and [shortest paths](#) were computed within it. Loops or paths in either direction that could not be resolved were excluded from the analysis. The gene detected by both [SLIPT](#) and [siRNA](#) (or resampling for them) were included in the analysis and the number of these were fixed to the number observed.

There does not appear to be a consensus on the directionality of [SLIPT](#) and [siRNA](#) candidates across pathways as distinct pathways showed stronger tendency for [siRNA](#) genes to be either upstream or downstream. Even related pathways such as [PI3K](#) and [PI3K/AKT](#) signalling showed directional events in opposite directions. The strongest pathway (among those tested) with support for directional pathways structure is [G_{αi} signalling](#) which showed significant downstream [siRNA](#) genes for both [SLIPT](#) and [mt-SLIPT](#) from a large number of [shortest paths](#) (in Table 5.4 and Appendix Table I.1). This would indicate that [SLIPT](#) detects upstream regulators of genes experimentally validated by [siRNA](#). However, these results are borderline significant (with raw permutation p-values) and are unlikely to be detected after adjusting for multiple comparisons across the 10 pathways presented here (nor in the 1652 Reactome pathways used previously in Chapter 4).

Therefore, there is insufficient evidence to determine whether there is [pathway](#) structure, gene detected upstream or downstream by either method, between the [SLIPT](#) and [siRNA](#) candidates in many of the [synthetic lethal](#) pathways (identified in Chapter 4). In particular, directional structure among [synthetic lethal](#) candidates for *CDH1* was not strongly supported in signalling pathways upon which the rationale for [pathway](#) structure hypotheses were based on. Despite the design of a robust resampling ap-

proach to test relationships between gene groups, this did not detect many structural relationships between [SLIPT](#) and [siRNA](#) gene candidates, although it may apply more broadly to gene networks. Furthermore, the pathway relationships are unlikely to be statistically supported by resampling when testing across the search space of Reactome pathways and adjusting for multiple comparisons. While there is statistically significant over-representation of many of these pathways in genes detected by both [SLIPT](#) and [siRNA](#) (as described in Chapter 4), these did not consistently show [pathway](#) structure. Furthermore, [pathway](#) structure did not account for the discrepancy between [SLIPT](#) and [siRNA](#) gene candidates which did not significantly intersect such as the [PI3K](#) cascade.

5.4 Discussion

These investigations used a functional pathway network that encapsulates protein complexes and functional modules. The Reactome network ([Croft *et al.*, 2014](#)) uses curated, experimentally identified pathways to determine relationships between genes and does not have the limitation of relying solely on protein binding or text-mining which are prone to false positives. While it is not documented whether these relationships are activating or inhibitory, the Reactome network ([Croft *et al.*, 2014](#)) is sufficient to test pathway relationships with directional information.

Synthetic lethal genes and pathways (for *CDH1* loss in cancer) were identified across [gene expression](#) and [mutation](#) datasets in Chapter 4. These [pathway](#) structure investigations extend those investigations into [synthetic lethal](#) gene candidates including exploring the discrepancy between [SLIPT](#) and [siRNA](#) candidate genes in a pathway such as [PI3K](#) in which they did not significantly intersect. Pathways with replicated [synthetic lethal](#) genes across these detection methods, breast and stomach cancer data, and patient and cell line data were also investigated including pathways from the extracellular microenvironment to core translational pathways and the signalling pathways between them.

Synthetic lethal gene candidates in the context of [pathway](#) structures can also be interpreted to provide additional mechanisms and support for belonging to a [synthetic lethal](#) pathway. Gene candidates with known mechanisms are ideal for triage of targets specific to *CDH1* deficient tumours and for further experimental validation in preclinical models. This chapter presents computational methods to use [pathway](#) structure in an attempt to detect genes with importance in a pathway and reconcile the differ-

ences between [SLIPT](#) and [siRNA](#) candidate genes with pathway relationships (e.g., one group being downstream of the other).

Many genes were detected by either method and the differences between the computational and experimental screening approaches could feasibly lead to differences in which genes within a [synthetic lethal](#) pathway are identified. Genes detected by [synthetic lethal](#) detection strategies included those of biological importance within [synthetic lethal](#) pathways, those which are actionable drug targets, and those with functional implications for the biological growth mechanisms or vulnerabilities of *CDH1* deficient tumours. It appeared that genes detected by both approaches were highly connected (or of importance) in the [network](#) structure or some pathways and that there may be some structure with [SLIPT](#) and [siRNA](#) upstream or downstream of each other. However, the complexity of biological pathways meant that relationships between gene candidates were difficult to discern without formal mathematical and computational approaches and thus these were used to analyse large biological networks.

Network analysis techniques were therefore applied to formalise and quantify the connectivity and importance (centrality) of genes within pathways (using [PI3K](#) as an example). However, these network techniques were unable to identify distinct differences in the network properties of genes detected as [synthetic lethal](#) candidates by computational or experimental methods. These network metrics support the application of synthetic detection across pathways (and the findings using pathways as gene sets in Chapter 4) as neither [synthetic lethal](#) detection approach was biased towards genes of higher importance or connectivity and neither approach was insensitive to genes of lower importance or connectivity. [SLIPT](#) is therefore not biased towards genes with more crucial role in the pathway as inferred by pathway connectivity and [centrality](#) measures and detects genes irrespective of [pathway](#) structure.

Similarly, a network hierarchy based on biological context (ordered from receiving extracellular stimuli to affecting downstream [gene expression](#) and cell growth) was devised to test whether [PI3K](#) genes of a particular upstream or downstream level were more frequently detected as [synthetic lethal](#) candidates. However, this approach was unable to ascertain whether genes detected by either method were further upstream or downstream in the pathway and there was no statistical evidence that either method differed in which levels of this structure were detected.

A measure of [pathway](#) structure between individual [SLIPT](#) and [siRNA](#) genes within a pathway was also devised using the direction of [shortest paths](#) in a directed [graph](#) structure. This is amenable to detecting the consensus directionality of the pathway

across pairs of genes detected by either method. The [pathway](#) structure methodology developed here is generally applicable to comparison of [node](#) groups (allowing overlapping) including genes in biological pathways and their detection by different methodologies. While the [pathway](#) structure measure alone is not able to detect structural relationships between gene groups (e.g., [SLIPT](#) and [siRNA](#) gene candidates), it is amenable to resampling to determine whether these relationships are statistically significant.

5.5 Summary

Together these analyses of biological pathways, network metrics, and statistical procedures devised specifically for this purpose were applied to Reactome [pathway](#) structures to test whether structural relationships exist between [synthetic lethal](#) candidates. Of particular interest was whether these relationships relate to the differences between the computational ([SLIPT](#)) and experimental ([siRNA](#)) [synthetic lethal](#) candidate partners of *CDH1* (in the pathways discussed in Chapter 4).

While biologically relevant relationships were observed in specific pathways, there were few detectable structural relationships between [SLIPT](#) and [siRNA](#) gene candidates. These candidates did not exhibit significant differences in network connectivity or [centrality](#) measures. Network analyses were also unable to ascertain whether the candidates detected by either method stratified into upstream and downstream genes on the pathway and they likely do not.

A statistical resampling procedure was applied to [shortest path](#) analysis to test whether pairs of [SLIPT](#) and [siRNA](#) gene candidates were more likely to be upstream or downstream of each other. This approach detected very few structural relationships in the [synthetic lethal](#) pathways identified in Chapter 4. Overall, support for [pathway](#) structure between [SLIPT](#) and [siRNA](#) gene candidates is weak and the direction is inconsistent between pathways. Therefore [pathway](#) structure does not account for the differences between the [SLIPT](#) and [siRNA](#) gene candidates, although this does support the validity of gene set analyses in Chapter 4 and the [synthetic lethal](#) pathways identified.

Furthermore, the resampling procedure demonstrated in this chapter is more widely applicable to gene states in [network](#) structures and may be of further utility in the analysis of biological pathways or networks. This approach was able to quantify structural relationships that were otherwise difficult to interpret and to conclusively exclude many

potential relationships. In this respect, the network resampling methodology may also be applicable to triage of experimental validation.

