

Celrr KO vs WT (Adult)

This file shows the wt-v-ko comparison for Celrr.

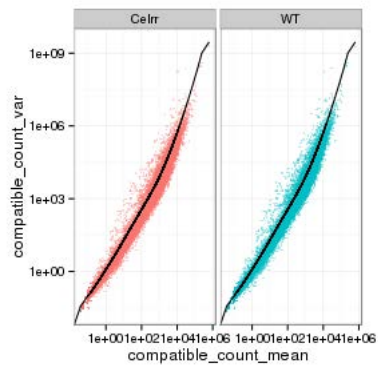
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

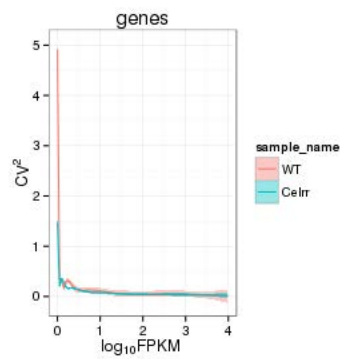
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



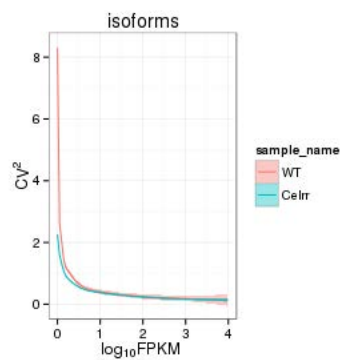
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

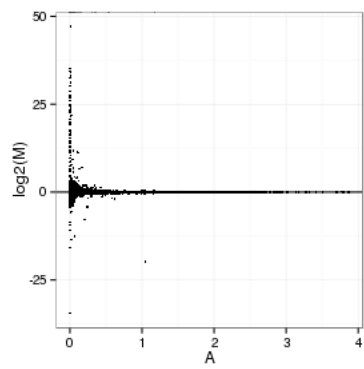
Genes:



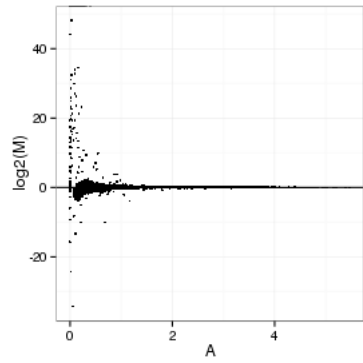
Isoforms:



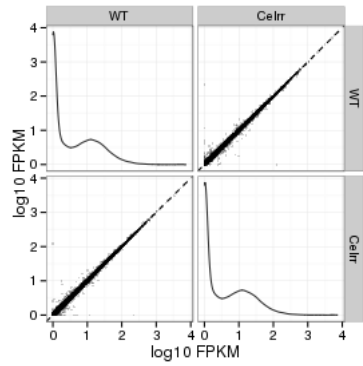
MvA plot



MvA plot counts



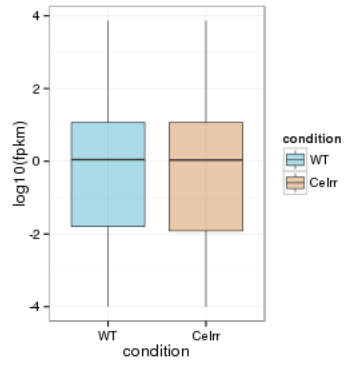
Scatterplot matrix



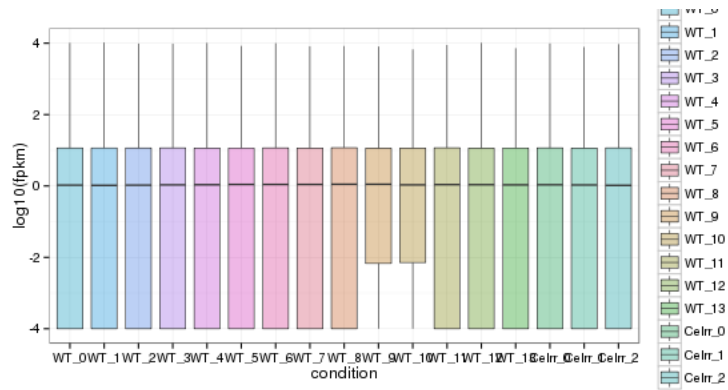
Distributions

Boxplots

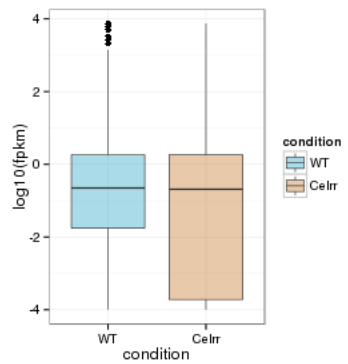
Boxplot (genes)



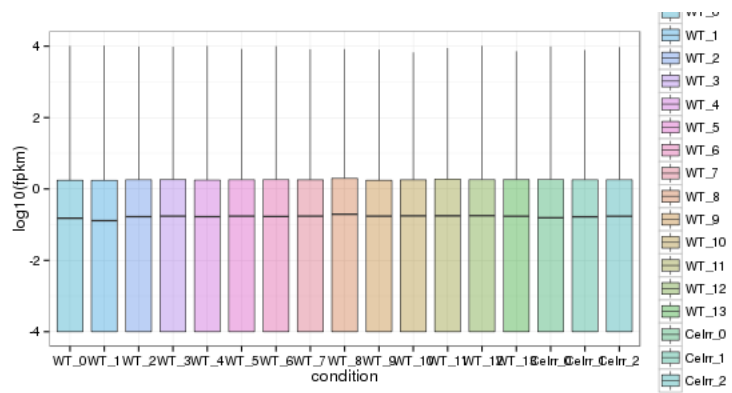
Boxplot (genes, replicates)



Boxplot (isoforms)

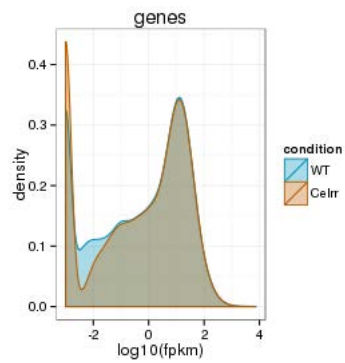


Boxplot (isoforms, replicates)

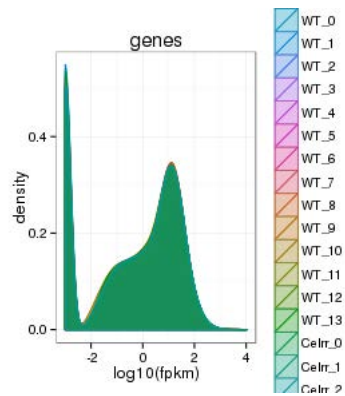


Density

Density (genes)

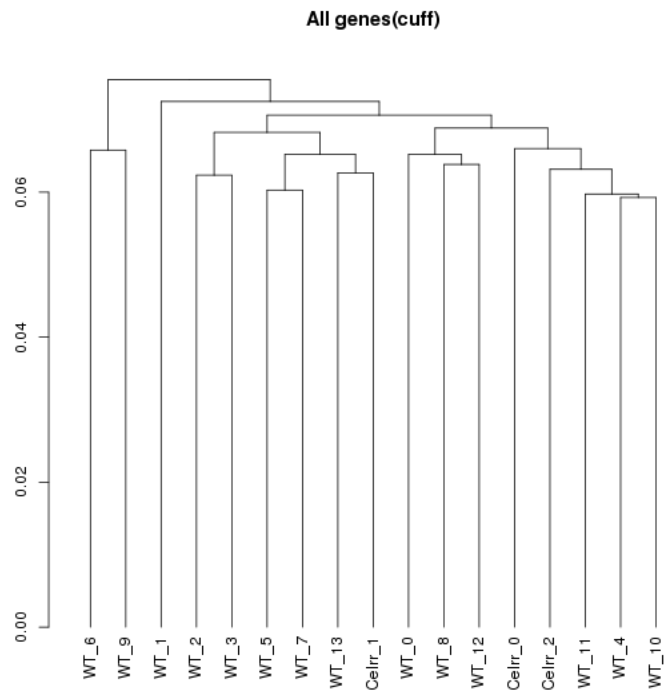


Density (genes, replicates)



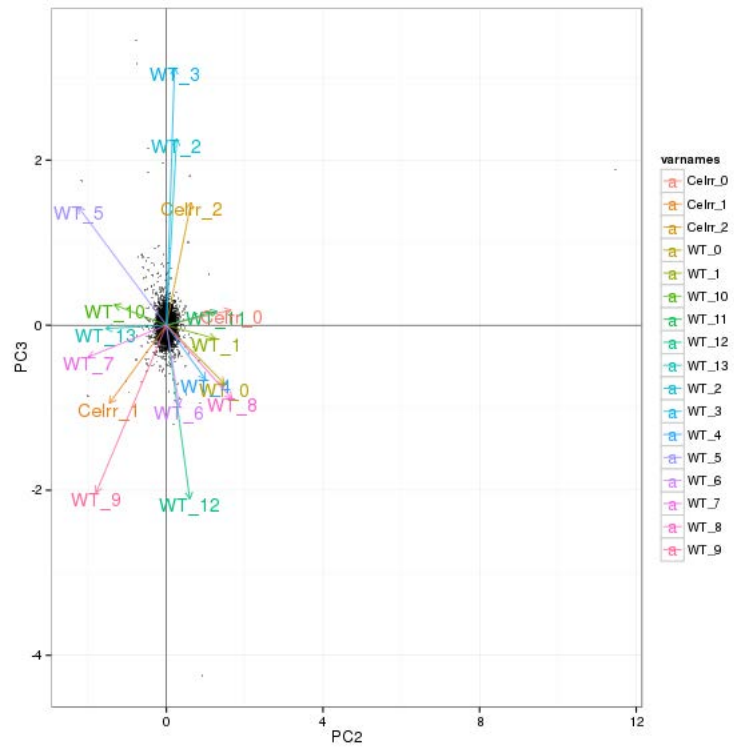
Clustering

Replicate Clusters

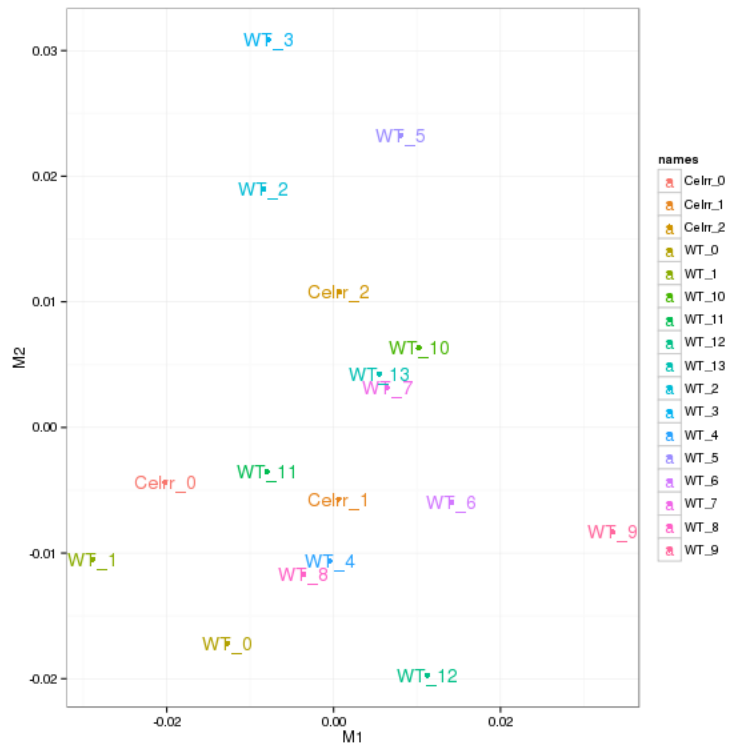


NULL

PCA (genes)

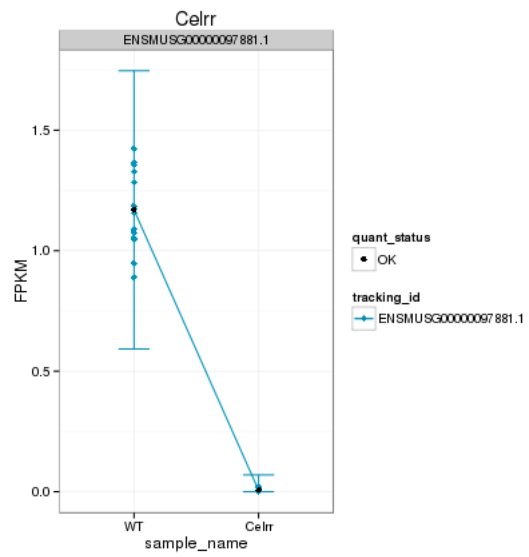


MDS (genes)

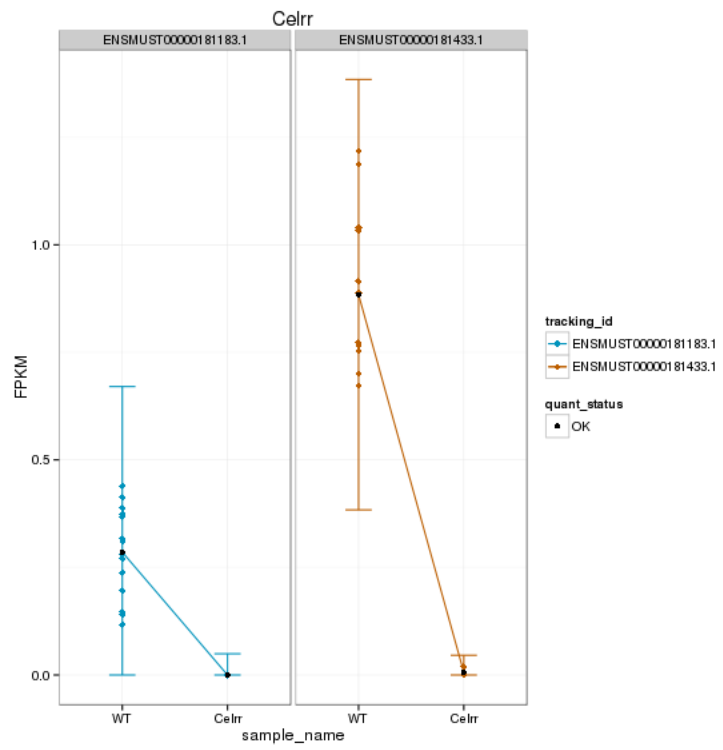


KO assessment

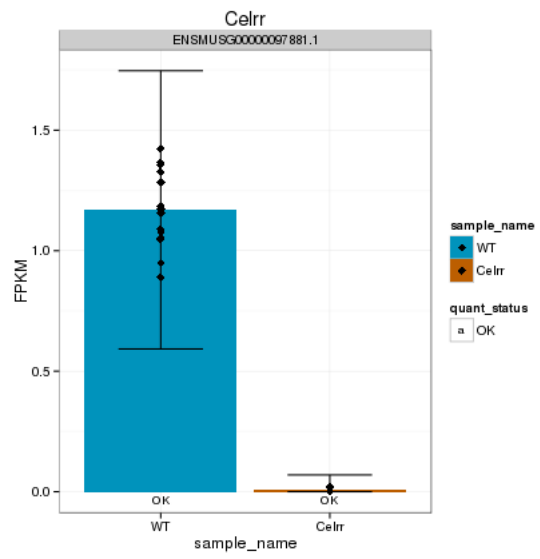
Endogenous lncRNA expression



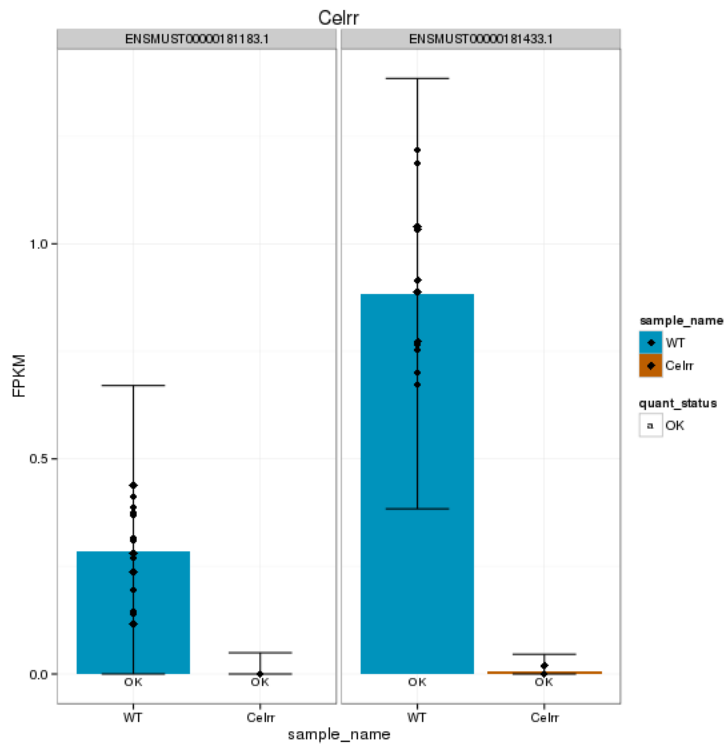
Endogenous expression of Celrr isoforms:



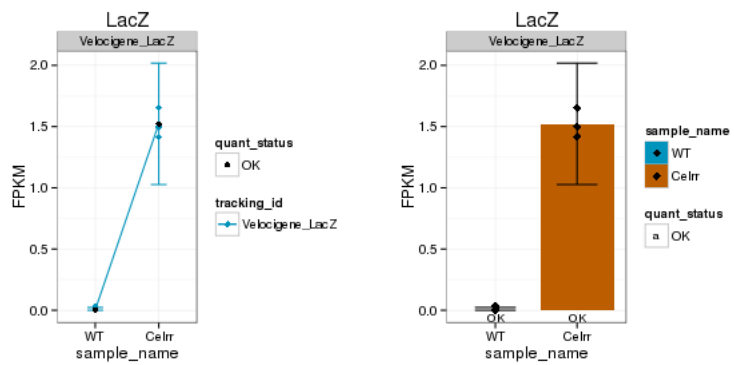
Barplot of gene expression:



Barplot of isoform expression:

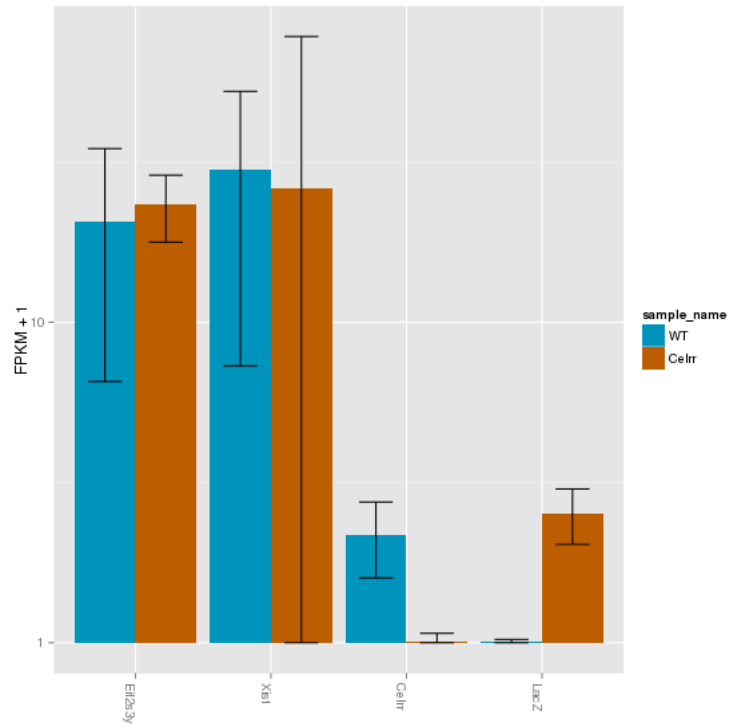


LacZ expression

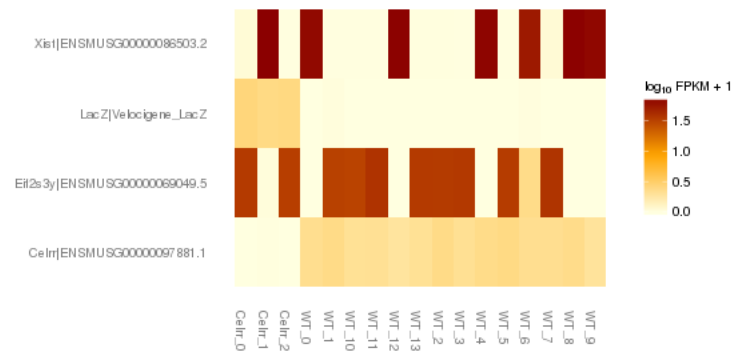


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

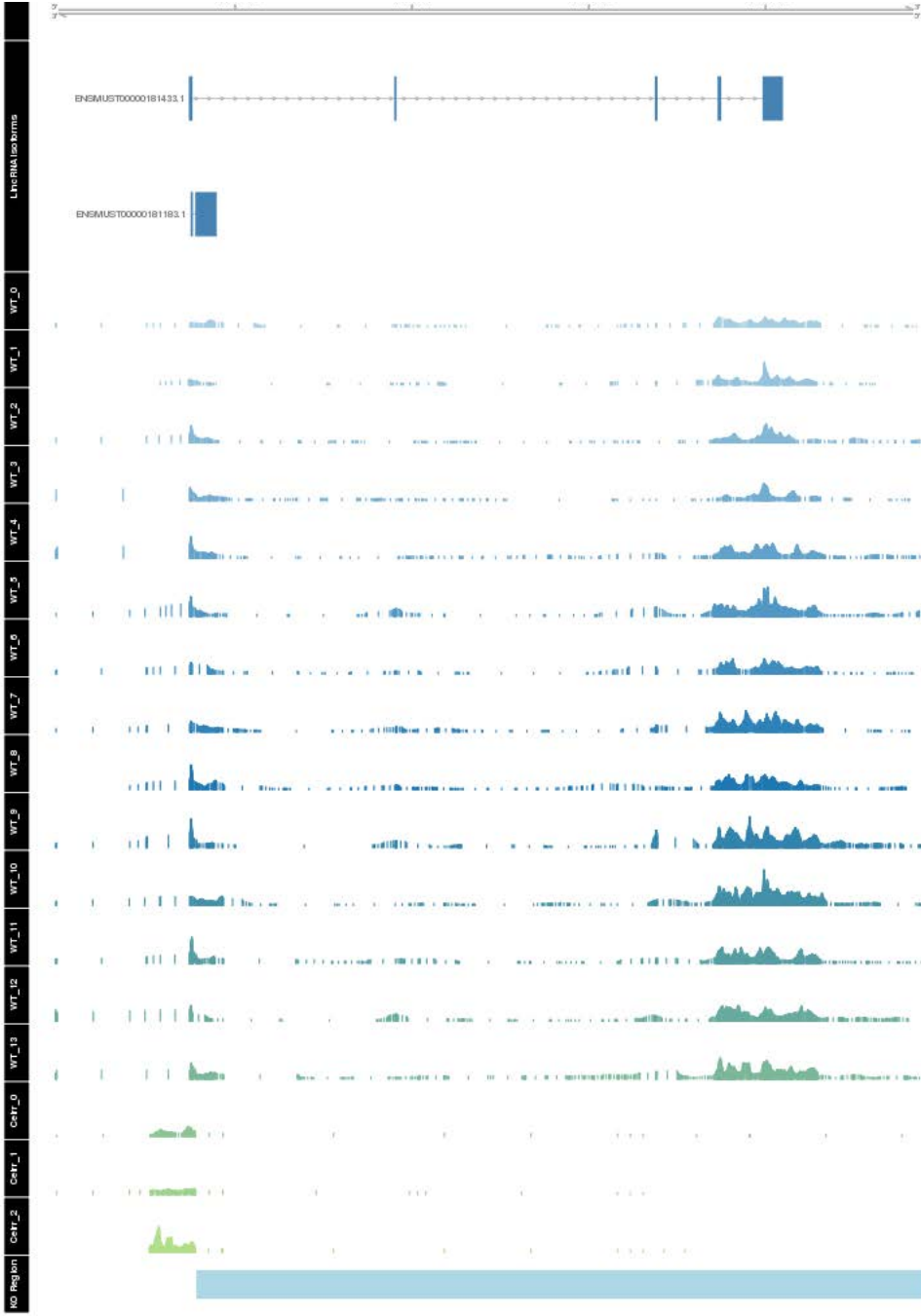


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 54 significantly differentially expressed genes. They are:

geneAnnot\$gene_short_name

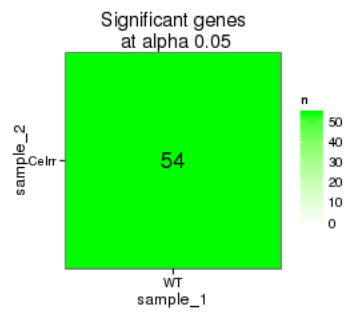
- 1 Chordc1
- 2 Nes
- 3 Dnajb1
- 4 Dio2
- 5 Slc38a3
- 6 P4ha1
- 7 Btg2
- 8 Ccdc117
- 9 Xbp1
- 10 Pdia6
- 11 Adi1
- 12 Serpina3n
- 13 Fos
- 14 Arc
- 15 Sdf2l1
- 16 Nr4a1
- 17 Creld2
- 18 Glo1
- 19 Dusp1
- 20 Pdia4
- 21 Lcn2
- 22 Hspa5
- 23 Vcam1
- 24 Hddc3
- 25 Tsc22d3
- 26 Mt2
- 27 Mt1
- 28 Pllp
- 29 Cryab
- 30 Ptgs2
- 31 Tlr13
- 32 Lars2
- 33 Egr1
- 34 Fmo2
- 35 Obfc1
- 36 Pcsk9
- 37 Gm9493

- 38 Acp1
- 39 Cirbp
- 40 Npas4
- 41 Tmem252
- 42 Opalin
- 43 Cd14

- 44 Tpt1
- 45 Zbtb40
- 46 Rpl34
- 47 Ppia
- 48 Gm7292
- 49 H2-BI
- 50 Wdfy1
- 51 Hspa1b
- 52 Hspa1a
- 53 Neat1
- 54 AA465934

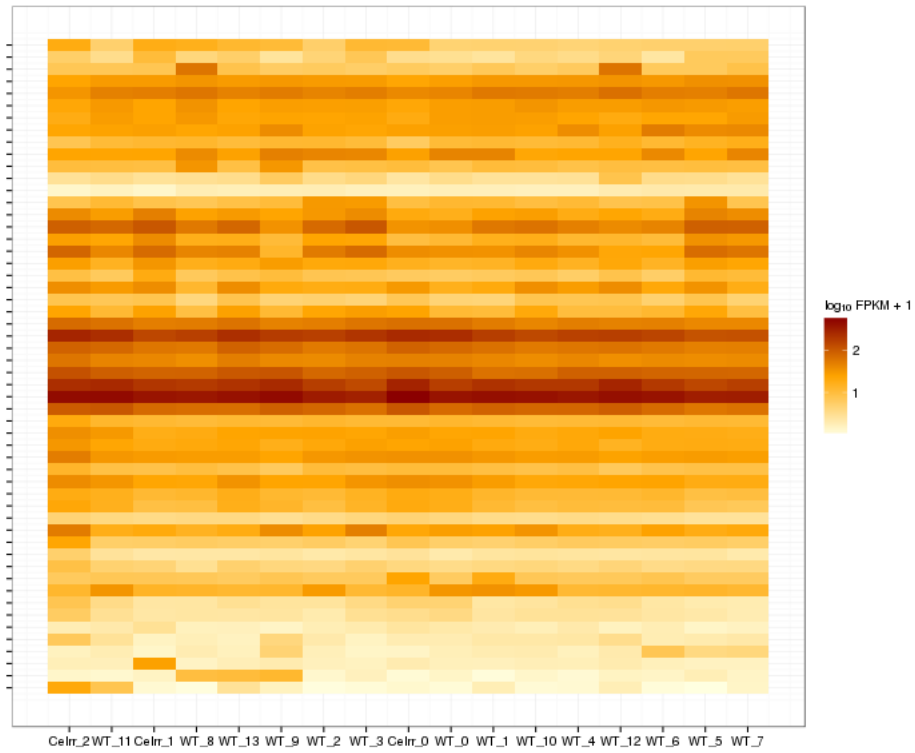
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

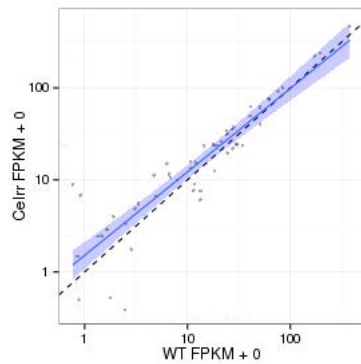


Significant genes with expression >50fpkm (any condition):(turned off)

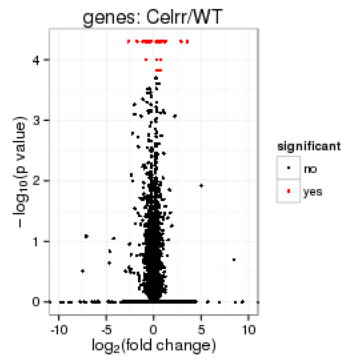
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

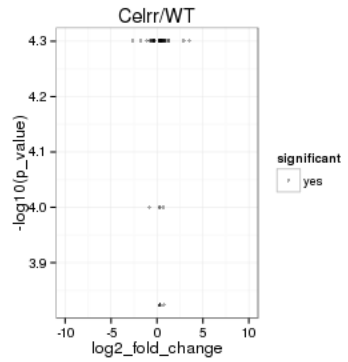
Scatter plot of significant genes only:



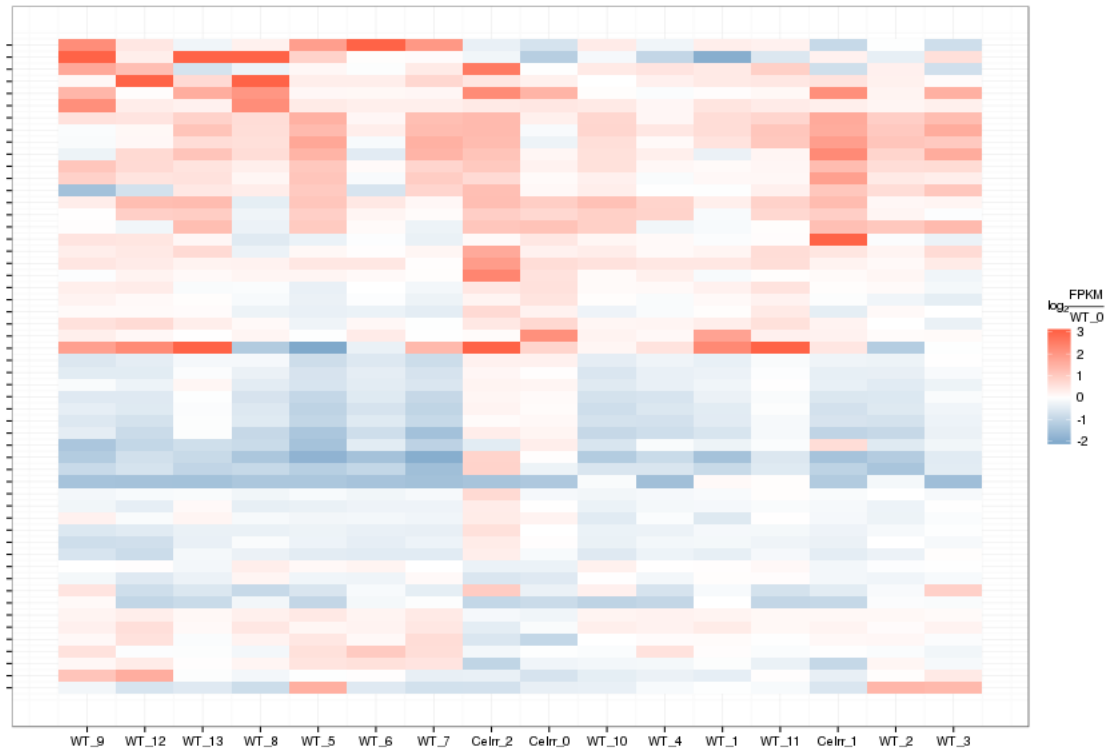
Volcano Plot



Volcano plot with significant genes only:



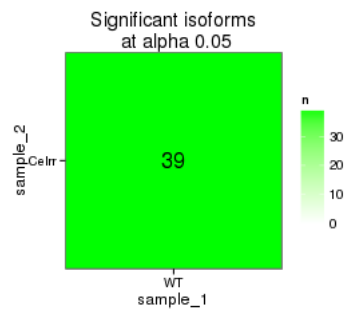
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

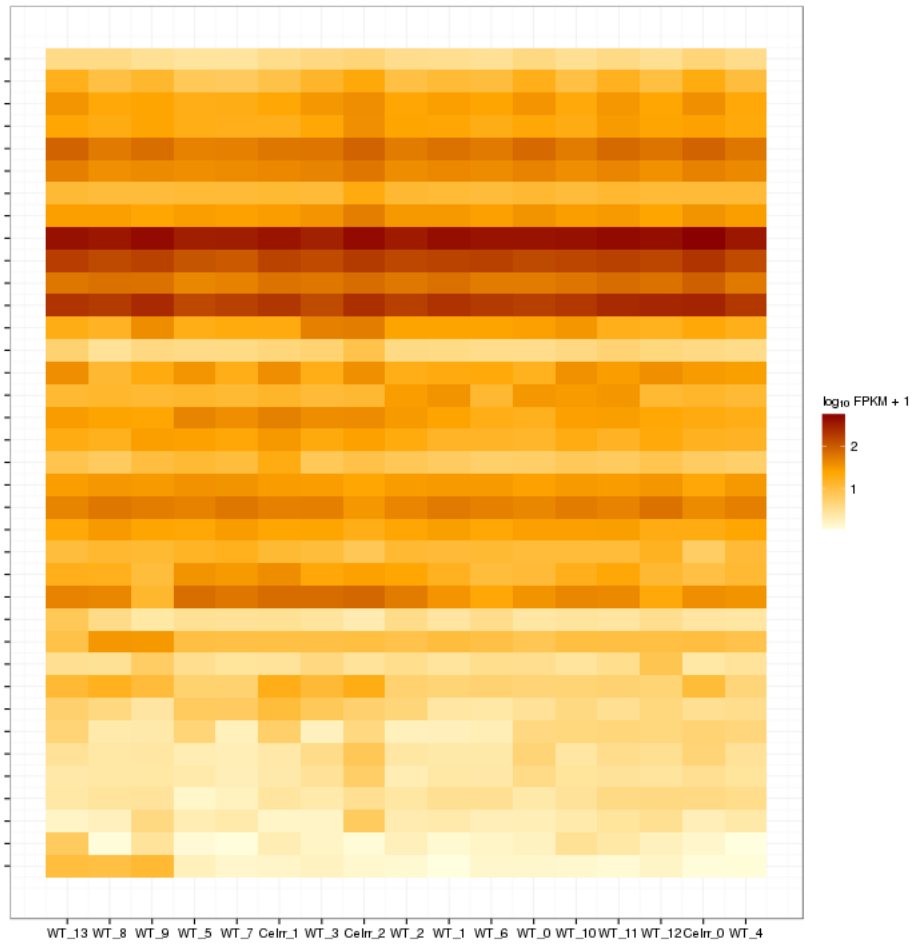
isoAnnot\$gene_short_name

- 1 Chordc1
- 2 Rec8
- 3 Hif3a
- 4 Nes
- 5 Dnajb1
- 6 Cyp2a5
- 7 Dio2
- 8 Btg2
- 9 Pdia6

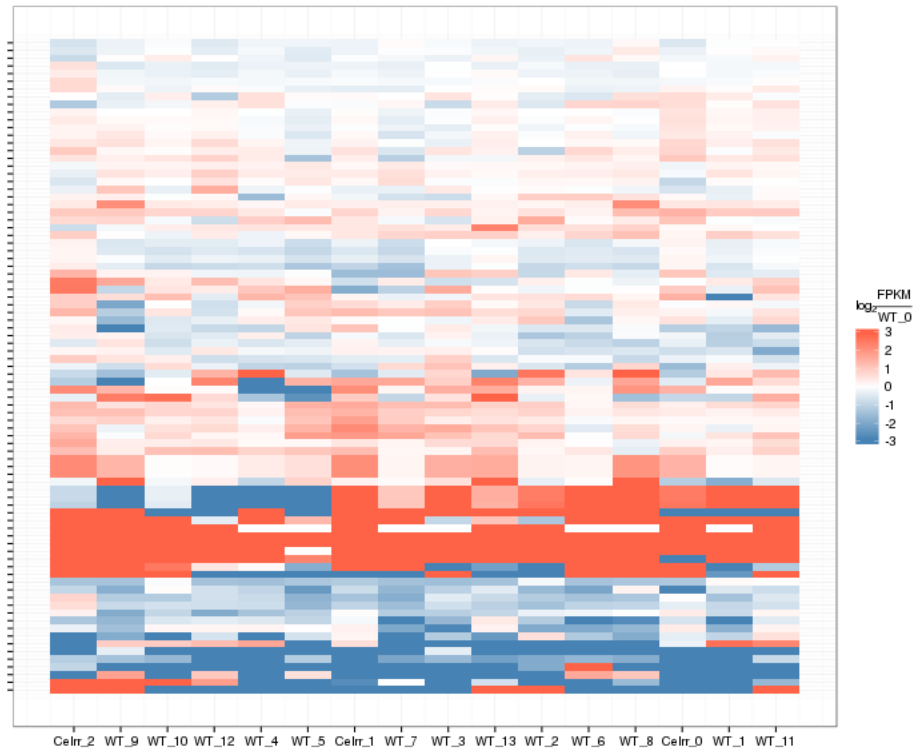
- 10 Serpina3n
- 11 Fos
- 12 Fam107a
- 13 Arc
- 14 Sdf2l1
- 15 Nr4a1
- 16 Dusp1
- 17 Pdia4
- 18 Vcam1
- 19 Hddc3
- 20 Mt2
- 21 Mt1
- 22 Pllp
- 23 Ptgs2
- 24 Lars2
- 25 Gjb6
- 26 Gm9493
- 27 Cirbp
- 28 Npas4
- 29 Tmem252
- 30 Opalin
- 31 Pla2g4e
- 32 Zbtb40
- 33 Gm7292
- 34 Wdfy1
- 35 Hspa1b
- 36 Hspa1a

- 37 Neat1

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

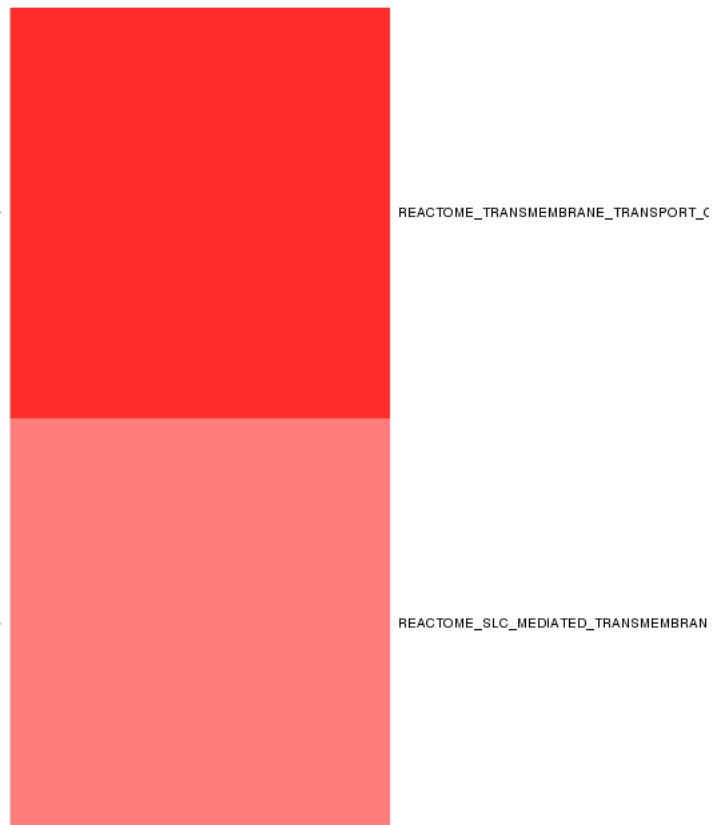
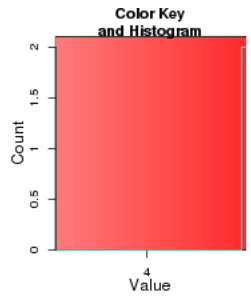
Biocarta enrichment:

```
## [1] "Not enough significant categories to print a heatmap!"
```

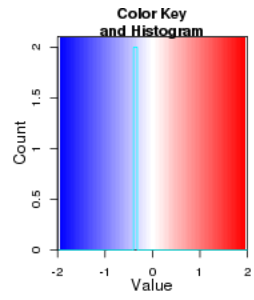
Biocarta zscore:

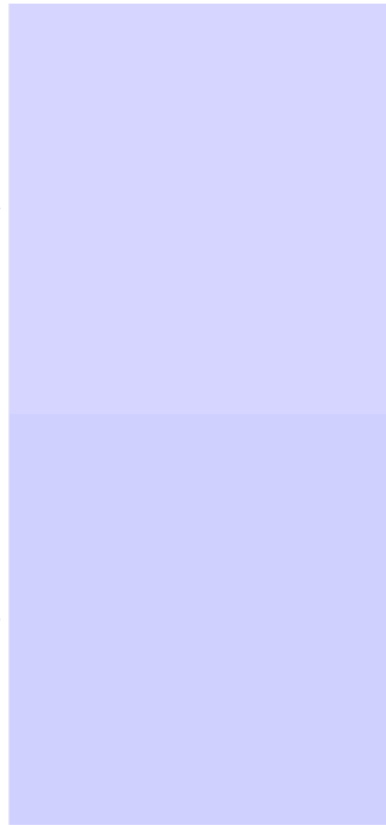
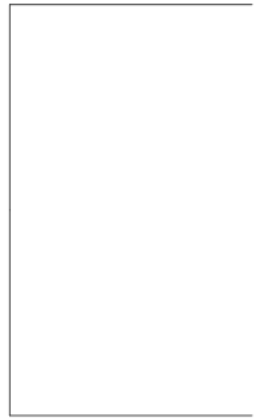
```
## [1] "Not enough significant categories to print a heatmap!"
```

Reactome enrichment:



Reactome zscore:





REACTOME_TRANSMEMBRANE_TRANSPORT_C

REACTOME_SLC_MEDIATED_TRANSMEMBRAN

Kegg enrichment:

Error: argument is of length zero

Kegg zscore:

```
## Error: incorrect number of dimensions
```

```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Interneuron enrichment:

```
## Error: argument is of length zero
```

Interneuron zscore:

```
## Error: incorrect number of dimensions
```

```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

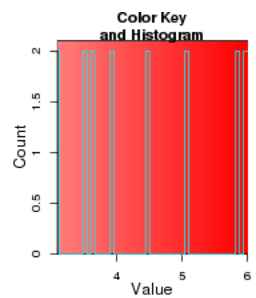
Oncogene enrichment:

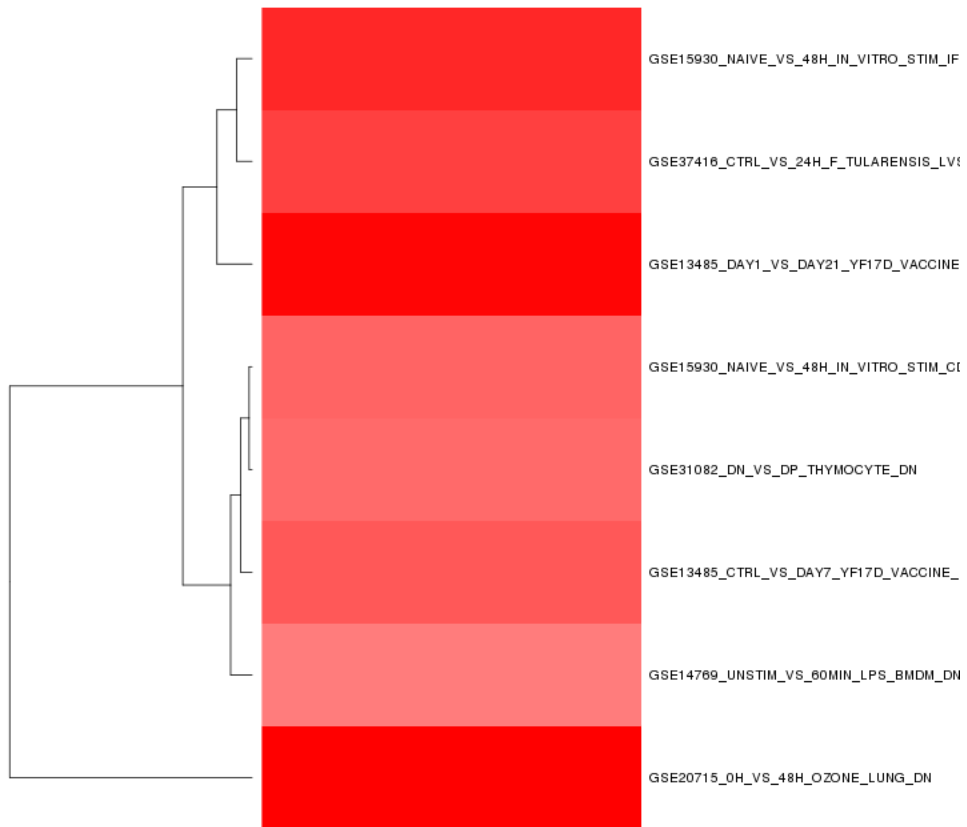
```
## [1] "Not enough significant categories to print a heatmap!"
```

Oncogene zscore:

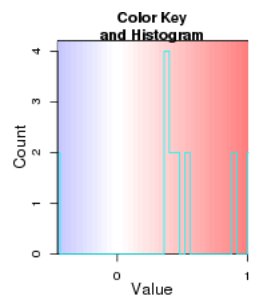
```
## [1] "Not enough significant categories to print a heatmap!"
```

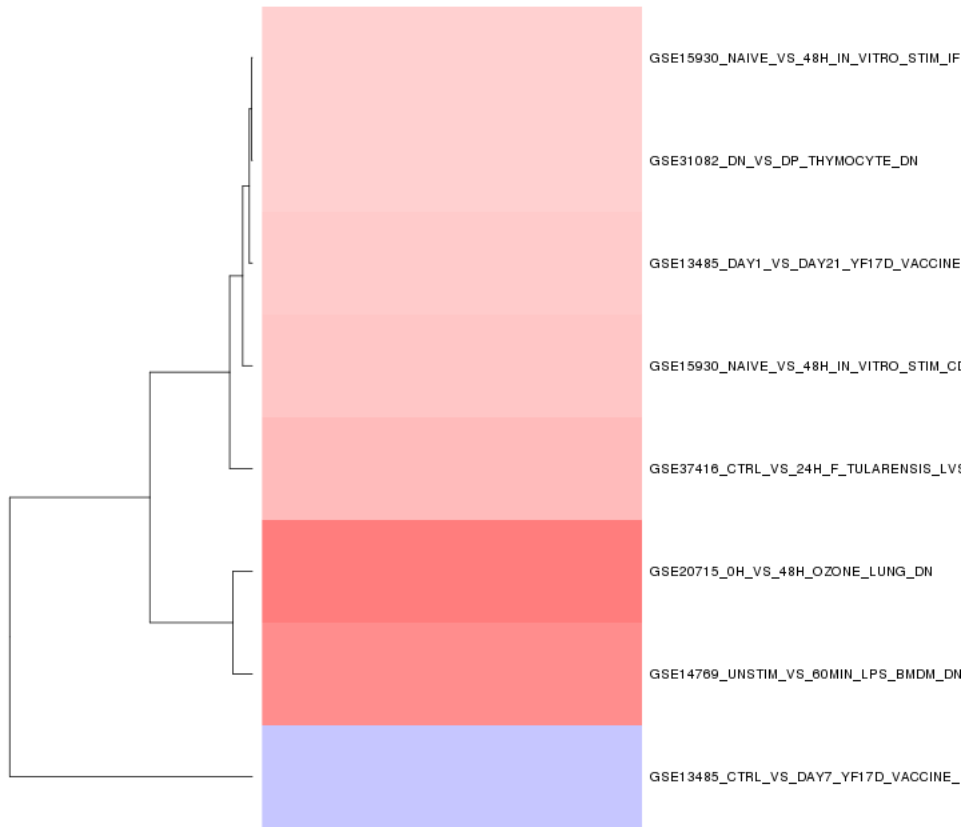
Immuno enrichment:



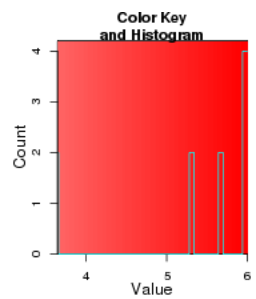


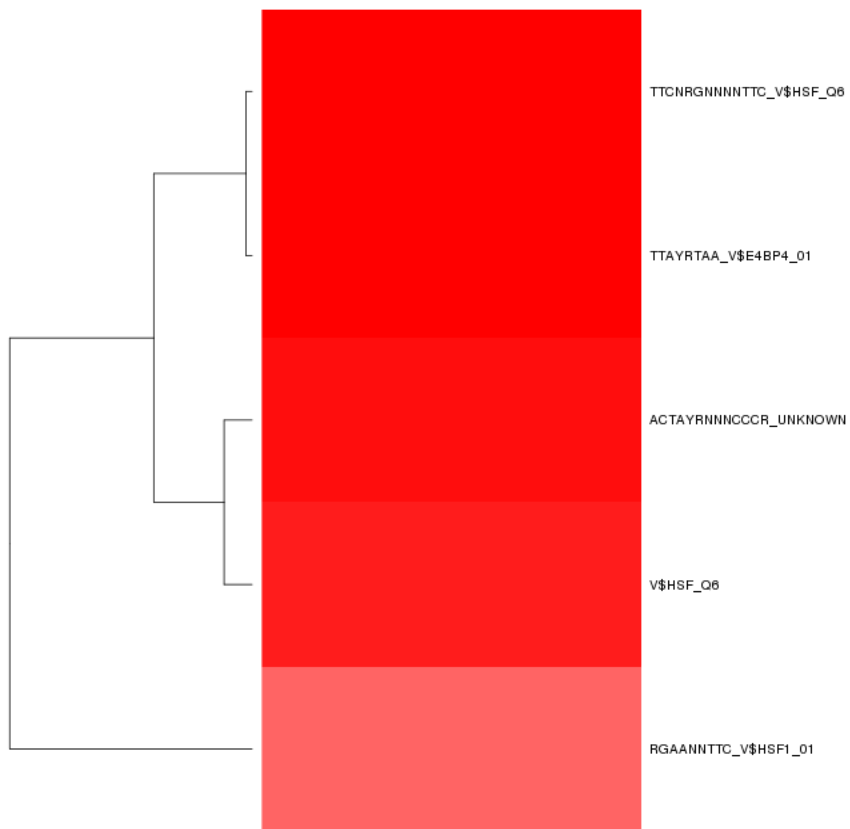
Immuno zscore:



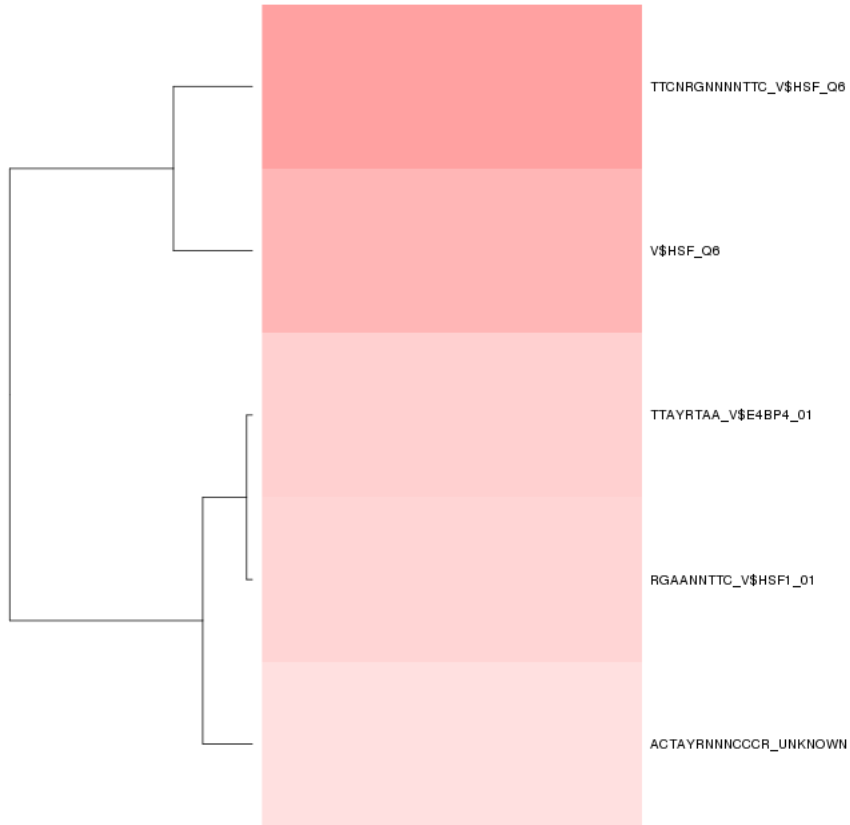
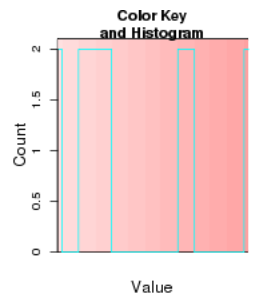


TF enrichment:





TF zscore:

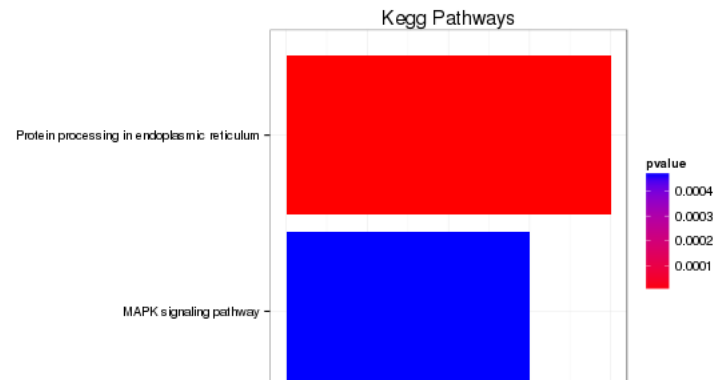
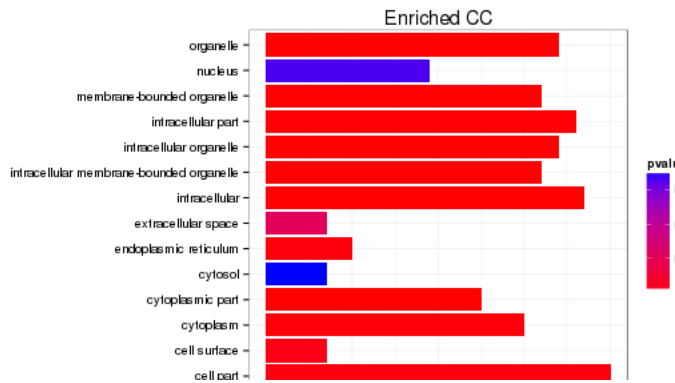
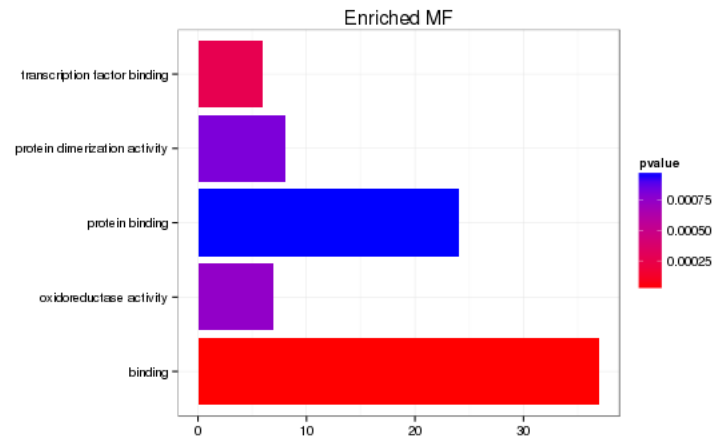
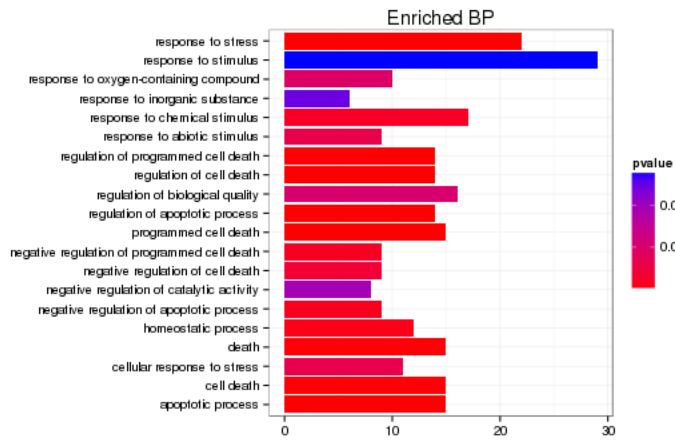


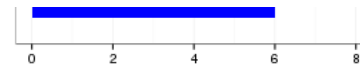
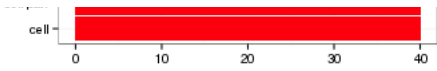
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```

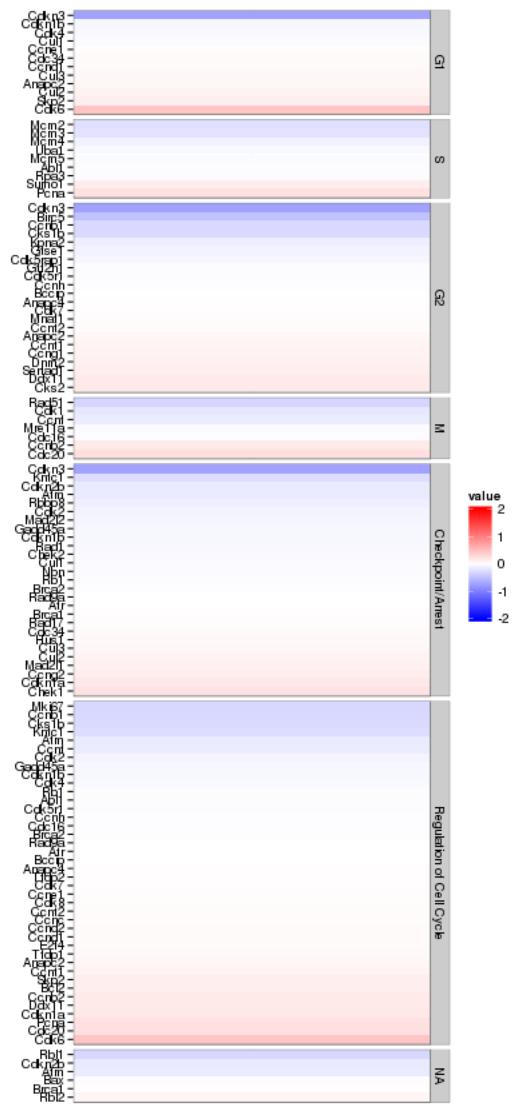
```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```



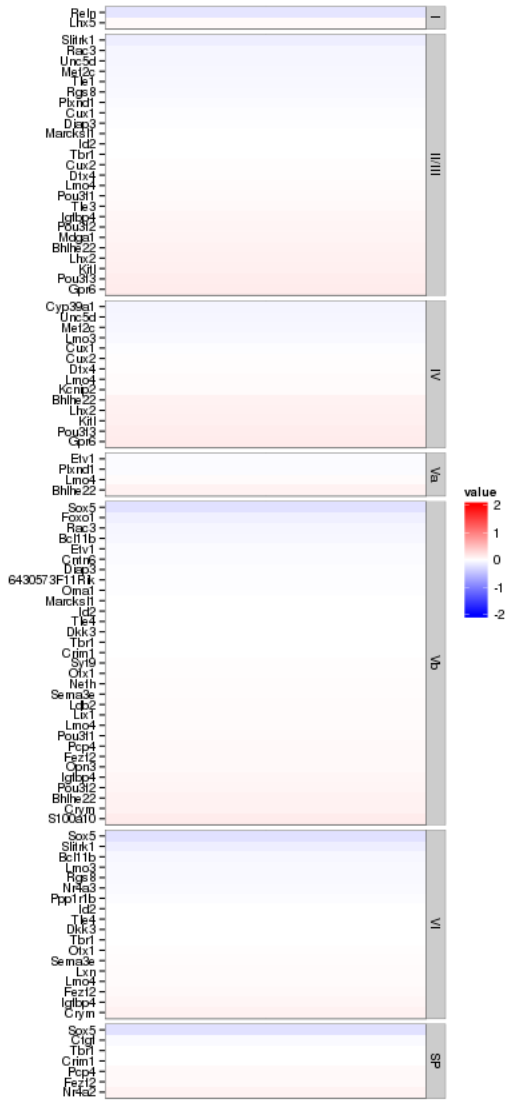


```
## Error: 'x' and 'units' must have length > 0
```

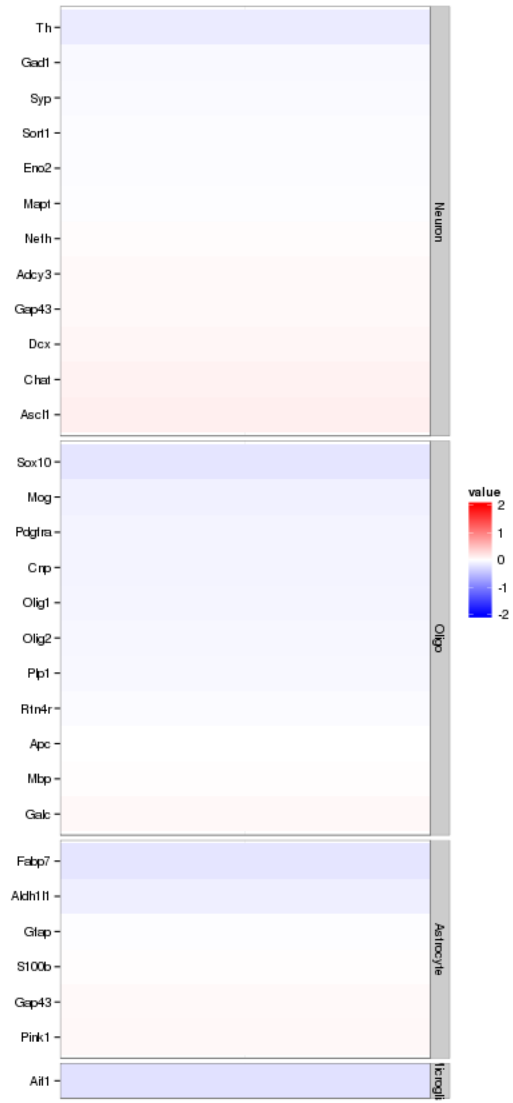
Enrichment or depletion for stage-specific cell cycle markers



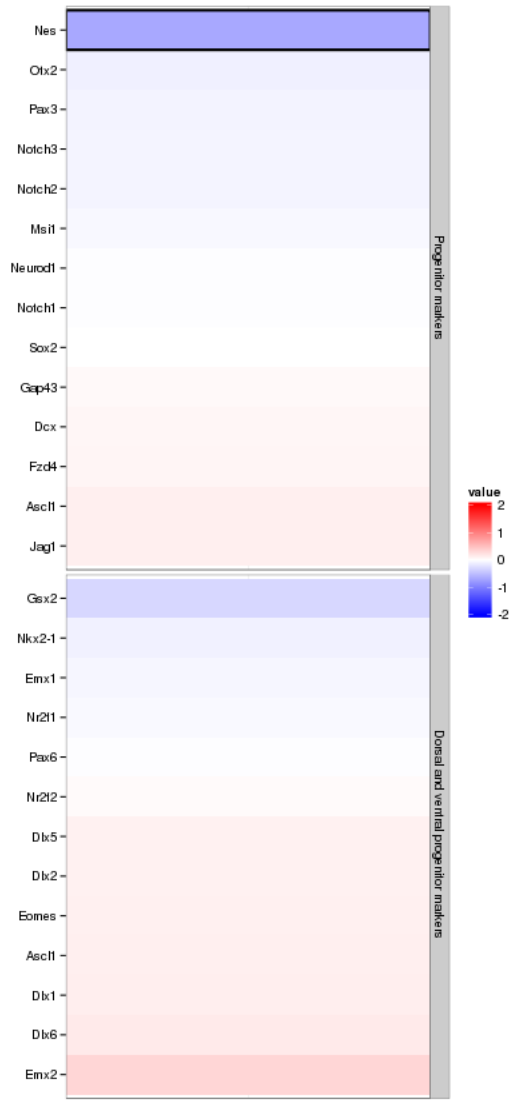
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



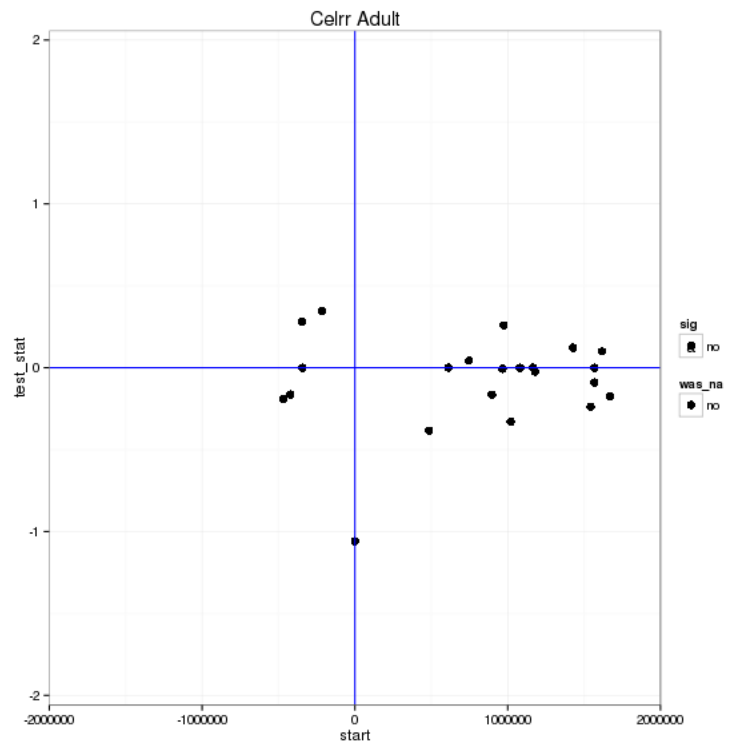
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR739
16 JR783
17 JR804

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	33868500.00	0.78	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	33868500.00	0.60	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	33868500.00	1.00	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	33868500.00	0.82	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	33868500.00	1.06	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	33868500.00	1.10	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	33868500.00	0.82	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	33868500.00	1.18	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	33868500.00	1.03	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	33868500.00	1.33	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	33868500.00	1.54	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	33868500.00	1.04	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	33868500.00	1.00	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	33868500.00	1.23	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR739/abundances.cxb	Celrr	0	Celrr_0	23325900.00	33868500.00	0.69	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR783/abundances.cxb	Celrr	1	Celrr_1	37161800.00	33868500.00	1.10	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR804/abundances.cxb	Celrr	2	Celrr_2	39715300.00	33868500.00	1.16	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1      KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1      reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1        zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Celrr -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Celrr_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/ar
## 2
## 3
## 4
## 5
```

Celrr KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Celrr.

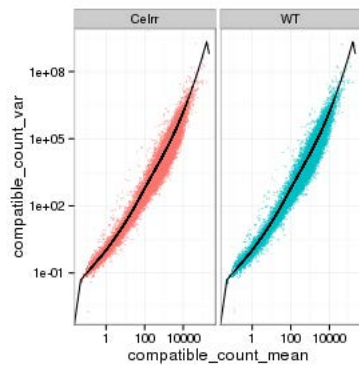
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

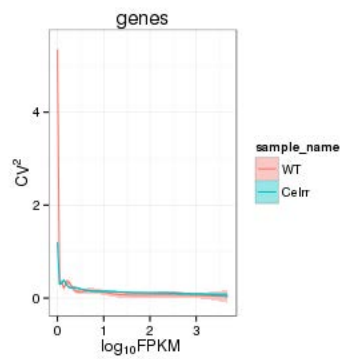
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



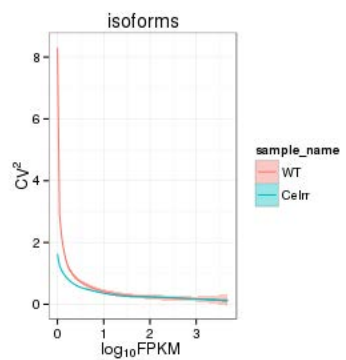
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

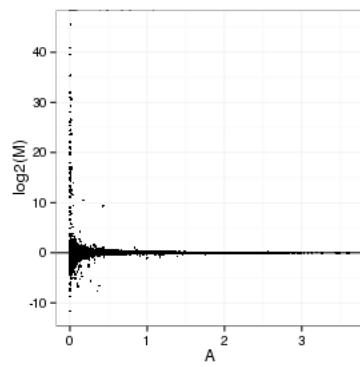
Genes:



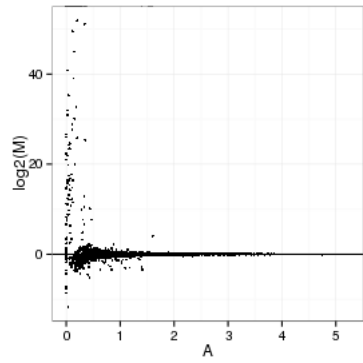
Isoforms:



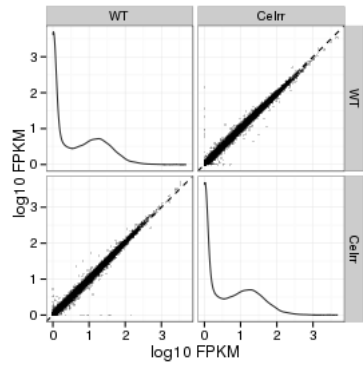
MvA plot



MvA plot counts



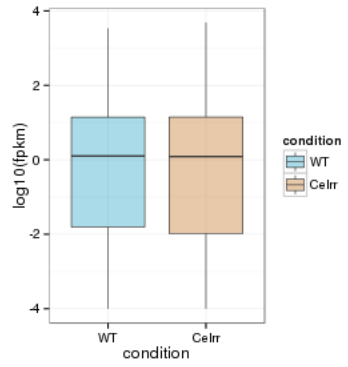
Scatterplot matrix



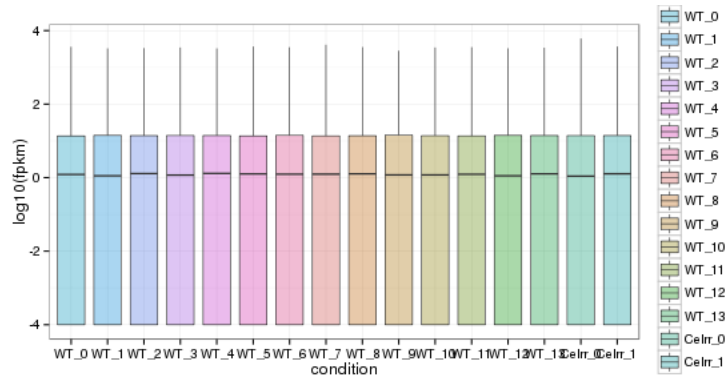
Distributions

Boxplots

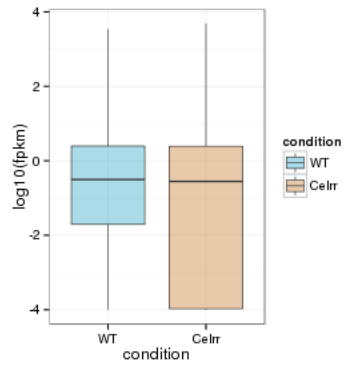
Boxplot (genes)



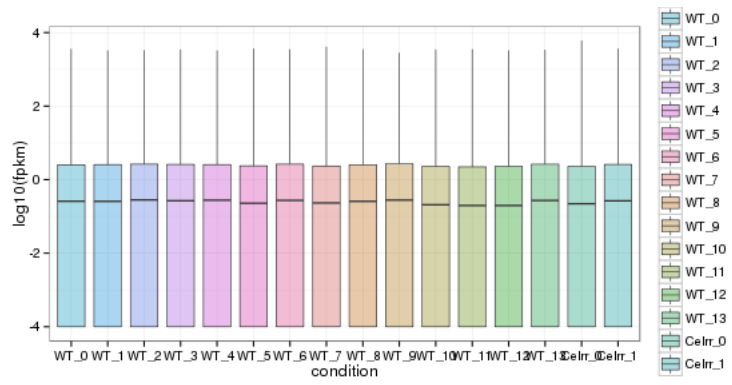
Boxplot (genes, replicates)



Boxplot (isoforms)

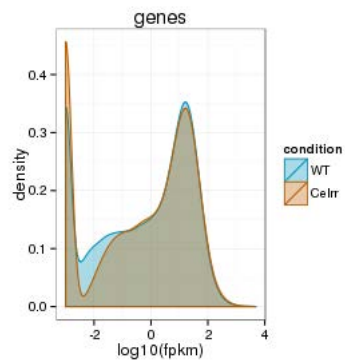


Boxplot (isoforms, replicates)

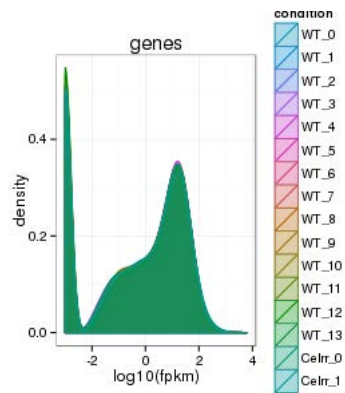


Density

Density (genes)

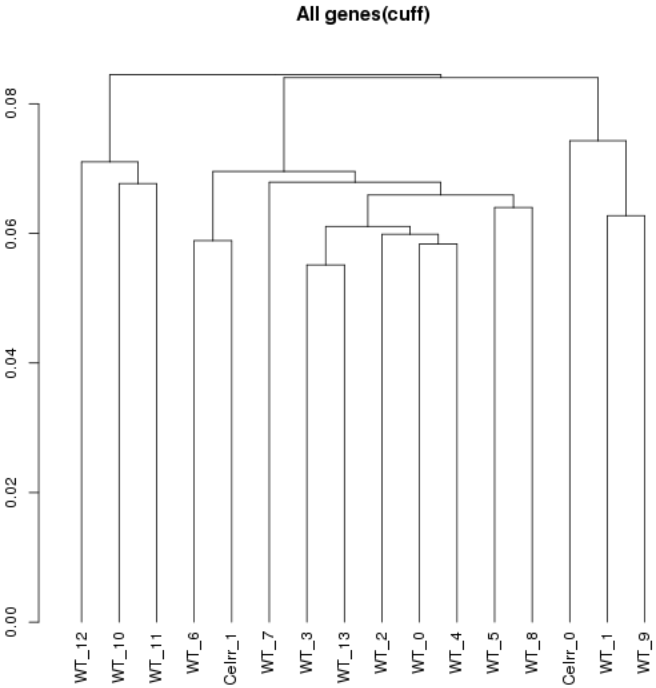


Density (genes, replicates)



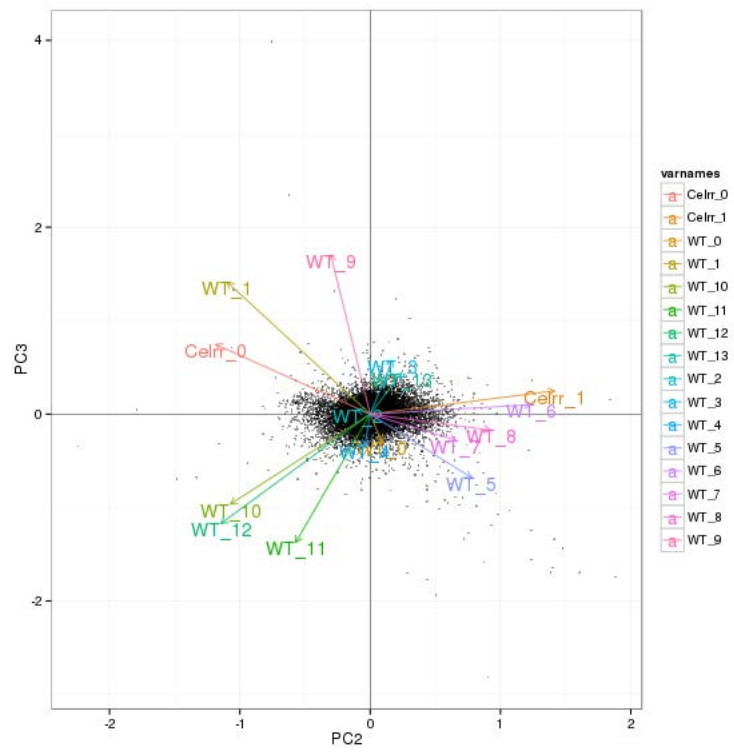
Clustering

Replicate Clusters

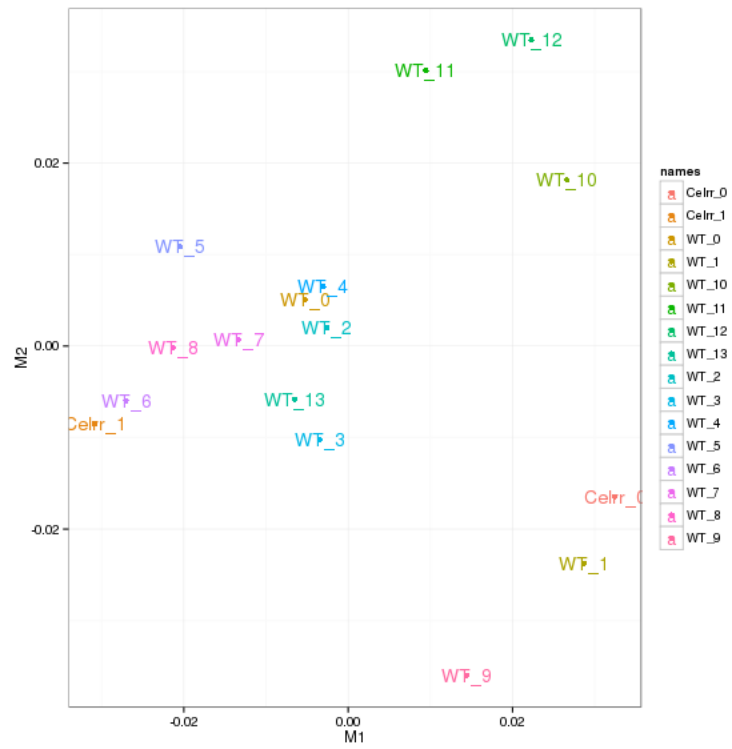


NULL

PCA (genes)

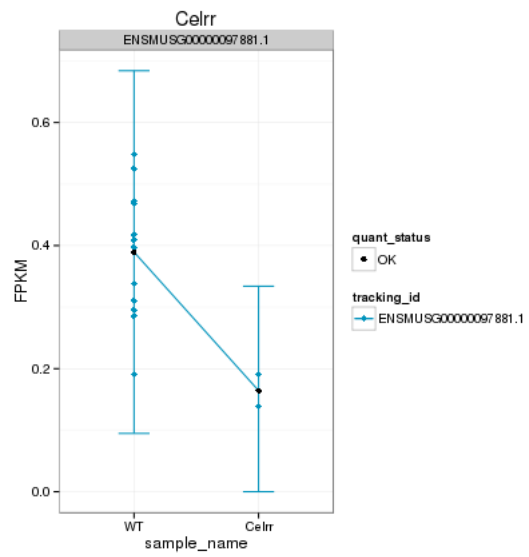


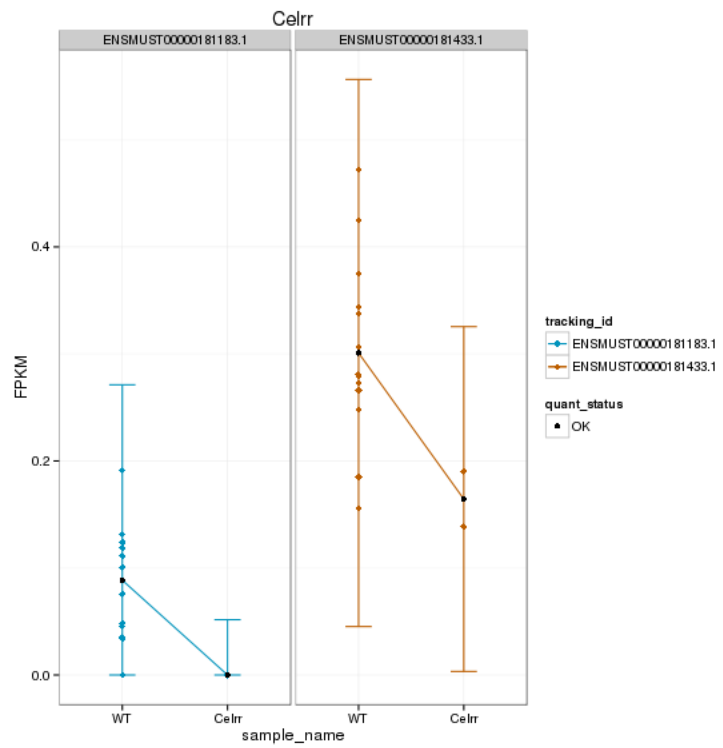
MDS (genes)



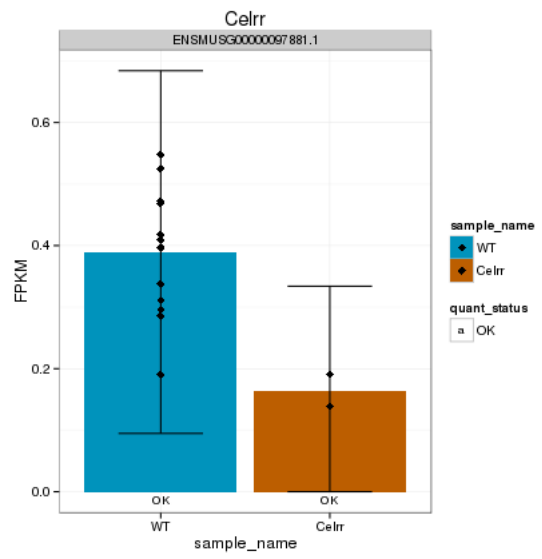
KO assessment

Endogenous lncRNA expression

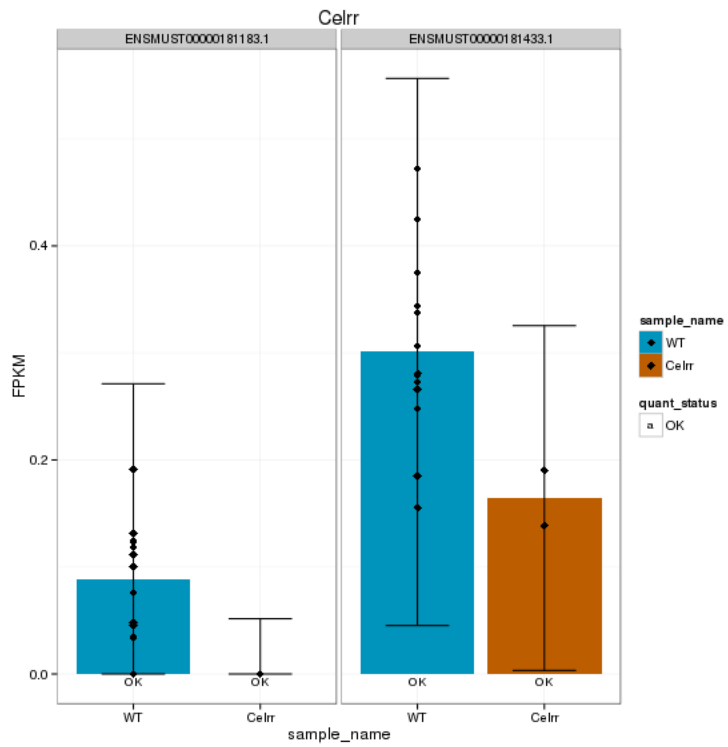




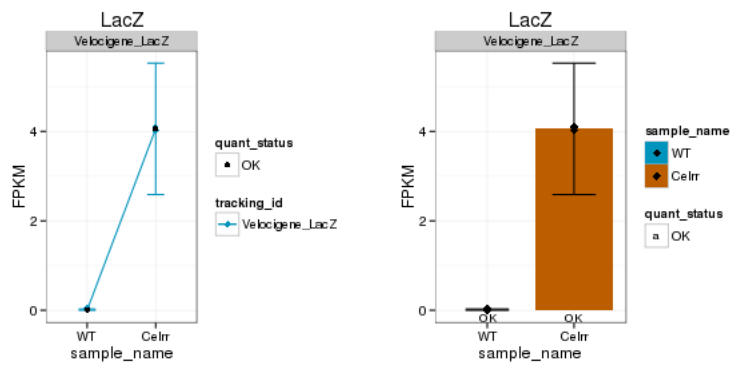
Barplot of gene expression:



Barplot of isoform expression:

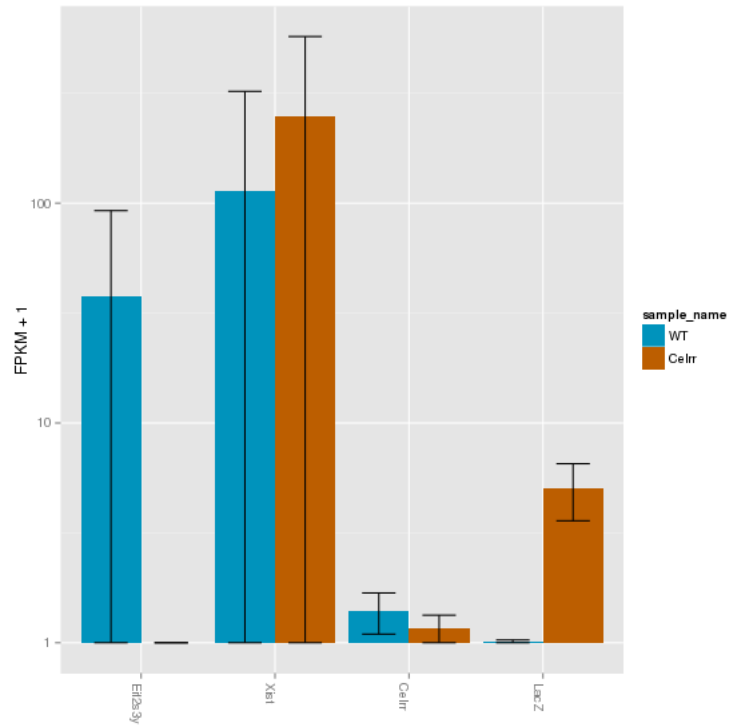


LacZ expression

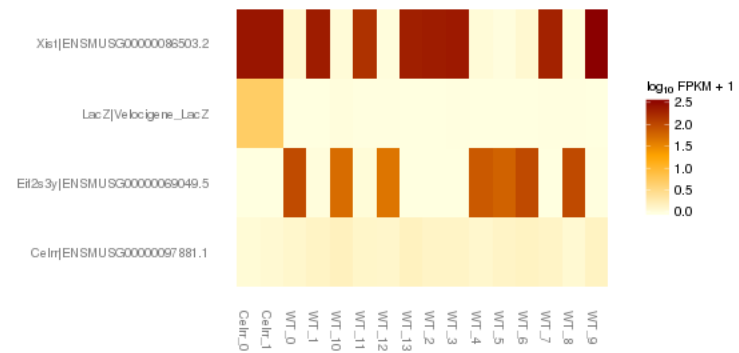


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

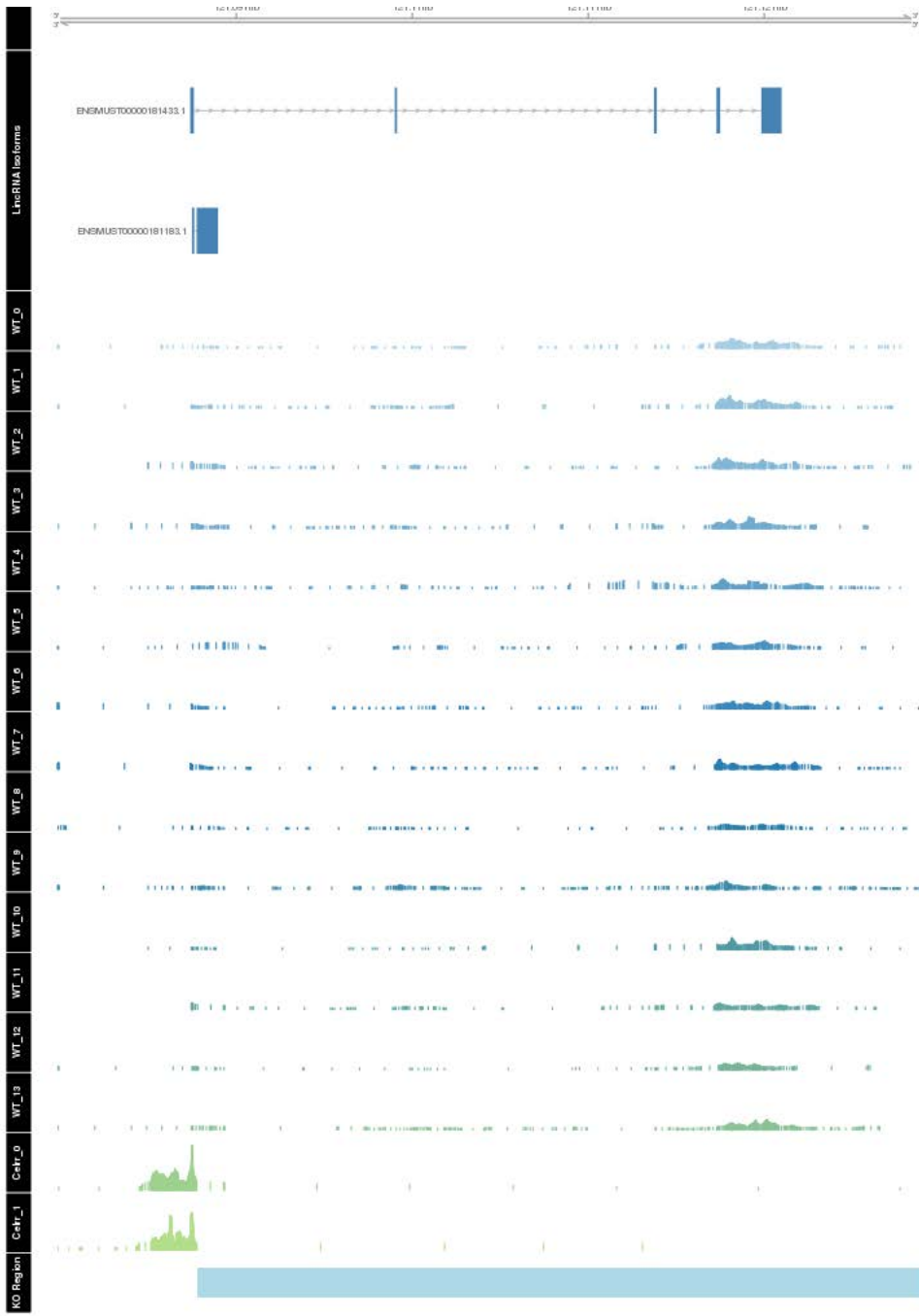


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 39 significantly differentially expressed genes. They are:

geneAnnot\$gene_short_name

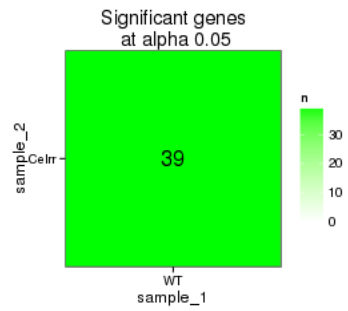
- 1 Mef2c
- 2 Phox2b
- 3 Sulf1
- 4 Slc16a10
- 5 Fam49a
- 6 Ntn1
- 7 Cxcl14
- 8 Sla
- 9 Clic6
- 10 Rps14
- 11 Fn1
- 12 Notch1
- 13 Notch2
- 14 Dync1i1
- 15 Dbx1
- 16 Slit2
- 17 Robo3
- 18 Lama1
- 19 Kirrel2
- 20 Neurod6
- 21 Notch3
- 22 Spon1
- 23 Satb2
- 24 Mpped1
- 25 Tuba1c
- 26 Cnpy1
- 27 Hbb-y
- 28 Krt15
- 29 Hba-x
- 30 Rpl32
- 31 Rpl26
- 32 Rps7
- 33 Rpl35
- 34 Cyp26b1
- 35 Ddx3y
- 36 Fat1
- 37 Rpl23

38 Rpl39

39 AA465934

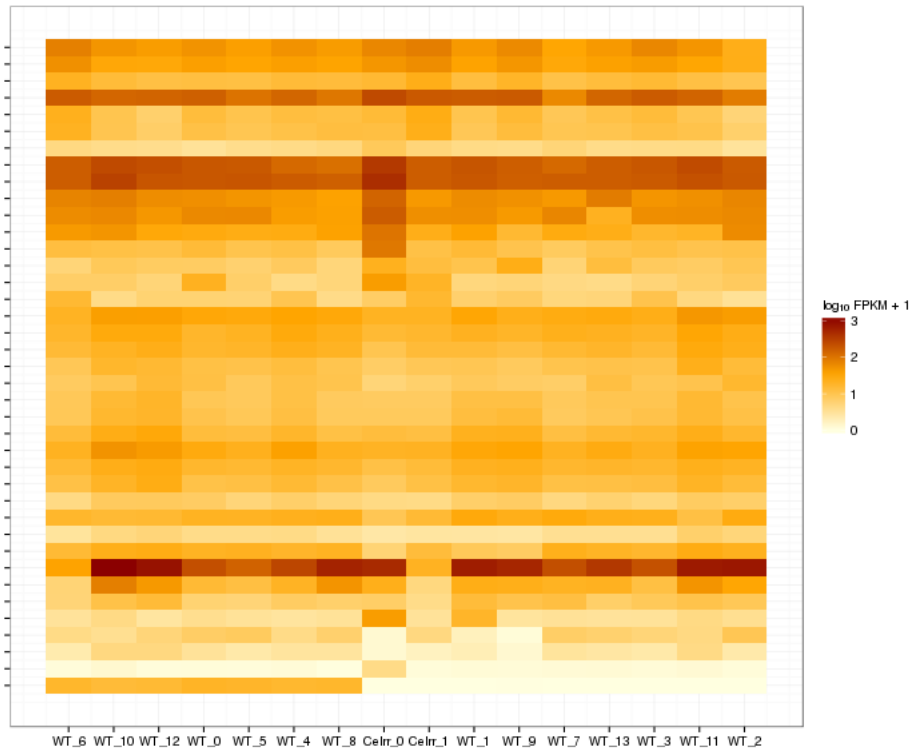
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

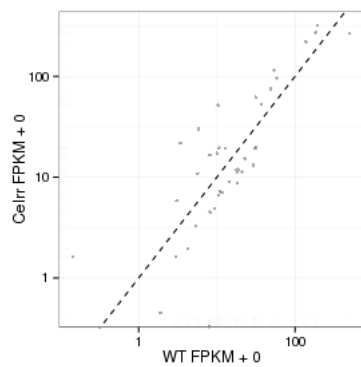


Significant genes with expression >50fpkm (any condition):(turned off)

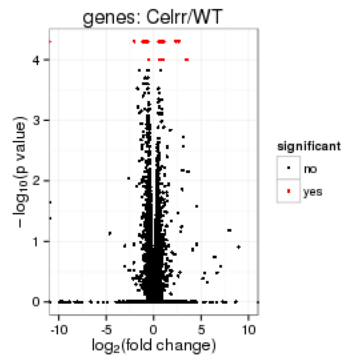
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

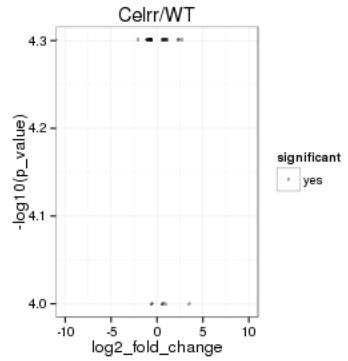
Scatter plot of significant genes only:



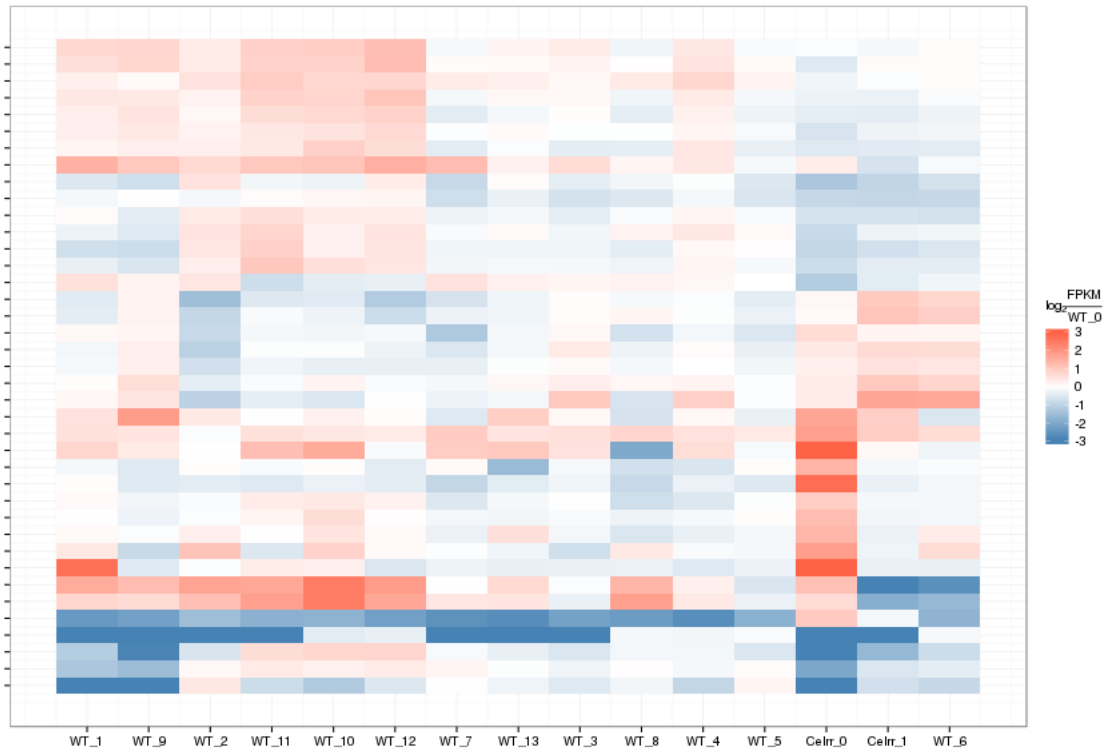
Volcano Plot



Volcano plot with significant genes only:



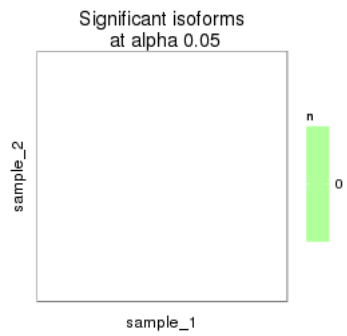
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

[1] "no sig isoforms"

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

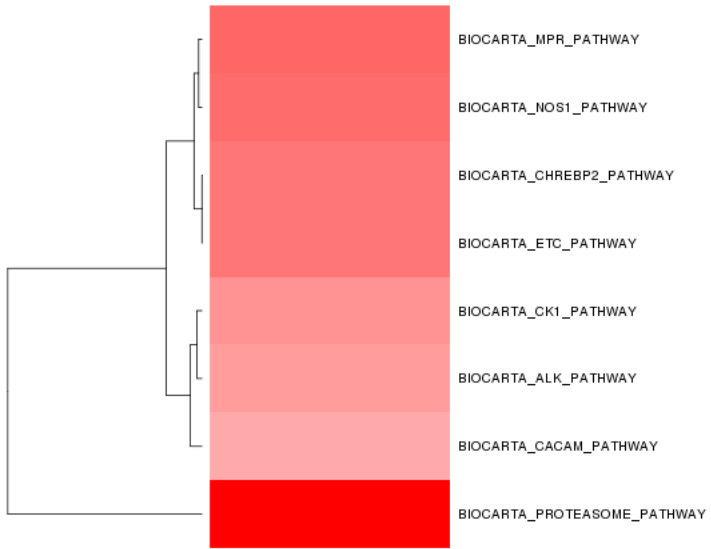
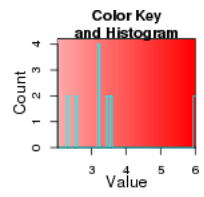
Gene/Pathway Analysis

GSEA

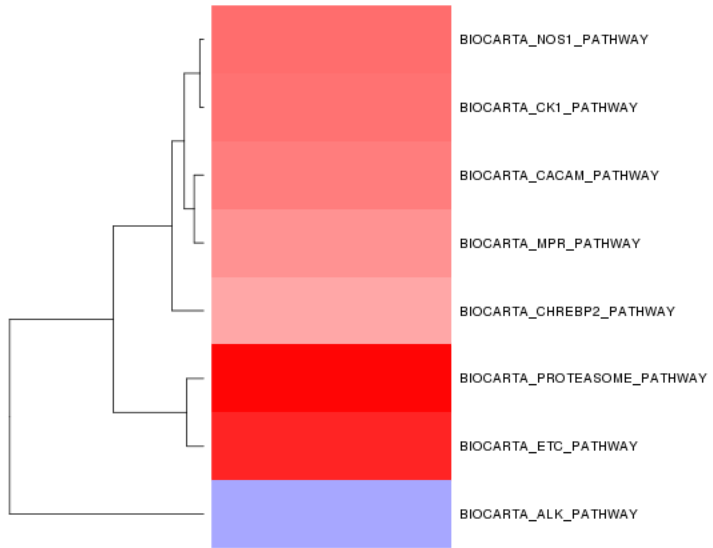
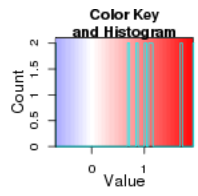
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

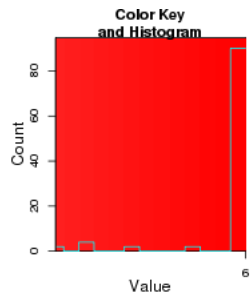
Biocarta enrichment:

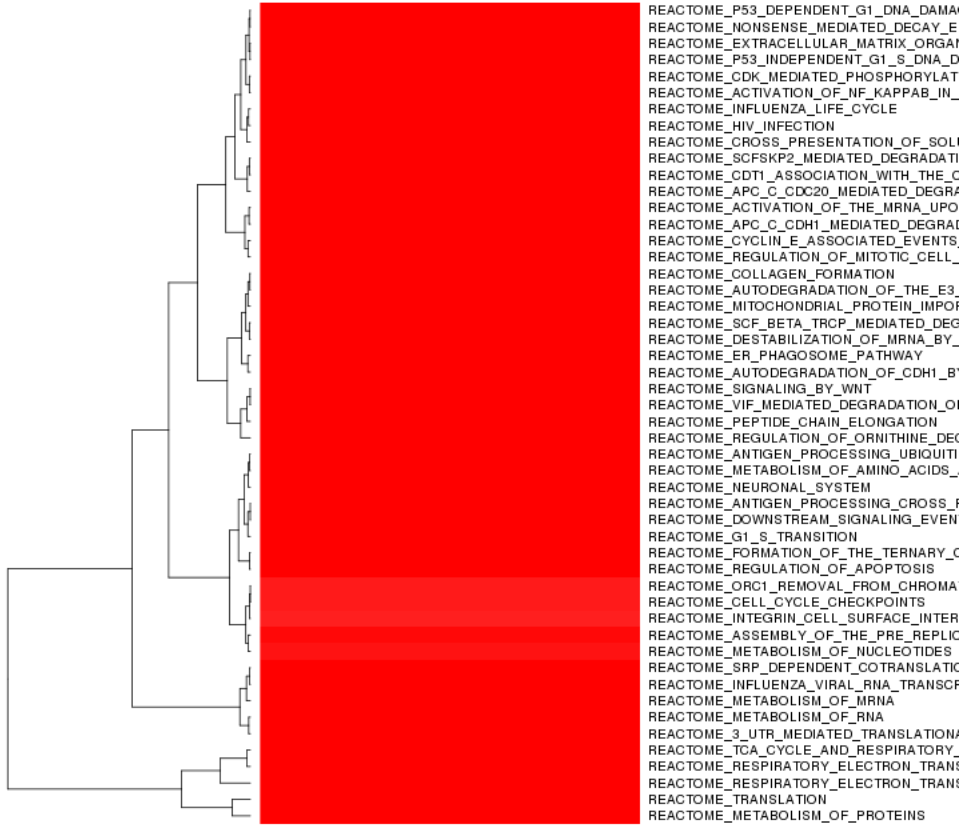


Biocarta zscore:

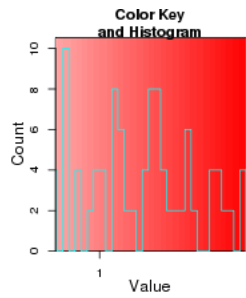


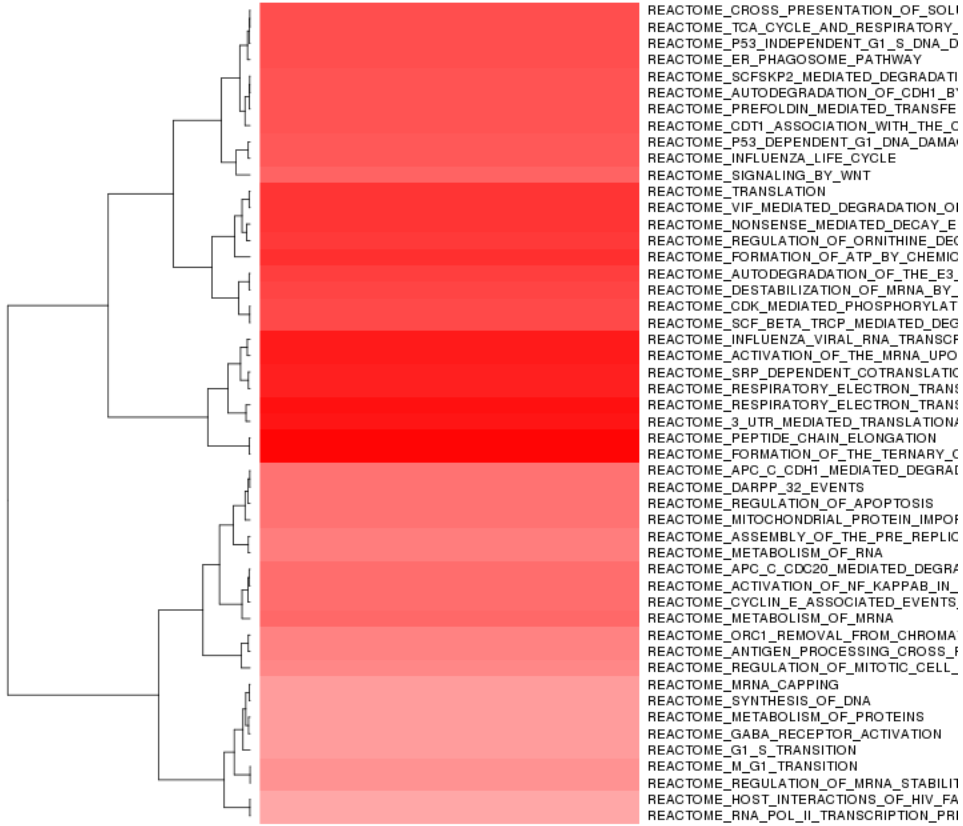
Reactome enrichment:



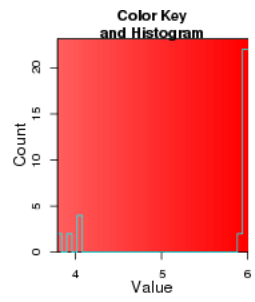


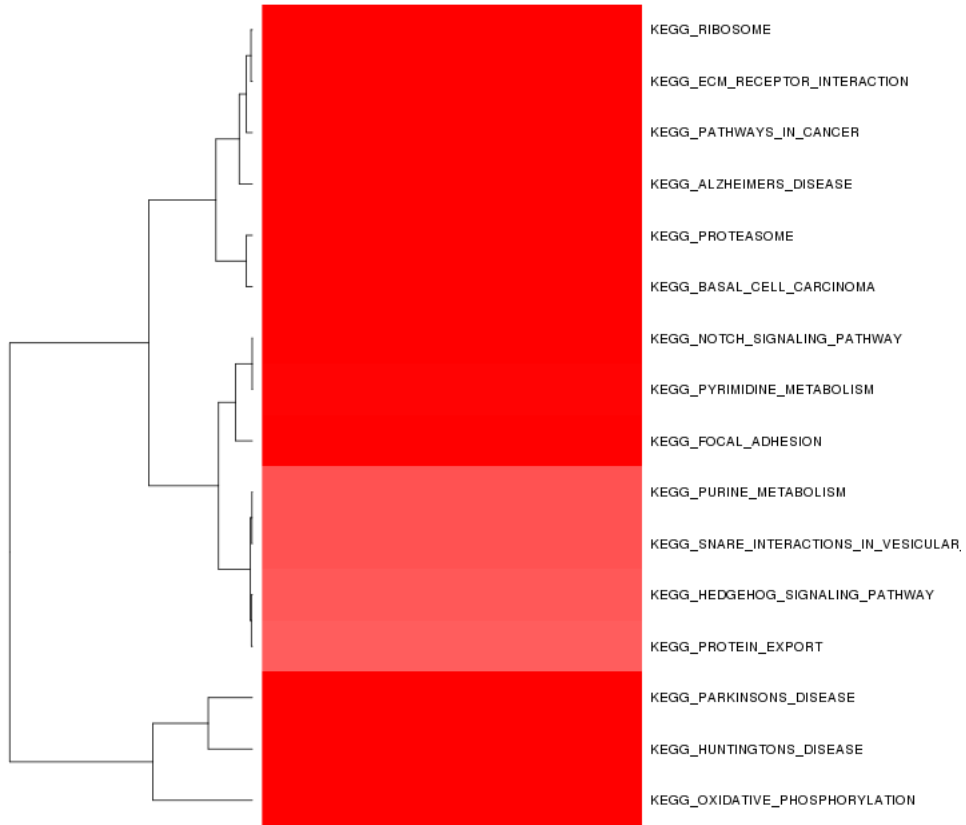
Reactome zscore:



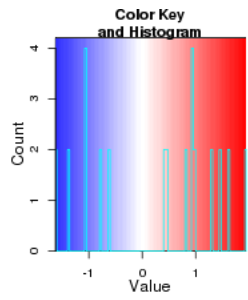


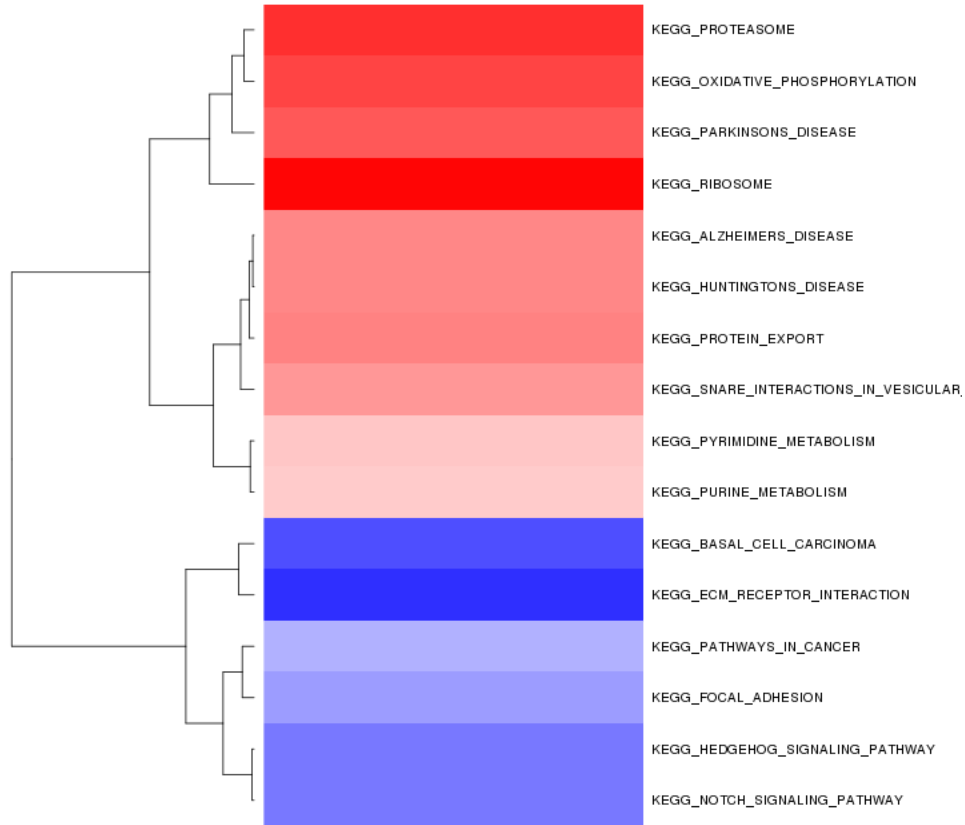
Kegg enrichment:





Kegg zscore:





Interneuron enrichment:

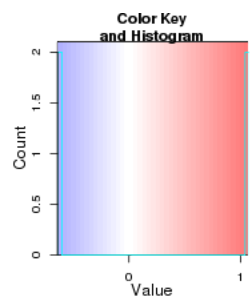
Error: no locations are finite

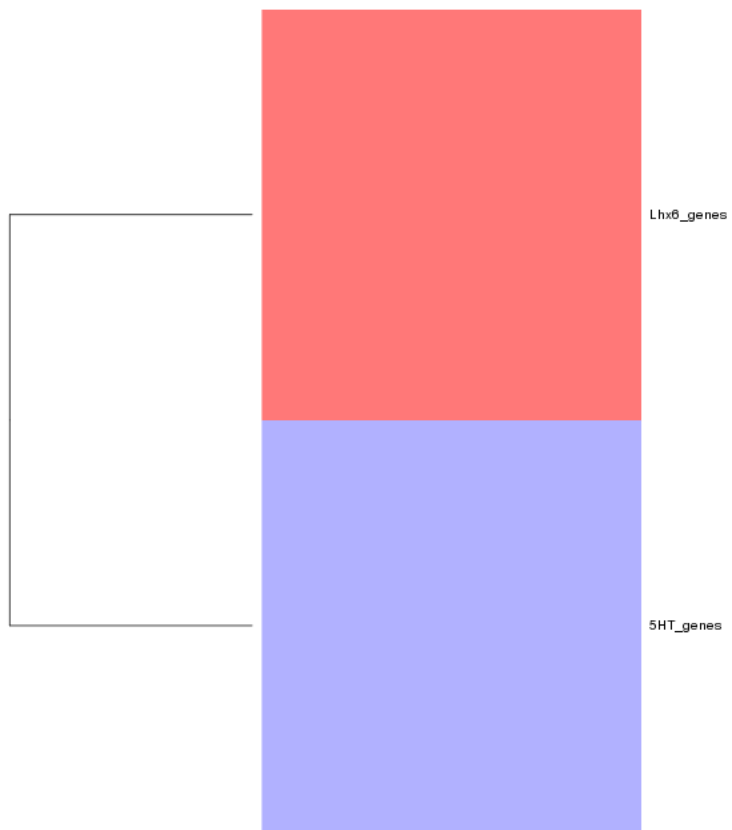


Lhx6_genes

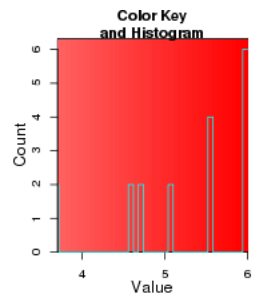
5HT_genes

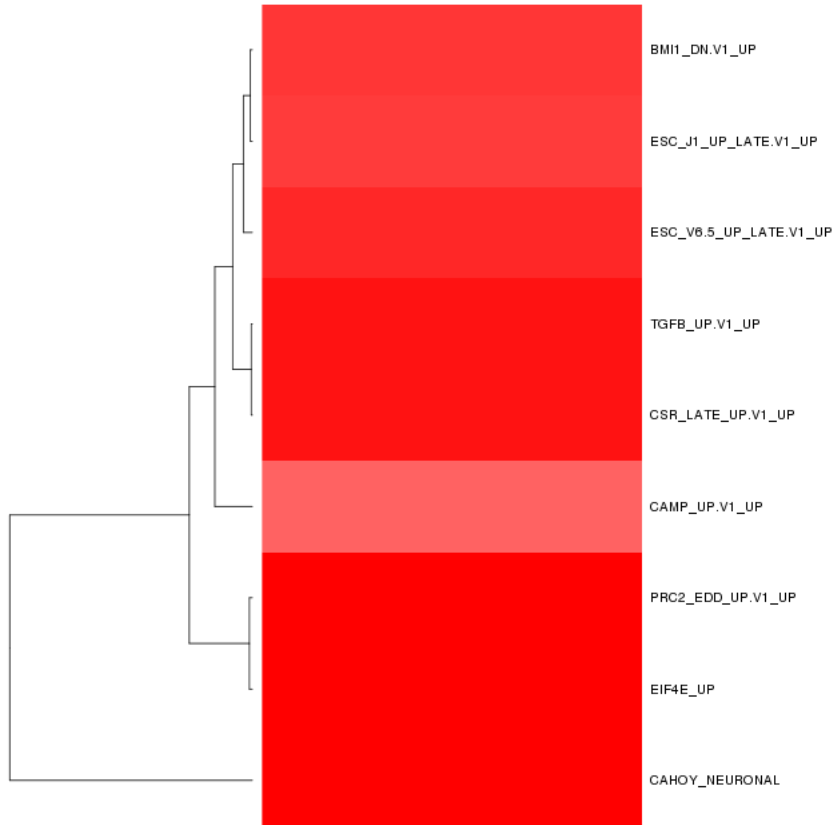
Interneuron zscore:



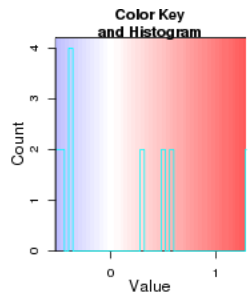


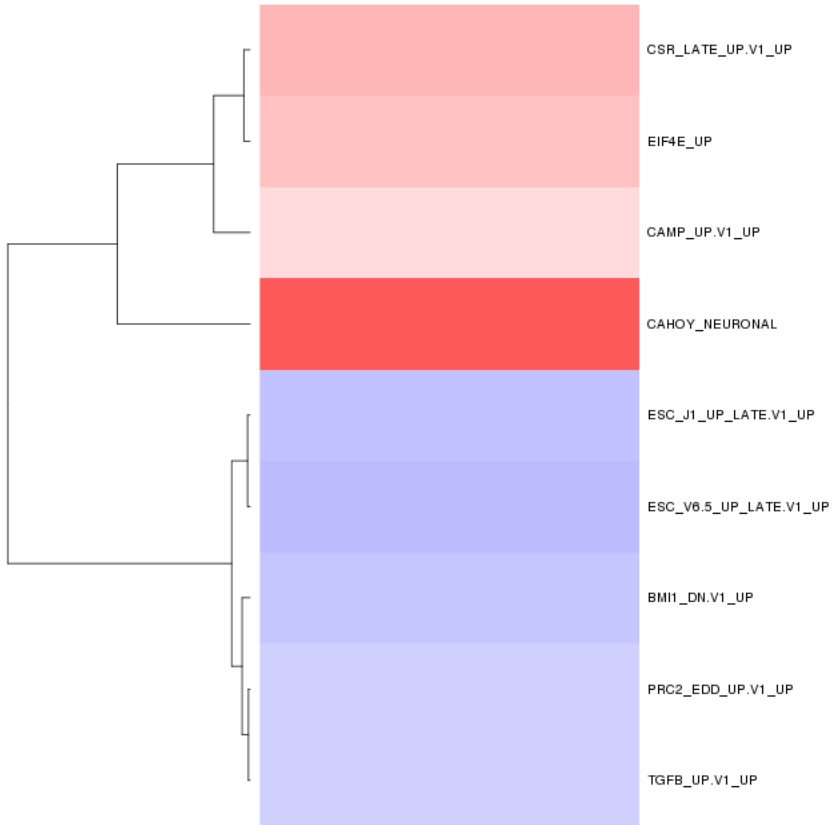
Oncogene enrichment:





Oncogene zscore:

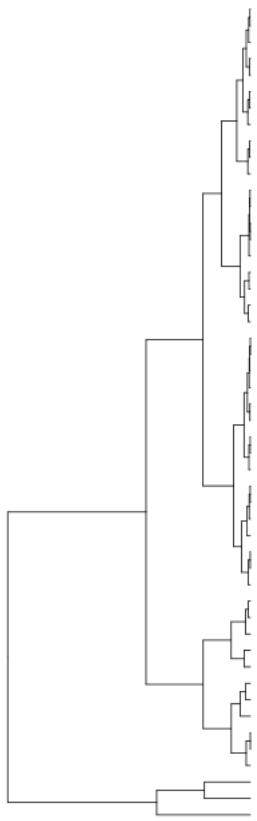




Immuno enrichment:

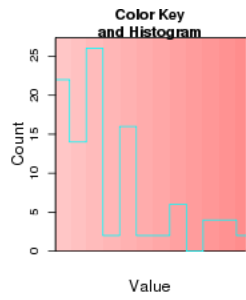
Error: no locations are finite

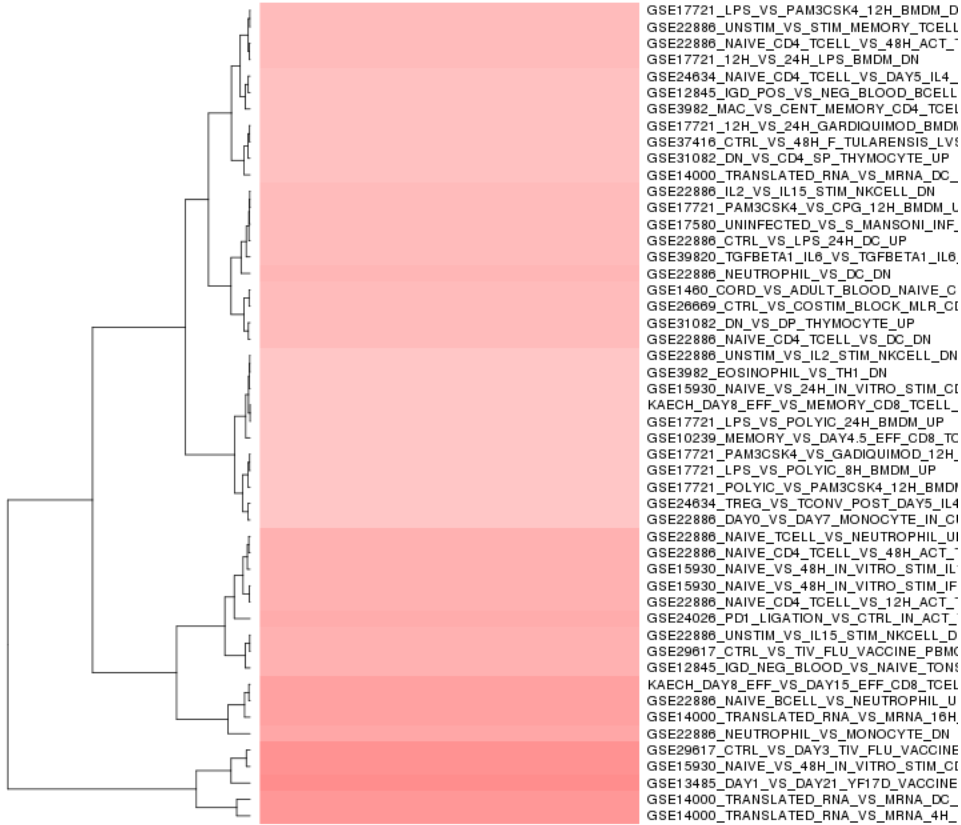




GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IN
GSE14000_TRANSLATED_RNA_VS_MRNA_DC_
GSE17721_LPS_VS_PAM3CSK4_12H_BMDM_D
GSE17721_12H_VS_24H_LPS_BMDM_DN
GSE17580_UNINFECTED_VS_S_MANSONI_INF.
GSE12845_IGD_POS_VS_NEG_BLOOD_BCELL
GSE14000_TRANSLATED_RNA_VS_MRNA_DC_
GSE22886_UNSTIM_VS_STIM_MEMORY_TCELL
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_CI
GSE24026_PD1_LIGATION_VS_CTRL_IN_ACT_
GSE17721_LPS_VS_PAM3CSK4_1H_BMDM_UP
GSE24634_TREG_VS_TCONV_POST_DAYS_IL4
GSE22886_NEUTROPHIL_VS_DC_DN
GSE22886_NAIVE_TCELL_VS_NEUTROPHIL_UI
GSE17721_PAM3CSK4_VS_CPG_12H_BMDM_L
GSE12845_IGD_NEG_BLOOD_VS_NAIVE_TONR
GSE39820_TGFBETA1_IL6_VS_TGFBETA1_IL6
GSE31082_DN_VS_DP_THYMOCYTE_UP
GSE17721_12H_VS_24H_GARDIQUIMOD_BMDM
GSE26669_CTRL_VS_COSTIM_BLOCK_MLR_CI
GSE22886_DAY1_VS_DAY7_MONOCYTE_IN_CI
GSE17721_LPS_VS_CPG_4H_BMDM_DN
GSE3982_EOSINOPHIL_VS_TH1_DN
GSE22886_DAY0_VS_DAY7_MONOCYTE_IN_CI
GSE17721_PAM3CSK4_VS_GADIOQUIMOD_12H
GSE22886_NAIVE_CD4_TCELL_VS_48H_ACT_
GSE17721_LPS_VS_POLYIC_24H_BMDM_UP
GSE13485_DAY7_VS_DAY21_YF17D_VACCINE
GSE37416_CTRL_VS_48H_F_TULARENSIS_LV1
GSE1460_CORD_VS_ADULT_BLOOD_NAIVE_C
GSE17974_CTRL_VS_ACT_IL4_AND_ANTI_IL12
GSE10239_MEMORY_VS_DAY4.5_EFF_CD8_TC
GSE22886_NEUTROPHIL_VS_MONOCYTE_DN
KAECH_DAY8_EFF_VS_MEMORY_CD8_TCELL_
GSE24634_TEFF_VS_TCONV_DAYS_IN_CULTU
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IL
GSE14000_TRANSLATED_RNA_VS_MRNA_4H_
GSE22886_NAIVE_CD4_TCELL_VS_48H_ACT_
GSE14000_TRANSLATED_RNA_VS_MRNA_16H
KAECH_DAY8_EFF_VS_DAY15_EFF_CD8_TCEL
GSE22886_NAIVE_CD4_TCELL_VS_12H_ACT_
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IF
GSE22886_UNSTIM_VS_IL15_STIM_NKCELL_D
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IL
GSE29617_CTRL_VS_TIV_FLU_VACCINE_PBMK
GSE22886_IL2_VS_IL15_STIM_NKCELL_DN
GSE24634_NAIVE_CD4_TCELL_VS_DAYS_IL4_
GSE13485_DAY1_VS_DAY21_YF17D_VACCINE
GSE29617_CTRL_VS_DAY3_TIV_FLU_VACCINE
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_CI

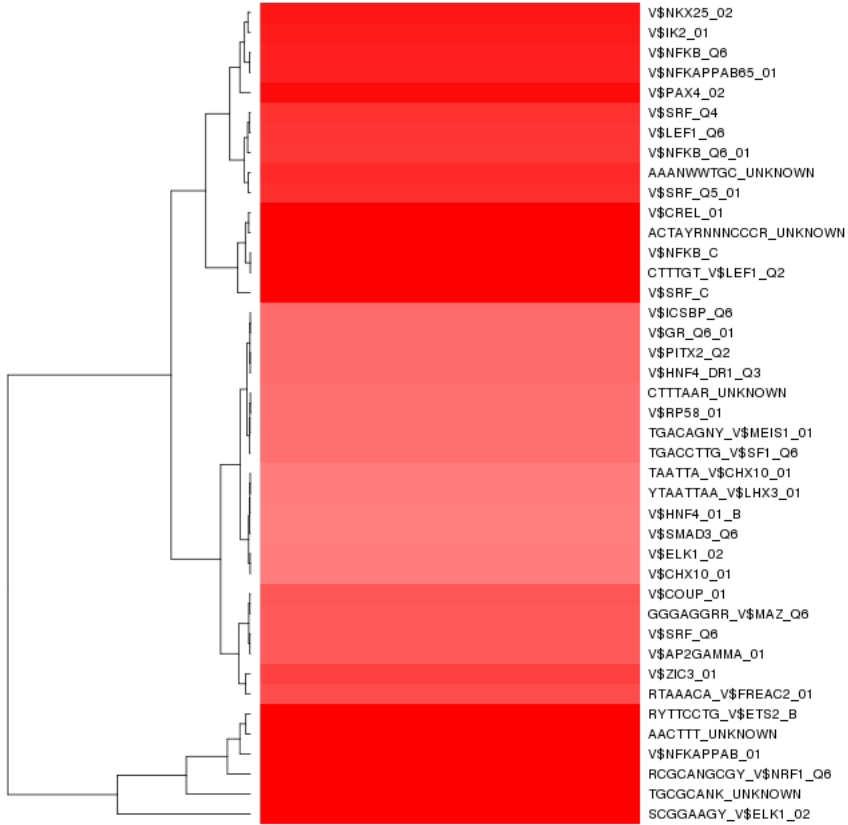
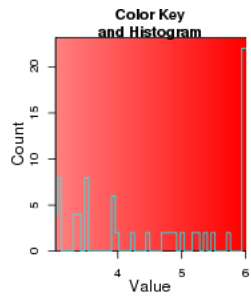
Immuno zscore:





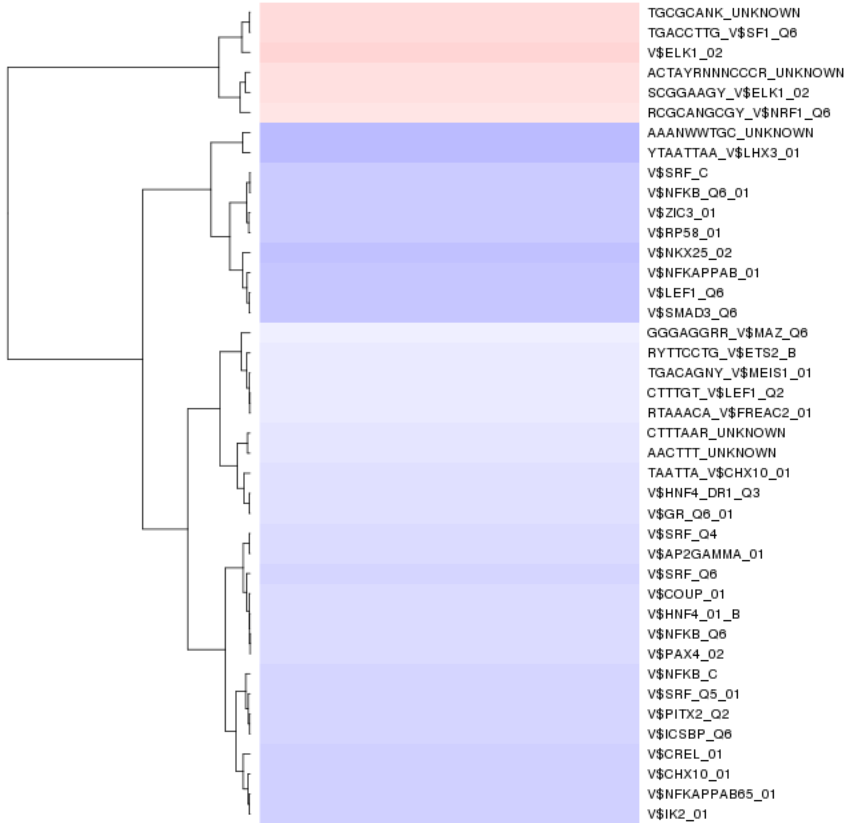
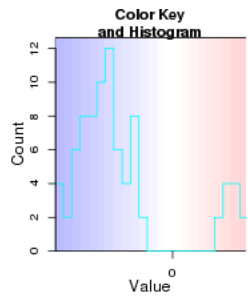
TF enrichment:

Error: subscript out of bounds



TF zscore:

Error: subscript out of bounds

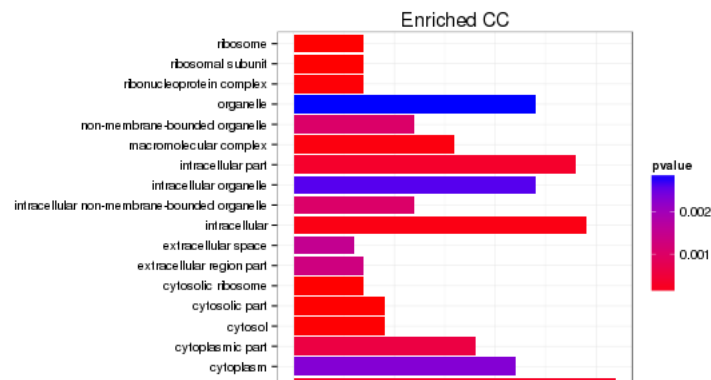
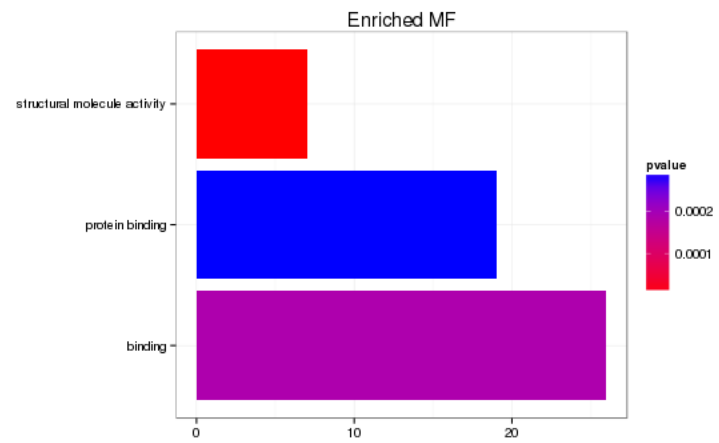
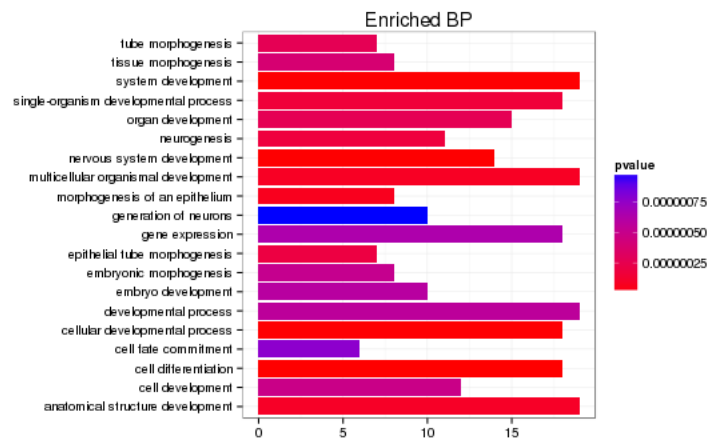


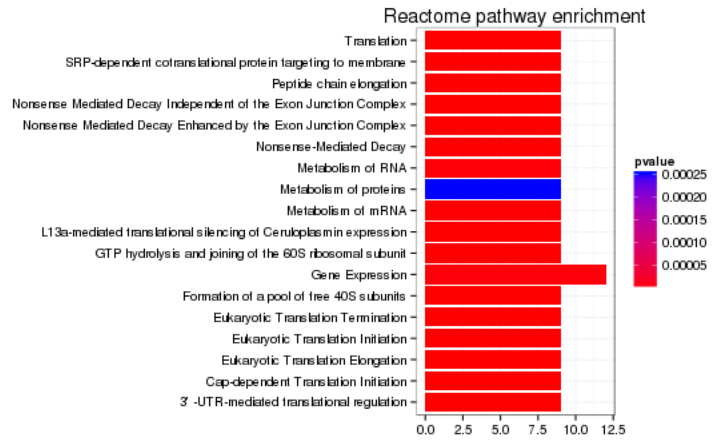
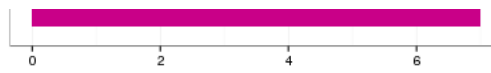
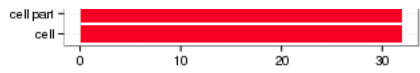
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

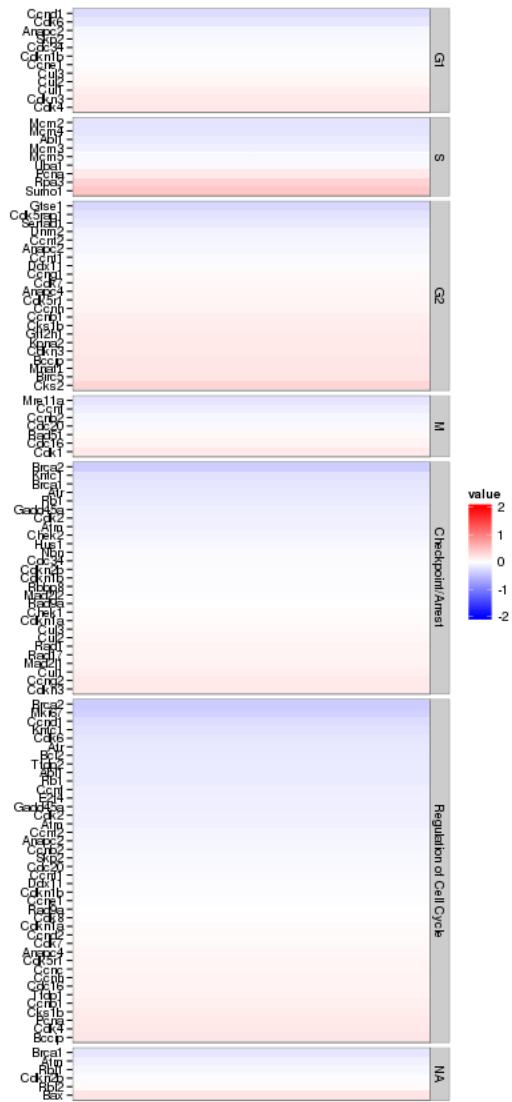
```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```

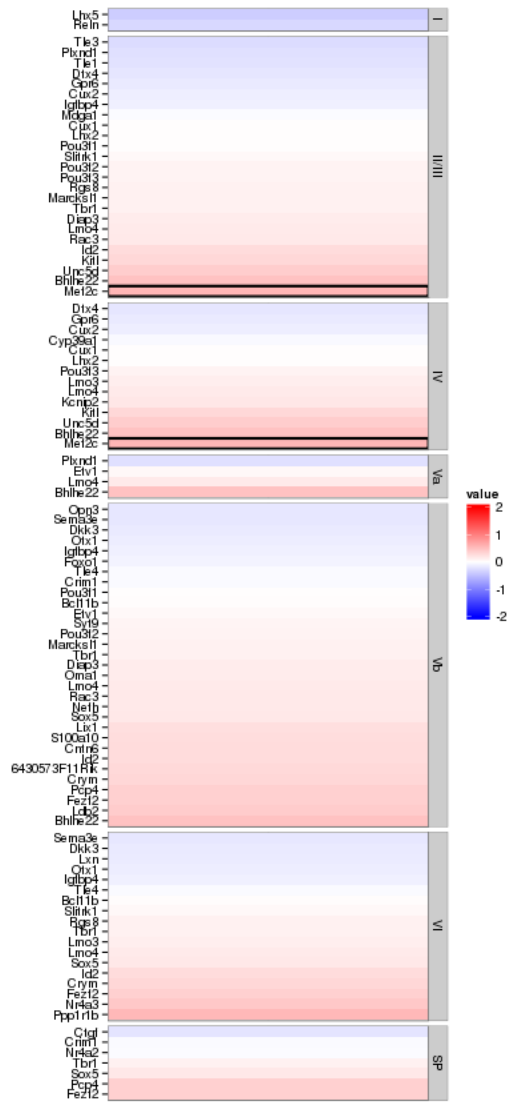
```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```



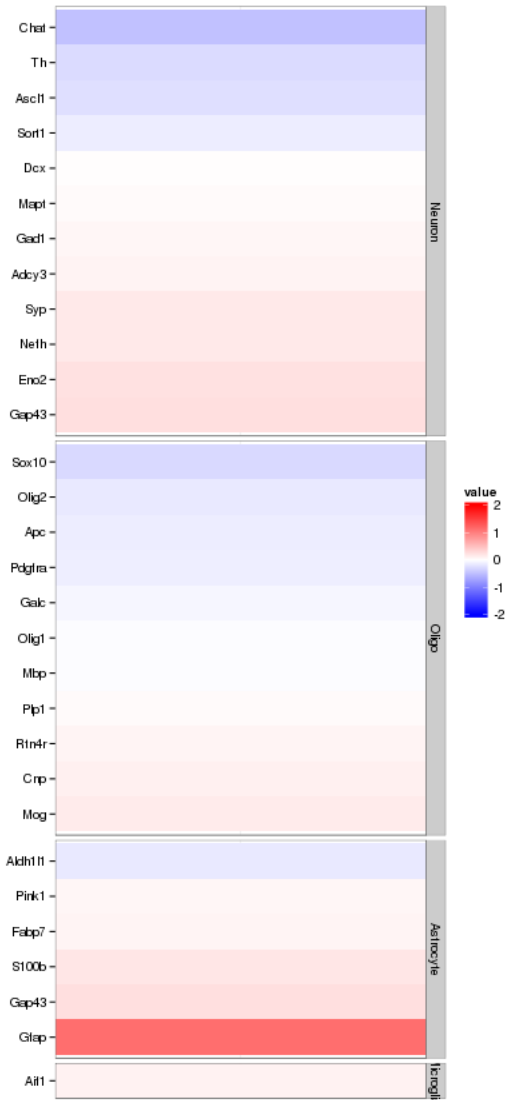


Enrichment or depletion for stage-specific cell cycle markers

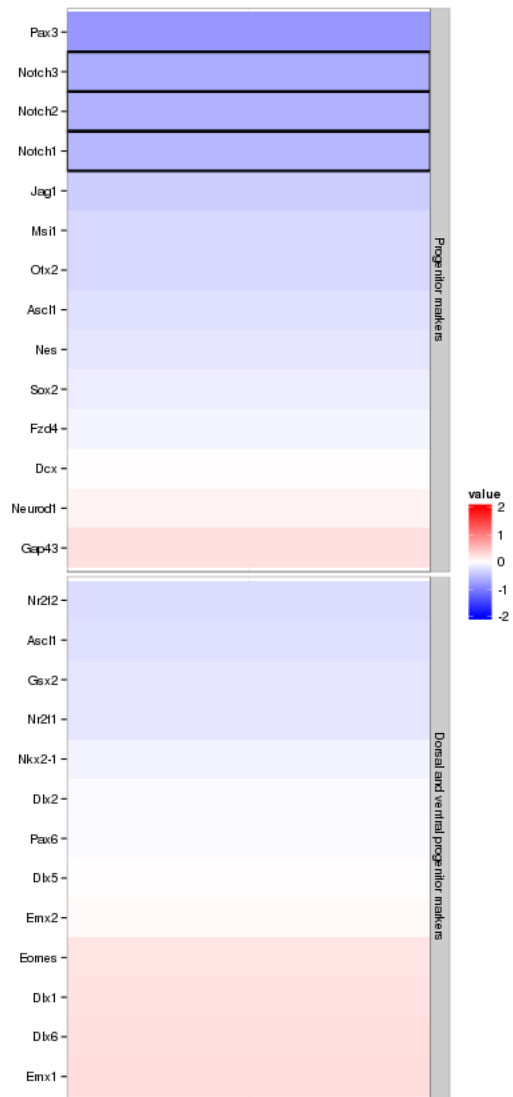




Enrichment or depletion for specific neural cell types



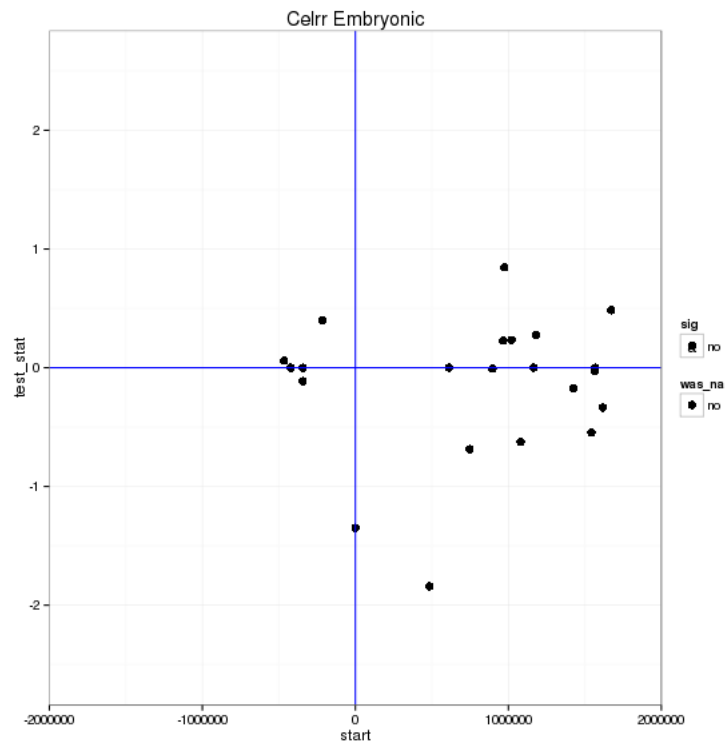
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR773

16 JR760

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR753/abundances.cxb WT		0	WT_0	39331900.00	32377800.00	1.26	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR750/abundances.cxb WT		1	WT_1	36031100.00	32377800.00	1.10	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR771/abundances.cxb WT		2	WT_2	38857100.00	32377800.00	1.23	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR755/abundances.cxb WT		3	WT_3	41811300.00	32377800.00	1.29	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR811/abundances.cxb WT		4	WT_4	38172800.00	32377800.00	1.20	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR768/abundances.cxb WT		5	WT_5	28375200.00	32377800.00	0.89	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR761/abundances.cxb WT		6	WT_6	31706200.00	32377800.00	0.97	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR815/abundances.cxb WT		7	WT_7	29733100.00	32377800.00	0.92	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR789/abundances.cxb WT		8	WT_8	27681500.00	32377800.00	0.86	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR748/abundances.cxb WT		9	WT_9	32367000.00	32377800.00	0.96	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR716/abundances.cxb WT		10	WT_10	26144700.00	32377800.00	0.81	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR717/abundances.cxb WT		11	WT_11	25565400.00	32377800.00	0.80	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR719/abundances.cxb WT		12	WT_12	22697800.00	32377800.00	0.68	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR756/abundances.cxb WT		13	WT_13	38948900.00	32377800.00	1.21	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR773/abundances.cxb Celrr		0	Celrr_0	37950800.00	32377800.00	1.13	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR760/abundances.cxb Celrr		1	Celrr_1	30671700.00	32377800.00	0.94	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
```

```

## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2     colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4       DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10       Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3    graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1       Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29    latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3       munSELL_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10       qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4       splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7    tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Celrr -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Celrr_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/BrainMap/dat
## 2
## 3
## 4
## 5
```

Crnde KO vs WT (Adult)

This file shows the wt-v-ko comparison for Crnde.

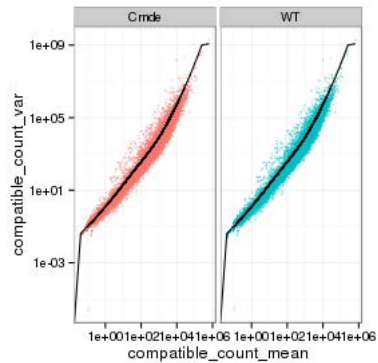
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

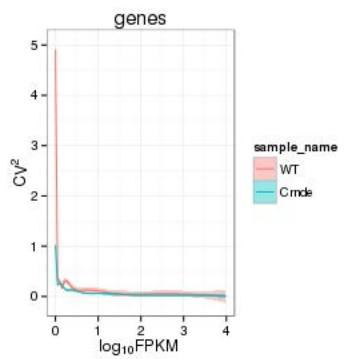
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



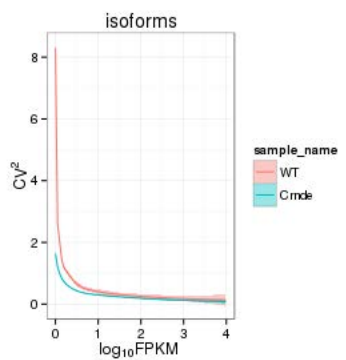
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

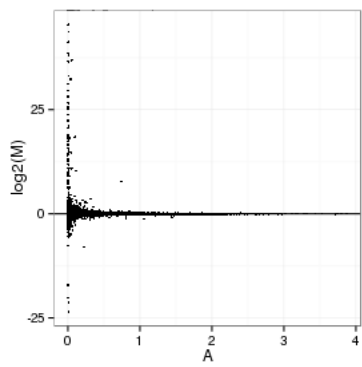
Genes:



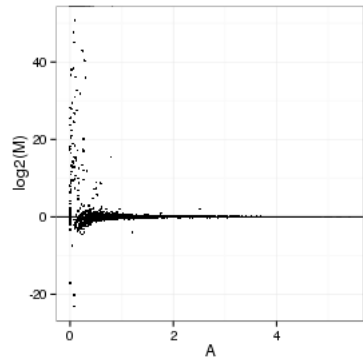
Isoforms:



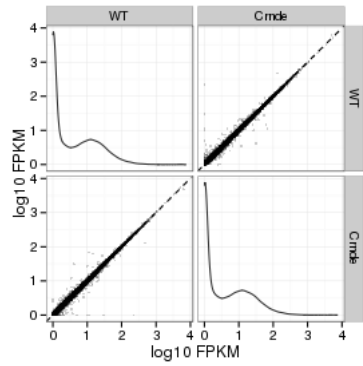
MvA plot



MvA plot counts



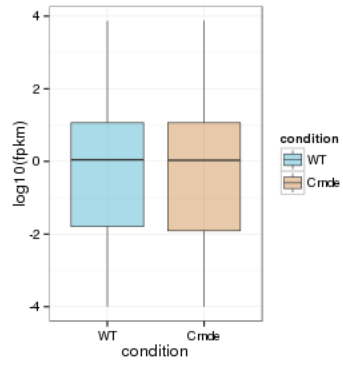
Scatterplot matrix



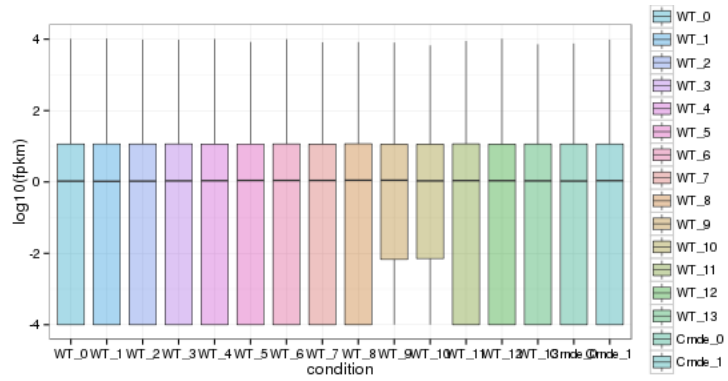
Distributions

Boxplots

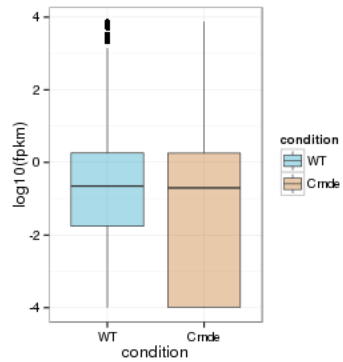
Boxplot (genes)



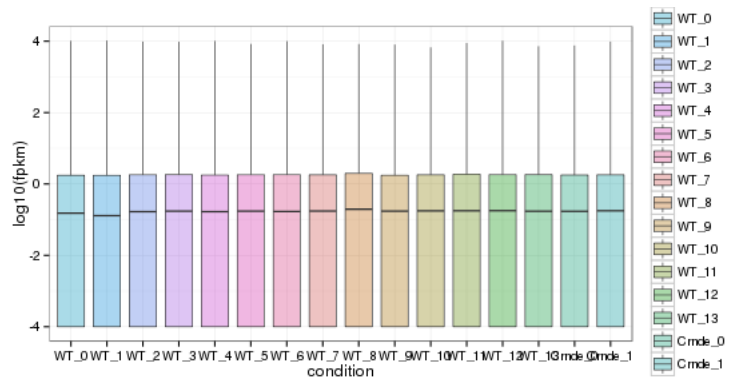
Boxplot (genes, replicates)



Boxplot (isoforms)

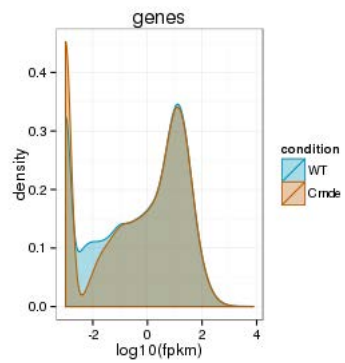


Boxplot (isoforms, replicates)

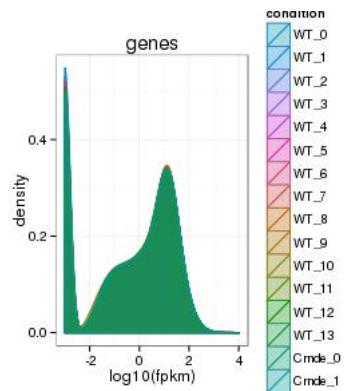


Density

Density (genes)

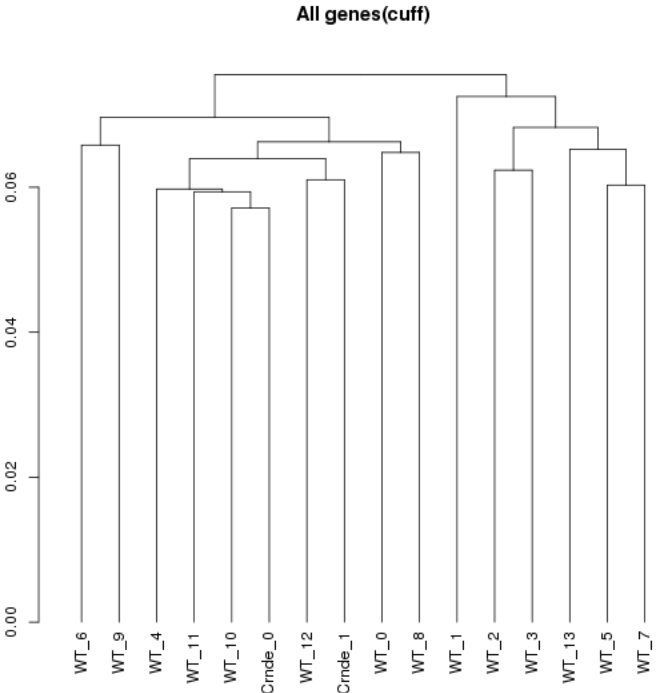


Density (genes, replicates)



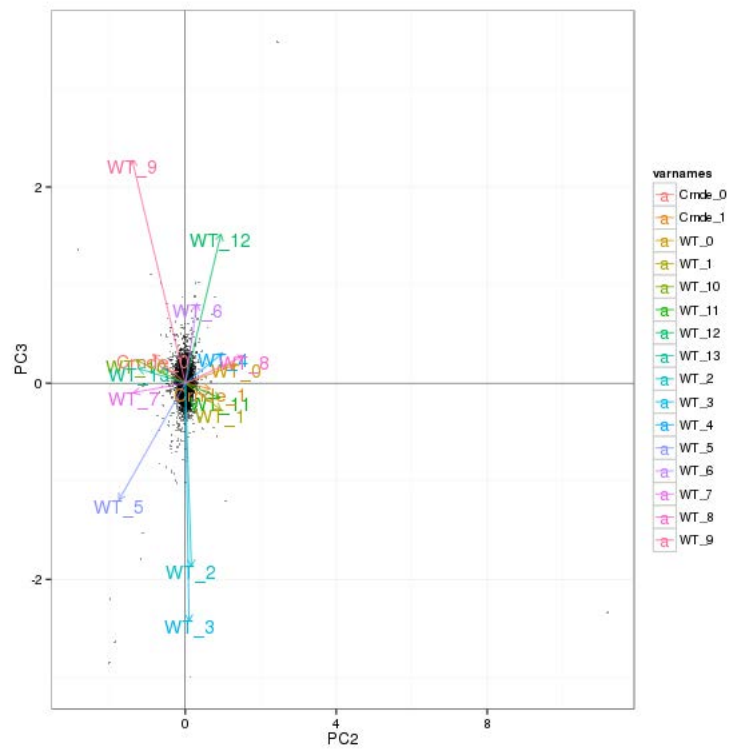
Clustering

Replicate Clusters

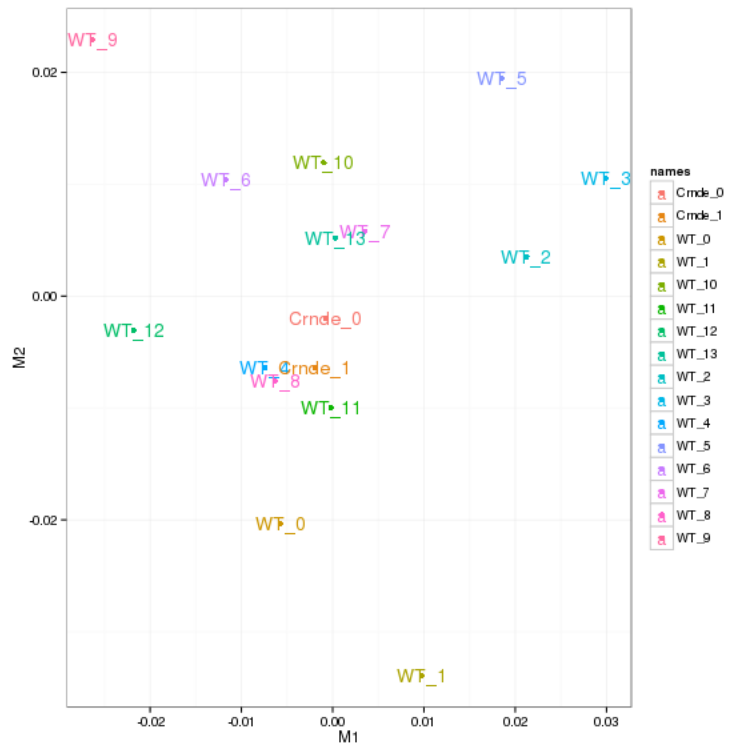


NULL

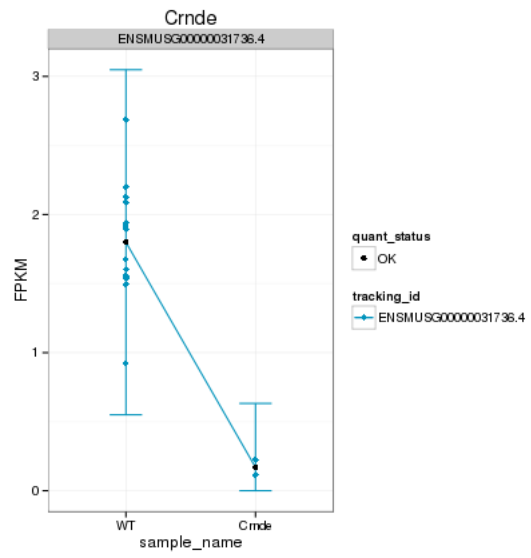
PCA (genes)



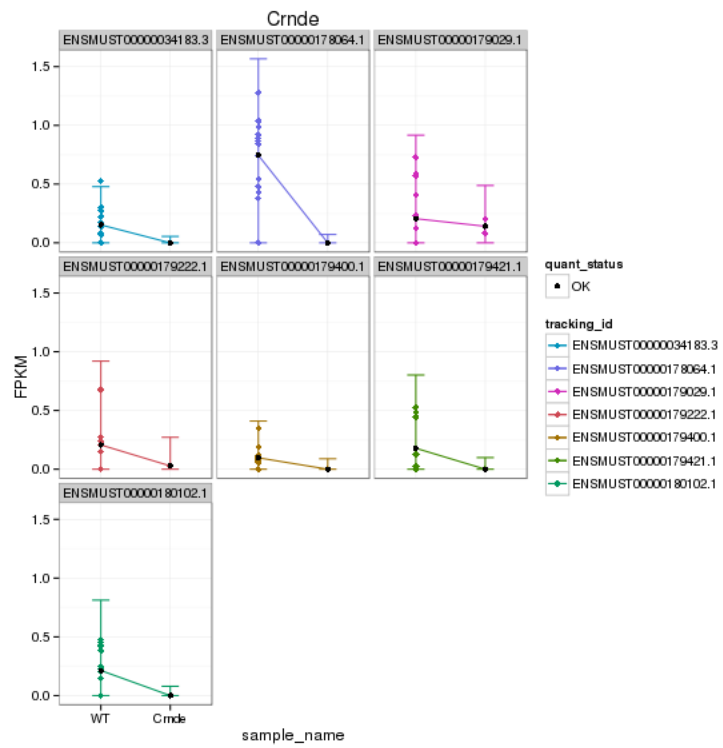
MDS (genes)



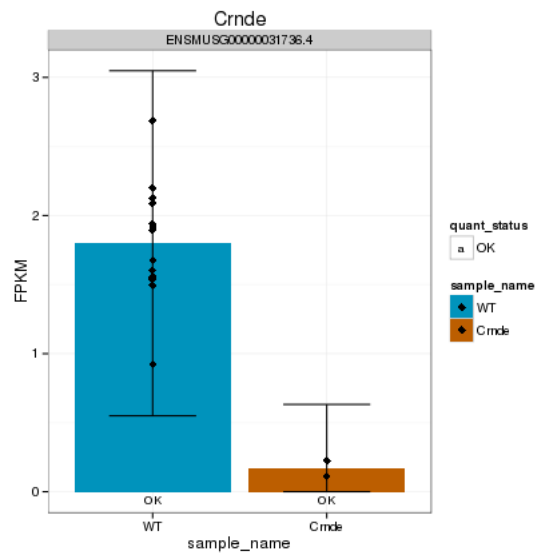
KO assessment
Endogenous lncRNA expression



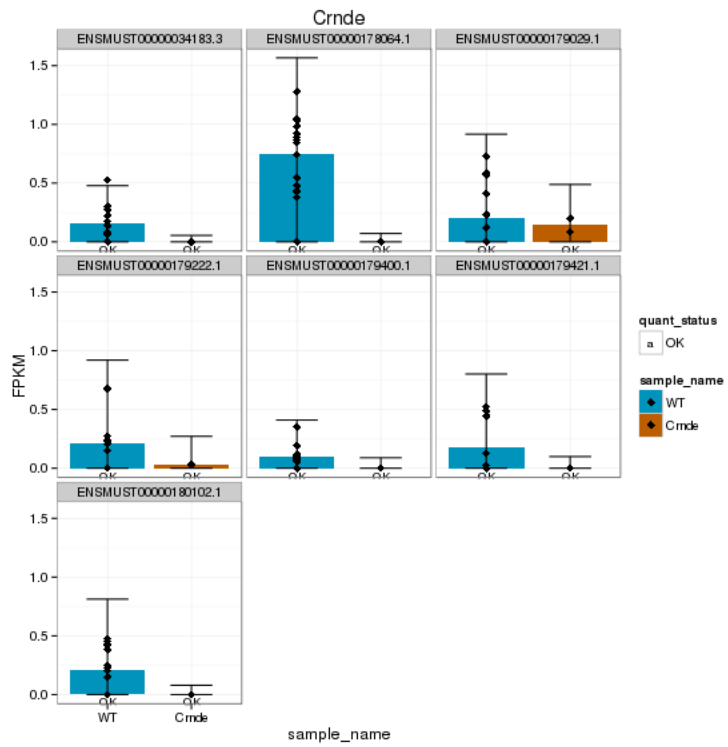
Endogenous expression of Crnde isoforms:



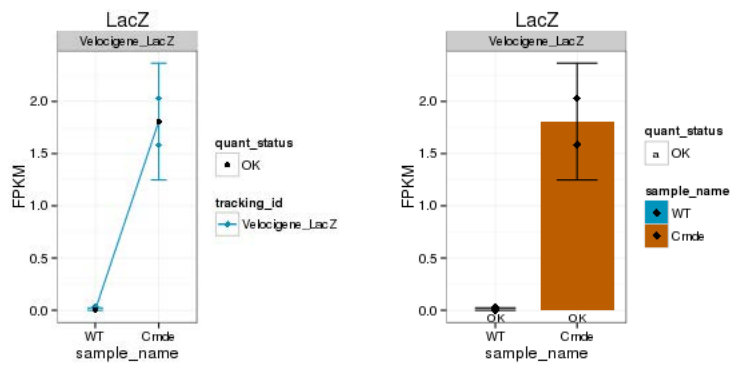
Barplot of gene expression:



Barplot of isoform expression:

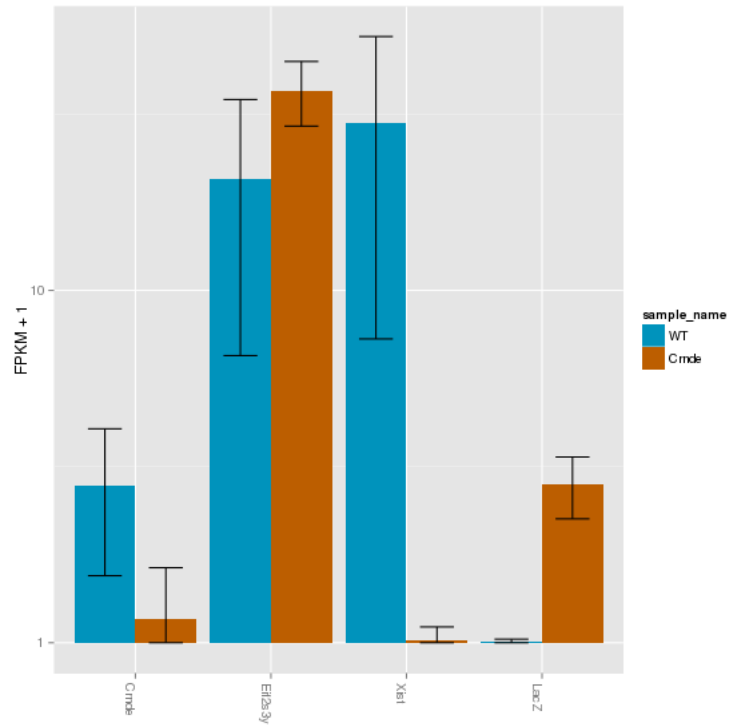


LacZ expression

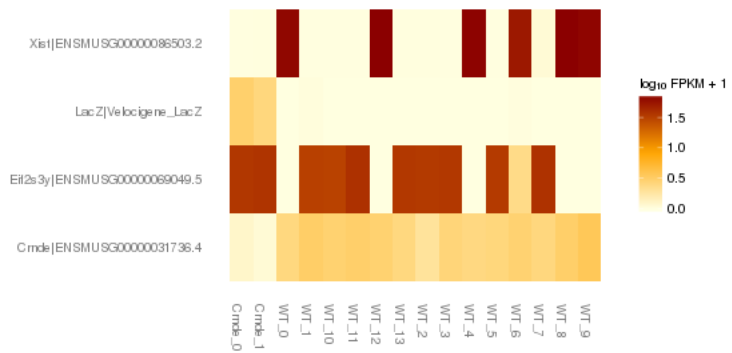


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

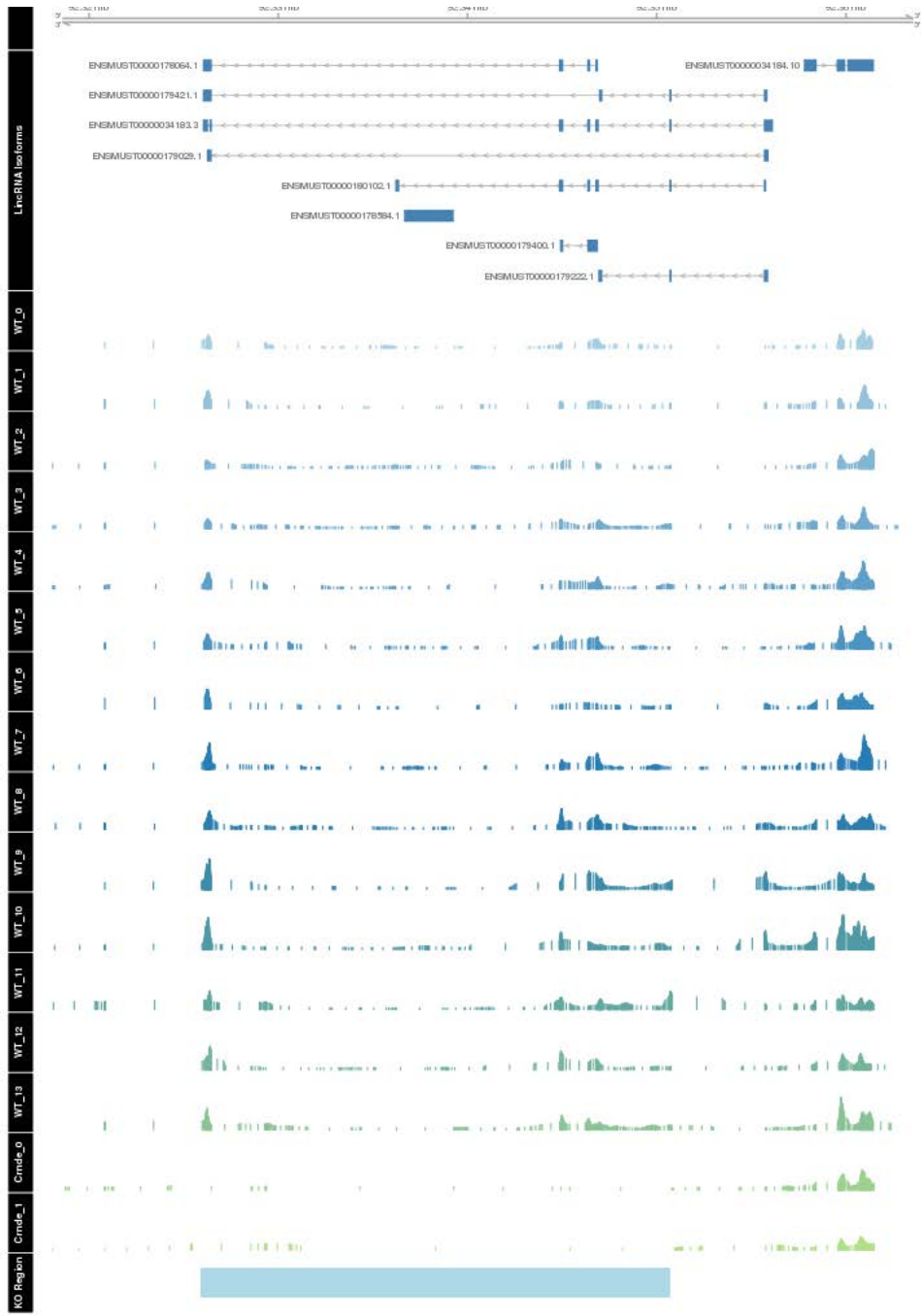


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 42 significantly differentially expressed genes. They are:

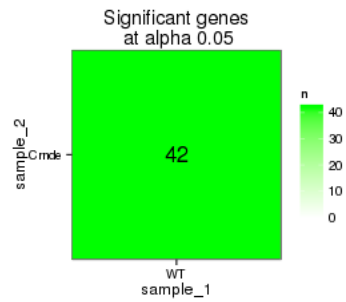
geneAnnot\$gene_short_name

- 1 Fosb
- 2 Merlk
- 3 Adi1
- 4 Fos
- 5 Arc
- 6 Nr4a1
- 7 Cdkn1a
- 8 Clic5
- 9 Fkbp5
- 10 Tma16
- 11 Unc5a
- 12 Sgk3
- 13 Nr4a3
- 14 Slc2a1
- 15 Mfsd2a
- 16 Tinagl1
- 17 Fosl2
- 18 Hddc3
- 19 Mt2
- 20 Sox10
- 21 Lpcat2
- 22 Arhgap27
- 23 Klf10
- 24 Egr2
- 25 Egr1
- 26 Pcsk1n
- 27 Cdc42ep4
- 28 Npas4
- 29 Cdh19
- 30 Junb
- 31 Kdm5d
- 32 Gm5506
- 33 Cox5b
- 34 Scoc
- 35 Ddx3y
- 36 Jund
- 37 Egr4

38 Gm7292
39 A830036E02Rik
40 Xist
41 AA465934
42 Ccdc85b

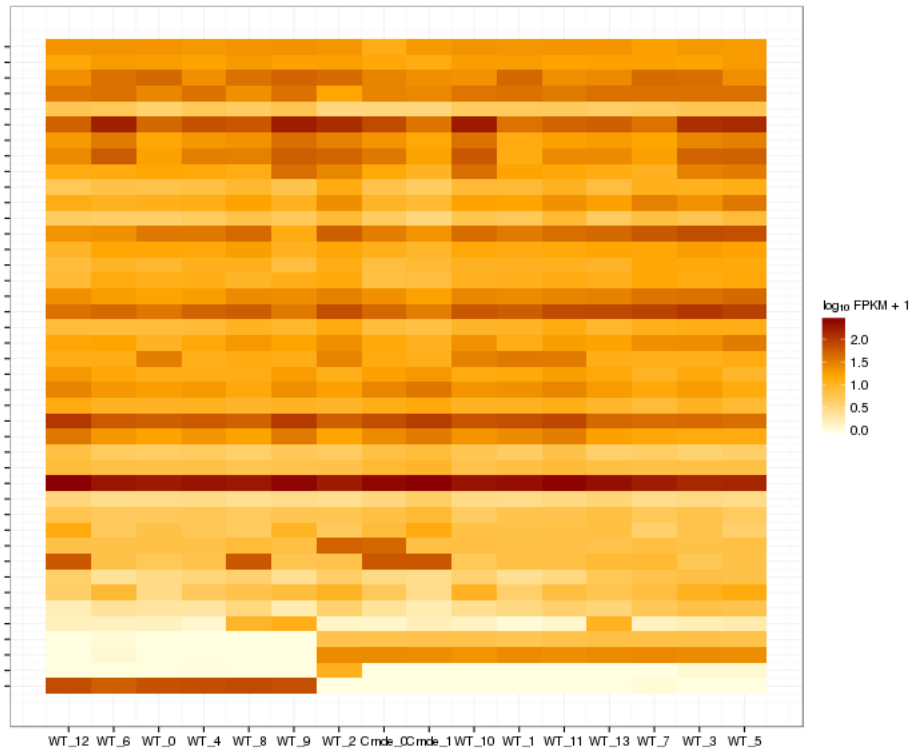
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

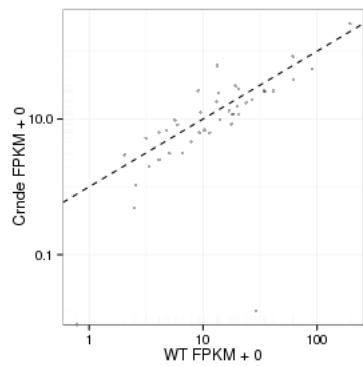


Significant genes with expression >50fpkm (any condition):(turned off)

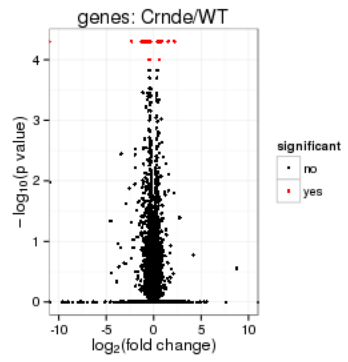
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

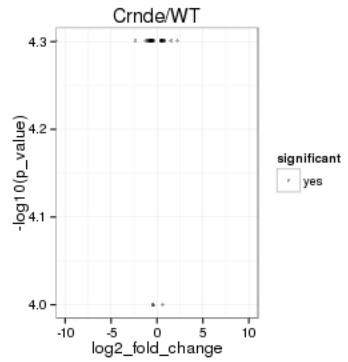
Scatter plot of significant genes only:



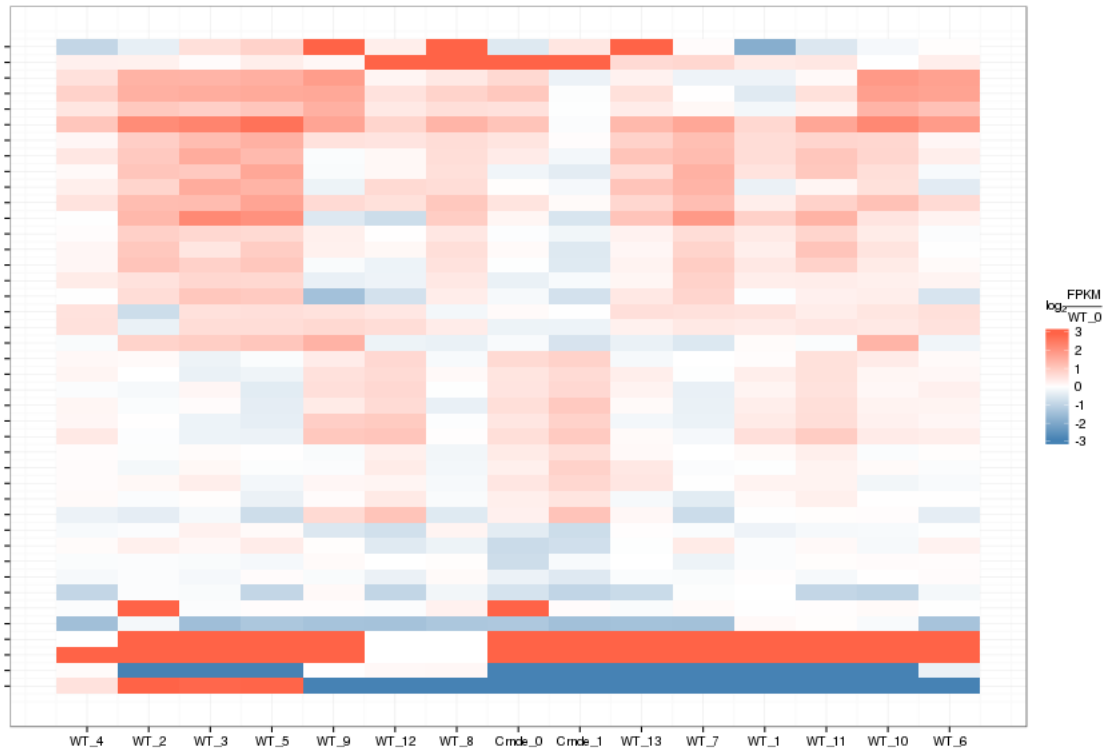
Volcano Plot



Volcano plot with significant genes only:



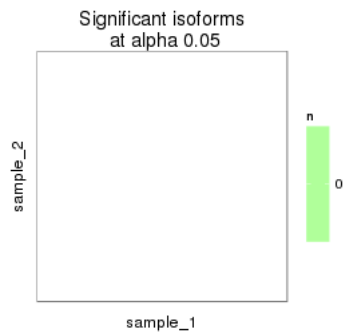
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

[1] "no sig isoforms"

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

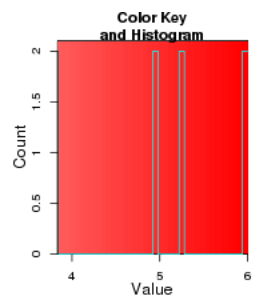
Biocarta enrichment:

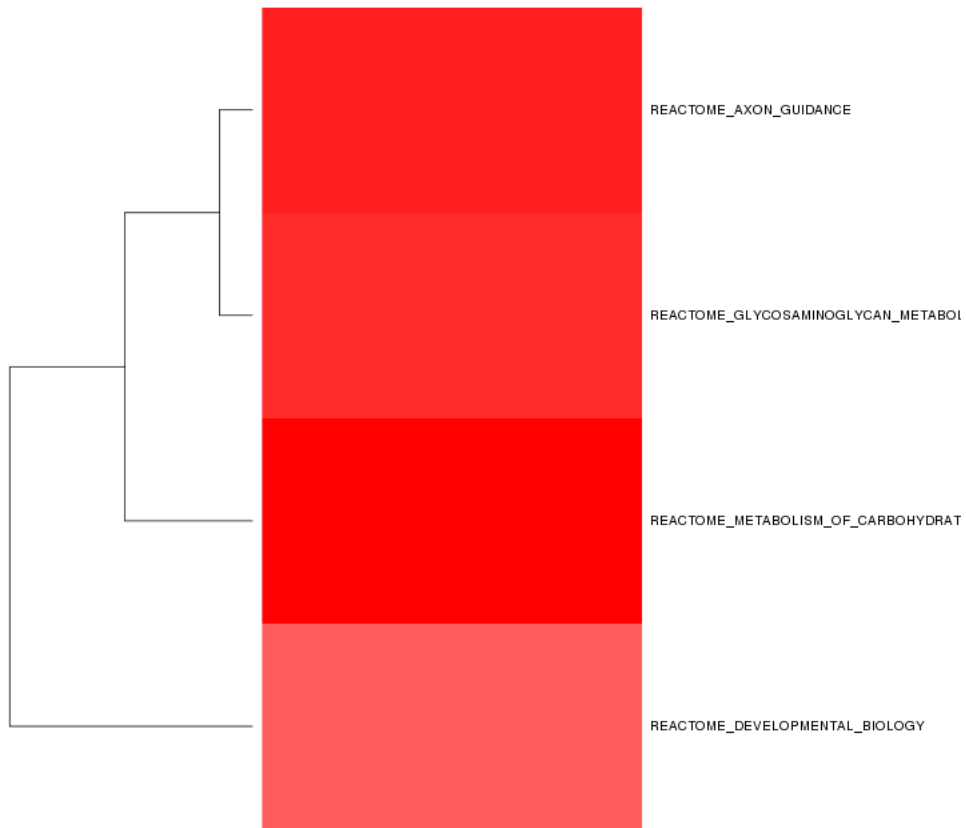
```
## [1] "Not enough significant categories to print a heatmap!"
```

Biocarta zscore:

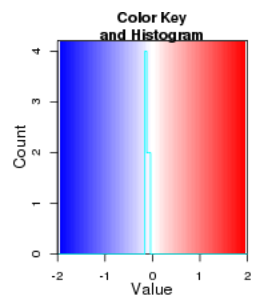
```
## [1] "Not enough significant categories to print a heatmap!"
```

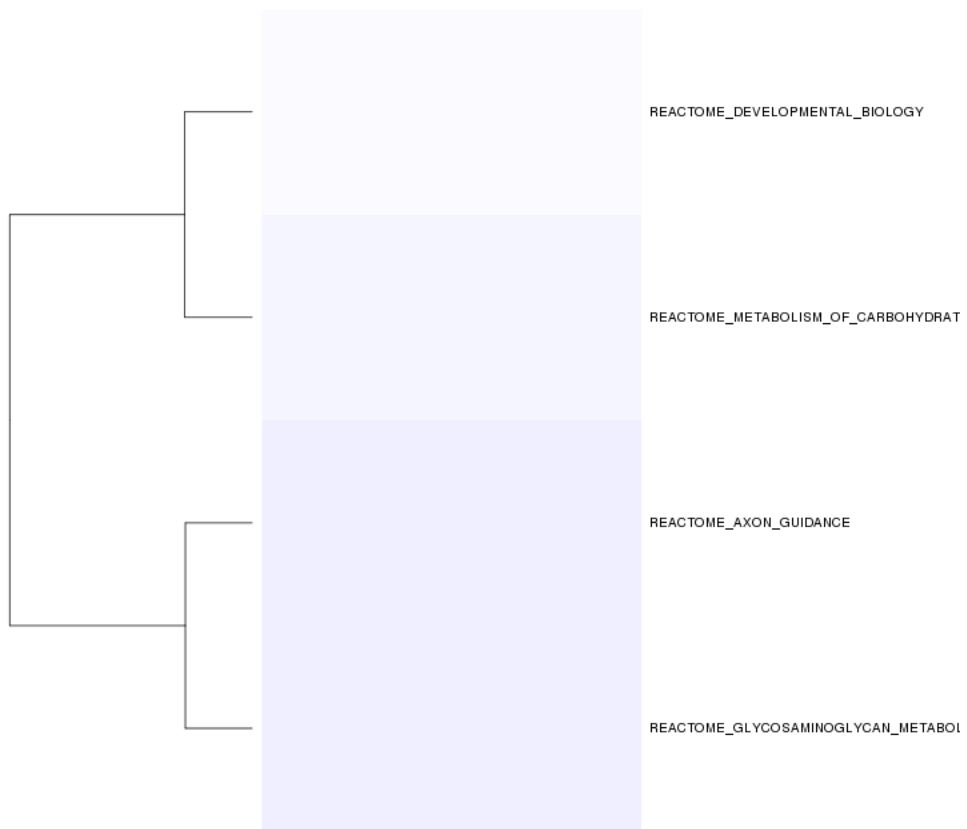
Reactome enrichment:



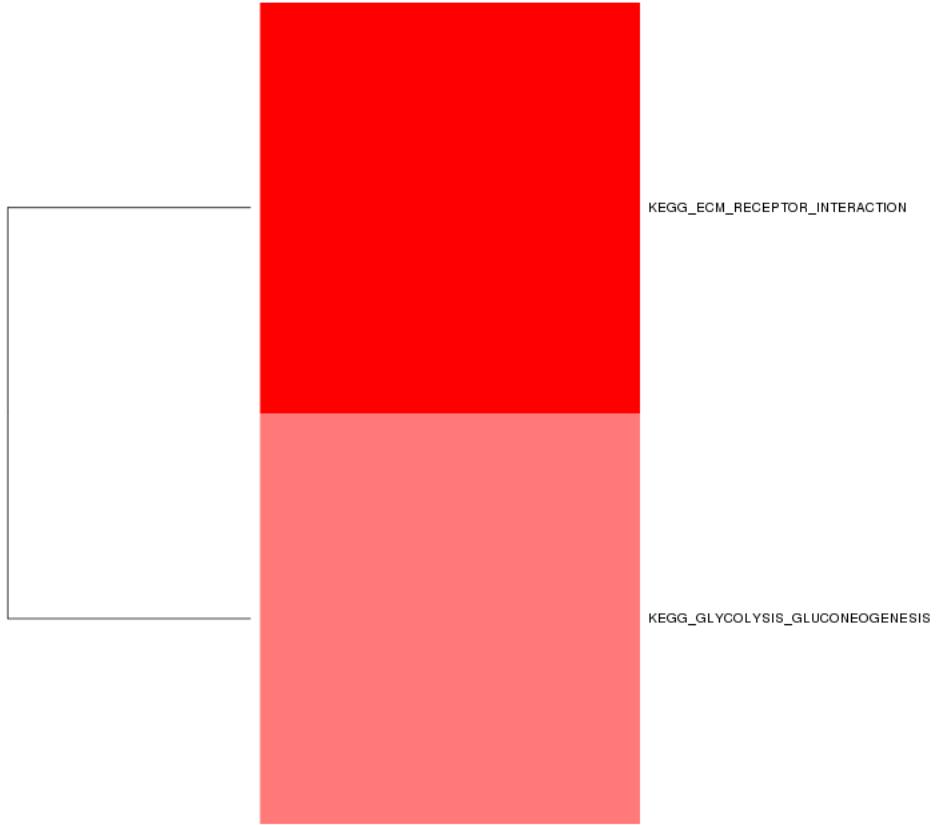
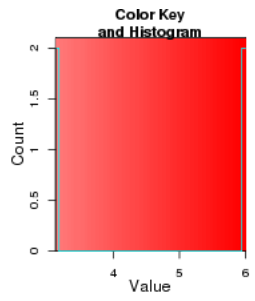


Reactome zscore:

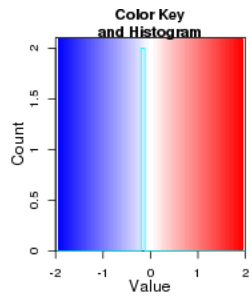


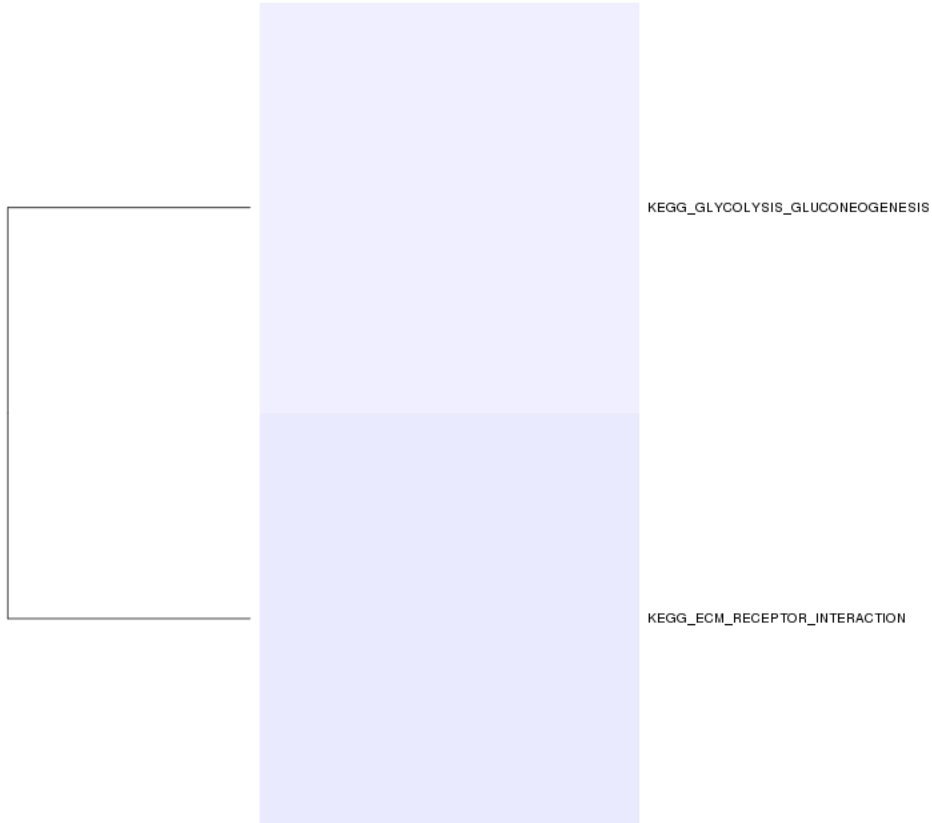


Kegg enrichment:



Kegg zscore:





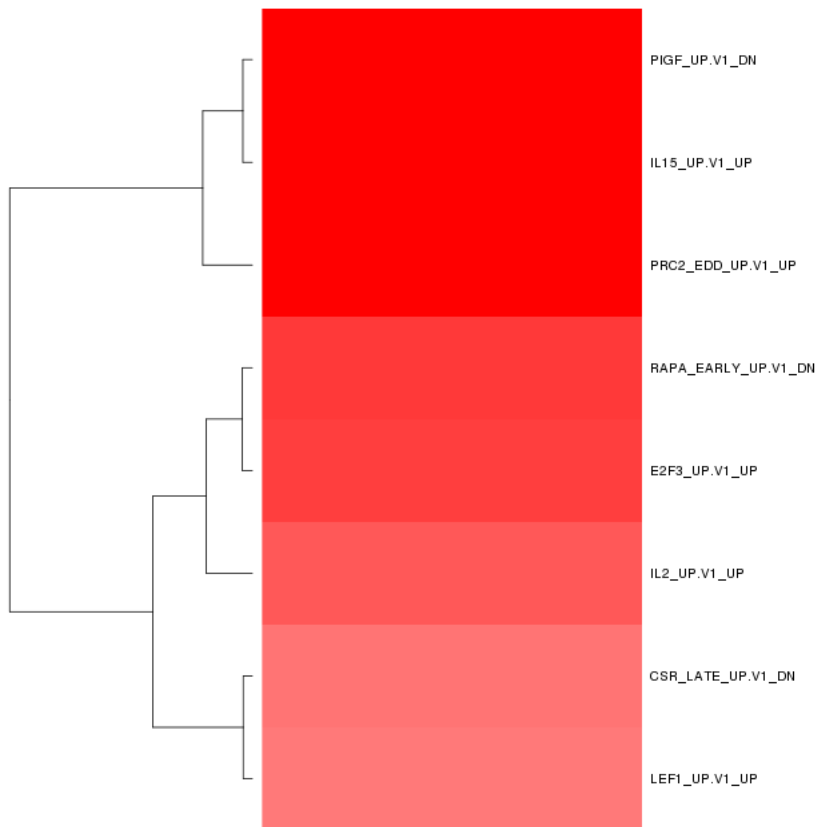
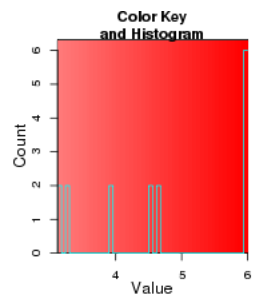
Interneuron enrichment:

```
## [1] "Not enough significant categories to print a heatmap!"
```

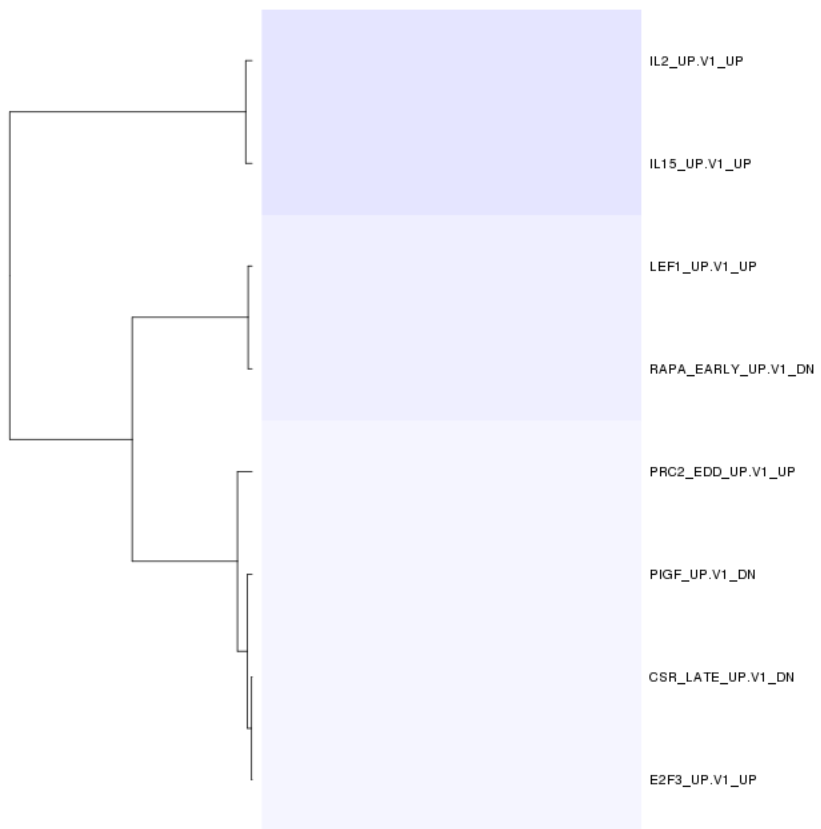
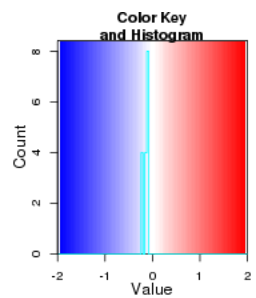
Interneuron zscore:

```
## [1] "Not enough significant categories to print a heatmap!"
```

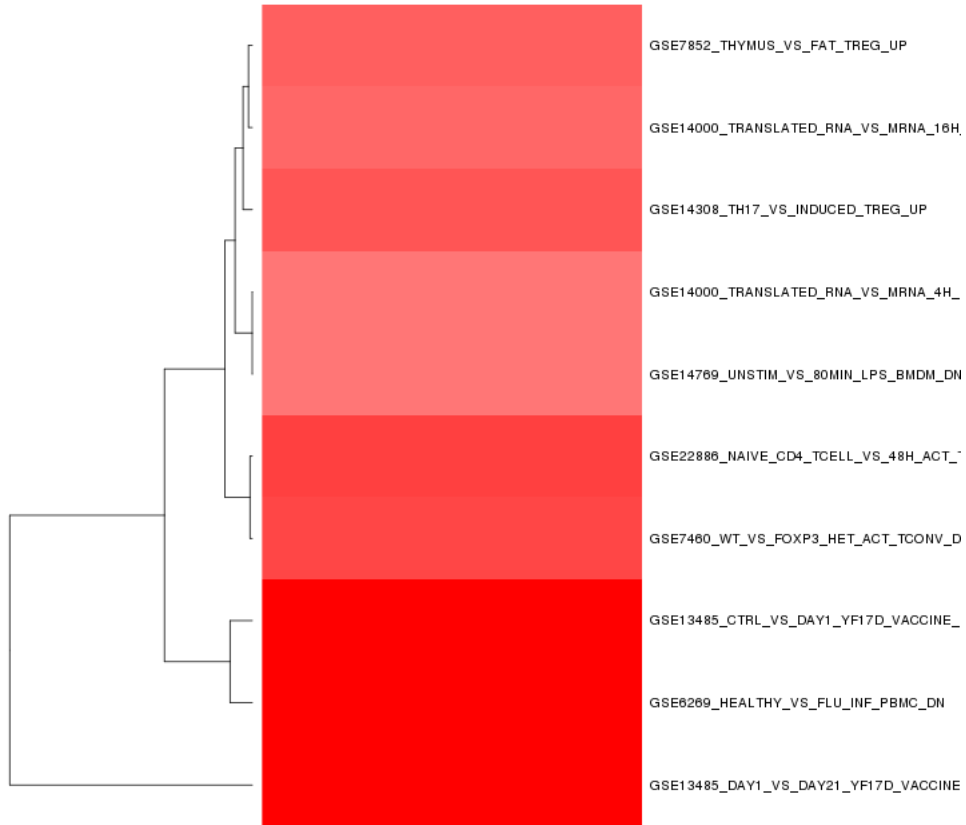
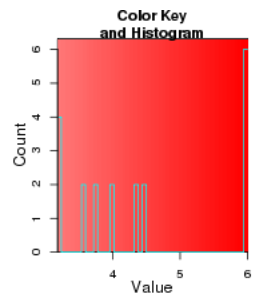
Oncogene enrichment:



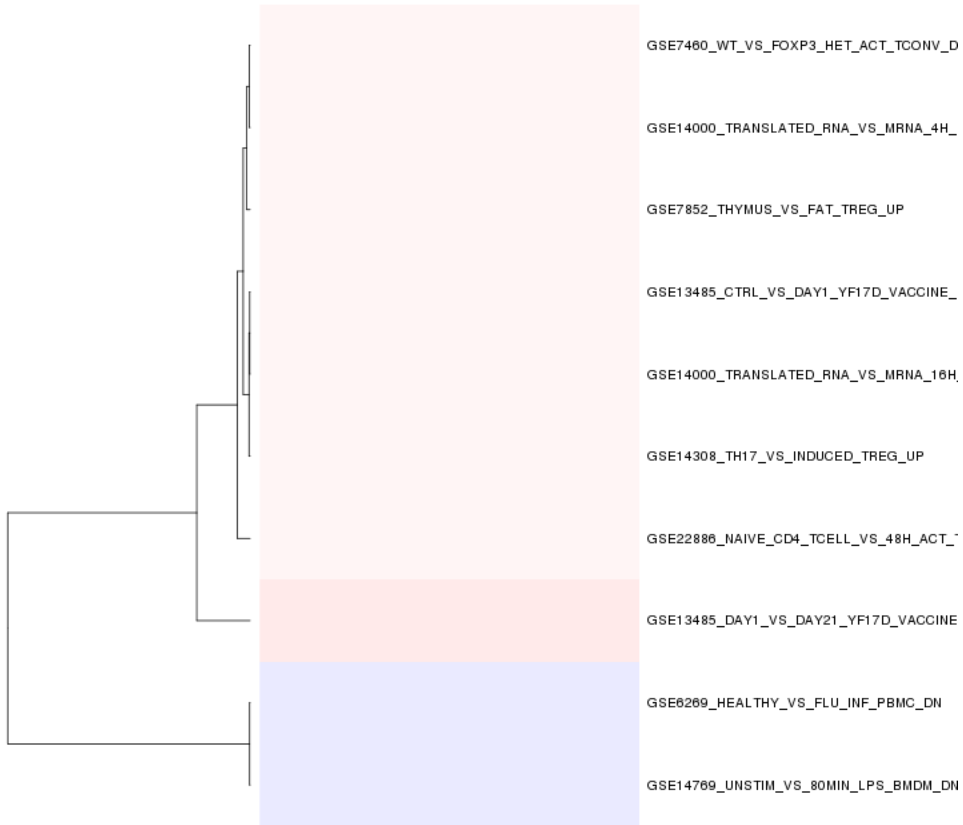
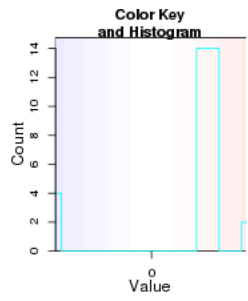
Oncogene zscore:



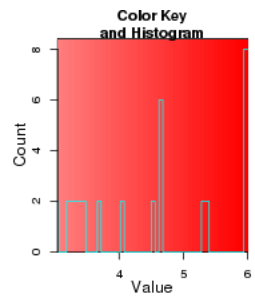
Immuno enrichment:

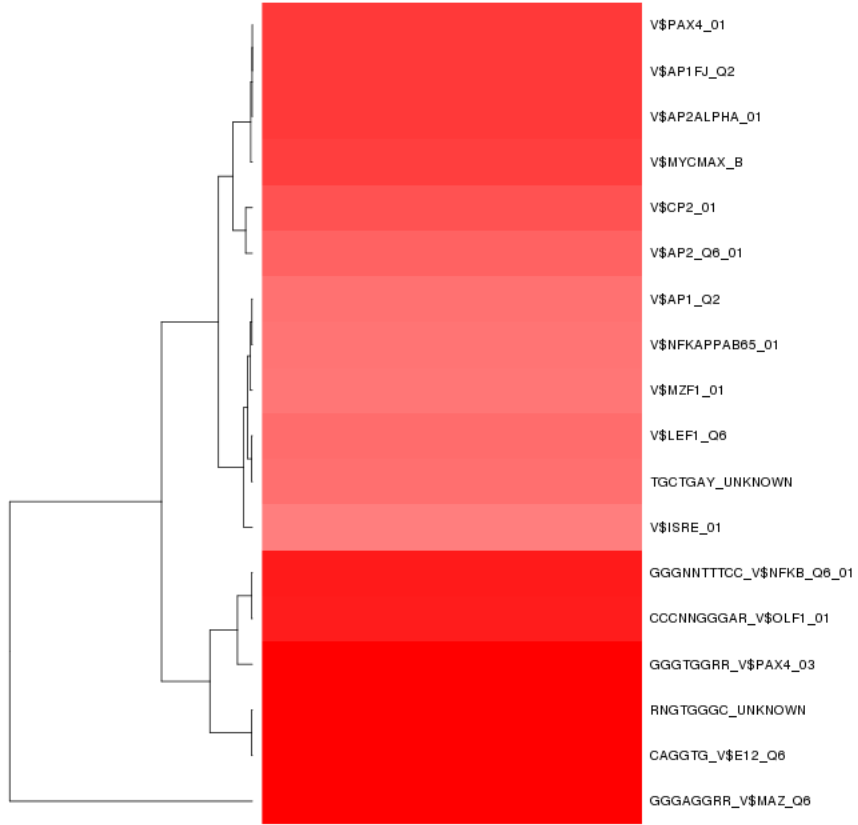


Immuno zscore:

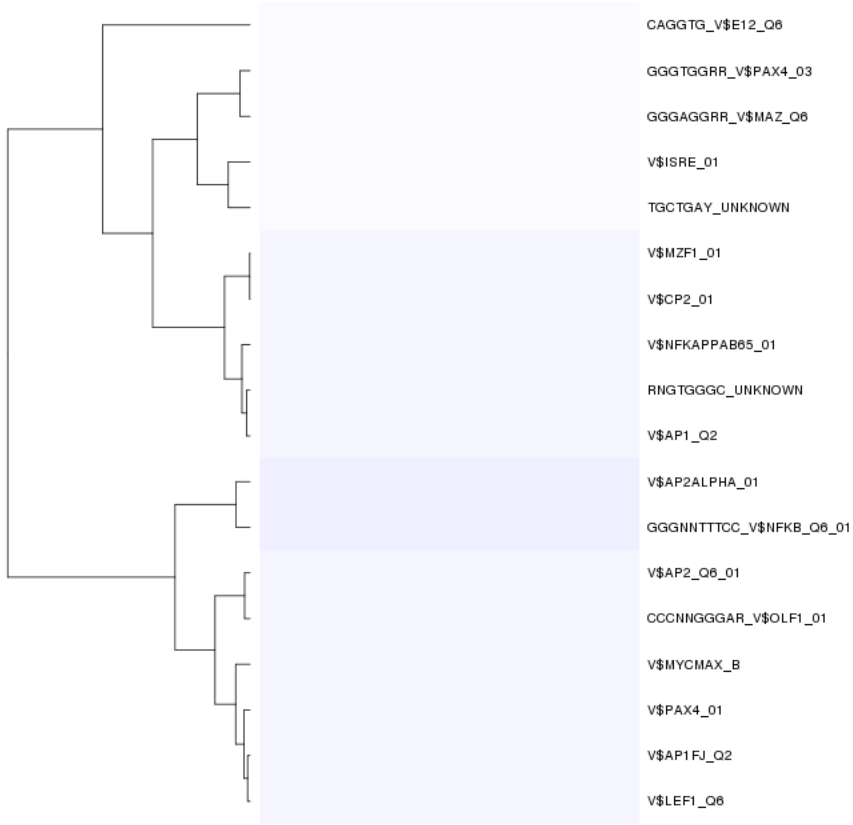
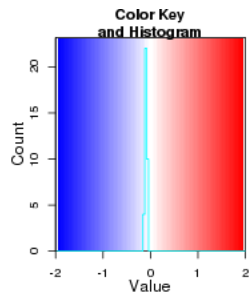


TF enrichment:





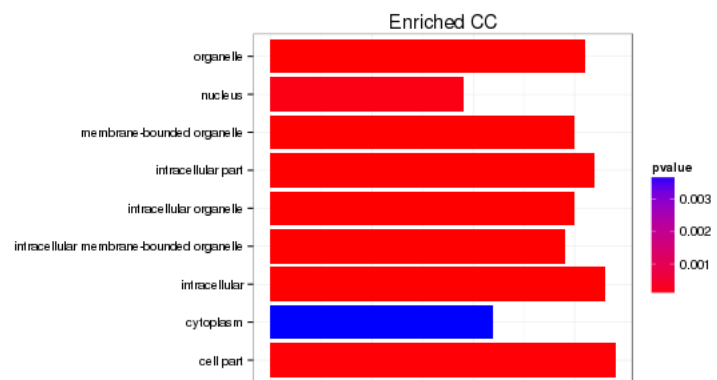
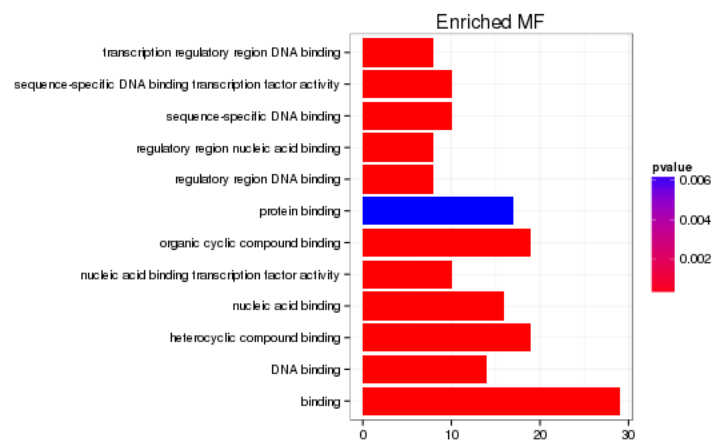
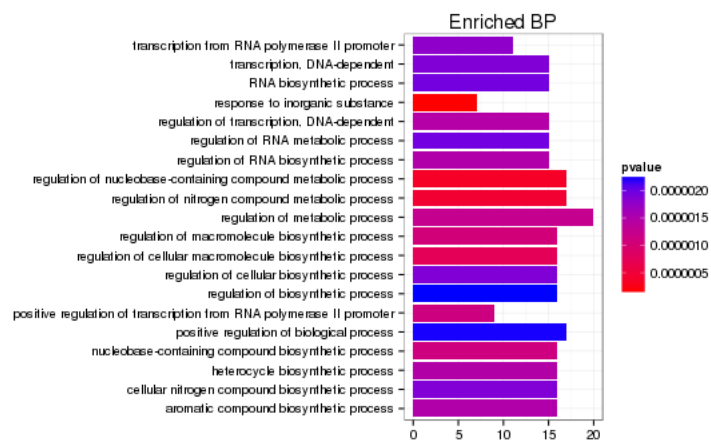
TF zscore:



GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```

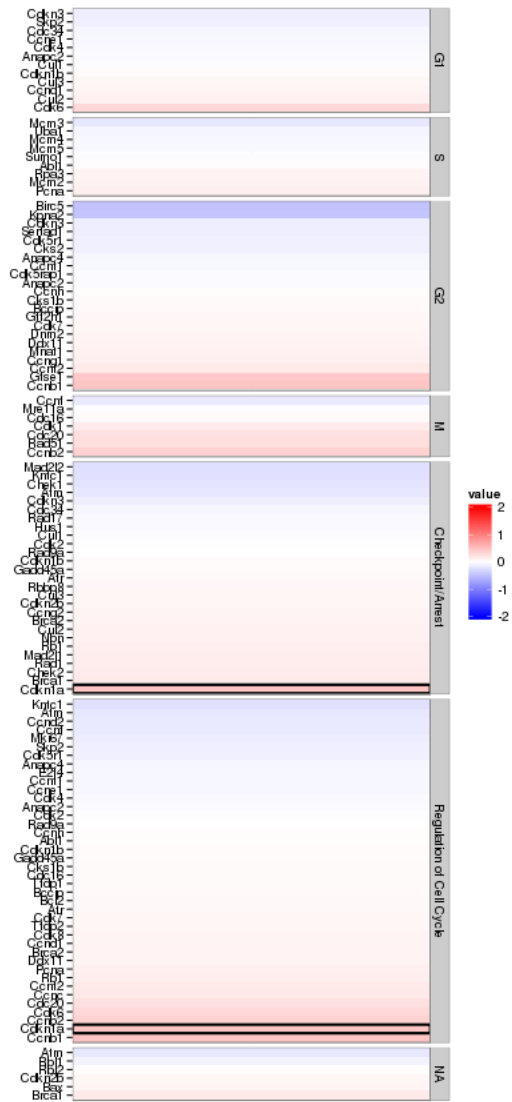




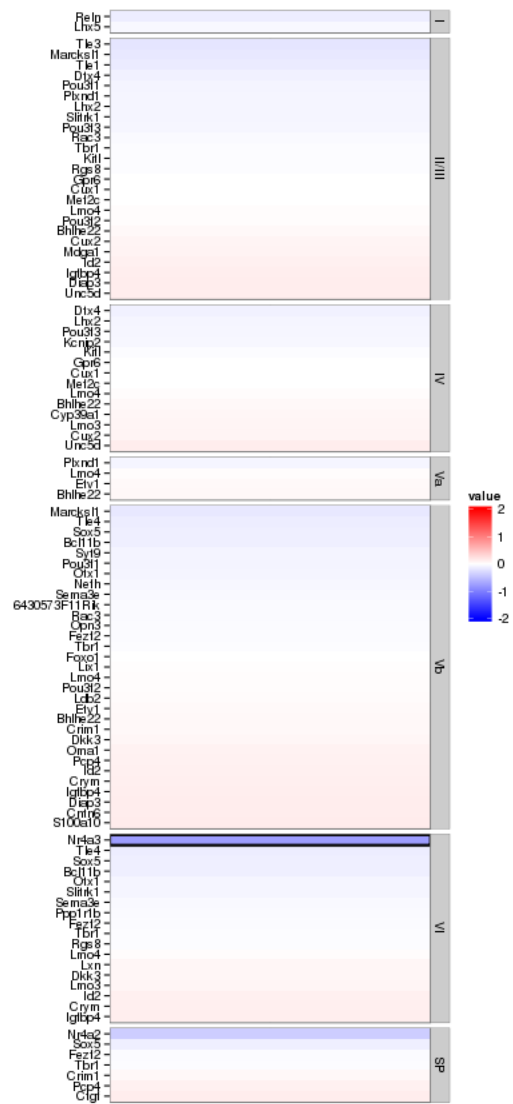
```
## Error: need finite 'xlim' values
```

```
## Error: 'x' and 'units' must have length > 0
```

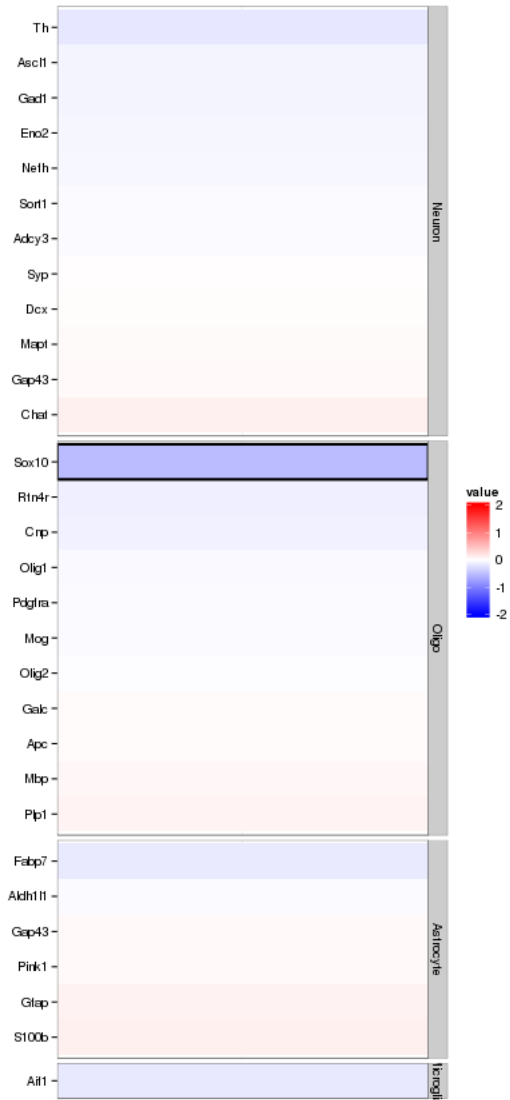
Enrichment or depletion for stage-specific cell cycle markers



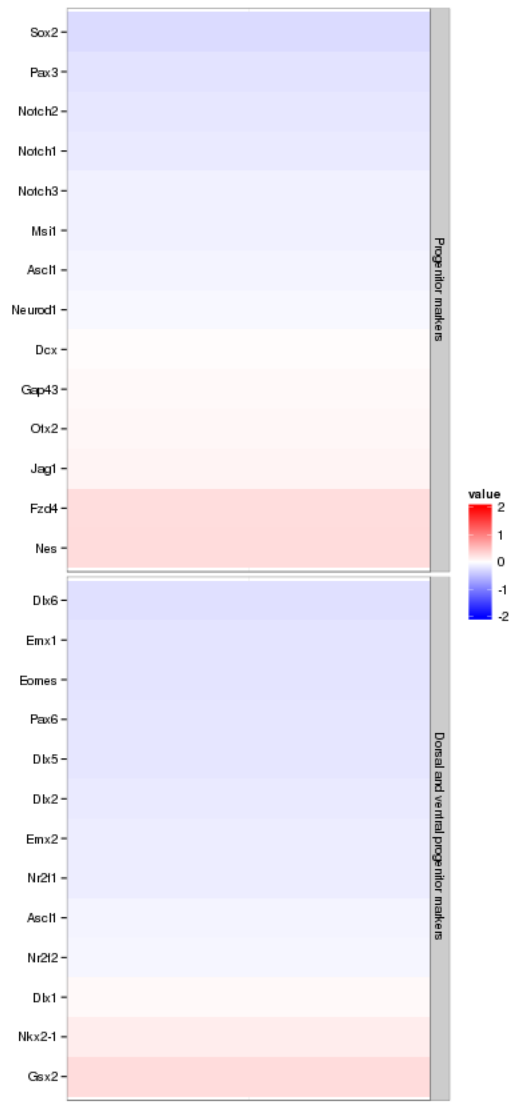
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



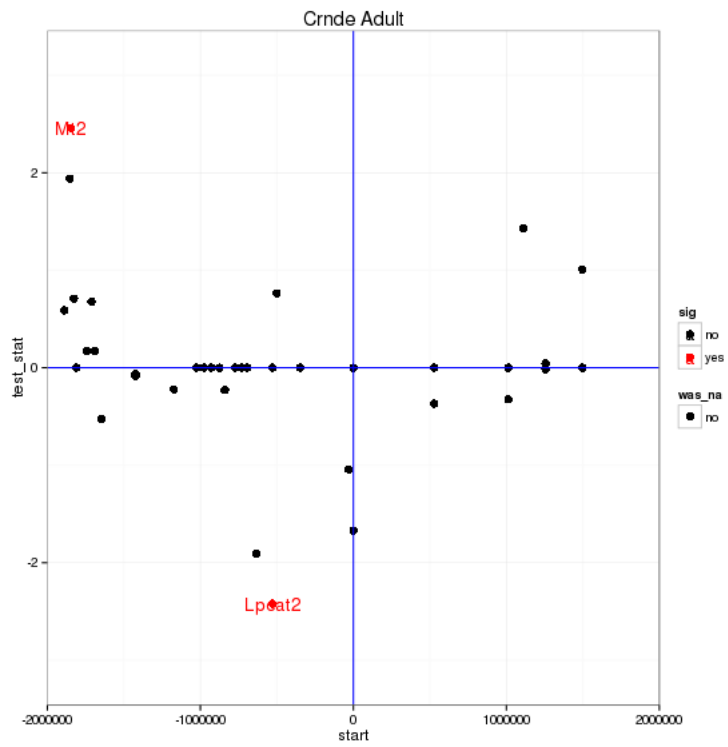
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 2 genes significantly regulated in a region this size is: 0.0189



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR741

16 JR784

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34590000.00	0.76	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34590000.00	0.59	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34590000.00	0.98	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34590000.00	0.80	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34590000.00	1.04	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34590000.00	1.07	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34590000.00	0.81	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34590000.00	1.16	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34590000.00	1.01	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34590000.00	1.30	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34590000.00	1.51	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34590000.00	1.02	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34590000.00	0.97	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34590000.00	1.21	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR741/abundances.cxb	Cmdc	0	Cmdc_0	37686800.00	34590000.00	1.09	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR784/abundances.cxb	Cmdc	1	Cmdc_1	37652200.00	34590000.00	1.09	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
```

```

## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33        Matrix_1.1-3        munSELL_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1        zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Crnde -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Crnde_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/ar
## 2
## 3
## 4
## 5
```

Crnde KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Crnde.

