## Analysis of Births Ratios from Muscle TSC1/Raptor Flies

### Isabelle Hatfield and Dave Bridges

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### Experimental Design

These data are stored in /Volumes/bridges\_lab/Hatfield/Drosophila/Births, with the raw data saved in ../Data/birth\_worksheet.csv. This script was most recently run on Mon Feb 24 09:12:45 2014

#### Strain Summaries

So far this analysis includes **6488** flies from 18 crosses. Statistics are based on a Chi Squared Test, and are adjusted for multiple observation by the method of Benjamini and Hochberg. These statistics exclude the genotypically WT (dual balancer chromosome) strains, which have significantly less progeny in nearly all cases (see Table 9). Counts and p-values are shown in Table 2 and Figures 1 and 3 for all drivers, Table 7 for all crosses and Table 8 with gendered crossed data. Summarized per cross the data are below.

#### Pre-Eclosure Lethality of Raptor Strains with 24B-Gal4

The three Raptor knockout strains all had much less progeny than would be expected with 24B-GAL4 driven expression. The adjusted p-values for these effects are 1.30925507135797e-31, 6.61466969174944e-33, 0.01059795534579 respectively for shRNA's 31529, 34814, 35144.

#### Crosses with C179-Gal4

Since there was embryonic lethality of the Raptor strains with 24B-Gal4 we repeated these crosses with C179-Gal4. These results are shown in Tables 3 and 4 as well as Figure ??. There is a gender specific effect for only one of these crosses (see Table 5 for Fisher tests comparing gender for each shRNA strain and Figure 5).

#### Effects of 18C

Since we observed few births at 24C, we repeated several crosses at 18C using the TSC shRNA lines. These data are summarised in Table 6 and Figure 2

Driver	Gene	KD	$\operatorname{shRNA}$	GAL4	WT	Total	pval	padj
24B-Gal4	Raptor	8	433	374	188	1003	0.00000	0.00000
24B-Gal4	Tsc1	457	416	322	168	1363	0.00001	0.00001
Hand-Gal4	Raptor	385	489	311	244	1429	0.00000	0.00000
Hand-Gal4	Tsc1	400	320	240	102	1062	0.00000	0.00000

Table 1: Gene Level Summarised Data

Driver	UAS	KD	shRNA	GAL4	WT	Total	pval	padj
24B-Gal4	31039	165	133	101	47	446	0.00045	0.00091
24B-Gal $4$	31314	126	89	82	36	333	0.00353	0.00471
24B-Gal $4$	31528	1	104	77	48	230	0.00000	0.00000
24B-Gal $4$	31529	0	154	133	55	342	0.00000	0.00000
24B-Gal $4$	34814	7	175	164	85	431	0.00000	0.00000
24B-Gal $4$	35144	166	194	139	85	584	0.01060	0.01060
Hand-Gal4	31039	195	180	112	44	531	0.00001	0.00001
Hand-Gal4	31314	80	56	41	19	196	0.00142	0.00213
Hand-Gal4	31528	105	174	102	65	446	0.00000	0.00001
Hand-Gal4	31529	116	147	89	72	424	0.00076	0.00131
Hand-Gal4	34814	164	168	120	107	559	0.00902	0.00984
Hand-Gal4	35144	125	84	87	39	335	0.00502	0.00603

Table 2: Driver Level Summarised Data

Driver	UAS	KD	shRNA	GAL4	WT	Total	pval	padj
C179-Gal4	31528	122	0	176	0	298	0.002	0.002
C179-Gal4	31529	109	0	225	0	334	0.000	0.000
C179-Gal4	34814	104	0	351	0	455	0.000	0.000

Table 3: Driver Level Summarized Data for C179-Gal4 Crosses

#### Crosses with no Knockdown Progeny

So far some crosses have produced no knockdown progeny. These are described in Table 10

#### **General Traits**

To test how many generations we can get out of 10 female flies in a vial, we examined the number of births in a vial, vs the generation of that vial. These data are in Figure 6. There is no detectable decrease in fecundity over time (p=0.523).

On the other hand, there was a significant difference between the number of Male flies born vs the number of Female flies born (p=0.00077). This is visulized in Figure 7

The key packages used in this analysis were R [1] and plyr [2]

#### References

- [1] R Core Team. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria, 2013.
- [2] Hadley Wickham. The split-apply-combine strategy for data analysis. *Journal of Statistical Software*, 40(1):1–29, 2011.