Bibliography

- Aarts, M., Bajrami, I., Herrera-Abreu, M.T., Elliott, R., Brough, R., Ashworth, A., Lord, C.J., and Turner, N.C. (2015) Functional genetic screen identifies increased sensitivity to weel inhibition in cells with defects in fanconi anemia and hr pathways. *Mol Cancer Ther*, **14**(4): 865–76.
- Abeshouse, A., Ahn, J., Akbani, R., Ally, A., Amin, S., Andry, C.D., Annala, M., Aprikian, A., Armenia, J., Arora, A., *et al.* (2015) The Molecular Taxonomy of Primary Prostate Cancer. *Cell*, **163**(4): 1011–1025.
- Adler, D. (2005) *vioplot: Violin plot*. R package version 0.2.
- Akbani, R., Akdemir, K.C., Aksoy, B.A., Albert, M., Ally, A., Amin, S.B., Arachchi, H., Arora, A., Auman, J.T., Ayala, B., *et al.* (2015) Genomic Classification of Cutaneous Melanoma. *Cell*, **161**(7): 1681–1696.
- Akobeng, A.K. (2007) Understanding diagnostic tests 3: receiver operating characteristic curves. *Acta Pdiatrica*, **96**(5): 644–647.
- American Cancer Society (2017) Genetics and cancer. <https://www.cancer.org/cancer/cancer-causes/genetics.html>. Accessed: 22/03/2017.
- Anjomshoaa, A., Lin, Y.H., Black, M.A., McCall, J.L., Humar, B., Song, S., Fukuzawa, R., Yoon, H.S., Holzmann, B., Friederichs, J., *et al.* (2008) Reduced expression of a gene proliferation signature is associated with enhanced malignancy in colon cancer. *Br J Cancer*, **99**(6): 966–973.
- Araki, H., Knapp, C., Tsai, P., and Print, C. (2012) GeneSetDB: A comprehensive meta-database, statistical and visualisation framework for gene set analysis. *FEBS Open Bio*, **2**: 76–82.

- Ashburner, M., Ball, C.A., Blake, J.A., Botstein, D., Butler, H., Cherry, J.M., Davis, A.P., Dolinski, K., Dwight, S.S., Eppig, J.T., *et al.* (2000) Gene ontology: tool for the unification of biology. The Gene Ontology Consortium. *Nat Genet*, **25**(1): 25–29.
- Ashworth, A. (2008) A synthetic lethal therapeutic approach: poly(adp) ribose polymerase inhibitors for the treatment of cancers deficient in dna double-strand break repair. *J Clin Oncol*, **26**(22): 3785–90.
- Ashworth, A., Lord, C.J., and Reis-Filho, J.S. (2011) Genetic interactions in cancer progression and treatment. *Cell*, **145**(1): 30–38.
- Audeh, M.W., Carmichael, J., Penson, R.T., Friedlander, M., Powell, B., Bell-McGuinn, K.M., Scott, C., Weitzel, J.N., Oaknin, A., Loman, N., *et al.* (2010) Oral poly(adp-ribose) polymerase inhibitor olaparib in patients with *BRCA1* or *BRCA2* mutations and recurrent ovarian cancer: a proof-of-concept trial. *Lancet*, **376**(9737): 245–51.
- Babyak, M.A. (2004) What you see may not be what you get: a brief, nontechnical introduction to overfitting in regression-type models. *Psychosom Med*, **66**(3): 411–21.
- Bamford, S., Dawson, E., Forbes, S., Clements, J., Pettett, R., Dogan, A., Flanagan, A., Teague, J., Futreal, P.A., Stratton, M.R., *et al.* (2004) The COSMIC (Catalogue of Somatic Mutations in Cancer) database and website. *Br J Cancer*, **91**(2): 355–358.
- Barabási, A.L. and Albert, R. (1999) Emergence of scaling in random networks. *Science*, **286**(5439): 509–12.
- Barabási, A.L., Gulbahce, N., and Loscalzo, J. (2011) Network medicine: a network-based approach to human disease. *Nat Rev Genet*, **12**(1): 56–68.
- Barabási, A.L. and Oltvai, Z.N. (2004) Network biology: understanding the cell’s functional organization. *Nat Rev Genet*, **5**(2): 101–13.
- Barrat, A. and Weigt, M. (2000) On the properties of small-world network models. *The European Physical Journal B - Condensed Matter and Complex Systems*, **13**(3): 547–560.

- Barretina, J., Caponigro, G., Stransky, N., Venkatesan, K., Margolin, A.A., Kim, S., Wilson, C.J., Lehar, J., Kryukov, G.V., Sonkin, D., *et al.* (2012) The Cancer Cell Line Encyclopedia enables predictive modelling of anticancer drug sensitivity. *Nature*, **483**(7391): 603–607.
- Barry, W.T. (2016) *safe: Significance Analysis of Function and Expression*. R package version 3.14.0.
- Baryshnikova, A., Costanzo, M., Dixon, S., Vizeacoumar, F.J., Myers, C.L., Andrews, B., and Boone, C. (2010a) Synthetic genetic array (sga) analysis in *saccharomyces cerevisiae* and *schizosaccharomyces pombe*. *Methods Enzymol*, **470**: 145–79.
- Baryshnikova, A., Costanzo, M., Kim, Y., Ding, H., Koh, J., Toufighi, K., Youn, J.Y., Ou, J., San Luis, B.J., Bandyopadhyay, S., *et al.* (2010b) Quantitative analysis of fitness and genetic interactions in yeast on a genome scale. *Nat Meth*, **7**(12): 1017–1024.
- Bass, A.J., Thorsson, V., Shmulevich, I., Reynolds, S.M., Miller, M., Bernard, B., Hinoue, T., Laird, P.W., Curtis, C., Shen, H., *et al.* (2014) Comprehensive molecular characterization of gastric adenocarcinoma. *Nature*, **513**(7517): 202–209.
- Bates, D. and Maechler, M. (2016) *Matrix: Sparse and Dense Matrix Classes and Methods*. R package version 1.2-7.1.
- Bateson, W. and Mendel, G. (1909) *Mendel's principles of heredity, by W. Bateson*. University Press, Cambridge [Eng.].
- Becker, K.F., Atkinson, M.J., Reich, U., Becker, I., Nekarda, H., Siewert, J.R., and Hfler, H. (1994) E-cadherin gene mutations provide clues to diffuse type gastric carcinomas. *Cancer Research*, **54**(14): 3845–3852.
- Bell, D., Berchuck, A., Birrer, M., Chien, J., Cramer, D., Dao, F., Dhir, R., DiSaia, P., Gabra, H., Glenn, P., *et al.* (2011) Integrated genomic analyses of ovarian carcinoma. *Nature*, **474**(7353): 609–615.
- Benjamini, Y. and Hochberg, Y. (1995) Controlling the false discovery rate: A practical and powerful approach to multiple testing. *Journal of the Royal Statistical Society Series B (Methodological)*, **57**(1): 289–300.

- Berx, G., Cleton-Jansen, A.M., Nollet, F., de Leeuw, W.J., van de Vijver, M., Cornelisse, C., and van Roy, F. (1995) E-cadherin is a tumour/invasion suppressor gene mutated in human lobular breast cancers. *EMBO J*, **14**(24): 6107–15.
- Berx, G., Cleton-Jansen, A.M., Strumane, K., de Leeuw, W.J., Nollet, F., van Roy, F., and Cornelisse, C. (1996) E-cadherin is inactivated in a majority of invasive human lobular breast cancers by truncation mutations throughout its extracellular domain. *Oncogene*, **13**(9): 1919–25.
- Berx, G. and van Roy, F. (2009) Involvement of members of the cadherin superfamily in cancer. *Cold Spring Harb Perspect Biol*, **1**: a003129.
- Bitler, B.G., Aird, K.M., Garipov, A., Li, H., Amatangelo, M., Kossenkova, A.V., Schultz, D.C., Liu, Q., Shih, Ie, M., Conejo-Garcia, J.R., *et al.* (2015) Synthetic lethality by targeting ezh2 methyltransferase activity in arid1a-mutated cancers. *Nat Med*, **21**(3): 231–8.
- Blake, J.A., Christie, K.R., Dolan, M.E., Drabkin, H.J., Hill, D.P., Ni, L., Sitnikov, D., Burgess, S., Buza, T., Gresham, C., *et al.* (2015) Gene Ontology Consortium: going forward. *Nucleic Acids Res*, **43**(Database issue): D1049–1056.
- Boettcher, M., Lawson, A., Ladenburger, V., Fredebohm, J., Wolf, J., Hoheisel, J.D., Frezza, C., and Shlomi, T. (2014) High throughput synthetic lethality screen reveals a tumorigenic role of adenylate cyclase in fumarate hydratase-deficient cancer cells. *BMC Genomics*, **15**: 158.
- Boone, C., Bussey, H., and Andrews, B.J. (2007) Exploring genetic interactions and networks with yeast. *Nat Rev Genet*, **8**(6): 437–49.
- Borgatti, S.P. (2005) Centrality and network flow. *Social Networks*, **27**(1): 55 – 71.
- Boucher, B. and Jenna, S. (2013) Genetic interaction networks: better understand to better predict. *Front Genet*, **4**: 290.
- Bozovic-Spasojevic, I., Azambuja, E., McCaskill-Stevens, W., Dinh, P., and Cardoso, F. (2012) Chemoprevention for breast cancer. *Cancer treatment reviews*, **38**(5): 329–339.
- Breiman, L. (2001) Random forests. *Machine Learning*, **45**(1): 5–32.