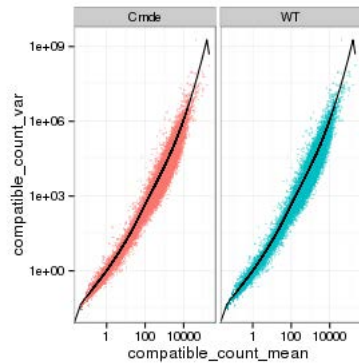
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

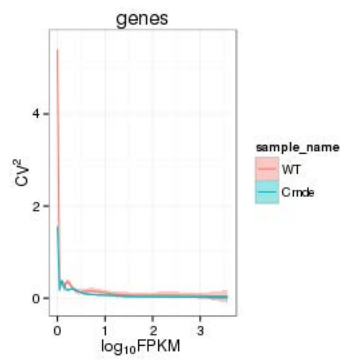
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



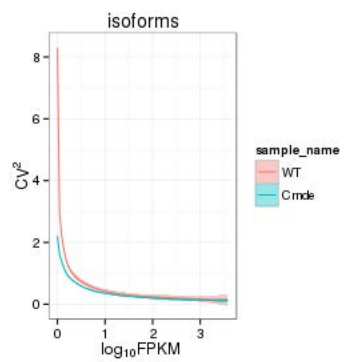
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

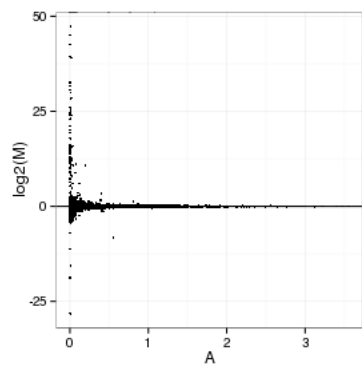
Genes:



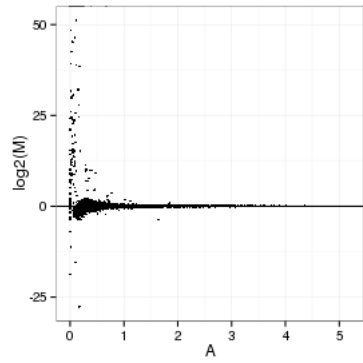
Isoforms:



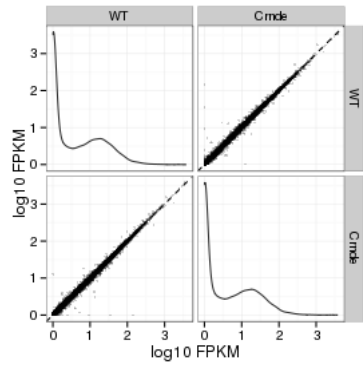
MvA plot



MvA plot counts



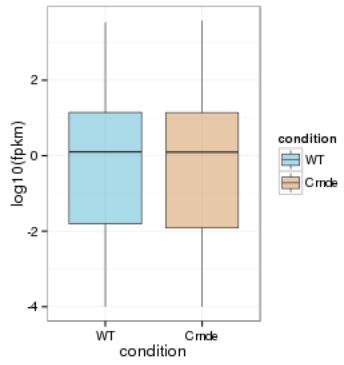
Scatterplot matrix



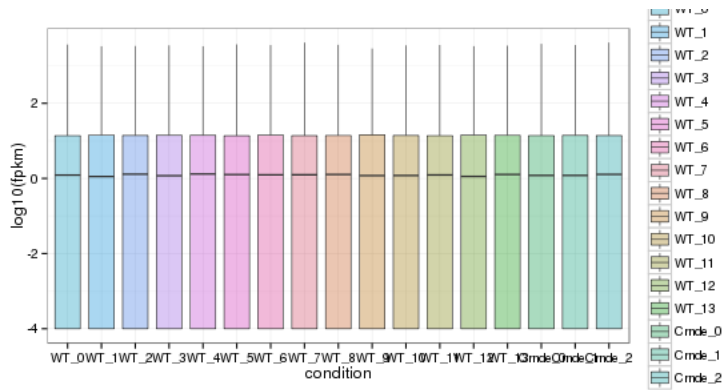
Distributions

Boxplots

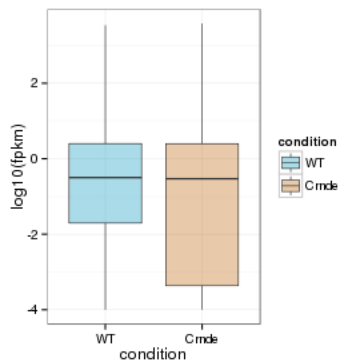
Boxplot (genes)



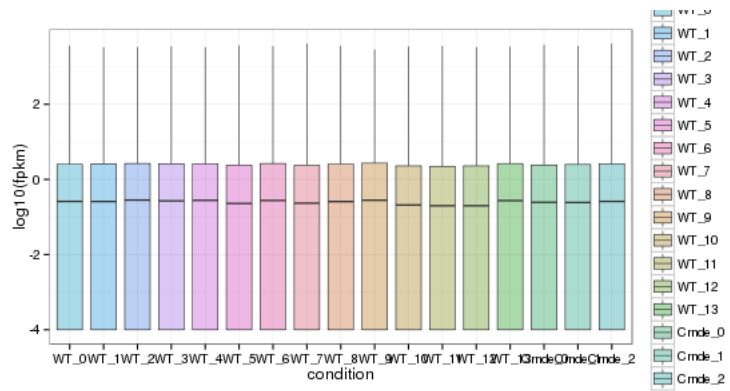
Boxplot (genes, replicates)



Boxplot (isoforms)

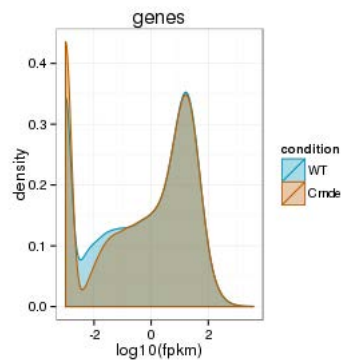


Boxplot (isoforms, replicates)

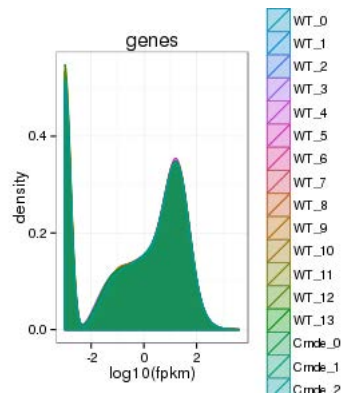


Density

Density (genes)

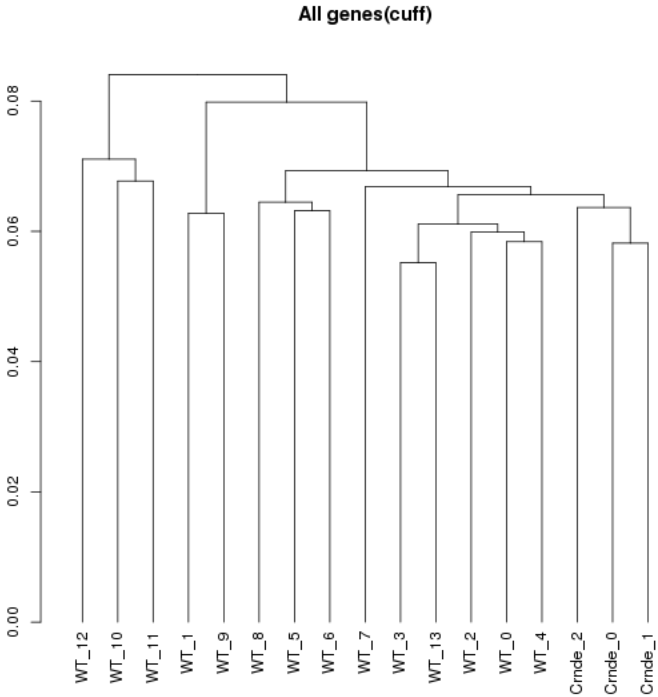


Density (genes, replicates)



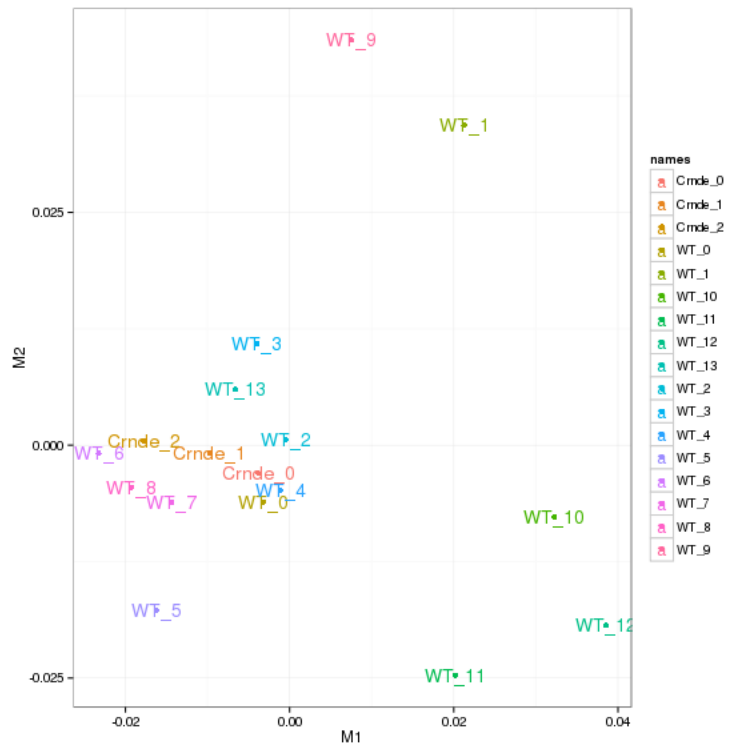
Clustering

Replicate Clusters



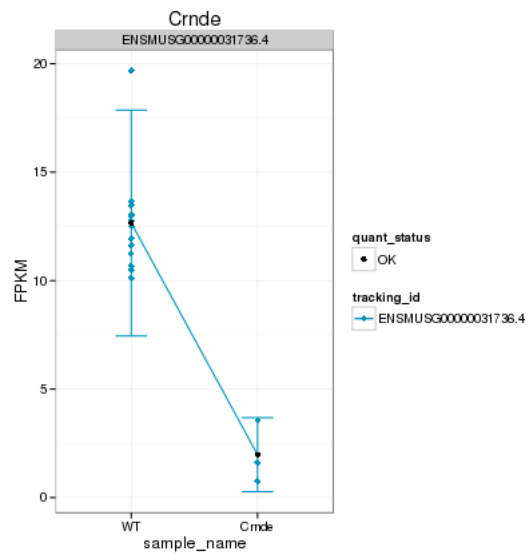
NULL

PCA (genes)

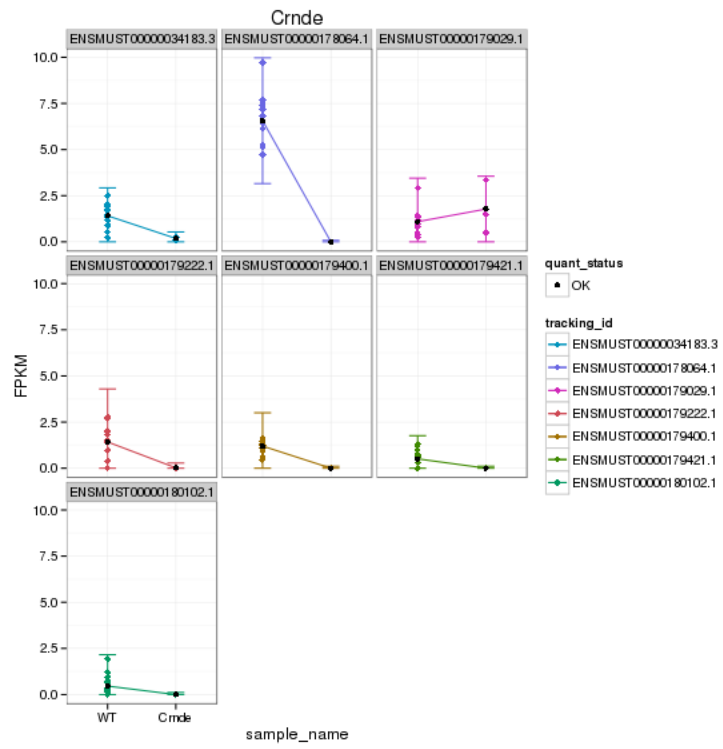


KO assessment

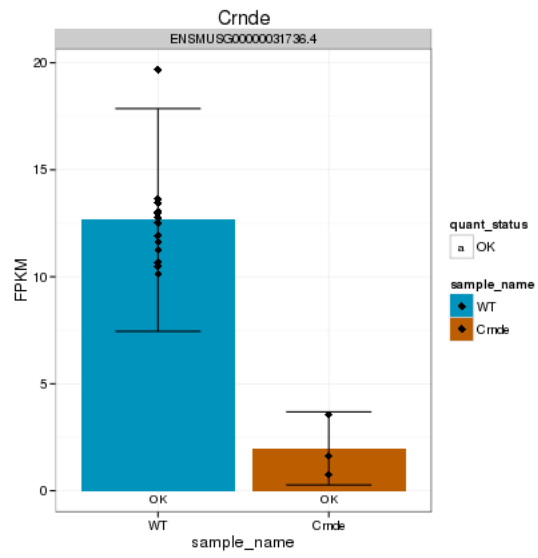
Endogenous lncRNA expression



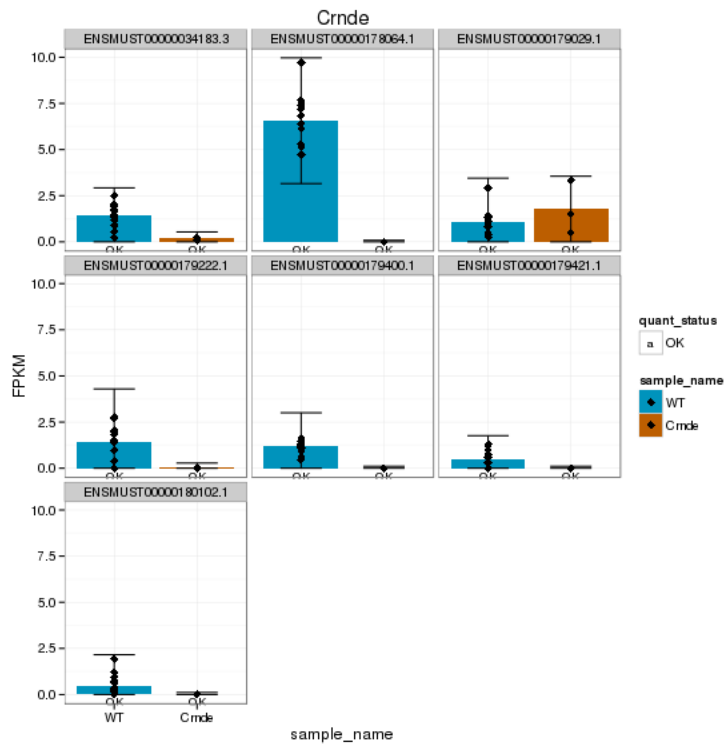
Endogenous expression of Crnde isoforms:



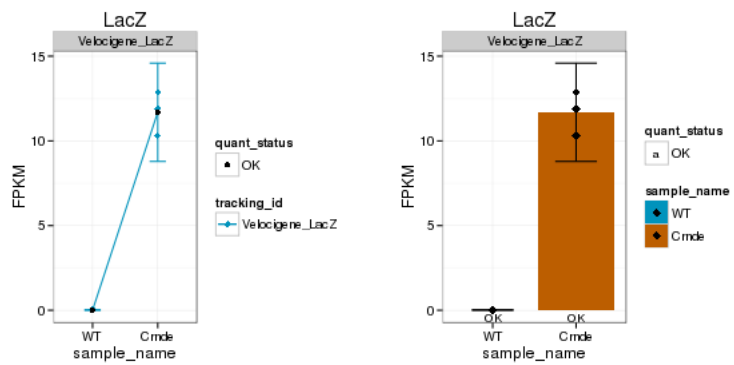
Barplot of gene expression:



Barplot of isoform expression:

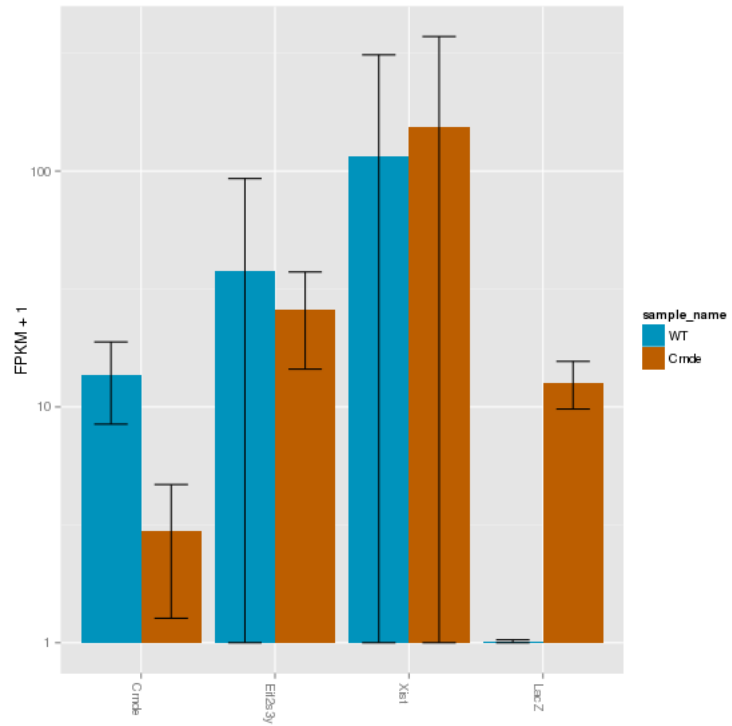


LacZ expression

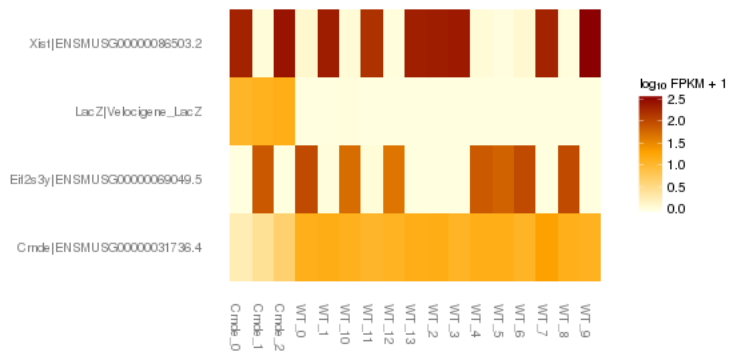


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



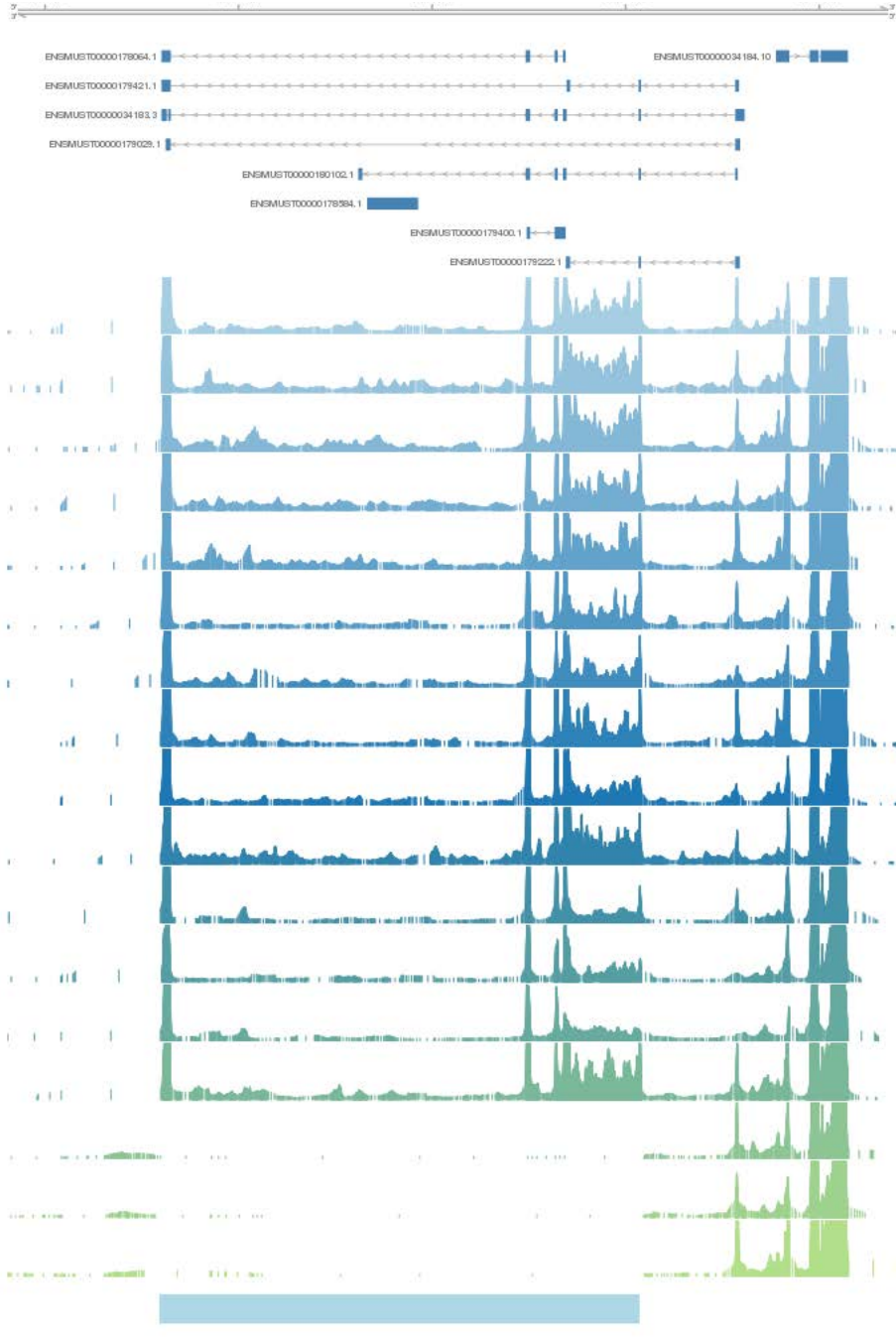
Expression heatmap:



Track visualization



NCBI RefSeq LincRNA isoforms WT_0 WT_1 WT_2 WT_3 WT_4 WT_5 WT_6 WT_7 WT_8 WT_9 WT_10 WT_11 WT_12 WT_13 Cntrl_0 Cntrl_1 Cntrl_2 Cntrl_3 KO Region



Differential Analysis

Differential Genes

There are 80 significantly differentially expressed genes. They are:

geneAnnot\$gene_short_name

- 1 Col6a1
- 2 Col18a1
- 3 Col1a1
- 4 Lamb1
- 5 Nes
- 6 Polr2a
- 7 Nid1
- 8 Rplp1
- 9 Hspa8
- 10 Sulf1
- 11 Dync1h1
- 12 Lama2
- 13 Col6a2
- 14 Mrc2
- 15 Prpf8
- 16 Ntn1
- 17 Iqgap2
- 18 Nid2
- 19 Myh9
- 20 Igf2r
- 21 Lox
- 22 Fbn2
- 23 Rps14
- 24 Pdgfrb
- 25 Rps26
- 26 Col5a2
- 27 Col3a1
- 28 Fn1
- 29 Lamc1
- 30 Col5a1
- 31 Lamc3
- 32 Notch1
- 33 Lrp2
- 34 Fbn1
- 35 Lrp4
- 36 Notch2
- 37 Tnc

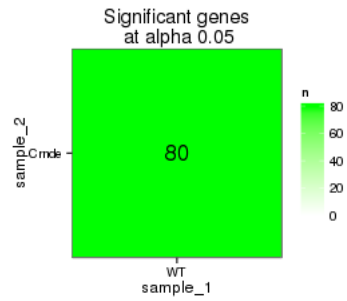
38 Slep1
39 Sorcs2
40 Col1a2
41 Aldh1l1
42 Emp1
43 Dera

44 Hddc3
45 Col4a5
46 Col4a1
47 Cmde
48 Itga11
49 Lama1
50 Fras1
51 Lum
52 Wnt8b
53 Frem2
54 Notch3
55 Spon1
56 Neurod2
57 Satb2
58 Atp5l
59 Bend6
60 Gm9493
61 Acp1
62 Mc4r
63 Gm9846
64 Hbb-y
65 Hbb-bh1
66 Lamb2
67 Hba-x
68 Capn11
69 Alox5ap
70 Dcc

71 Rps17
72 Rpl35
73 Scoc
74 Rpl36-ps3
75 3110035E14Rik
76 Flnc
77 Ptpnz1
78 Fat1
79 Gm26924
80 Gm19980

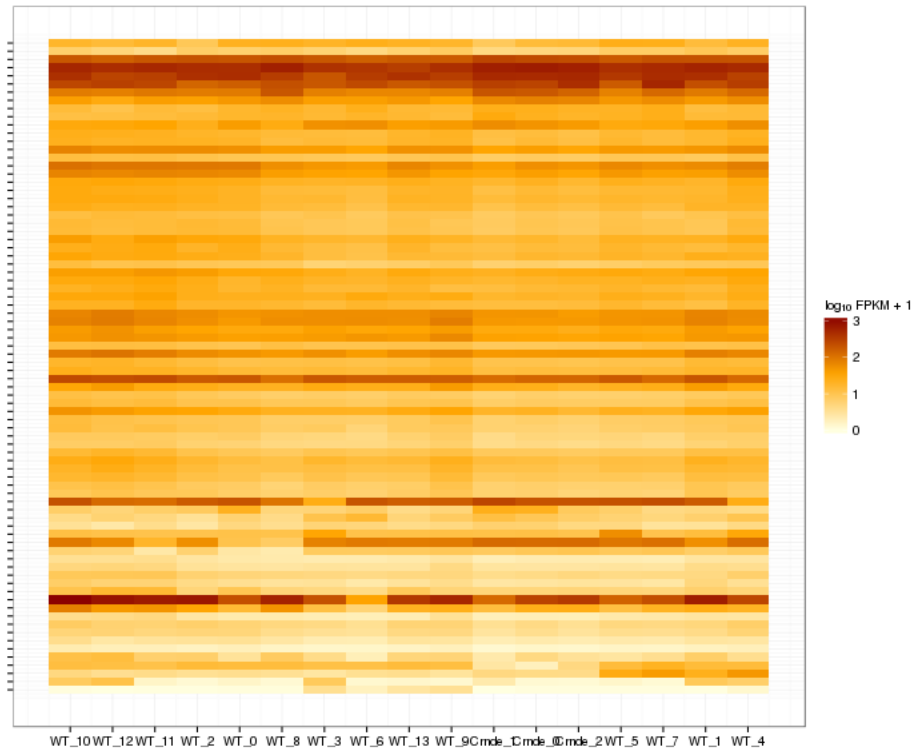
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

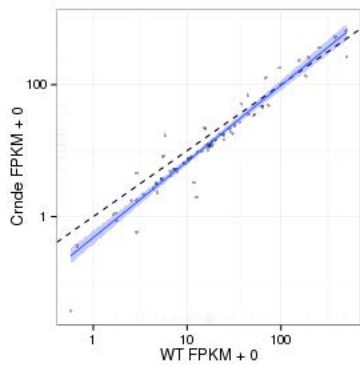


Significant genes with expression >50fpm (any condition):(turned off)

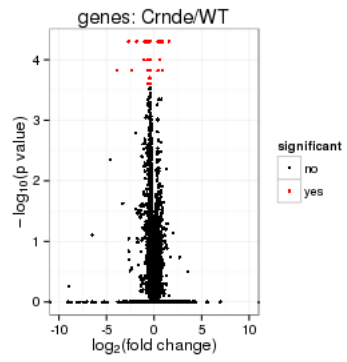
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

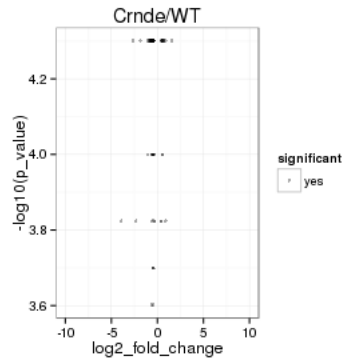
Scatter plot of significant genes only:



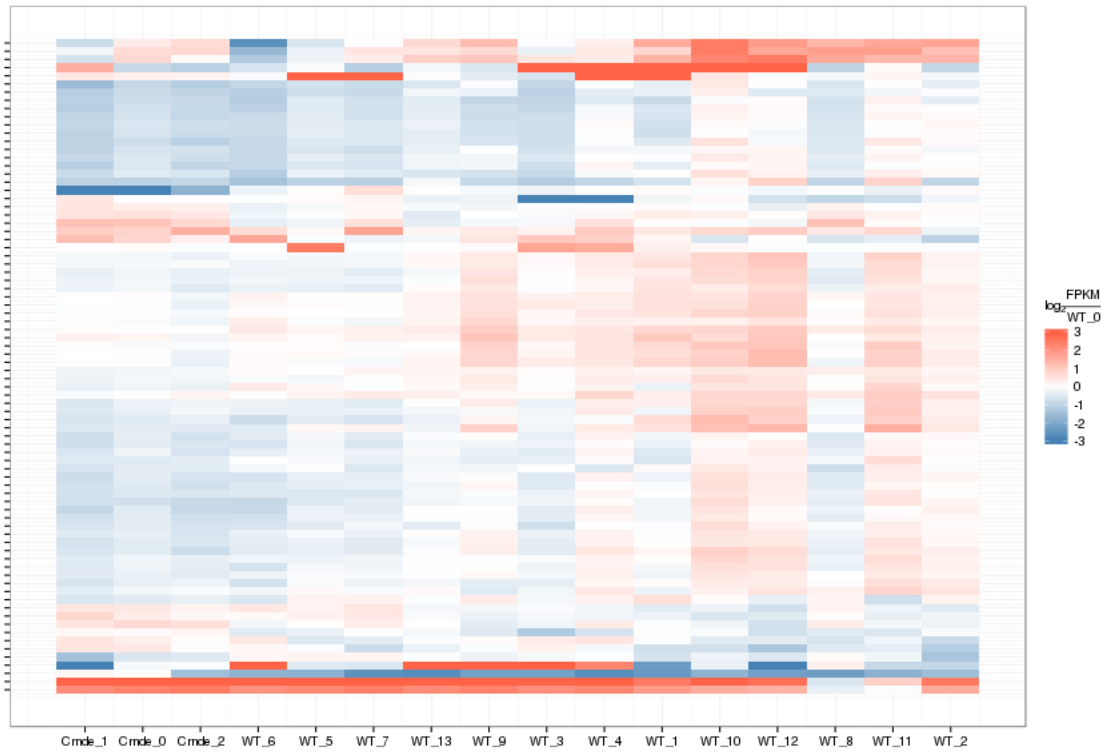
Volcano Plot



Volcano plot with significant genes only:



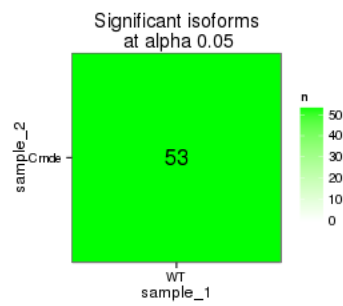
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:

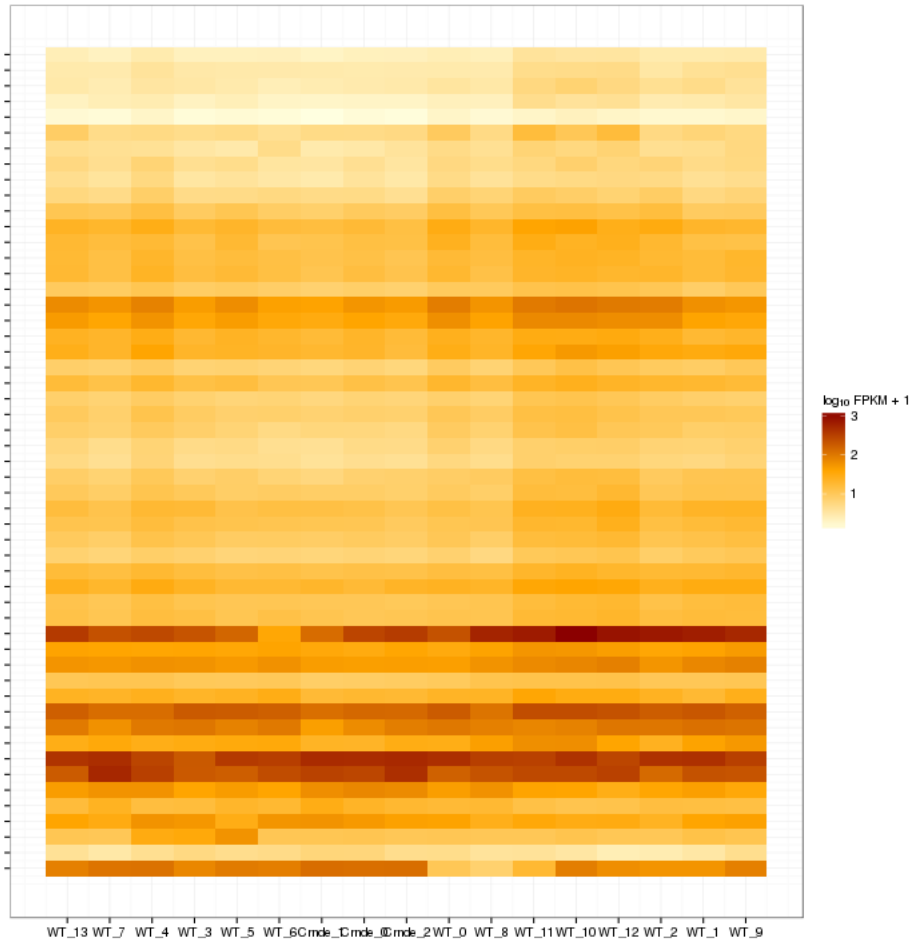


These isoforms are:

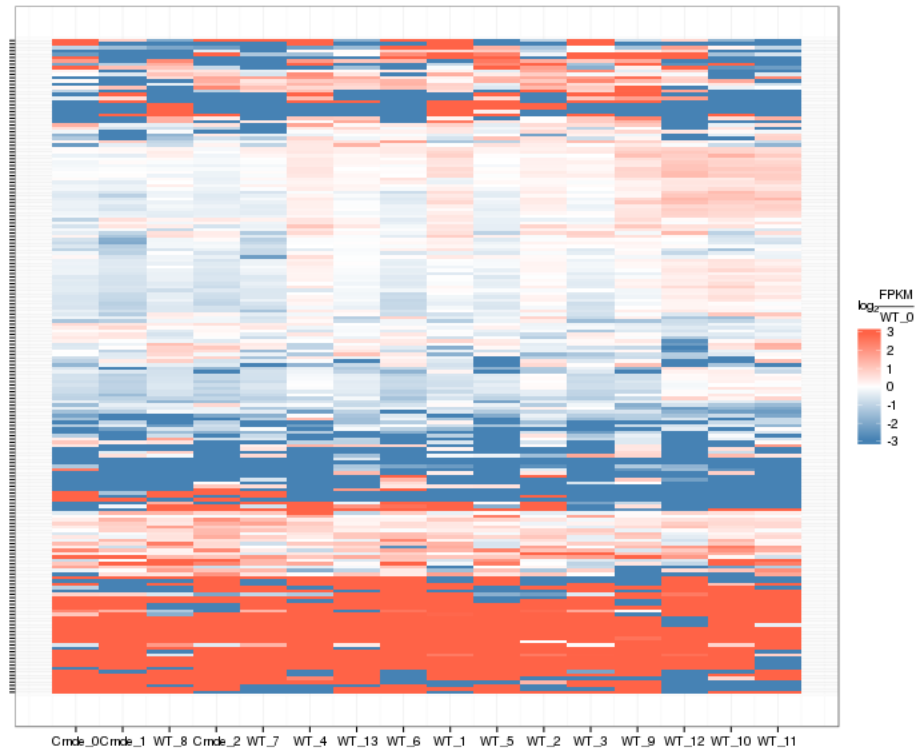
isoAnnot	gene_short_name
1	Col6a1
2	Col1a1
3	Lamb1
4	Nid1
5	Rplp1
6	Cad
7	Dync1h1
8	Lama2
9	Mrc2
10	Myh9
11	Col2a1
12	Igf2r
13	Fbn2
14	Rps14
15	Pdgfrb
16	Col5a2
17	Col3a1
18	Fn1
19	Lamc1
20	Col5a1
21	Lamc3
22	Notch1
23	Lrp2
24	Fbn1
25	Lrp4
26	Notch2
27	Tnc
28	Svep1
29	Sorcs2
30	Col1a2
31	Emp1
32	Dera
33	Hddc3
34	Lama1
35	Fras1
36	Wnt8b
37	Frem2
38	Notch3
39	Neurod2
40	Lrp1

- 41 Reln
- 42 Gm9493
- 43 Hjurp
- 44 Mc4r
- 45 Col6a3
- 46 Hbb-y
- 47 Lamb2
- 48 Rpl36-ps3
- 49 3110035E14Rik
- 50 Gpr98
- 51 Fat1
- 52 Col22a1
- 53 Gm26924

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

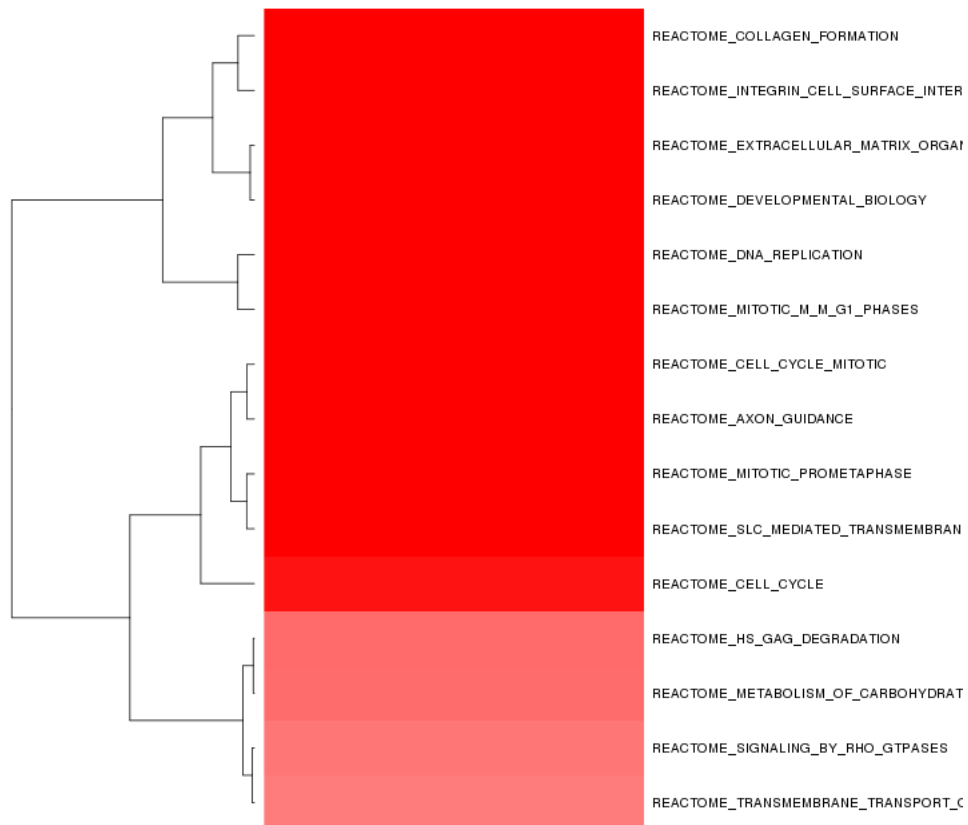
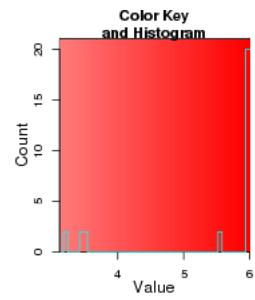
Biocarta enrichment:

```
## [1] "Not enough significant categories to print a heatmap!"
```

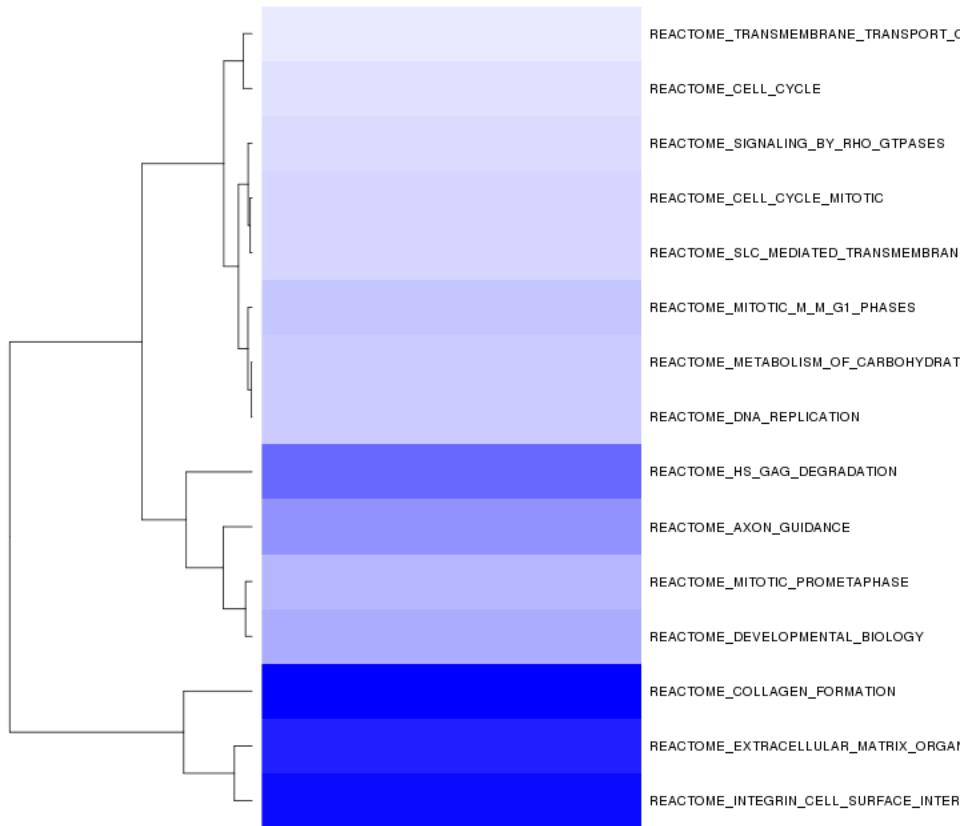
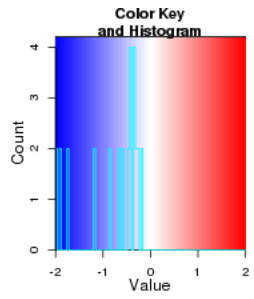
Biocarta zscore:

```
## [1] "Not enough significant categories to print a heatmap!"
```

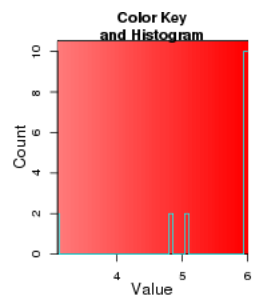
Reactome enrichment:

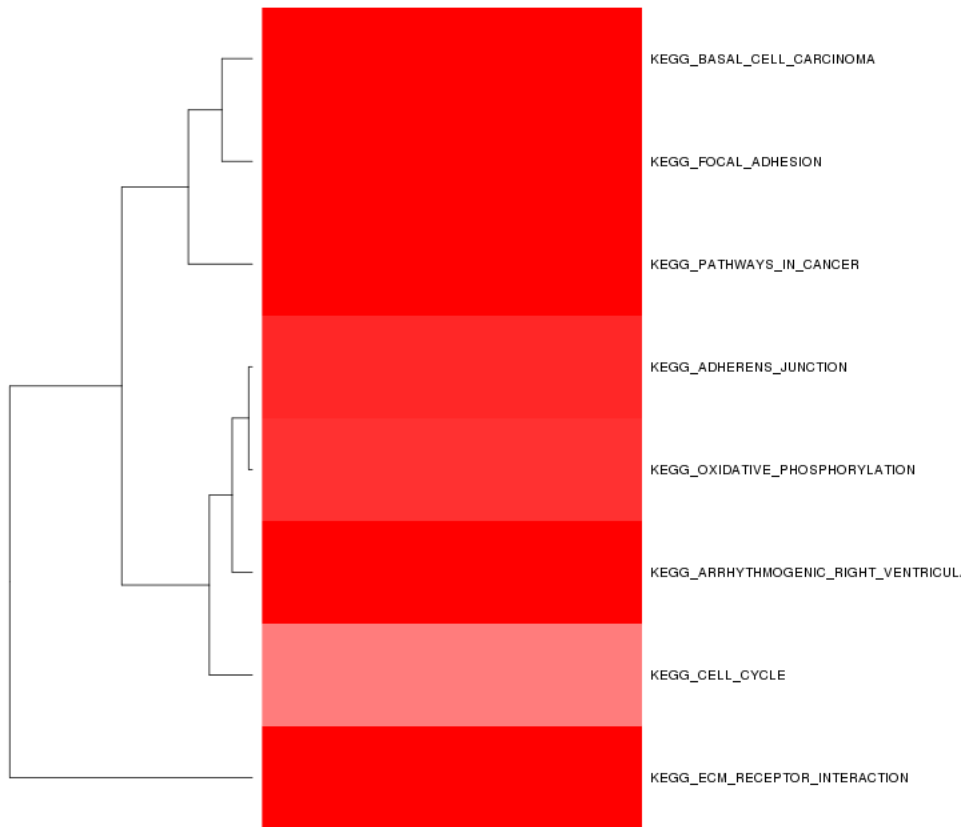


Reactome zscore:

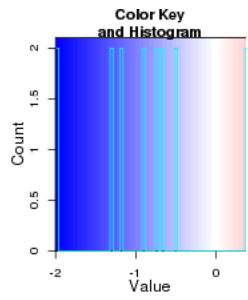


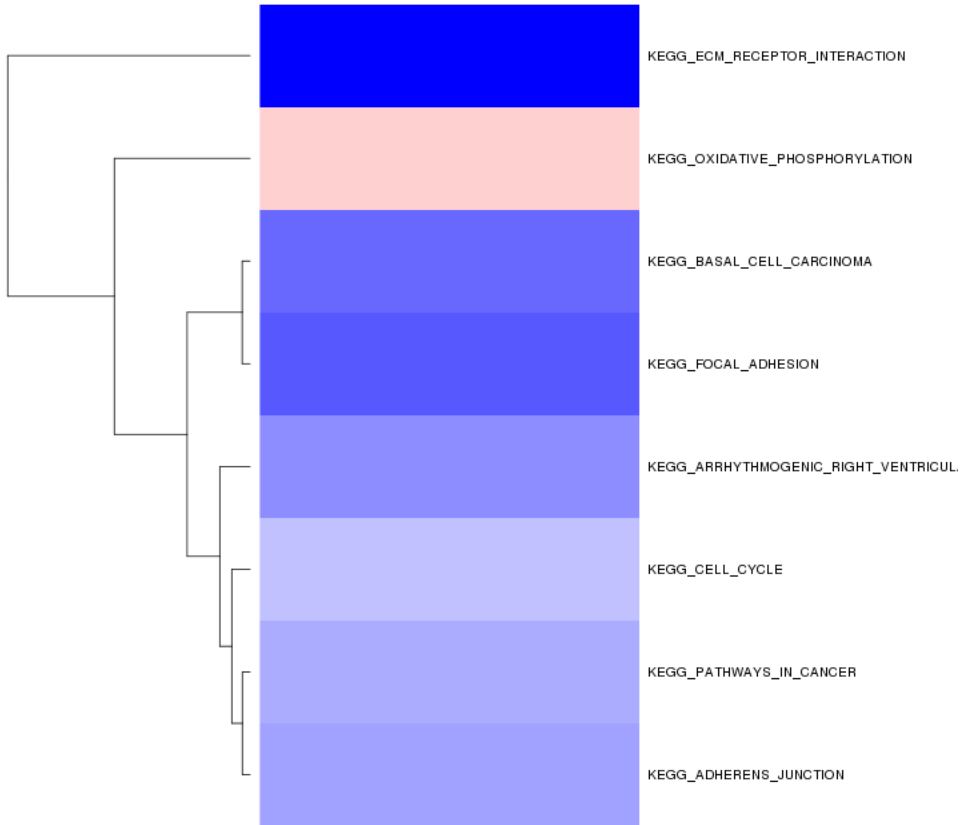
Kegg enrichment:





Kegg zscore:





Interneuron enrichment:

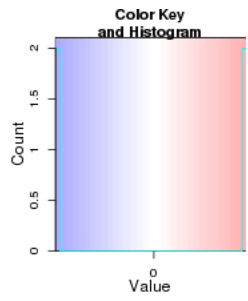
Error: no locations are finite

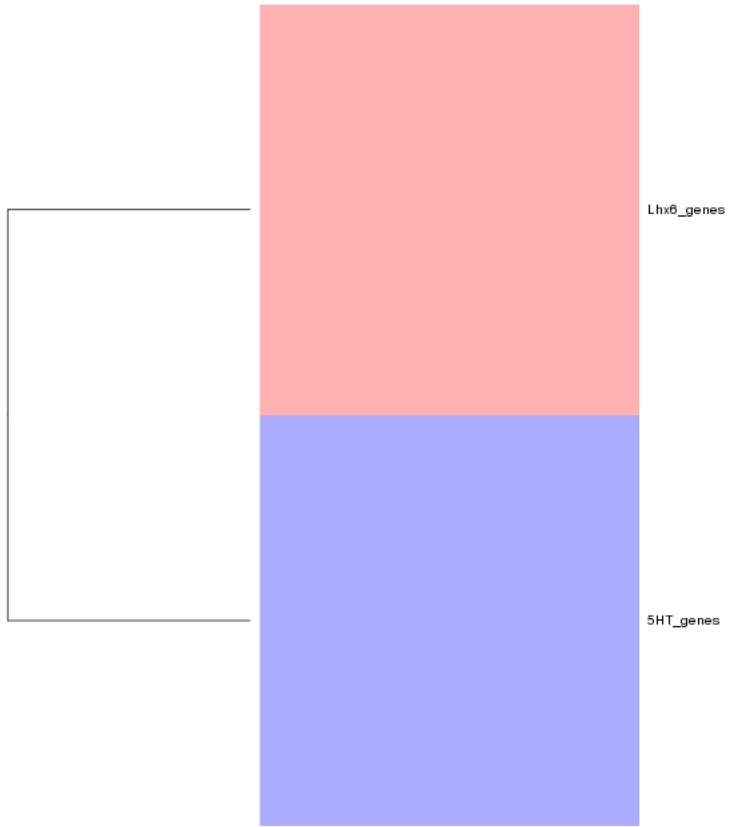


Lhx6_genes

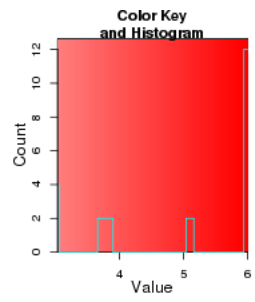
5HT_genes

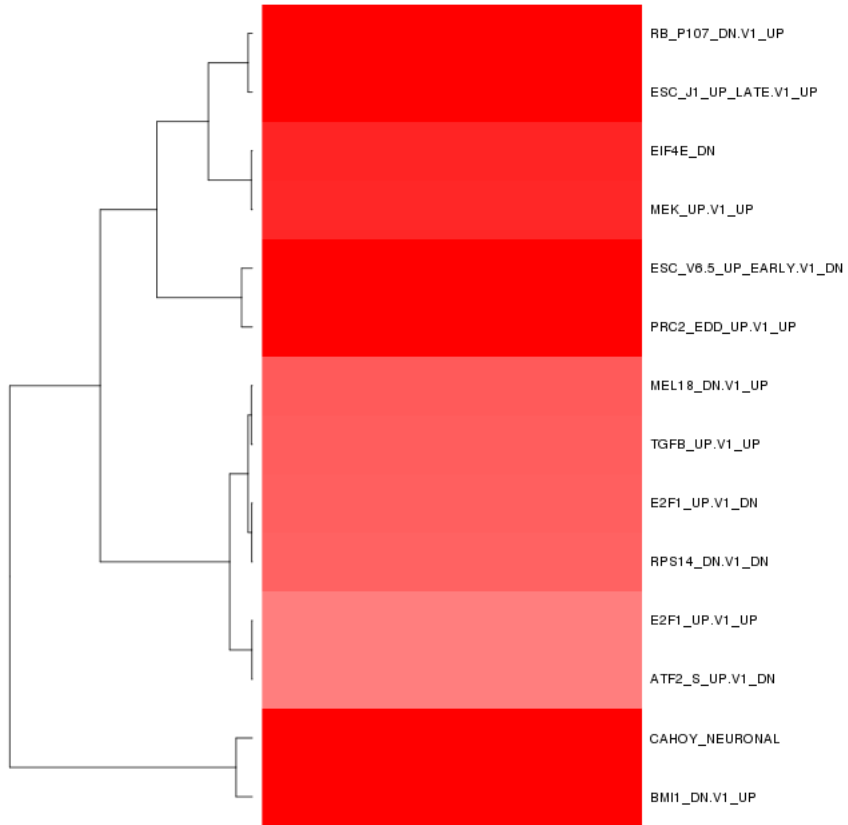
Interneuron zscore:



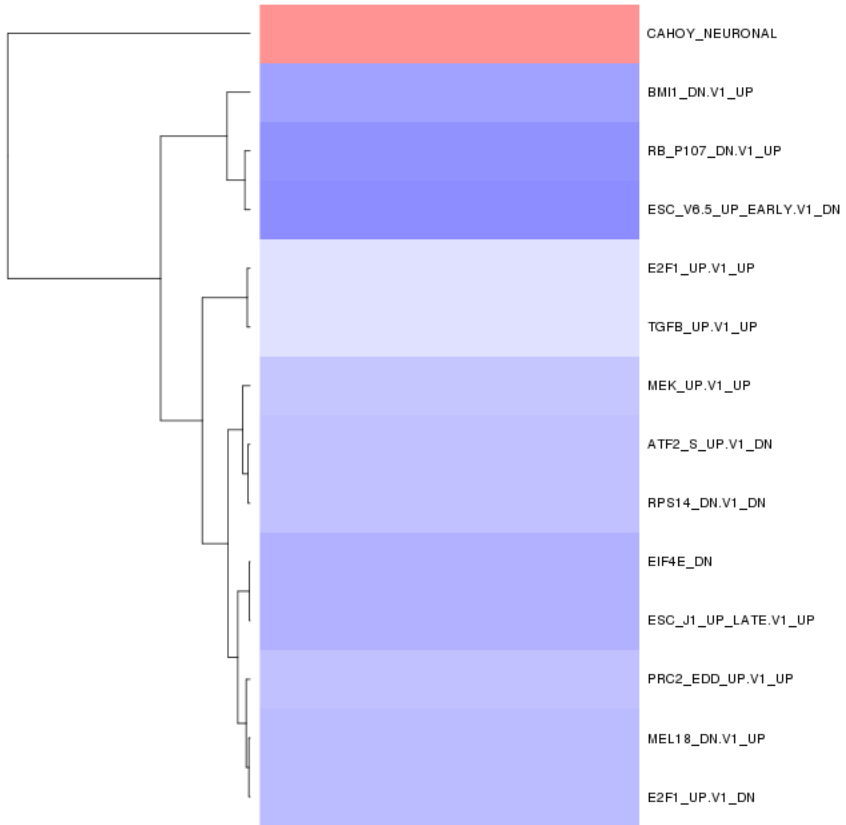
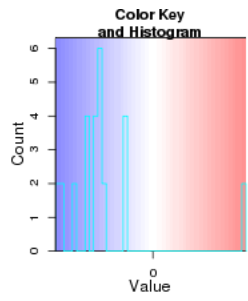


Oncogene enrichment:

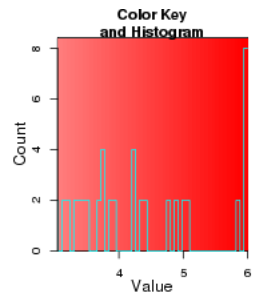


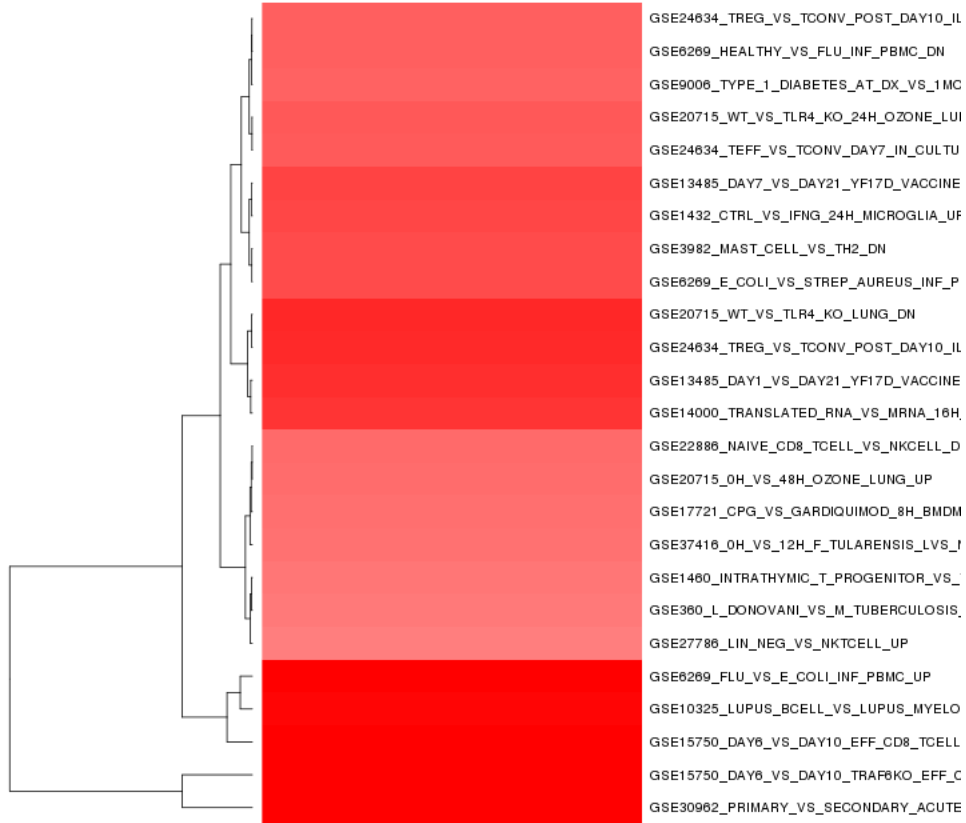


Oncogene zscore:

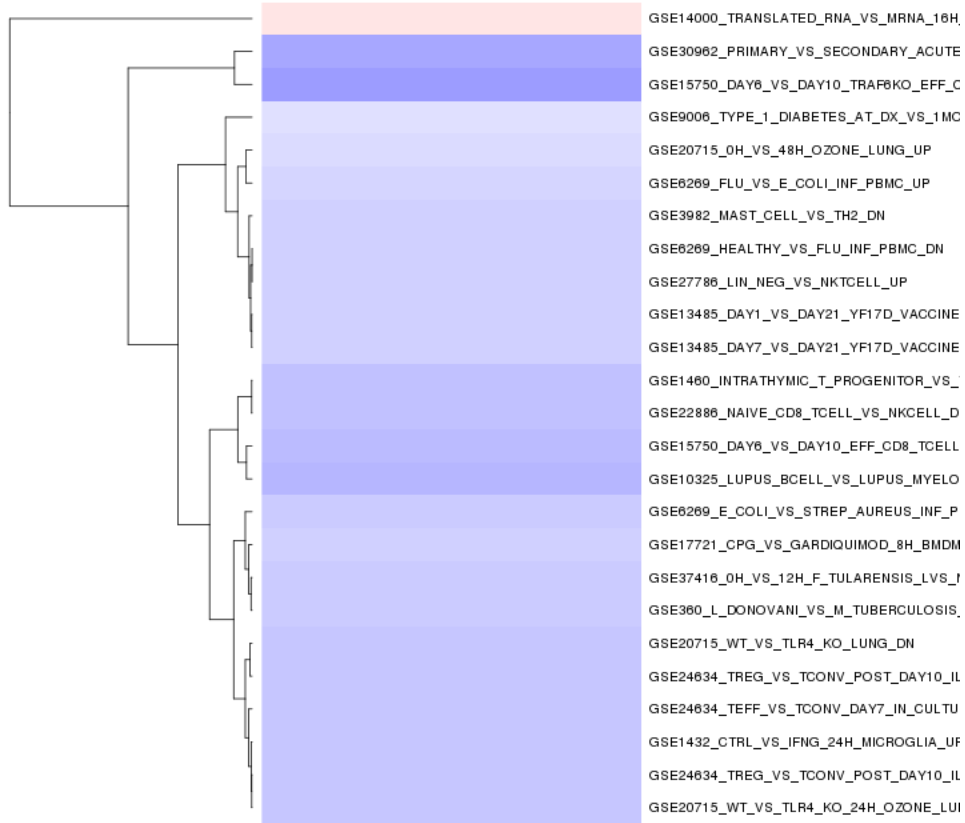
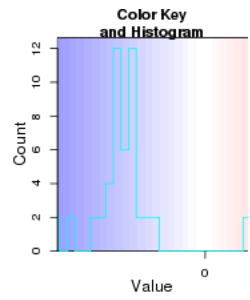


Immuno enrichment:

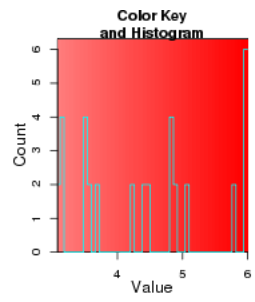


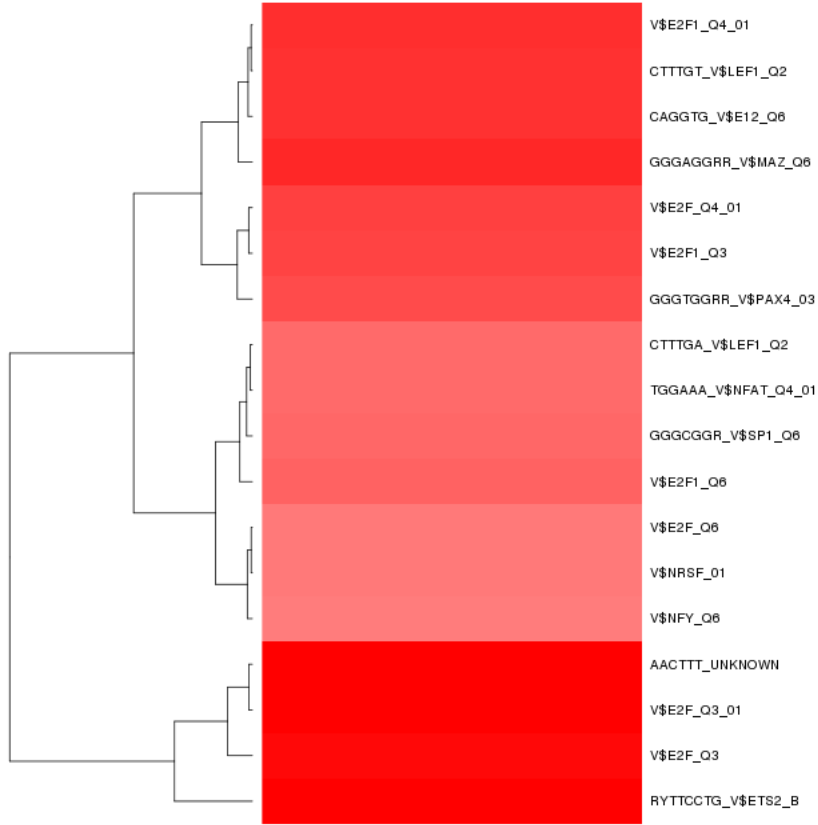


Immuno zscore:

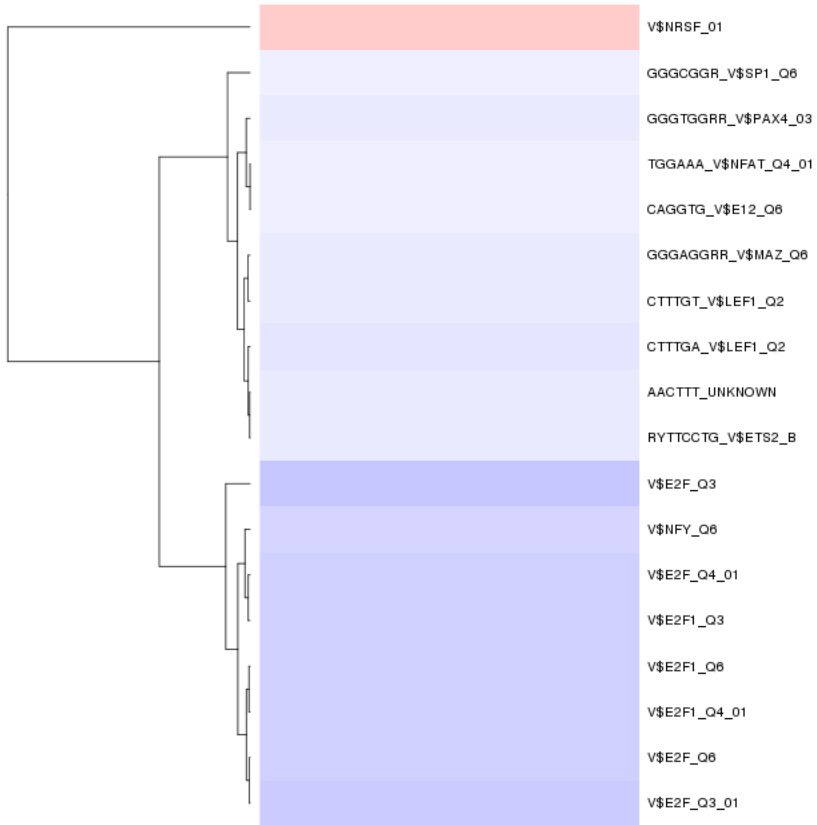
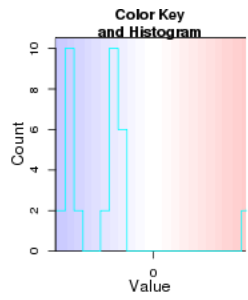


TF enrichment:





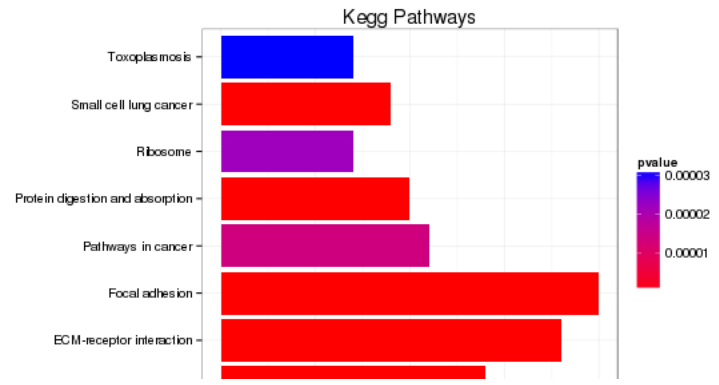
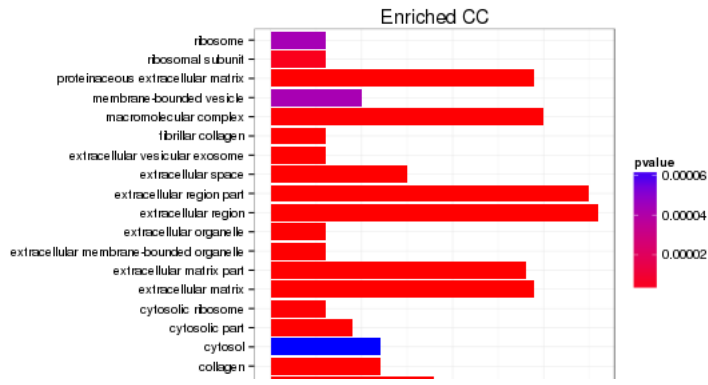
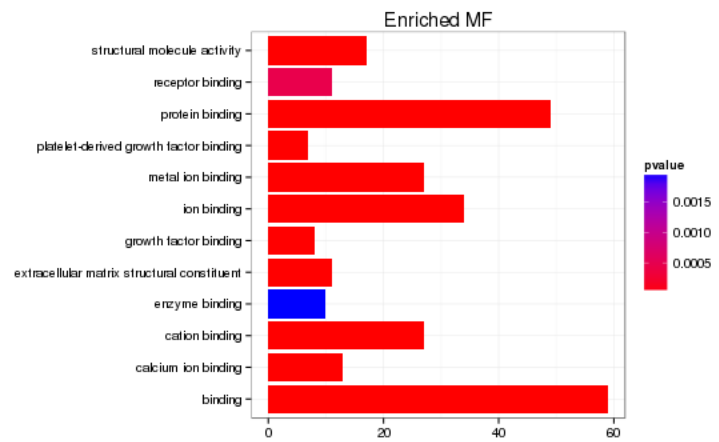
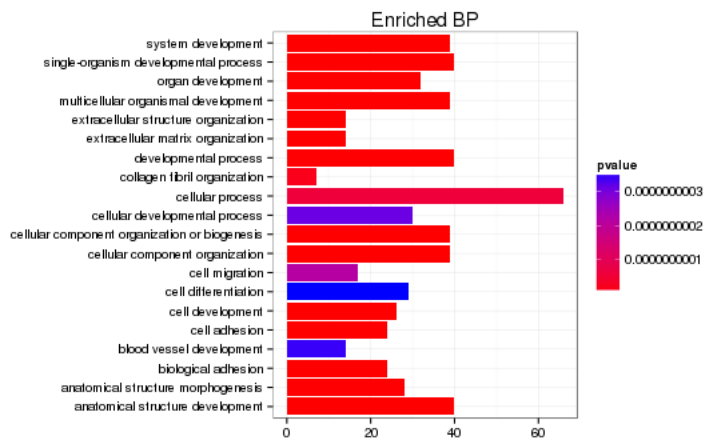
TF zscore:

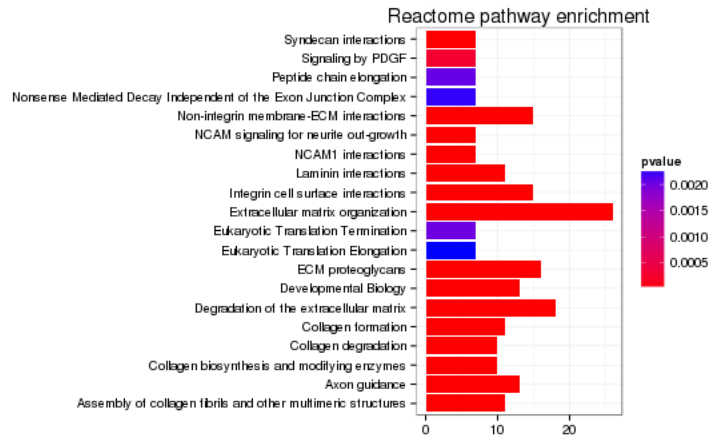
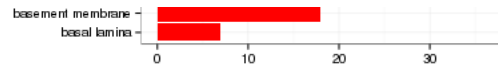


GO enrichment

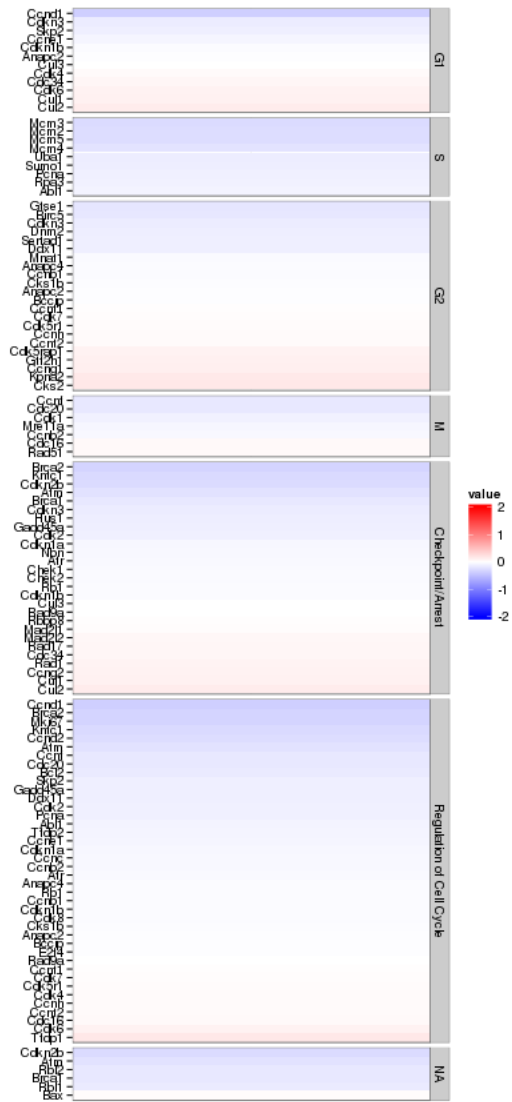
Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```

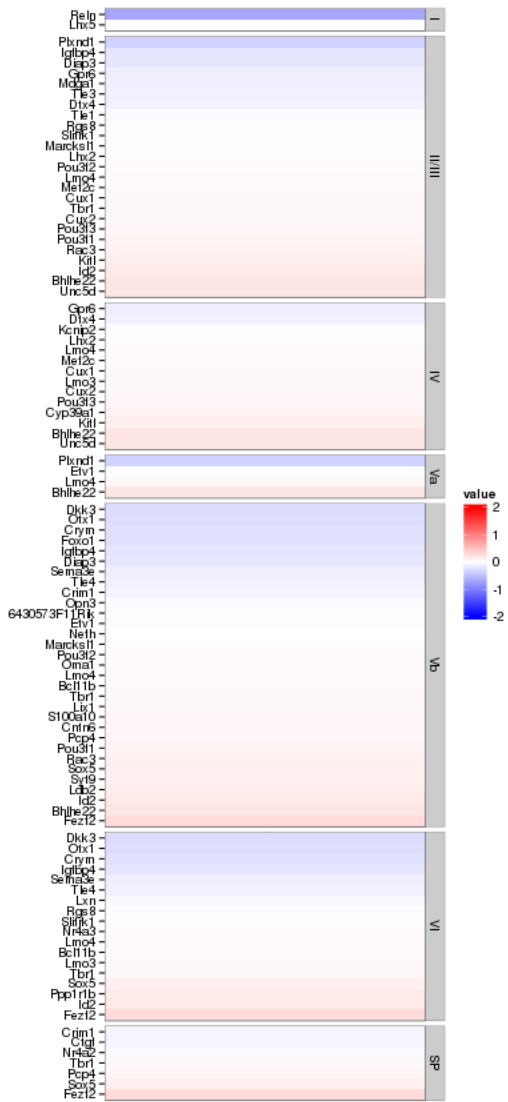




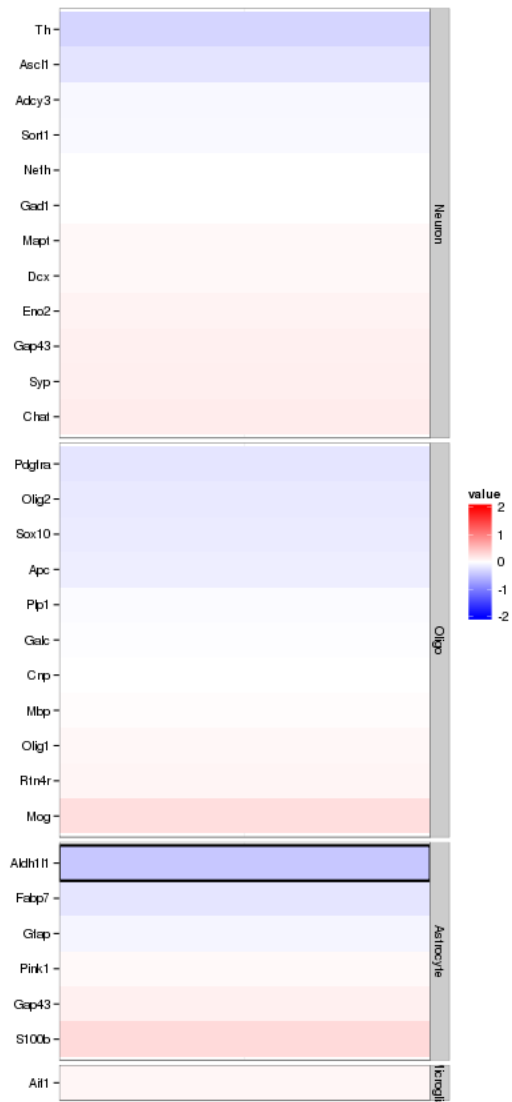
Enrichment or depletion for stage-specific cell cycle markers



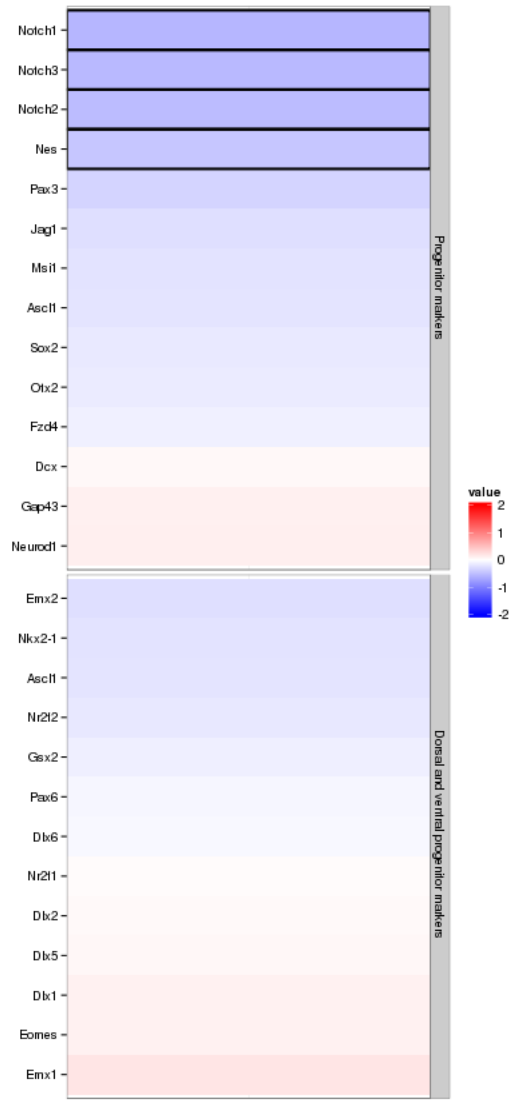
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



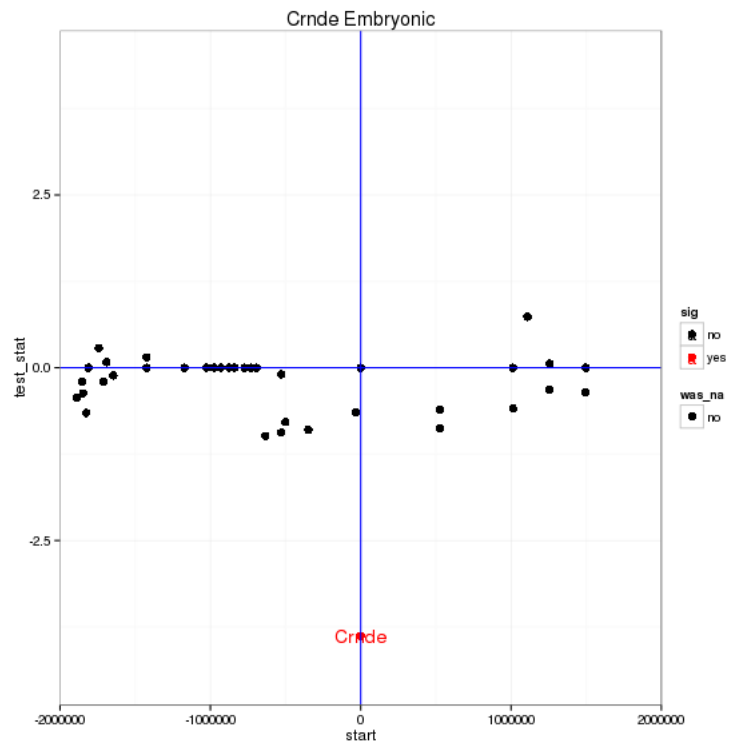
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR793
16 JR790
17 JR830

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	31799500.00	1.28	1.00
2 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	31799500.00	1.11	1.00
3 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	31799500.00	1.24	1.00
4 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	31799500.00	1.30	1.00
5 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	31799500.00	1.22	1.00
6 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	31799500.00	0.90	1.00
7 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	31799500.00	0.99	1.00
8 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	31799500.00	0.93	1.00
9 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	31799500.00	0.87	1.00
10 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	31799500.00	0.97	1.00
11 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	31799500.00	0.82	1.00
12 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	31799500.00	0.81	1.00
13 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	31799500.00	0.69	1.00
14 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	31799500.00	1.23	1.00
15 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR793/abundances.cxb	Cmde	0	Cmde_0	36520100.00	31799500.00	1.16	1.00
16 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR790/abundances.cxb	Cmde	1	Cmde_1	27555700.00	31799500.00	0.87	1.00
17 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR830/abundances.cxb	Cmde	2	Cmde_2	28630200.00	31799500.00	0.91	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Crnde -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Crnde_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/BrainMap/dat
## 2
## 3
## 4
## 5
```

Eldr KO vs WT (Adult)

This file shows the wt-v-ko comparison for Eldr.

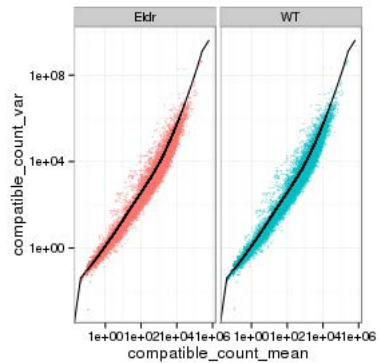
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

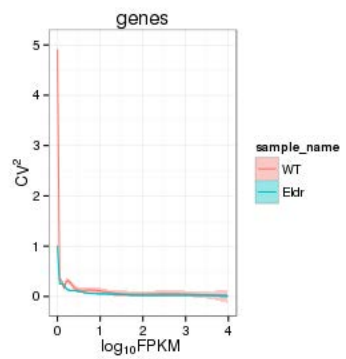
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



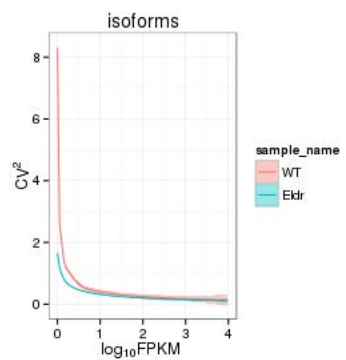
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

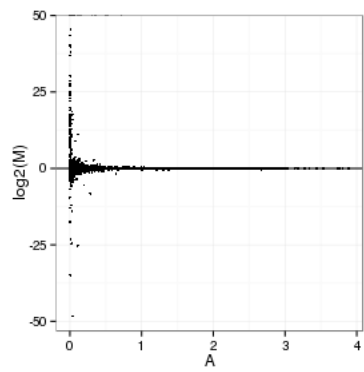
Genes:



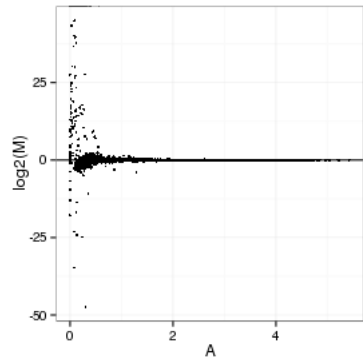
Isoforms:



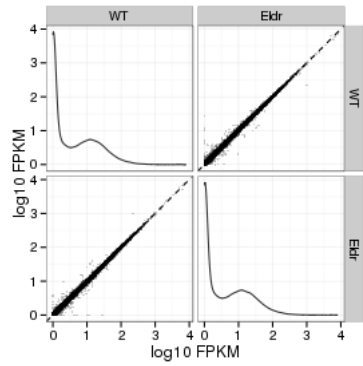
MvA plot



MvA plot counts



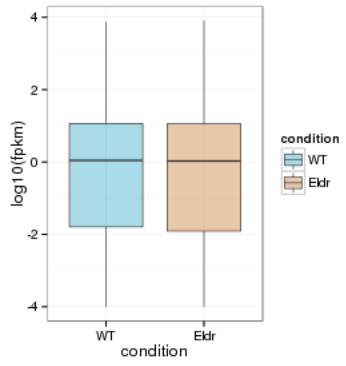
Scatterplot matrix



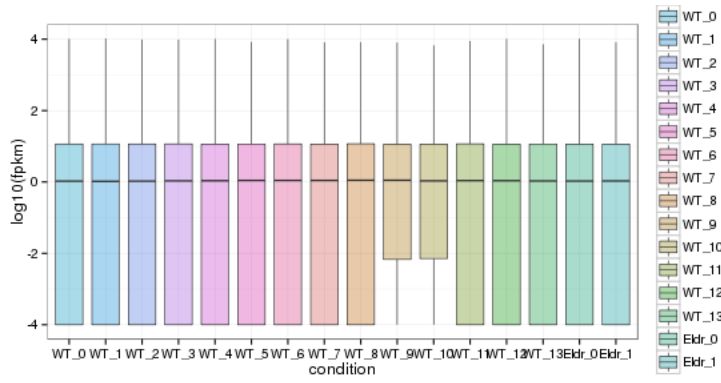
Distributions

Boxplots

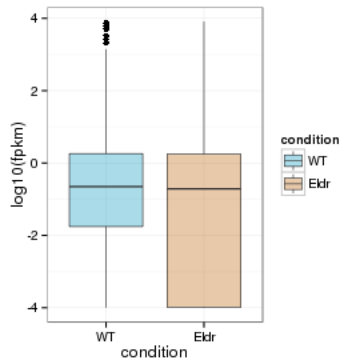
Boxplot (genes)



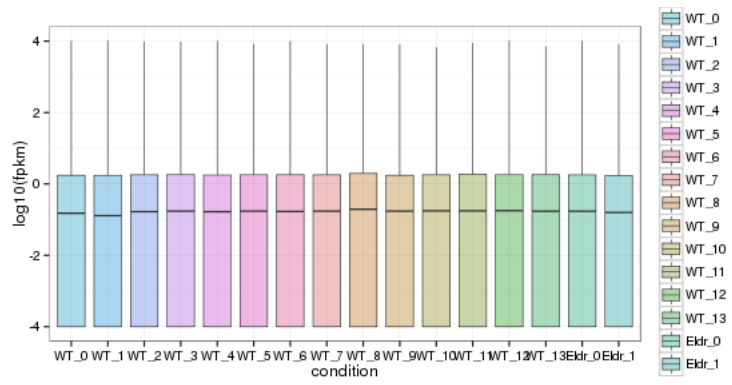
Boxplot (genes, replicates)



Boxplot (isoforms)

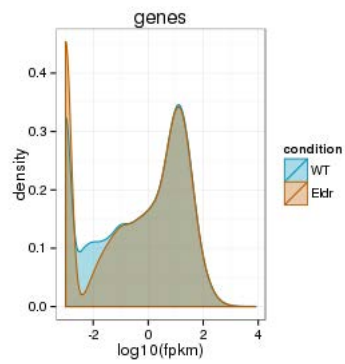


Boxplot (isoforms, replicates)

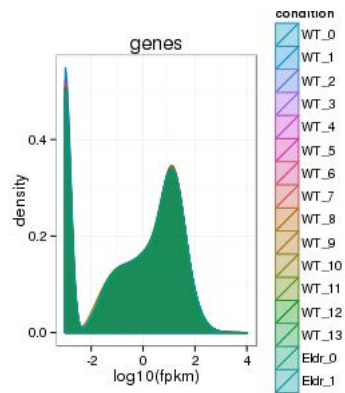


Density

Density (genes)

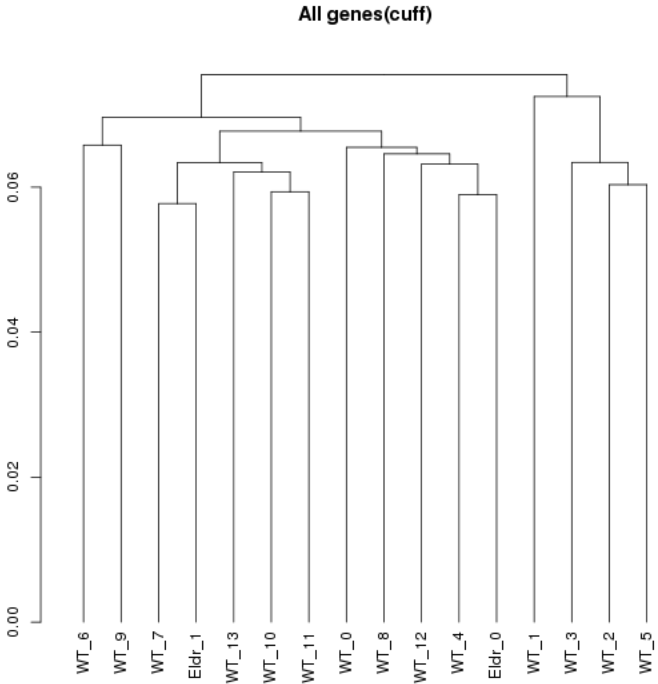


Density (genes, replicates)



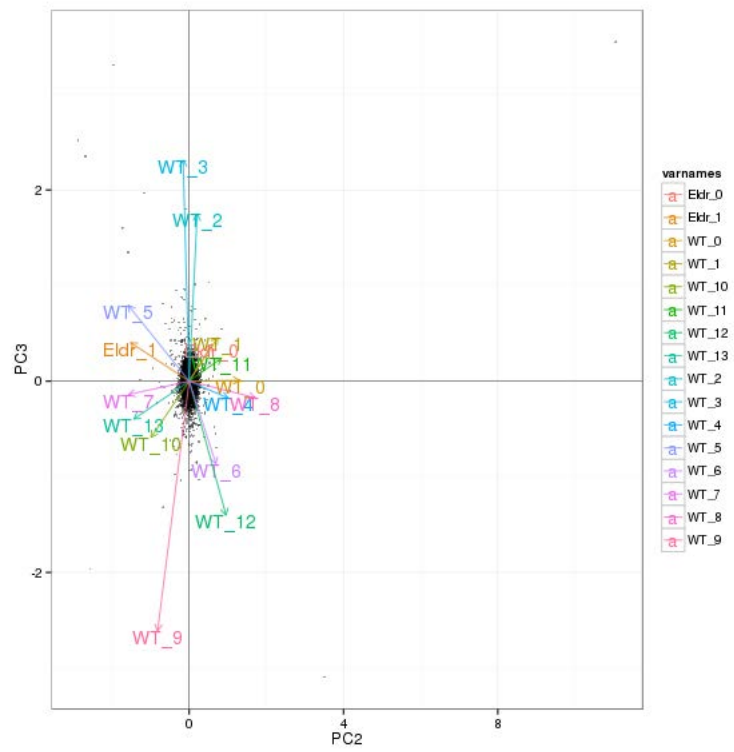
Clustering

Replicate Clusters

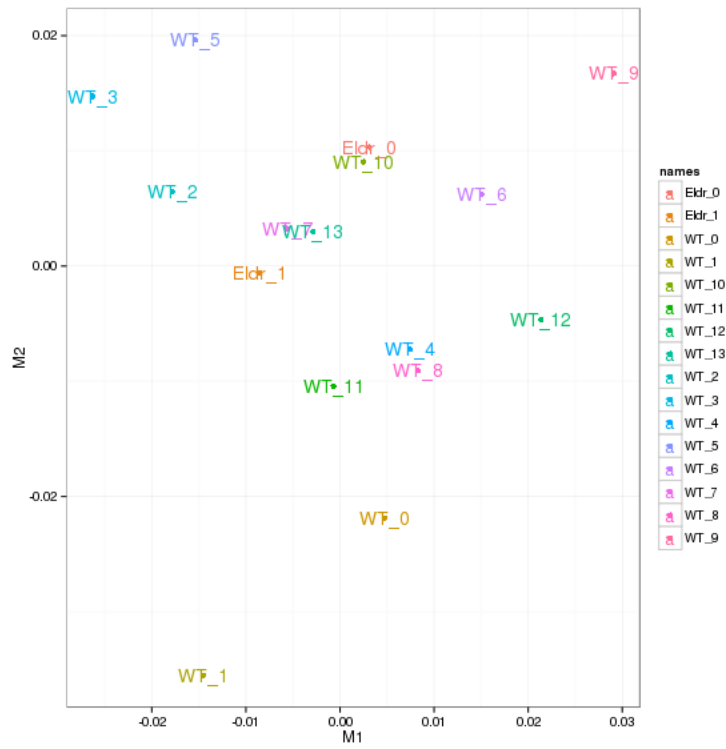


NULL

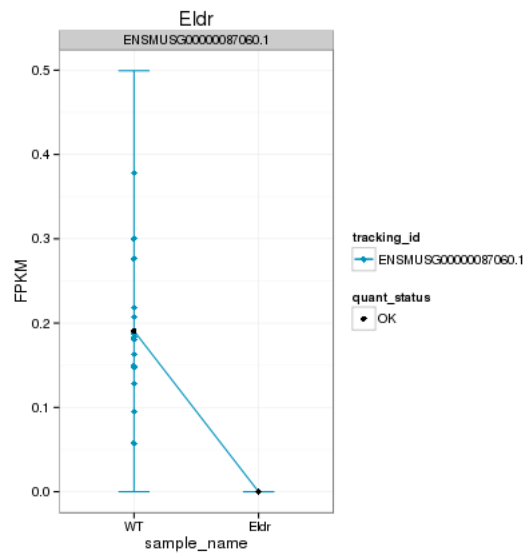
PCA (genes)



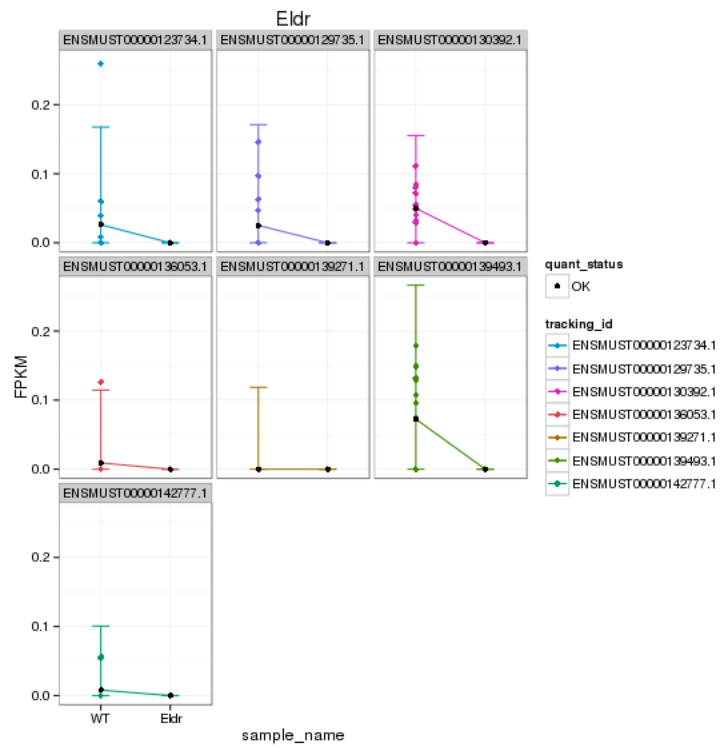
MDS (genes)



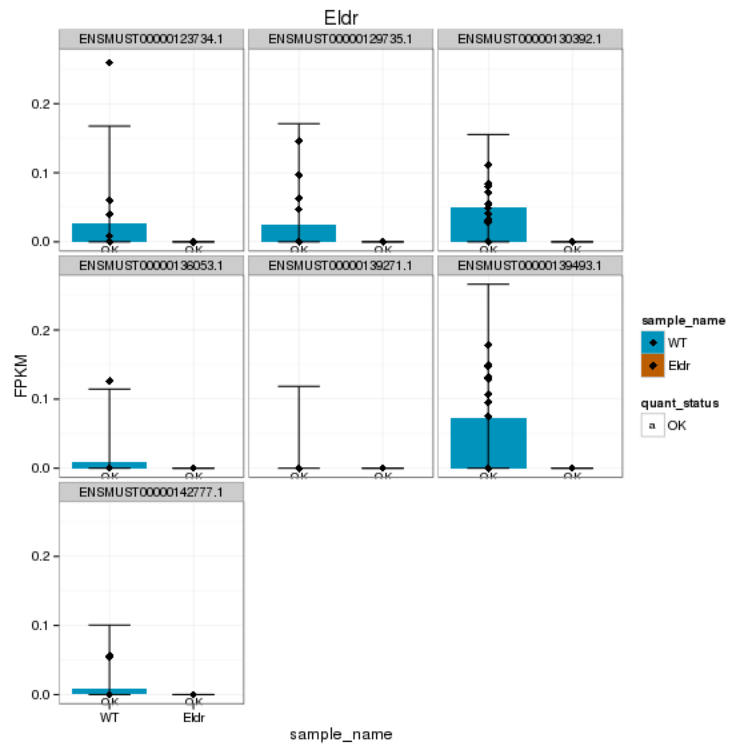
KO assessment
Endogenous lncRNA expression



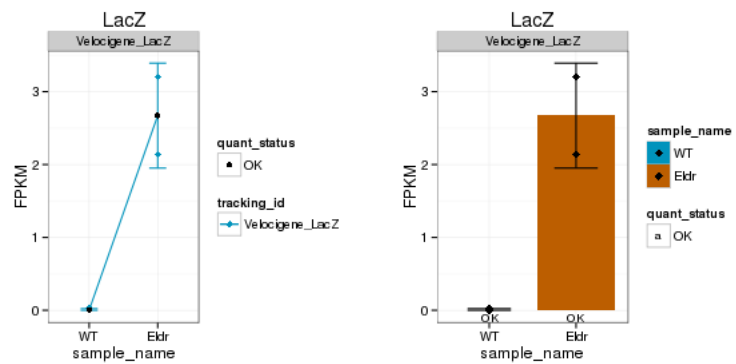
Endogenous expression of Eldr isoforms:



Barplot of gene expression:

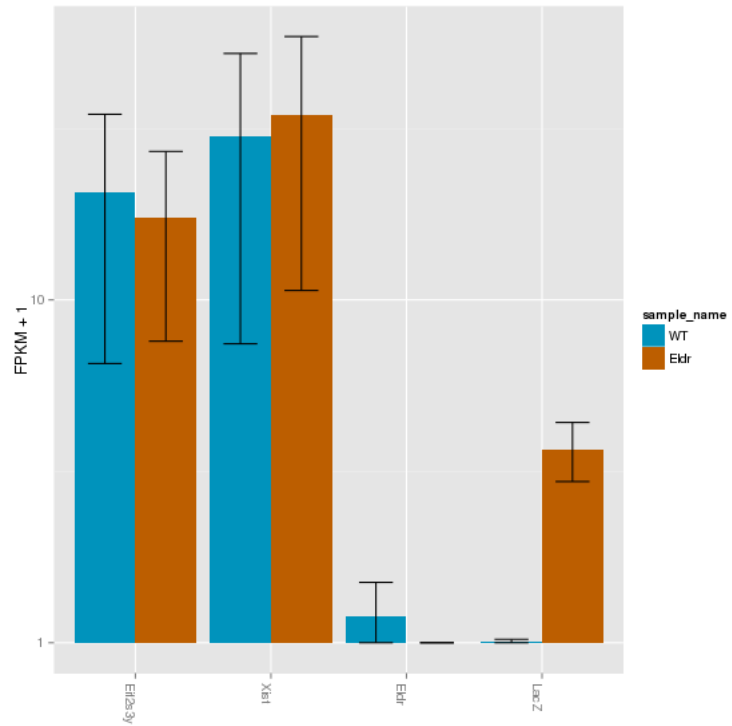


LacZ expression

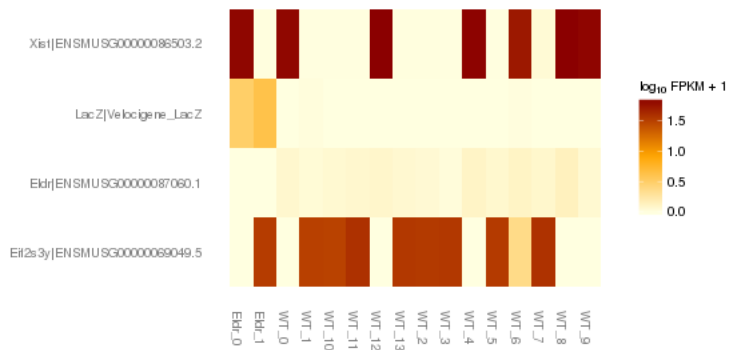


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

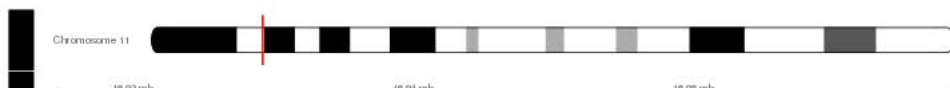
Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

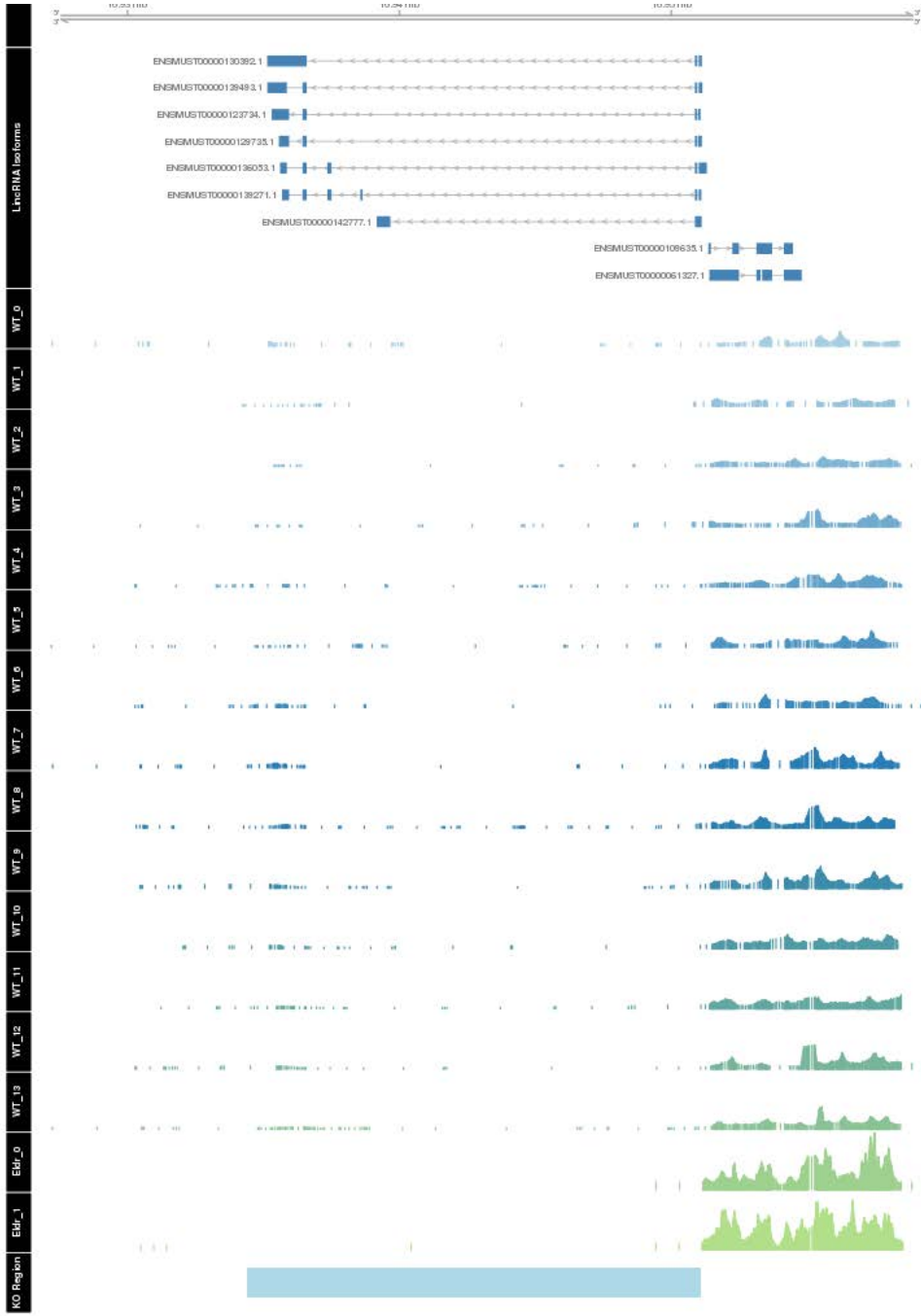


Expression heatmap:



Track visualization





Differential Analysis

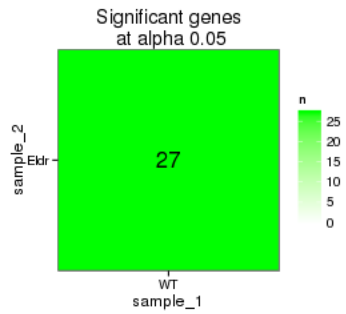
Differential Genes

There are 27 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 Nes
2 Ubc
3 Tpgs1
4 Adi1
5 Fos
6 Nr4a1
7 Rps14
8 Rps24
9 Nr4a3
10 Ndufs5
11 Arhgap27
12 Egr2
13 Egr1
14 Pcsk1n
15 Kihl42
16 C1qtnf4
17 Acp1
18 Fbxo48
19 Junb
20 Fat2
21 Capn11
22 Tpt1
23 Egr4
24 Gm9008
25 Wdfy1
26 Apold1
27 AA465934
```

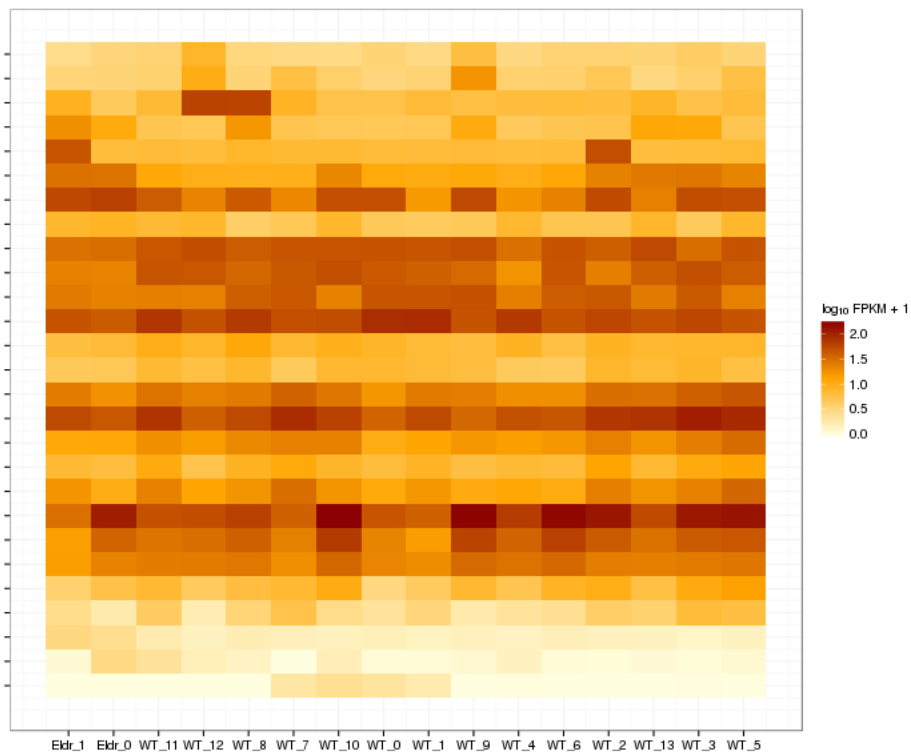
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

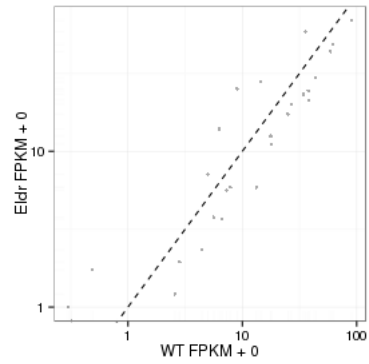


Significant genes with expression >50fpm (any condition):(turned off)

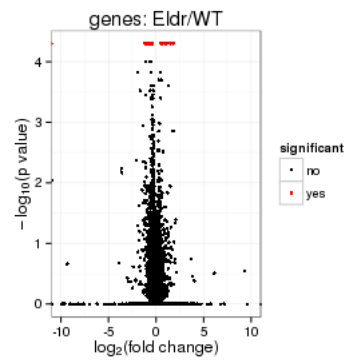
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

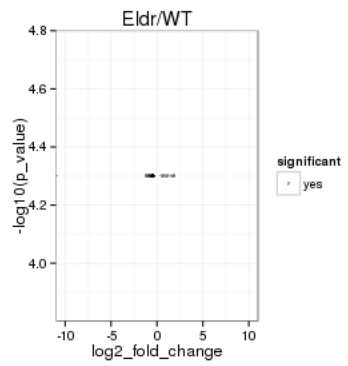
Scatter plot of significant genes only:



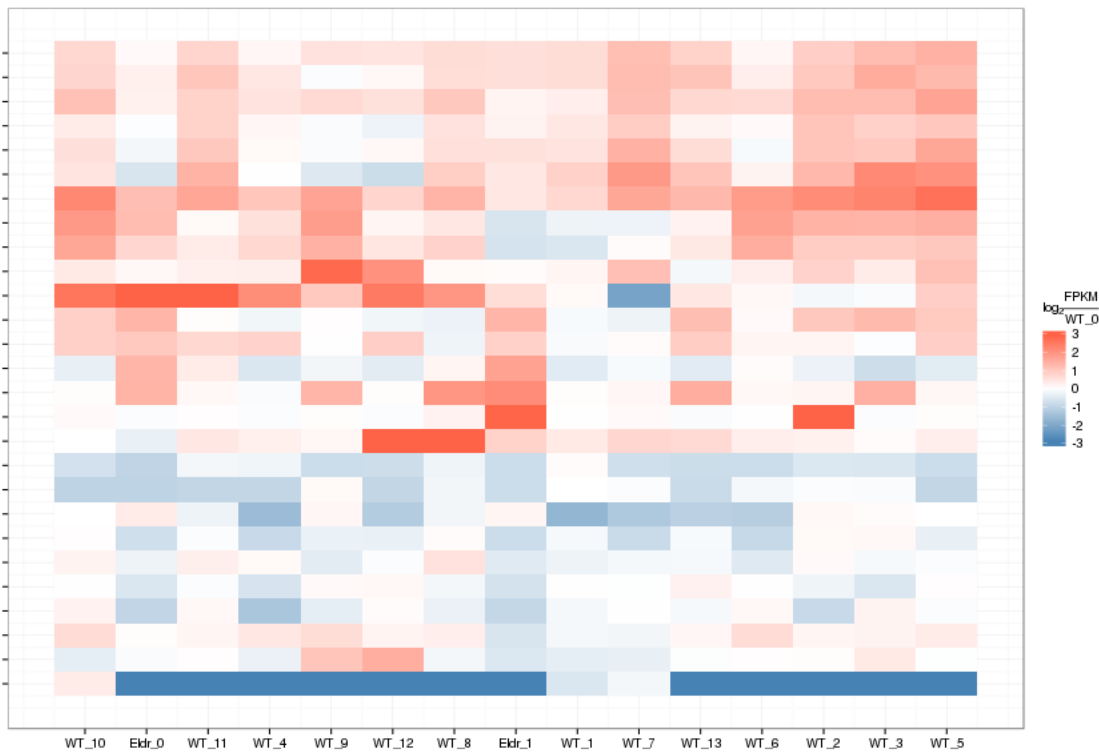
Volcano Plot



Volcano plot with significant genes only:



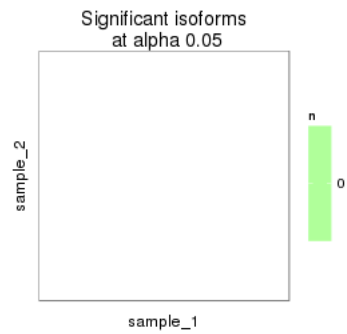
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

```
[1] "no sig isoforms"
```

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

Biocarta enrichment:


```
## [1] "Not enough significant categories to print a heatmap!"
```

Biocarta zscore:

```
## [1] "Not enough significant categories to print a heatmap!"
```

Reactome enrichment:

```
## [1] "Not enough significant categories to print a heatmap!"
```

Reactome zscore:

```
## [1] "Not enough significant categories to print a heatmap!"
```

Kegg enrichment:

```
## [1] "Not enough significant categories to print a heatmap!"
```

Kegg zscore:

```
## [1] "Not enough significant categories to print a heatmap!"
```

Interneuron enrichment:

```
## Error: argument is of length zero
```

Interneuron zscore:

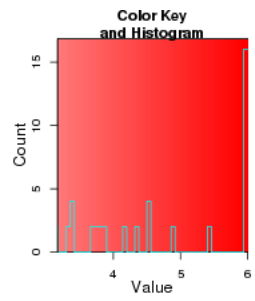
```
## Error: incorrect number of dimensions
```

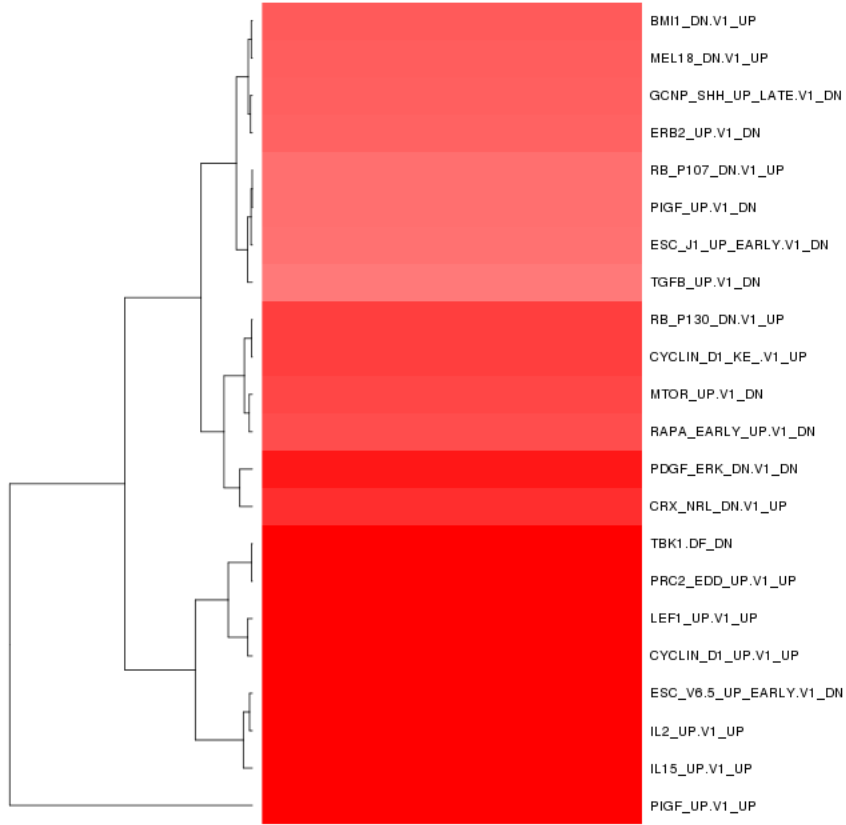
```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

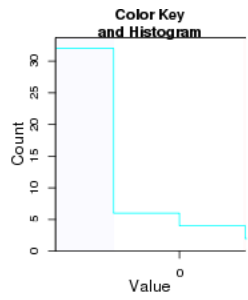
```
## Error: object 'x_ordered' not found
```

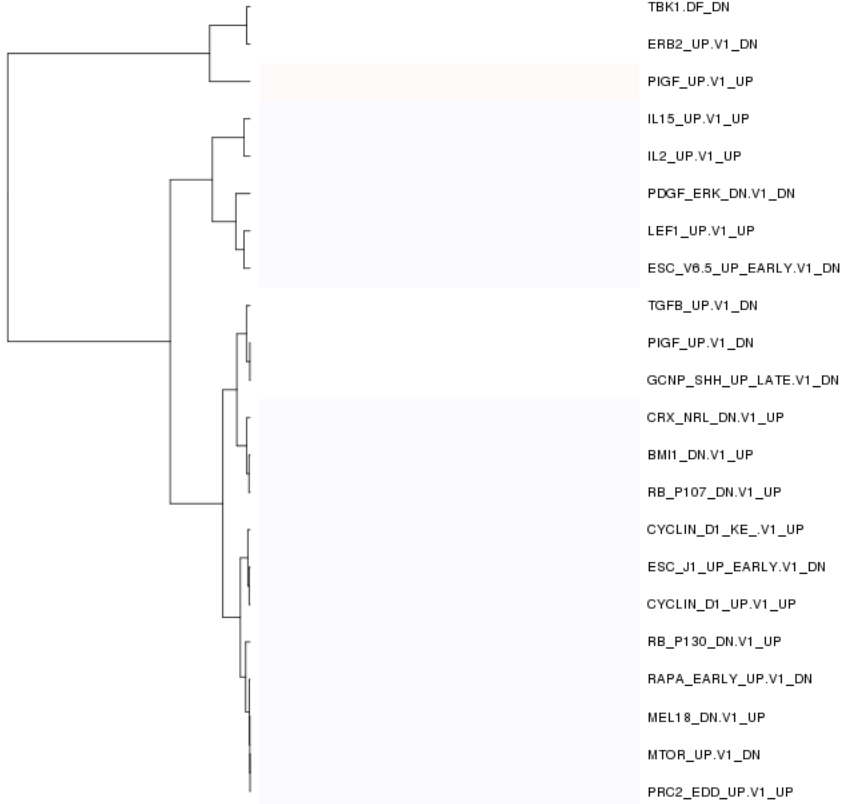
Oncogene enrichment:





Oncogene zscore:

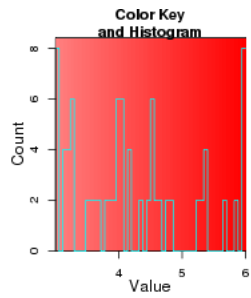


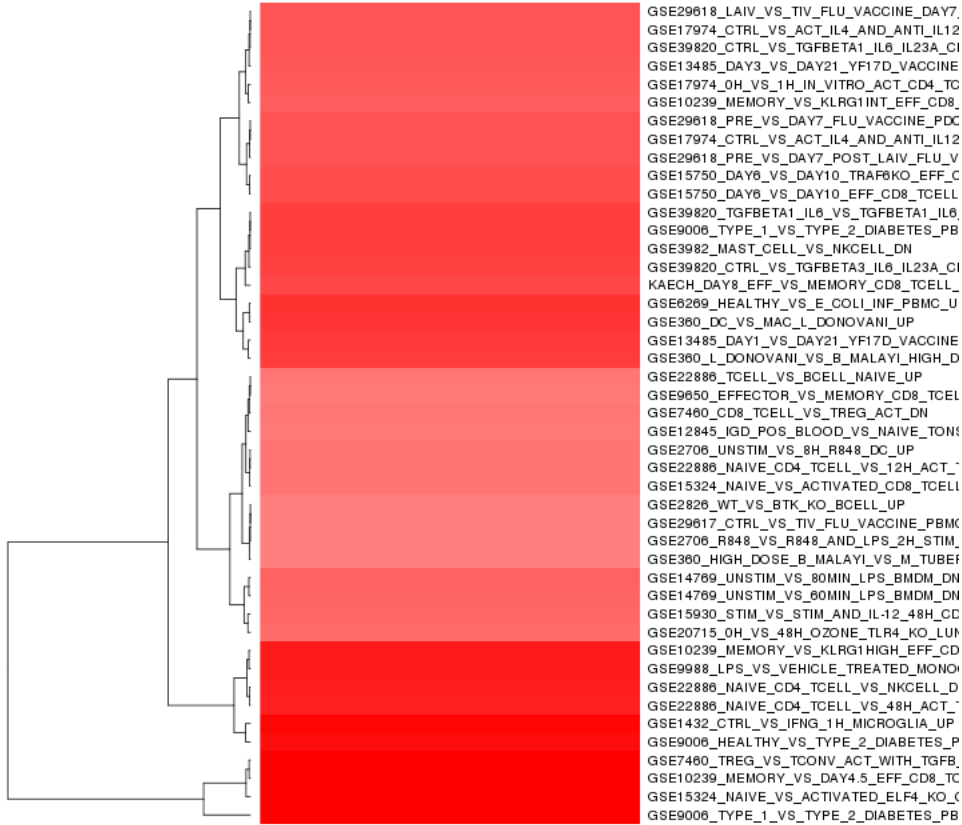


TBK1.DF_DN
 ERB2_UP.V1_DN
 PIGF_UP.V1_UP
 IL15_UP.V1_UP
 IL2_UP.V1_UP
 PDGF_ERK_DN.V1_DN
 LEF1_UP.V1_UP
 ESC_V6.5_UP_EARLY.V1_DN
 TGFB_UP.V1_DN
 PIGF_UP.V1_DN
 GCNP_SHH_UP_LATE.V1_DN
 CRX_NRL_DN.V1_UP
 BMI1_DN.V1_UP
 RB_P107_DN.V1_UP
 CYCLIN_D1_KE_.V1_UP
 ESC_J1_UP_EARLY.V1_DN
 CYCLIN_D1_UP.V1_UP
 RB_P130_DN.V1_UP
 RAPA_EARLY_UP.V1_DN
 MEL18_DN.V1_UP
 MTOR_UP.V1_DN
 PRC2_EDD_UP.V1_UP

Immuno enrichment:

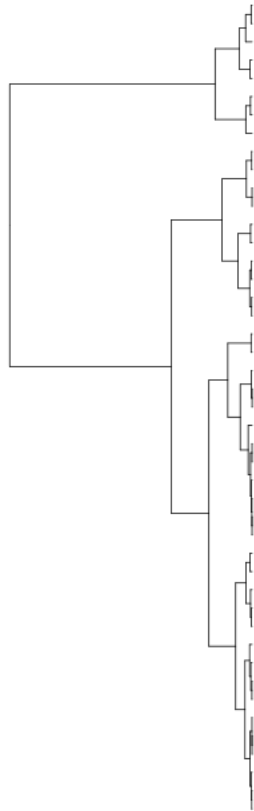
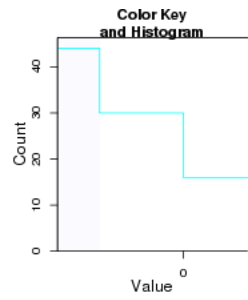
Error: subscript out of bounds





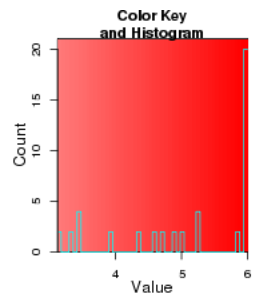
Immuno zscore:

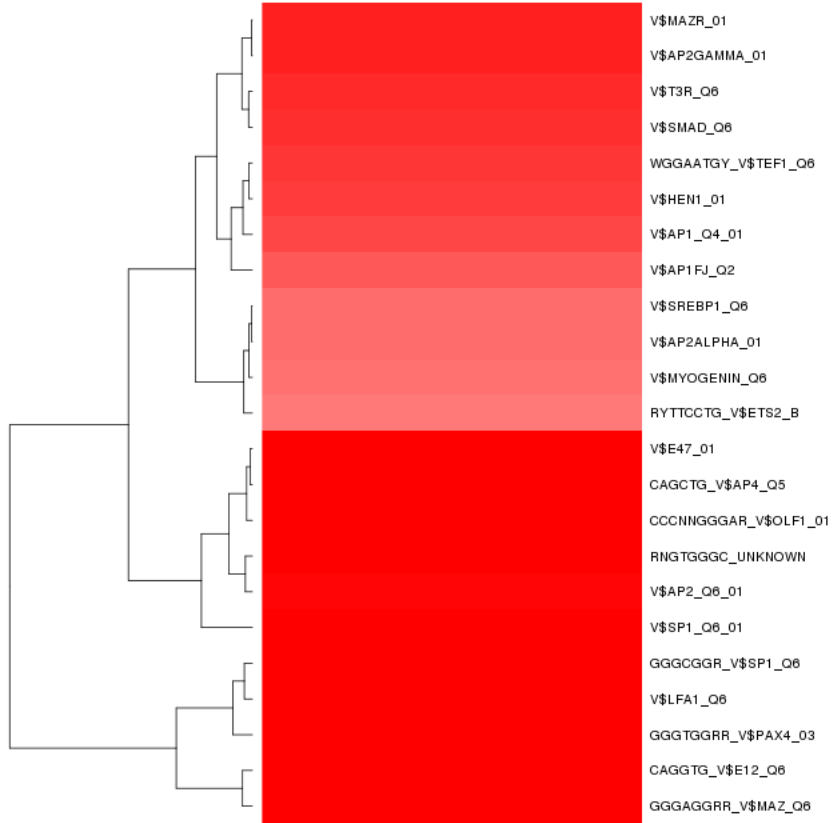
Error: subscript out of bounds



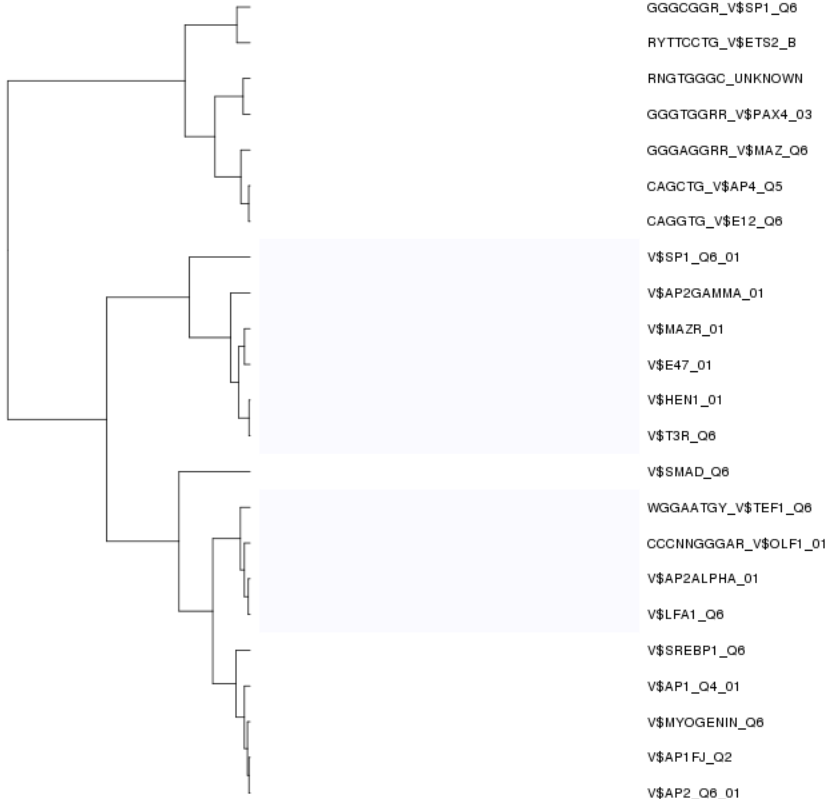
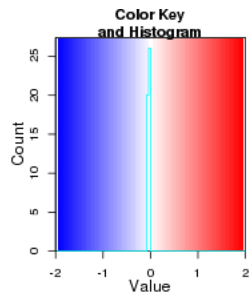
GSE39820_CTRL_VS_TGFBETA1_IL6_IL23A_CI
 GSE29618_PRE_VS_DAY7_FLU_VACCINE_PDC
 GSE7460_CD8_TCELL_VS_TREG_ACT_DN
 GSE13485_DAY1_VS_DAY21_YF17D_VACCINE
 GSE9006_TYPE_1_VS_TYPE_2_DIABETES_PB
 GSE29617_CTRL_VS_TIV_FLU_VACCINE_PBM
 GSE22886_NAIVE_CD4_TCELL_VS_NKCELL_D
 GSE29618_PRE_VS_DAY7_POST_LAIV_FLU_V
 GSE17974_CTRL_VS_ACT_IL4_AND_ANTI_IL12
 GSE9050_EFFECTOR_VS_MEMORY_CD8_TCEL
 GSE17974_CTRL_VS_ACT_IL4_AND_ANTI_IL12
 GSE17974_0H_VS_1H_IN_VITRO_ACT_CD4_TC
 GSE9006_TYPE_1_VS_TYPE_2_DIABETES_PB
 GSE14709_UNSTIM_VS_80MIN_LPS_BMDM_DN
 GSE14709_UNSTIM_VS_80MIN_LPS_BMDM_DN
 KAECH_DAY8_EFF_VS_MEMORY_CD8_TCELL_
 GSE15324_NAIVE_VS_ACTIVATED_ELF4_KO_C
 GSE10239_MEMORY_VS_DAY4.5_EFF_CD8_TC
 GSE300_HIGH_DOSE_B_MALAYI_VS_M_TUBEI
 GSE9988_LPS_VS_VEHICLE_TREATED_MONO
 GSE22886_TCELL_VS_BCELL_NAIVE_UP
 GSE300_DC_VS_MAC_L_DONOVANI_UP
 GSE39820_TGFBETA1_IL6_VS_TGFBETA1_IL6
 GSE22886_NAIVE_CD4_TCELL_VS_48H_ACT_
 GSE15930_STIM_VS_STIM_AND_IL12_48H_CD
 GSE7460_TREG_VS_TCONV_ACT_WITH_TGFB
 GSE13485_DAY3_VS_DAY21_YF17D_VACCINE
 GSE29618_LAIV_VS_TIV_FLU_VACCINE_DAY7
 GSE2706_R848_VS_R848_AND_LPS_2H_STIM
 GSE3982_MAST_CELL_VS_NKCELL_DN
 GSE1432_CTRL_VS_IFNG_1H_MICROGLIA_UP
 GSE9006_HEALTHY_VS_TYPE_2_DIABETES_P
 GSE15750_DAY6_VS_DAY10_TRAF6KO_EFF_C
 GSE20715_0H_VS_48H_OZONE_TLR4_KO_LUN
 GSE10239_MEMORY_VS_KLRG1INT_EFF_CD8
 GSE0209_HEALTHY_VS_E_COLI_INF_PBMC_U
 GSE12845_IJD_POS_BLOOD_VS_NAIVE_TON
 GSE300_L_DONOVANI_VS_B_MALAYI_HIGH_D
 GSE10239_MEMORY_VS_KLRG1HIGH_EFF_CD
 GSE15324_NAIVE_VS_ACTIVATED_CD8_TCELL
 GSE2706_UNSTIM_VS_8H_R848_DC_UP
 GSE2826_WT_VS_BTK_KO_BCELL_UP
 GSE22886_NAIVE_CD4_TCELL_VS_12H_ACT_
 GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL
 GSE39820_CTRL_VS_TGFBETA3_IL6_IL23A_CI

TF enrichment:



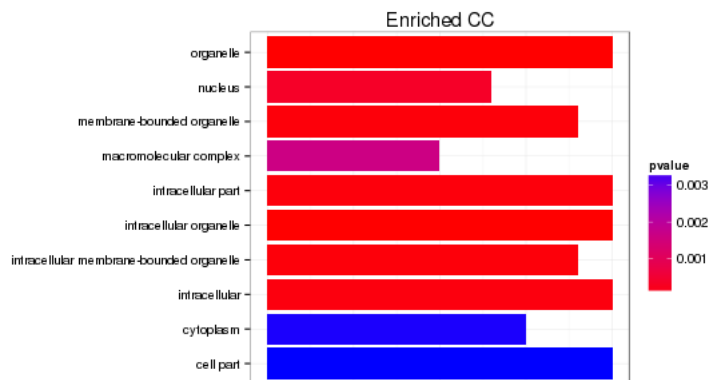
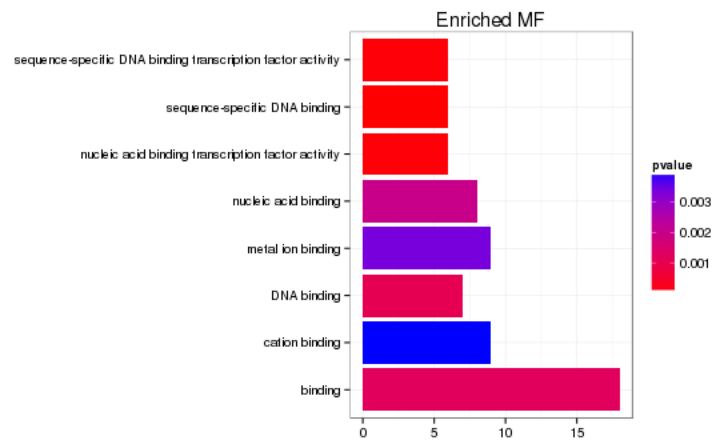
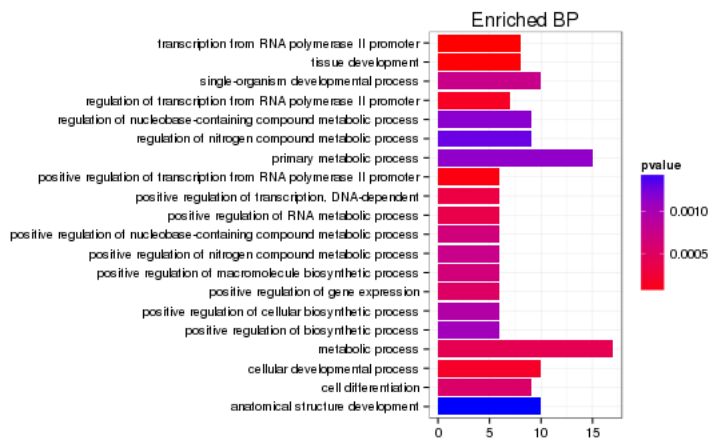


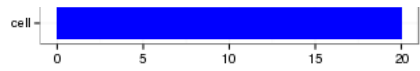
TF zscore:



GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

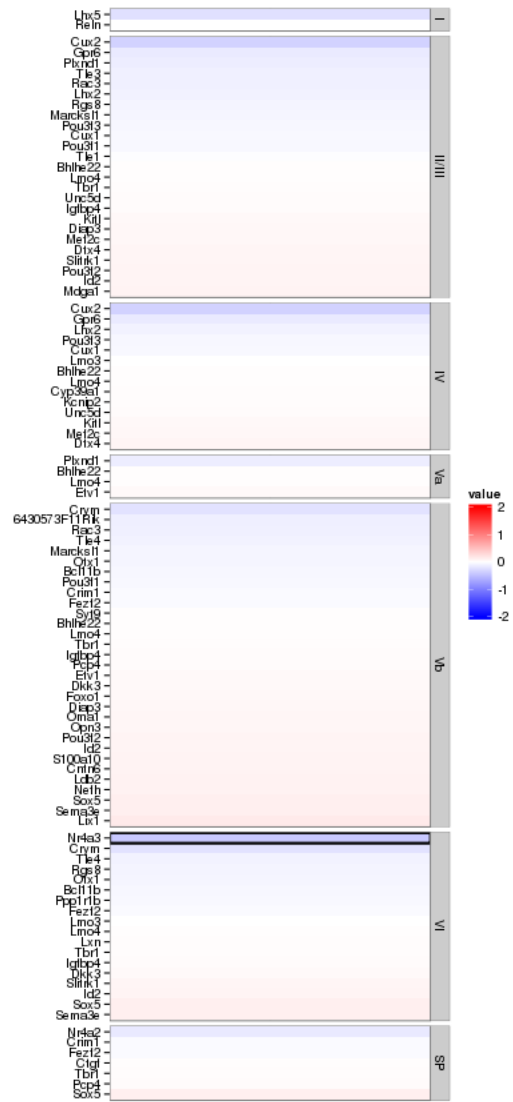




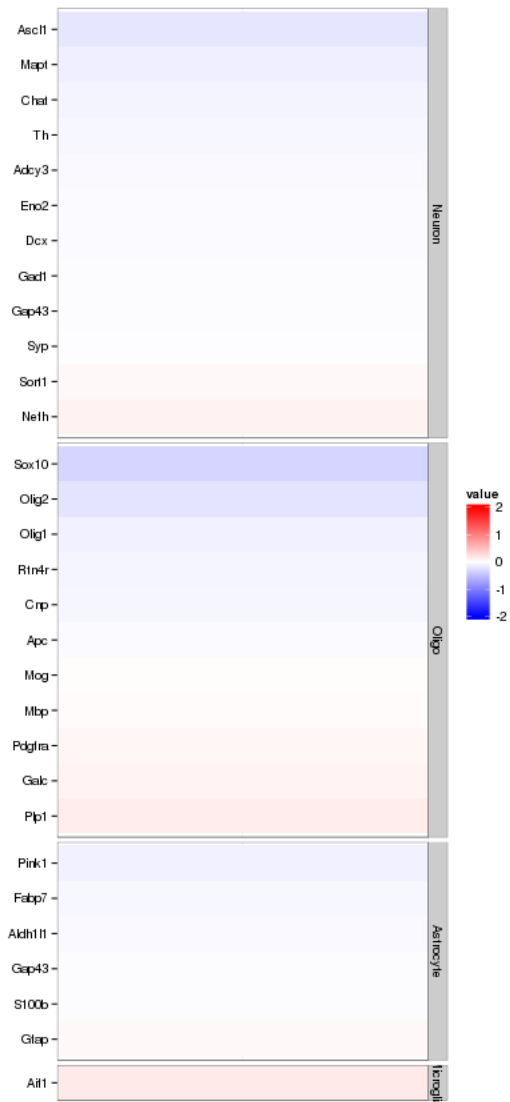
```
## Error: need finite 'xlim' values
```

```
## Error: need finite 'xlim' values
```

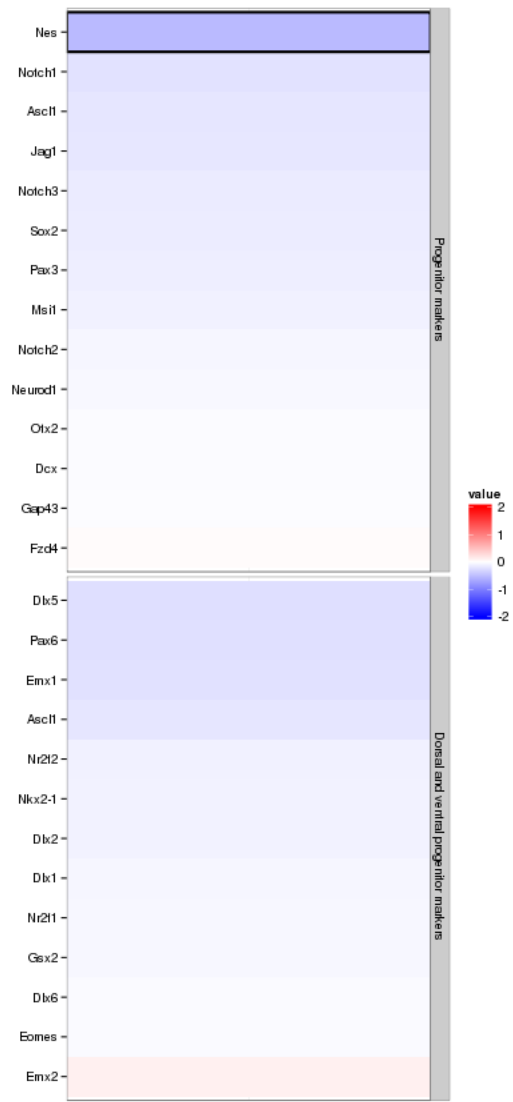
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



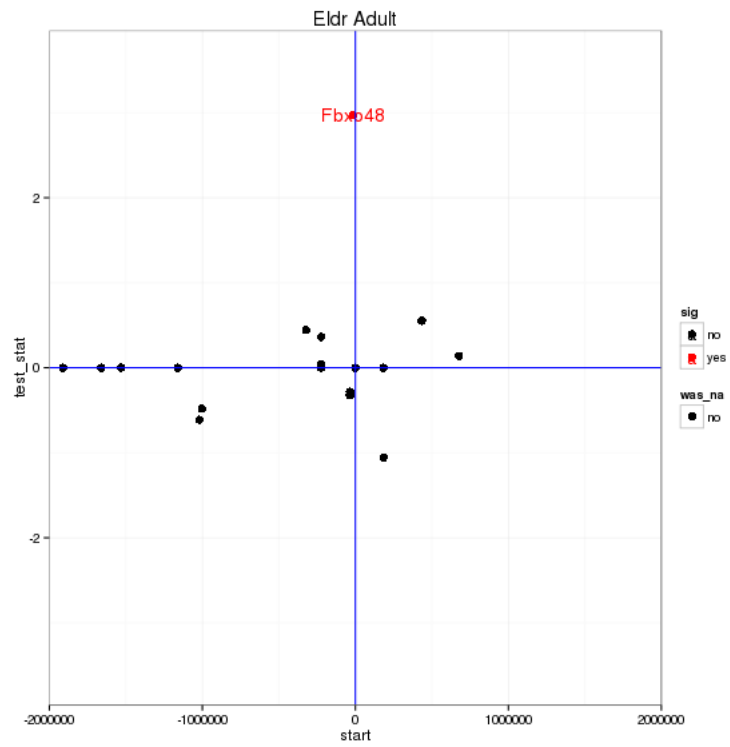
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 1 genes significantly regulated in a region this size is: 0.0788



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR799

16 JR777

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34779900.00	0.76	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34779900.00	0.58	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34779900.00	0.98	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34779900.00	0.80	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34779900.00	1.03	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34779900.00	1.07	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34779900.00	0.80	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34779900.00	1.15	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34779900.00	1.01	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34779900.00	1.30	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34779900.00	1.50	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34779900.00	1.01	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34779900.00	0.97	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34779900.00	1.20	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR799/abundances.cxb	Eldr	0	Eldr_0	36453400.00	34779900.00	1.03	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR777/abundances.cxb	Eldr	1	Eldr_1	42739700.00	34779900.00	1.23	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
```

```

## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2     colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4       DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10       Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3    graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1       Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29    latticeExtra_0.6-26
## [25] MASS_7.3-33        Matrix_1.1-3       munSELL_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10       qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3   scales_0.2.4       splines_3.0.2
## [37] stats4_3.0.2       survival_2.37-7    tcltk_3.0.2
## [40] tools_3.0.2        XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info


```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Eldr -o /n/rinn_data1/seq/lgofff/Projects/BrainMap/data/diffs/Eldr_vs_WT_Adult /n/rinn_data1/seq/lgofff/Projects/BrainMap/data/annc
## 2
## 3
## 4
## 5
```

Eldr KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Eldr.

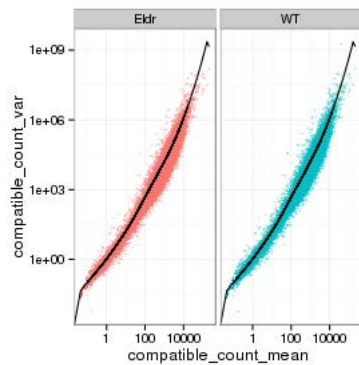
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

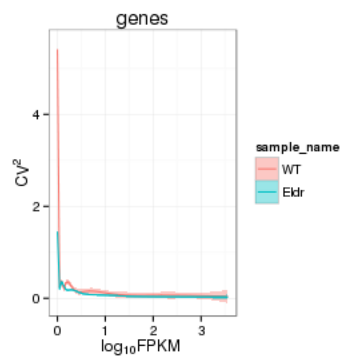
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



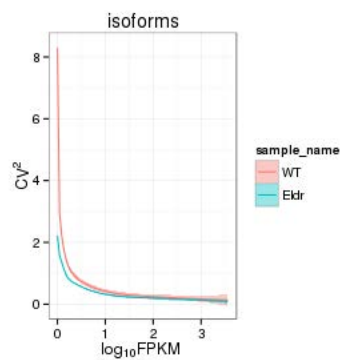
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

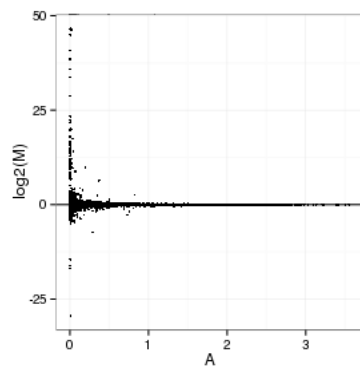
Genes:



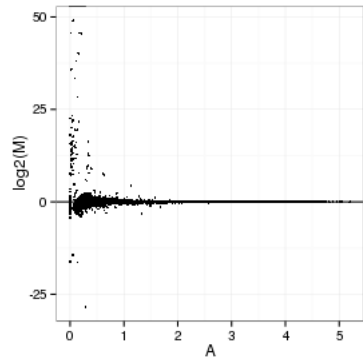
Isoforms:



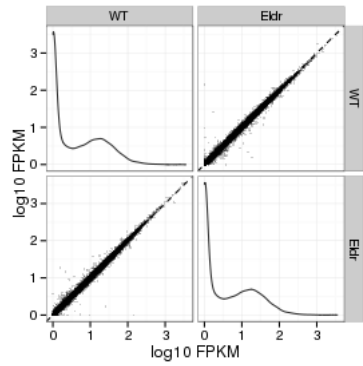
MvA plot



MvA plot counts



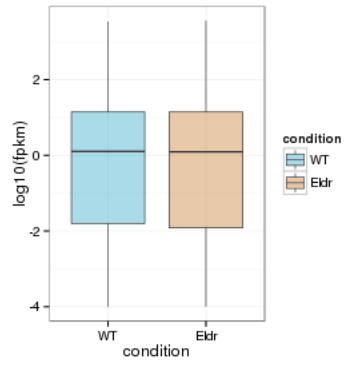
Scatterplot matrix



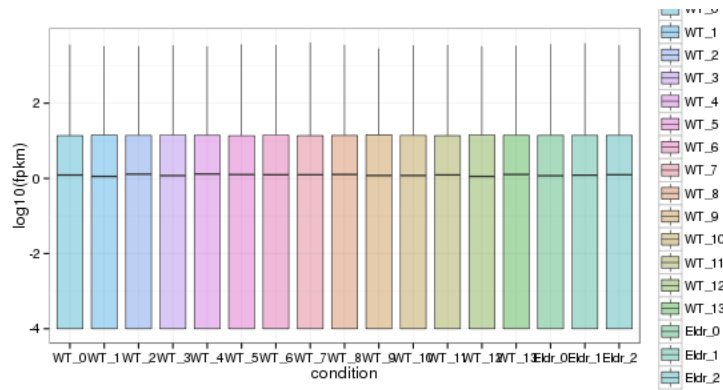
Distributions

Boxplots

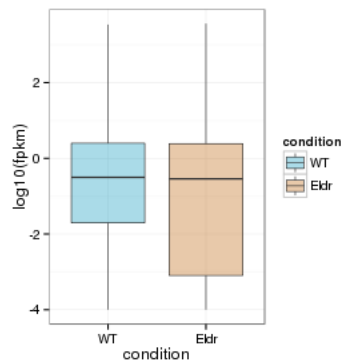
Boxplot (genes)



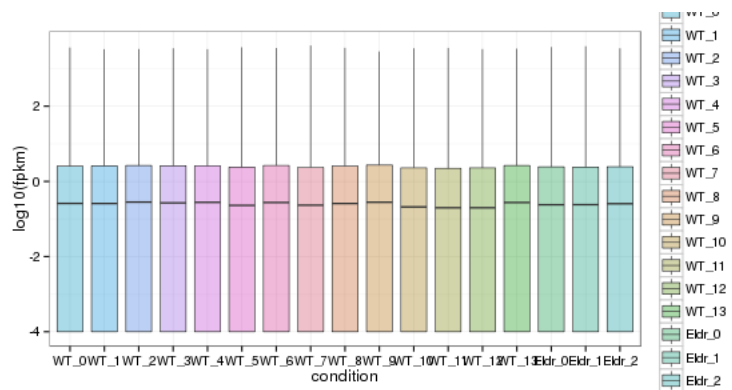
Boxplot (genes, replicates)



Boxplot (isoforms)

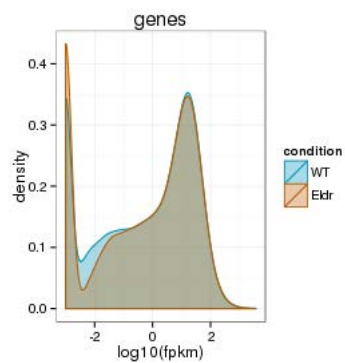


Boxplot (isoforms, replicates)

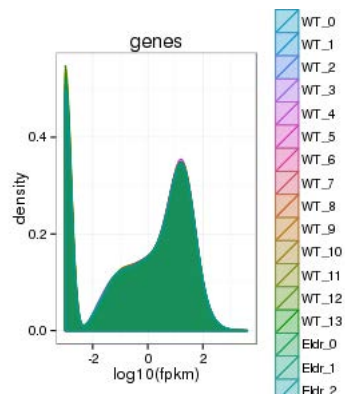


Density

Density (genes)

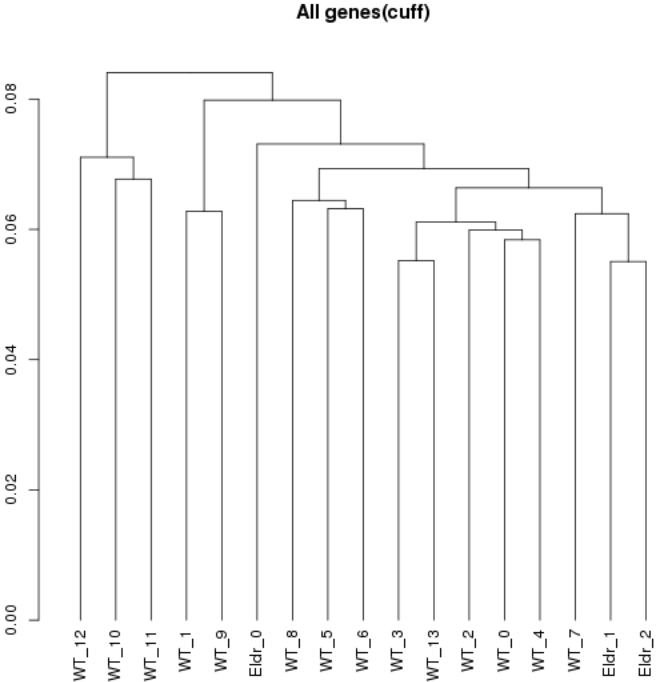


Density (genes, replicates)



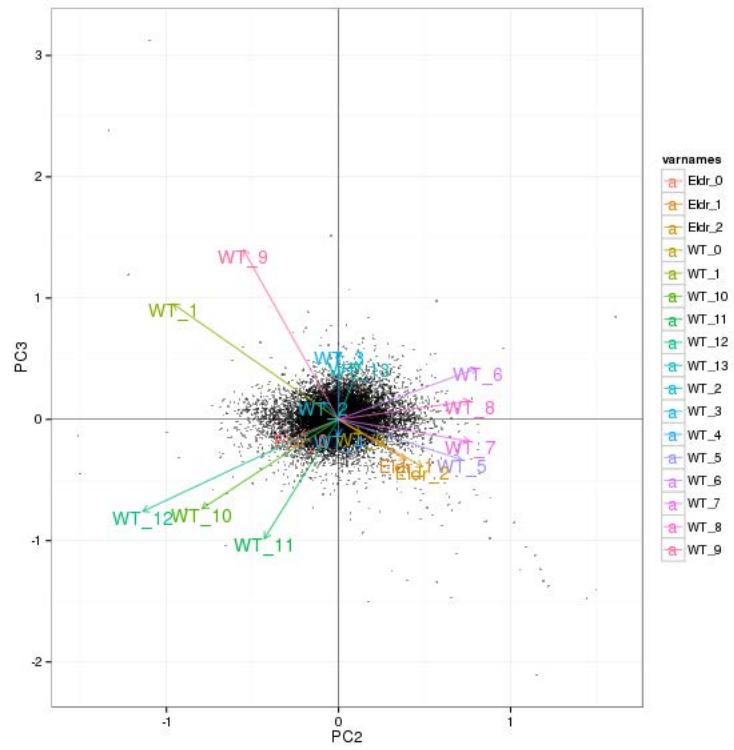
Clustering

Replicate Clusters

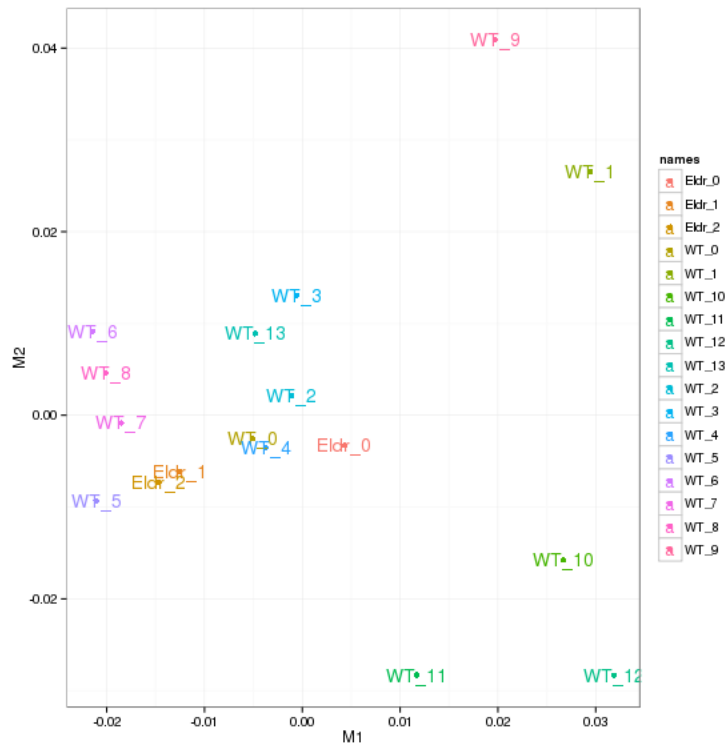


NULL

PCA (genes)

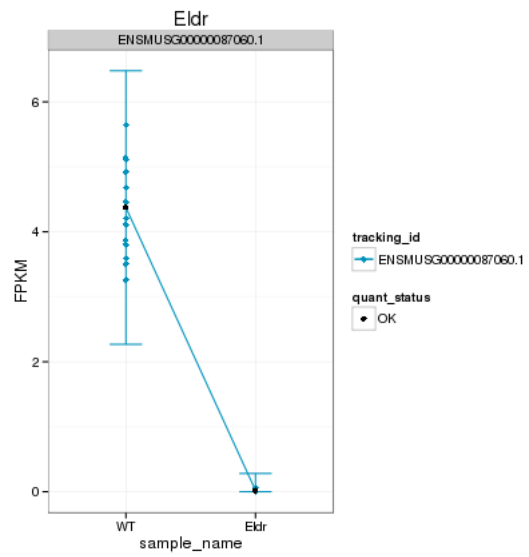


MDS (genes)

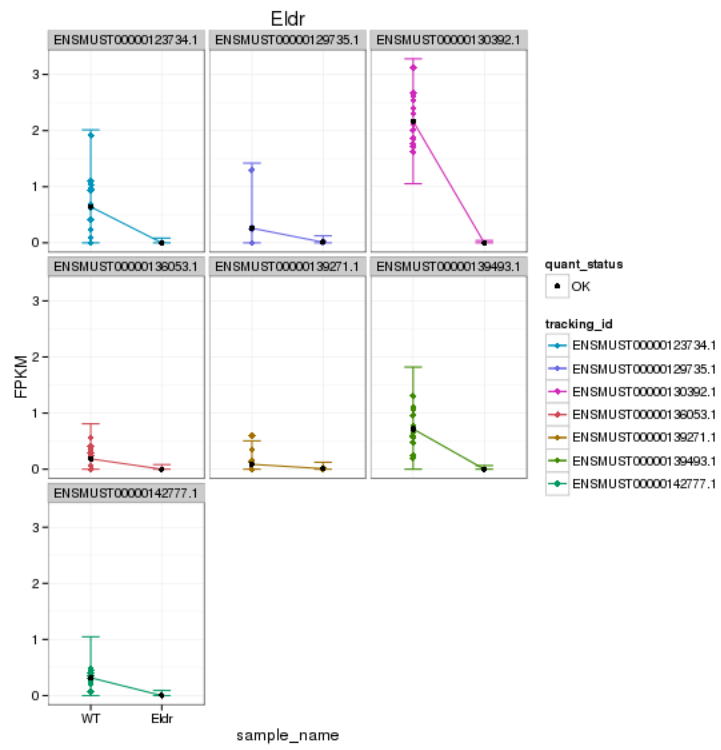


KO assessment

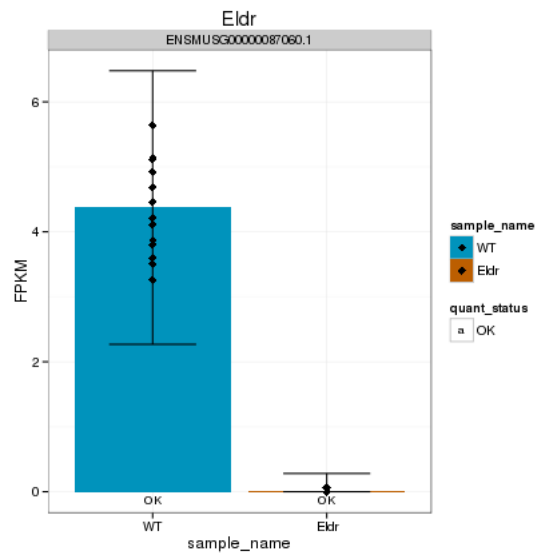
Endogenous IncRNA expression



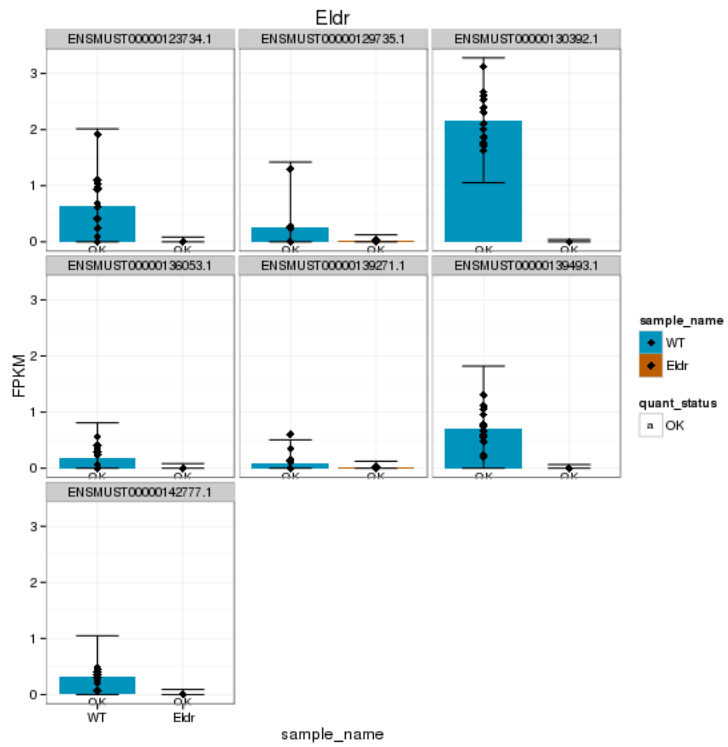
Endogenous expression of Eldr isoforms:



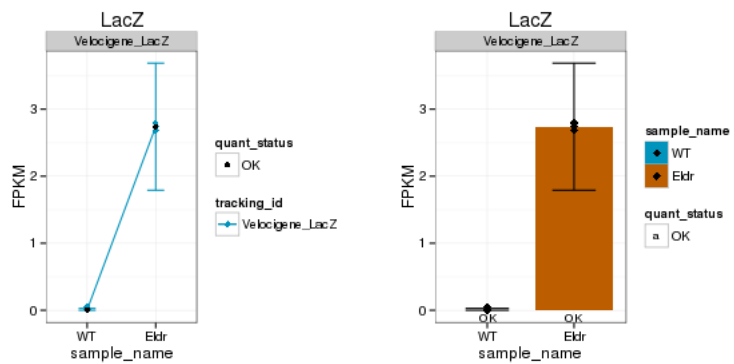
Barplot of gene expression:



Barplot of isoform expression:

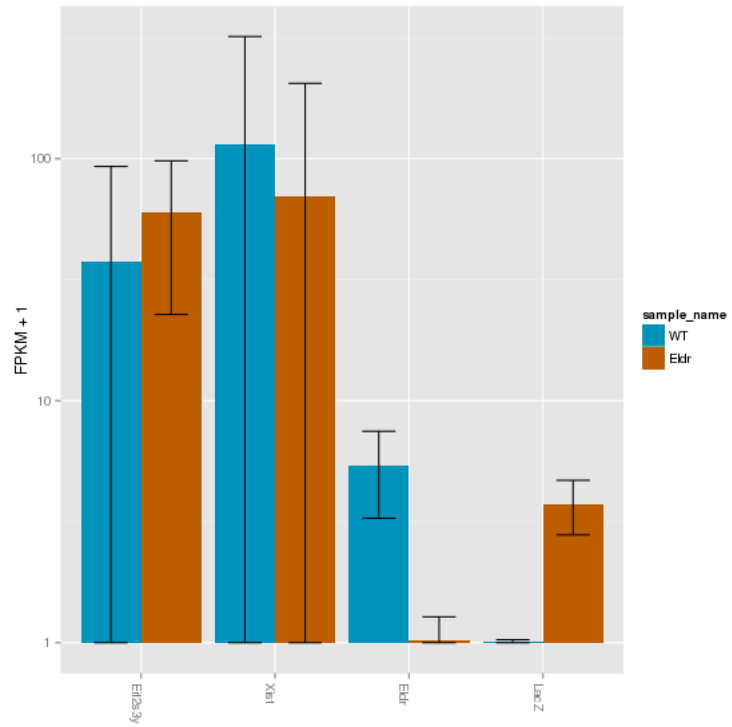


LacZ expression

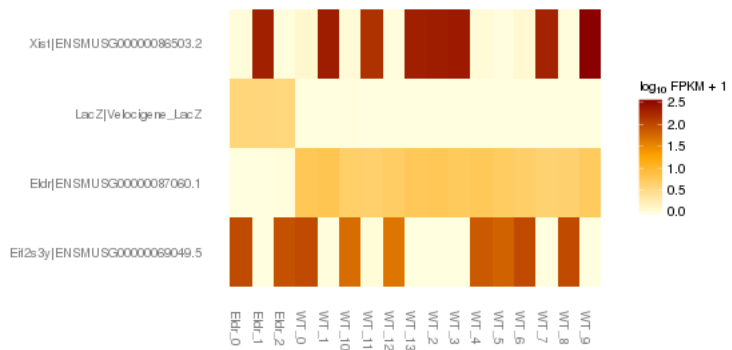


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

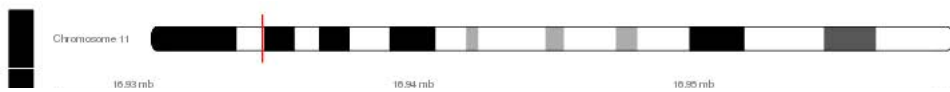
Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



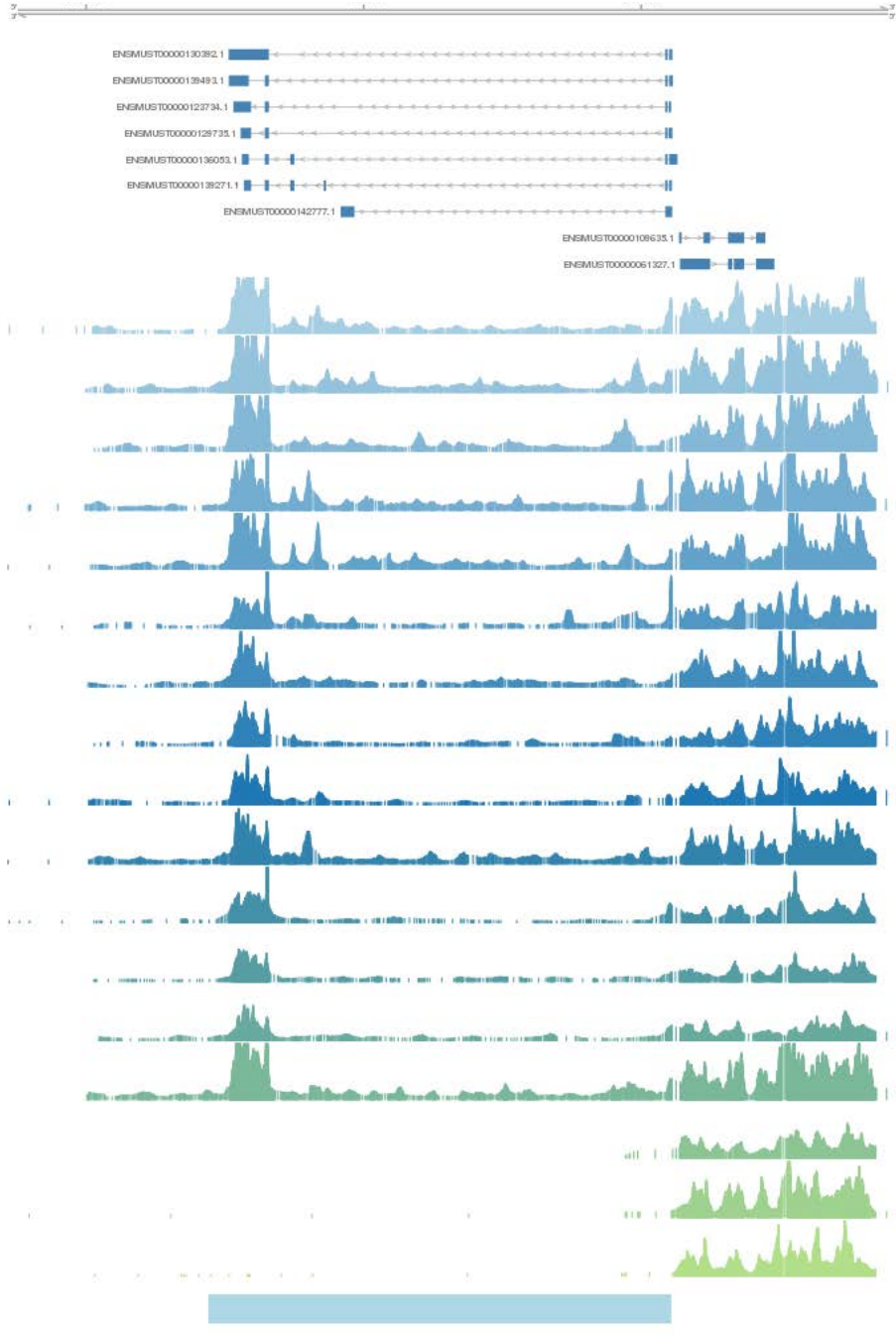
Expression heatmap:



Track visualization



NO Region LncRNA isoforms WT_0 WT_1 WT_2 WT_3 WT_4 WT_5 WT_6 WT_7 WT_8 WT_9 WT_10 WT_11 WT_12 WT_13 EB1_0 EB1_1 EB1_2



Differential Analysis

Differential Genes

There are 151 significantly differentially expressed genes. They are:

	geneAnnot\$gene_short_name
1	Cdh1
2	Col1a1
3	Cacna1e
4	Utp20
5	Polr2a
6	Nid1
7	Vax1
8	Rplp1
9	Ttc9b
10	Ptgds
11	Abca1
12	Dync1h1
13	D10Bwg1379e
14	Lama2
15	Snrpf
16	Egfr
17	Pold2
18	Aebp1
19	Prpf8
20	Hoxb9
21	Pdhb
22	Nid2
23	Sla
24	Myh9
25	Col2a1
26	Dlx2
27	Igf2r
28	Prss41
29	Fbn2
30	Rps14
31	Lbx1
32	Rps26
33	Pgm2
34	Fn1
35	Lamc1
36	Ddr2
37	Lrp2

38 Fbn1
39 Notch2
40 Col11a1
41 Tnc
42 Svep1
43 Snapc3

44 Scp2
45 Kcnq4
46 Ndufs5
47 Wdr65
48 Nkx1-1
49 Pdgfra
50 Uncx
51 Lhx5
52 Col1a2
53 Slc13a4
54 Aldh1l1
55 Itpr1
56 Dera
57 Herc2
58 Hddc3
59 Iqgap1
60 Eif3f
61 Lama1
62 3830406C13Rik
63 Adamts15
64 Gbx2
65 Prr7
66 Fras1
67 Vps13c
68 Figl1
69 Cox8a
70 Frem2

71 Rps16
72 Vstm2l
73 Notch3
74 Ifit172
75 Hoxb4
76 Atp5l
77 Srrm2
78 Pcsk1n
79 Klhl42
80 Tlx3

81 C1qtnf4
82 Dnah7b
83 2310036O22Rik
84 Fmod
85 Hebp1
86 Tuba1c
87 Lor
88 Pgp
89 Gm9493
90 Rps2
91 Acp1
92 Sox3
93 Wdr89
94 Scand1
95 Sox18
96 Gm7729
97 Fat4

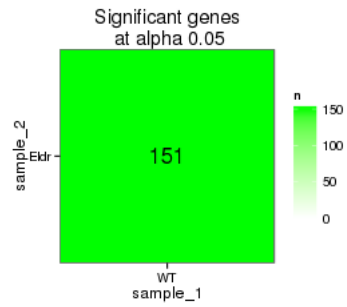
98 Dync2h1

99 Kmt2d
100 Sct1
101 Rpl29
102 H2afx
103 Ccdc171
104 Dnah6
105 Lamb2
106 Tusc1
107 Zfp771
108 Mrfap1
109 Gm10020
110 Rpl36
111 Capn11
112 H2afj
113 Tpt1
114 Rps12
115 Rpl35
116 Sox11
117 Zfp428
118 Rpl36-ps3
119 Finc
120 Ptpz1
121 Ddx3y
122 Nkain2
123 Ahnak

124 Fat1
125 Gm1673
126 Kihl9
127 Jund
128 Gm10340
129 Gm7292
130 Gpr27
131 Srp54a
132 Wdfy1
133 Hbb-bt
134 Fat3
135 Gm3558
136 Gm2974
137 Gm3411
138 Gm2897
139 Gm11273
140 Ccdc85c
141 Ier5l
142 Gm3739
143 Malat1
144 Gm10406
145 Gm3373
146 Gm3526
147 Ccdc85b
148 Gm3500
149 Gm9825
150 Gm5796
151 Gm3591

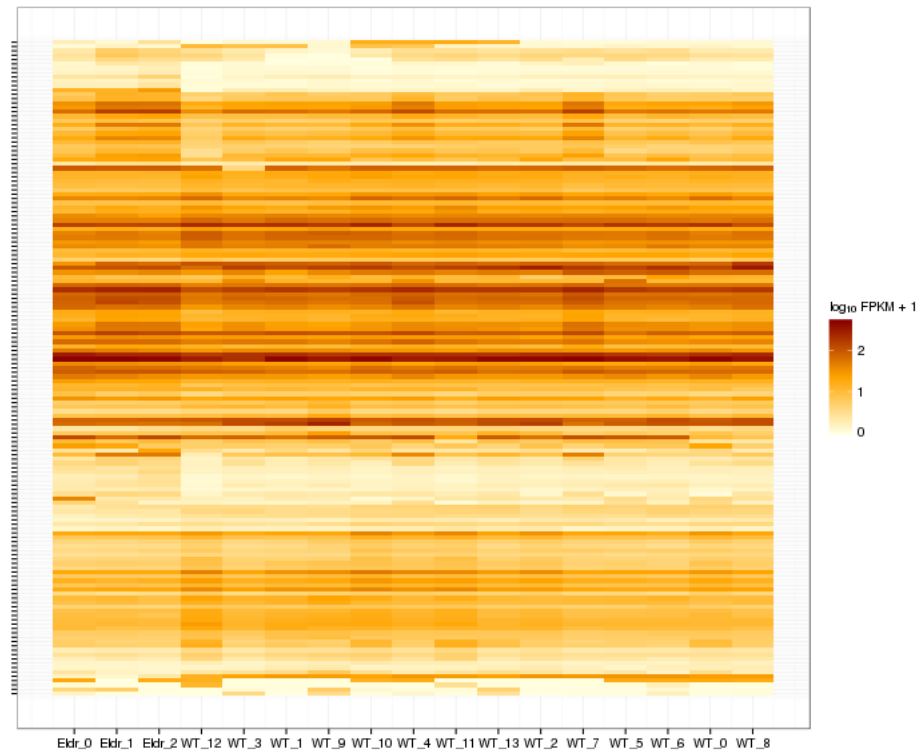
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

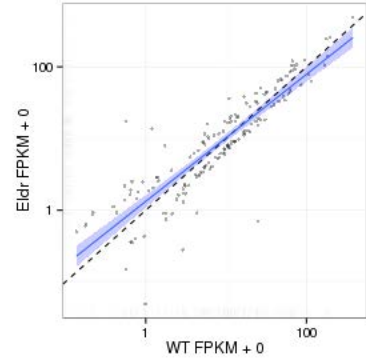


Significant genes with expression >50fpkm (any condition):(turned off)

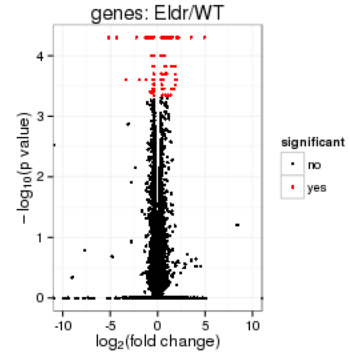
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

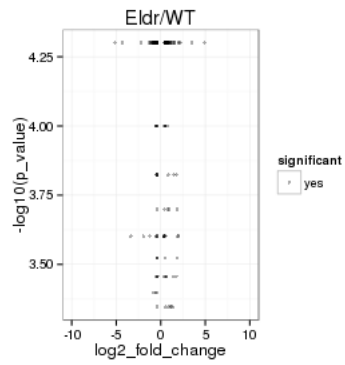
Scatter plot of significant genes only:



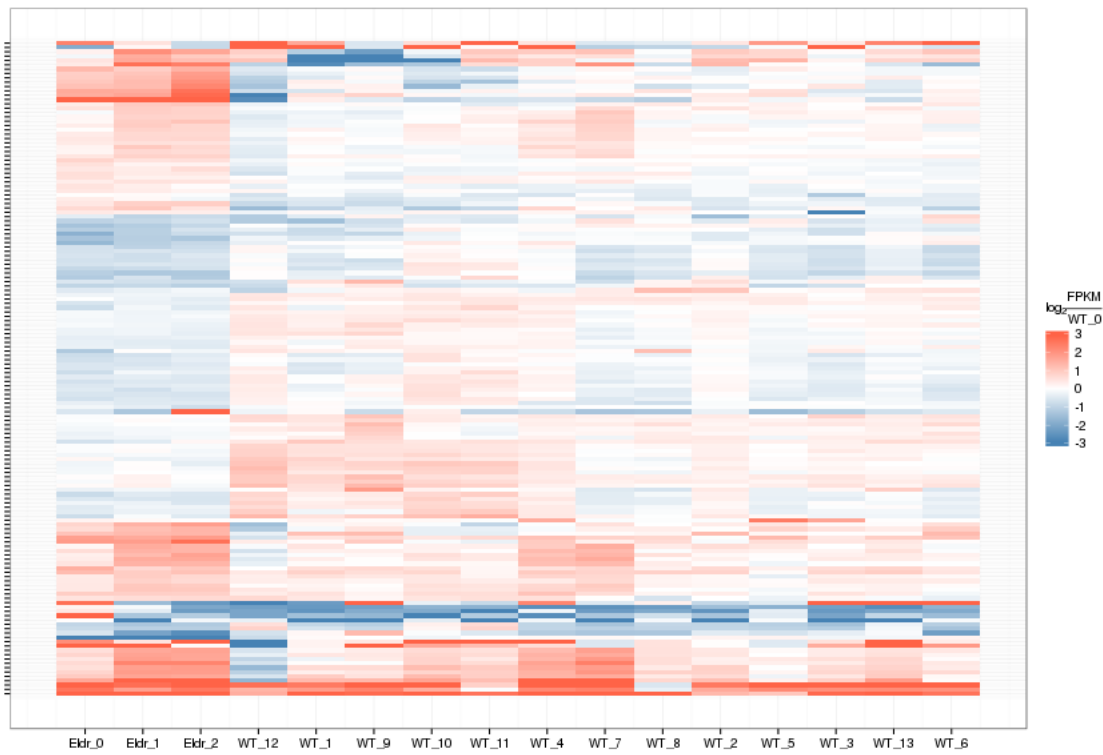
Volcano Plot



Volcano plot with significant genes only:



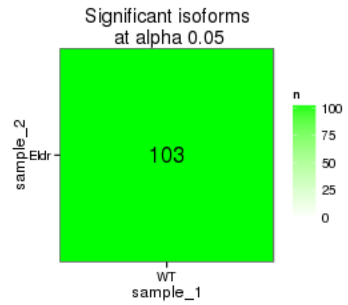
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

iso	Annotation	gene_short_name
1	Cdh1	Cdh1
2	Col1a1	Col1a1
3	Lamb1	Lamb1
4	Nid1	Nid1
5	Ttc9b	Ttc9b
6	Cad	Cad
7	Abca1	Abca1
8	Lama5	Lama5
9	Dync1h1	Dync1h1
10	Lama2	Lama2
11	Gm872	Gm872
12	Egfr	Egfr
13	Pold2	Pold2
14	Mrc2	Mrc2
15	Hoxb9	Hoxb9
16	Nid2	Nid2
17	Myh9	Myh9
18	Dlx2	Dlx2
19	Igf2r	Igf2r
20	Fbn2	Fbn2
21	Rps14	Rps14
22	Fasn	Fasn
23	Lbx1	Lbx1
24	Pgm2	Pgm2
25	Dst	Dst
26	Fn1	Fn1

27 Lamc1
28 Col5a1
29 Notch1
30 Lrp2
31 Fbn1
32 Notch2
33 Tnc
34 Svep1
35 Snapc3
36 Scp2

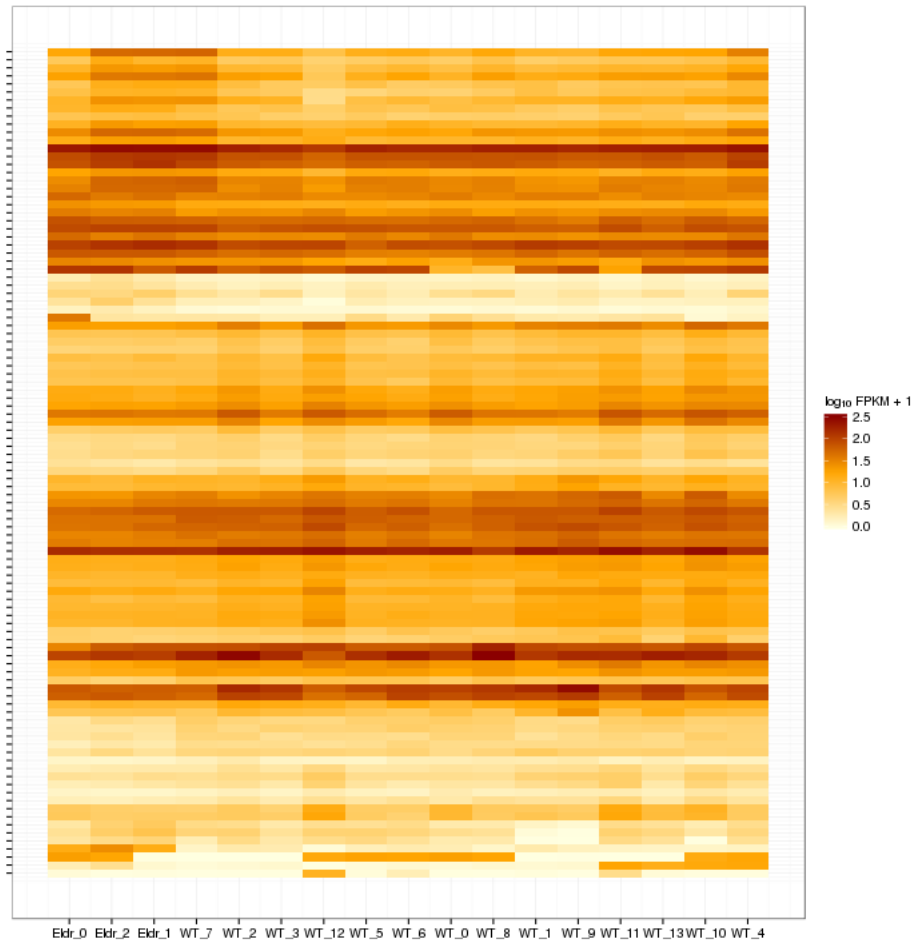
37 Kcnq4
38 Trp73
39 Uncx
40 Col1a2
41 Slc13a4
42 Itpr1
43 Dera
44 Eif3f
45 Col12a1
46 Lama1
47 Szt2
48 Adamts15
49 Gbx2
50 Prr7
51 Fras1
52 Leng8
53 Cox8a
54 Frem2
55 Vstm2l
56 Notch3
57 Hoxb4
58 Pcsk1n
59 Kihl42
60 Lrp1
61 Tlx3
62 C1qtnf4
63 Dnah7b

64 2310036O22Rik

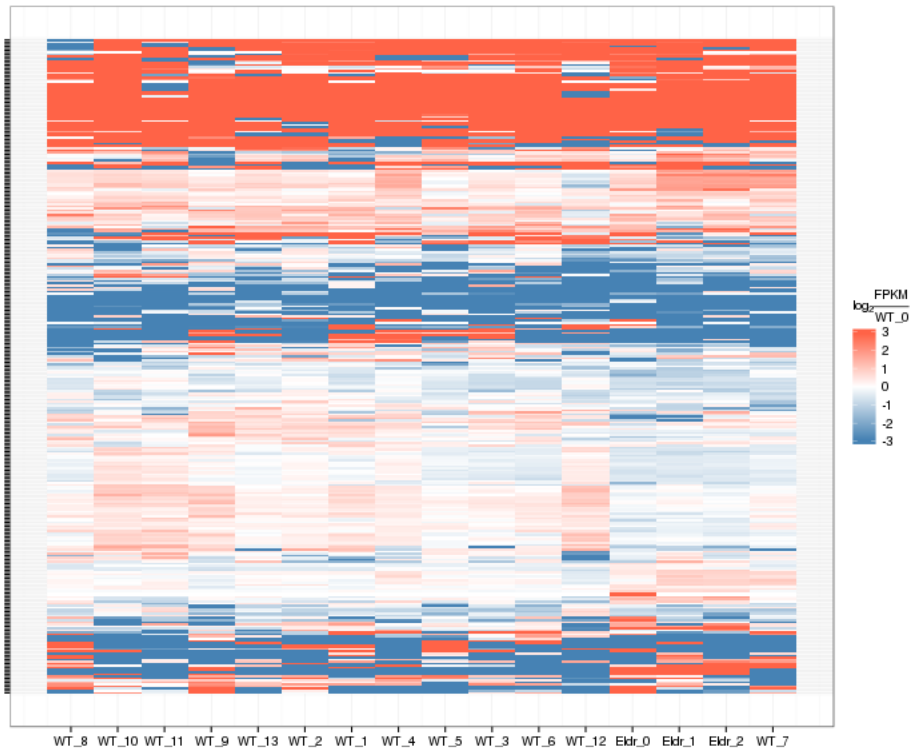
65 Reln
66 Hebp1
67 Tuba1c
68 Gm9493

69 Wdr89
70 Fat4
71 Col6a3
72 H2afx
73 Ccdc171
74 Lamb2
75 Tusc1
76 Zfp771
77 Gm10020
78 Rpl36
79 H2afj
80 Tpt1
81 Rps12
82 Sox11
83 Ptpnz1
84 Ddx3y
85 Gpr98
86 Ahnak
87 Fat1
88 Kih19
89 Jund
90 Gpr27
91 Srp54a
92 Hbb-bt
93 Fat3
94 Gm3558
95 Gm2897
96 Gm11273
97 Ccdc85c
98 Ier5l
99 Malat1
100 Ccdc85b
101 Gm3500
102 Gm9825
103 Gm3591

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

Biocarta enrichment:

```
## Error: `x` must have at least 2 rows and 2 columns
```

Biocarta zscore:

```
## Error: incorrect number of dimensions
```

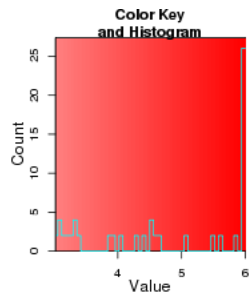
```
## Error: object 'x_ordered' not found
```

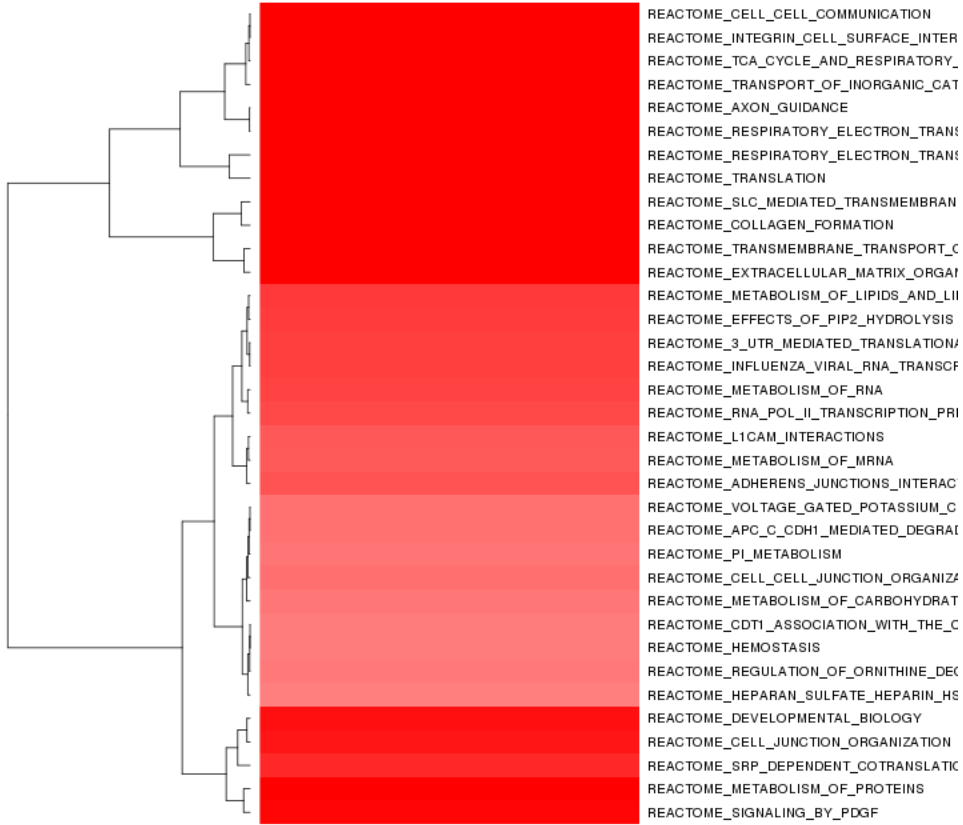
```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Reactome enrichment:

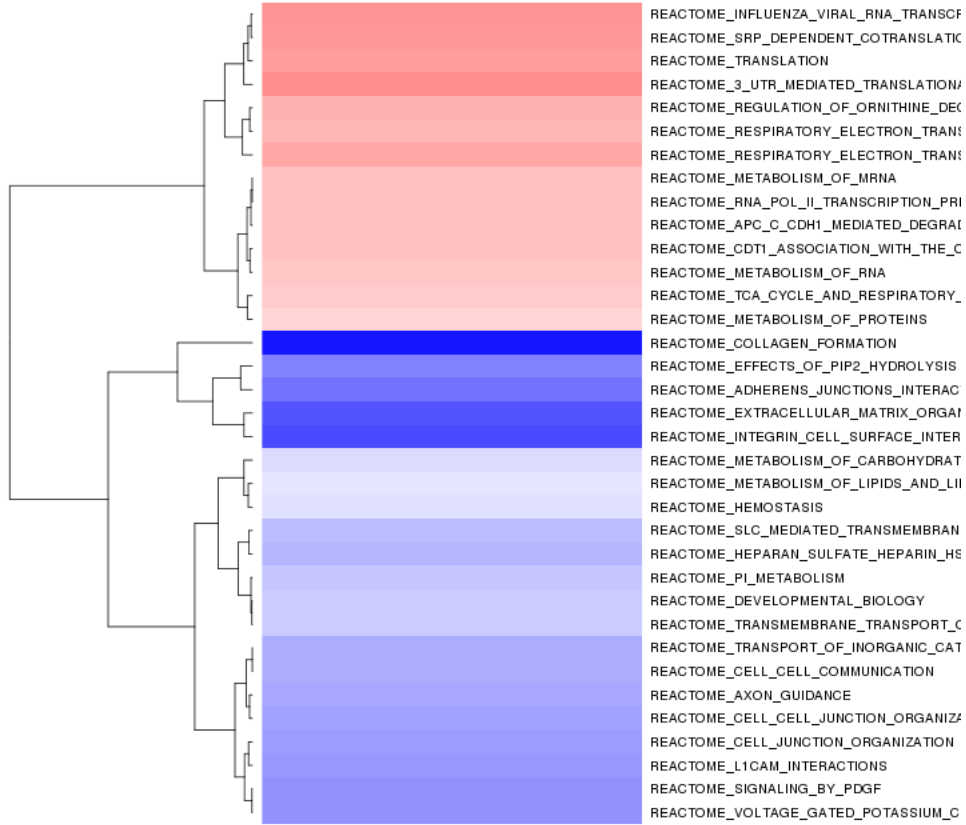
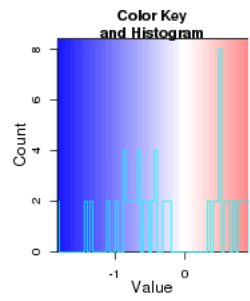
```
## Error: subscript out of bounds
```



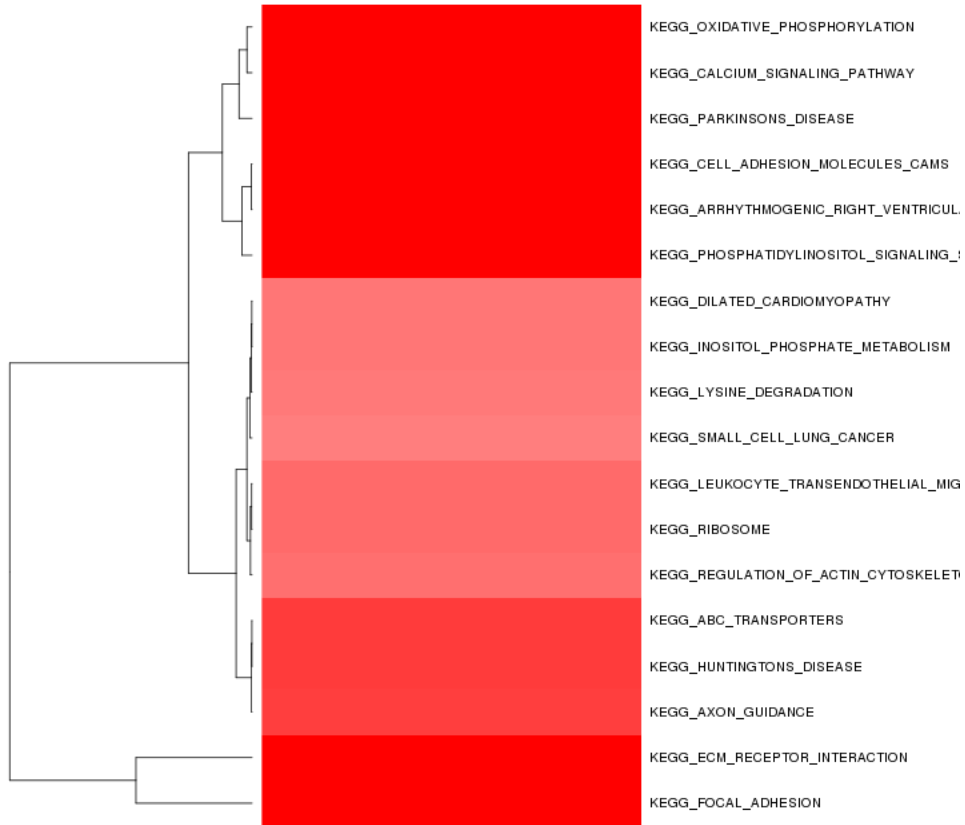
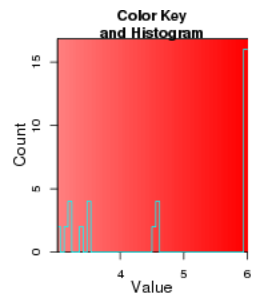


Reactome zscore:

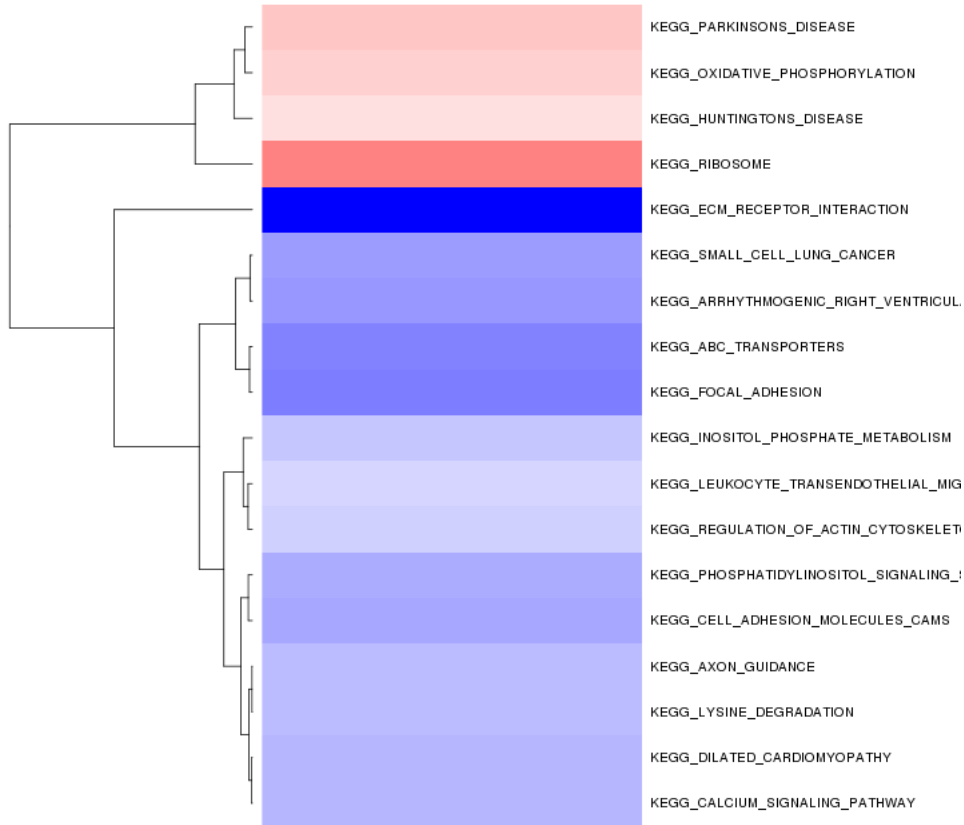
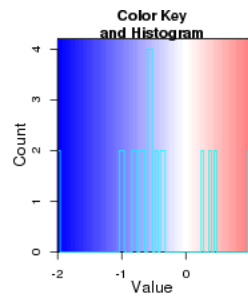
Error: subscript out of bounds



Kegg enrichment:

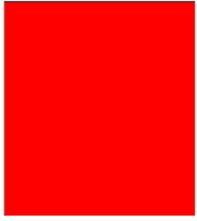


Kegg zscore:



Interneuron enrichment:

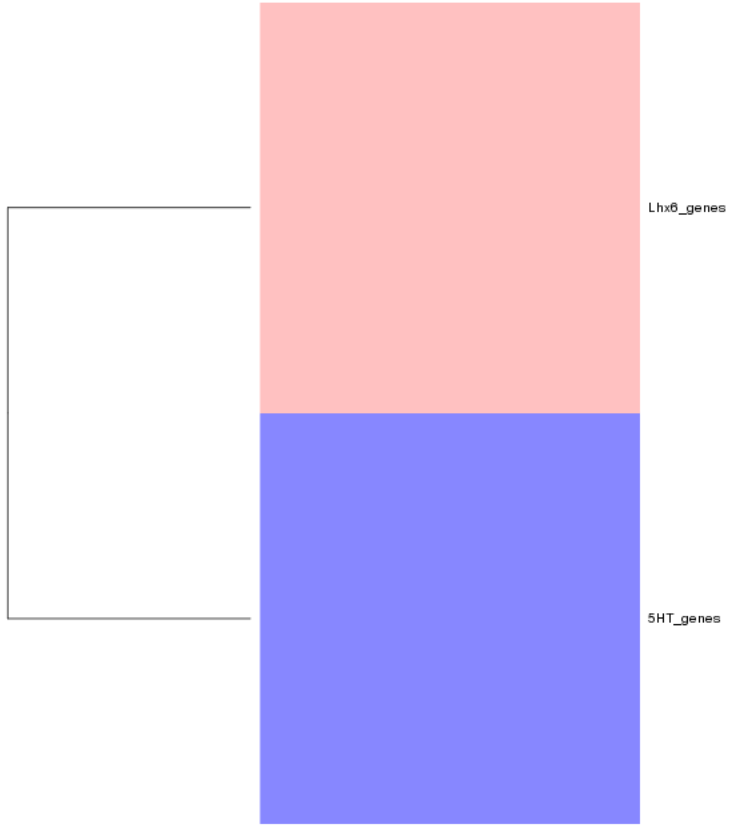
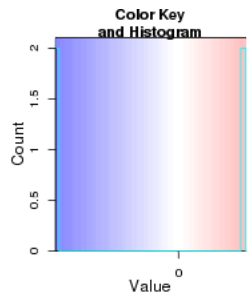
Error: no locations are finite



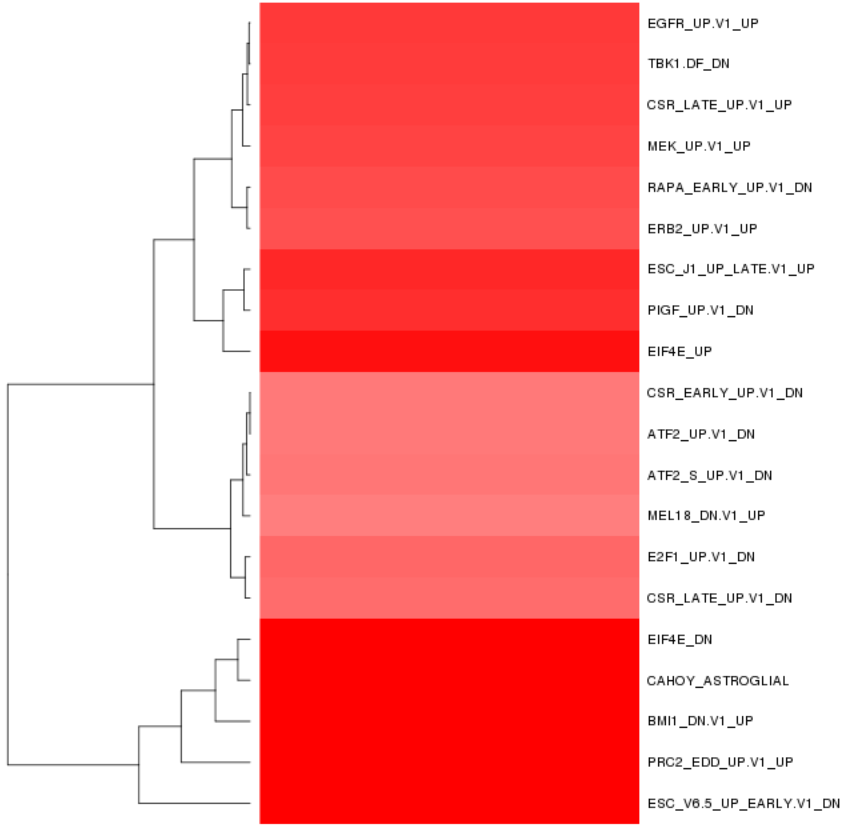
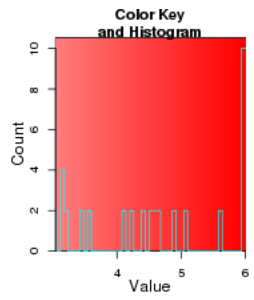
5HT_genes

Lhx6_genes

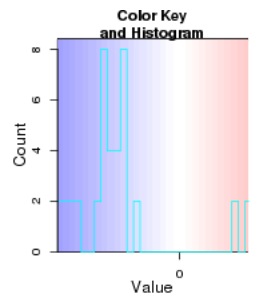
Interneuron zscore:

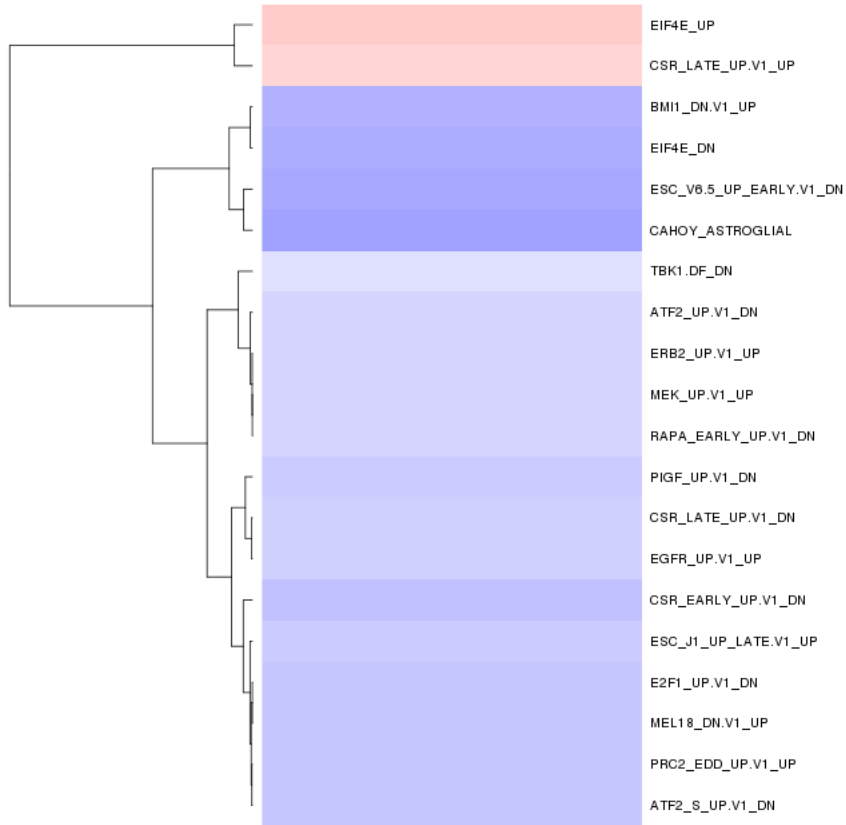


Oncogene enrichment:

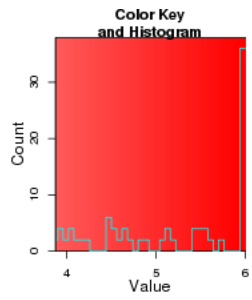


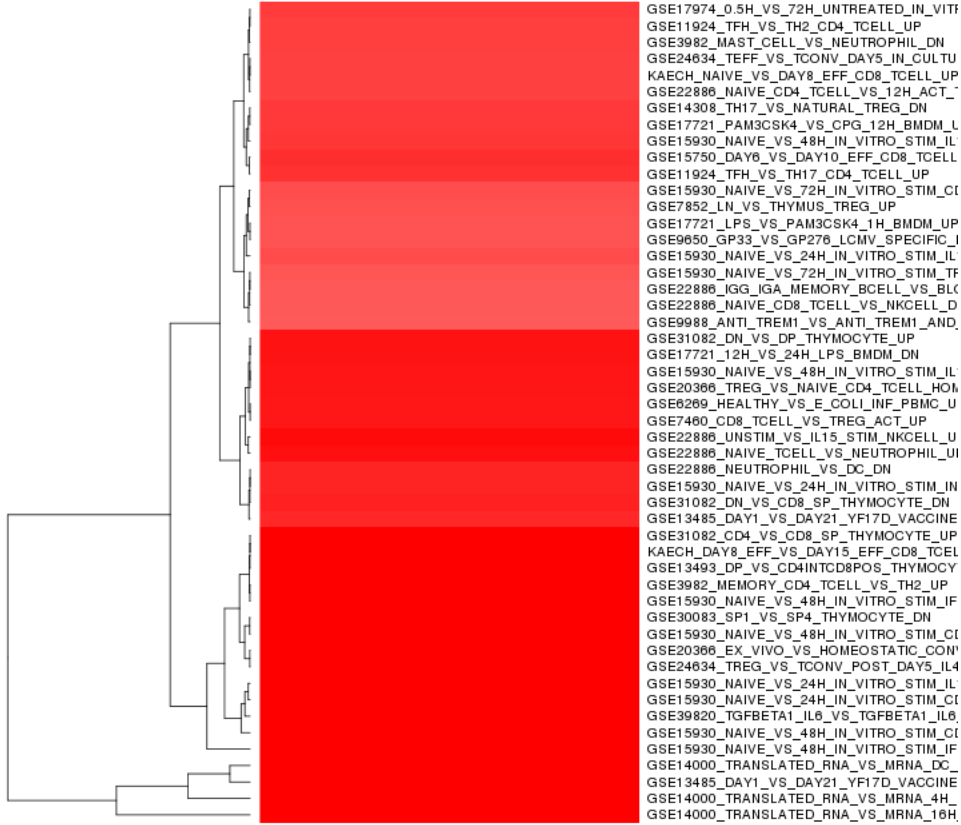
Oncogene zscore:



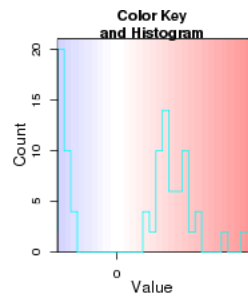


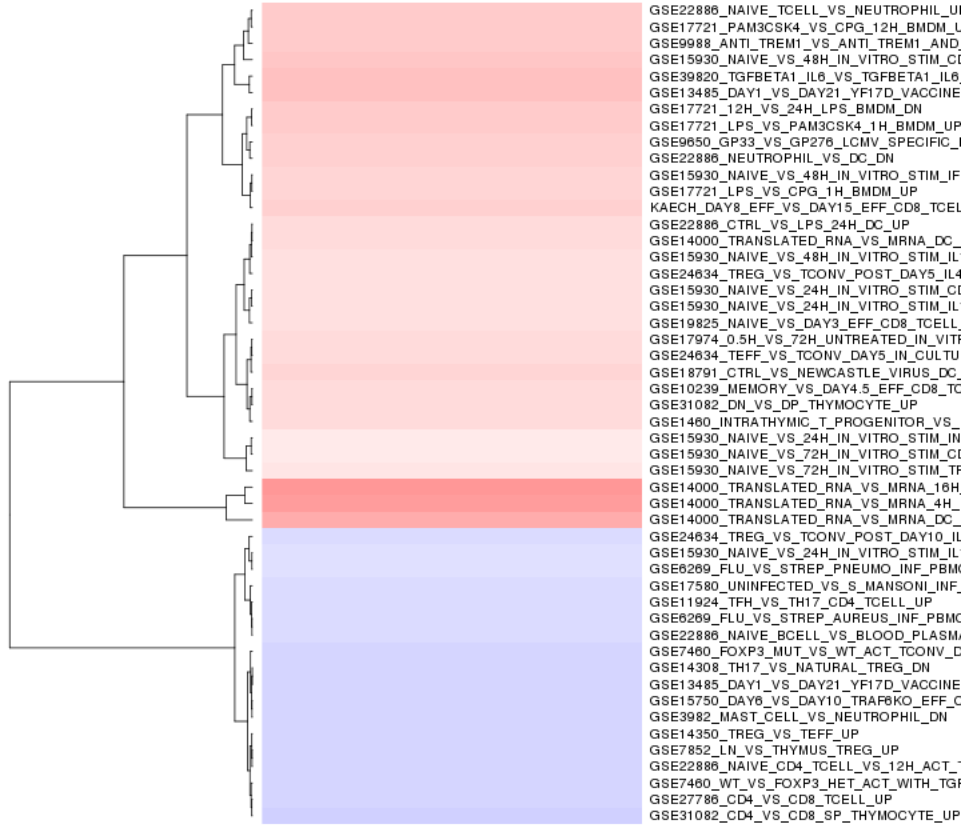
Immuno enrichment:





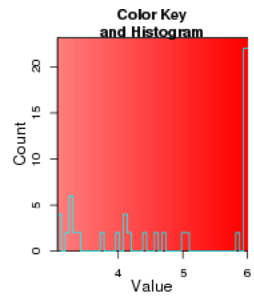
Immuno zscore:

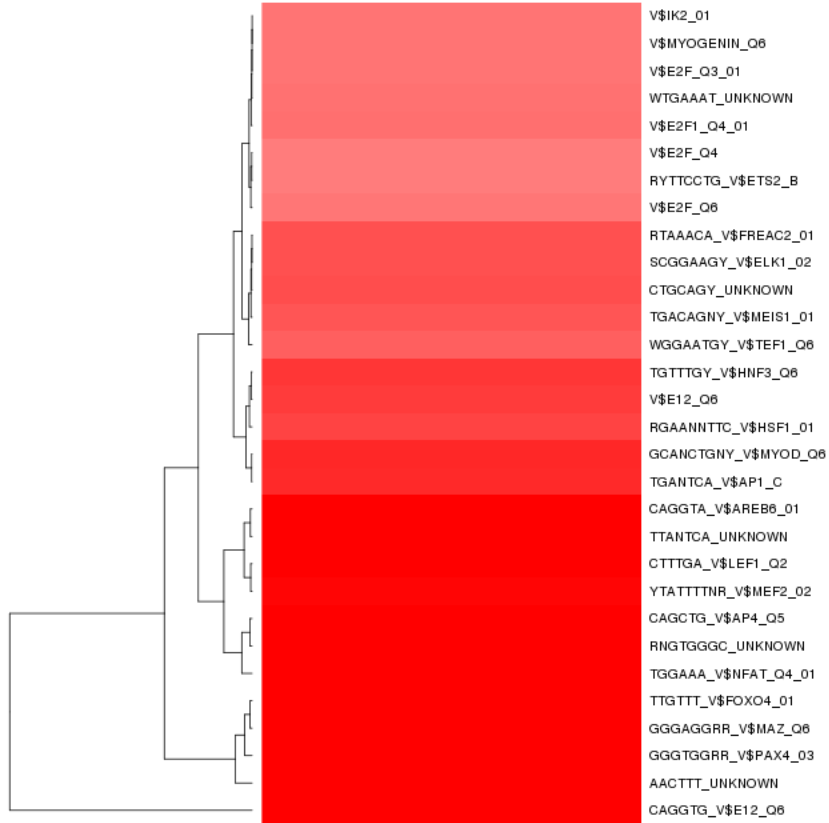




TF enrichment:

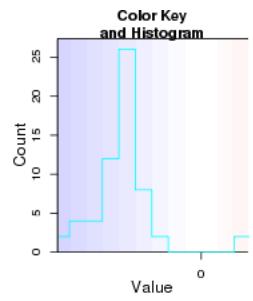
Error: subscript out of bounds

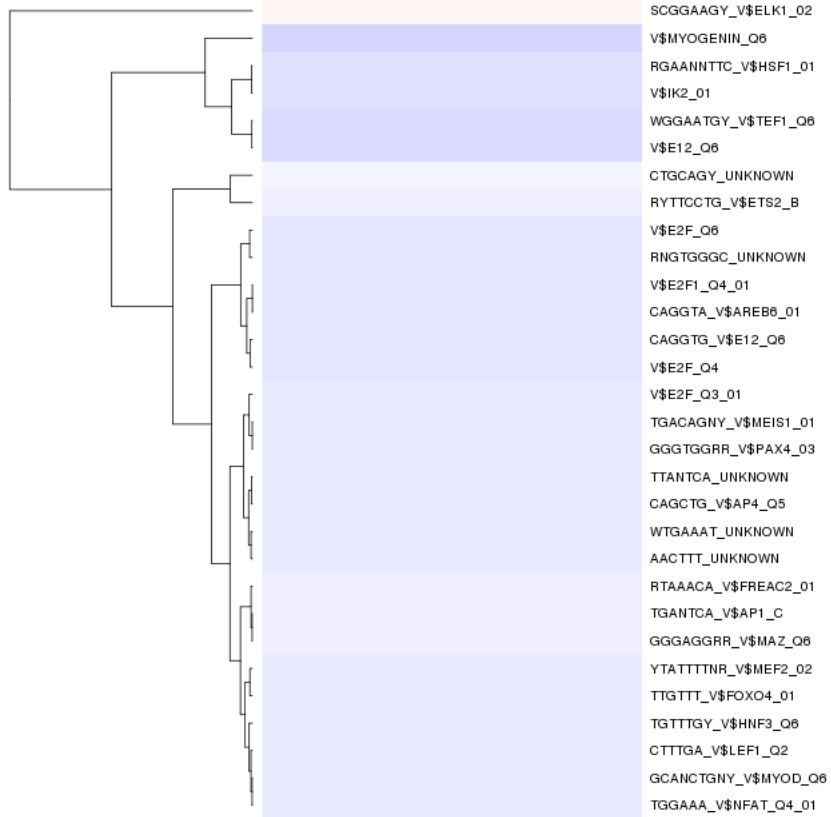




TF zscore:

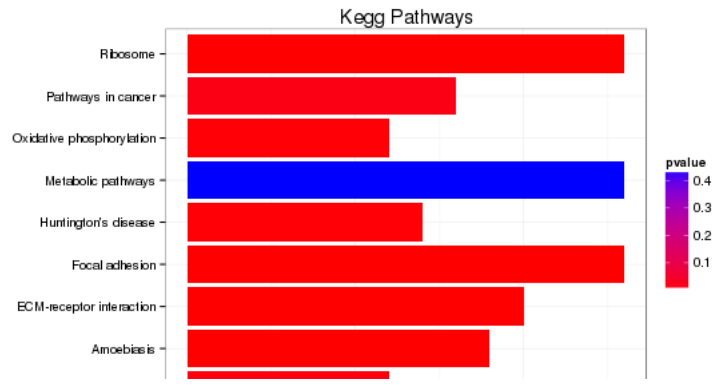
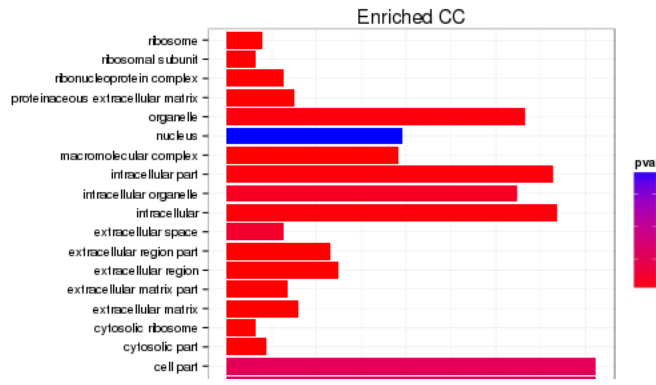
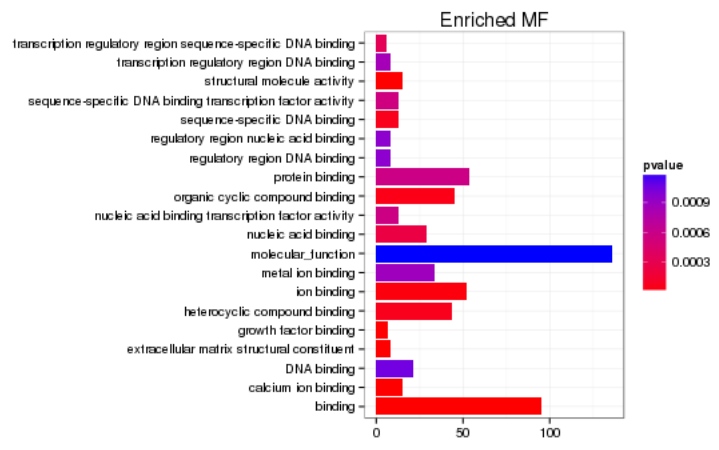
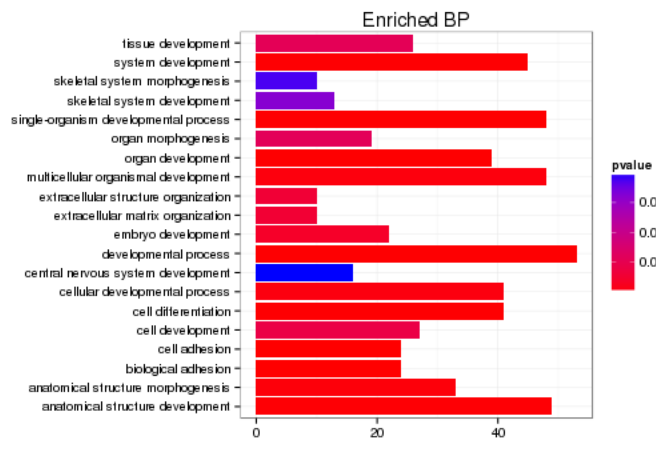
Error: subscript out of bounds

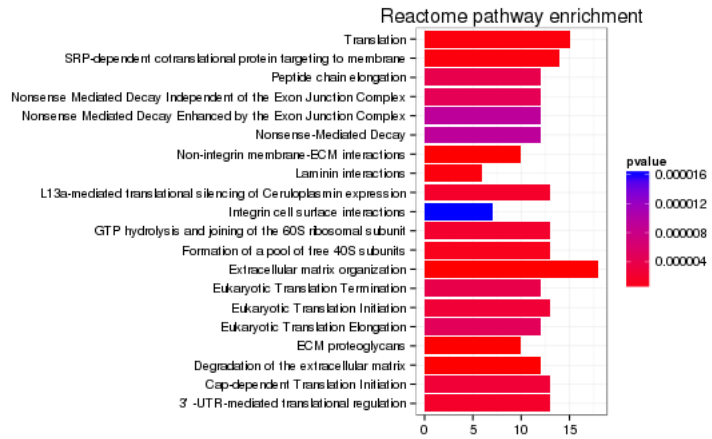
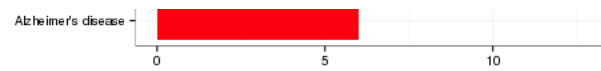
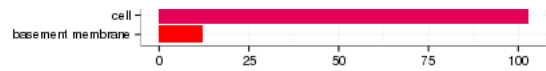




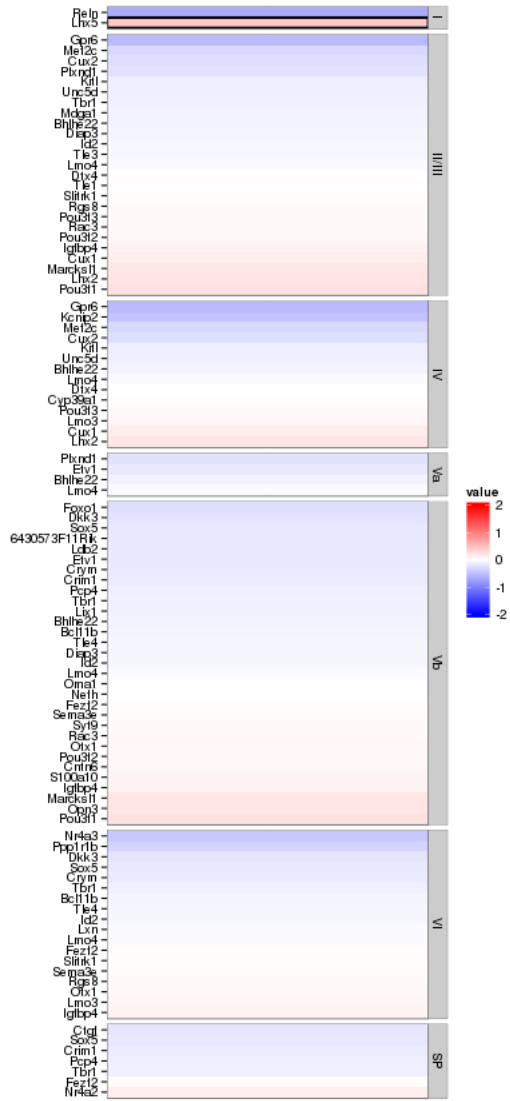
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

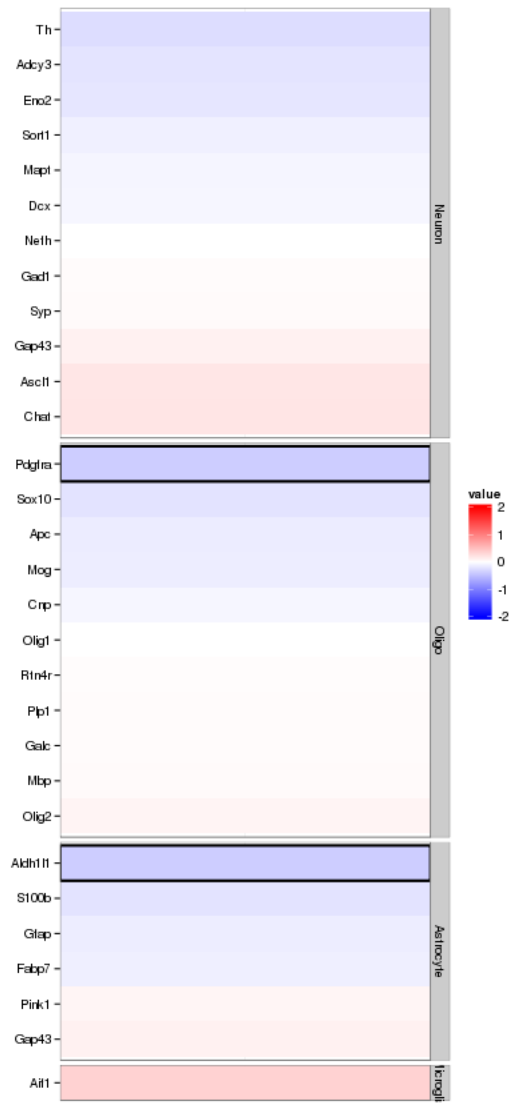




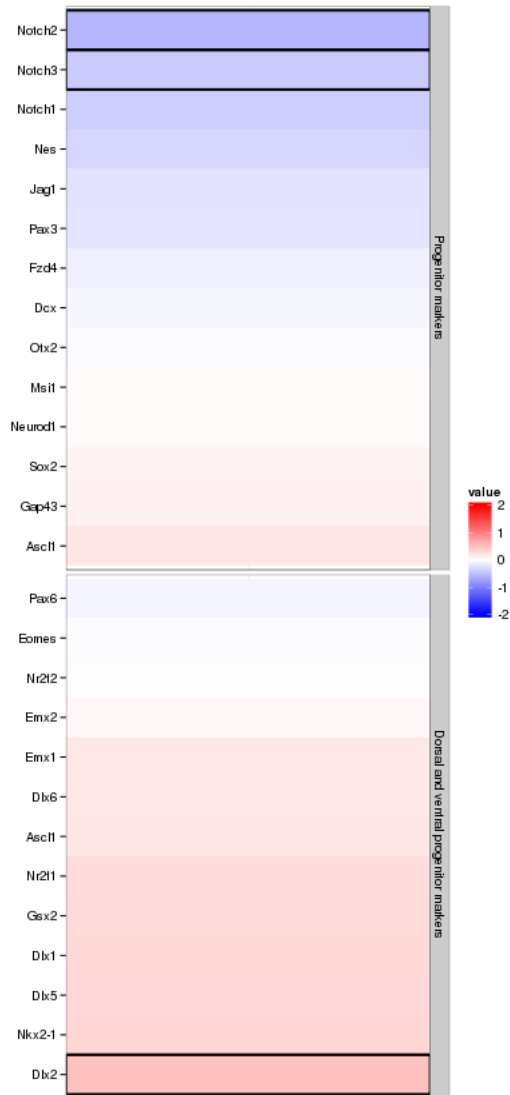
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



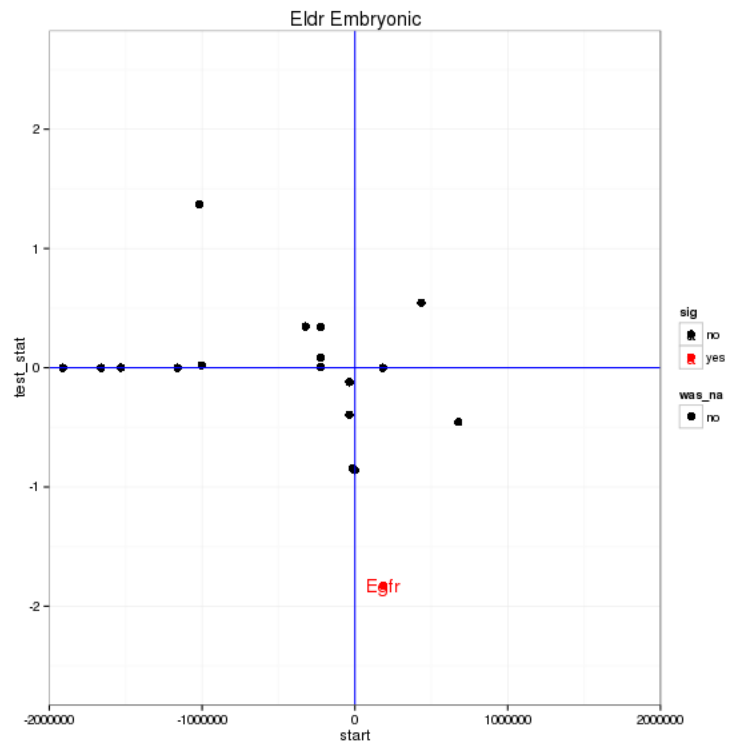
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 1 genes significantly regulated in a region this size is: 0.3083



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR787
16 JR810
17 JR838

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	32543000.00	1.25	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	32543000.00	1.09	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	32543000.00	1.22	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	32543000.00	1.27	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	32543000.00	1.19	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	32543000.00	0.88	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	32543000.00	0.96	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	32543000.00	0.91	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	32543000.00	0.85	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	32543000.00	0.95	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	32543000.00	0.80	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	32543000.00	0.79	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	32543000.00	0.68	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	32543000.00	1.20	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR787/abundances.cxb	Eldr	0	Eldr_0	26894700.00	32543000.00	0.83	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR810/abundances.cxb	Eldr	1	Eldr_1	41828800.00	32543000.00	1.29	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR838/abundances.cxb	Eldr	2	Eldr_2	37002600.00	32543000.00	1.15	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```



```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Eldr -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Eldr_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/
## 2
## 3
## 4
## 5
```

Haunt KO vs WT (Adult)

This file shows the wt-v-ko comparison for Haunt.

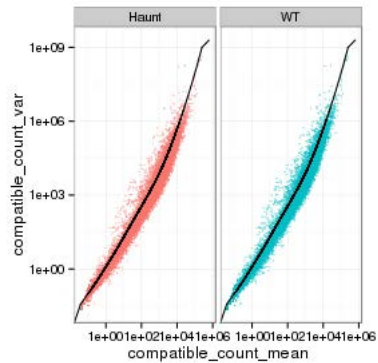
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

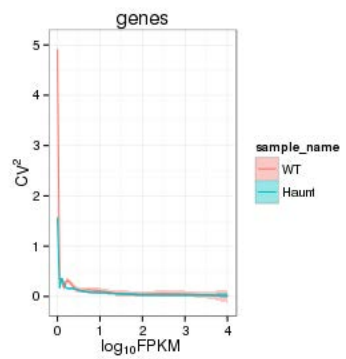
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



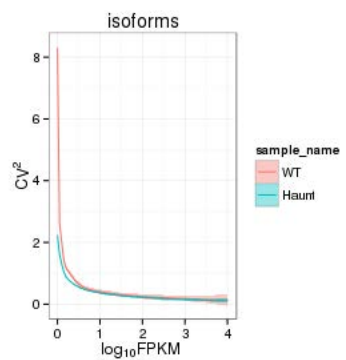
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

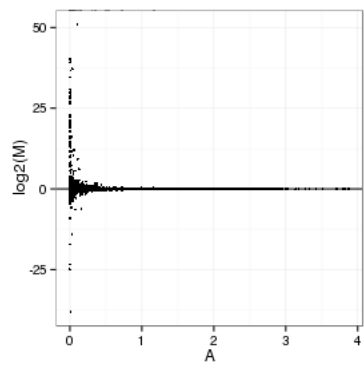
Genes:



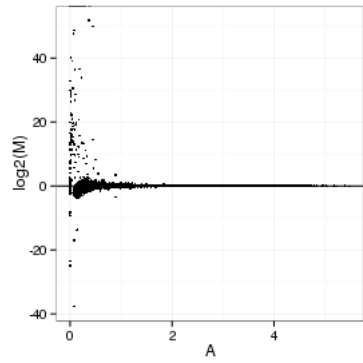
Isoforms:



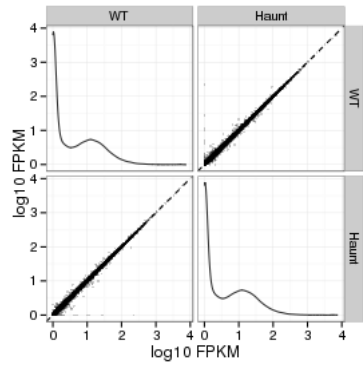
MvA plot



MvA plot counts



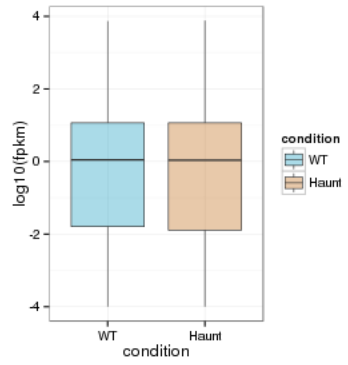
Scatterplot matrix



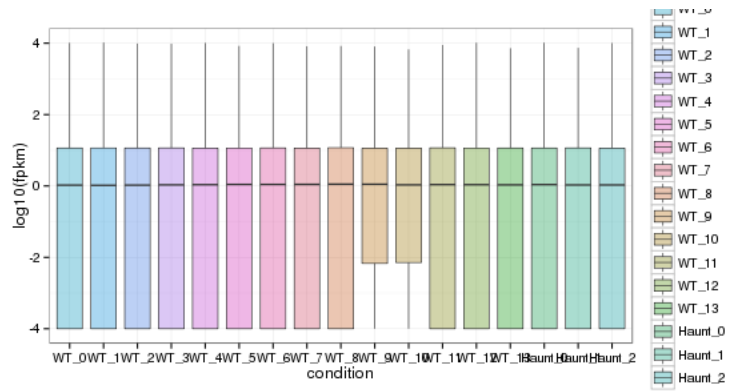
Distributions

Boxplots

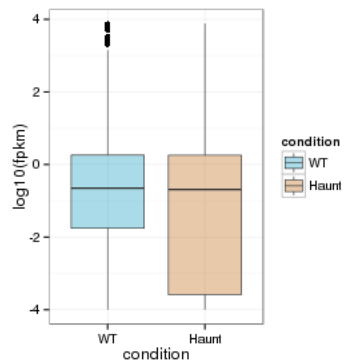
Boxplot (genes)



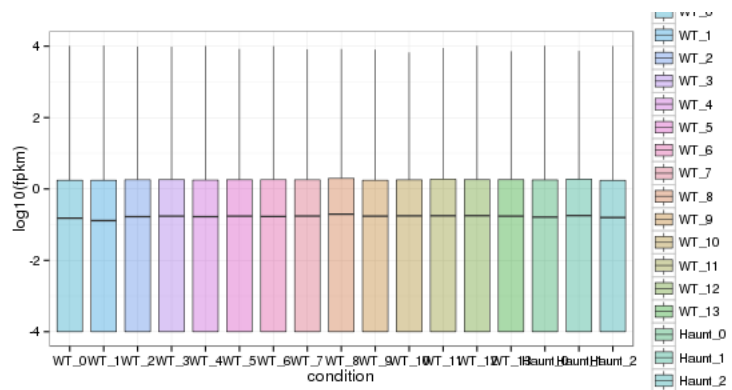
Boxplot (genes, replicates)



Boxplot (isoforms)

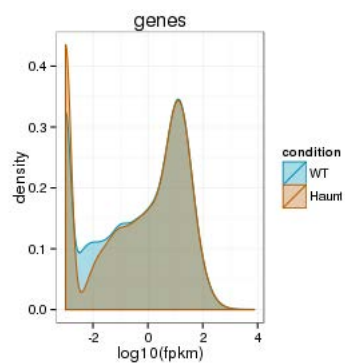


Boxplot (isoforms, replicates)

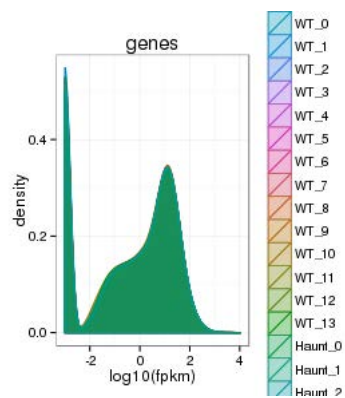


Density

Density (genes)

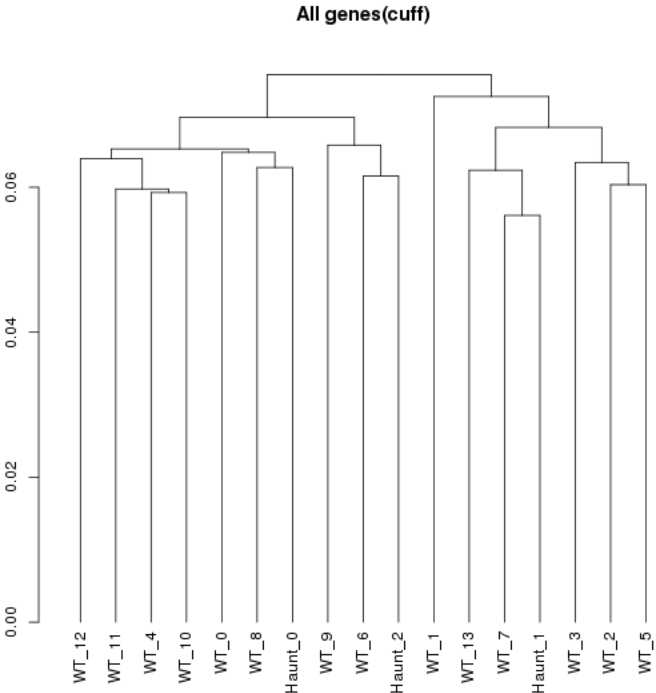


Density (genes, replicates)



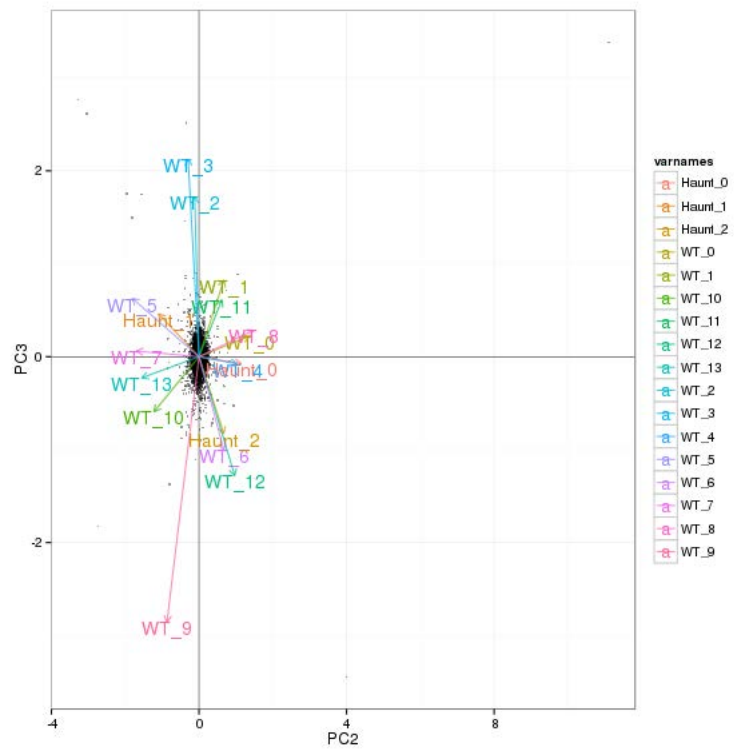
Clustering

Replicate Clusters

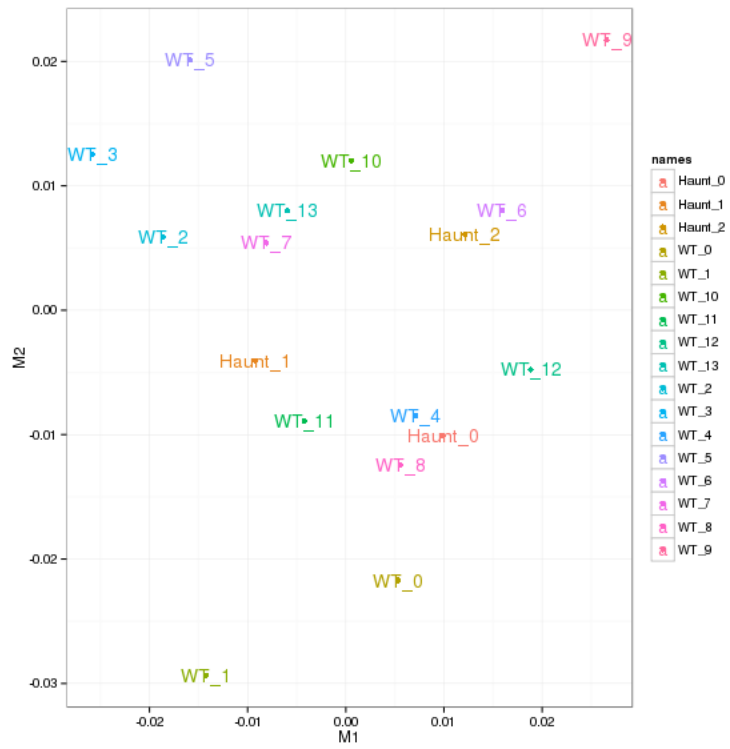


NULL

PCA (genes)

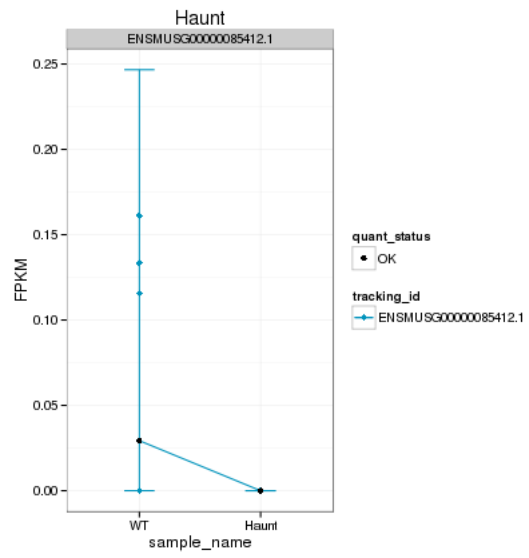


MDS (genes)

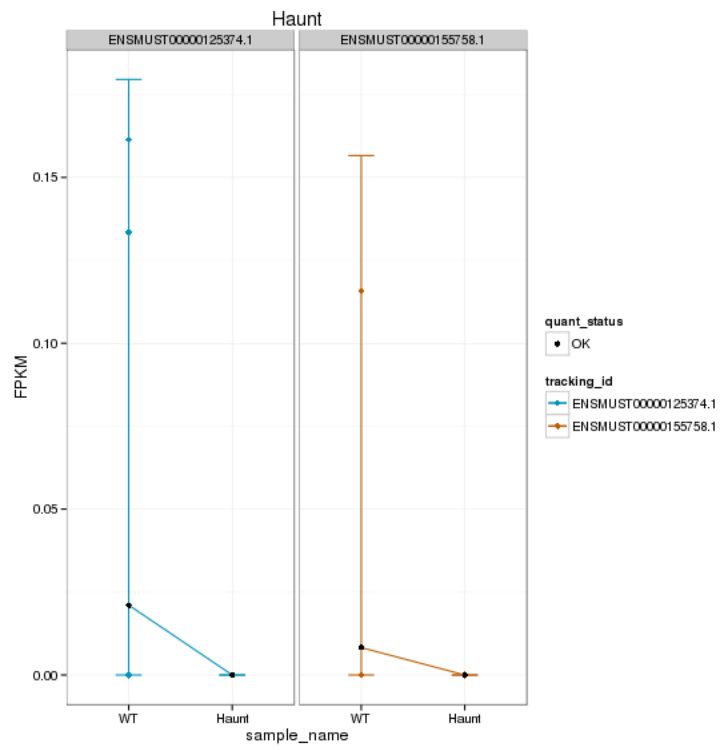


KO assessment

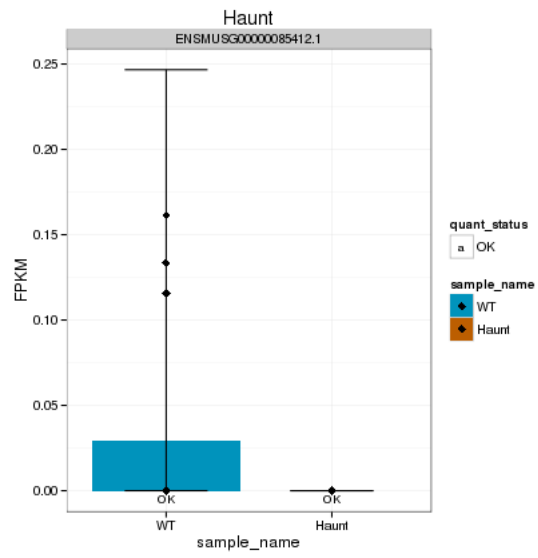
Endogenous lncRNA expression



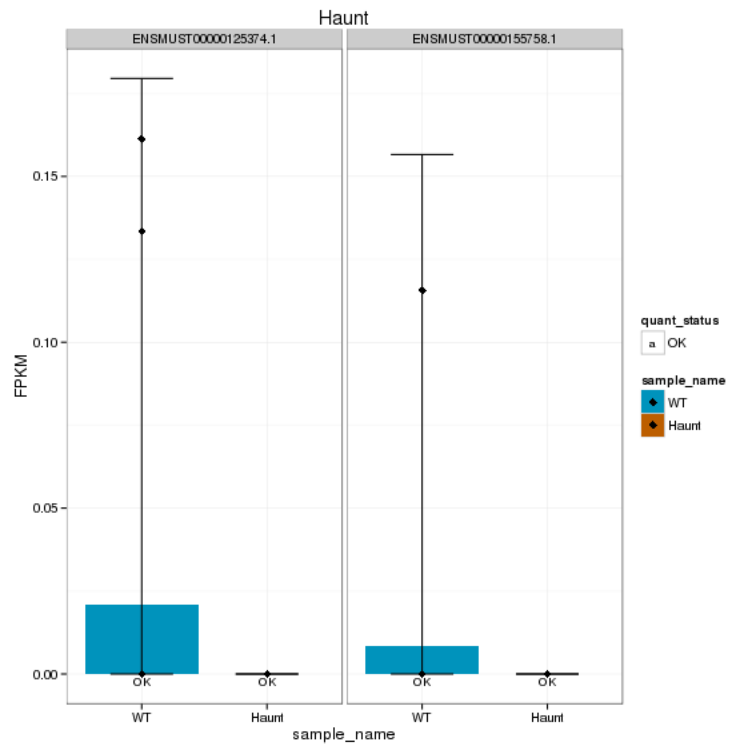
Endogenous expression of Haunt isoforms:



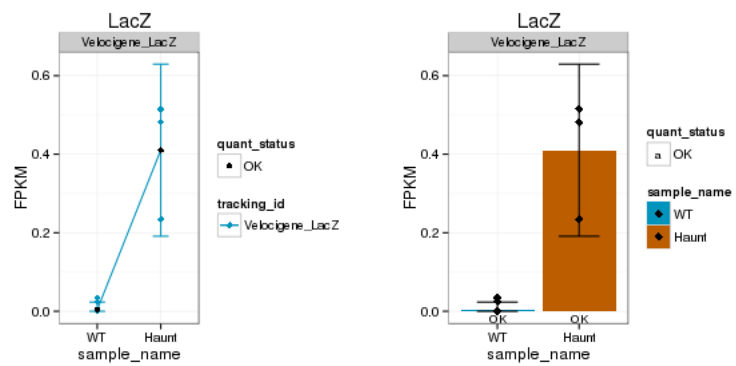
Barplot of gene expression:



Barplot of isoform expression:

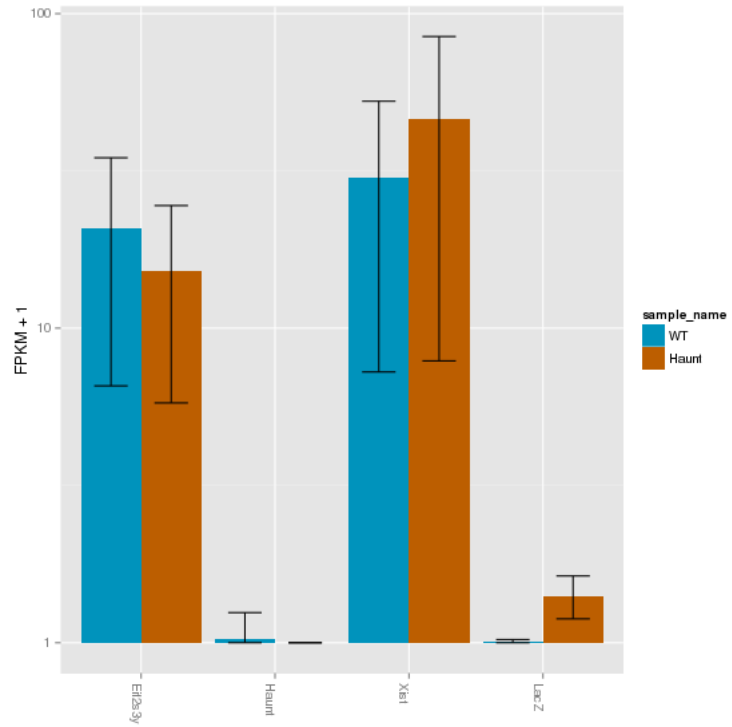


LacZ expression

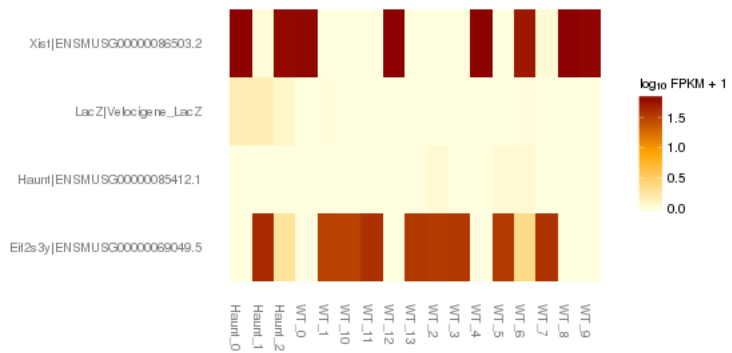


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



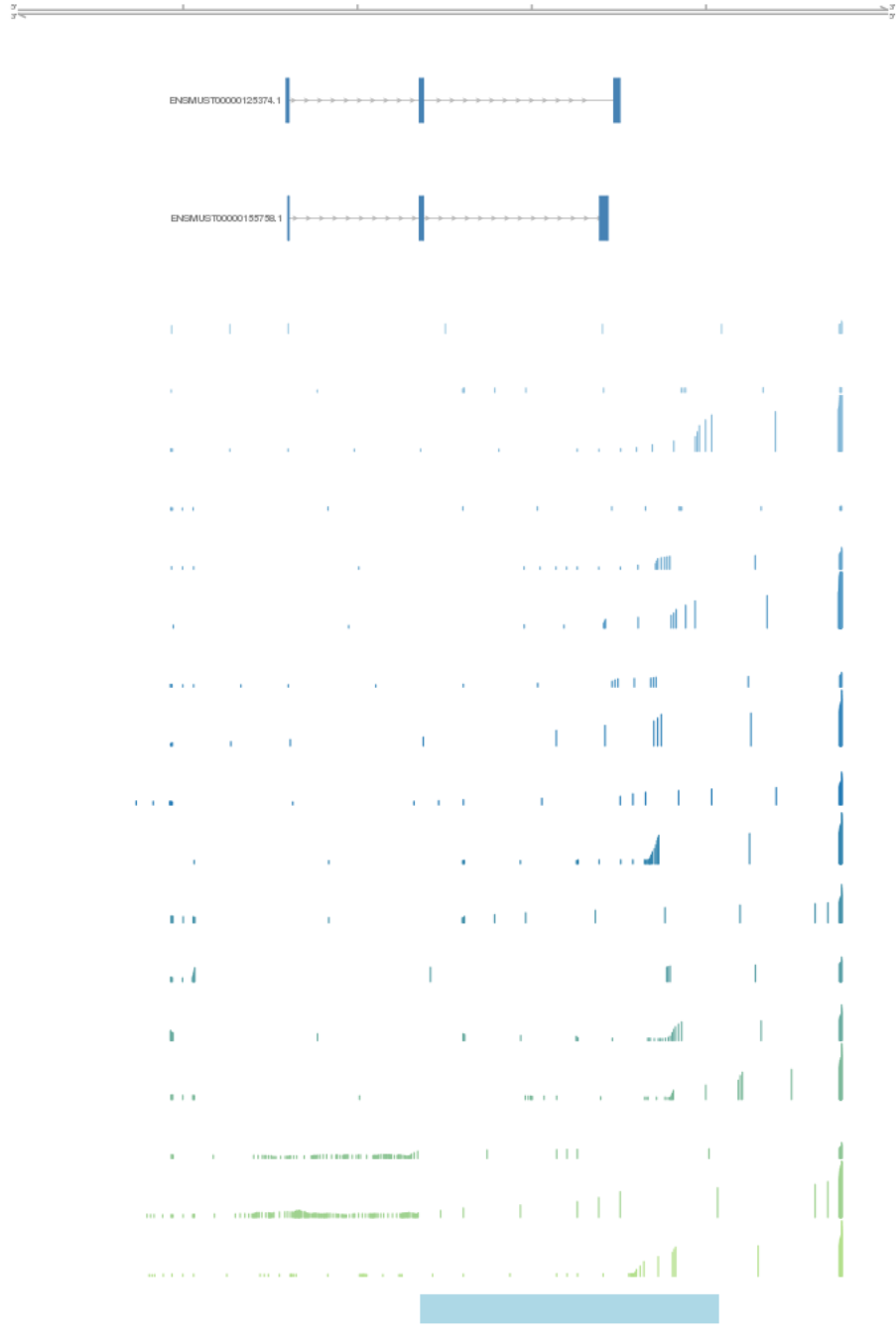
Expression heatmap:



Track visualization



KO Region Hsm1L2 Hsm1L1 Hsm1L0 WT_15 WT_12 WT_11 WT_10 WT_9 WT_8 WT_7 WT_6 WT_5 WT_4 WT_3 WT_2 WT_1 WT_0 LncRNA Isoforms



Differential Analysis

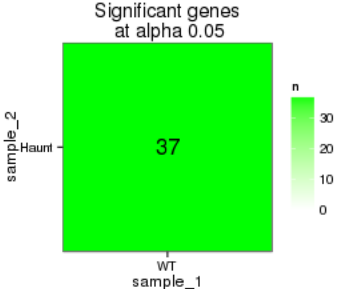
Differential Genes

There are 37 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 Hspa8
2 Ubb
3 Dusp6
4 Pttg1
5 Adi1
6 Serpina3n
7 Fos
8 Arc
9 Nr4a1
10 Glo1
11 Xdh
12 Nr4a3
13 Hddc3
14 Cryab
15 Arhgap27
16 Fam214a
17 Filip1
18 Klf10
19 Egr2
20 Txnip
21 Egr1
22 Akap12
23 Atp5l
24 Maff
25 Gm9493
26 Acp1
27 Npas4
28 Hbb-bs
29 Junb
30 Kdm5d
31 Tpm3-rs7
32 Rpl34
33 Ddx3y
34 Egr4
35 Gm7292
36 AA465934
37 Gm26825
```

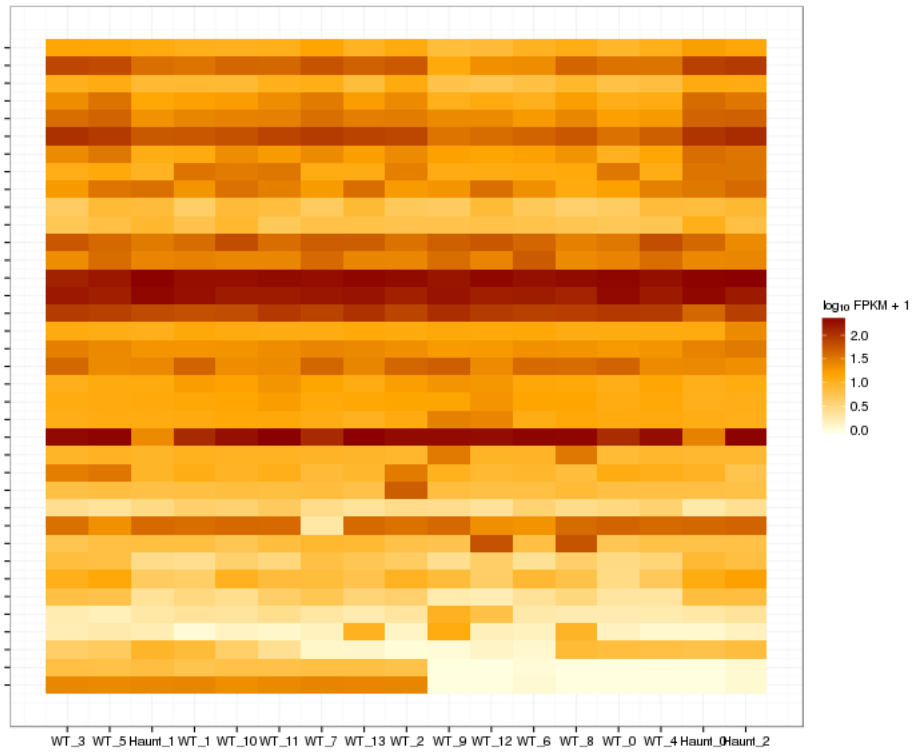
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

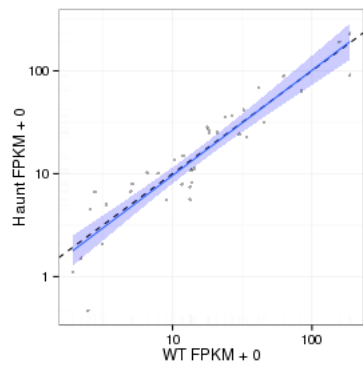


Significant genes with expression >50fpkm (any condition):(turned off)

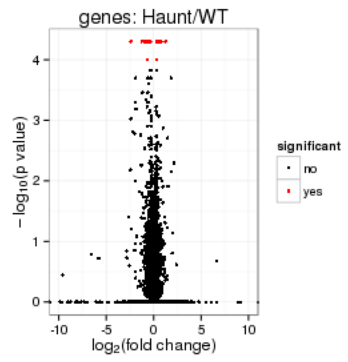
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

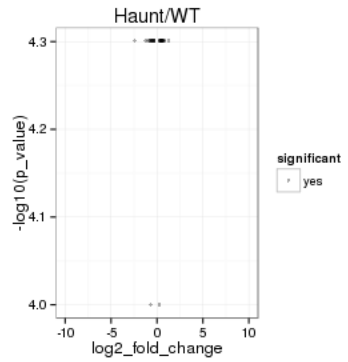
Scatter plot of significant genes only:



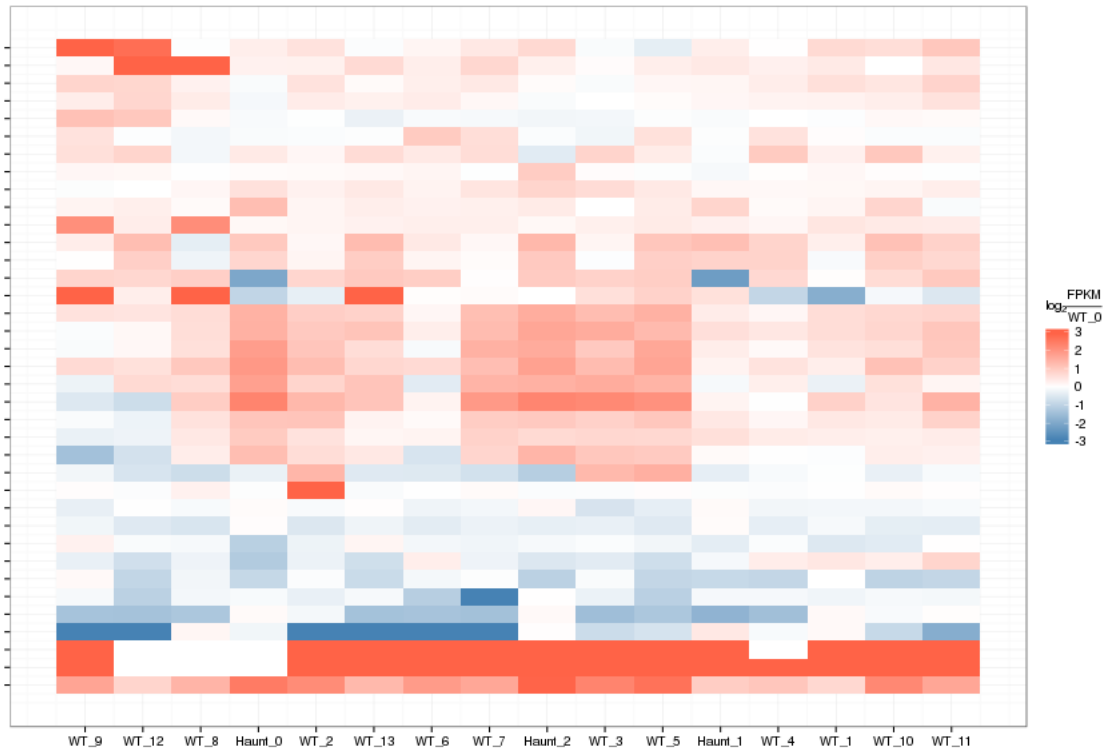
Volcano Plot



Volcano plot with significant genes only:



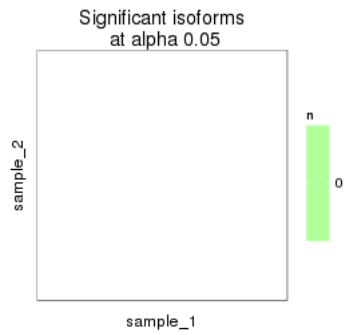
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

[1] "no sig isoforms"

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

Biocarta enrichment:

```
## Error: `x` must have at least 2 rows and 2 columns
```

Biocarta zscore:

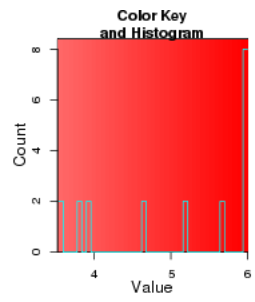
```
## Error: incorrect number of dimensions
```

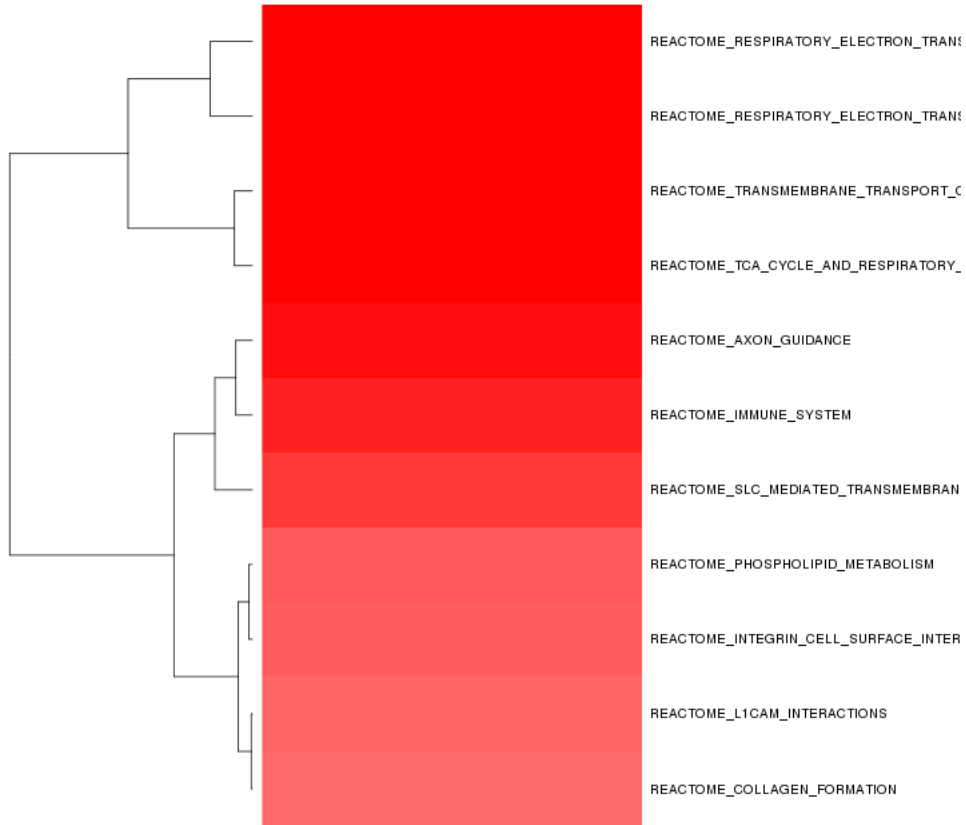
```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

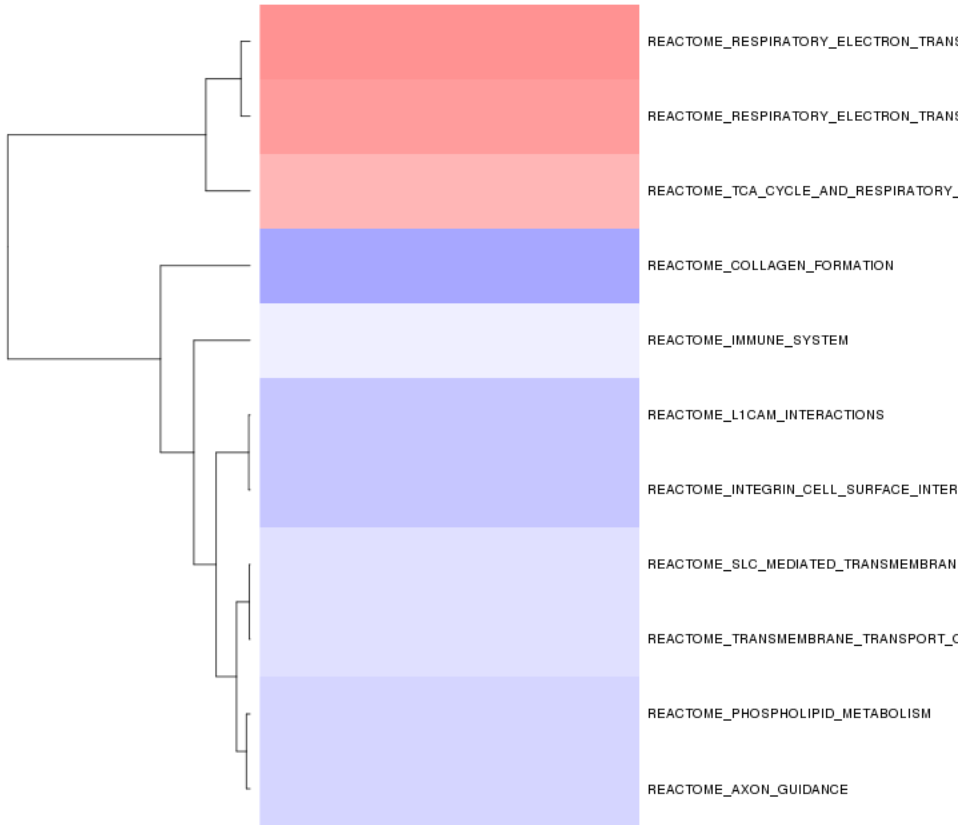
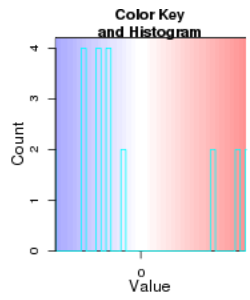
```
## Error: object 'x_ordered' not found
```

Reactome enrichment:

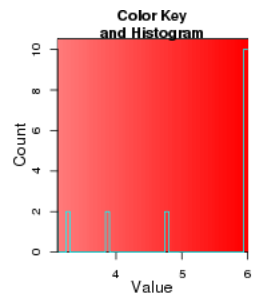


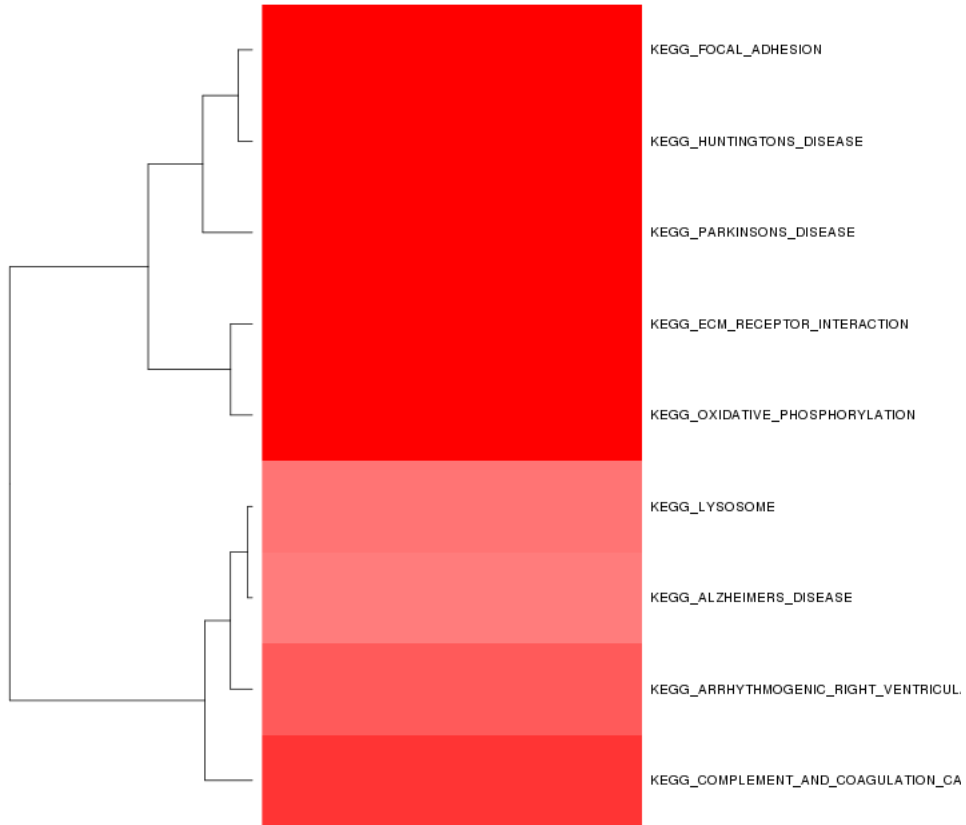


Reactome zscore:

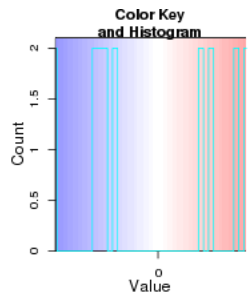


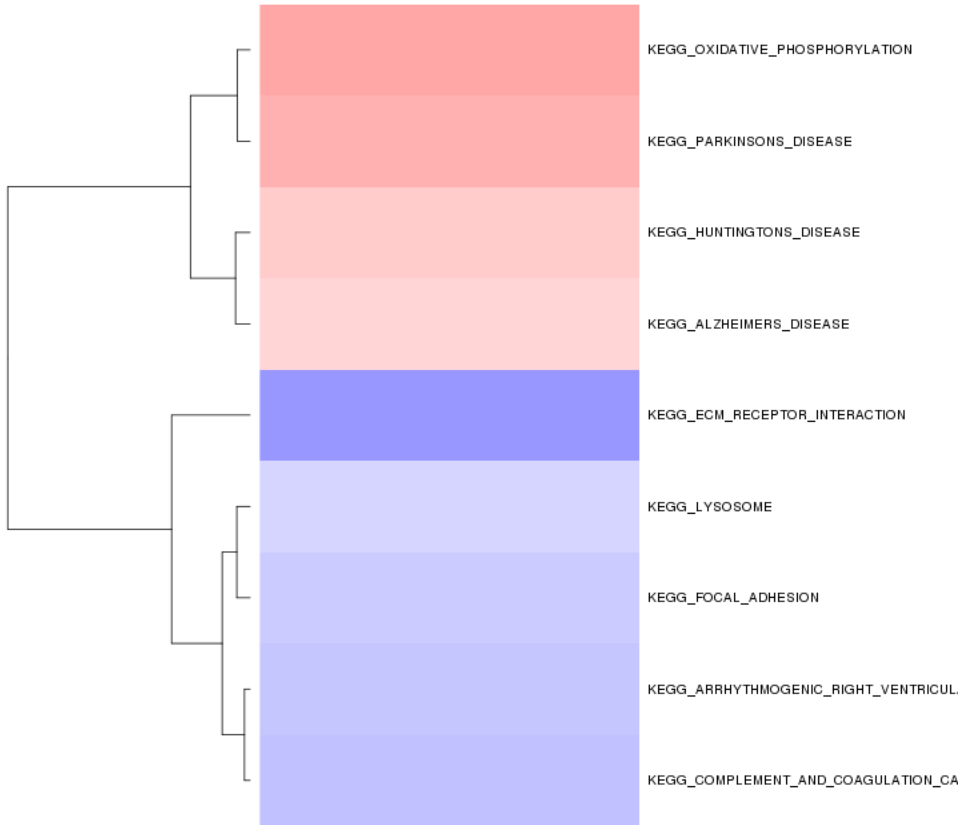
Kegg enrichment:



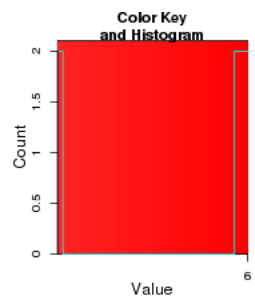


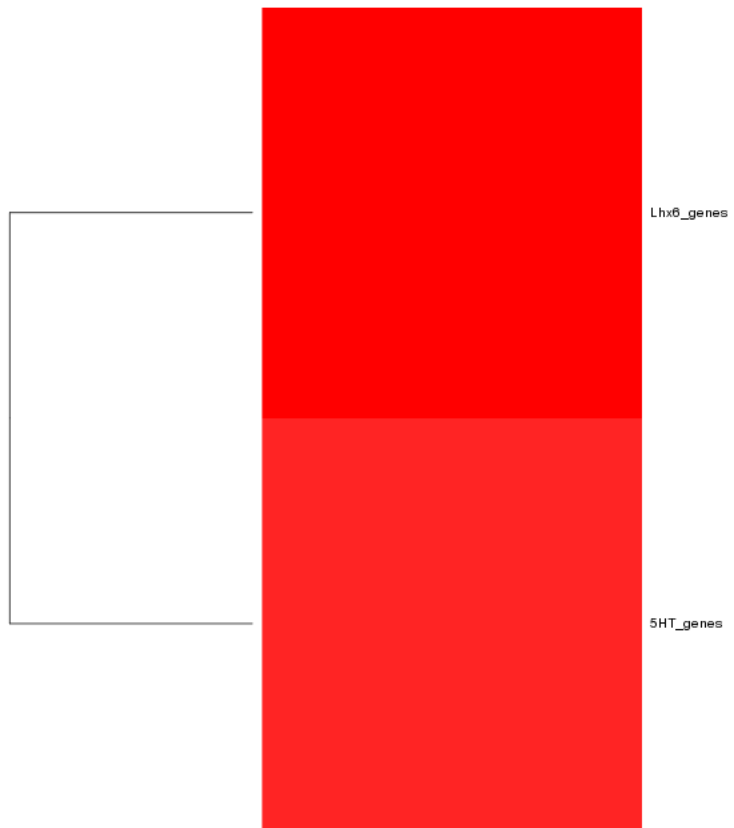
Kegg zscore:



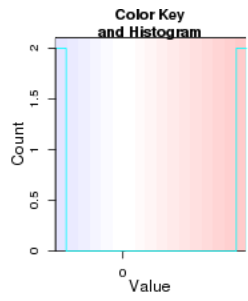


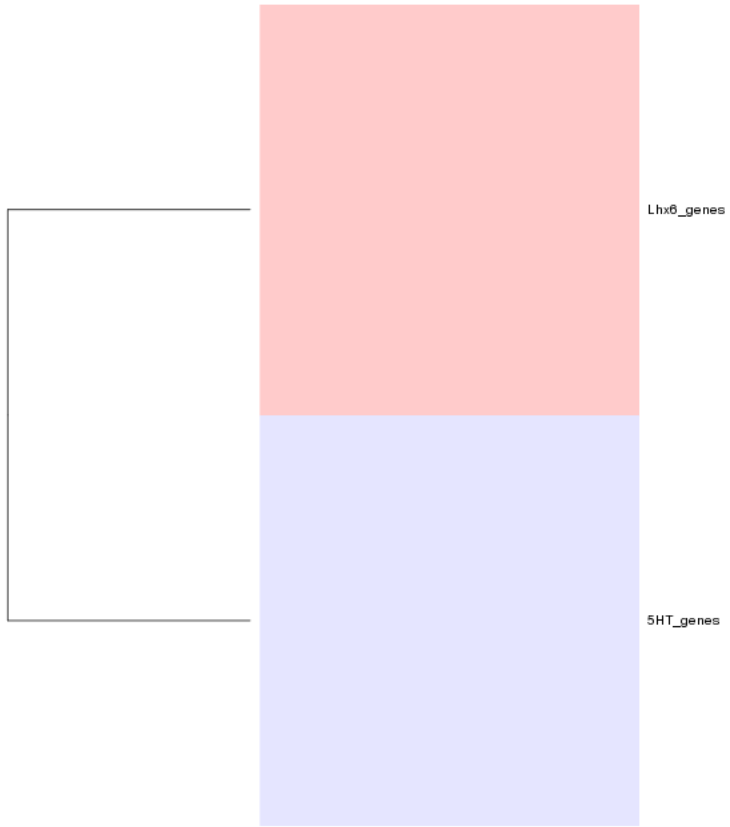
Interneuron enrichment:



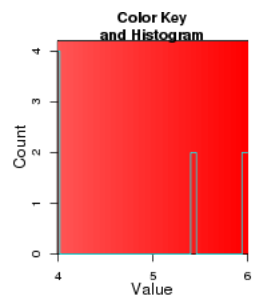


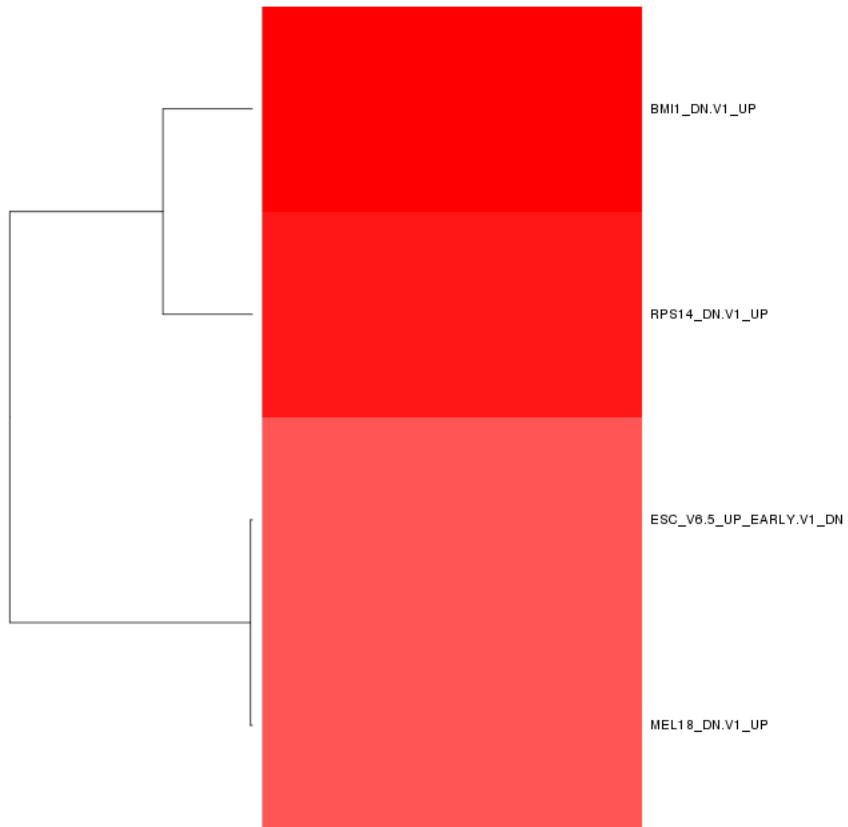
Interneuron zscore:



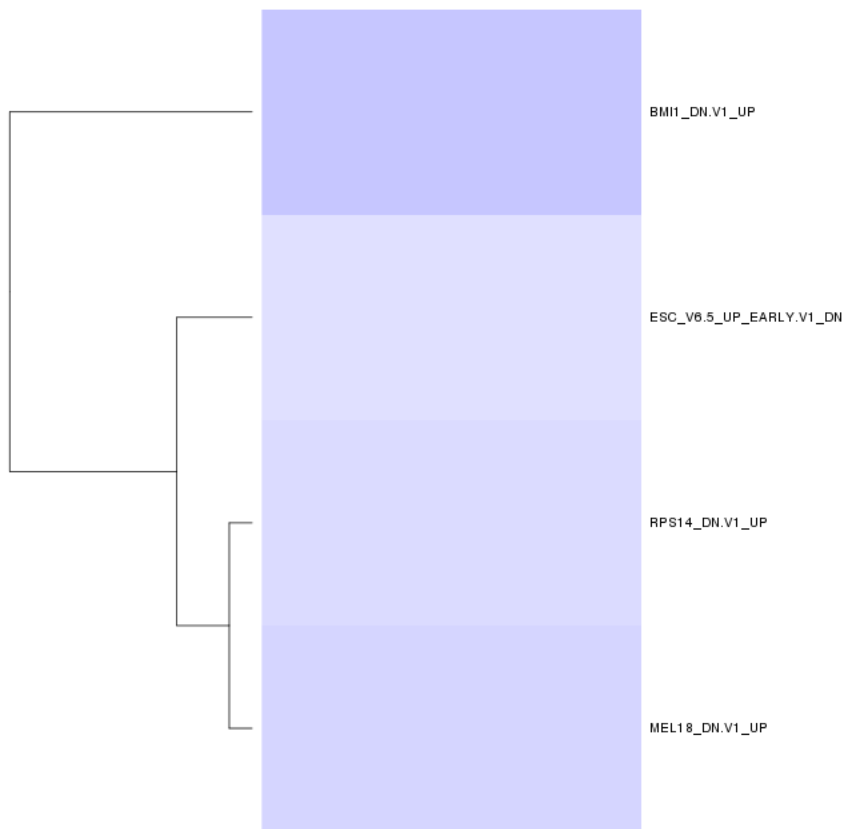
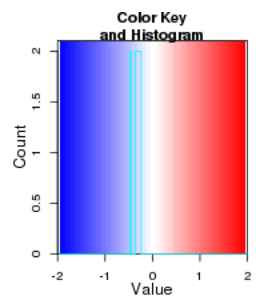


Oncogene enrichment:

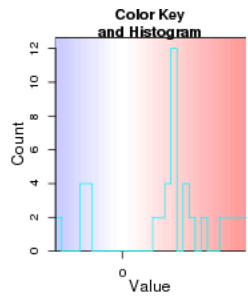


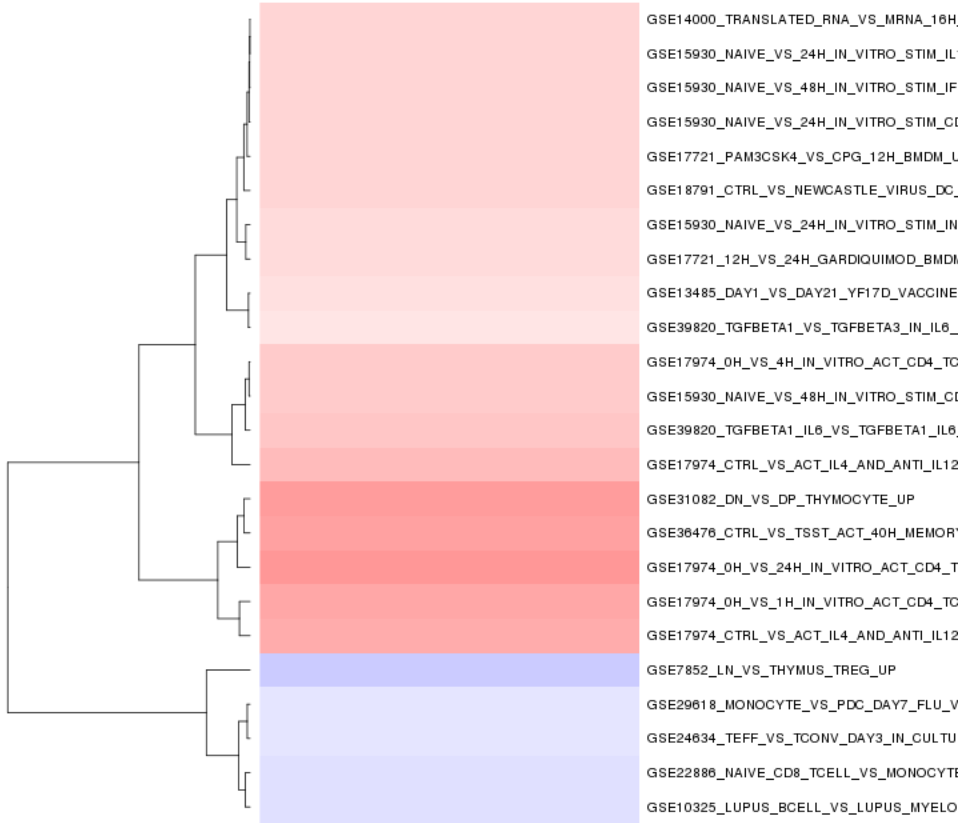


Oncogene zscore:

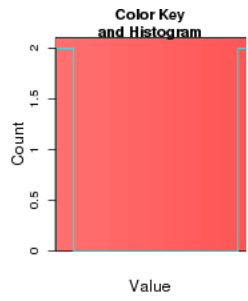


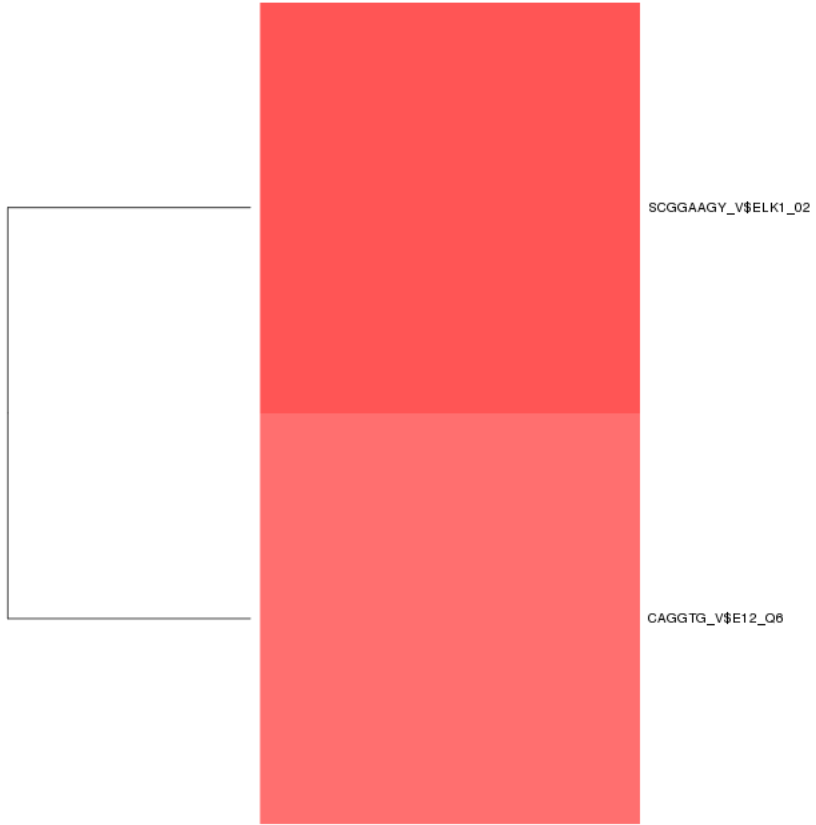
Immuno enrichment:



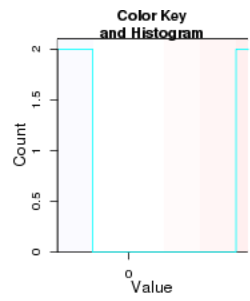


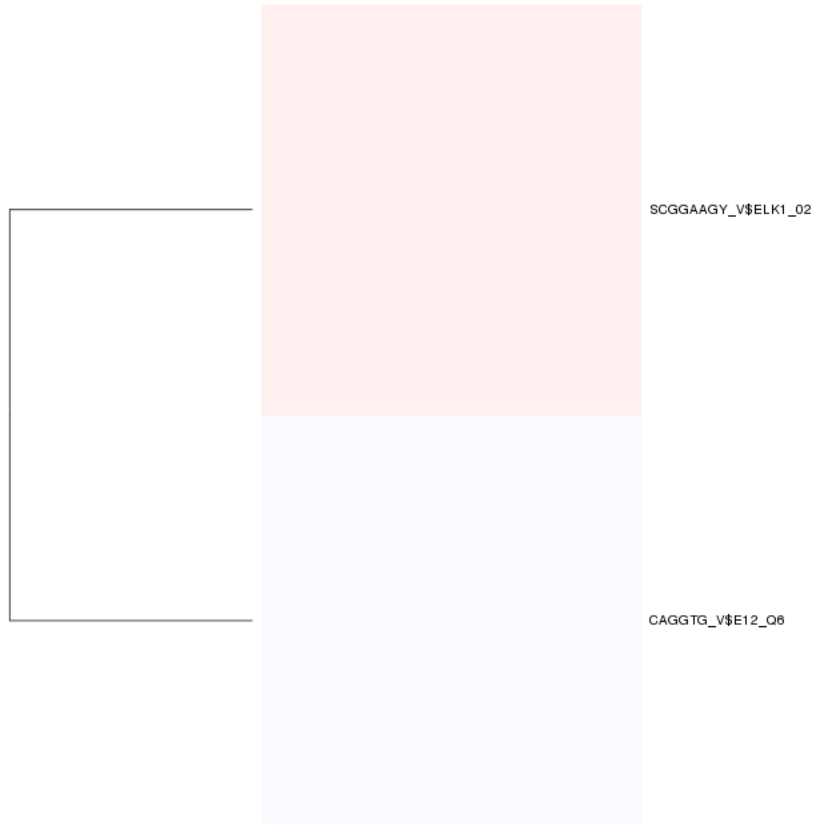
TF enrichment:





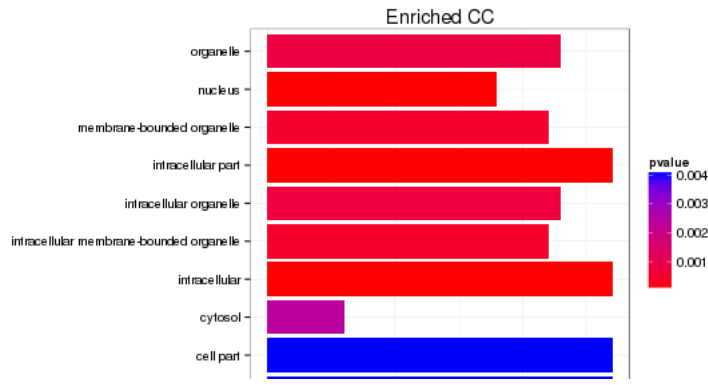
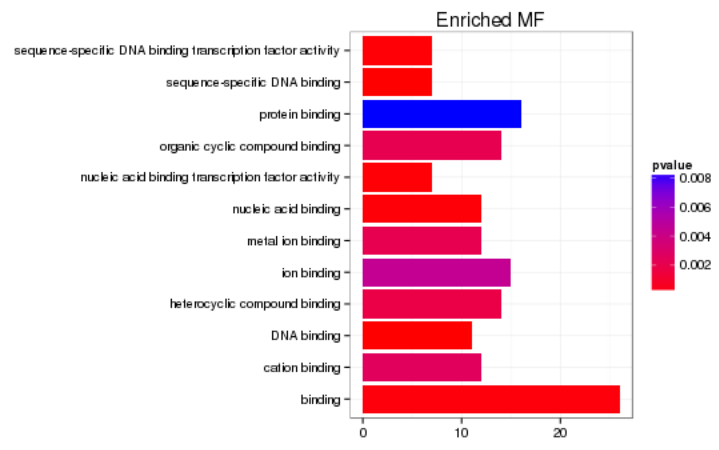
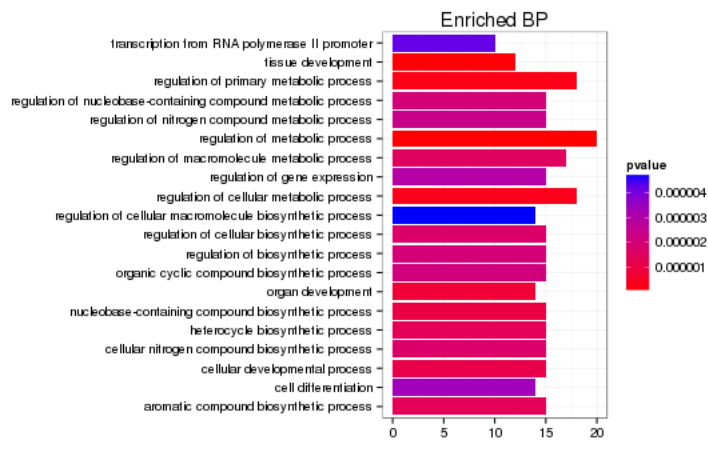
TF zscore:

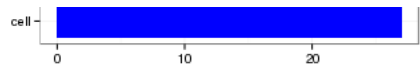




GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

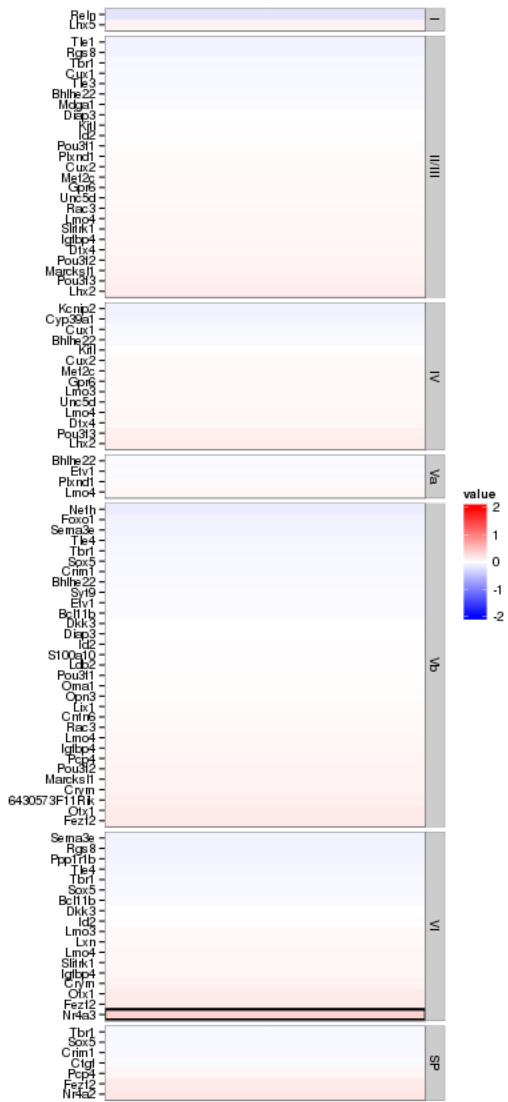




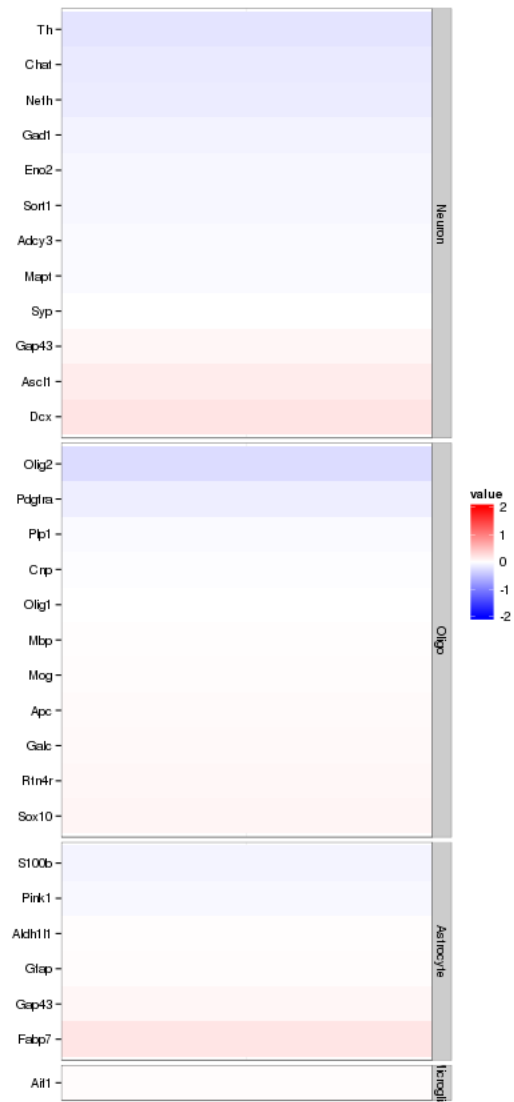
```
## Error: need finite 'xlim' values
```

```
## Error: need finite 'xlim' values
```

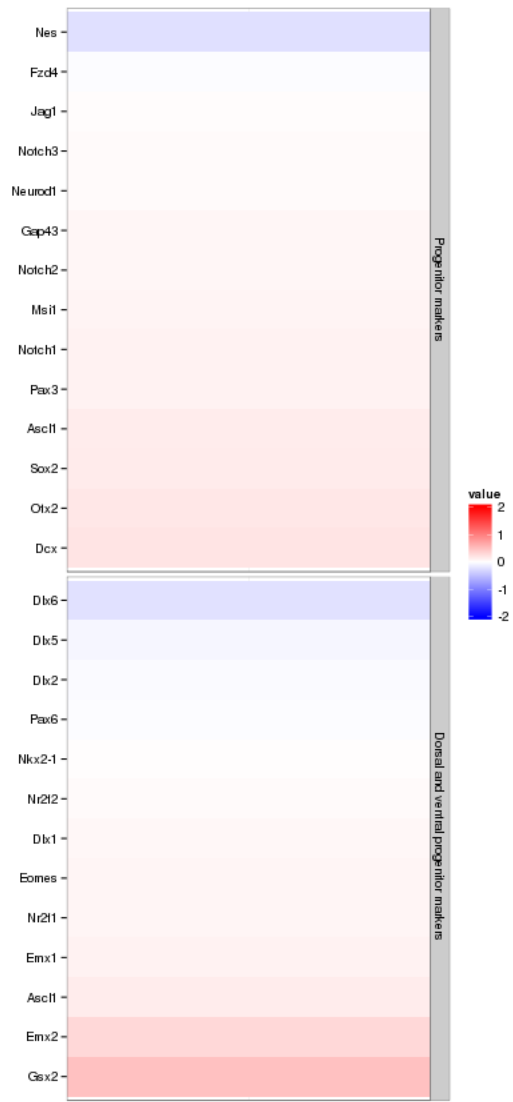
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



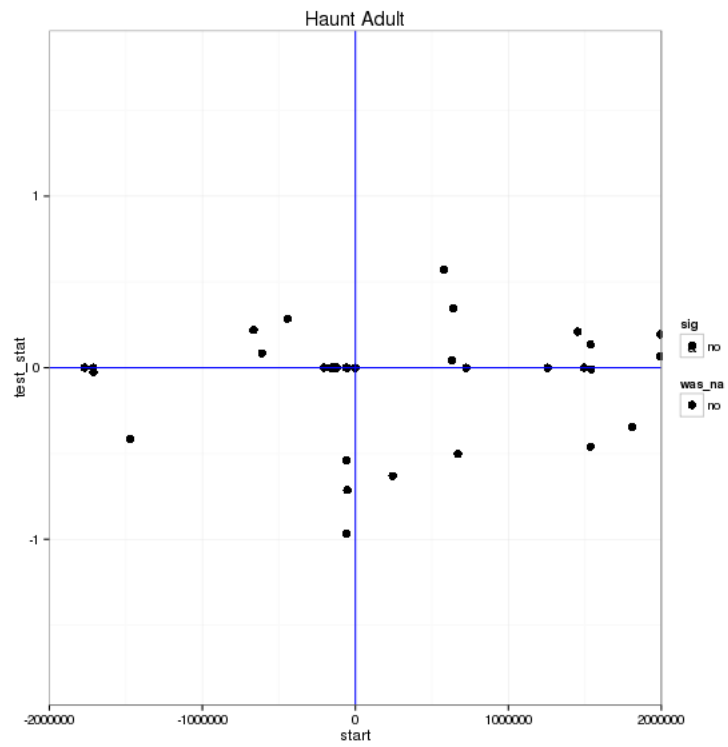
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

log2 Foldchange and test statistic are calculated with the ratio of fpkm(KO)/fpkm(WT), thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR738
16 JR782
17 JR826

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34083500.00	0.77	1.00
2 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34083500.00	0.59	1.00
3 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34083500.00	1.00	1.00
4 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34083500.00	0.81	1.00
5 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34083500.00	1.05	1.00
6 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34083500.00	1.09	1.00
7 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34083500.00	0.82	1.00
8 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34083500.00	1.17	1.00
9 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34083500.00	1.03	1.00
10 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34083500.00	1.32	1.00
11 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34083500.00	1.53	1.00
12 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34083500.00	1.03	1.00
13 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34083500.00	0.99	1.00
14 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34083500.00	1.22	1.00
15 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR738/abundances.cxb	Haunt	0	Haunt_0	26084100.00	34083500.00	0.77	1.00
16 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR782/abundances.cxb	Haunt	1	Haunt_1	43523200.00	34083500.00	1.28	1.00
17 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR826/abundances.cxb	Haunt	2	Haunt_2	33775800.00	34083500.00	0.99	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Haunt -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Haunt_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/ar
## 2
## 3
## 4
## 5
```

Haunt KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Haunt.

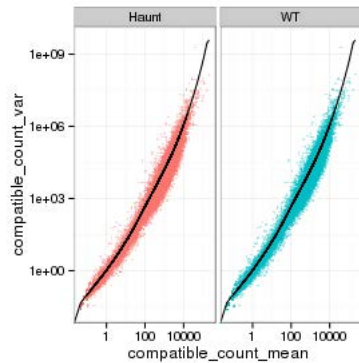
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

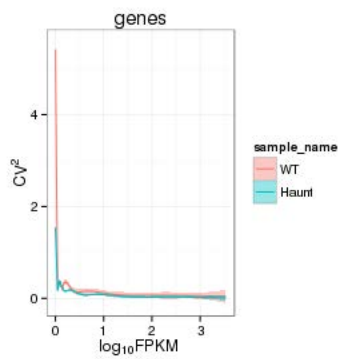
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



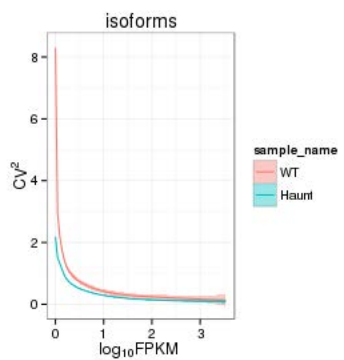
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

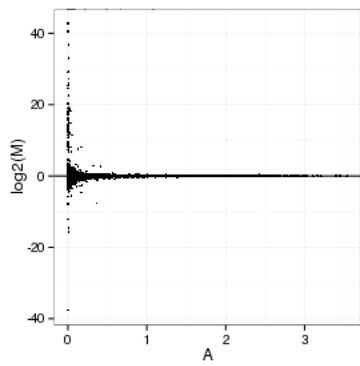
Genes:



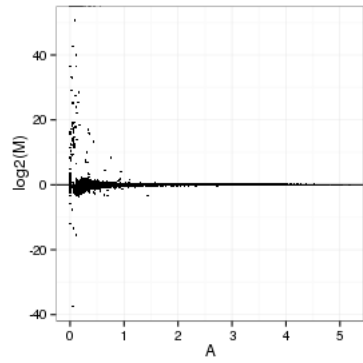
Isoforms:



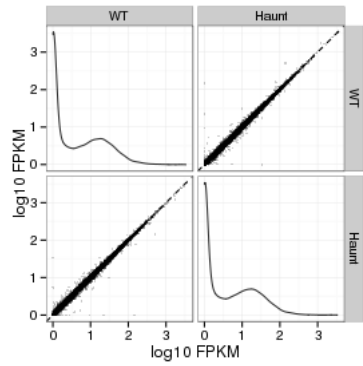
MvA plot



MvA plot counts



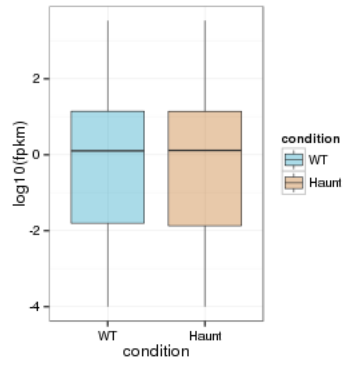
Scatterplot matrix



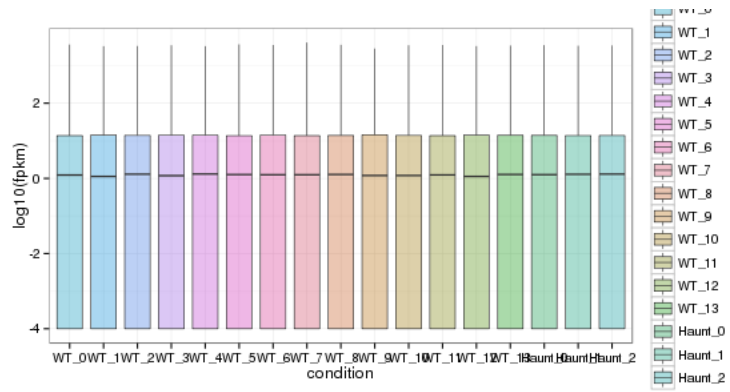
Distributions

Boxplots

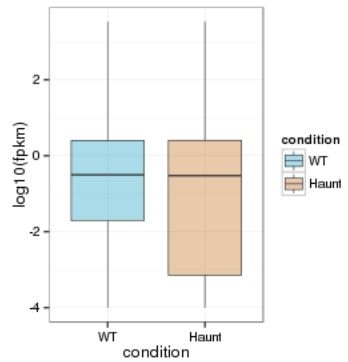
Boxplot (genes)



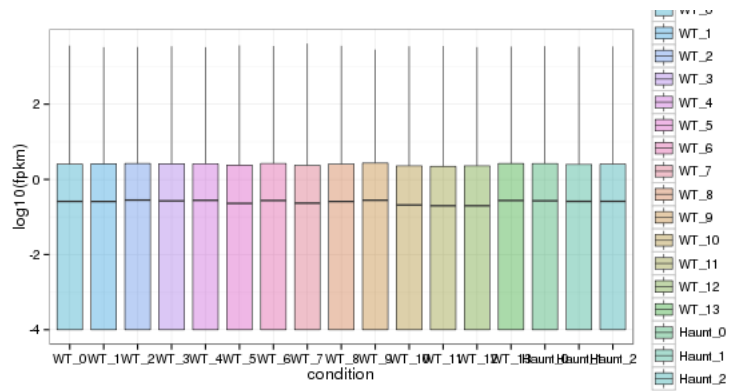
Boxplot (genes, replicates)



Boxplot (isoforms)

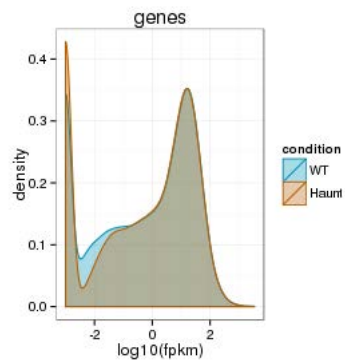


Boxplot (isoforms, replicates)

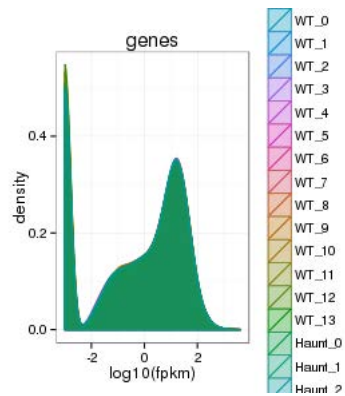


Density

Density (genes)

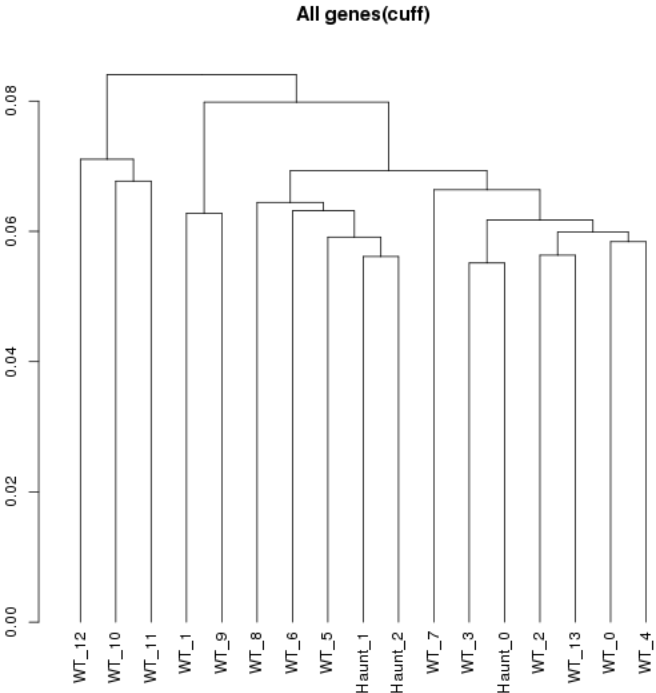


Density (genes, replicates)



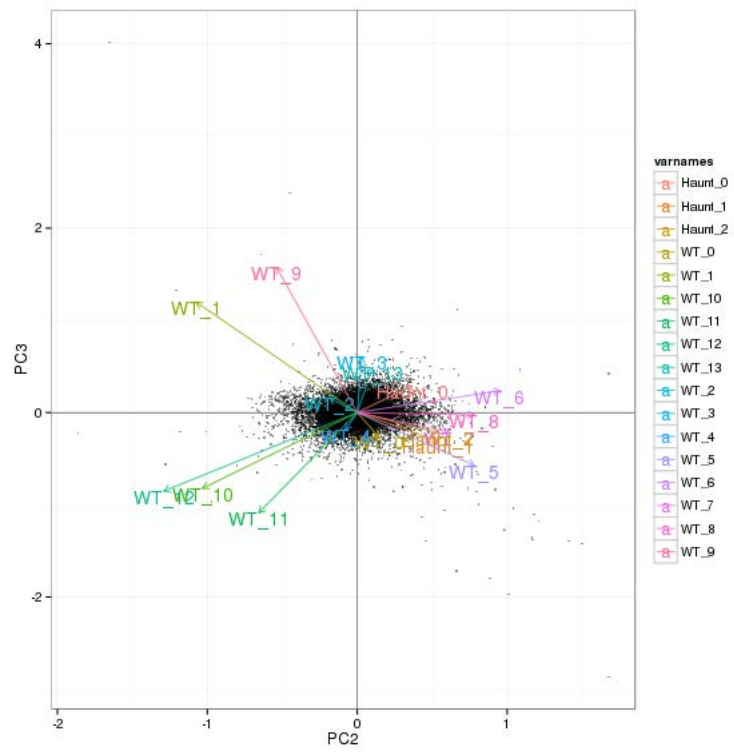
Clustering

Replicate Clusters

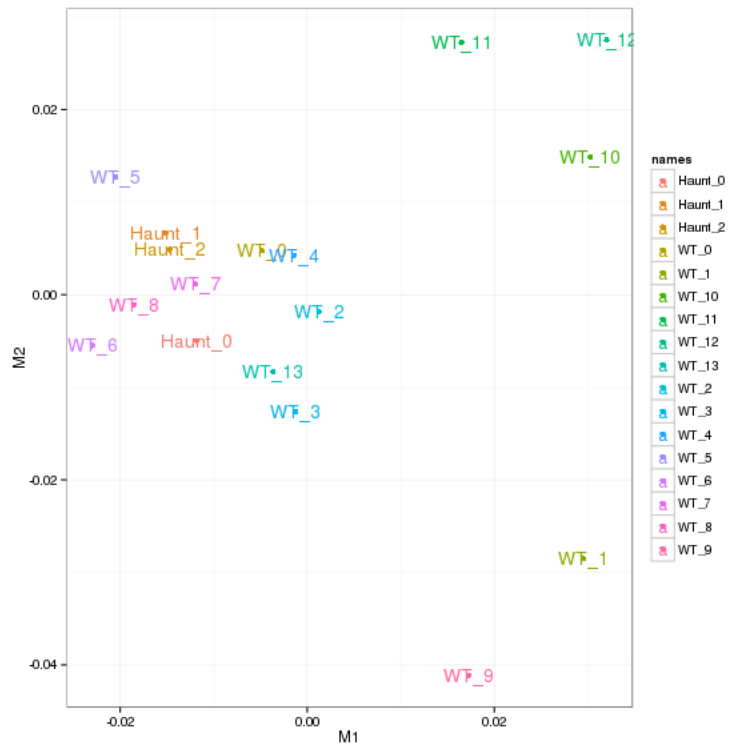


NULL

PCA (genes)

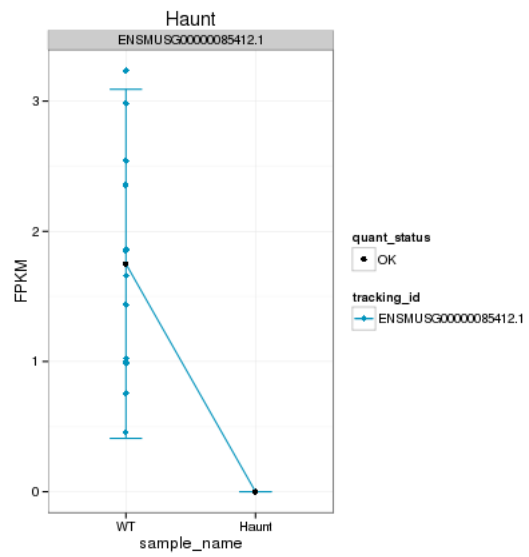


MDS (genes)

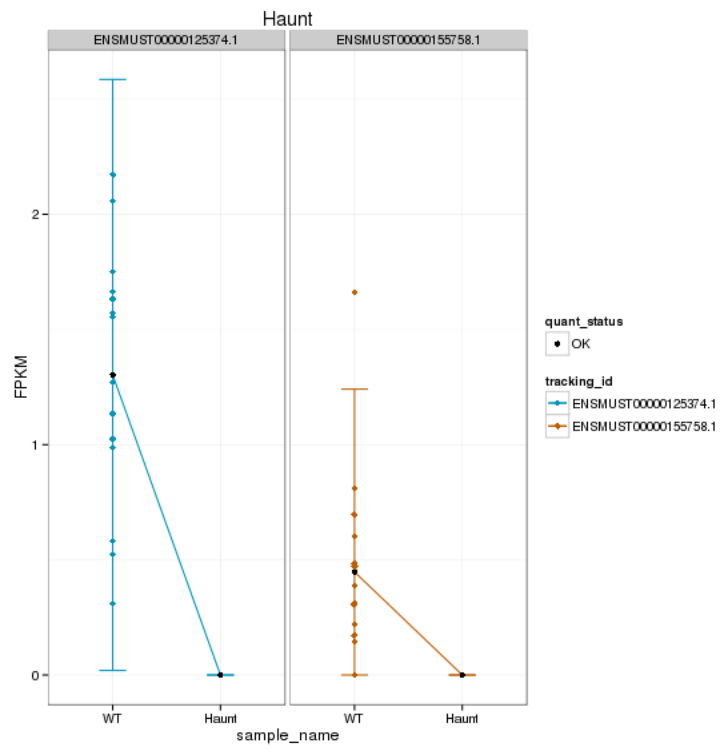


KO assessment

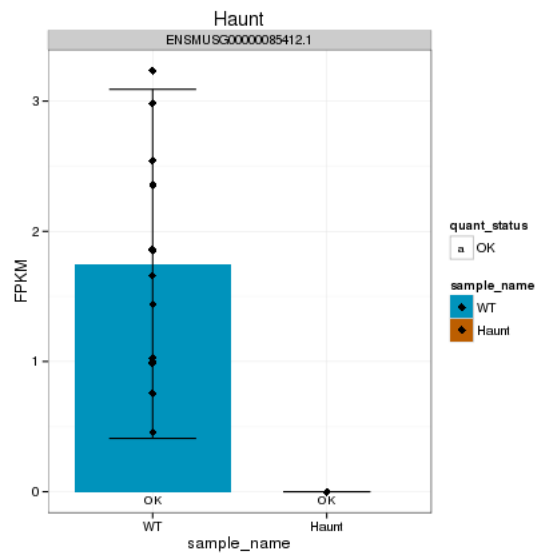
Endogenous lncRNA expression



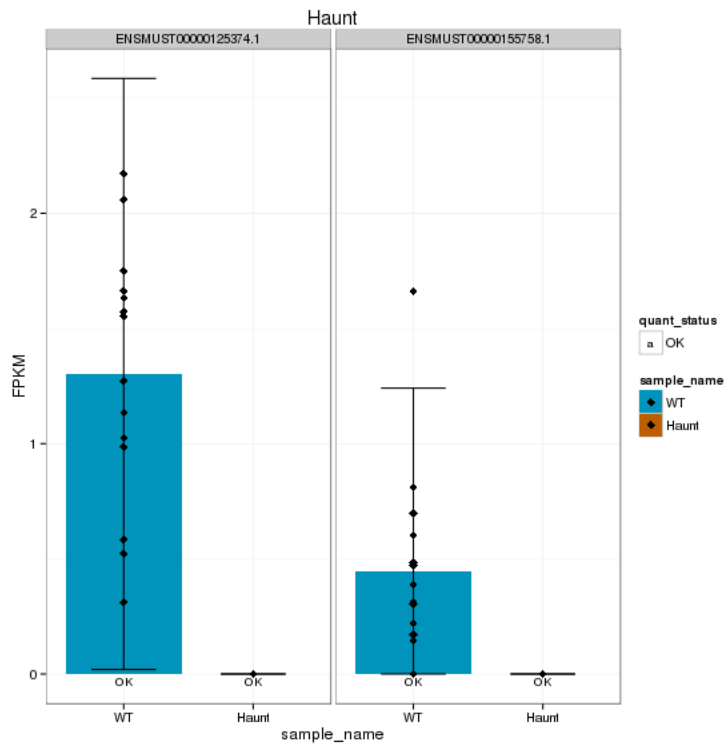
Endogenous expression of Haunt isoforms:



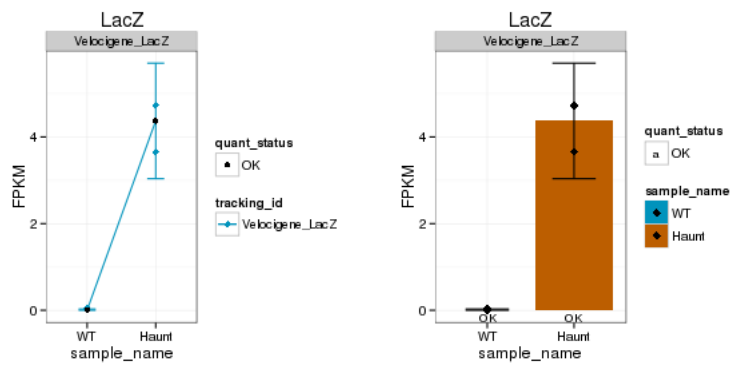
Barplot of gene expression:



Barplot of isoform expression:

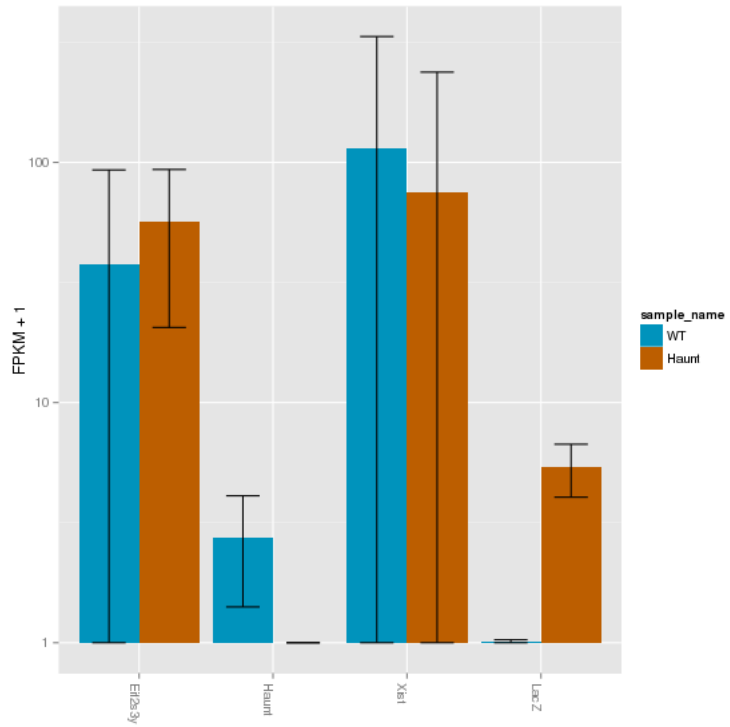


LacZ expression

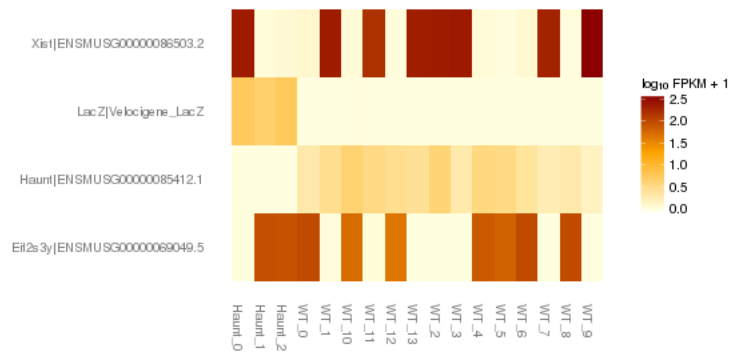


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



Expression heatmap:

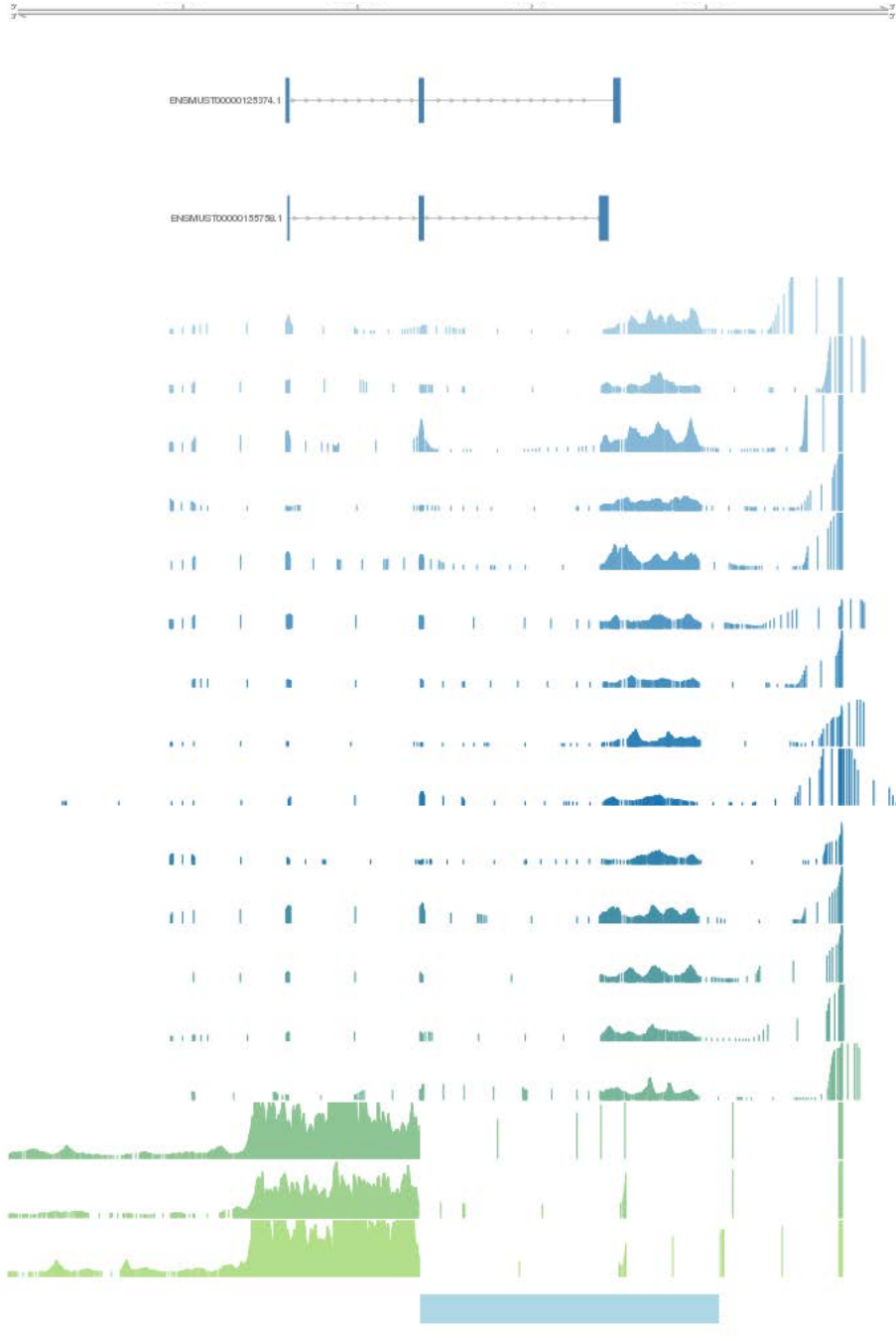


Track visualization



KO Region HsmU1 HsmU2 HsmU3 HsmU4 HsmU5 HsmU6 HsmU7 HsmU8 HsmU9 HsmU10 HsmU11 HsmU12 HsmU13 HsmU0 WT_0 WT_1 WT_2 WT_3 WT_4 WT_5 WT_6 WT_7 WT_8 WT_9 WT_10 WT_11 WT_12 WT_13

LncRNA isoforms



Differential Analysis

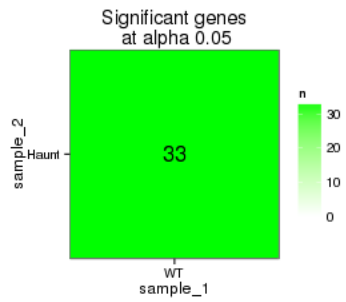
Differential Genes

There are 33 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 Nkx2-1
2 Rplp1
3 Hspa8
4 Tshz3
5 Dlx2
6 Clec3b
7 Lhx6
8 Hsd3b6
9 Dlx5
10 Dera
11 Hddc3
12 Mki67
13 Chma3
14 Cbl
15 Arx
16 Aox4
17 Pcsk1n
18 Dnah7b
19 Tuba1c
20 Rpl29
21 Creg2
22 Hbb-y
23 Hbb-bh1
24 Rnf26
25 Aldh1a1
26 Vsnl1
27 Hba-x
28 Sox11
29 Gm5089
30 Ddx3y
31 Gm9008
32 Haunt
33 Lhx8
```

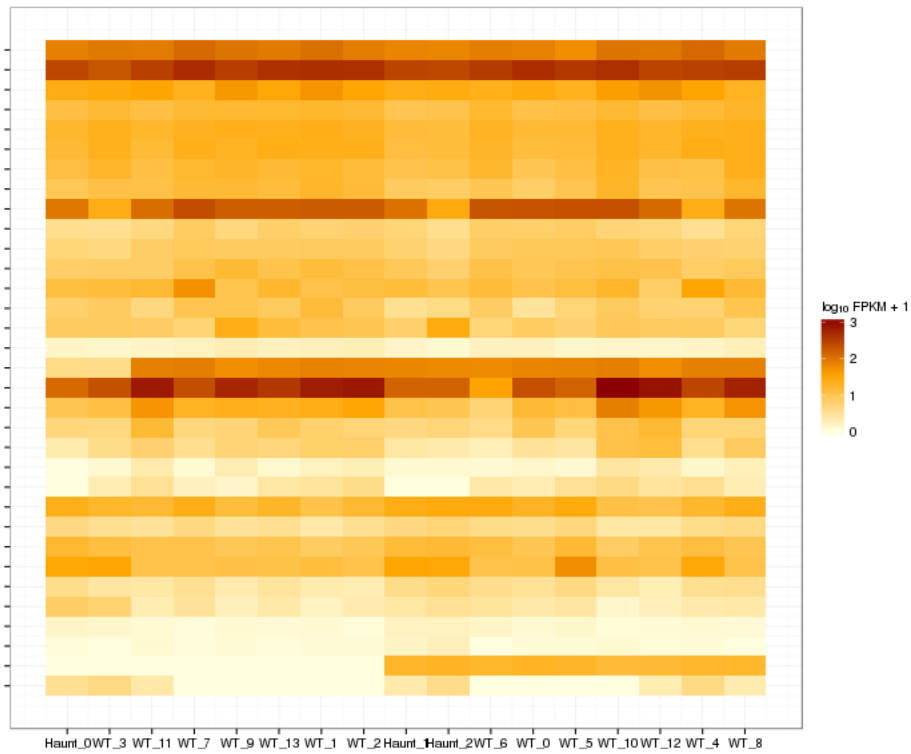
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

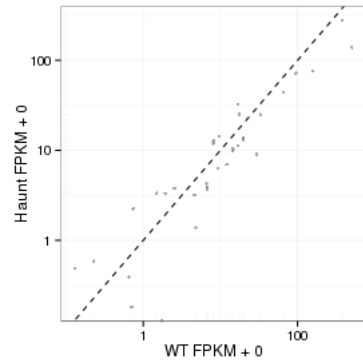


Significant genes with expression >50fpkm (any condition):(turned off)

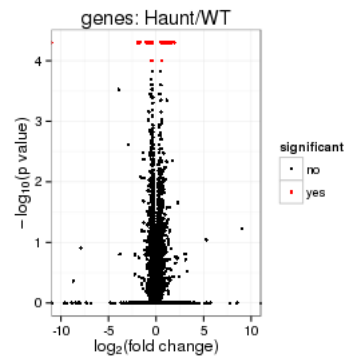
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

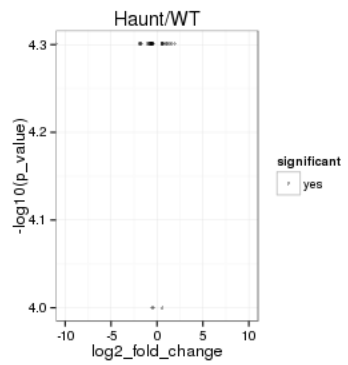
Scatter plot of significant genes only:



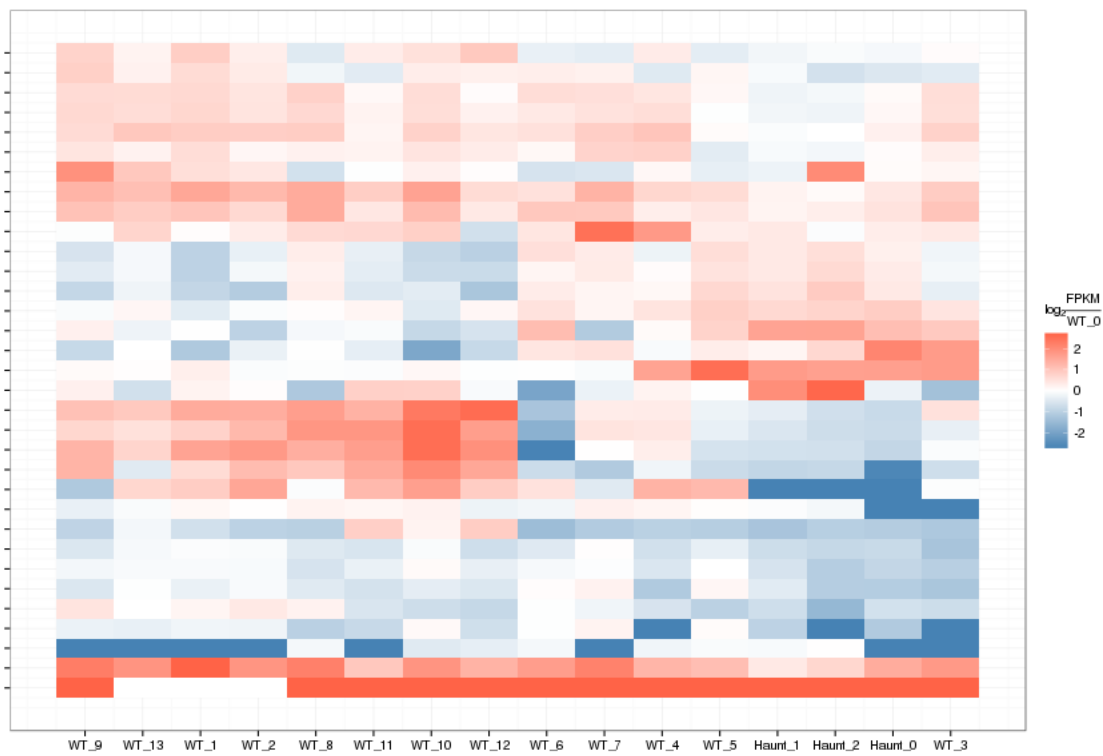
Volcano Plot



Volcano plot with significant genes only:



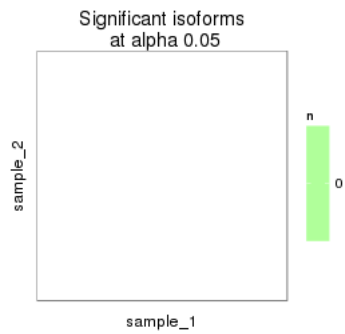
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

```
[1] "no sig isoforms"
```

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

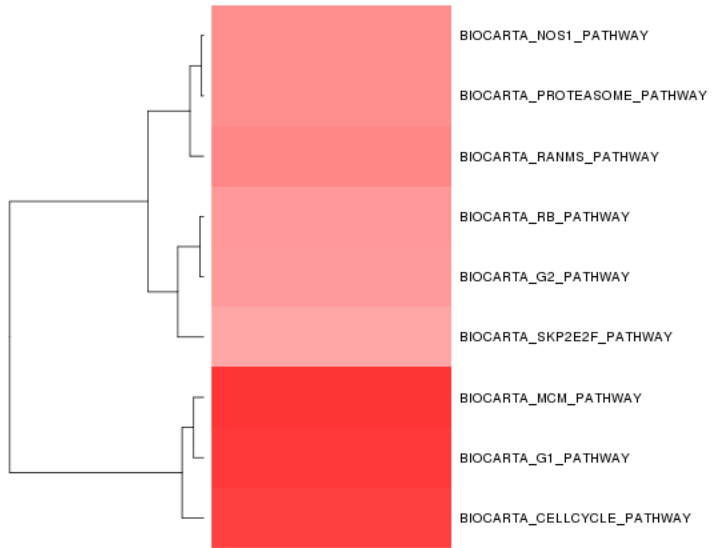
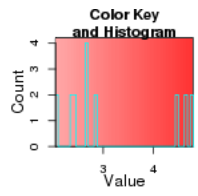
Gene/Pathway Analysis

GSEA

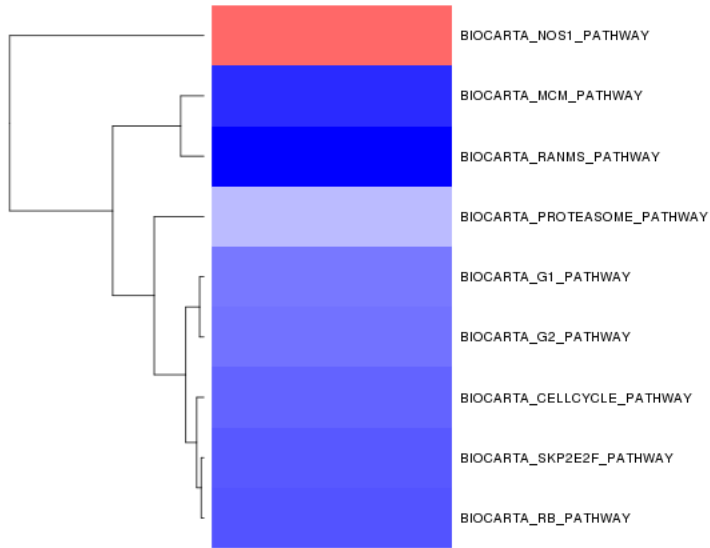
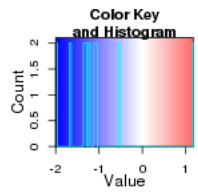
Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

Biocarta enrichment:

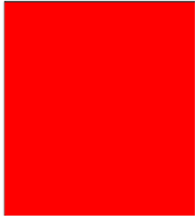


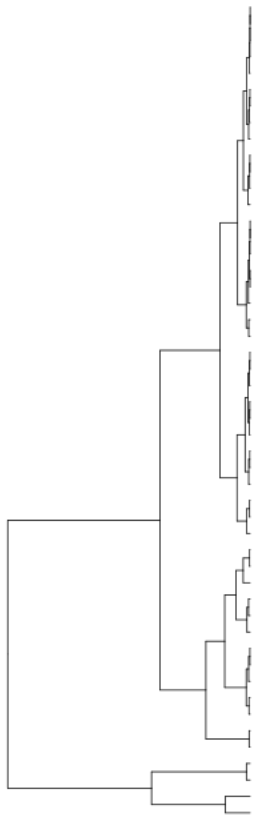
Biocarta zscore:



Reactome enrichment:

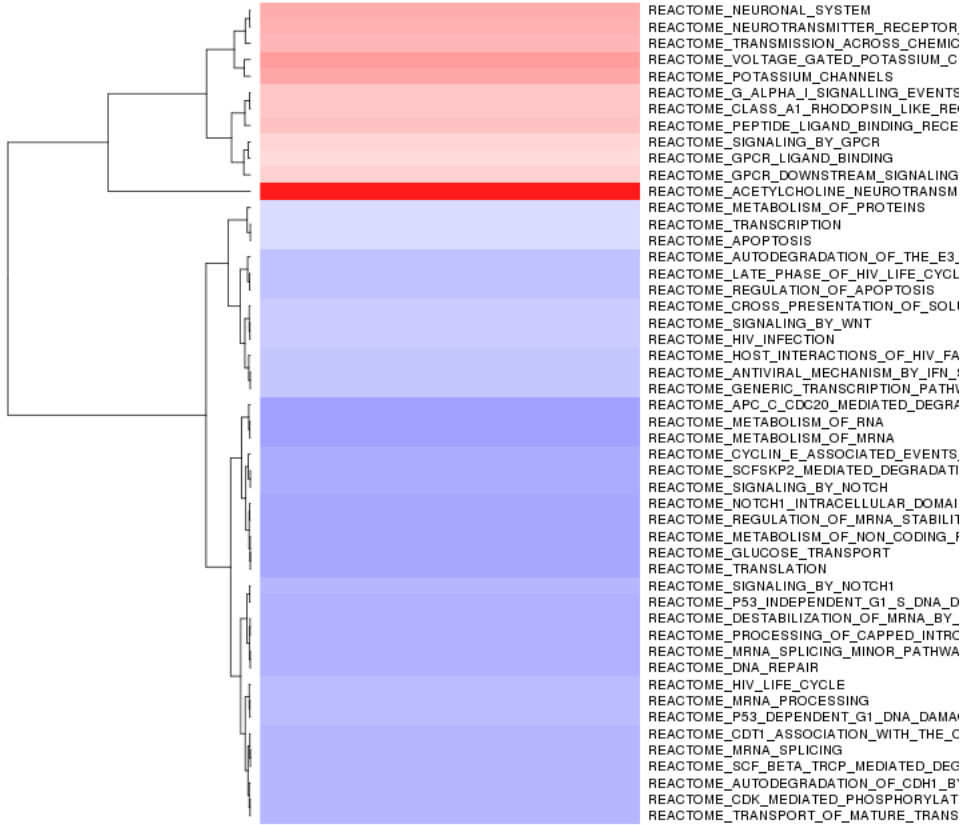
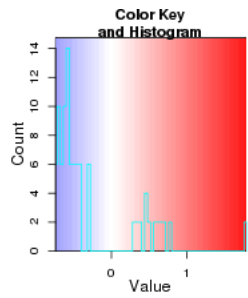
```
## Error: no locations are finite
```



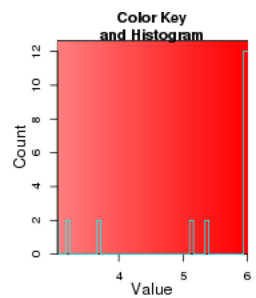


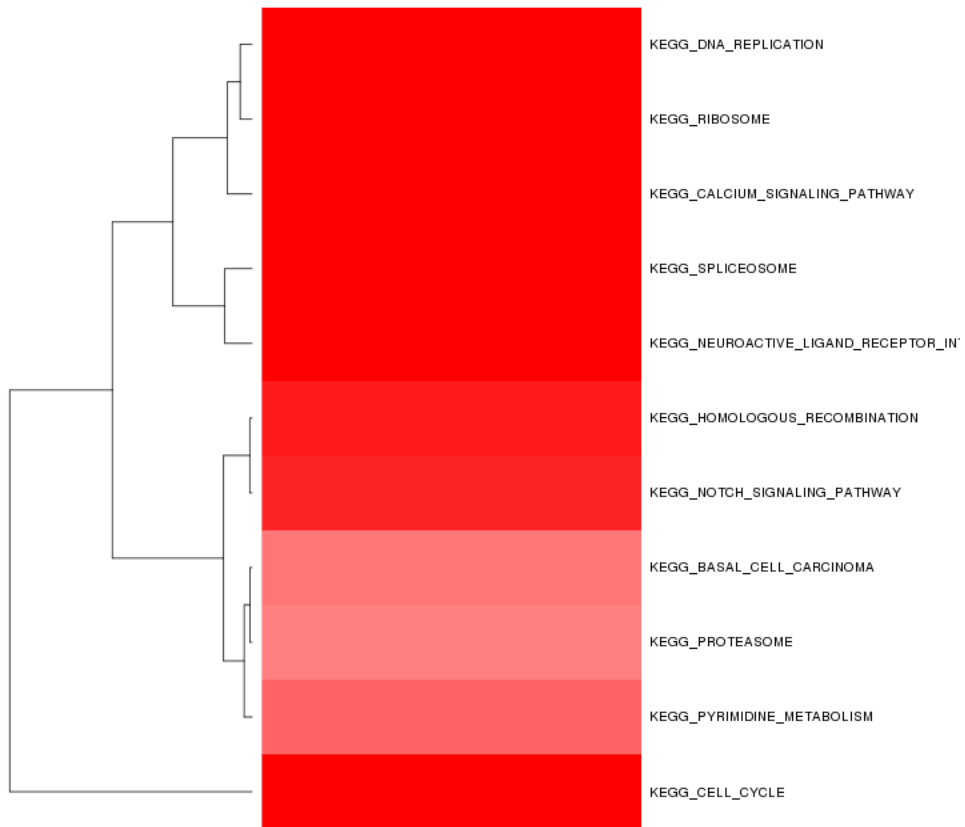
REACTOME_METABOLISM_OF_PROTEINS
REACTOME_SIGNALING_BY_NOTCH
REACTOME_CYCLIN_E_ASSOCIATED_EVENTS
REACTOME_CDT1_ASSOCIATION_WITH_THE_C
REACTOME_AUTODEGRADATION_OF_CDH1_B
REACTOME_REGULATION_OF_MRNA_STABILIT
REACTOME_TRANSMISSION_ACROSS_CHEMIC
REACTOME_SCF5KIP2_MEDIATED_DEGRADATI
REACTOME_SRP_DEPENDENT_COTRANSLATIC
REACTOME_ACTIVATION_OF_ATR_IN_RESPON
REACTOME_SIGNALING_BY_GPCR
REACTOME_HOST_INTERACTIONS_OF_HIV_FA
REACTOME_HIV_LIFE_CYCLE
REACTOME_NONSENSE_MEDIATED_DECAY_E
REACTOME_PEPTIDE_CHAIN_ELONGATION
REACTOME_APC_C_CDC20_MEDIATED_DEGRA
REACTOME_ACTIVATION_OF_THE_PRE_REPLI
REACTOME_GPCR_DOWNSTREAM_SIGNALING
REACTOME_INFLUENZA_VIRAL_RNA_TRANSCF
REACTOME_APC_C_CDH1_MEDIATED_DEGRAI
REACTOME_POTASSIUM_CHANNELS
REACTOME_HIV_INFECTION
REACTOME_ACTIVATION_OF_THE_MRNA_UPO
REACTOME_TRANSLATION
REACTOME_G2_M_CHECKPOINTS
REACTOME_FORMATION_OF_THE_TERNARY_C
REACTOME_ASSEMBLY_OF_THE_PRE_REPLIC
REACTOME_DNA_STRAND_ELONGATION
REACTOME_CHROMOSOME_MAINTENANCE
REACTOME_ORC1_REMOVAL_FROM_CHROMA
REACTOME_MRNA_SPLICING
REACTOME_REGULATION_OF_MITOTIC_CELL_
REACTOME_3_UTR_MEDIATED_TRANSLATION
REACTOME_S_PHASE
REACTOME_CELL_CYCLE_CHECKPOINTS
REACTOME_METABOLISM_OF_MRNA
REACTOME_PROCESSING_OF_CAPPED_INTRC
REACTOME_G1_S_TRANSITION
REACTOME_SYNTHESIS_OF_DNA
REACTOME_INFLUENZA_LIFE_CYCLE
REACTOME_M_G1_TRANSITION
REACTOME_MRNA_PROCESSING
REACTOME_NEURONAL_SYSTEM
REACTOME_MITOTIC_PROMETAPHASE
REACTOME_METABOLISM_OF_RNA
REACTOME_MITOTIC_G1_G1_S_PHASES
REACTOME_CELL_CYCLE
REACTOME_CELL_CYCLE_MITOTIC
REACTOME_DNA_REPLICATION
REACTOME_MITOTIC_M_M_G1_PHASES

Reactome zscore:

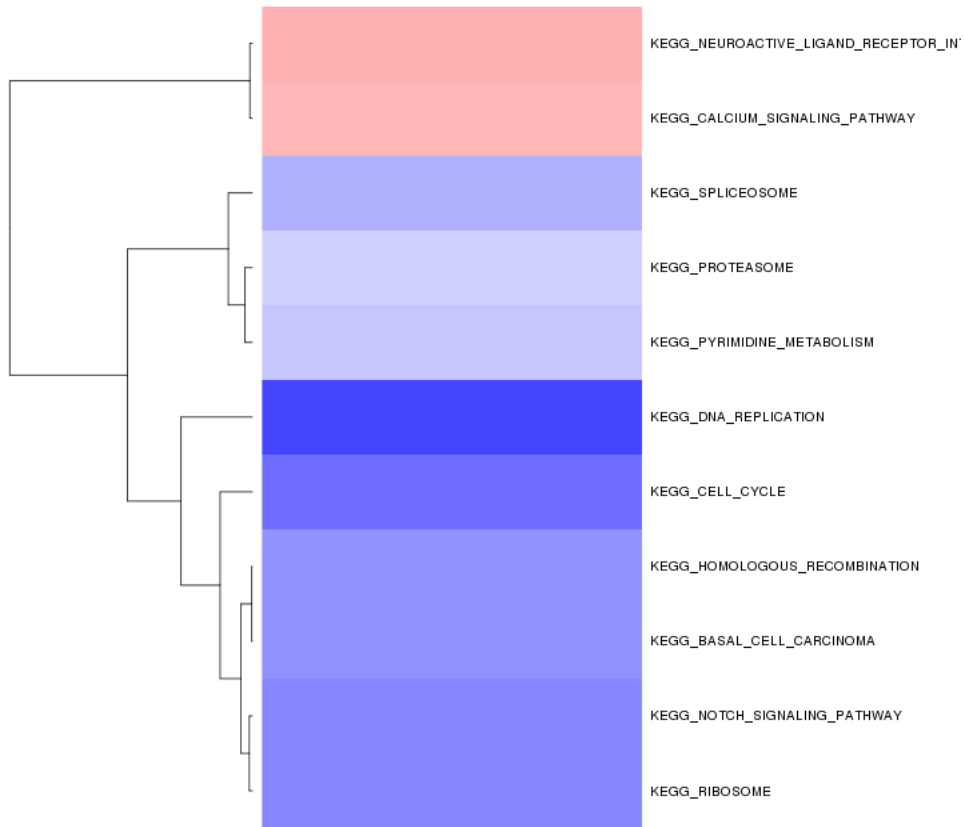
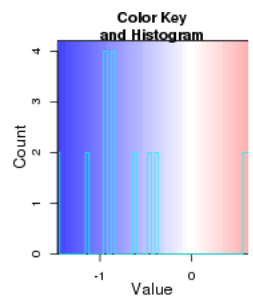


Kegg enrichment:



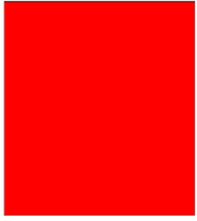


Kegg zscore:



Interneuron enrichment:

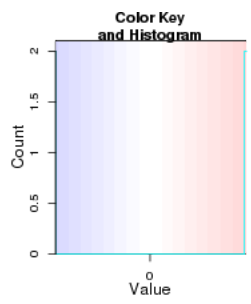
Error: no locations are finite



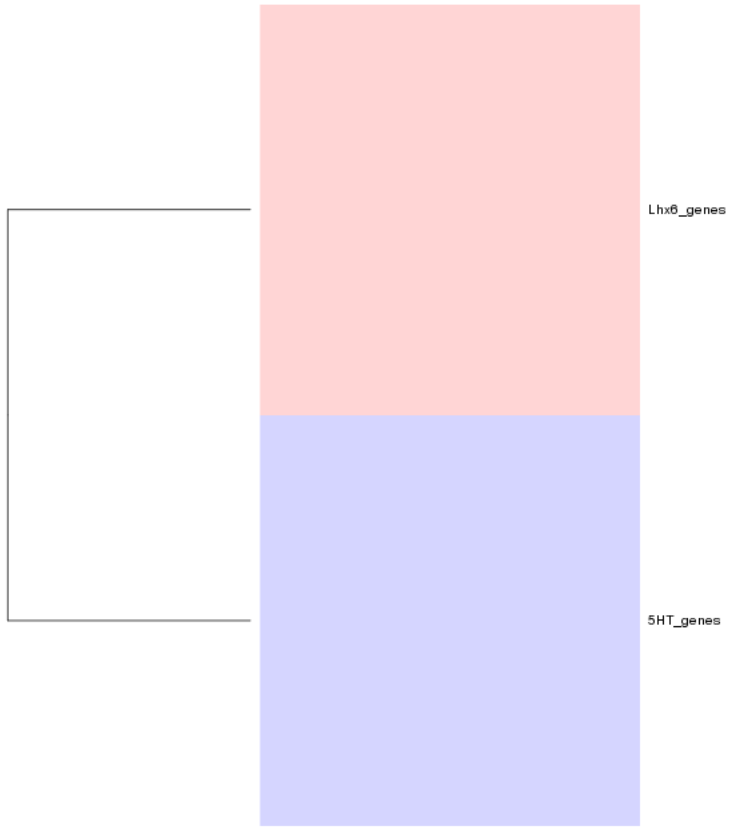
5HT_genes

Lhx6_genes

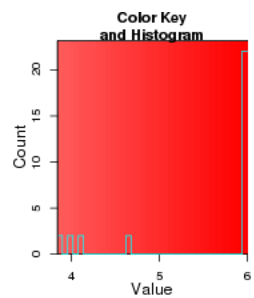
Interneuron zscore:

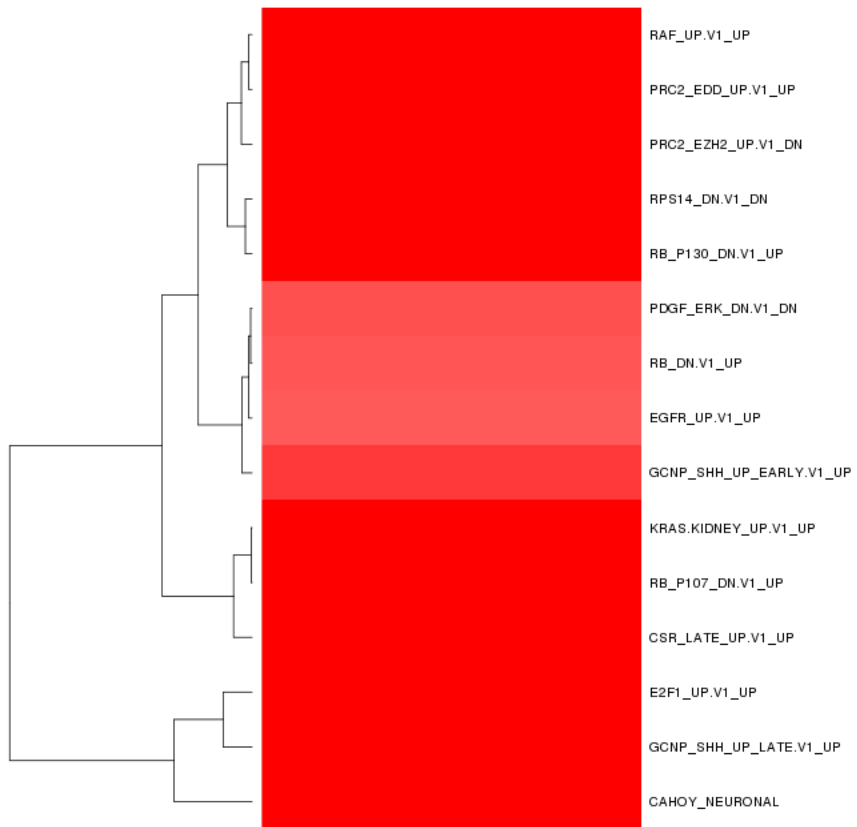


—

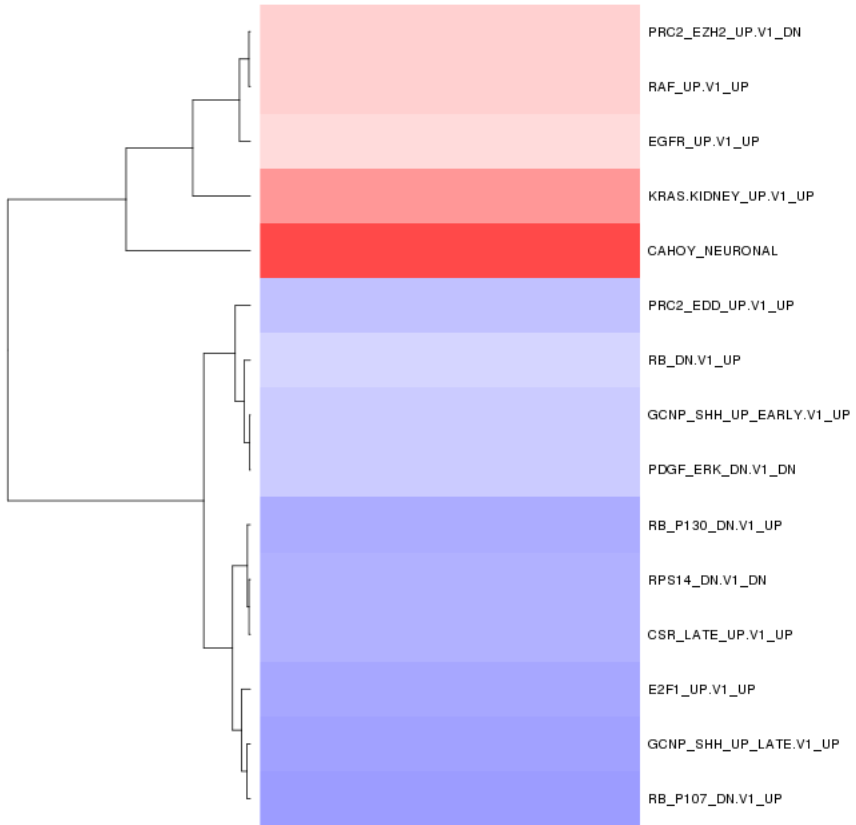
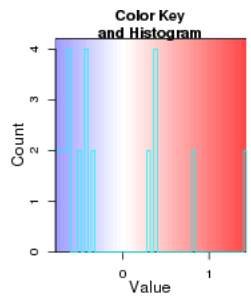


Oncogene enrichment:



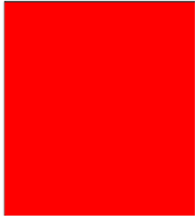


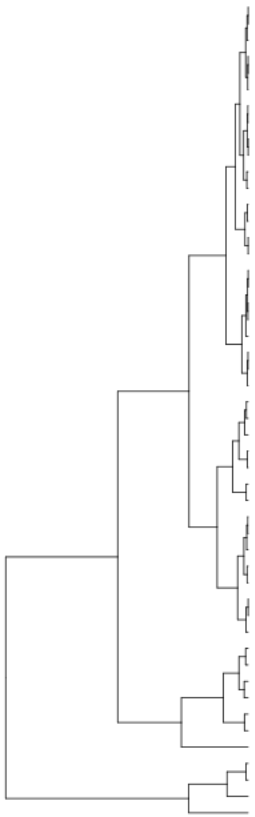
Oncogene zscore:



Immuno enrichment:

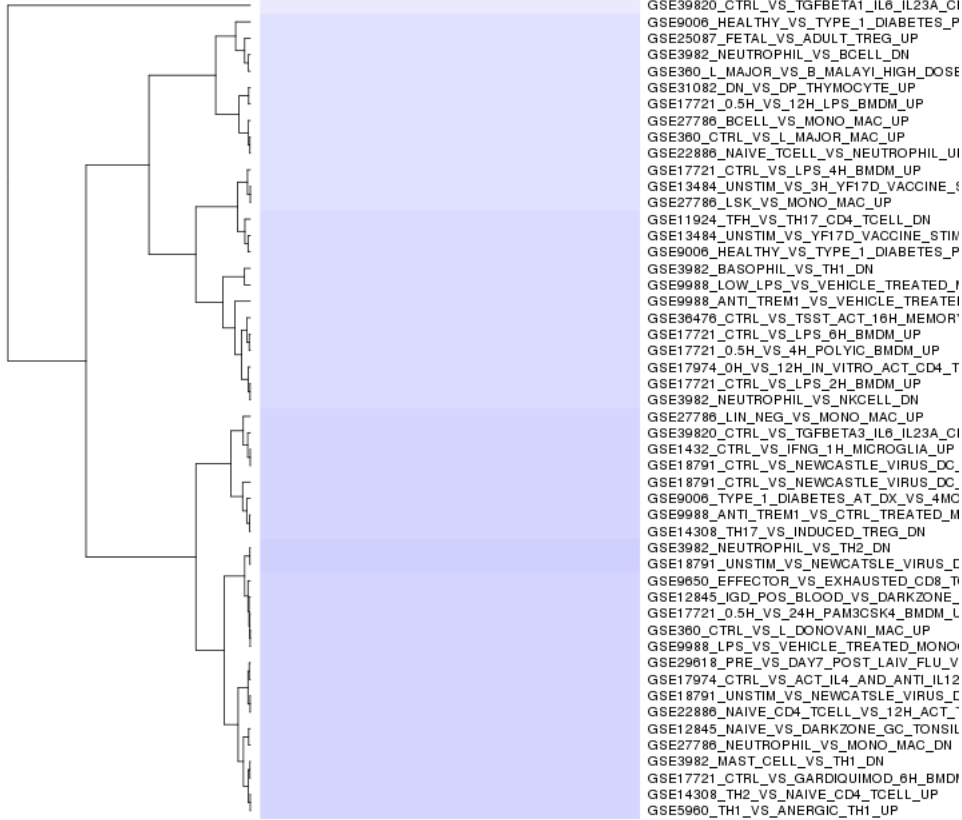
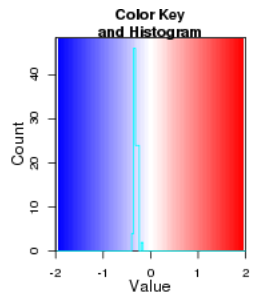
Error: no locations are finite





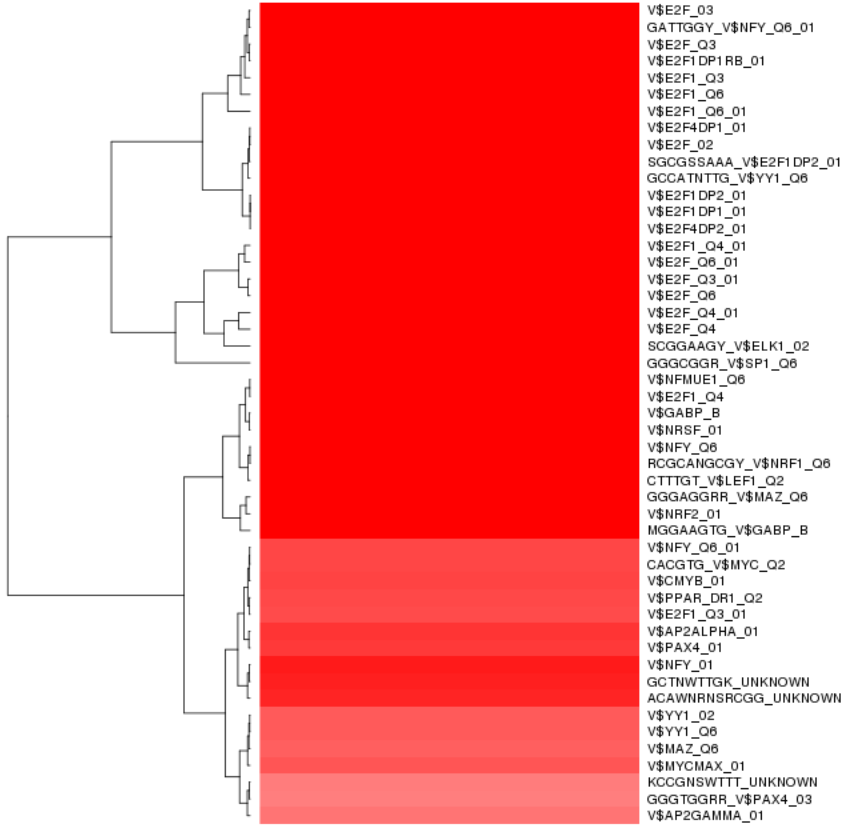
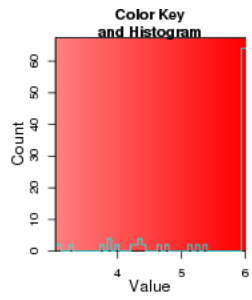
GSE12845_NAIVE_VS_PRE_GC_TONSIL_BCEL
GSE3982_EFF_MEMORY_CD4_TCELL_VS_TH2
GSE9006_TYPE_1_DIABETES_AT_DX_VS_1MC
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IN
GSE300_L_MAJOR_VS_T_GONDII_MAC_DN
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IL
GSE17974_CTRL_VS_ACT_IL4_AND_ANTI_IL12
GSE24634_TEFF_VS_TCONV_DAY3_IN_CULTU
GSE31082_DP_VS_CD8_SP_THYMOCYTE_UP
GSE12306_GC_VS_NAIVE_BCELL_UP
GSE3982_MAC_VS_TH1_DN
GSE12306_GC_VS_MEMORY_BCELL_UP
GOLDRATH_NAIVE_VS_EFF_CD8_TCELL_DN
GSE22886_NAIVE_CD4_TCELL_VS_48H_ACT_
GSE10239_NAIVE_VS_KLRG1HIGH_EFF_CD8_
GSE3982_MEMORY_CD4_TCELL_VS_TH2_DN
GSE19825_CD24LOW_VS_IL2RA_HIGH_DAY3_
GSE24634_TREG_VS_TCONV_POST_DAYS_IL4
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_CI
GSE24634_TEFF_VS_TCONV_DAY10_IN_CULTI
GSE22886_UNSTIM_VS_STIM_MEMORY_TCELL
GSE9988_LOW_LPS_VS_CTRL_TREATED_MON
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IL
GSE1460_INTRATHYMIC_T_PROGENITOR_VS_
GSE24634_TEFF_VS_TCONV_DAY7_IN_CULTU
GSE12845_IGD_POS_BLOOD_VS_PRE_GC_TO
GOLDRATH_EFF_VS_MEMORY_CD8_TCELL_UP
GSE17974_0H_VS_24H_IN_VITRO_ACT_CD4_T
GSE24634_TREG_VS_TCONV_POST_DAY3_IL4
GSE9006_TYPE_1_VS_TYPE_2_DIABETES_PB
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_CI
GSE24026_PDI_LIGATION_VS_CTRL_IN_ACT_
GSE19825_NAIVE_VS_IL2RAHIGH_DAY3_EFF_
GSE24634_TEFF_VS_TCONV_DAYS_IN_CULTU
GSE10239_NAIVE_VS_DAY4.5_EFF_CD8_TCEL
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IF
GSE31082_DN_VS_CD4_SP_THYMOCYTE_UP
KAECH_DAY8_EFF_VS_DAY15_EFF_CD8_TOEL
GSE15930_NAIVE_VS_72H_IN_VITRO_STIM_TF
GSE22886_UNSTIM_VS_IL15_STIM_NKCELL_D
GSE7764_IL15_TREATED_VS_CTRL_NK_CELL_
GSE24634_TREG_VS_TCONV_POST_DAY7_IL4
GSE30476_CTRL_VS_TSST_ACT_40H_MEMOR
GSE30476_CTRL_VS_TSST_ACT_72H_MEMOR
GSE22886_UNSTIM_VS_IL2_STIM_NKCELL_DN
GSE30476_CTRL_VS_TSST_ACT_40H_MEMOR
GSE30902_PRIMARY_VS_SECONDARY_ACUTE
GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL
GSE30476_CTRL_VS_TSST_ACT_72H_MEMOR
GSE15750_DAY6_VS_DAY10_TRAF6KO_EFF_C

Immuno zscore:



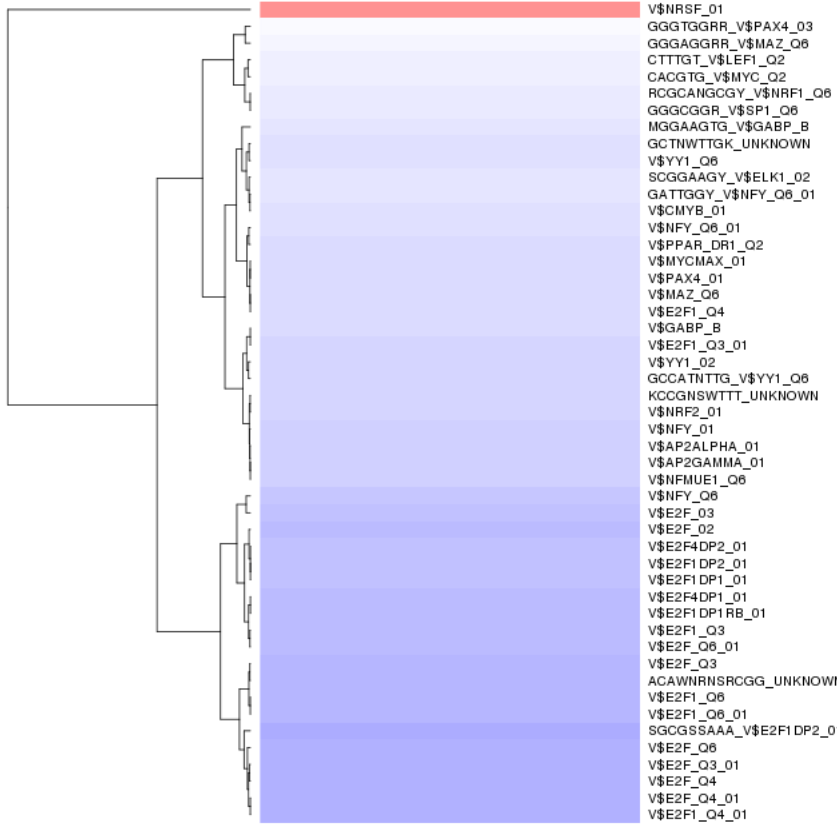
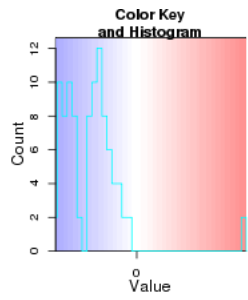
TF enrichment:

Error: subscript out of bounds



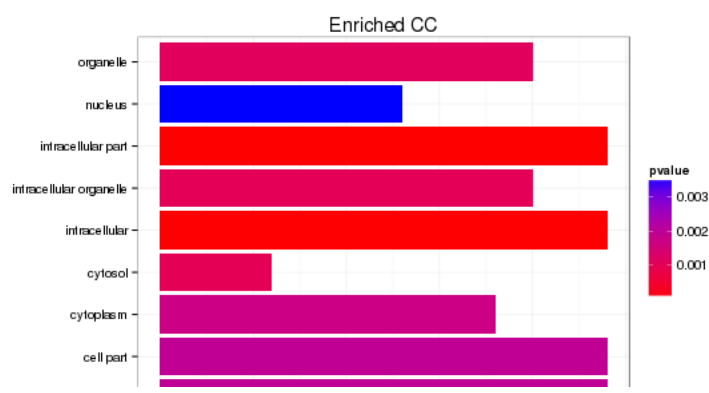
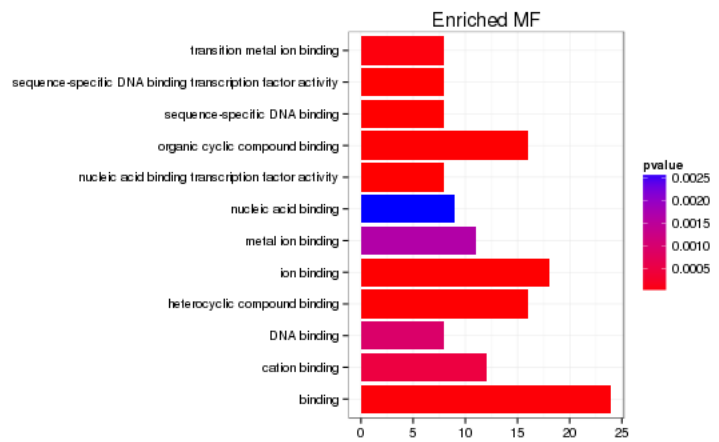
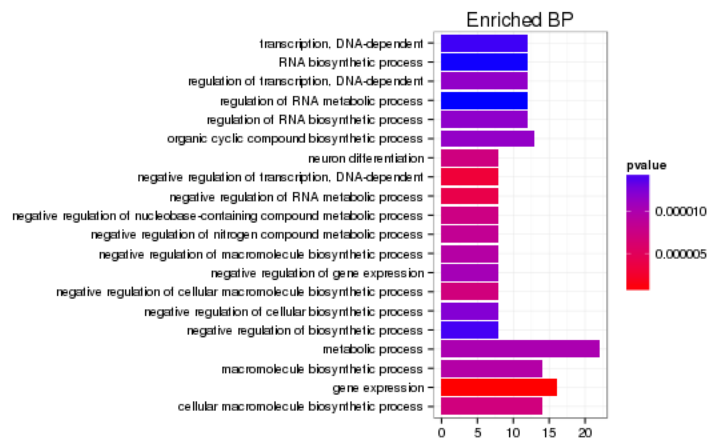
TF zscore:

Error: subscript out of bounds



GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

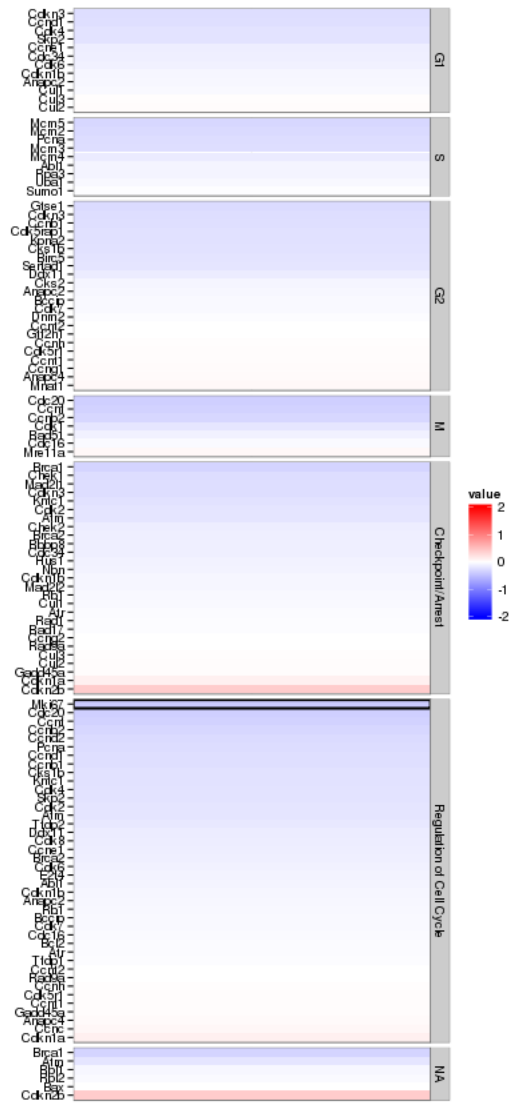


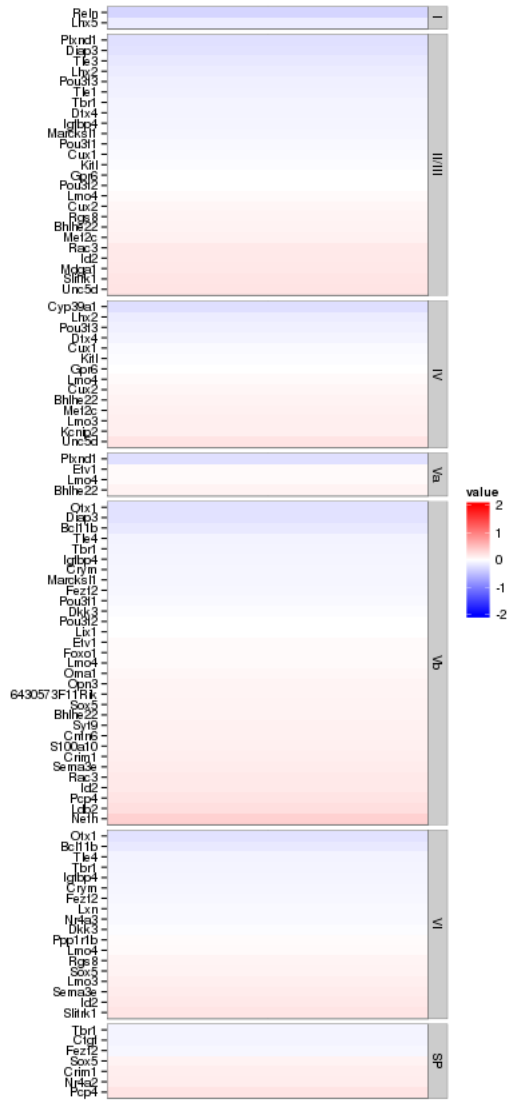


```
## Error: need finite 'xlim' values
```

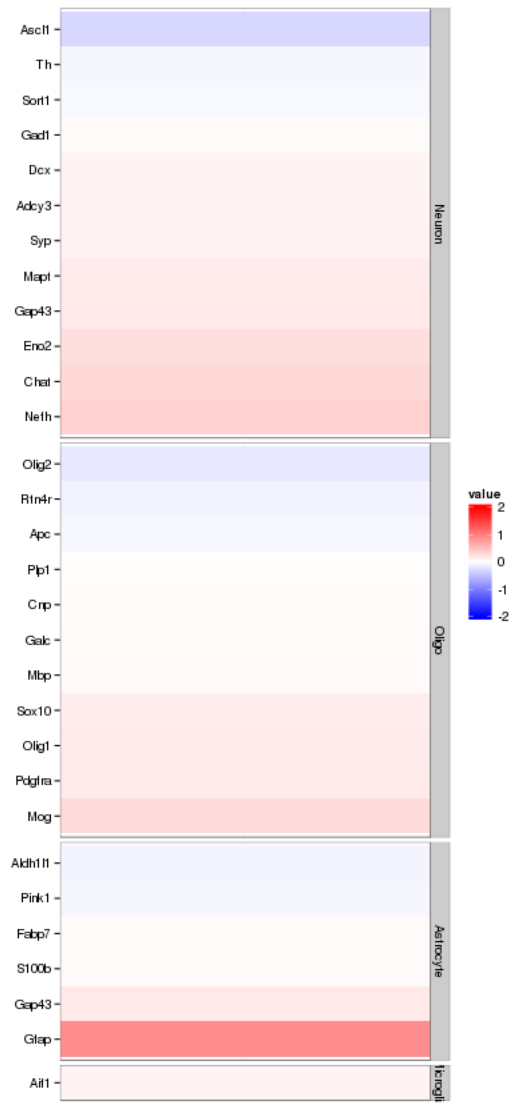
```
## Error: need finite 'xlim' values
```

Enrichment or depletion for stage-specific cell cycle markers

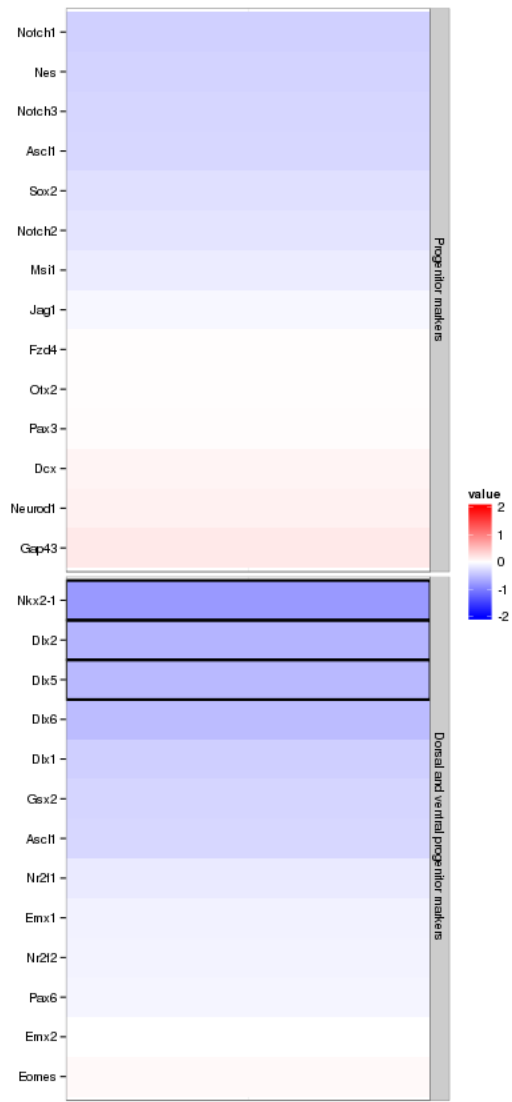




Enrichment or depletion for specific neural cell types



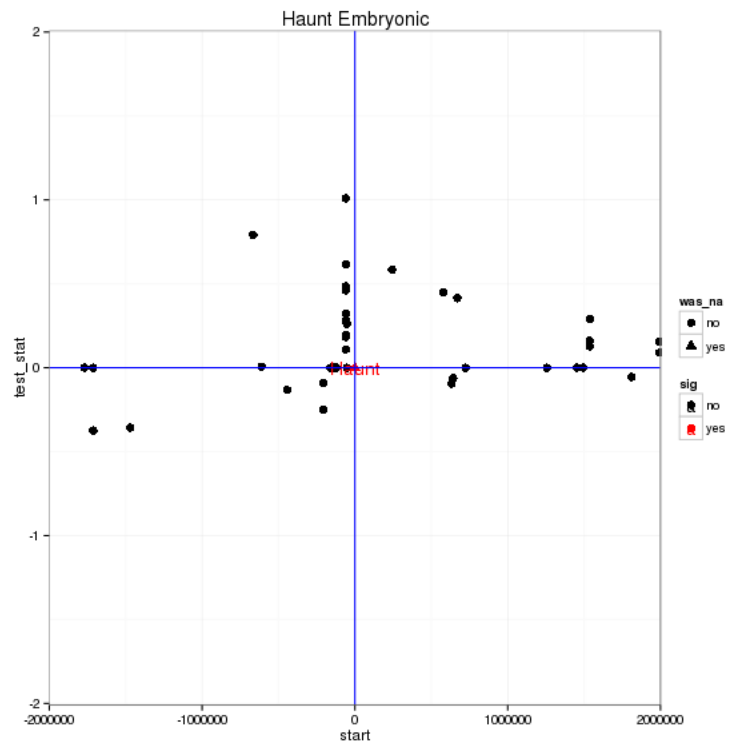
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR754
16 JR767
17 JR759