#### **Session Information**

- R version 3.0.2 (2013-09-25), x86\_64-apple-darwin10.8.0
- Locale: en\_US.UTF-8/en\_US.UTF-8/en\_US.UTF-8/en\_US.UTF-8/en\_US.UTF-8
- Base packages: base, datasets, graphics, grDevices, methods, stats, utils

## 24B-Gal4 Driver

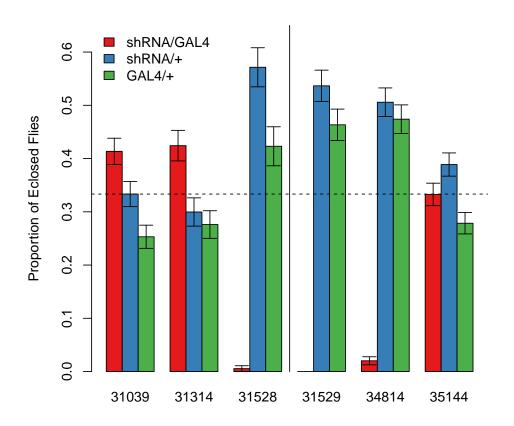


Figure 1: Distribution of Strains for 24B-Gal4

### 24B-Gal4 Driver at 18C

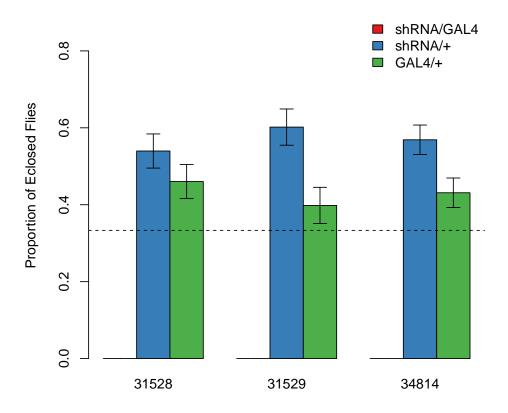


Figure 2: Distribution of Strains for 24B-Gal4 at 18C

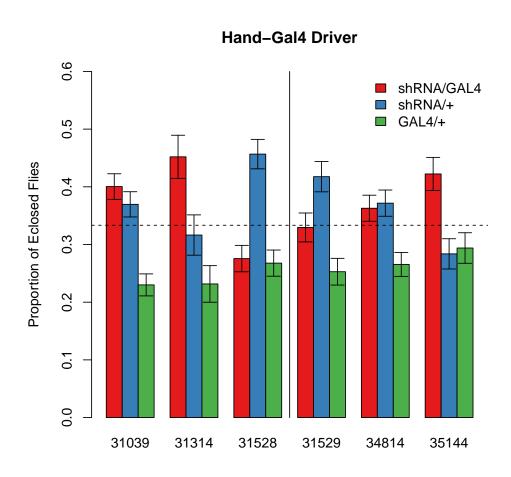


Figure 3: Distribution of Strains for Hand-Gal4

# C157 Driver

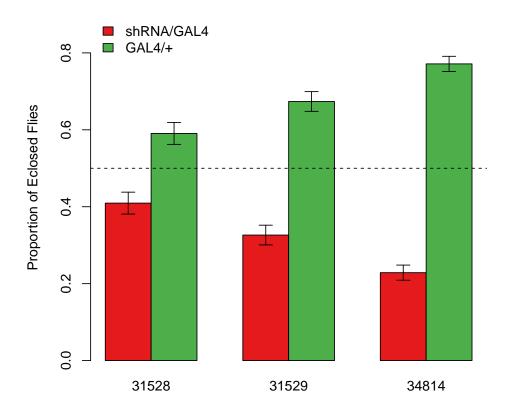


Figure 4: C179-Gal4 Driven Raptor Knockdown Strains

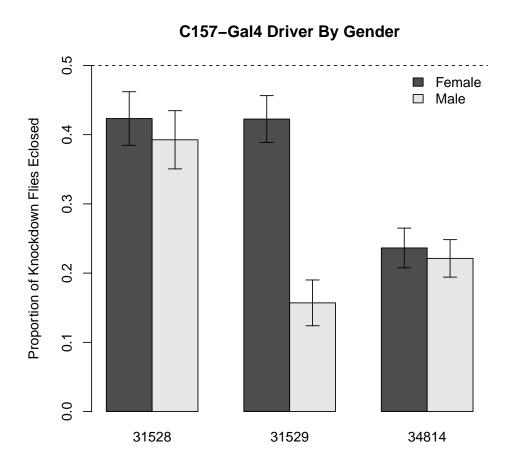


Figure 5: Gender Specific Differences Between C517-Gal4 Driven Raptor Knockdown Strains

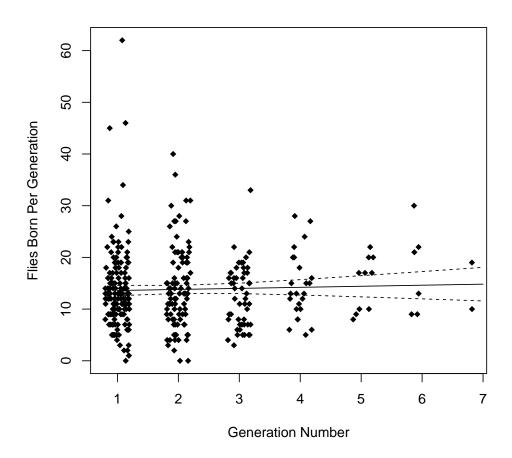


Figure 6: Number of flies eclosed over the course of several generations. Dashed lines indicate 95% confidence intervals, Male and female flies are separated in this analysis.