- Brin, S. and Page, L. (1998) The anatomy of a large-scale hypertextual web search engine. *Computer Networks and ISDN Systems*, **30**(1): 107 – 117.
- Brouxhon, S.M., Kyrkanides, S., Teng, X., Athar, M., Ghazizadeh, S., Simon, M., O'Banion, M.K., and Ma, L. (2014) Soluble E-cadherin: a critical oncogene modulating receptor tyrosine kinases, MAPK and PI3K/Akt/mTOR signaling. *Oncogene*, **33**(2): 225–235.
- Brückner, A., Polge, C., Lentze, N., Auerbach, D., and Schlattner, U. (2009) Yeast two-hybrid, a powerful tool for systems biology. *Int J Mol Sci*, **10**(6): 2763–2788.
- Bryant, H.E., Schultz, N., Thomas, H.D., Parker, K.M., Flower, D., Lopez, E., Kyle, S., Meuth, M., Curtin, N.J., and Helleday, T. (2005) Specific killing of *BRCA2*-deficient tumours with inhibitors of polyadprbose polymerase. *Nature*, **434**(7035): 913–7.
- Bussey, H., Andrews, B., and Boone, C. (2006) From worm genetic networks to complex human diseases. *Nat Genet*, **38**(8): 862–3.
- Butland, G., Babu, M., Diaz-Mejia, J.J., Bohdana, F., Phanse, S., Gold, B., Yang, W., Li, J., Gagarinova, A.G., Pogoutse, O., *et al.* (2008) esga: E. coli synthetic genetic array analysis. *Nat Methods*, **5**(9): 789–95.
- cBioPortal for Cancer Genomics (cBioPortal) (2017) cBioPortal for Cancer Genomics. <http://www.cbioportal.org/>. Accessed: 26/03/2017.
- Cerami, E.G., Gross, B.E., Demir, E., Rodchenkov, I., Babur, O., Anwar, N., Schultz, N., Bader, G.D., and Sander, C. (2011) Pathway Commons, a web resource for biological pathway data. *Nucleic Acids Res*, **39**(Database issue): D685–690.
- Chen, A., Beetham, H., Black, M.A., Priya, R., Telford, B.J., Guest, J., Wiggins, G.A.R., Godwin, T.D., Yap, A.S., and Guilford, P.J. (2014) E-cadherin loss alters cytoskeletal organization and adhesion in non-malignant breast cells but is insufficient to induce an epithelial-mesenchymal transition. *BMC Cancer*, **14**(1): 552.
- Chen, S. and Parmigiani, G. (2007) Meta-analysis of BRCA1 and BRCA2 penetrance. *J Clin Oncol*, **25**(11): 1329–1333.
- Chipman, K. and Singh, A. (2009) Predicting genetic interactions with random walks on biological networks. *BMC Bioinformatics*, **10**(1): 17.

- Christofori, G. and Semb, H. (1999) The role of the cell-adhesion molecule E-cadherin as a tumour-suppressor gene. *Trends in Biochemical Sciences*, **24**(2): 73 – 76.
- Ciriello, G., Gatza, M.L., Beck, A.H., Wilkerson, M.D., Rhie, S.K., Pastore, A., Zhang, H., McLellan, M., Yau, C., Kandoth, C., *et al.* (2015) Comprehensive Molecular Portraits of Invasive Lobular Breast Cancer. *Cell*, **163**(2): 506–519.
- Clark, M.J. (2004) Endogenous Regulator of G Protein Signaling Proteins Suppress G α -Dependent μ -Opioid Agonist-Mediated Adenylyl Cyclase Supersensitization. *Journal of Pharmacology and Experimental Therapeutics*, **310**(1): 215–222.
- Collingridge, D.S. (2013) A primer on quantitized data analysis and permutation testing. *Journal of Mixed Methods Research*, **7**(1): 81–97.
- Collins, F.S. and Barker, A.D. (2007) Mapping the cancer genome. Pinpointing the genes involved in cancer will help chart a new course across the complex landscape of human malignancies. *Sci Am*, **296**(3): 50–57.
- Collisson, E., Campbell, J., Brooks, A., Berger, A., Lee, W., Chmielecki, J., Beer, D., Cope, L., Creighton, C., Danilova, L., *et al.* (2014) Comprehensive molecular profiling of lung adenocarcinoma. *Nature*, **511**(7511): 543–550.
- Costanzo, M., Baryshnikova, A., Bellay, J., Kim, Y., Spear, E.D., Sevier, C.S., Ding, H., Koh, J.L., Toufighi, K., Mostafavi, S., *et al.* (2010) The genetic landscape of a cell. *Science*, **327**(5964): 425–31.
- Costanzo, M., Baryshnikova, A., Myers, C.L., Andrews, B., and Boone, C. (2011) Charting the genetic interaction map of a cell. *Curr Opin Biotechnol*, **22**(1): 66–74.
- Courtney, K.D., Corcoran, R.B., and Engelman, J.A. (2010) The PI3K pathway as drug target in human cancer. *J Clin Oncol*, **28**(6): 1075–1083.
- Creighton, C.J., Morgan, M., Gunaratne, P.H., Wheeler, D.A., Gibbs, R.A., Robertson, A., Chu, A., Beroukhi, R., Cibulskis, K., Signoretti, S., *et al.* (2013) Comprehensive molecular characterization of clear cell renal cell carcinoma. *Nature*, **499**(7456): 43–49.
- Croft, D., Mundo, A.F., Haw, R., Milacic, M., Weiser, J., Wu, G., Caudy, M., Garapati, P., Gillespie, M., Kamdar, M.R., *et al.* (2014) The Reactome pathway knowledge-base. *Nucleic Acids Res*, **42**(database issue): D472D477.

- Crunkhorn, S. (2014) Cancer: Predicting synthetic lethal interactions. *Nat Rev Drug Discov*, **13**(11): 812.
- Csardi, G. and Nepusz, T. (2006) The igraph software package for complex network research. *InterJournal, Complex Systems*: 1695.
- Dai, X., Li, T., Bai, Z., Yang, Y., Liu, X., Zhan, J., and Shi, B. (2015) Breast cancer intrinsic subtype classification, clinical use and future trends. *Am J Cancer Res*, **5**(10): 2929–2943.
- Davierwala, A.P., Haynes, J., Li, Z., Brost, R.L., Robinson, M.D., Yu, L., Mnaimneh, S., Ding, H., Zhu, H., Chen, Y., *et al.* (2005) The synthetic genetic interaction spectrum of essential genes. *Nat Genet*, **37**(10): 1147–1152.
- De Leeuw, W.J., Berx, G., Vos, C.B., Peterse, J.L., Van de Vijver, M.J., Litvinov, S., Van Roy, F., Cornelisse, C.J., and Cleton-Jansen, A.M. (1997) Simultaneous loss of E-cadherin and catenins in invasive lobular breast cancer and lobular carcinoma in situ. *J Pathol*, **183**(4): 404–11.
- De Santis, G., Miotti, S., Mazzi, M., Canevari, S., and Tomassetti, A. (2009) E-cadherin directly contributes to PI3K/AKT activation by engaging the PI3K-p85 regulatory subunit to adherens junctions of ovarian carcinoma cells. *Oncogene*, **28**(9): 1206–1217.
- Demir, E., Babur, O., Rodchenkov, I., Aksoy, B.A., Fukuda, K.I., Gross, B., Sumer, O.S., Bader, G.D., and Sander, C. (2013) Using biological pathway data with Pax-tools. *PLoS Comput Biol*, **9**(9): e1003194.
- Deshpande, R., Asiedu, M.K., Klebig, M., Sutor, S., Kuzmin, E., Nelson, J., Piotrowski, J., Shin, S.H., Yoshida, M., Costanzo, M., *et al.* (2013) A comparative genomic approach for identifying synthetic lethal interactions in human cancer. *Cancer Res*, **73**(20): 6128–36.
- Dickson, D. (1999) Wellcome funds cancer database. *Nature*, **401**(6755): 729.
- Dijkstra, E.W. (1959) A note on two problems in connexion with graphs. *Numerische Mathematik*, **1**(1): 269–271.
- Dixon, S.J., Andrews, B.J., and Boone, C. (2009) Exploring the conservation of synthetic lethal genetic interaction networks. *Commun Integr Biol*, **2**(2): 78–81.