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	32555300.00	1.25	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	32555300.00	1.09	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	32555300.00	1.22	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	32555300.00	1.27	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	32555300.00	1.19	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	32555300.00	0.88	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	32555300.00	0.96	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	32555300.00	0.91	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	32555300.00	0.85	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	32555300.00	0.95	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	32555300.00	0.80	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	32555300.00	0.79	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	32555300.00	0.68	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	32555300.00	1.20	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR754/abundances.cxb	Haunt	0	Haunt_0	34277200.00	32555300.00	1.05	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR767/abundances.cxb	Haunt	1	Haunt_1	30963400.00	32555300.00	0.96	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR759/abundances.cxb	Haunt	2	Haunt_2	40179300.00	32555300.00	1.25	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1      KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1      reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1        zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Haunt -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Haunt_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/BrainMap/dat
## 2
## 3
## 4
## 5
```

Kantr KO vs WT (Adult)

This file shows the wt-v-ko comparison for Kantr.

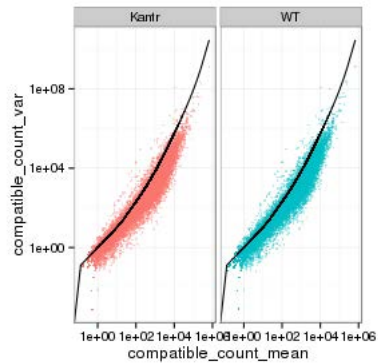
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

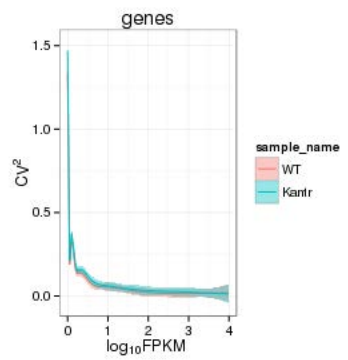
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



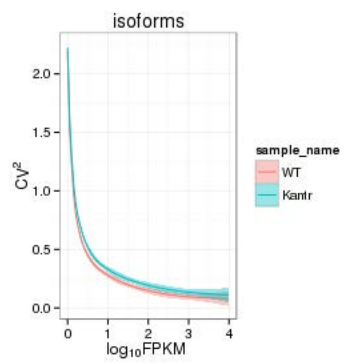
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

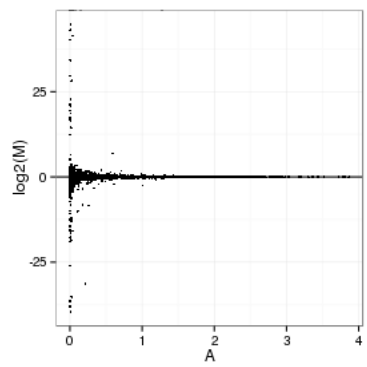
Genes:



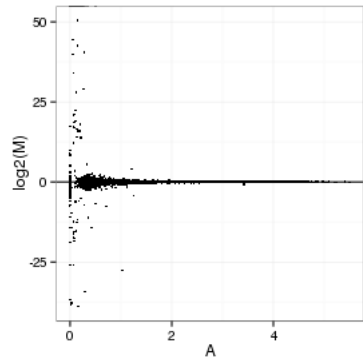
Isoforms:



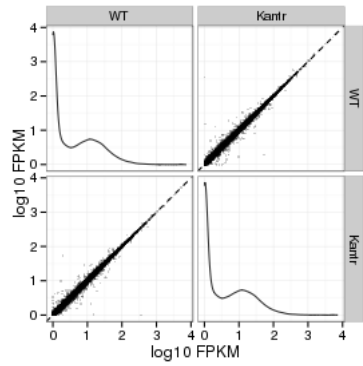
MvA plot



MvA plot counts



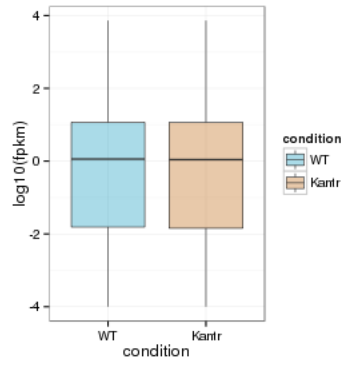
Scatterplot matrix



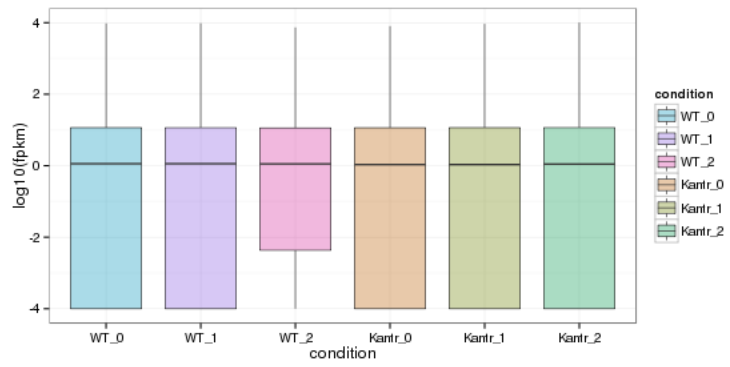
Distributions

Boxplots

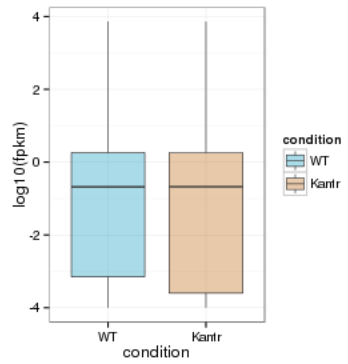
Boxplot (genes)



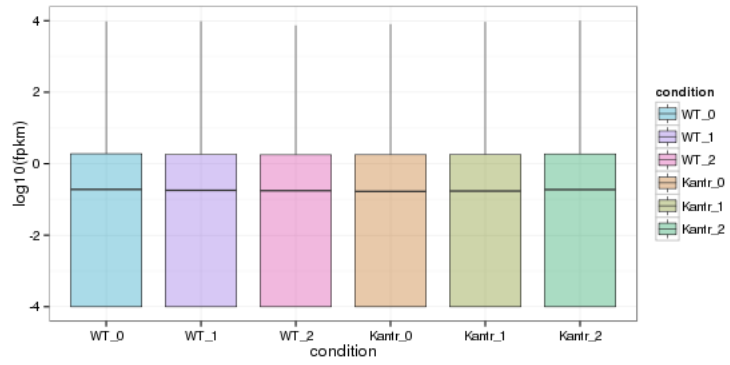
Boxplot (genes, replicates)



Boxplot (isoforms)

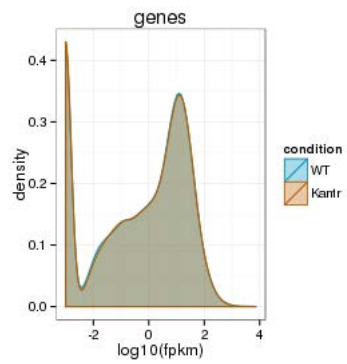


Boxplot (isoforms, replicates)

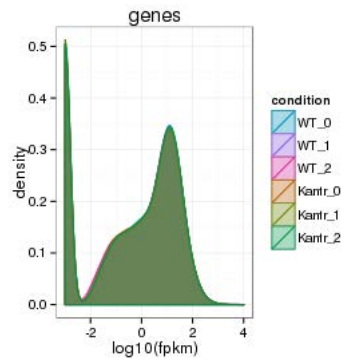


Density

Density (genes)

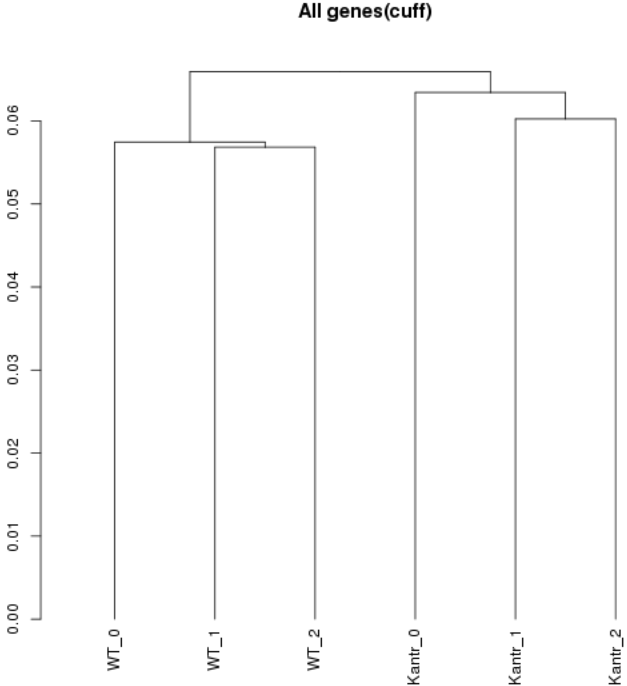


Density (genes, replicates)



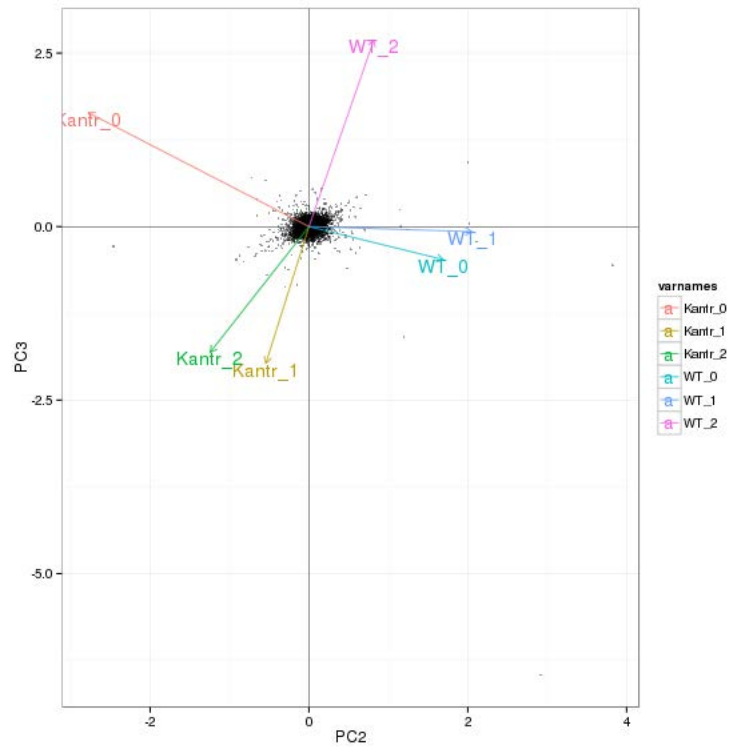
Clustering

Replicate Clusters

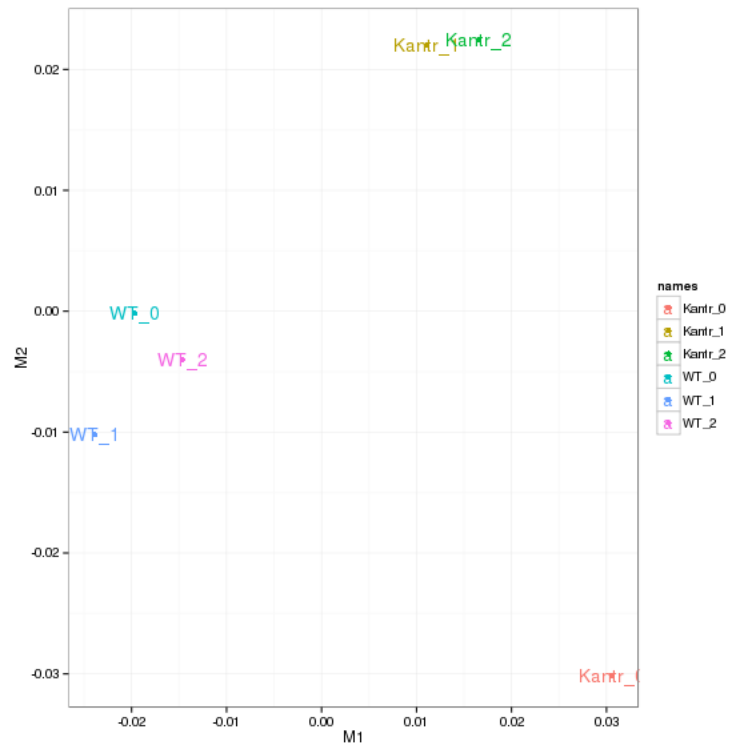


NULL

PCA (genes)

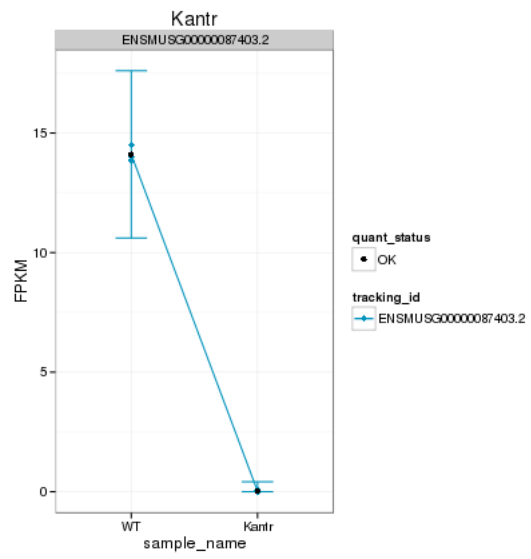


MDS (genes)

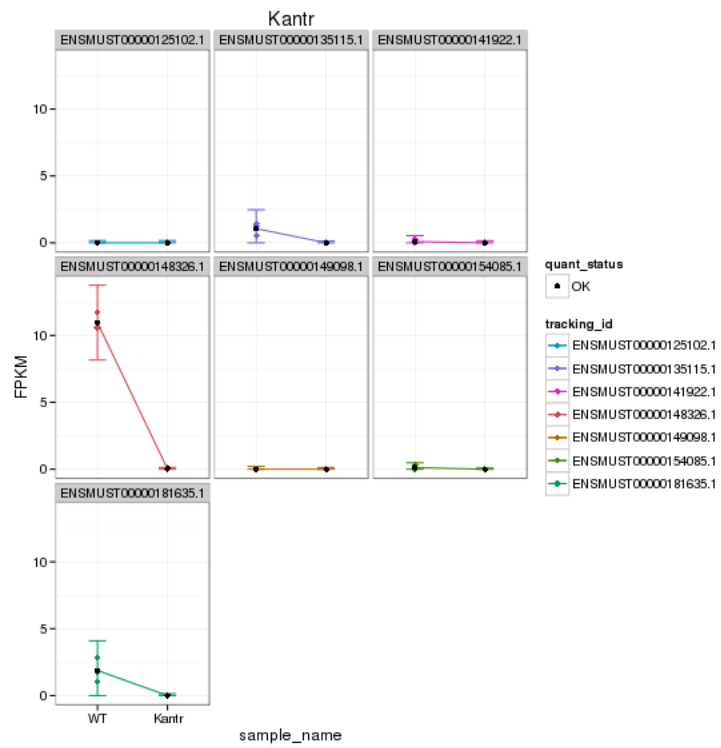


KO assessment

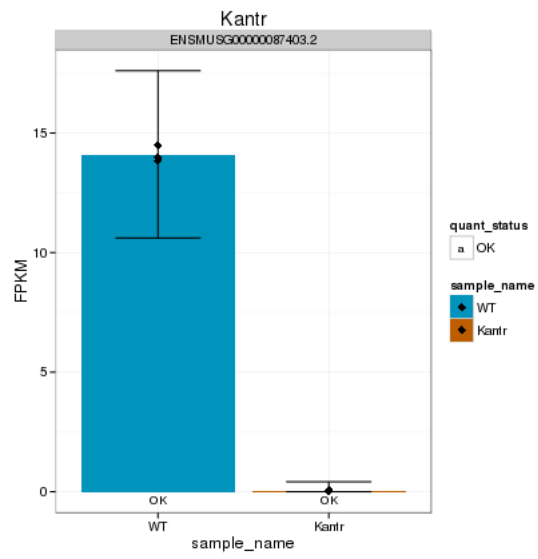
Endogenous IncRNA expression



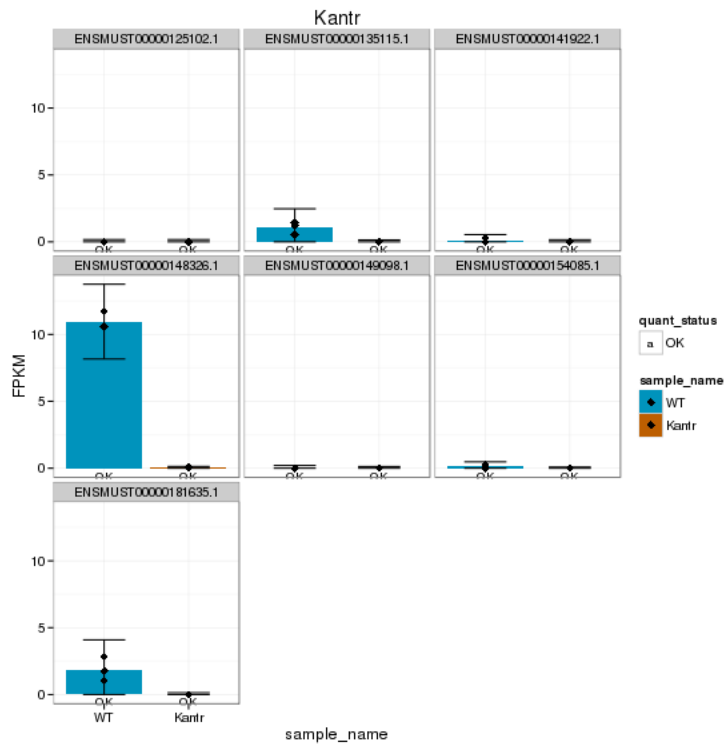
Endogenous expression of Kantr isoforms:



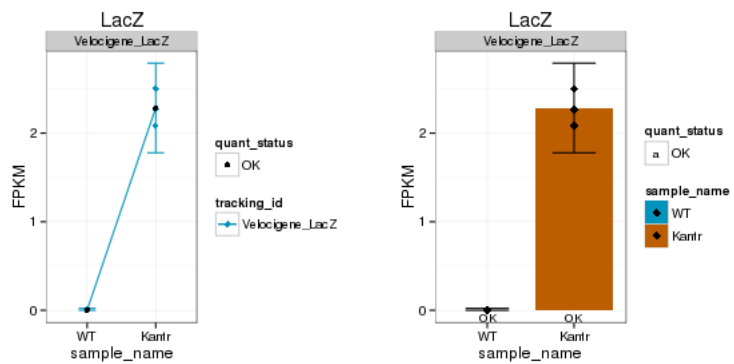
Barplot of gene expression:



Barplot of isoform expression:

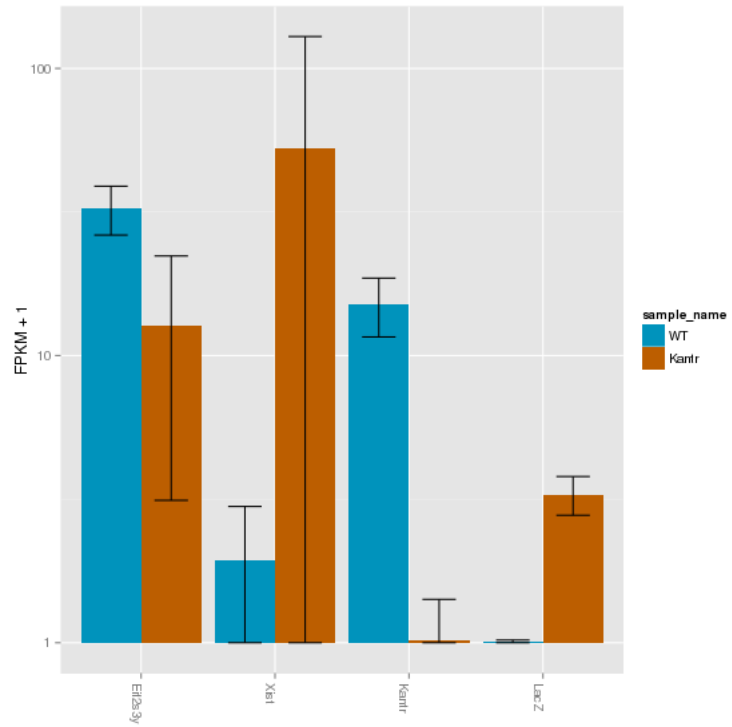


LacZ expression

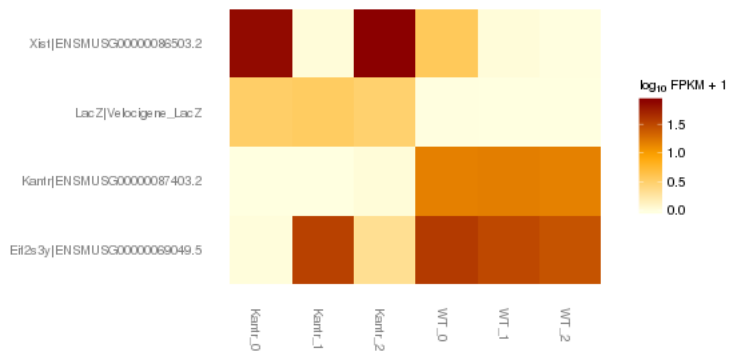


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



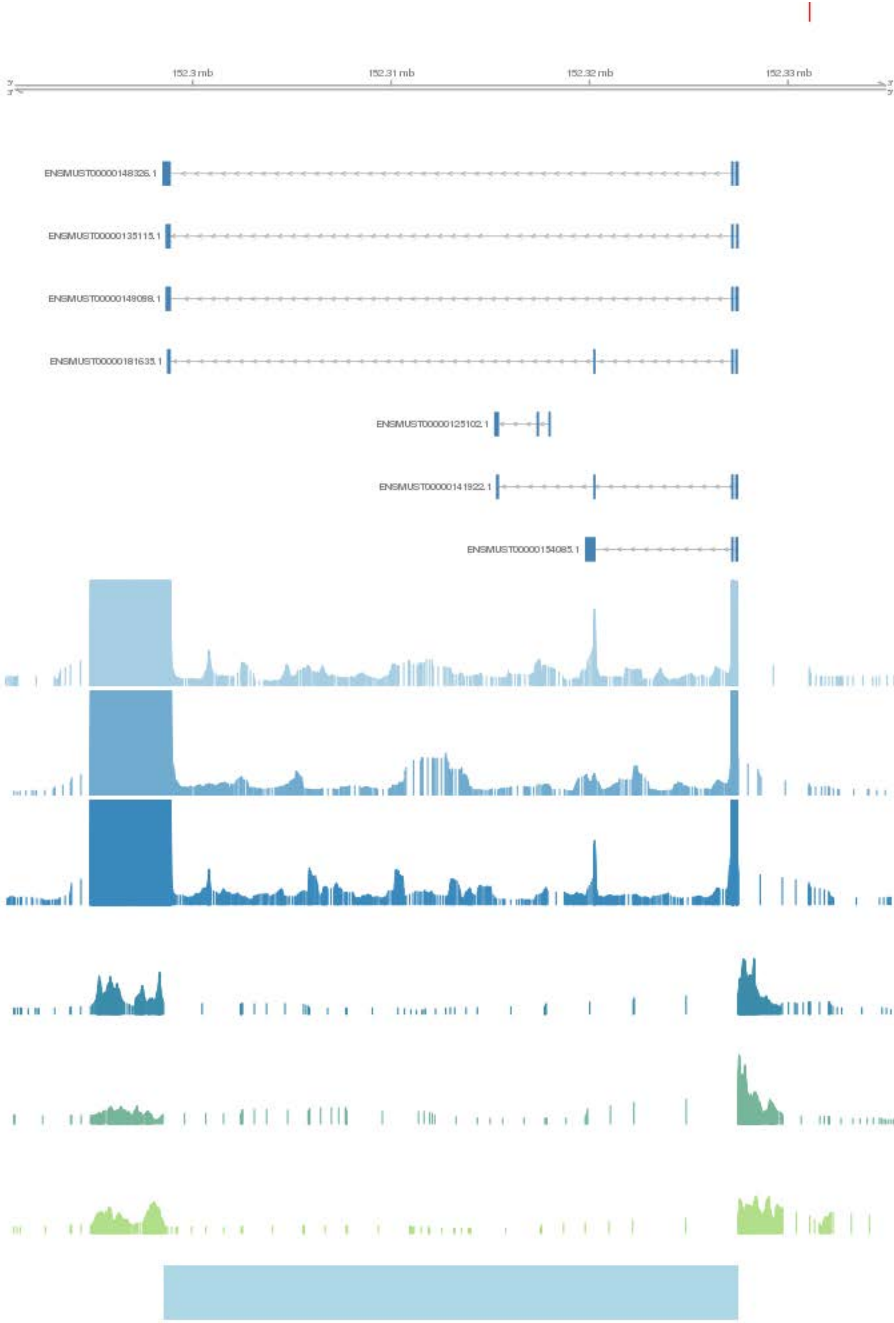
Expression heatmap:



Track visualization



KO Region Kantr_2 Kantr_1 Kantr_0 WT_2 WT_1 WT_0 LincRNA isoforms



Differential Analysis

Differential Genes

There are 315 significantly differentially expressed genes. They are:

	geneAnnot\$gene_short_name
1	Wnt9a
2	Sox9
3	Il12rb1
4	Mmp14
5	S100a5
6	N4bp3
7	Ramp2
8	Meox1
9	Gramd3
10	Plin4
11	Arrdc2
12	Fosb
13	Hif3a
14	9330159F19Rik
15	Cd33
16	Sh2b2
17	Osgep
18	Bcl2l1
19	Dio2
20	Id3
21	Fgfr1
22	Cd163
23	Derl3
24	Tbx1
25	Gm266
26	Tjap1
27	Trpv4
28	Mertk
29	Gata2
30	Slc25a13
31	Cnksr3
32	Sdc4
33	Rhbdl3
34	Ada
35	ErbB3
36	Ksr1
37	Etnppl

38 Pdk4
39 Lama4
40 Arid5b
41 Sgk1
42 Ddit4
43 Cpm

44 Nipal4
45 Btg2
46 Gatsl3
47 Igfbp3
48 Pomc
49 Gh
50 Nfkbia
51 Zfp3611
52 Net1
53 Fos
54 Dapk1
55 Elovl7
56 4930452B06Rik
57 Fst
58 Ldb3
59 Galnt15
60 Sema3g
61 Ednrb
62 Psm5
63 Fzd6
64 Fbxo32
65 Slc38a2
66 Bcl6
67 Crebbp
68 Arc
69 Nde1
70 Adamts1

71 Nr4a1
72 Cdkn1a
73 Tmem52
74 Clic5
75 Trem2
76 Glo1
77 Sik1
78 Xdh
79 Atp6v0c
80 Tek14

81 Dusp1
82 Fkbp5
83 Nedd4l
84 Cd74
85 Pdgfrb
86 Rab3il1
87 Fas
88 Frmd8
89 Hhex
90 Atp10a
91 Kdm5c
92 Pnpla2
93 D3Erttd751e
94 Sgk3
95 Paqr8
96 Slc40a1
97 Il1r1

98 2810459M11Rik

99 Myoc
100 Lcn2
101 Col5a1
102 Depdc7
103 Dll4
104 Bpifa1
105 Fam83d
106 Anxa5
107 Exosc9
108 Dclre1b
109 Tmem144
110 Cyr61
111 Trp53inp1
112 Gbp3
113 Gbp2
114 Nr4a3
115 Slc44a5
116 Rps6
117 Slc2a1
118 Mfsd2a
119 Alpl
120 Tinagl1
121 Map3k6
122 Errfi1
123 Ugt2a1

124 Gbp9
125 Aff1
126 Rasgef1b
127 Bmp3
128 Rilpl1
129 Uncx
130 Lfng
131 Nfe2l3
132 Gkn3
133 Klf15
134 Lag3
135 Clec2d
136 Adipor2
137 Dusp16
138 Rhpn2
139 Sult1a1
140 Lyve1
141 Cckbr
142 Slc38a5
143 Tsc22d3
144 Plat
145 Sall1
146 Gpt2
147 Gab1
148 Mt2
149 Mt1
150 Cmtm3
151 Ets1
152 Mcam
153 Ldlr

154 Paqr5
155 Igdcc3
156 Heyl
157 Gng11
158 Alox12b
159 Slc16a1
160 Sox10
161 Tbc1d4
162 Tlr13
163 Ston1
164 Klf9
165 Trim36
166 Neu4

167 Igsf9b
168 Trim59
169 Cbl
170 Pla2g3
171 Nostrin
172 Fam214a
173 Eva1a
174 Lars2
175 Tmem98
176 Prr5
177 Tmem260
178 Micall2
179 Kirrel2
180 Tenc1
181 Ppp1r14a
182 Mxd4
183 Galnt6
184 Uvssa
185 Avp
186 Dzip11
187 Egr2
188 Smim3
189 Foxf2
190 Egr1
191 Olfml2b
192 Atp5l
193 Mboat1
194 Pcsk1n
195 Tnfrsf10
196 Hlx
197 Atp8b1
198 Zfp189
199 Gpr37
200 Spsb1
201 Gbp5
202 Rps19
203 Cables1
204 Nkx6-2
205 Cldn5
206 Syt15
207 Cdc42ep4
208 Pkp2
209 Kihl6

210 Gjc2
211 Dmrt1
212 Zfp488
213 Tlr7
214 Serpinb1a
215 Lsm11
216 Rassf9
217 S1pr5
218 Cirbp
219 Insig1
220 Zfp36l2
221 Npas4
222 Clec14a
223 Gjb2
224 Hmga1
225 Kcne4
226 Gjb1
227 Kmt2d
228 Ccdc6
229 Dact2
230 Acap2
231 Cdc42ep1
232 Cyp2g1
233 Rasd1
234 Opalin
235 Fzd2
236 Ppp1r3g
237 Plekho2
238 Hs3st1
239 Olfml1
240 Lingo3
241 Islr2
242 Gfod1
243 BC068281
244 Klf13
245 Hbb-y
246 Gpr17
247 Hbb-bs
248 Nrros
249 Jun
250 Junb
251 Creb5
252 Ier2

253 Iigp1
254 Lifr
255 Lrrc8c
256 Zcchc24
257 Per2
258 S100a9
259 1810011O10Rik

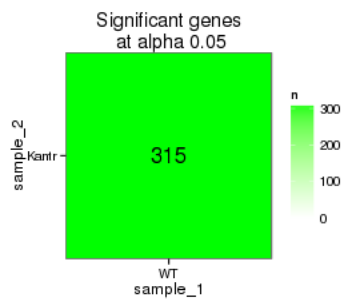
260 Kdm5d
261 Chsy3
262 Sclt1
263 Fhit

264 Zbtb40
265 Rpl26
266 Cox5b
267 Ksr2
268 Kdr
269 D630045J12Rik
270 Srrm4
271 Phactr4
272 Zbtb16
273 Ppp1r3c
274 Uty
275 Il3ra
276 Ddx3y
277 Eif2s3y
278 Ahnak
279 Hba-a1
280 1700084C01Rik
281 Rasgrp3
282 Jund
283 Egr4
284 Ccnb1ip1
285 Ppp1r3e
286 Gm7292
287 H2-BI
288 H2-Ab1
289 Tmem88b
290 Hbb-bt
291 Plekhf1
292 Adh1
293 Sox2
294 Bpifb4
295 B3galt5

- 296 Nrarp
- 297 Gm17414
- 298 Gbp4
- 299 Ms4a6c
- 300 Gm15631
- 301 Xist
- 302 BC064078
- 303 Slc5a3
- 304 Apold1
- 305 Sox2ot
- 306 Myl6
- 307 A930018M24Rik
- 308 Neat1
- 309 Gm10406
- 310 Gm7094
- 311 Fam181a
- 312 4732491K20Rik
- 313 Gm26782
- 314 Bin2
- 315 Gm26924

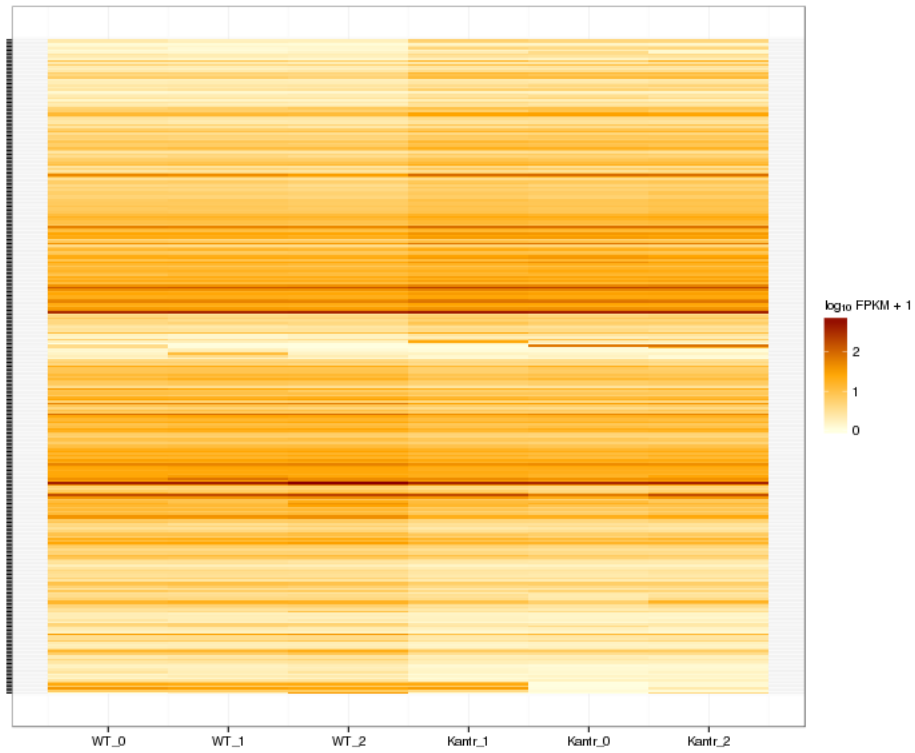
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

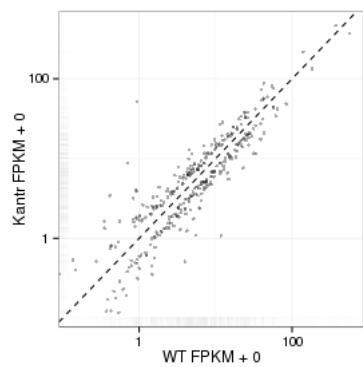


Significant genes with expression >50fpkm (any condition):(turned off)

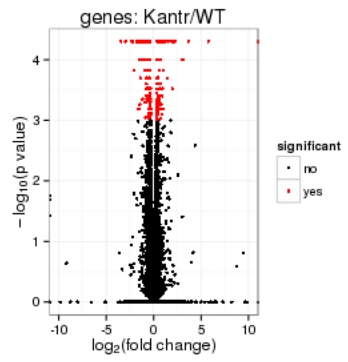
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

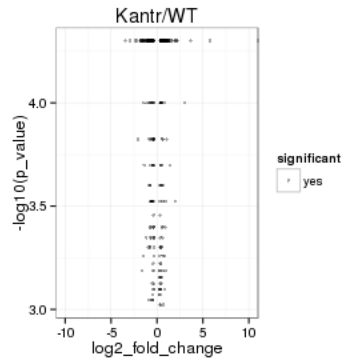
Scatter plot of significant genes only:



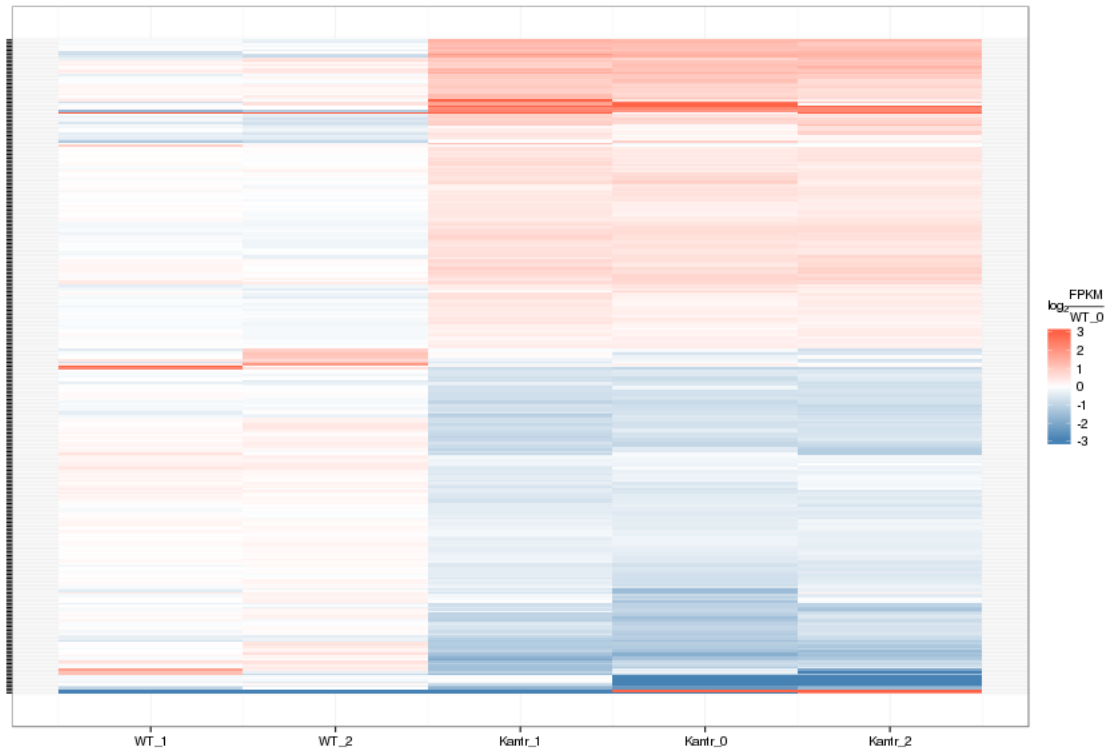
Volcano Plot



Volcano plot with significant genes only:



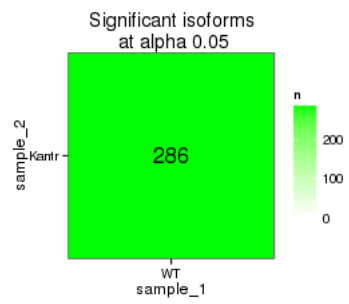
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

isoAnnot	gene_short_name
1	Sox9
2	Mmp14
3	N4bp3
4	Gramd3
5	Angptl4
6	Slc9a3r2
7	Golga2
8	Plin4
9	Arrdc2
10	Fosb
11	Hif3a
12	9330159F19Rik
13	Sh2b2
14	Prlr
15	Cyp2a5
16	Lrrc8a
17	Bcl2l1
18	Dio2
19	Id3
20	Derl3
21	Gm266
22	Med15
23	Tjap1
24	Mertk
25	Slc25a13
26	Cnksr3
27	Notch4
28	Camk1g
29	Sdc4
30	Ada
31	ErbB3
32	Ksr1
33	Pdk4
34	Sema6a
35	Lama4
36	Arid5b
37	Sgk1
38	Ddit4
39	Vps54
40	Ddc

41 Cpm
42 Nipal4
43 Tns3
44 Btg2
45 Gatsl3
46 Pomc
47 Pxdn
48 Gh
49 Helz
50 Nfkbia
51 Zfp36l1
52 Otub2
53 Net1
54 Fos
55 Zkscan3
56 Elovl7
57 4930452B06Rik
58 Fst
59 Galnt15
60 Sema3g
61 Psmb5
62 Myo10
63 Slc38a2

64 Litaf

65 Bcl6
66 Arc
67 Adamts1
68 Slc15a2
69 Nr4a1
70 Cdkn1a
71 Vegfa
72 Clic5
73 Glo1
74 Sik1
75 Xdh
76 Atp6v0c
77 Tekt4
78 Dusp1
79 Fkbp5
80 Sncaip
81 Pdgrfb
82 Fas
83 Frmd8

84 Uhrf2
85 Hhex
86 Alas2
87 Atp10a
88 Tma16
89 D3Ertid751e
90 Sgk3
91 Paqr8
92 Slc40a1
93 Lamc1
94 Myoc
95 Lypd6b
96 Lcn2
97 Depdc7
98 Jag1
99 Dll4
100 Nkx2-2
101 Bpifa1
102 Fam83d
103 Anxa5
104 Exosc9
105 Dclre1b
106 Fam198b
107 Alpk1
108 Cyr61
109 Gbp2
110 Nr4a3
111 Slc2a1
112 Mfsd2a
113 Hspg2
114 Tinagl1
115 Map3k6
116 Arhgef19
117 Errfi1
118 Ugt2a1
119 Aff1

120 Rasgef1b
121 Lfng
122 Nfe2l3
123 Gkn3
124 Klif15
125 Lag3
126 Clec2d

127 Adipor2
128 Dusp16
129 Slco1a4
130 Rhpn2
131 Sult1a1
132 Disp1
133 Lyve1
134 Cckbr
135 Slc38a5
136 Col4a5
137 Mtm1
138 Tsc22d3
139 Plat
140 Sall1
141 Gpt2
142 Gab1
143 Mt2
144 Mt1
145 Cmtm3
146 Ets1
147 Ldlr
148 Heyl
149 Gng11
150 Alox12b
151 Rnft2
152 Slc16a1
153 Sox10
154 Tbc1d4
155 Ucp2
156 Klf9
157 Trim36
158 Igsf9b
159 Trim59
160 Nostrin
161 Eva1a
162 Lars2
163 Tmem98
164 Prr5
165 Tmem260
166 Micall2
167 Kirrel2
168 Ppp1r14a
169 Mxd4
170 4932438A13Rik

171 Ovol2
172 Avp
173 Egr2
174 Sh3rf3

175 Smim3
176 Galnt11
177 Foxf2
178 Egr1
179 Olfml2b
180 Atp5l
181 Mboat1
182 Pcsk1n
183 Tnfsf10
184 Atp8b1
185 Zfp189
186 Cercam
187 Gpr37
188 Spsb1
189 Gjb6
190 Gbp5
191 Erf
192 Cables1
193 Nkx6-2
194 Cldn5
195 Syt15
196 Sdk2
197 Cdc42ep4
198 Pkp2
199 Greb1l
200 Aplnr
201 Zfp488
202 Tlr7
203 Serpinb1a
204 Rassf9
205 Cirbp
206 Insig1
207 Zfp36l2
208 Npas4
209 Clec14a
210 Gjb2
211 Kcne4
212 Gjb1
213 Kmt2d

214 Ranbp3l
215 Ccdc6
216 Dact2
217 Acap2
218 Cdc42ep1
219 Rasd1
220 Opalin
221 Fzd2
222 Ppp1r3g
223 Plekho2
224 Olfml1
225 Klf13

226 Hbb-y
227 Gpr17
228 Hbb-bs
229 Junb

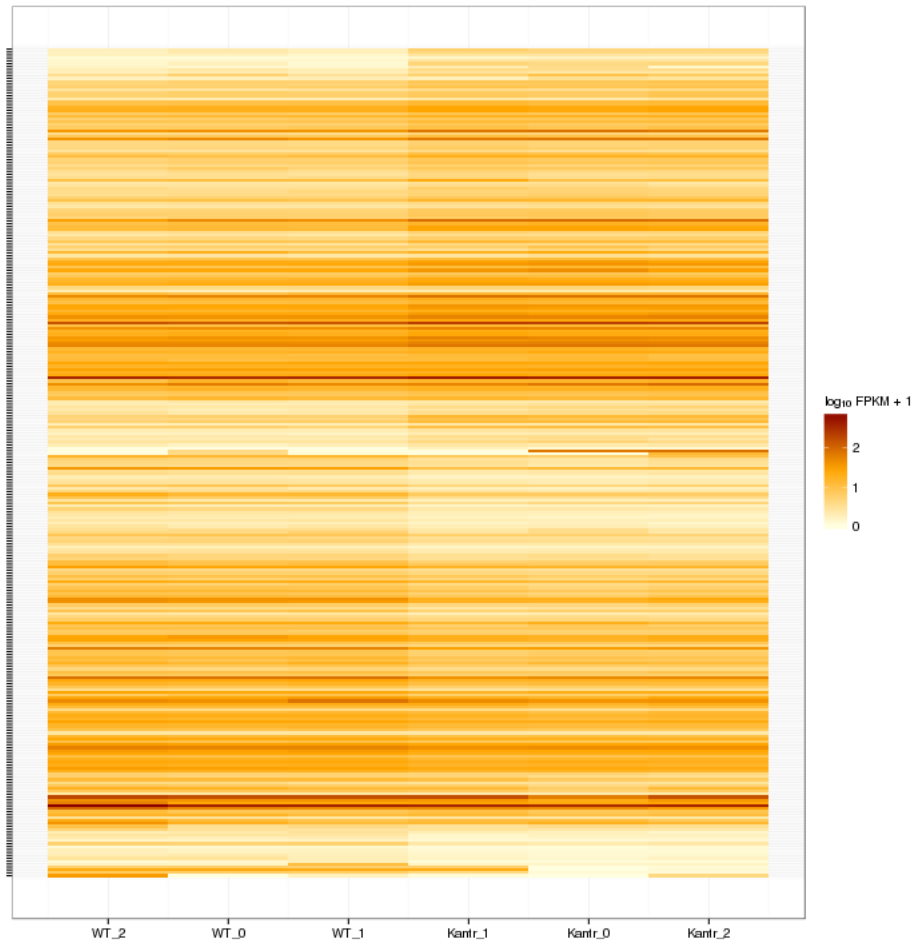
230 Creb5
231 Ier2
232 Lifr
233 Lrrc8c
234 Wdr86
235 Per2
236 Pla2g4a
237 Ncoa4
238 1810011O10Rik
239 Kdm5d
240 Chsy3
241 Calcr1
242 Zbtb40
243 Rpl26
244 Cxcl12
245 Cox5b
246 Kdr
247 Hspb11
248 Slc22a8
249 Srrm4
250 Zfp428
251 Cntn4
252 Zbtb16
253 Ppp1r3c
254 Ddx3y
255 Ahnak
256 170084C01Rik

257 Jund
258 Egr4
259 Ccnb1ip1
260 Ppp1r3e
261 Gm7292
262 H2-BI
263 Tmem88b
264 Hbb-bt
265 Plekhf1
266 Adh1
267 B3galt5
268 Nrarp
269 Aoc2
270 Gm3411
271 Nhs12
272 Gm15631
273 Xist
274 BC064078
275 Apold1
276 Sox2ot
277 Myl6
278 Neat1
279 Malat1

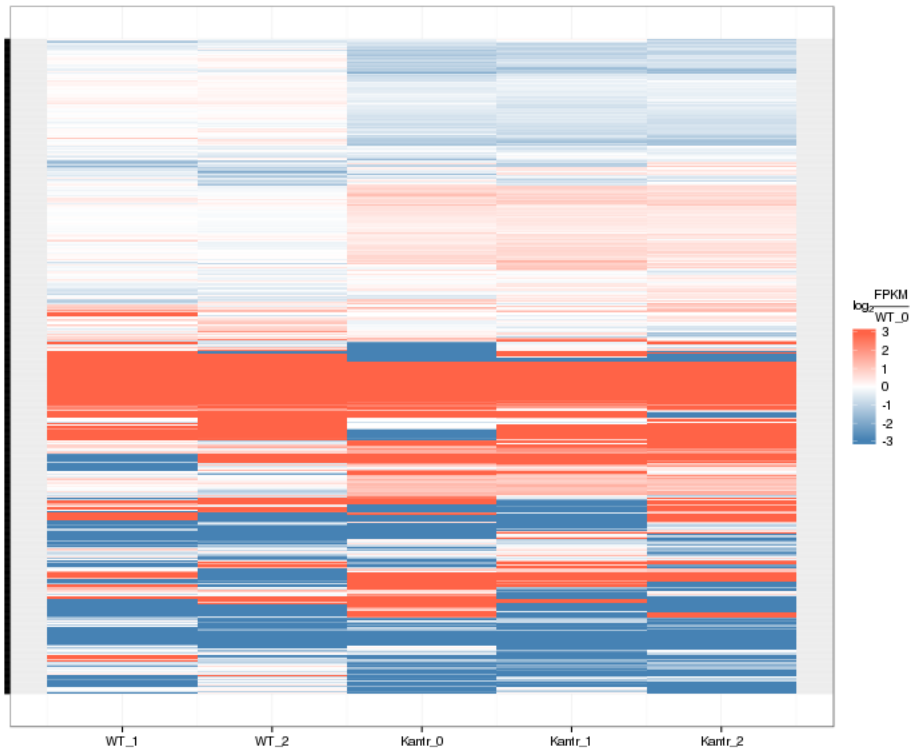
280 Gm21092
281 Gm7094
282 Fam181a
283 Gm26782
284 Bin2

285 Gm26924

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

Biocarta enrichment:

```
## Error: `x` must have at least 2 rows and 2 columns
```

Biocarta zscore:

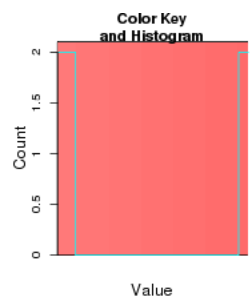
```
## Error: incorrect number of dimensions
```

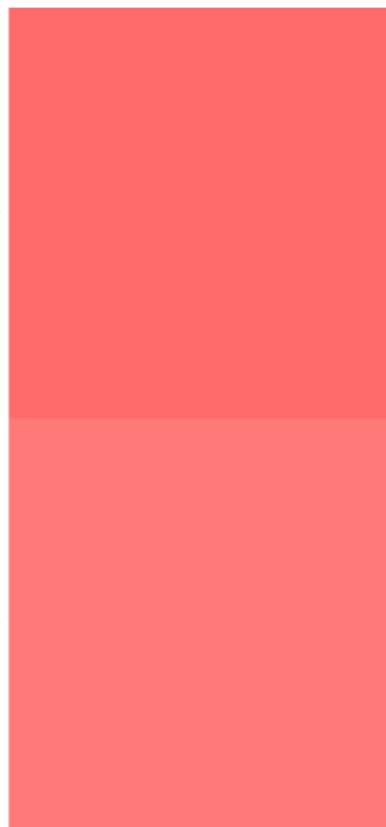
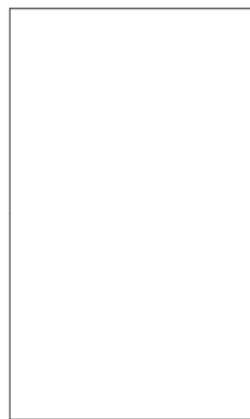
```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Reactome enrichment:

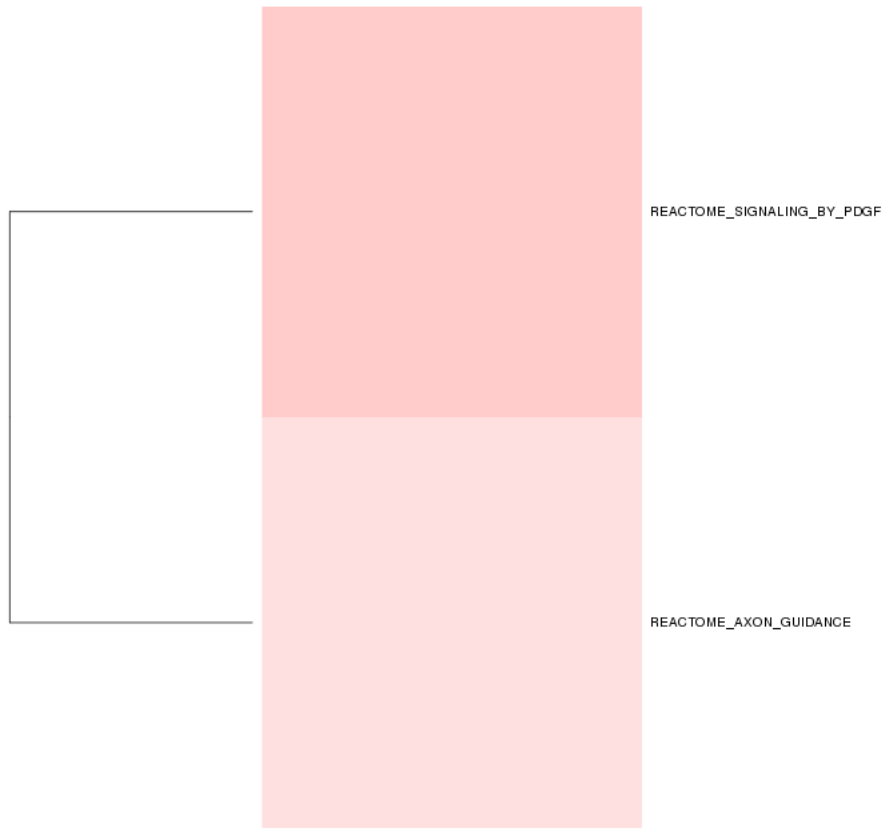
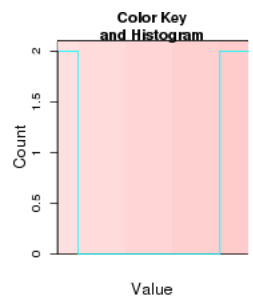




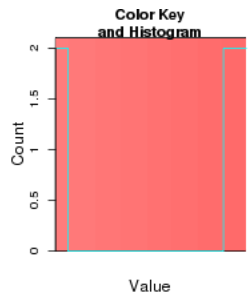
REACTOME_SIGNALING_BY_PDGF

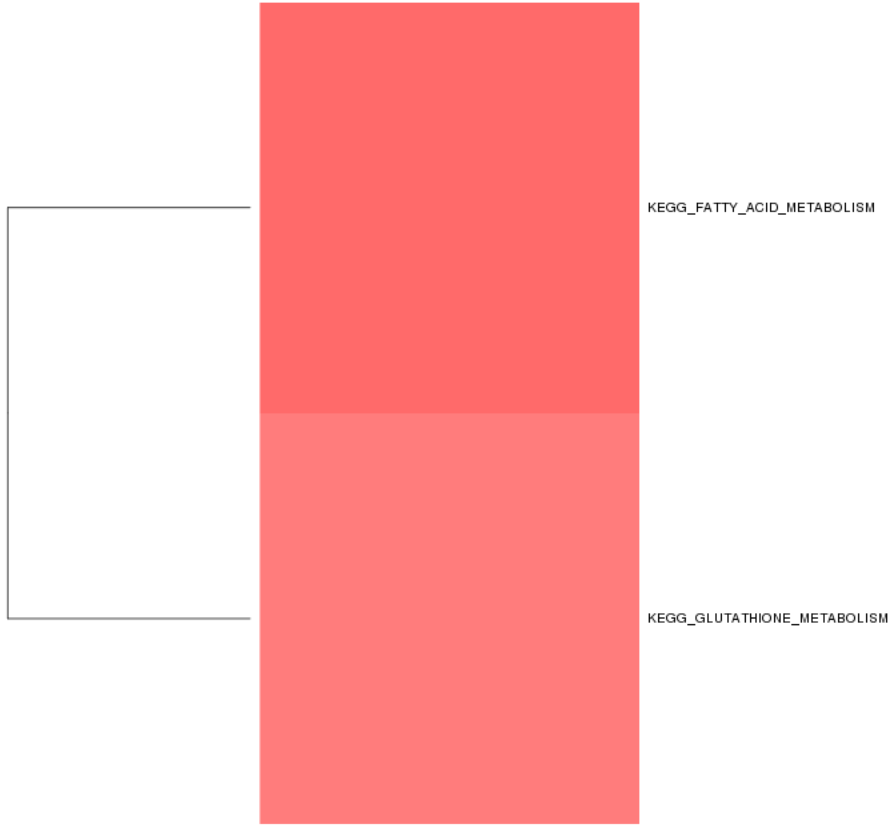
REACTOME_AXON_GUIDANCE

Reactome zscore:

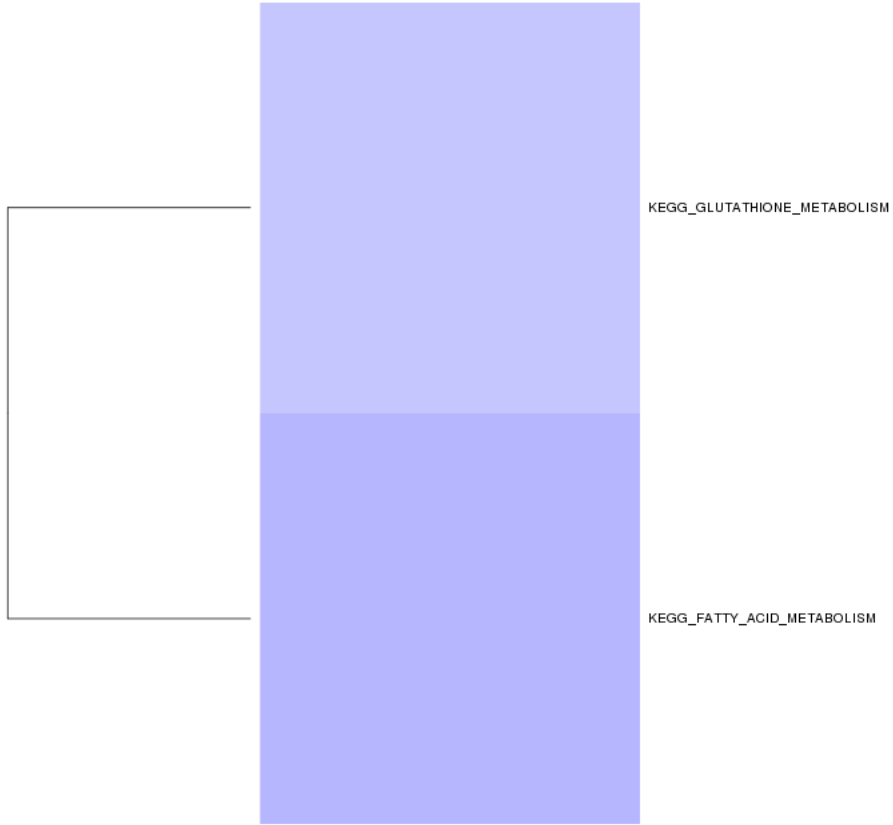
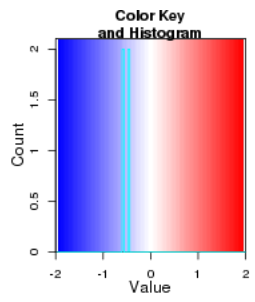


Kegg enrichment:



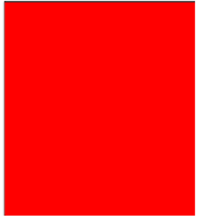


Kegg zscore:



Interneuron enrichment:

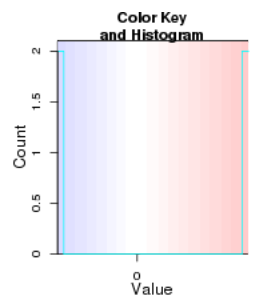
```
## Error: no locations are finite
```

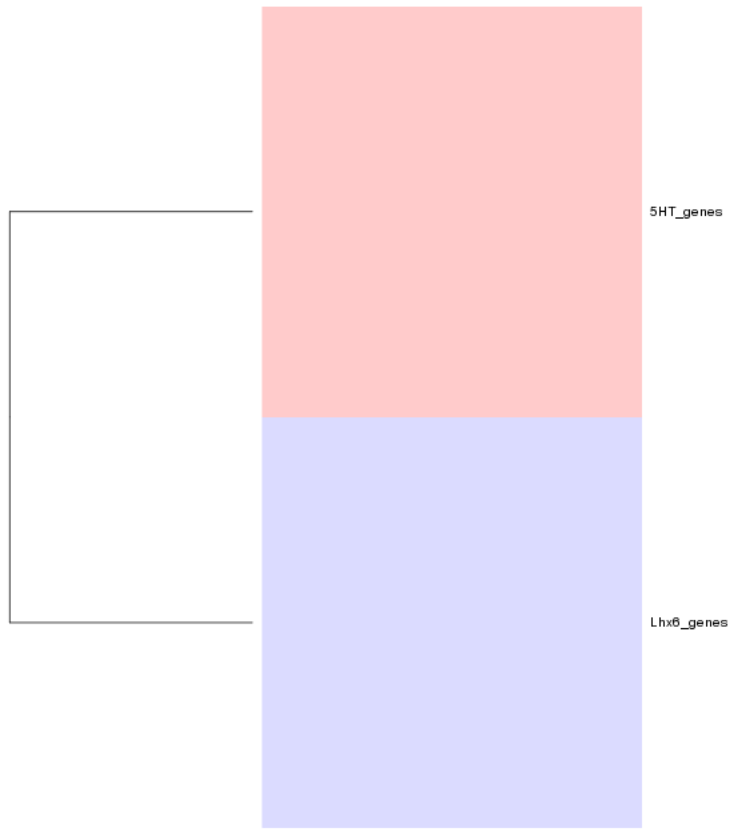


5HT_genes

Lhx6_genes

Interneuron zscore:





Oncogene enrichment:

```
## Error: argument is of length zero
```

Oncogene zscore:

```
## Error: incorrect number of dimensions
```

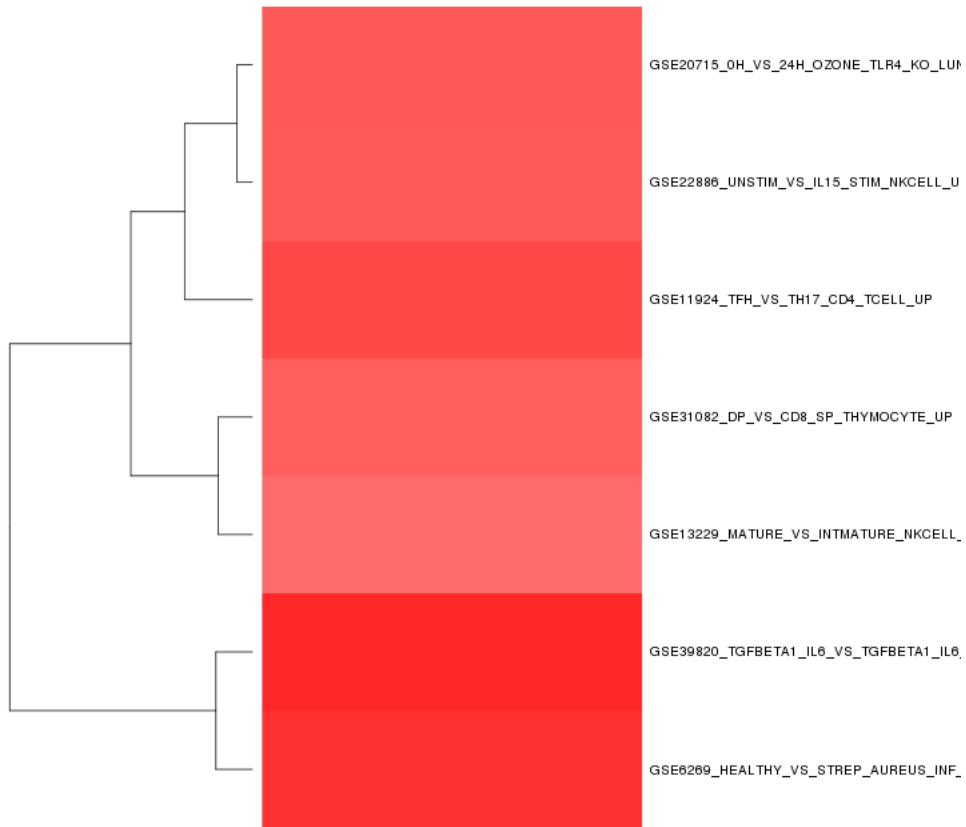
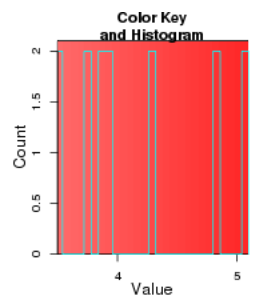


```
## Error: object 'x_ordered' not found
```

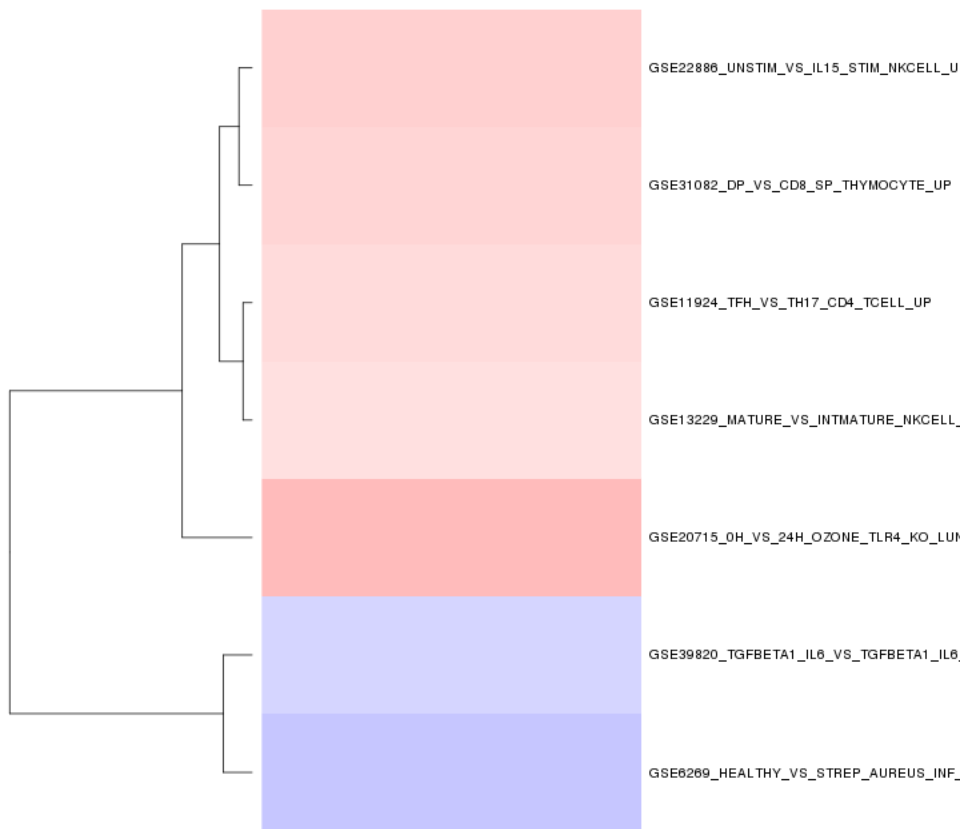
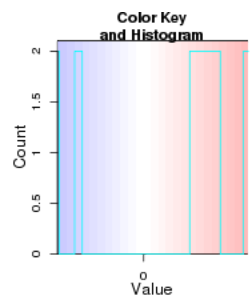
```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

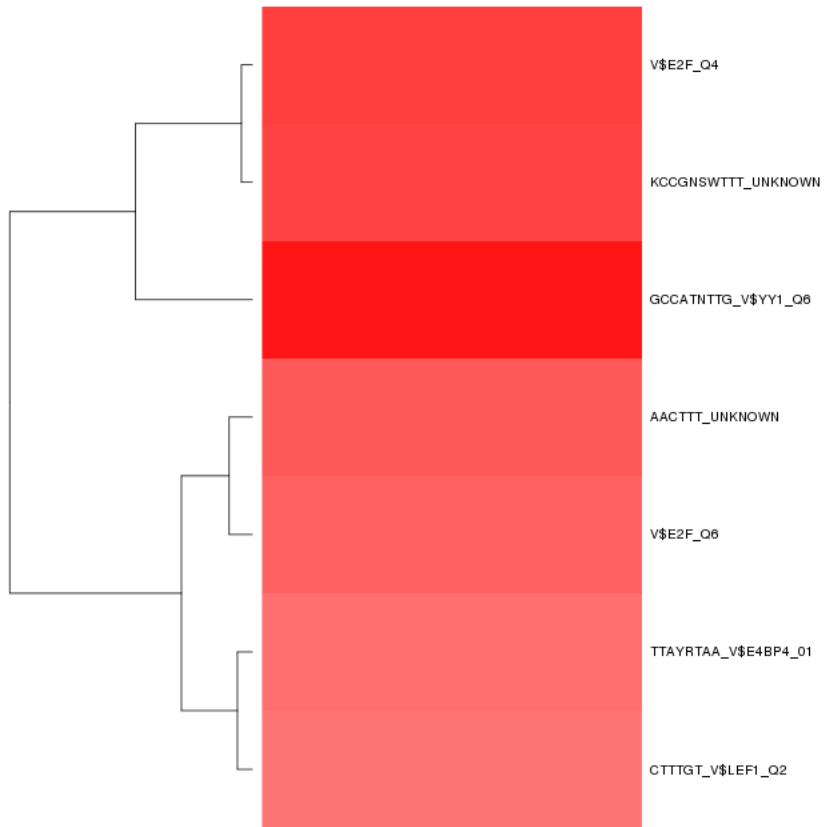
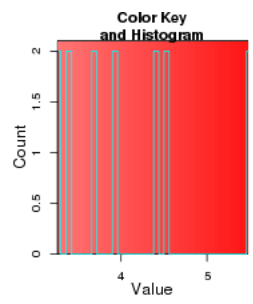
Immuno enrichment:



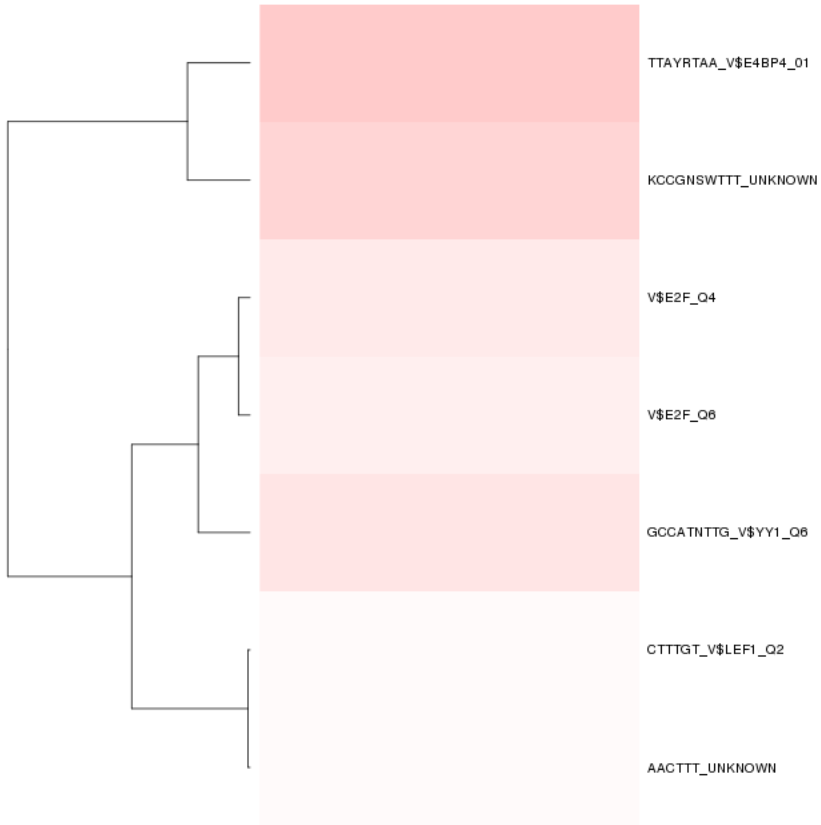
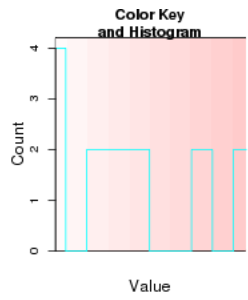
Immuno zscore:



TF enrichment:

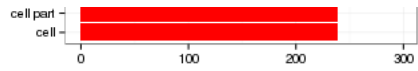


TF zscore:



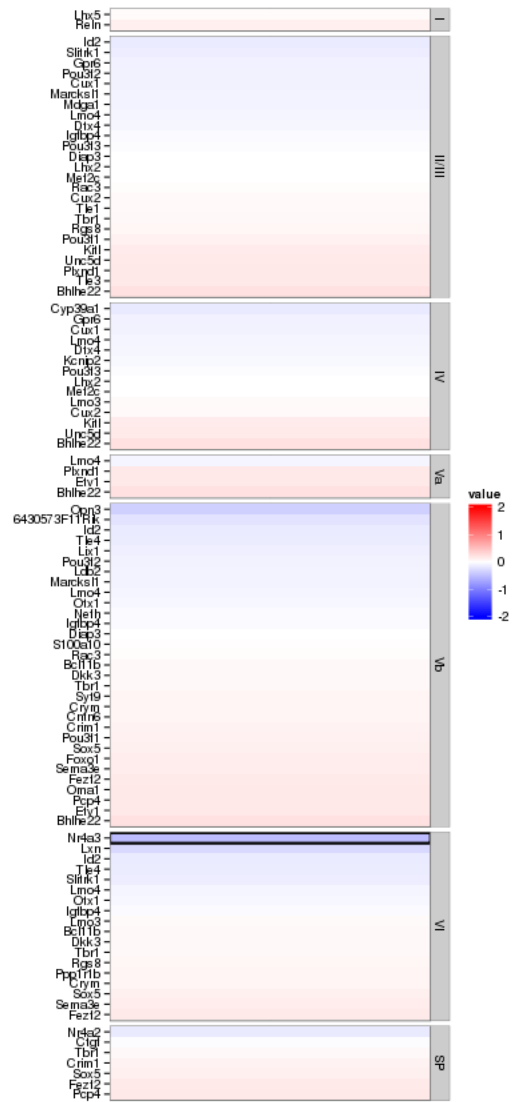
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

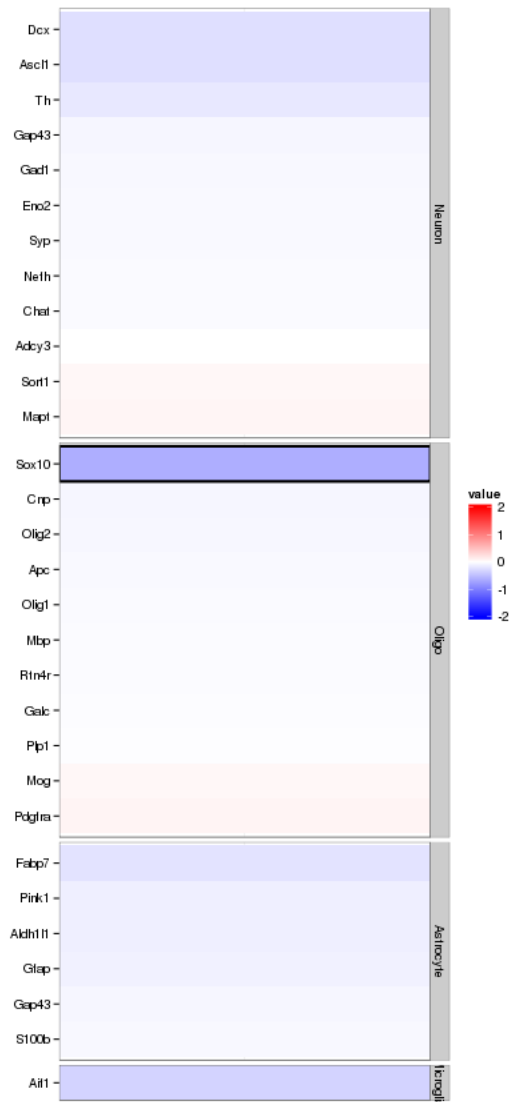


```
## Error: 'x' and 'units' must have length > 0
```

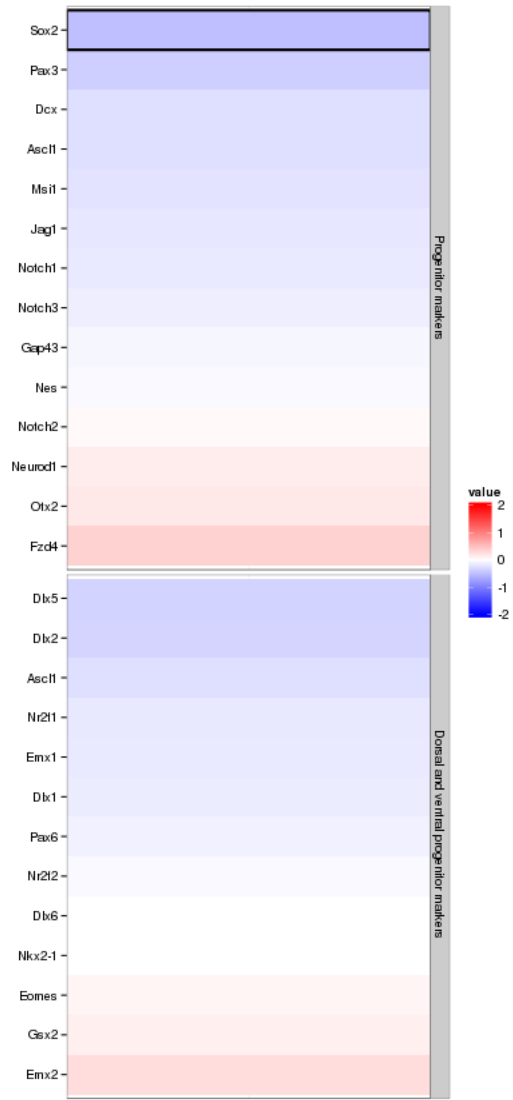
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



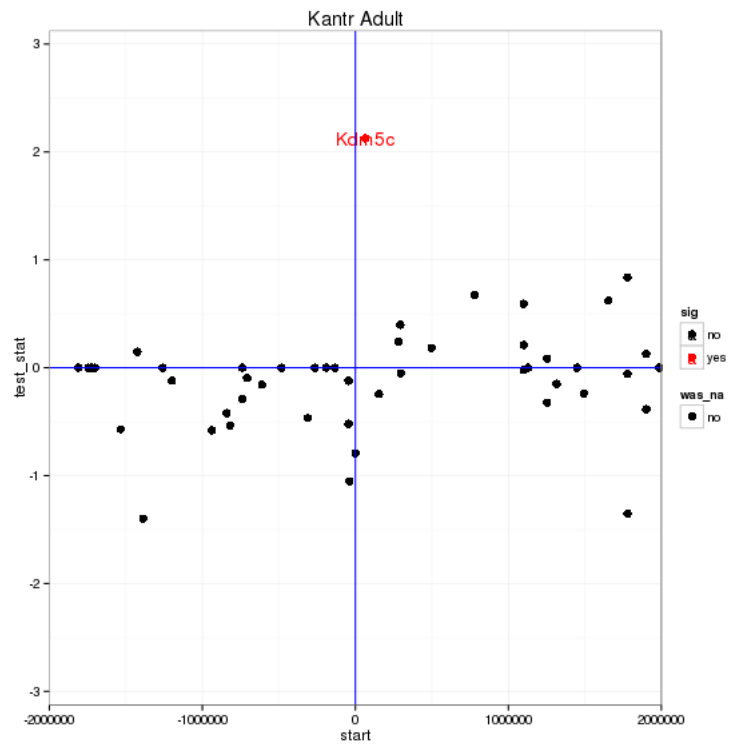
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

log2 Foldchange and test statistic are calculated with the ratio of fpkm(KO)/fpkm(WT), thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 1 genes significantly regulated in a region this size is: 0.4783



Notes

Samples used are:

10

- 1 JR824
- 2 JR820
- 3 JR821
- 4 JR742
- 5 JR819
- 6 JR823

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/loff/Projects/BrainMap/data/quants/JR824/abundances.cxb	WT	0	WT_0	34306900.00	36012300.00	0.96	1.00
2 /n/rinn_data1/seq/loff/Projects/BrainMap/data/quants/JR820/abundances.cxb	WT	1	WT_1	36256500.00	36012300.00	1.01	1.00
3 /n/rinn_data1/seq/loff/Projects/BrainMap/data/quants/JR821/abundances.cxb	WT	2	WT_2	45848800.00	36012300.00	1.29	1.00
4 /n/rinn_data1/seq/loff/Projects/BrainMap/data/quants/JR742/abundances.cxb	Kantr	0	Kantr_0	35615300.00	36012300.00	0.98	1.00

```
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR819/abundances.cxb Kantr
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR823/abundances.cxb Kantr
```

```
1 Kantr_1 32408700.00 36012300.00 0.89 1.00
2 Kantr_2 33428000.00 36012300.00 0.92 1.00
```

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8       LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8   LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C              LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
```

```

## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRT_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7    tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```

##      param
## 1    cmd_line
## 2    version
## 3  SVN_revision
## 4  boost_version
## 5    genome
##
## 1 cuffdiff -p 10 -L WT,Kantr -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Kantr_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/ar
## 2
## 3
## 4
## 5

```

Kantr KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Kantr.

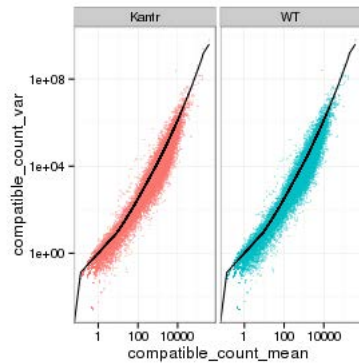
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

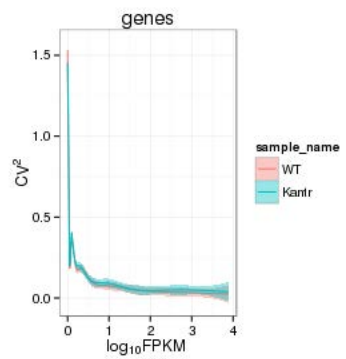
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



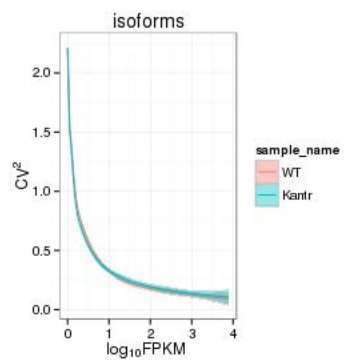
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

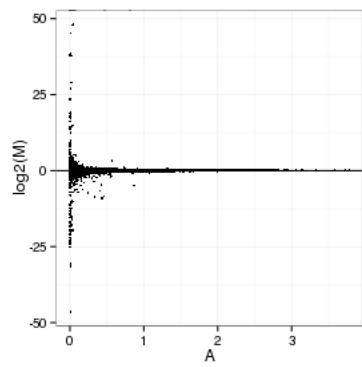
Genes:



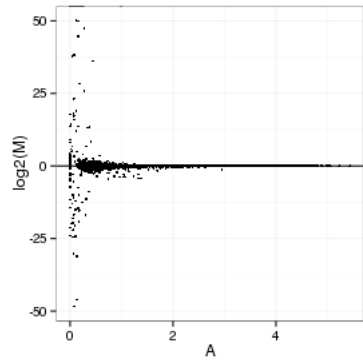
Isoforms:



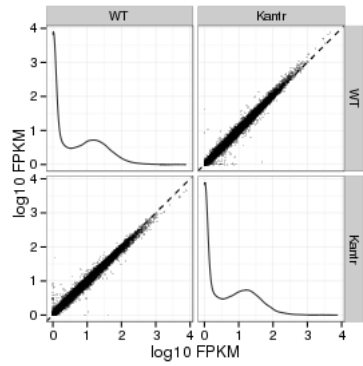
MvA plot



MvA plot counts



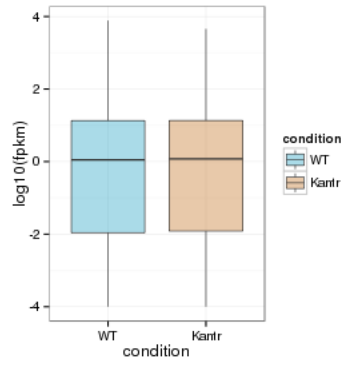
Scatterplot matrix



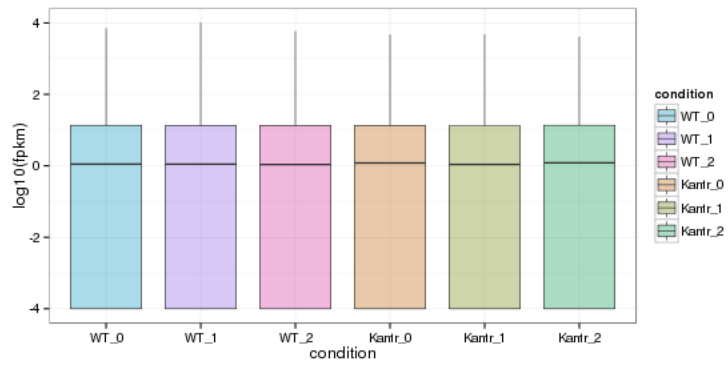
Distributions

Boxplots

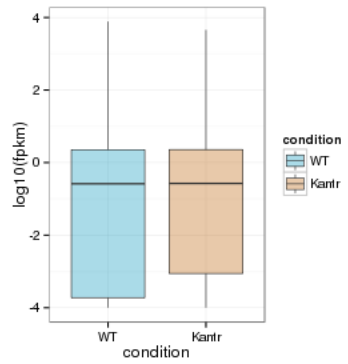
Boxplot (genes)



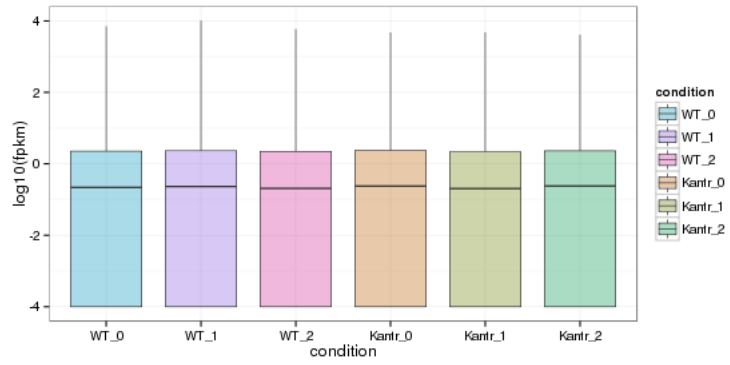
Boxplot (genes, replicates)



Boxplot (isoforms)

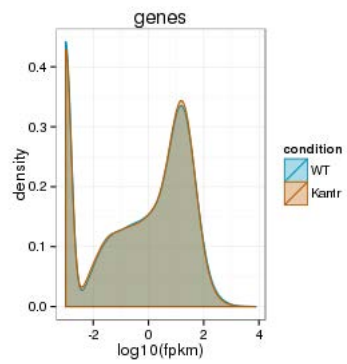


Boxplot (isoforms, replicates)

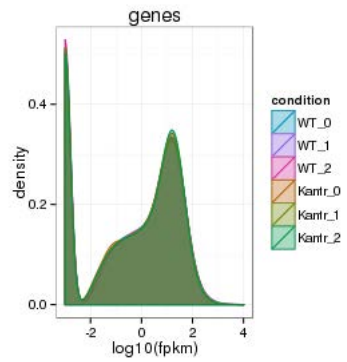


Density

Density (genes)

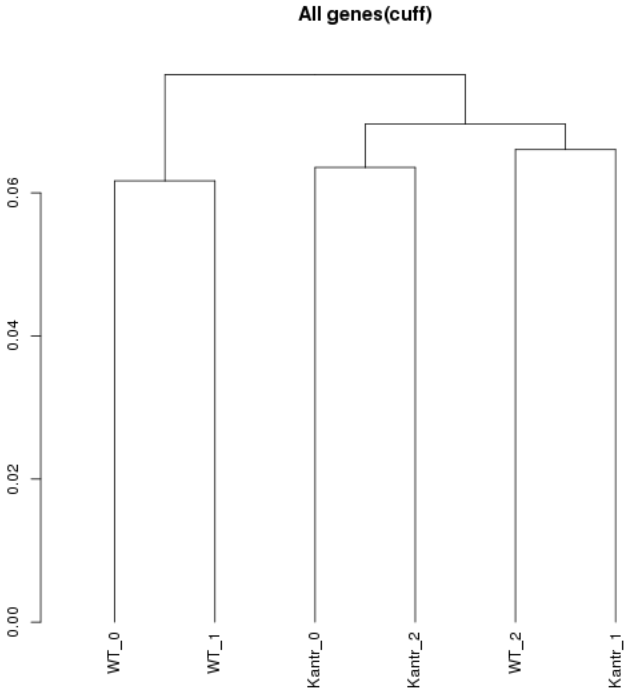


Density (genes, replicates)



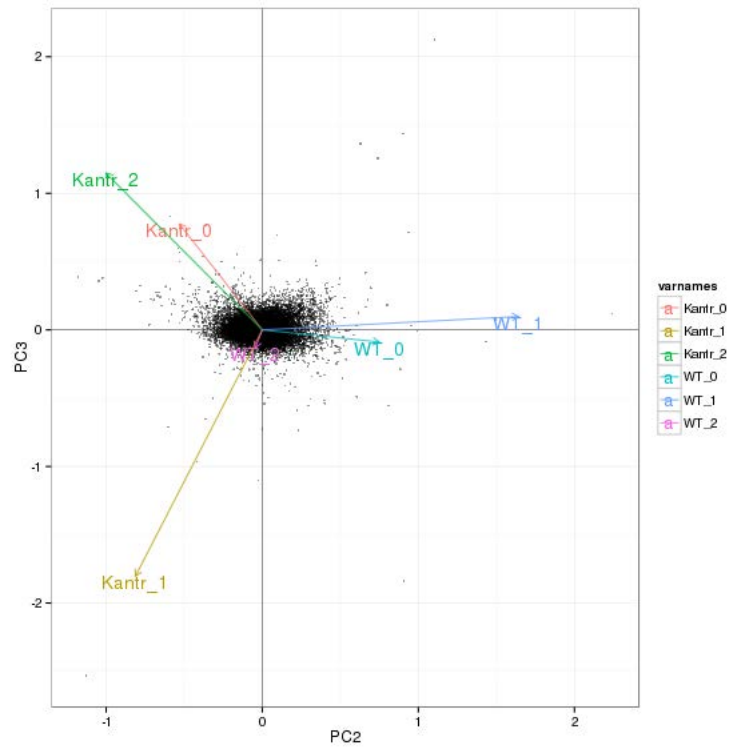
Clustering

Replicate Clusters

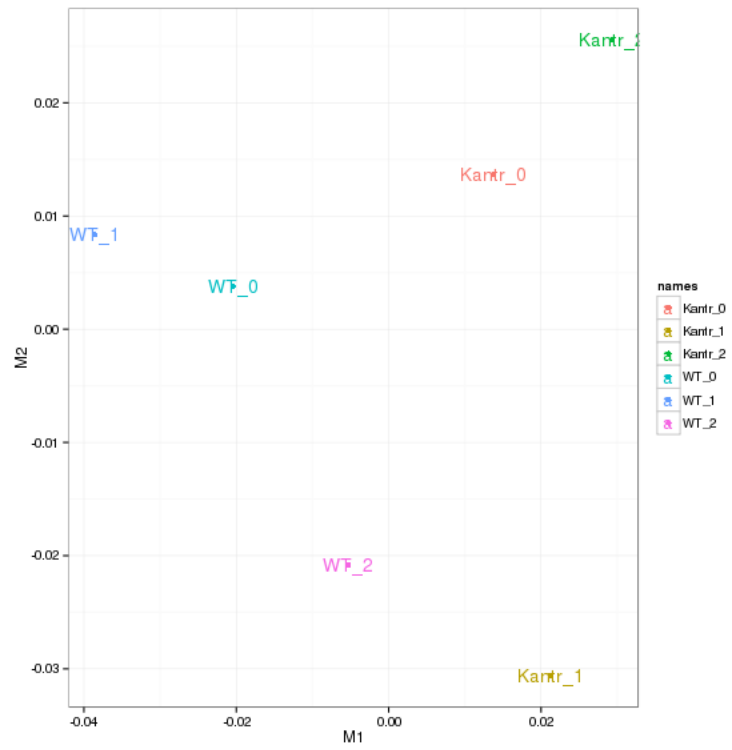


NULL

PCA (genes)

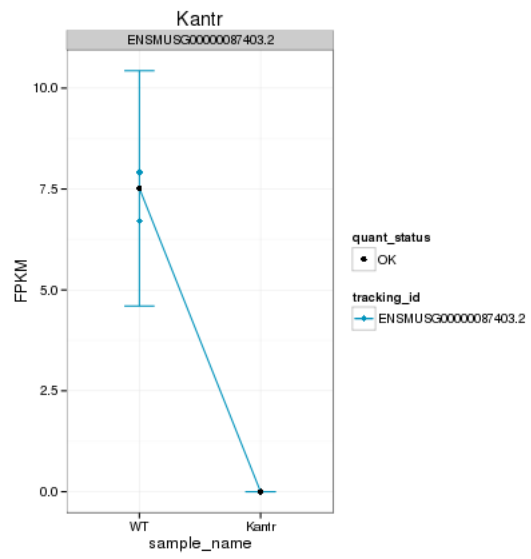


MDS (genes)

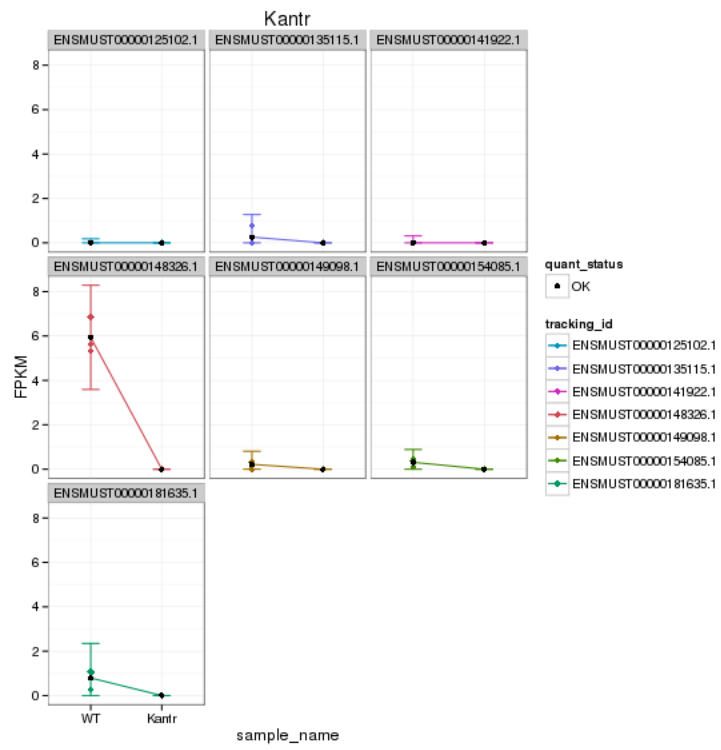


KO assessment

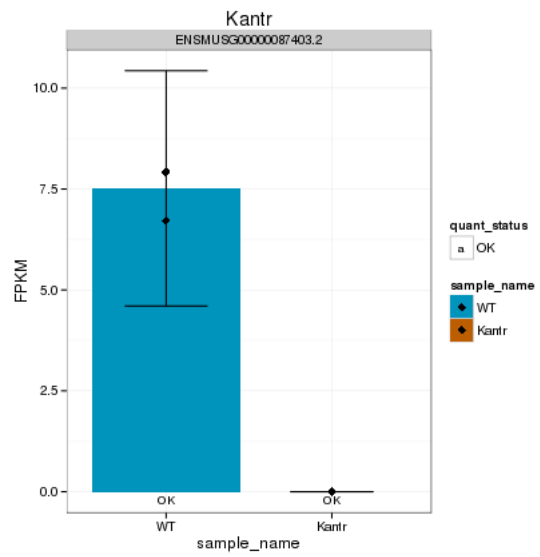
Endogenous lncRNA expression



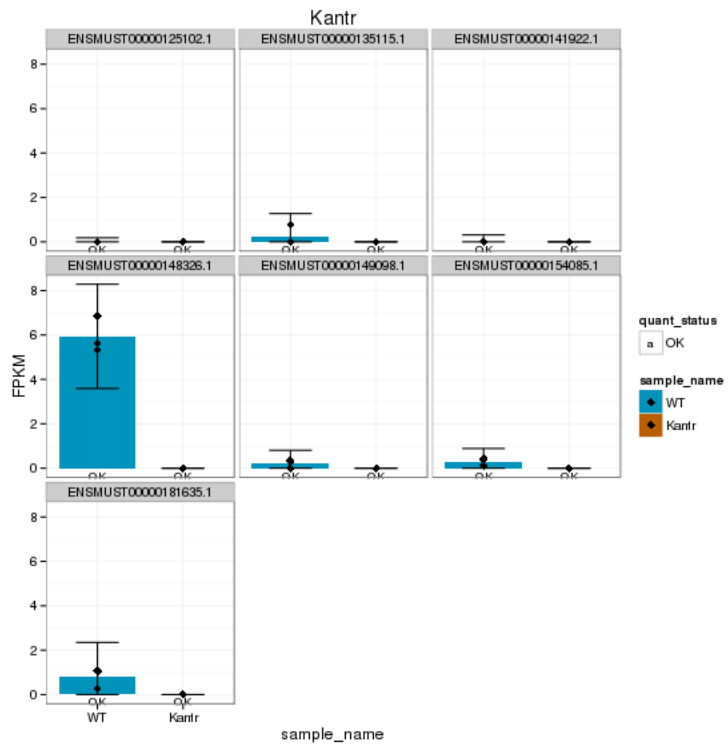
Endogenous expression of Kantr isoforms:



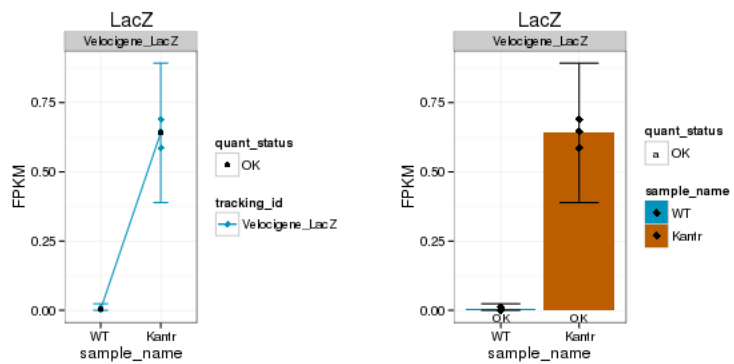
Barplot of gene expression:



Barplot of isoform expression:

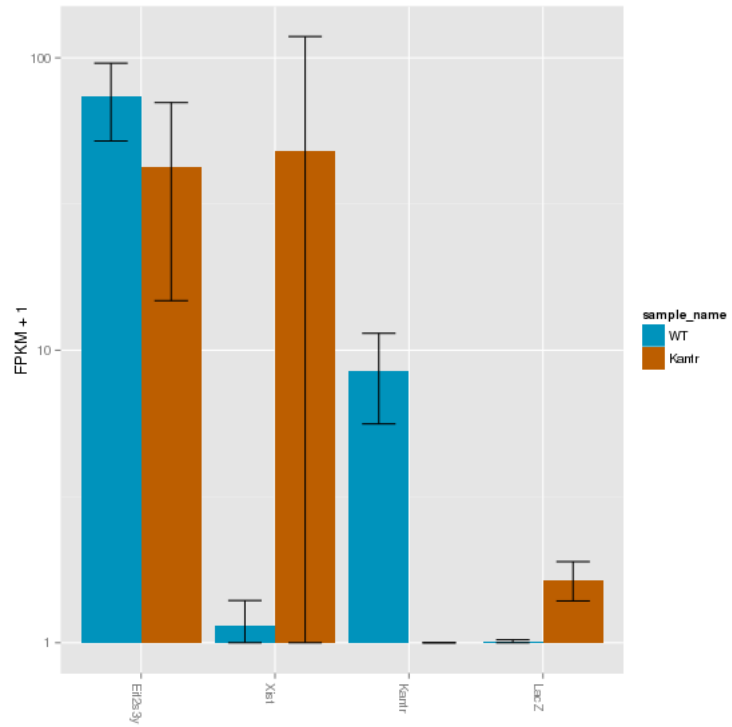


LacZ expression

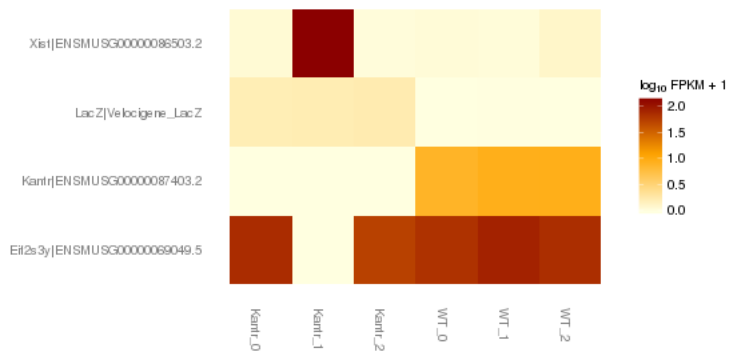


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



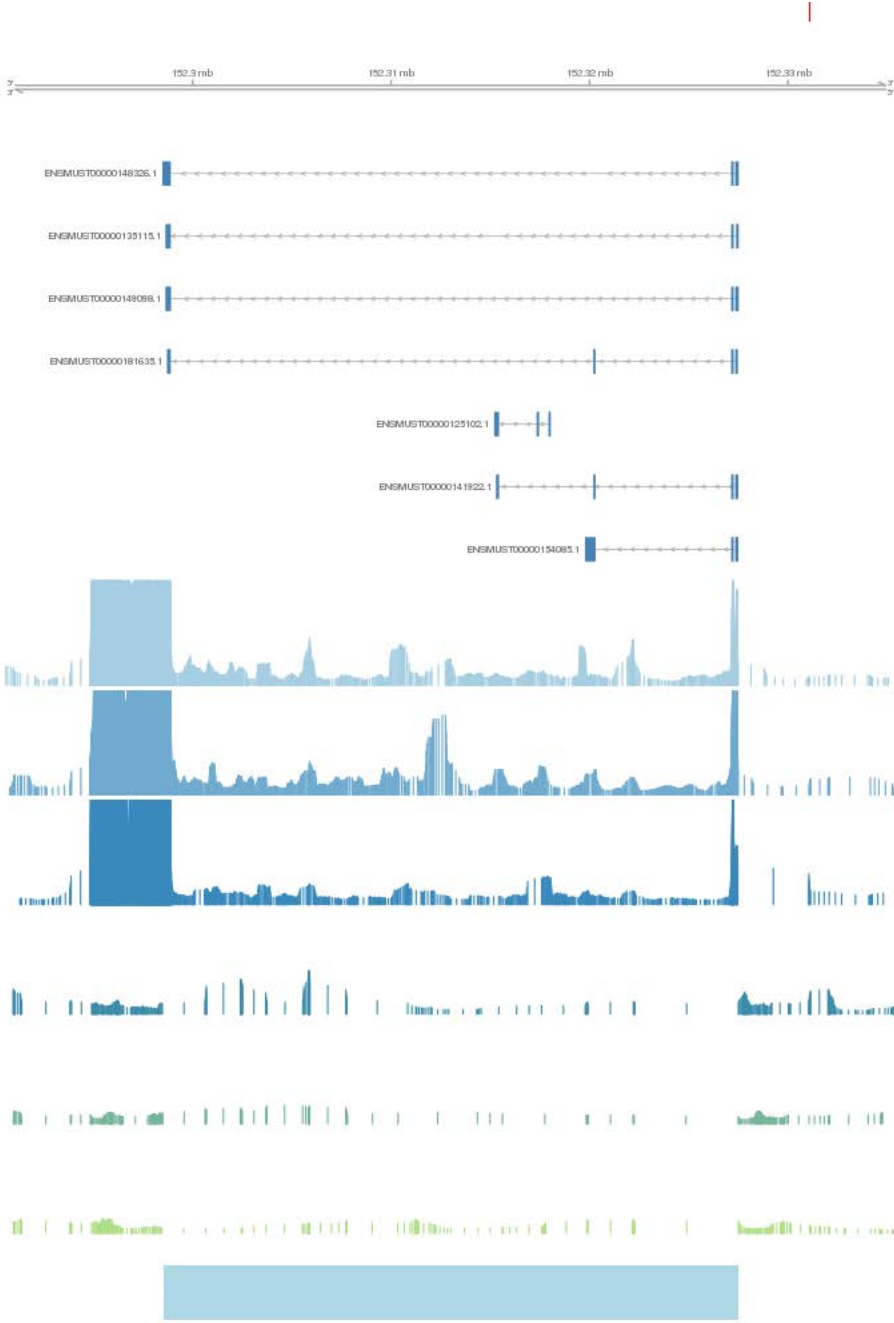
Expression heatmap:



Track visualization



KO Region Kantr_2 Kantr_1 Kantr_0 WT_2 WT_1 WT_0 LincRNA isoforms



Differential Analysis

Differential Genes

There are 766 significantly differentially expressed genes. They are:

	geneAnnot\$gene_short_name
1	H19
2	Tbx2
3	Ngfr
4	Gira1
5	Hddc2
6	Cdh1
7	Mcts1
8	Itga5
9	Gabra2
10	Col6a1
11	Snmp27
12	Gabrg1
13	Col18a1
14	Col1a1
15	Eef1e1
16	Sept7
17	Use1
18	Hltf
19	Zfp40
20	Shh
21	Lamb1
22	Fcgrt
23	Bicd1
24	Calb2
25	Zfp81
26	Ptrf
27	Pax2
28	Sst
29	Col26a1
30	Aqp1
31	Tmem242
32	Wisp1
33	Polr2a
34	Corin
35	Nid1
36	Evx1
37	Tep1

38 Fbln1
39 Nphs1
40 Acot13
41 Gabrb2
42 Khsrp
43 Dio2

44 0610009B22Rik
45 Zmiz1
46 Arid1a
47 Rplp1
48 Snrpb2
49 Bmp7
50 Sdhd
51 Slc38a3
52 Mrpl52
53 Ebf3
54 Gabra1
55 Scarf2
56 Rpa3
57 Phox2b
58 Rps5
59 Slc41a1
60 Aldh1a2
61 Tctex1d2
62 Cacybp
63 Ndufa2
64 Cox6c
65 Srp19
66 Hoxa2
67 Gata2
68 Ptgds
69 Psma2
70 Mrpl32

71 Capza2
72 Lcorl
73 Lbp
74 Atp5e
75 Ndufa1
76 Plgrkt
77 Mrps18c
78 Sulf1
79 Etaa1
80 Vtn

81 Erbb3
82 Trove2
83 Anxa6
84 Gira2
85 Slc35g3
86 Slc16a10
87 Lama4
88 Fabp7
89 Nts
90 Lama2
91 Actr6
92 Kittl
93 Enpp3
94 Meis1
95 Grb10
96 Snrpd3
97 Phlda1

98 Col6a2

99 Hint1
100 Hmnr
101 Fit4
102 Sar1b
103 Nefh
104 Gabrg2
105 Rps27a
106 Ccdc104
107 Aebp1
108 Smc6
109 Bcap29
110 Txndc17
111 Ntn1
112 Kihl28
113 Fkbp3
114 Scfd1
115 Nemf
116 Psmc6
117 Mpp5
118 Eif2s1
119 Smoc1
120 2010107E04Rik
121 Tfap2a
122 Dek
123 Nol8

124 Ror2
125 A530054K11Rik
126 Cetn3
127 Irx4
128 Slc6a3
129 Serf1
130 Otp
131 Trim23
132 Cenpk
133 Ndufs4
134 Fam213a
135 Cdhr1
136 Psmc6
137 Chat
138 Zdihc20
139 Dnajc15
140 Sugt1
141 Bnip3l
142 Nefm
143 Uchl3
144 Dab2
145 Sub1
146 Npr3
147 Skor1
148 Oxr1
149 Angpt1
150 Eif3h
151 Matn2
152 Eif3e
153 Emc2

154 Kcnv1
155 Ndufb9
156 Sla
157 Lrrc6
158 Enpp2
159 Myh9
160 Ndufa6
161 Plec
162 Ube2v2
163 B3gnt5
164 Sidt1
165 Ncam2
166 Arhgap31

167 Sncg
168 Ndufa5
169 Scg5
170 Prph
171 Igf2r
172 Thbs2
173 Zfp51
174 Pla2g7
175 Slc5a7
176 Arhgap28
177 Cyp1b1
178 Ndufv2
179 Epas1
180 Cript
181 Uqcc2
182 Cox7a2l
183 Slc25a46
184 Rock1
185 Esco1
186 Rnf138
187 Pgg1b
188 Yip5
189 Sec11c
190 C330018D20Rik
191 Fbn2
192 Rps14
193 Tmx3
194 Pdgfrb
195 Slc22a6
196 Aldh1a7
197 Kif20b
198 Capn1
199 Slit1
200 Kazald1
201 Tlx1
202 Lbx1
203 Alas2
204 Rps24
205 Itgb8
206 Rps26
207 Esyt1
208 Zfp711
209 Gaa

210 Tmem47
211 Gdap1
212 Itih5
213 Rpl14
214 Xiap
215 Kbtbd3
216 Cwf19l2
217 Mybl1
218 Mob4
219 Nop58
220 Sumo1
221 Ndufb3
222 Sgol2
223 Col5a2
224 Col3a1
225 Tex30
226 Pdcl3
227 Mrpl30
228 Ormdl1
229 1110058L19Rik
230 Fn1
231 Nppc
232 Cfh
233 Tfcp2l1
234 Dbi
235 Syt2
236 Lamc1
237 Kif26b
238 Dusp27
239 Ptpn14
240 Vamp4
241 Mrc1
242 Rabgap1l
243 Yme1l1
244 Acbd5
245 Ermn
246 Lamc3
247 Gsn
248 Pax8
249 Kif18a
250 Lin7c
251 B230118H07Rik
252 Eif3m

253 Fbn1
254 Cops2
255 Creb3l1
256 Map1a
257 Lamp5
258 Nusap1
259 Casc5

260 Otor
261 Pkia
262 Snx16
263 Lrrcc1

264 Psma7
265 Sec62
266 Dcun1d1
267 Hmgcs2
268 Olfm3
269 Sfrp2
270 Enpep
271 Aimp1
272 Rps3a1
273 Depdc1a
274 Rps20
275 Casp8ap2
276 Smc2
277 Svep1
278 Zfp37
279 Bspry
280 Nudt2
281 Rps6
282 Ift74
283 Magoh
284 Ccdc23
285 Cmpk1
286 Rpl22
287 Tomm7
288 Orc5
289 Slc30a3
290 Emilin1
291 Gabra4
292 Pdgfra
293 Epha5
294 Tgfb3
295 Sparcl1

296 Alb
297 Ndufa4
298 Col1a2
299 Nt5c3
300 Slc13a4
301 Zyx
302 Mrps33
303 Zfml
304 Kbtbd8
305 Plxna1
306 Slc6a13
307 Ret
308 A2m
309 Dera
310 Slc6a11
311 Zfp606
312 Gipr
313 Zfp273

314 Dbx1
315 Mphosph10
316 Ccdc90b
317 Tmem126a
318 Zfand6

319 Ndufc2
320 Pik3c2a
321 Rps3
322 Bag3
323 Ndufb11
324 Mrgprf
325 Vbp1
326 Rab39b
327 Cox7b
328 Hmgn5
329 Sh3bgrl
330 Col4a6
331 Col4a5
332 Cetn2
333 Ap1s2
334 Bgn
335 Gpr124
336 Col4a1
337 Col4a2
338 Polb

339 Slit2
340 Pcm1
341 Scrg1
342 Irx3
343 Irx5
344 Mmp2
345 Mt3
346 Cdh5
347 Hsd11b2
348 Ankrd49
349 Cfdp1
350 Tbx20
351 Pts
352 Mpzl2
353 Robo4
354 Robo3
355 Kank2
356 Dock6
357 Itga11
358 Phip
359 Crabp1
360 Isl2
361 Stra6
362 Tmem30a
363 Cox7a2
364 Snapc5
365 Tgfbr2
366 Rpsa
367 Mst1r
368 Gbbp1
369 Lama1
370 Sox10
371 Mzt1
372 Wnt6
373 Antxr1

374 Lgj3
375 Adra2a
376 Tlr13
377 9430020K01Rik
378 Zbtb41
379 Zfp944
380 Gria2
381 Lsm3

382 Smc4
383 Gbx2
384 Hdx
385 Ankrd12
386 Lrn1
387 Ppp1r1c
388 Pcdh11x
389 Plvap
390 Pam1
391 Arhgap5
392 Nkx6-1
393 Lars2
394 Trim13
395 Ssc5d
396 Lysmd3
397 Fnip1
398 Colec12
399 Pnpla8
400 Rpl39-ps
401 Lum
402 March1
403 Rps27l
404 Neto2
405 Bcor1
406 Anapc10
407 Tenc1
408 Sep15
409 Islr
410 Matr3
411 0610009D07Rik
412 Thoc2
413 Cldn11
414 Cdkn3
415 Avp
416 Neurod6
417 2700029M09Rik
418 Cntln
419 Notch3
420 Spon1
421 Fam126b
422 Fau
423 Foxf2
424 Akap12

425 Gng10
426 Med30
427 Creb3l2
428 Atp5j2

429 Lmx1b
430 Rps21
431 Timm8b
432 Anpep
433 Zfp503
434 Rpl22l1
435 Pcsk1n
436 Raly1
437 Slc6a5
438 Gjb6
439 Nxph4
440 Zfp770
441 Cdh12
442 Tlx3
443 St8sia4
444 Snmp25
445 Slc22a2
446 Npnt
447 Trim62
448 Ostc
449 Nbl1
450 Arhgap11a
451 Car8
452 Tmem29
453 Piezo2
454 Fmod
455 Gucy1a2
456 Fcho2
457 Prxl1
458 Slc24a4
459 Abca9
460 Tbca
461 Zfp386
462 Bend6
463 Nxt2
464 Rbm7
465 Dctpp1
466 Klhl14
467 Shfm1

468 Sh2b3
469 Tfap2d
470 Itpr3
471 Imp11
472 Selk
473 Wbp5
474 B630005N14Rik
475 Hebp1
476 Tuba1c
477 Hic1
478 Aqp6
479 Smco3
480 Rab9b
481 Tril
482 Rpl7
483 Pura

484 Sptssa
485 Gm9493
486 Rin3
487 Zfp758
488 Trmt10c
489 Rpl18a
490 Dpy19l4
491 Cenpe
492 Defb11
493 Gprin3
494 Onecut3
495 Penk
496 Olig3
497 Esf1
498 Zfp36l2
499 Polr2k
500 Zfp62
501 Gjb2
502 C77370
503 Olfml2a
504 Ppp1r3b
505 Ccdc108
506 Zfp654
507 Dynlrb1
508 Adamts12
509 Mafa
510 Rps8

511 Rpsa-ps10
512 Zcchc5
513 Tmem196
514 Pirt
515 Crebrf
516 Sacs
517 Zfp738
518 Osr1
519 Ranbp3l
520 Igf2
521 Myof
522 C1galt1c1
523 Mterfd3
524 Gm8186
525 Plcx3
526 Zfp518a
527 Tceal1
528 Adamts16
529 2410015M20Rik

530 Tmsb4x
531 Shisa3
532 Foxc1
533 Gm9843
534 Foxl2
535 Gm9846
536 Atp5k
537 Zfp366
538 Gja1

539 Gprc5c
540 Rgs13
541 Sv2c
542 Commd3
543 Nhlh1
544 Gm6563
545 Tceal8
546 Coa6
547 Gypa
548 Xkr4
549 Pfdn4
550 Rps10
551 Gpr17
552 Hbb-bs
553 Lamb2

554 C130021120Rik

555 Aldh1a1

556 Sec61b

557 Zfp943

558 Thoc7

559 Atf7ip

560 Spock3

561 Atpif1

562 Adam12

563 Mmm1

564 Hmgb2

565 Zfp182

566 Fsd1l

567 Zfp935

568 Zfp101

569 Tns1

570 Fut9

571 Maf

572 Zfp458

573 Gpc3

574 Zfp277

575 Triqk

576 Al504432

577 Col8a2

578 Uts2b

579 Cd248

580 Foxk1

581 Mpz

582 Kdm5d

583 Foxb2

584 Gadl1

585 Gm7536

586 Npm1

587 Zfp759

588 Mab2112

589 Zfp119a

590 Rpl32

591 AA987161

592 Gm5148

593 Zfp68

594 Zfp85-rs1

595 Gpc6

596 Zfp958

597 Col13a1
598 Zfp708
599 Gm10053
600 Zfp945
601 Rpl11
602 Zfp933
603 Zfp930
604 Unc5c
605 Gm10073
606 Rp2h
607 Tpt1
608 Gm10076
609 BC002059
610 Scrt2
611 2610008E11Rik
612 linc-Bm1a
613 Rps10-ps1
614 Tsga10
615 Rpl26
616 Zfp873
617 Rps7
618 Zic2
619 Zfp229
620 Tac1
621 Rps12
622 Rpl34
623 Rpl9-ps6
624 Rps3a2
625 Zfp677
626 Zfp938
627 Gm10123
628 Kdr
629 Mrpl42
630 Sox7
631 3300002I08Rik
632 Zfp947
633 Cyp26b1
634 Rps15
635 Col23a1
636 Chchd1
637 Uqcrh
638 Fbln2
639 Gm10146

640 mt-Nd1
641 mt-Nd2
642 mt-Co1
643 mt-Co2
644 mt-Co3
645 mt-Nd4
646 Rpl36-ps3
647 Cntnap5b
648 Foxd3

649 Tspan6
650 Zfp763
651 Gm10221
652 Zfp760
653 Zfp948
654 Zfp160
655 Zfp442
656 Zfp120
657 Ndufaf2
658 Col8a1
659 Gm11808
660 Slc4a5
661 Efcab1
662 Flnc
663 Gm10250
664 Rap1a
665 Ssb
666 Ddx3y
667 Gm10260
668 Nxph2
669 Zfp72
670 Gm10275
671 Gm5595
672 Zfp820
673 Hba-a2
674 Hba-a1
675 Gm10288
676 Tmem256
677 Smim15
678 Zfp946
679 Zfp942
680 Zfp71-rs1
681 Rpl23
682 Phf2011

683 Gm13826
684 4930522L14Rik
685 Vma21
686 Trim10
687 Dok6
688 Myeov2
689 Nbeal1
690 Hbb-bt
691 9830147E19Rik
692 Zfp788
693 Mafb
694 Gm10742
695 Thbd
696 Gm561
697 Zfp808
698 Ctxn2
699 Accs1
700 Hoxc4
701 Nynrin
702 Selt
703 Rpl23a-ps3

704 Gm2000
705 Gm3940
706 Foxd1
707 Samp
708 AU041133
709 Abracl
710 2210404O09Rik
711 Zfp931
712 Gm14326
713 Gm14322
714 Gm14418
715 Gm14305
716 Gm14391
717 D130040H23Rik
718 Jrkl
719 Ccdc82
720 Haus3
721 Hoxa3
722 Rpl39
723 Tceb1
724 Zfp703
725 Gm12688

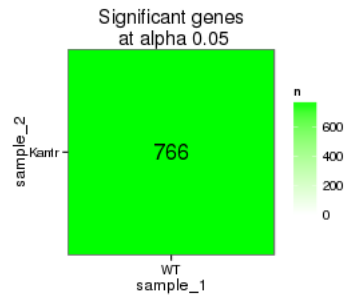
726 Xist
727 Gm15500
728 Kantr
729 Gm15501
730 Tmsb15b2
731 Dlx6os1
732 Gm10177
733 Pcp4
734 2810055G20Rik
735 Snrpe
736 Gm5611
737 Zfp712
738 Myl6
739 Lrrc32
740 Gm5141
741 Gm10269
742 Skor2
743 Tma7
744 Lsm5
745 Cox16

746 Gm4540
747 Zfp804b
748 Zfp141
749 Rpl41
750 Zfp955a
751 Gm14403
752 Rps2-ps6
753 Figl2
754 Gm6472
755 Gm7094
756 Gm4944
757 Zfp955b
758 Foxl2os

759 A330076H08Rik
760 Gm26917
761 Gm17750
762 Gm26924
763 A430106G13Rik
764 Gm27031
765 Snhg6
766 RP23-442I7.1

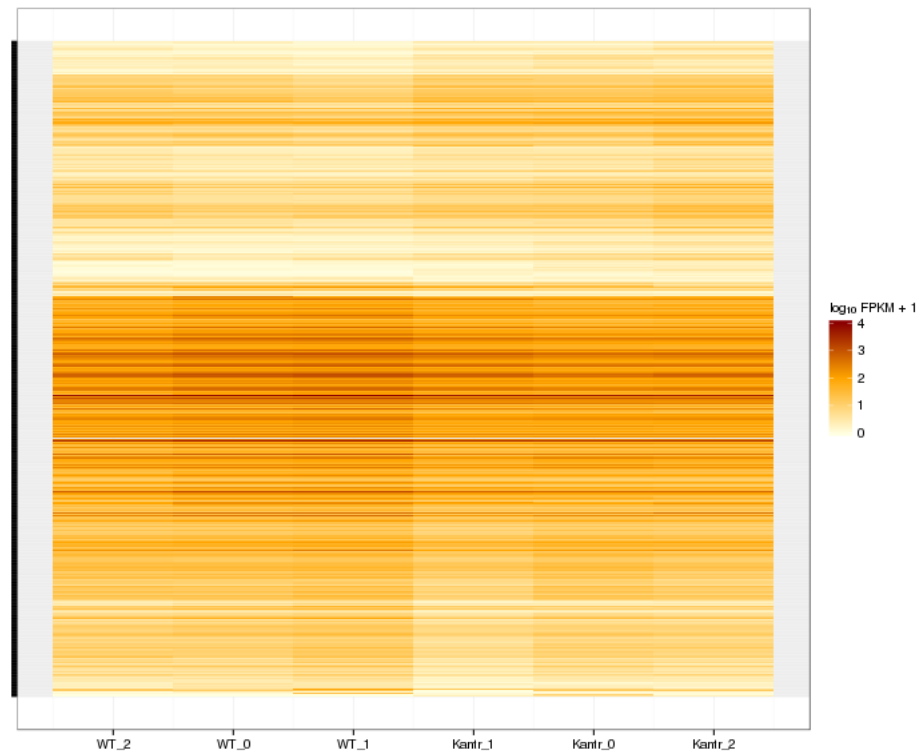
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

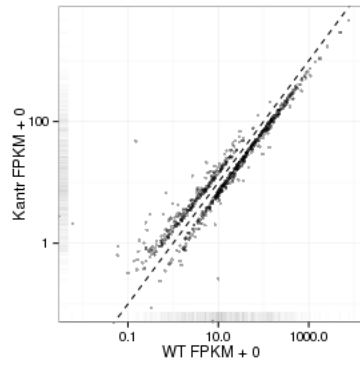


Significant genes with expression >50fpm (any condition):(turned off)

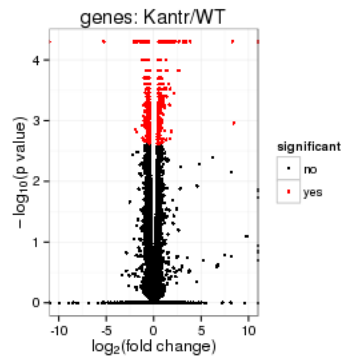
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

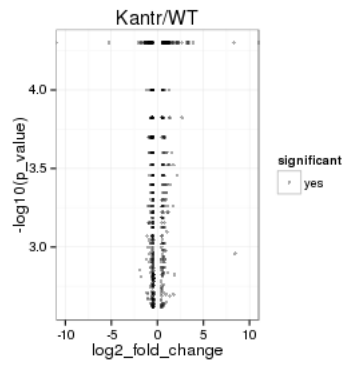
Scatter plot of significant genes only:



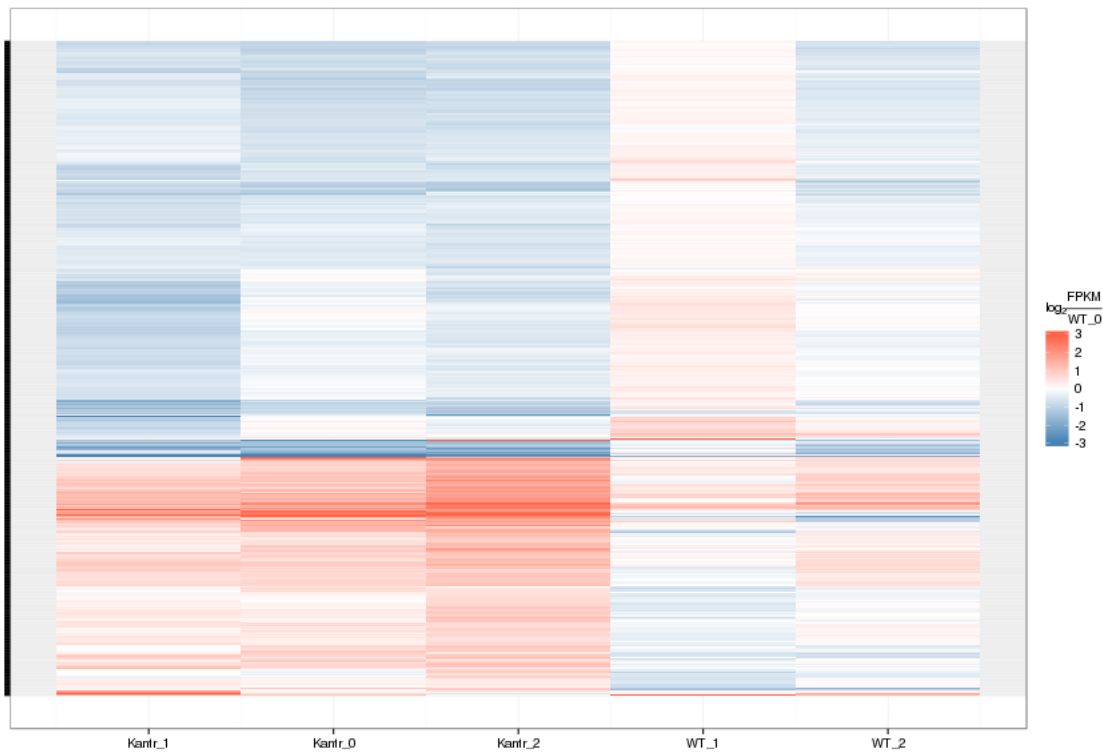
Volcano Plot



Volcano plot with significant genes only:



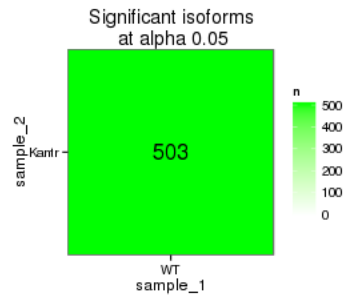
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

	isoAnnot	gene_short_name
1	H19	
2	Tbx2	
3	Ngfr	
4	Hddc2	
5	Cdh1	
6	Mcts1	
7	Gabra2	
8	C1d	
9	Serpinf1	
10	Col6a1	
11	Gabrg1	
12	Col18a1	
13	Col1a1	
14	Eef1e1	
15	Use1	
16	Zfp40	
17	Lamb1	
18	Dvl3	
19	Fcgrt	
20	Bicd1	
21	Zfp81	
22	Ptrf	
23	Pax2	
24	Hif3a	
25	Sst	
26	Aqp1	

27 Tmem242
28 Wisp1
29 Corin
30 Nid1
31 Kif5b
32 Gabrb2
33 Dio2
34 0610009B22Rik
35 Rplp1
36 Snrpb2

37 Bmp7
38 Gabra1
39 Scarf2
40 Phox2b
41 Rps5
42 Cacybp
43 Cox6c
44 Hoxa2
45 Gata2
46 Ptgds
47 Psm2
48 Mrpl32
49 Capza2
50 Atp5e
51 Ndufa1
52 Mrps18c
53 Vtn
54 Erbb3
55 Trove2
56 Glra2
57 Slc35g3
58 Slc16a10
59 Lama4
60 Fabp7
61 Nts
62 Lama2
63 Actr6

64 Kitl

65 Moxd1
66 Meis1
67 Col6a2
68 Hmnr

69 Flt4
70 Sar1b
71 Nefh
72 Rps27a
73 Ccdc104
74 Aebp1
75 Smc6
76 Fam20a
77 Bcap29
78 Mrc2
79 Ntn1
80 Fkbp3
81 Scfd1
82 Mpp5
83 Eif2s1
84 Hsp90aa1
85 2010107E04Rik
86 Dek
87 Nol8
88 A530054K11Rik
89 Cetn3
90 Ankrd32
91 Irx4
92 Otp
93 Trim23
94 Ndufs4
95 Cdhr1
96 Psmc6
97 Chat
98 Zdhhc20
99 Dnajc15
100 Sugt1
101 Nefm
102 Uchl3
103 Dab2
104 Npr3
105 Oxr1
106 Angpt1
107 Eif3h
108 Eif3e
109 Emc2
110 Kcnv1
111 Col14a1
112 Enpp2

113 Myh9
114 Ndufa6
115 Ube2v2
116 B3gnt5
117 Zbtb20

118 Fgd4
119 Arhgap31

120 Sncg
121 Prph
122 Thbs2
123 Zfp51
124 Pla2g7
125 Slc5a7
126 Arhgap28
127 Cyp1b1
128 Ndufv2
129 Epas1
130 Uqcc2
131 Esco1
132 Fbn2
133 Rps14
134 Tmx3
135 Pdgfrb
136 Slc22a6
137 Kif20b
138 Lbx1
139 Rps24
140 Rps26
141 Esyt1
142 Zfp711
143 Tmem47
144 Itih5
145 Stag2
146 Cwf19l2
147 Lypla1
148 Rb1cc1
149 Mob4
150 Nop58
151 Sumo1
152 Ndufb3
153 Col5a2
154 Col3a1
155 Mrpl30

156 1110058L19Rik
157 Fn1
158 Nppc
159 Dbi
160 Syt2
161 Lamc1
162 Cenpf
163 Mrc1
164 Itga8
165 Col5a1
166 Lamc3
167 Pax8
168 Kif18a
169 Eif3m
170 Fbn1
171 Cops2
172 Lamp5
173 Otor
174 Pkia

175 Dcun1d1
176 Pdcc10
177 Sfrp2
178 Enpep
179 Aimp1
180 Rps3a1
181 Abca4
182 Tmem68
183 Casp8ap2
184 Svep1
185 Zfp37
186 Bspry
187 Rps6
188 Ifit4
189 Magoh
190 Col9a2
191 Tomm7
192 Orc5
193 Mxra8
194 Slc30a3
195 Tgfb3
196 Ndufa4
197 Col1a2
198 Slc13a4

199 Zyx
200 Slc6a13
201 Ret
202 A2m
203 Dera
204 Dbx1
205 Mphosph10
206 Ccdc90b
207 Zfand6
208 Ndufc2
209 Ndufb11
210 Zfp300
211 Vbp1
212 Atrx
213 Cox7b
214 Hmgn5
215 Sh3bgr1
216 Col4a6
217 Bgn
218 Col4a1
219 Pcm1
220 Mmp2
221 Cdh5
222 Tbx20
223 Mpzl2
224 Robo3
225 Kank2

226 Dock6
227 Rora
228 Itga11
229 Phip

230 Crabp1
231 Tmem30a
232 Cox7a2
233 Tgfb2
234 Rpsa
235 Ccdc88a
236 Lama1
237 Sox10
238 Wnt6
239 Lgi3
240 Zfp944
241 Gria2

242 Lsm3
243 Smc4
244 Gbx2
245 Pcdh11x
246 Pam1
247 Wnk4
248 Arhgap5
249 1110059E24Rik
250 Nkx6-1
251 Lars2
252 Trim13
253 Scai
254 G2e3
255 Lysmd3
256 Rpl39-ps
257 Rps27l
258 Neto2
259 Anapc10
260 Islr
261 Matr3
262 Itih2
263 0610009D07Rik
264 Thoc2
265 Cldn11
266 Neurod6
267 Spon1
268 Fam126b
269 Foxf2
270 Akap12
271 Atp5j2
272 Lmx1b
273 Rps21
274 Timm8b
275 Zfp503
276 Rpl22l1
277 Slc6a5
278 Gjb6
279 Nxp4
280 Cdh12
281 Tlx3
282 St8sia4
283 Slc22a2
284 Phf8

285 Car8
286 Fmod
287 Gucy1a2
288 Prrx1
289 Abca9
290 Tbca
291 Zfp386
292 Bend6
293 Ehbp1
294 Dctpp1
295 Shfm1
296 Itpr3
297 Imp1
298 Selk
299 Wbp5
300 B630005N14Rik
301 Hebp1
302 Egflam
303 Tuba1c
304 Hic1
305 Aqp6
306 Tril
307 Rpl7
308 Sptssa
309 Gm9493
310 Trmt10c
311 Cenpe
312 Onecut3
313 Penk
314 Olig3
315 Esf1
316 Zfp322a
317 Gjb2
318 Olfml2a
319 Tefm
320 Zfp654
321 Dynlrb1
322 Adamts12
323 Mafa
324 Rps8
325 Rpsa-ps10
326 Zcchc5
327 Sned1
328 Tmem196

329 Pirt
330 Col6a3
331 Zfp738
332 Osr1
333 Ranbp3l

334 Igf2
335 Plcx3
336 Zfp518a
337 Tmsb4x
338 Shisa3
339 Foxc1

340 Gm9843
341 Gm9846
342 Vstm4
343 Atp5k
344 Gja1
345 Sv2c
346 Commd3
347 Nhlh1
348 Gm6563
349 Six1
350 Tceal8
351 Coa6
352 Gypa
353 Pfdn4
354 Il1rap1
355 C130021120Rik
356 Aldh1a1
357 Sec61b
358 Zfp943
359 Thoc7
360 Atf7ip
361 Atpif1
362 Adam12
363 Mmm1
364 Kihl18
365 Tns1
366 Fut9
367 Gpc3
368 Triqk
369 Al504432
370 Col8a2
371 Uts2b

372 Slit3
373 Cd248
374 Mpz
375 Kdm5d
376 Foxb2
377 Gad1
378 Gm7536
379 Zfp759
380 Mab21l2
381 Rpl32
382 AA987161
383 Gda
384 Zfp958
385 Zfp708
386 Gm10053
387 Zfp933
388 Zfp930
389 Gm10073
390 Rp2h
391 Sct2
392 2610008E11Rik
393 Zfp868
394 Rps10-ps1

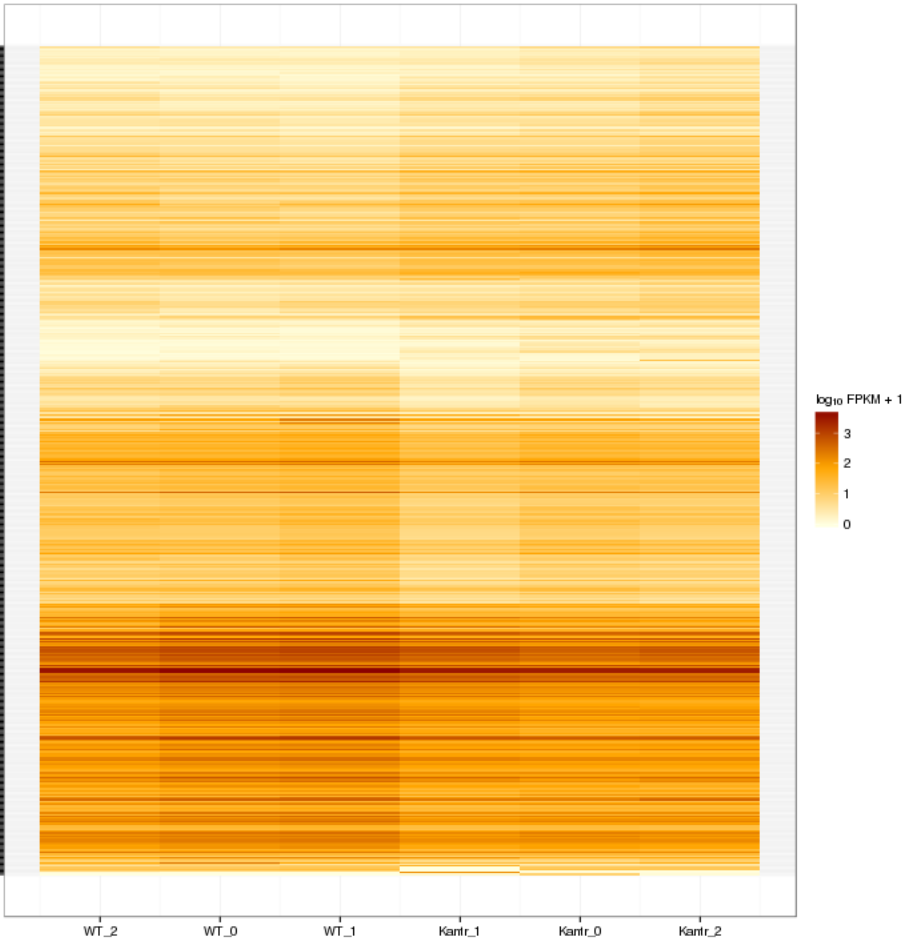
395 Rpl26
396 Zfp873
397 Rps7
398 Zic2
399 Rps12
400 Rpl34
401 Rpl9-ps6
402 Rps3a2
403 Zfp938
404 Kdr
405 Mrpl42
406 Cyp26b1
407 Rps15
408 Col23a1
409 Chchd1
410 Uqcrh
411 Fbln2
412 Gm10146
413 mt-Nd1
414 mt-Nd2

415 mt-Nd4
416 Foxd3
417 Zfp763
418 Gm10221
419 Zfp760
420 Zfp948
421 Zfp160
422 Ndufaf2
423 Col8a1
424 Gm11808
425 Slc4a5
426 Efcab1
427 Rap1a
428 Ssb
429 Ddx3y
430 Gm10260
431 Nxph2
432 Zfp72
433 Gm10275
434 Gm10288
435 Smim15
436 Zfp946
437 Zfp942
438 Zfp71-rs1
439 Rpl23
440 Gm13826
441 Dok6
442 Myeov2
443 Nbeal1
444 Hbb-bt
445 Rpl13a
446 9830147E19Rik
447 Ehd2
448 Mafb
449 Gm10742

450 Thbd
451 Gm561
452 Zfp808
453 Scn2a1
454 Hoxc4
455 Rpl23a-ps3
456 Gm2000
457 Gm3940

458 Foxd1
459 AU041133
460 Abracl
461 2210404O09Rik
462 Zfp931
463 Gm14326
464 Gm14418
465 Jrkl
466 Haus3
467 Rpl39
468 Tceb1
469 Zfp703
470 Gm12688
471 Xist
472 Gm15500
473 Kantr
474 Tmsb15b2
475 3110047P20Rik
476 Gm10177
477 Pcp4
478 2810055G20Rik
479 Snrpe
480 Zfp712
481 Myl6
482 Lrrc32
483 Gm5141
484 Gm10269
485 Tma7
486 Gm4540
487 Zfp804b
488 Zfp141
489 Rpl41
490 Zfp955a
491 Gm14325
492 Zfp748
493 Gm6472
494 Gm4944
495 Foxl2os
496 Gm17750
497 Gm26924
498 A430106G13Rik
499 Gm27031
500 Snhg6

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

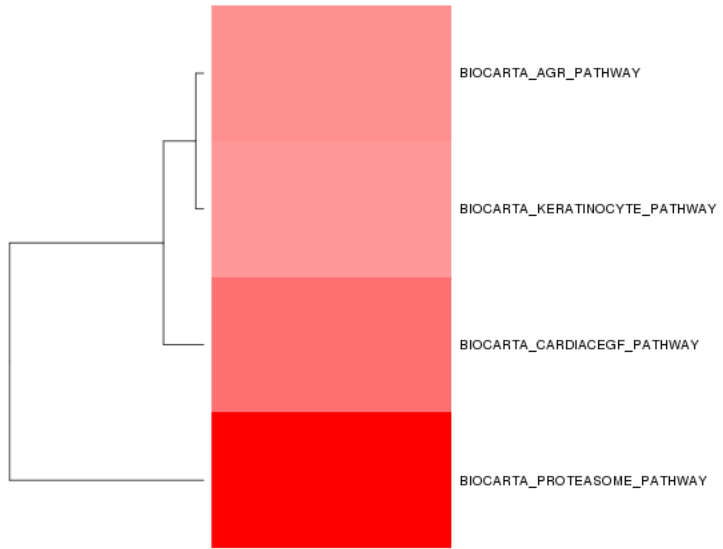
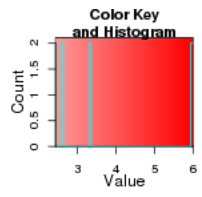
Gene/Pathway Analysis

GSEA

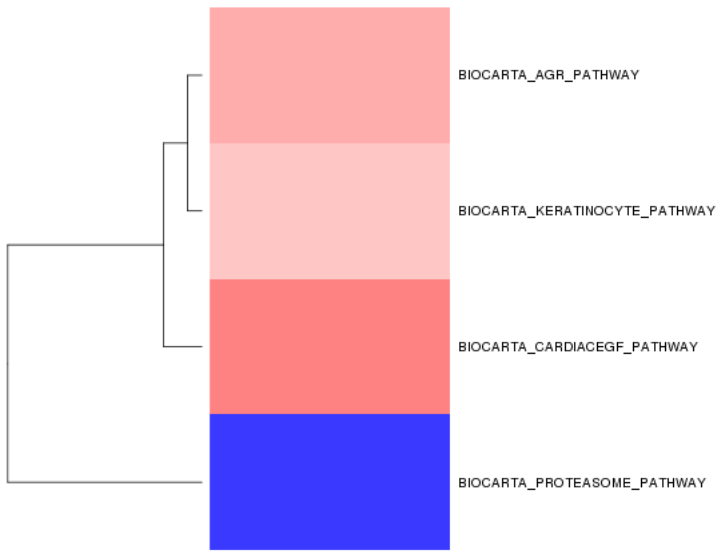
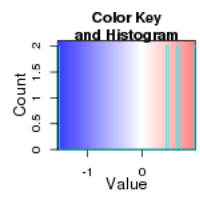
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

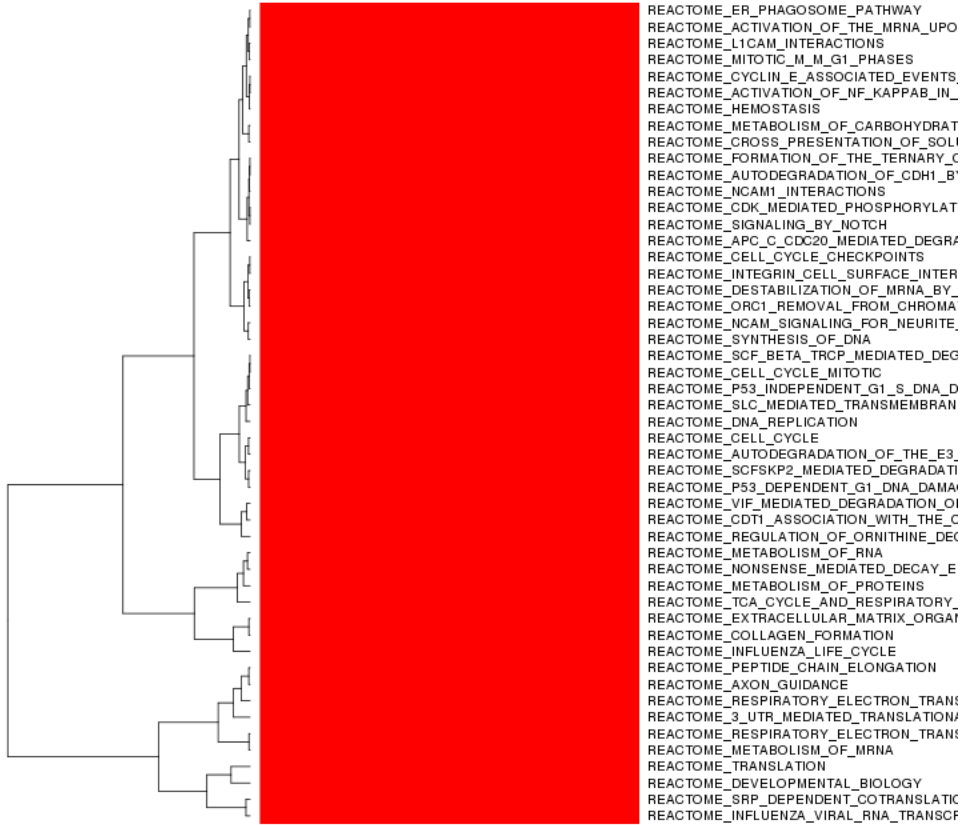
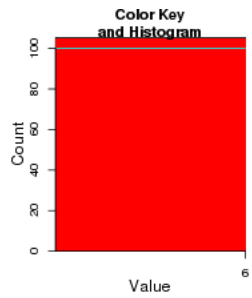
Biocarta enrichment:



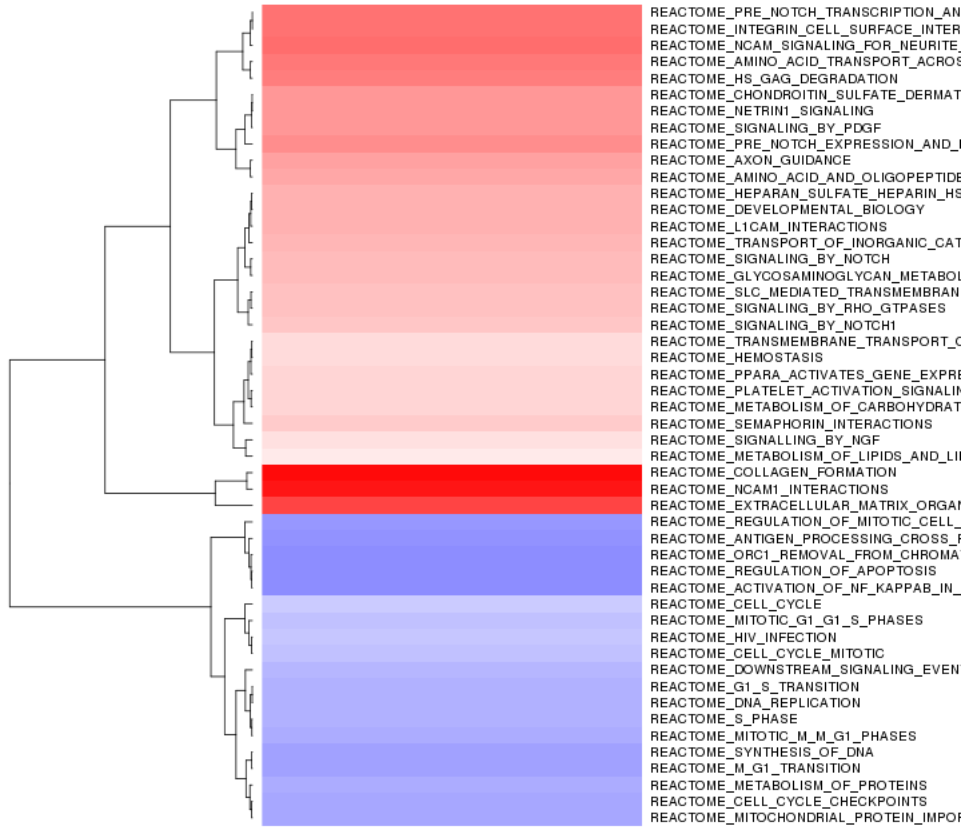
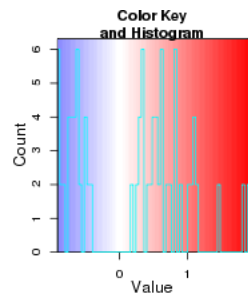
Biocarta zscore:



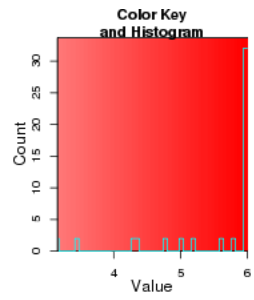
Reactome enrichment:

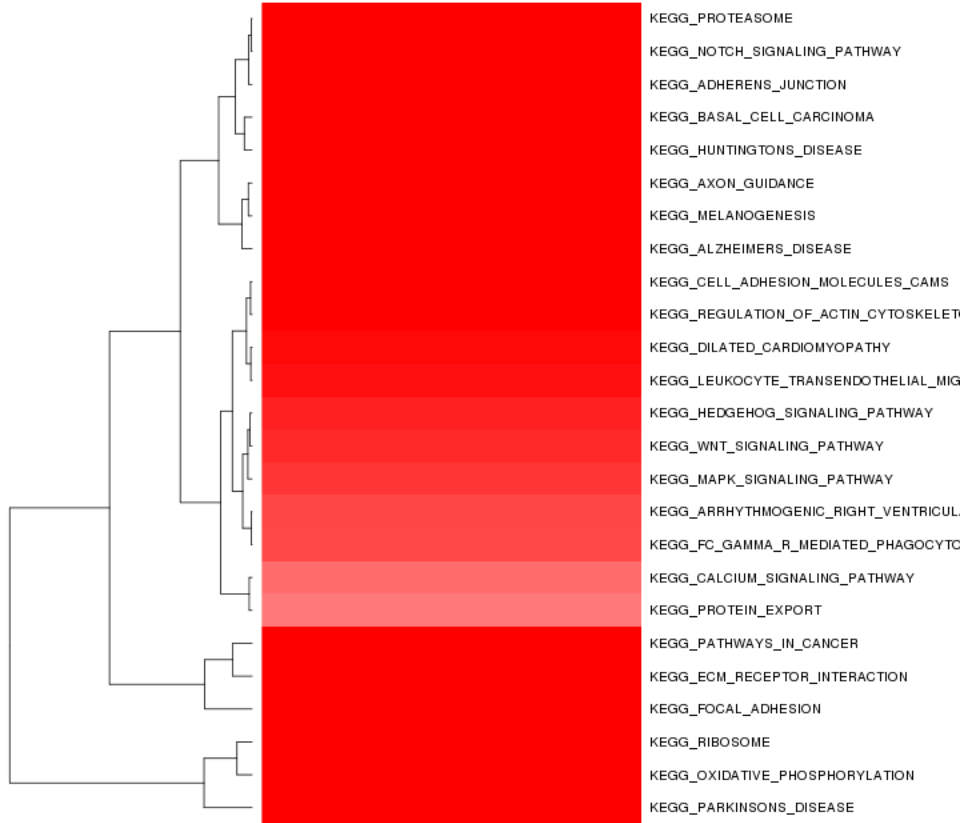


Reactome zscore:

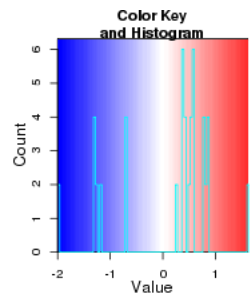


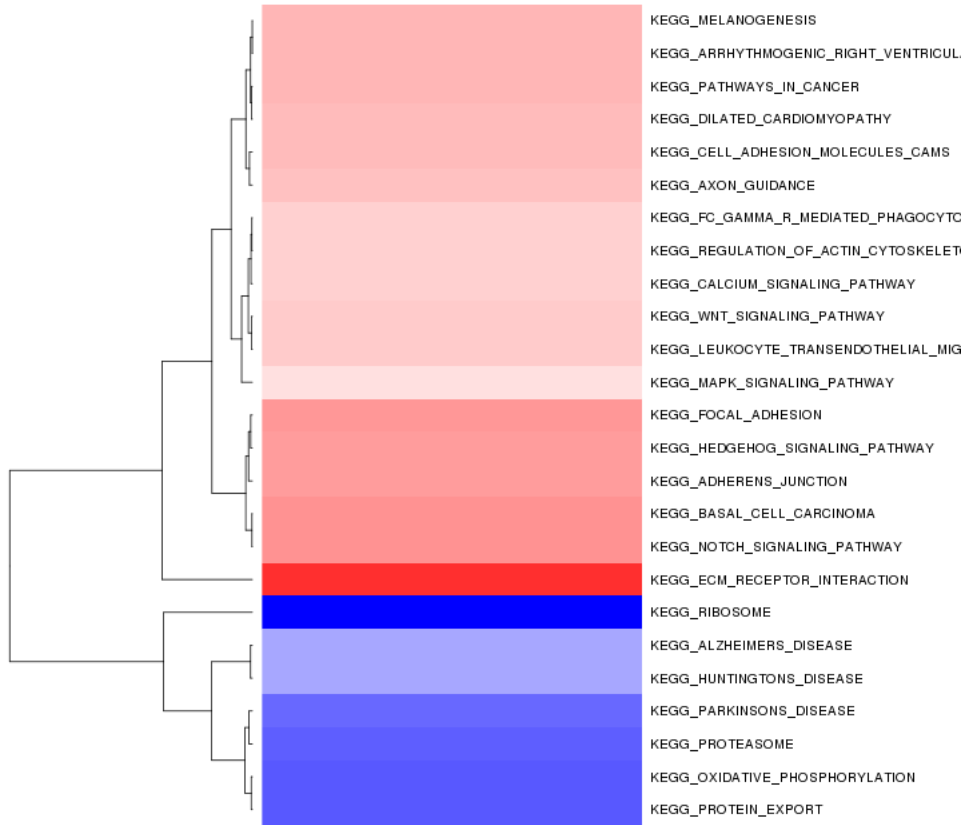
Kegg enrichment:





Kegg zscore:





Interneuron enrichment:

```
## Error: argument is of length zero
```

Interneuron zscore:

```
## Error: incorrect number of dimensions
```

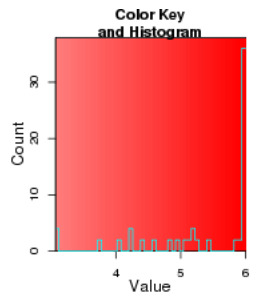
```
## Error: object 'x_ordered' not found
```

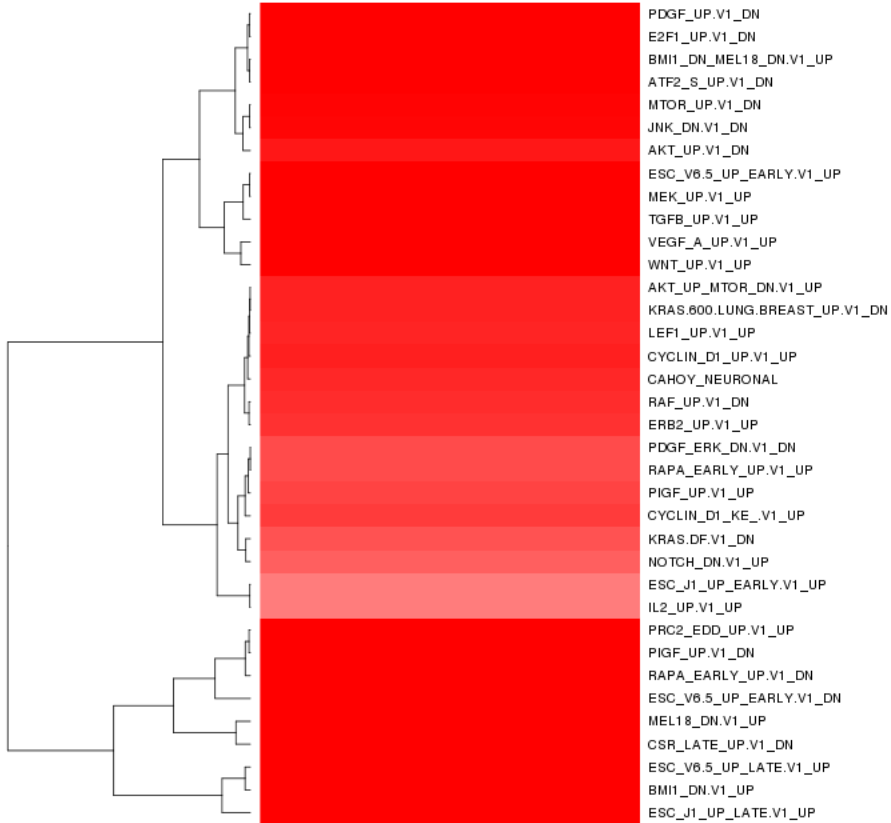
```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Oncogene enrichment:

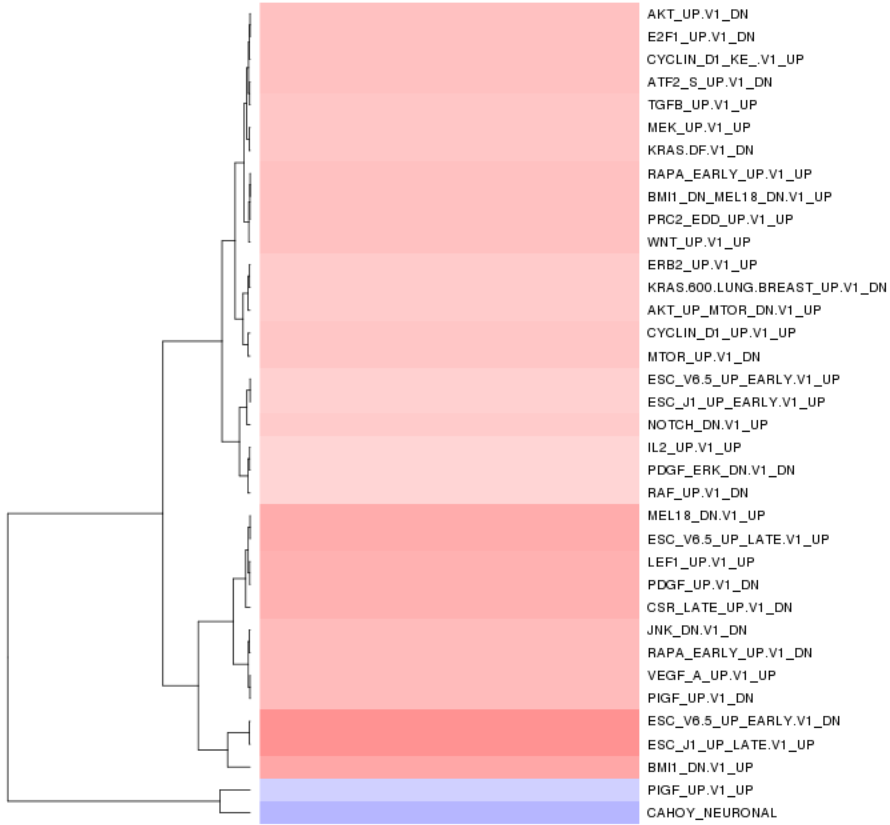
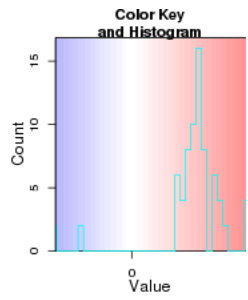
```
## Error: subscript out of bounds
```





Oncogene zscore:

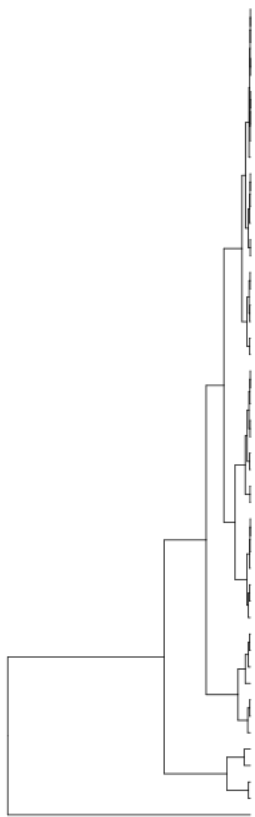
Error: subscript out of bounds



Immuno enrichment:

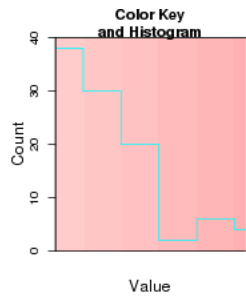
Error: no locations are finite

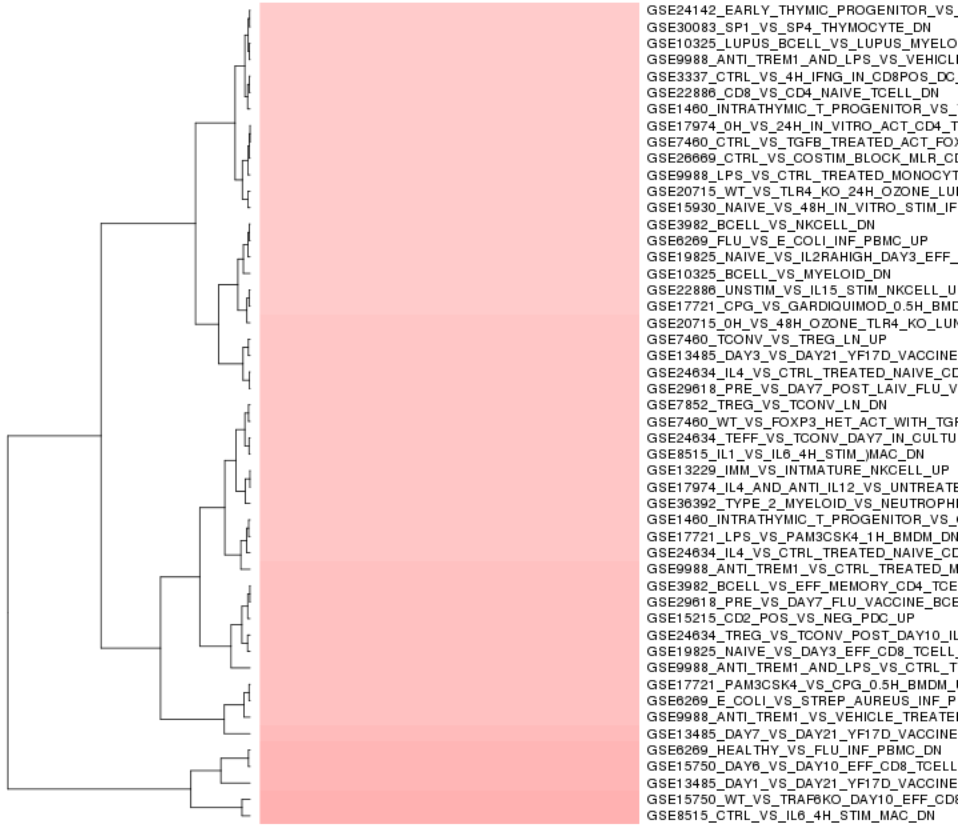




GSE11057_CD4_CENT_MEM_VS_PBMC_UP
GSE22886_CD8_VS_CD4_NAIVE_TCELL_UP
GSE22886_IL2_VS_IL15_STIM_NKCELL_DN
GSE15750_WT_TRAF6KO_DAY10_EFF_CD8
GSE17721_LPS_VS_PAM3CSK4_1H_BMDM_UP
GSE17974_CTRL_VS_ACT_IL4_AND_ANTI_IL12
GSE17721_CPG_VS_GARDIQUIMOD_0.5H_BMC
GSE24142_DN2_VS_DN3_THYMOCYTE_DN
GSE8515_IL1_VS_IL6_4H_STIM_MAC_DN
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IF
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_CI
GSE26669_CTRL_VS_COSTIM_BLOCK_MLR_CI
GSE15215_CD2_POS_VS_NEG_PDC_UP
GSE29618_BCELL_VS_MONOCYTE_DAY7_FLU
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IL
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IF
GSE24634_TREG_VS_TCONV_POST_DAYS_IL4
GSE13485_DAY3_VS_DAY21_YF17D_VACCINE
GSE22886_NAIVE_CD4_TCELL_VS_MEMORY_T
GSE9988_LOW_LPS_VS_CTRL_TREATED_MONO
GSE17721_CPG_VS_GARDIQUIMOD_4H_BMDM
GSE360_HIGH_DOSE_B_MALAYI_VS_M_TUBE1
GSE9988_ANTI_TREM1_VS_CTRL_TREATED_M
GSE36392_TYPE_2_MYELOID_VS_NEUTROPHI
GSE6269_E_COLL_VS_STREP_AUREUS_INF_P
GSE9988_LPS_VS_VEHICLE_TREATED_MONO
GSE9988_LPS_VS_CTRL_TREATED_MONOCYT
GSE6269_HEALTHY_VS_E_COLL_INF_PBMC_U
GSE3982_MAC_VS_NEUTROPHIL_LPS_STIM_U
GSE7400_CTRL_VS_CSF3_IN_VIVO_TREATED
GSE6269_FLU_VS_E_COLL_INF_PBMC_UP
GSE17721_PAM3CSK4_VS_CPG_0.5H_BMDM_I
GSE13485_DAY7_VS_DAY21_YF17D_VACCINE
GSE9988_LOW_LPS_VS_VEHICLE_TREATED_I
GSE7400_WT_VS_FOXP3_HET_ACT_WITH_TG1
GSE8515_CTRL_VS_IL6_4H_STIM_MAC_DN
GSE17721_LPS_VS_PAM3CSK4_1H_BMDM_DN
GSE22886_NAIVE_CD4_TCELL_VS_48H_ACT_T
GSE9988_ANTI_TREM1_VS_VEHICLE_TREAT
GSE6269_HEALTHY_VS_FLU_INF_PBMC_DN
GSE9988_ANTI_TREM1_AND_LPS_VS_VEHICLI
GSE14000_TRANSLATED_RNA_VS_MRNA_4H
GSE13485_DAY1_VS_DAY21_YF17D_VACCINE
GSE14000_TRANSLATED_RNA_VS_MRNA_16H
GSE9988_ANTI_TREM1_AND_LPS_VS_CTRL_T
GSE29617_CTRL_VS_DAY3_TIV_FLU_VACCINE
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_CI
GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL
GSE29617_CTRL_VS_TIV_FLU_VACCINE_PBMC
GSE13485_DAY1_VS_DAY21_YF17D_VACCINE

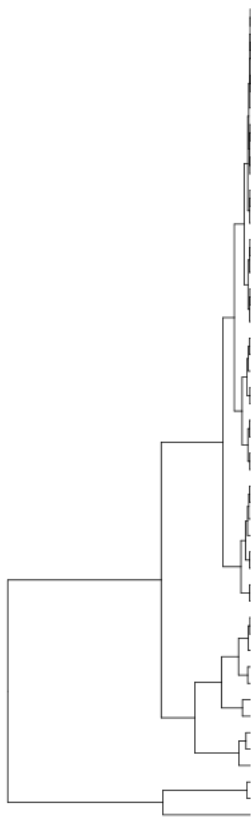
Immuno zscore:





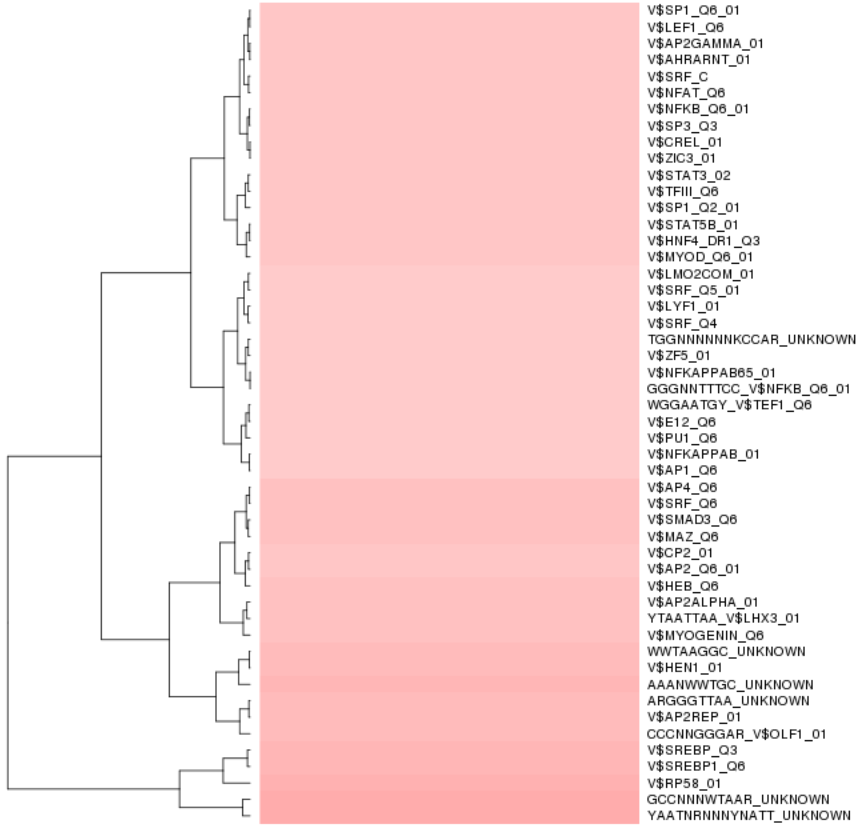
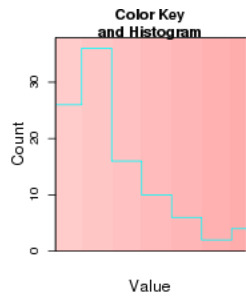
TF enrichment:

Error: no locations are finite



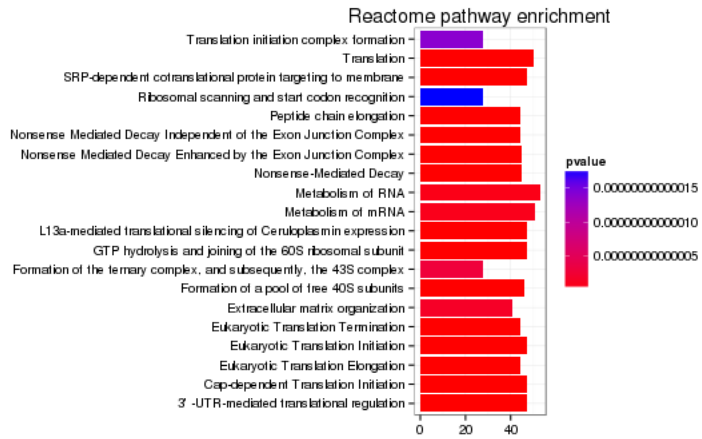
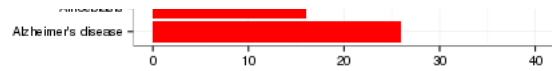
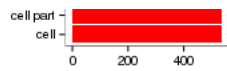
V\$LMO2COM_01
V\$ZIC3_01
V\$NFKAPPAB05_01
V\$HEN1_01
V\$HNF4_DR1_Q3
V\$LYF1_01
V\$PAX4_03
TGCCAAAR_V\$NFI_Q6
V\$E12_Q6
V\$PAX4_01
V\$AP1_Q6
CAGGTA_V\$AREB6_01
TAATTA_V\$CHX10_01
GCCNNNWTAAR_UNKNOWN
V\$PUI_Q6
V\$RP58_01
V\$NFKAPPAB_01
V\$AP2ALPHA_01
CTTTGA_V\$LEF1_Q2
V\$LEF1_Q6
CTTTAAR_UNKNOWN
RNGTGGGC_UNKNOWN
V\$AP2GAMMA_01
GGGYGTGNY_UNKNOWN
V\$AP2_Q3
WGGAAATGY_V\$TEF1_Q6
V\$CREL_01
TGCTGAY_UNKNOWN
GGGCGGR_V\$SP1_Q6
CTGCAGY_UNKNOWN
GCANCTGNY_V\$MYOD_Q6
V\$SP3_Q3
V\$HEB_Q6
V\$CP2_01
TTGTTT_V\$FOXO4_01
RGAGGAARY_V\$PUI_Q6
V\$SREBP1_Q6
TGANTCA_V\$AP1_C
V\$AP2_Q6_01
AACTTT_UNKNOWN
TGTTTG_V\$HNF3_Q6
TGGAAA_V\$NFAT_Q4_01
CCCNNGGAR_V\$OLF1_01
TGGNNNNNNKCCAR_UNKNOWN
CAGCTG_V\$AP4_Q5
CTTTGT_V\$LEF1_Q2
RYTTCCTG_V\$ETS2_B
CAGGTG_V\$E12_Q6
GGGAGGRR_V\$MAZ_Q6
GGGTGRRR_V\$PAX4_Q3

TF zscore:

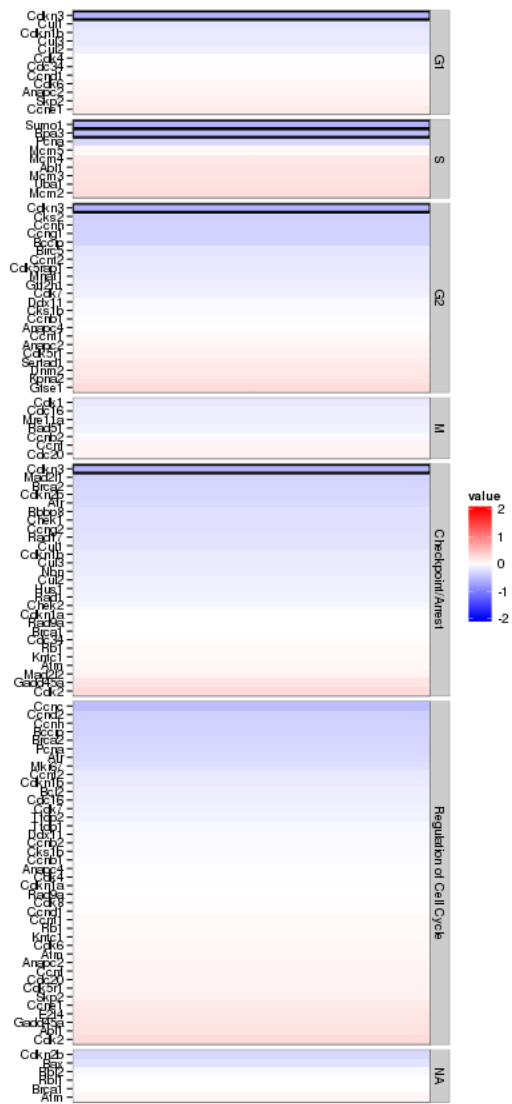


GO enrichment

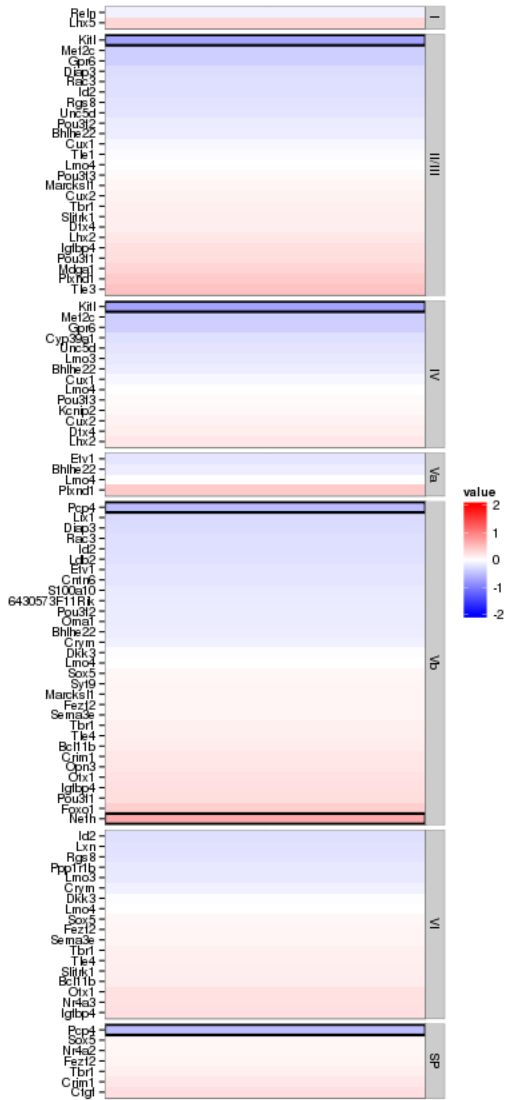
Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.



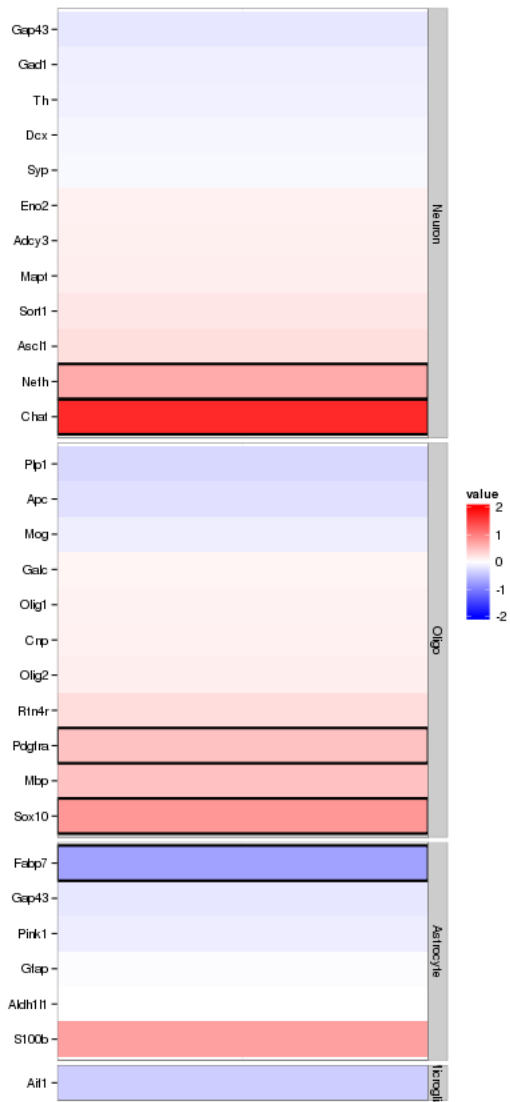
Enrichment or depletion for stage-specific cell cycle markers



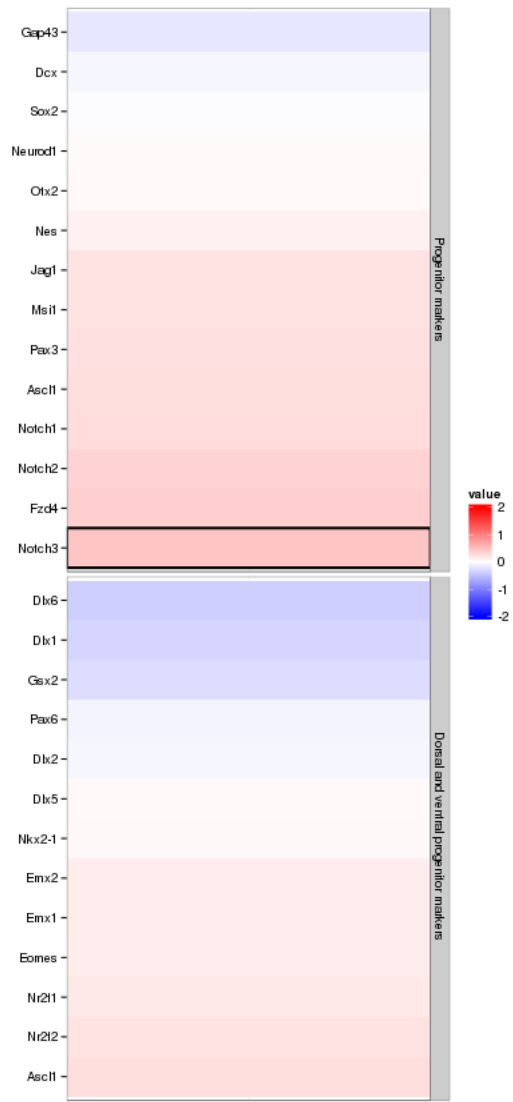
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



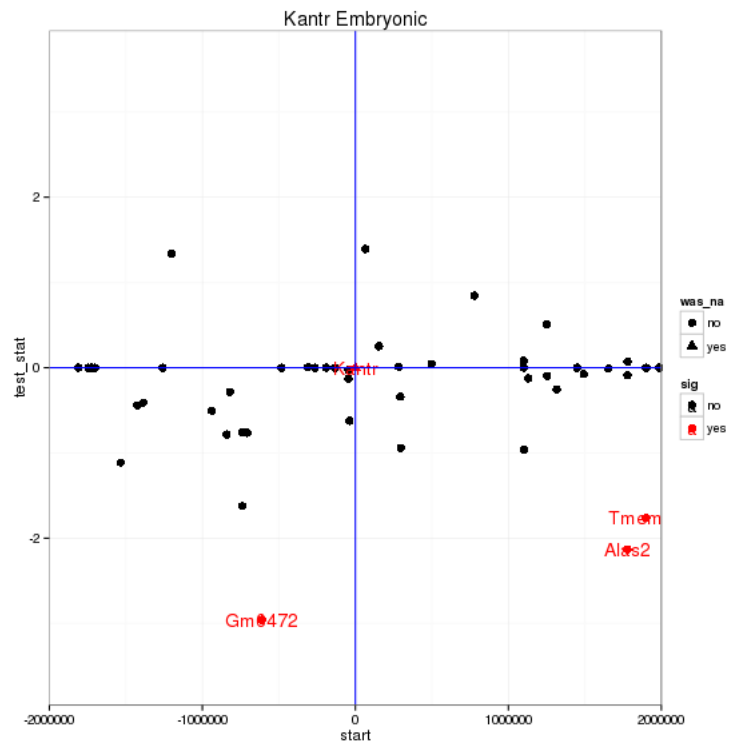
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 3 genes significantly regulated in a region this size is: 0.3615



Notes

Samples used are:

10

- 1 JR762
- 2 JR774
- 3 JR746
- 4 JR775
- 5 JR747
- 6 JR812

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR762/abundances.cxb	WT	0	WT_0	32218100.00	33340200.00	0.95	1.00
2 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR774/abundances.cxb	WT	1	WT_1	32505000.00	33340200.00	0.95	1.00
3 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR746/abundances.cxb	WT	2	WT_2	27986400.00	33340200.00	0.83	1.00
4 /n/rinn_data1/seq/igoff/Projects/BrainMap/data/quants/JR775/abundances.cxb	Kantr	0	Kantr_0	41486000.00	33340200.00	1.27	1.00

5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR747/abundances.cxb Kantr
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR812/abundances.cxb Kantr

1 Kantr_1	33582200.00	33340200.00	1.03	1.00
2 Kantr_2	34934000.00	33340200.00	1.08	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8    LC_NAME=C
## [9] LC_ADDRESS=C            LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
```

```

## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRT_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17       cluster_1.15.2     colorspace_1.2-4
## [7] dichromat_2.0-0    digest_0.6.4       DO.db_2.7
## [10] evaluate_0.5.5     formatR_0.10       Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3    graph_1.40.1
## [16] graphite_1.8.1     gtools_3.4.1       Hmisc_3.14-4
## [19] igraph_0.7.1       KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2       lattice_0.20-29    latticeExtra_0.6-26
## [25] MASS_7.3-33        Matrix_1.1-3       munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10       qvalue_1.36.0
## [31] Rcpp_0.11.2        RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3  scales_0.2.4       splines_3.0.2
## [37] stats4_3.0.2       survival_2.37-7    tcltk_3.0.2
## [40] tools_3.0.2        XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```

##      param
## 1    cmd_line
## 2    version
## 3  SVN_revision
## 4  boost_version
## 5    genome
##
## 1 cuffdiff -p 10 -L WT,Kantr -o /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/diffs/Kantr_vs_WT_Embryonic /n/rinn_data1/seq/lgooff/Projects/BrainMap/dat
## 2
## 3
## 4
## 5

```

linc-Brn1a KO vs WT (Adult)

This file shows the wt-v-ko comparison for linc-Brn1a.

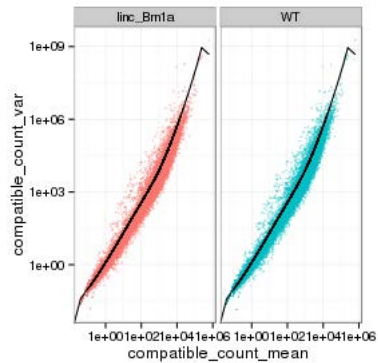
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

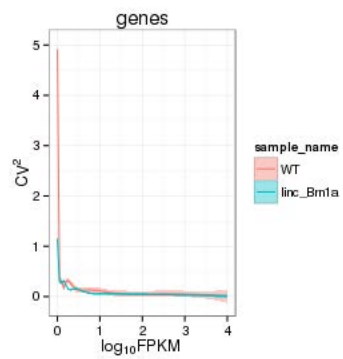
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



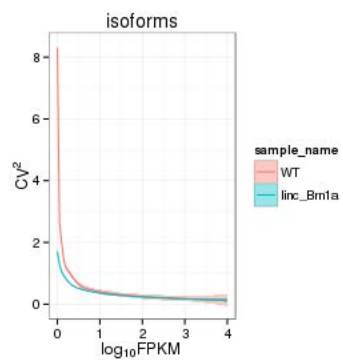
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

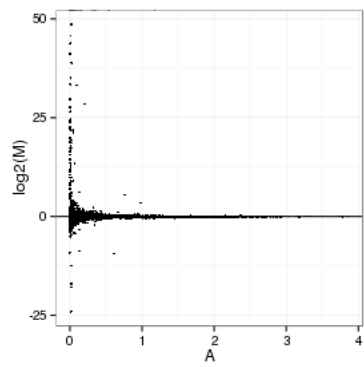
Genes:



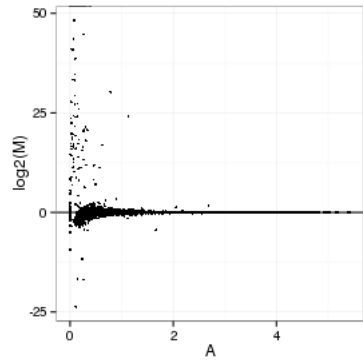
Isoforms:



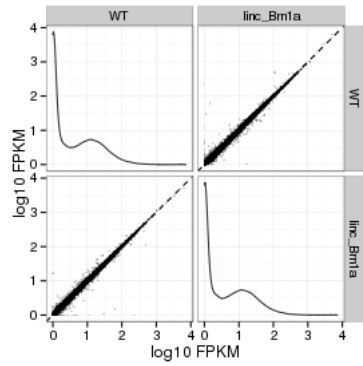
MvA plot



MvA plot counts



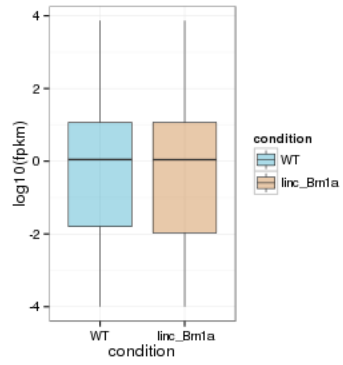
Scatterplot matrix



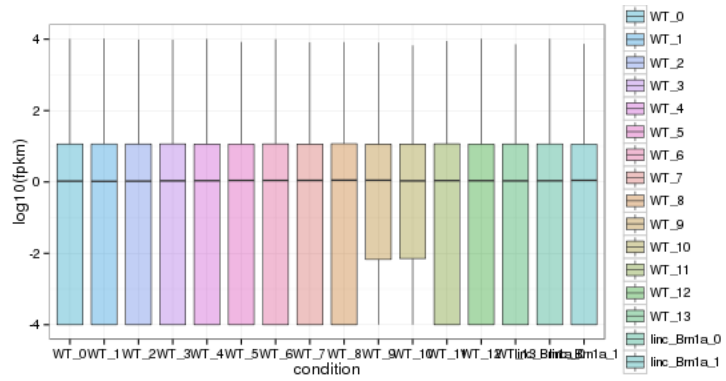
Distributions

Boxplots

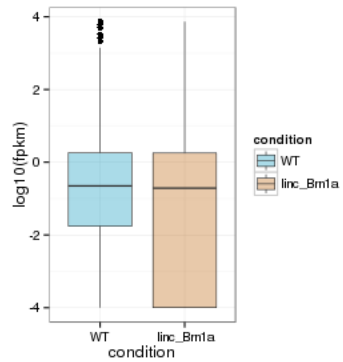
Boxplot (genes)



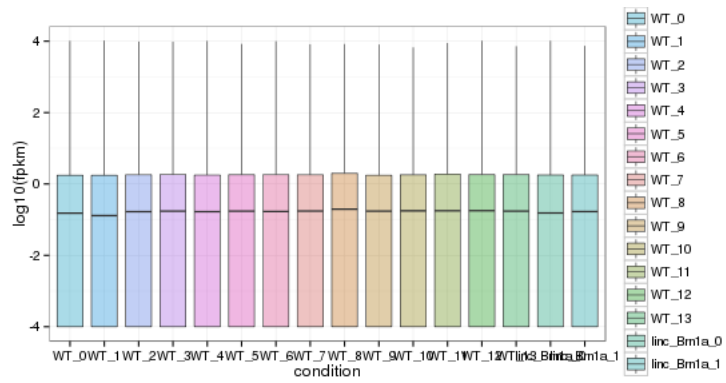
Boxplot (genes, replicates)



Boxplot (isoforms)

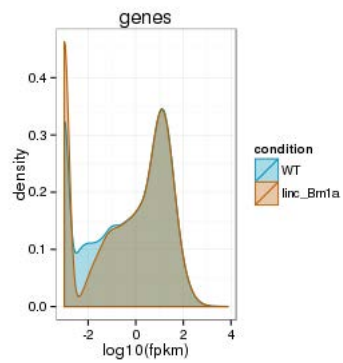


Boxplot (isoforms, replicates)

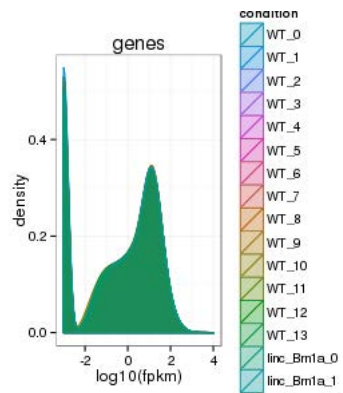


Density

Density (genes)

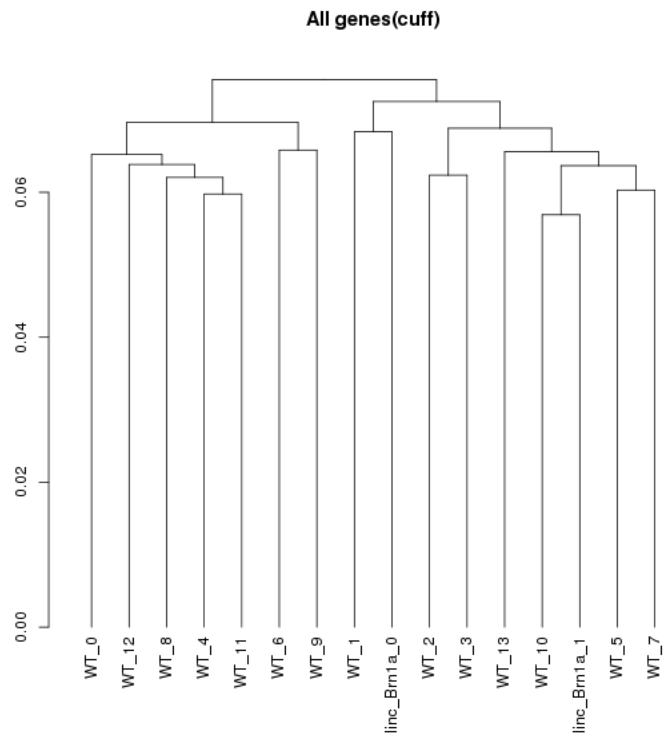


Density (genes, replicates)



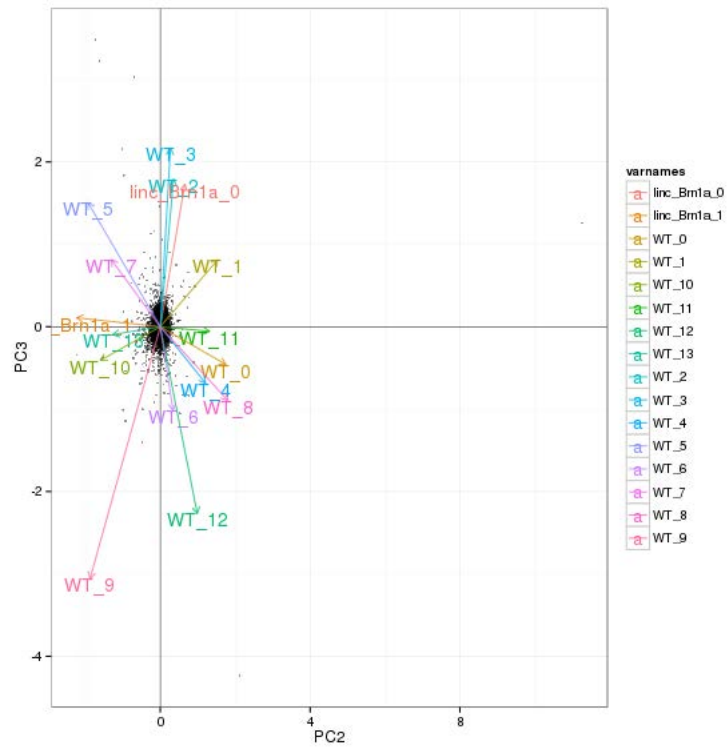
Clustering

Replicate Clusters

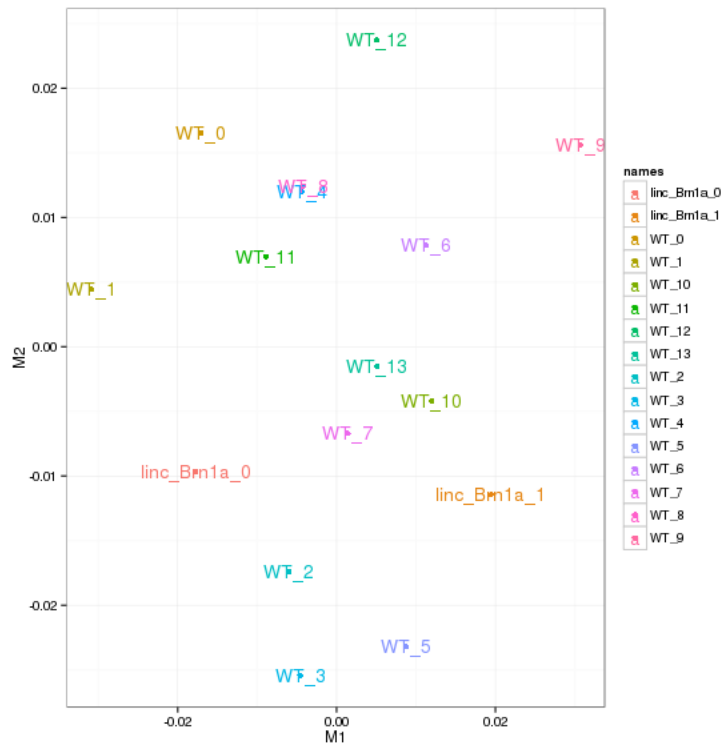


NULL

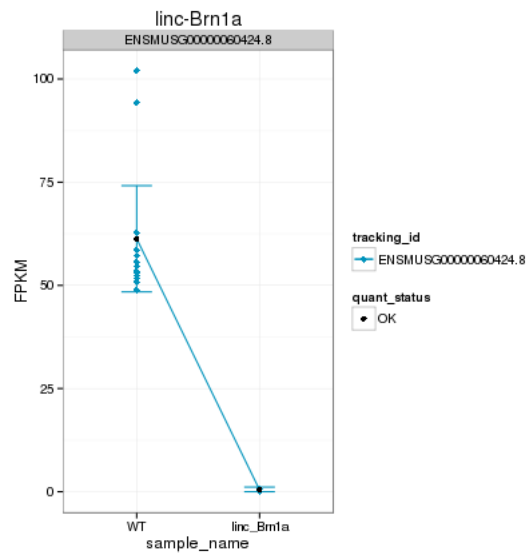
PCA (genes)



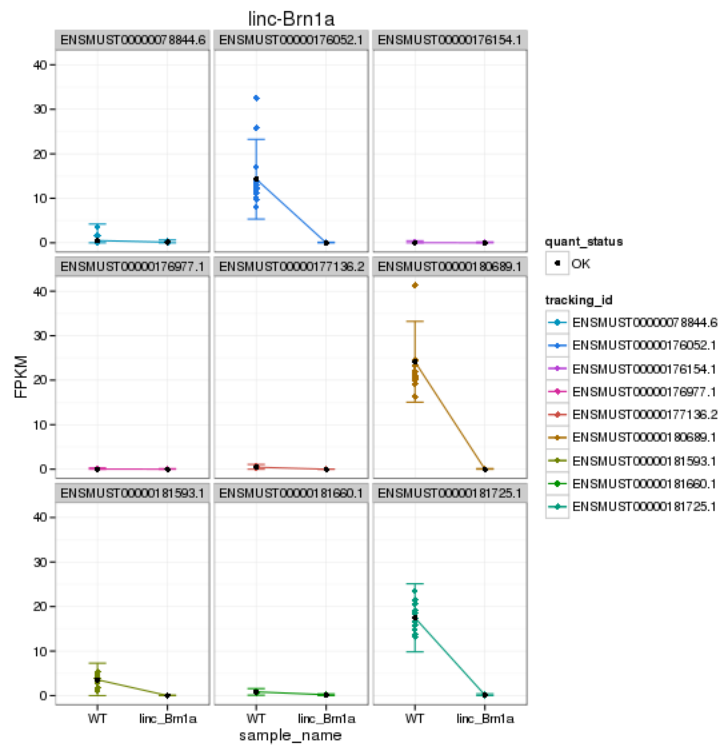
MDS (genes)



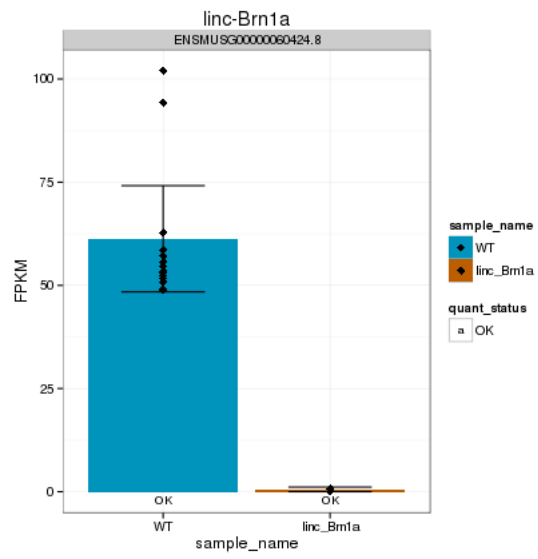
KO assessment
Endogenous lncRNA expression



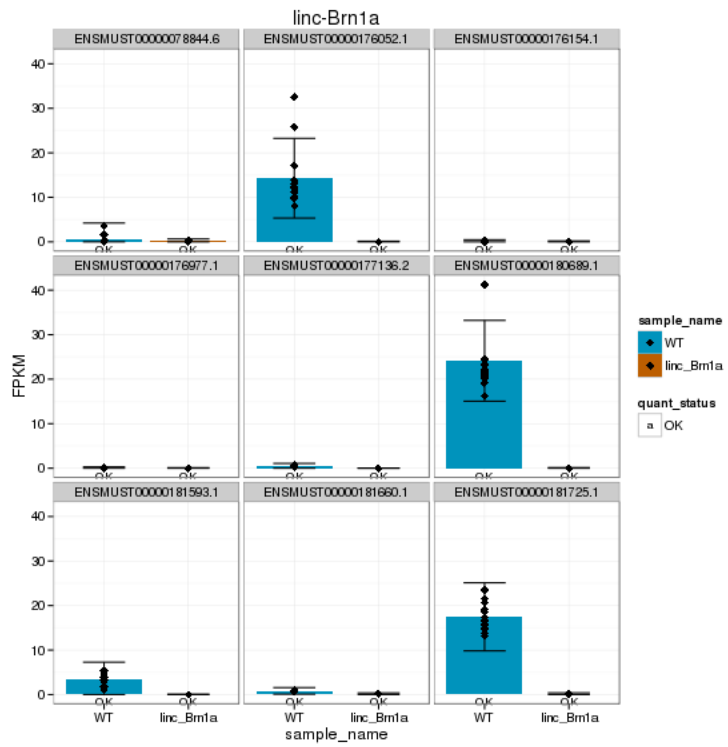
Endogenous expression of linc-Brn1a isoforms:



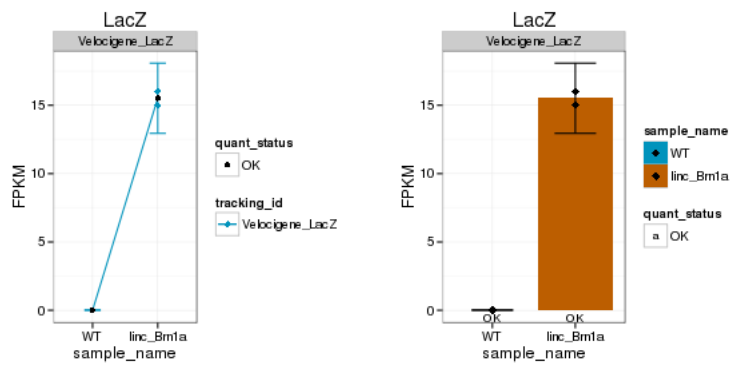
Barplot of gene expression:



Barplot of isoform expression:

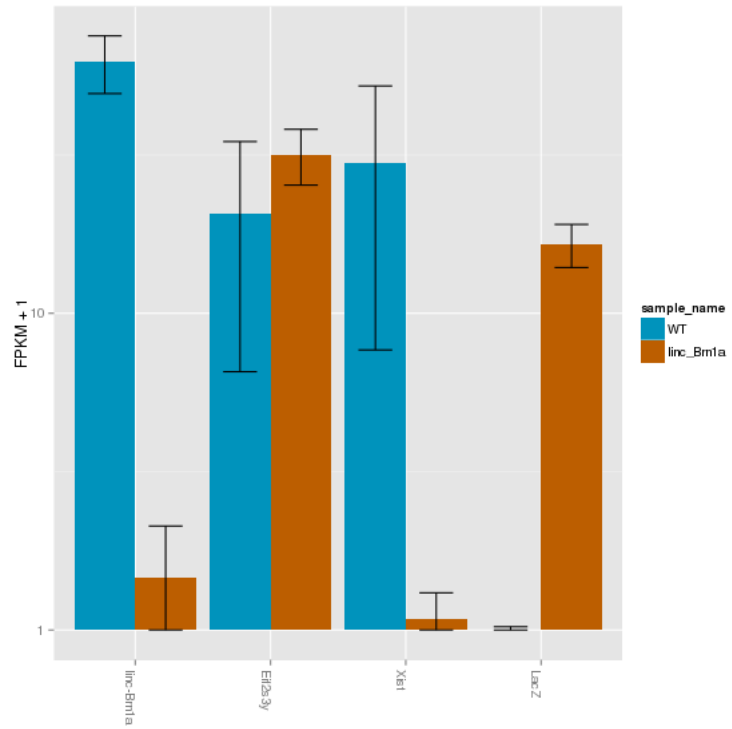


LacZ expression

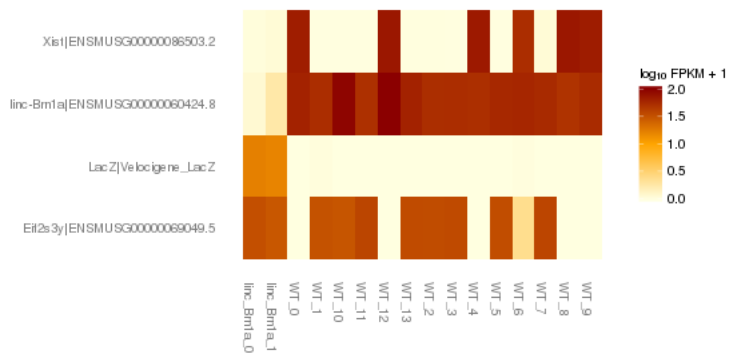


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

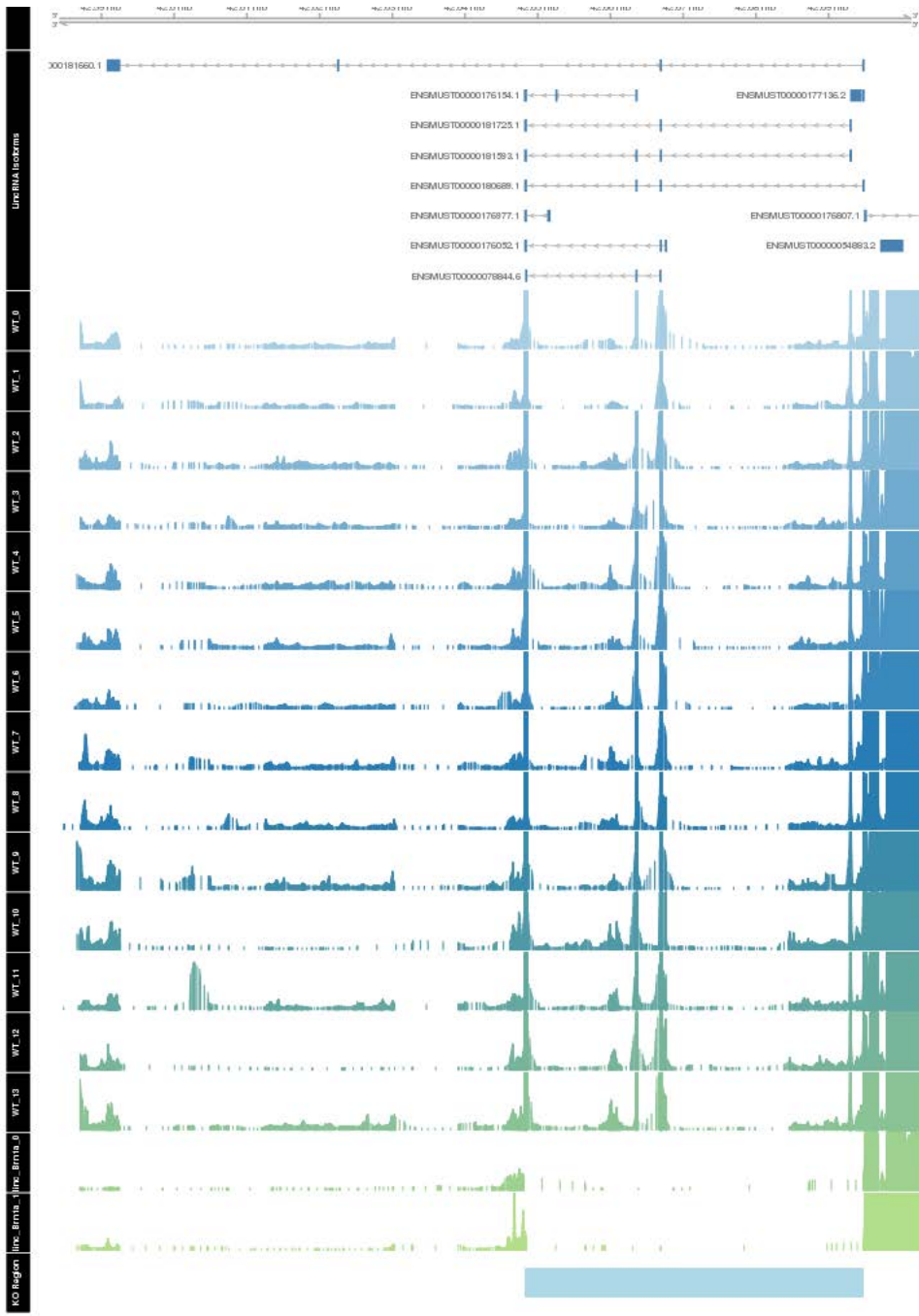


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 74 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 Rec8
2 Arrdc2
3 Fosb
4 Dio2
5 Mertk
6 Hspa8
7 Vip
8 Fabp7
9 Sgk1
10 Mybpc1
11 Btg2
12 Xbp1
13 Fos
14 Rgcc
15 Dnajc3
16 Arc
17 Sdf2l1
18 Nr4a1
19 Cdkn1a
20 Sik1
21 Xdh
22 Dusp1
23 Fkbp5
24 Il33
25 Pdia4
26 Paqr8
27 Mcm6
28 Tfcp2l1
29 Hspa5
30 Cyr61
31 Nr4a3
32 Alad
33 Slc2a1
34 Mfsd2a
35 Adipor2
36 Abcc9
37 Pglyrp1
```

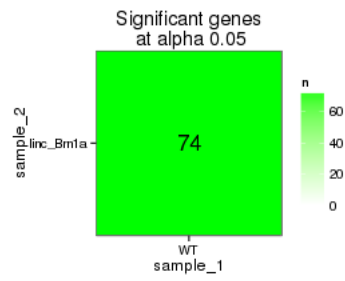
38 Hddc3
39 Tsc22d3
40 Cryab
41 Ldlr
42 Ndn
43 Igsf9b

44 Cbl
45 Nostrin
46 Lars2
47 Anln
48 Tob1
49 Egr2
50 Smim3
51 Egr1
52 Hdhd3
53 Daam2
54 Tlr7
55 Npas4
56 Nrros
57 Ccdc171
58 Jun
59 Junb
60 Ier2
61 Per2
62 Kdm5d
63 linc-Bm1a
64 Usp31
65 Zbtb16
66 Ddx3y
67 Kihl9
68 Egr4
69 Gm7292
70 Xist

71 Cdr1
72 AA465934
73 Gm6472
74 Gm26924

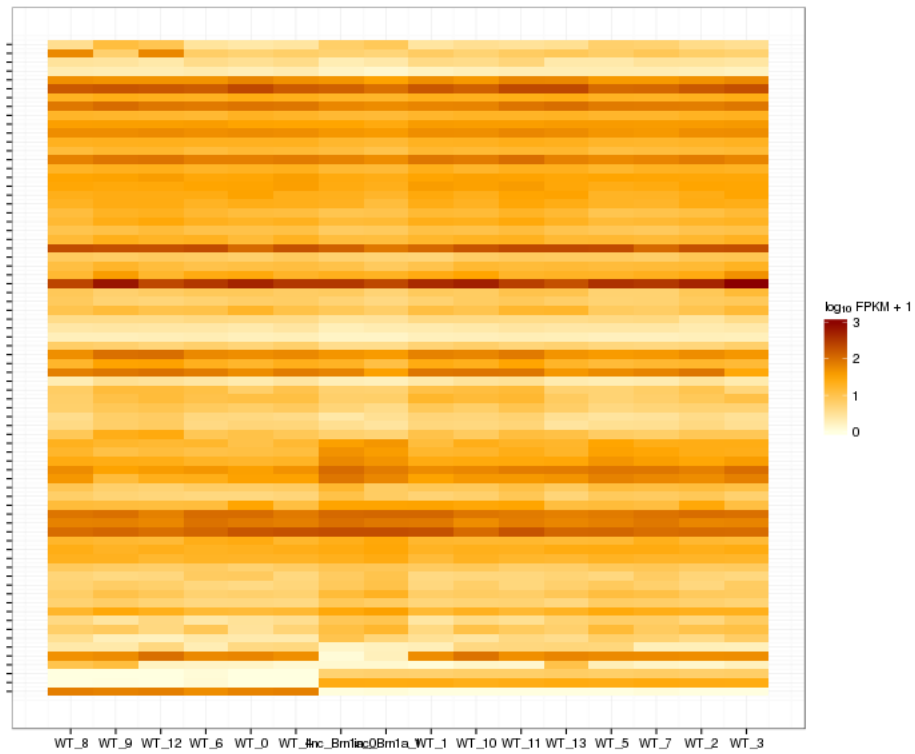
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

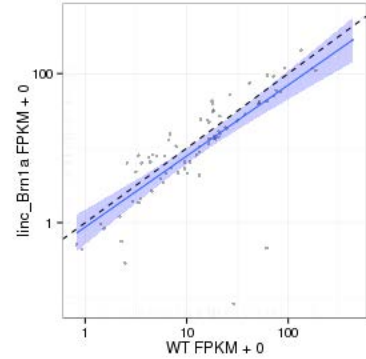


Significant genes with expression >50fpkm (any condition):(turned off)

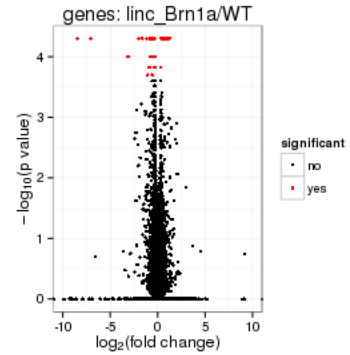
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

Scatter plot of significant genes only:

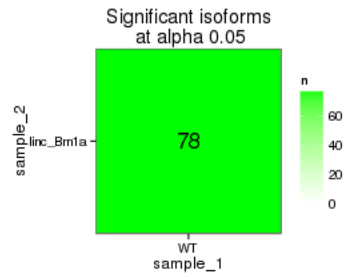


Volcano Plot



Volcano plot with significant genes only:

Per isoform difference between conditions:



These isoforms are:

isoAnnot\$gene_short_name

- 1 Rec8
- 2 Arrdc2
- 3 Fosb
- 4 Hif3a
- 5 Dio2
- 6 Mertk
- 7 Hspa8
- 8 Vip
- 9 Fabp7

- 10 Cpm
- 11 Tns3
- 12 Btg2
- 13 Xbp1
- 14 Atp2a3
- 15 Fos
- 16 Arc
- 17 Zbtb20
- 18 Sdf2l1
- 19 Nr4a1
- 20 Cdkn1a
- 21 Sik1
- 22 Xdh
- 23 Atp6v0c
- 24 Dusp1
- 25 Fkbp5
- 26 Il33

27 Pdia4
28 Paqr8
29 Tfcp2l1
30 Hspa5
31 Cyr61
32 Alad
33 Slc2a1
34 Mfsd2a
35 Ago3
36 Map3k6

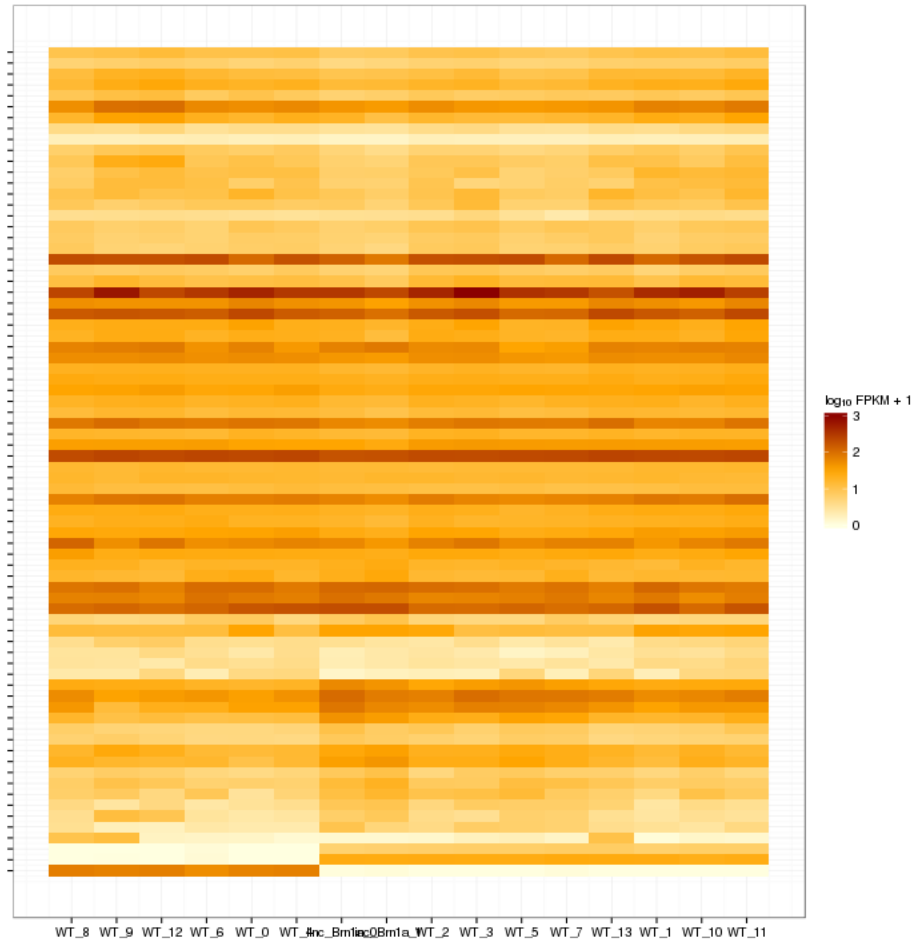
37 Ccnl2
38 Adipor2
39 Pglyrp1
40 Hddc3
41 Tsc22d3
42 Cryab
43 Kdm4a
44 Ndn
45 Vps8
46 Igsf9b
47 Cbl
48 Anln
49 Kirrel2
50 Egr2
51 Smim3
52 Egr1
53 Hdhd3
54 Eltd1
55 Daam2
56 Dmtf1
57 Npas4
58 Ezr
59 Jun
60 Junb
61 Ier2
62 Per2
63 Kdm5d

64 Usp31

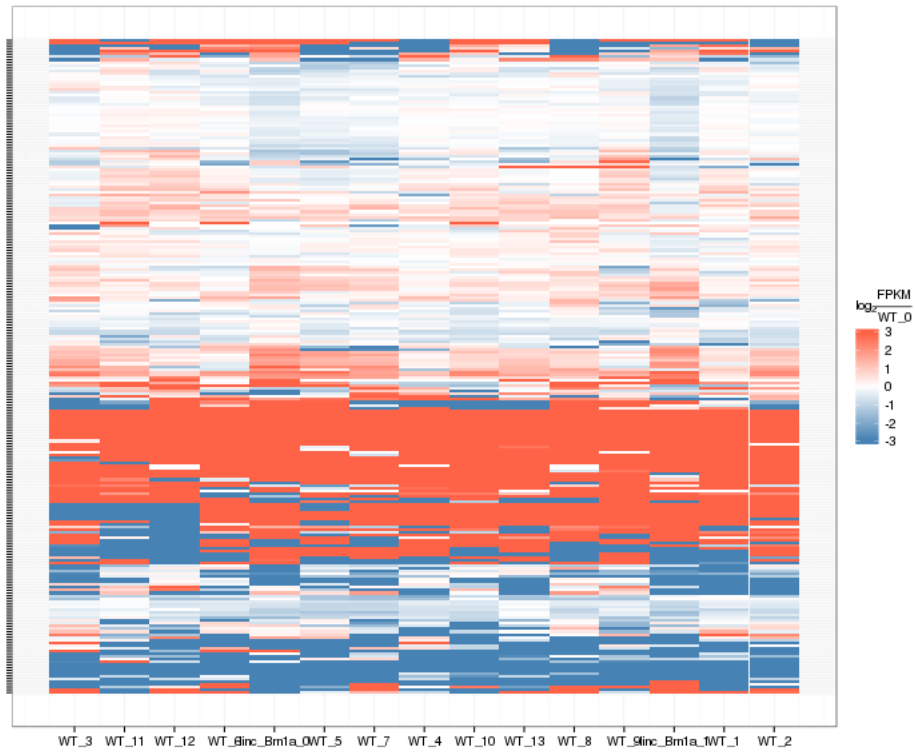
65 Sepp1
66 Zbtb16
67 Ddx3y
68 Ahnak

- 69 Kihl9
- 70 Egr4
- 71 Gm7292
- 72 Xist
- 73 Cdr1
- 74 Malat1
- 75 Gm6472
- 76 Gm26924

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

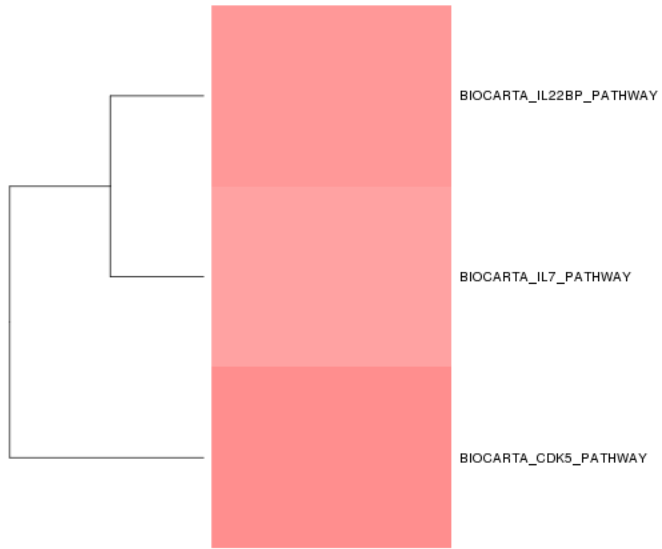
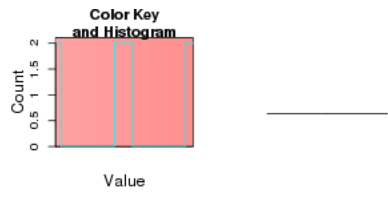
Gene/Pathway Analysis

GSEA

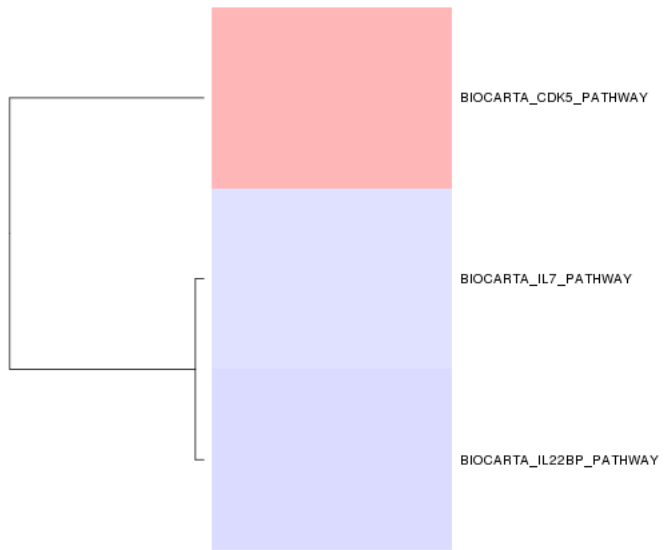
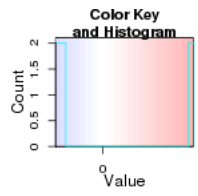
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkm}_{\text{KO}}/\text{fpkm}_{\text{WT}}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

Biocarta enrichment:

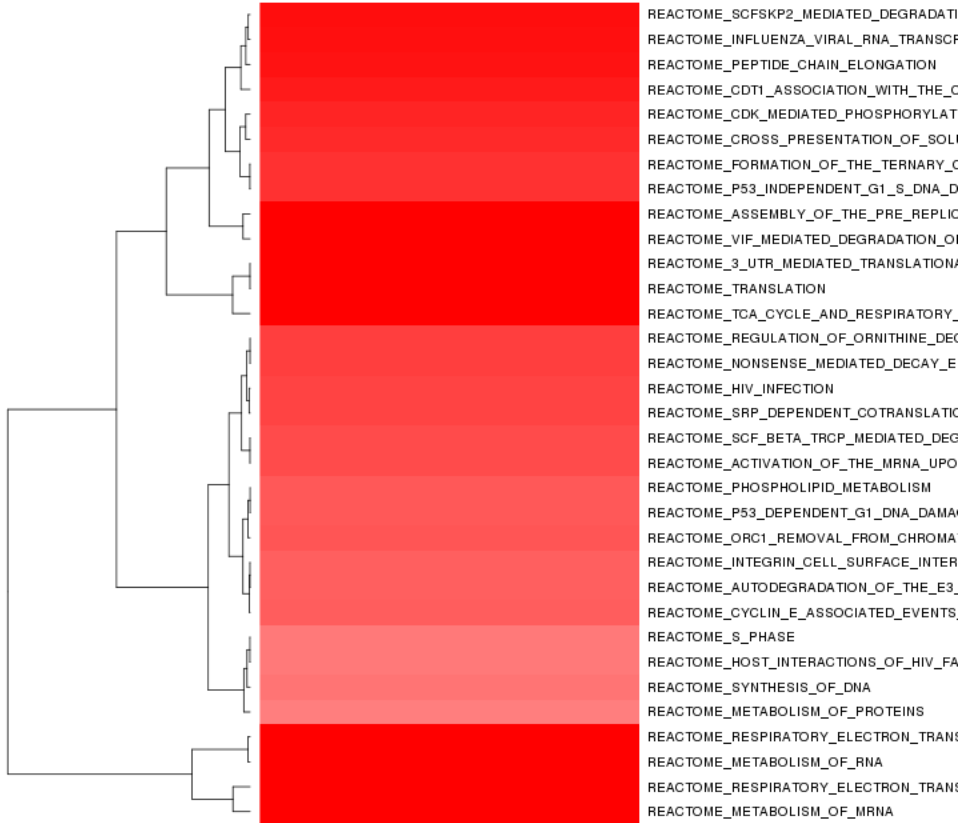
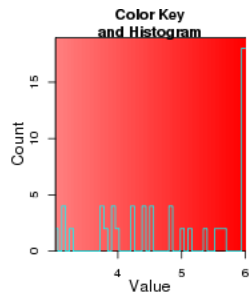


Biocarta zscore:



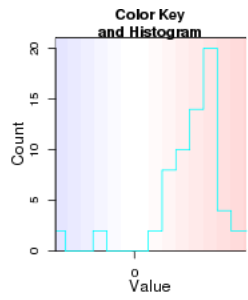
Reactome enrichment:

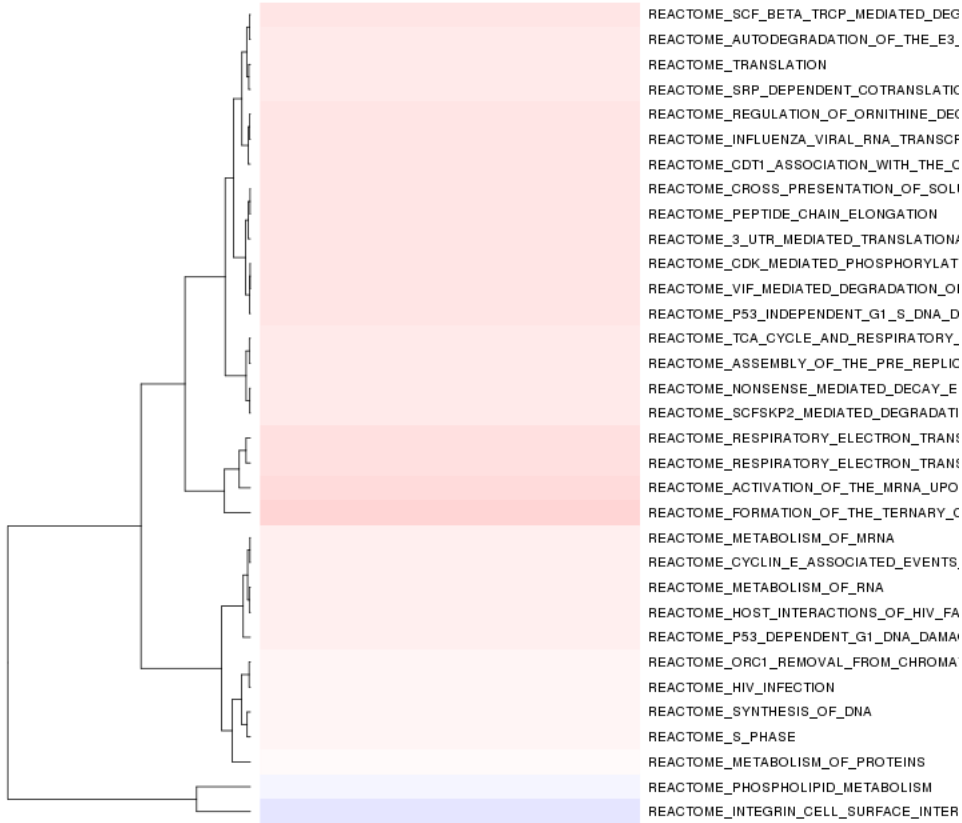
```
## Error: subscript out of bounds
```



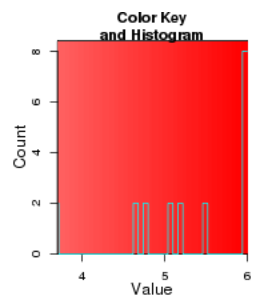
Reactome zscore:

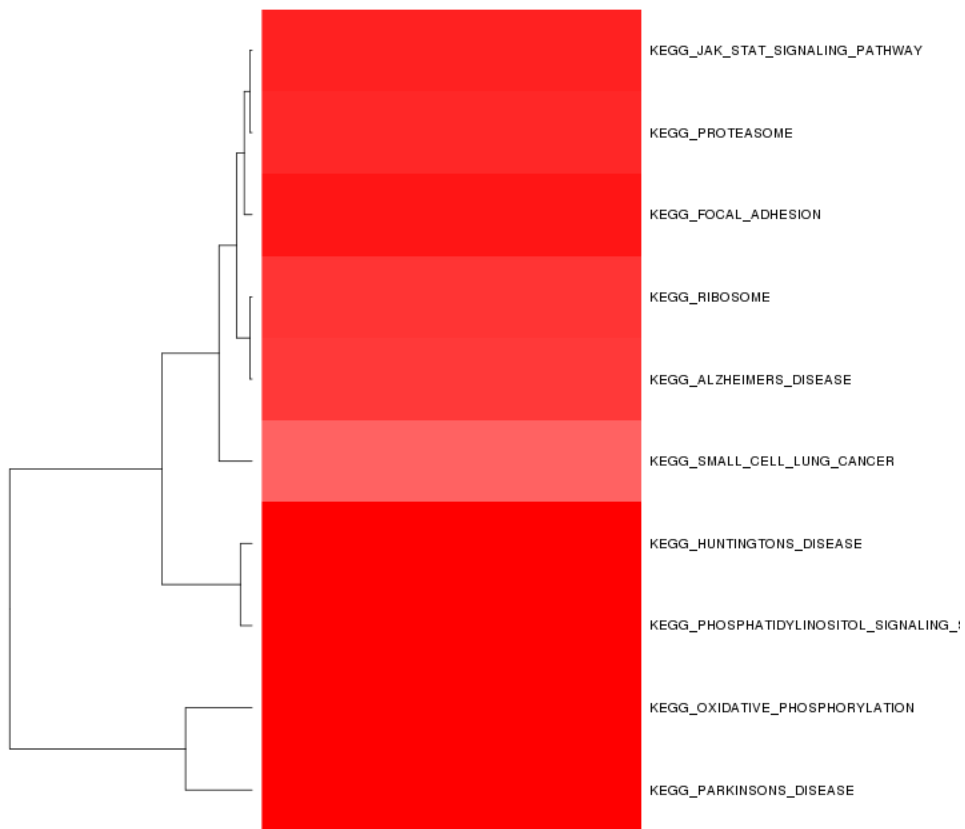
Error: subscript out of bounds



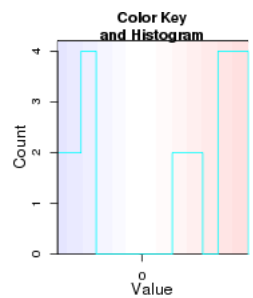


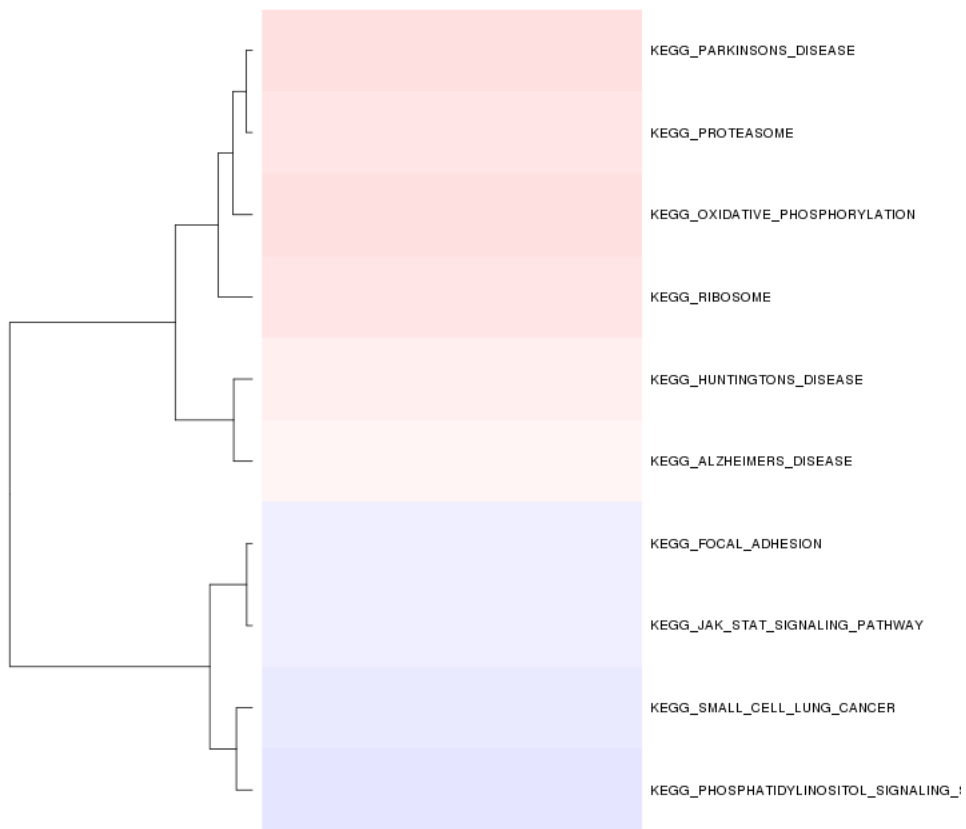
Kegg enrichment:





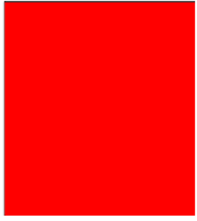
Kegg zscore:





Interneuron enrichment:

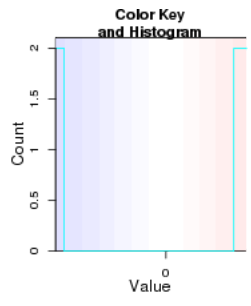
Error: no locations are finite



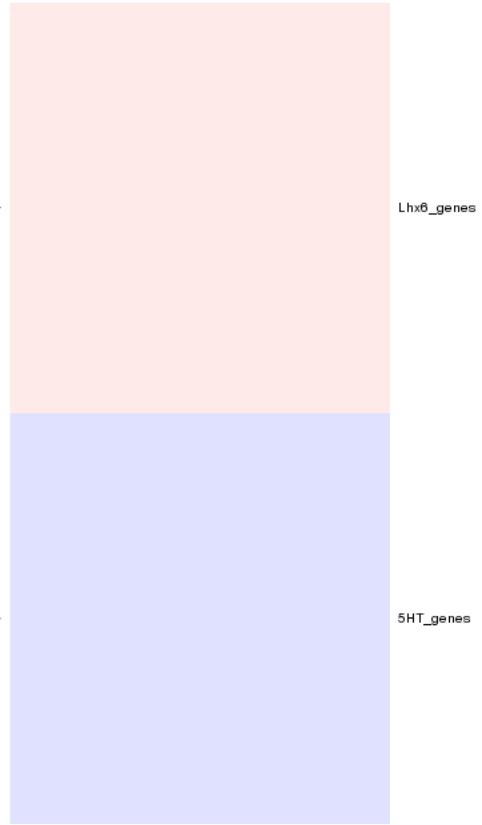
5HT_genes

Lhx6_genes

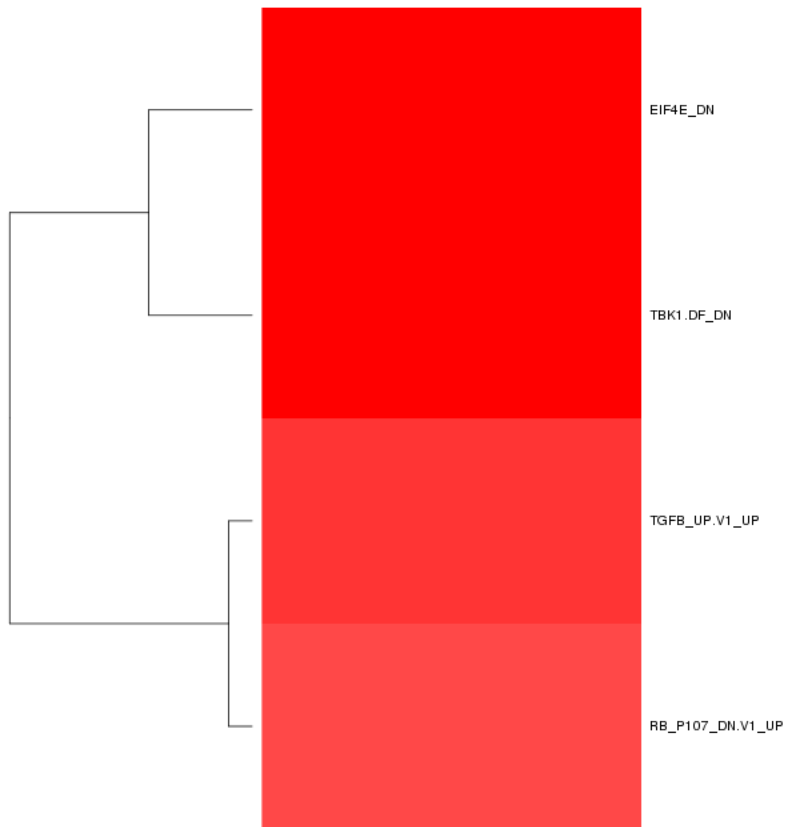
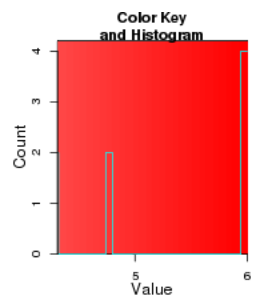
Interneuron zscore:



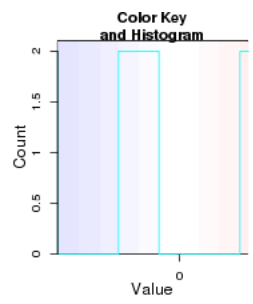
—

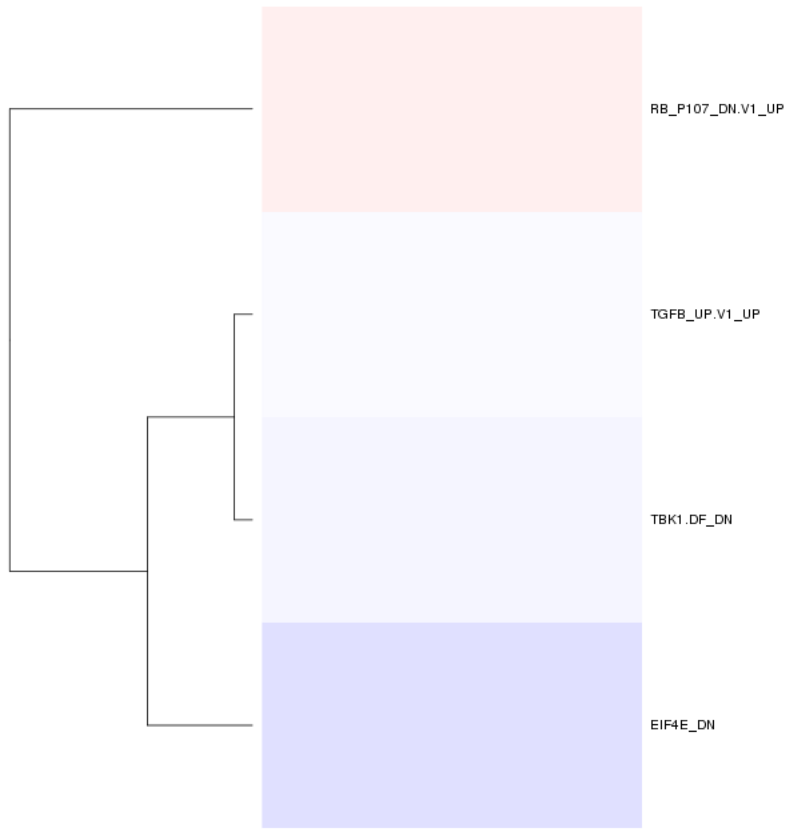


Oncogene enrichment:

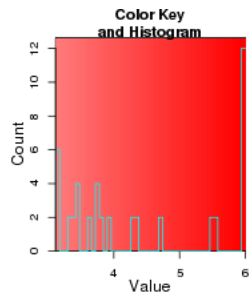


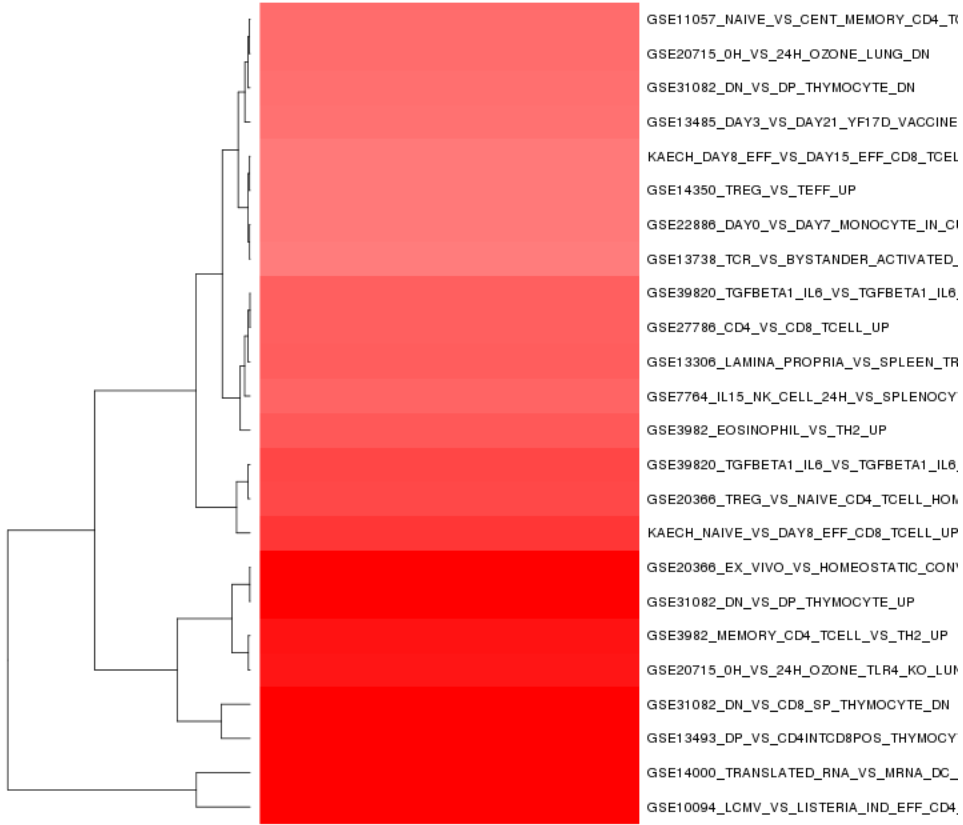
Oncogene zscore:



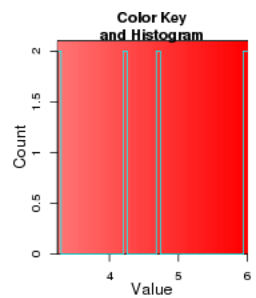


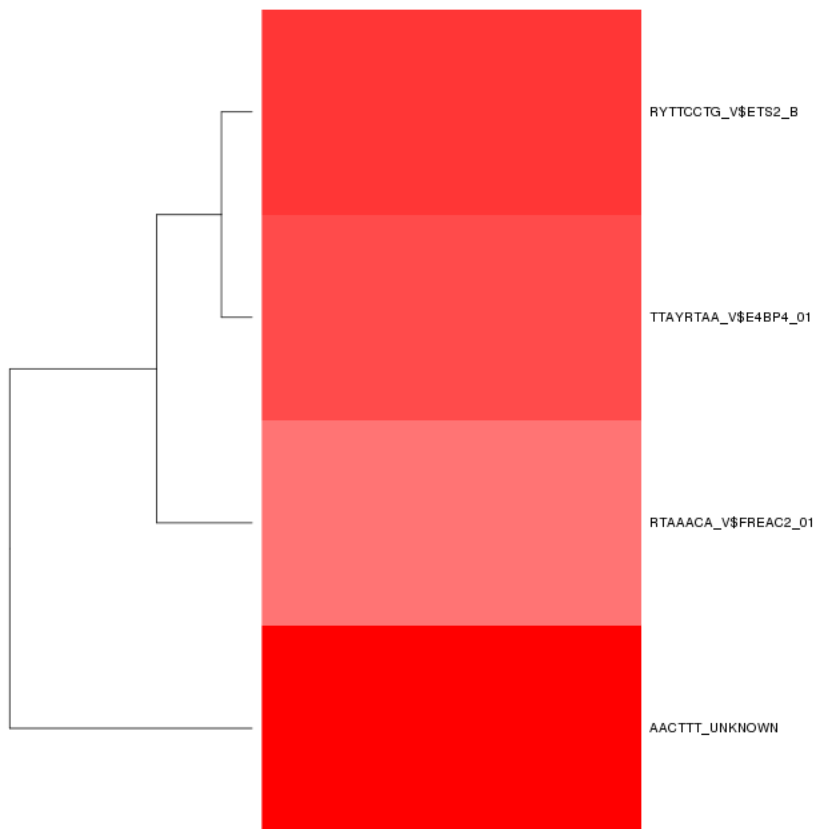
Immuno enrichment:



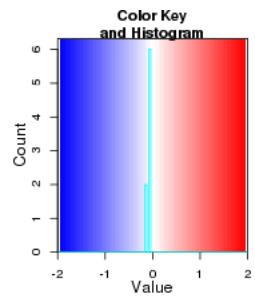


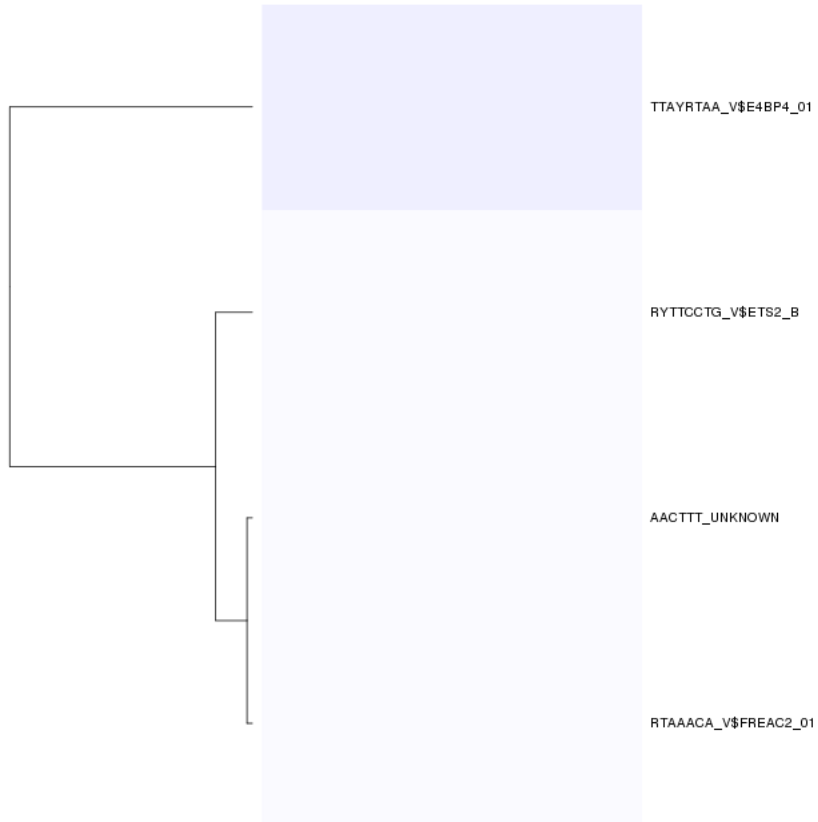
Immuno zscore:





TF zscore:

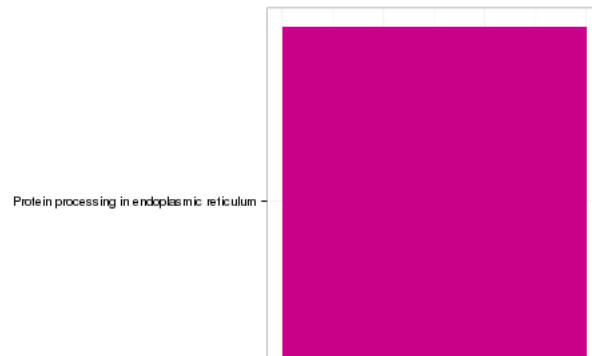
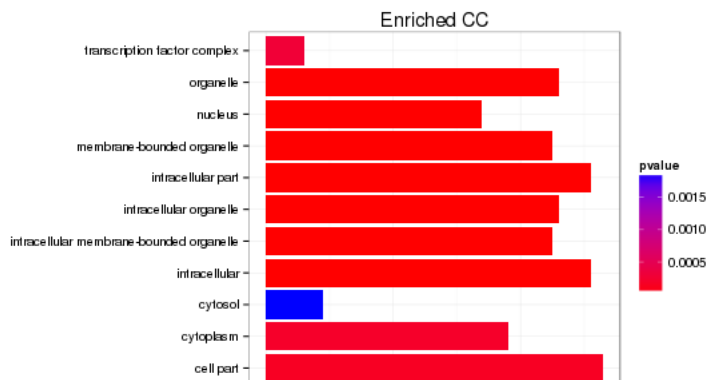
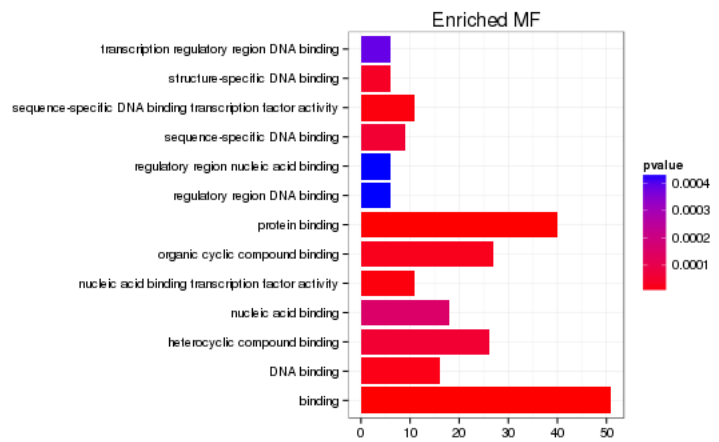
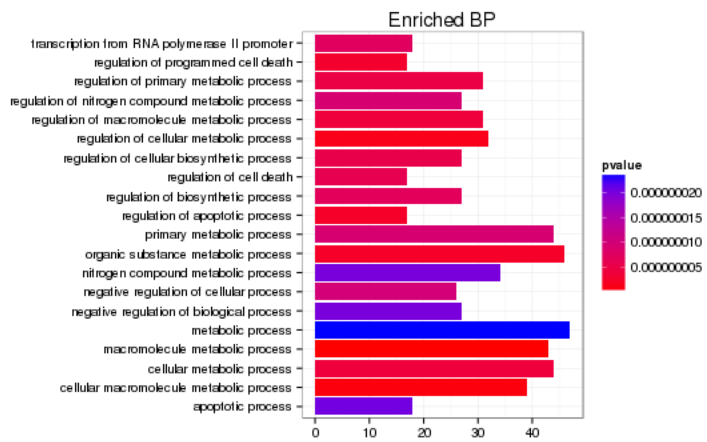


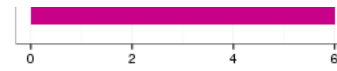
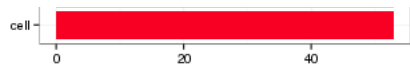


GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

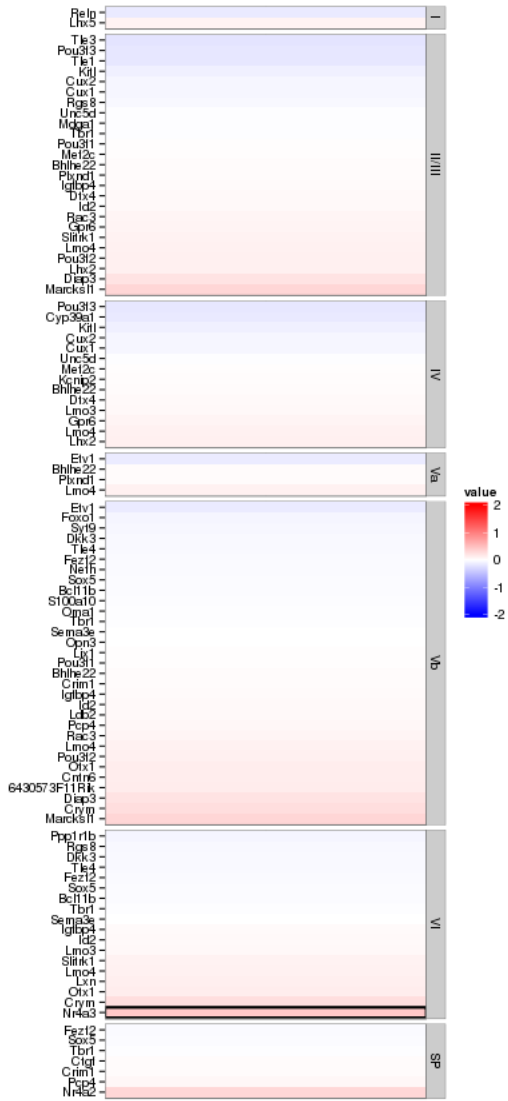
```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```



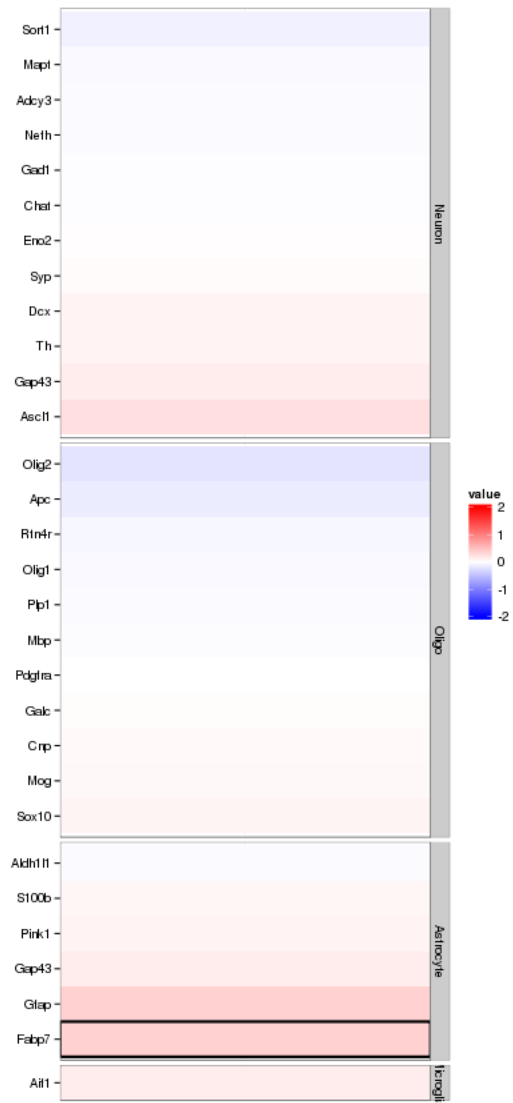


```
## Error: 'x' and 'units' must have length > 0
```

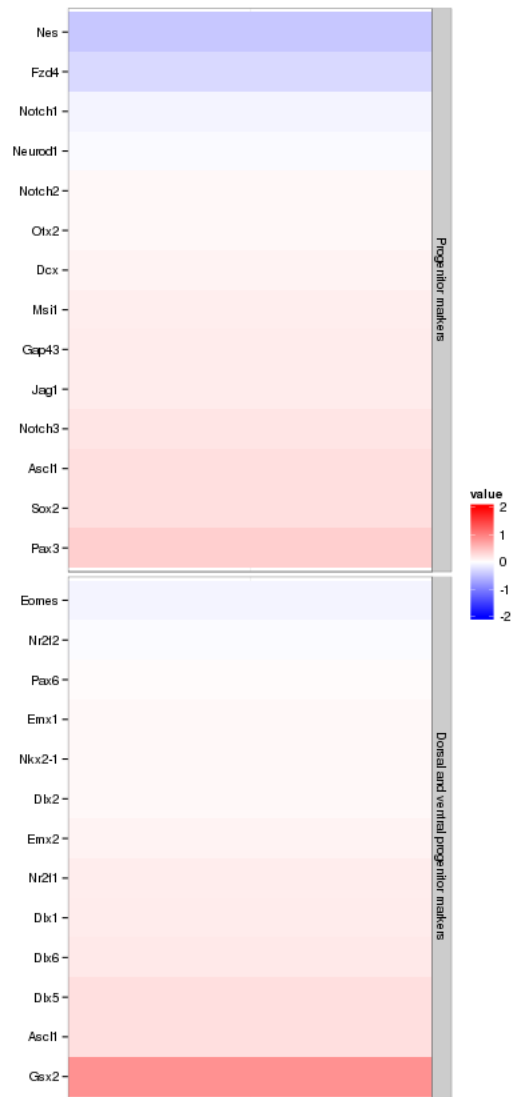
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



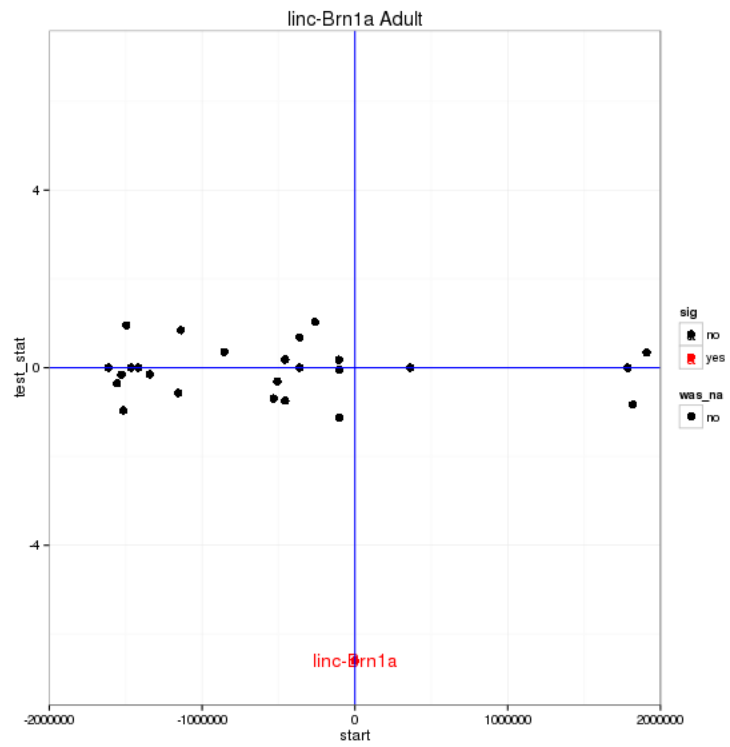
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR730

16 JR837

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	33756400.00	0.78	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	33756400.00	0.60	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	33756400.00	1.00	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	33756400.00	0.82	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	33756400.00	1.06	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	33756400.00	1.10	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	33756400.00	0.83	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	33756400.00	1.18	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	33756400.00	1.04	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	33756400.00	1.33	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	33756400.00	1.54	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	33756400.00	1.04	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	33756400.00	1.00	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	33756400.00	1.24	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR730/abundances.cxb	linc_Brn1a	0	linc_Brn1a_0	24947000.00	33756400.00	0.74	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR837/abundances.cxb	linc_Brn1a	1	linc_Brn1a_1	38529800.00	33756400.00	1.16	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostings_2.30.1
```

```

## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17       cluster_1.15.2     colorspace_1.2-4
## [7] dichromat_2.0-0    digest_0.6.4       DO.db_2.7
## [10] evaluate_0.5.5     formatR_0.10       Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3    graph_1.40.1
## [16] graphite_1.8.1     gtools_3.4.1       Hmisc_3.14-4
## [19] igraph_0.7.1       KEGG.db_2.10.1    KernSmooth_2.23-12
## [22] labeling_0.2       lattice_0.20-29    latticeExtra_0.6-26
## [25] MASS_7.3-33       Matrix_1.1-3       munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10       qvalue_1.36.0
## [31] Rcpp_0.11.2        RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3  scales_0.2.4       splines_3.0.2
## [37] stats4_3.0.2      survival_2.37-7    tcltk_3.0.2
## [40] tools_3.0.2       XML_3.98-1.1      zlibbioc_1.8.0

```

Run Info

```
##      param
## 1    cmd_line
## 2      version
## 3  SVN_revision
## 4  boost_version
## 5      genome
##
## 1 cuffdiff -p 10 -L WT,linc-Brn1a -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/linc-Brn1a_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainM
## 2
## 3
## 4
## 5
```

linc-Brn1a KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for linc-Brn1a.

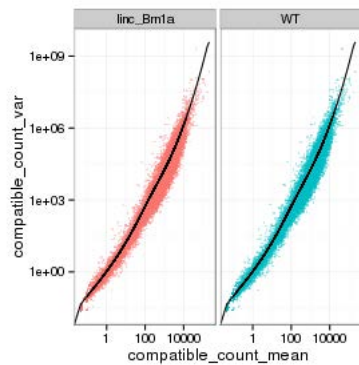
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

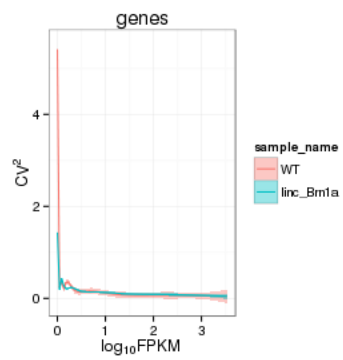
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



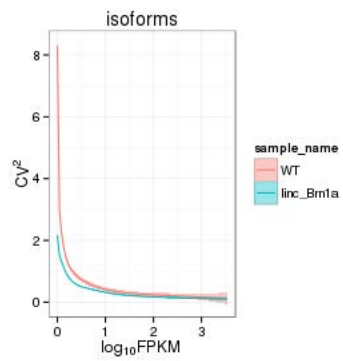
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

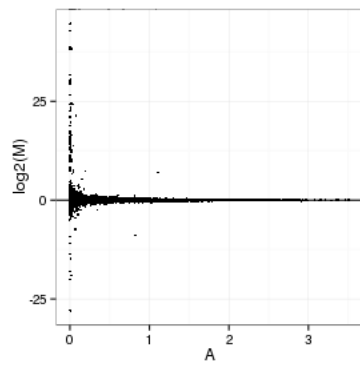
Genes:



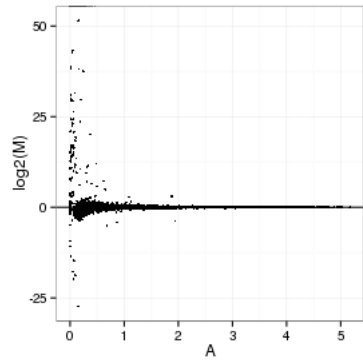
Isoforms:



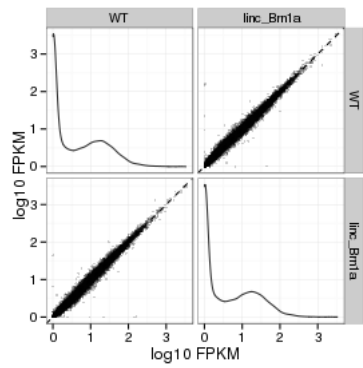
MvA plot



MvA plot counts



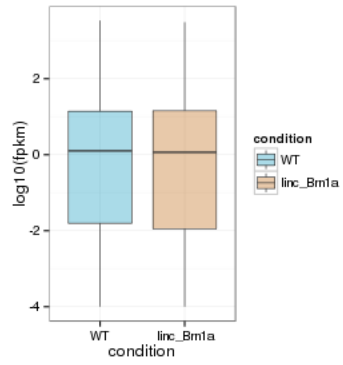
Scatterplot matrix



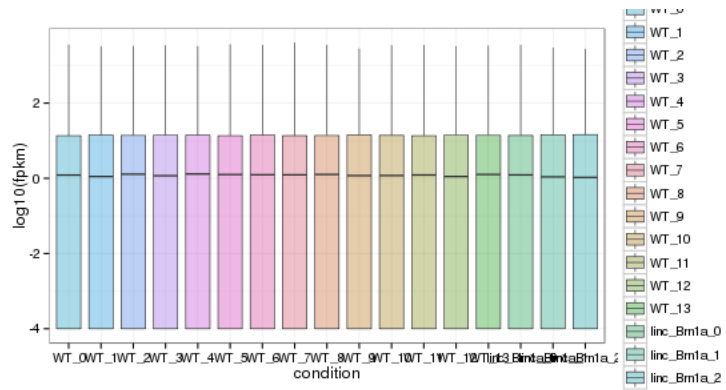
Distributions

Boxplots

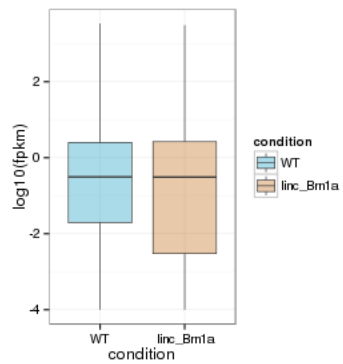
Boxplot (genes)



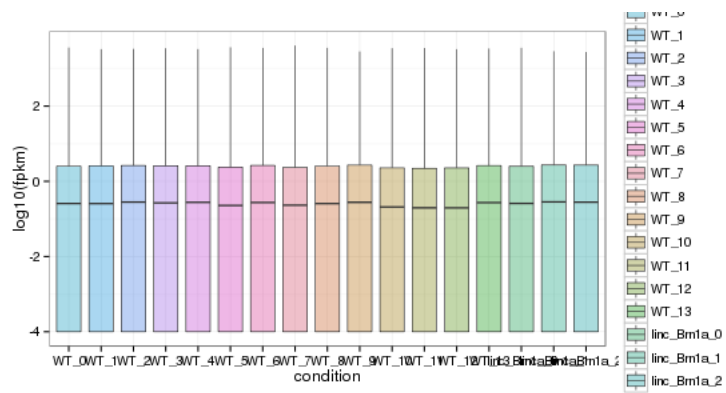
Boxplot (genes, replicates)



Boxplot (isoforms)

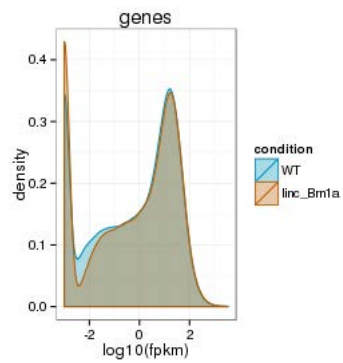


Boxplot (isoforms, replicates)

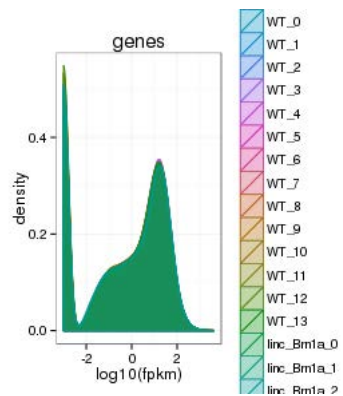


Density

Density (genes)

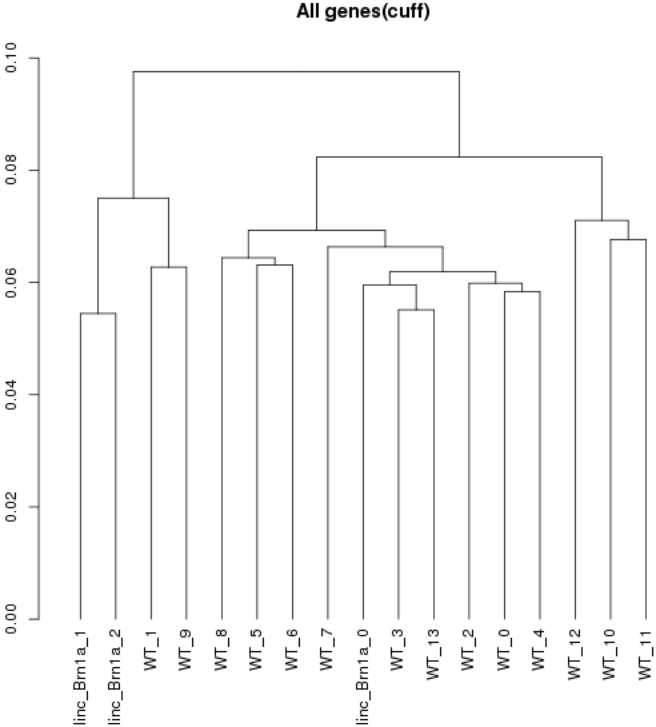


Density (genes, replicates)



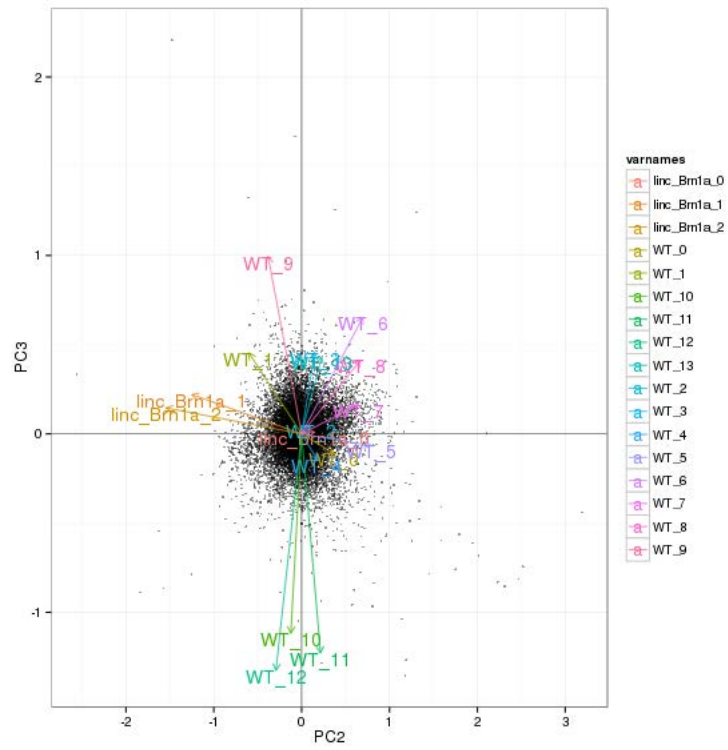
Clustering

Replicate Clusters

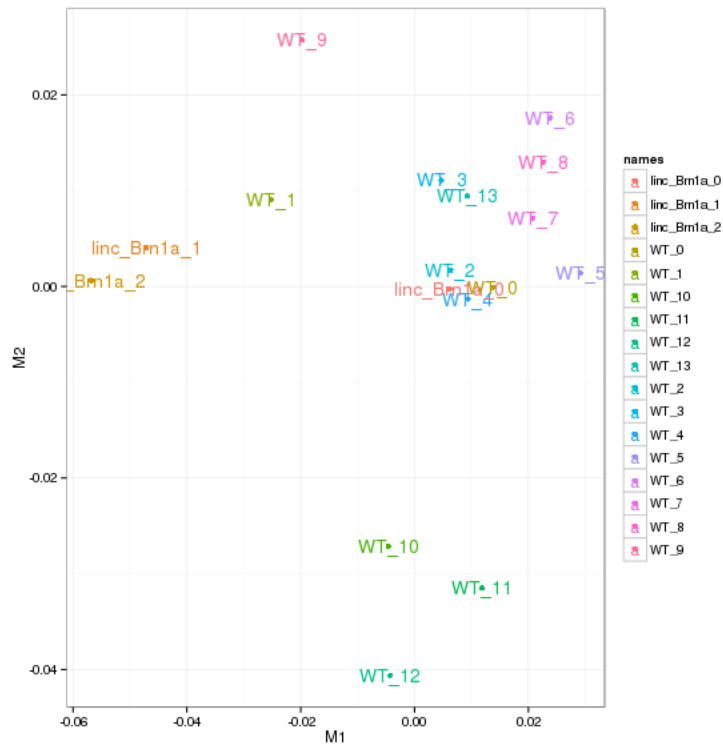


NULL

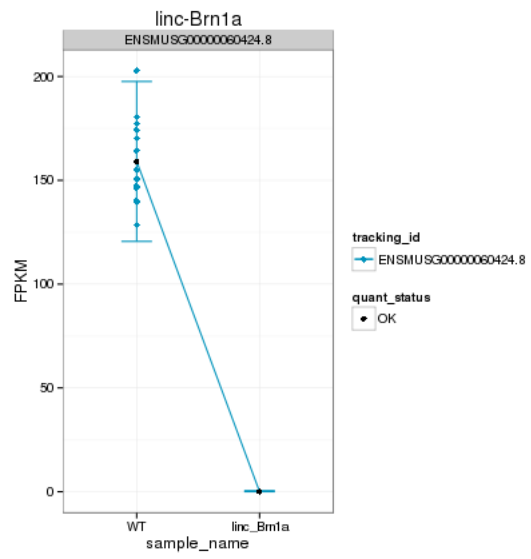
PCA (genes)



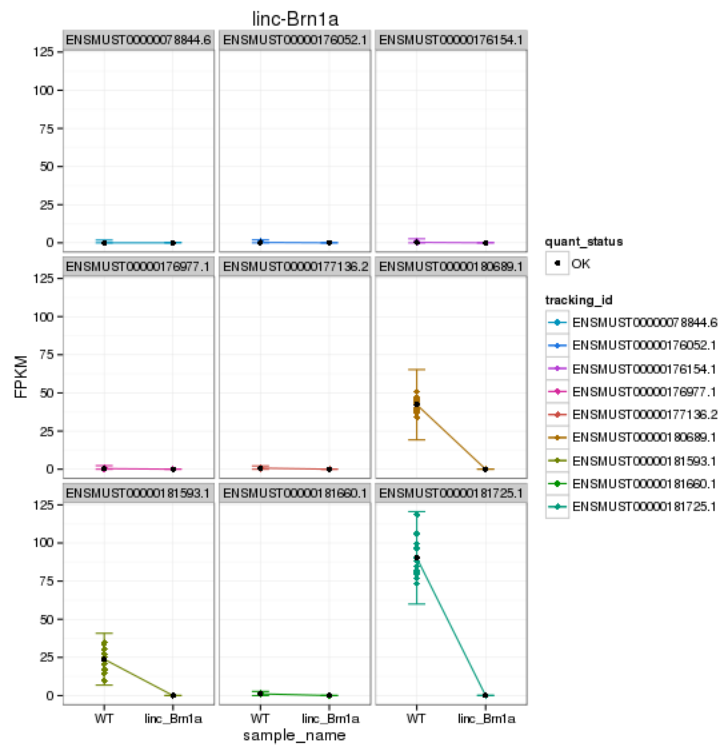
MDS (genes)



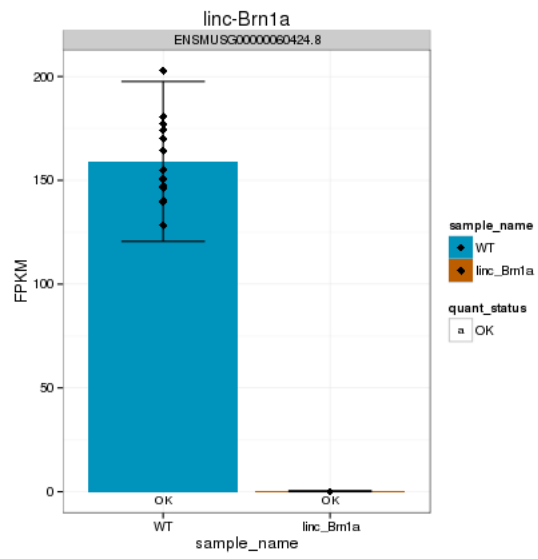
KO assessment
Endogenous lncRNA expression



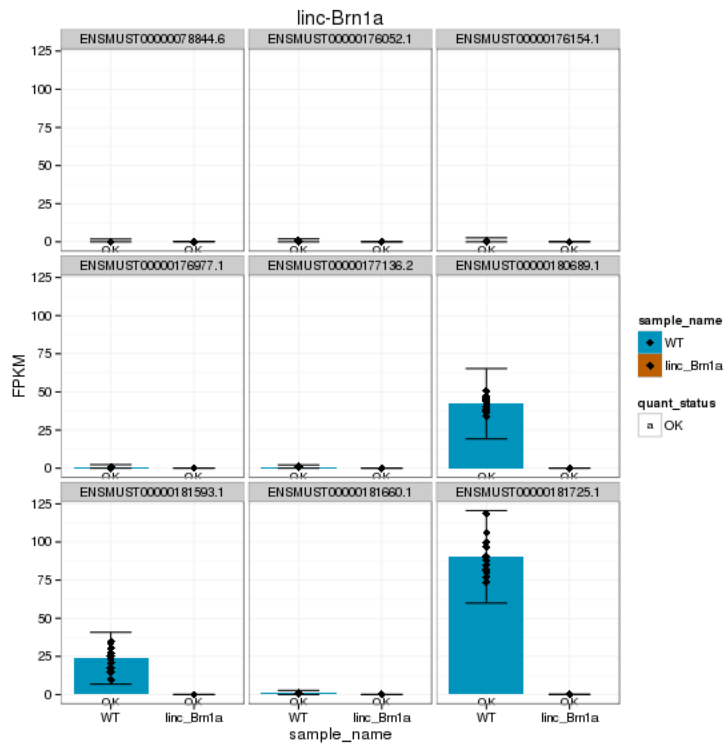
Endogenous expression of linc-Brn1a isoforms:



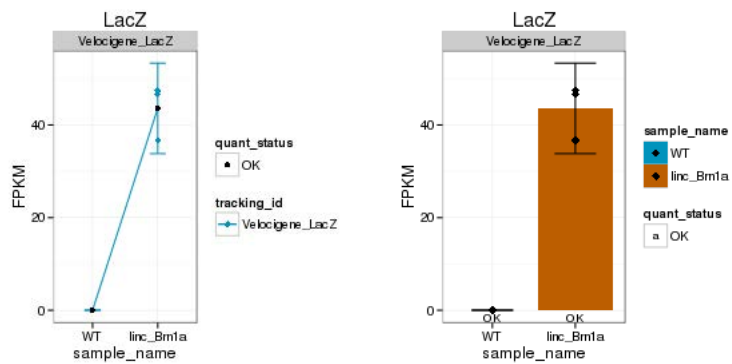
Barplot of gene expression:



Barplot of isoform expression:

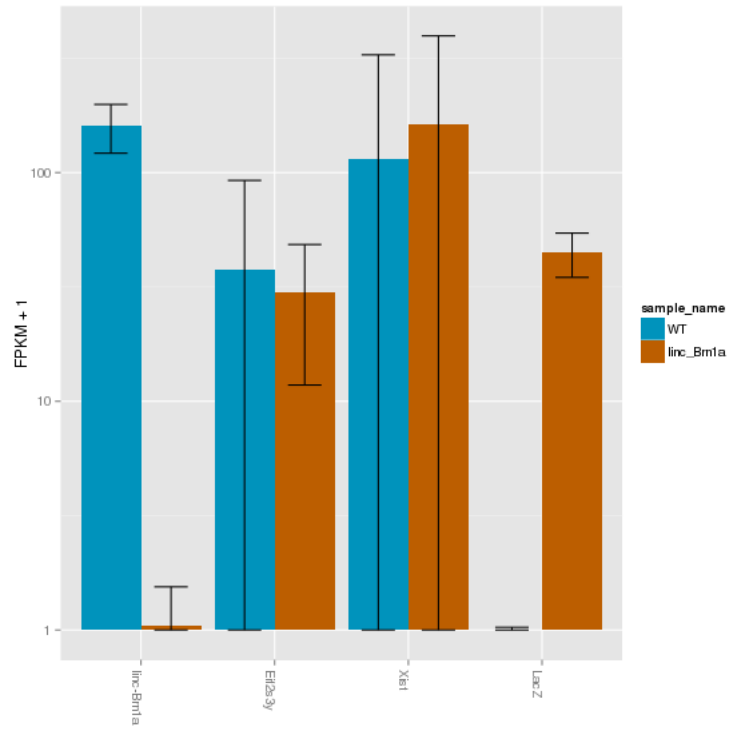


LacZ expression

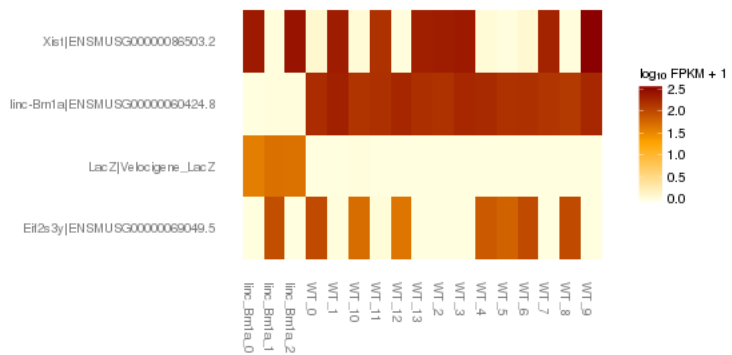


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

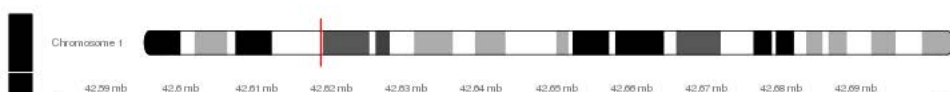
Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



Expression heatmap:



Track visualization



Differential Analysis

Differential Genes

There are 1536 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 H19
2 Nalcn
3 Lhx2
4 Cdh1
5 Ckmt1
6 Sox9
7 Sema4f
8 Hk2
9 Rpa1
10 S100a6
11 Col6a1
12 Pcnt
13 Uhrf1
14 Gabrg1
15 Ckb
16 Sp1
17 Fndc5
18 Ube2c
19 Nkx2-1
20 Col1a1
21 Foxm1
22 Smo
23 Fbxo7
24 Folr1
25 Slc1a5
26 Pnck
27 Spag5
28 Tead3
29 Dbf4
30 Rec8
31 Baz1b
32 Chaf1a
33 Mcm2
34 Kcnn1
35 Ppp1r17
36 Cd36
37 Apoe
```

38 Prkar2b
39 Hmgn2
40 Cdk12
41 Slc2a3
42 Dlgap1
43 Plag1

44 Fcgrt
45 Dll3
46 Phyh1p
47 Ddr1
48 Calb2
49 Snmp200
50 Man1a
51 Hlf
52 Grm3
53 Brinp2
54 Ptrf
55 Dnmt1
56 Cacna1e
57 Kifc2
58 Pax2
59 Eno2
60 Utp20
61 9330159F19Rik
62 Sst
63 Ndrp2
64 Aqp1
65 Anid3b
66 Emr1
67 Chmp2b
68 Pax3
69 Lbr
70 Crabp2

71 Nes
72 Bcan
73 Slc25a18
74 Matk
75 Syt5
76 Prkacb
77 Chd5
78 Wisp1
79 Ndrp1
80 Polr2a

81 Spc25
82 Slc1a6
83 Slc1a3
84 Nid1
85 Mcm5
86 Asf1b
87 Mef2c
88 Pan2
89 Ampd3
90 Pvalb
91 Slc30a4
92 Reep5
93 Otx1
94 Fam131c
95 Vax1
96 Cdc20
97 Neurl1a
98 Epha2

99 Slc4a1
100 Nphs1
101 Pola1
102 Gmnn
103 Cdk4
104 B4galnt1
105 Tph2
106 Klk4
107 Syng3
108 Pole
109 Pcdha2
110 Tinf2
111 Tgfbr1
112 Khgrp
113 Rplp1
114 Fhl2
115 Rbfox1
116 Ewsr1
117 Bcl2l13
118 Fam163b
119 Slc16a12
120 Syn2
121 Nuak2
122 Slc38a3
123 Pifo

124 Raver1
125 Eya4
126 Myt1
127 Lmbr1
128 Odc1
129 Sptbn4
130 Gltf
131 Kif11
132 Phox2b
133 Enkd1
134 Igf2bp1
135 Aldh1a2
136 Cad
137 Dll1
138 Ptgds
139 Slc25a13
140 Ube2i
141 Nolc1
142 Abca1
143 Cd83
144 Hivep2
145 Anp32e
146 Eps8
147 Tnr
148 Rxrg
149 Ncapp
150 Stk32c
151 Celsr1

152 Slc25a5
153 Eef1a2

154 Mapkapk2
155 Phf21b
156 Matn4
157 Il13ra1
158 Rnd3
159 Brca1
160 Vtn
161 Traf4
162 Aldoc
163 Timp2
164 Rarb
165 Cdc6
166 Atad5

167 Zfp334
168 Rhbdl3
169 Birc5
170 Wfdc2
171 Mmp9
172 Slc12a7
173 Mybl2
174 Rac3
175 Erbb3
176 Mfng
177 Pmp22
178 Anxa6
179 Kpna2
180 Mapt
181 Gira2
182 Sparc
183 Crrh1
184 Ikzf1
185 Cbx1
186 Lhx1
187 Sfrp5
188 Sult4a1
189 E2f2
190 Cacng2
191 Scn1b
192 Lhx9
193 Syne1
194 Fbxo5
195 Nr2e1
196 Plagl1
197 Grm1
198 Perp
199 Smpdl3a
200 Fabp7
201 Nts
202 Slc6a15
203 Sim1
204 Dcn
205 Cdk1
206 Actr6
207 Tmpo
208 Kitl
209 Ahi1

210 Nedd1
211 Mtrf2
212 Stx7
213 Moxd1
214 Usp44
215 Ascl1
216 Ptpr
217 Mum1
218 Meis1
219 Tcf3
220 Cobl
221 Grb10
222 E2f7
223 Mdm1
224 Col6a2
225 Glyctk
226 Hmnr
227 Hcn2
228 Slc22a4
229 Rasgef1c
230 Sar1b
231 Nefh
232 Btg2
233 Igfbp3
234 Adcy1
235 Aebp1
236 Prr11
237 Gria1
238 Shmt1
239 Ligl1
240 Pctp
241 Fam49a
242 Fam84a
243 Id2
244 Rrm2
245 Ace
246 Rasl10b
247 Asic2
248 Slc9a3r1
249 Fam64a
250 Cygb
251 Pld2
252 Slc6a4

253 Doc2b
254 Prpf8
255 Aurkb
256 Pfas
257 Top2a
258 Higd1b
259 Pole2

260 Ptpn21
261 Ngb
262 Sptb
263 Zfp3611

264 Efcab11
265 Ccdc88c
266 Fbln5
267 Lgmn
268 Tshz3
269 Rgs6
270 Ppp1r13b
271 Kif26a
272 Gng4
273 Ryr2
274 Amph
275 Gli3
276 Dek
277 Ogn
278 Gadd45g
279 Ror2
280 Habp4
281 Drd1a
282 Adcy2
283 Golm1
284 Trip13
285 Cep72
286 Slc6a3
287 Ocln
288 Arhgef28
289 Iqgap2
290 Elovl7
291 Depdc1b
292 Rasgrf2
293 Nln
294 Rgs7bp
295 Htr1a

296 Hcn1
297 Cdhr1
298 Dnajc9
299 Otx2
300 Slc35f4
301 Rnase4
302 Sh3bp5
303 Arhgef3
304 Gdf10
305 Xpo4
306 Ska3
307 Ints9
308 Cacna2d3
309 Wnt5a
310 Diap3
311 Pbk
312 Esco2
313 Clu

314 Ephx2
315 Stmn4
316 Ebf2
317 Bora
318 Kihl1

319 Fgf17
320 Osmr
321 Haus4
322 Ajuba
323 Psmb5
324 Efs
325 Cpne6
326 Sema5a
327 Cmb1
328 Rai14
329 Baalc
330 Csmc3
331 Cdh10
332 Shcbp1
333 Matn2
334 Kcnc1
335 Atad2
336 Sla
337 Adcy8
338 Gtse1

339 Syng1
340 Dsc1
341 Enpp2
342 Col2a1
343 Shisa9
344 Emp2
345 Cldn1
346 Fgf12
347 Hrasl
348 Apod
349 Ly6h
350 Lynx1
351 Mapk12
352 Cd200
353 Prkdc
354 Mcm4
355 Nde1
356 Mpv17l
357 Boc
358 Sidt1
359 Slc7a4
360 Ncam2
361 Itgb5
362 Ccdc14
363 Adcy5
364 Grik1
365 Chaf1b
366 Clic6
367 Gart
368 Tmem50b
369 Ifngr2
370 Faim2
371 Racgap1
372 Krt18
373 Sncg
374 Rtn
375 Serinc2
376 Kcnk5
377 Dlx2
378 Prph
379 Cdca3
380 Tiam2
381 Thbs2

382 Smoc2
383 Pla2g7
384 Slc5a7
385 Enpp5
386 Pim1
387 Ndc80
388 Crim1
389 Cyp1b1
390 Lhcgr
391 Hn1l
392 Syt4
393 Kifc5b
394 Aqp4
395 Lama3
396 Ndfip1
397 Mal2
398 Hbegf
399 Ppp2r2b
400 Rab27b
401 Gnal
402 Lox
403 Cep192
404 Slc14a2
405 Lmnb1
406 Megf10
407 Slc22a6
408 Anxa1
409 Incenp
410 Fads3
411 Fen1
412 Tmem2
413 Stambpl1
414 Cdca5
415 Kif20b
416 Htr7
417 Glc
418 Pola2
419 Eif1ad
420 Apba1
421 Slc1a1
422 Rcor2
423 Cep55
424 Rbp4

425 Hells
426 Aldh18a1
427 Lcor
428 Maa

429 Pdc11
430 Nhrc2
431 Afap1l2
432 Ablm1
433 Hspa12a
434 Bhlhe22
435 Arhgap19
436 Lbx1
437 Kcnp2
438 Limd1
439 Itga7
440 Cdk2
441 Cdh9
442 Aatk
443 Agap2
444 Caly
445 Ifitm3
446 Lrdd
447 Dock9
448 Tk1
449 Cbx2
450 Klhl4
451 Sdc3
452 Tys
453 Plk4
454 Gdap1
455 Itih5
456 Clec3b
457 Ccr1
458 Nrp1
459 Prkar1b
460 Snca
461 Gria4
462 Paqr8
463 Slco5a1
464 Slc40a1
465 Ica11
466 Sgol2
467 Col3a1

468 1500015O10Rik
469 2010300C02Rik
470 Tmeff2
471 Sphkap
472 Bard1
473 2810459M11Rik
474 Nppc
475 Serpine2
476 Lypd1
477 Mcm6
478 Cfh
479 Epb4.1l5
480 Dbi
481 Cd55
482 Tnni1
483 Slc45a3

484 Pik3c2b
485 Ppfia4
486 Lamc2
487 Parp1
488 Cnih3
489 Rgs7
490 Uck2
491 Atp1b1
492 Kifap3
493 Cenpf
494 Nek2
495 Mcm10
496 Optn
497 Ddr2
498 Hsd17b7
499 Rgs5
500 Nuf2
501 Prdx6
502 Mrc1
503 Rabgap1l
504 Nr5a1
505 Prkcq
506 Mastl
507 Stxbp1
508 Traf1
509 Grb14
510 Slc4a10

511 Notch1
512 Sapcd2
513 Grin1
514 Hnmt
515 Cybrd1
516 Lrp2
517 Slc43a1
518 Kif18a
519 Pax6
520 Dut
521 Mdk
522 Wdr76
523 Lrp4
524 Lamp5
525 Snap25
526 Nusap1
527 Gfra4
528 Rad51
529 Casc5

530 Knstrn
531 Pcn
532 Bub1
533 Pdyn
534 Nkx2-2
535 Acss1
536 Gins1
537 Tpx2
538 E2f1

539 Aurka
540 Pkia
541 Rbm38
542 Snx16
543 Atp9a
544 Sall4
545 Col9a3
546 Map1lc3a
547 Mmp24
548 Dsn1
549 Rbl1
550 Tti1
551 Fam83d
552 Gnb4
553 Ect2

554 Anxa5
555 Ccna2
556 Slc7a11
557 Postn
558 Tm4sf1
559 Shox2
560 Hsd3b6
561 Hmgcs2
562 Sypl2
563 Slc6a17
564 Kcnc4
565 Neurog2
566 Gucy1b3

567 Glib
568 Enpep
569 Cks1b
570 Pmf1
571 Iqgap3
572 Chd1l
573 Them4
574 Cdh17
575 Smc2
576 Anp32b
577 Nr4a3
578 Brinp1
579 Tnc
580 Pappa
581 Bspry
582 Mpdz
583 Tal2

584 Kif24
585 Cntfr
586 Sh3gl2
587 Prkaa2
588 Pde4b
589 Dnajc6
590 Jak1
591 Usp1
592 Dmrtb1
593 Col9a2

594 Tfp2c
595 Slc2a1
596 Ephb2

597 Kif2c
598 Plk3
599 Nasp
600 Rad54l
601 Stil
602 Pax7
603 HpcA
604 Rspo1
605 Cdca8
606 Rpa2
607 Rcc1
608 Epb4.1
609 Abcf2
610 Prom1
611 Kcnip4
612 Gpr125
613 Ablim2
614 Ppp2r2c
615 Stk32b
616 Nsg1
617 Rnf32
618 Fosl2
619 Cgref1
620 Cenpa
621 Rfc1
622 Pds5a
623 Gabra4
624 Gabrb1
625 Tec
626 Pdgfra
627 Tgfbr3
628 Spp1
629 Sparcl1
630 Hsd17b11
631 Nos1
632 Dck
633 Pf4
634 Shroom3
635 Cdkl2
636 Abcb9
637 Kntc1
638 Rimbp2
639 Uncx

640 Ung
641 Lhx5
642 Rph3a
643 Col1a2
644 Fam3c
645 Eln
646 Ezh2
647 Fezf1
648 Asns

649 Dync1i1
650 Akr1b8
651 Igf2bp3
652 Npy
653 Ptn
654 Slc13a4
655 Kel
656 Dbpht2
657 Mad2l1
658 Tgfa
659 Fbxl14
660 Cntn3
661 Chl1
662 Aldh1l1
663 Cntn6
664 Grip2
665 Slc6a13
666 Slc6a12
667 Ret
668 A2m
669 Rasgef1a
670 Dusp16
671 Emp1
672 Plbd1
673 Mgp
674 Slco1c1
675 Rad18
676 Tmtc1
677 Slc6a11
678 Cand2
679 Mbd4
680 Pianp
681 Cd9
682 Rad51ap1

683 Tspan11
684 U2af2
685 Dbx1
686 Blm
687 Hddc3
688 Idh2
689 Mfge8
690 Ndufc2
691 Ipo5

692 Kif22
693 Nupr1
694 Syt3
695 Dkk3
696 Lyve1
697 Tead2
698 Plk1
699 Rrm1
700 Mki67
701 Wee1
702 Mrgprf
703 Fgf15

704 Ano1
705 Fgf13
706 Syp
707 Slc38a5
708 Msn
709 Efnb1
710 Magee1
711 Itm2a
712 2610002M06Rik
713 Phka2
714 Gabra3
715 Ap1s2
716 Zfp92
717 Atp2b3
718 Tceal6
719 Rnf128
720 Gas6
721 Chma6
722 Gpm6a
723 Tmem66
724 Plat
725 Sfrp1

726 Scrg1
727 Sall1
728 Itfg1
729 Irx3
730 Irx5
731 Il34
732 Cenpn
733 Mt3
734 Cntnap4
735 Gpr56
736 Rab3a
737 Cdh13
738 BC021891
739 Bean1
740 Hsd11b2
741 Jam3
742 Pdgfd
743 Thy1
744 Kirrel3
745 Cryab

746 Mpzl2
747 Robo3
748 Olfm2
749 Ldlr
750 Ccnb2
751 Anxa2
752 Kif23
753 Pkm
754 1700017B05Rik
755 Nptn
756 Mlip
757 Car12
758 Igdcc3

759 Tipin
760 Zwlch
761 Tfdp2
762 Clstn2
763 Cdc25a
764 Cspg5
765 DclK3
766 Scn5a
767 Slco2a1
768 Topbp1

769 Thsd7a
770 Colgalt2
771 Ablim3
772 Inpp1
773 Chrm1
774 Troap
775 Pdxk
776 Ccdc85a
777 Rims3
778 Itgbl1
779 Camkv
780 Nup93
781 Rasgrp2
782 Sox10
783 Cdo1
784 C330027C09Rik
785 Cntnap3
786 Gas7
787 Mgl1
788 S100b
789 Ttf2
790 Wnt6
791 Ttll4
792 Galnt9
793 4833424O15Rik
794 Snap91
795 Adamts15
796 Prss35
797 Cdc14a
798 Bai3
799 Ndn

800 Rfwd3
801 Cplx1
802 Gabrb3
803 Adra2a
804 Emx1
805 Fndc3c1
806 Recql4
807 D930015E06Rik
808 Map9
809 Gucy1a3
810 Zswim5
811 Aspm

812 9430020K01Rik

813 Gria2

814 Kcnk1

815 Fancd2

816 Rpap1

817 Fstl5

818 Polq

819 Rhov

820 Slc25a37

821 Tmem132d

822 Kif4

823 Cbl

824 Smc4

825 Ramp1

826 Barhl2

827 Gbx2

828 Slc41a2

829 Pbx2

830 Tdg

831 Fras1

832 Mfrp

833 Pcdh11x

834 BC030867

835 Nrip3

836 Larp6

837 Plvap

838 Sncb

839 Ncaph

840 Htr2a

841 Dpp4

842 Ncapd3

843 Brinp3

844 Lars2

845 Rims4

846 Arx

847 Mid1

848 Lzts2

849 Tmem98

850 Fign1

851 Pcdh17

852 Lrln5

853 Zswim4

854 Melk

855 Mic1
856 Ddx11
857 Ids
858 Syt1
859 Tmem59l
860 Colec12
861 Prickle1
862 Sostdc1
863 Ska1
864 Lrm3
865 Cep135
866 Npy1r
867 March1
868 Greb1

869 Fxyd1
870 Fxyd7
871 Aaas
872 Zcchc12
873 Tubb4b
874 Dner
875 Kif15
876 Anln
877 Dpp10
878 Plch1
879 Wnt4
880 Dna2
881 C1qa
882 C1qc
883 Neto2
884 Fzd8
885 Slc39a12
886 Wnt8b
887 Spryd3
888 Hk1
889 Sstr4
890 1110059M19Rik
891 Mgarp
892 Ppp1r14a
893 Pacrg
894 Islr
895 Clic4
896 Itih2
897 Tacc3

898 Slc9a7
899 Paqr7
900 Nov
901 Endod1
902 Vgf
903 4930427A07Rik
904 Dtl
905 Dlgap5
906 Wdhd1
907 Cldn11
908 Ckap2
909 Phyhipl
910 Ppp1r16b
911 Slc16a9
912 Ctnna1
913 Neurod6
914 Rmi2
915 Dhx38
916 Igsf9
917 Cramp1l
918 Haus6
919 2410066E13Rik
920 Kcna6
921 AW551984
922 Cdon
923 Camk4

924 Notch3
925 Cep250
926 Ncapd2
927 Glra3
928 Fau
929 Satb2
930 Ttk
931 Sv2a
932 Rgs4
933 Akap12
934 Pold1
935 Hoxb4
936 Trhr
937 Ephx1
938 Prc1
939 Tspyl5
940 Bmp6

941 Cpq
942 Myo16
943 Anpep
944 En2
945 Htr5a
946 F13a1
947 Gpr126
948 Prune2
949 Camk1d
950 Akna
951 Tekt5
952 Rbpj
953 Suv39h1
954 AF529169
955 Rnf122
956 March4
957 Neil3
958 Tspy14
959 Cntn5
960 Mmd2
961 Grin3a
962 Tmem246
963 Strip2
964 Kcne2
965 Ncoa7
966 Ldb2
967 Raly1
968 Slc6a5
969 Exo1
970 Cpeb2
971 Gabbr2
972 Xkr5
973 Arhgap29
974 Fam60a
975 Timeless
976 Baz2a
977 Bub1b
978 Gpr176

979 Abcc8
980 Kcns1
981 2810417H13Rik
982 Sema3d
983 Dnm3

984 Hey1
985 Cacng5
986 Cdh18
987 Cdh12
988 Prdm13
989 Necab1
990 Pitpnm3
991 C3ar1
992 Abcb1a
993 Ildr2
994 Erc2
995 Ppp1r14c
996 Rad54l2
997 St8sia4
998 Il17rd
999 Hnmpul1
1000 Six5
1001 Reps2
1002 Atp1a3
1003 Tet2
1004 Rcc2
1005 Slc22a2
1006 Npnt
1007 Nrg3
1008 Fam117b
1009 Pif1
1010 Grid1
1011 Nbl1
1012 H2afv
1013 Dnah7b
1014 Brca2
1015 Arhgap11a
1016 Chd7
1017 Rbbp8
1018 Car8
1019 Nkx6-2
1020 Inhba
1021 Atp1b2
1022 Wrap53
1023 Rap1gap
1024 4930506M07Rik
1025 Cldn5
1026 Dicer1

1027 Pik3r1
1028 Ccnb1
1029 Gpr12
1030 Kif14
1031 Ago1
1032 Fmod
1033 Nlrp5-ps

1034 Cdc42ep4
1035 Rims1
1036 Kcnj2
1037 Mpped1
1038 Trpc5
1039 Kirrel
1040 Plekha6
1041 Myrip
1042 Abca9
1043 Ptpre
1044 Mcm3
1045 Ipo9
1046 S100a10
1047 Spidr
1048 Elmod1
1049 Foxn4
1050 Ncapg2
1051 Tbca
1052 Bend6
1053 Slc35f2
1054 Specc1
1055 Grap2
1056 Dmrt3
1057 Adora1
1058 Clspn
1059 Kcnj12
1060 Golga7b
1061 Jhdm1d
1062 Mrap2
1063 Hebp1
1064 Smg7
1065 Muc1
1066 Egflam
1067 Onecut1
1068 Smco3
1069 B3galnt1

1070 Kcnj6
1071 Tmem130
1072 E130012A19Rik
1073 Eifn2
1074 Acpl2
1075 Adamts3
1076 Diras1
1077 Dmrt1
1078 1700048O20Rik
1079 Cim1
1080 Fam19a2
1081 Cdc25c
1082 Snhg11
1083 Shisa2
1084 Fam46c
1085 Rps2
1086 Cnrip1
1087 D3Bwg0562e
1088 Fzd1

1089 Cnpy1
1090 Ccnjl
1091 Kcnj10
1092 Fads6
1093 Syt16
1094 Napepld
1095 Ankrd34b
1096 Prkce
1097 Pcdhb7
1098 Lingo2
1099 Sox3
1100 Shroom2
1101 Lrrc3b
1102 Cenph
1103 Lhfpl2
1104 Cenpe
1105 Gprin3
1106 Bhlhe23
1107 Penk
1108 Frrs1l
1109 Olig3
1110 Dbx2
1111 Chrm2
1112 Zfp629

1113 Vwc2l
1114 Plekha7
1115 Cdc42ep2
1116 Mms22l
1117 C2cd4c
1118 Sdpr
1119 Onecut2
1120 Hpcal4
1121 Chrm3
1122 Olig1
1123 Nxph1

1124 E2f8
1125 Gsg1l
1126 Iqub
1127 Hepacam
1128 Ankle1
1129 Hs3st2
1130 Gjb2
1131 Camk2n1
1132 C77370
1133 Ticrr
1134 Hrk
1135 Olfm12a
1136 Hmga1
1137 Nxph3
1138 Ppp1r3b
1139 Fam211b
1140 Vat1l
1141 Fbl
1142 Spsb4
1143 Zfp41

1144 Fbxo41
1145 Dmrt2
1146 Helt
1147 C2cd3
1148 Mc4r
1149 Gap43
1150 Mettl21c
1151 Fam180a
1152 Rnf152
1153 Mis18bp1
1154 Cltb
1155 Fgfbp3

1156 Lix1
1157 Cdca4
1158 Diras2
1159 Tshz2
1160 Hes5
1161 Ldlrad3
1162 Pirt
1163 Tmem72
1164 Rbm15
1165 Col6a3
1166 Dmrt2
1167 Nwd1
1168 Gpr85
1169 Slitrk3
1170 Ckap2l
1171 Pou4f1
1172 Arxes1
1173 Fgfbp1
1174 Gli2
1175 Nhlh2
1176 Sp8
1177 Igf2
1178 Myof
1179 Rhno1
1180 Rpl29
1181 Lonrf2
1182 Vstm2a
1183 Neurog1
1184 Cdca2
1185 Atp13a5
1186 Nrsn1
1187 Rapgef4
1188 Oxtr
1189 Frmd3
1190 Frmpd4
1191 Lrp1b
1192 Kcnk3
1193 Scn3b
1194 Zfp219
1195 Rps23
1196 Lingo1
1197 Grm5
1198 Crh

1199 Armcx4
1200 Fam26e
1201 2610318N02Rik
1202 H2afx
1203 Ankrd55
1204 Hic2
1205 Fzd2
1206 Neto1
1207 Lgals3
1208 Tcf19
1209 Pcdh20
1210 Adra1b
1211 Trhde
1212 Vstm4
1213 Emilin3
1214 Scg2
1215 Sgms2
1216 Gja1
1217 Kcns2
1218 Lin28a
1219 Olfm1
1220 P4ha3
1221 Lingo3
1222 Mblac2
1223 Sv2c
1224 Ercc6l
1225 Gen1
1226 Msrb3
1227 Nhlh1
1228 4930422G04Rik
1229 Gfod1
1230 Ncald
1231 Plagl2
1232 Kcnj16
1233 Arhgef39
1234 Map3k19
1235 Mest
1236 Sox6
1237 Xkr4
1238 Slc6a7
1239 Zfp217
1240 Sema5b
1241 Hbb-y

1242 Hbb-bh1
1243 Gpr17
1244 Hbb-bs
1245 Dlg2
1246 Lrrtm4
1247 Jun
1248 Reep1
1249 Dnah6
1250 Prkcb
1251 Gas1
1252 Ube2ql1
1253 Sv2b

1254 Yap1
1255 E330013P04Rik
1256 Rnf26
1257 Gsx1
1258 Mapk11
1259 Ptprt
1260 Arhgap20
1261 Aldh1a1
1262 Al854703
1263 Phgdh
1264 Kdm3a
1265 Kcnp1
1266 Plxnb1
1267 Exoc6
1268 Ppfia2
1269 Cnn3
1270 6330403A02Rik
1271 Tmem179
1272 Nt5dc3
1273 Skp2
1274 Spock3
1275 4922502B01Rik
1276 Fgfr3
1277 Msi1
1278 Cadps
1279 Sytl5
1280 Vsnl1
1281 Slc8a1
1282 Hmgb2
1283 6330419J24Rik
1284 Tmem158

1285 Nyap2
1286 Lrtm2
1287 Cntn1
1288 Gabra5
1289 Ntrk2
1290 Nell1
1291 Maf
1292 Alk
1293 Epha6
1294 Hba-x
1295 Cdca7
1296 Gpc3
1297 Nap1l3
1298 Abcd2
1299 Car10
1300 Col8a2
1301 Synpr
1302 Lig1
1303 Adap1
1304 Pign
1305 Mpz
1306 Trnp1
1307 Fry
1308 Grm7

1309 Hmga2
1310 Pou3f4
1311 Jakmip3
1312 Hist3h2ba
1313 Magel2
1314 Slc35f3
1315 Cep110
1316 4922502B01Rik
1317 Rit2
1318 Gnai1
1319 Ldoc1
1320 A830018L16Rik
1321 Lepr
1322 Camk2b
1323 Palld
1324 Gm10036
1325 Gstm1
1326 Tspan7
1327 Espl1

1328 Spock2
1329 Nrf1
1330 Znf2
1331 Gpc6
1332 Anks1b
1333 Gda
1334 Capn11
1335 Samd12
1336 Tubb2a
1337 Zfp516
1338 Pde1a
1339 Skap2

1340 Fam19a1
1341 Il1rap12
1342 Hopx
1343 Fcgr3
1344 Trp53
1345 Rcan3
1346 Zfp422
1347 Ntm
1348 Alox5ap
1349 Cend1
1350 Sct2
1351 Caln1
1352 Rnf14
1353 Dcc
1354 Hist1h4i
1355 Kcnd2
1356 Acyp2
1357 Csmc1
1358 Slc4a4
1359 Cdh3
1360 Gm10093
1361 Maml3
1362 Fiz1
1363 Acvr2b

1364 Hipk2
1365 Snrpa
1366 Sox21
1367 Pclo
1368 Mdc1
1369 Tmem91
1370 Ppp1r1b

1371 Tac1
1372 Ttr
1373 Suclg2
1374 Galnt16
1375 Myt1l
1376 Unc13c
1377 Tgif2
1378 Cks2
1379 Opcml
1380 Trank1
1381 Otof
1382 Dgkk
1383 Nsl1
1384 Sim2
1385 Mtap
1386 Kcnma1
1387 Tmem117
1388 Luzp2
1389 Cyp26b1
1390 Sema3e
1391 Jazf1
1392 Unc5d
1393 Sox11
1394 Jakmip1
1395 Crb1
1396 Slc22a8
1397 Alms1
1398 Slc24a3
1399 Gm7964
1400 Fbln2
1401 Ifi27
1402 Cntn4
1403 Clasp1
1404 Scn1a
1405 Sepp1
1406 Dhrs3
1407 Phactr4
1408 Knkc1
1409 Numa1
1410 Lefty2
1411 Psg16
1412 Gm10184
1413 Cntnap5b

1414 Hes6
1415 Xpo5
1416 Lgi1
1417 Foxd3
1418 Ppp1r3c

1419 Cbln4
1420 Pcnaps2
1421 Cenpm
1422 Ninl
1423 Col8a1
1424 Gm608
1425 Slc4a5
1426 Cep152
1427 Gpr88
1428 Trp53i11
1429 Celsr2
1430 Psrc1
1431 Col28a1
1432 Ddx3y
1433 Slc7a14
1434 Nxph2
1435 Prdm6
1436 Lyz2
1437 Nkain2
1438 Sifn9
1439 Hba-a2
1440 Ccnd1
1441 Fam217b
1442 Tmem132b
1443 Peg12
1444 Cntnap5a
1445 Gm10282
1446 Zfp804a
1447 Jund

1448 Syndig1l
1449 2410004P03Rik
1450 Rpl23
1451 Grid2
1452 Ccnf
1453 Fam84b
1454 Gm7292
1455 Sod3
1456 Drc1

1457 Arhgdig
1458 Cep76
1459 Wdfy1
1460 5730409E04Rik
1461 4632427E13Rik
1462 Snmp40
1463 Spc24
1464 Mafb
1465 Zcchc3
1466 Thbd
1467 Gas2l3
1468 Ctxn2
1469 Pak6
1470 Ptar1
1471 Fibin
1472 Heg1
1473 Pde11a

1474 Scn2a1
1475 Selm
1476 2700094K13Rik
1477 Gm16485
1478 Gm12184
1479 Nrap
1480 Fam43b
1481 Al593442
1482 Aunip
1483 Hs3st4
1484 Rad54b
1485 Slc8a3
1486 Gm14446
1487 Kifc1
1488 Clec2l
1489 Pprt4
1490 Fzd10
1491 Nap1l2
1492 Ccdc85c
1493 Gm12371
1494 Gm14342
1495 D030055H07Rik
1496 9130020K20Rik
1497 Mia
1498 Ier5l
1499 Gm16586

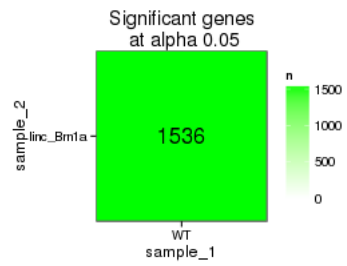
1500 3110047P20Rik
1501 Pcp4

1502 Mndal
1503 Cdr1
1504 Gm340
1505 Hspa1b
1506 Tcerg1l
1507 Smim13
1508 D17H6S56E-5
1509 Gm19345
1510 Six3os1
1511 Gm9866
1512 Pou3f2
1513 Fignl2
1514 Sox1
1515 Kcnj11
1516 H1f0
1517 Fam181a
1518 Galntl6
1519 Gm26872
1520 Al854517
1521 Gm3764
1522 1700037F24Rik
1523 A930011O12Rik
1524 Gm17388
1525 Miat
1526 Gm5101
1527 Gm26809
1528 2810011L19Rik

1529 Gm26981
1530 Gm27032
1531 Gm17750
1532 Gm27031
1533 Pclo
1534 RP23-45G16.5
1535 KCTD12
1536 RP24-312B12.1

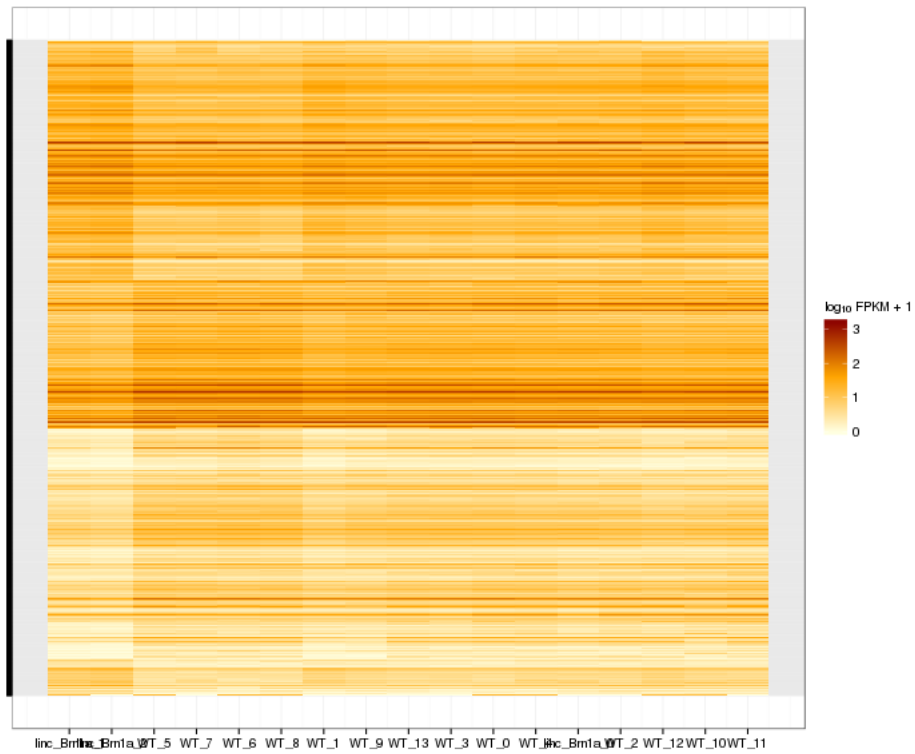
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

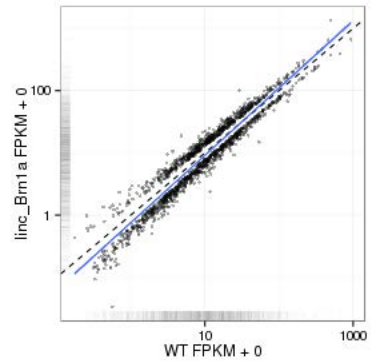


Significant genes with expression >50fpkm (any condition):(turned off)

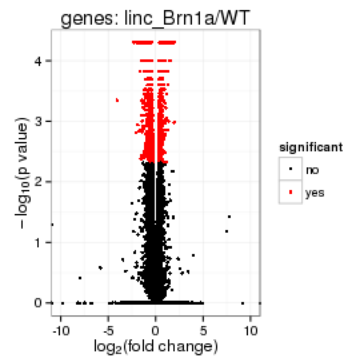
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

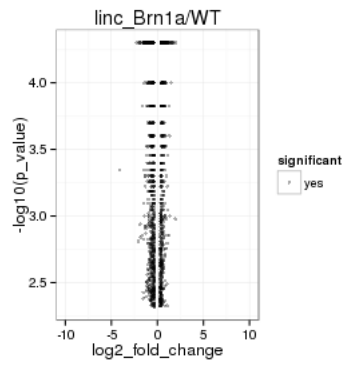
Scatter plot of significant genes only:



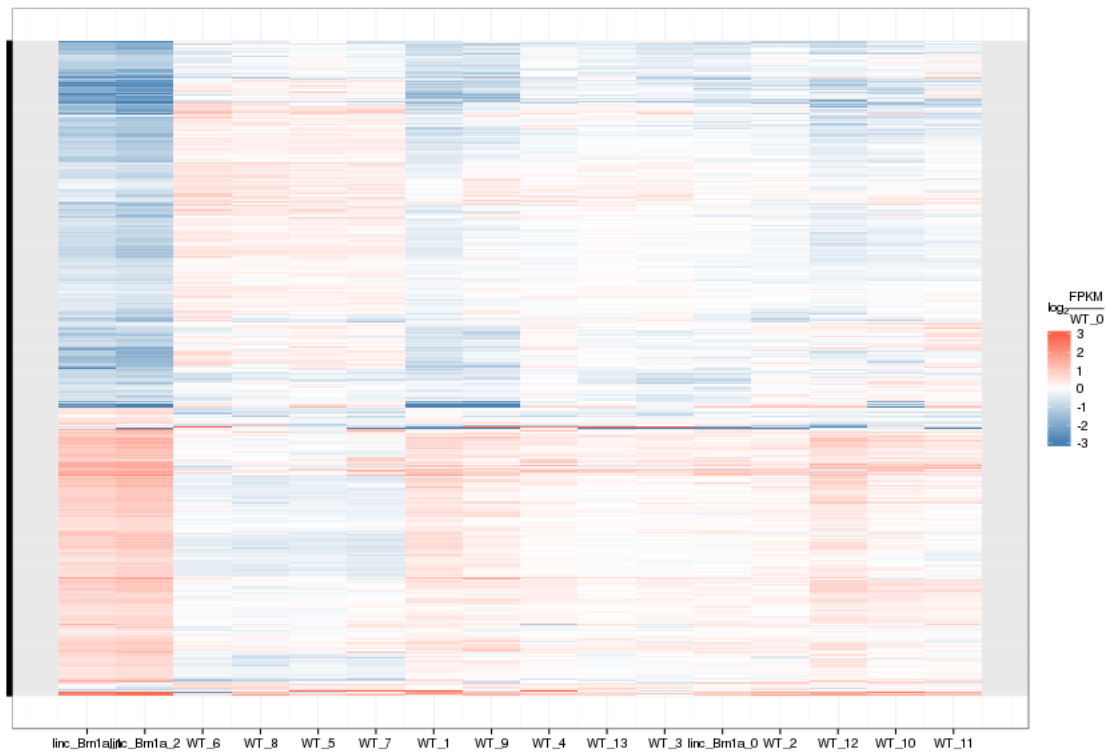
Volcano Plot



Volcano plot with significant genes only:



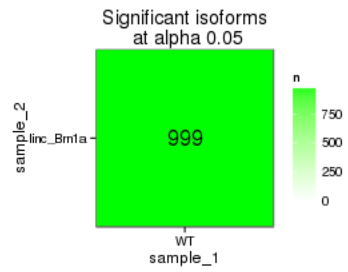
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

isoAnnot	gene_short_name
1	H19
2	Nalcn
3	Lhx2
4	Cdh1
5	Ckmt1
6	C1d
7	Hk2
8	Rpa1
9	Snrpn
10	S100a6
11	Col6a1
12	Pcnt
13	Uhrf1
14	Gabrg1
15	Ckb
16	Ube2c
17	Col1a1
18	Foxm1
19	Smo
20	Nfix
21	Gria3
22	Spag5
23	Ccne1
24	Rec8
25	Grin2d
26	Mcm2

27 Kcnn1
28 Apoe
29 Hmgn2
30 Slc2a3
31 Dlgap1
32 Fcgrt
33 Dll3
34 Calb2
35 Snmp200
36 Man1a

37 Hlf
38 Grm3
39 Ptrf
40 Cacna1e
41 Kifc2
42 Eno2
43 9330159F19Rik
44 Sst
45 Ndr2
46 Aqp1
47 Chmp2b
48 Lbr
49 Crabp2
50 Nes
51 Bcan
52 Slc25a18
53 Syt5
54 Ndr1
55 Polr2a
56 Spc25
57 Slc1a6
58 Slc1a3
59 Nid1
60 Mcm5
61 Asf1b
62 Mef2c
63 Ampd3

64 Reep5

65 Otx1
66 Fam131c
67 Cdc20
68 Slc4a1

69 Nphs1
70 Pola1
71 Gmnn
72 Tph2
73 Syngn3
74 Pole
75 Stx1a
76 Tinf2
77 Khsrp
78 Fhl2
79 Rbfox1
80 Fam163b
81 Slc16a12
82 Syn2
83 Myt1
84 Gabra1
85 Odc1
86 Kif11
87 Phox2b
88 Igf2bp1
89 Aldh1a2
90 Cad
91 Dll1
92 Slc25a13
93 Abca1
94 Cd83
95 Hivep2
96 Rxrg
97 Celsr1
98 Eef1a2
99 E2f3
100 Mapkapk2
101 Brca1
102 Vtn
103 Timp2
104 Rarb
105 Cdc6
106 Zfp334
107 Rhbdl3
108 Birc5
109 Wfdc2
110 Mmp9
111 Slc12a5
112 Slc12a7

113 Mybl2
114 Rac3
115 Erbb3
116 Mfng
117 Pmp22
118 Kpna2
119 Mapt

120 G3bp1
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122 Cchr1
123 Lhx1
124 Abca5
125 Sfrp5
126 Sult4a1
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129 Chtf18
130 Syne1
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133 Nr2e1
134 Plagl1
135 Grm1
136 Perp
137 Fabp7
138 Nts
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142 Cdk1
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158 Btg2
159 Igfbp3
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161 Aebp1
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163 Gria1
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165 Fam84a
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167 Rrm2
168 Ace
169 Fam64a
170 Pld2
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172 Aurkb
173 Pfas
174 Top2a

175 Sptb
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184 Gli3
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187 Adcy2
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193 Nln
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197 Sh3bp5
198 Arhgef3

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202 Pbk
203 Esco2
204 Clu
205 Bora
206 Fgf17
207 Haus4
208 Ajuba
209 Efs
210 Sema5a
211 Cmb1
212 Rai14
213 Cdh10
214 Shcbp1
215 Kcnv1
216 Atad2
217 Adcy8
218 Gtse1
219 Syng1
220 Enpp2
221 Shisa9
222 Emp2
223 Cldn1
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225 Apod

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229 Alcam

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250 Prph
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253 Thbs2
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291 Bhlhe22
292 Arhgap19
293 Lbx1
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295 Cdh9
296 Cbx2
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323 Vangl2
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328 Ddr2

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332 Prkcq
333 Mastl

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339 Cybrd1

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427 Akr1b8
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626 Neil3
627 Tspyl4
628 Cntn5
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870 Znf2
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872 Capn11
873 Tubb2a
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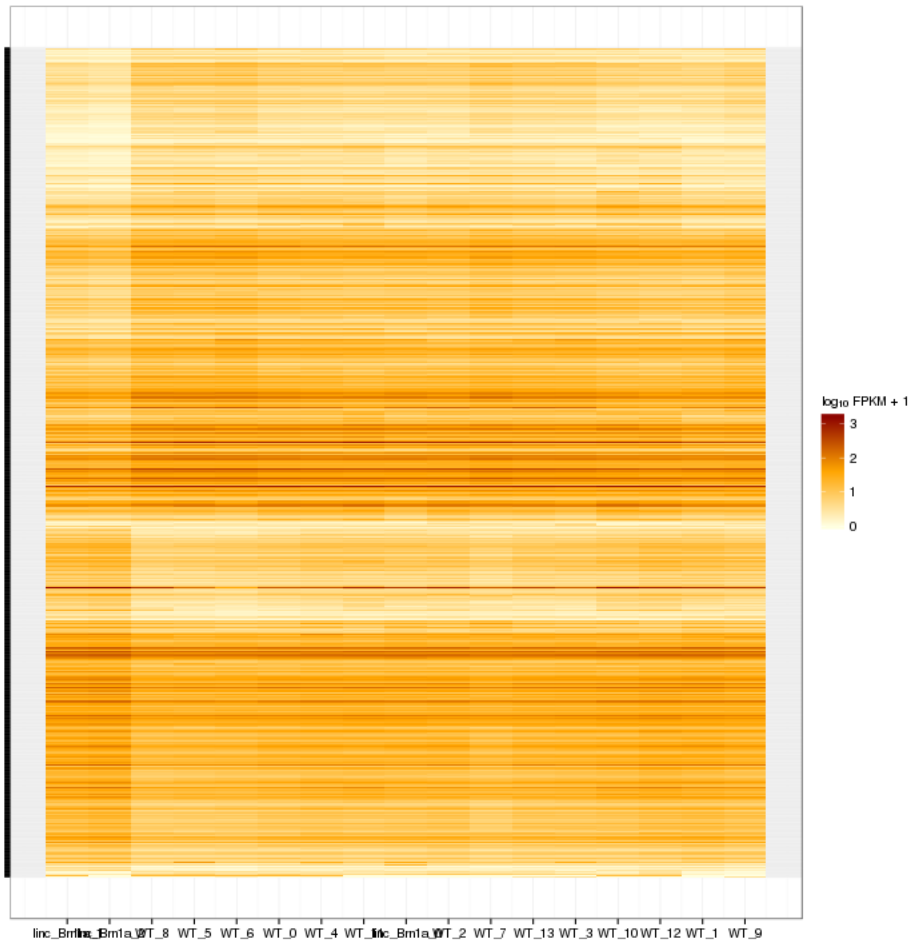
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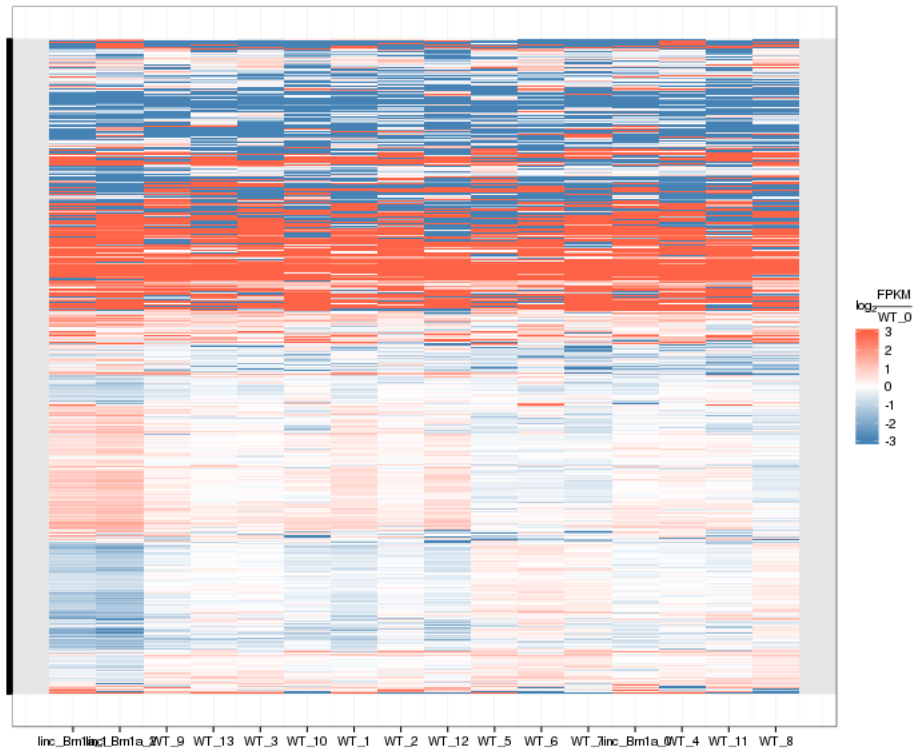
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954 Pak6
955 Fbin
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958 Fam43b
959 AI593442
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966 Gm14342
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983 Miat
984 Gm5101
985 Gm26981
986 Gm17750
987 Gm27031
988 Pclo
989 KCTD12
990 RP24-312B12.1

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

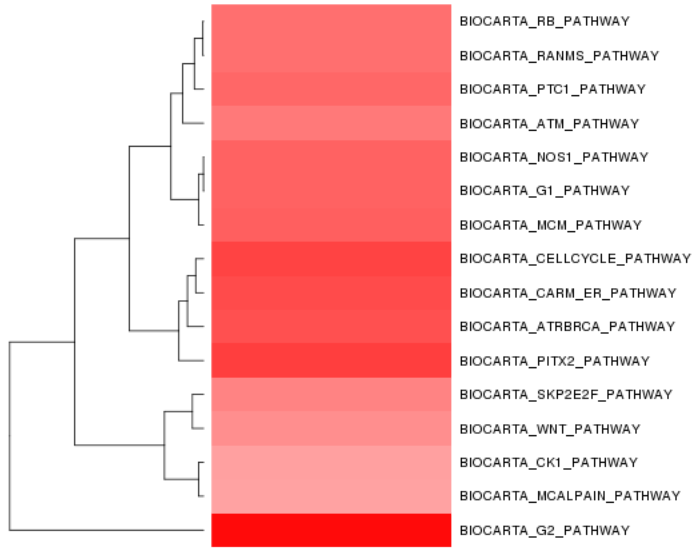
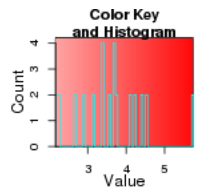
Gene/Pathway Analysis

GSEA

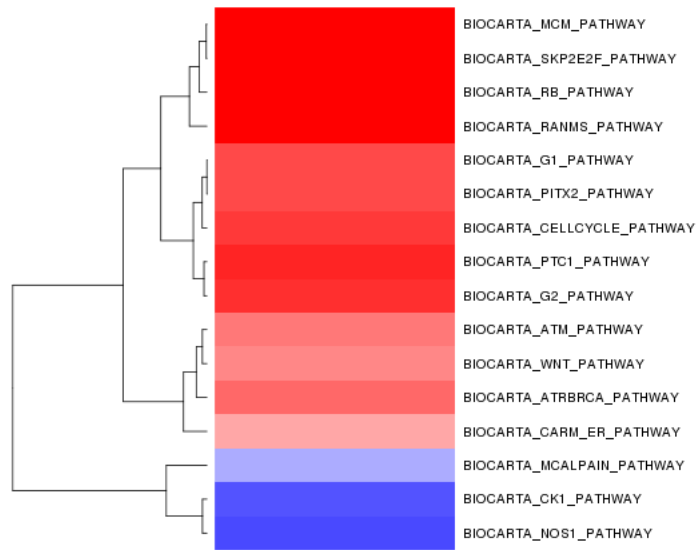
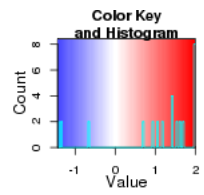
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

Biocarta enrichment:

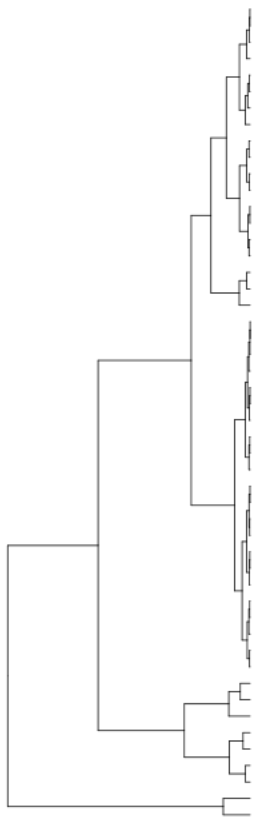


Biocarta zscore:



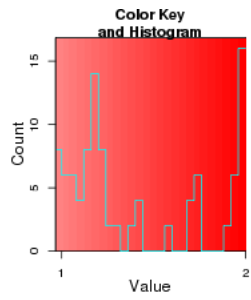
Reactome enrichment:

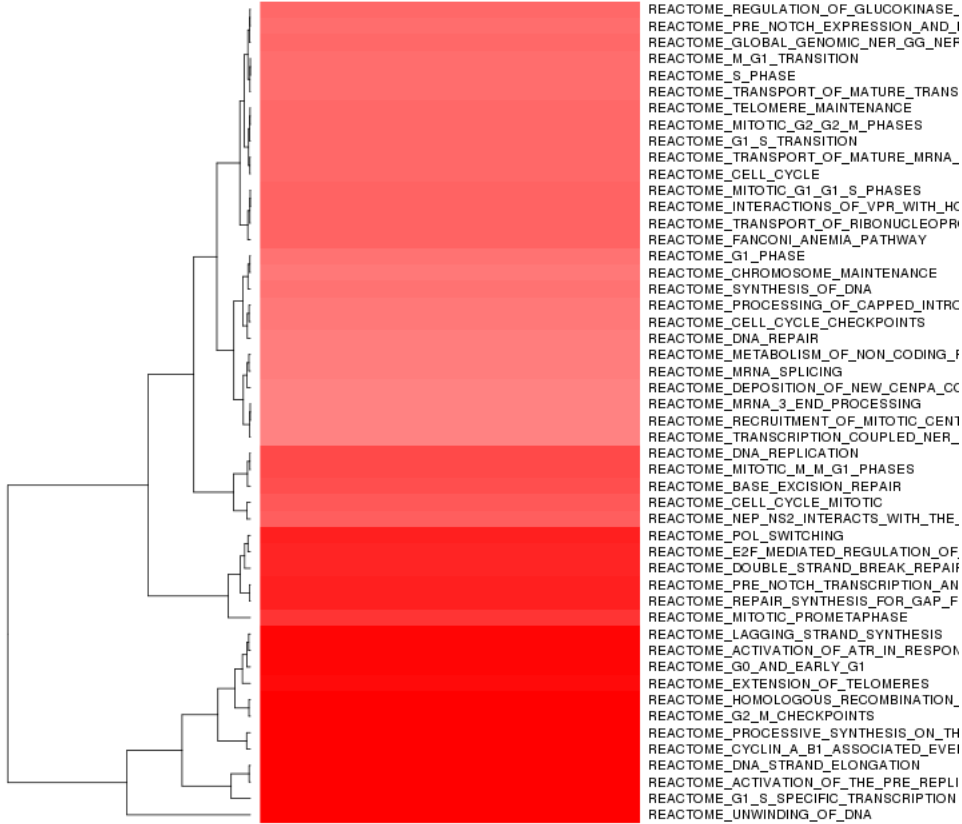
Error: no locations are finite



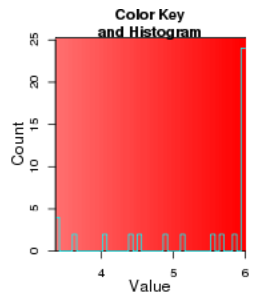
REACTOME_CHROMOSOME_MAINTENANCE
REACTOME_SIGNALING_BY_GPCR
REACTOME_GPCR_DOWNSTREAM_SIGNALING
REACTOME_TRANSCRIPTION
REACTOME_METABOLISM_OF_MRNA
REACTOME_TRANSPORT_OF_MATURE_TRANS
REACTOME_TRANSMISSION_ACROSS_CHEMIC
REACTOME_HIV_LIFE_CYCLE
REACTOME_LATE_PHASE_OF_HIV_LIFE_CYCL
REACTOME_S_PHASE
REACTOME_METABOLISM_OF_NON_CODING_F
REACTOME_G2_M_CHECKPOINTS
REACTOME_DNA_STRAND_ELONGATION
REACTOME_CELL_CYCLE_CHECKPOINTS
REACTOME_RNA_POL_I_TRANSCRIPTION
REACTOME_G1_S_TRANSITION
REACTOME_DNA_REPAIR
REACTOME_MITOTIC_G1_G1_S_PHASES
REACTOME_MITOTIC_PROMETAPHASE
REACTOME_TRANSPORT_OF_MATURE_MRNA
REACTOME_INFLUENZA_LIFE_CYCLE
REACTOME_EXTENSION_OF_TELOMERES
REACTOME_ACTIVATION_OF_ATR_IN_RESPON
REACTOME_SYNTHESIS_OF_DNA
REACTOME_POTASSIUM_CHANNELS
REACTOME_MITOTIC_G2_G2_M_PHASES
REACTOME_TELOMERE_MAINTENANCE
REACTOME_ACTIVATION_OF_THE_PRE_REPLI
REACTOME_HIV_INFECTION
REACTOME_TRANSPORT_OF_RIBONUCLEOPR
REACTOME_CLEAVAGE_OF_GROWING_TRANS
REACTOME_G0_AND_EARLY_G1
REACTOME_NEUROTRANSMITTER_RECEPTOR
REACTOME_PLATELET_ACTIVATION_SIGNALIN
REACTOME_INTERACTIONS_OF_VPR_WITH_HC
REACTOME_DEADENYLATION_DEPENDENT_MI
REACTOME_TRANSMEMBRANE_TRANSPORT_C
REACTOME_NEP_NS2_INTERACTS_WITH_THE
REACTOME_GPCR_LIGAND_BINDING
REACTOME_CLASS_A1_RHODOPSIN_LIKE_RE
REACTOME_M_G1_TRANSITION
REACTOME_PROCESSING_OF_CAPPED_INTRC
REACTOME_MRNA_PROCESSING
REACTOME_DNA_REPLICATION
REACTOME_MITOTIC_M_M_G1_PHASES
REACTOME_MRNA_SPLICING
REACTOME_METABOLISM_OF_RNA
REACTOME_NEURONAL_SYSTEM
REACTOME_CELL_CYCLE
REACTOME_CELL_CYCLE_MITOTIC

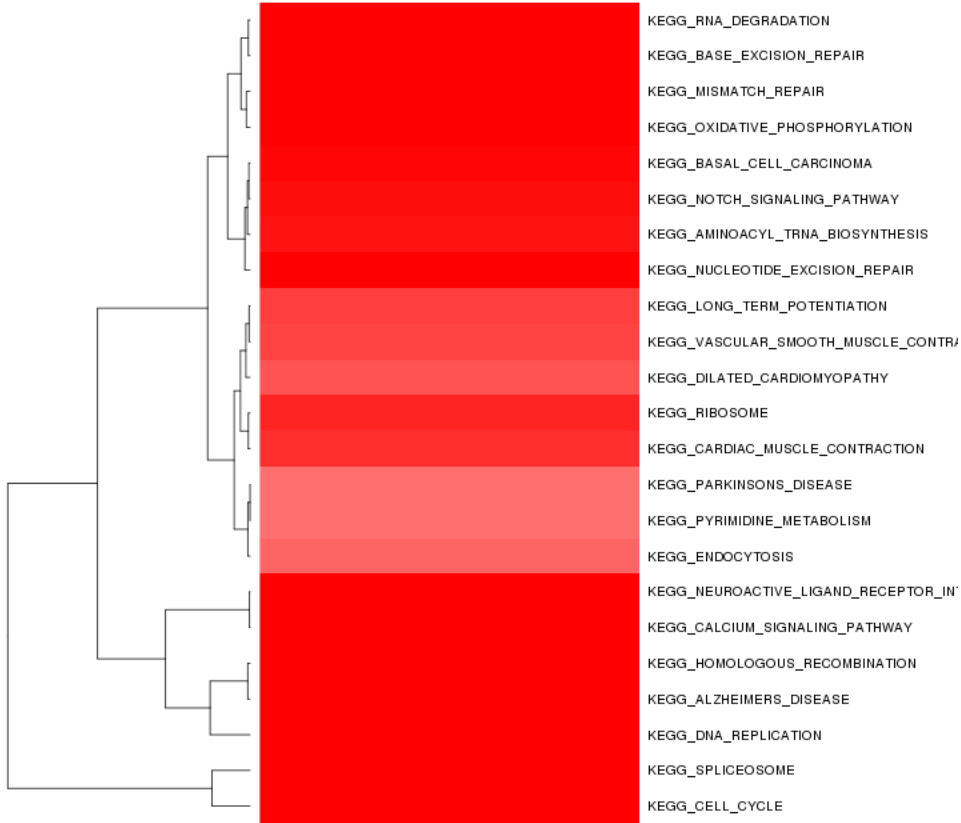
Reactome zscore:



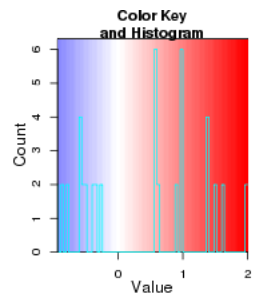


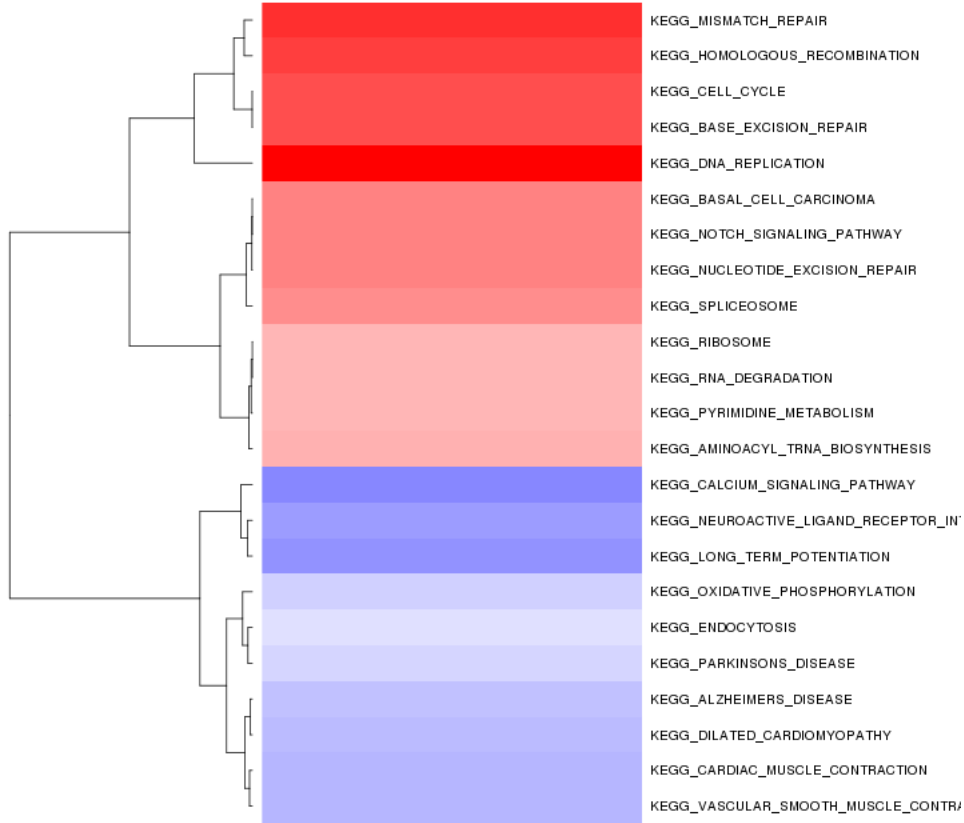
Kegg enrichment:





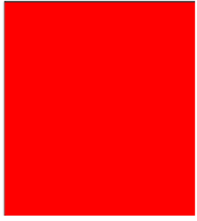
Kegg zscore:





Interneuron enrichment:

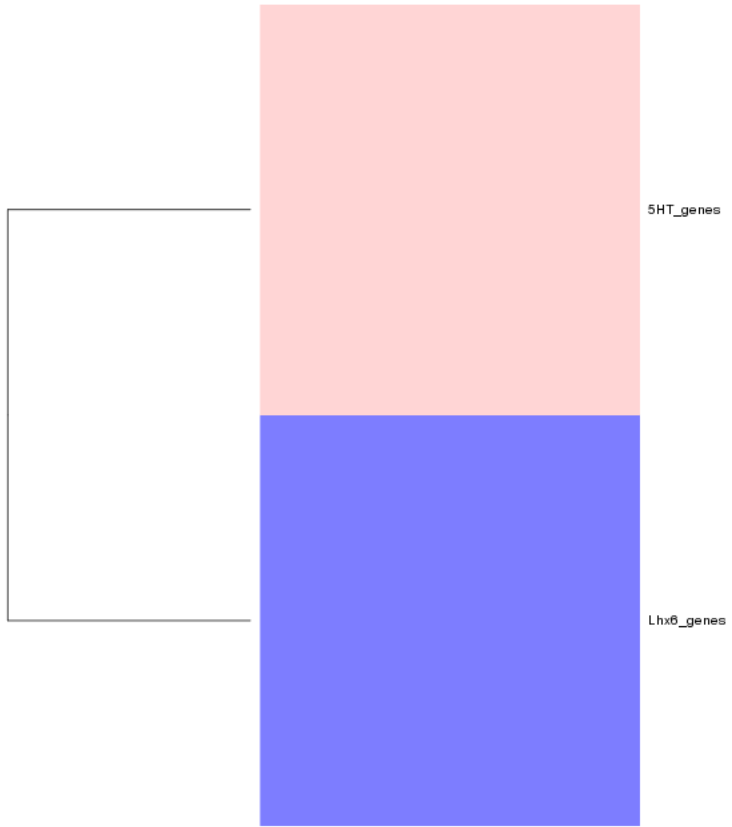
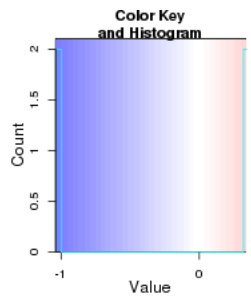
Error: no locations are finite



Lhx6_genes

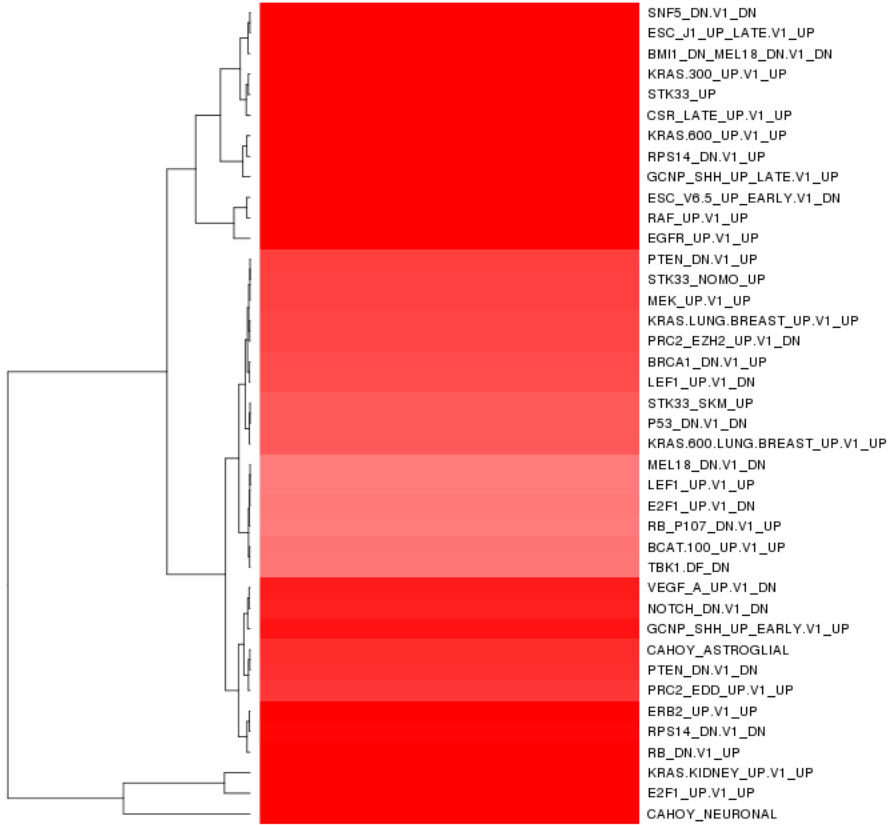
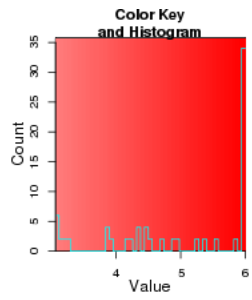
5HT_genes

Interneuron zscore:



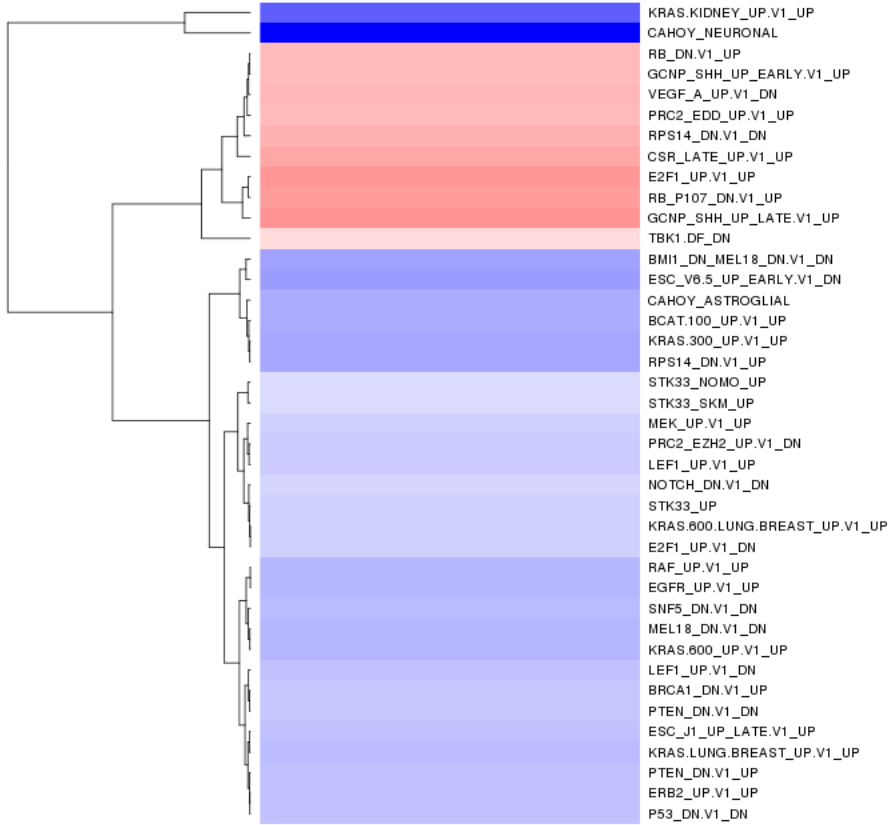
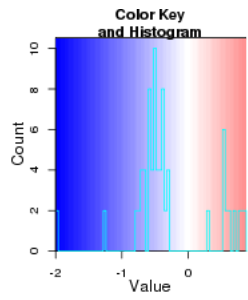
Oncogene enrichment:

```
## Error: subscript out of bounds
```



Oncogene zscore:

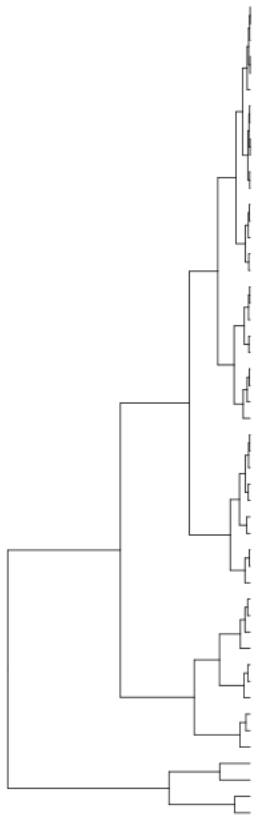
Error: subscript out of bounds



Immuno enrichment:

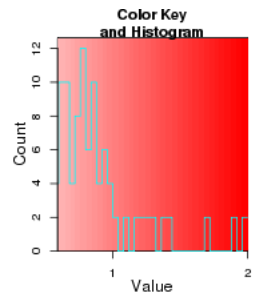
Error: no locations are finite

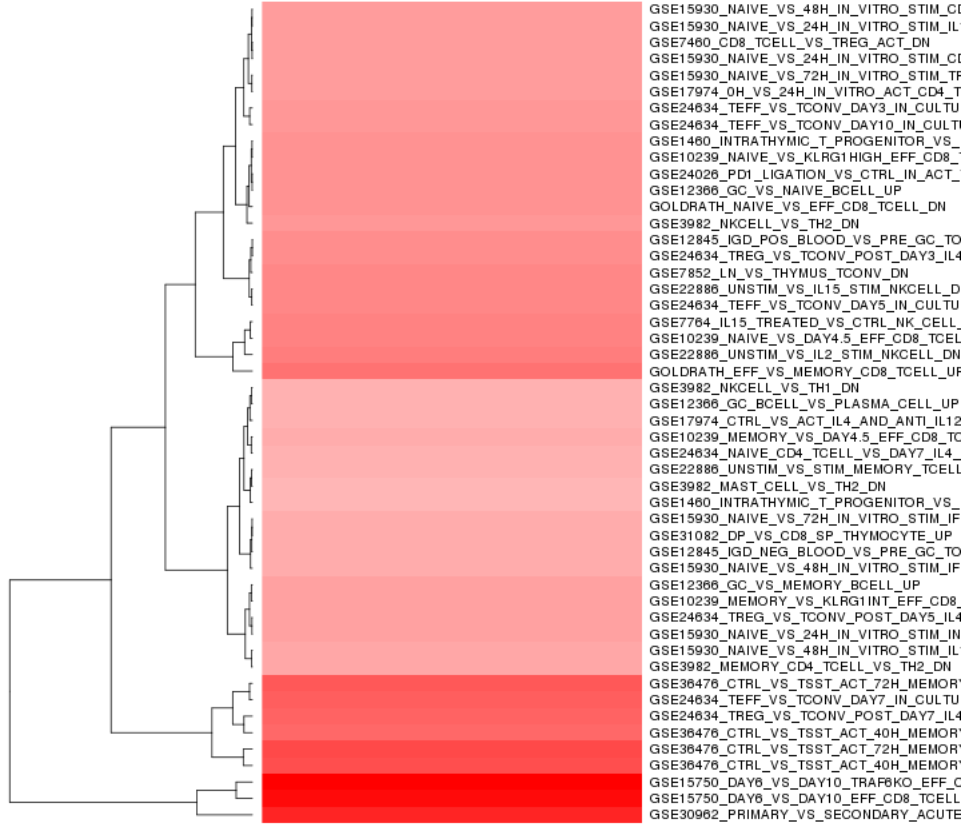




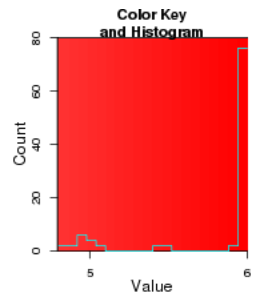
GSE15930_NAIVE_VS_72H_IN_VITRO_STIM_TF
GSE19825_NAIVE_VS_IL2RAHIGH_DAY3_EFF_
GSE22886_UNSTIM_VS_STIM_MEMORY_TCELL
GSE17974_0H_VS_24H_IN_VITRO_ACT_CD4_T
GOLDRATH_EFF_VS_MEMORY_CD8_TCELL_UF
GSE15930_NAIVE_VS_72H_IN_VITRO_STIM_IF
GSE10239_MEMORY_VS_KLRG1INT_EFF_CD8_
GSE3982_MAST_CELL_VS_TH2_DN
GSE27786_LIN_NEG_VS_MONO_MAC_UP
GSE31082_DP_VS_CD8_SP_THYMOCYTE_UP
GSE10239_NAIVE_VS_DAY4.5_EFF_CD8_TCEL
GSE3982_NKCELL_VS_TH2_DN
GSE10325_LUPUS_CD4_TCELL_VS_LUPUS_M
GSE12845_IGD_POS_BLOOD_VS_DARKZONE_
GSE1460_INTRATHYMIC_T_PROGENITOR_VS_
GSE12845_IGD_NEG_BLOOD_VS_PRE_GC_TO
GSE15767_MED_VS_SCS_MAC_LN_UP
GSE31082_DN_VS_CD4_SP_THYMOCYTE_UP
GSE12845_IGD_NEG_BLOOD_VS_DARKZONE_
GSE24634_TREG_VS_TCONV_POST_DAY3_IL4
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IL
GSE19825_CD24LOW_VS_IL2RA_HIGH_DAY3_
GSE7852_LN_VS_THYMUS_TCONV_DN
GSE9006_TYPE_1_VS_TYPE_2_DIABETES_PB
GSE12845_IGD_POS_BLOOD_VS_PRE_GC_TO
GSE7460_CD8_TCELL_VS_TREG_ACT_DN
GSE24634_TEFF_VS_TCONV_DAY3_IN_CULTU
GSE24634_TREG_VS_TCONV_POST_DAYS_IL4
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IN
GSE14000_TRANSLATED_RNA_VS_MRNA_4H_
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IF
GSE1460_INTRATHYMIC_T_PROGENITOR_VS_
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_CI
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IL
GSE24634_TEFF_VS_TCONV_DAYS_IN_CULTU
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_CI
GSE24634_TREG_VS_TCONV_POST_DAY3_IL4
GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR
GSE22886_UNSTIM_VS_IL15_STIM_NKCELL_D
GSE7764_IL15_TREATED_VS_CTRL_NK_CELL_
GSE24634_TEFF_VS_TCONV_DAY7_IN_CULTU
GSE24634_TREG_VS_TCONV_POST_DAY7_IL4
GSE24026_PDI_LIGATION_VS_CTRL_IN_ACT_
GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR
GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR
GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR
GSE15750_DAY6_VS_DAY10_TRAF6KO_EFF_C
GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL
GSE22886_UNSTIM_VS_IL2_STIM_NKCELL_DN
GSE30962_PRIMARY_VS_SECONDARY_ACUTE

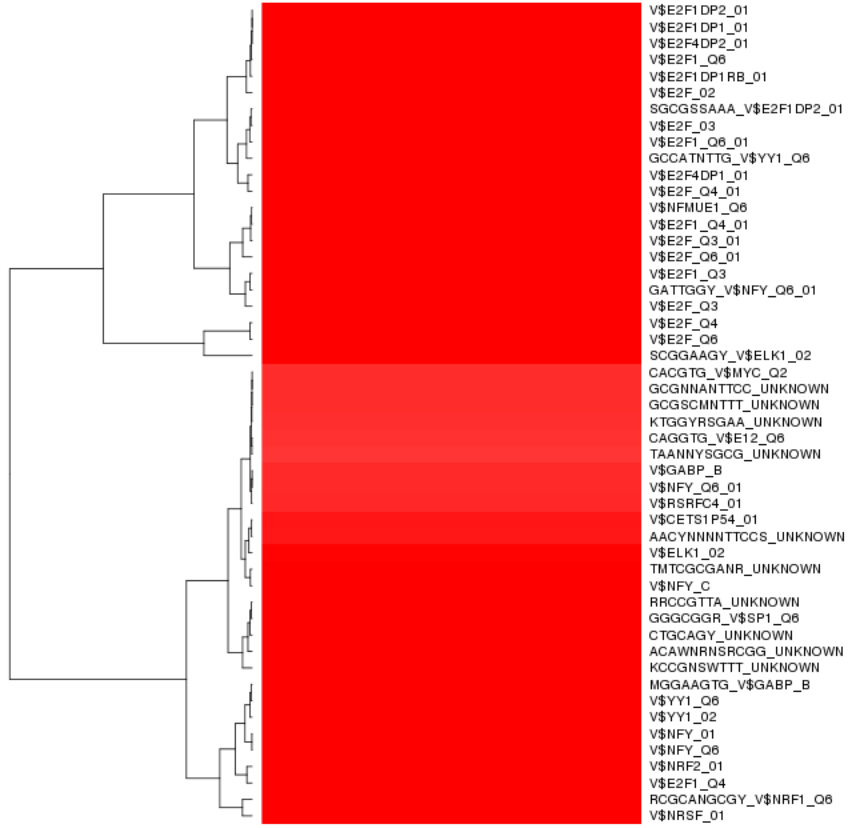
Immuno zscore:



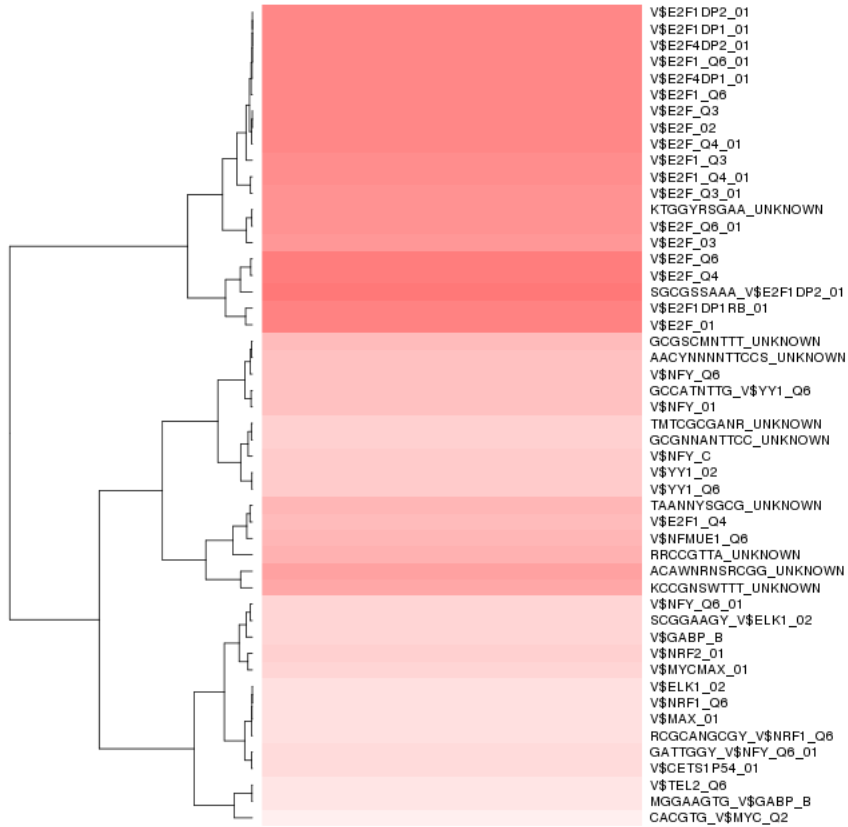
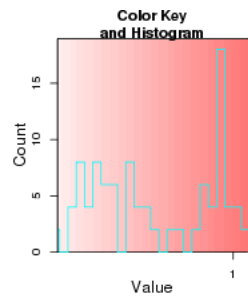


TF enrichment:





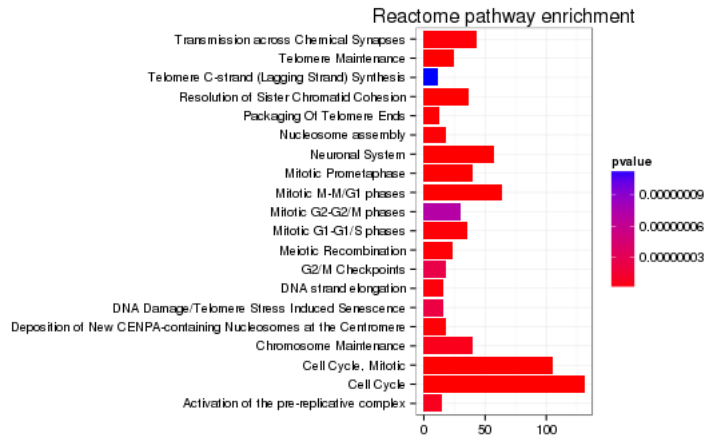
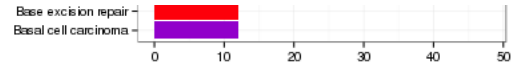
TF zscore:



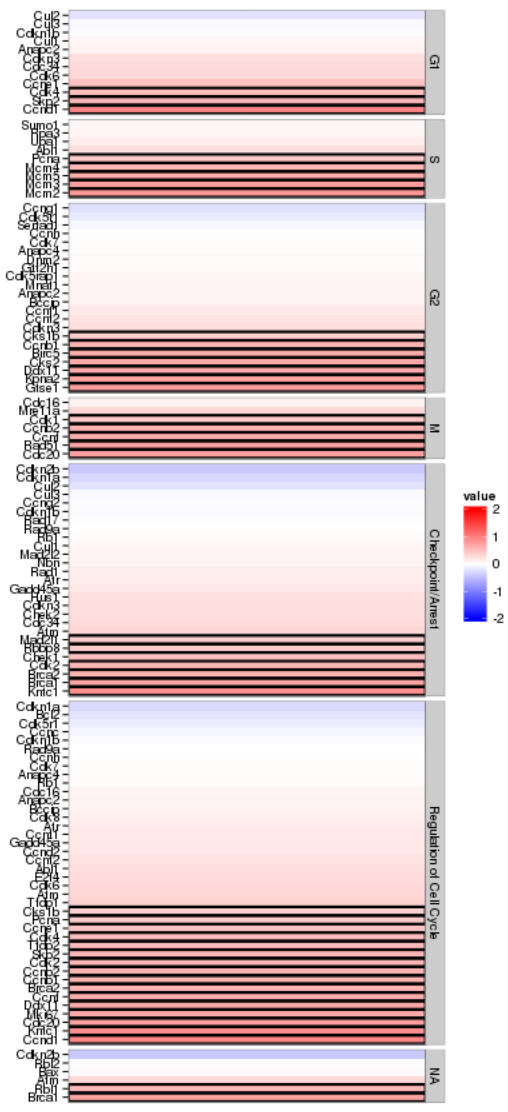
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

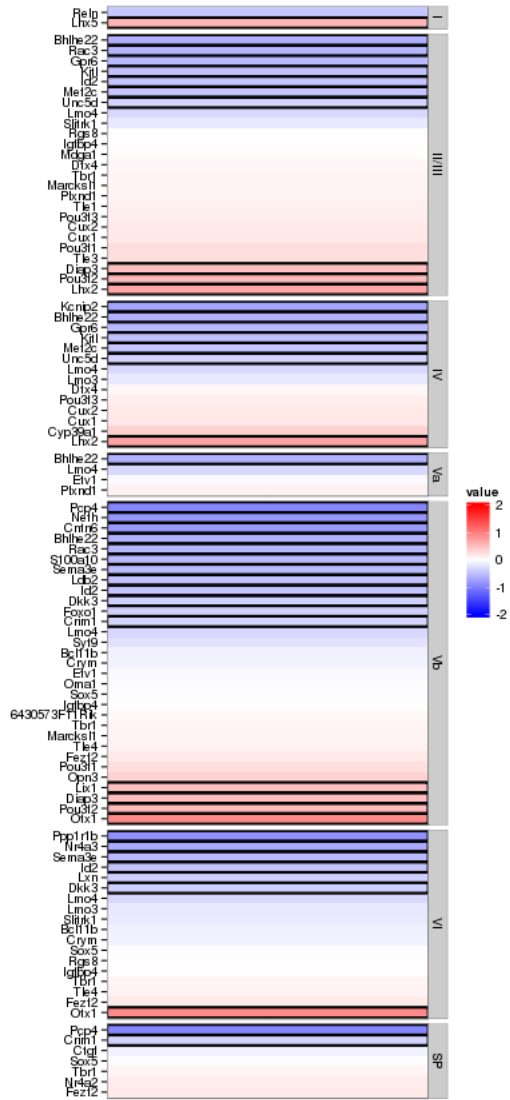
cell part
cell
GO



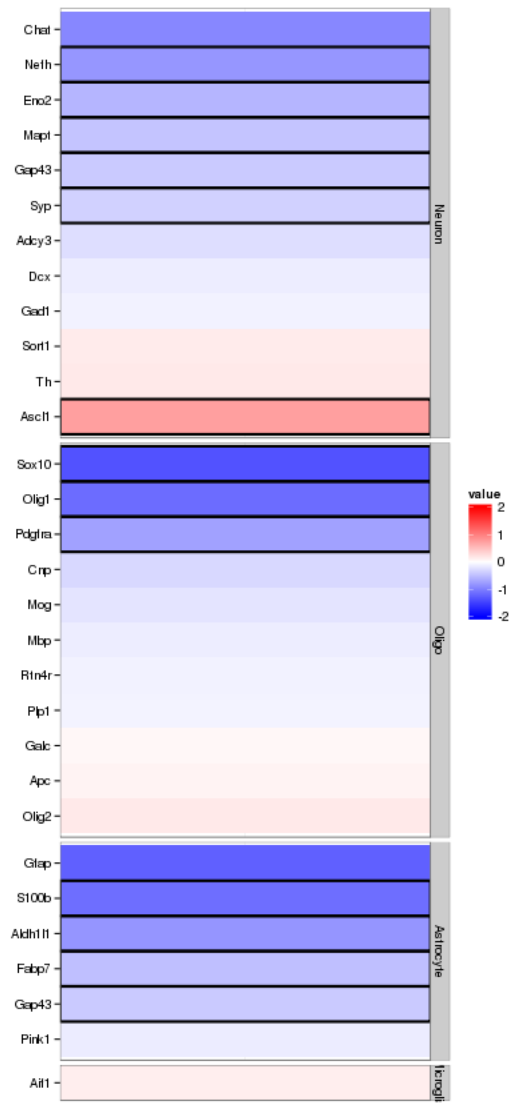
Enrichment or depletion for stage-specific cell cycle markers



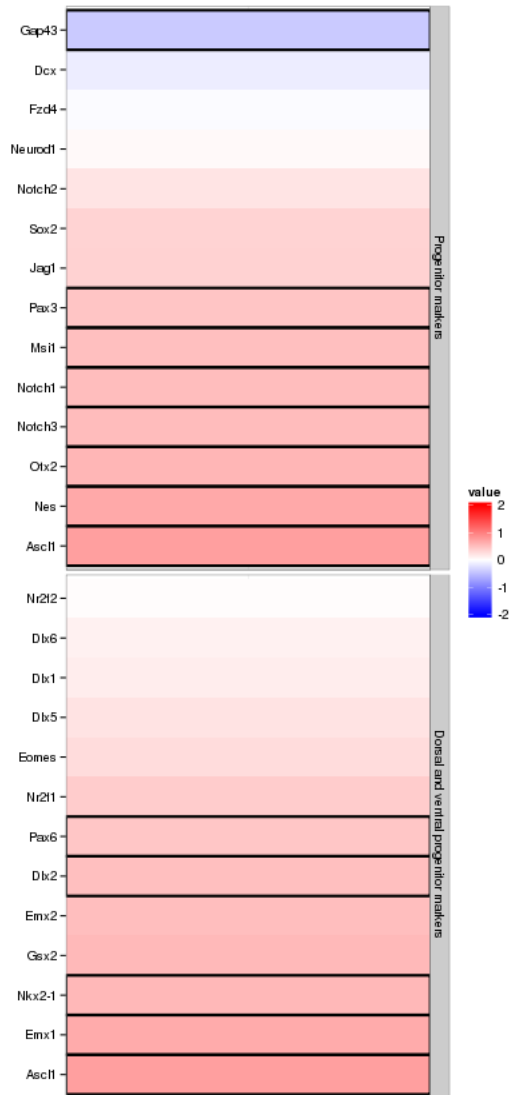
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



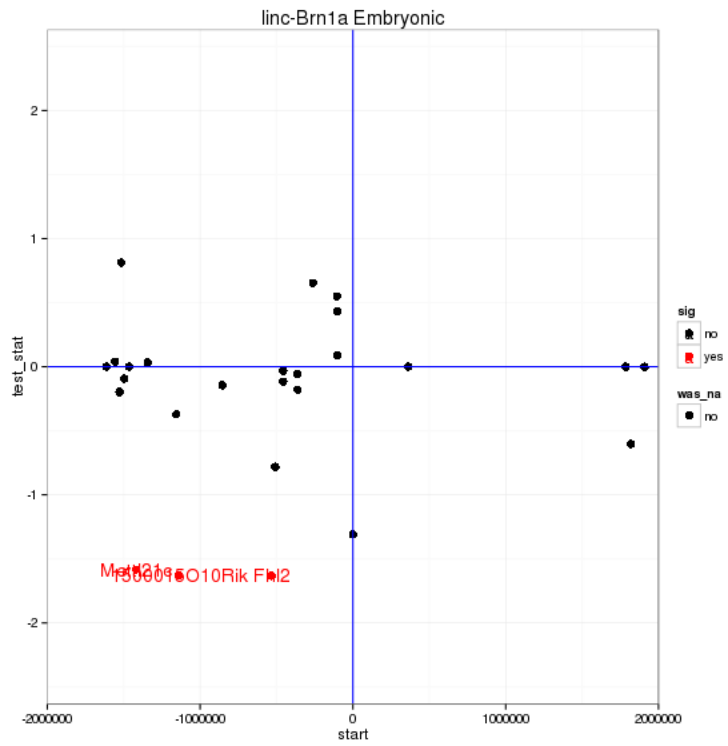
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 3 genes significantly regulated in a region this size is: 0.6938



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR858
16 JR859
17 JR860

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	33371600.00	1.23	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	33371600.00	1.07	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	33371600.00	1.19	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	33371600.00	1.25	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	33371600.00	1.16	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	33371600.00	0.87	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	33371600.00	0.94	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	33371600.00	0.90	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	33371600.00	0.84	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	33371600.00	0.93	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	33371600.00	0.78	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	33371600.00	0.78	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	33371600.00	0.67	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	33371600.00	1.18	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR858/abundances.cxb	linc_Bm1a	0	linc_Bm1a_0	34867700.00	33371600.00	1.08	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR859/abundances.cxb	linc_Bm1a	1	linc_Bm1a_1	45282400.00	33371600.00	1.30	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR860/abundances.cxb	linc_Bm1a	2	linc_Bm1a_2	41604700.00	33371600.00	1.20	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1      KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1      reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1        zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,linc-Brn1a -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/linc-Brn1a_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/Br
## 2
## 3
## 4
## 5
```

linc-Brn1b KO vs WT (Adult)

This file shows the wt-v-ko comparison for linc-Brn1b.

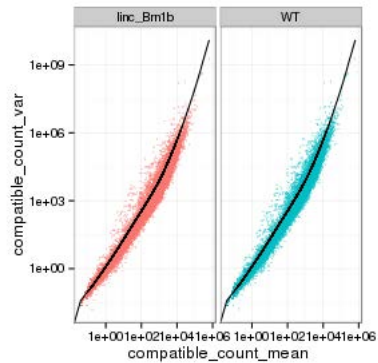
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

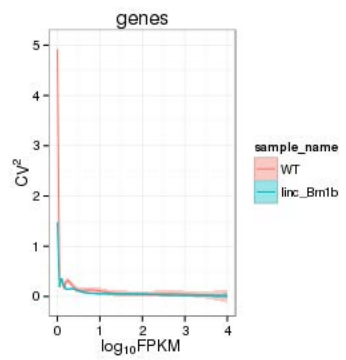
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



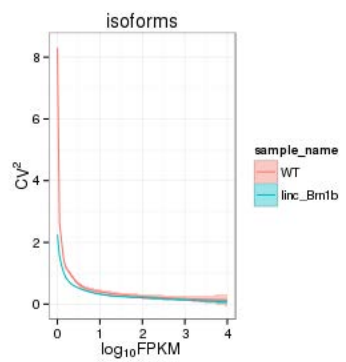
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

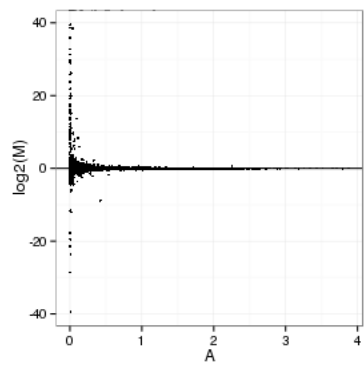
Genes:



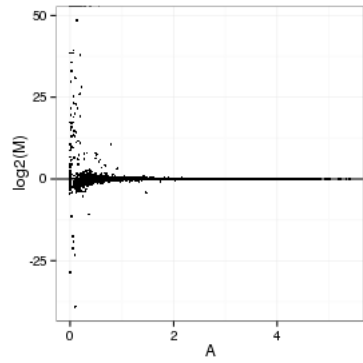
Isoforms:



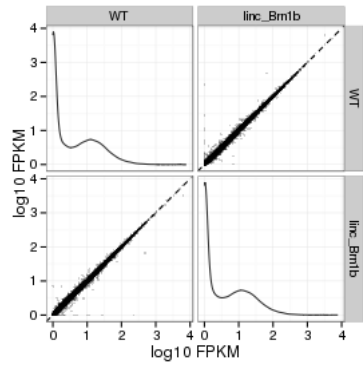
MvA plot



MvA plot counts



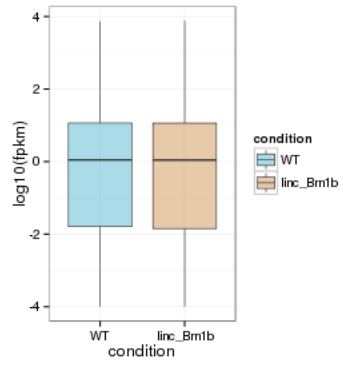
Scatterplot matrix



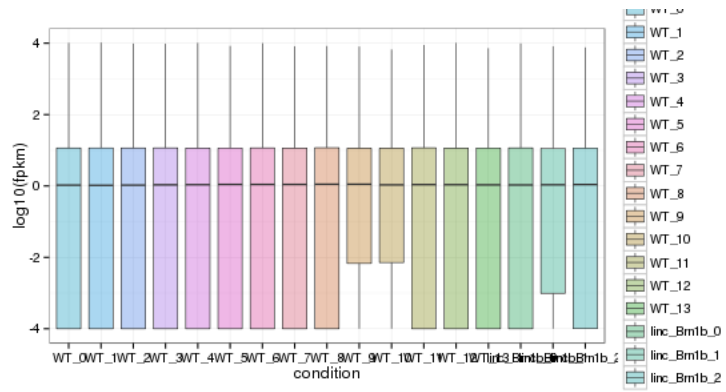
Distributions

Boxplots

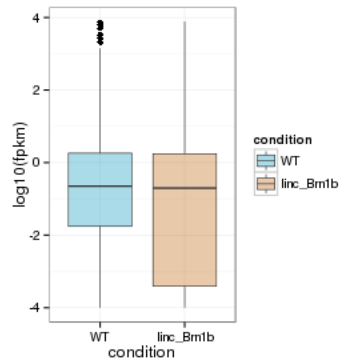
Boxplot (genes)



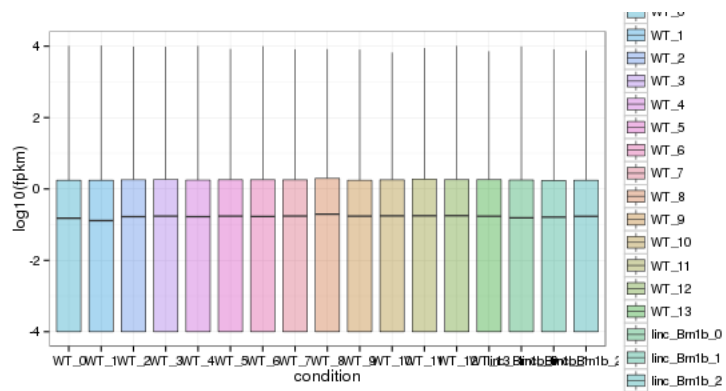
Boxplot (genes, replicates)



Boxplot (isoforms)

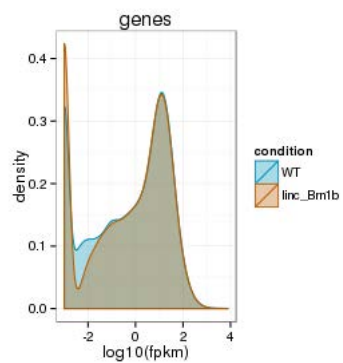


Boxplot (isoforms, replicates)

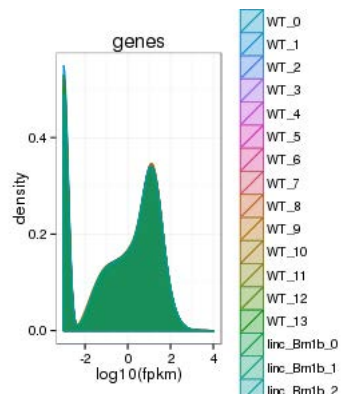


Density

Density (genes)

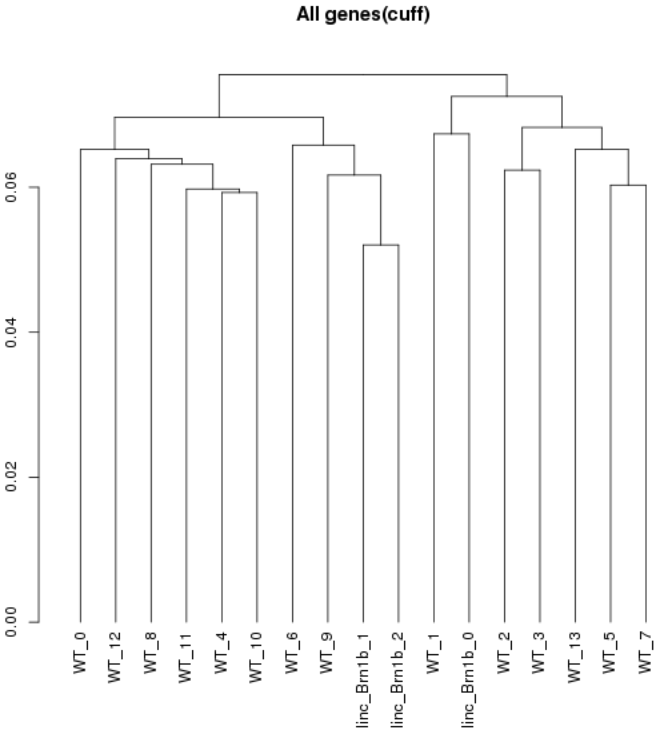


Density (genes, replicates)



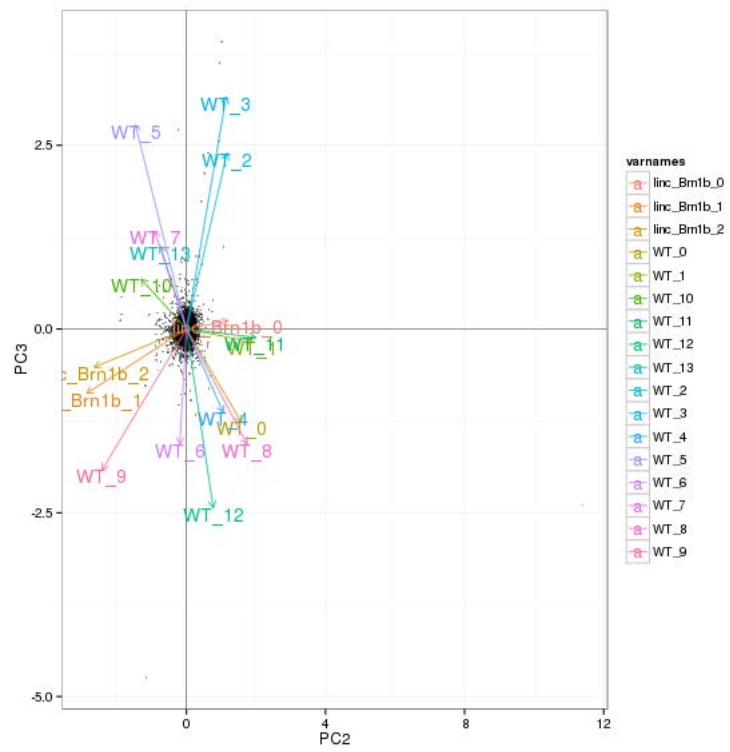
Clustering

Replicate Clusters

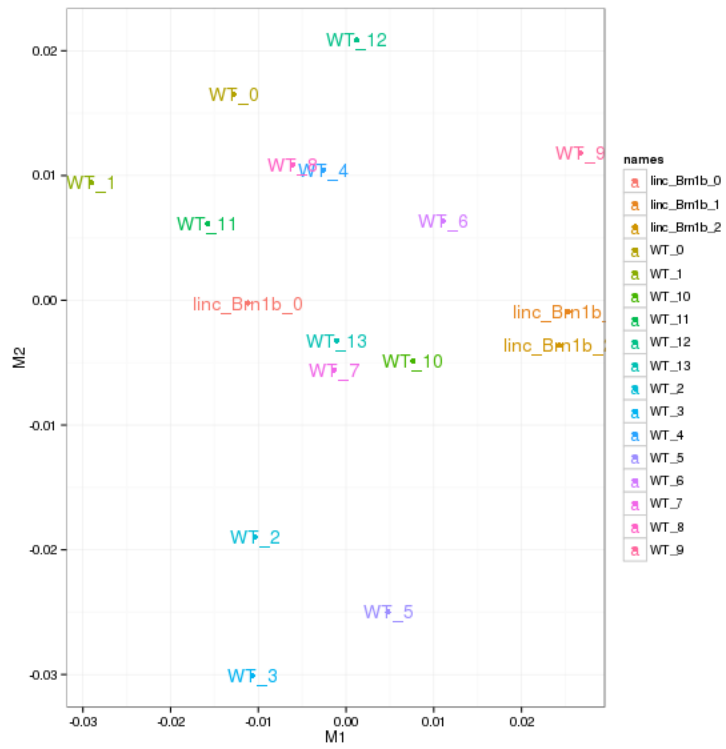


NULL

PCA (genes)

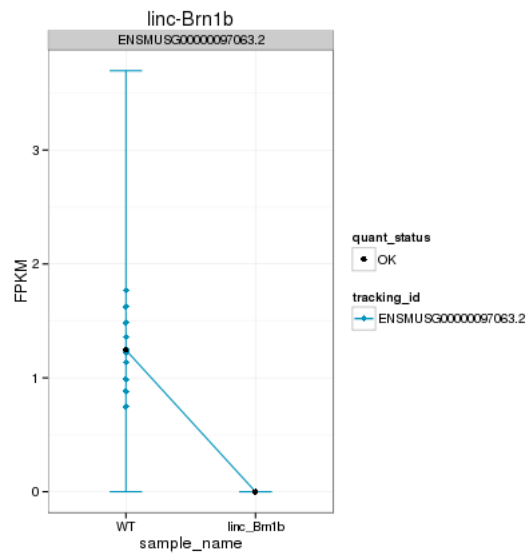


MDS (genes)

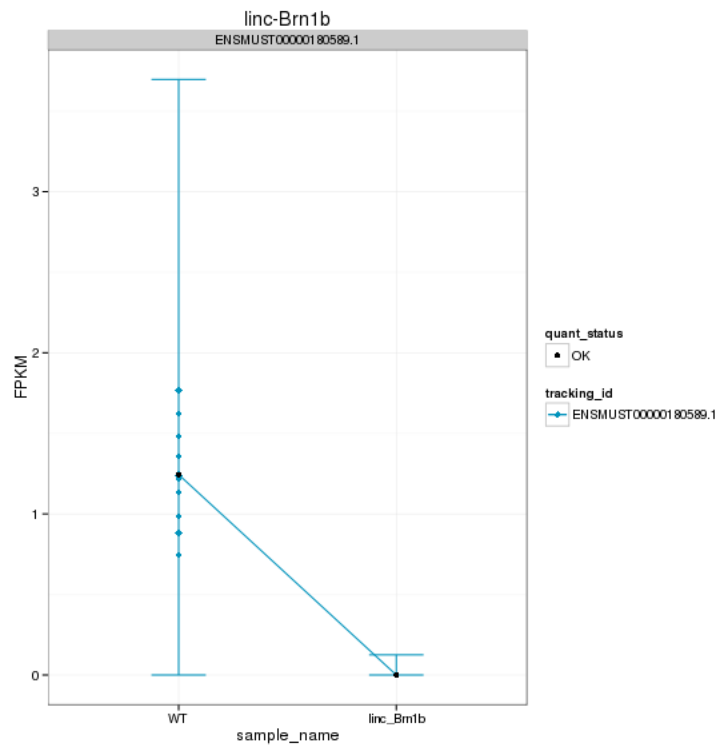


KO assessment

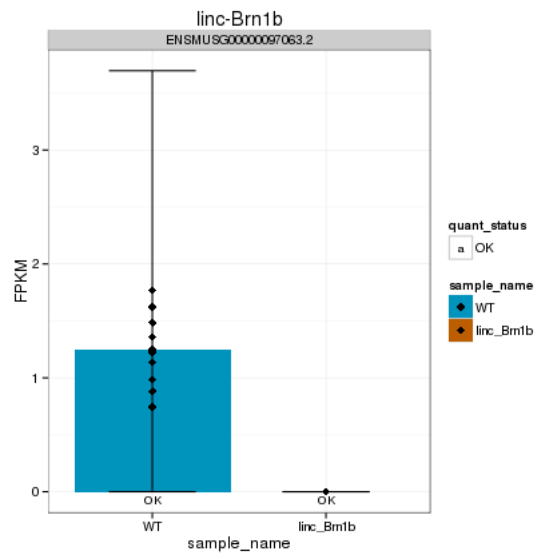
Endogenous lincRNA expression



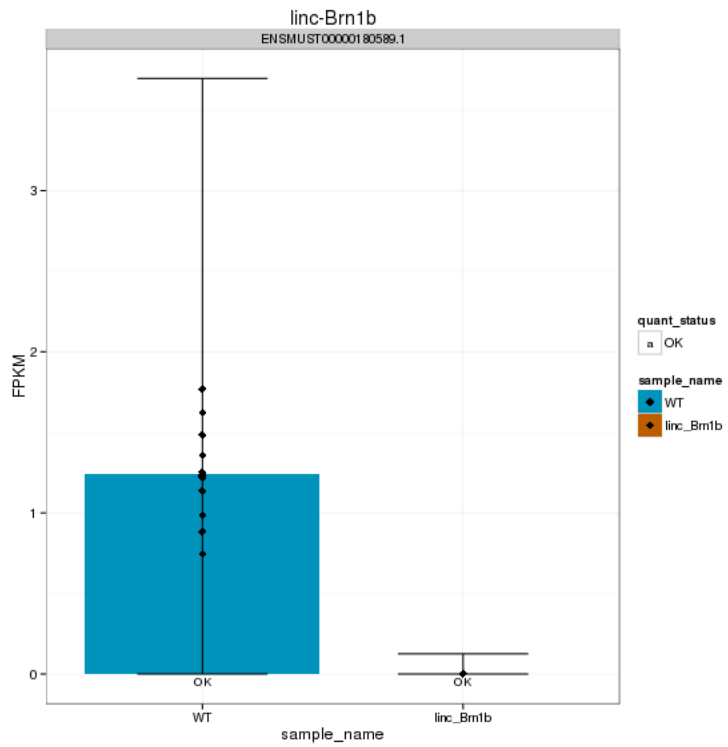
Endogenous expression of linc-Brn1b isoforms:



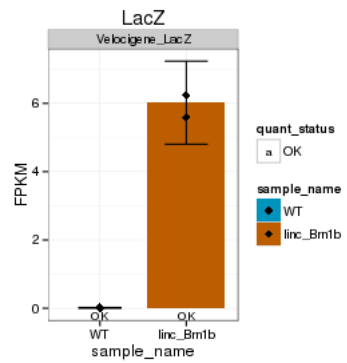
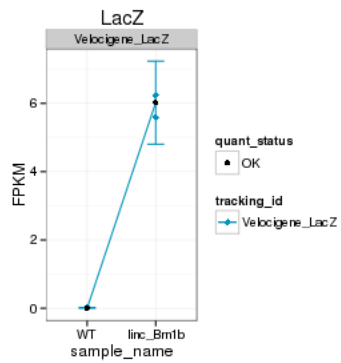
Barplot of gene expression:



Barplot of isoform expression:

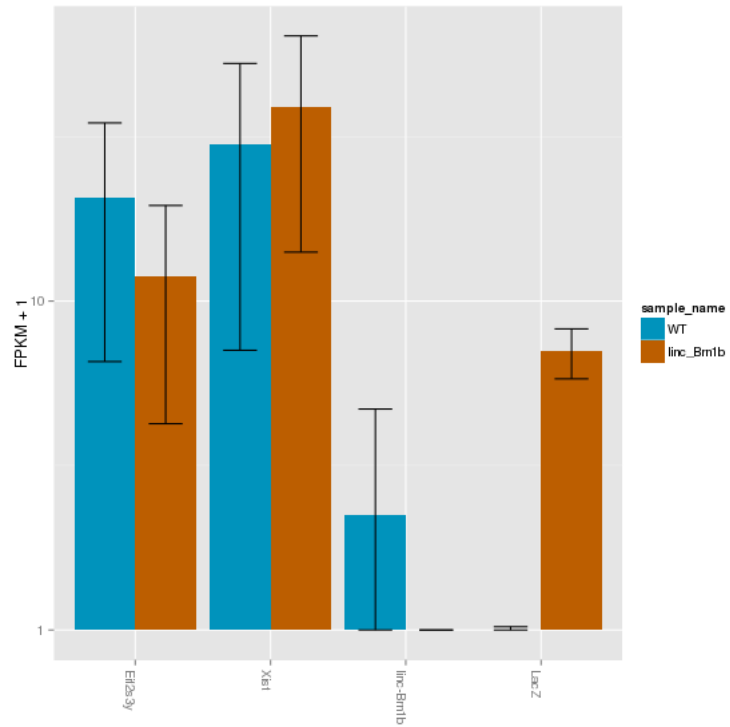


LacZ expression

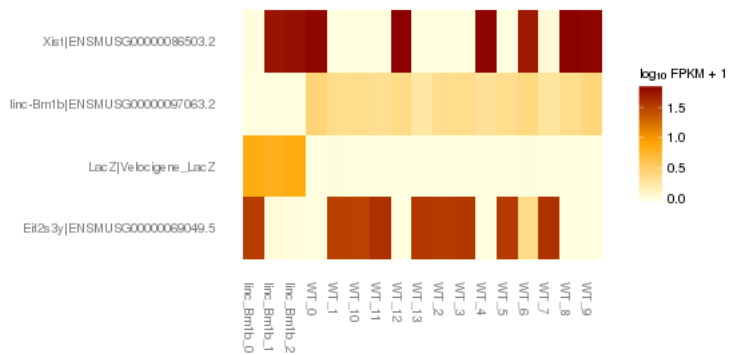


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

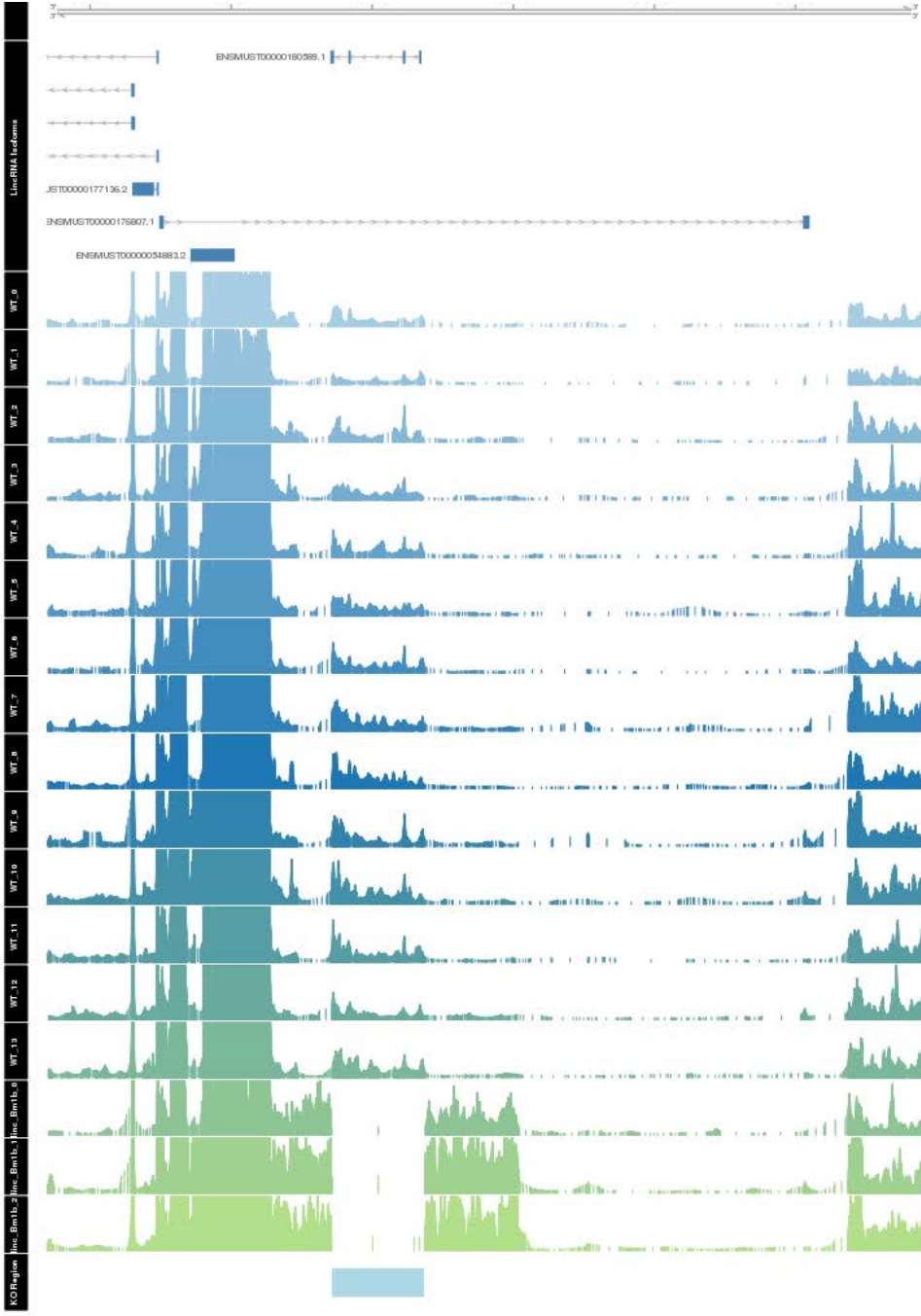


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 193 significantly differentially expressed genes. They are:

	geneAnnot\$gene_short_name
1	Gabra2
2	Sox9
3	Folh1
4	Kmt2a
5	Rec8
6	Arrdc2
7	Calr
8	Gpx6
9	Pde1c
10	Nes
11	Epha2
12	Homer1
13	Dio2
14	Hnmpa0
15	Ttc9b
16	Carhsp1
17	Tppp3
18	Etnppl
19	Pdk4
20	P4ha1
21	Arid5b
22	Sgk1
23	Timp3
24	Ddit4
25	Btg2
26	Xbp1
27	Pdia6
28	Pxdn
29	Nfkbia
30	Serpina3n
31	Fos
32	Gpr137b
33	Zkscan3
34	Esd
35	Dnajc3
36	Slc38a2
37	Bcl6

38 Sdf2l1
39 Cdkn1a
40 Creld2
41 Glo1
42 Xdh
43 Atp6v0c

44 Dusp1
45 Fkbp5
46 Syt4
47 Rps24
48 Pdia4
49 Slc40a1
50 Clk1
51 2810459M11Rik
52 Bok
53 Mlph
54 Serpinb8
55 Dbi
56 Atf3
57 Lcn2
58 Hspa5
59 Itpka
60 Spred1
61 F3
62 Trp53inp1
63 Nr4a3
64 Slc2a1
65 Tinagl1
66 Errfi1
67 Ccnl2
68 Fosl2
69 Uncx
70 Klf15

71 Adipor2
72 Pglyrp1
73 Tmem238
74 Hddc3
75 Polr3e
76 Tsc22d3
77 Plat
78 Mt2
79 Tdgf1
80 Mlh1

81 Cck
82 Trf
83 Ugt8a
84 Tbc1d4
85 Egr3
86 Tlr13
87 Slc25a37
88 Arhgap27
89 Pla2g3
90 Tiparp
91 Prr7
92 Nostrin
93 Filip1
94 Arl4d
95 Cox8a
96 H2-Q4
97 Anln

98 Tob1

99 Avp
100 Vstm2l
101 Smim3
102 Camk4
103 Txnip
104 Foxq1
105 Akap12
106 Irs2
107 Pcsk1n
108 Tnfsf10
109 Zfp189
110 Slc6a5
111 C1qtnf4
112 Nkx6-2
113 Cldn5
114 Kcnj2
115 Maff
116 Tuba1c
117 Lor
118 Nxpe4
119 Ctla2a
120 Snhg11
121 Rps2
122 Hjurp
123 Pcdhb7

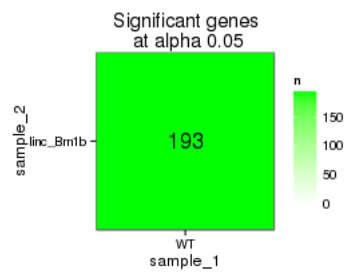
124 Cirbp
125 Nudt18
126 Pcdhb3
127 Zfp36l2
128 Npas4
129 Clec14a
130 Plaur
131 Mpeg1
132 Cdh19
133 Gimap6
134 Hes5
135 Tmem252
136 Ctxn1
137 Ccdc6
138 Dact2
139 Chchd10
140 Rasd1
141 Rasl11b
142 Klk6
143 Kcnk12
144 Plekho2
145 Pcdhb6
146 Hbb-bs
147 Junb
148 Ier2
149 Mat2a
150 Iigp1
151 Tmem158
152 5033411D12Rik
153 Zcchc24

154 Pign
155 Kdm5d
156 Ero1lb
157 Mex3b
158 C030014I23Rik
159 Cox5b
160 Rpl34
161 Scoc
162 BC048507
163 Zbtb16
164 Bpifb9b
165 Bpifb3
166 Ddx3y

167 Irgm2
168 Gm1673
169 Junb
170 Egr4
171 Gm7292
172 Gpr27
173 H2-BI
174 Wdfy1
175 Plekha7
176 Arrdc3
177 Fjx1
178 Rpm1
179 Nrarp
180 Gbp4
181 Gm11273
182 Slc5a3
183 Cdr1
184 Apold1
185 Hspa1b
186 Malat1
187 AA465934
188 Gm21967
189 Ccdc85b
190 Gm6472
191 Al848285
192 4930480K23Rik
193 Gpr137b-ps

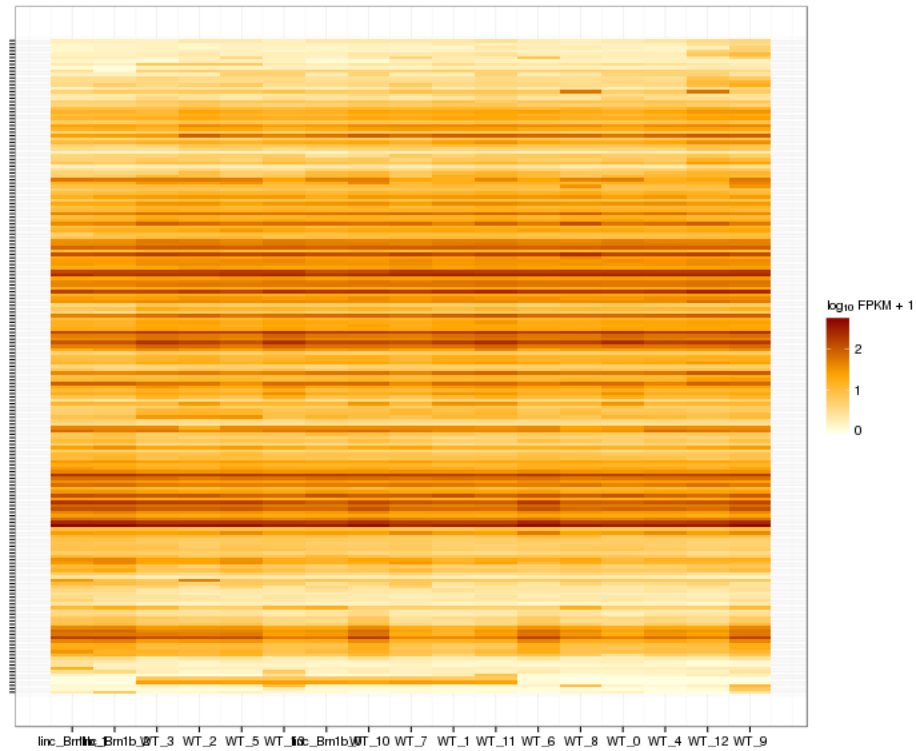
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

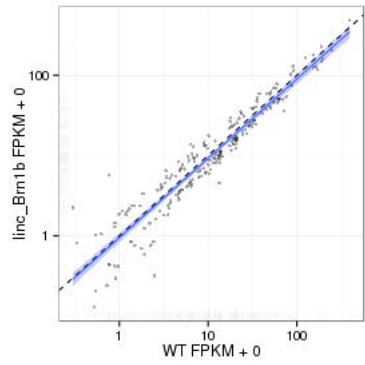


Significant genes with expression >50fpm (any condition):(turned off)

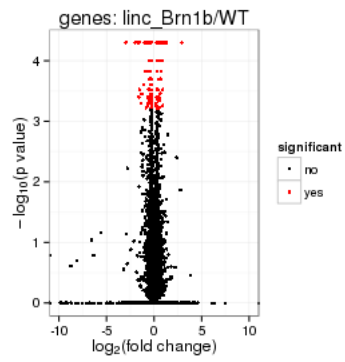
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

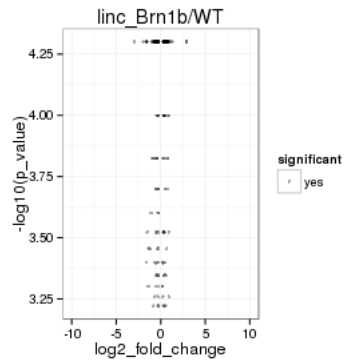
Scatter plot of significant genes only:



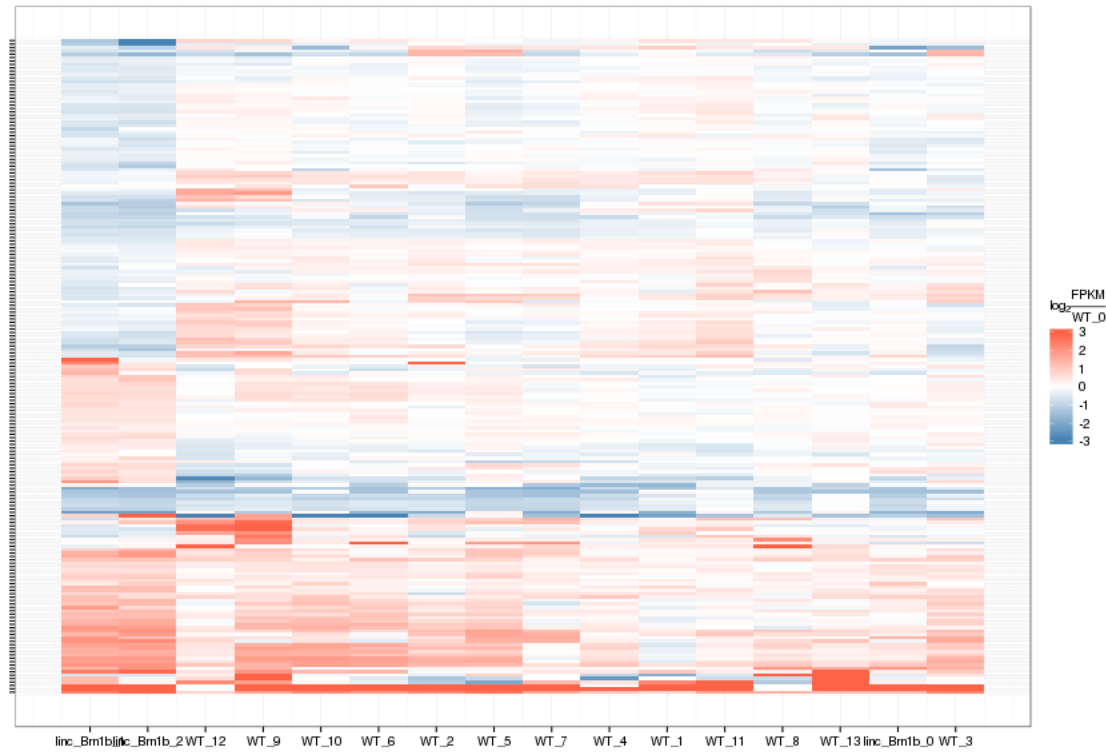
Volcano Plot



Volcano plot with significant genes only:



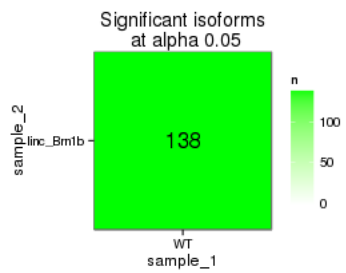
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

isoAnnot\$gene_short_name

- 1 Gabra2
- 2 Sox9
- 3 Rec8
- 4 Arrdc2
- 5 Calr
- 6 Hif3a
- 7 Nes
- 8 Ndrg1
- 9 Dio2

- 10 Ttc9b
- 11 Pdk4
- 12 Sgk1
- 13 Tmcc3
- 14 Timp3
- 15 Ddit4
- 16 Cpm
- 17 Tns3
- 18 Btg2
- 19 Xbp1
- 20 Pxdn
- 21 Nfkbia
- 22 Serpina3n
- 23 Fos
- 24 Gpr137b
- 25 Thbs4
- 26 Kat6b
- 27 Dnajc3
- 28 Slc38a2
- 29 Bcl6
- 30 Sdf2l1
- 31 Tfrc
- 32 Cdkn1a
- 33 Glo1
- 34 Xdh
- 35 Atp6v0c
- 36 Dusp1

- 37 Fkbp5
- 38 Rps24
- 39 Pdia4
- 40 Bok

41 Mlph
42 Hspa5
43 Spred1
44 F3
45 Slc2a1
46 Ago3
47 Map3k6
48 Errfi1
49 Ccnl2
50 Fosl2
51 Adipor2
52 Pglyrp1
53 Hddc3
54 Tsc22d3
55 Mt2
56 Mlh1
57 Cck
58 Ugt8a
59 Fndc3a
60 Akr1c14
61 Egr3
62 Slc25a37
63 Tiparp
64 Prr7

65 Arl4d
66 Cox8a
67 Anln
68 Tob1
69 Avp
70 Vstm2l
71 Acer2
72 Smim3
73 Camk4
74 Slc43a2
75 Aox4
76 Txnip
77 Foxq1
78 Akap12
79 Pcsk1n
80 Tnfsf10
81 Slc6a5
82 Gjb6
83 C1qtnf4

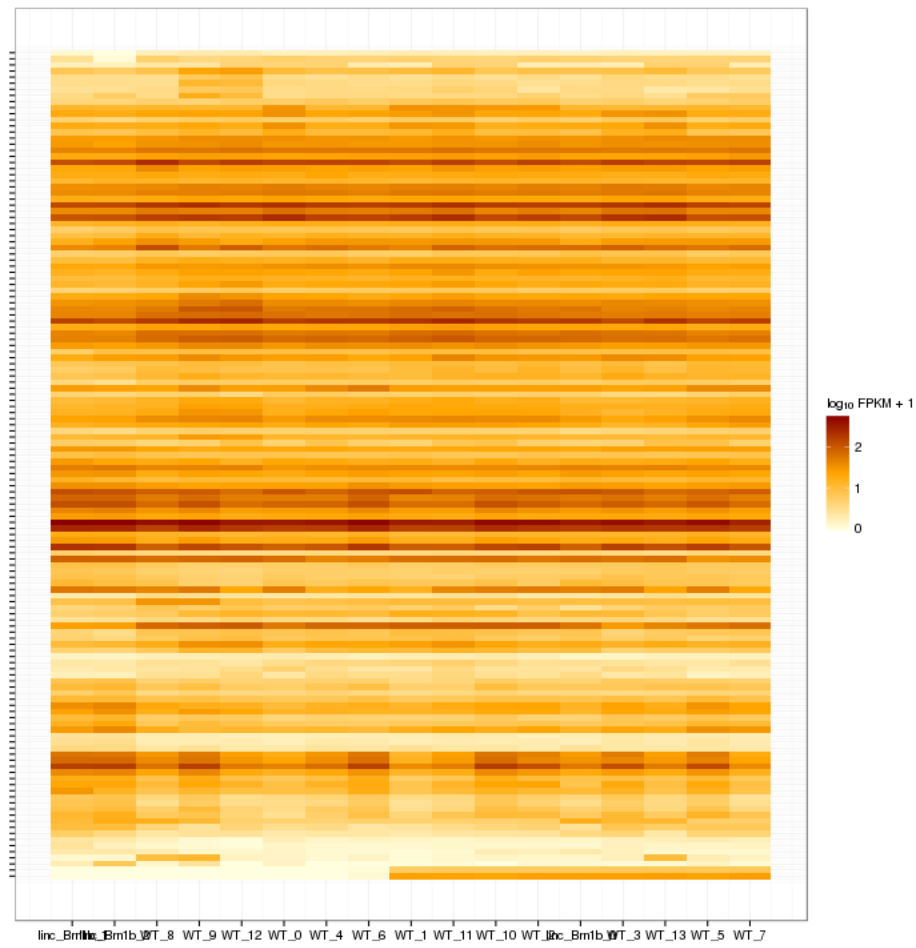
84 Nkx6-2
85 Cldn5
86 Kcnj2
87 Ctla2a
88 Shhg11
89 Acp1
90 Hjurp
91 Pcdhb7
92 Nudt18
93 Zfp36l2
94 Npas4
95 Cdh19
96 Hes5
97 Dact2
98 Chchd10
99 Kcnk12
100 Pcdhb6
101 Adarb2
102 Junb
103 Ier2
104 Mat2a
105 Zfp771
106 Tmem158
107 5033411D12Rik
108 Pign
109 Kdm5d
110 Ero1lb
111 Mex3b
112 Zbtb16
113 Bpifb9b
114 Ddx3y
115 Ahnak
116 Jund
117 Egr4

118 Gm7292
119 Gpr27

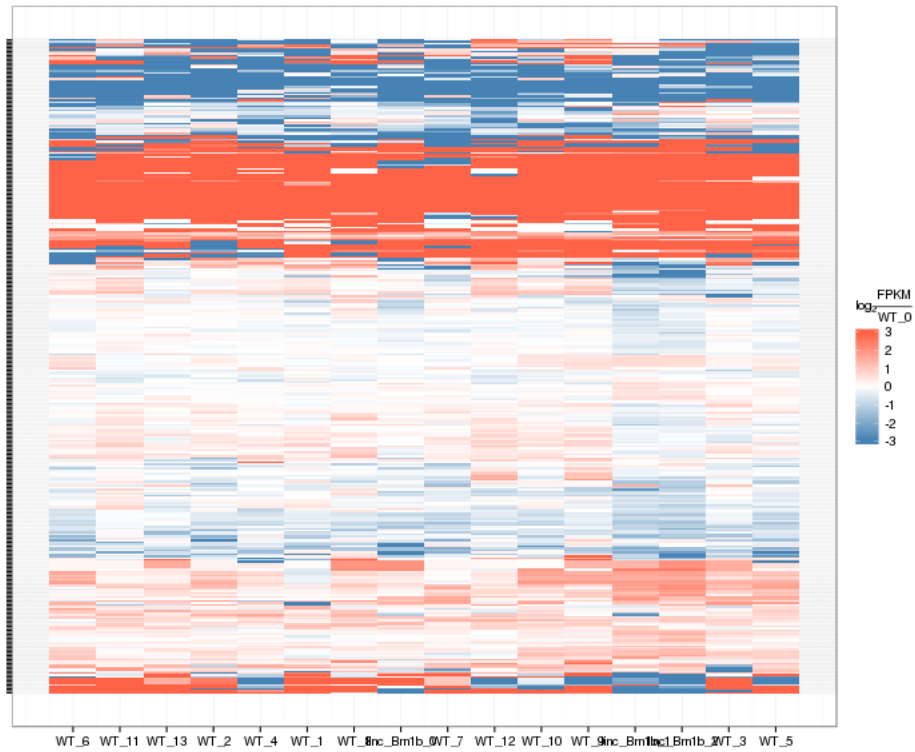
120 Wdfy1
121 Plekha7
122 Arrdc3
123 Fjx1
124 Rprm
125 Nrarp
126 Gbp4

- 127 Gm11273
- 128 Cdr1
- 129 Apold1
- 130 Hspa1b
- 131 Malat1
- 132 Ccdc85b
- 133 Gm6472
- 134 Al848285
- 135 4930480K23Rik
- 136 Gpr137b-ps

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

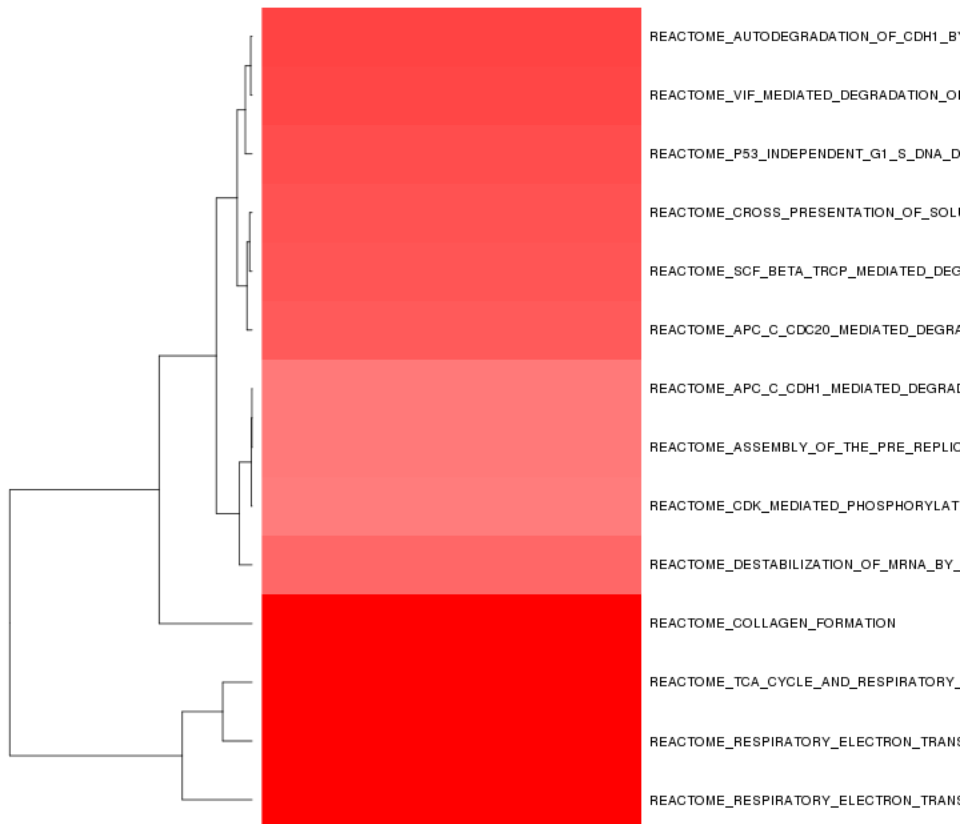
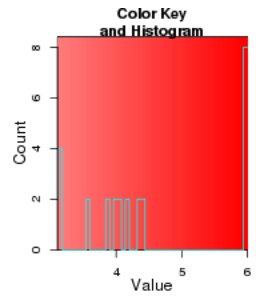
Biocarta enrichment:

```
## [1] "Not enough significant categories to print a heatmap!"
```

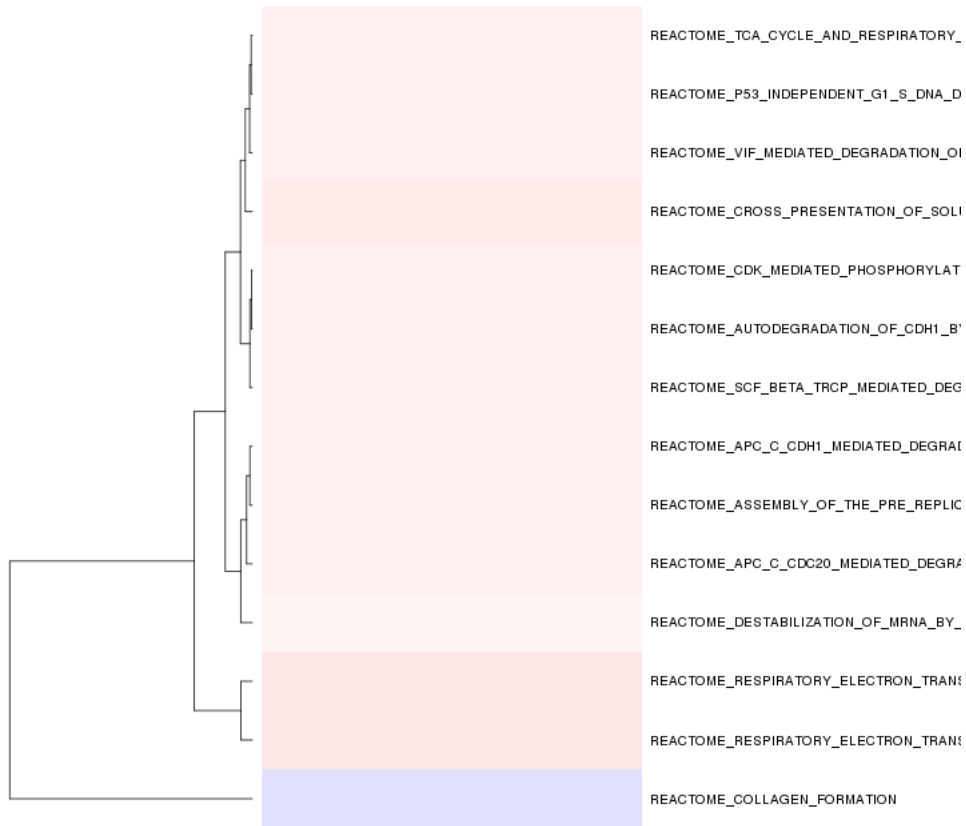
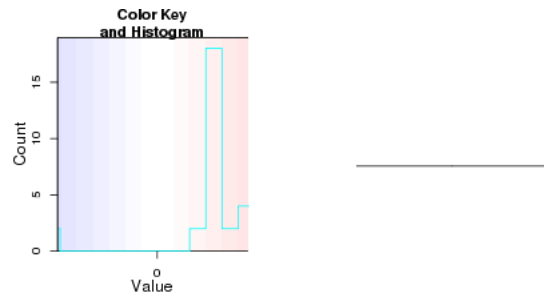
Biocarta zscore:

```
## [1] "Not enough significant categories to print a heatmap!"
```

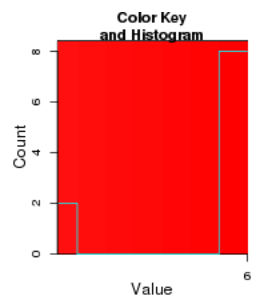
Reactome enrichment:

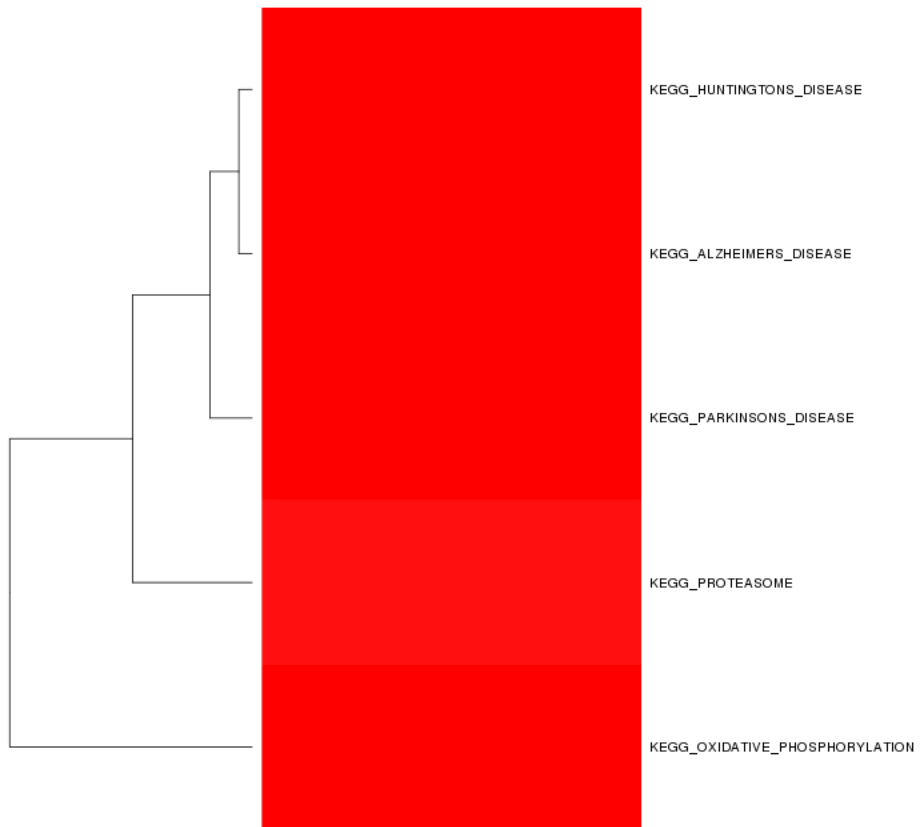


Reactome zscore:

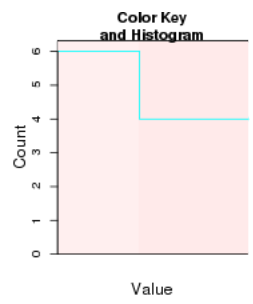


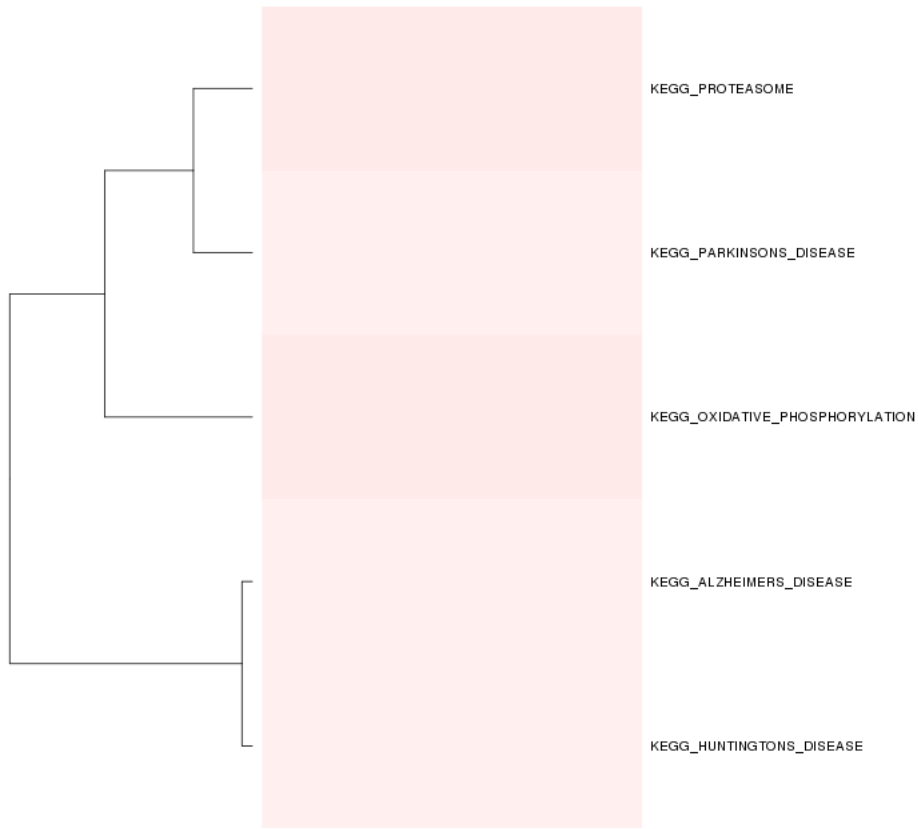
Kegg enrichment:





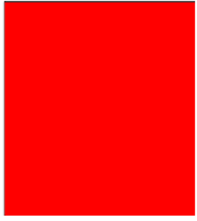
Kegg zscore:





Interneuron enrichment:

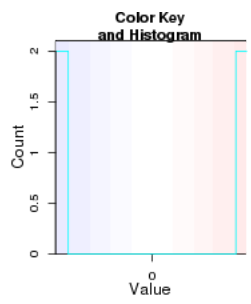
Error: no locations are finite



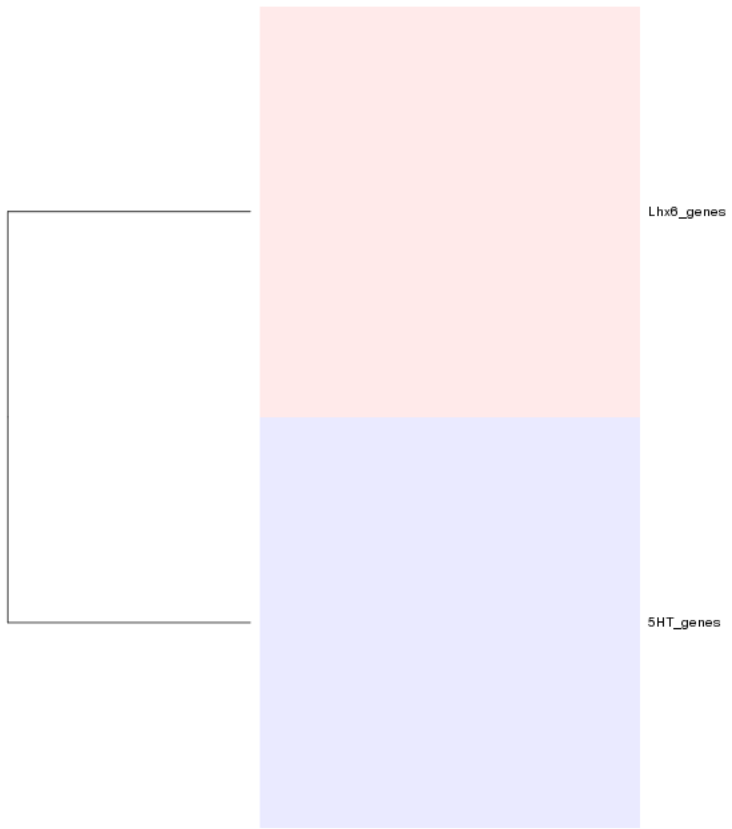
Lhx6_genes

5HT_genes

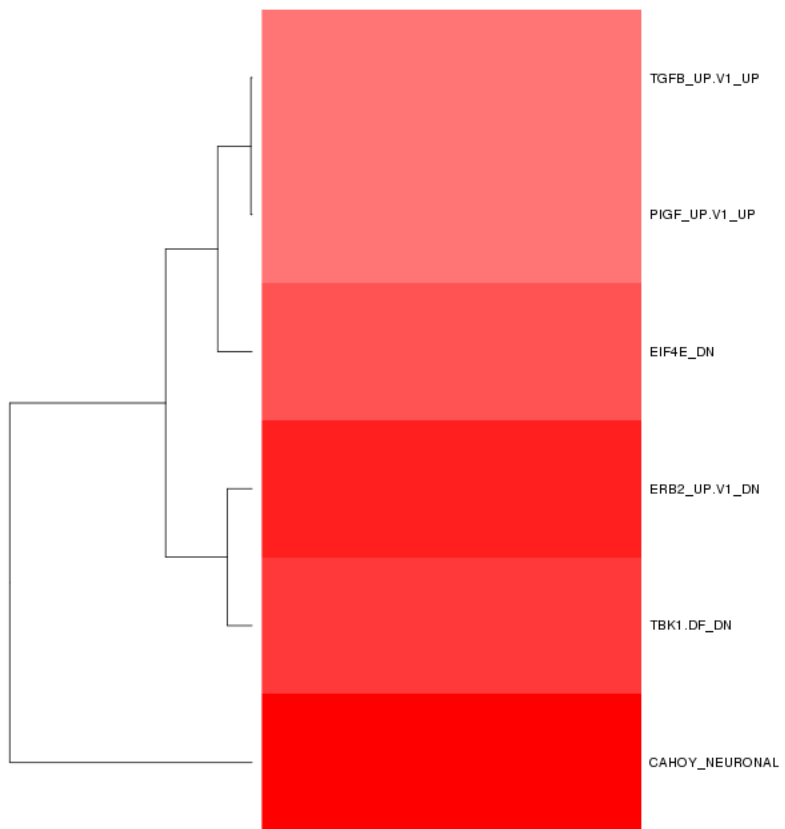
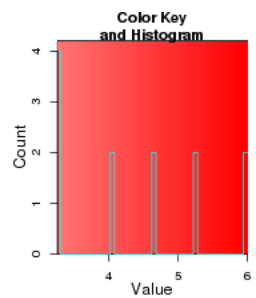
Interneuron zscore:



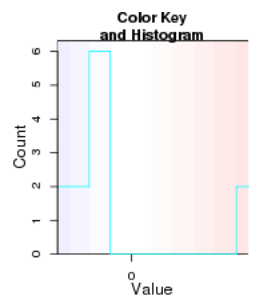
—

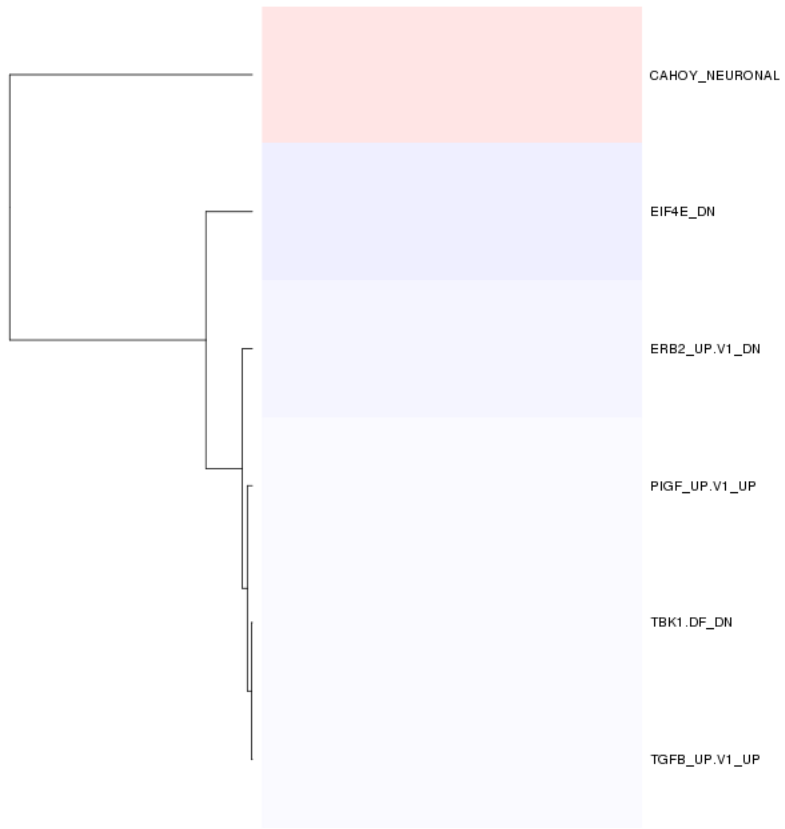


Oncogene enrichment:

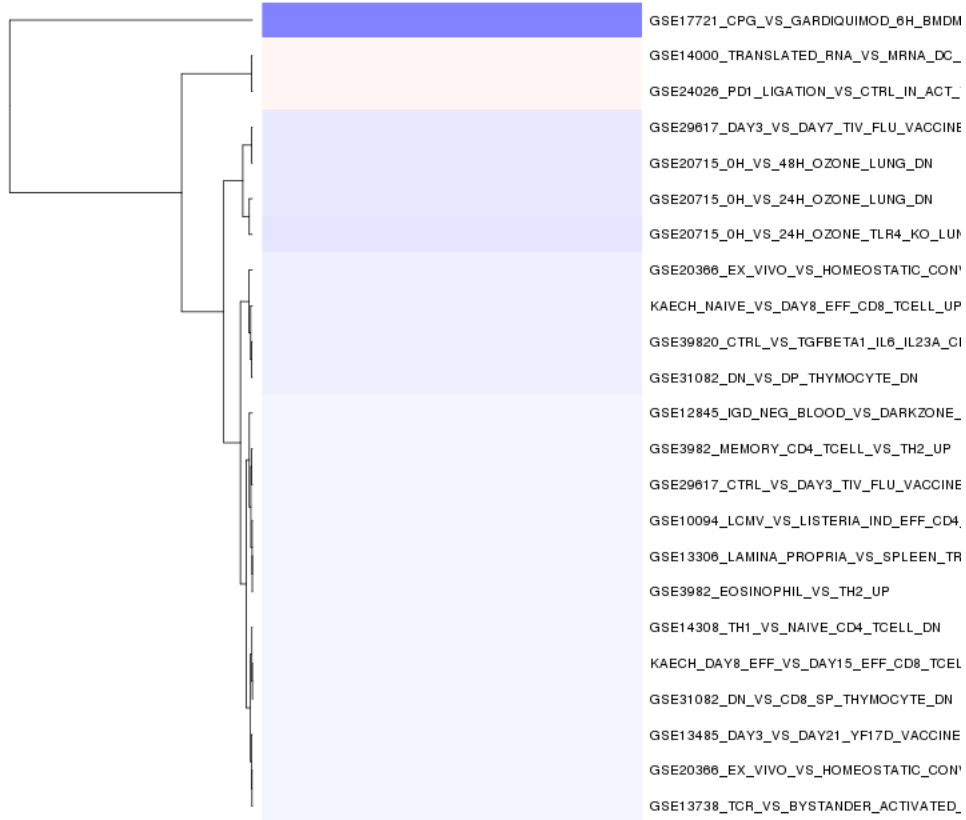
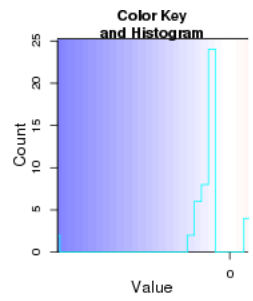


Oncogene zscore:

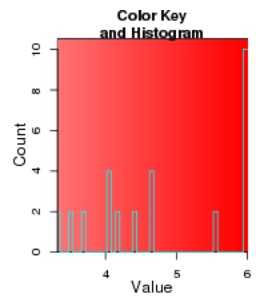


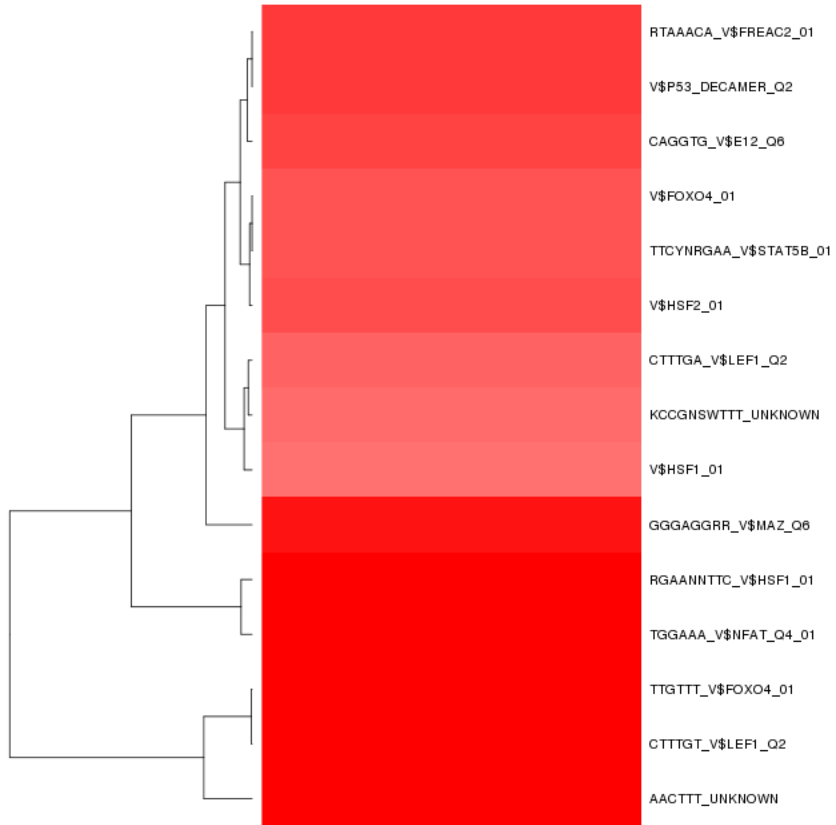


Immuno enrichment:

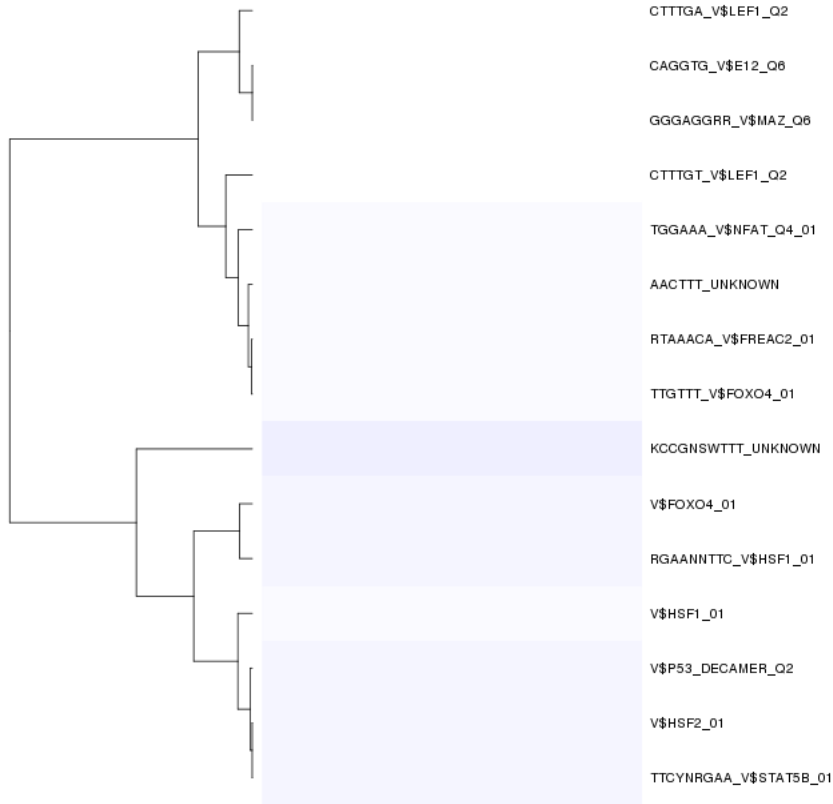
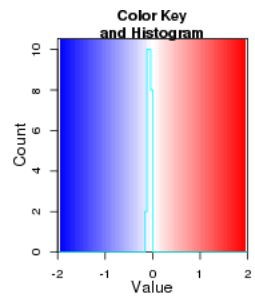


TF enrichment:



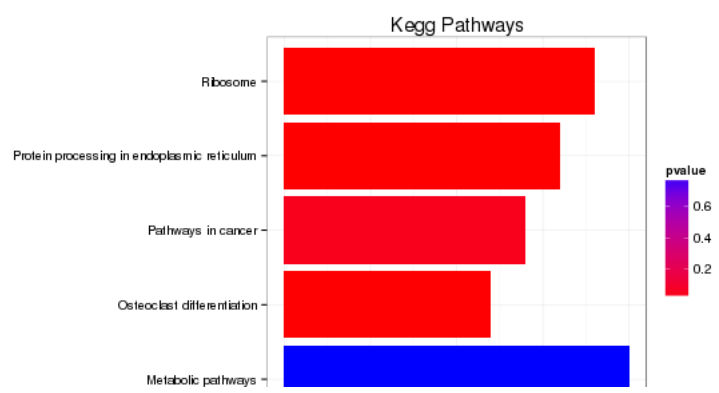
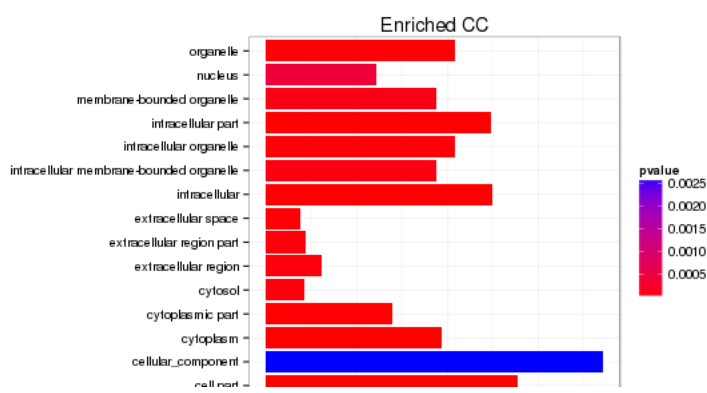
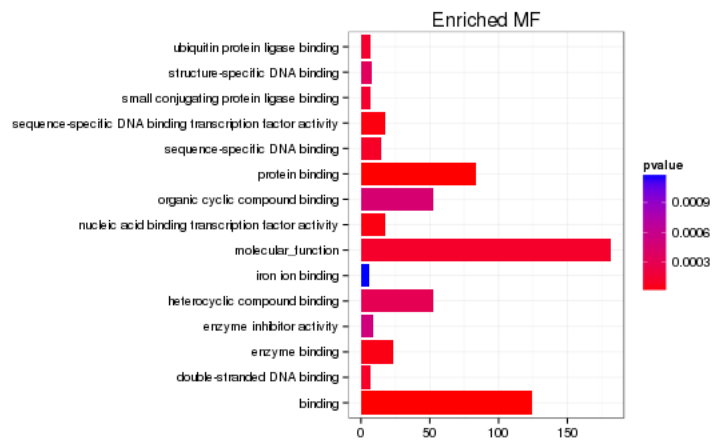
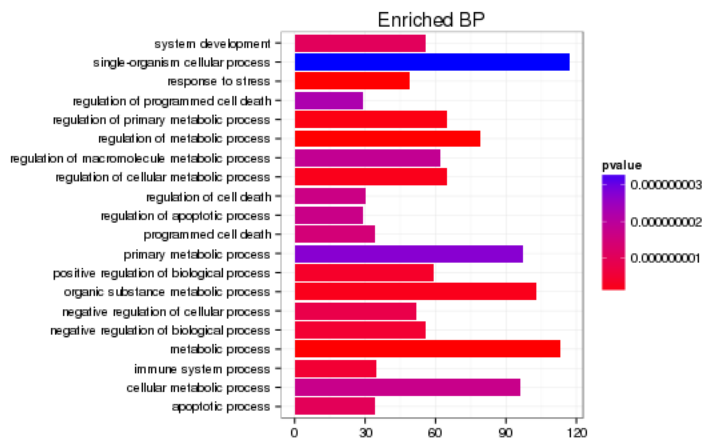


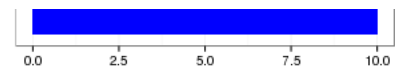
TF zscore:



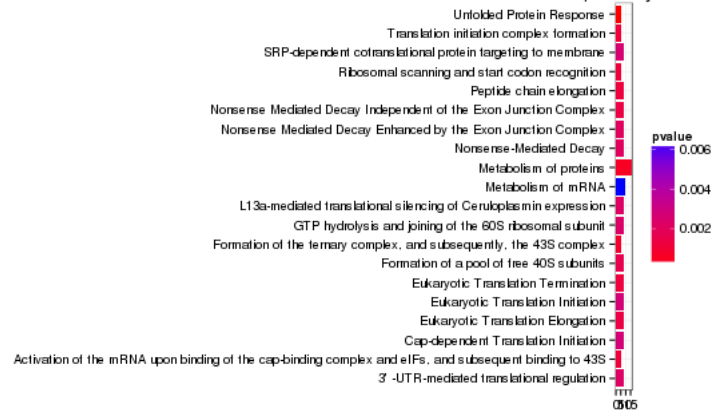
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

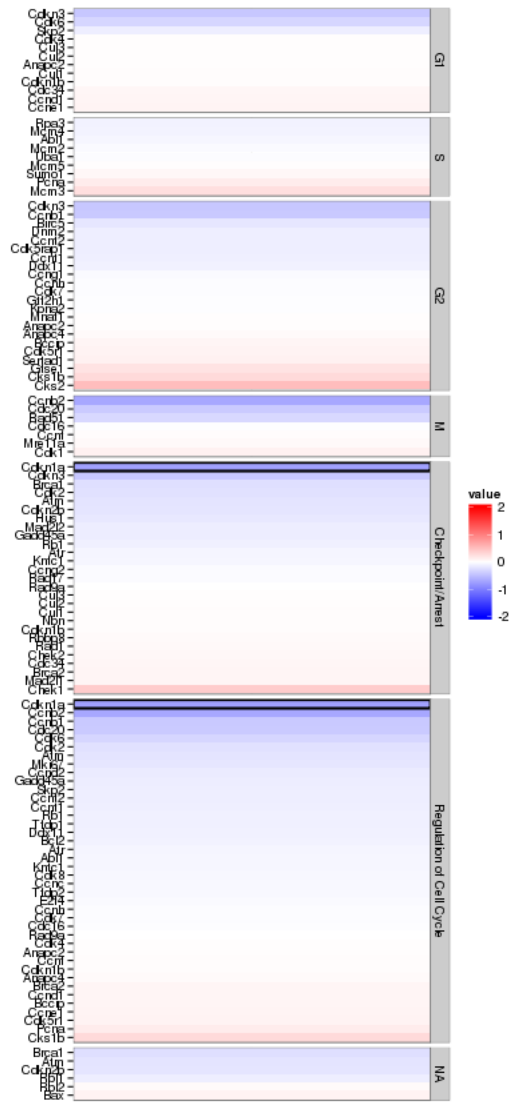




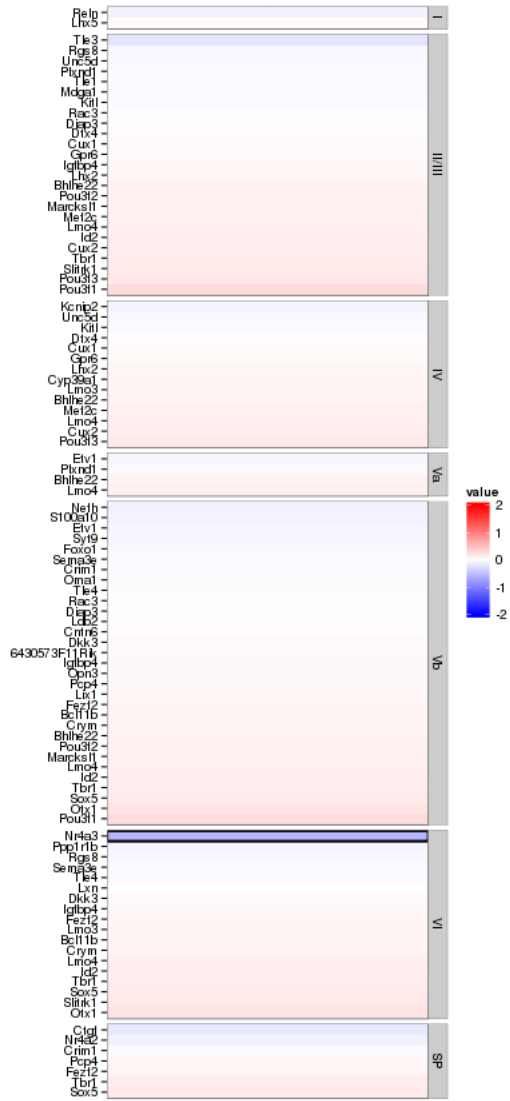
Reactome pathway enrichment



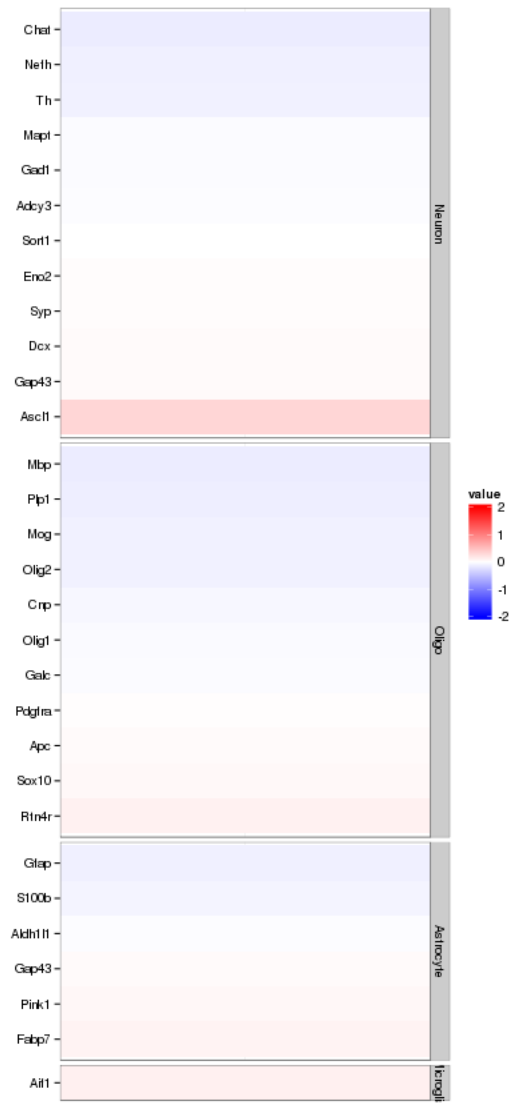
Enrichment or depletion for stage-specific cell cycle markers



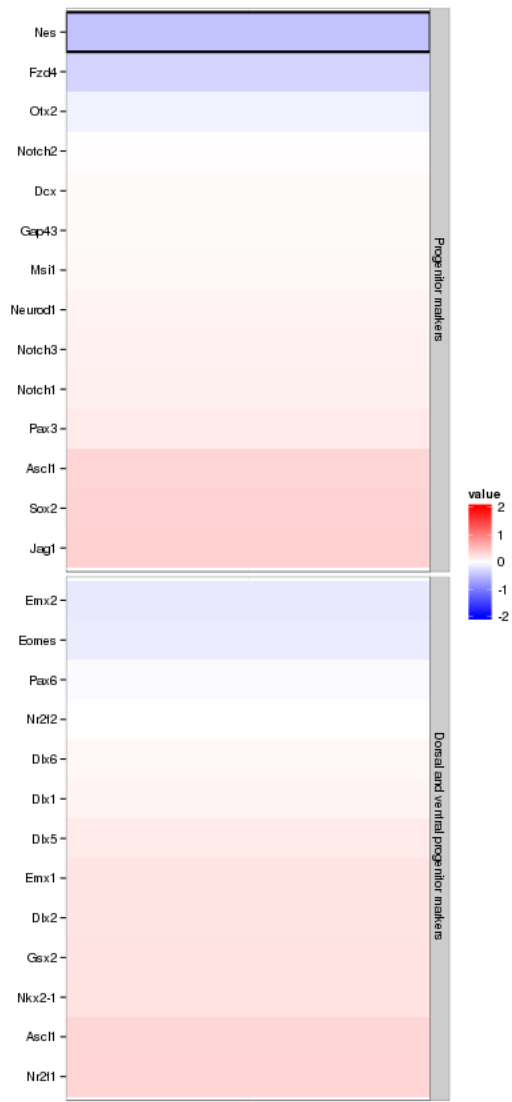
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



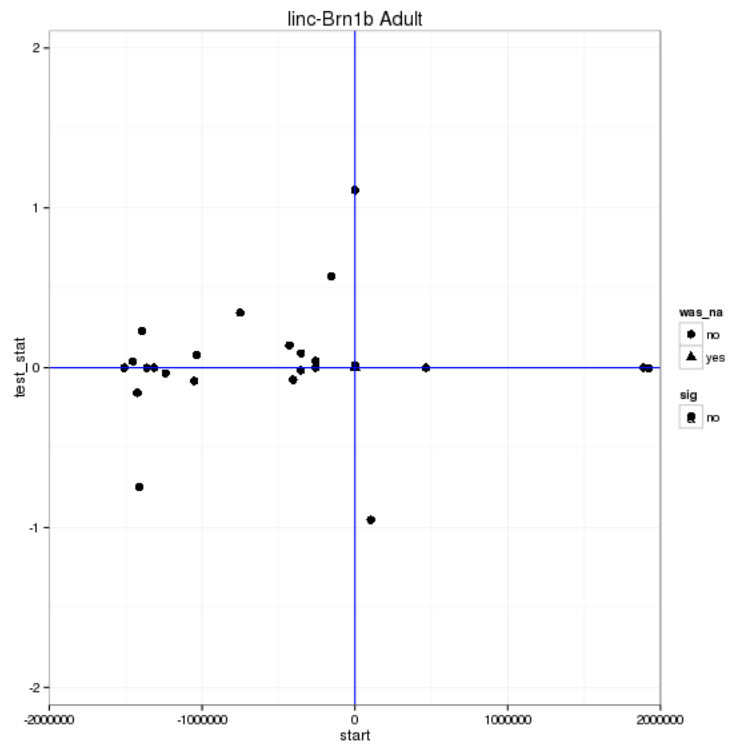
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR731
16 JR835
17 JR836

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34504400.00	0.76	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34504400.00	0.59	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34504400.00	0.98	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34504400.00	0.80	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34504400.00	1.04	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34504400.00	1.07	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34504400.00	0.81	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34504400.00	1.16	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34504400.00	1.01	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34504400.00	1.30	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34504400.00	1.51	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34504400.00	1.02	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34504400.00	0.98	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34504400.00	1.21	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR731/abundances.cxb	linc_Bm1b	0	linc_Bm1b_0	24288100.00	34504400.00	0.70	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR835/abundances.cxb	linc_Bm1b	1	linc_Bm1b_1	44561200.00	34504400.00	1.30	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR836/abundances.cxb	linc_Bm1b	2	linc_Bm1b_2	43675800.00	34504400.00	1.27	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,linc-Brn1b -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/linc-Brn1b_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainV
## 2
## 3
## 4
## 5
```

linc-Brn1b KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for linc-Brn1b.

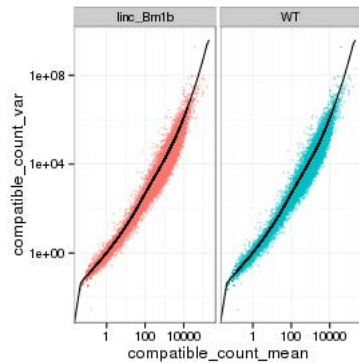
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

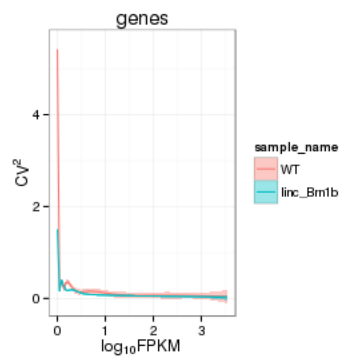
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



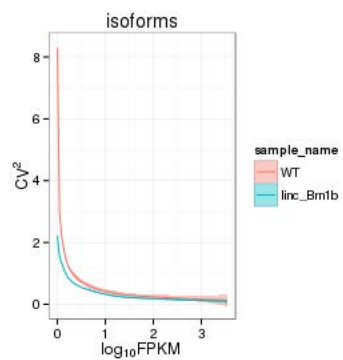
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

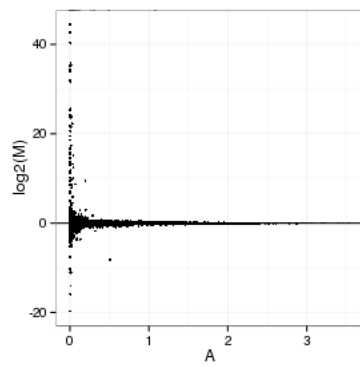
Genes:



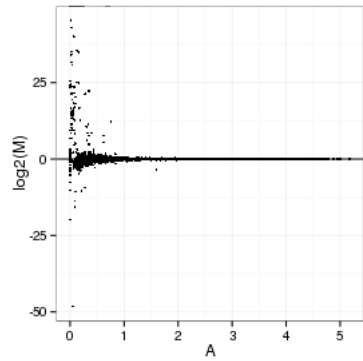
Isoforms:



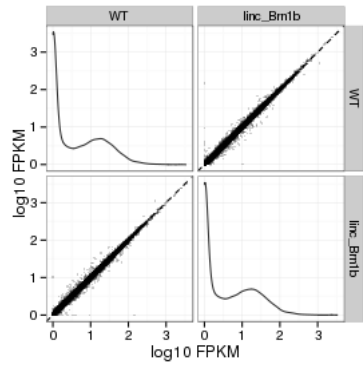
MvA plot



MvA plot counts



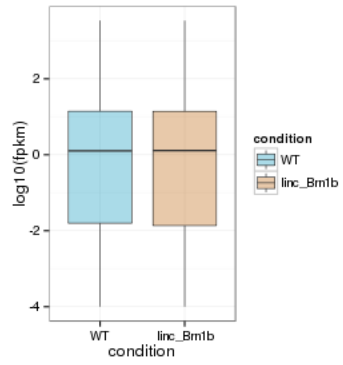
Scatterplot matrix



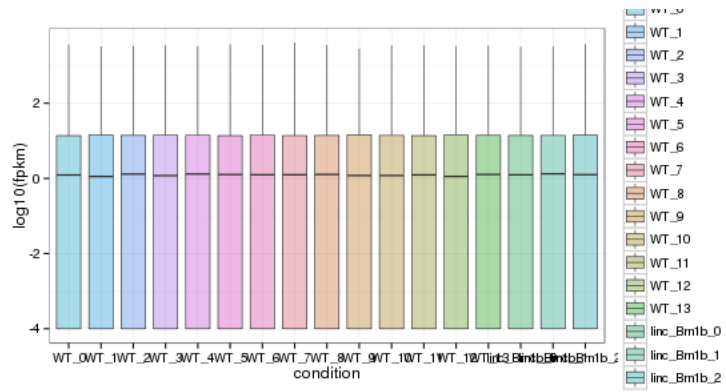
Distributions

Boxplots

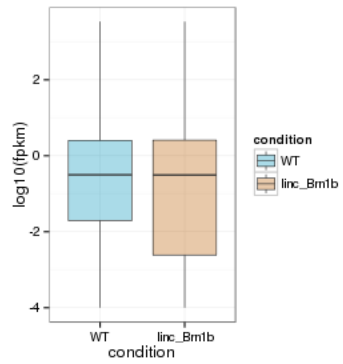
Boxplot (genes)



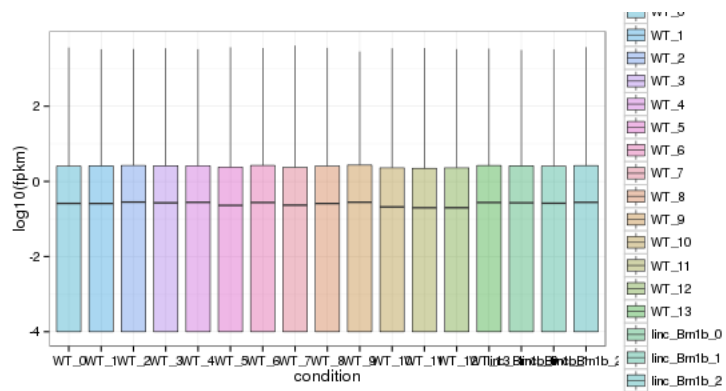
Boxplot (genes, replicates)



Boxplot (isoforms)

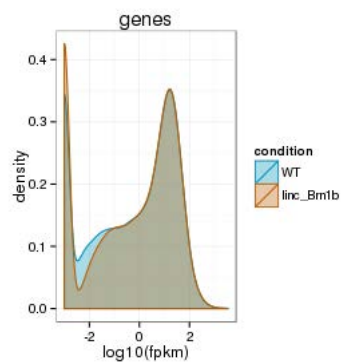


Boxplot (isoforms, replicates)

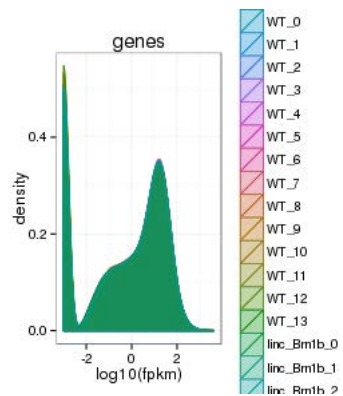


Density

Density (genes)

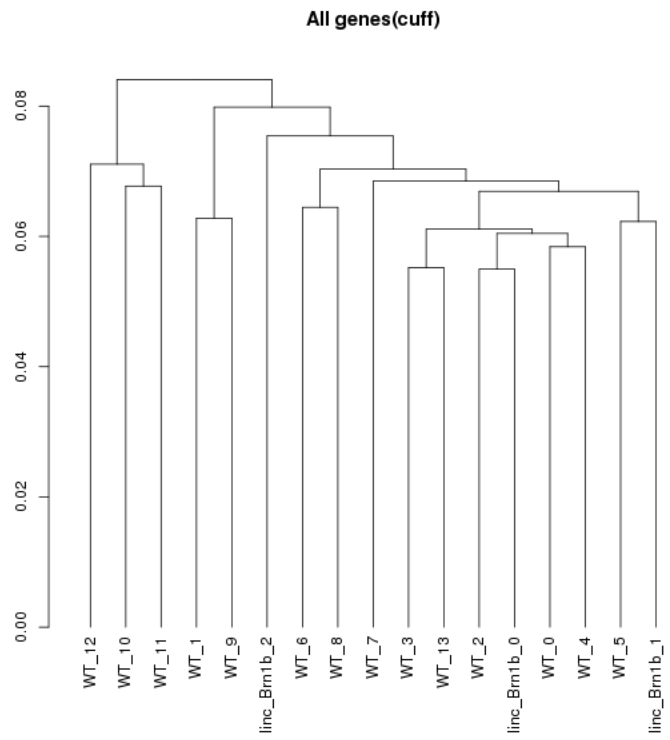


Density (genes, replicates)



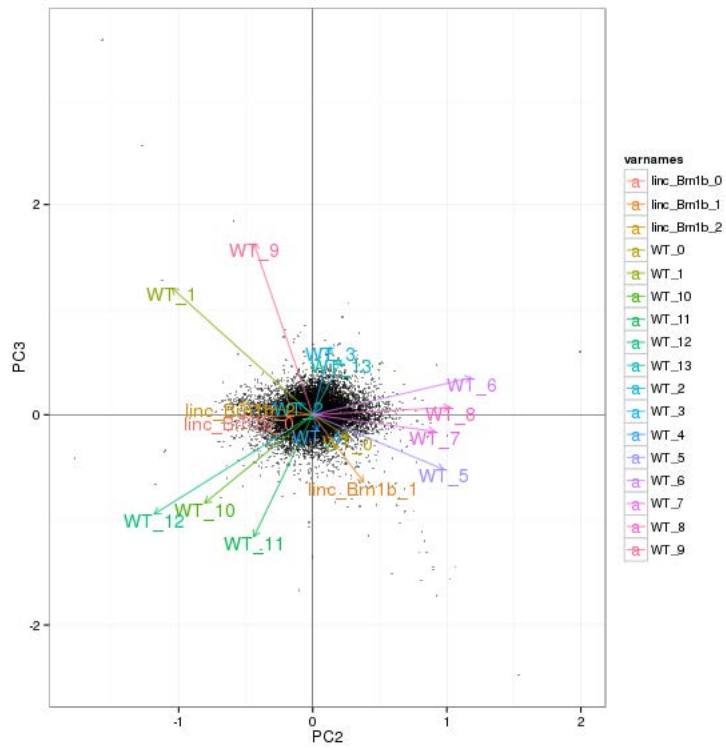
Clustering

Replicate Clusters

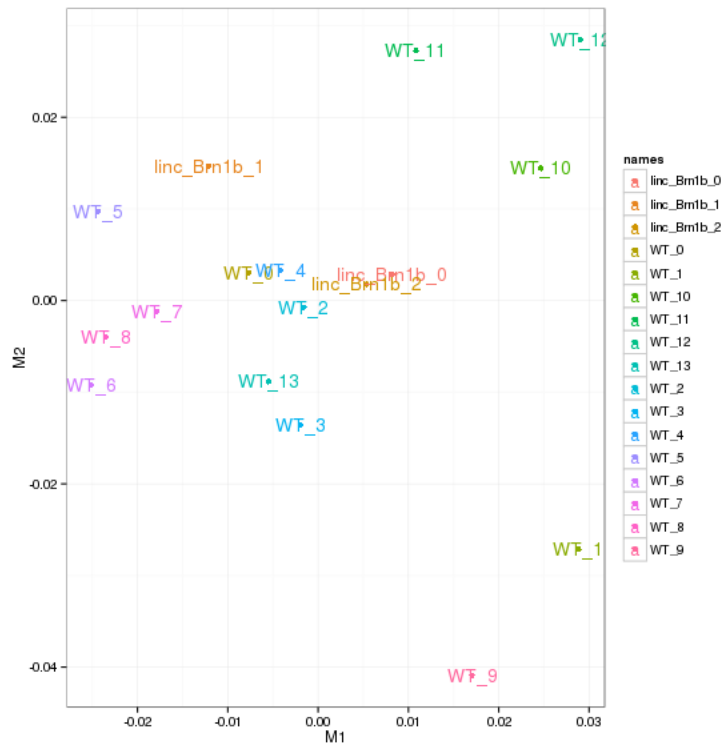


NULL

PCA (genes)

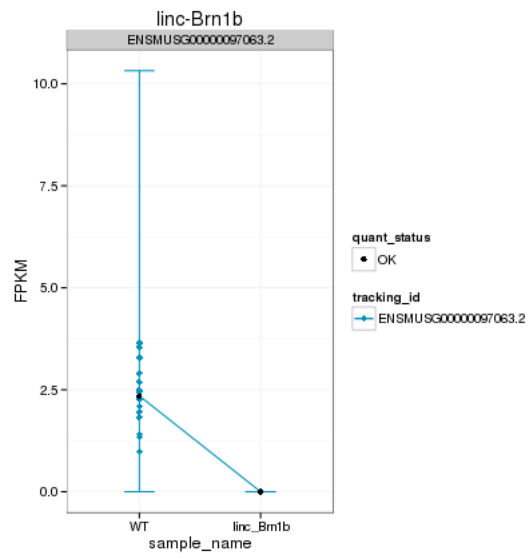


MDS (genes)

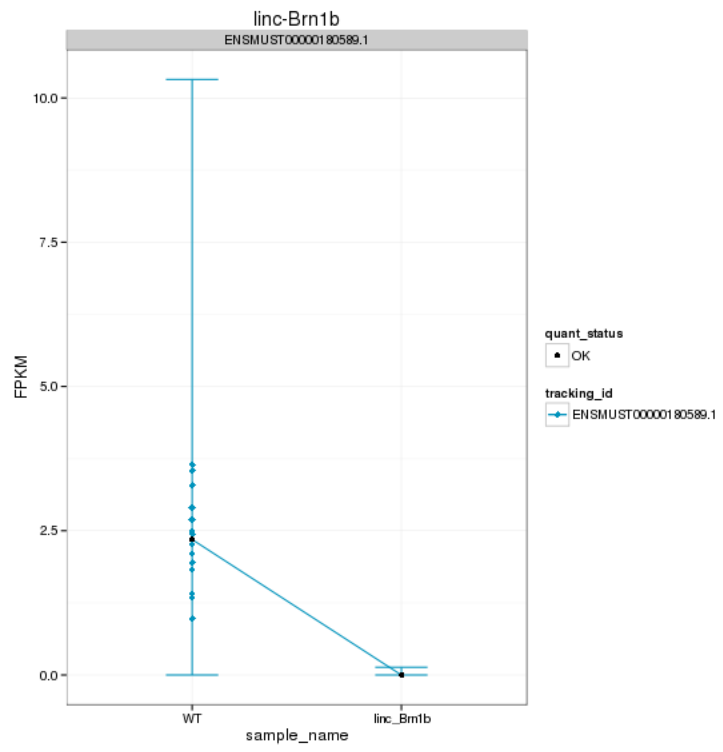


KO assessment

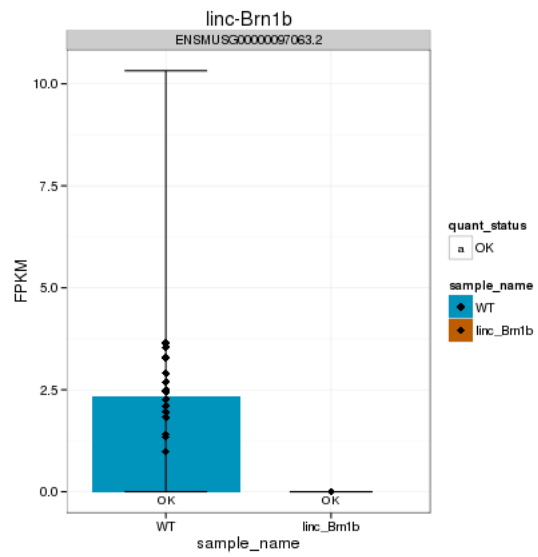
Endogenous lincRNA expression



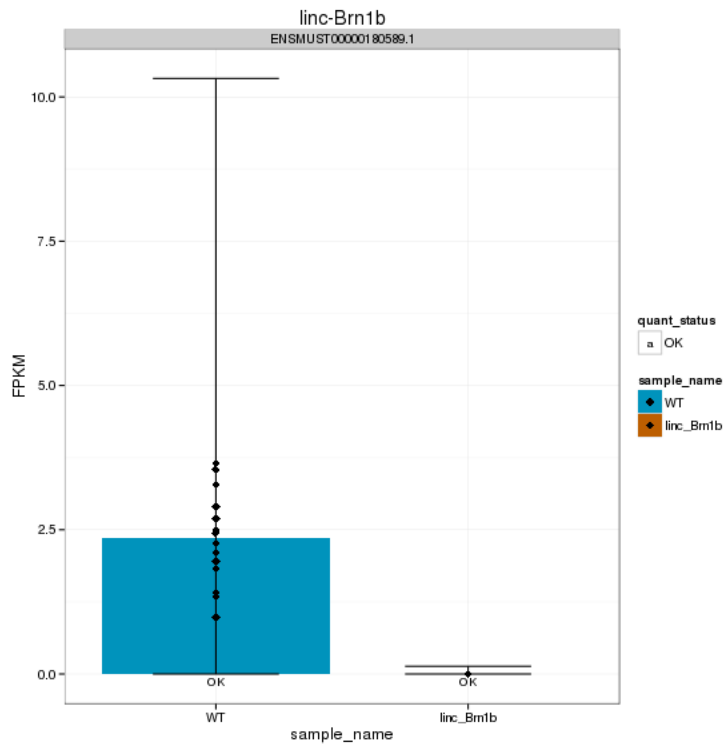
Endogenous expression of linc-Brn1b isoforms:



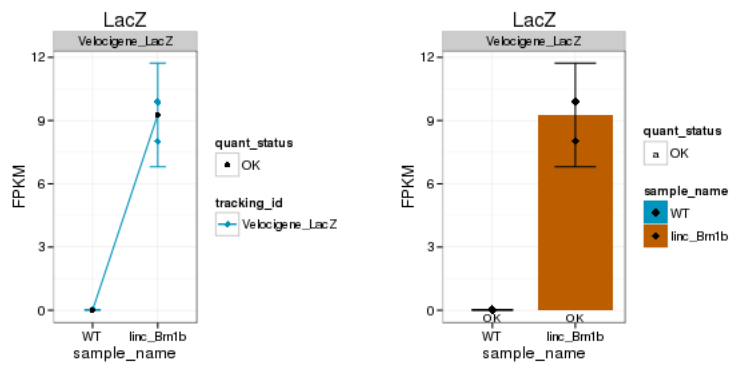
Barplot of gene expression:



Barplot of isoform expression:

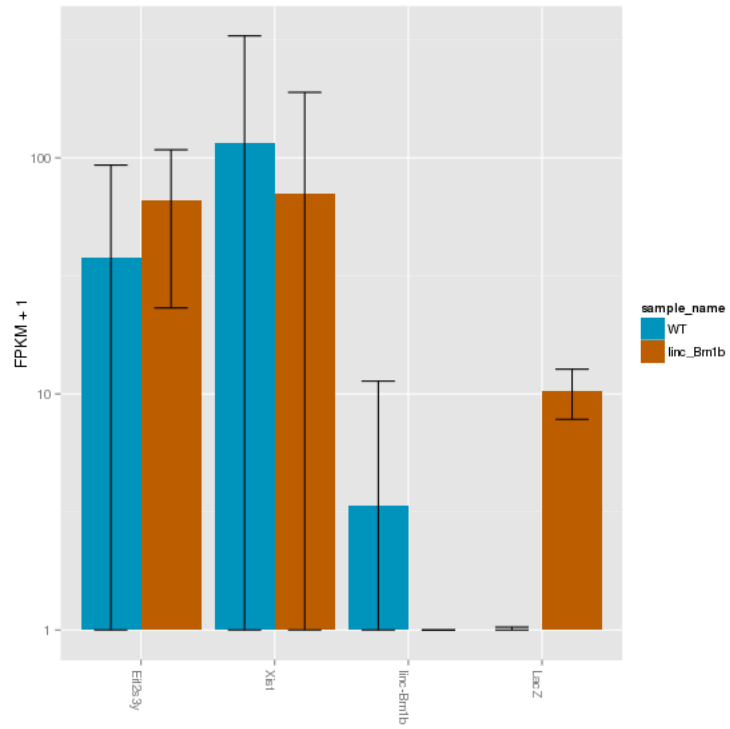


LacZ expression

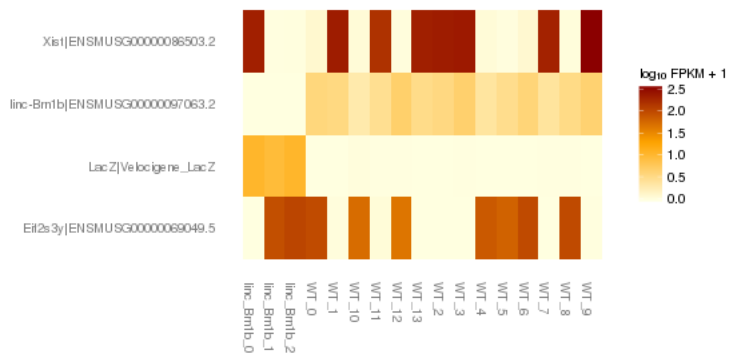


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

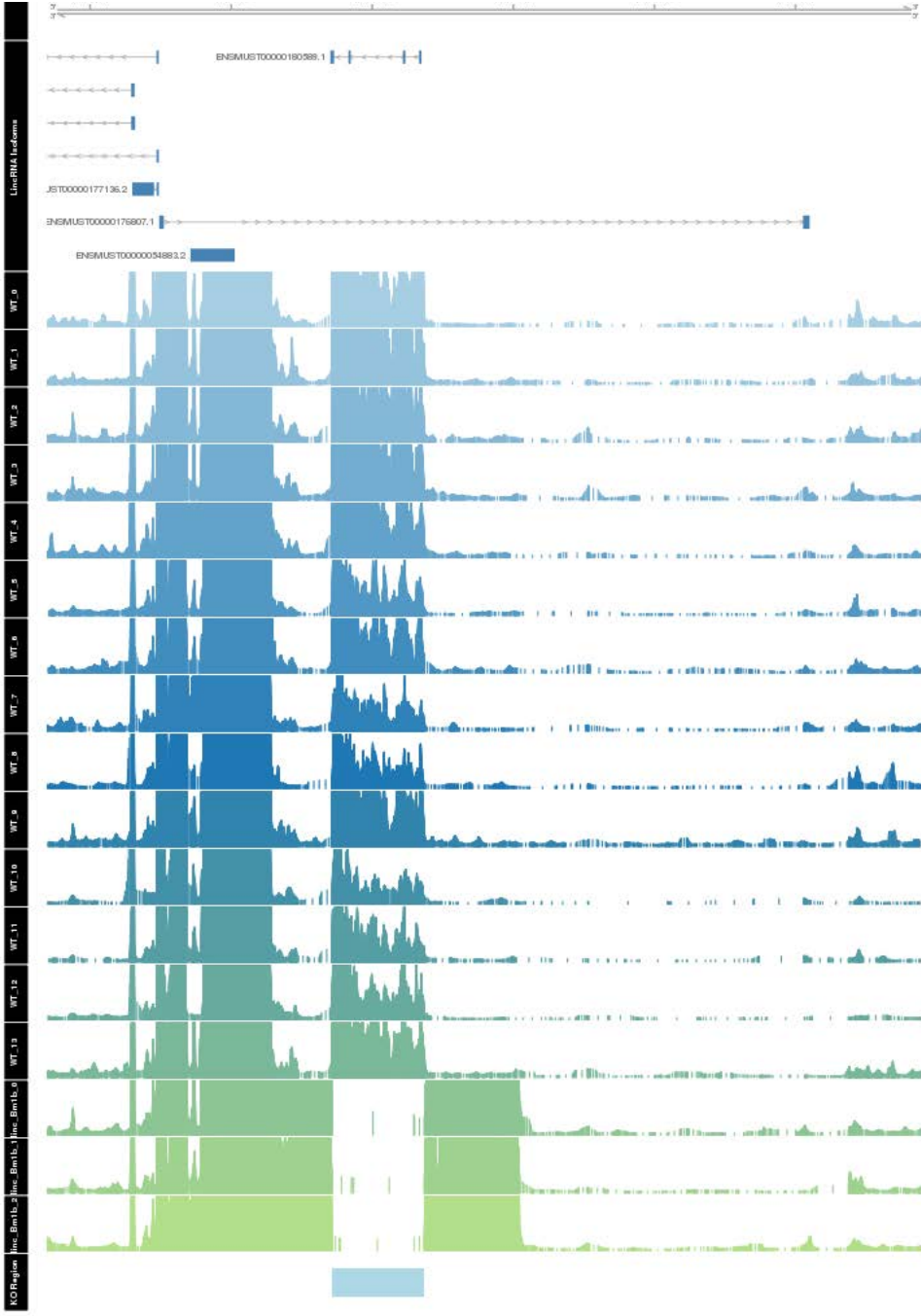


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 130 significantly differentially expressed genes. They are:

geneAnnot\$gene_short_name

- 1 Gira1
- 2 Sez6
- 3 Nfix
- 4 Grik3
- 5 Dlgap1
- 6 Cacna1e
- 7 Pax2
- 8 Sst
- 9 Crabp2
- 10 Ndrg1
- 11 Mef2c
- 12 Syngn3
- 13 Syn2
- 14 Prox1
- 15 Ebf3
- 16 Phox2b
- 17 Ptgds
- 18 Hivep2
- 19 Rxrg
- 20 Celsr1
- 21 Lhx1
- 22 Kitl
- 23 Gria1
- 24 Fam49a
- 25 Foxg1
- 26 Tfp2a
- 27 Fezf2
- 28 Skor1
- 29 Sla
- 30 Sncg
- 31 Dlx2
- 32 Prph
- 33 Tiam2
- 34 Lbx1
- 35 Nnt
- 36 Msx3
- 37 Asb1

38 Plxna2
39 Lhx6
40 Syt13
41 Chst1
42 Lamp5
43 Ntsr1

44 Hsd3b6
45 Nr4a3
46 Tal2
47 Ppp2r2c
48 Fosl2
49 Nos1
50 Mmp17
51 Uncx
52 Rph3a
53 Dlx5
54 Dync1i1
55 Rasgef1a
56 Hdcd3
57 Mfge8
58 Tbx20
59 Thy1
60 Kirrel3
61 Dscaml1
62 Robo3
63 Camkv
64 Cntnap3
65 Gas7
66 Tbc1d4
67 Ttc28
68 4833424O15Rik
69 Gucy1a3
70 Snca

71 Arl4d
72 Tbr1
73 Lars2
74 Arx
75 Vgf
76 Kcnh3
77 Neurod6
78 Neurod2
79 Satb2
80 Ttk

81 Hoxb4
82 Slc6a5
83 Abcc8
84 Zfp704
85 Tlx3
86 Mpped1
87 Dlx1
88 Adamts3
89 Gm9493
90 D3Bwg0562e
91 Cnpy1
92 Prkce
93 Hpcal4
94 Olig1
95 Nxph3
96 Fat4
97 Mc4r

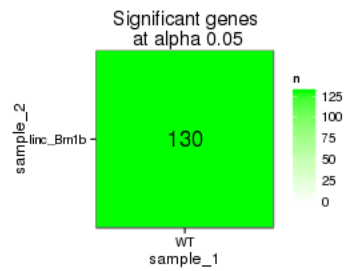
98 Pirt

99 Bcl11b
100 Pou4f1
101 Rpl29
102 Pla2g4e
103 Slc6a7
104 Hbb-y
105 Hbb-bh1
106 Adamts18
107 Hba-x
108 Pign
109 Kdm5d
110 Sez6l
111 Gda
112 Ppp1r1b
113 Trank1
114 Zbtb18
115 D430041D05Rik
116 Gpr88
117 Ptprz1
118 Ddx3y
119 Slc17a7
120 Gm9008
121 Wdfy1
122 Gm10709
123 Kcng1

- 124 Hs3st4
- 125 Dlx6os1
- 126 Myl6
- 127 Skor2
- 128 Lhx8
- 129 Gm26924
- 130 RP24-312G4.2

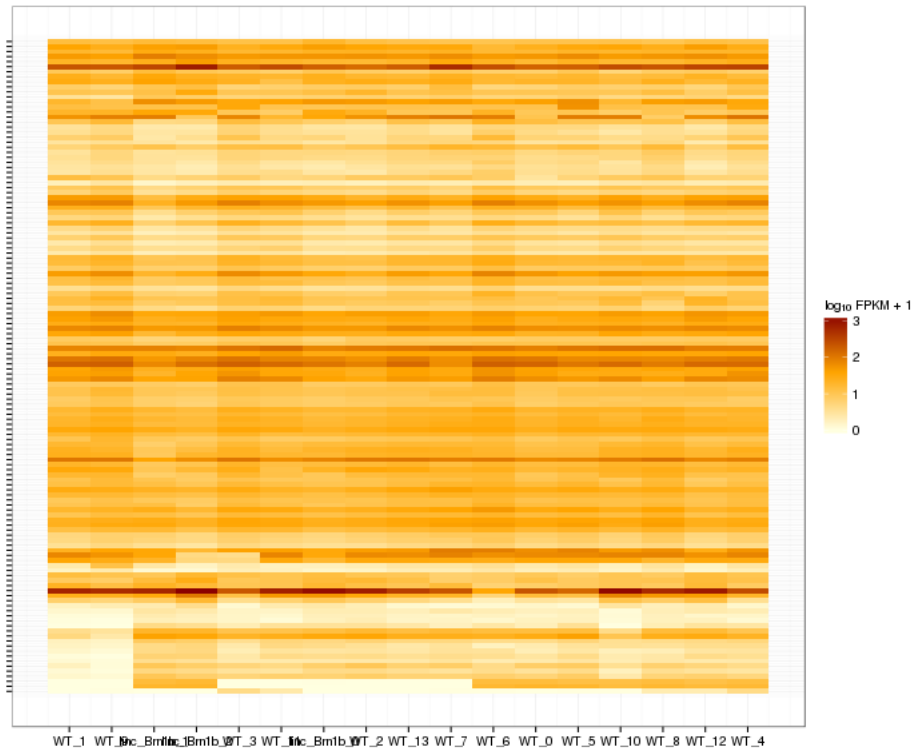
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

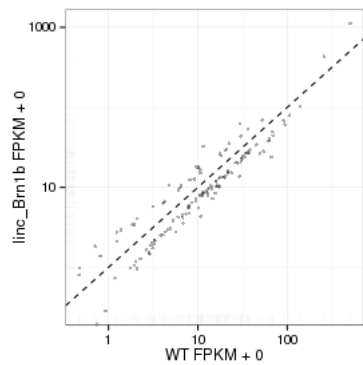


Significant genes with expression >50fpm (any condition):(turned off)

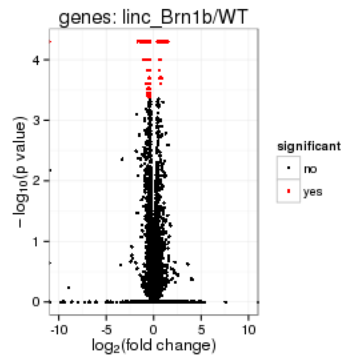
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

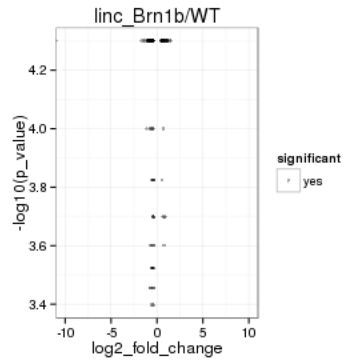
Scatter plot of significant genes only:



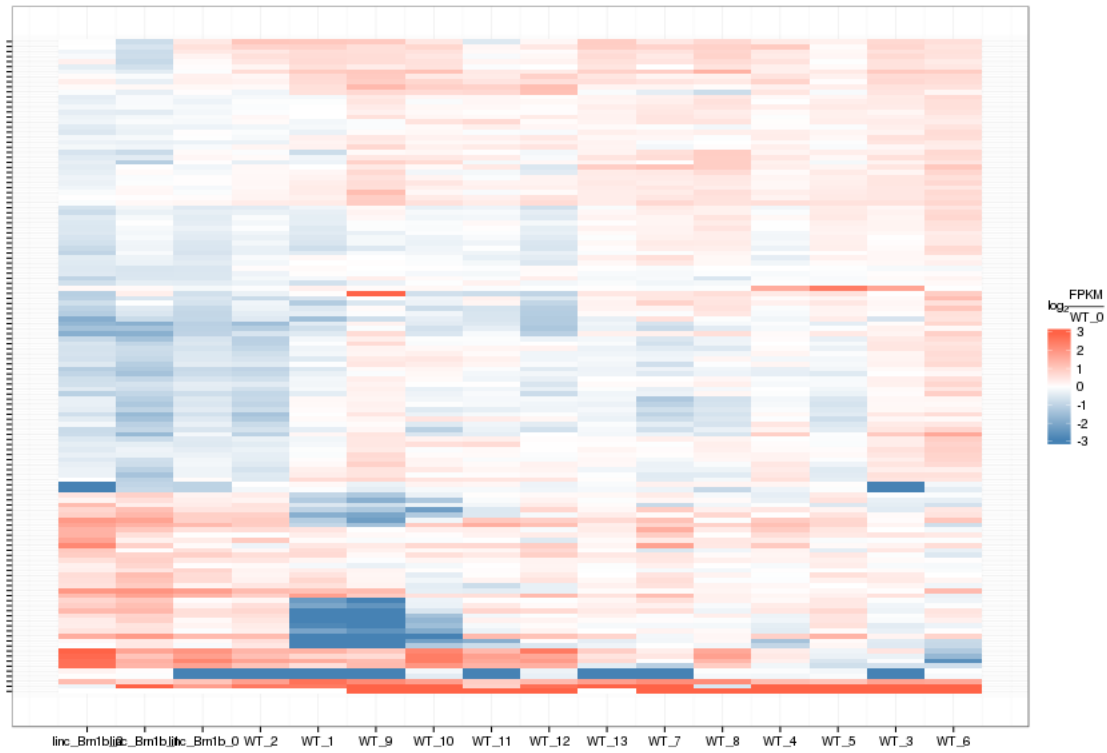
Volcano Plot



Volcano plot with significant genes only:



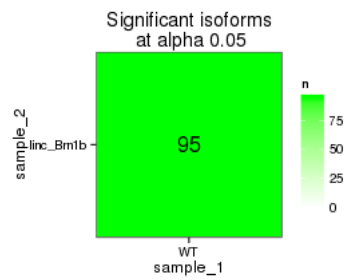
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

isoAnnot\$gene_short_name

- 1 Nfix
- 2 Grik3
- 3 Rec8
- 4 Crabp2
- 5 Ndrp1
- 6 Mef2c
- 7 Phox2b
- 8 Hivep2
- 9 Celsr1

- 10 Lhx1
- 11 Kitl
- 12 Foxg1
- 13 Fezf2
- 14 Sla
- 15 Sncg
- 16 Dlx2
- 17 Prph
- 18 Tiam2
- 19 Lbx1
- 20 Nnt
- 21 Lancl1
- 22 Ngef
- 23 Asb1
- 24 Lypd1
- 25 Lhx6
- 26 Syt13
- 27 Ntsr1
- 28 Tal2
- 29 Fosl2
- 30 Nos1
- 31 Dlx5
- 32 Dync111
- 33 Rasgef1a
- 34 Hddc3
- 35 Thy1
- 36 Kirrel3

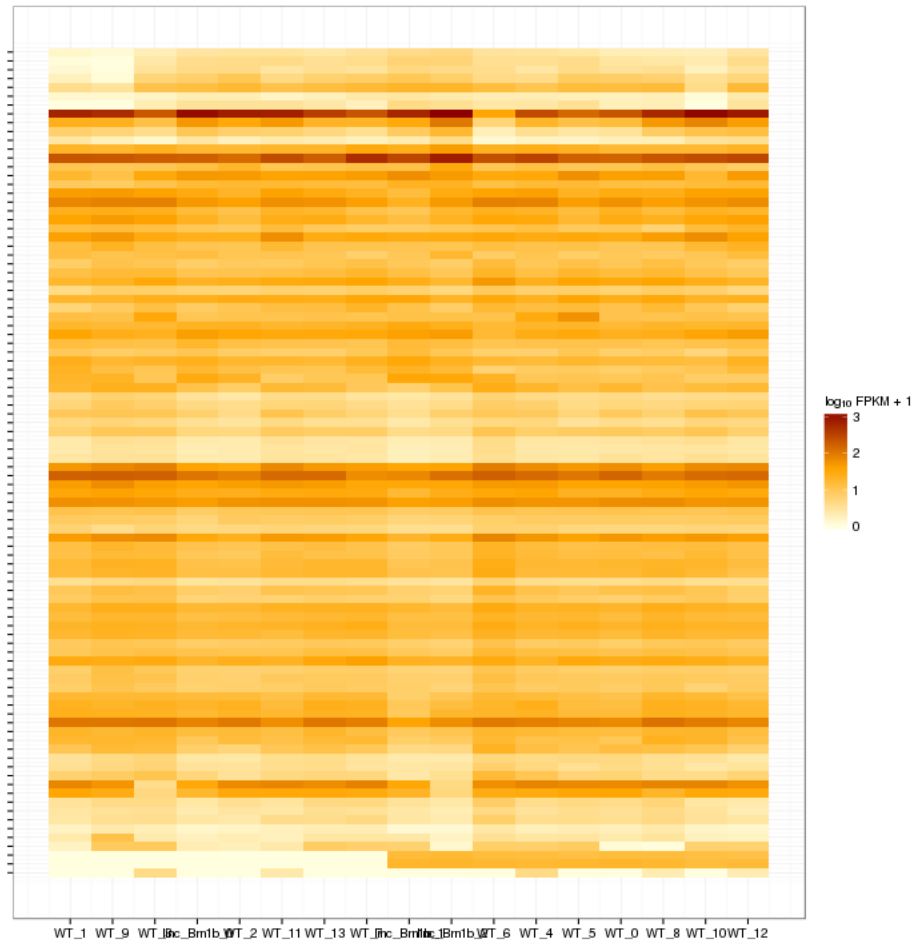
- 37 Dscaml1
- 38 Camkv
- 39 Gas7
- 40 Fndc3c1

41 Gucy1a3
42 Sncb
43 Arl4d
44 Tbr1
45 Lars2
46 Arx
47 Kcnh3
48 Neurod6
49 Sh3rf3
50 Satb2
51 Ttk
52 Hoxb4
53 Slc6a5
54 Abcc8
55 Tlx3
56 Sox5
57 Mpped1
58 Dlx1
59 Reln
60 Adamts3
61 Prkce
62 Olig1
63 Nxph3
64 Fat4

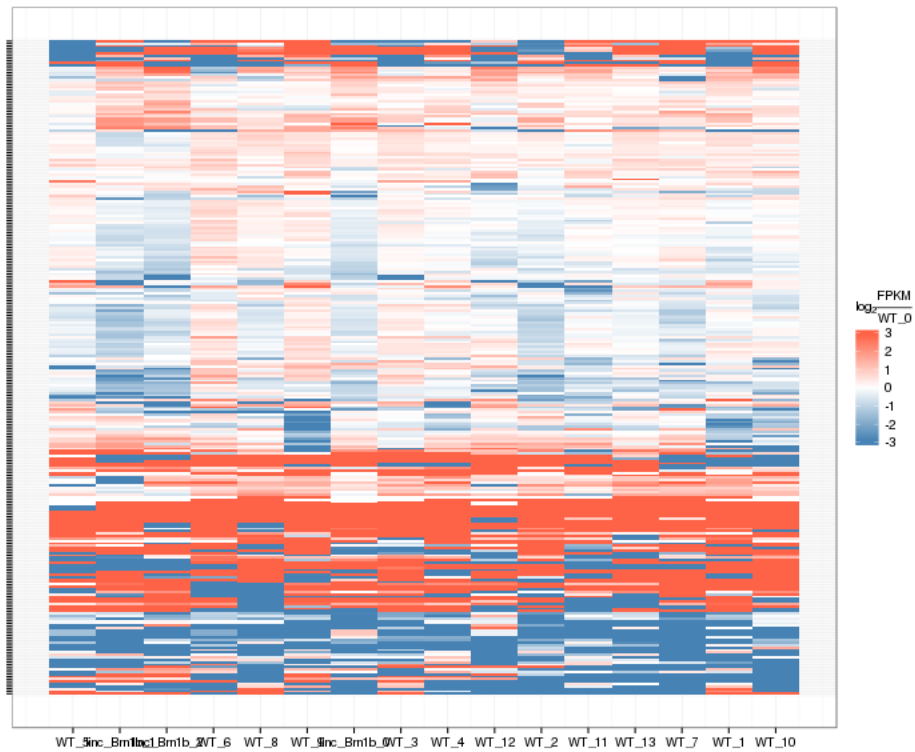
65 Mc4r
66 Pirt
67 Bcl11b
68 Pou4f1
69 Rpl29
70 Pla2g4e
71 Slc6a7
72 Hbb-y
73 Hbb-bh1
74 C130021120Rik
75 Adamts18
76 Hba-x
77 Kcnq3
78 Pign
79 Kdm5d
80 Gda
81 Opcml
82 Ptprz1
83 Ddx3y

84 Tmem132b
85 Slc17a7
86 Gm9008
87 D2hgdh
88 Wdfy1
89 Gm10709
90 Kcng1
91 Hs3st4
92 Myl6
93 Gm26924
94 RP24-312G4.2

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

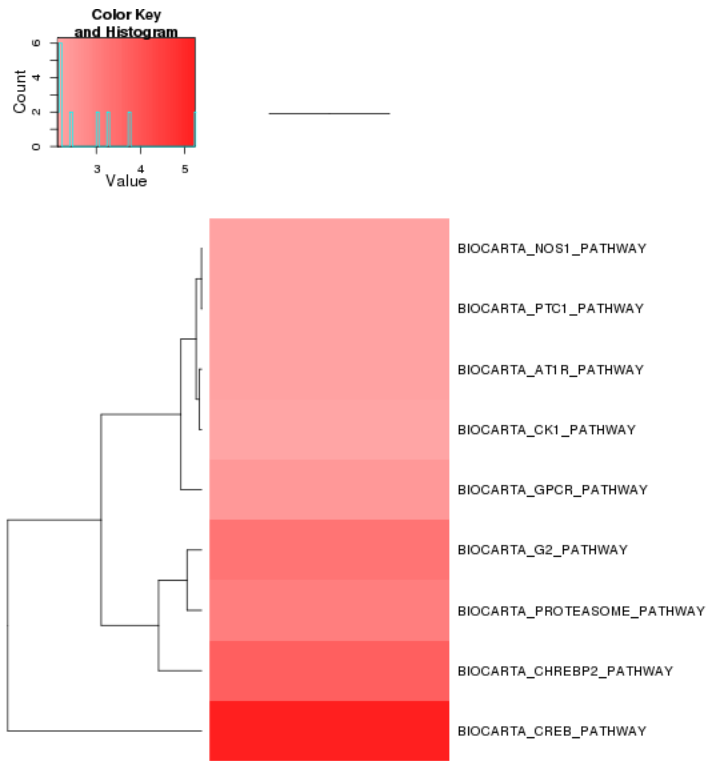
Gene/Pathway Analysis

GSEA

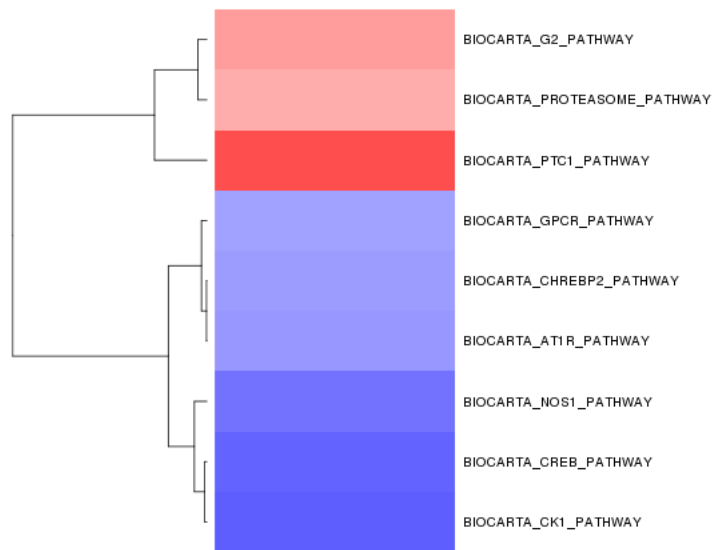
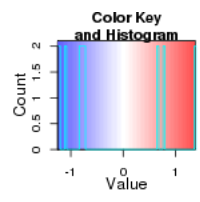
Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

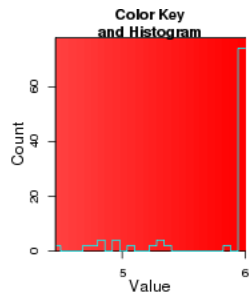
Biocarta enrichment:

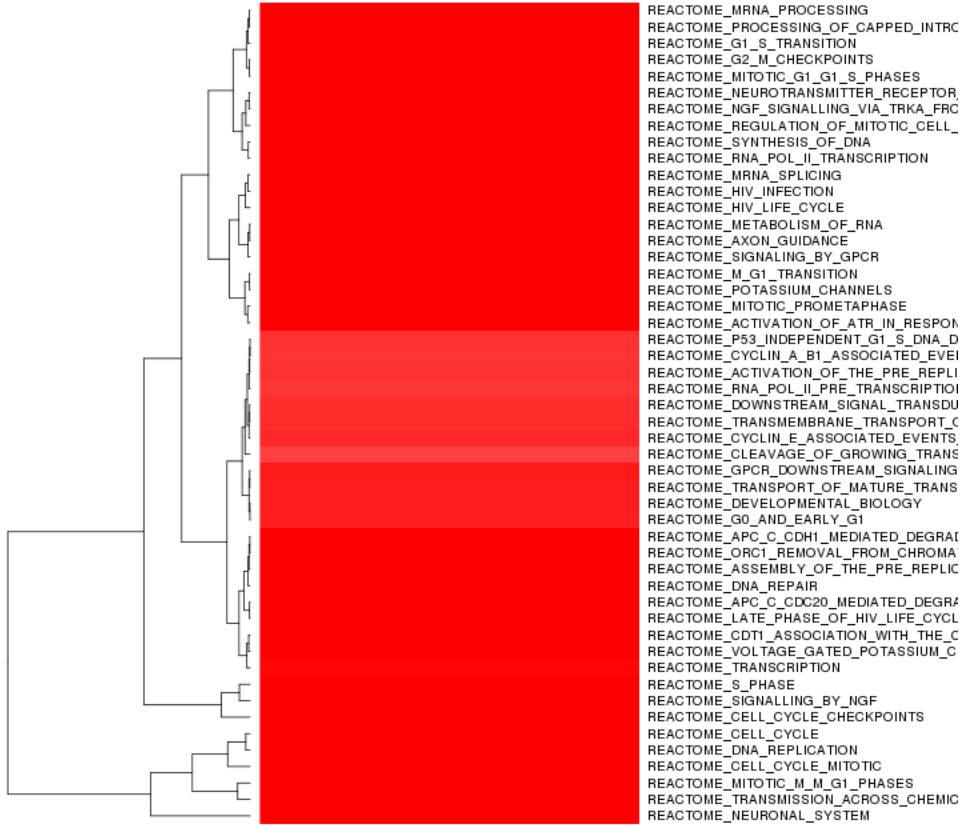


Biocarta zscore:

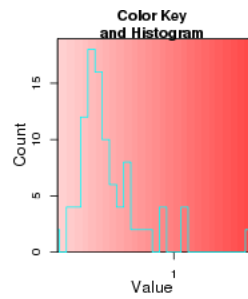


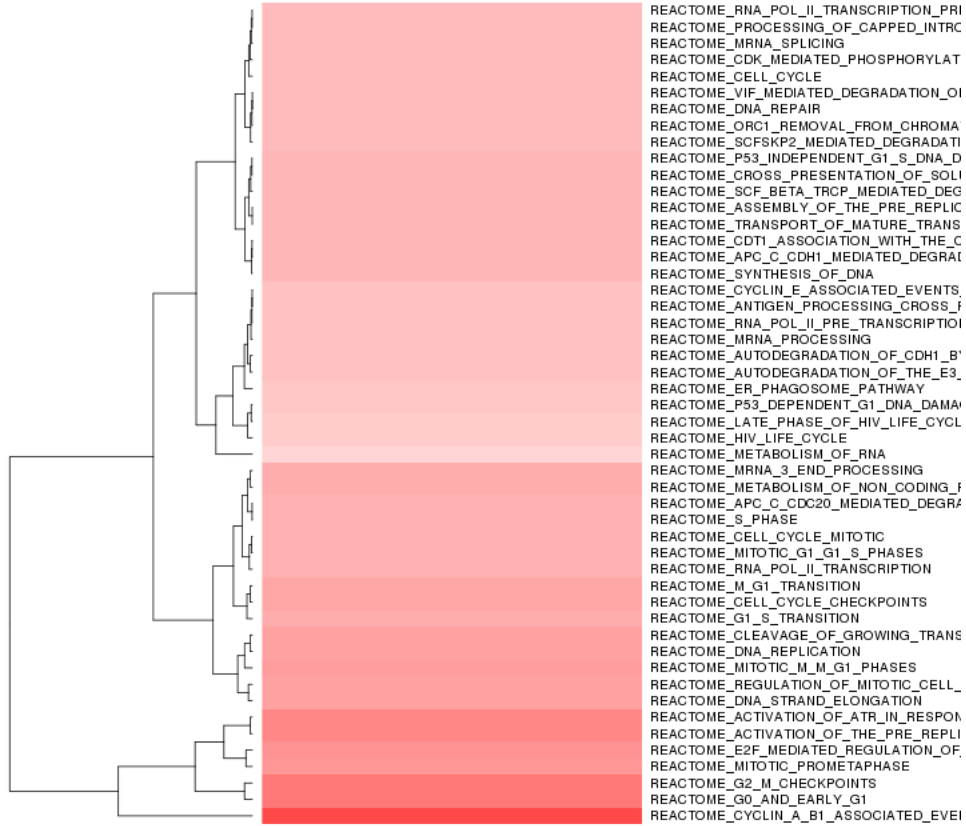
Reactome enrichment:



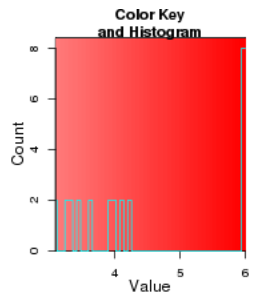


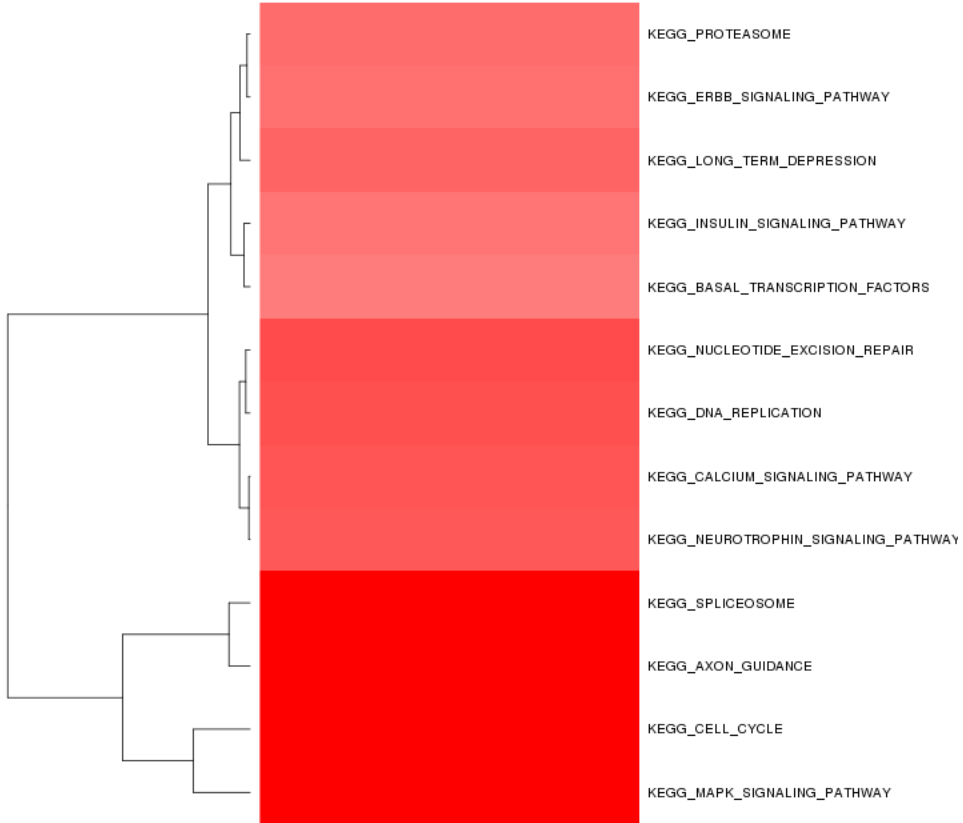
Reactome zscore:



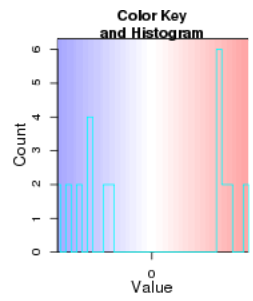


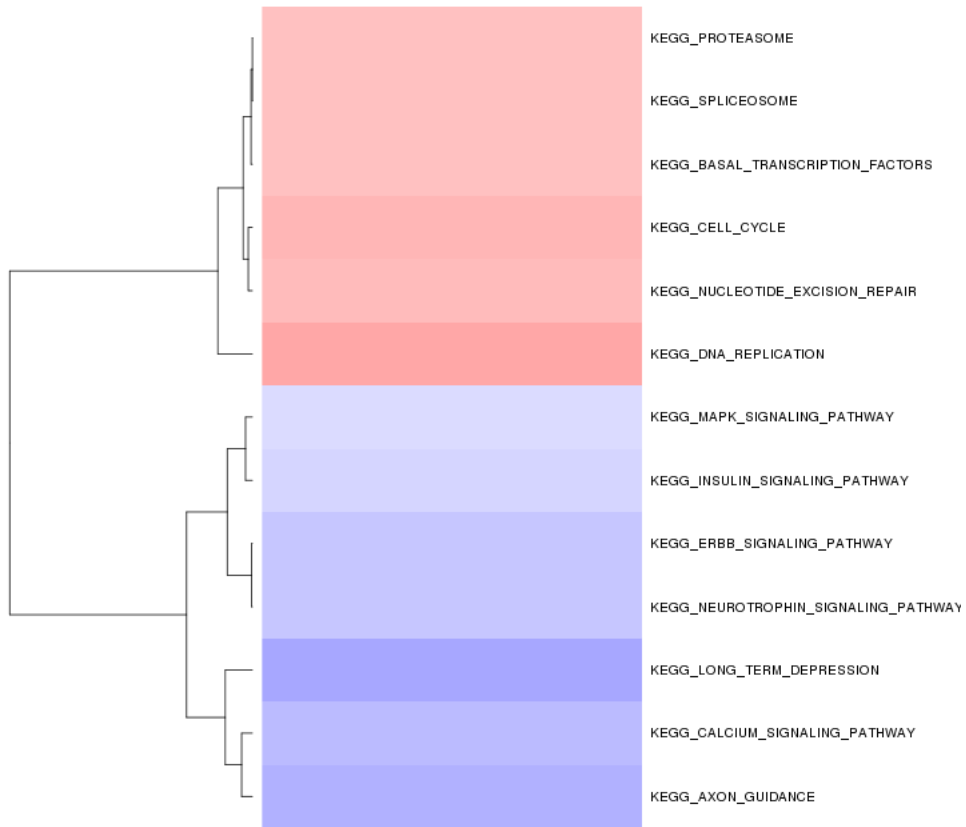
Kegg enrichment:





Kegg zscore:





Interneuron enrichment:

Error: argument is of length zero

Interneuron zscore:

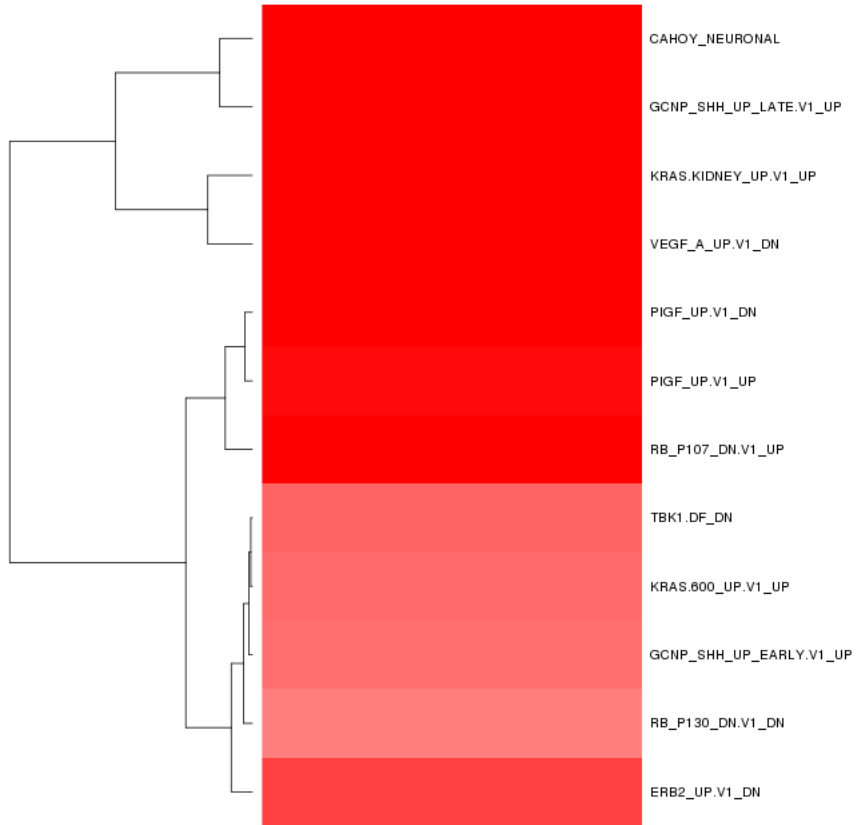
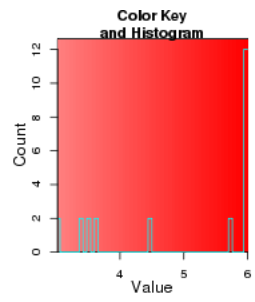
Error: incorrect number of dimensions

```
## Error: object 'x_ordered' not found
```

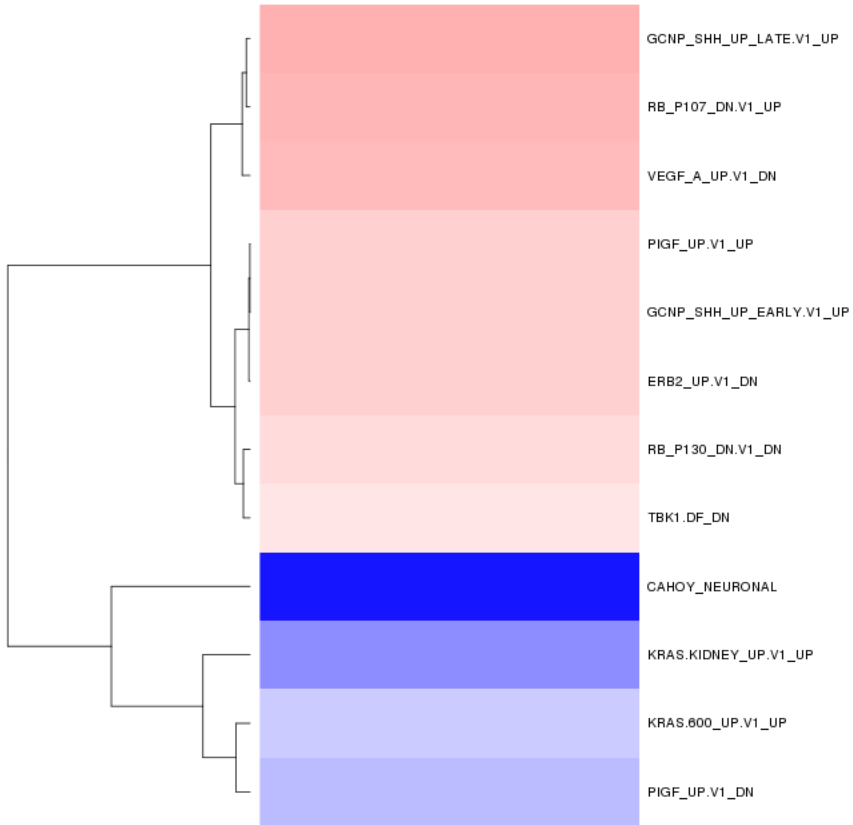
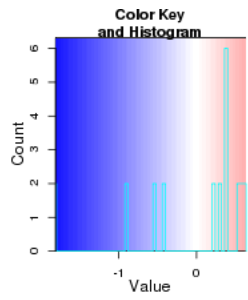
```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Oncogene enrichment:

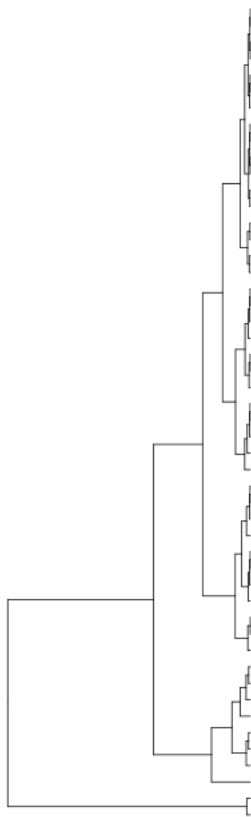


Oncogene zscore:



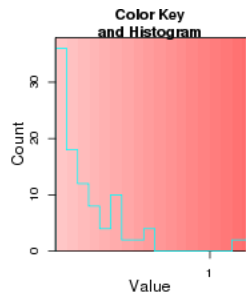
Immuno enrichment:

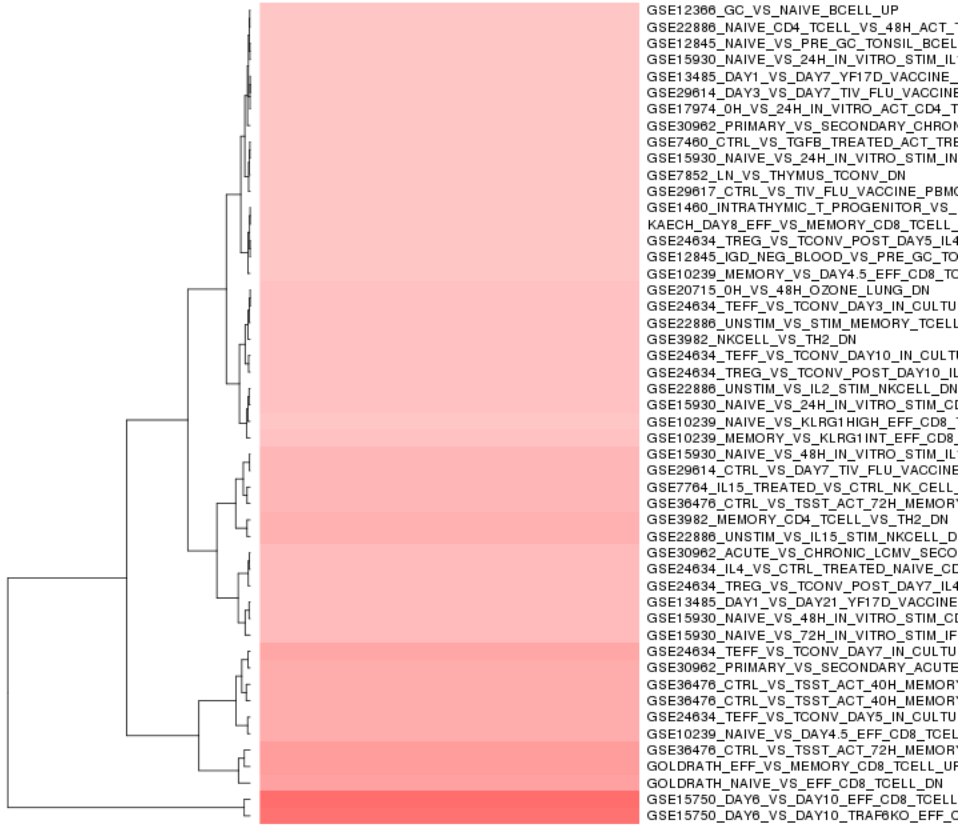
Error: no locations are finite



GSE19825_CD24LOW_VS_IL2RA_HIGH_DAY3_
GSE24634_IL4_VS_CTRL_TREATED_NAIVE_CI
GSE15930_STIM_VS_STIM_AND_IL12_24H_CD
GSE13485_DAY1_VS_DAY3_YF17D_VACCINE_
GSE30962_ACUTE_VS_CHRONIC_LCMV_SECO
GSE30962_PRIMARY_VS_SECONDARY_CHRON
GSE22886_UNSTIM_VS_IL2_STIM_NKCELL_DN
GSE13485_DAY1_VS_DAY7_YF17D_VACCINE_
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IF
GSE3982_NKCELL_VS_TH2_DN
GSE30962_ACUTE_VS_CHRONIC_LCMV_PRIMA
GSE24634_TREG_VS_TCONV_POST_DAY10_IL
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IN
KAECH_DAY8_EFF_VS_MEMORY_CD8_TCELL_
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IL
GSE15930_STIM_VS_STIM_AND_IFNAB_72H_C
GSE24634_NAIVE_CD4_TCELL_VS_DAY3_IL4_
GSE22886_UNSTIM_VS_STIM_MEMORY_TCELL
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IN
GSE7460_CTRL_VS_TGFB_TREATED_ACT_TRE
GSE24634_TREG_VS_TCONV_POST_DAYS_IL4
GSE15930_NAIVE_VS_72H_IN_VITRO_STIM_IF
GSE12845_NAIVE_VS_PRE_GC_TONSIL_BCEL
GSE10239_MEMORY_VS_KLRG1INT_EFF_CD8_
GSE24634_TEFF_VS_TCONV_DAY10_IN_CULTI
GSE22886_NAIVE_CD4_TCELL_VS_48H_ACT_
GSE22886_NEUTROPHIL_VS_DC_DN
GSE24634_TREG_VS_TCONV_POST_DAY7_IL4
GSE29617_CTRL_VS_TIV_FLU_VACCINE_PBMK
GSE1460_INTRATHYMIC_T_PROGENITOR_VS_
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IL
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_CI
GSE3982_MEMORY_CD4_TCELL_VS_TH2_DN
GSE10239_MEMORY_VS_DAY4.5_EFF_CD8_TC
GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR'
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_CI
GSE7764_IL15_TREATED_VS_CTRL_NK_CELL
GSE30962_PRIMARY_VS_SECONDARY_ACUTE
GSE24634_TEFF_VS_TCONV_DAY7_IN_CULTU
GOLDRATH_NAIVE_VS_EFF_CD8_TCELL_DN
GSE24634_TEFF_VS_TCONV_DAYS_IN_CULTU
GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR'
GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR'
GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR'
GSE10239_NAIVE_VS_DAY4.5_EFF_CD8_TCEL
GSE22886_UNSTIM_VS_IL15_STIM_NKCELL_D
GSE13485_DAY1_VS_DAY21_YF17D_VACCINE
GOLDRATH_EFF_VS_MEMORY_CD8_TCELL_UF
GSE15750_DAY6_VS_DAY10_TRAFKO_EFF_C
GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL

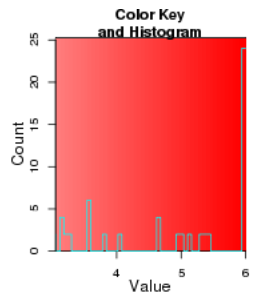
Immuno zscore:

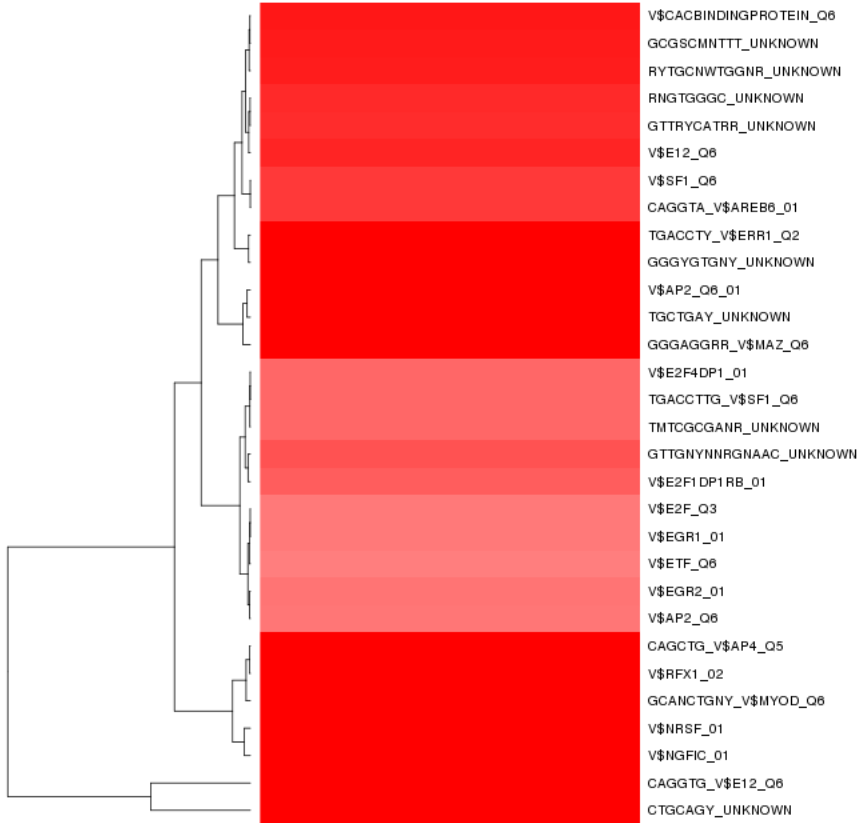




TF enrichment:

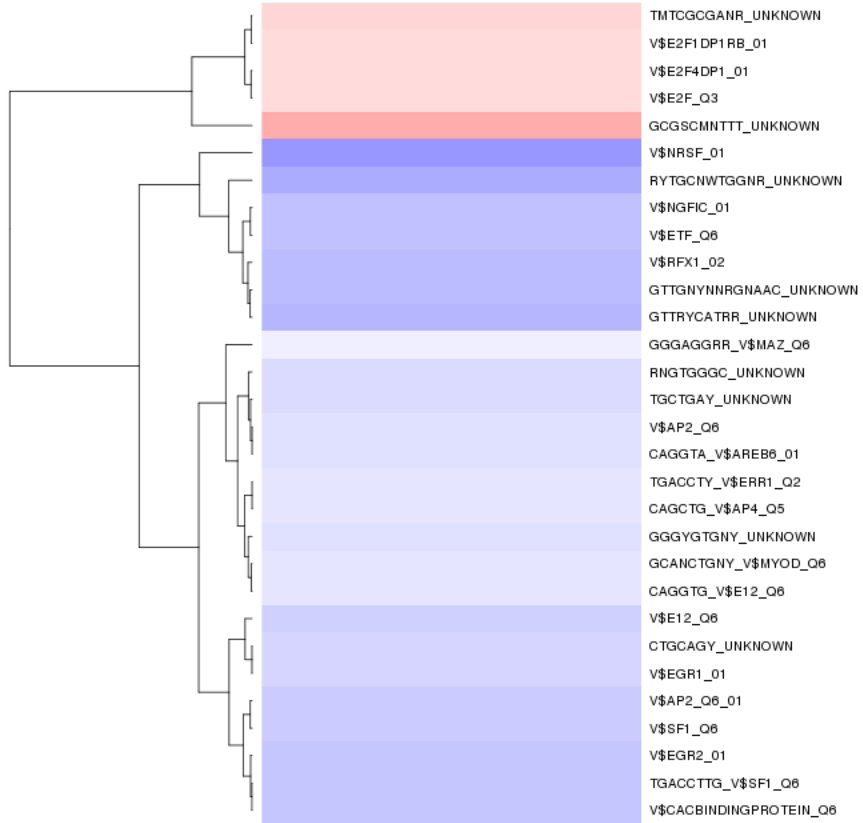
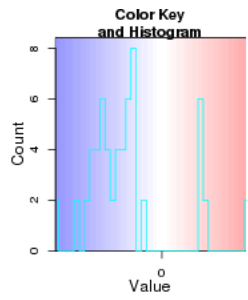
Error: subscript out of bounds





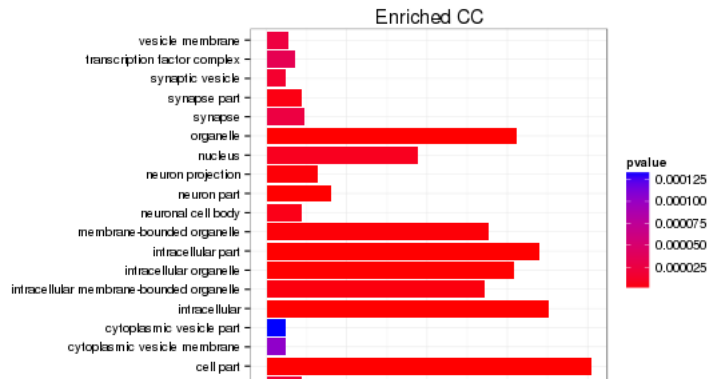
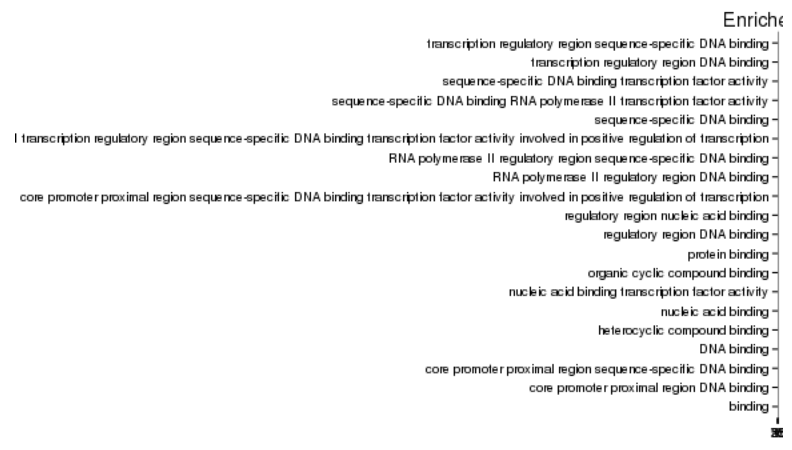
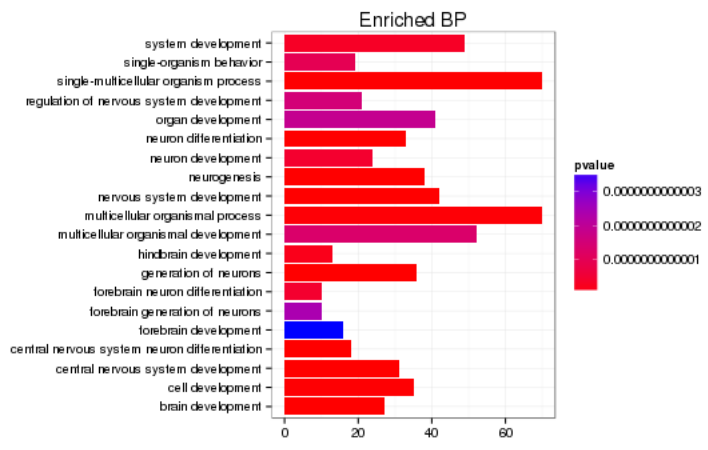
TF zscore:

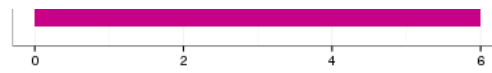
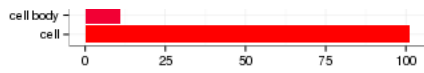
Error: subscript out of bounds



GO enrichment

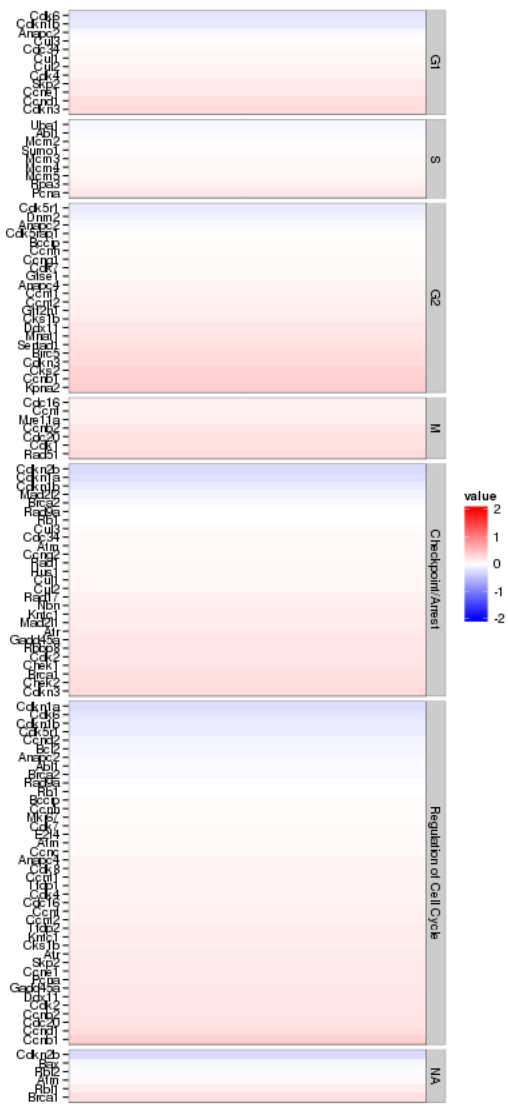
Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.



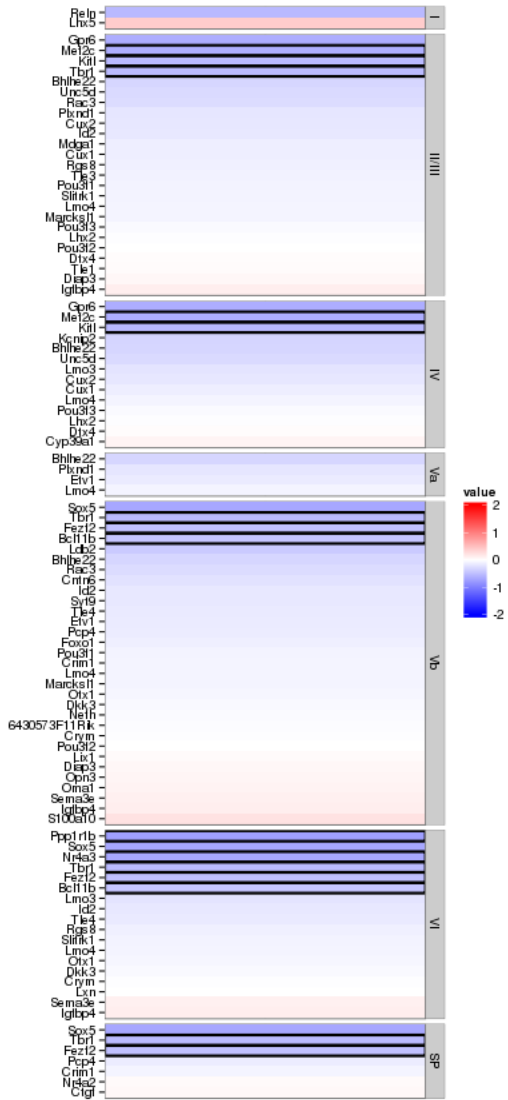


```
## Error: 'x' and 'units' must have length > 0
```

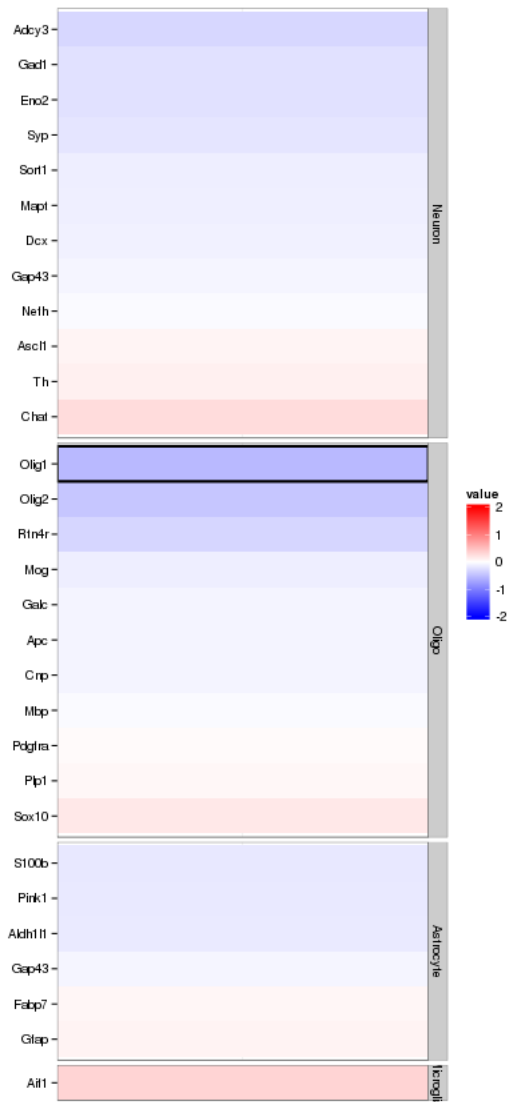
Enrichment or depletion for stage-specific cell cycle markers



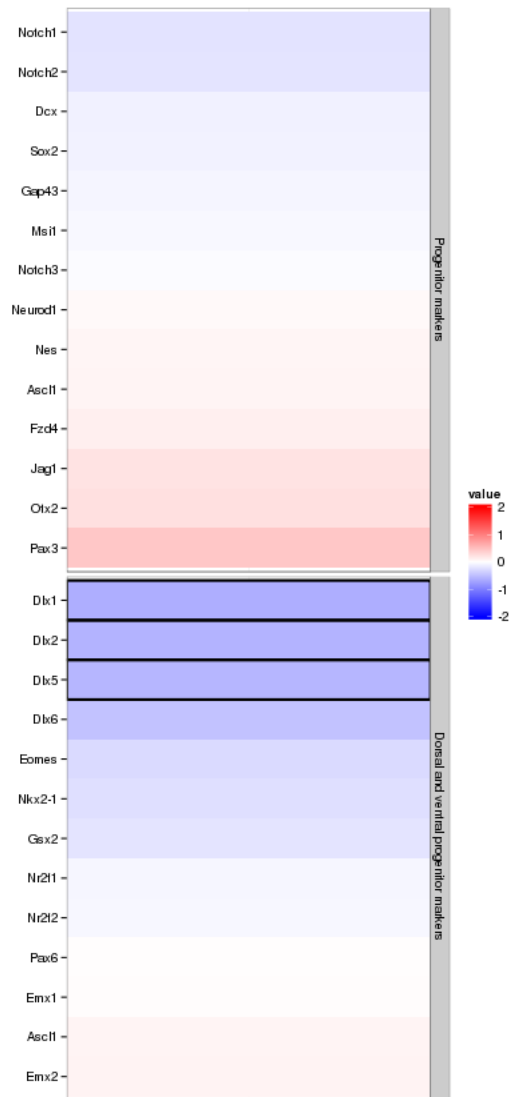
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



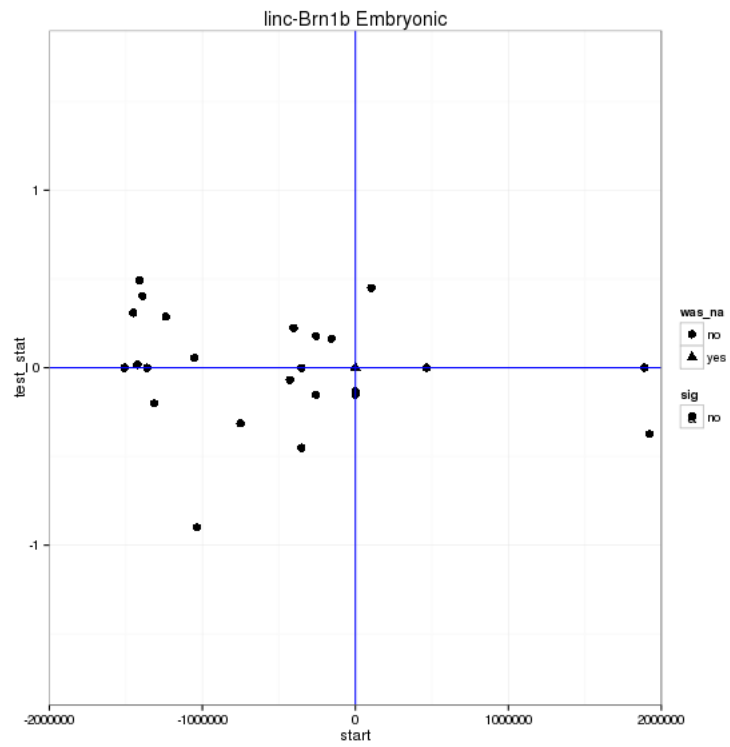
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR758
16 JR764
17 JR808

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	32398700.00	1.25	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	32398700.00	1.09	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	32398700.00	1.22	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	32398700.00	1.28	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	32398700.00	1.19	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	32398700.00	0.89	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	32398700.00	0.97	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	32398700.00	0.91	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	32398700.00	0.85	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	32398700.00	0.95	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	32398700.00	0.80	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	32398700.00	0.79	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	32398700.00	0.68	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	32398700.00	1.20	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR758/abundances.cxb	linc_Bm1b	0	linc_Bm1b_0	41595700.00	32398700.00	1.31	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR764/abundances.cxb	linc_Bm1b	1	linc_Bm1b_1	30294500.00	32398700.00	0.95	1.00
17 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR808/abundances.cxb	linc_Bm1b	2	linc_Bm1b_2	31223700.00	32398700.00	0.97	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igrph_0.7.1         KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,linc-Brn1b -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/linc-Brn1b_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/Br
## 2
## 3
## 4
## 5
```

linc-Cox2 KO vs WT (Adult)

This file shows the wt-v-ko comparison for linc-Cox2.

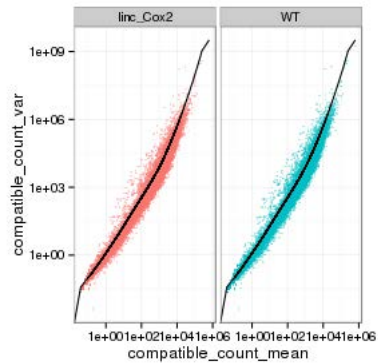
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

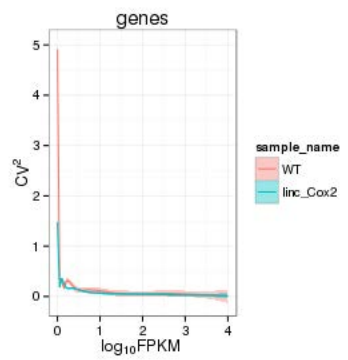
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



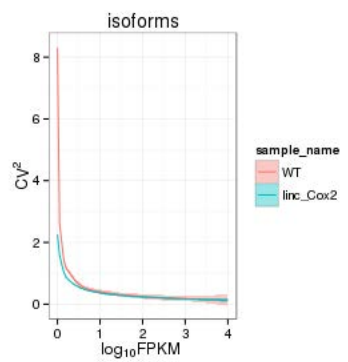
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

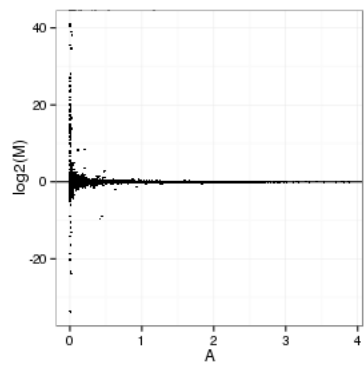
Genes:



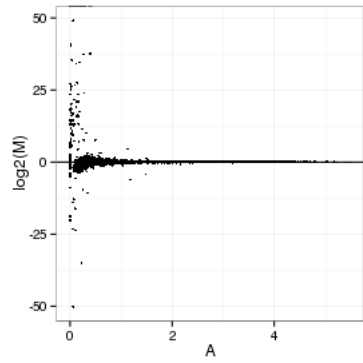
Isoforms:



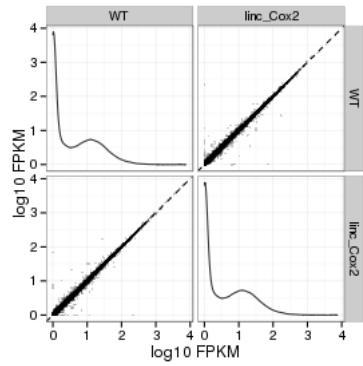
MvA plot



MvA plot counts



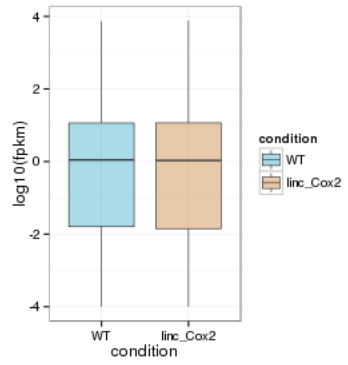
Scatterplot matrix



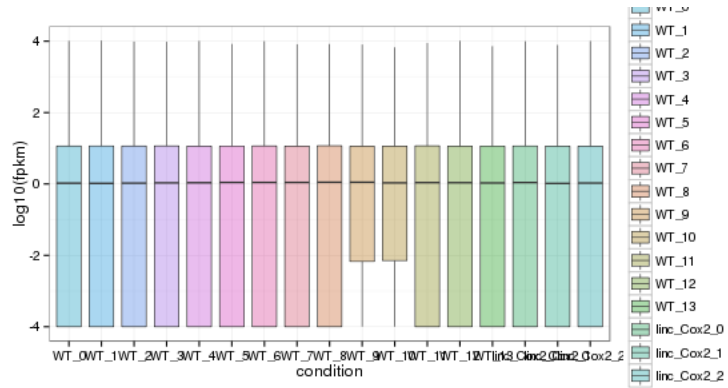
Distributions

Boxplots

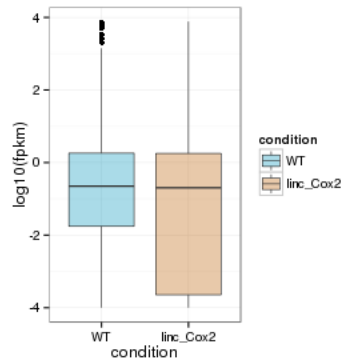
Boxplot (genes)



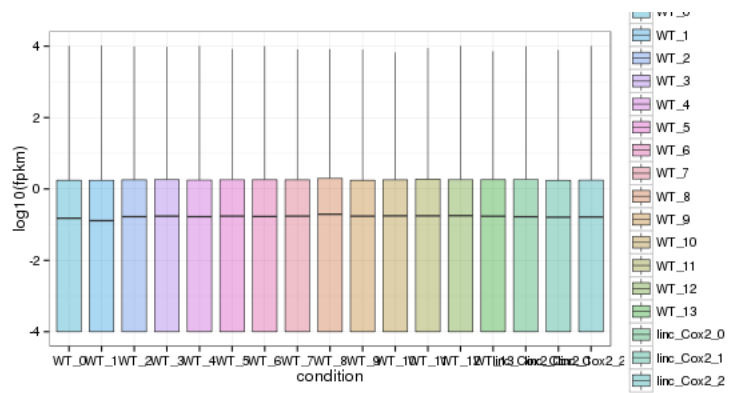
Boxplot (genes, replicates)



Boxplot (isoforms)

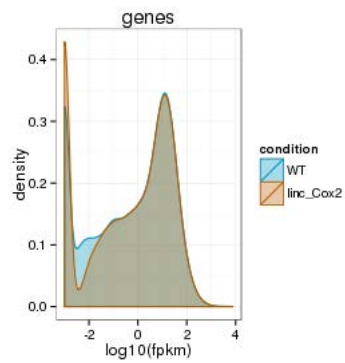


Boxplot (isoforms, replicates)

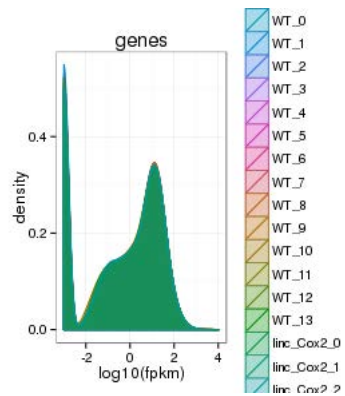


Density

Density (genes)

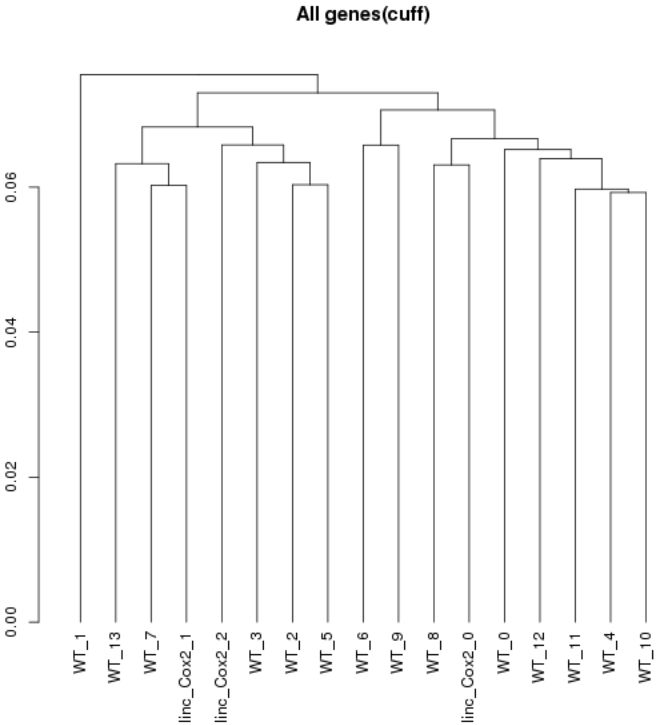


Density (genes, replicates)



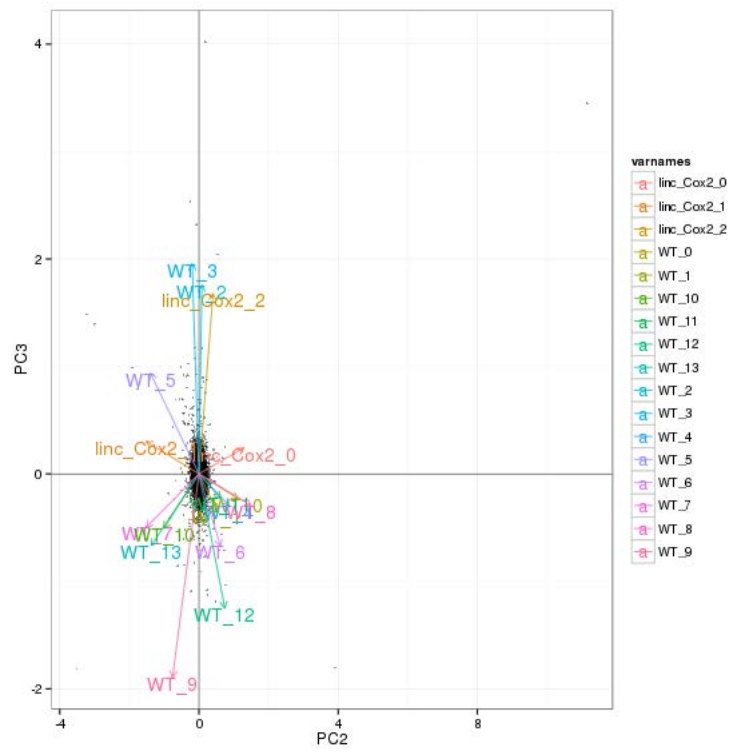
Clustering

Replicate Clusters

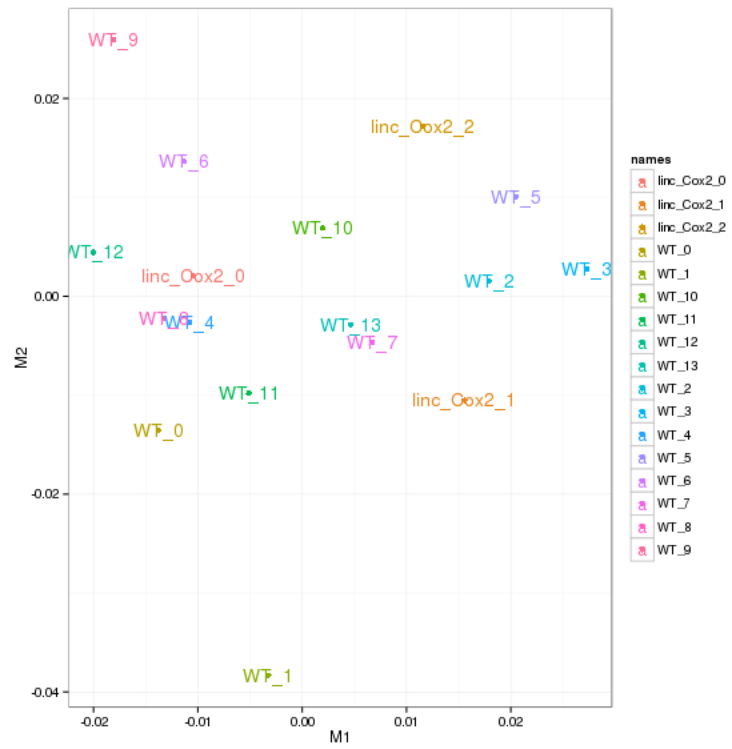


NULL

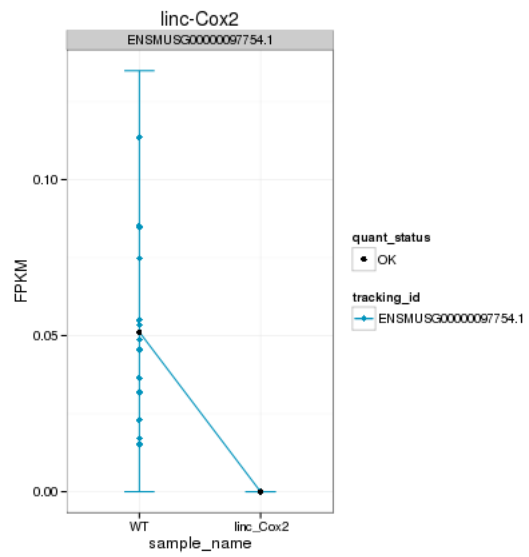
PCA (genes)



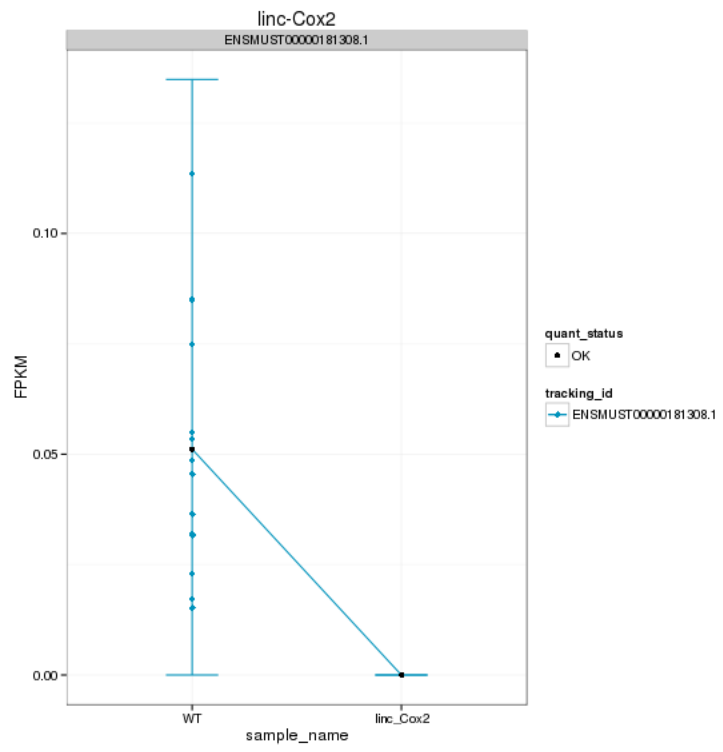
MDS (genes)



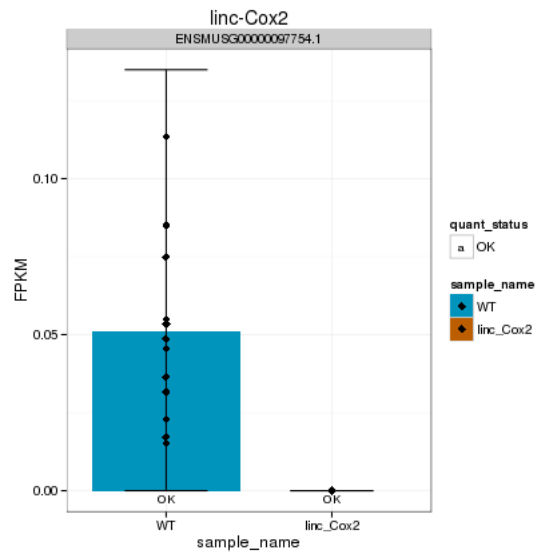
KO assessment
Endogenous lincRNA expression



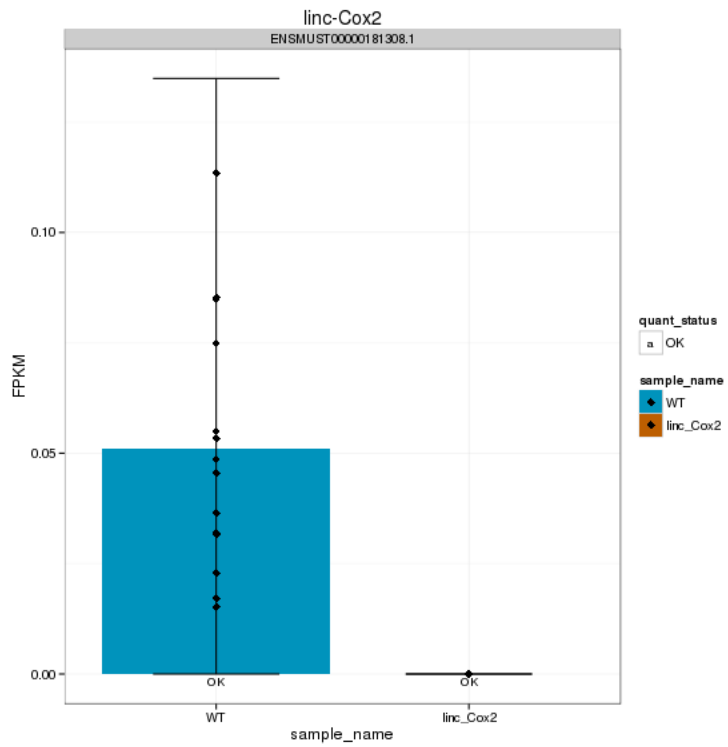
Endogenous expression of linc-Cox2 isoforms:



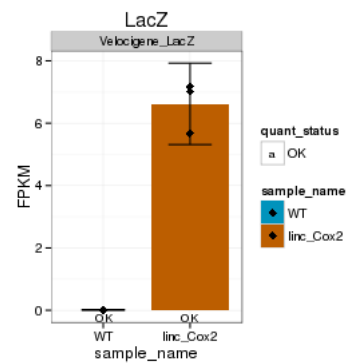
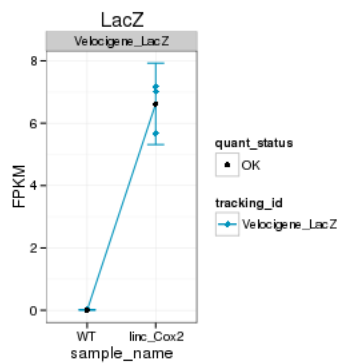
Barplot of gene expression:



Barplot of isoform expression:

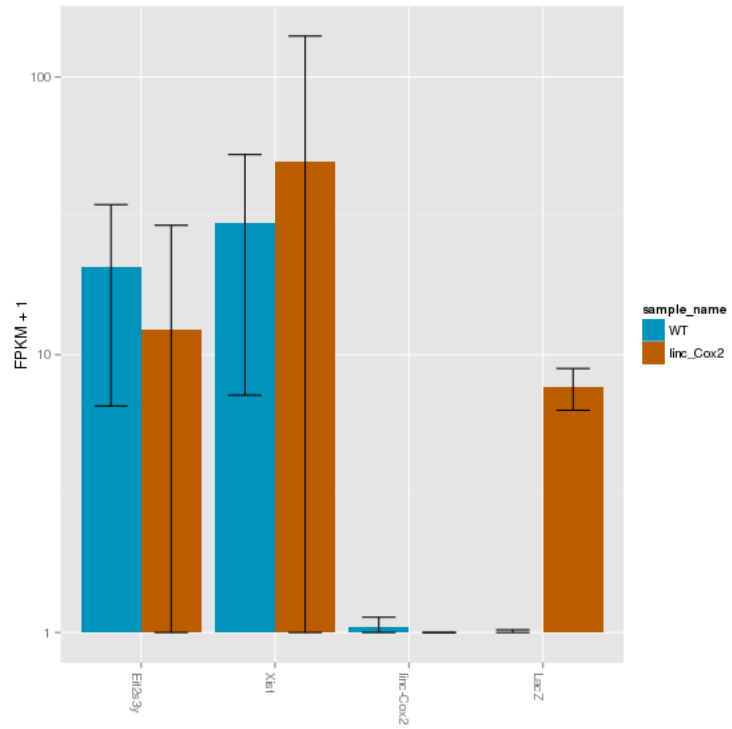


LacZ expression

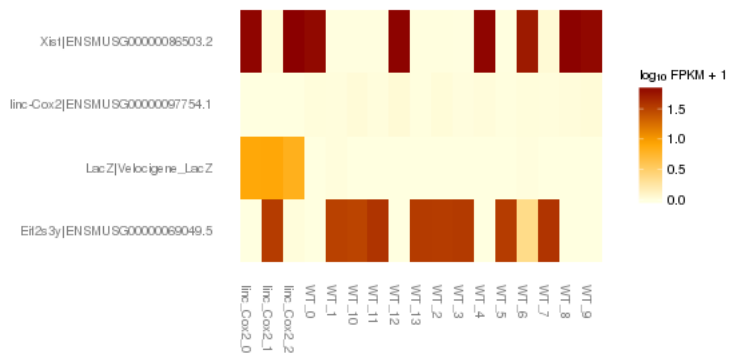


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



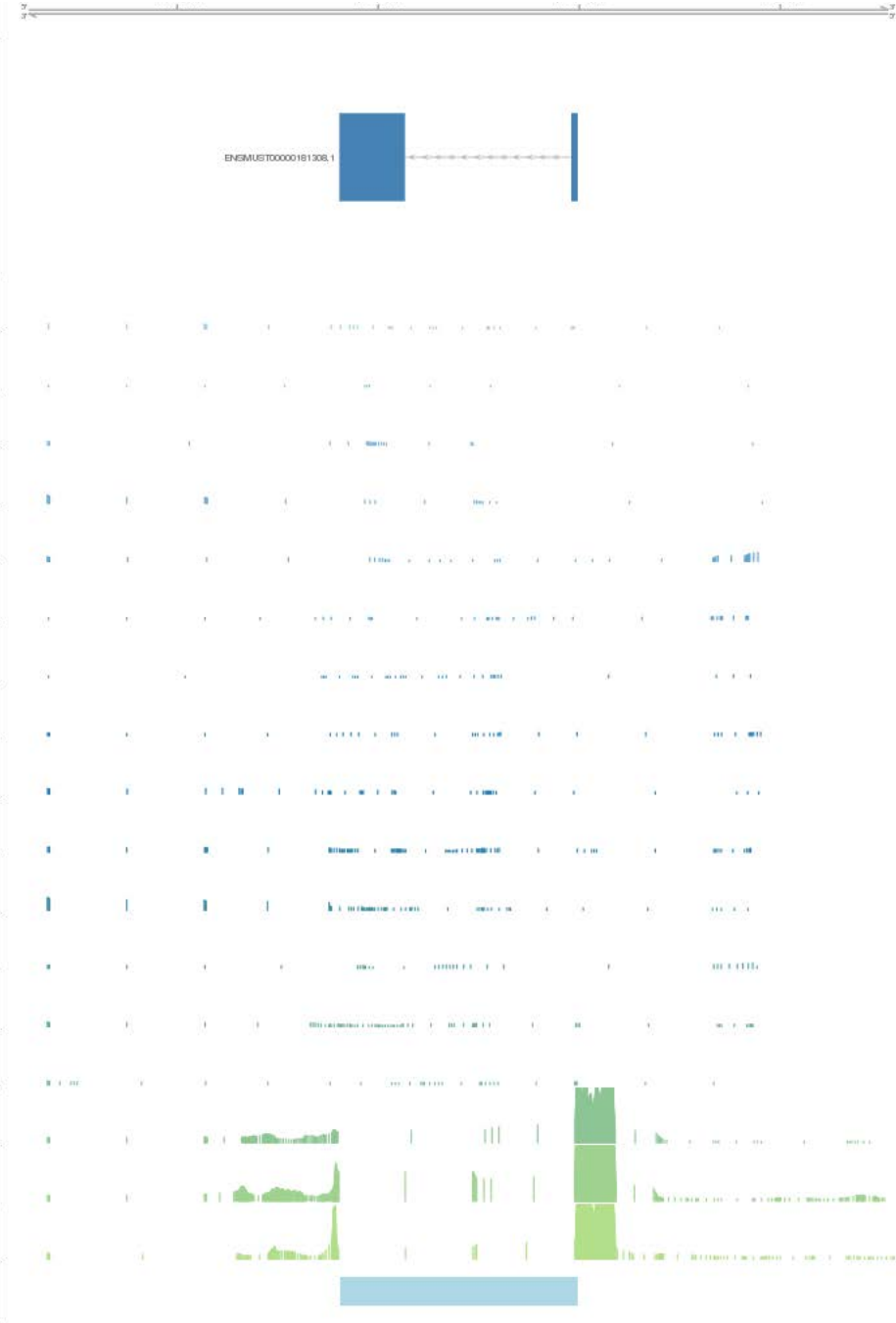
Expression heatmap:



Track visualization



NO Region | line_Coord_1 | line_Coord_2 | line_Coord_3 | WT_0 | WT_1 | WT_2 | WT_3 | WT_4 | WT_5 | WT_6 | WT_7 | WT_8 | WT_9 | WT_10 | WT_11 | WT_12 | WT_13



Differential Analysis

Differential Genes

There are 94 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 Tubb6
2 Folh1
3 Nes
4 Epha2
5 Nav1
6 Vtn
7 Fabp7
8 Adi1
9 Serpina3n
10 Fos
11 Emp2
12 Arc
13 Sdf2l1
14 Nr4a1
15 Cdkn1a
16 Park2
17 Glo1
18 Prss41
19 Atp6v0c
20 Eci1
21 Lars
22 Camk2a
23 Cnksr2
24 Mcm6
25 Dbi
26 Tnni1
27 Lamc2
28 Rgs5
29 Myoc
30 Lypd6b
31 Lcn2
32 Hspa5
33 Olfm13
34 Rps3a1
35 Nr4a3
36 Tinagl1
37 Padi2
```

38 Hes3
39 Fbxo44
40 Fosl2
41 Gkn3
42 Hddc3
43 Col4a1

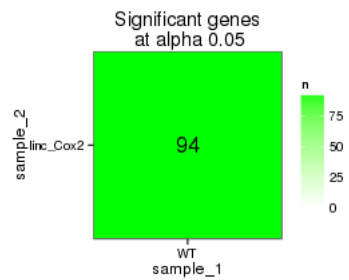
44 Ptgs2
45 Ndn
46 Arhgap27
47 Rnps1
48 Prr7
49 Zranb3
50 Arl5a
51 Mgat5
52 Decr2
53 Psenen
54 Egr1
55 Akap12
56 Pcsk1n
57 Trub2
58 Fmo2
59 Slc25a34
60 Cldn5
61 Gucy1a2
62 Kif21b
63 Shisa4
64 Maff
65 Id1
66 Acp1
67 4930503L19Rik
68 Pcdhb3
69 Olfml2a
70 Tnrc6b

71 Lmod1
72 Tagap1
73 Aldh7a1
74 Per2
75 S100a9
76 Kcnq3
77 Pign
78 Kdm5d
79 Tpm3-rs7
80 Capn11

- 81 Gm5506
- 82 Pyroxd2
- 83 Eno1
- 84 Chi3l1
- 85 Clasp1
- 86 Ddx3y
- 87 Gm7292
- 88 Wdfy1
- 89 Apold1
- 90 Hspa1b
- 91 Scnm1
- 92 AA465934
- 93 Gm10524
- 94 RP24-390G17.1

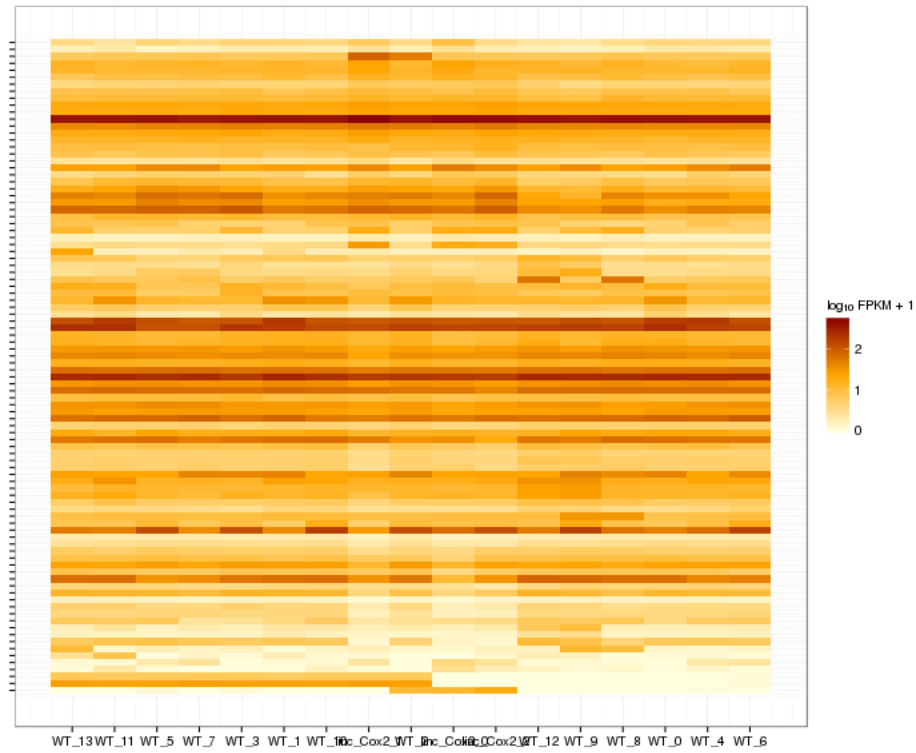
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

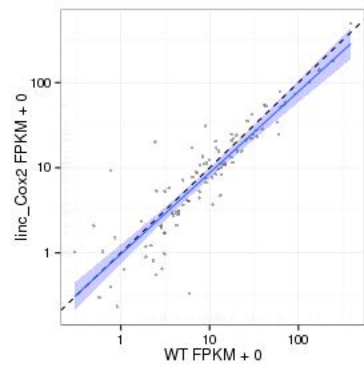


Significant genes with expression >50fpm (any condition):(turned off)

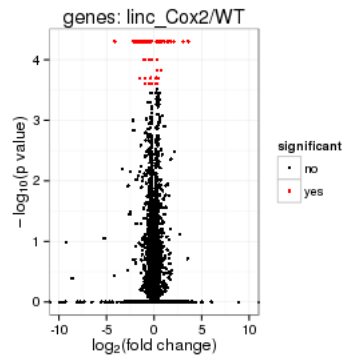
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

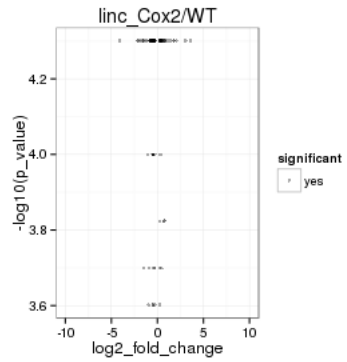
Scatter plot of significant genes only:



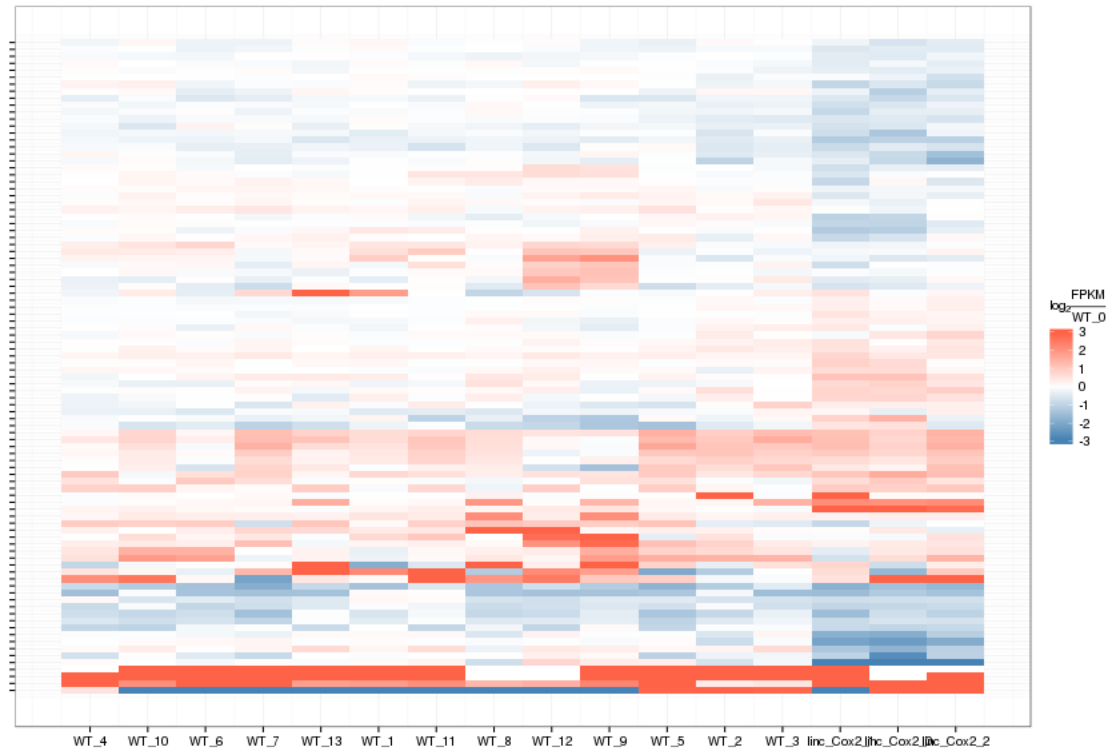
Volcano Plot



Volcano plot with significant genes only:



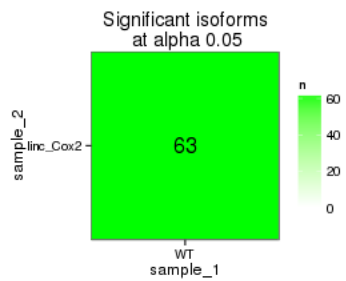
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:

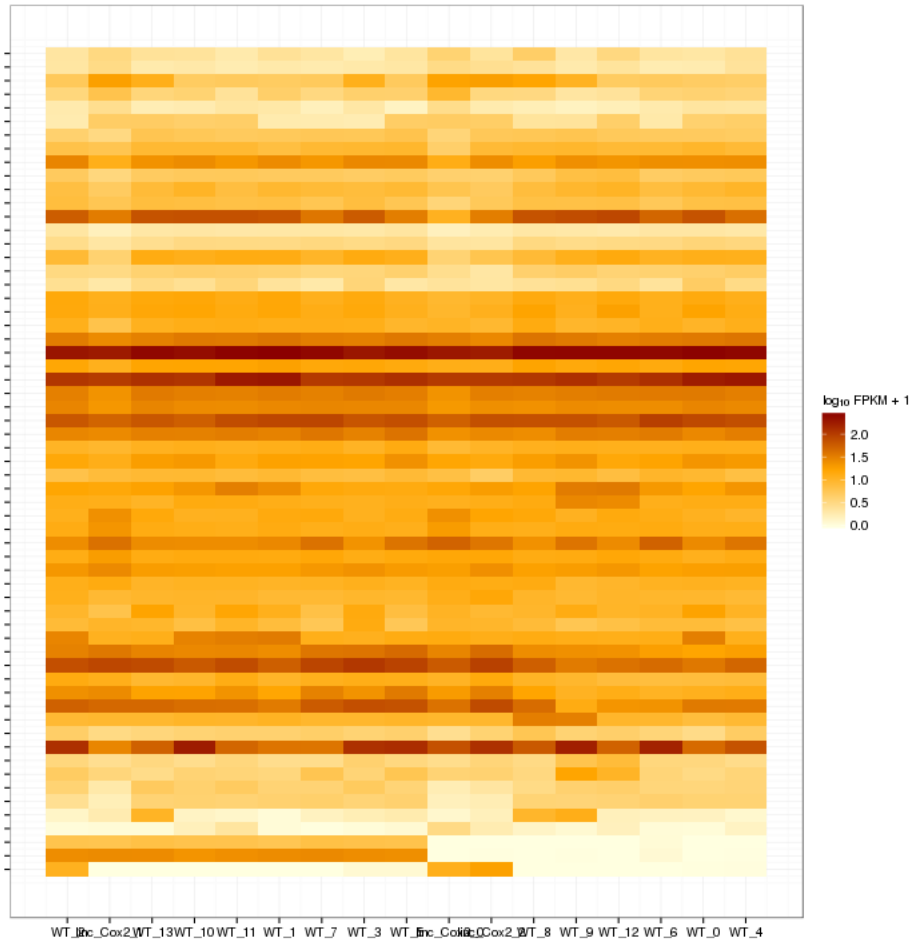


These isoforms are:

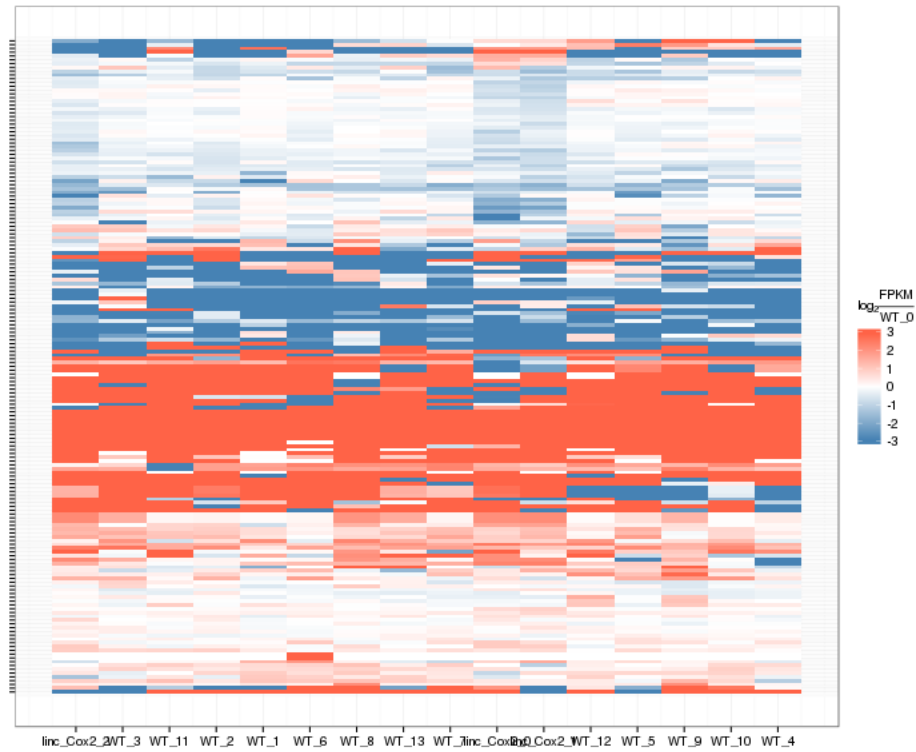
isoAnnot	gene_short_name
1	Tubb6
2	Rec8
3	Nes
4	Vtn
5	Fabp7
6	Serpina3n
7	Fos
8	Emp2
9	Arc
10	Sdf2l1
11	Nr4a1
12	Cdkn1a
13	Glo1
14	Atp6v0c
15	Eci1
16	Lars
17	Cdh7
18	Mcm6
19	Dbi
20	Lamc2
21	Rgs5
22	Myoc
23	Lypd6b
24	Olfml3
25	Tpm3
26	Rps3a1
27	Arhgef19
28	Padi2
29	Hes3
30	Fosl2
31	Gkn3
32	Hddc3
33	Ptgs2
34	Ndn
35	Rnps1
36	Arl5a
37	Decr2
38	Egr1
39	Akap12
40	Pcsk1n

41 Cercam
42 Trub2
43 Fmo2
44 Zfp236
45 Cldn5
46 Kif21b
47 Pm20d1
48 Olfm12a
49 Tagap1
50 Per2
51 Kcnq3
52 Kdm5d
53 Tpm3-rs7
54 Capn11
55 Gm5506
56 Ddx3y
57 Gm7292
58 Wdfy1
59 Apold1
60 Hspa1b
61 Ptpv

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

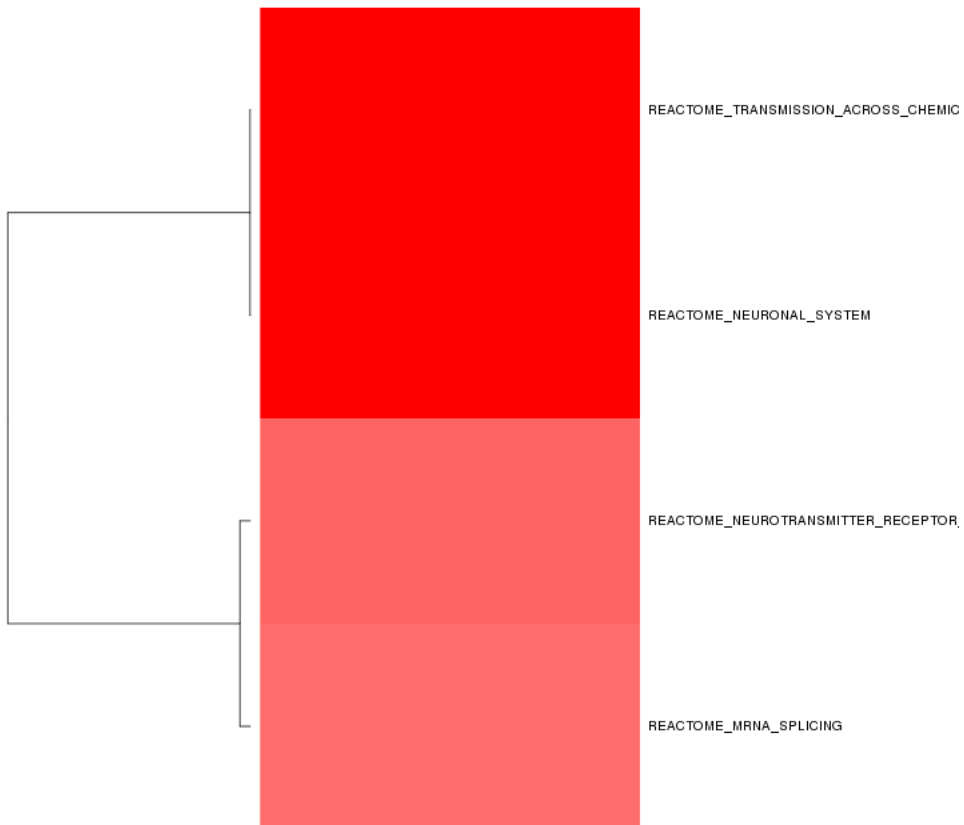
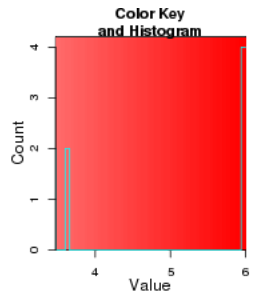
Biocarta enrichment:

```
## [1] "Not enough significant categories to print a heatmap!"
```

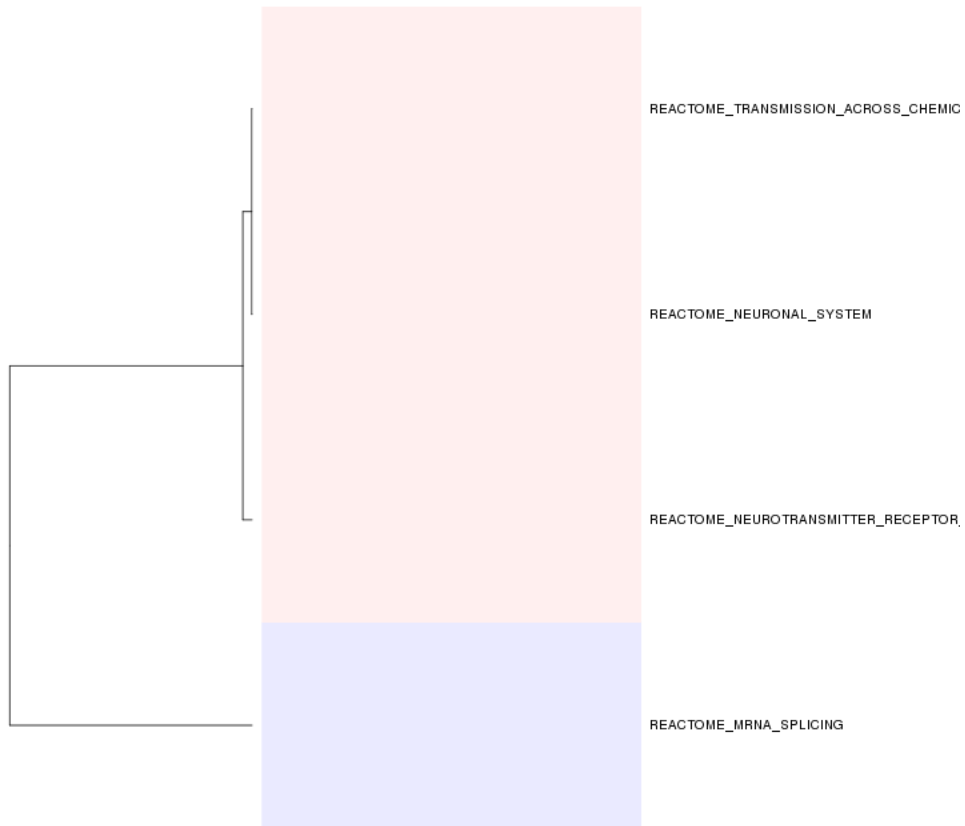
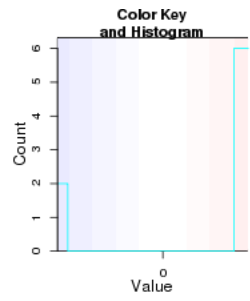
Biocarta zscore:

```
## [1] "Not enough significant categories to print a heatmap!"
```

Reactome enrichment:



Reactome zscore:



Kegg enrichment:

```
## Error: argument is of length zero
```

Kegg zscore:

```
## Error: incorrect number of dimensions
```

```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Interneuron enrichment:

```
## Error: argument is of length zero
```

Interneuron zscore:

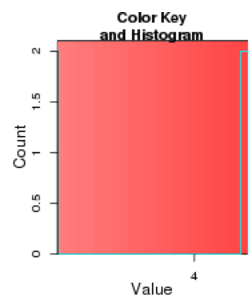
```
## Error: incorrect number of dimensions
```

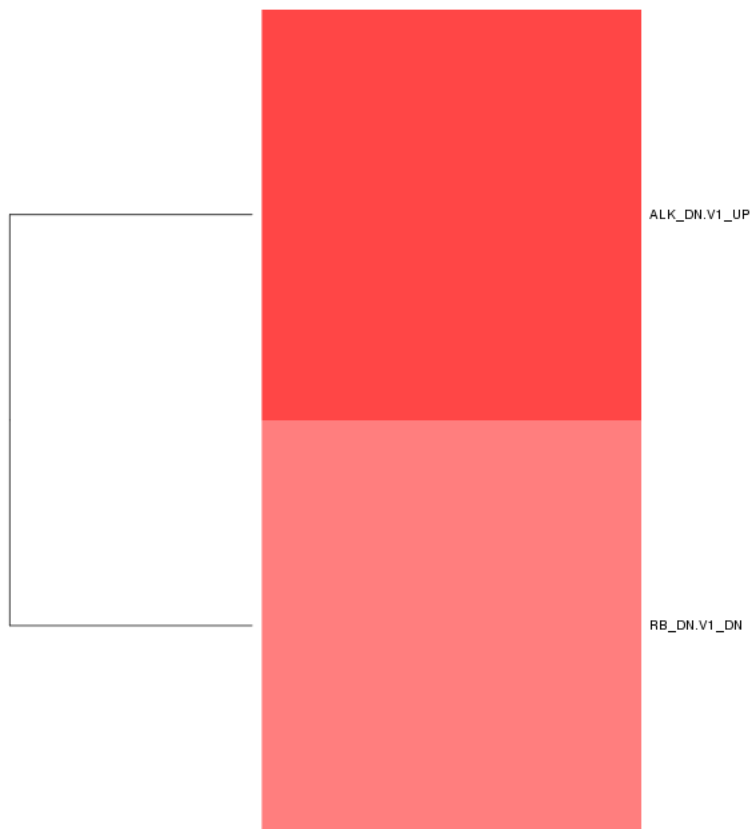
```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

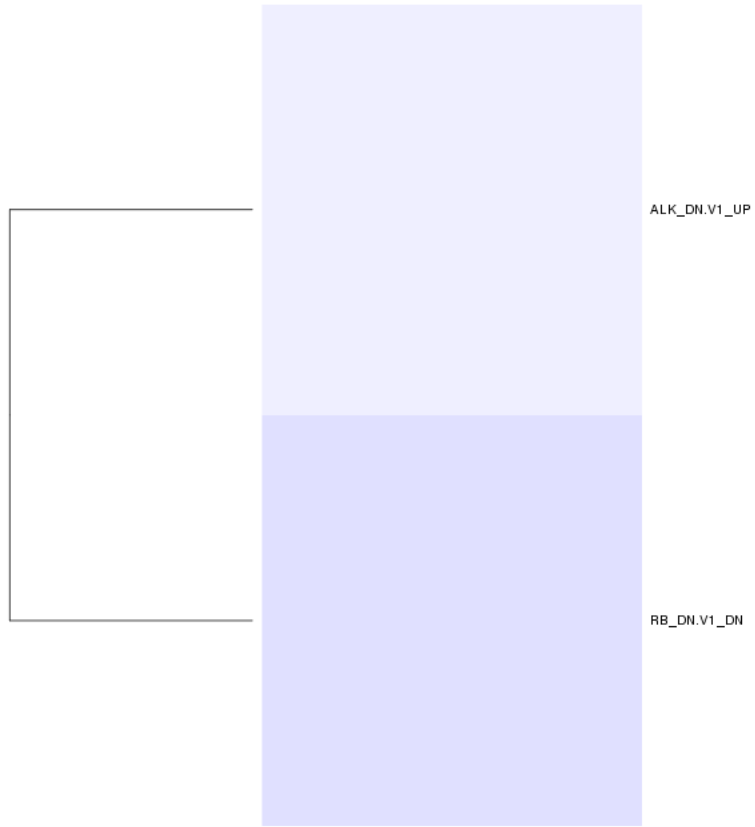
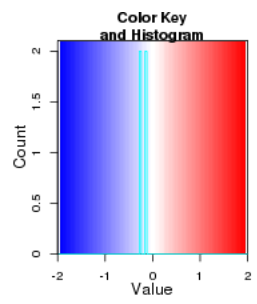
```
## Error: object 'x_ordered' not found
```

Oncogene enrichment:

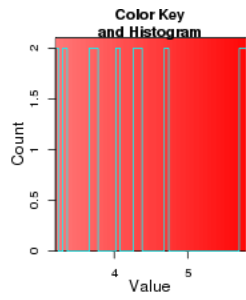


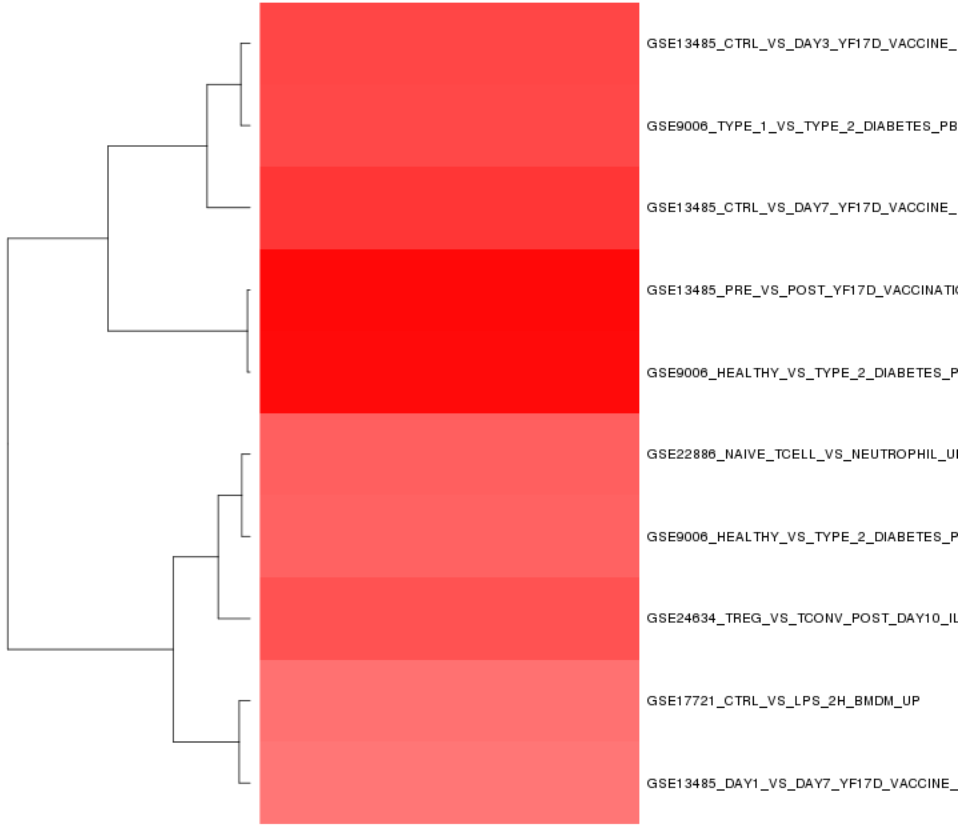


Oncogene zscore:

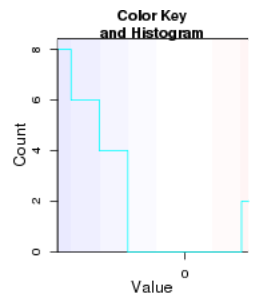


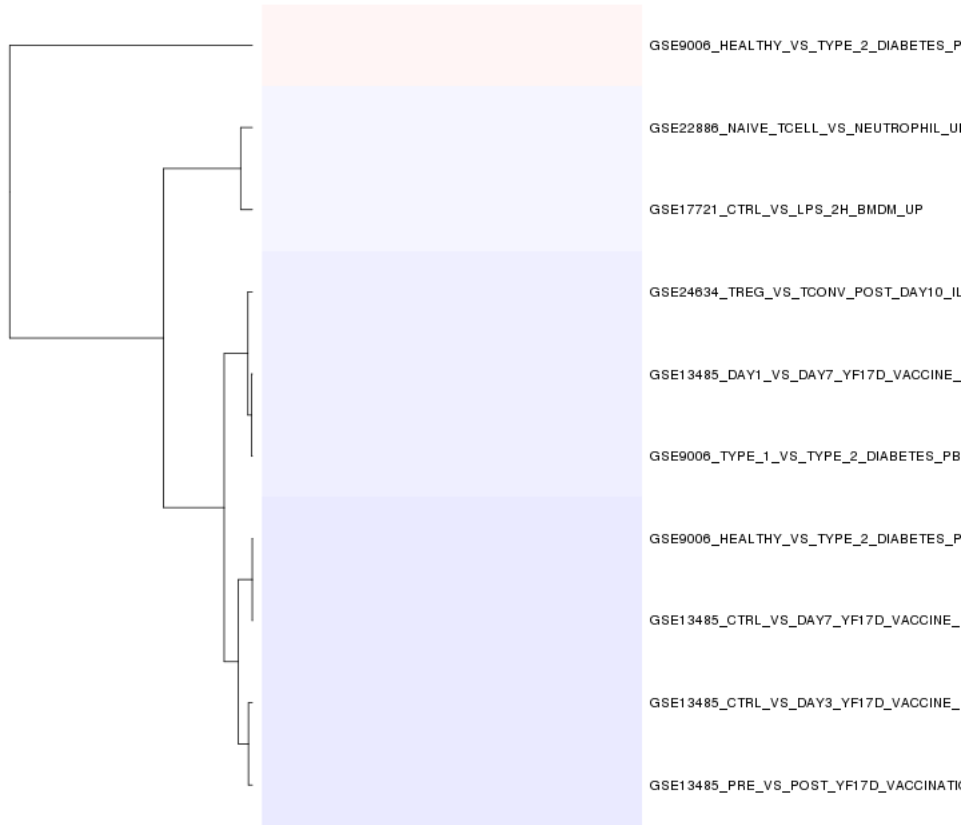
Immuno enrichment:



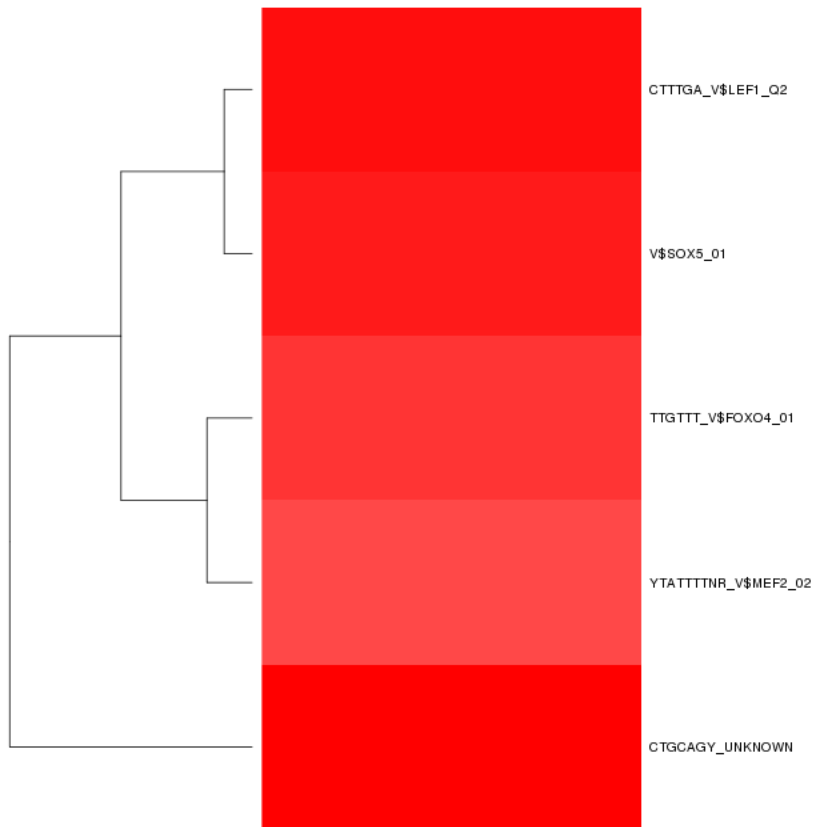
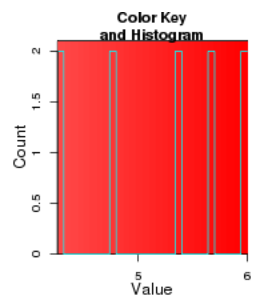


Immuno zscore:

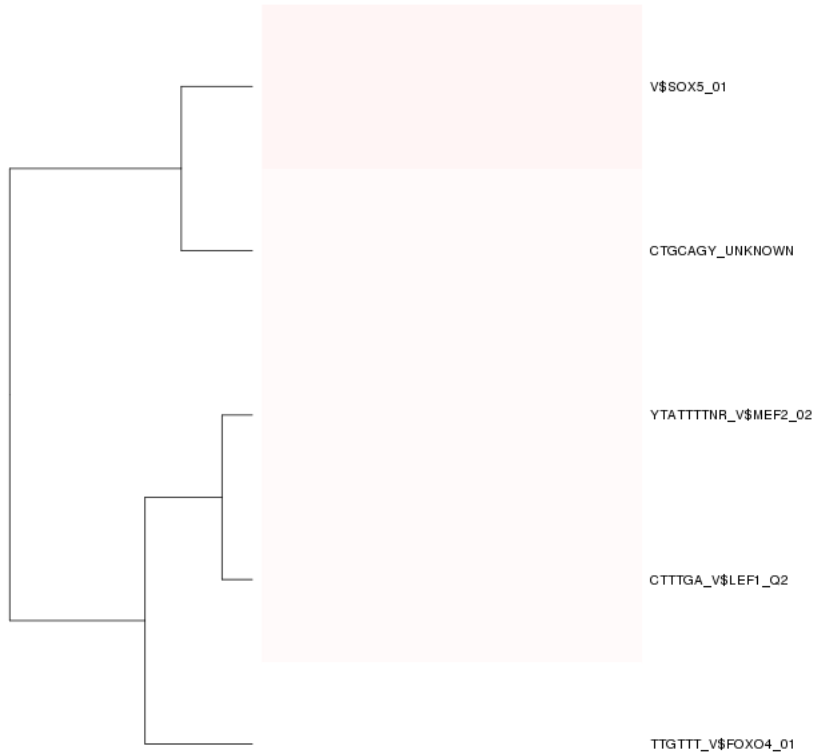
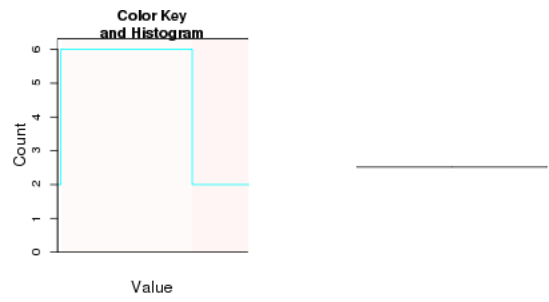




TF enrichment:

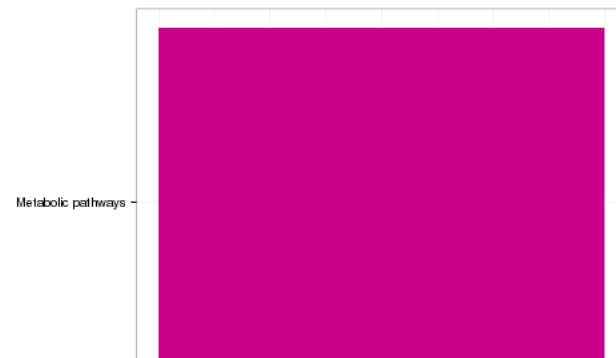
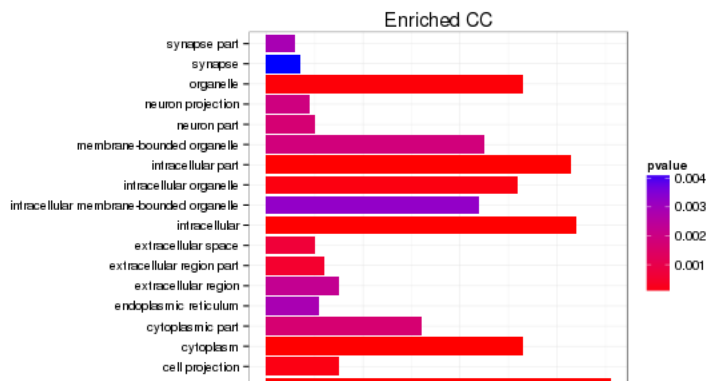
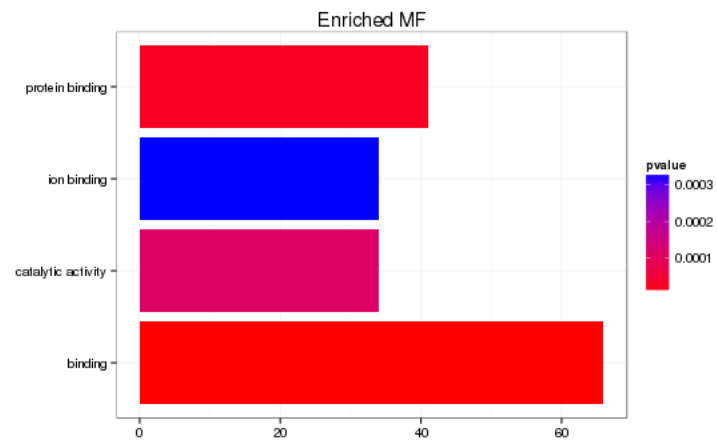
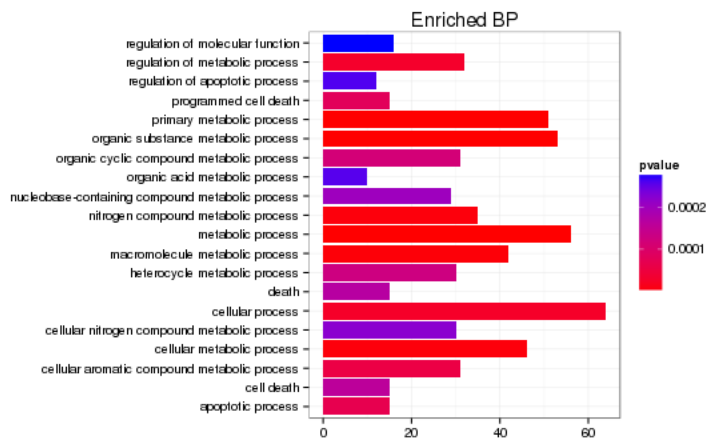


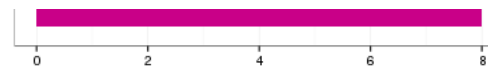
TF zscore:



GO enrichment

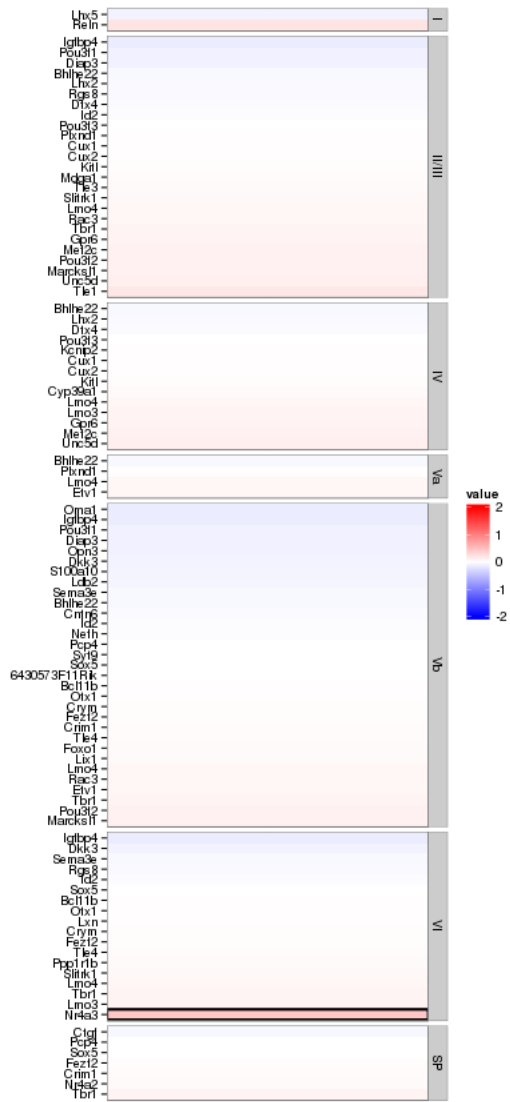
Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.



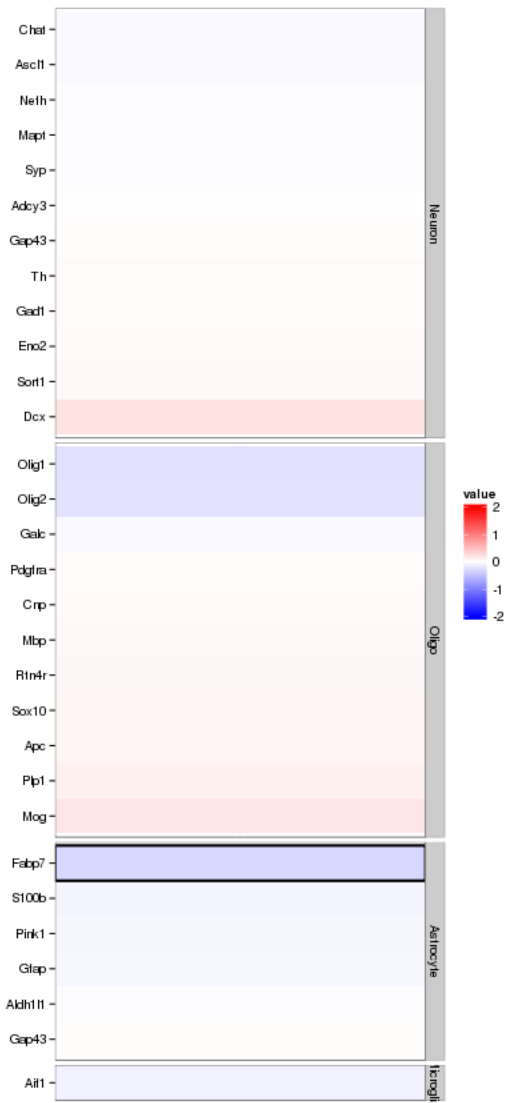


```
## Error: 'x' and 'units' must have length > 0
```

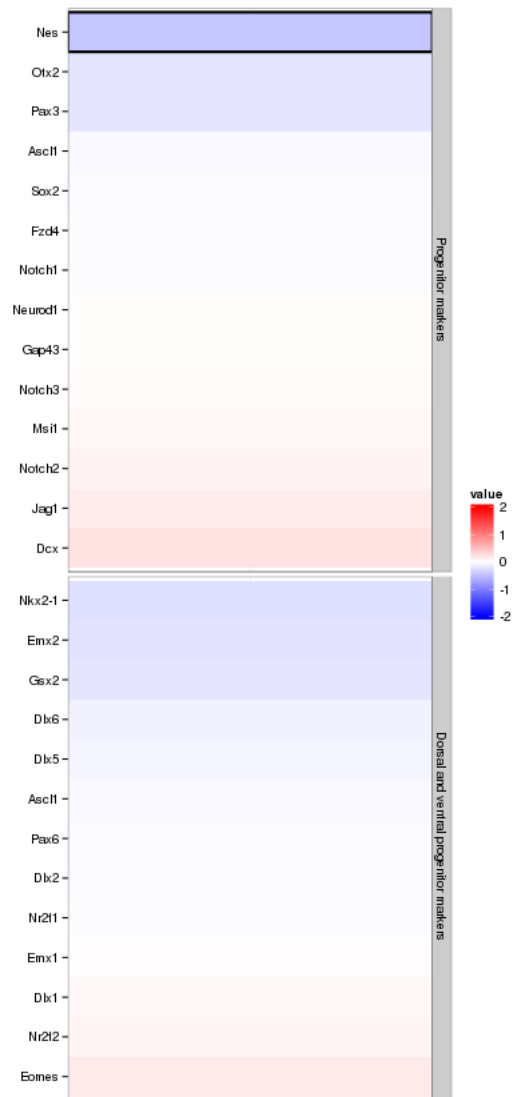
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



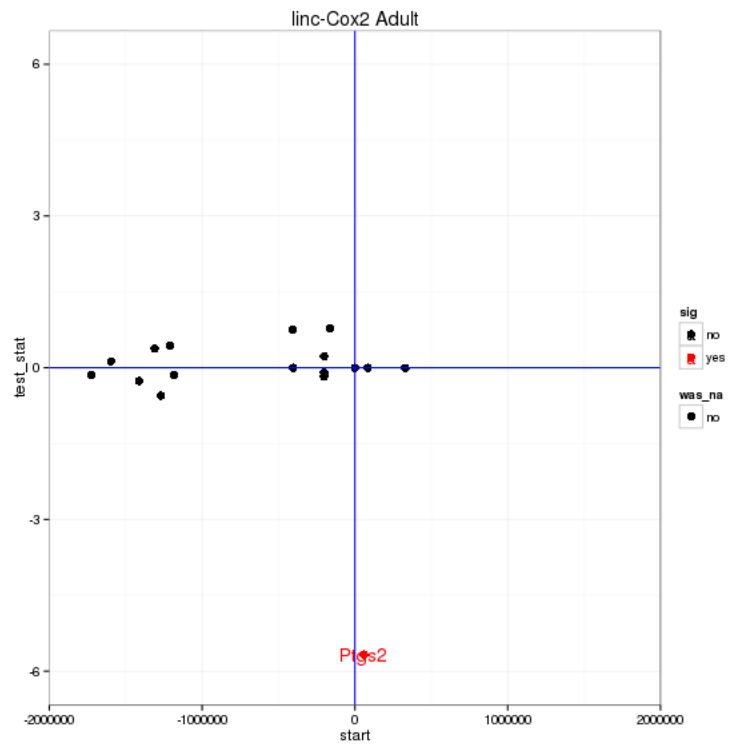
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 1 genes significantly regulated in a region this size is: 0.1821



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR732
16 JR776
17 JR798

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34095400.00	0.77	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34095400.00	0.59	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34095400.00	1.00	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34095400.00	0.82	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34095400.00	1.05	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34095400.00	1.09	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34095400.00	0.82	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34095400.00	1.18	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34095400.00	1.03	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34095400.00	1.32	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34095400.00	1.53	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34095400.00	1.03	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34095400.00	0.99	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34095400.00	1.23	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR732/abundances.cxb	linc_Cox2	0	linc_Cox2_0	27439100.00	34095400.00	0.81	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR776/abundances.cxb	linc_Cox2	1	linc_Cox2_1	39517800.00	34095400.00	1.14	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR798/abundances.cxb	linc_Cox2	2	linc_Cox2_2	35636800.00	34095400.00	1.03	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2     colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4       DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10       Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3    graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1       Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29    latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3       munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10       qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4       splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7    tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,linc-Cox2 -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/linc-Cox2_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap
## 2
## 3
## 4
## 5
```

linc-Cox2 KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for linc-Cox2.