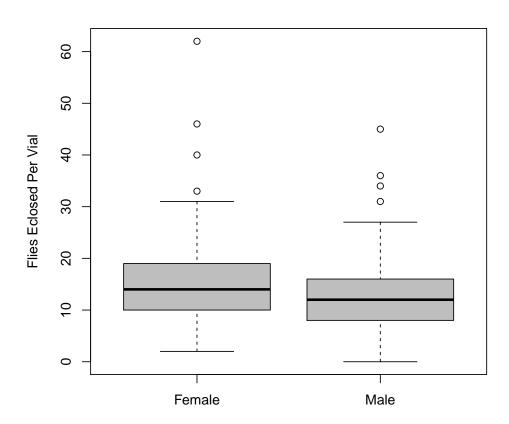


Figure 7: Gender distribution of eclosed flies.

UAS	Driver	Gender	KD	$\operatorname{shRNA}$	GAL4	WT	Total	pval	padj
31528	C179-Gal4	Female	69	0	94	0	163	0.0502	0.0502
31528	C179-Gal4	Male	53	0	82	0	135	0.0126	0.0188
31529	C179-Gal4	Female	90	0	123	0	213	0.0238	0.0285
31529	C179-Gal4	Male	19	0	102	0	121	0.0000	0.0000
34814	C179-Gal4	Female	52	0	168	0	220	0.0000	0.0000
34814	C179-Gal4	Male	52	0	183	0	235	0.0000	0.0000

Table 4: Driver Level, Gender Separated Summarized Data for C179-Gal4 Crosses

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pval	pagi
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Table 5: Fisher Tests Comparing Gender Effects on C157/Raptor shRNA Crosses

- Other packages: bibtex 0.3-6, plyr 1.8, RColorBrewer 1.0-5, reshape2 1.2.2, xtable 1.7-1
- Loaded via a namespace (and not attached): stringr 0.6.2, tools 3.0.2

Driver	UAS	KD	shRNA	GAL4	WT	Total	pval	padj
24B-Gal4	31528	0	68	58	51	177	0.0000000	0.0000000
24B-Gal4	31529	0	65	43	31	139	0.0000000	0.0000000
24B-Gal $4$	34814	0	95	72	61	228	0.0000000	0.0000000

Table 6: Driver Level Summarized Data for 18C Crosses

Cross	KD	shRNA	GAL4	WT	Total	pval	padj
$31039/\text{Tm}6\text{B} \times 24\text{B-Gal}4/\text{Tm}3\text{-sb}$	83	55	36	4	178	0.00007	0.00013
$31039/Tm6B \times 24B-Gal4/Tm3-ser$	82	78	65	43	268	0.34877	0.36929
$31039/Tm6B \times Hand-Gal4/Tm3-ser$	195	180	112	44	531	0.00001	0.00001
$31314/\mathrm{Tm}6\mathrm{B} \ge 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	80	57	45	18	200	0.00544	0.00753
$31314/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{ser}$	46	32	37	18	133	0.26900	0.30263
$31314/\mathrm{Tm}6\mathrm{B}$ x Hand-Gal4/Tm3-ser	80	56	41	19	196	0.00142	0.00232
$31528/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	0	58	41	17	116	0.00000	0.00000
$31528/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{ser}$	1	46	36	31	114	0.00000	0.00000
$31528/\mathrm{Tm}6\mathrm{B}$ x Hand-Gal4/Tm3-ser	105	174	102	65	446	0.00000	0.00001
$31529/\mathrm{Tm}6\mathrm{B} \ge 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	0	82	57	14	153	0.00000	0.00000
$31529/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{ser}$	0	72	76	41	189	0.00000	0.00000
$31529/Tm6B \times Hand-Gal4/Tm3-ser$	116	147	89	72	424	0.00076	0.00137
$34814/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	0	96	77	37	210	0.00000	0.00000
$34814/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{ser}$	7	79	87	48	221	0.00000	0.00000
$34814/\mathrm{Tm}6\mathrm{B}$ x Hand-Gal4/Tm3-ser	164	168	120	107	559	0.00902	0.01083
$35144/\mathrm{Tm}6\mathrm{B} \ge 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	50	55	48	19	172	0.77499	0.77499
$35144/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{ser}$	116	139	91	66	412	0.00676	0.00869
$35144/\mathrm{Tm}6\mathrm{B}$ x Hand-Gal4/Tm3-ser	125	84	87	39	335	0.00502	0.00753