- Dixon, S.J., Fedyszyn, Y., Koh, J.L., Prasad, T.S., Chahwan, C., Chua, G., Toufighi, K., Baryshnikova, A., Hayles, J., Hoe, K.L., *et al.* (2008) Significant conservation of synthetic lethal genetic interaction networks between distantly related eukaryotes. *Proc Natl Acad Sci U S A*, **105**(43): 16653–8.
- Dong, L.L., Liu, L., Ma, C.H., Li, J.S., Du, C., Xu, S., Han, L.H., Li, L., and Wang, X.W. (2012) E-cadherin promotes proliferation of human ovarian cancer cells in vitro via activating MEK/ERK pathway. *Acta Pharmacol Sin*, **33**(6): 817–822.
- Dorsam, R.T. and Gutkind, J.S. (2007) G-protein-coupled receptors and cancer. *Nat Rev Cancer*, **7**(2): 79–94.
- Erdős, P. and Rényi, A. (1959) On random graphs I. *Publ Math Debrecen*, **6**: 290–297.
- Erdős, P. and Rényi, A. (1960) On the evolution of random graphs. In *Publ. Math. Inst. Hung. Acad. Sci*, volume 5, 17–61.
- Eroles, P., Bosch, A., Perez-Fidalgo, J.A., and Lluch, A. (2012) Molecular biology in breast cancer: intrinsic subtypes and signaling pathways. *Cancer Treat Rev*, **38**(6): 698–707.
- Farmer, H., McCabe, N., Lord, C.J., Tutt, A.N., Johnson, D.A., Richardson, T.B., Santarosa, M., Dillon, K.J., Hickson, I., Knights, C., *et al.* (2005) Targeting the dna repair defect in BRCA mutant cells as a therapeutic strategy. *Nature*, **434**(7035): 917–21.
- Fawcett, T. (2006) An introduction to ROC analysis. *Pattern Recognition Letters*, **27**(8): 861 – 874. {ROC} Analysis in Pattern Recognition.
- Fece de la Cruz, F., Gapp, B.V., and Nijman, S.M. (2015) Synthetic lethal vulnerabilities of cancer. *Annu Rev Pharmacol Toxicol*, **55**: 513–531.
- Ferlay, J., Soerjomataram, I., Dikshit, R., Eser, S., Mathers, C., Rebelo, M., Parkin, D.M., Forman, D., and Bray, F. (2015) Cancer incidence and mortality worldwide: sources, methods and major patterns in GLOBOCAN 2012. *Int J Cancer*, **136**(5): E359–386.
- Fisher, R.A. (1919) Xv.the correlation between relatives on the supposition of mendelian inheritance. *Earth and Environmental Science Transactions of the Royal Society of Edinburgh*, **52**(02): 399–433.

- Fong, P.C., Boss, D.S., Yap, T.A., Tutt, A., Wu, P., Mergui-Roelvink, M., Mortimer, P., Swaisland, H., Lau, A., O'Connor, M.J., *et al.* (2009) Inhibition of poly(adp-ribose) polymerase in tumors from BRCA mutation carriers. *N Engl J Med*, **361**(2): 123–34.
- Fong, P.C., Yap, T.A., Boss, D.S., Carden, C.P., Mergui-Roelvink, M., Gourley, C., De Greve, J., Lubinski, J., Shanley, S., Messiou, C., *et al.* (2010) Poly(adp)-ribose polymerase inhibition: frequent durable responses in BRCA carrier ovarian cancer correlating with platinum-free interval. *J Clin Oncol*, **28**(15): 2512–9.
- Forbes, S.A., Beare, D., Gunasekaran, P., Leung, K., Bindal, N., Boutselakis, H., Ding, M., Bamford, S., Cole, C., Ward, S., *et al.* (2015) COSMIC: exploring the world's knowledge of somatic mutations in human cancer. *Nucleic Acids Res*, **43**(Database issue): D805–811.
- Fraser, A. (2004) Towards full employment: using RNAi to find roles for the redundant. *Oncogene*, **23**(51): 8346–52.
- Fromental-Ramain, C., Warot, X., Lakkaraju, S., Favier, B., Haack, H., Birling, C., Dierich, A., Dollé, P., and Chambon, P. (1996) Specific and redundant functions of the paralogous Hoxa-9 and Hoxd-9 genes in forelimb and axial skeleton patterning. *Development*, **122**(2): 461–472.
- Futreal, P.A., Coin, L., Marshall, M., Down, T., Hubbard, T., Wooster, R., Rahman, N., and Stratton, M.R. (2004) A census of human cancer genes. *Nat Rev Cancer*, **4**(3): 177–183.
- Futreal, P.A., Kasprzyk, A., Birney, E., Mullikin, J.C., Wooster, R., and Stratton, M.R. (2001) Cancer and genomics. *Nature*, **409**(6822): 850–852.
- Gao, B. and Roux, P.P. (2015) Translational control by oncogenic signaling pathways. *Biochimica et Biophysica Acta*, **1849**(7): 753–65.
- Gatza, M.L., Kung, H.N., Blackwell, K.L., Dewhirst, M.W., Marks, J.R., and Chi, J.T. (2011) Analysis of tumor environmental response and oncogenic pathway activation identifies distinct basal and luminal features in HER2-related breast tumor subtypes. *Breast Cancer Res*, **13**(3): R62.

- Gatza, M.L., Lucas, J.E., Barry, W.T., Kim, J.W., Wang, Q., Crawford, M.D., Datto, M.B., Kelley, M., Mathey-Prevot, B., Potti, A., *et al.* (2010) A pathway-based classification of human breast cancer. *Proc Natl Acad Sci USA*, **107**(15): 6994–6999.
- Gatza, M.L., Silva, G.O., Parker, J.S., Fan, C., and Perou, C.M. (2014) An integrated genomics approach identifies drivers of proliferation in luminal-subtype human breast cancer. *Nat Genet*, **46**(10): 1051–1059.
- Gentleman, R.C., Carey, V.J., Bates, D.M., Bolstad, B., Dettling, M., Dudoit, S., Ellis, B., Gautier, L., Ge, Y., Gentry, J., *et al.* (2004) Bioconductor: open software development for computational biology and bioinformatics. *Genome Biol*, **5**(10): R80.
- Genz, A. and Bretz, F. (2009) Computation of multivariate normal and t probabilities. In *Lecture Notes in Statistics*, volume 195. Springer-Verlag, Heidelberg.
- Genz, A., Bretz, F., Miwa, T., Mi, X., Leisch, F., Scheipl, F., and Hothorn, T. (2016) *mvtnorm: Multivariate Normal and t Distributions*. R package version 1.0-5. URL.
- Glaire, M.A., Brown, M., Church, D.N., and Tomlinson, I. (2017) Cancer predisposition syndromes: lessons for truly precision medicine. *J Pathol*, **241**(2): 226–235.
- Globus (Globus) (2017) Research data management simplified. <https://www.globus.org/>. Accessed: 25/03/2017.
- Goodwin, S., McPherson, J.D., and McCombie, W.R. (2016) Coming of age: ten years of next-generation sequencing technologies. *Nat Rev Genet*, **17**(6): 333–351.
- Grady, W.M., Willis, J., Guilford, P.J., Dunbier, A.K., Toro, T.T., Lynch, H., Wiesner, G., Ferguson, K., Eng, C., Park, J.G., *et al.* (2000) Methylation of the CDH1 promoter as the second genetic hit in hereditary diffuse gastric cancer. *Nat Genet*, **26**(1): 16–17.
- Graziano, F., Humar, B., and Guilford, P. (2003) The role of the E-cadherin gene (*CDH1*) in diffuse gastric cancer susceptibility: from the laboratory to clinical practice. *Annals of Oncology*, **14**(12): 1705–1713.
- Guaragnella, N., Palermo, V., Galli, A., Moro, L., Mazzoni, C., and Giannattasio, S. (2014) The expanding role of yeast in cancer research and diagnosis: insights into the function of the oncosuppressors p53 and BRCA1/2. *FEMS Yeast Res*, **14**(1): 2–16.