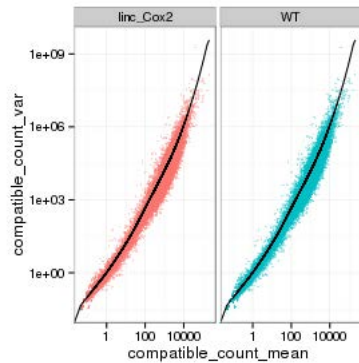
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

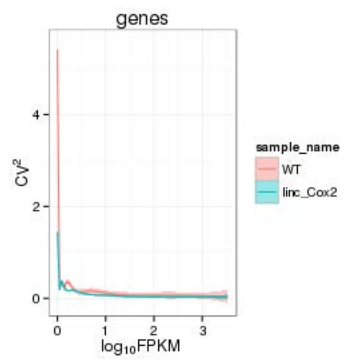
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



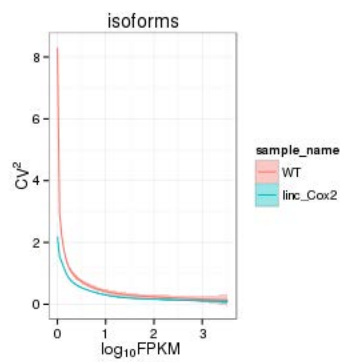
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

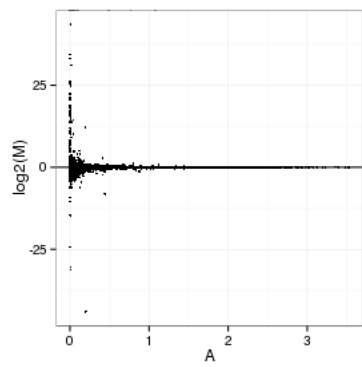
Genes:



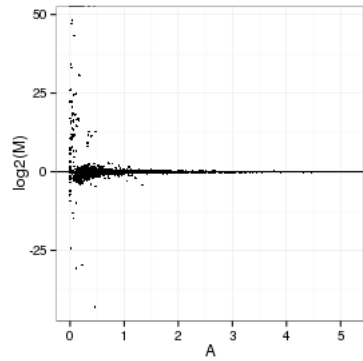
Isoforms:



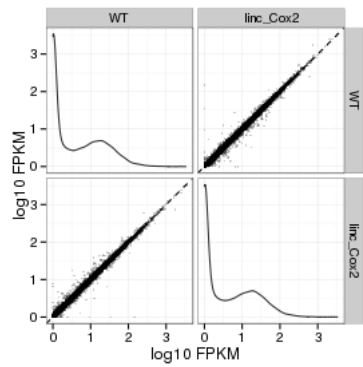
MvA plot



MvA plot counts



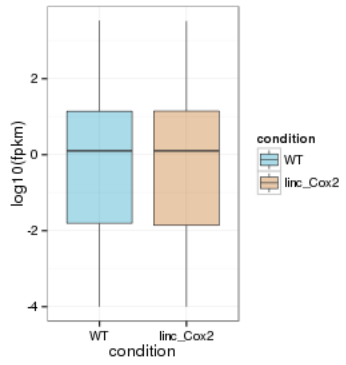
Scatterplot matrix



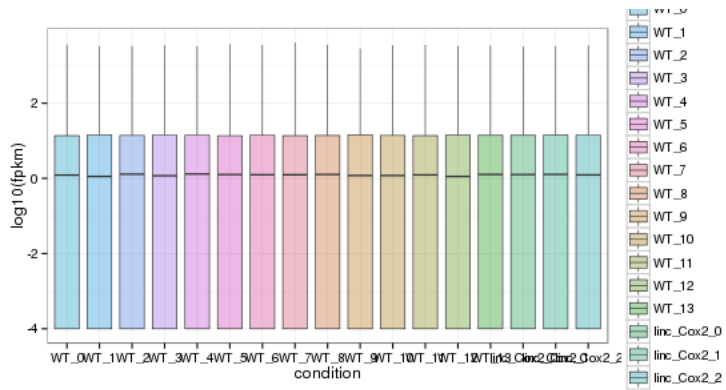
Distributions

Boxplots

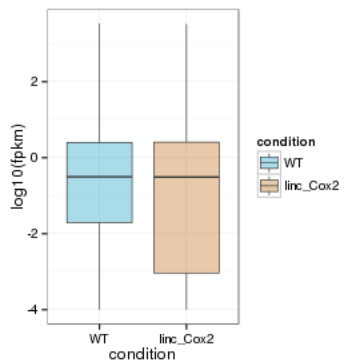
Boxplot (genes)



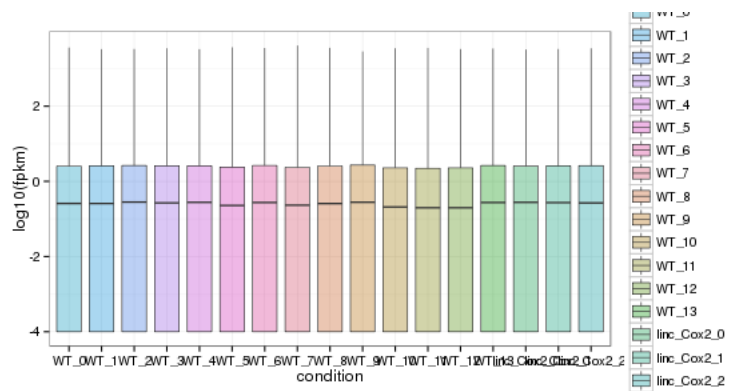
Boxplot (genes, replicates)



Boxplot (isoforms)

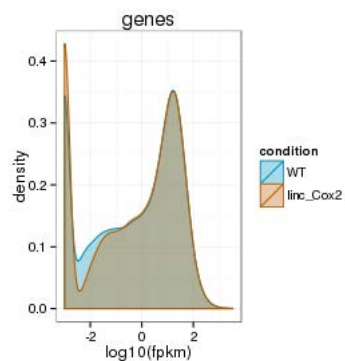


Boxplot (isoforms, replicates)

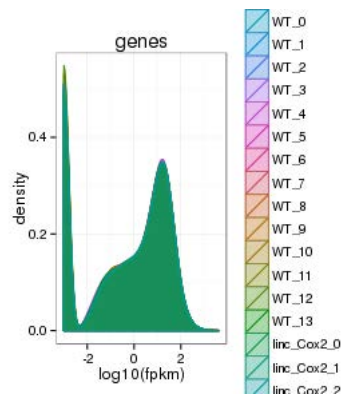


Density

Density (genes)

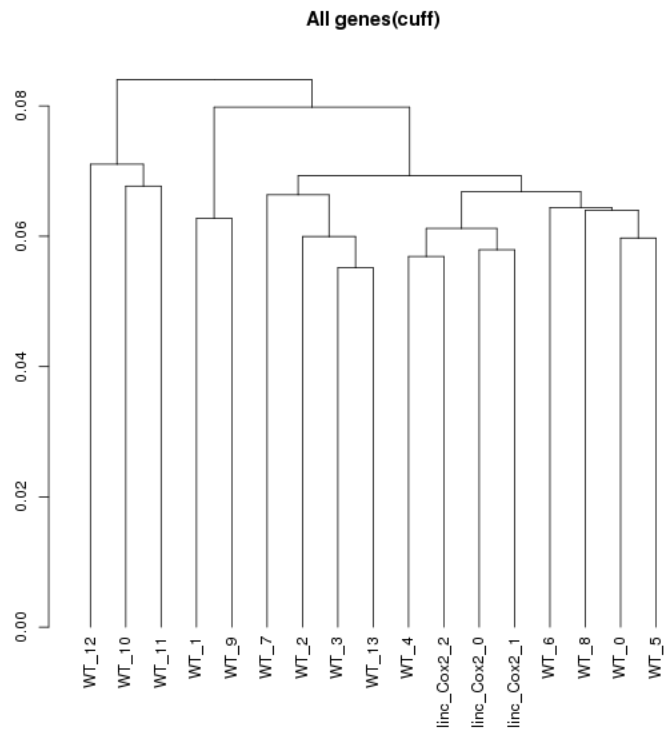


Density (genes, replicates)



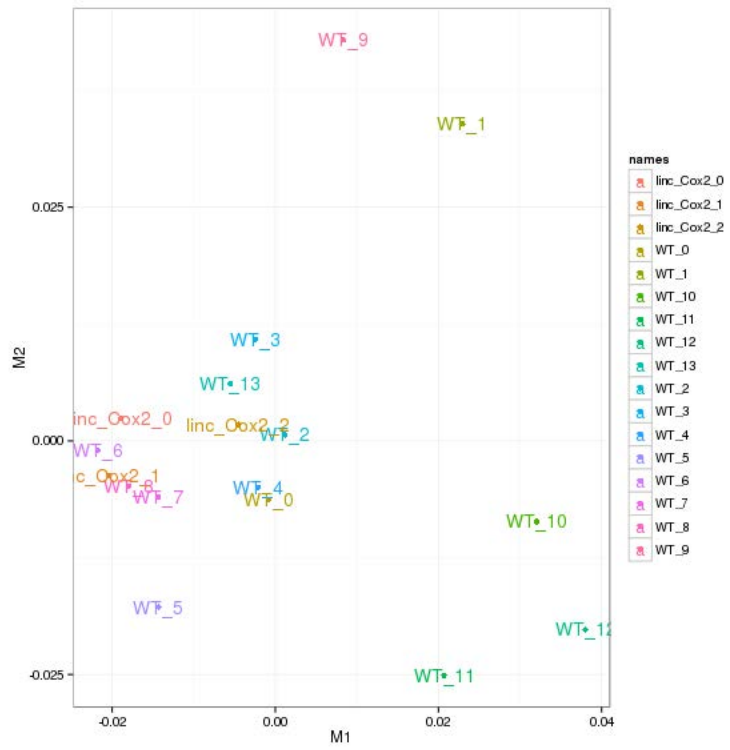
Clustering

Replicate Clusters



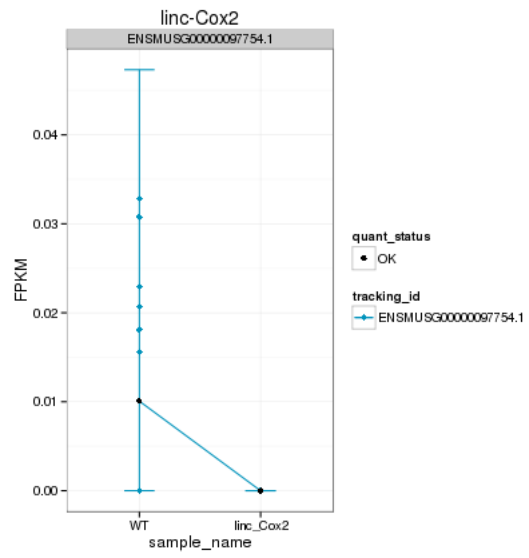
NULL

PCA (genes)

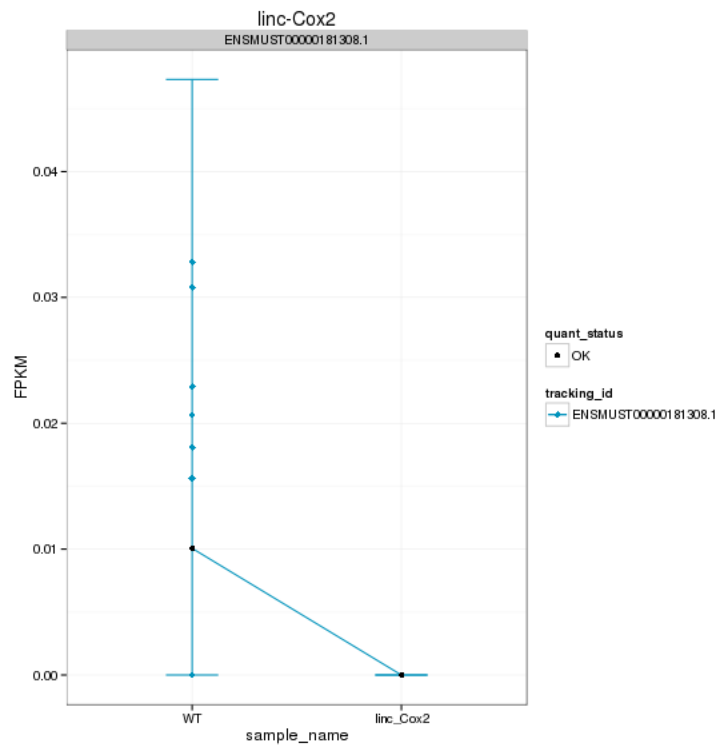


KO assessment

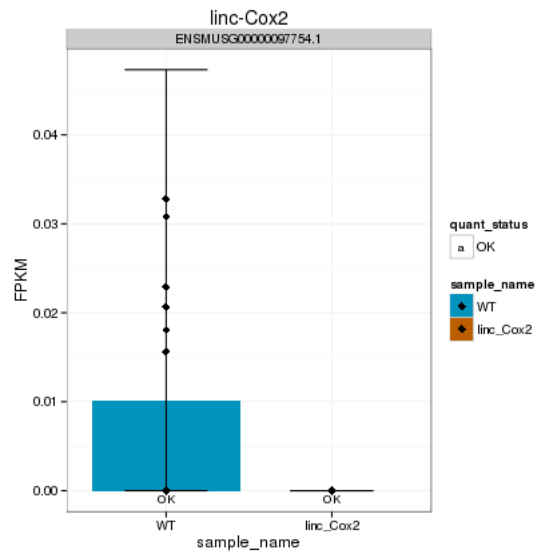
Endogenous lincRNA expression



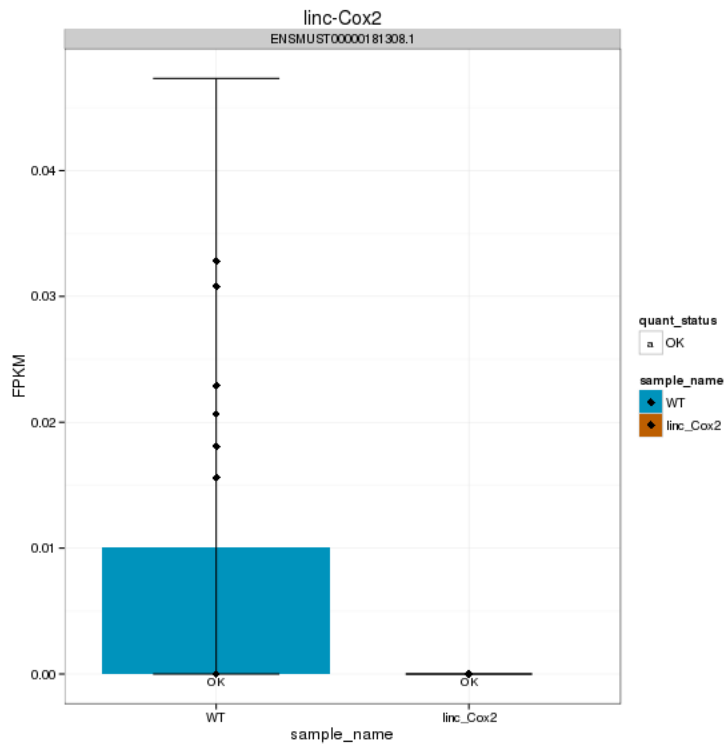
Endogenous expression of linc-Cox2 isoforms:



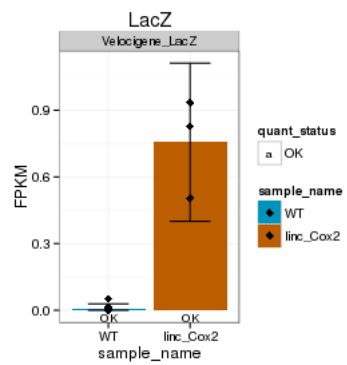
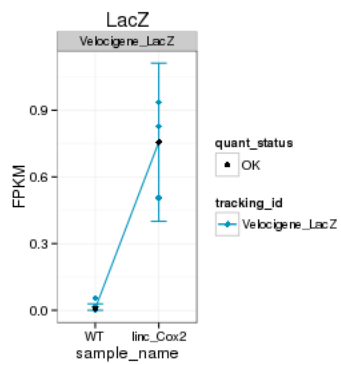
Barplot of gene expression:



Barplot of isoform expression:

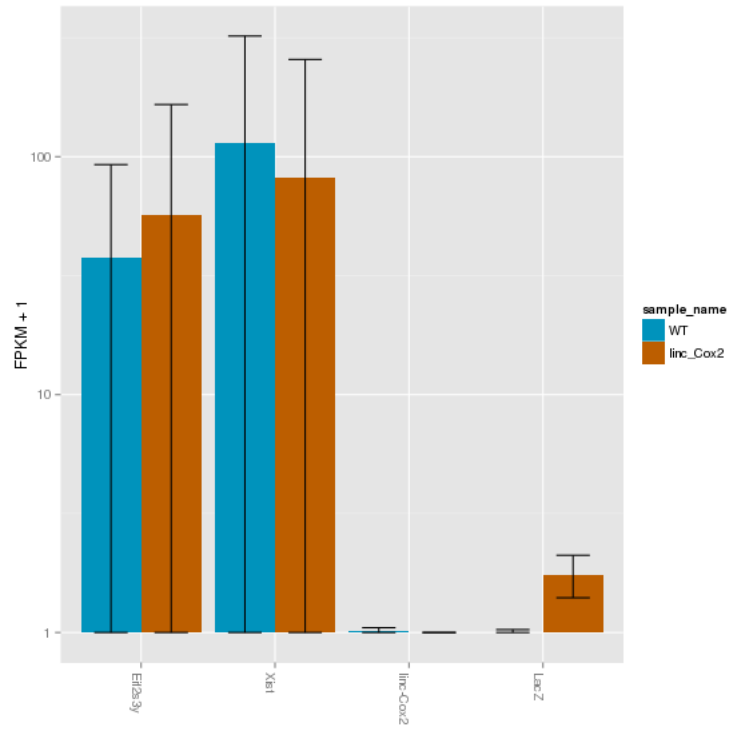


LacZ expression

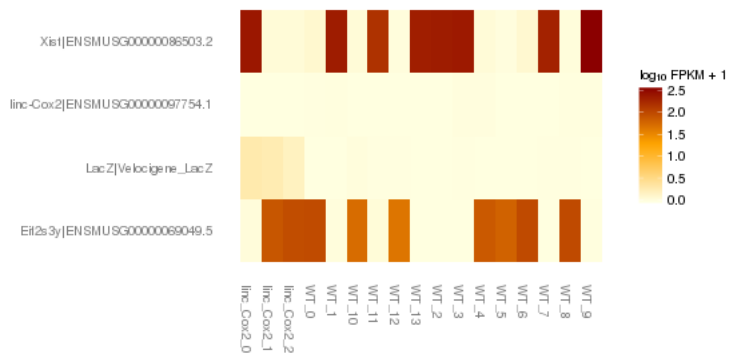


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

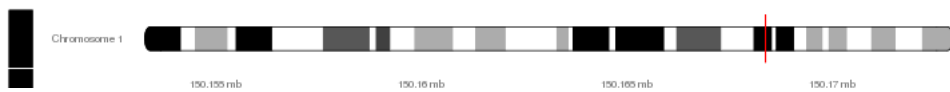
Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



Expression heatmap:



Track visualization



RG Region | line_Coord_1 | line_Coord_2 | line_Coord_3 | WT_0 | WT_1 | WT_2 | WT_3 | WT_4 | WT_5 | WT_6 | WT_7 | WT_8 | WT_9 | WT_10 | WT_11 | WT_12 | WT_13



Differential Analysis

Differential Genes

There are 97 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 Col1a1
2 Rec8
3 Zfp40
4 Fam131c
5 Vax1
6 Ttc9b
7 Lama2
8 Snrpf
9 Skor1
10 Dlx2
11 Park2
12 Glo1
13 Prss41
14 Atp6v0c
15 Hn1l
16 Lox
17 Fbn2
18 Rps14
19 Lbx1
20 Col3a1
21 Fn1
22 Bok
23 Mcm6
24 Dbi
25 Fbn1
26 Syt13
27 Rps3a1
28 Svep1
29 Uncx
30 Emp1
31 Dera
32 Hddc3
33 Pgl3
34 Col12a1
35 Dnah8
36 Irf2bpl
37 Arhgap27
```

38 Gbx2
39 Rnps1
40 Prr7
41 Lars2
42 Arx
43 3110052M02Rik

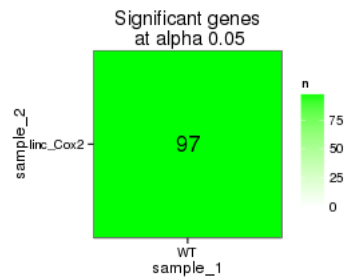
44 Mgat5
45 Lum
46 Decr2
47 Cldn11
48 Vstm2l
49 Hoxb4
50 Shank1
51 Pcsk1n
52 Tlx3
53 C1qtnf4
54 2310036O22Rik
55 Fmod
56 Shisa4
57 Hebp1
58 Pgp
59 Gm9493
60 Rps2
61 Acp1
62 Olfml2a
63 Hmga1
64 Lrrc4b
65 Dmrt2
66 Mafa
67 Tagap1
68 Hbb-y
69 Zfp771
70 Pign

71 Kdm5d
72 Capn11
73 H2afj
74 Alox5ap
75 Rpl26
76 Sox11
77 Zfp428
78 Sp9
79 Nkain2
80 Ssbp4

- 81 Gm1673
- 82 Jund
- 83 Gpr27
- 84 9030025P20Rik
- 85 Wdfy1
- 86 Hbb-bt
- 87 Rpl39
- 88 Gm11273
- 89 Ccdc85c
- 90 Ier5l
- 91 Myl6
- 92 Rnaset2b
- 93 Ccdc85b
- 94 Gm6472
- 95 Gm9825
- 96 Gm26917
- 97 Gm26924

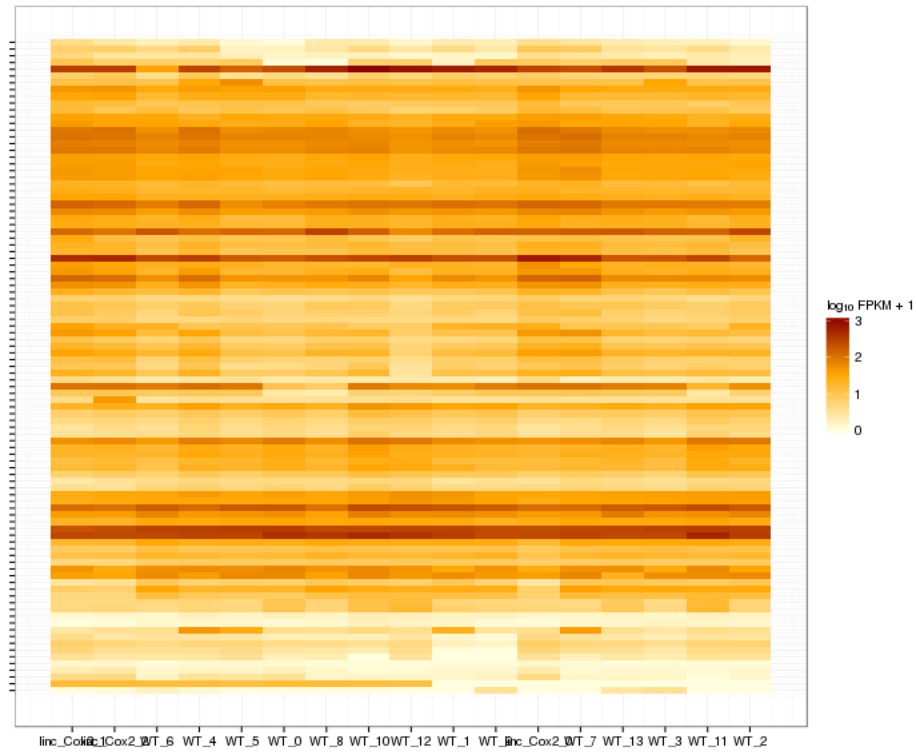
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

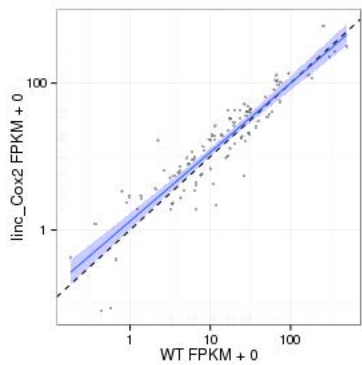


Significant genes with expression >50fpm (any condition):(turned off)

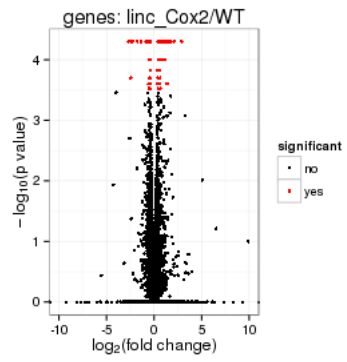
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

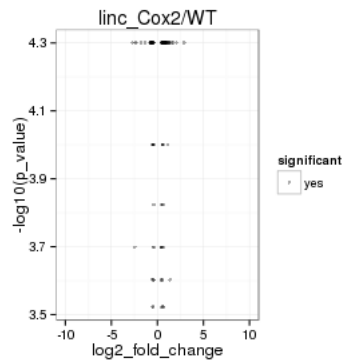
Scatter plot of significant genes only:



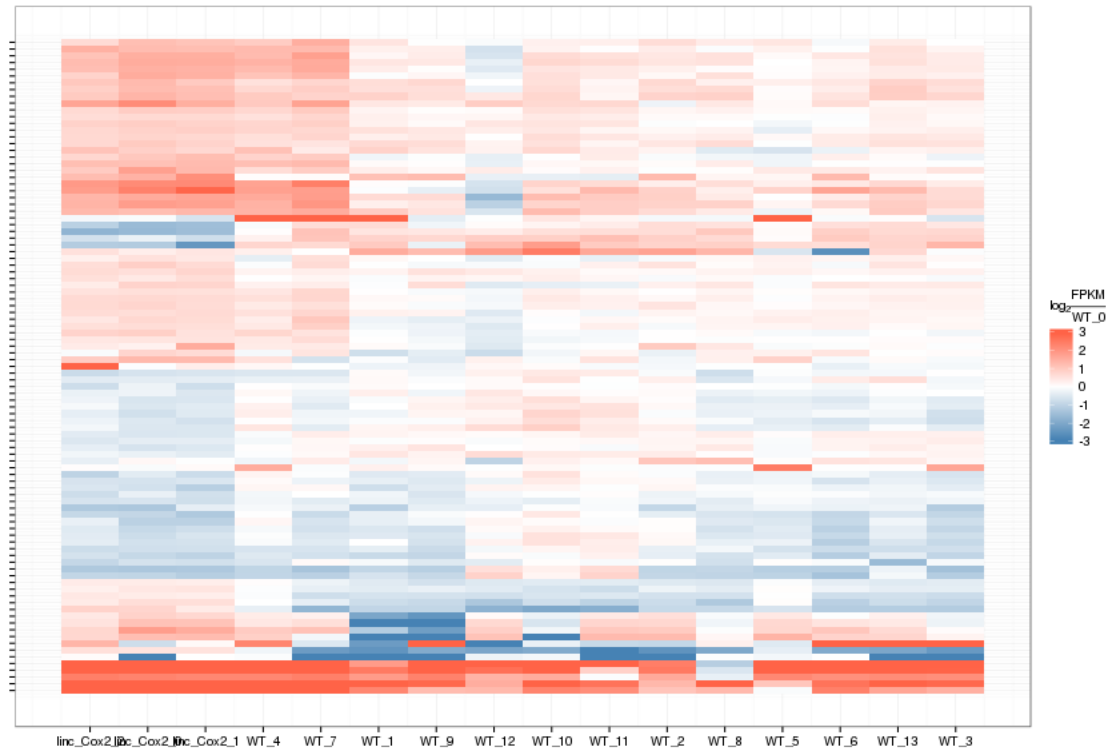
Volcano Plot



Volcano plot with significant genes only:



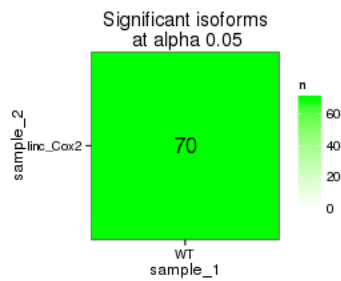
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

isoAnnot\$gene_short_name

- 1 Dynl1c
- 2 Col1a1
- 3 Rec8
- 4 Zfp40
- 5 Lama2
- 6 Snrpf
- 7 Prpf8
- 8 Dlx2
- 9 Hcfc1r1

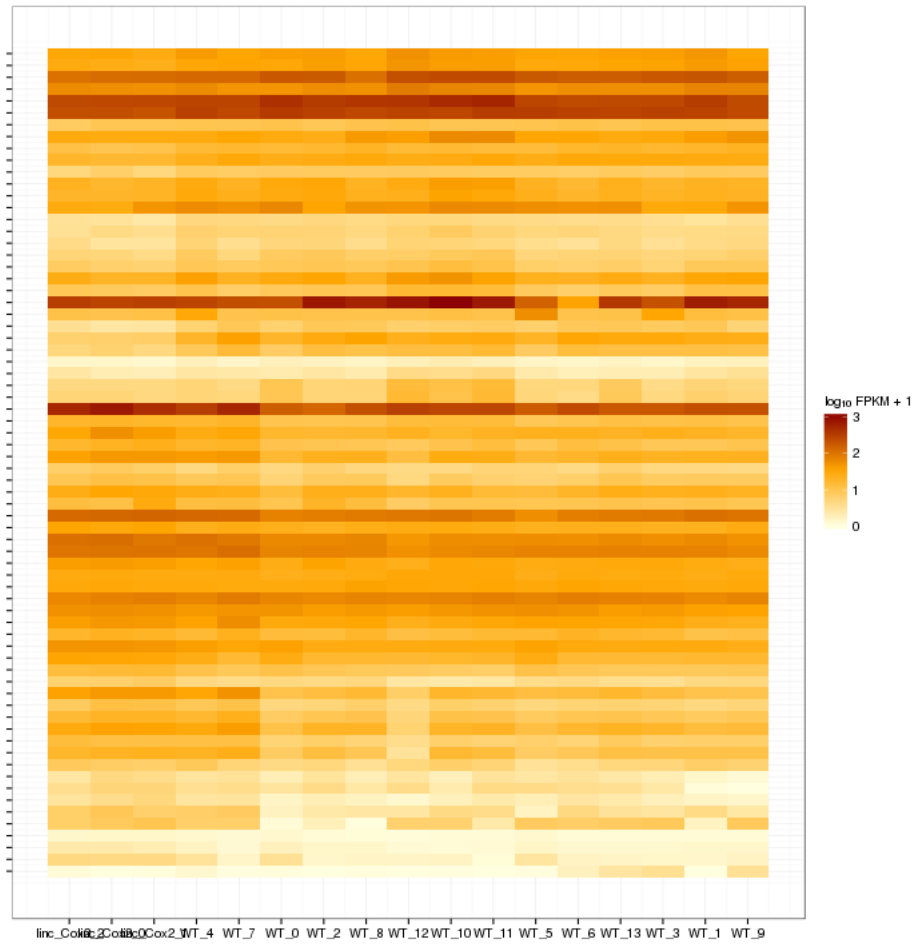
- 10 Glo1
- 11 Prss41
- 12 Atp6v0c
- 13 Hn1l
- 14 Lox
- 15 Rps14
- 16 Lbx1
- 17 Fn1
- 18 Bok
- 19 Asb1
- 20 Mcm6
- 21 Dbi
- 22 Lrp2
- 23 Rps3a1
- 24 Svep1
- 25 Ski
- 26 Dera
- 27 Hddc3
- 28 Dnah8
- 29 Gbx2
- 30 Rnps1
- 31 Fam171a2
- 32 Prr7
- 33 Lars2
- 34 3110052M02Rik
- 35 Cldn11
- 36 Vstm2l

- 37 Pcsk1n
- 38 Dgkz
- 39 Tlx3
- 40 C1qtnf4

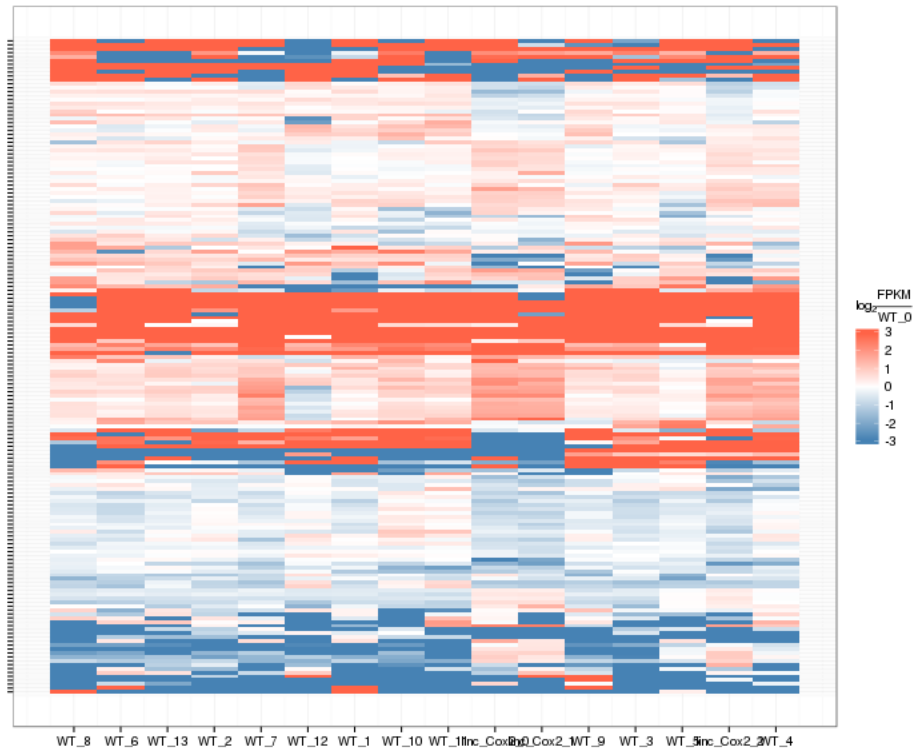
41 2310036O22Rik
42 Fmod
43 Dlgap3
44 Reln
45 Hebp1
46 Olfm12a
47 Hmga1
48 Dmrta2
49 Mafa
50 Col6a3
51 Tagap1
52 Hbb-y
53 Zfp771
54 Pign
55 Capn11
56 Sox11
57 Sp9
58 Gpr98
59 Ssbp4
60 Jund
61 Gpr27
62 9030025P20Rik
63 Gm11273
64 Ccdc85c

65 Myl6
66 Ccdc85b
67 Gm6472
68 Gm9825
69 Gm26917
70 Gm26924

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

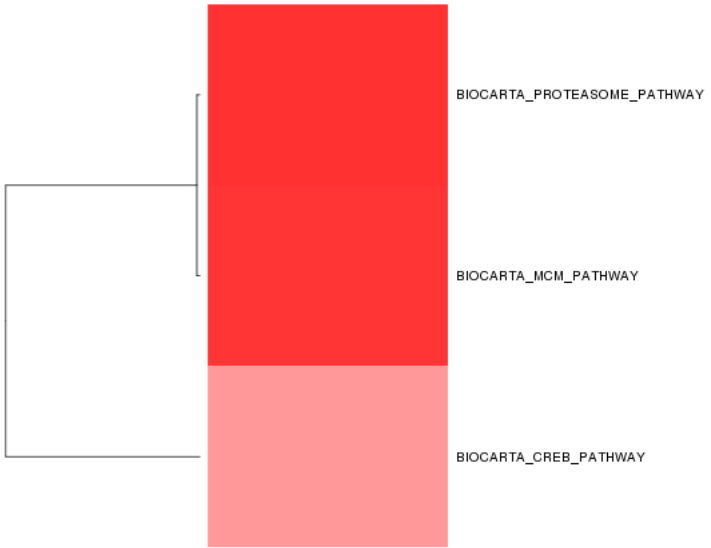
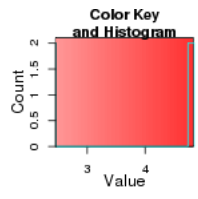
Gene/Pathway Analysis

GSEA

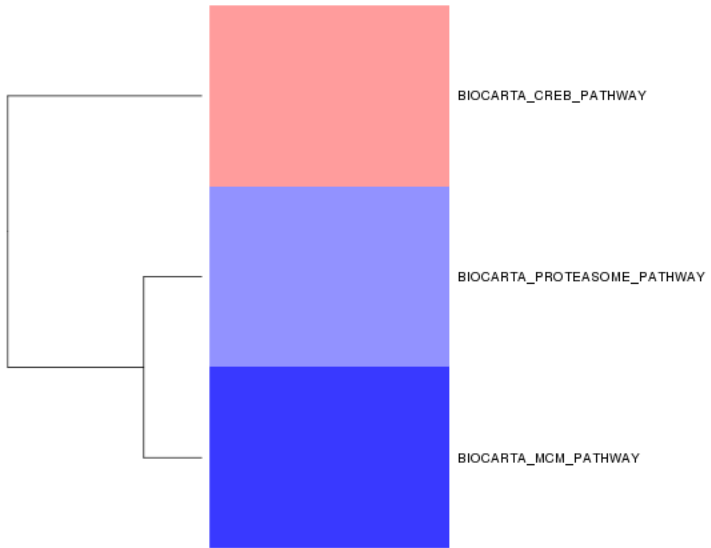
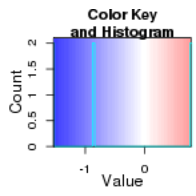
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

Biocarta enrichment:

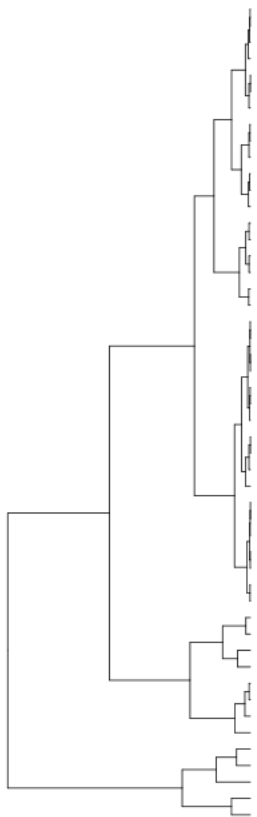


Biocarta zscore:



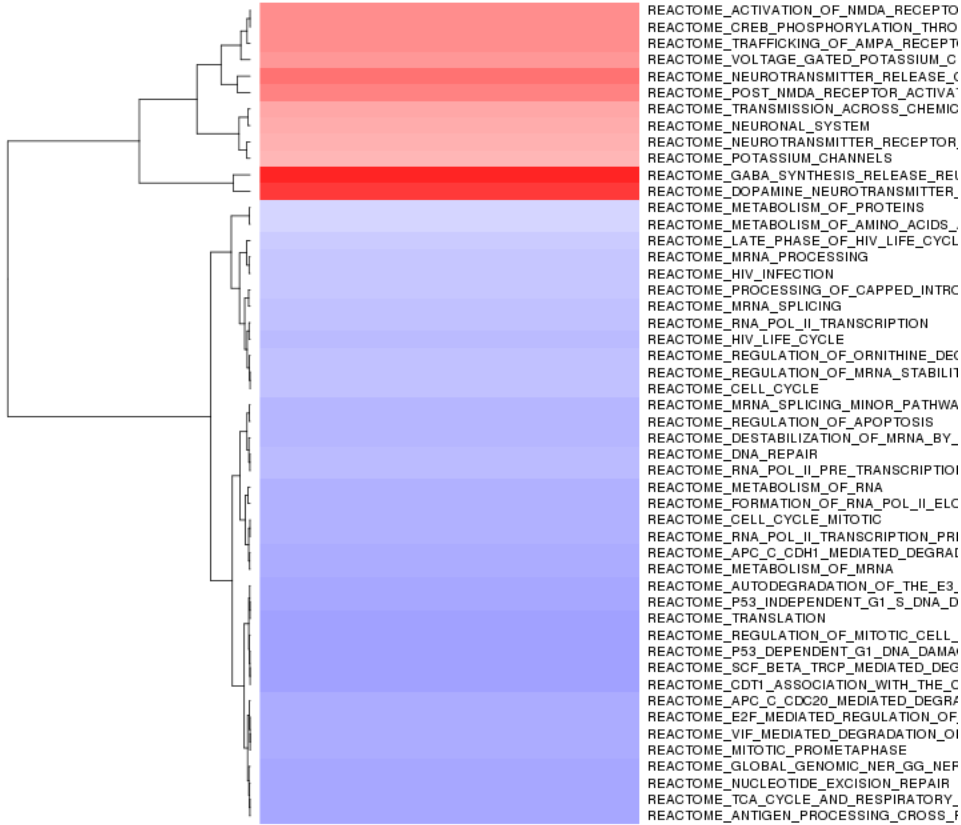
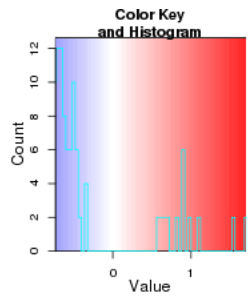
Reactome enrichment:

```
## Error: no locations are finite
```

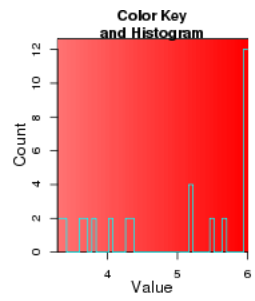


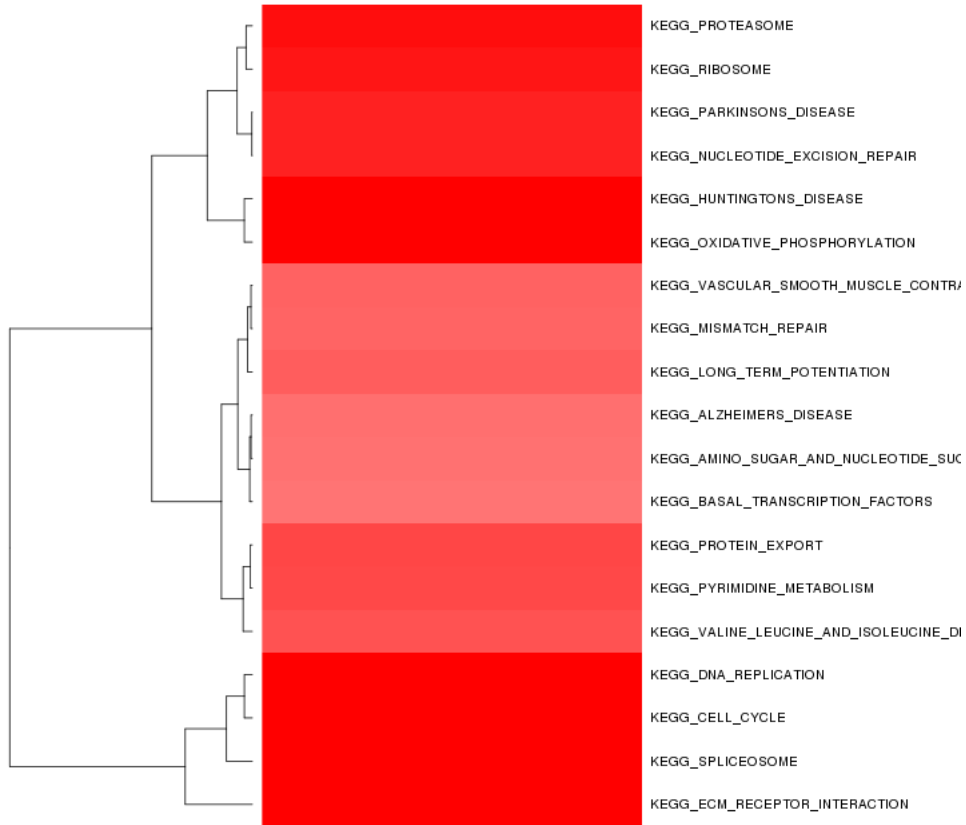
REACTOME_REGULATION_OF_MITOTIC_CELL_
REACTOME_DNA_STRAND_ELONGATION
REACTOME_DNA_REPAIR
REACTOME_MRNA_SPLICING
REACTOME_CYCLIN_E_ASSOCIATED_EVENTS
REACTOME_INFLUENZA_LIFE_CYCLE
REACTOME_COLLAGEN_FORMATION
REACTOME_ACTIVATION_OF_THE_PRE_REPLI
REACTOME_G2_M_CHECKPOINTS
REACTOME_TRANSLATION
REACTOME_ASSEMBLY_OF_THE_PRE_REPLIC
REACTOME_SRP_DEPENDENT_COTRANSLATI
REACTOME_HIV_INFECTION
REACTOME_METABOLISM_OF_MRNA
REACTOME_PROCESSING_OF_CAPPED_INTRC
REACTOME_MITOTIC_PROMETAPHASE
REACTOME_ORC1_REMOVAL_FROM_CHROMA
REACTOME_MRNA_PROCESSING
REACTOME_NEUROTRANSMITTER_RECEPTOR
REACTOME_METABOLISM_OF_PROTEINS
REACTOME_GABA_SYNTHESIS_RELEASE_REI
REACTOME_HIV_LIFE_CYCLE
REACTOME_INFLUENZA_VIRAL_RNA_TRANSCF
REACTOME_ANTIGEN_PROCESSING_CROSS_f
REACTOME_SCF5KP2_MEDIATED_DEGRADATI
REACTOME_NEUROTRANSMITTER_RELEASE_C
REACTOME_ER_PHAGOSOME_PATHWAY
REACTOME_RESPIRATORY_ELECTRON_TRANF
REACTOME_CDT1_ASSOCIATION_WITH_THE_C
REACTOME_ACTIVATION_OF_ATR_IN_RESPON
REACTOME_RNA_POL_II_TRANSCRIPTION
REACTOME_TCA_CYCLE_AND_RESPIRATORY_
REACTOME_RNA_POL_II_TRANSCRIPTION_PRI
REACTOME_REGULATION_OF_MRNA_STABILIT
REACTOME_TRANSCRIPTION_COUPLED_NER_
REACTOME_METABOLISM_OF_AMINO_ACIDS_
REACTOME_CROSS_PRESENTATION_OF_SOLI
REACTOME_S_PHASE
REACTOME_TRANSMISSION_ACROSS_CHEMIC
REACTOME_G1_S_TRANSITION
REACTOME_SYNTHESIS_OF_DNA
REACTOME_METABOLISM_OF_RNA
REACTOME_CELL_CYCLE_CHECKPOINTS
REACTOME_M_G1_TRANSITION
REACTOME_MITOTIC_G1_G1_S_PHASES
REACTOME_MITOTIC_M_M_G1_PHASES
REACTOME_NEURONAL_SYSTEM
REACTOME_CELL_CYCLE
REACTOME_DNA_REPLICATION
REACTOME_CELL_CYCLE_MITOTIC

Reactome zscore:

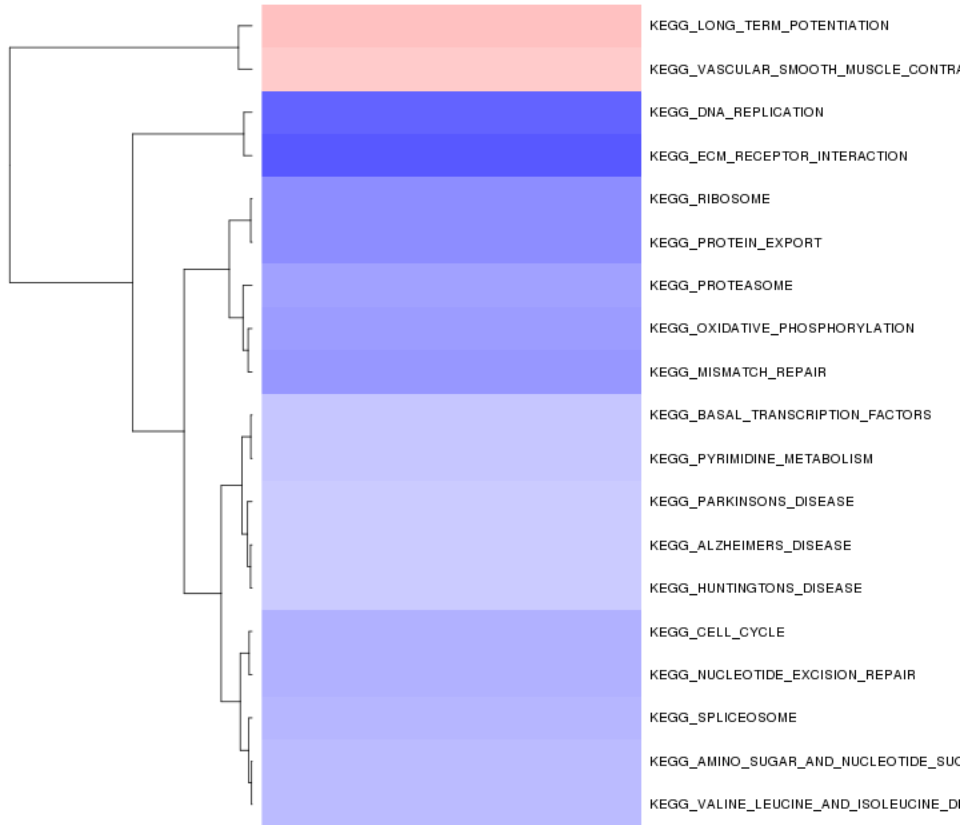
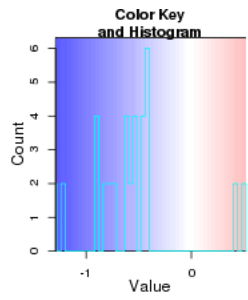


Kegg enrichment:





Kegg zscore:



Interneuron enrichment:

```
## Error: argument is of length zero
```

Interneuron zscore:

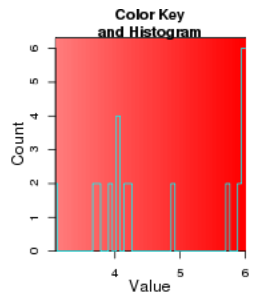
```
## Error: incorrect number of dimensions
```

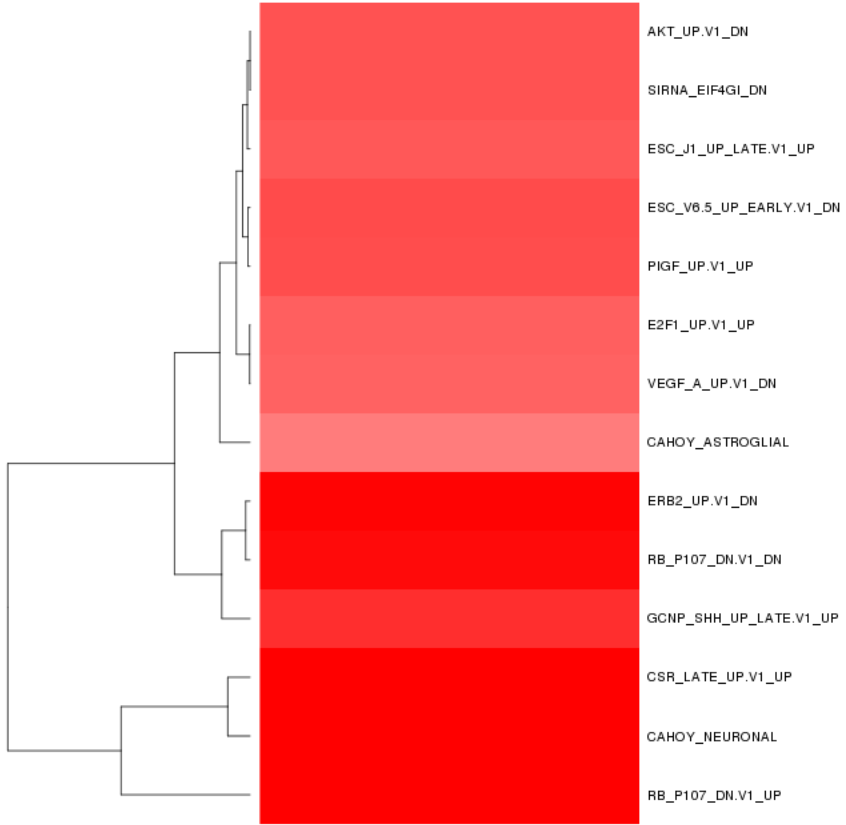
```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

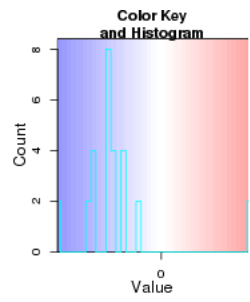
```
## Error: object 'x_ordered' not found
```

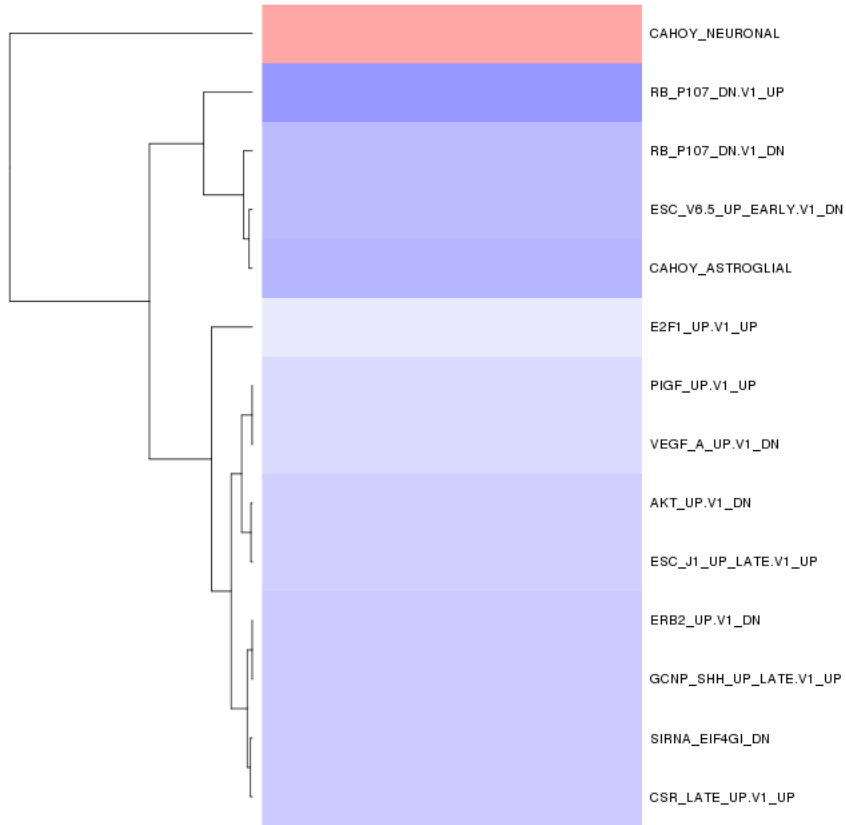
Oncogene enrichment:





Oncogene zscore:

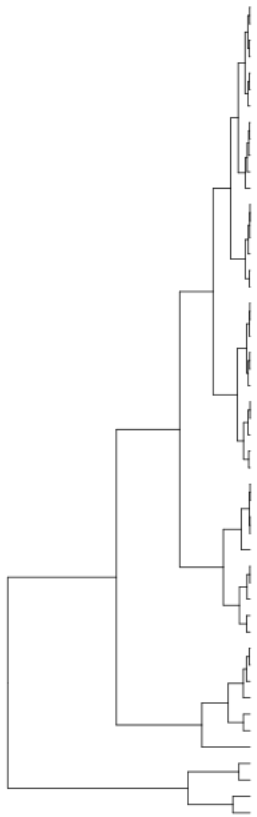




Immuno enrichment:

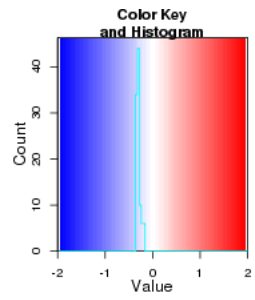
Error: no locations are finite

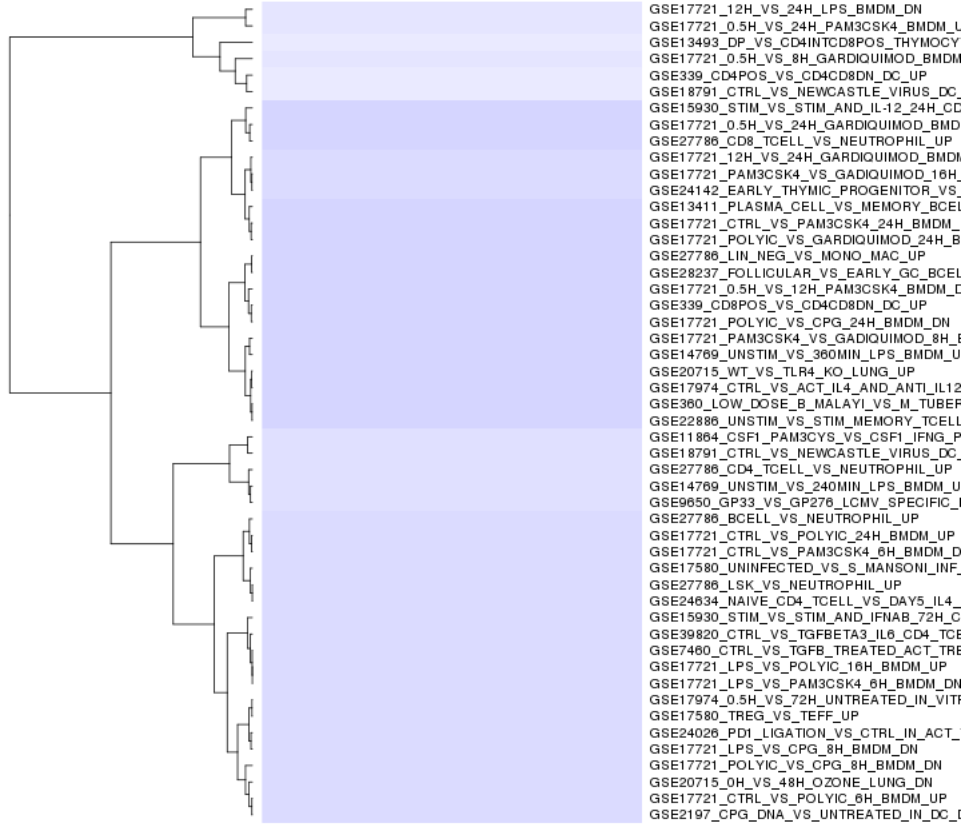




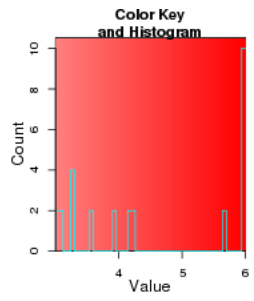
GSE17974_0H_VS_24H_IN_VITRO_ACT_CD4_T
GSE20609_CTRL_VS_COSTIM_BLOCK_MLR_CI
GSE3337_4H_VS_16H_IFNG_IN_CD8POS_DC_I
GSE30962_ACUTE_VS_CHRONIC_LCMV_SECO
GSE24634_TREG_VS_TCONV_POST_DAYS_IL4
GSE31082_DN_VS_CD8_SP_THYMOCYTE_UP
GSE15930_NAIVE_VS_72H_IN_VITRO_STIM_TF
GOLDRATH_EFF_VS_MEMORY_CD8_TCELL_UP
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IL
GSE29617_CTRL_VS_DAY3_TIV_FLU_VACCINE
GSE24634_NAIVE_CD4_TCELL_VS_DAY3_IL4_
GSE30476_CTRL_VS_TSST_ACT_16H_MEMOR
GSE9050_NAIVE_VS_EFF_CD8_TCELL_DN
GSE31082_DN_VS_CD4_SP_THYMOCYTE_UP
GSE10239_NAIVE_VS_DAY4.5_EFF_CD8_TCEL
GSE22886_UNSTIM_VS_IL2_STIM_NKCELL_DN
GSE17974_CTRL_VS_ACT_IL4_AND_ANTI_IL12
GSE14000_TRANSLATED_RNA_VS_MRNA_DC_
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_CI
GSE24634_TEFF_VS_TCONV_DAYS_IN_CULTU
KAECH_NAIVE_VS_DAY8_EFF_CD8_TCELL_DN
GSE24634_NAIVE_CD4_TCELL_VS_DAY7_IL4_
GSE29617_CTRL_VS_TIV_FLU_VACCINE_PBM
GSE3982_CENT_MEMORY_CD4_TCELL_VS_TH
GSE3982_MAC_VS_NEUTROPHIL_LPS_STIM_U
GSE24634_TEFF_VS_TCONV_DAY10_IN_CULTI
GSE3982_MEMORY_CD4_TCELL_VS_TH2_DN
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IL
GSE30962_PRIMARY_VS_SECONDARY_ACUTE
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IF
GSE9050_EFFECTOR_VS_MEMORY_CD8_TCEL
GSE22886_NAIVE_CD4_TCELL_VS_48H_ACT_
GSE7764_IL15_TREATED_VS_CTRL_NK_CELL_
GSE29614_CTRL_VS_DAY7_TIV_FLU_VACCINE
GSE24634_TREG_VS_TCONV_POST_DAY10_IL
GSE12845_NAIVE_VS_PRE_GC_TONSIL_BCEL
GSE22886_UNSTIM_VS_STIM_MEMORY_TCELL
GSE30476_CTRL_VS_TSST_ACT_40H_MEMOR
GSE22886_UNSTIM_VS_IL15_STIM_NKCELL_D
KAECH_DAY8_EFF_VS_MEMORY_CD8_TCELL_
GSE30476_CTRL_VS_TSST_ACT_72H_MEMOR
GSE22886_NAIVE_CD4_TCELL_VS_48H_ACT_
GSE30476_CTRL_VS_TSST_ACT_72H_MEMOR
GSE17974_CTRL_VS_ACT_IL4_AND_ANTI_IL12
GSE30476_CTRL_VS_TSST_ACT_40H_MEMOR
GSE13485_DAY1_VS_DAY21_YF17D_VACCINE
GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL
GSE15750_DAY6_VS_DAY10_TRAF6KO_EFF_C
KAECH_DAY8_EFF_VS_DAY15_EFF_CD8_TCEL
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_CI

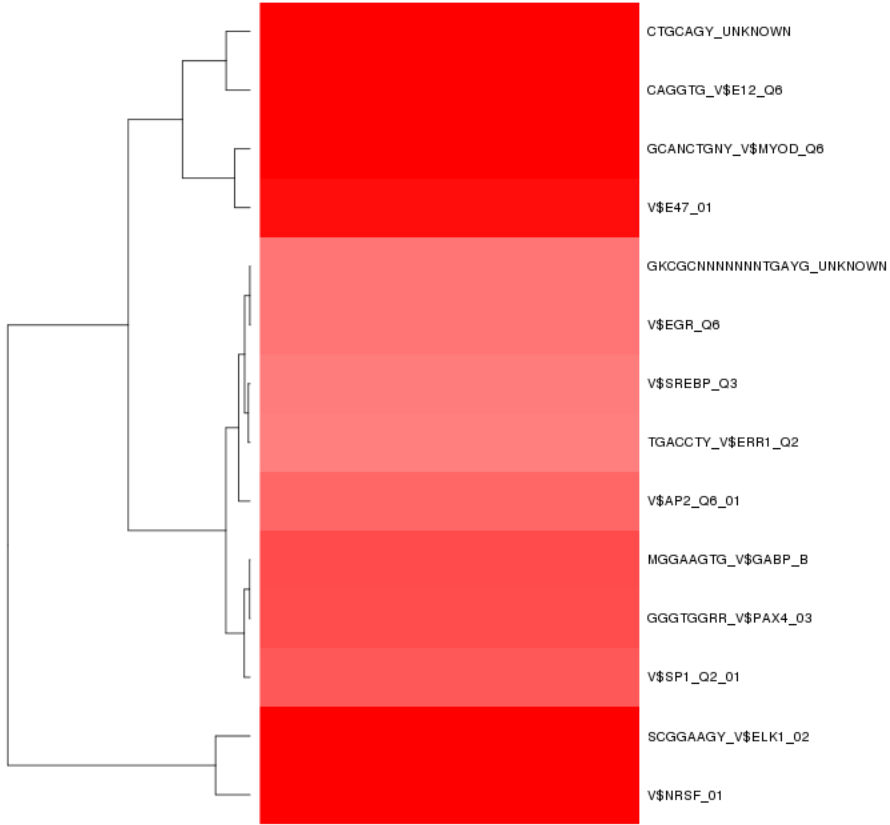
Immuno zscore:



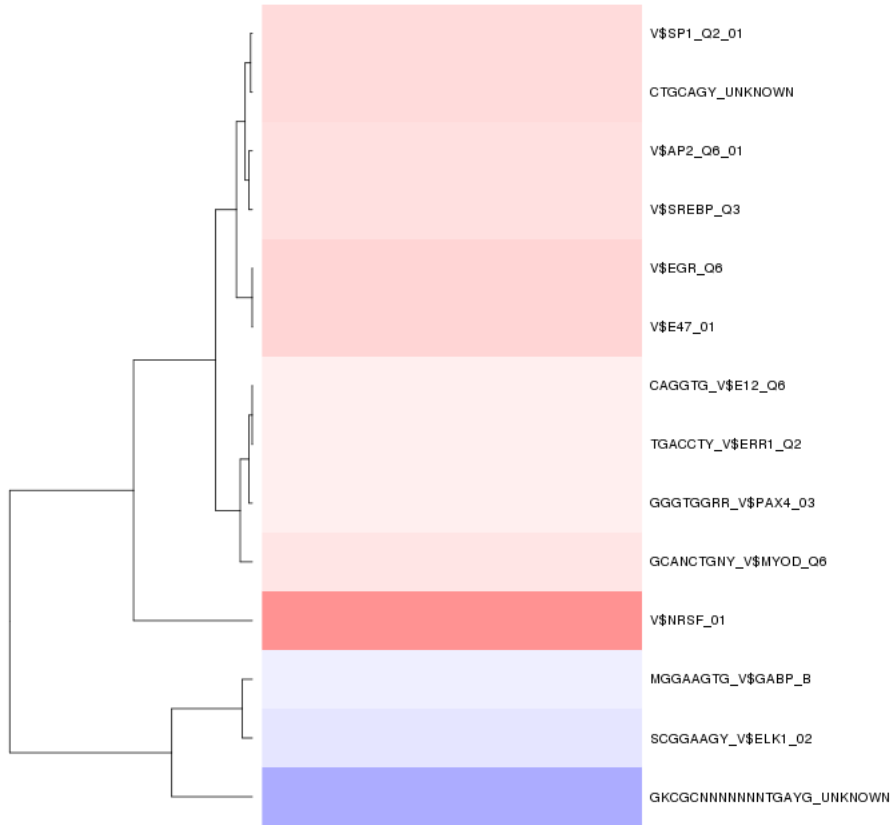
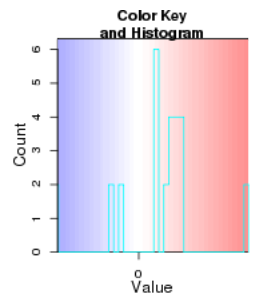


TF enrichment:



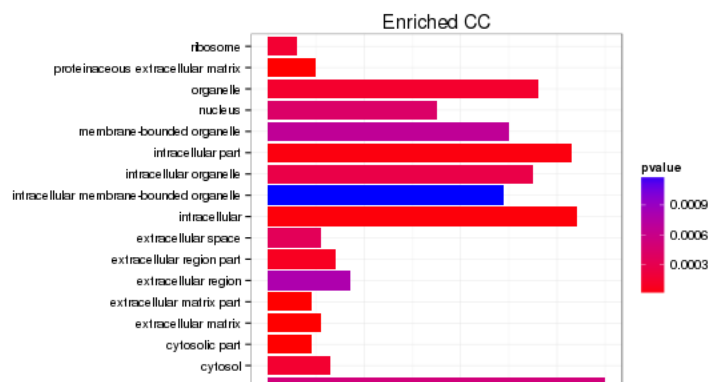
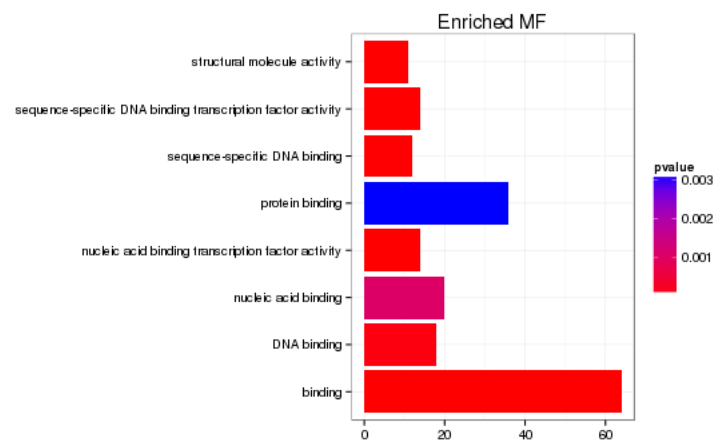
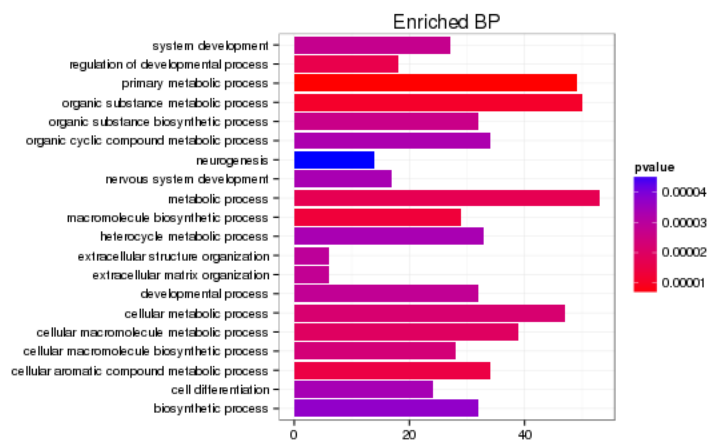


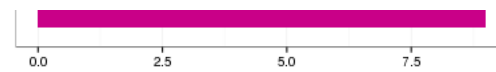
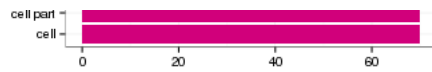
TF zscore:



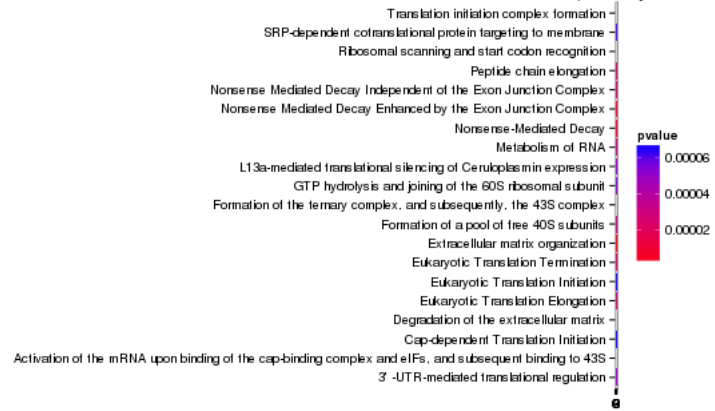
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

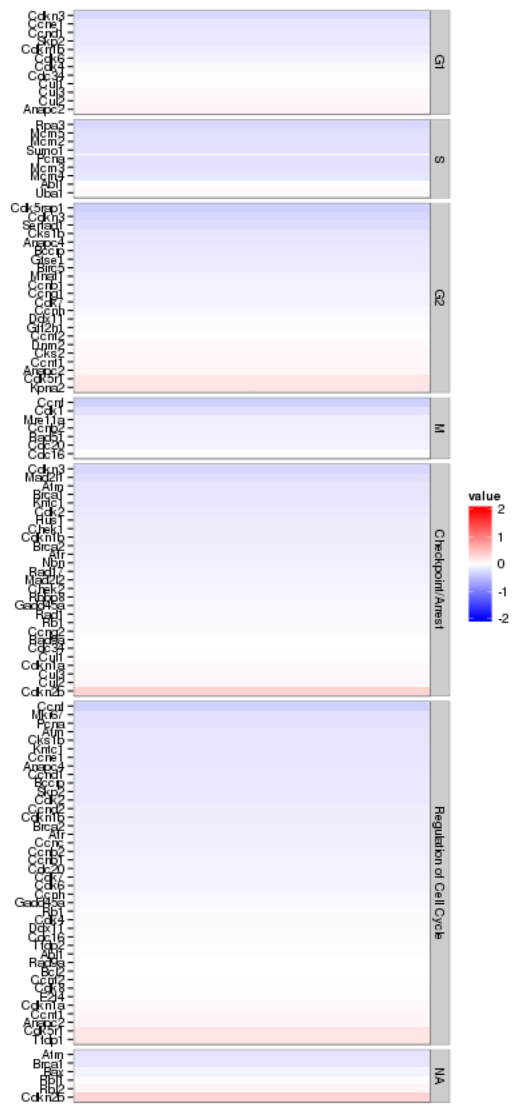




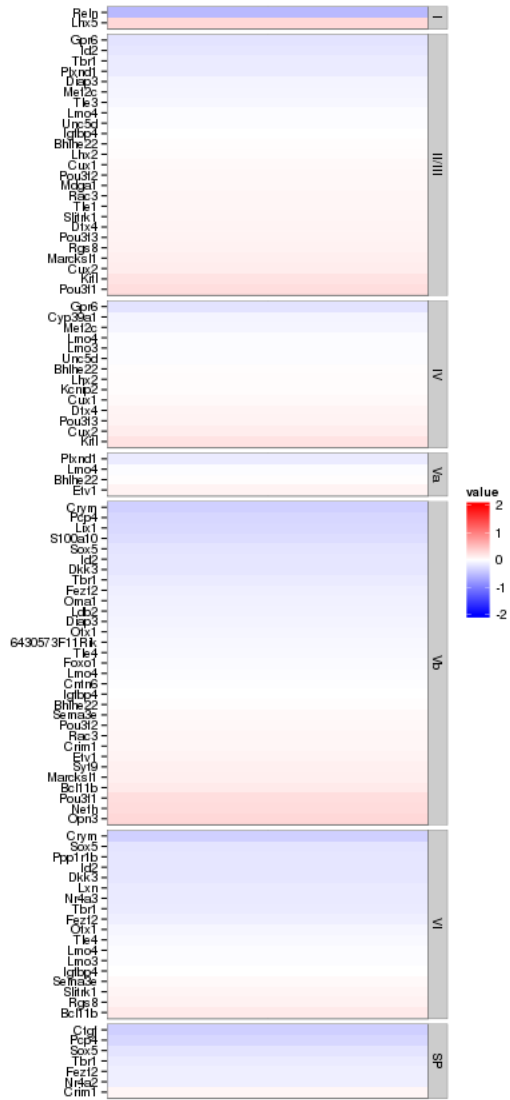
Reactome pathway enrichment



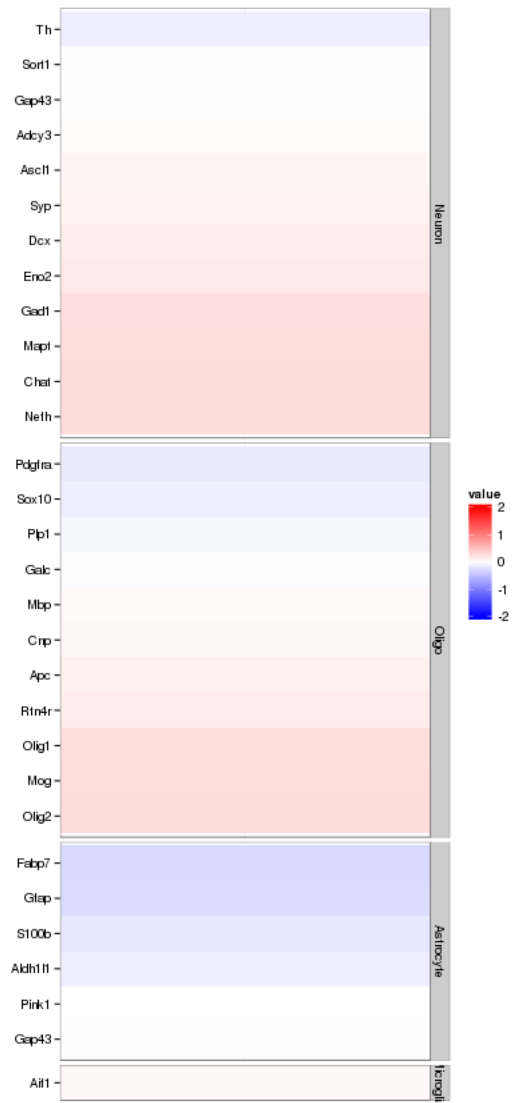
Enrichment or depletion for stage-specific cell cycle markers



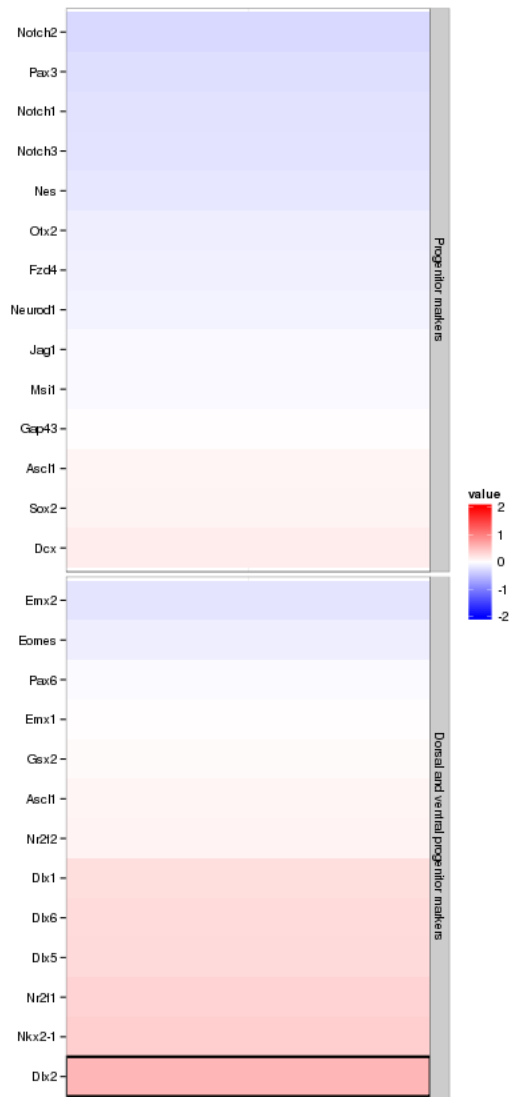
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



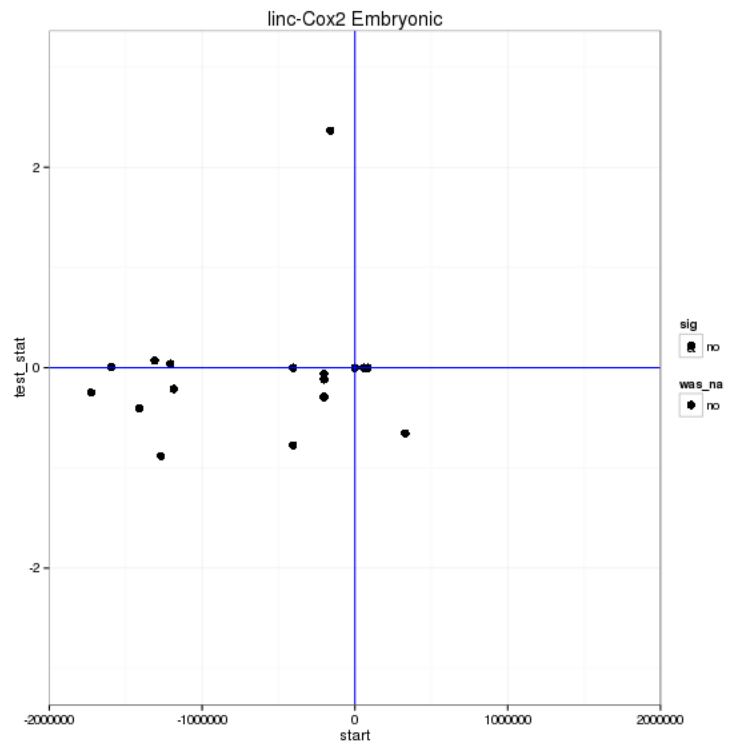
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR809
16 JR816
17 JR829

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	32194900.00	1.27	1.00
2 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	32194900.00	1.10	1.00
3 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	32194900.00	1.23	1.00
4 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	32194900.00	1.29	1.00
5 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	32194900.00	1.21	1.00
6 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	32194900.00	0.90	1.00
7 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	32194900.00	0.98	1.00
8 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	32194900.00	0.93	1.00
9 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	32194900.00	0.86	1.00
10 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	32194900.00	0.96	1.00
11 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	32194900.00	0.81	1.00
12 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	32194900.00	0.80	1.00
13 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	32194900.00	0.69	1.00
14 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	32194900.00	1.22	1.00
15 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR809/abundances.cxb	linc_Cox2	0	linc_Cox2_0	36051200.00	32194900.00	1.11	1.00
16 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR816/abundances.cxb	linc_Cox2	1	linc_Cox2_1	31829200.00	32194900.00	0.98	1.00
17 /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/quants/JR829/abundances.cxb	linc_Cox2	2	linc_Cox2_2	30833100.00	32194900.00	0.95	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats     graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,linc-Cox2 -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/linc-Cox2_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/Brai
## 2
## 3
## 4
## 5
```

linc-Enc1 KO vs WT (Adult)

This file shows the wt-v-ko comparison for linc-Enc1.

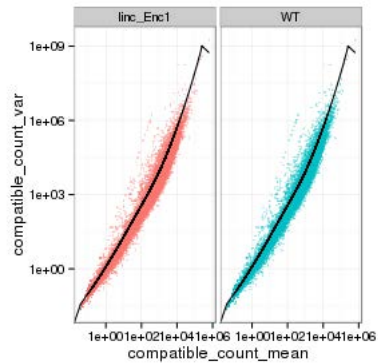
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

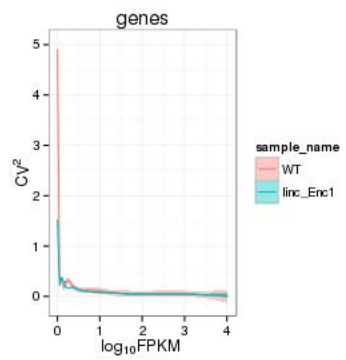
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



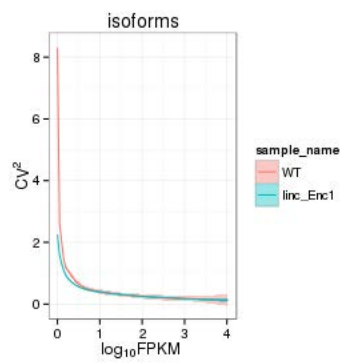
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

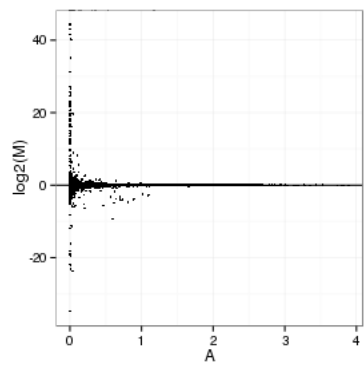
Genes:



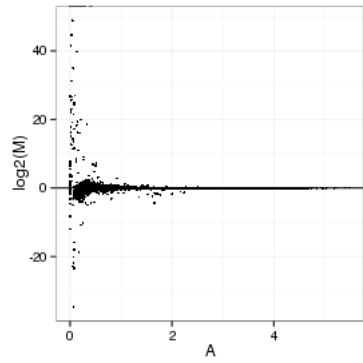
Isoforms:



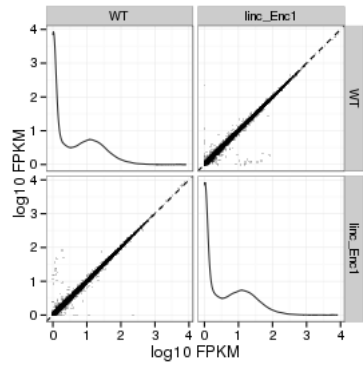
MvA plot



MvA plot counts



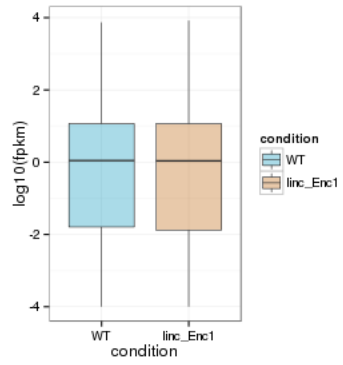
Scatterplot matrix



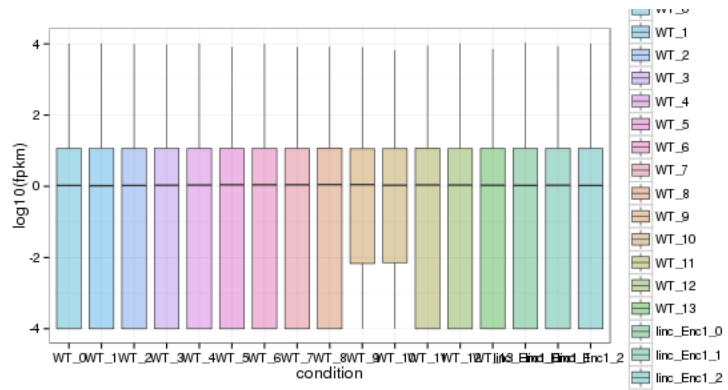
Distributions

Boxplots

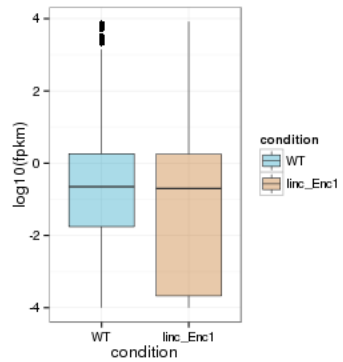
Boxplot (genes)



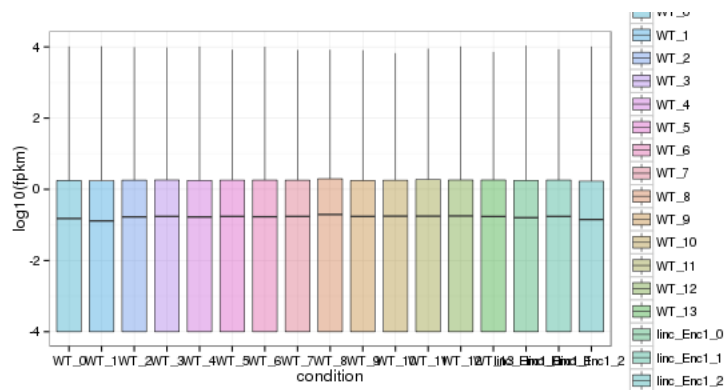
Boxplot (genes, replicates)



Boxplot (isoforms)

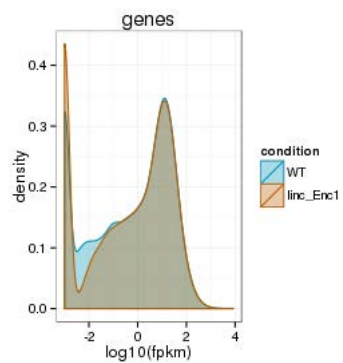


Boxplot (isoforms, replicates)

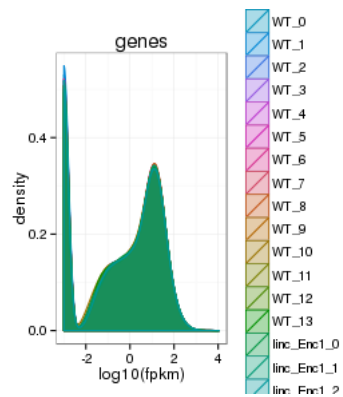


Density

Density (genes)

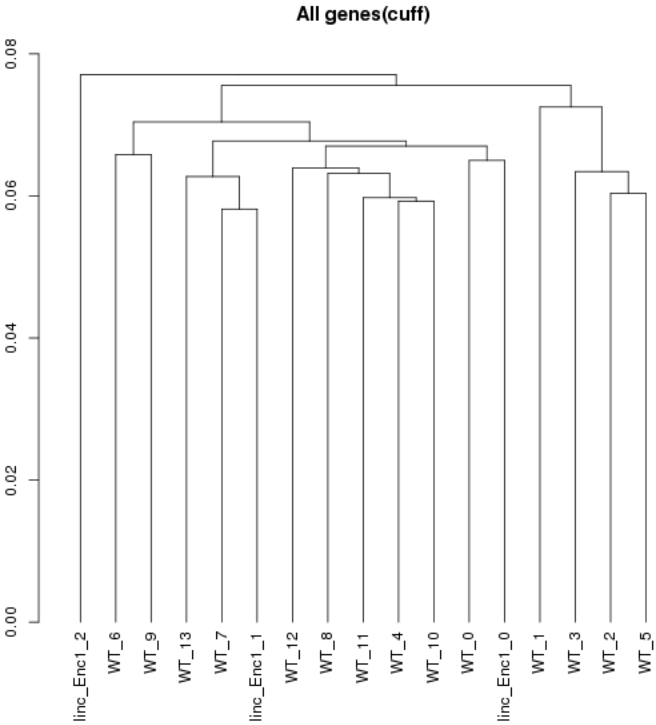


Density (genes, replicates)



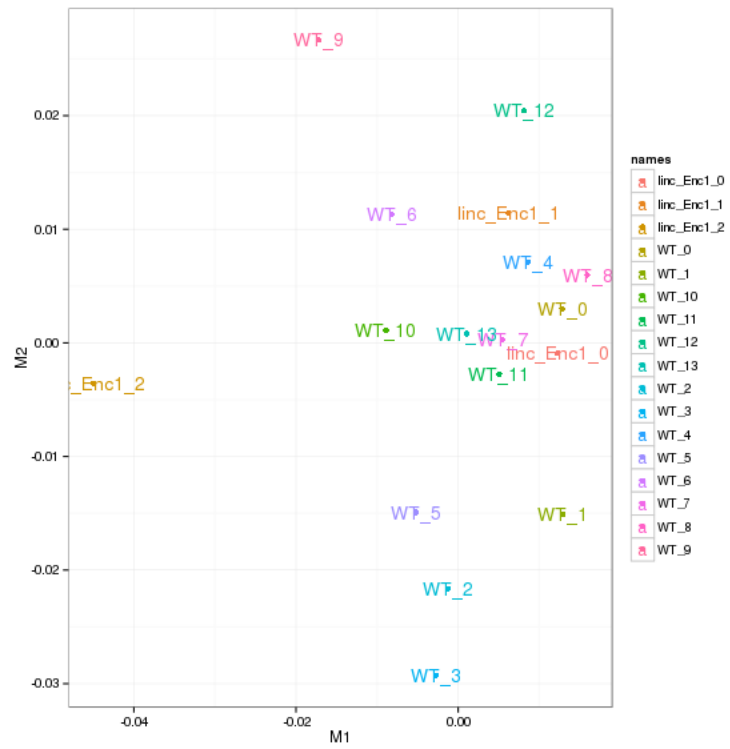
Clustering

Replicate Clusters

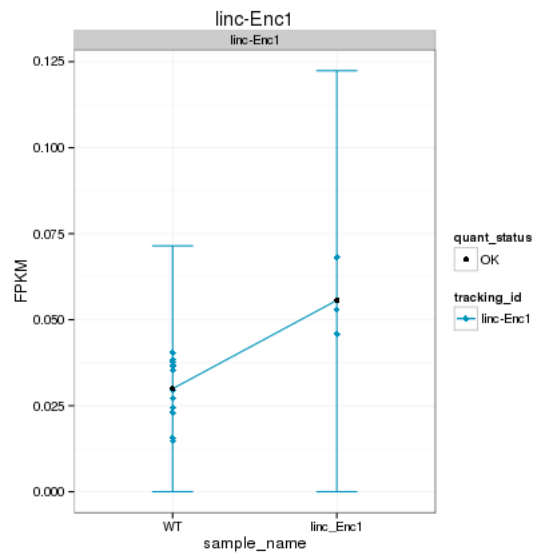


NULL

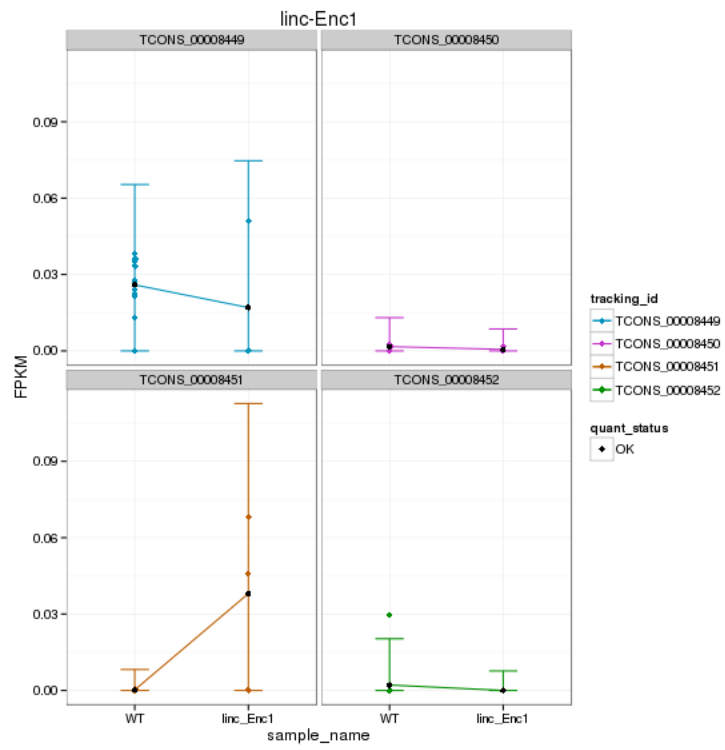
PCA (genes)



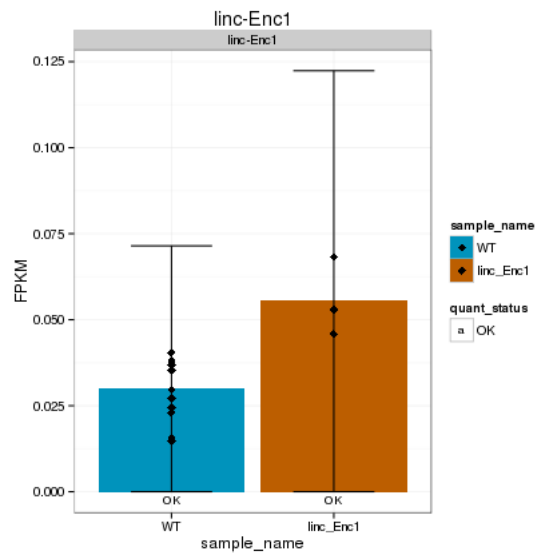
KO assessment
Endogenous lincRNA expression



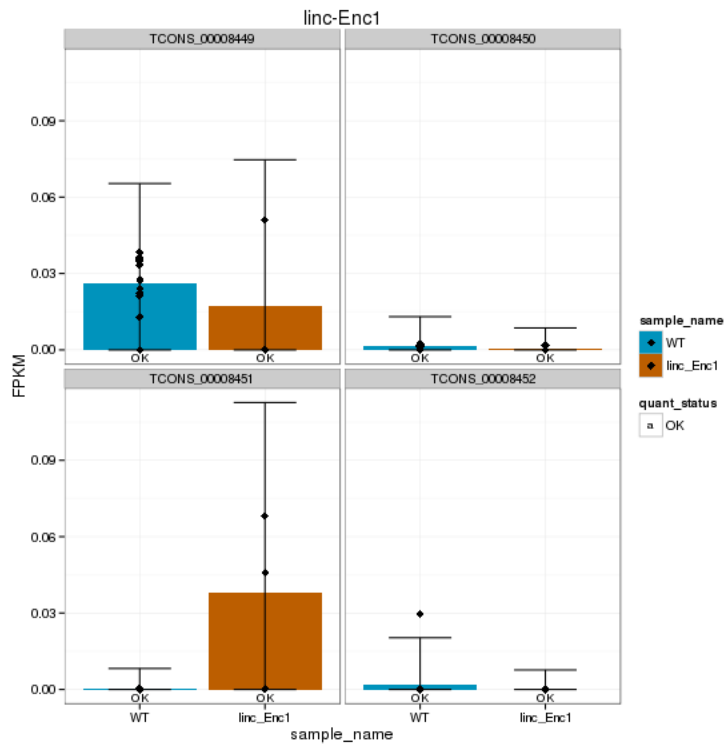
Endogenous expression of linc-Enc1 isoforms:



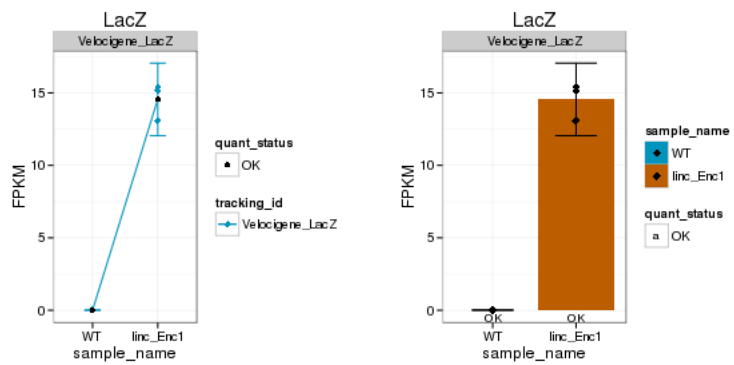
Barplot of gene expression:



Barplot of isoform expression:

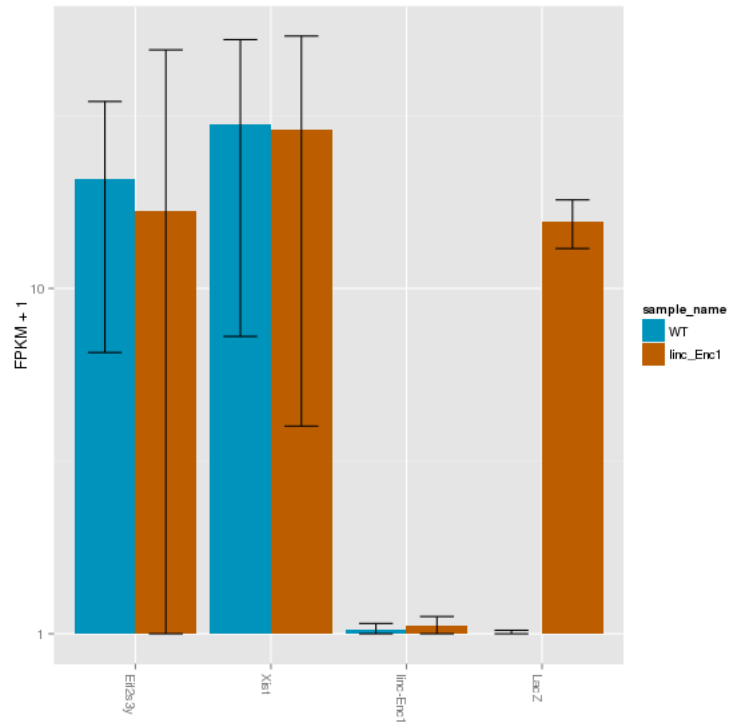


LacZ expression

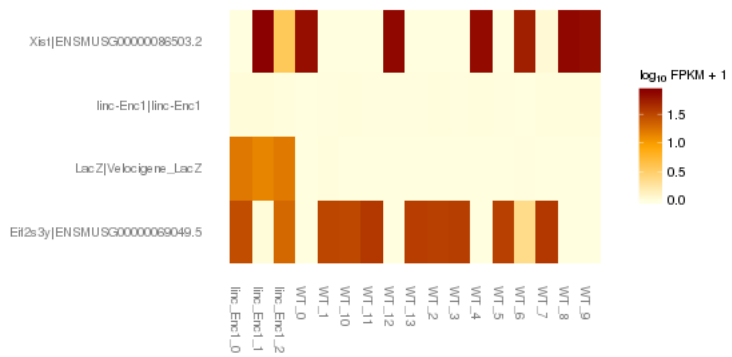


Digital Genotyping (LacZ vs Endogenous IncRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

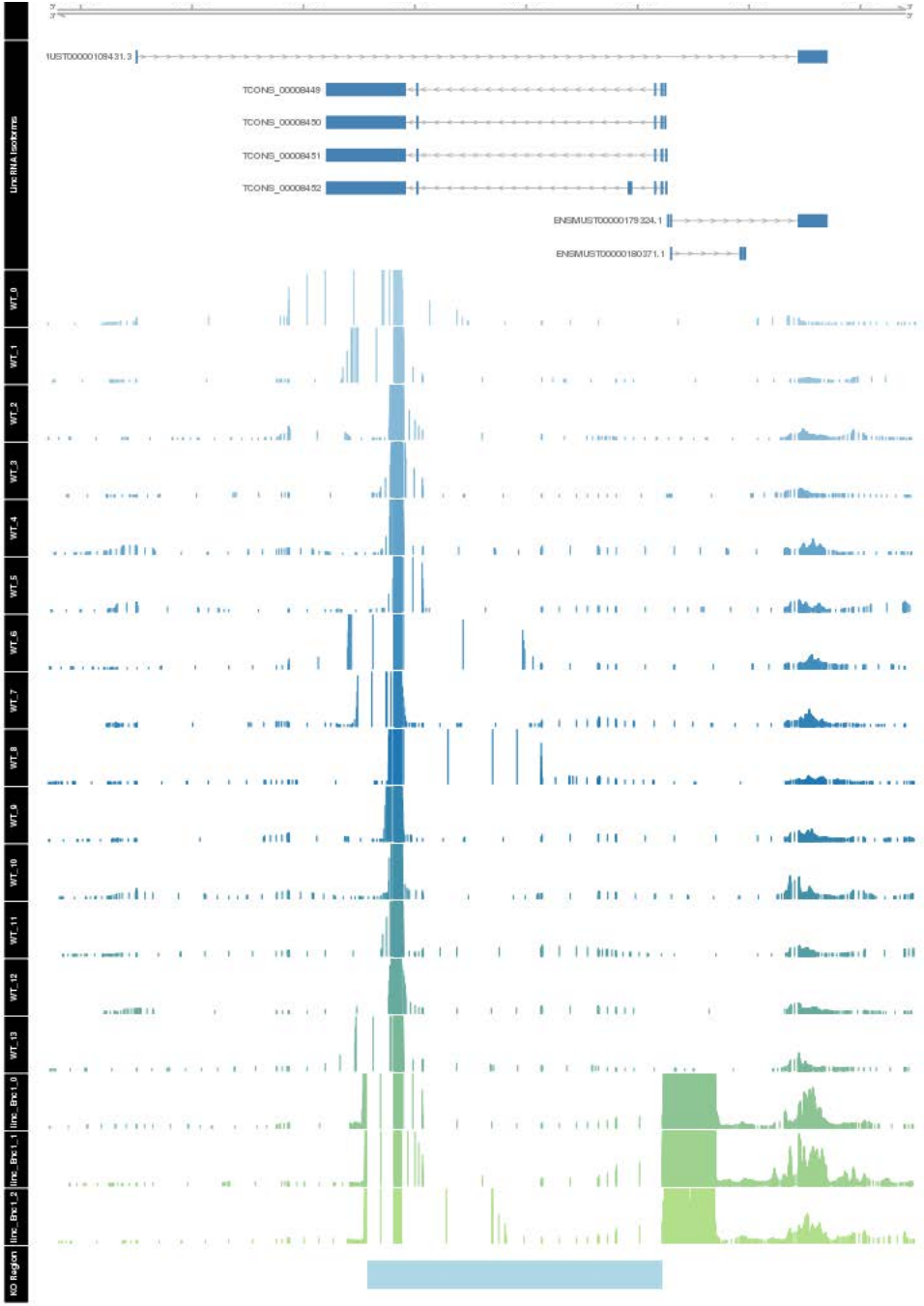


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 52 significantly differentially expressed genes. They are:

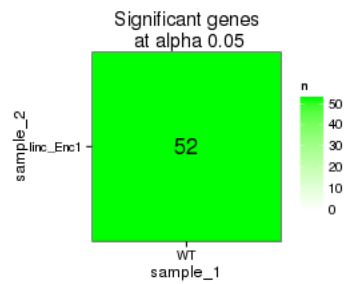
```
geneAnnot$gene_short_name
1 Nes
2 Dnajb1
3 Pttg1
4 Adi1
5 Psmb5
6 Arc
7 Mut
8 Glo1
9 Flnb
10 Nnt
11 Lcn2
12 Cyr61
13 Alad
14 Ugt2a1
15 Hddc3
16 Arhgap27
17 Tmem260
18 Hdhd3
19 Car8
20 Mup4
21 Enc1
22 Tuba1c
23 5430402E10Rik
24 Gm9493
25 Zfp488
26 Acp1
27 Cd24a
28 BC048546
29 Myh7
30 BC051076
31 Mup5
32 Cox5b
33 Rpl34
34 Obp2a
35 Obp1b
36 Obp1a
37 Bpifb9b
```

- 38 Bpifb3
- 39 Ppp1r3e
- 40 Gm7292
- 41 H2-BI
- 42 Ccl28
- 43 Gm2373

- 44 Gm14744
- 45 Obp2b
- 46 4930412C18Rik
- 47 Hspa1b
- 48 Hspa1a
- 49 AA465934
- 50 4930480K23Rik
- 51 Gm26825
- 52 Gm26924

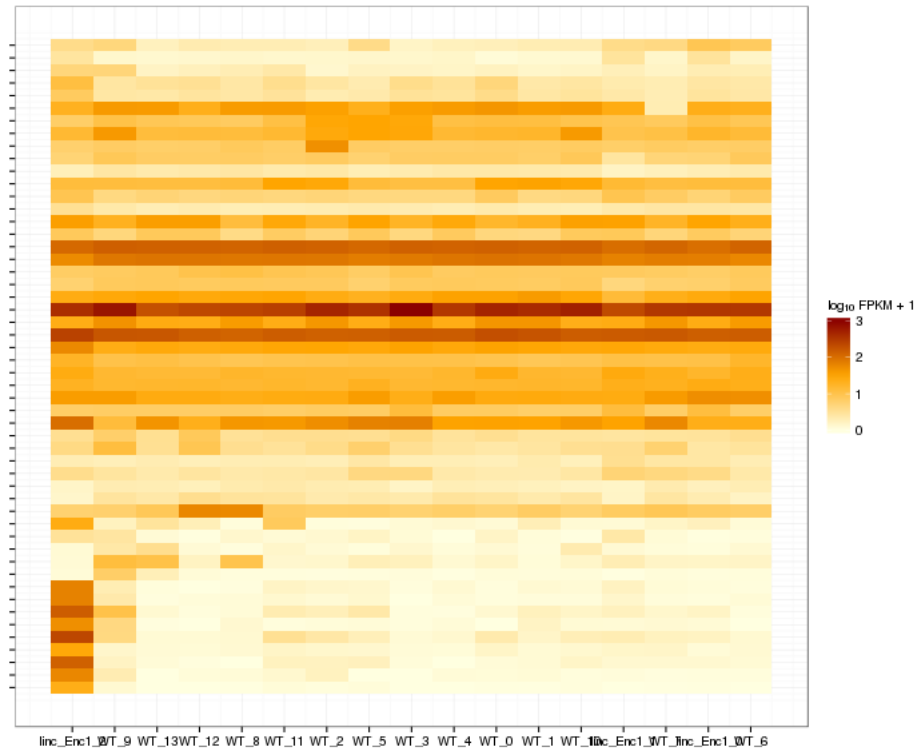
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

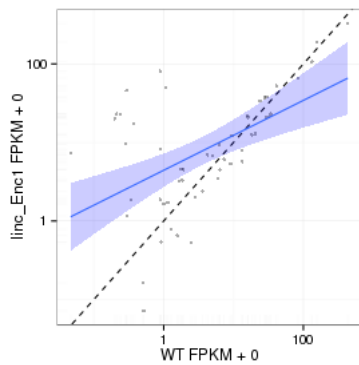


Significant genes with expression >50fpm (any condition):(turned off)

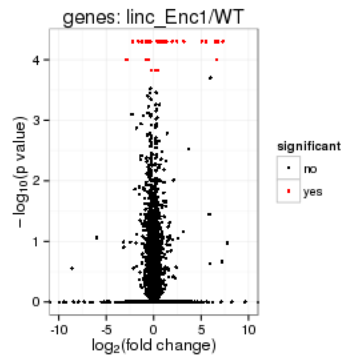
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

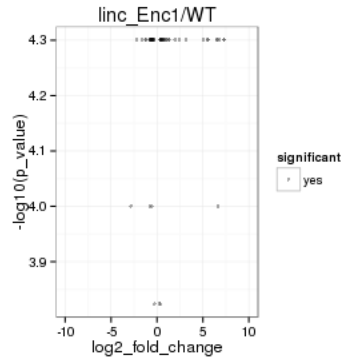
Scatter plot of significant genes only:



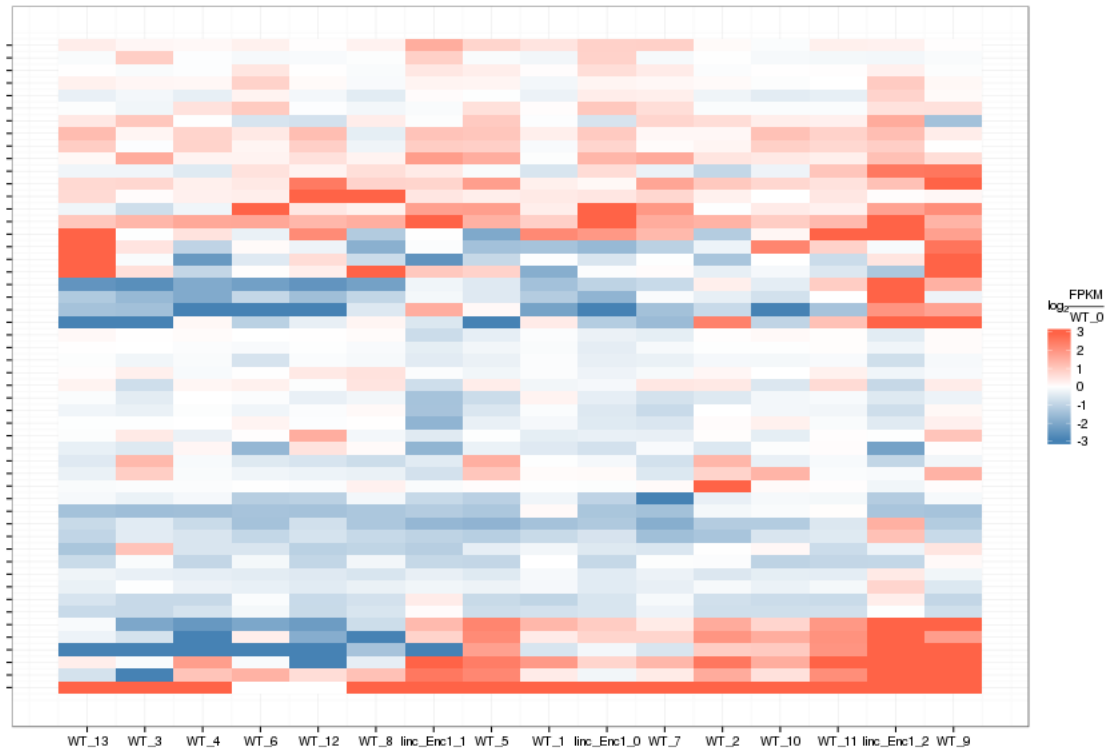
Volcano Plot



Volcano plot with significant genes only:



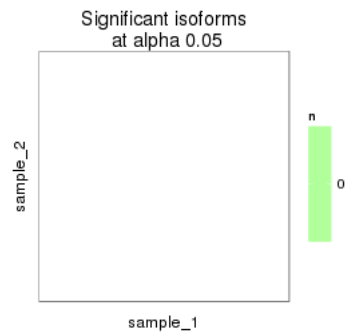
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

[1] "no sig isoforms"

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

Biocarta enrichment:

```
## Error: `x` must have at least 2 rows and 2 columns
```

Biocarta zscore:

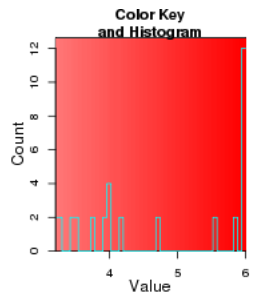
```
## Error: incorrect number of dimensions
```

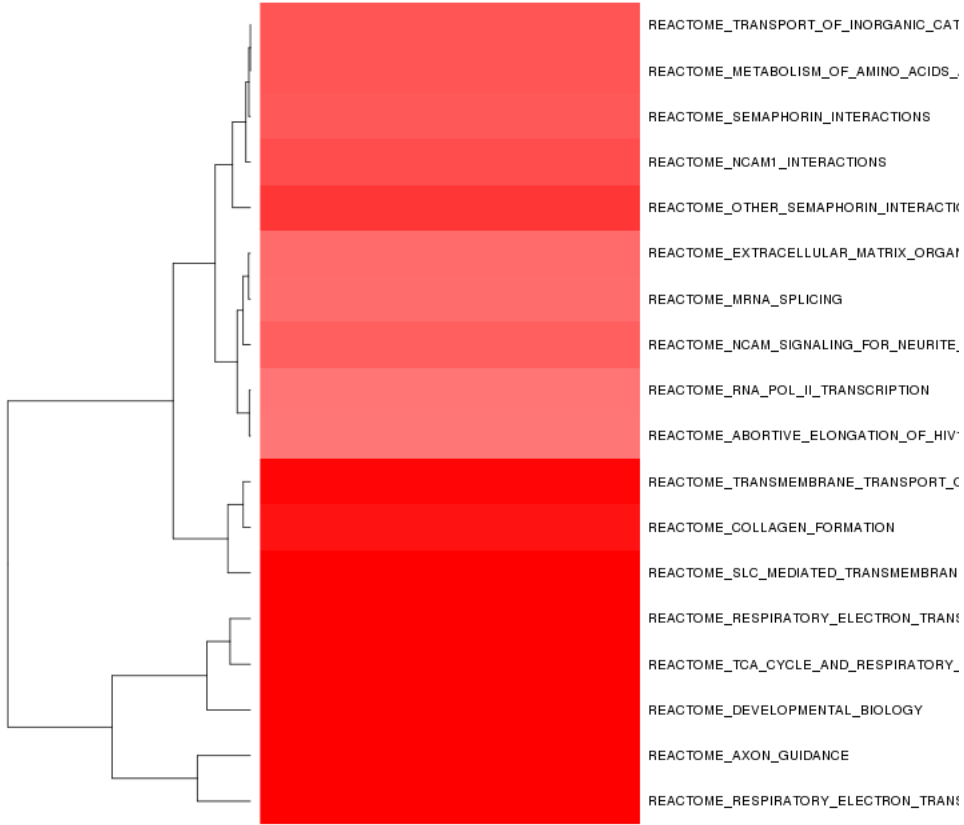
```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

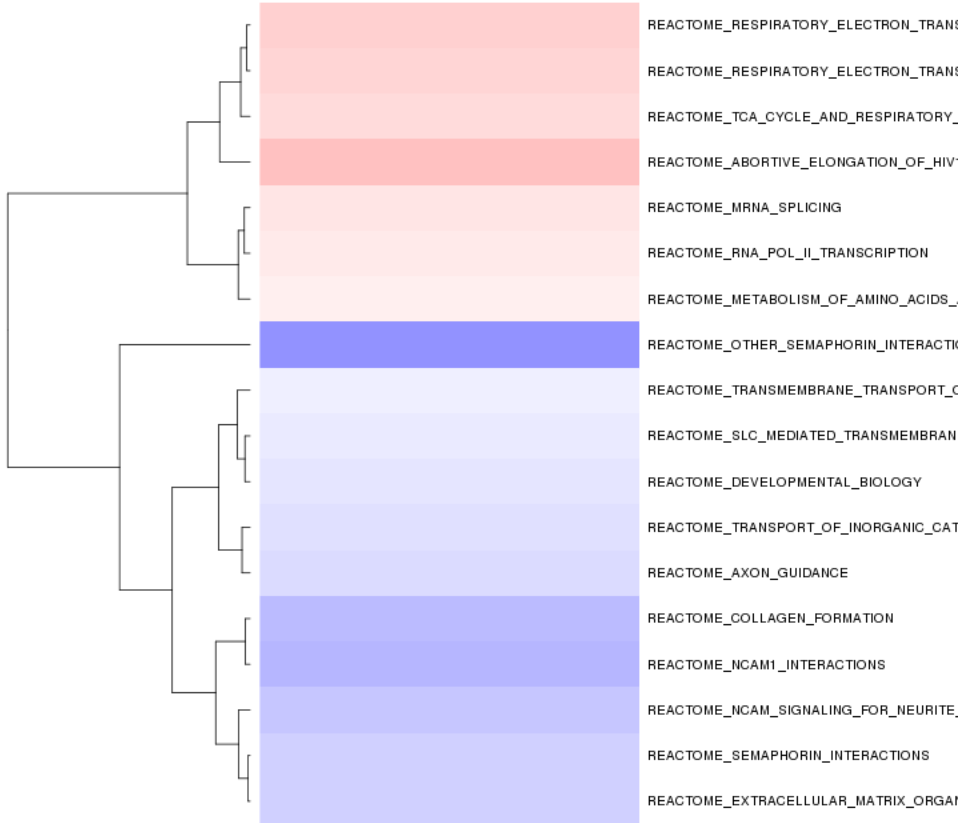
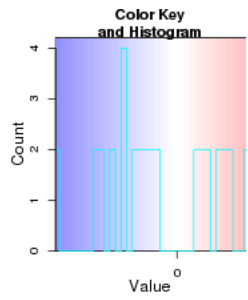
```
## Error: object 'x_ordered' not found
```

Reactome enrichment:

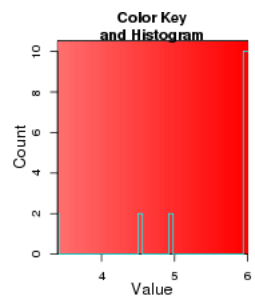


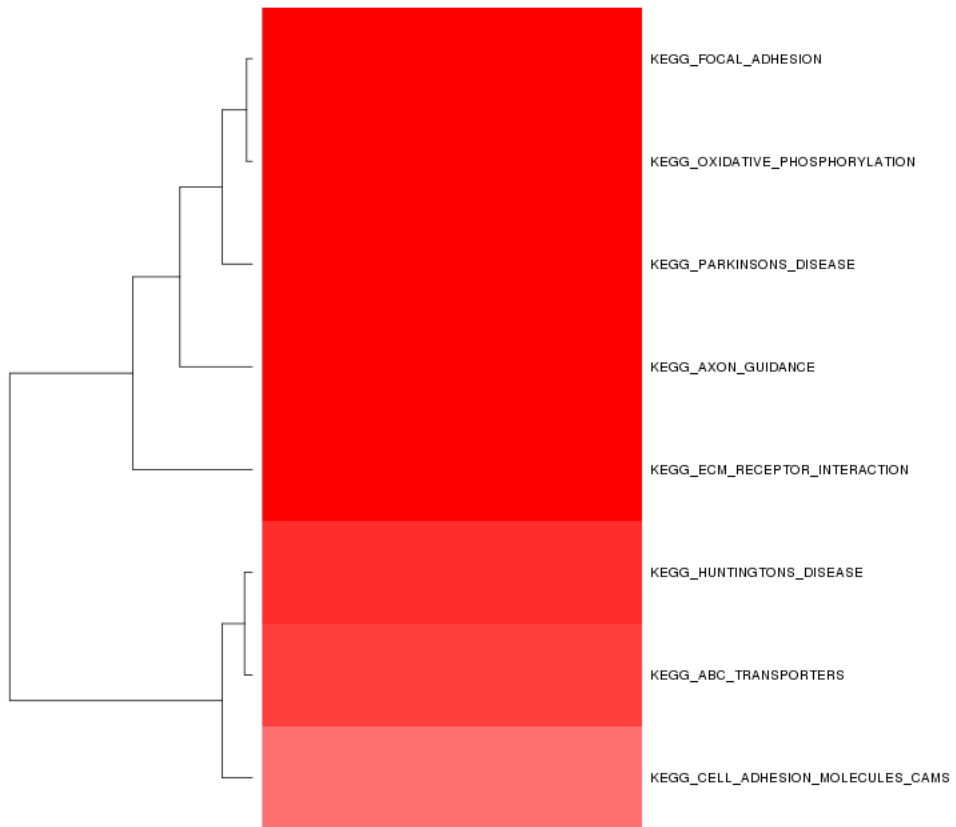


Reactome zscore:

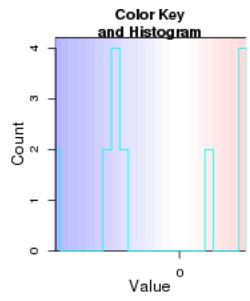


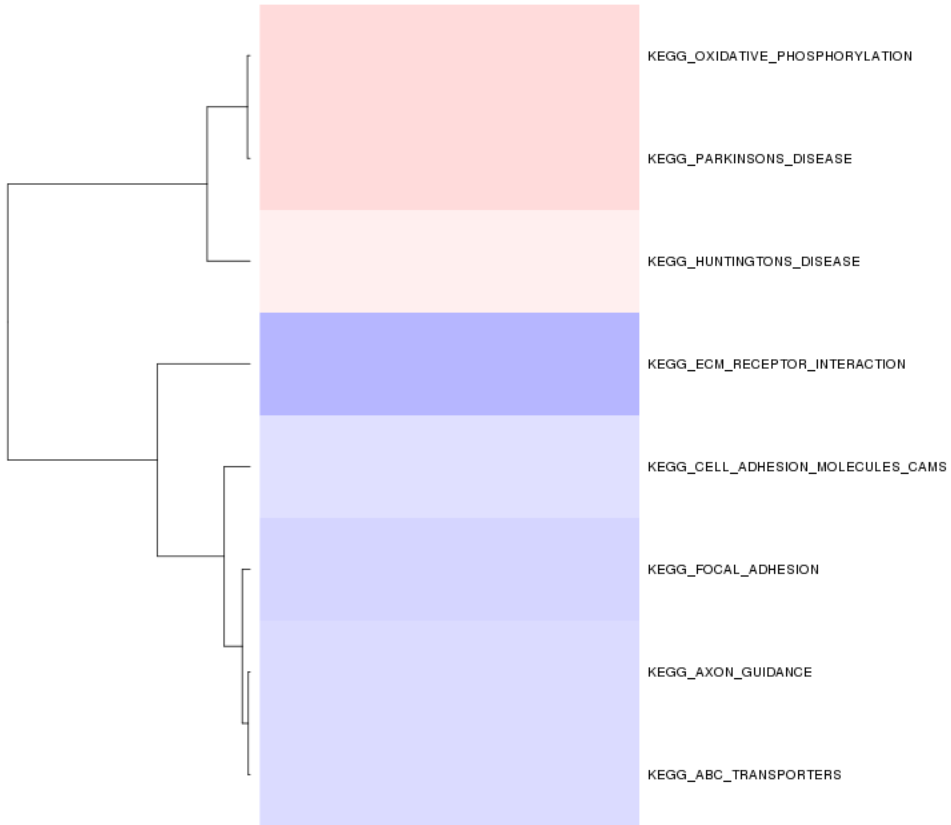
Kegg enrichment:





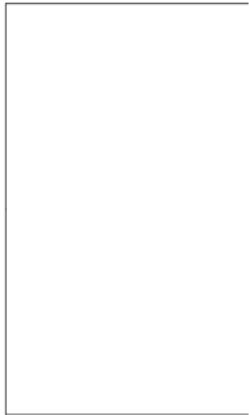
Kegg zscore:





Interneuron enrichment:

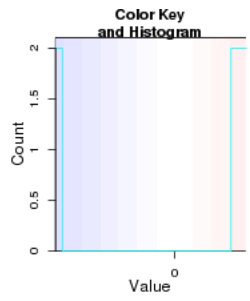
Error: no locations are finite

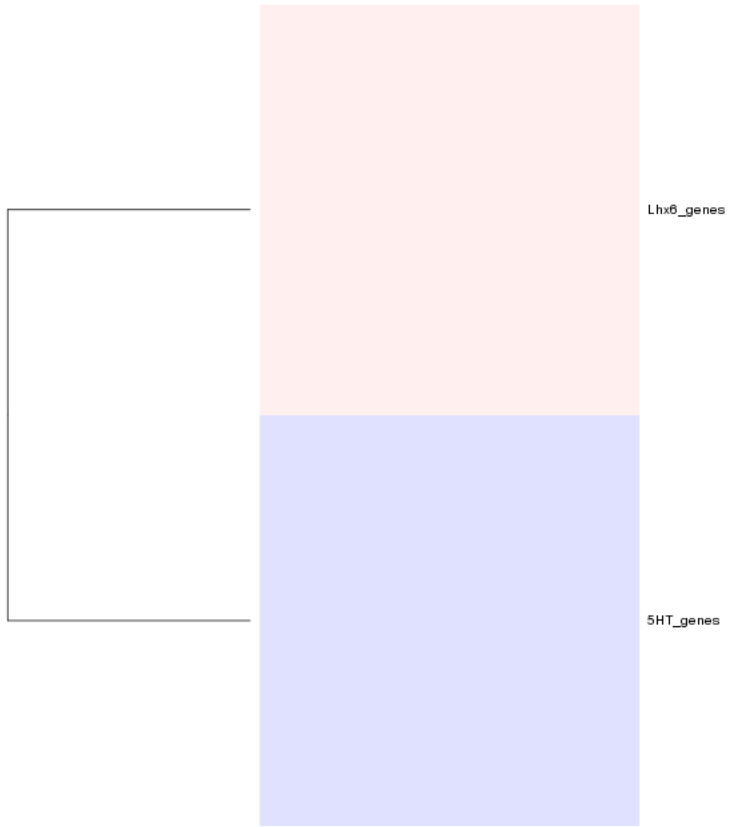


5HT_genes

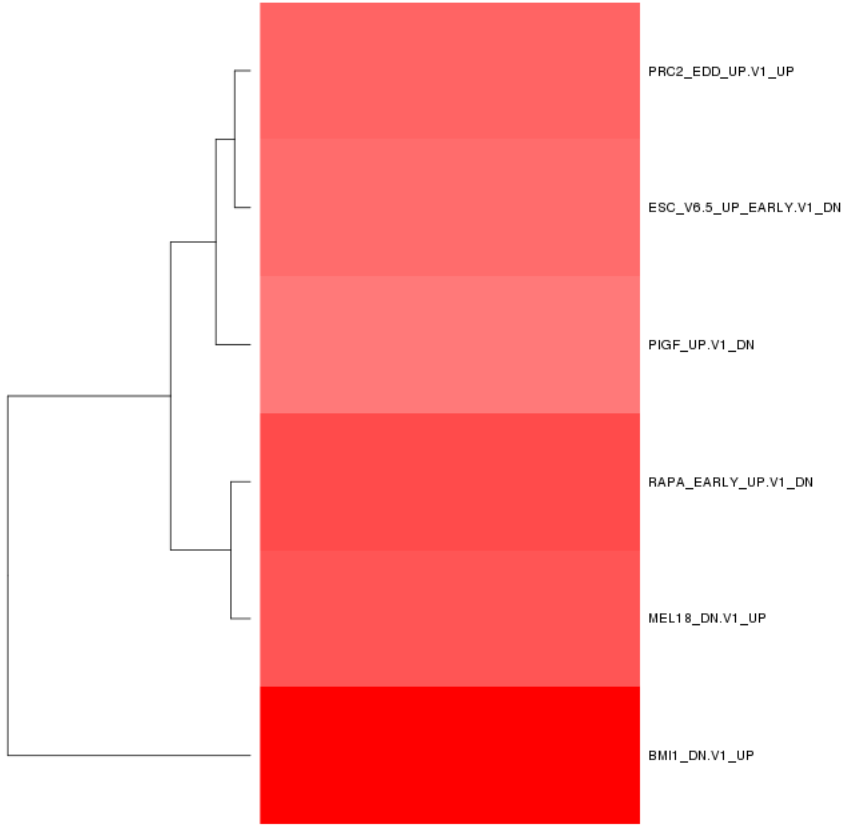
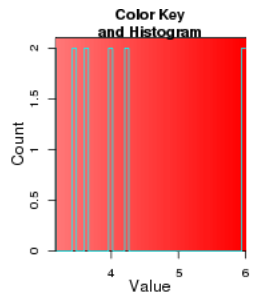
Lhx0_genes

Interneuron zscore:

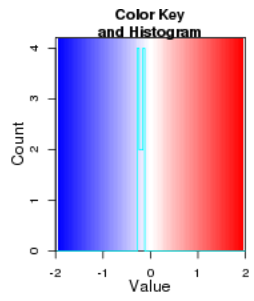


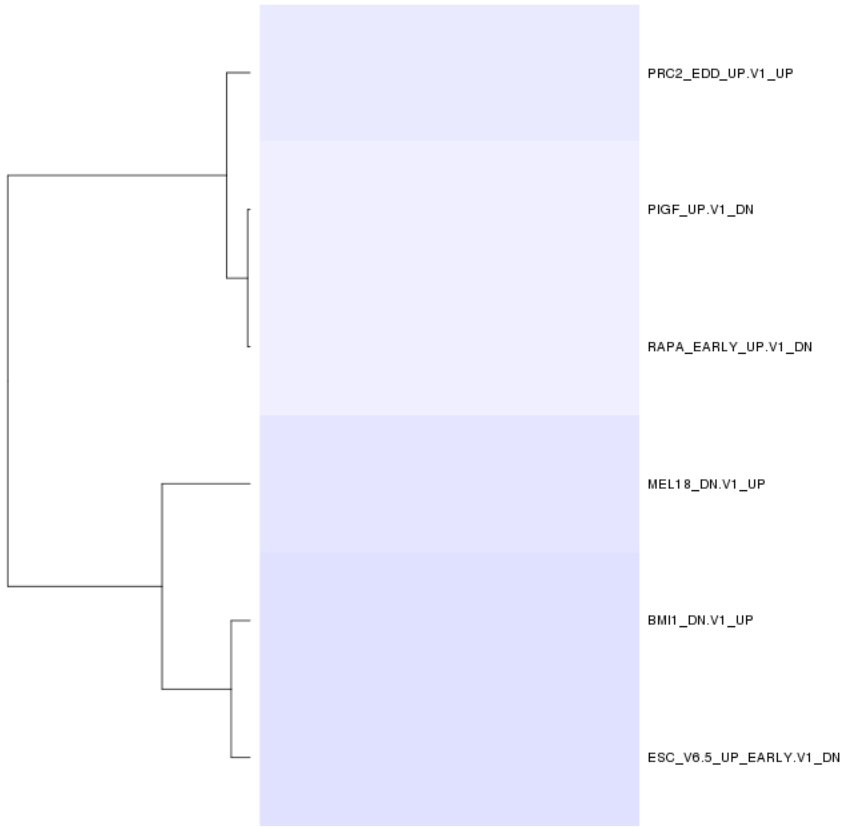


Oncogene enrichment:



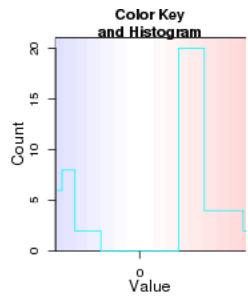
Oncogene zscore:

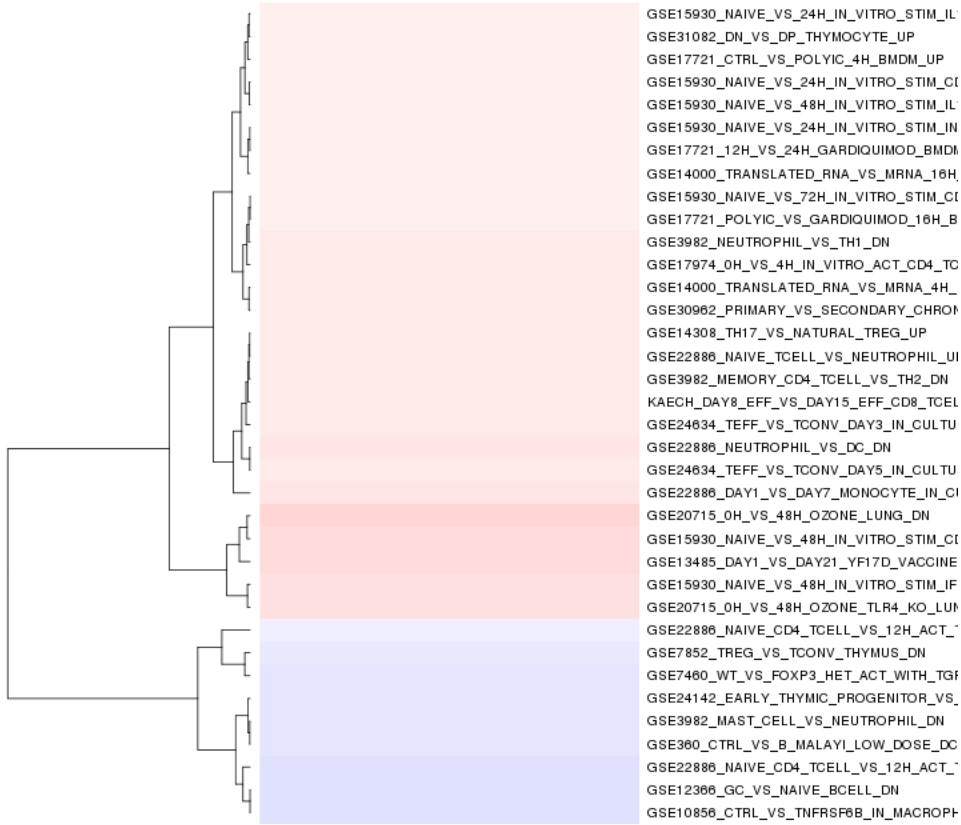




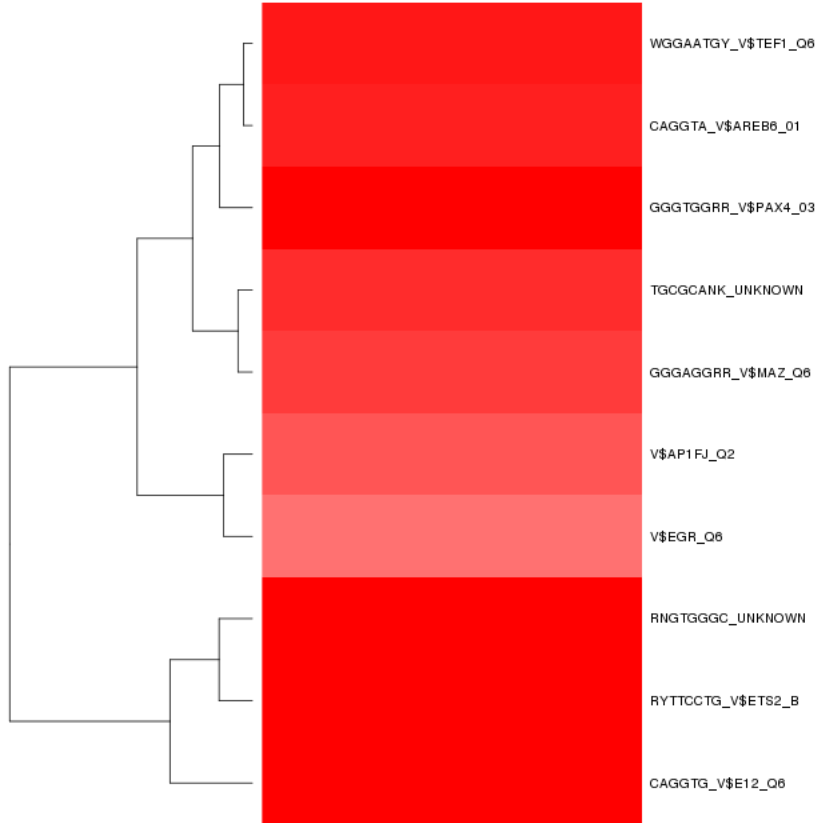
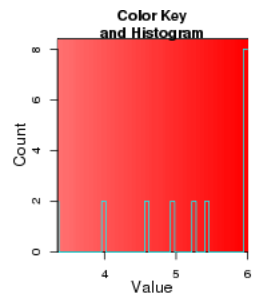
Immuno enrichment:

Error: subscript out of bounds

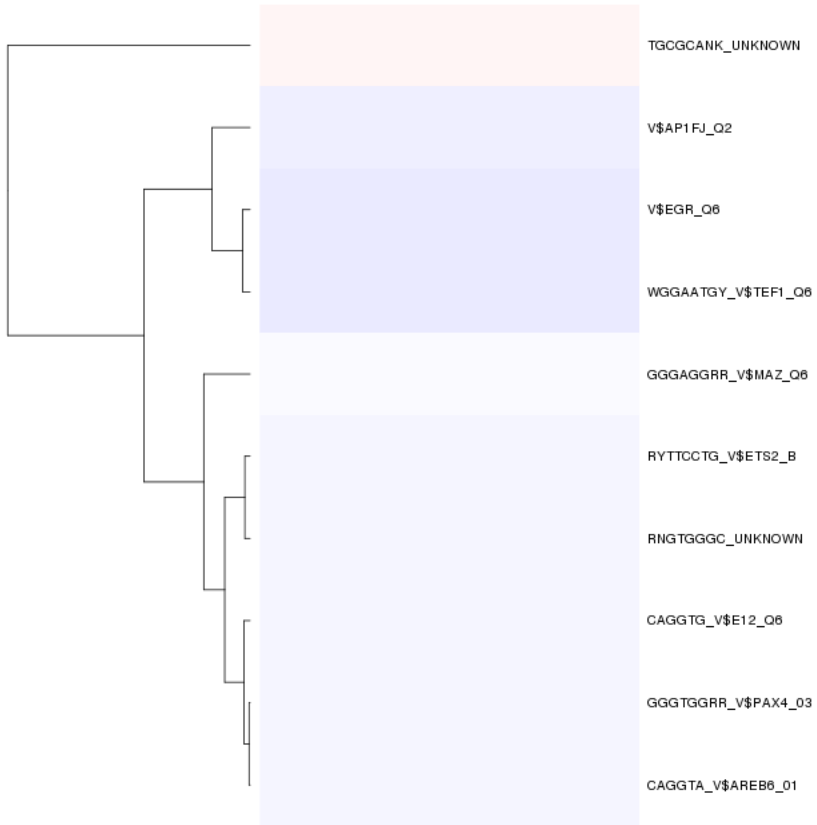
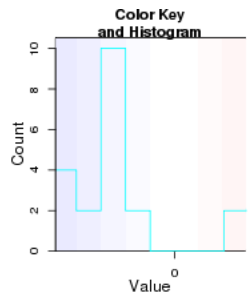




TF enrichment:



TF zscore:



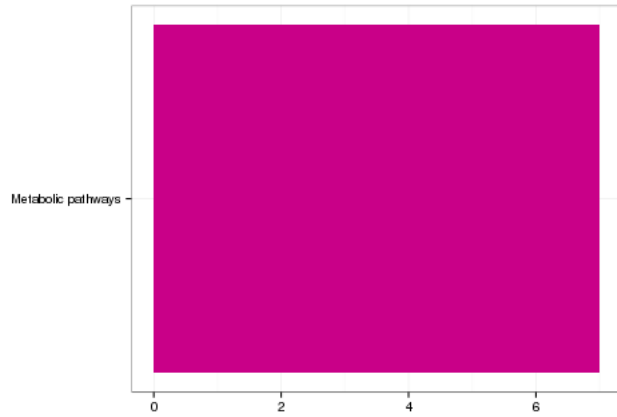
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

```
## Error: 'x' and 'units' must have length > 0
```

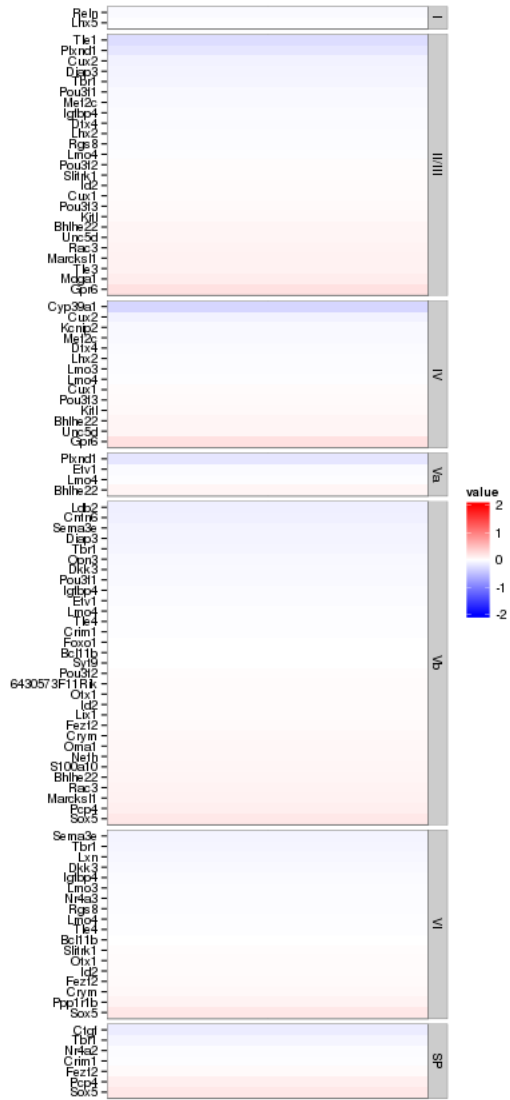
```
## Error: 'x' and 'units' must have length > 0
```

```
## Error: 'x' and 'units' must have length > 0
```

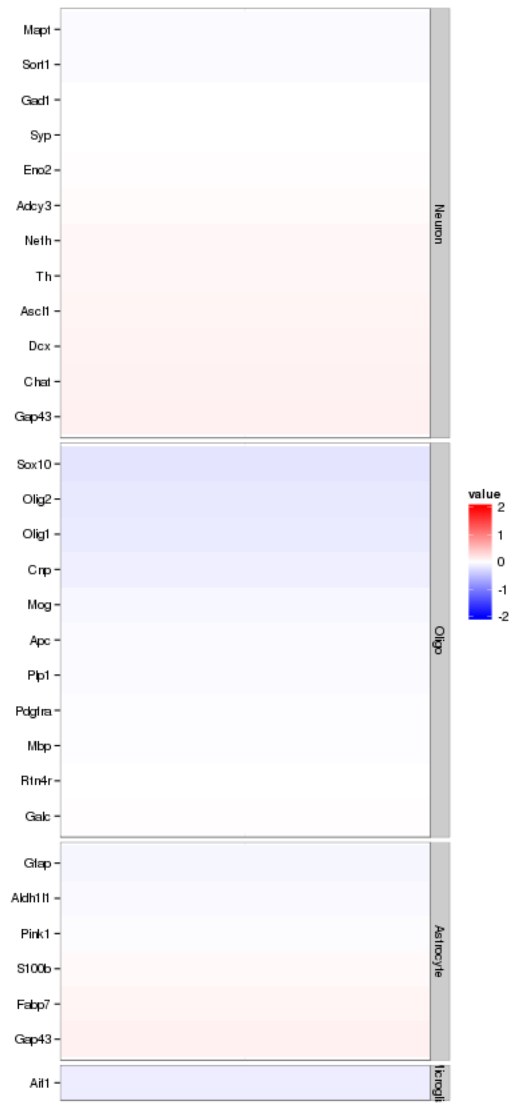


```
## Error: 'x' and 'units' must have length > 0
```

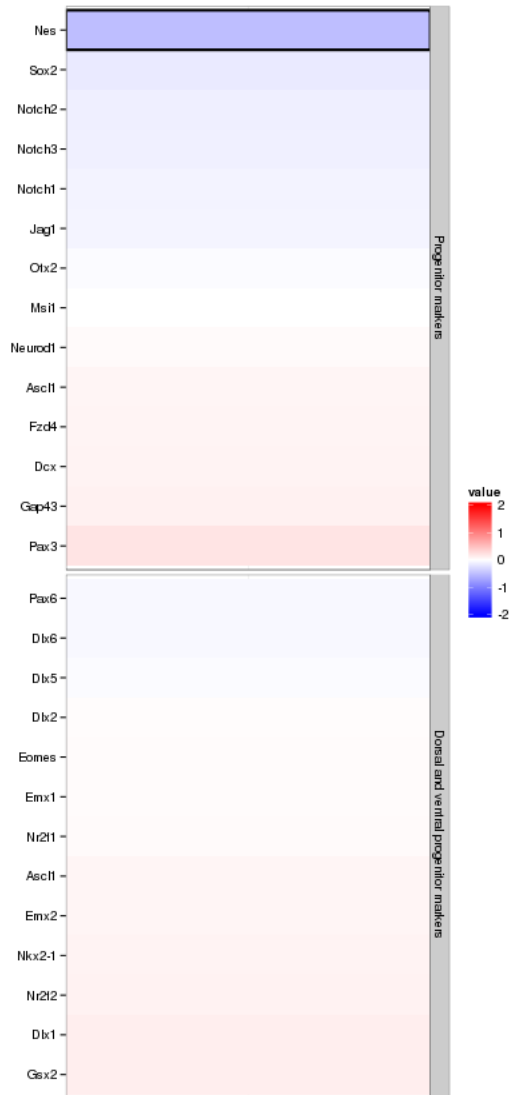
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



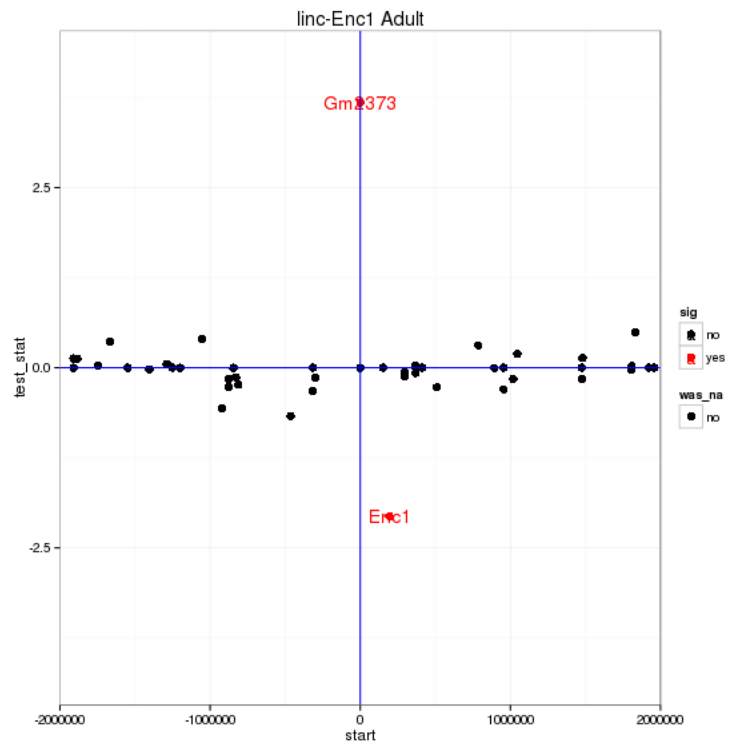
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 1 genes significantly regulated in a region this size is: 0.0947



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR736
16 JR779
17 JR825

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34110000.00	0.77	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34110000.00	0.59	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34110000.00	1.00	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34110000.00	0.82	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34110000.00	1.05	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34110000.00	1.09	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34110000.00	0.82	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34110000.00	1.17	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34110000.00	1.03	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34110000.00	1.32	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34110000.00	1.53	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34110000.00	1.03	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34110000.00	0.99	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34110000.00	1.23	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR736/abundances.cxb	linc_Enc1	0	linc_Enc1_0	30101500.00	34110000.00	0.88	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR779/abundances.cxb	linc_Enc1	1	linc_Enc1_1	38936900.00	34110000.00	1.14	1.00
17 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR825/abundances.cxb	linc_Enc1	2	linc_Enc1_2	33174000.00	34110000.00	0.96	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2     colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4       DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10       Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3    graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1       Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29    latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3       munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10       qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4       splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7    tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,linc-Enc1 -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/linc-Enc1_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap
## 2
## 3
## 4
## 5
```

linc-Enc1 KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for linc-Enc1.

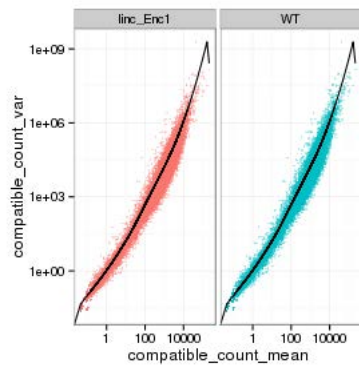
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

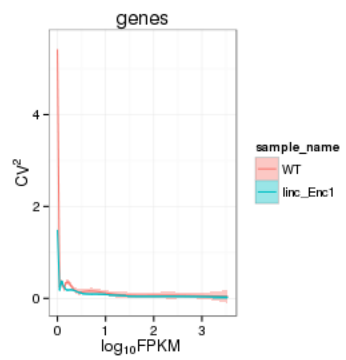
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



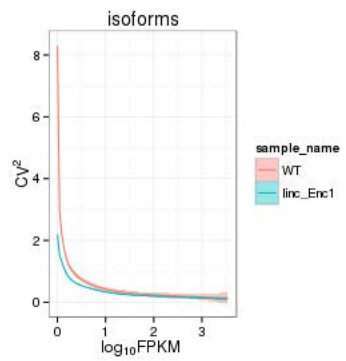
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

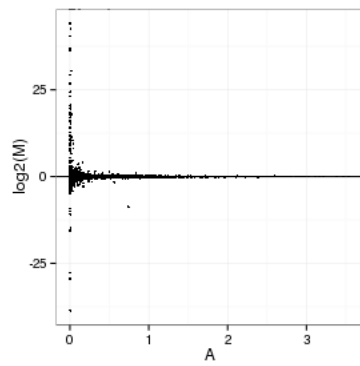
Genes:



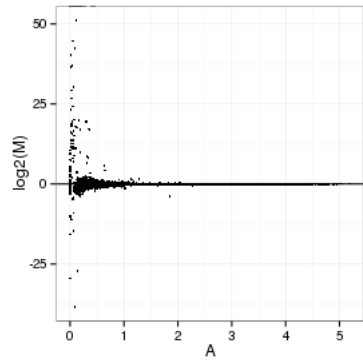
Isoforms:



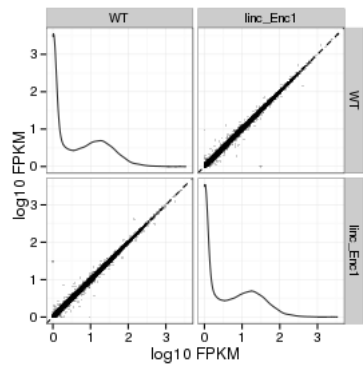
MvA plot



MvA plot counts



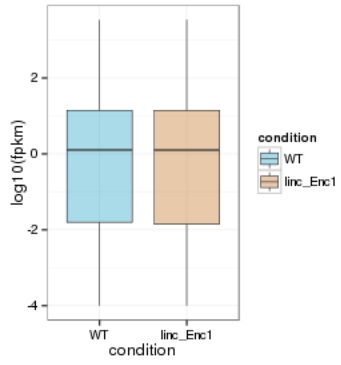
Scatterplot matrix



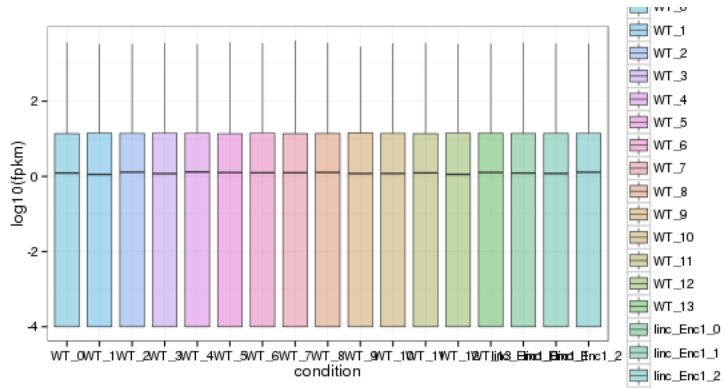
Distributions

Boxplots

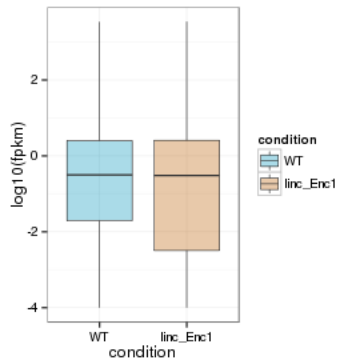
Boxplot (genes)



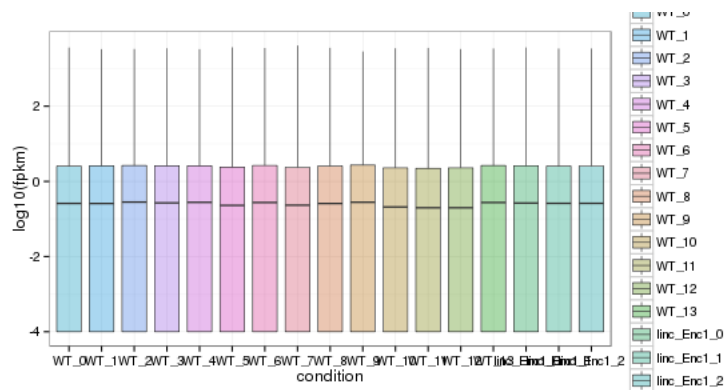
Boxplot (genes, replicates)



Boxplot (isoforms)

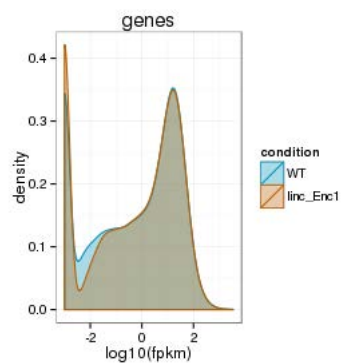


Boxplot (isoforms, replicates)

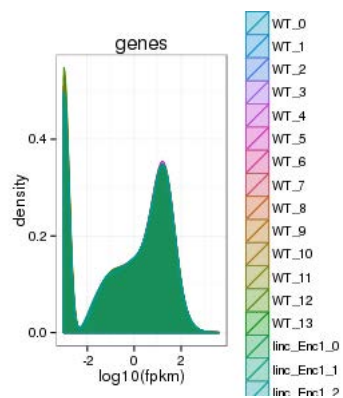


Density

Density (genes)

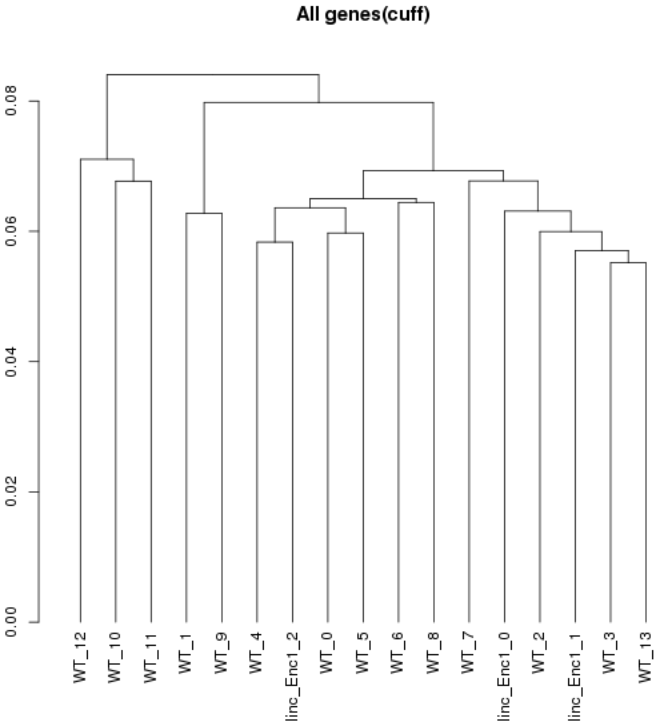


Density (genes, replicates)



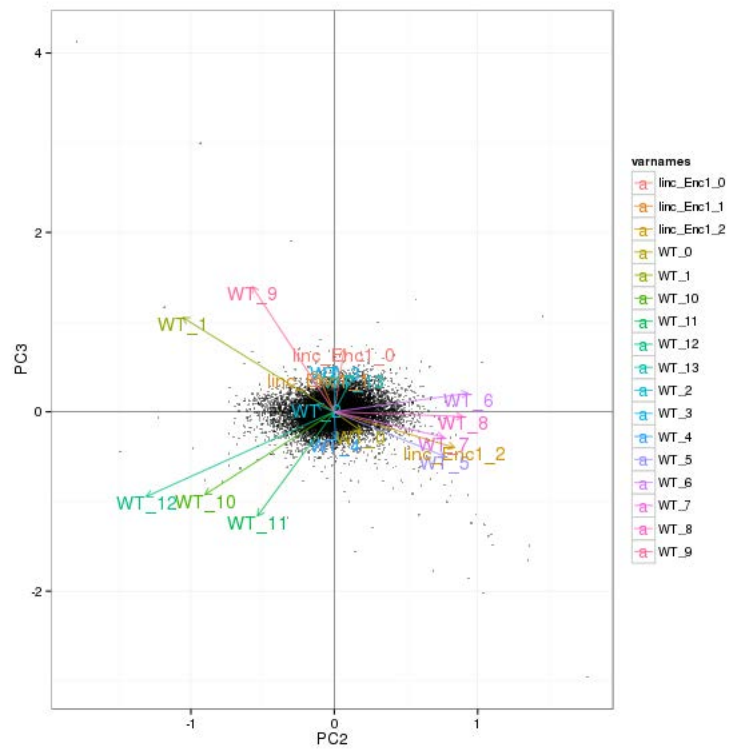
Clustering

Replicate Clusters

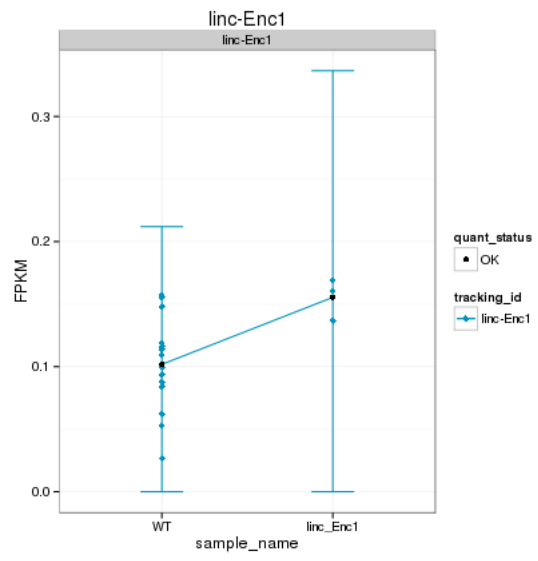


NULL

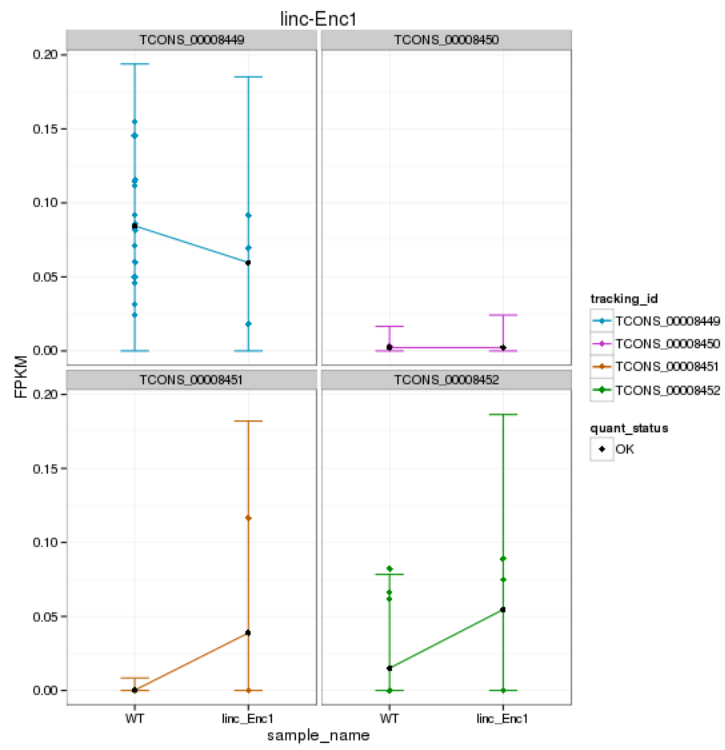
PCA (genes)



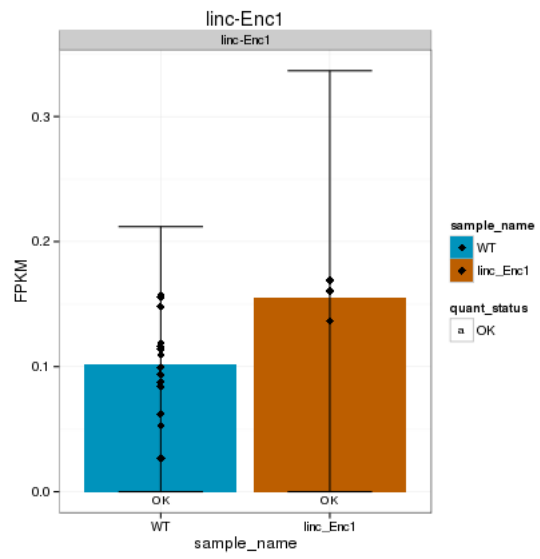
MDS (genes)



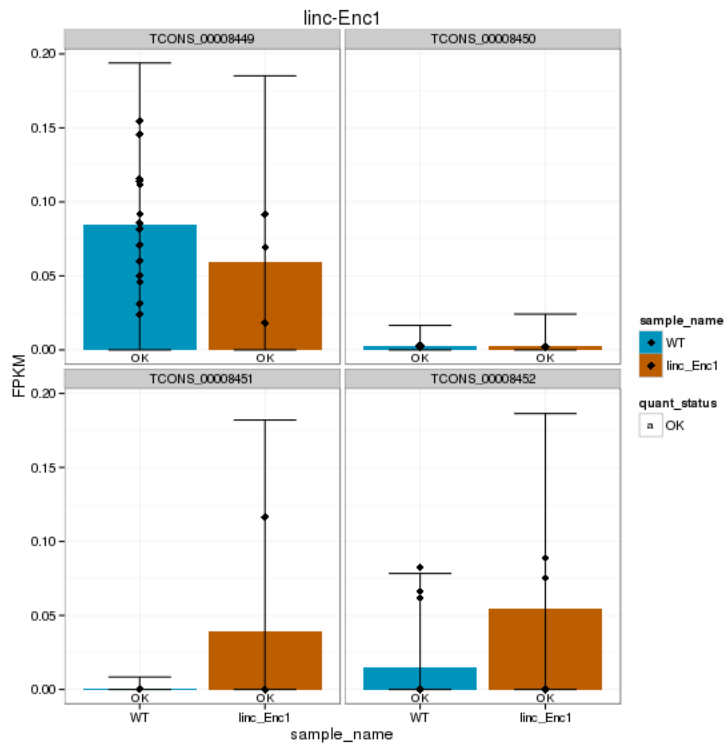
Endogenous expression of linc-Enc1 isoforms:



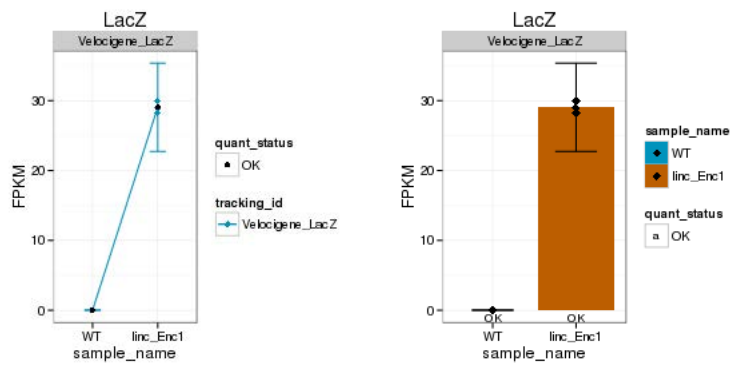
Barplot of gene expression:



Barplot of isoform expression:

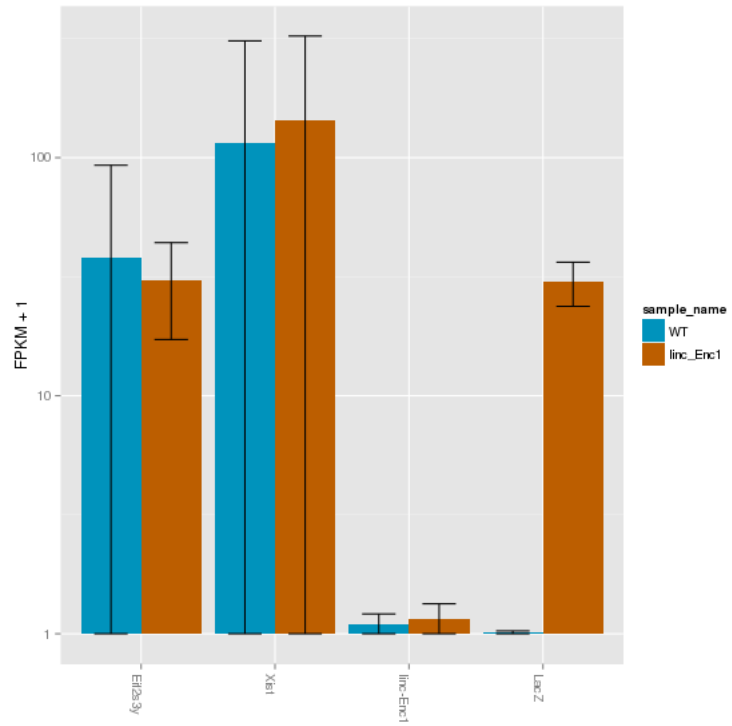


LacZ expression

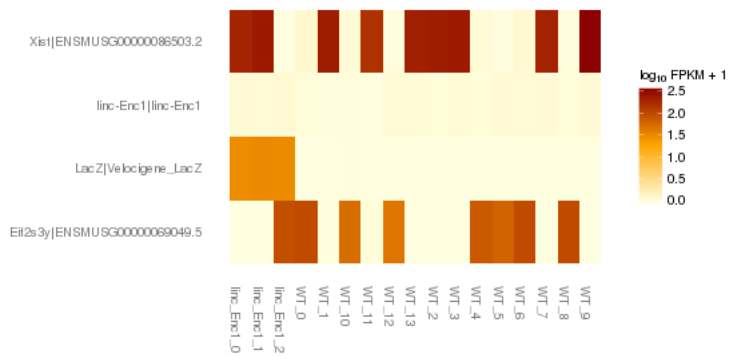


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

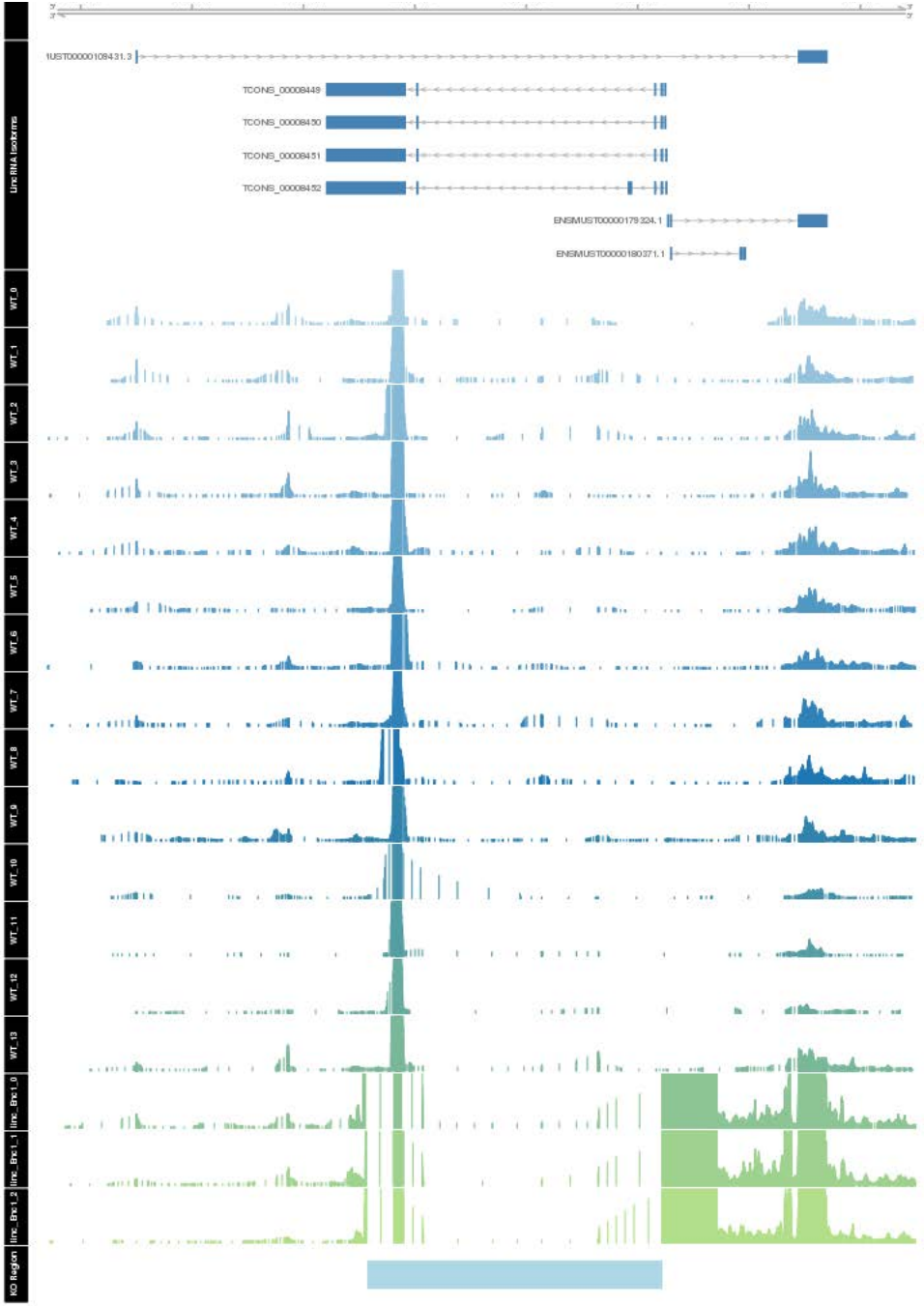


Expression heatmap:



Track visualization





Differential Analysis

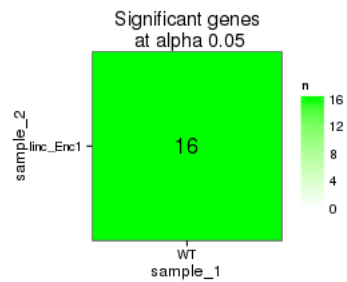
Differential Genes

There are 16 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 Rec8
2 Nnt
3 Fn1
4 Bspry
5 Hddc3
6 Foxj1
7 Enc1
8 Gm9493
9 Fat4
10 Hbb-y
11 Hbb-bh1
12 Hba-x
13 Alox5ap
14 Gstm6
15 Fat1
16 Gm2373
```

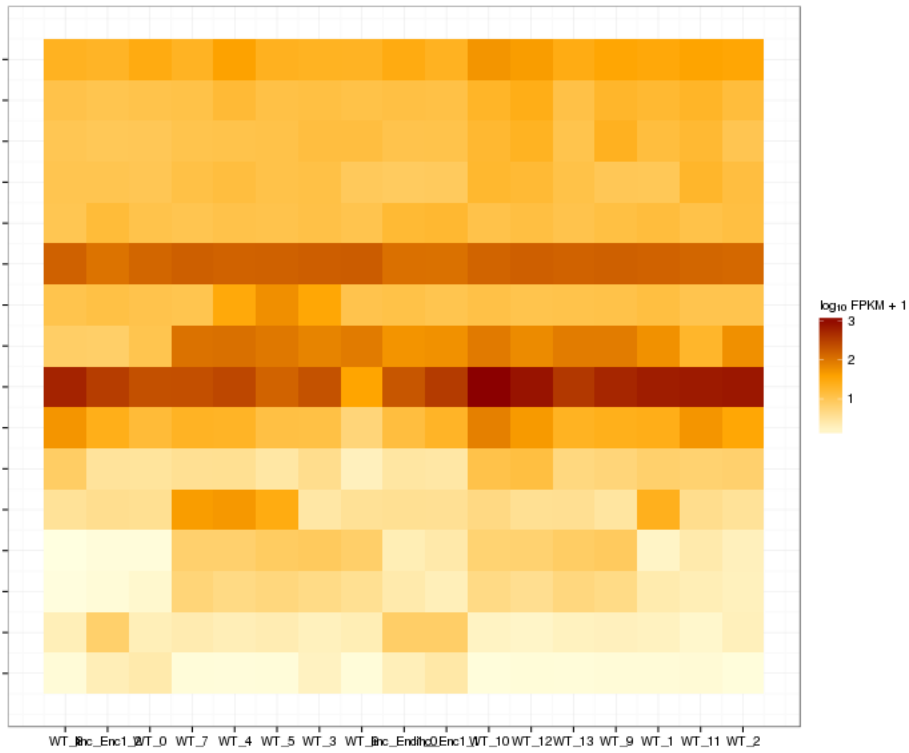
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

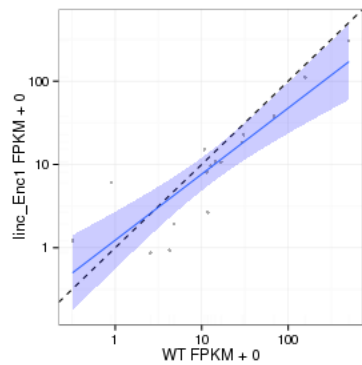


Significant genes with expression >50fpm (any condition):(turned off)

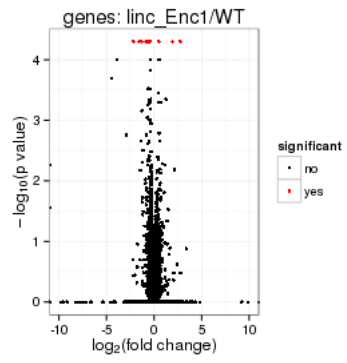
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

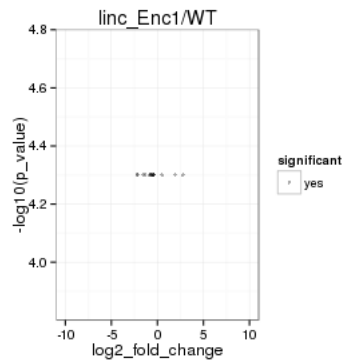
Scatter plot of significant genes only:



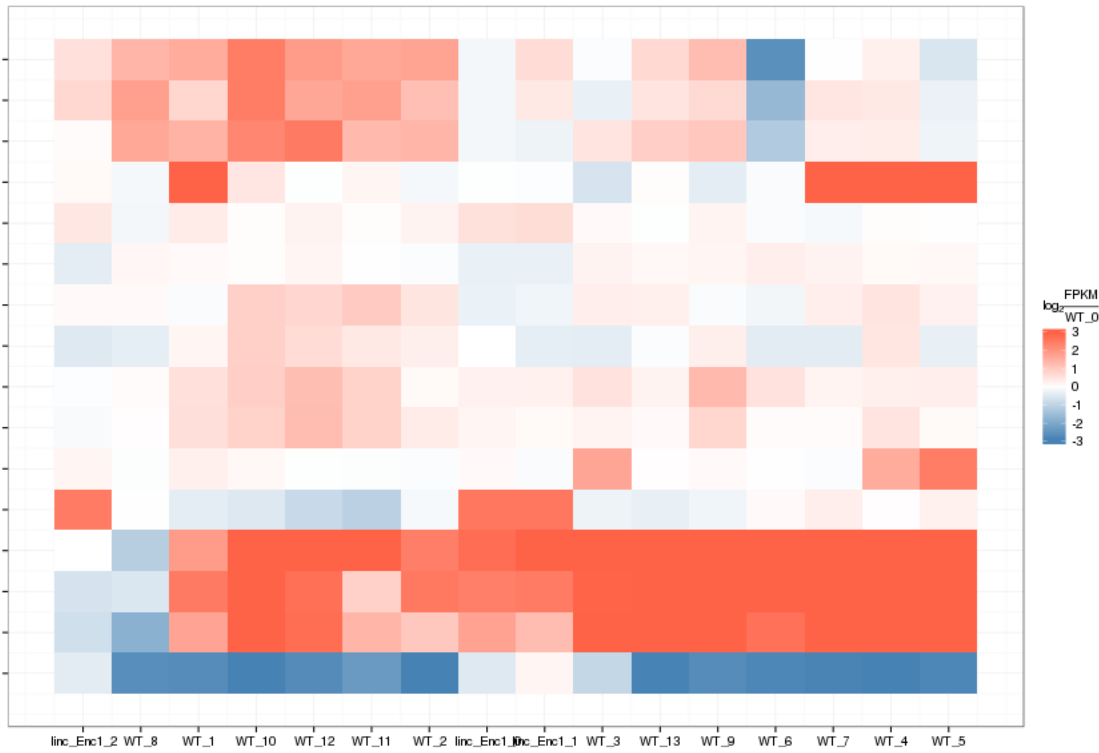
Volcano Plot



Volcano plot with significant genes only:



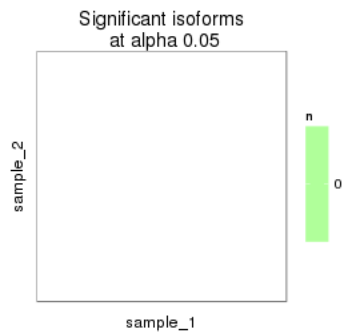
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

[1] "no sig isoforms"

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

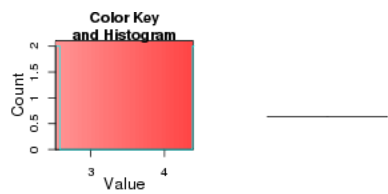
Gene/Pathway Analysis

GSEA

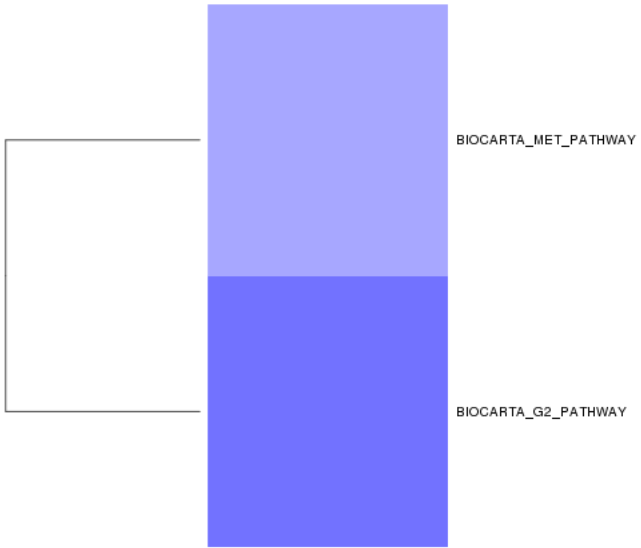
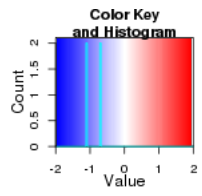
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

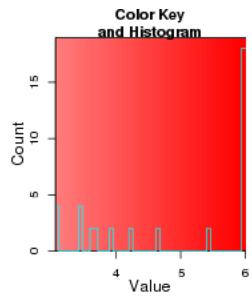
Biocarta enrichment:

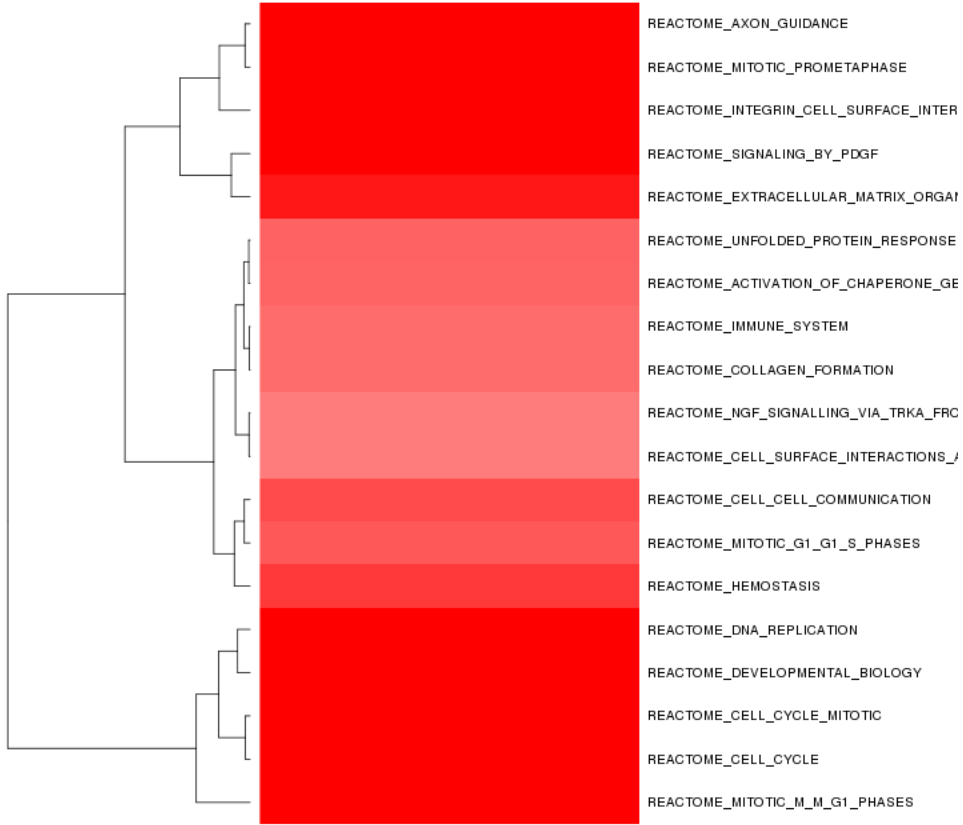


Biocarta zscore:

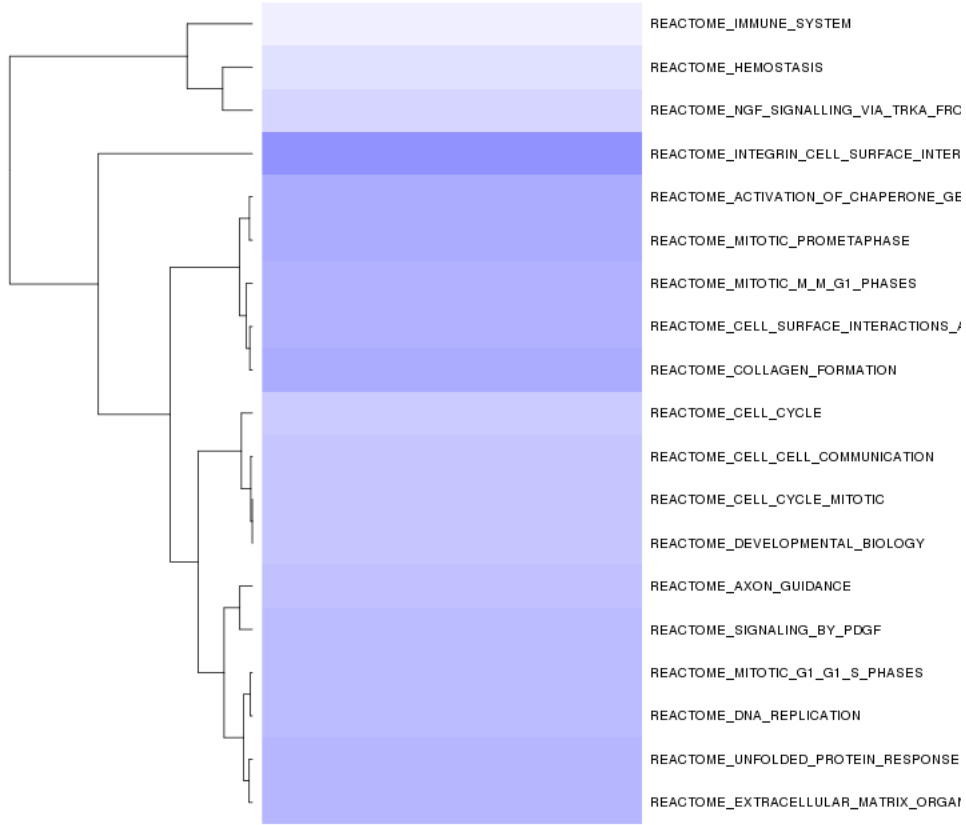
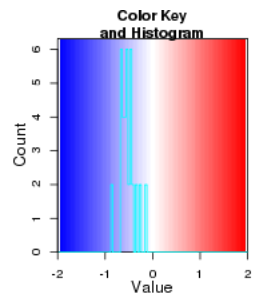


Reactome enrichment:

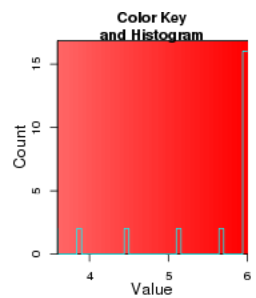


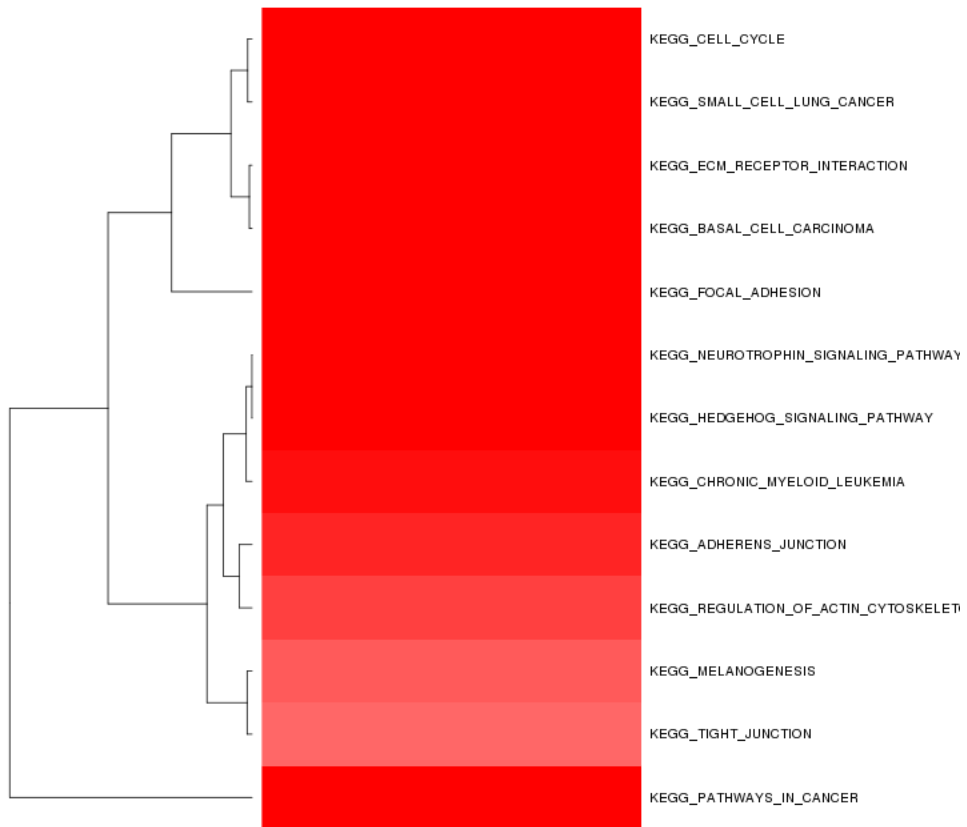


Reactome zscore:

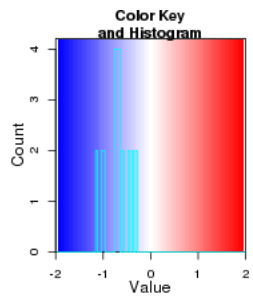


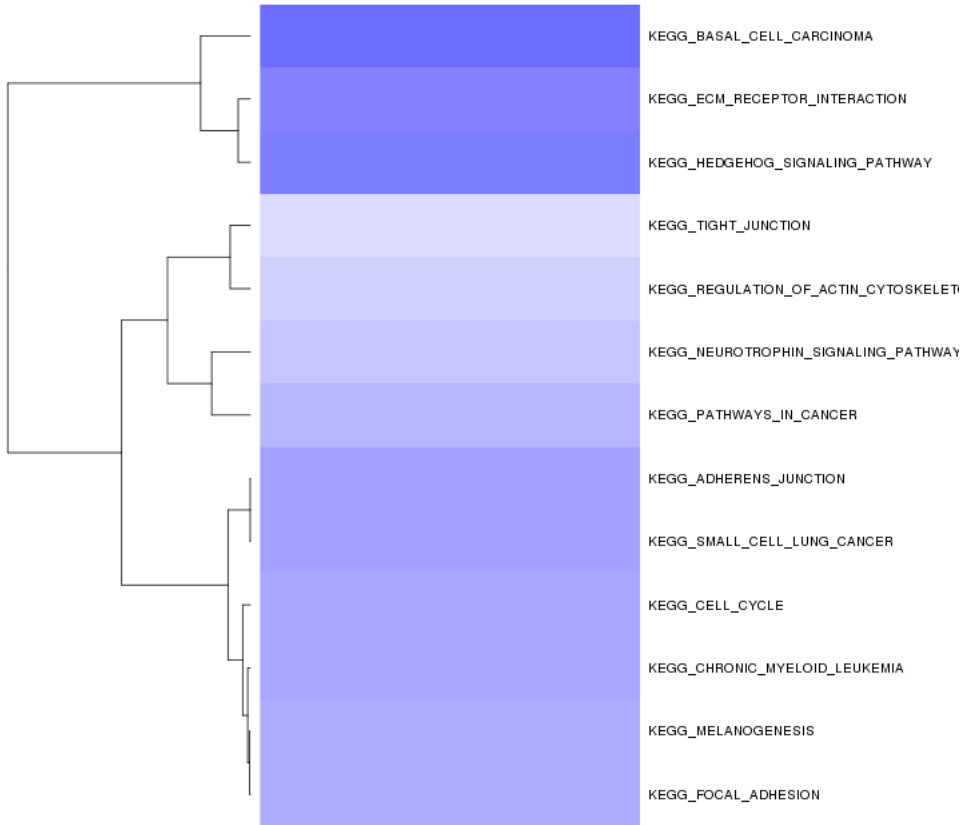
Kegg enrichment:





Kegg zscore:





Interneuron enrichment:

Error: argument is of length zero

Interneuron zscore:

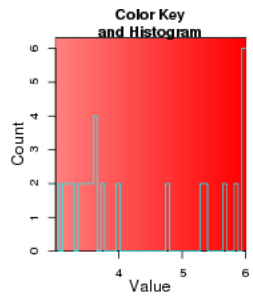
Error: incorrect number of dimensions

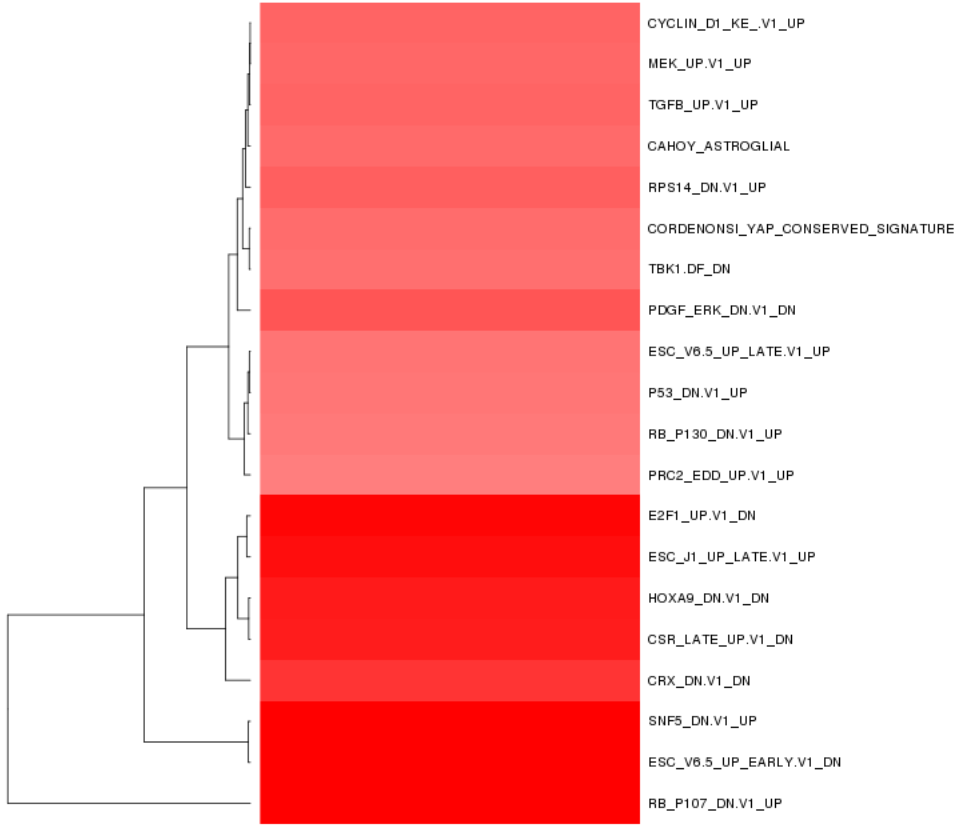

```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

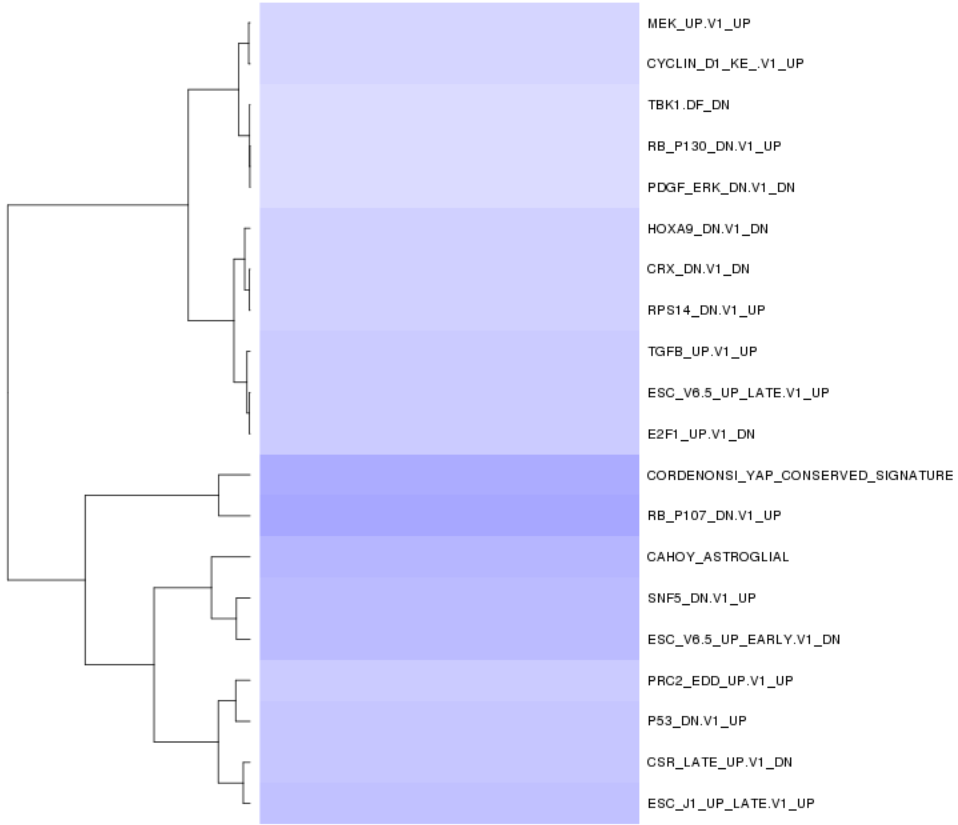
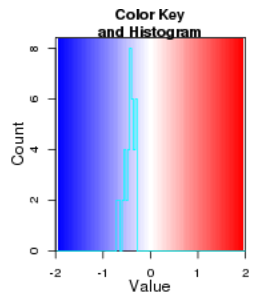
```
## Error: object 'x_ordered' not found
```

Oncogene enrichment:

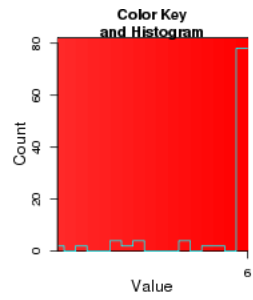




Oncogene zscore:

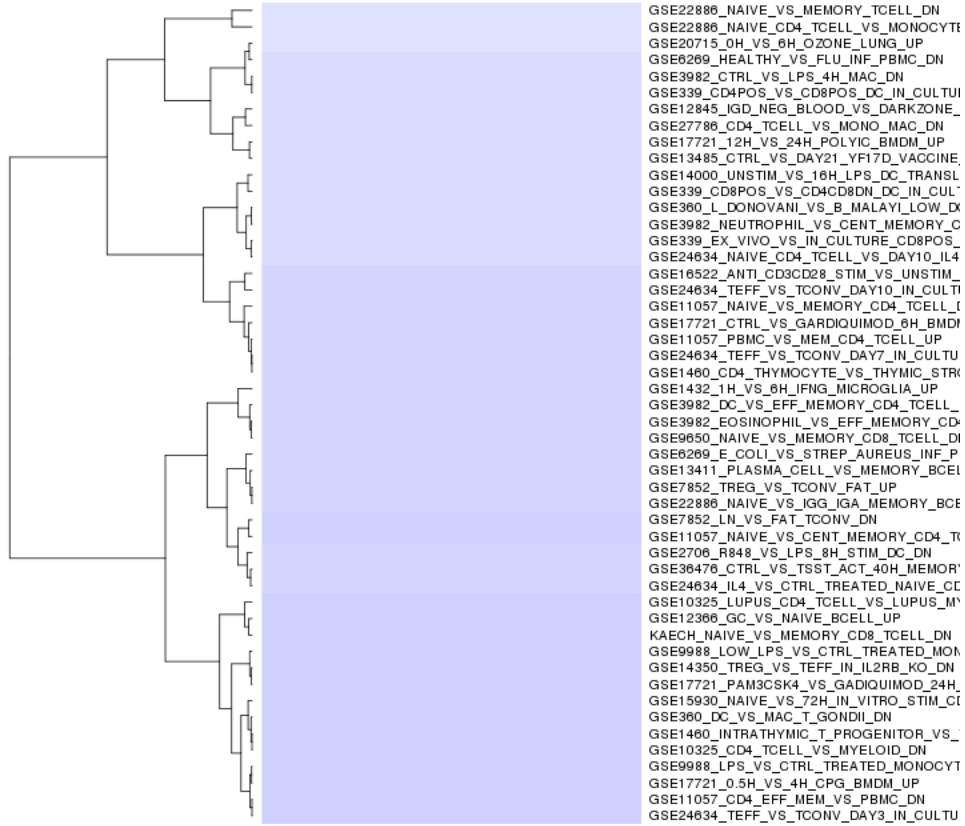
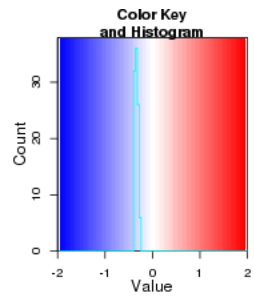


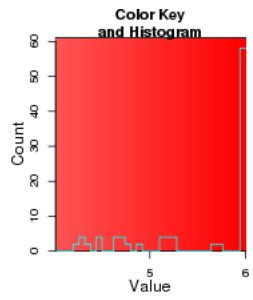
Immuno enrichment:

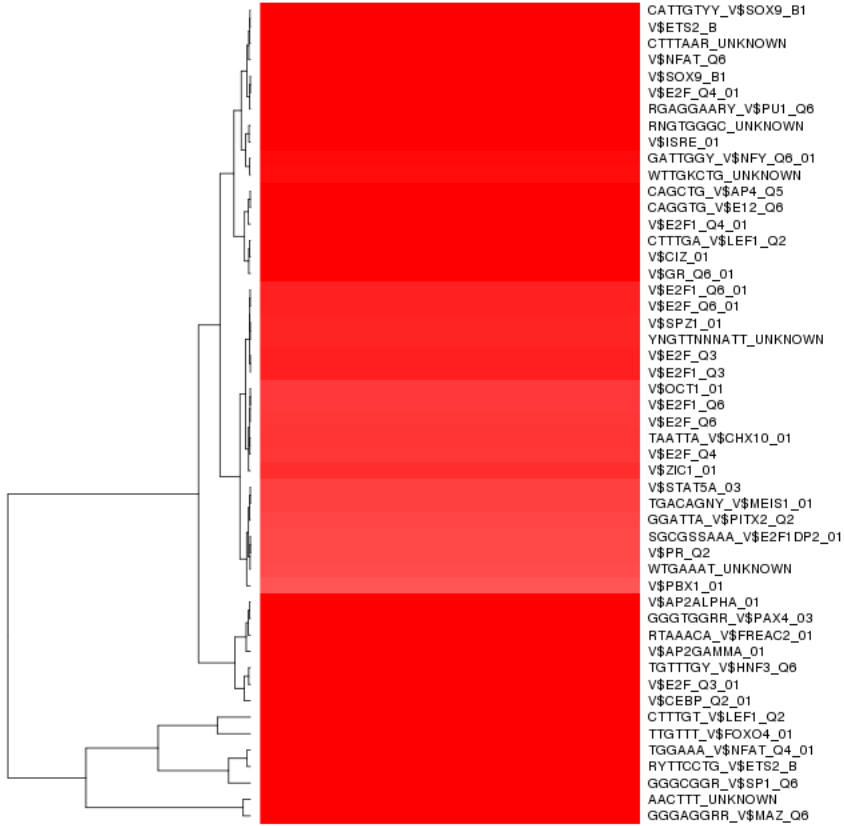


GSE339_EX_VIVO_VS_IN_CULTURE_CD8POS_
 GSE15930_NAIVE_VS_72H_IN_VITRO_STIM_TF
 GSE10325_LUPUS_BCELL_VS_LUPUS_MYELO
 GSE2826_WT_VS_BTK_KO_BCELL_DN
 GSE24634_TEFF_VS_TCONV_DAY10_IN_CULTI
 GSE29614_CTRL_VS_TIV_FLU_VACCINE_PBMK
 GSE13485_DAY7_VS_DAY21_YF17D_VACCINE
 GSE9988_ANTI_TREM1_VS_CTRL_TREATED_M
 GSE14350_TREG_VS_TEFF_IN_IL2RB_KO_DN
 GSE15215_CD2_POS_VS_NEG_PDC_UP
 GSE11057_NAIVE_VS_CENT_MEMORY_CD4_TI
 GSE1432_1H_VS_24H_IFNG_MICROGLIA_UP
 GSE339_CD4POS_VS_CD4CD8DN_DC_DN
 GSE2706_R848_VS_LPS_8H_STIM_DC_DN
 GSE9988_ANTI_TREM1_AND_LPS_VS_CTRL_T
 GSE29618_MONOCYTE_VS_MDC_UP
 GSE9988_LOW_LPS_VS_CTRL_TREATED_MON
 GSE22886_NAIVE_CD8_TCELL_VS_MONOCYTE
 GSE20715_0H_VS_24H_OZONE_TLR4_KO_LUM
 GSE29618_MONOCYTE_VS_MDC_DAY7_FLU_V
 GSE12366_GC_VS_MEMORY_BCELL_UP
 GSE22886_NAIVE_CD8_TCELL_VS_NKCELL_D
 GSE29614_DAY3_VS_DAY7_TIV_FLU_VACCINE
 GSE17721_0.5H_VS_4H_CPG_BMDM_UP
 GSE22886_NAIVE_TCELL_VS_NKCELL_DN
 GSE22886_NAIVE_BCELL_VS_MONOCYTE_DN
 GSE9650_EFFECTOR_VS_MEMORY_CD8_TCEL
 GSE9650_NAIVE_VS_EFF_CD8_TCELL_DN
 GSE3982_DC_VS_MAC_DN
 GSE8289_HEALTHY_VS_STREP_AUREUS_INF
 GSE14000_4H_VS_16H_LPS_DC_TRANSLATEC
 GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR
 KAECH_NAIVE_VS_DAY8_EFF_CD8_TCELL_DN
 GSE20715_0H_VS_48H_OZONE_LUNG_DN
 KAECH_DAY8_EFF_VS_MEMORY_CD8_TCELL_
 GSE24634_TREG_VS_TCONV_POST_DAY10_IL
 GSE29618_MONOCYTE_VS_PDC_UP
 GOLDRATH_NAIVE_VS_EFF_CD8_TCELL_DN
 GSE9988_ANTI_TREM1_AND_LPS_VS_VEHICLI
 GSE9988_LOW_LPS_VS_VEHICLE_TREATED_I
 KAECH_DAY8_EFF_VS_DAY15_EFF_CD8_TCEL
 GSE9988_ANTI_TREM1_VS_VEHICLE_TREATEI
 GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR
 GSE9988_LPS_VS_VEHICLE_TREATED_MONO
 GSE29614_CTRL_VS_DAY7_TIV_FLU_VACCINE
 GSE8289_HEALTHY_VS_STREP_PNEUMO_INF
 GOLDRATH_EFF_VS_MEMORY_CD8_TCELL_UF
 GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL
 GSE15750_DAY6_VS_DAY10_TRAFKO_EFF_C
 GSE30962_PRIMARY_VS_SECONDARY_ACUTE

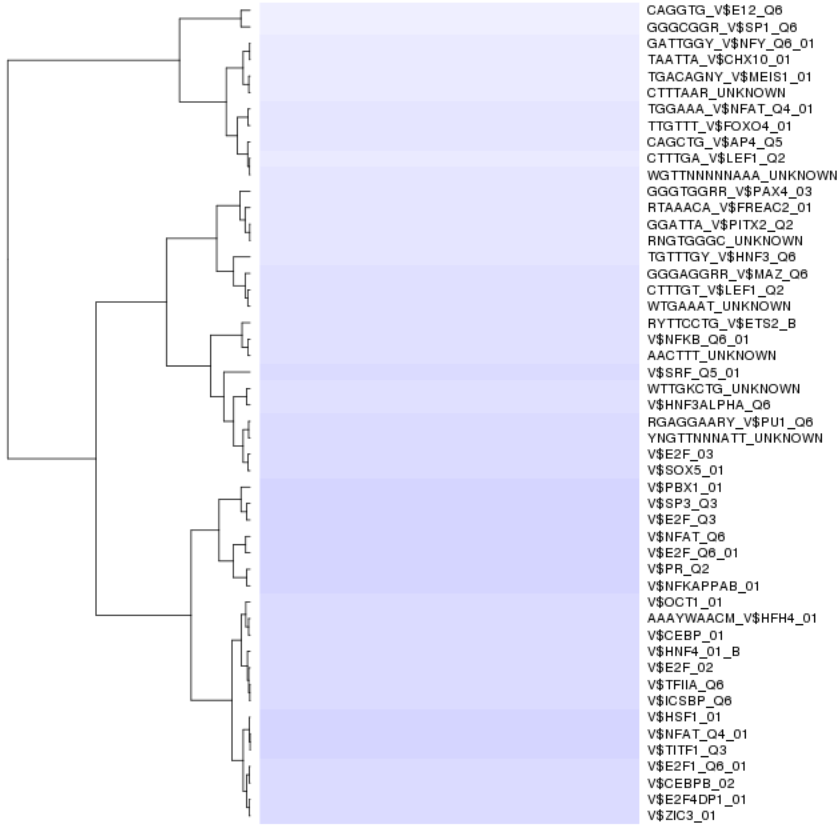
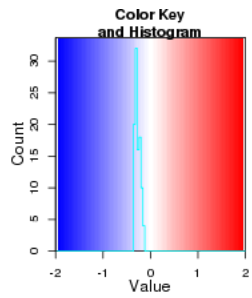
Immuno zscore:







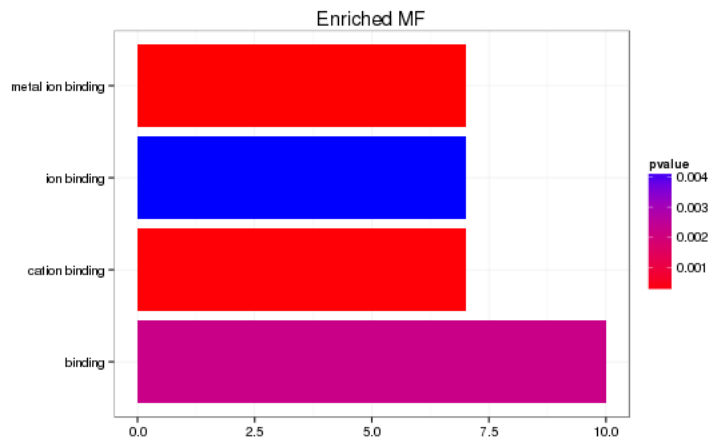
TF zscore:



GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

```
## Error: 'x' and 'units' must have length > 0
```

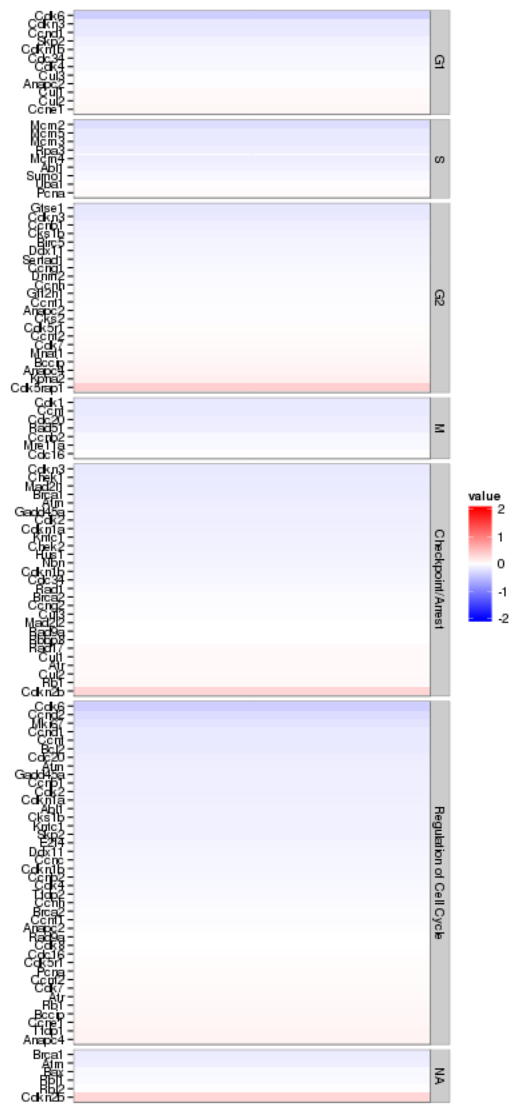


```
## Error: 'x' and 'units' must have length > 0
```

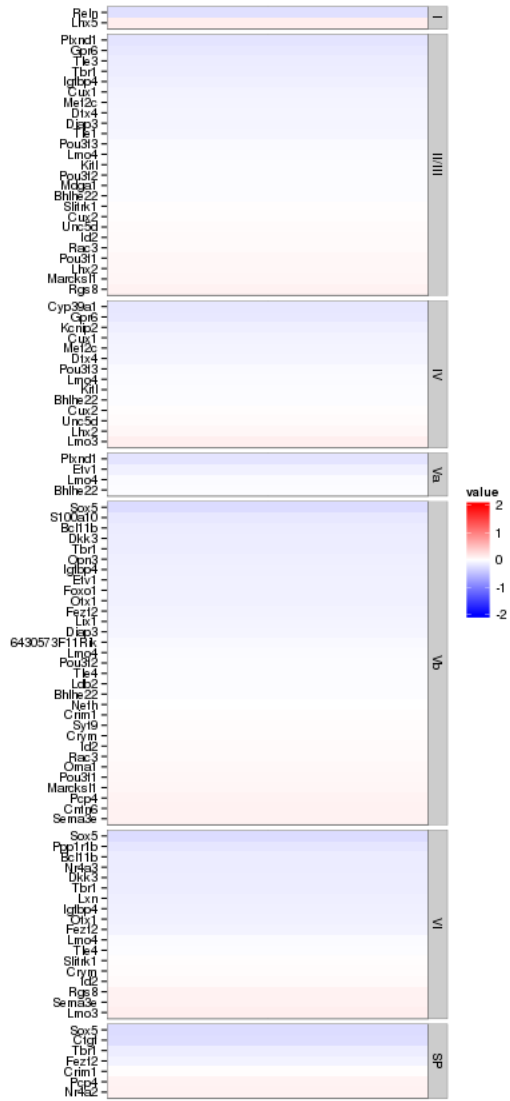
```
## Error: need finite 'xlim' values
```

```
## Error: need finite 'xlim' values
```

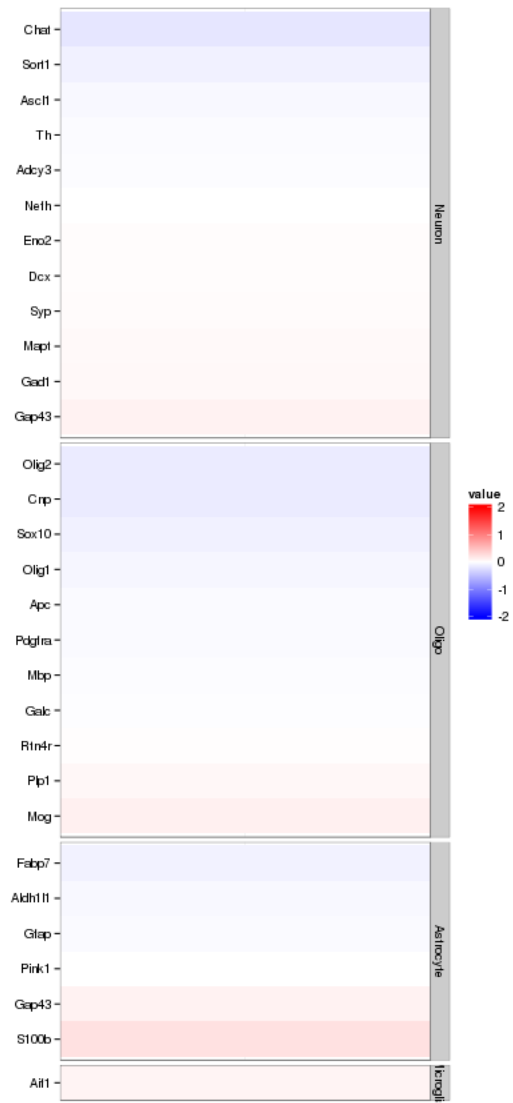
Enrichment or depletion for stage-specific cell cycle markers



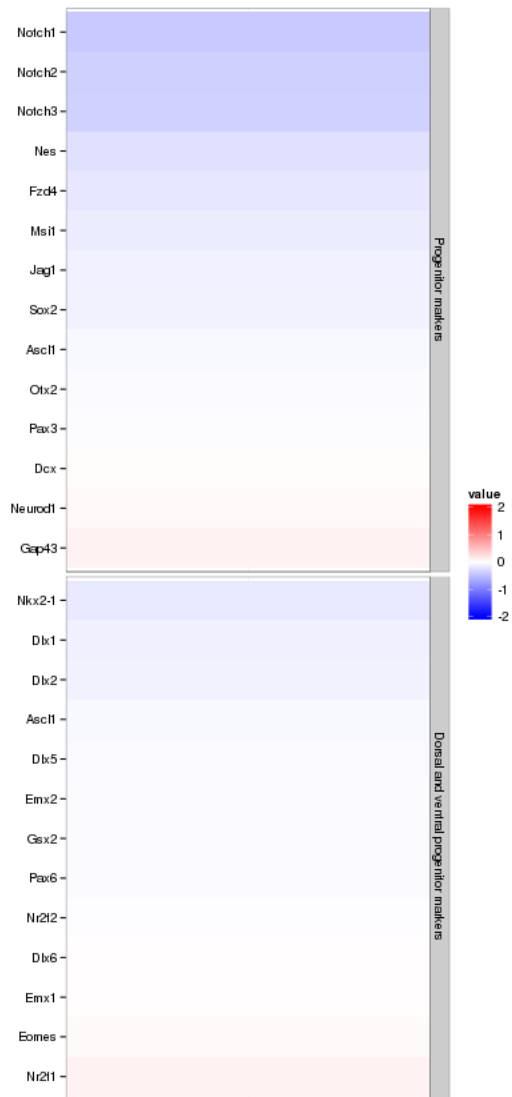
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



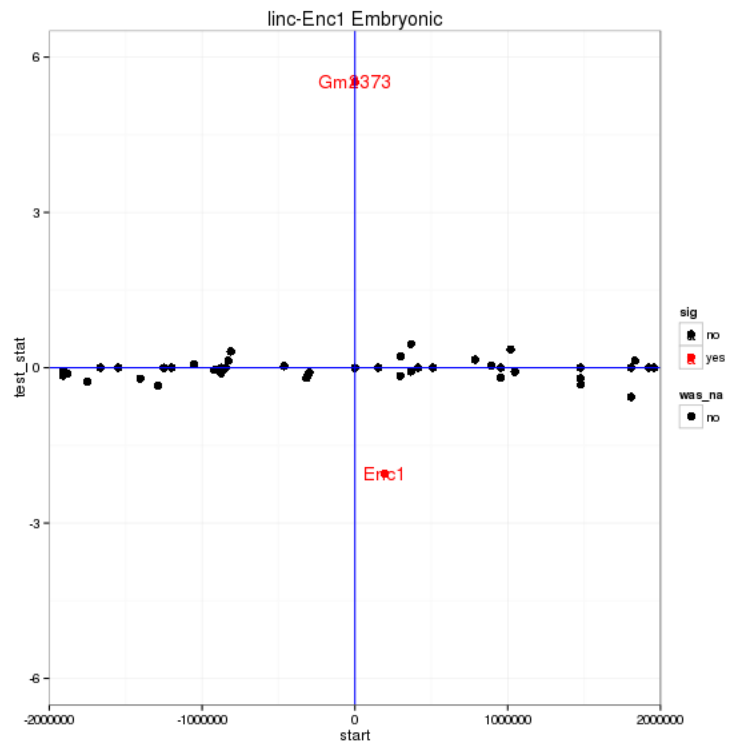
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 1 genes significantly regulated in a region this size is: 0.0409



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR752
16 JR770
17 JR839

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	33003900.00	1.23	1.00
2 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	33003900.00	1.07	1.00
3 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	33003900.00	1.20	1.00
4 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	33003900.00	1.26	1.00
5 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	33003900.00	1.17	1.00
6 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	33003900.00	0.87	1.00
7 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	33003900.00	0.95	1.00
8 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	33003900.00	0.90	1.00
9 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	33003900.00	0.84	1.00
10 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	33003900.00	0.94	1.00
11 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	33003900.00	0.79	1.00
12 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	33003900.00	0.78	1.00
13 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	33003900.00	0.67	1.00
14 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	33003900.00	1.18	1.00
15 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR752/abundances.cxb	linc_Enc1	0	linc_Enc1_0	37182200.00	33003900.00	1.13	1.00
16 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR770/abundances.cxb	linc_Enc1	1	linc_Enc1_1	42845600.00	33003900.00	1.30	1.00
17 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR839/abundances.cxb	linc_Enc1	2	linc_Enc1_2	33810100.00	33003900.00	1.03	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1      KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1      reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1        zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,linc-Enc1 -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/linc-Enc1_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/Brai
## 2
## 3
## 4
## 5
```


Manr KO vs WT (Adult)

This file shows the wt-v-ko comparison for Manr.