Table 7: Cross Level Summarised Data

UAS	Driver	Gender	KD	shRNA	GAL4	WT	Total	pval	padj
31039	24B-Gal4	Female	95	67	57	31	250	0.00492	0.01180
31039	24B-Gal $4$	Male	70	66	44	16	196	0.03813	0.05384
31314	24B-Gal $4$	Female	56	49	43	16	164	0.42396	0.42396
31314	24B-Gal $4$	Male	70	40	39	20	169	0.00193	0.00516
31528	24B-Gal $4$	Female	1	55	36	34	126	0.00000	0.00000
31528	24B-Gal $4$	Male	0	49	41	14	104	0.00000	0.00000
31529	24B-Gal $4$	Female	0	85	83	29	197	0.00000	0.00000
31529	24B-Gal $4$	Male	0	69	50	26	145	0.00000	0.00000
34814	24B-Gal $4$	Female	5	88	87	45	225	0.00000	0.00000
34814	24B-Gal $4$	Male	2	87	77	40	206	0.00000	0.00000
35144	24B-Gal $4$	Female	94	113	80	51	338	0.05684	0.07052
35144	24B-Gal $4$	Male	72	81	59	34	246	0.17708	0.18478
31039	Hand-Gal4	Female	104	95	70	26	295	0.03140	0.05254
31039	Hand-Gal4	Male	91	85	42	18	236	0.00005	0.00016
31314	Hand-Gal4	Female	43	24	23	7	97	0.01450	0.02678
31314	Hand-Gal4	Male	37	32	18	12	99	0.03527	0.05290
31528	Hand-Gal4	Female	64	89	63	33	249	0.04910	0.06547
31528	Hand-Gal4	Male	41	85	39	32	197	0.00000	0.00002
31529	Hand-Gal4	Female	64	78	51	40	233	0.05877	0.07052
31529	Hand-Gal4	Male	52	69	38	32	191	0.01060	0.02119
34814	Hand-Gal4	Female	94	90	71	57	312	0.16923	0.18478
34814	Hand-Gal4	Male	70	78	49	50	247	0.03284	0.05254
35144	Hand-Gal4	Female	70	38	50	19	177	0.00700	0.01527
35144	Hand-Gal4	Male	55	46	37	20	158	0.17190	0.18478

Table 8: Gendered and Cross Level Summarised Data

	pval	padj
31039/Tm6B x 24B-Gal4/Tm3-sb	0.00000	0.00000
$31039/Tm6B \times 24B-Gal4/Tm3-ser$	0.00316	0.00335
$31039/Tm6B \times Hand-Gal4/Tm3-ser$	0.00000	0.00000
$31314/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	0.00000	0.00000
$31314/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{ser}$	0.00627	0.00627
$31314/\mathrm{Tm}6\mathrm{B}$ x Hand-Gal4/Tm3-ser	0.00000	0.00000
$31528/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	0.00000	0.00000
$31528/Tm6B \times 24B-Gal4/Tm3-ser$	0.00000	0.00000
$31528/\mathrm{Tm}6\mathrm{B}$ x Hand-Gal4/Tm3-ser	0.00000	0.00000
$31529/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	0.00000	0.00000
$31529/Tm6B \times 24B-Gal4/Tm3-ser$	0.00000	0.00000
$31529/Tm6B \times Hand-Gal4/Tm3-ser$	0.00000	0.00000
$34814/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{sb}$	0.00000	0.00000
$34814/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-}\mathrm{Gal}4/\mathrm{Tm}3\text{-}\mathrm{ser}$	0.00000	0.00000
$34814/\mathrm{Tm}6\mathrm{B}$ x Hand-Gal4/Tm3-ser	0.00014	0.00017
$35144/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-Gal}4/\mathrm{Tm}3\text{-sb}$	0.00035	0.00040
$35144/\mathrm{Tm}6\mathrm{B} \times 24\mathrm{B}\text{-Gal}4/\mathrm{Tm}3\text{-ser}$	0.00000	0.00000
$35144/\mathrm{Tm}6\mathrm{B}$ x Hand-Gal4/Tm3-ser	0.00000	0.00000

Table 9: Summarized Data Including Double Balancer Strains

	UAS	Driver	Gender	KD	$\operatorname{shRNA}$	GAL4
10	31528	24B-Gal $4$	Male	0	49	41
13	31529	24B-Gal $4$	Female	0	85	83
14	31529	24B-Gal $4$	Male	0	69	50

Table 10: Crosses with no Knockdown Flies