- Güell, O., Sagus, F., and Serrano, M. (2014) Essential plasticity and redundancy of metabolism unveiled by synthetic lethality analysis. *PLoS Comput Biol*, **10**(5): e1003637.
- Guilford, P. (1999) E-cadherin downregulation in cancer: fuel on the fire? *Molecular Medicine Today*, **5**(4): 172 – 177.
- Guilford, P., Hopkins, J., Harraway, J., McLeod, M., McLeod, N., Harawira, P., Taite, H., Scoular, R., Miller, A., and Reeve, A.E. (1998) E-cadherin germline mutations in familial gastric cancer. *Nature*, **392**(6674): 402–5.
- Guilford, P., Humar, B., and Blair, V. (2010) Hereditary diffuse gastric cancer: translation of *CDH1* germline mutations into clinical practice. *Gastric Cancer*, **13**(1): 1–10.
- Guilford, P.J., Hopkins, J.B., Grady, W.M., Markowitz, S.D., Willis, J., Lynch, H., Rajput, A., Wiesner, G.L., Lindor, N.M., Burgart, L.J., *et al.* (1999) E-cadherin germline mutations define an inherited cancer syndrome dominated by diffuse gastric cancer. *Hum Mutat*, **14**(3): 249–55.
- Guo, J., Liu, H., and Zheng, J. (2016) SynLethDB: synthetic lethality database toward discovery of selective and sensitive anticancer drug targets. *Nucleic Acids Res*, **44**(D1): D1011–1017.
- Hajian-Tilaki, K. (2013) Receiver Operating Characteristic (ROC) Curve Analysis for Medical Diagnostic Test Evaluation. *Caspian J Intern Med*, **4**(2): 627–635.
- Hall, M., Frank, E., Holmes, G., Pfahringer, B., Reutemann, P., and Witten, I.H. (2009) The weka data mining software: an update. *SIGKDD Explor Newsl*, **11**(1): 10–18.
- Hammerman, P.S., Lawrence, M.S., Voet, D., Jing, R., Cibulskis, K., Sivachenko, A., Stojanov, P., McKenna, A., Lander, E.S., Gabriel, S., *et al.* (2012) Comprehensive genomic characterization of squamous cell lung cancers. *Nature*, **489**(7417): 519–525.
- Hanahan, D. and Weinberg, R.A. (2000) The hallmarks of cancer. *Cell*, **100**(1): 57–70.
- Hanahan, D. and Weinberg, R.A. (2011) Hallmarks of cancer: the next generation. *Cell*, **144**(5): 646–674.

- Hanna, S. (2003) Cancer incidence in new zealand (2003-2007). In D. Forman, D. Bray F Brewster, C. Gombe Mbalawa, B. Kohler, M. Piñeros, E. Steliarova-Foucher, R. Swaminathan, and J. Ferlay (editors), *Cancer Incidence in Five Continents*, volume X, 902–907. International Agency for Research on Cancer, Lyon, France. Electronic version <http://ci5.iarc.fr> Accessed 22/03/2017.
- Hansford, S., Kaurah, P., Li-Chang, H., Woo, M., Senz, J., Pinheiro, H., Schrader, K.A., Schaeffer, D.F., Shumansky, K., Zogopoulos, G., *et al.* (2015) Hereditary Diffuse Gastric Cancer Syndrome: CDH1 Mutations and Beyond. *JAMA Oncol*, **1**(1): 23–32.
- Heiskanen, M.A. and Aittokallio, T. (2012) Mining high-throughput screens for cancer drug targets-lessons from yeast chemical-genomic profiling and synthetic lethality. *Wiley Interdisciplinary Reviews: Data Mining and Knowledge Discovery*, **2**(3): 263–272.
- Hell, P. (1976) Graphs with given neighbourhoods i. problèmes combinatoires at theorie des graphes. *Proc Coil Int CNRS, Orsay*, **260**: 219–223.
- Higgins, M.E., Claremont, M., Major, J.E., Sander, C., and Lash, A.E. (2007) CancerGenes: a gene selection resource for cancer genome projects. *Nucleic Acids Res*, **35**(Database issue): D721–726.
- Hillenmeyer, M.E. (2008) The chemical genomic portrait of yeast: uncovering a phenotype for all genes. *Science*, **320**: 362–365.
- Hoadley, K.A., Yau, C., Wolf, D.M., Cherniack, A.D., Tamborero, D., Ng, S., Leiserson, M.D., Niu, B., McLellan, M.D., Uzunangelov, V., *et al.* (2014) Multiplatform analysis of 12 cancer types reveals molecular classification within and across tissues of origin. *Cell*, **158**(4): 929–944.
- Hoehndorf, R., Hardy, N.W., Osumi-Sutherland, D., Tweedie, S., Schofield, P.N., and Gkoutos, G.V. (2013) Systematic analysis of experimental phenotype data reveals gene functions. *PLoS ONE*, **8**(4): e60847.
- Holm, S. (1979) A simple sequentially rejective multiple test procedure. *Scandinavian Journal of Statistics*, **6**(2): 65–70.
- Hopkins, A.L. (2008) Network pharmacology: the next paradigm in drug discovery. *Nat Chem Biol*, **4**(11): 682–690.

- Hu, Z., Fan, C., Oh, D.S., Marron, J.S., He, X., Qaqish, B.F., Livasy, C., Carey, L.A., Reynolds, E., Dressler, L., *et al.* (2006) The molecular portraits of breast tumors are conserved across microarray platforms. *BMC Genomics*, **7**: 96.
- Huang, E., Cheng, S., Dressman, H., Pittman, J., Tsou, M., Horng, C., Bild, A., Iversen, E., Liao, M., Chen, C., *et al.* (2003) Gene expression predictors of breast cancer outcomes. *Lancet*, **361**: 1590–1596.
- Hutchison, C.A., Chuang, R.Y., Noskov, V.N., Assad-Garcia, N., Deerinck, T.J., Ellisman, M.H., Gill, J., Kannan, K., Karas, B.J., Ma, L., *et al.* (2016) Design and synthesis of a minimal bacterial genome. *Science*, **351**(6280): aad6253.
- International HapMap 3 Consortium (HapMap) (2003) The International HapMap Project. *Nature*, **426**(6968): 789–796.
- Jeanes, A., Gottardi, C.J., and Yap, A.S. (2008) Cadherins and cancer: how does cadherin dysfunction promote tumor progression? *Oncogene*, **27**(55): 6920–6929.
- Jerby-Arnon, L., Pfetzer, N., Waldman, Y., McGarry, L., James, D., Shanks, E., Seashore-Ludlow, B., Weinstock, A., Geiger, T., Clemons, P., *et al.* (2014) Predicting cancer-specific vulnerability via data-driven detection of synthetic lethality. *Cell*, **158**(5): 1199–1209.
- Joachims, T. (1999) Making large-scale support vector machine learning practical. In S. Bernhard, I. Kropf, J.C.B. Christopher, and J.S. Alexander (editors), *Advances in kernel methods*, 169–184. MIT Press.
- Ju, Z., Liu, W., Roebuck, P.L., Siwak, D.R., Zhang, N., Lu, Y., Davies, M.A., Akbani, R., Weinstein, J.N., Mills, G.B., *et al.* (2015) Development of a robust classifier for quality control of reverse-phase protein arrays. *Bioinformatics*, **31**(6): 912.
- Kaelin, Jr, W. (2005) The concept of synthetic lethality in the context of anticancer therapy. *Nat Rev Cancer*, **5**(9): 689–98.
- Kaelin, Jr, W. (2009) Synthetic lethality: a framework for the development of wiser cancer therapeutics. *Genome Med*, **1**: 99.
- Kamada, T. and Kawai, S. (1989) An algorithm for drawing general undirected graphs. *Information Processing Letters*, **31**(1): 7–15.

- Kawai, J., Shinagawa, A., Shibata, K., Yoshino, M., Itoh, M., Ishii, Y., Arakawa, T., Hara, A., Fukunishi, Y., Konno, H., *et al.* (2001) Functional annotation of a full-length mouse cDNA collection. *Nature*, **409**(6821): 685–690.
- Kelley, R. and Ideker, T. (2005) Systematic interpretation of genetic interactions using protein networks. *Nat Biotech*, **23**(5): 561–566.
- Kelly, S.T. (2013) *Statistical Predictions of Synthetic Lethal Interactions in Cancer*. Dissertation, University of Otago.
- Keshava Prasad, T.S., Goel, R., Kandasamy, K., Keerthikumar, S., Kumar, S., Mathivanan, S., Telikicherla, D., Raju, R., Shafreen, B., Venugopal, A., *et al.* (2009) Human Protein Reference Database–2009 update. *Nucleic Acids Res*, **37**(Database issue): D767–772.
- Kim, N.G., Koh, E., Chen, X., and Gumbiner, B.M. (2011) E-cadherin mediates contact inhibition of proliferation through Hippo signaling-pathway components. *Proc Natl Acad Sci USA*, **108**(29): 11930–11935.
- Koboldt, D.C., Fulton, R.S., McLellan, M.D., Schmidt, H., Kalicki-Veizer, J., McMichael, J.F., Fulton, L.L., Dooling, D.J., Ding, L., Mardis, E.R., *et al.* (2012) Comprehensive molecular portraits of human breast tumours. *Nature*, **490**(7418): 61–70.
- Kockel, L., Zeitlinger, J., Staszewski, L.M., Mlodzik, M., and Bohmann, D. (1997) Jun in drosophila development: redundant and nonredundant functions and regulation by two mapk signal transduction pathways. *Genes & Development*, **11**(13): 1748–1758.
- Kozlov, K.N., Gursky, V.V., Kulakovskiy, I.V., and Samsonova, M.G. (2015) Sequence-based model of gap gene regulation network. *BMC Genomics*, **15**(Suppl 12): S6.
- Kranthi, S., Rao, S., and Manimaran, P. (2013) Identification of synthetic lethal pairs in biological systems through network information centrality. *Mol BioSyst*, **9**(8): 2163–2167.
- Kroepil, F., Fluegen, G., Totikov, Z., Baldus, S.E., Vay, C., Schauer, M., Topp, S.A., Esch, J.S., Knoefel, W.T., and Stoecklein, N.H. (2012) Down-regulation of CDH1 is associated with expression of SNAI1 in colorectal adenomas. *PLoS ONE*, **7**(9): e46665.

- Lander, E.S. (2011) Initial impact of the sequencing of the human genome. *Nature*, **470**(7333): 187–197.
- Lander, E.S., Linton, L.M., Birren, B., Nusbaum, C., Zody, M.C., Baldwin, J., Devon, K., Dewar, K., Doyle, M., FitzHugh, W., *et al.* (2001) Initial sequencing and analysis of the human genome. *Nature*, **409**(6822): 860–921.
- Langmead, B., Trapnell, C., Pop, M., and Salzberg, S.L. (2009) Ultrafast and memory-efficient alignment of short DNA sequences to the human genome. *Genome Biol*, **10**(3): R25.
- Latora, V. and Marchiori, M. (2001) Efficient behavior of small-world networks. *Phys Rev Lett*, **87**: 198701.
- Laufer, C., Fischer, B., Billmann, M., Huber, W., and Boutros, M. (2013) Mapping genetic interactions in human cancer cells with RNAi and multiparametric phenotyping. *Nat Methods*, **10**(5): 427–31.
- Law, C.W., Chen, Y., Shi, W., and Smyth, G.K. (2014) voom: precision weights unlock linear model analysis tools for RNA-seq read counts. *Genome Biol*, **15**(2): R29.
- Le Meur, N. and Gentleman, R. (2008) Modeling synthetic lethality. *Genome Biol*, **9**(9): R135.
- Le Meur, N., Jiang, Z., Liu, T., Mar, J., and Gentleman, R.C. (2014) Slgi: Synthetic lethal genetic interaction. r package version 1.26.0.
- Lee, A.Y., Perreault, R., Harel, S., Boulier, E.L., Suderman, M., Hallett, M., and Jenna, S. (2010a) Searching for signaling balance through the identification of genetic interactors of the rab guanine-nucleotide dissociation inhibitor gdi-1. *PLoS ONE*, **5**(5): e10624.
- Lee, I., Lehner, B., Vavouri, T., Shin, J., Fraser, A.G., and Marcotte, E.M. (2010b) Predicting genetic modifier loci using functional gene networks. *Genome Research*, **20**(8): 1143–1153.
- Lee, I. and Marcotte, E.M. (2009) Effects of functional bias on supervised learning of a gene network model. *Methods Mol Biol*, **541**: 463–75.

- Lee, M.J., Ye, A.S., Gardino, A.K., Heijink, A.M., Sorger, P.K., MacBeath, G., and Yaffe, M.B. (2012) Sequential application of anticancer drugs enhances cell death by rewiring apoptotic signaling networks. *Cell*, **149**(4): 780–94.
- Lehner, B., Crombie, C., Tischler, J., Fortunato, A., and Fraser, A.G. (2006) Systematic mapping of genetic interactions in *caenorhabditis elegans* identifies common modifiers of diverse signaling pathways. *Nat Genet*, **38**(8): 896–903.
- Li, B., Ruotti, V., Stewart, R.M., Thomson, J.A., and Dewey, C.N. (2010) RNA-Seq gene expression estimation with read mapping uncertainty. *Bioinformatics*, **26**(4): 493–500.
- Li, X.J., Mishra, S.K., Wu, M., Zhang, F., and Zheng, J. (2014) Syn-lethality: An integrative knowledge base of synthetic lethality towards discovery of selective anticancer therapies. *Biomed Res Int*, **2014**: 196034.
- Linehan, W.M., Spellman, P.T., Ricketts, C.J., Creighton, C.J., Fei, S.S., Davis, C., Wheeler, D.A., Murray, B.A., Schmidt, L., Vocke, C.D., *et al.* (2016) Comprehensive Molecular Characterization of Papillary Renal-Cell Carcinoma. *N Engl J Med*, **374**(2): 135–145.
- Lokody, I. (2014) Computational modelling: A computational crystal ball. *Nature Reviews Cancer*, **14**(10): 649–649.
- Lord, C.J., Tutt, A.N., and Ashworth, A. (2015) Synthetic lethality and cancer therapy: lessons learned from the development of PARP inhibitors. *Annu Rev Med*, **66**: 455–470.
- Lu, X., Kensche, P.R., Huynen, M.A., and Notebaart, R.A. (2013) Genome evolution predicts genetic interactions in protein complexes and reveals cancer drug targets. *Nat Commun*, **4**: 2124.
- Lu, X., Megchelenbrink, W., Notebaart, R.A., and Huynen, M.A. (2015) Predicting human genetic interactions from cancer genome evolution. *PLoS One*, **10**(5): e0125795.
- Lum, P.Y., Armour, C.D., Stepaniants, S.B., Cavet, G., Wolf, M.K., Butler, J.S., Hinshaw, J.C., Garnier, P., Prestwich, G.D., Leonardson, A., *et al.* (2004) Discovering modes of action for therapeutic compounds using a genome-wide screen of yeast heterozygotes. *Cell*, **116**(1): 121–137.

- Luo, J., Solimini, N.L., and Elledge, S.J. (2009) Principles of Cancer Therapy: Oncogene and Non-oncogene Addiction. *Cell*, **136**(5): 823–837.
- Machado, J., Olivera, C., Carvalh, R., Soares, P., Berx, G., Caldas, C., Sercuca, R., Carneiro, F., and Sorbrinho-Simoes, M. (2001) E-cadherin gene (*CDH1*) promoter methylation as the second hit in sporadic diffuse gastric carcinoma. *Oncogene*, **20**: 1525–1528.
- Markowetz, F. (2017) All biology is computational biology. *PLoS Biol*, **15**(3): e2002050.
- Masciari, S., Larsson, N., Senz, J., Boyd, N., Kaurah, P., Kandel, M.J., Harris, L.N., Pinheiro, H.C., Troussard, A., Miron, P., *et al.* (2007) Germline E-cadherin mutations in familial lobular breast cancer. *J Med Genet*, **44**(11): 726–31.
- Mattison, J., van der Weyden, L., Hubbard, T., and Adams, D.J. (2009) Cancer gene discovery in mouse and man. *Biochim Biophys Acta*, **1796**(2): 140–161.
- McLachlan, J., George, A., and Banerjee, S. (2016) The current status of parp inhibitors in ovarian cancer. *Tumori*, **102**(5): 433–440.
- McLendon, R., Friedman, A., Bigner, D., Van Meir, E.G., Brat, D.J., Mastrogianakis, G.M., Olson, J.J., Mikkelsen, T., Lehman, N., Aldape, K., *et al.* (2008) Comprehensive genomic characterization defines human glioblastoma genes and core pathways. *Nature*, **455**(7216): 1061–1068.
- Miles, D.W. (2001) Update on HER-2 as a target for cancer therapy: herceptin in the clinical setting. *Breast Cancer Res*, **3**(6): 380–384.
- Muzny, D.M., Bainbridge, M.N., Chang, K., Dinh, H.H., Drummond, J.A., Fowler, G., Kovar, C.L., Lewis, L.R., Morgan, M.B., Newsham, I.F., *et al.* (2012) Comprehensive molecular characterization of human colon and rectal cancer. *Nature*, **487**(7407): 330–337.
- Nagalla, S., Chou, J.W., Willingham, M.C., Ruiz, J., Vaughn, J.P., Dubey, P., Lash, T.L., Hamilton-Dutoit, S.J., Bergh, J., Sotiriou, C., *et al.* (2013) Interactions between immunity, proliferation and molecular subtype in breast cancer prognosis. *Genome Biol*, **14**(4): R34.
- Neeley, E.S., Kornblau, S.M., Coombes, K.R., and Baggerly, K.A. (2009) Variable slope normalization of reverse phase protein arrays. *Bioinformatics*, **25**(11): 1384.

- Novomestky, F. (2012) *matrixcalc: Collection of functions for matrix calculations*. R package version 1.0-3.
- Nowak, M.A., Boerlijst, M.C., Cooke, J., and Smith, J.M. (1997) Evolution of genetic redundancy. *Nature*, **388**(6638): 167–171.
- Oliveira, C., Senz, J., Kaurah, P., Pinheiro, H., Sanges, R., Haegert, A., Corso, G., Schouten, J., Fitzgerald, R., Vogelsang, H., *et al.* (2009) Germline *CDH1* deletions in hereditary diffuse gastric cancer families. *Human Molecular Genetics*, **18**(9): 1545–1555.
- Oliveira, C., Seruca, R., Hoogerbrugge, N., Ligtenberg, M., and Carneiro, F. (2013) Clinical utility gene card for: Hereditary diffuse gastric cancer (HDGC). *Eur J Hum Genet*, **21**(8).
- Pandey, G., Zhang, B., Chang, A.N., Myers, C.L., Zhu, J., Kumar, V., and Schadt, E.E. (2010) An integrative multi-network and multi-classifier approach to predict genetic interactions. *PLoS Comput Biol*, **6**(9).
- Parker, J., Mullins, M., Cheung, M., Leung, S., Voduc, D., Vickery, T., Davies, S., Fauron, C., He, X., Hu, Z., *et al.* (2009) Supervised risk predictor of breast cancer based on intrinsic subtypes. *Journal of Clinical Oncology*, **27**(8): 1160–1167.
- Pereira, B., Chin, S.F., Rueda, O.M., Volland, H.K., Provenzano, E., Bardwell, H.A., Pugh, M., Jones, L., Russell, R., Sammut, S.J., *et al.* (2016) Erratum: The somatic mutation profiles of 2,433 breast cancers refine their genomic and transcriptomic landscapes. *Nat Commun*, **7**: 11908.
- Perou, C.M., Sørli, T., Eisen, M.B., van de Rijn, M., Jeffrey, S.S., Rees, C.A., Pollack, J.R., Ross, D.T., Johnsen, H., Akslen, L.A., *et al.* (2000) Molecular portraits of human breast tumours. *Nature*, **406**(6797): 747–752.
- Polyak, K. and Weinberg, R.A. (2009) Transitions between epithelial and mesenchymal states: acquisition of malignant and stem cell traits. *Nat Rev Cancer*, **9**(4): 265–73.
- R Core Team (2016) *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria. R version 3.3.2.
- Ritchie, M.E., Phipson, B., Wu, D., Hu, Y., Law, C.W., Shi, W., and Smyth, G.K. (2015) limma powers differential expression analyses for RNA-sequencing and microarray studies. *Nucleic Acids Research*, **43**(7): e47.

- Roguev, A., Bandyopadhyay, S., Zofall, M., Zhang, K., Fischer, T., Collins, S.R., Qu, H., Shales, M., Park, H.O., Hayles, J., *et al.* (2008) Conservation and rewiring of functional modules revealed by an epistasis map in fission yeast. *Science*, **322**(5900): 405–10.
- Roychowdhury, S. and Chinnaiyan, A.M. (2016) Translating cancer genomes and transcriptomes for precision oncology. *CA Cancer J Clin*, **66**(1): 75–88.
- Rung, J. and Brazma, A. (2013) Reuse of public genome-wide gene expression data. *Nat Rev Genet*, **14**(2): 89–99.
- Ryan, C., Lord, C., and Ashworth, A. (2014) Daisy: Picking synthetic lethals from cancer genomes. *Cancer Cell*, **26**(3): 306–308.
- Schena, M. (1996) Genome analysis with gene expression microarrays. *Bioessays*, **18**(5): 427–431.
- Scheuer, L., Kauff, N., Robson, M., Kelly, B., Barakat, R., Satagopan, J., Ellis, N., Hensley, M., Boyd, J., Borgen, P., *et al.* (2002) Outcome of preventive surgery and screening for breast and ovarian cancer in BRCA mutation carriers. *J Clin Oncol*, **20**(5): 1260–1268.
- Semb, H. and Christofori, G. (1998) The tumor-suppressor function of E-cadherin. *Am J Hum Genet*, **63**(6): 1588–93.
- Sing, T., Sander, O., Beerenwinkel, N., and Lengauer, T. (2005) Rocr: visualizing classifier performance in r. *Bioinformatics*, **21**(20): 7881.
- Slurm development team (Slurm) (2017) Slurm workload manager. <https://slurm.schedmd.com/>. Accessed: 25/03/2017.
- Sørli, T., Perou, C.M., Tibshirani, R., Aas, T., Geisler, S., Johnsen, H., Hastie, T., Eisen, M.B., van de Rijn, M., Jeffrey, S.S., *et al.* (2001) Gene expression patterns of breast carcinomas distinguish tumor subclasses with clinical implications. *Proc Natl Acad Sci USA*, **98**(19): 10869–10874.
- Srihari, S., Singla, J., Wong, L., and Ragan, M.A. (2015) Inferring synthetic lethal interactions from mutual exclusivity of genetic events in cancer. *Biology Direct*, **10**(1): 57.

- Stajich, J.E. and Lapp, H. (2006) Open source tools and toolkits for bioinformatics: significance, and where are we? *Brief Bioinformatics*, **7**(3): 287–296.
- Stratton, M.R., Campbell, P.J., and Futreal, P.A. (2009) The cancer genome. *Nature*, **458**(7239): 719–724.
- Ström, C. and Helleday, T. (2012) Strategies for the use of poly(adenosine diphosphate ribose) polymerase (parp) inhibitors in cancer therapy. *Biomolecules*, **2**(4): 635–649.
- Tarazona, S., Garcia-Alcalde, F., Dopazo, J., Ferrer, A., and Conesa, A. (2011) Differential expression in RNA-seq: a matter of depth. *Genome Res*, **21**(12): 2213–2223.
- Telford, B.J., Chen, A., Beetham, H., Frick, J., Brew, T.P., Gould, C.M., Single, A., Godwin, T., Simpson, K.J., and Guilford, P. (2015) Synthetic lethal screens identify vulnerabilities in gpcr signalling and cytoskeletal organization in E-cadherin-deficient cells. *Mol Cancer Ther*, **14**(5): 1213–1223.
- The 1000 Genomes Project Consortium (1000 Genomes) (2010) A map of human genome variation from population-scale sequencing. *Nature*, **467**(7319): 1061–1073.
- The Cancer Genome Atlas Research Network (TCGA) (2017) The Cancer Genome Atlas Project. <https://cancergenome.nih.gov/>. Accessed: 26/03/2017.
- The Catalogue Of Somatic Mutations In Cancer (COSMIC) (2016) Cosmic: The catalogue of somatic mutations in cancer. <http://cancer.sanger.ac.uk/cosmic>. Release 79 (23/08/2016), Accessed: 05/02/2017.
- The Comprehensive R Archive Network (CRAN) (2017) Cran. <https://cran.r-project.org/>. Accessed: 24/03/2017.
- The ENCODE Project Consortium (ENCODE) (2004) The ENCODE (ENCyclopedia Of DNA Elements) Project. *Science*, **306**(5696): 636–640.
- The National Cancer Institute (NCI) (2015) The genetics of cancer. <https://www.cancer.gov/about-cancer/causes-prevention/genetics>. Published: 22/04/2015, Accessed: 22/03/2017.
- The New Zealand eScience Infrastructure (NeSI) (2017) NeSI. <https://www.nesi.org.nz/>. Accessed: 25/03/2017.
- Tierney, L., Rossini, A.J., Li, N., and Sevcikova, H. (2015) *snow: Simple Network of Workstations*. R package version 0.4-2.

- Tiong, K.L., Chang, K.C., Yeh, K.T., Liu, T.Y., Wu, J.H., Hsieh, P.H., Lin, S.H., Lai, W.Y., Hsu, Y.C., Chen, J.Y., *et al.* (2014) Csnk1e/ctnnb1 are synthetic lethal to tp53 in colorectal cancer and are markers for prognosis. *Neoplasia*, **16**(5): 441–50.
- Tischler, J., Lehner, B., and Fraser, A.G. (2008) Evolutionary plasticity of genetic interaction networks. *Nat Genet*, **40**(4): 390–391.
- Tomasetti, C. and Vogelstein, B. (2015) Cancer etiology. Variation in cancer risk among tissues can be explained by the number of stem cell divisions. *Science*, **347**(6217): 78–81.
- Tong, A.H., Evangelista, M., Parsons, A.B., Xu, H., Bader, G.D., Page, N., Robinson, M., Raghibizadeh, S., Hogue, C.W., Bussey, H., *et al.* (2001) Systematic genetic analysis with ordered arrays of yeast deletion mutants. *Science*, **294**(5550): 2364–8.
- Tong, A.H., Lesage, G., Bader, G.D., Ding, H., Xu, H., Xin, X., Young, J., Berriz, G.F., Brost, R.L., Chang, M., *et al.* (2004) Global mapping of the yeast genetic interaction network. *Science*, **303**(5659): 808–13.
- Tran, B., Dancey, J.E., Kamel-Reid, S., McPherson, J.D., Bedard, P.L., Brown, A.M., Zhang, T., Shaw, P., Onetto, N., Stein, L., *et al.* (2012) Cancer genomics: technology, discovery, and translation. *J Clin Oncol*, **30**(6): 647–660.
- Travers, J. and Milgram, S. (1969) An experimental study of the small world problem. *Sociometry*, **32**(4): 425–443.
- Tunggal, J.A., Helfrich, I., Schmitz, A., Schwarz, H., Gunzel, D., Fromm, M., Kemler, R., Krieg, T., and Niessen, C.M. (2005) E-cadherin is essential for in vivo epidermal barrier function by regulating tight junctions. *EMBO J*, **24**(6): 1146–1156.
- Tutt, A., Robson, M., Garber, J.E., Domchek, S.M., Audeh, M.W., Weitzel, J.N., Friedlander, M., Arun, B., Loman, N., Schmutzler, R.K., *et al.* (2010) Oral poly(adp-ribose) polymerase inhibitor olaparib in patients with *BRCA1* or *BRCA2* mutations and advanced breast cancer: a proof-of-concept trial. *Lancet*, **376**(9737): 235–44.
- University of California, Santa Cruz (UCSC) (2012) Uscs cancer browser. Accessed 29/03/2012.
- van der Meer, R., Song, H.Y., Park, S.H., Abdulkadir, S.A., and Roh, M. (2014) RNAi screen identifies a synthetic lethal interaction between PIM1 overexpression and PLK1 inhibition. *Clinical Cancer Research*, **20**(12): 3211–3221.

- van der Post, R.S., Vogelaar, I.P., Carneiro, F., Guilford, P., Huntsman, D., Hoogerbrugge, N., Caldas, C., Schreiber, K.E., Hardwick, R.H., Ausems, M.G., *et al.* (2015) Hereditary diffuse gastric cancer: updated clinical guidelines with an emphasis on germline CDH1 mutation carriers. *J Med Genet*, **52**(6): 361–374.
- van Steen, K. (2012) Travelling the world of genegene interactions. *Briefings in Bioinformatics*, **13**(1): 1–19.
- van Steen, M. (2010) *Graph Theory and Complex Networks: An Introduction*. Maarten van Steen, VU Amsterdam.
- Vapnik, V.N. (1995) *The nature of statistical learning theory*. Springer-Verlag New York, Inc.
- Vizeacoumar, F.J., Arnold, R., Vizeacoumar, F.S., Chandrashekhar, M., Buzina, A., Young, J.T., Kwan, J.H., Sayad, A., Mero, P., Lawo, S., *et al.* (2013) A negative genetic interaction map in isogenic cancer cell lines reveals cancer cell vulnerabilities. *Mol Syst Biol*, **9**: 696.
- Vogelstein, B., Papadopoulos, N., Velculescu, V.E., Zhou, S., Diaz, L.A., and Kinzler, K.W. (2013) Cancer genome landscapes. *Science*, **339**(6127): 1546–1558.
- Vos, C.B., Cleton-Jansen, A.M., Berx, G., de Leeuw, W.J., ter Haar, N.T., van Roy, F., Cornelisse, C.J., Peterse, J.L., and van de Vijver, M.J. (1997) E-cadherin inactivation in lobular carcinoma in situ of the breast: an early event in tumorigenesis. *Br J Cancer*, **76**(9): 1131–3.
- Waldron, D. (2016) Cancer genomics: A multi-layer omics approach to cancer. *Nat Rev Genet*, **17**(8): 436–437.
- Wang, K., Singh, D., Zeng, Z., Coleman, S.J., Huang, Y., Savich, G.L., He, X., Mieczkowski, P., Grimm, S.A., Perou, C.M., *et al.* (2010) MapSplice: accurate mapping of RNA-seq reads for splice junction discovery. *Nucleic Acids Res*, **38**(18): e178.
- Wang, X. and Simon, R. (2013) Identification of potential synthetic lethal genes to p53 using a computational biology approach. *BMC Medical Genomics*, **6**(1): 30.
- Wappett, M. (2014) Bisep: Toolkit to identify candidate synthetic lethality. r package version 2.0.

- Wappett, M., Dulak, A., Yang, Z.R., Al-Watban, A., Bradford, J.R., and Dry, J.R. (2016) Multi-omic measurement of mutually exclusive loss-of-function enriches for candidate synthetic lethal gene pairs. *BMC Genomics*, **17**: 65.
- Warnes, G.R., Bolker, B., Bonebakker, L., Gentleman, R., Liaw, W.H.A., Lumley, T., Maechler, M., Magnusson, A., Moeller, S., Schwartz, M., *et al.* (2015) *gplots: Various R Programming Tools for Plotting Data*. R package version 2.17.0.
- Watts, D.J. and Strogatz, S.H. (1998) Collective dynamics of 'small-world' networks. *Nature*, **393**(6684): 440–2.
- Weinstein, I.B. (2000) Disorders in cell circuitry during multistage carcinogenesis: the role of homeostasis. *Carcinogenesis*, **21**(5): 857–864.
- Weinstein, J.N., Akbani, R., Broom, B.M., Wang, W., Verhaak, R.G., McConkey, D., Lerner, S., Morgan, M., Creighton, C.J., Smith, C., *et al.* (2014) Comprehensive molecular characterization of urothelial bladder carcinoma. *Nature*, **507**(7492): 315–322.
- Weinstein, J.N., Collisson, E.A., Mills, G.B., Shaw, K.R., Ozenberger, B.A., Ellrott, K., Shmulevich, I., Sander, C., Stuart, J.M., Chang, K., *et al.* (2013) The Cancer Genome Atlas Pan-Cancer analysis project. *Nat Genet*, **45**(10): 1113–1120.
- Wickham, H. and Chang, W. (2016) *devtools: Tools to Make Developing R Packages Easier*. R package version 1.12.0.
- Wickham, H., Danenberg, P., and Eugster, M. (2017) *roxygen2: In-Line Documentation for R*. R package version 6.0.1.
- Wong, S.L., Zhang, L.V., Tong, A.H.Y., Li, Z., Goldberg, D.S., King, O.D., Lesage, G., Vidal, M., Andrews, B., Bussey, H., *et al.* (2004) Combining biological networks to predict genetic interactions. *Proceedings of the National Academy of Sciences of the United States of America*, **101**(44): 15682–15687.
- World Health Organization (WHO) (2017) Fact sheet: Cancer. <http://www.who.int/mediacentre/factsheets/fs297/en/>. Updated February 2017, Accessed: 22/03/2017.
- Wu, M., Li, X., Zhang, F., Li, X., Kwoh, C.K., and Zheng, J. (2014) In silico prediction of synthetic lethality by meta-analysis of genetic interactions, functions, and pathways in yeast and human cancer. *Cancer Inform*, **13**(Suppl 3): 71–80.

- Yu, H. (2002) Rmpi: Parallel statistical computing in r. *R News*, **2**(2): 10–14.
- Zhang, F., Wu, M., Li, X.J., Li, X.L., Kwoh, C.K., and Zheng, J. (2015) Predicting essential genes and synthetic lethality via influence propagation in signaling pathways of cancer cell fates. *J Bioinform Comput Biol*, **13**(3): 1541002.
- Zhang, J., Baran, J., Cros, A., Guberman, J.M., Haider, S., Hsu, J., Liang, Y., Rivkin, E., Wang, J., Whitty, B., *et al.* (2011) International cancer genome consortium data portal a one-stop shop for cancer genomics data. *Database: The Journal of Biological Databases and Curation*, **2011**: bar026.
- Zhong, W. and Sternberg, P.W. (2006) Genome-wide prediction of c. elegans genetic interactions. *Science*, **311**(5766): 1481–1484.
- Zweig, M.H. and Campbell, G. (1993) Receiver-operating characteristic (roc) plots: a fundamental evaluation tool in clinical medicine. *Clinical Chemistry*, **39**(4): 561–577.