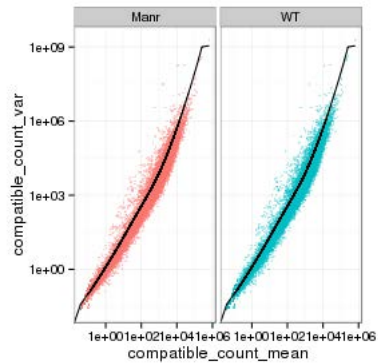
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

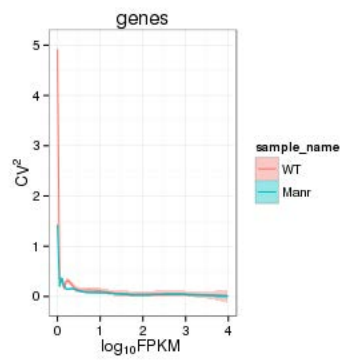
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



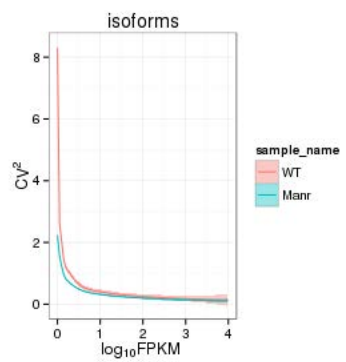
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

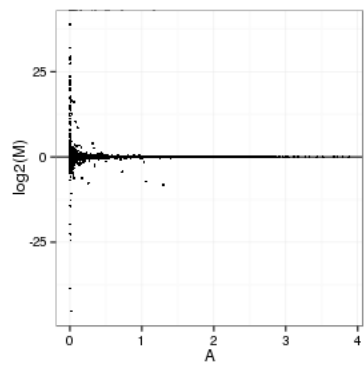
Genes:



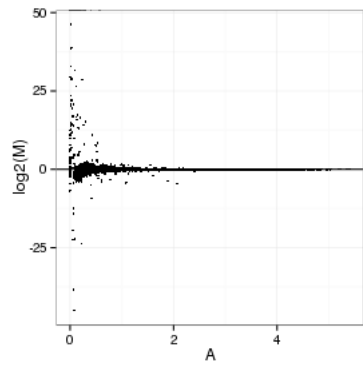
Isoforms:



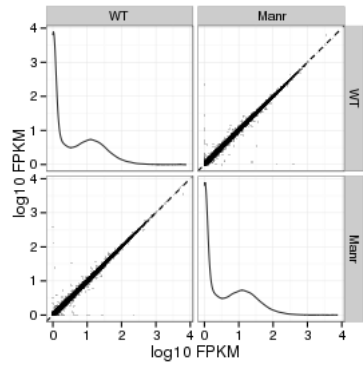
MvA plot



MvA plot counts



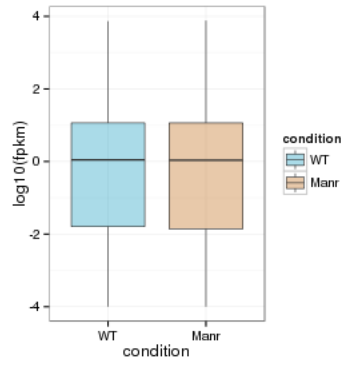
Scatterplot matrix



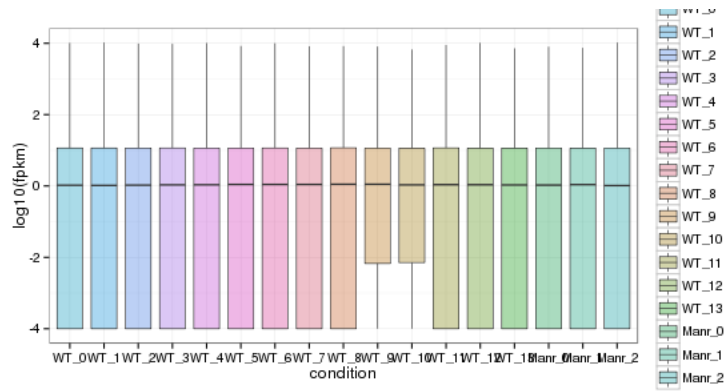
Distributions

Boxplots

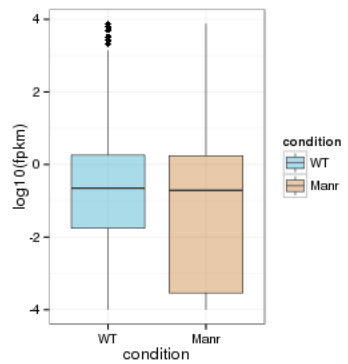
Boxplot (genes)



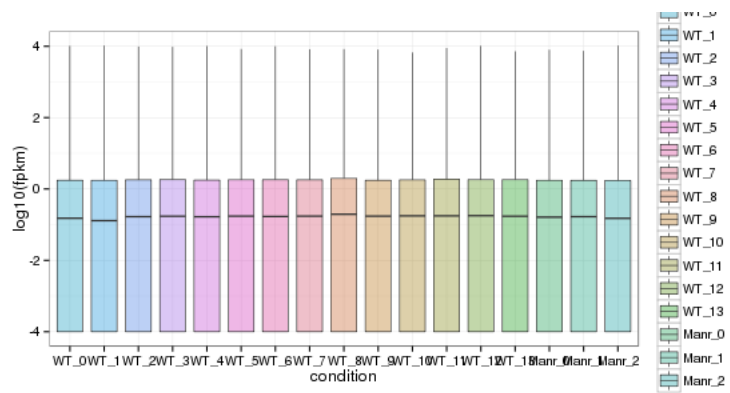
Boxplot (genes, replicates)



Boxplot (isoforms)

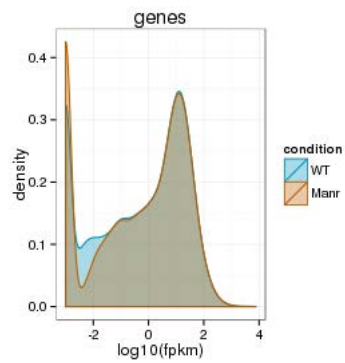


Boxplot (isoforms, replicates)

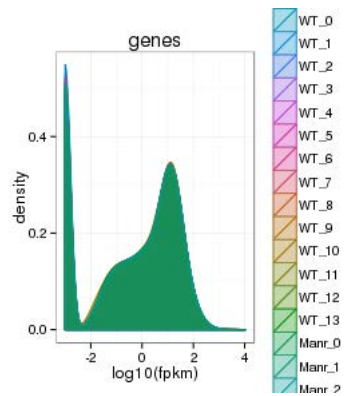


Density

Density (genes)

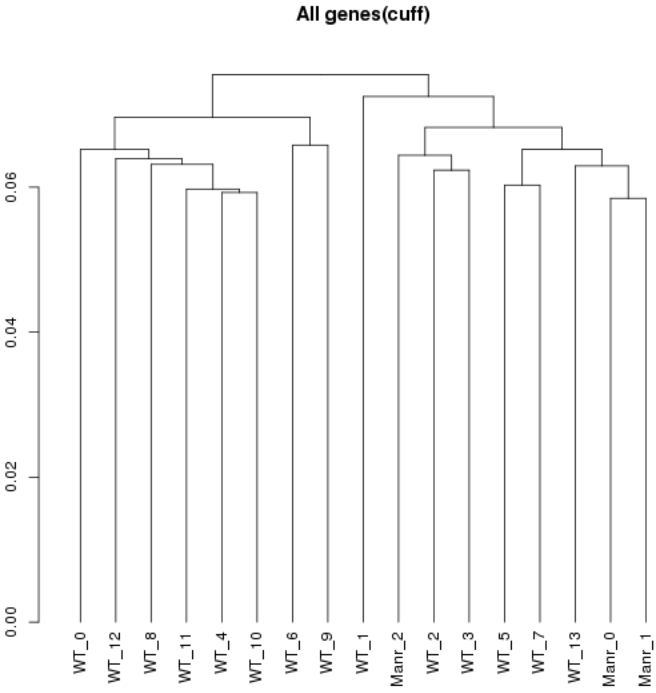


Density (genes, replicates)



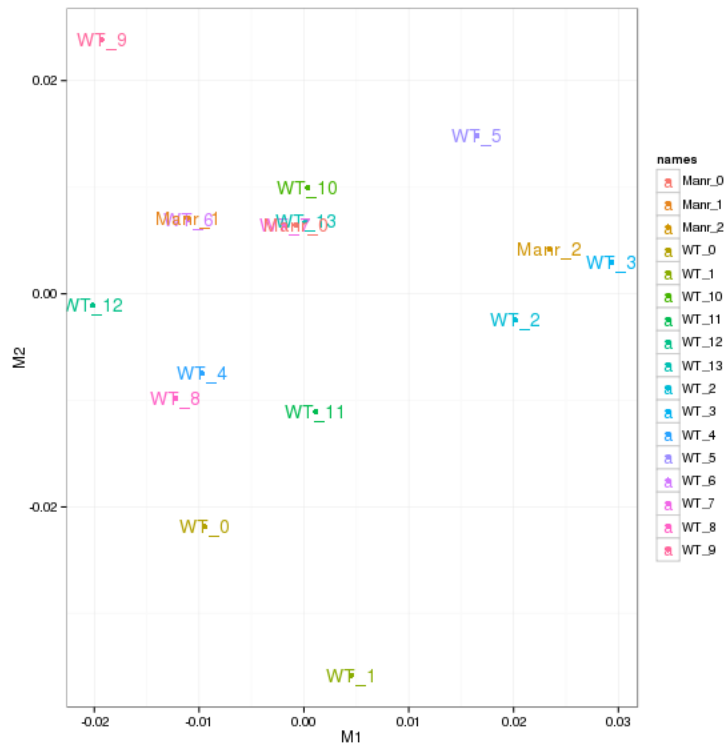
Clustering

Replicate Clusters

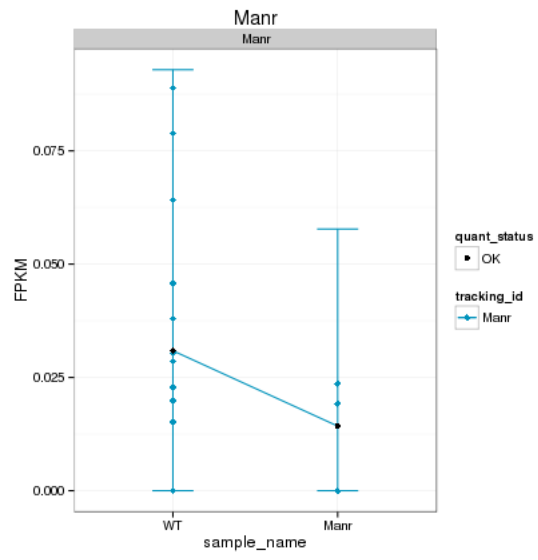


NULL

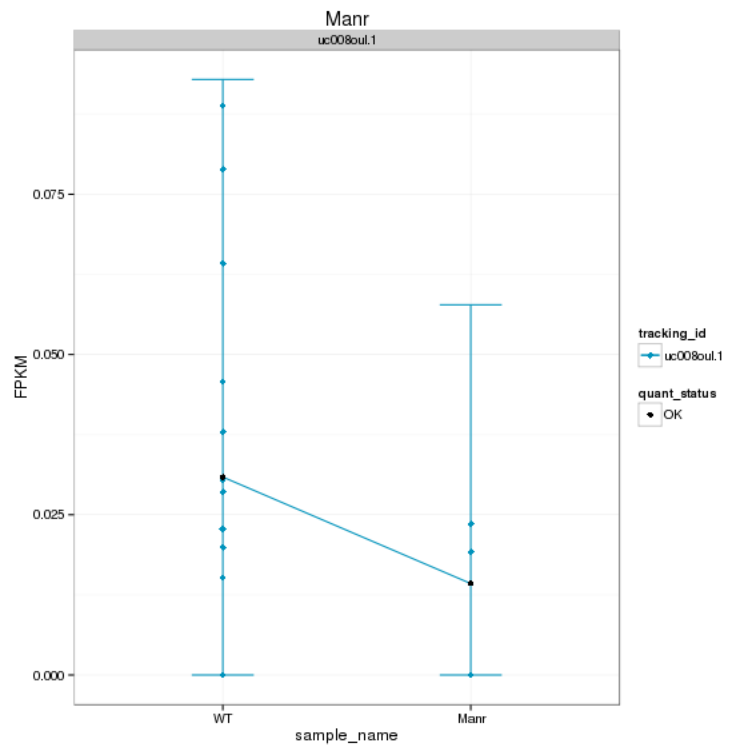
PCA (genes)



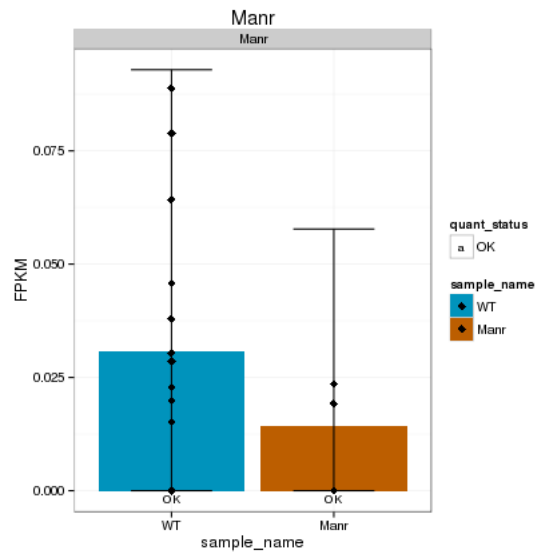
KO assessment
Endogenous lncRNA expression



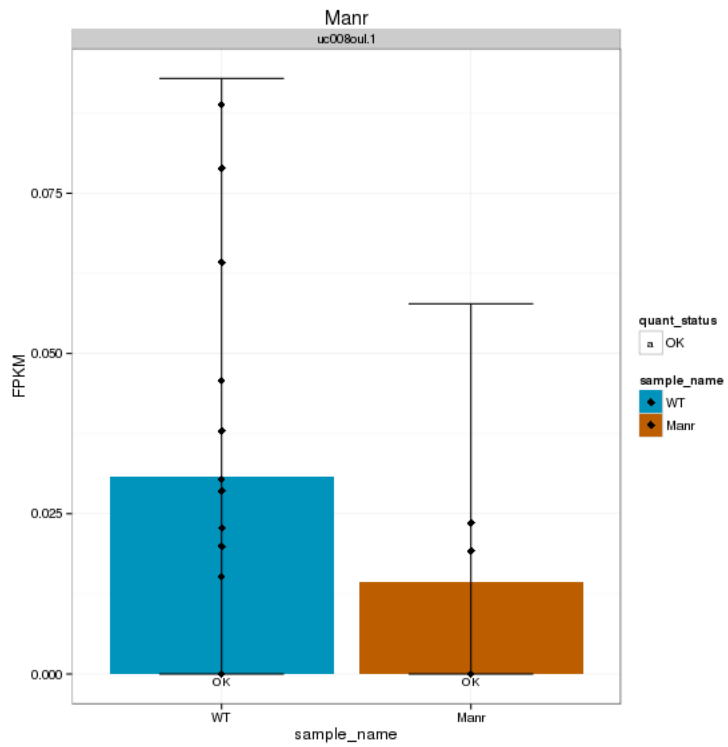
Endogenous expression of Manr isoforms:



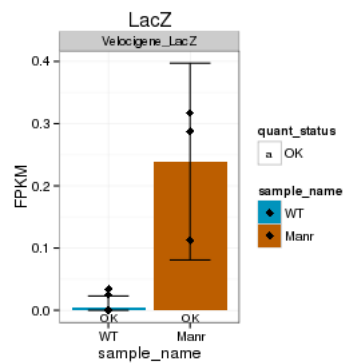
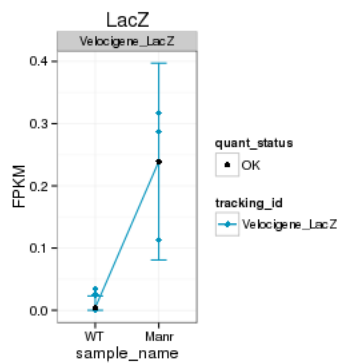
Barplot of gene expression:



Barplot of isoform expression:

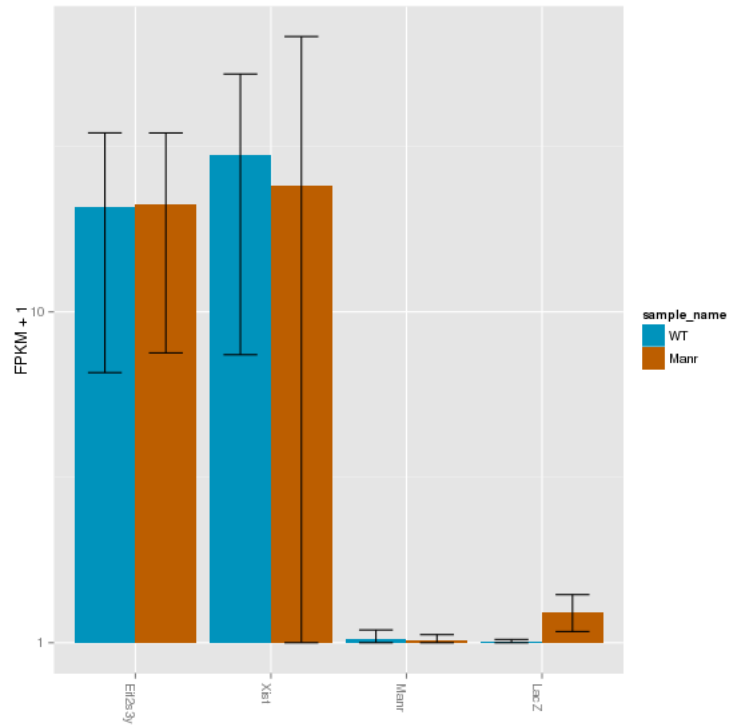


LacZ expression

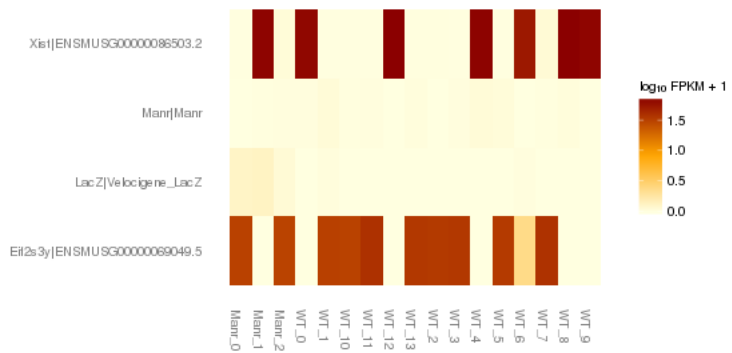


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 49 significantly differentially expressed genes. They are:

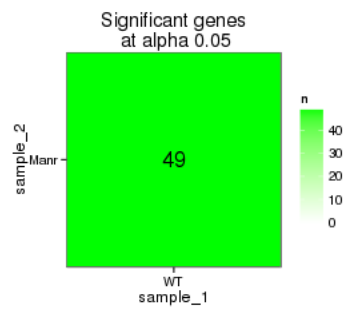
	geneAnnot\$gene_short_name
1	Slc5a5
2	Col6a1
3	Arrdc2
4	Nes
5	Lpl
6	Pdk4
7	Pttg1
8	Adi1
9	Pomc
10	Serpina3n
11	Fos
12	Arc
13	Nr4a1
14	Podxl
15	Lcn2
16	Fabp5
17	Anxa5
18	Tshb
19	Tmem144
20	Rps3a1
21	Nr4a3
22	Tinagl1
23	Hddc3
24	Dis3l
25	Arhgap27
26	Pla2g3
27	Plvap
28	Lars2
29	Tmem59l
30	Egr2
31	Egr1
32	Sgsm1
33	Tuba1c
34	Snhg11
35	Acp1
36	Epcam
37	Junb

- 38 Tpm3-rs7
- 39 Trank1
- 40 Gm4
- 41 Rps15
- 42 Bpifb3
- 43 Gm9008

- 44 Gm7292
- 45 Srp54a
- 46 Wdfy1
- 47 Scnm1
- 48 AA465934
- 49 Gm26924

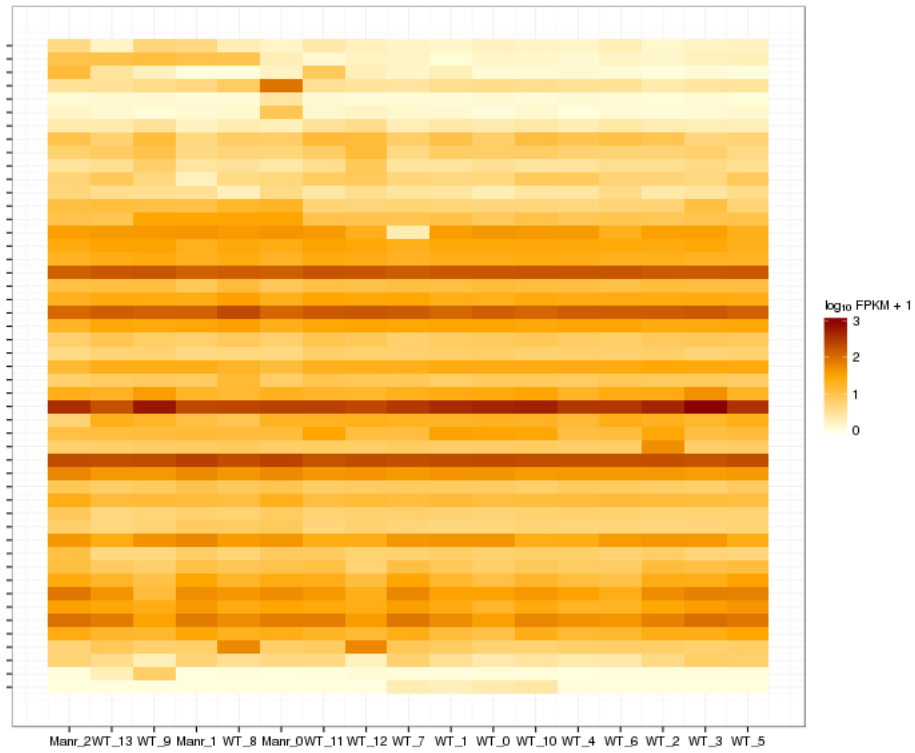
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

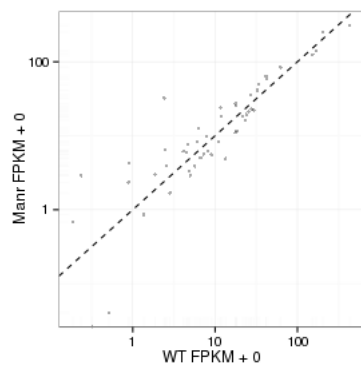


Significant genes with expression >50fpkm (any condition):(turned off)

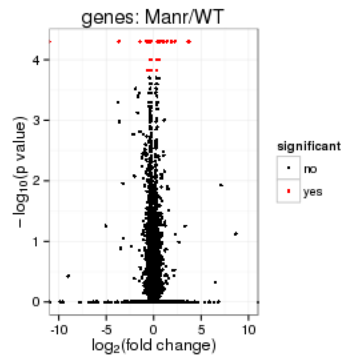
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

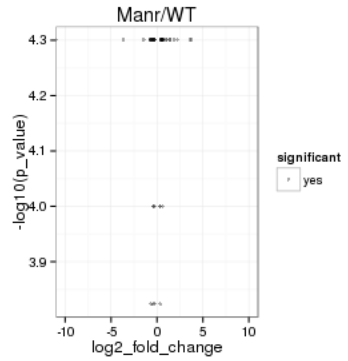
Scatter plot of significant genes only:



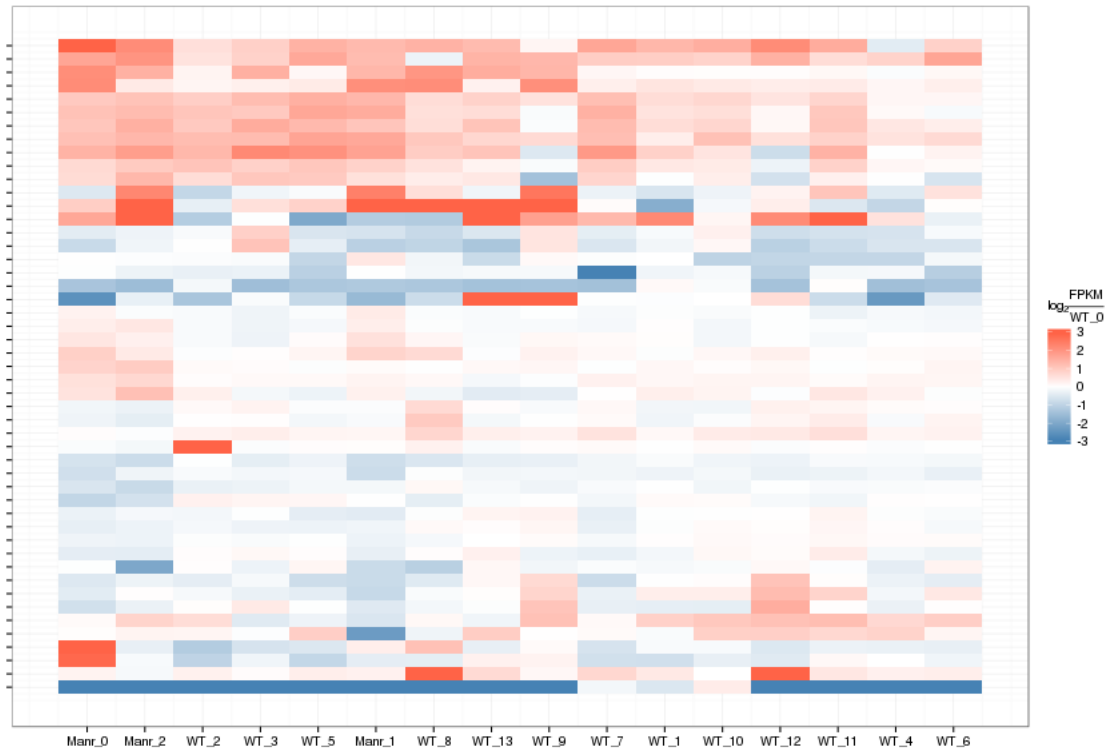
Volcano Plot



Volcano plot with significant genes only:



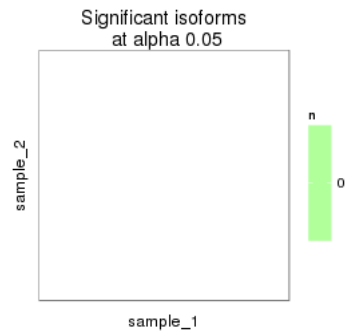
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

[1] "no sig isoforms"

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

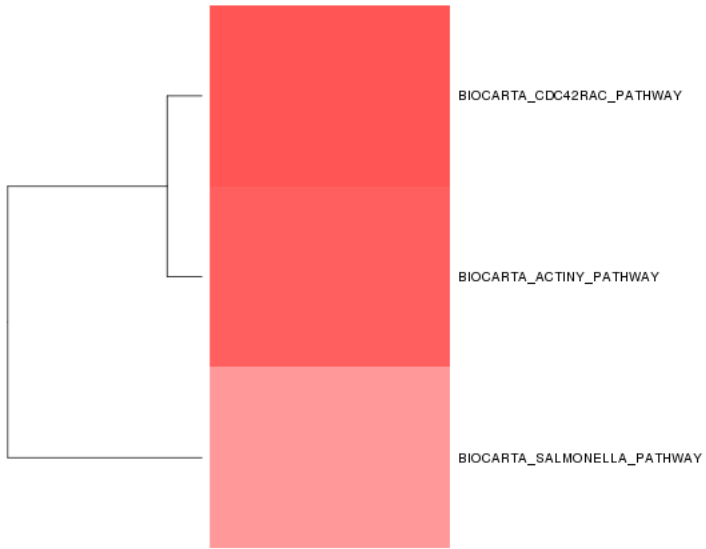
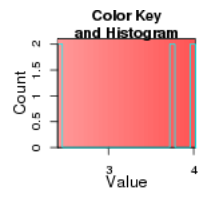
Gene/Pathway Analysis

GSEA

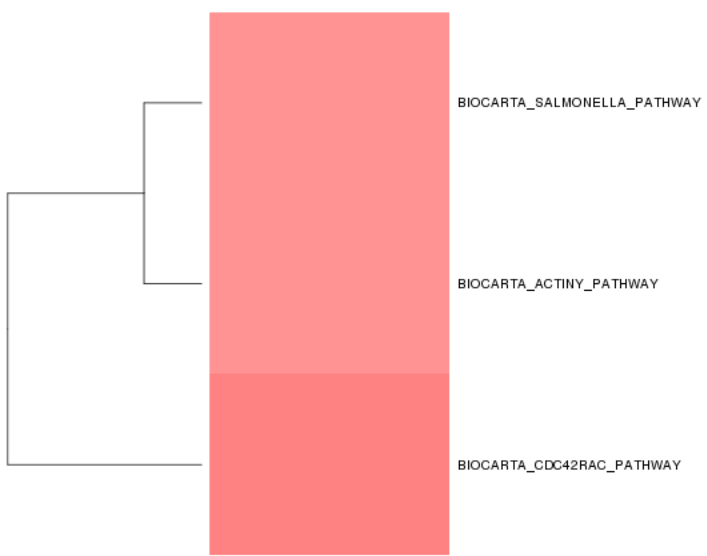
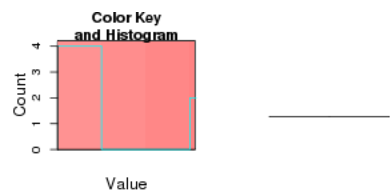
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

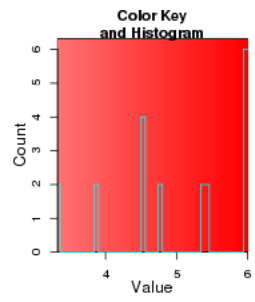
Biocarta enrichment:

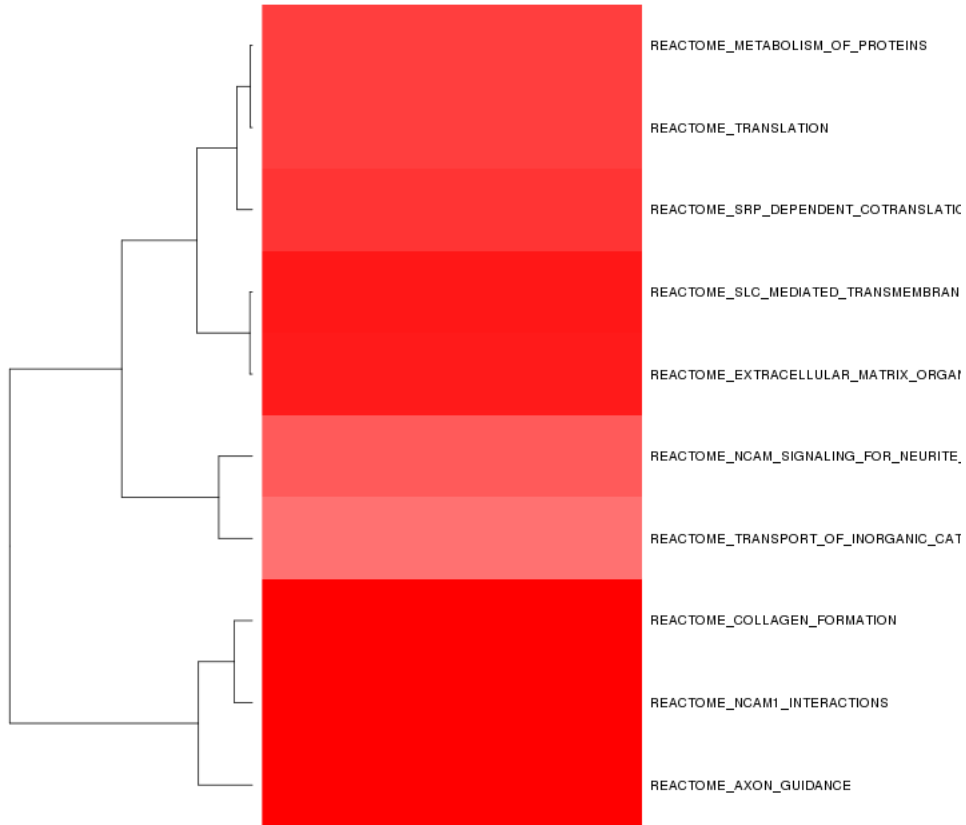


Biocarta zscore:

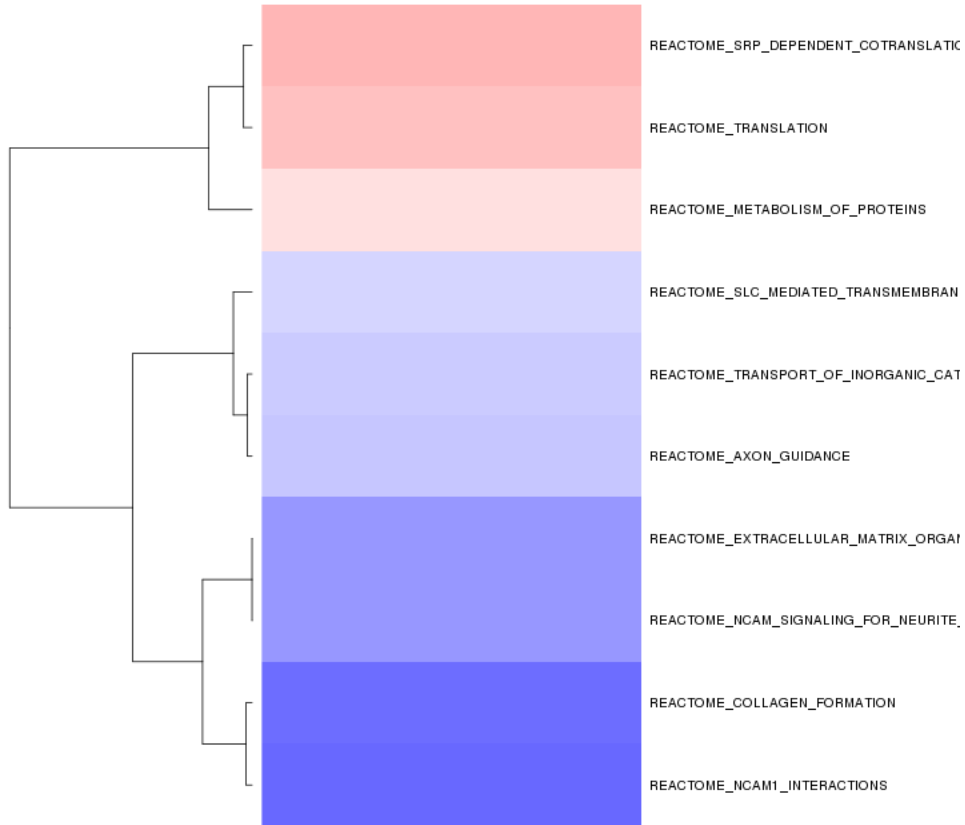
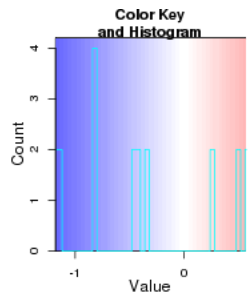


Reactome enrichment:





Reactome zscore:



Kegg enrichment:

Error: argument is of length zero

Kegg zscore:

Error: incorrect number of dimensions


```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

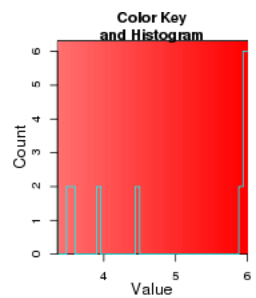
Interneuron enrichment:

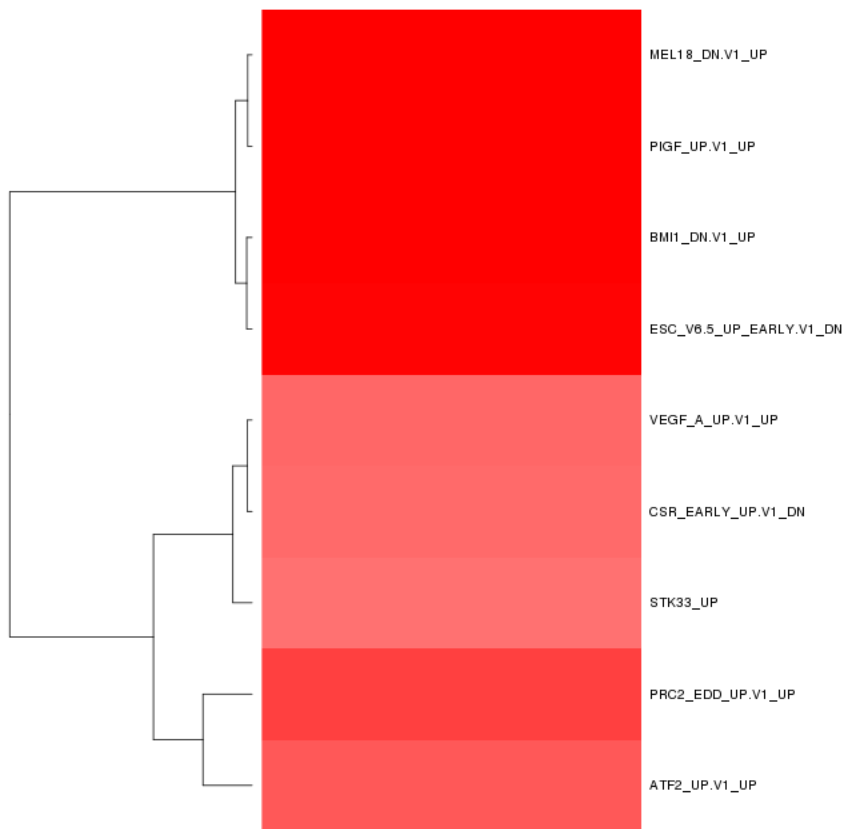
```
## [1] "Not enough significant categories to print a heatmap!"
```

Interneuron zscore:

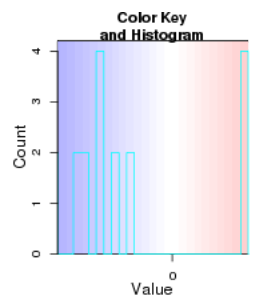
```
## [1] "Not enough significant categories to print a heatmap!"
```

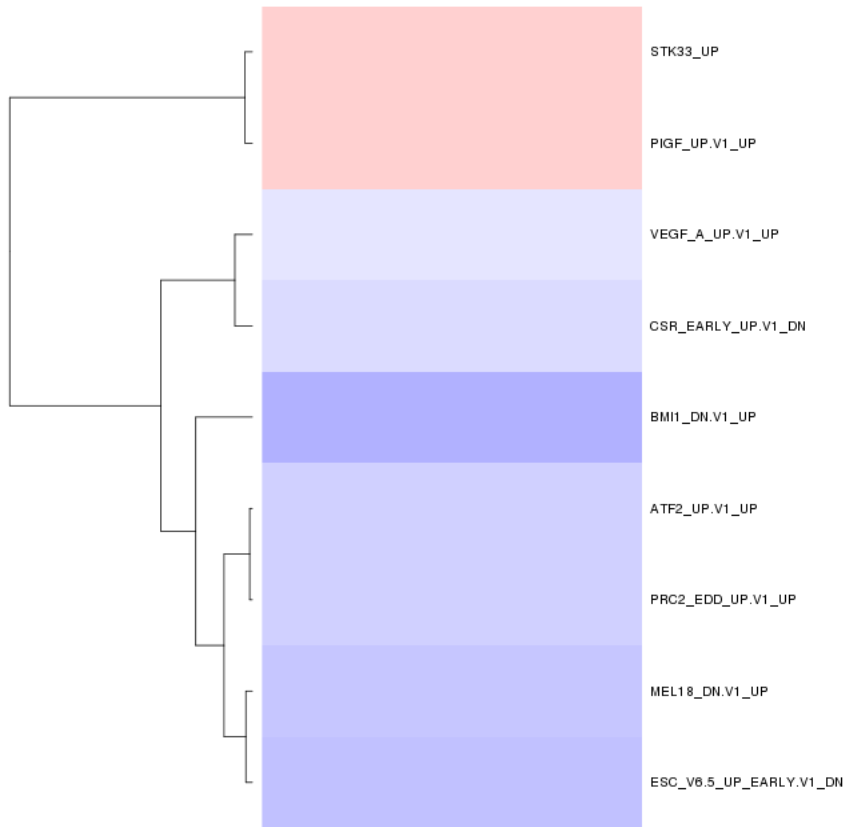
Oncogene enrichment:





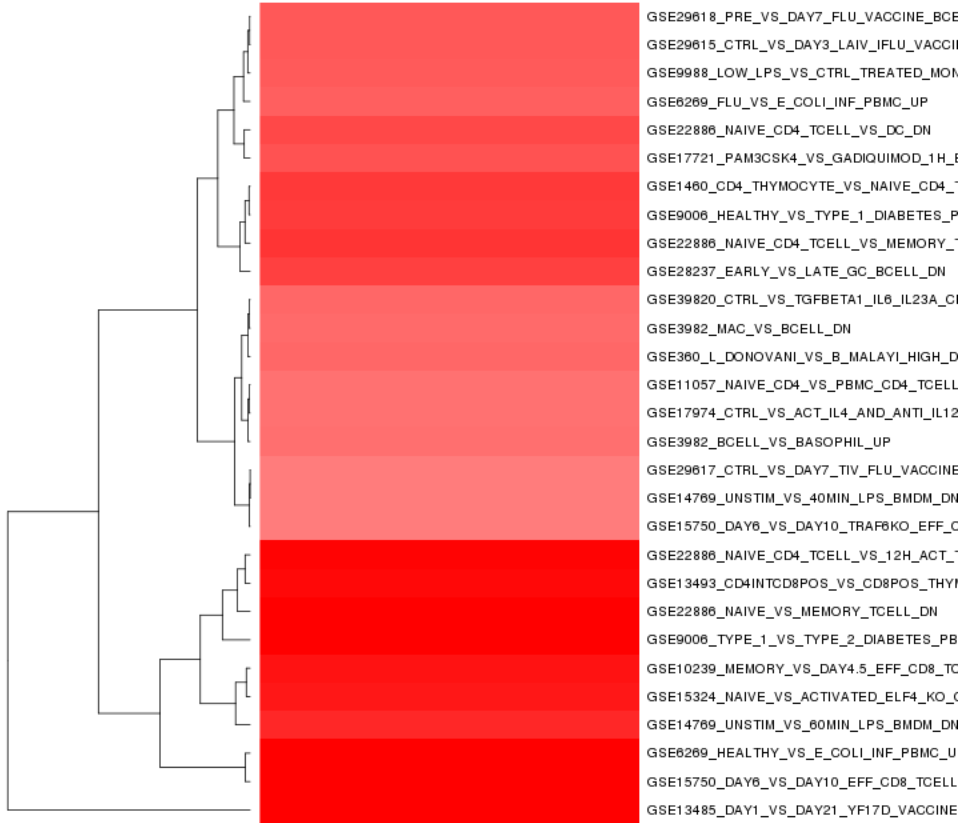
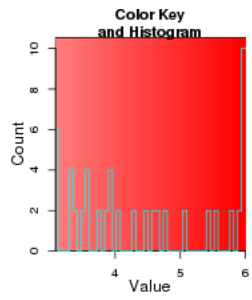
Oncogene zscore:





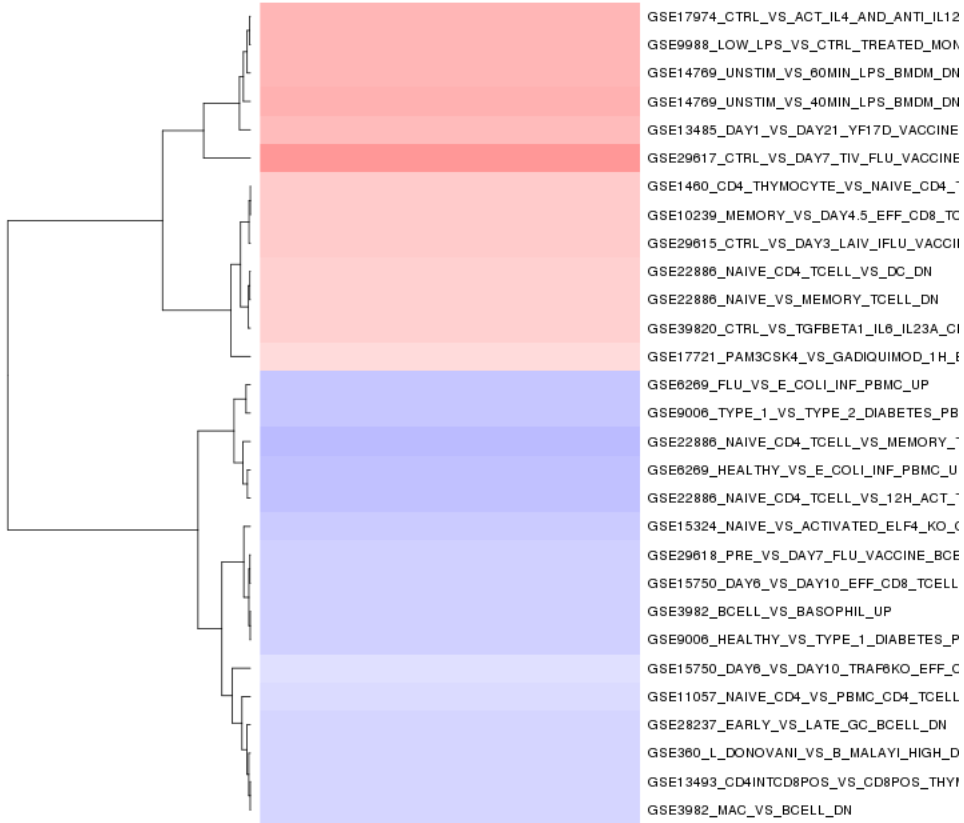
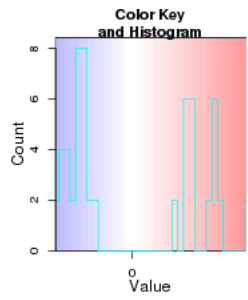
Immuno enrichment:

Error: subscript out of bounds

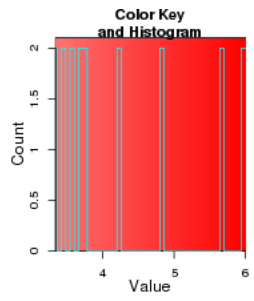


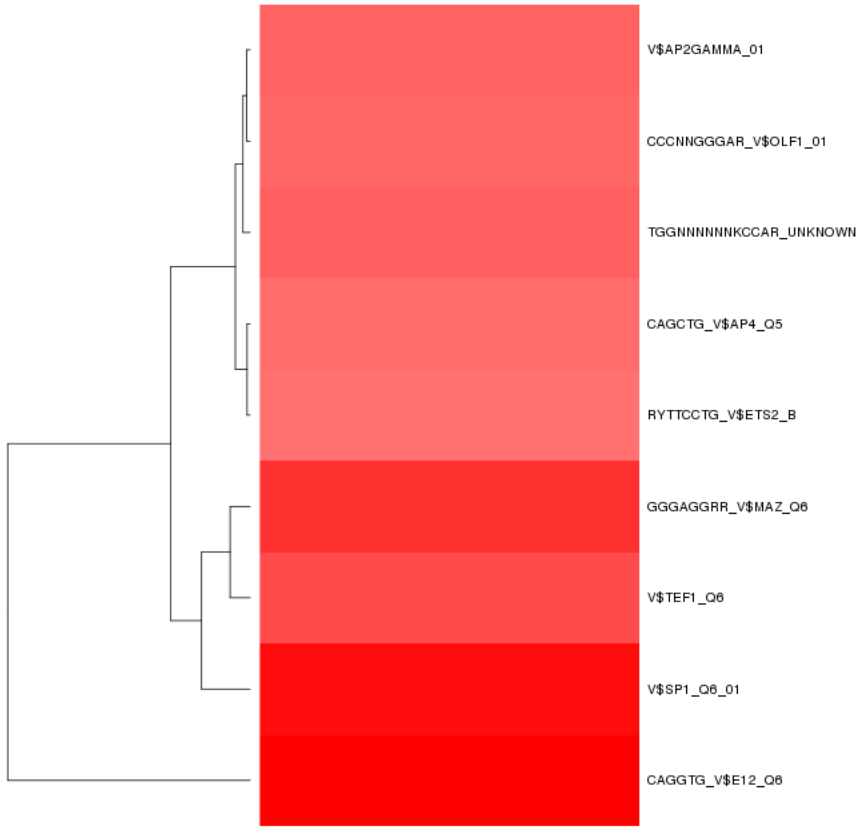
Immuno zscore:

Error: subscript out of bounds

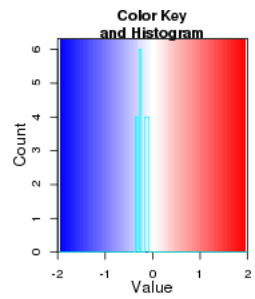


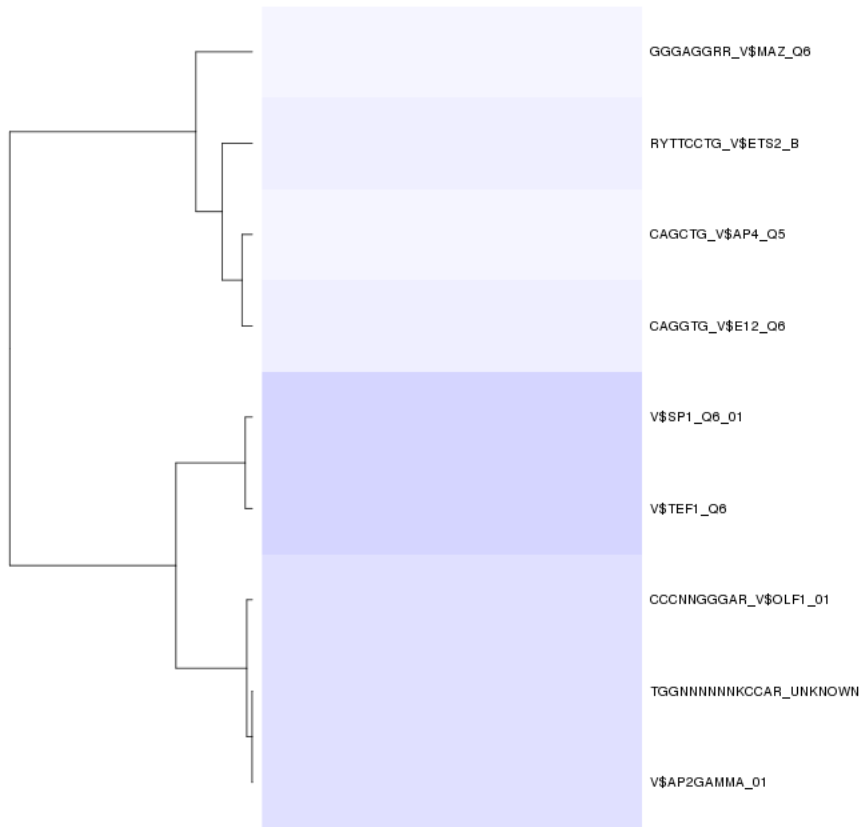
TF enrichment:





TF zscore:

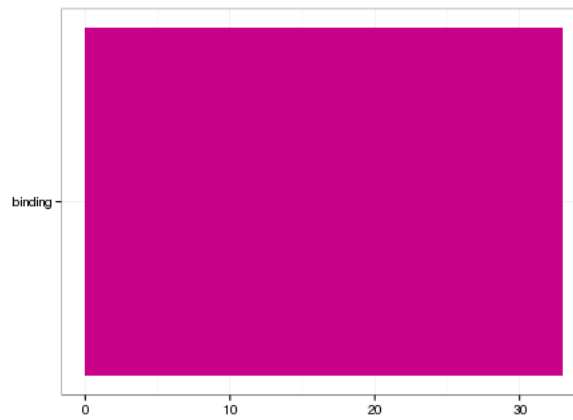
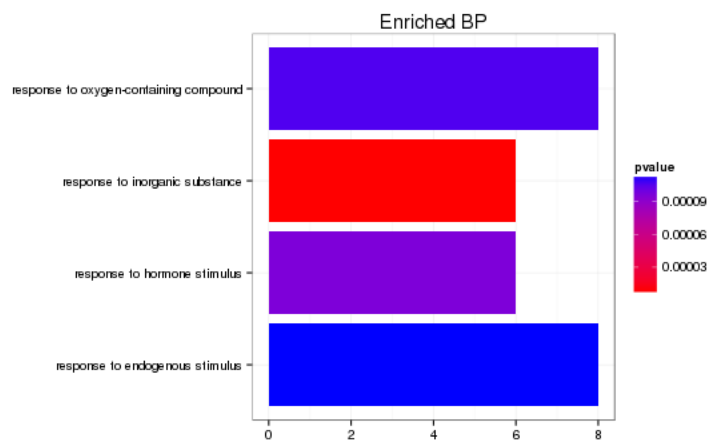




GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

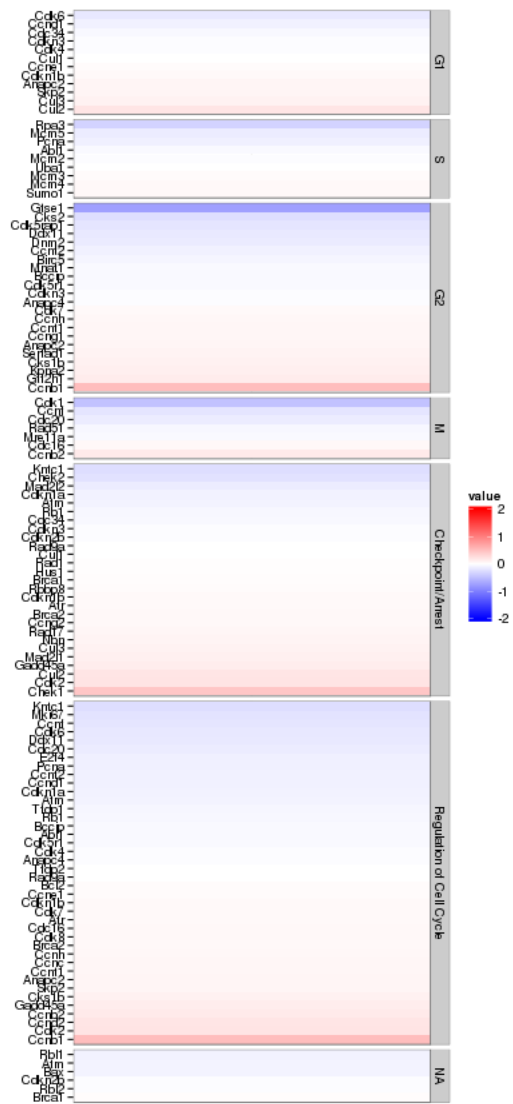
```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```

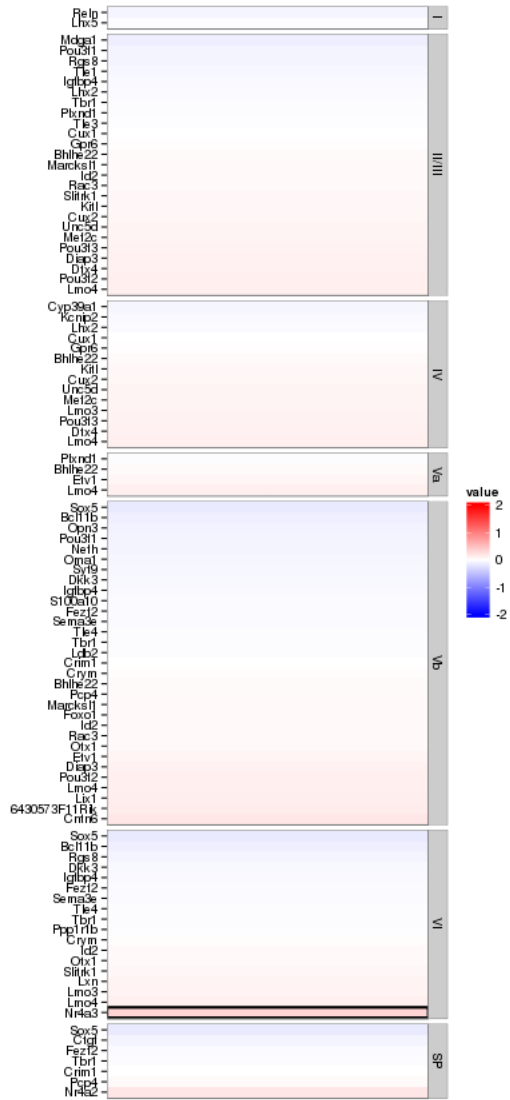




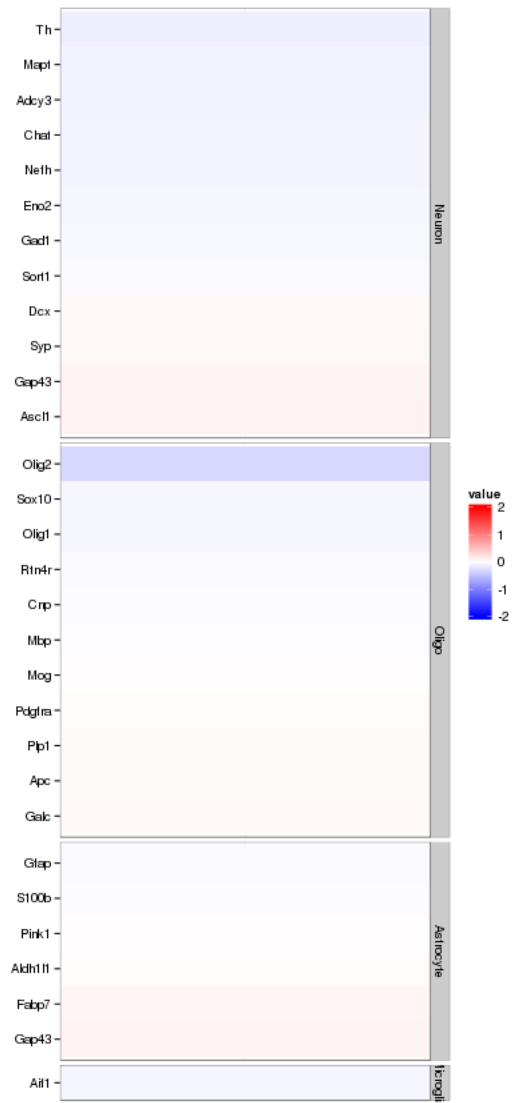
```
## Error: need finite 'xlim' values
```

Enrichment or depletion for stage-specific cell cycle markers

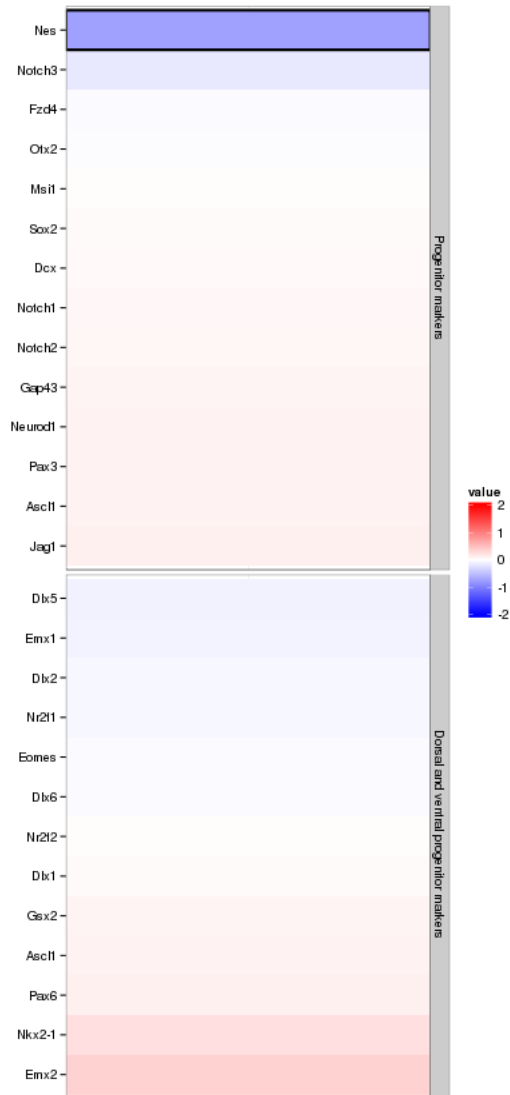




Enrichment or depletion for specific neural cell types



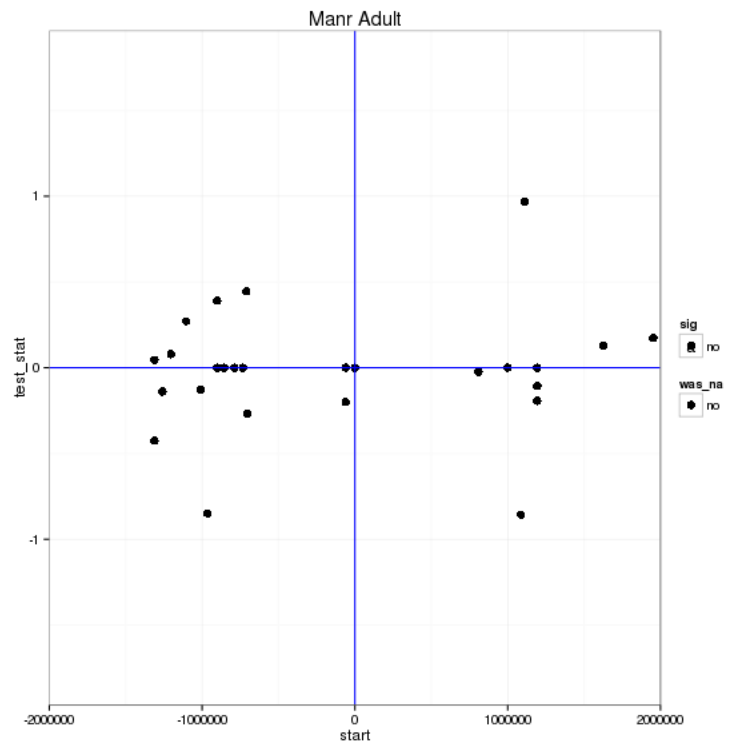
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR780
16 JR737
17 JR801

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34899700.00	0.75	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34899700.00	0.58	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34899700.00	0.97	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34899700.00	0.80	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34899700.00	1.03	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34899700.00	1.06	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34899700.00	0.80	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34899700.00	1.15	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34899700.00	1.00	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34899700.00	1.29	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34899700.00	1.49	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34899700.00	1.01	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34899700.00	0.97	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34899700.00	1.20	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR780/abundances.cxb	Manr	0	Manr_0	40782700.00	34899700.00	1.17	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR737/abundances.cxb	Manr	1	Manr_1	40835900.00	34899700.00	1.19	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR801/abundances.cxb	Manr	2	Manr_2	34495500.00	34899700.00	0.96	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1      reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1        zlibbioc_1.8.0

```


Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Manr -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Manr_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/ann
## 2
## 3
## 4
## 5
```

Manr KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Manr.

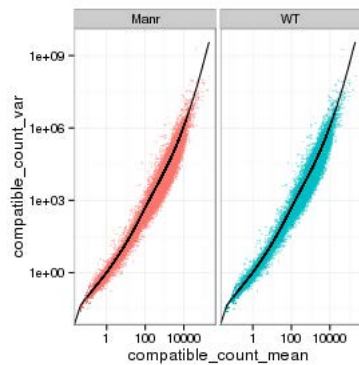
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

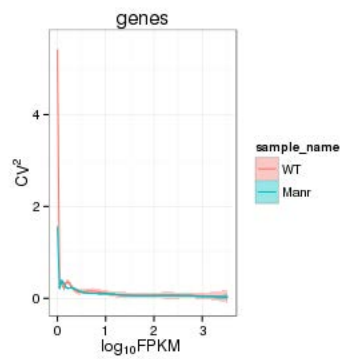
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



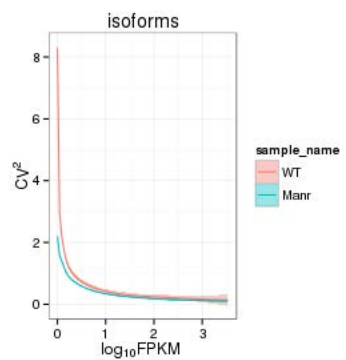
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

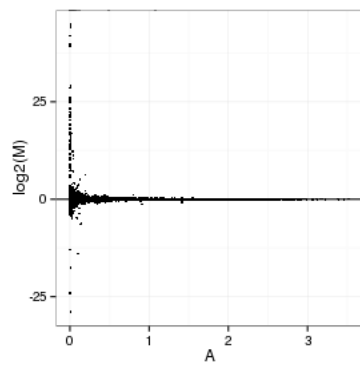
Genes:



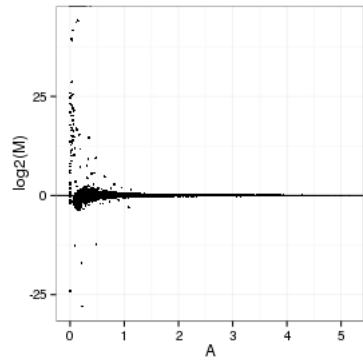
Isoforms:



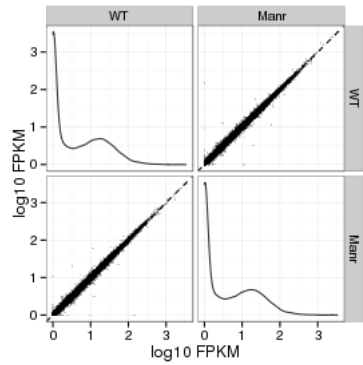
MvA plot



MvA plot counts



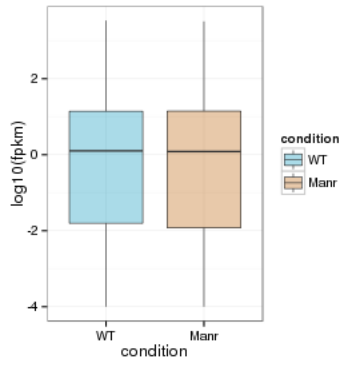
Scatterplot matrix



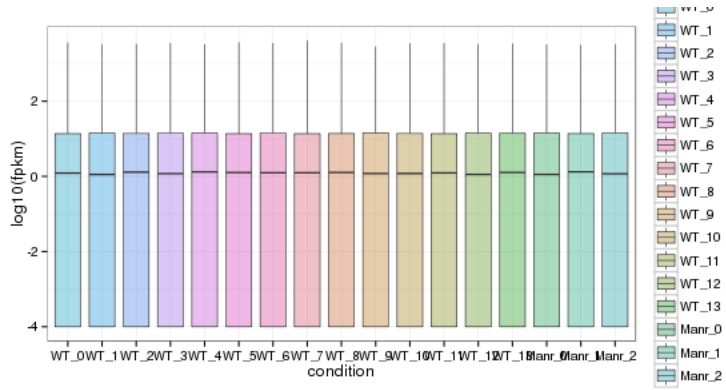
Distributions

Boxplots

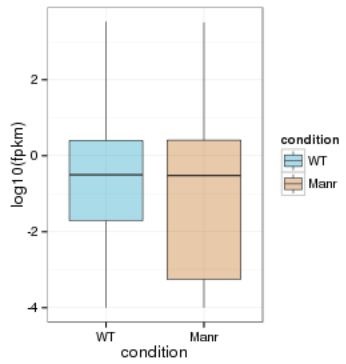
Boxplot (genes)



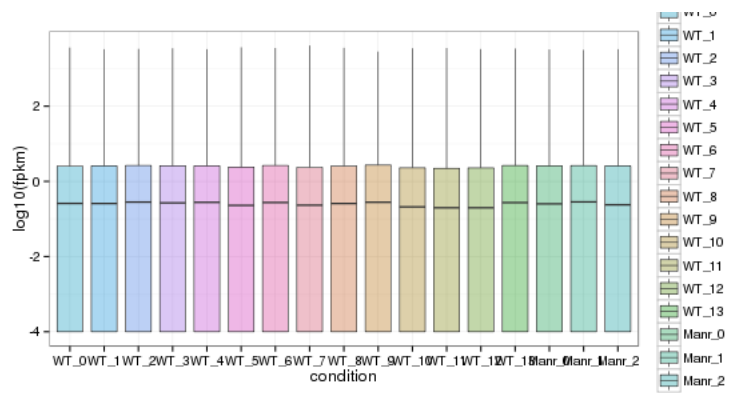
Boxplot (genes, replicates)



Boxplot (isoforms)

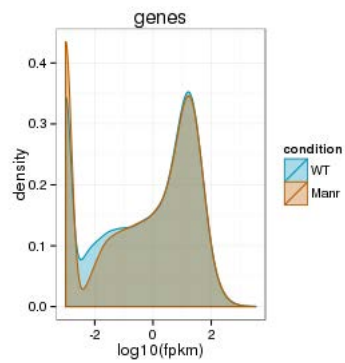


Boxplot (isoforms, replicates)

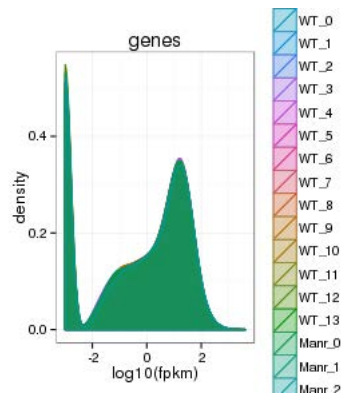


Density

Density (genes)

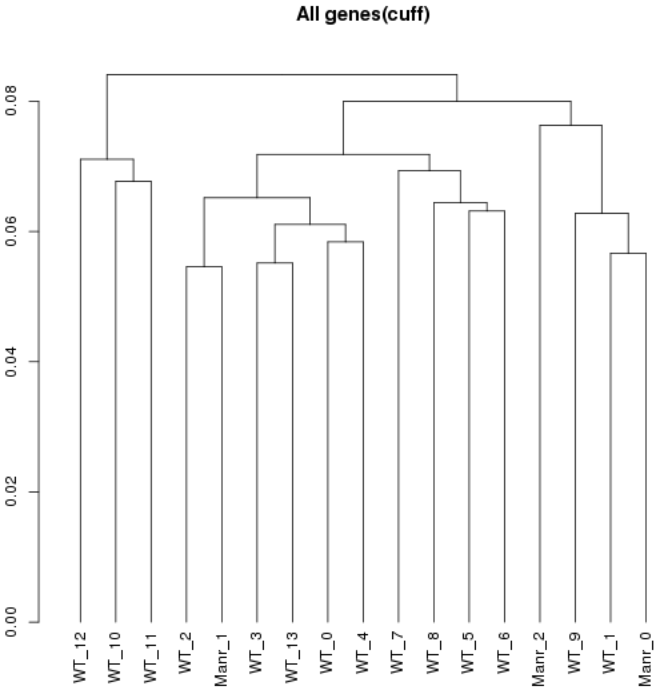


Density (genes, replicates)



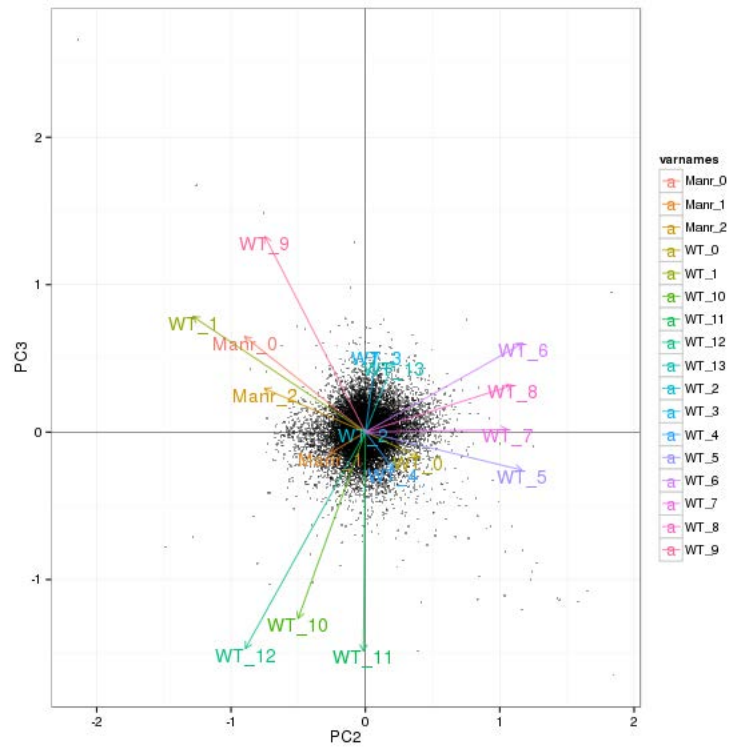
Clustering

Replicate Clusters

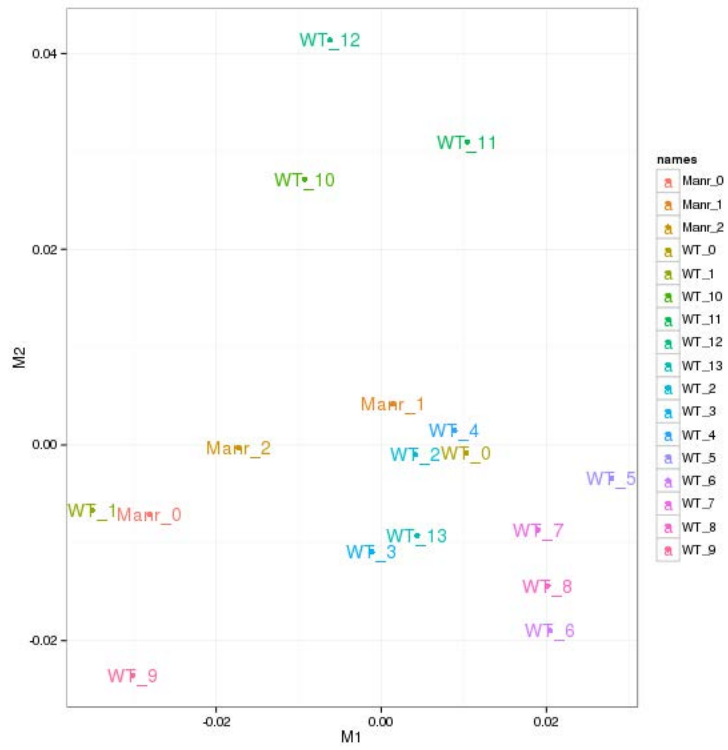


NULL

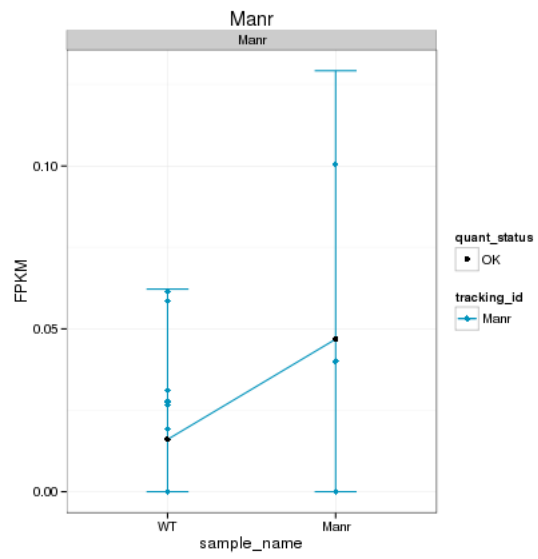
PCA (genes)



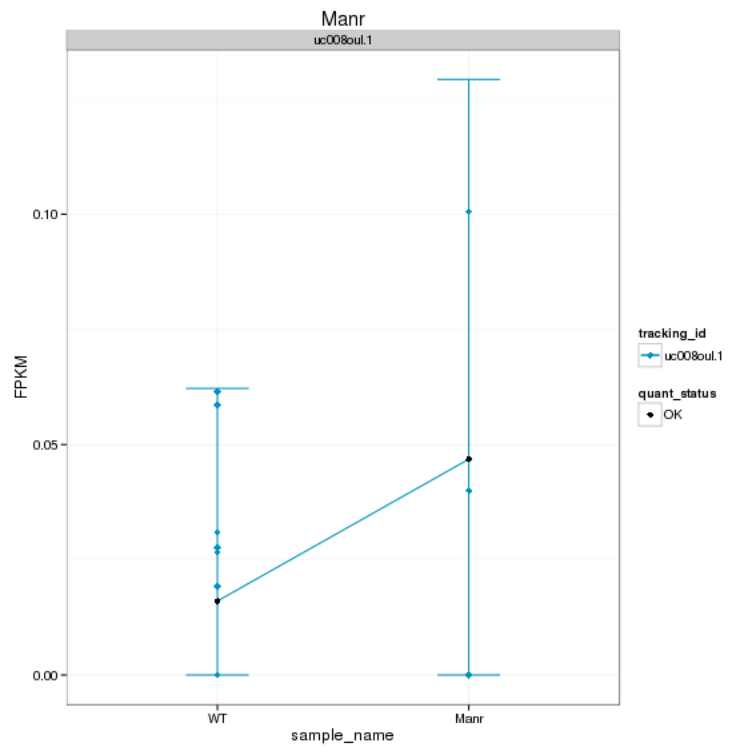
MDS (genes)



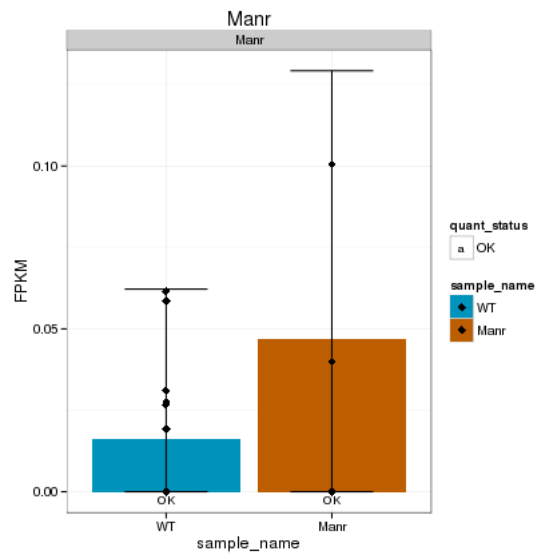
KO assessment
Endogenous lncRNA expression



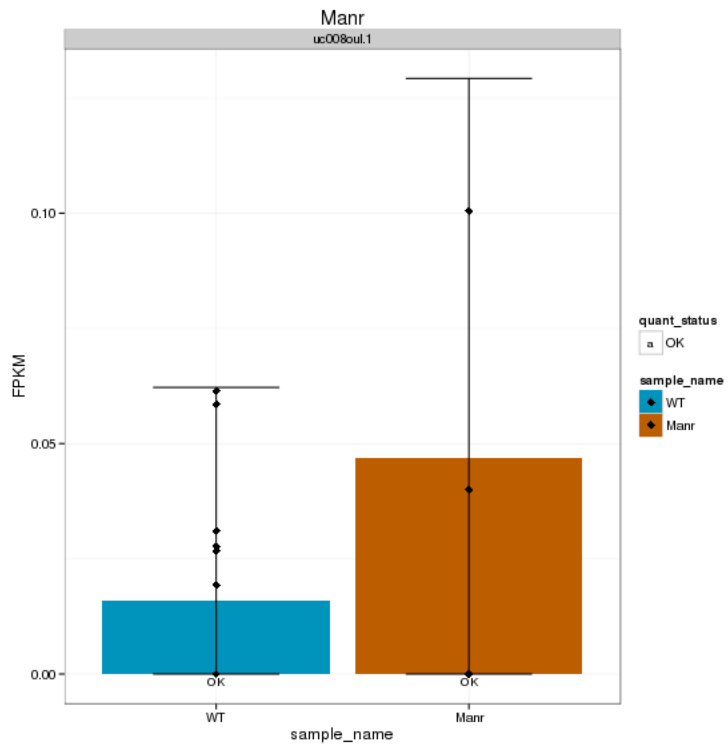
Endogenous expression of Manr isoforms:



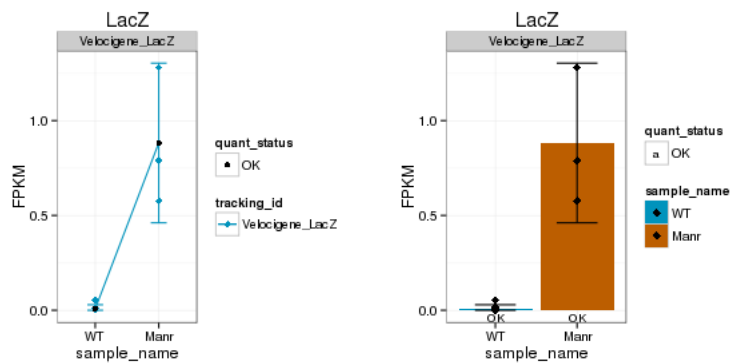
Barplot of gene expression:



Barplot of isoform expression:

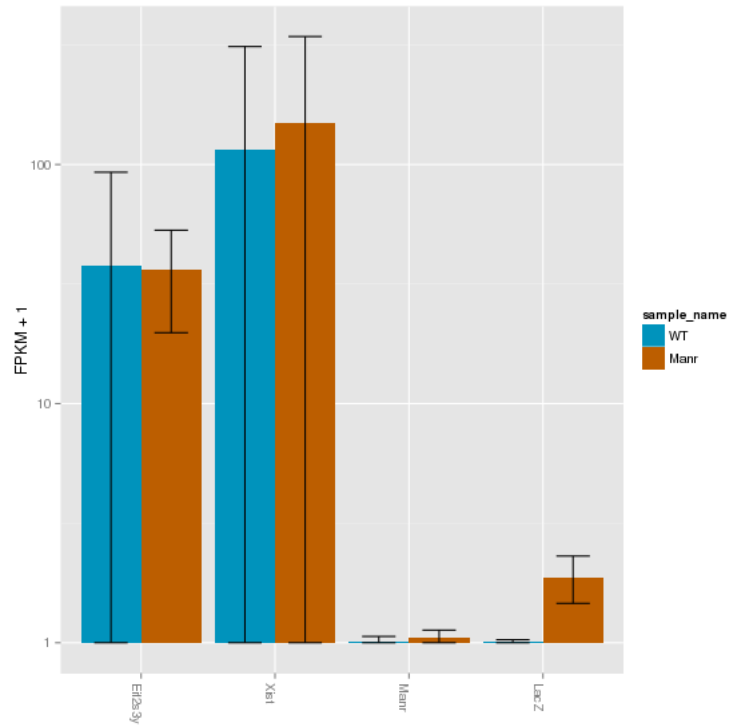


LacZ expression

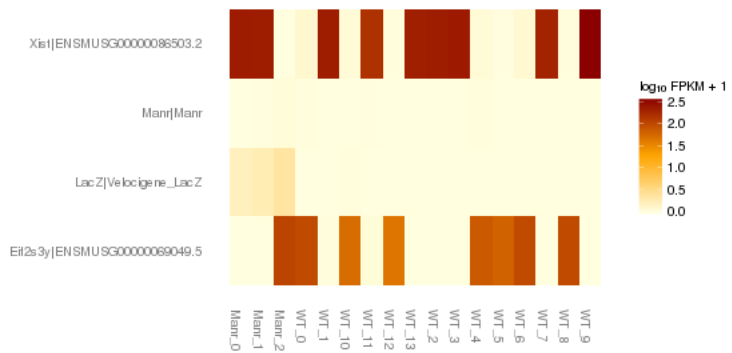


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



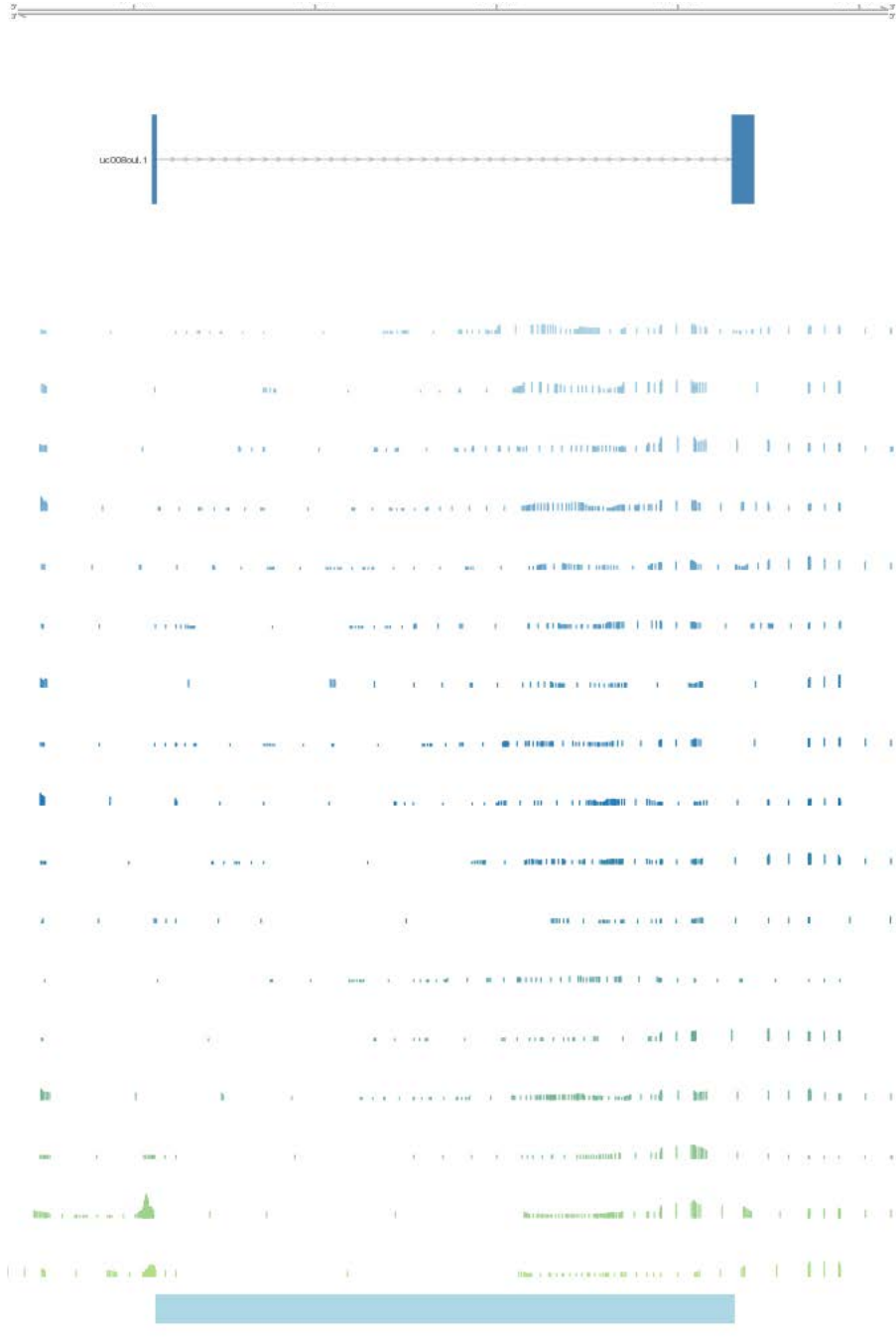
Expression heatmap:



Track visualization



KO Region Manti_2 Manti_3 Manti_0 Manti_1 WT_13 WT_12 WT_11 WT_10 WT_9 WT_8 WT_7 WT_6 WT_5 WT_4 WT_3 WT_2 WT_1 WT_0 LncRNA Isoforms



Differential Analysis

Differential Genes

There are 86 significantly differentially expressed genes. They are:

```
geneAnnot$gene_short_name
1 Gabra2
2 Col1a1
3 Nfix
4 Phyhip
5 Cacna1e
6 Mef2c
7 Fam131c
8 Ptgds
9 Hivep2
10 Eef1a2
11 Ascl1
12 Adcy1
13 Doc2b
14 Clu
15 Sla
16 Shisa9
17 Lynx1
18 Mal2
19 Gnal
20 Snca
21 Sphkap
22 Cybrd1
23 Fabp5
24 Slc7a11
25 Hmgcs2
26 Rps3a1
27 Nr4a3
28 Tnc
29 Tal2
30 Hpca
31 Ppp2r2c
32 Fosl2
33 Rph3a
34 Eln
35 Dync1i1
36 Slc6a11
37 Dbx1
```


38 Hddc3
39 Mfge8
40 Thy1
41 Robo3
42 Cspg5
43 Ablim3

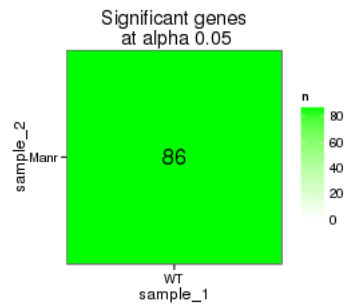
44 Rims3
45 Camkv
46 Mgl1
47 Arhgap27
48 Sncb
49 Mid1
50 Nov
51 Vgf
52 Camk4
53 Pcsk1n
54 Grin3a
55 Gabbr2
56 Necab1
57 Abcb1a
58 Mpped1
59 Bend6
60 Hebp1
61 Tuba1c
62 Cnpy1
63 Prkce
64 Olig1
65 Nxph3
66 Mc4r
67 Tmem72
68 Grm5
69 Slc6a7
70 Gpr17

71 Hbb-bs
72 Aldh1a1
73 AI854703
74 6330403A02Rik
75 Vsnl1
76 Gabra5
77 Adap1
78 Hmga2
79 Rps12
80 Pcna-ps2

- 81 Rpl23
- 82 Wdfy1
- 83 Hbb-bt
- 84 A1593442
- 85 Hs3st4
- 86 AA465934

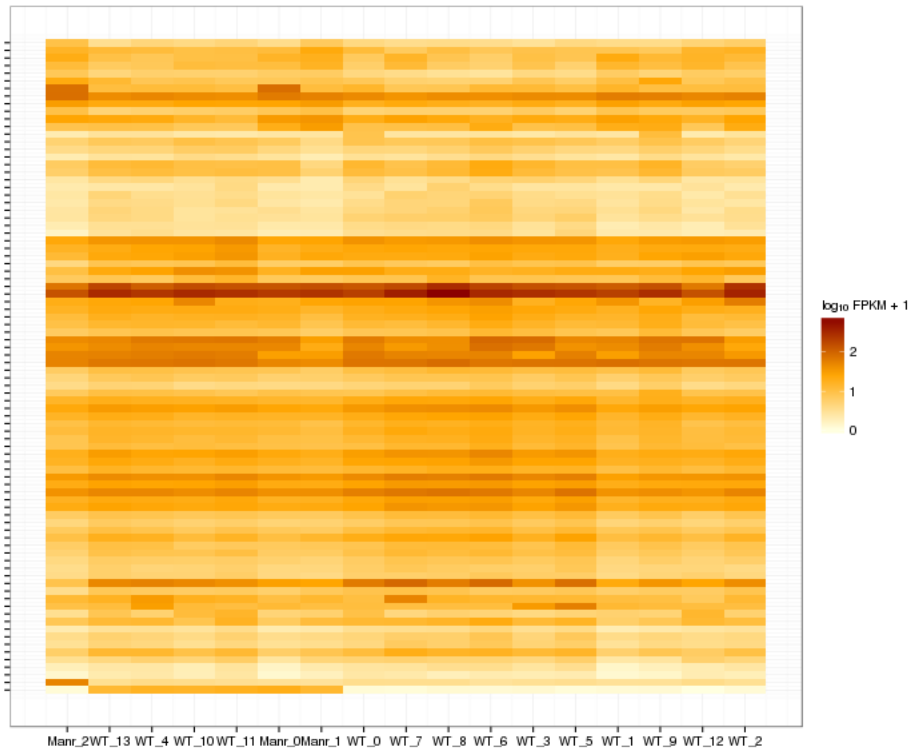
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

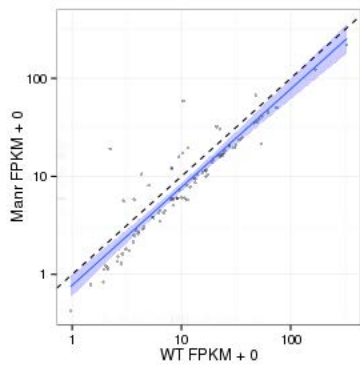


Significant genes with expression >50fpm (any condition):(turned off)

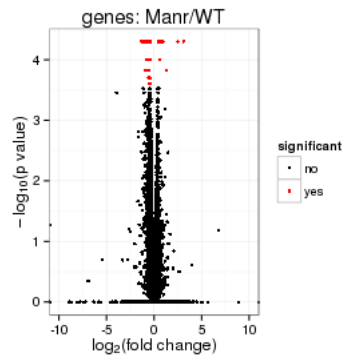
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

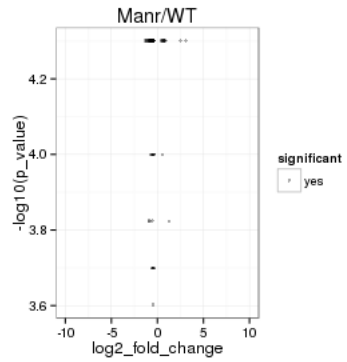
Scatter plot of significant genes only:



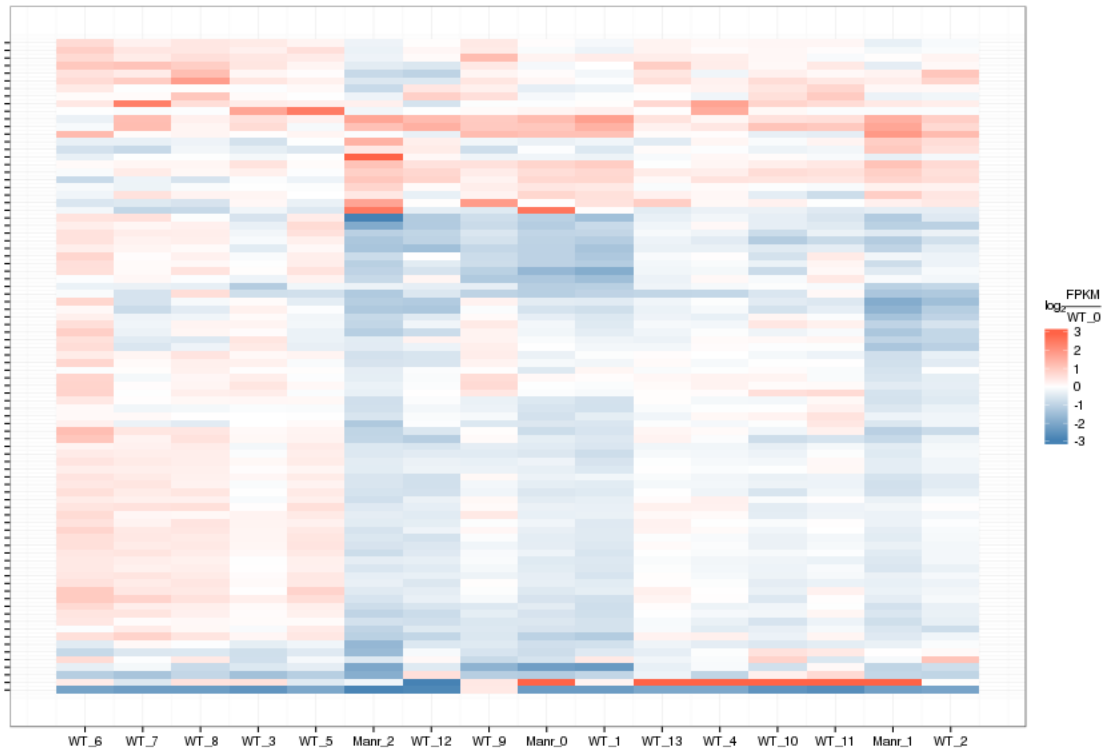
Volcano Plot



Volcano plot with significant genes only:



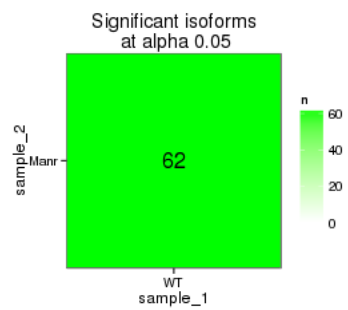
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:

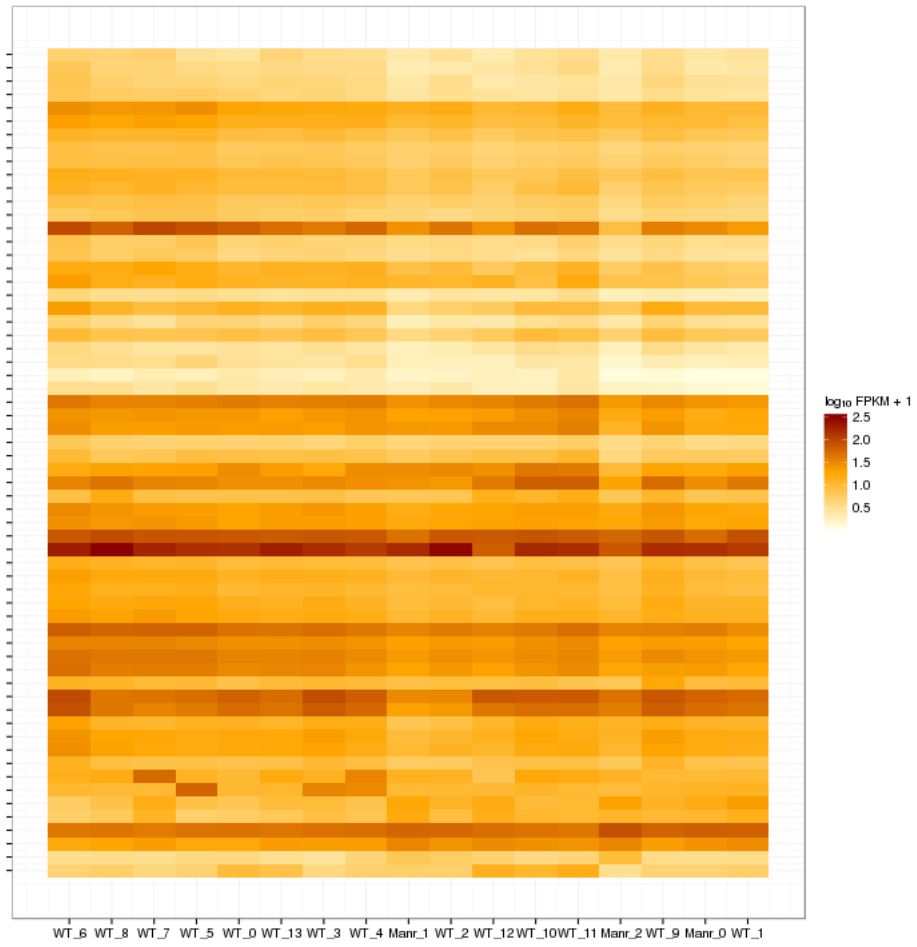


These isoforms are:

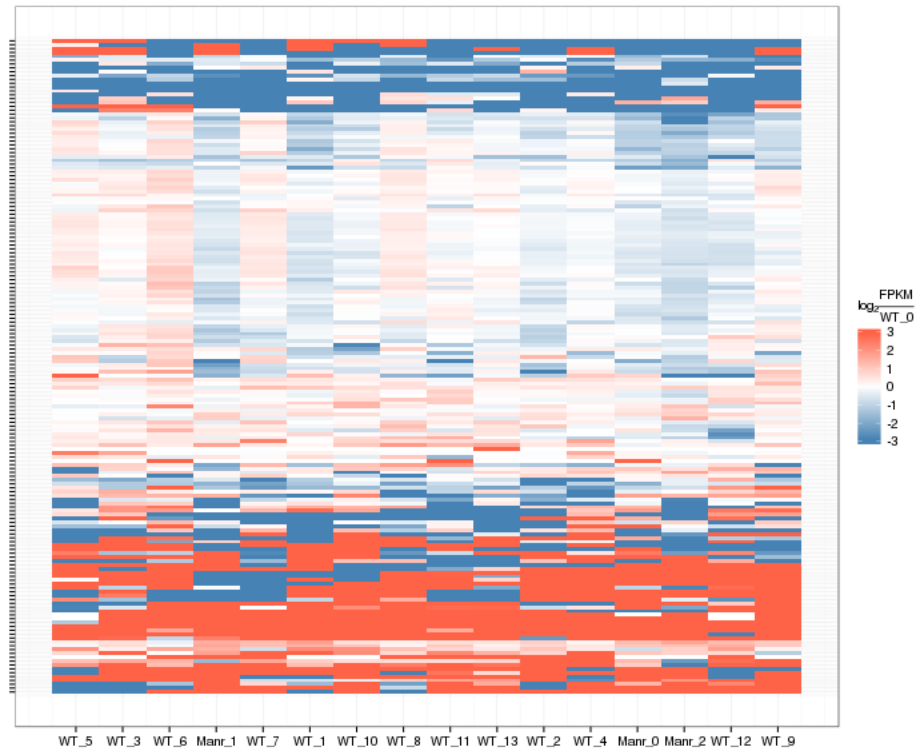
isoAnnot	gene_short_name
1	Gabra2
2	Col1a1
3	Nfix
4	Cacna1e
5	Ndrg1
6	Fam131c
7	Ptgds
8	Hivep2
9	Eef1a2
10	Kitl
11	Ascl1
12	Adcy1
13	Doc2b
14	Gng4
15	Clu
16	Sla
17	Lynx1
18	Mal2
19	Fabp5
20	Slc7a11
21	Tnc
22	Tal2
23	Ppp2r2c
24	Fosl2
25	Ein
26	Slc6a11
27	Dbx1
28	Hddc3
29	Mfge8
30	Thy1
31	Cspg5
32	Camkv
33	Gas7
34	Sncb
35	Vgf
36	Pcsk1n
37	Gabbr2
38	Necab1
39	Mpped1
40	Bend6

41 Reln
42 Hebp1
43 Prkce
44 Hpcal4
45 Olig1
46 Nxph3
47 Mc4r
48 Tmem72
49 Grm5
50 Slc6a7
51 Gpr17
52 Aldh1a1
53 AI854703
54 6330403A02Rik
55 Vsnl1
56 Gabra5
57 Slc4a4
58 Opcml
59 Pcna-ps2
60 Hbb-bt
61 AI593442
62 Hs3st4

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

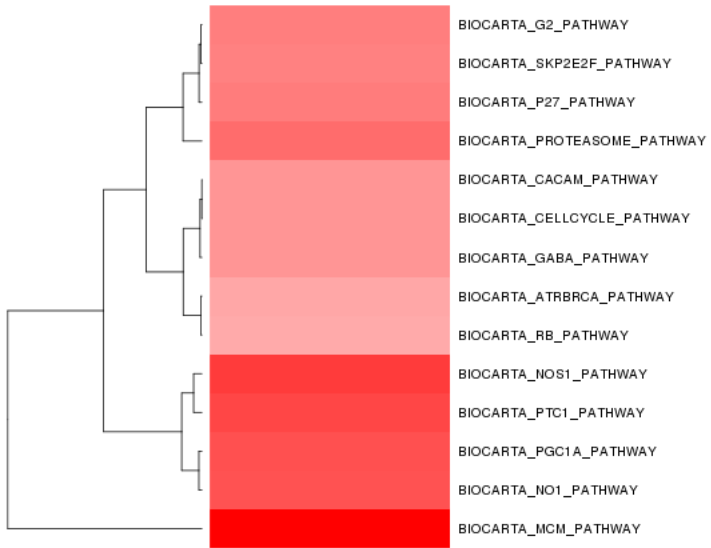
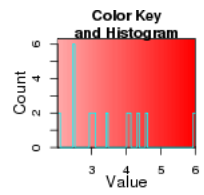
Gene/Pathway Analysis

GSEA

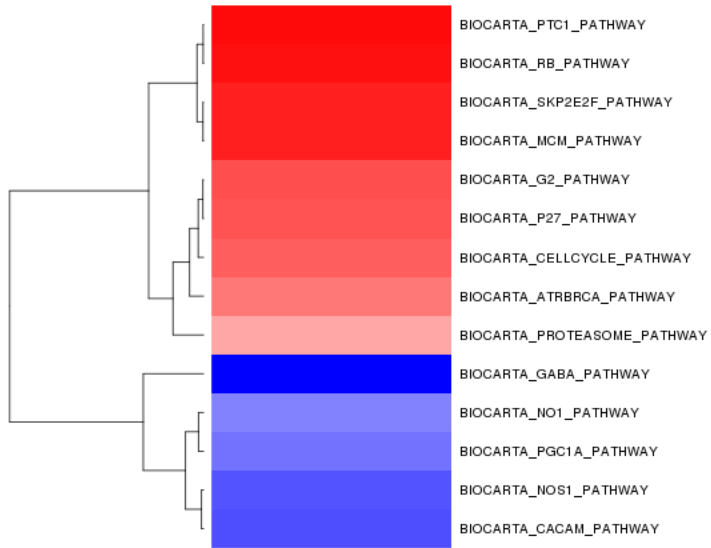
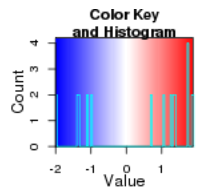
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

Biocarta enrichment:

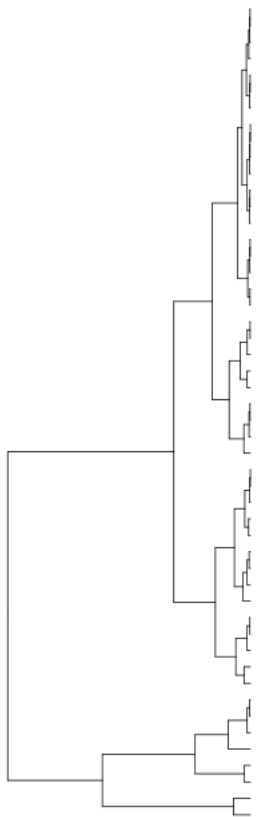


Biocarta zscore:



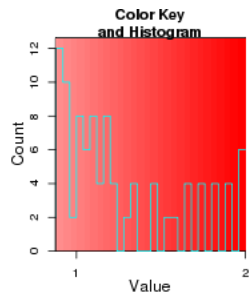
Reactome enrichment:

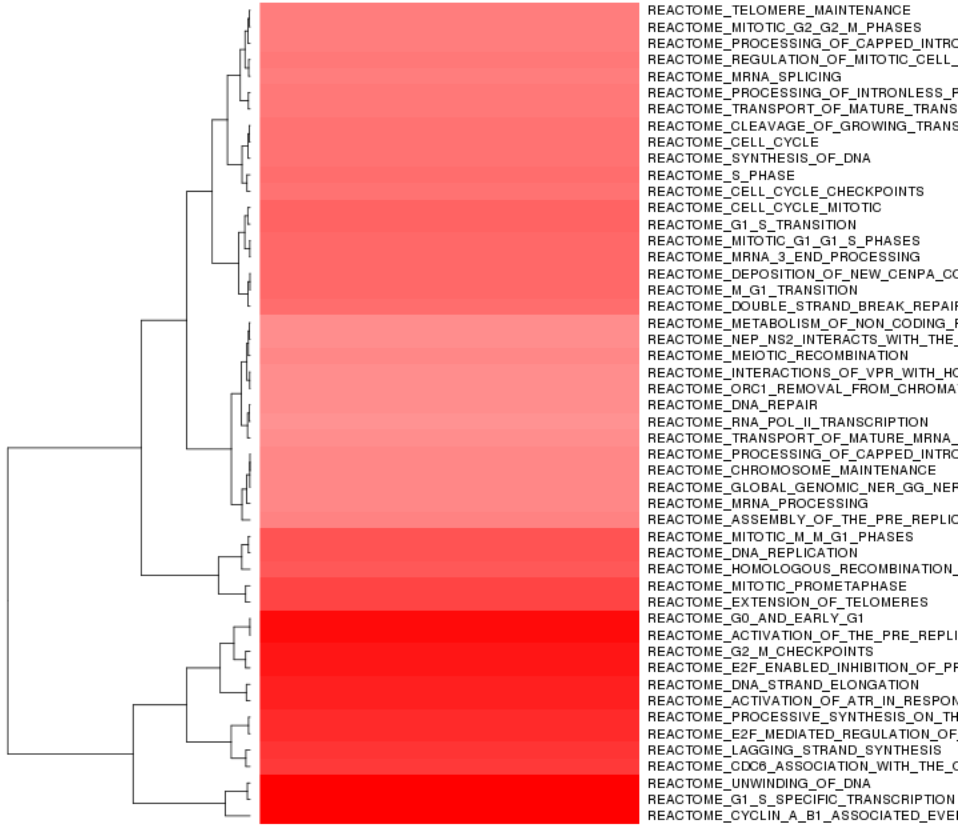
Error: no locations are finite



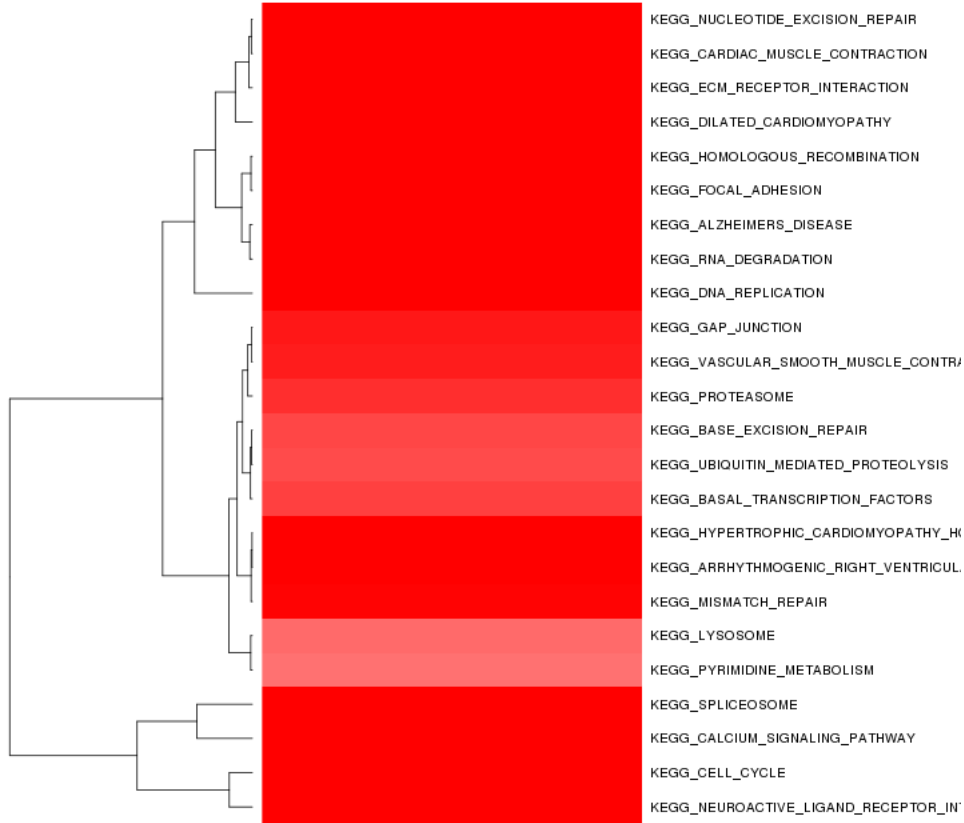
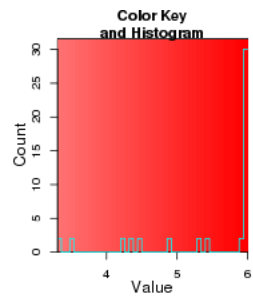
REACTOME_NUCLEOTIDE_EXCISION_REPAIR
REACTOME_MITOTIC_G2_G2_M_PHASES
REACTOME_E2F_MEDIATED_REGULATION_OF
REACTOME_ASSEMBLY_OF_THE_PRE_REPLIC
REACTOME_CLEAVAGE_OF_GROWING_TRANS
REACTOME_RNA_POL_II_PRE_TRANSCRIPTIO
REACTOME_DNA_STRAND_ELONGATION
REACTOME_ORC1_REMOVAL_FROM_CHROMA
REACTOME_PLATELET_ACTIVATION_SIGNALIN
REACTOME_POTASSIUM_CHANNELS
REACTOME_GPCR_LIGAND_BINDING
REACTOME_TRANSCRIPTION_COUPLED_NER_
REACTOME_METABOLISM_OF_NON_CODING_F
REACTOME_AXON_GUIDANCE
REACTOME_MRNA_3_END_PROCESSING
REACTOME_METABOLISM_OF_MRNA
REACTOME_REGULATION_OF_MITOTIC_CELL_
REACTOME_NEUROTRANSMITTER_RECEPTOR
REACTOME_HEMOSTASIS
REACTOME_LATE_PHASE_OF_HIV_LIFE_CYCLE
REACTOME_HIV_INFECTION
REACTOME_M_G1_TRANSITION
REACTOME_G2_M_CHECKPOINTS
REACTOME_TRANSPORT_OF_MATURE_TRANS
REACTOME_TRANSMEMBRANE_TRANSPORT_C
REACTOME_ACTIVATION_OF_ATR_IN_RESPON
REACTOME_CHROMOSOME_MAINTENANCE
REACTOME_ACTIVATION_OF_THE_PRE_REPLI
REACTOME_GPCR_DOWNSTREAM_SIGNALING
REACTOME_METABOLISM_OF_RNA
REACTOME_HIV_LIFE_CYCLE
REACTOME_SYNTHESIS_OF_DNA
REACTOME_MITOTIC_PROMETAPHASE
REACTOME_DNA_REPAIR
REACTOME_TRANSMISSION_ACROSS_CHEMIC
REACTOME_SIGNALING_BY_GPCR
REACTOME_CELL_CYCLE_CHECKPOINTS
REACTOME_RNA_POL_II_TRANSCRIPTION
REACTOME_G1_S_TRANSITION
REACTOME_S_PHASE
REACTOME_MITOTIC_G1_G1_S_PHASES
REACTOME_TRANSCRIPTION
REACTOME_PROCESSING_OF_CAPPED_INTRC
REACTOME_MRNA_PROCESSING
REACTOME_MITOTIC_M_M_G1_PHASES
REACTOME_DNA_REPLICATION
REACTOME_NEURONAL_SYSTEM
REACTOME_MRNA_SPLICING
REACTOME_CELL_CYCLE
REACTOME_CELL_CYCLE_MITOTIC

Reactome zscore:

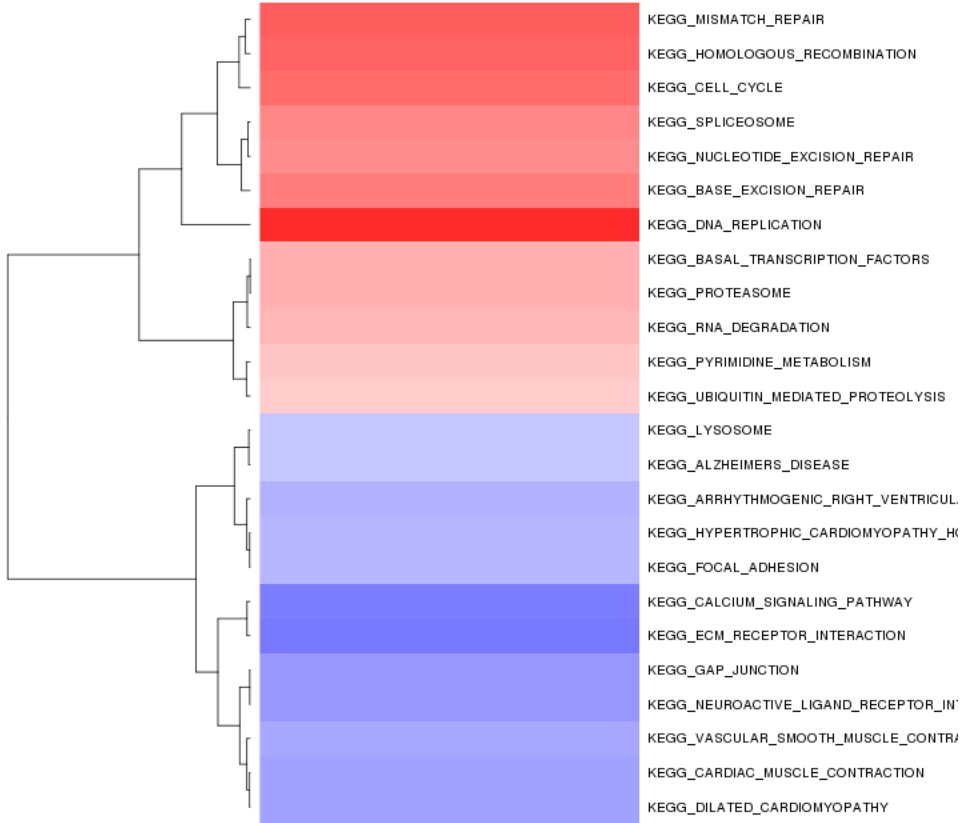
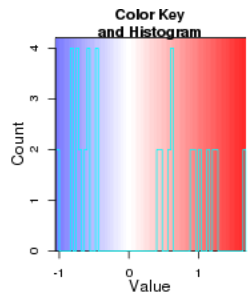




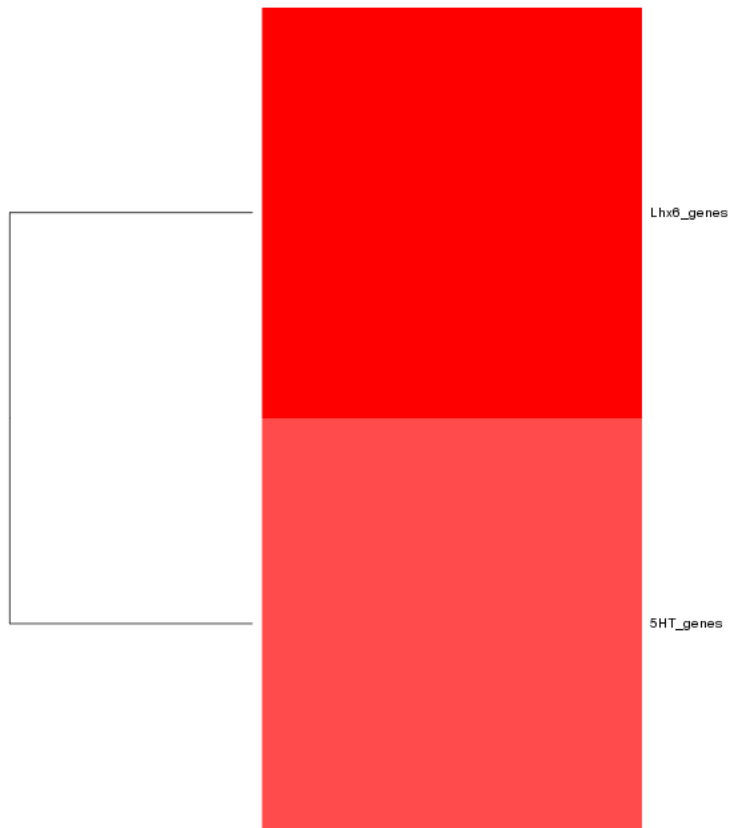
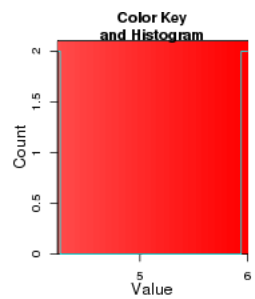
Kegg enrichment:



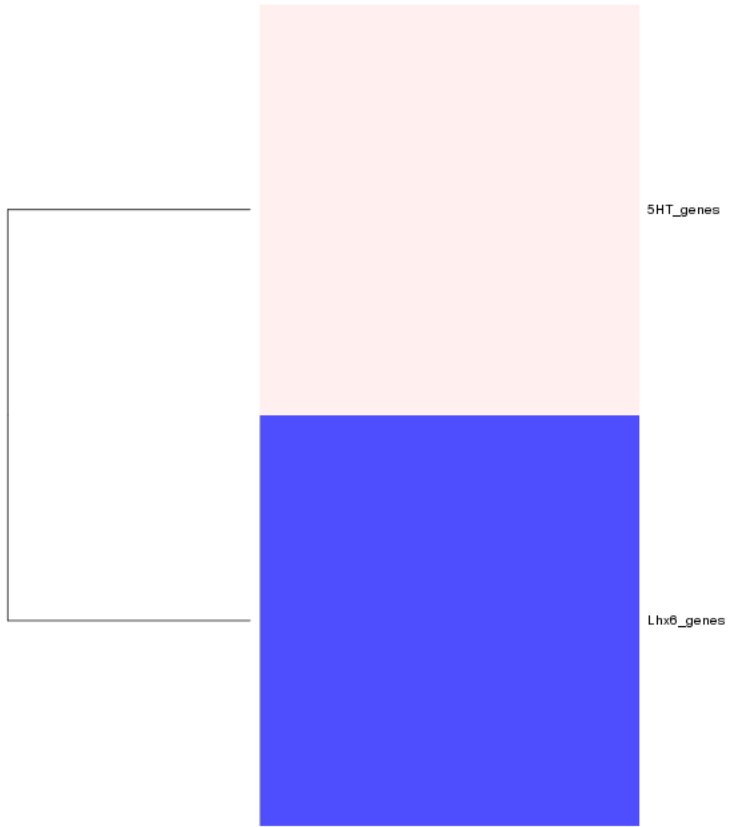
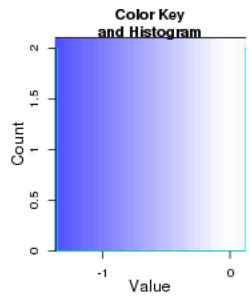
Kegg zscore:



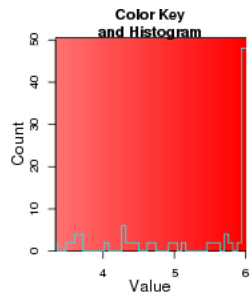
Interneuron enrichment:

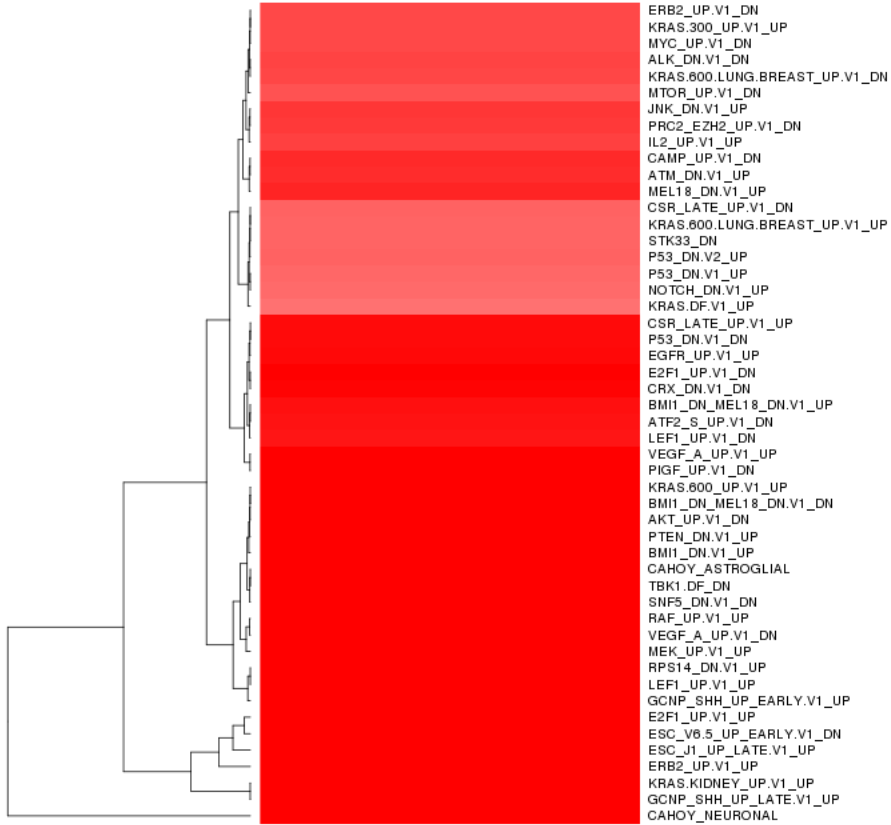


Interneuron zscore:

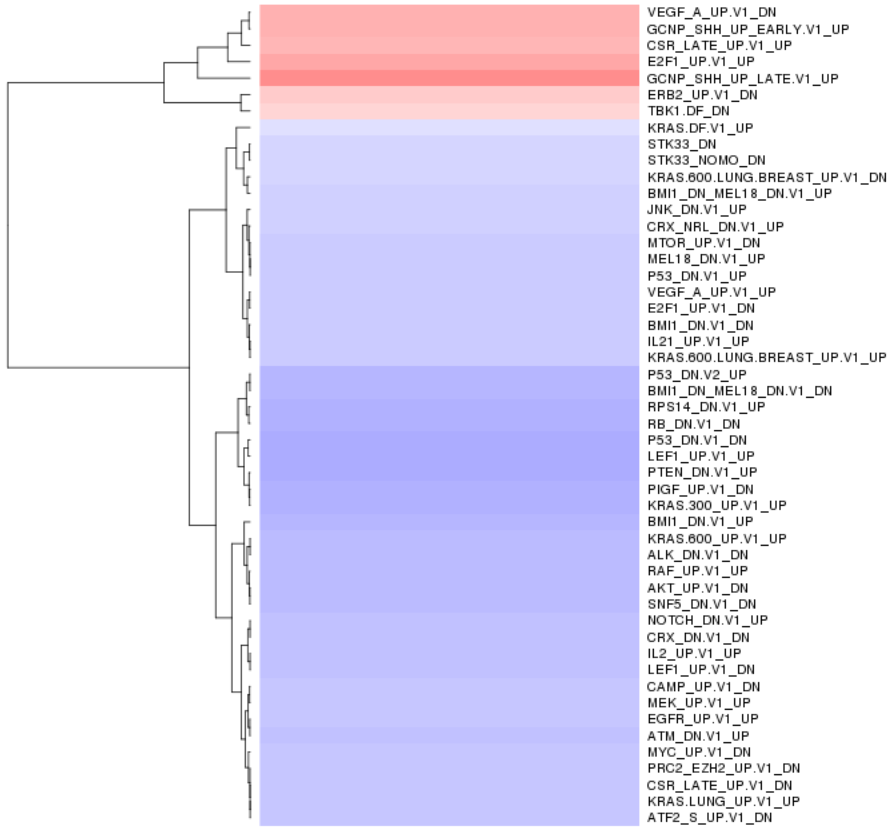
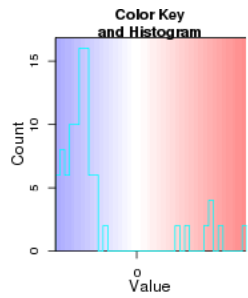


Oncogene enrichment:





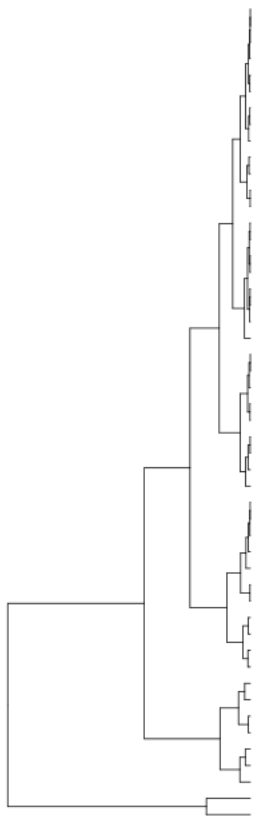
Oncogene zscore:



Immuno enrichment:

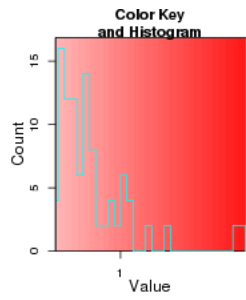
Error: no locations are finite

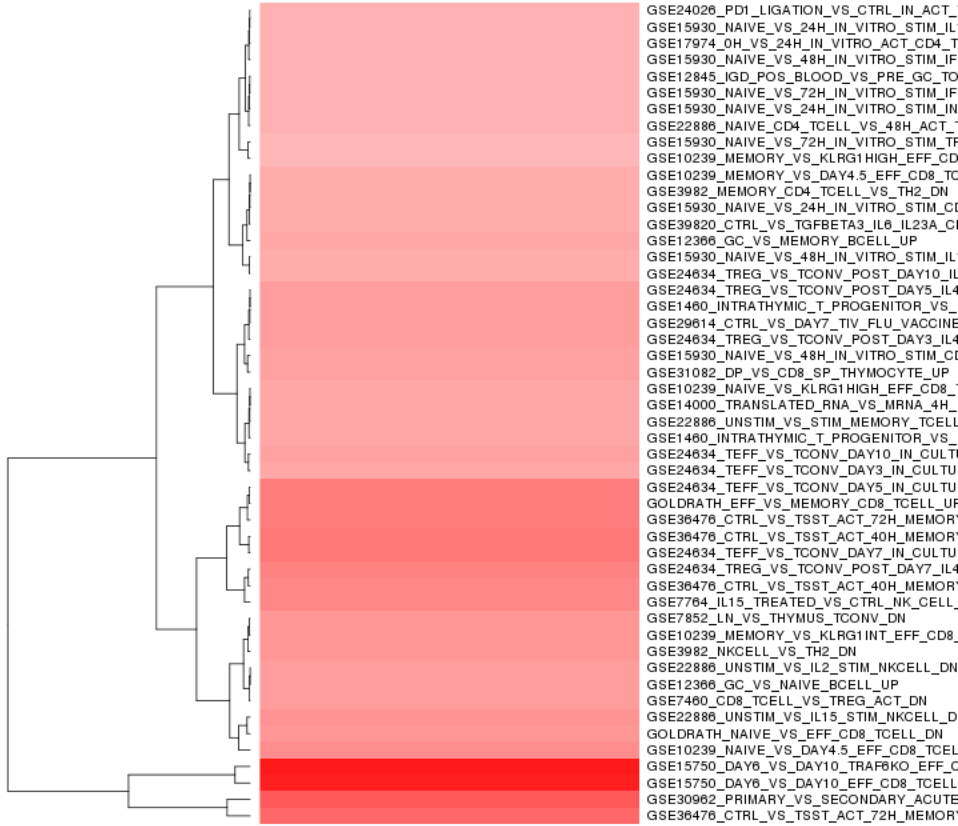




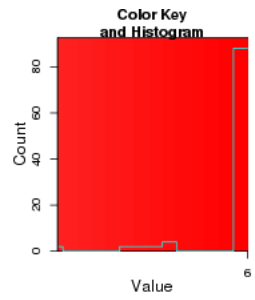
GSE30962_PRIMARY_VS_SECONDARY_CHRO
GSE9050_GP33_VS_GP276_LCMV_SPECIFIC_I
GSE15767_MED_VS_SCS_MAC_LN_UP
GSE24634_TEFF_VS_TCONV_DAY3_IN_CULTU
GSE10239_MEMORY_VS_KLRG1HIGH_EFF_CD
GSE12845_IGD_POS_BLOOD_VS_PRE_GC_TO
GSE19825_CD24LOW_VS_IL2RA_HIGH_DAY3_
GSE31082_DP_VS_CD4_SP_THYMOCYTE_UP
GSE27786_LIN_NEG_VS_MONO_MAC_UP
GSE10325_LUPUS_BCELL_VS_LUPUS_MYELO
GSE22886_NAIVE_BCELL_VS_MONOCYTE_DN
GSE31082_DN_VS_CD4_SP_THYMOCYTE_UP
GSE24634_TEFF_VS_TCONV_DAY10_IN_CULTI
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IN
GOLDRATH_EFF_VS_MEMORY_CD8_TCELL_UF
GSE10325_BCELL_VS_MYELOID_DN
GSE10325_LUPUS_CD4_TCELL_VS_LUPUS_M'
GSE24026_PD1_LIGATION_VS_CTRL_IN_ACT_
GSE24634_TREG_VS_TCONV_POST_DAY10_IL
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_IL
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IL
GSE12366_GC_VS_NAIVE_BCELL_UP
GSE24634_TREG_VS_TCONV_POST_DAY7_IL4
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_IF
GSE1480_INTRATHYMIC_T_PROGENITOR_VS_
GSE31082_DP_VS_CD8_SP_THYMOCYTE_UP
GSE15930_NAIVE_VS_24H_IN_VITRO_STIM_CI
GSE10239_NAIVE_VS_DAY4_5_EFF_CD8_TCEL
GSE1480_INTRATHYMIC_T_PROGENITOR_VS_
GSE22886_UNSTIM_VS_STIM_MEMORY_TCELL
GSE10239_MEMORY_VS_KLRG1INT_EFF_CD8
GSE24634_TREG_VS_TCONV_POST_DAY3_IL4
GSE24634_TEFF_VS_TCONV_DAY7_IN_CULTU
GSE7460_CD8_TCELL_VS_TREG_ACT_DN
GSE15930_NAIVE_VS_48H_IN_VITRO_STIM_CI
GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR
GSE39820_CTRL_VS_TGFBETA3_IL6_IL23A_CI
GSE24634_TREG_VS_TCONV_POST_DAYS_IL4
GSE10239_MEMORY_VS_DAY4_5_EFF_CD8_TC
GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR
GSE22886_UNSTIM_VS_IL2_STIM_NKCELL_DN
GSE7764_IL15_TREATED_VS_CTRL_NK_CELL
GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR
GSE14000_TRANSLATED_RNA_VS_MRNA_4H_
GSE30962_PRIMARY_VS_SECONDARY_ACUTE
GSE24634_TEFF_VS_TCONV_DAYS_IN_CULTU
GSE22886_UNSTIM_VS_IL15_STIM_NKCELL_D
GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR
GSE15750_DAY6_VS_DAY10_TRAFKO_EFF_C
GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL

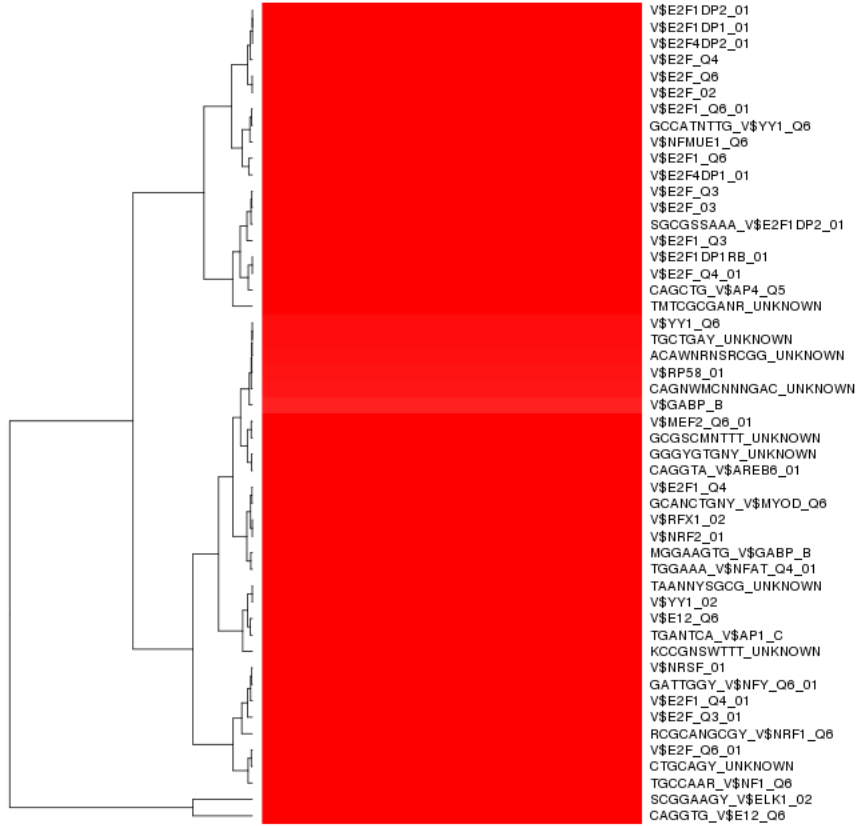
Immuno zscore:

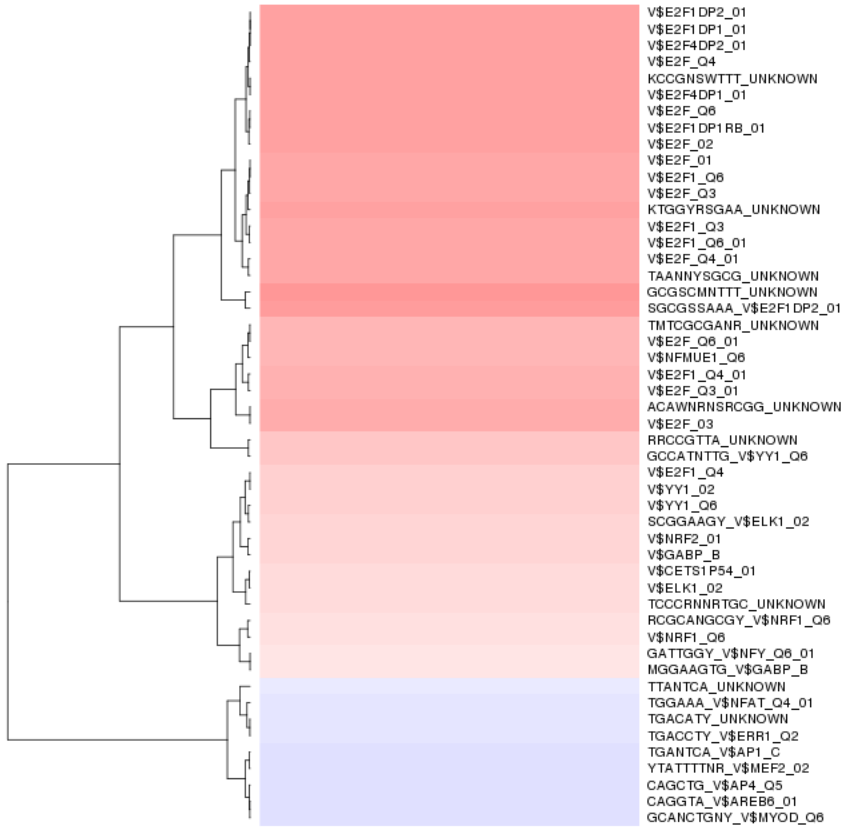
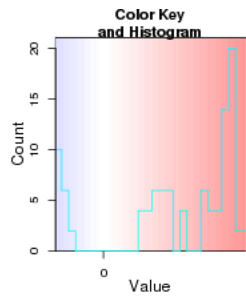




TF enrichment:



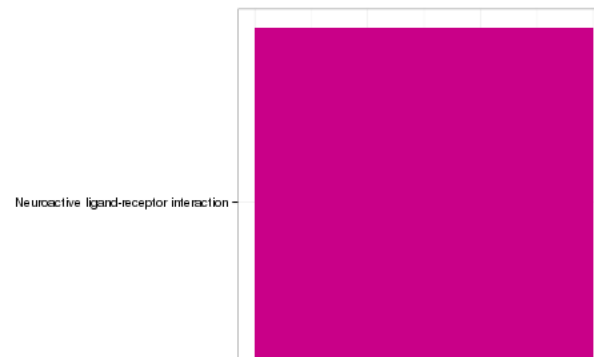
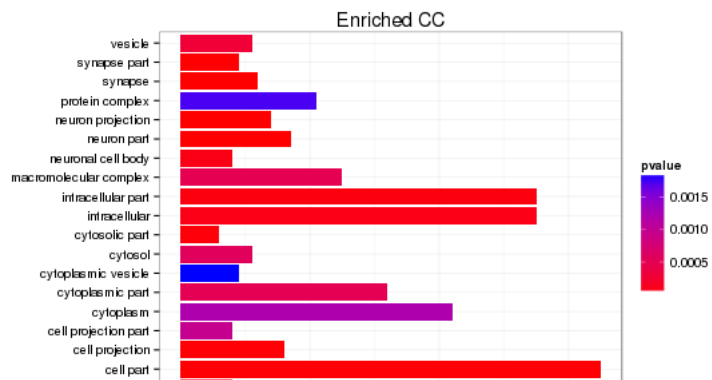
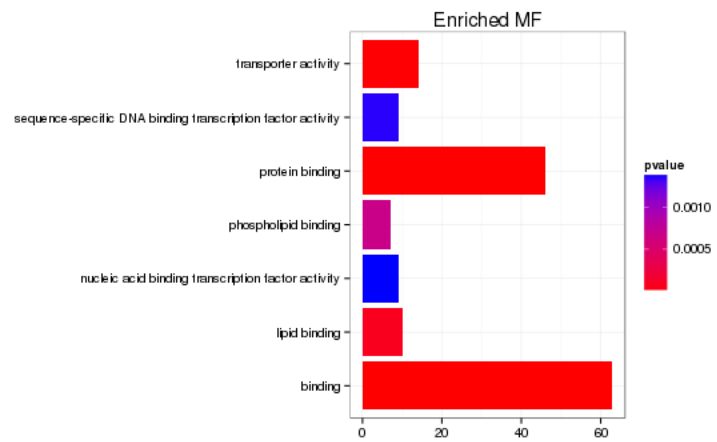
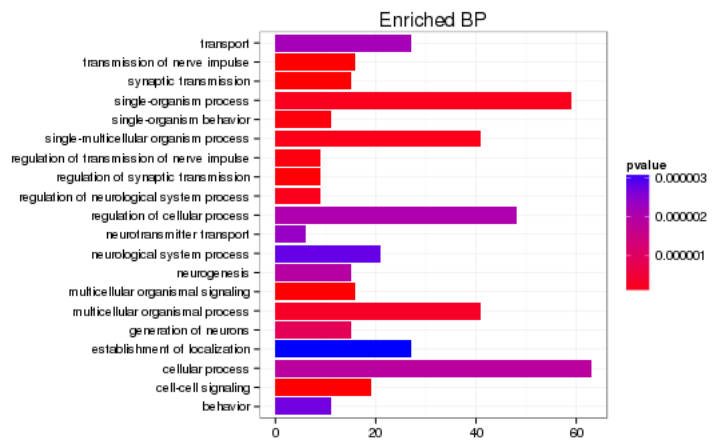


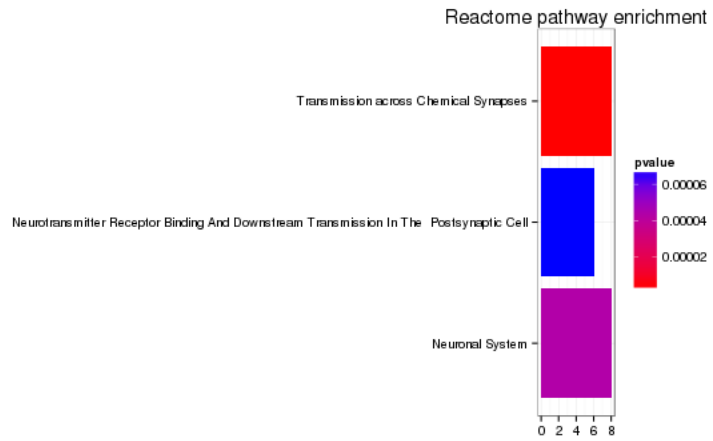
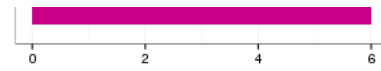


GO enrichment

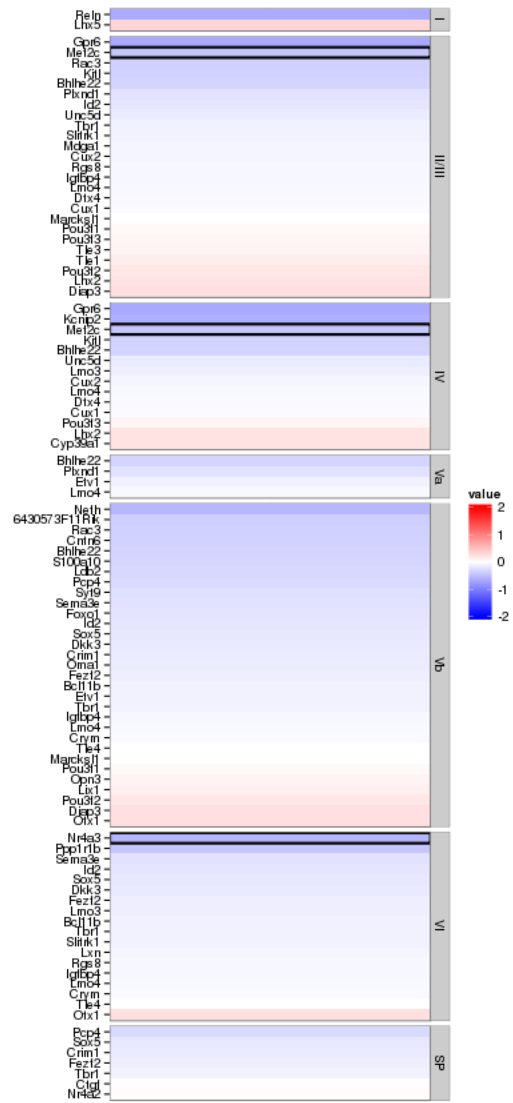
Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

```
## [1] "ERROR: The estimated pi0 <= 0. Check that you have valid p-values or use another lambda method."
```

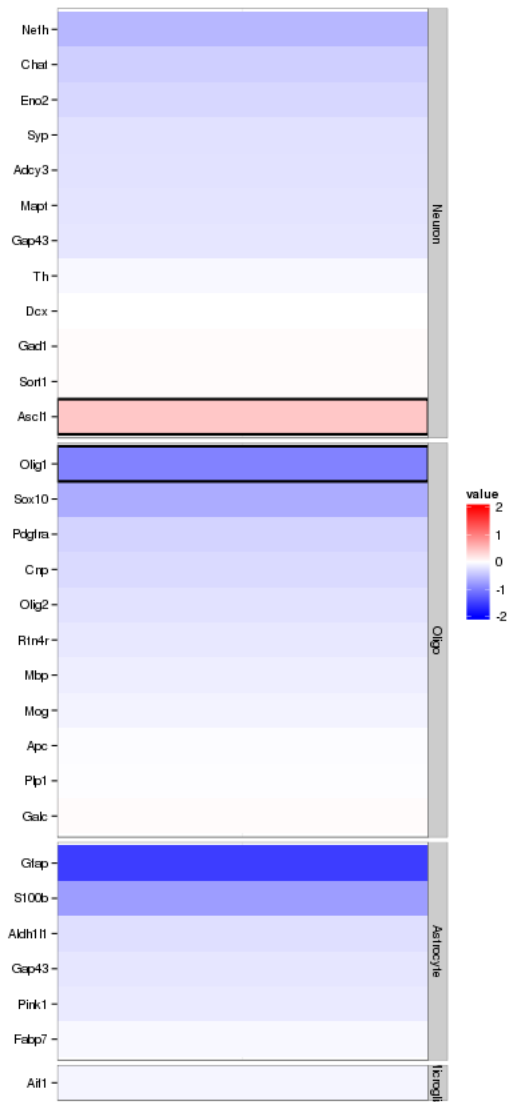




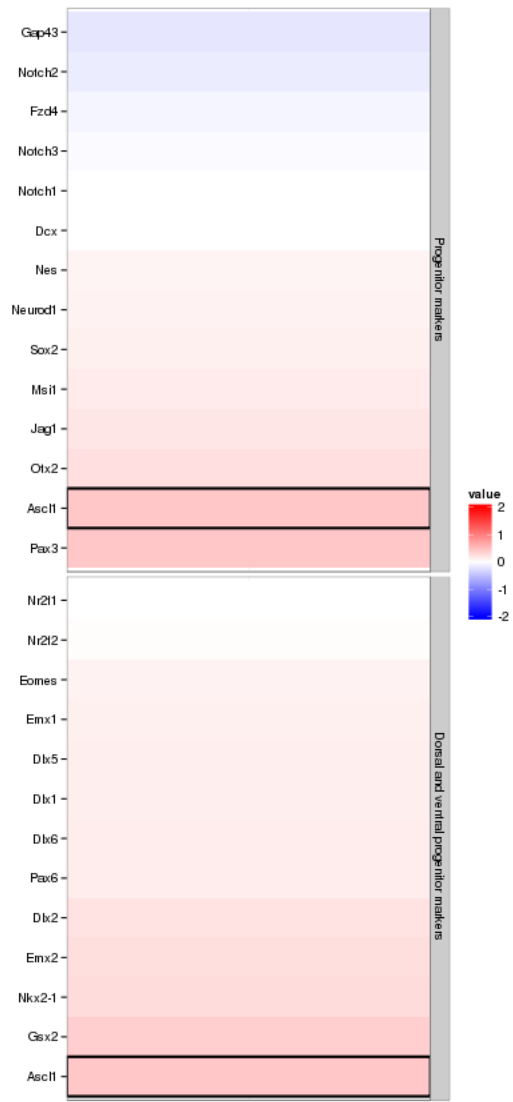
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



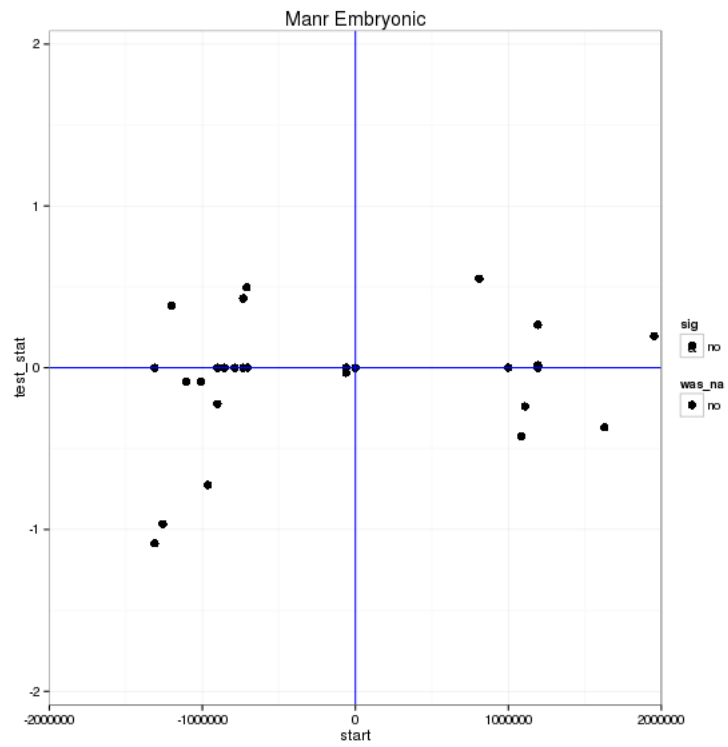
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR751
16 JR772
17 JR788

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR753/abundances.cxb WT	0	WT_0		39331900.00	31913100.00	1.28	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR750/abundances.cxb WT	1	WT_1		36031100.00	31913100.00	1.11	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR771/abundances.cxb WT	2	WT_2		38857100.00	31913100.00	1.24	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR755/abundances.cxb WT	3	WT_3		41811300.00	31913100.00	1.30	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR811/abundances.cxb WT	4	WT_4		38172800.00	31913100.00	1.21	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR768/abundances.cxb WT	5	WT_5		28375200.00	31913100.00	0.90	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR761/abundances.cxb WT	6	WT_6		31706200.00	31913100.00	0.98	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR815/abundances.cxb WT	7	WT_7		29733100.00	31913100.00	0.93	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR789/abundances.cxb WT	8	WT_8		27681500.00	31913100.00	0.87	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR748/abundances.cxb WT	9	WT_9		32367000.00	31913100.00	0.97	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR716/abundances.cxb WT	10	WT_10		26144700.00	31913100.00	0.82	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR717/abundances.cxb WT	11	WT_11		25565400.00	31913100.00	0.81	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR719/abundances.cxb WT	12	WT_12		22697800.00	31913100.00	0.69	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR756/abundances.cxb WT	13	WT_13		38948900.00	31913100.00	1.23	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR751/abundances.cxb Manr	0	Manr_0		33259800.00	31913100.00	1.03	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR772/abundances.cxb Manr	1	Manr_1		39063700.00	31913100.00	1.25	1.00
17 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR788/abundances.cxb Manr	2	Manr_2		23795600.00	31913100.00	0.74	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igrph_0.7.1         KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```


Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Manr -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Manr_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/
## 2
## 3
## 4
## 5
```

Peril KO vs WT (Adult)

This file shows the wt-v-ko comparison for Peril.

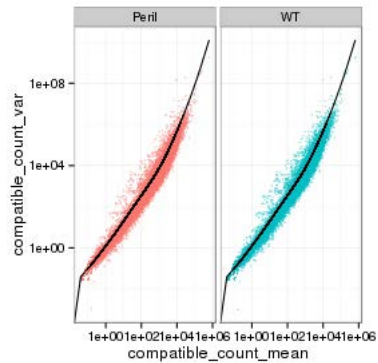
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

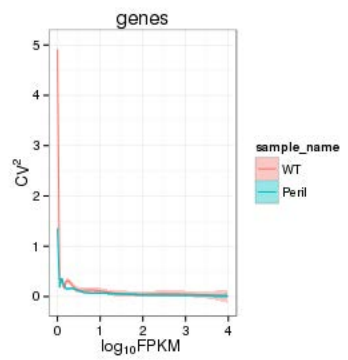
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



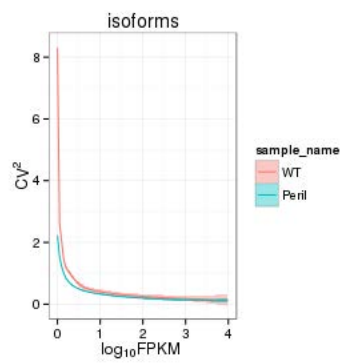
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

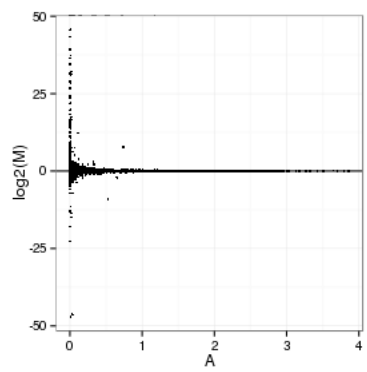
Genes:



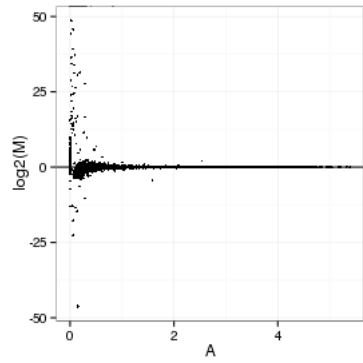
Isoforms:



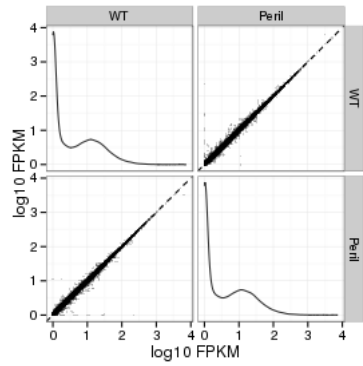
MvA plot



MvA plot counts



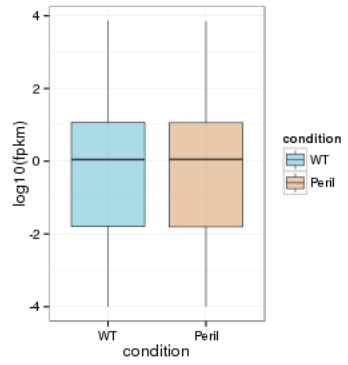
Scatterplot matrix



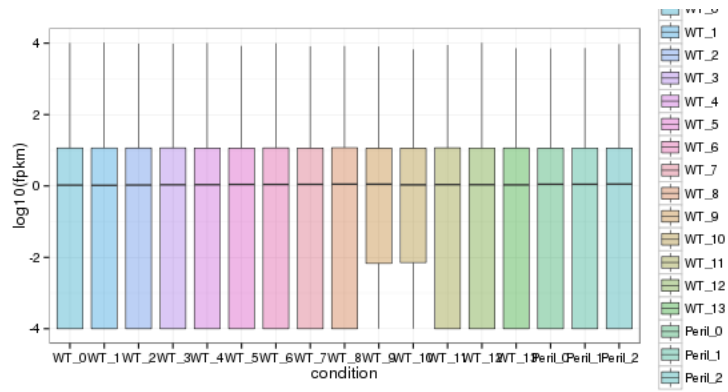
Distributions

Boxplots

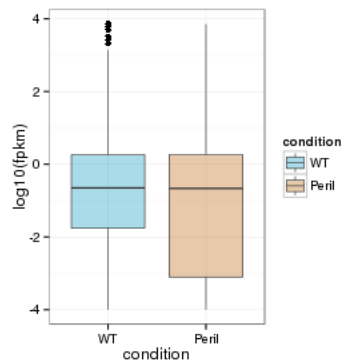
Boxplot (genes)



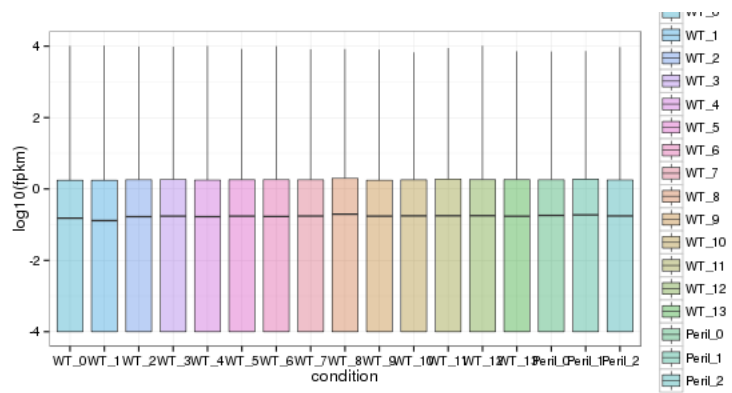
Boxplot (genes, replicates)



Boxplot (isoforms)

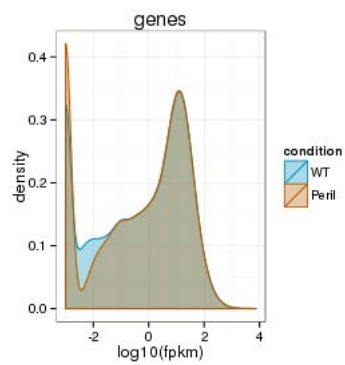


Boxplot (isoforms, replicates)

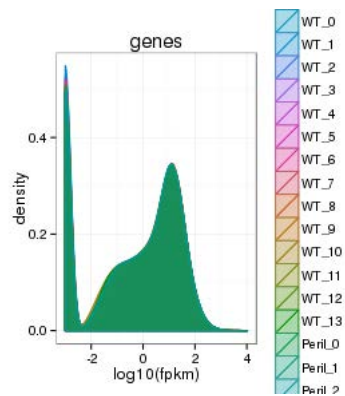


Density

Density (genes)

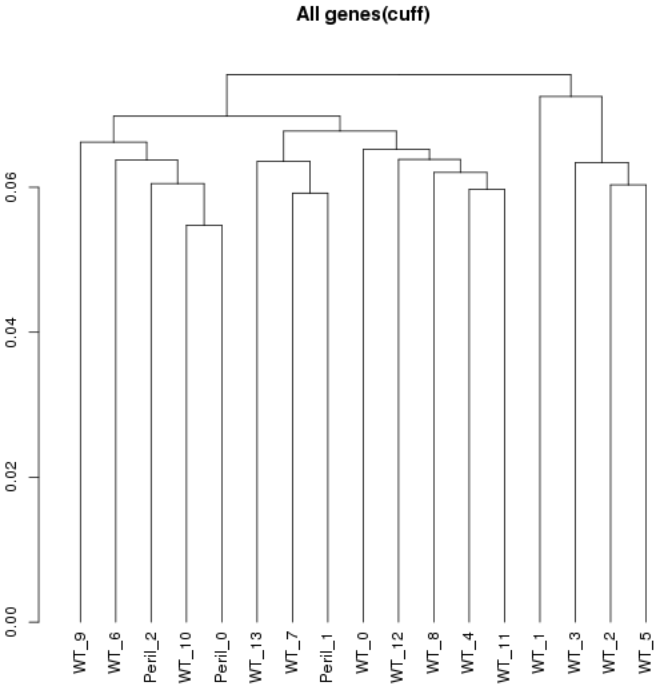


Density (genes, replicates)



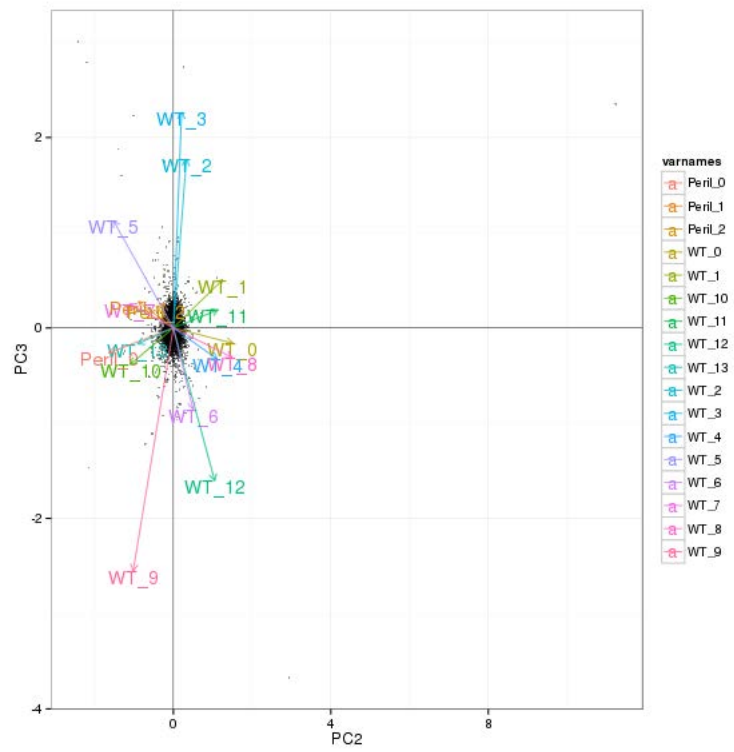
Clustering

Replicate Clusters

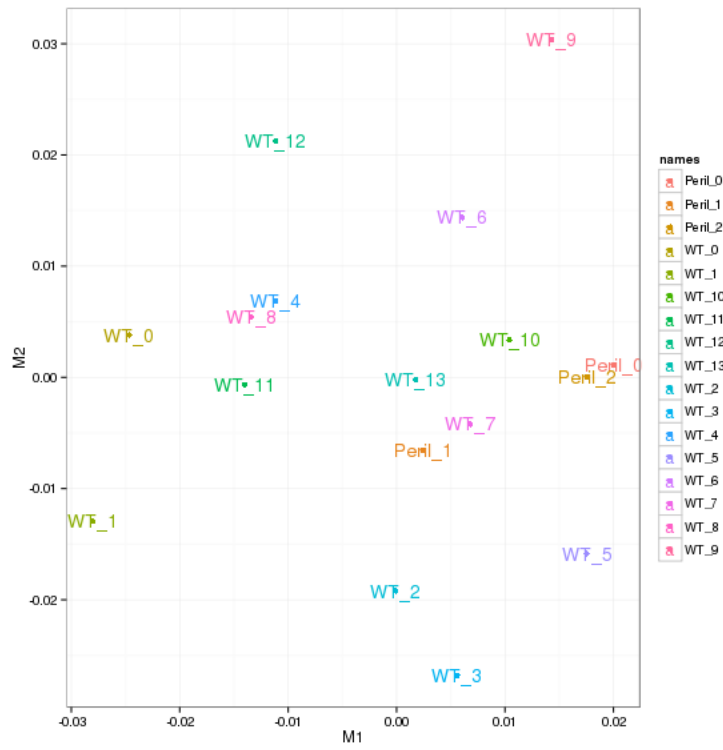


NULL

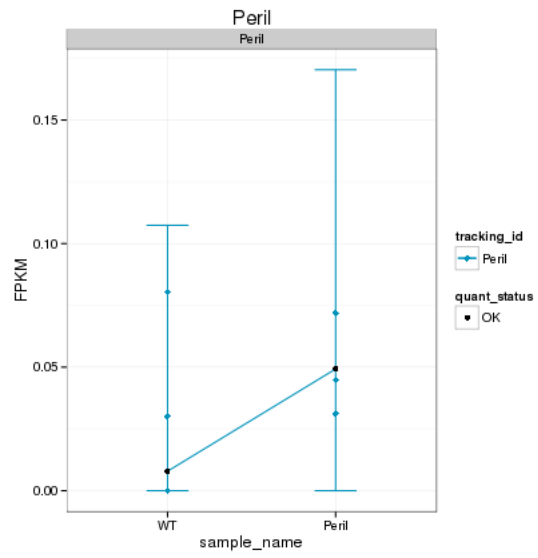
PCA (genes)



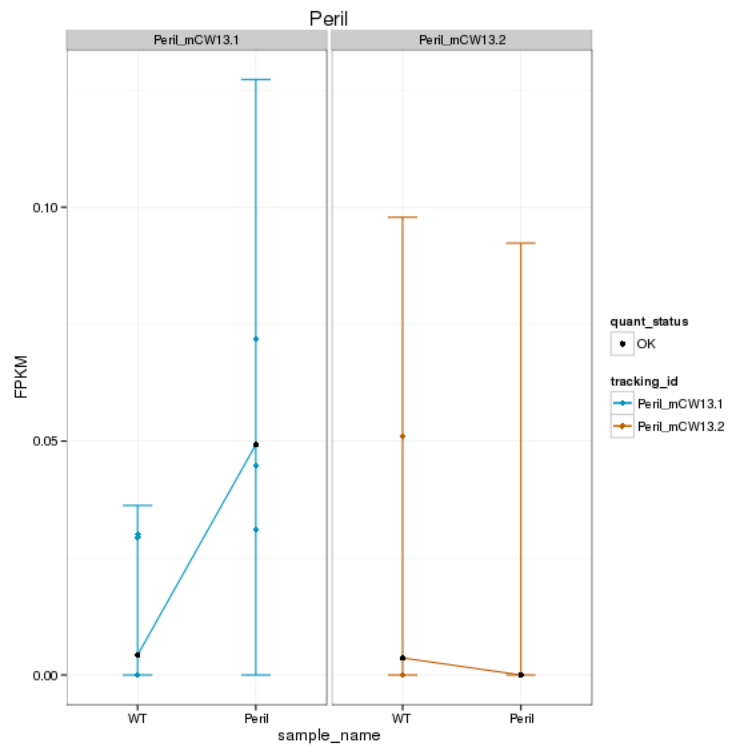
MDS (genes)



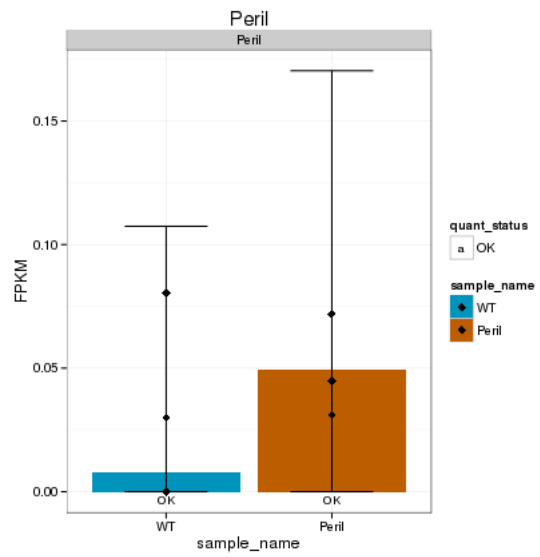
KO assessment
Endogenous IncRNA expression



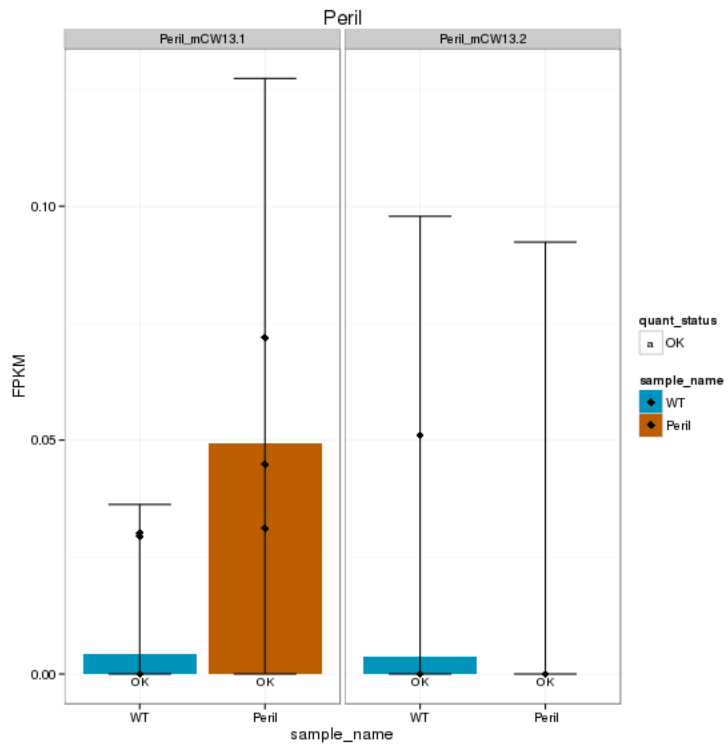
Endogenous expression of Peril isoforms:



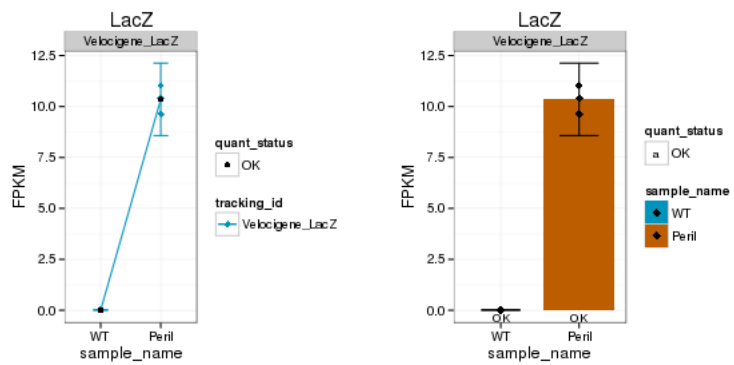
Barplot of gene expression:



Barplot of isoform expression:

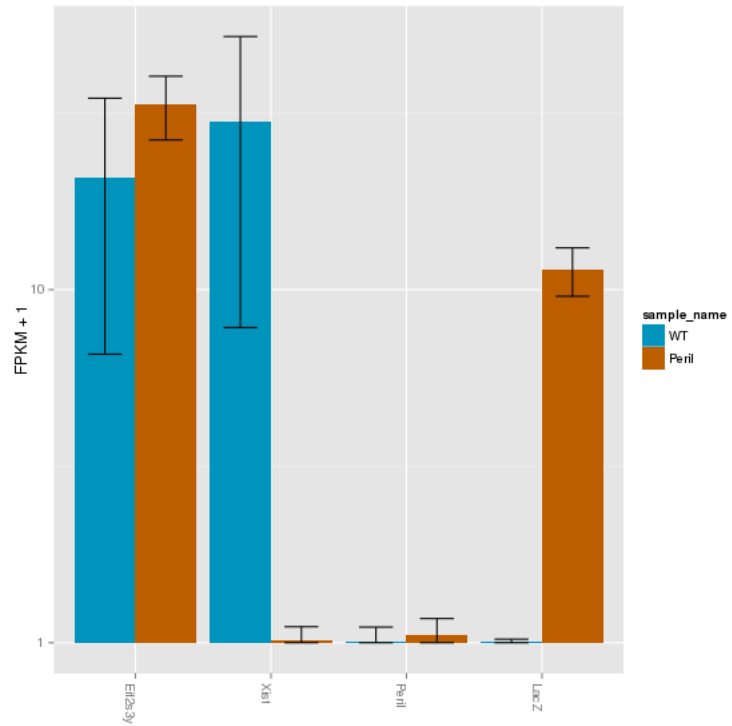


LacZ expression

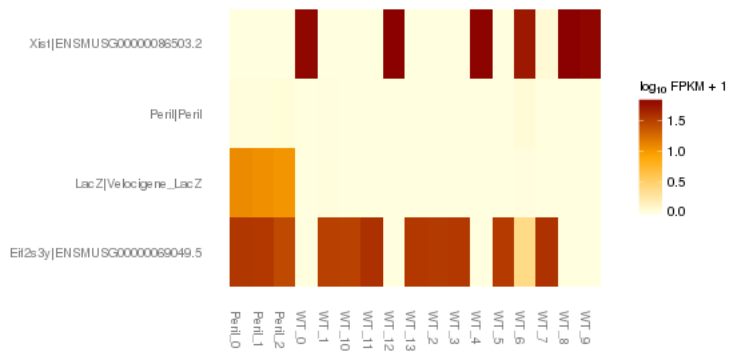


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



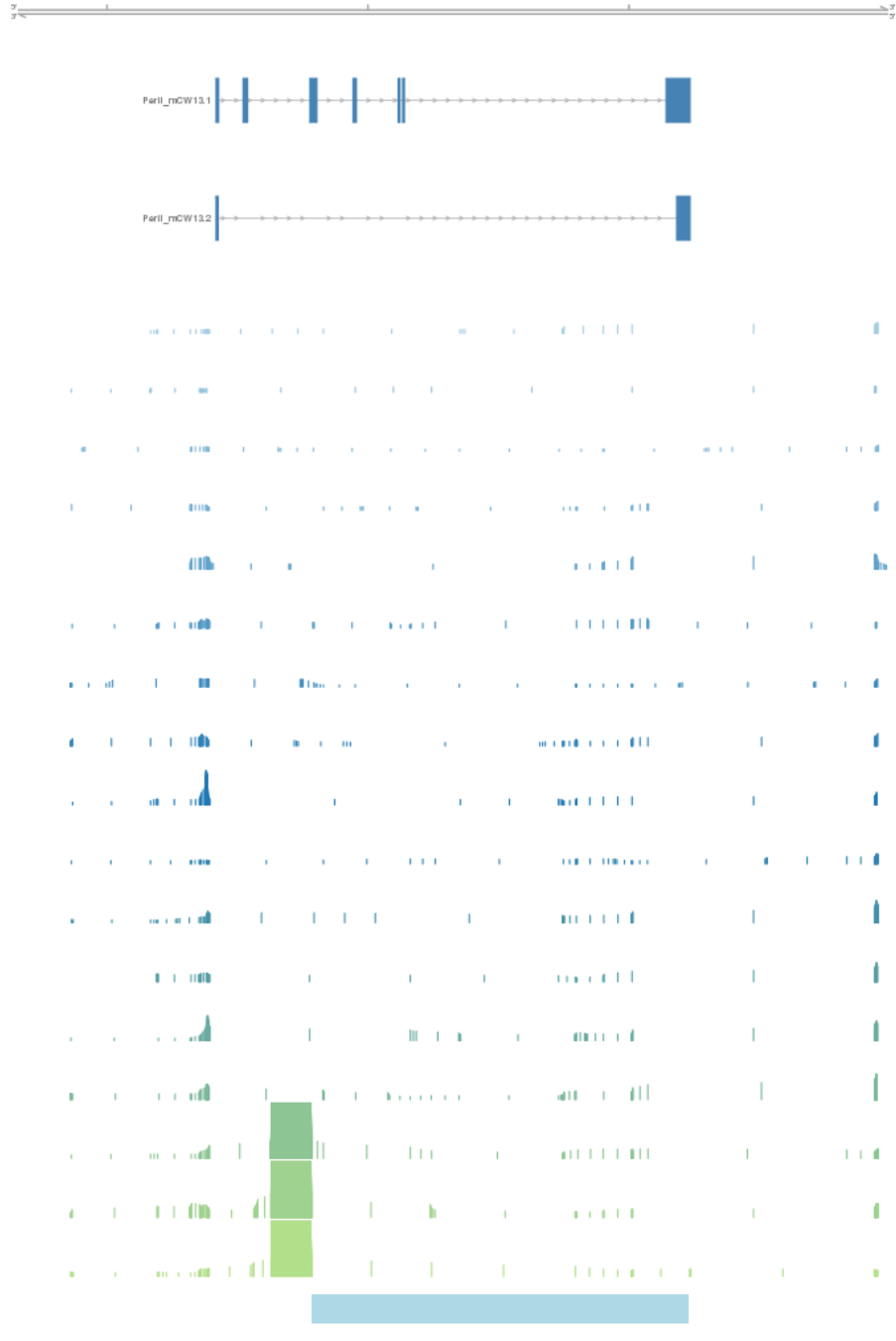
Expression heatmap:



Track visualization



KO Region Per1_Q2 Per1_Q3 Per1_Q0 WT_Q13 WT_Q12 WT_Q11 WT_Q10 WT_Q9 WT_Q8 WT_Q7 WT_Q6 WT_Q5 WT_Q4 WT_Q3 WT_Q2 WT_Q1 WT_Q0 LncRNA Isoforms



Differential Analysis

Differential Genes

There are 215 significantly differentially expressed genes. They are:

	geneAnnot\$gene_short_name
1	Wnt9a
2	Gabra2
3	Sox9
4	Mmp14
5	Chordc1
6	Plin4
7	Arrdc2
8	Klf4
9	Calr
10	Hif3a
11	9330159F19Rik
12	Nes
13	Epha2
14	Lrrc8a
15	Homer1
16	Bcl2l1
17	Dio2
18	Mertk
19	Camk1g
20	Sdc4
21	Ada
22	Etnppl
23	P4ha1
24	Arid5b
25	Sgk1
26	Gm872
27	Ndufa12
28	Timp3
29	Pttg1
30	Btg2
31	Gatsl3
32	Xbp1
33	Pdia6
34	Fam20a
35	Adi1
36	Camkk1
37	Per1

38 Nfkbia
39 Serpina3n
40 Zfp361l
41 Fos
42 Pcsk1
43 Fam107a

44 Dnajc3
45 Fzd6
46 Myh9
47 Slc38a2
48 Bcl6
49 Arc
50 Sdf2l1
51 Arhgap31
52 Ifnar1
53 Cdkn1a
54 Creld2
55 Tnfrsf12a
56 Xdh
57 Tekt4
58 Dusp1
59 Fkbp5
60 Spry4
61 Pdgfrb
62 Atp10a
63 Kdm5c
64 D3Ert751e
65 Pdia4
66 Paqr8
67 Slc40a1
68 Pfkfb3
69 Lcn2
70 Hspa5

71 Spred1
72 Fam83d
73 Anxa5
74 Exosc9
75 Schip1
76 F3
77 Trp53inp1
78 Nr4a3
79 Slc2a1
80 Mfsd2a

81 Tinagl1
82 Errfi1
83 Fosl2
84 Aff1
85 Rasgef1b
86 Lfng
87 Hsph1
88 Klf15
89 Slc6a6
90 Bhlhe40
91 Adipor2
92 Emp1
93 Pglyrp1
94 Rhpn2
95 Hdcd3
96 Sult1a1
97 Lyve1
98 Polr3e
99 Tsc22d3
100 Dusp4
101 Plat
102 Galnt7
103 Gpt2
104 Mt2
105 Herpud1
106 Ldlr
107 Eomes
108 Slc16a1
109 Tbc1d4
110 Galnt9
111 Egr3
112 Tlr13
113 Klf9
114 Trim36
115 Trim59
116 Pla2g3
117 Prr7
118 Nostrin
119 Dusp5
120 Arl4d
121 Lars2
122 Adrb1
123 Dnajb5

124 Micall2
125 Anln
126 Psenen
127 Mxd4
128 Tob1
129 Avp
130 Smim3
131 Bend3
132 Txnip
133 Irs2
134 Eltd1
135 Pcsk1n
136 Tnfsf10
137 C1qtnf4
138 Rps19
139 Cables1
140 Nkx6-2
141 Mup4
142 Cldn5
143 Pkp2
144 Sgsm1
145 Jhdm1d
146 Maff
147 Id1
148 Tuba1c
149 Gjc2
150 1700048O20Rik
151 Nxpe4
152 Ctla2a
153 Cirbp

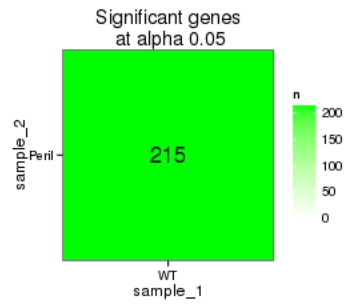
154 Zfp36l2
155 Npas4
156 Plaur
157 Fat4
158 Marcks11
159 Stbd1
160 Hes5
161 Bdnf
162 Mex3d
163 Dact2
164 Pdp1
165 Cdc42ep1
166 Rasd1

167 Opalin
168 Ppp1r3g
169 Plekho2
170 Islr2
171 Klf13
172 Jun
173 Junb
174 Tnfaip6
175 Ier2
176 Mat2a
177 1810037117Rik
178 A930005H10Rik
179 Mettl7a1
180 Per2
181 1810011O10Rik
182 Kdm5d
183 Mex3b
184 Tpm3-rs7
185 Rpl30
186 Gm5506
187 Sclt1
188 Alox5ap
189 Zbtb40
190 Rps7
191 Cox5b
192 Rpl34
193 Kdr
194 Aldoart2
195 mt-Co1
196 Zbtb16
197 Zfp810
198 Ddx3y
199 Jund
200 Egr4
201 Gm7292
202 Plekhf1
203 B3galt5
204 Fjx1
205 Rprm
206 Ly6a
207 Ly6c1
208 Gm11273
209 Xist

- 210 Slc5a3
- 211 A930033H14Rik
- 212 Apold1
- 213 Hspa1b
- 214 AA465934
- 215 Gm7094

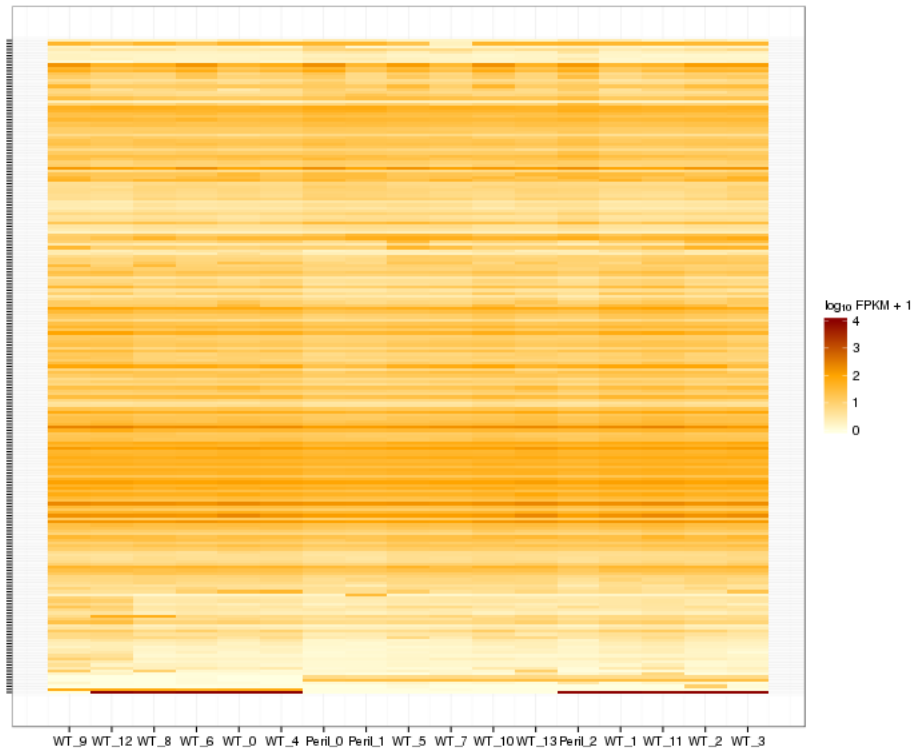
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

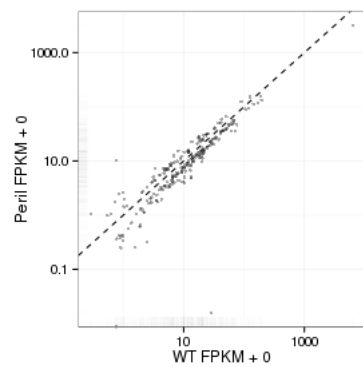


Significant genes with expression >50fpm (any condition):(turned off)

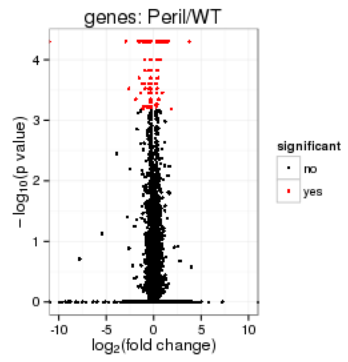
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

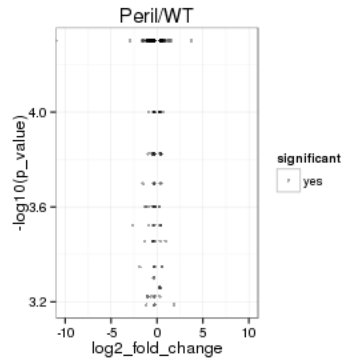
Scatter plot of significant genes only:



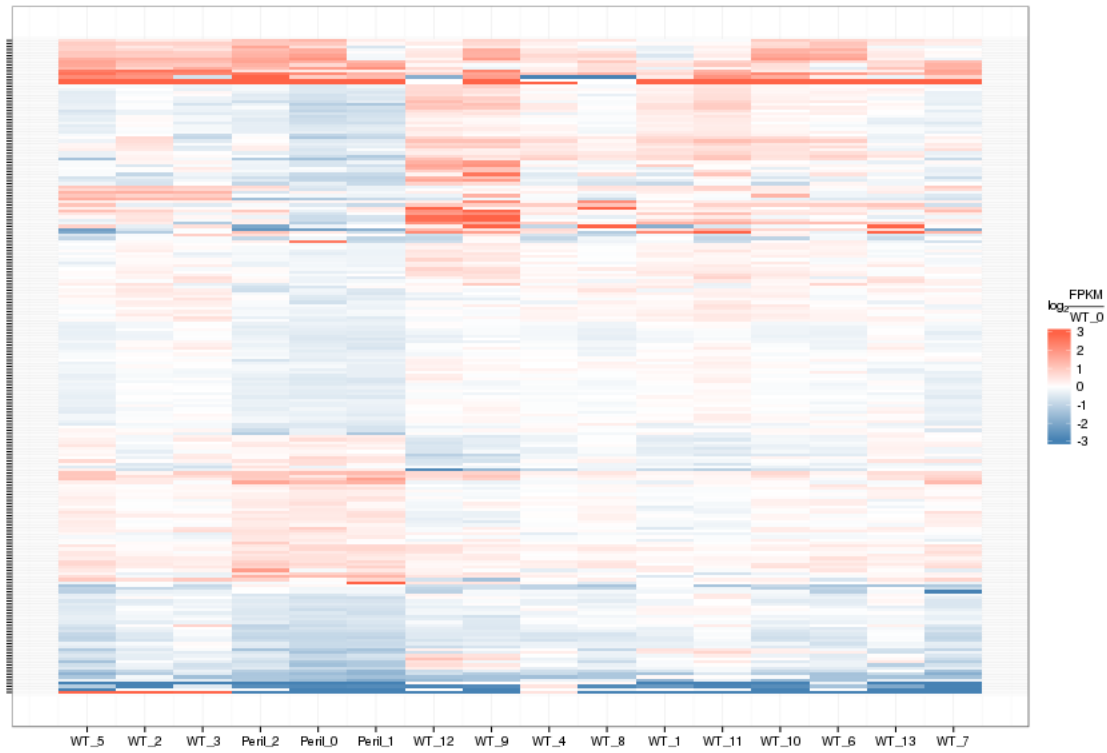
Volcano Plot



Volcano plot with significant genes only:



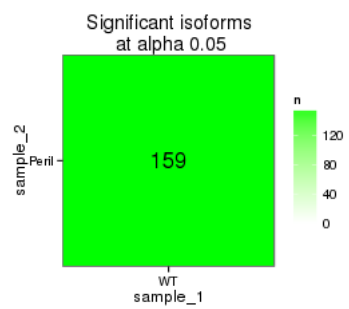
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

	isoAnnot\$gene_short_name
1	Gabra2
2	Sox9
3	Mmp14
4	Arrdc2
5	Klf4
6	Hif3a
7	9330159F19Rik
8	Nes
9	Prir
10	Epha2
11	Lrrc8a
12	Homer1
13	Dio2
14	Mertk
15	Camk1g
16	Sdc4
17	Ada
18	Arid5b
19	Sgk1
20	Gm872
21	Ntn4
22	Fgd6
23	Timp3
24	Cpm
25	Tns3
26	Btg2
27	Xbp1
28	Pdia6
29	Fam20a
30	Nfkbia
31	Serpina3n
32	Zfp36l1
33	Otub2
34	Fos
35	Pcsk1
36	Fam107a
37	Dnajc3
38	Myh9
39	Slc38a2
40	Bcl6

41 Sdf2l1
42 Arhgap31
43 Ap2m1
44 Cdkn1a
45 Tnfrsf12a
46 Xdh
47 Fkbp5
48 Pdgfrb
49 Kdm5c
50 D3Erttd751e
51 Pdia4
52 Paqr8
53 Slc40a1
54 Hspa5
55 Fam83d
56 Mccc1
57 Anxa5
58 Exosc9
59 Trpc3
60 F3
61 Slc2a1
62 Mfsd2a
63 Tinagl1
64 Map3k6

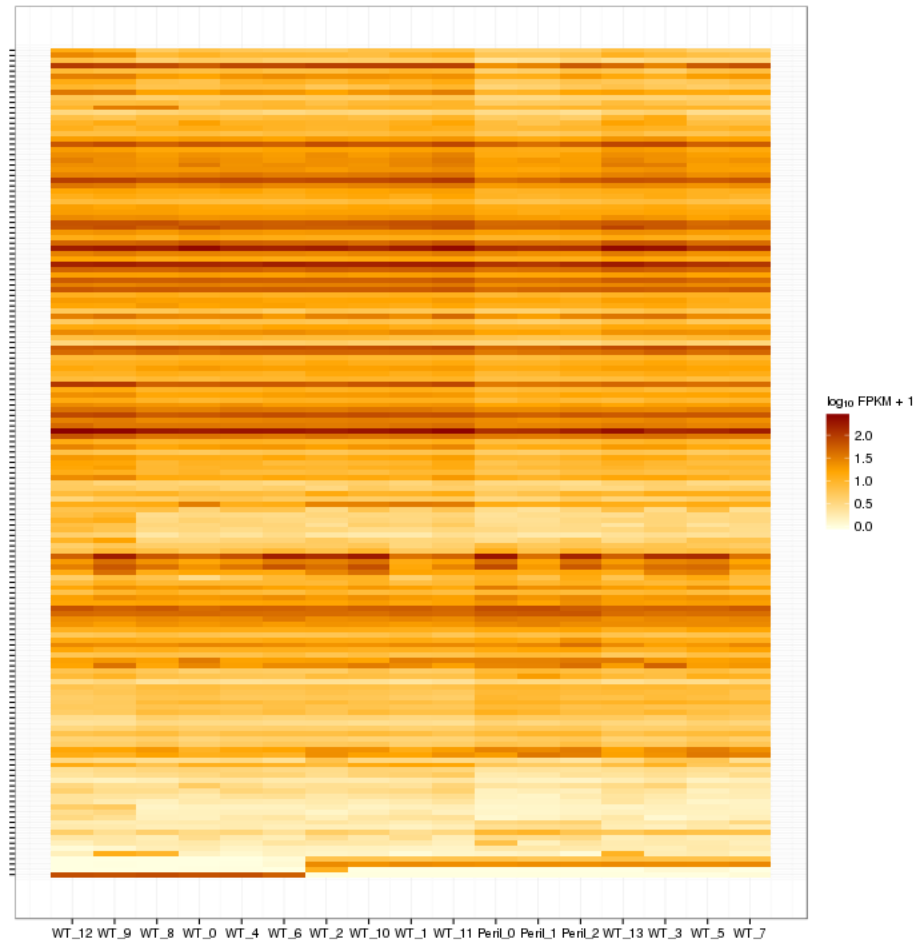
65 Errfi1
66 Fosl2
67 Lfng
68 Bhlhe40
69 Adipor2
70 Emp1
71 Pglyrp1
72 Hddc3
73 Sult1a1
74 Lyve1
75 Tsc22d3
76 Dusp4
77 Plat
78 Gpt2
79 Mt2
80 Ldlr
81 Paqr5
82 Smad3
83 Heyl

84 Slc16a1
85 Egr3
86 Klf9
87 Trim59
88 Nostrin
89 Dusp5
90 Lars2
91 Uba6
92 Dnajb5
93 Anln
94 Psenen
95 Mxd4
96 Tob1
97 Acer2
98 Smim3
99 Irs2
100 Eltd1
101 Pcsk1n
102 Tnfsf10
103 Spsb1
104 Gjb6
105 Fmo2
106 C1qtnf4
107 Nkx6-2
108 Cldn5
109 Sgsm1
110 Hps4
111 Jhdm1d
112 Id1
113 Gjc2
114 Nxpe4
115 Ctla2a
116 Cirbp
117 Zfp36l2
118 Npas4
119 Plaur

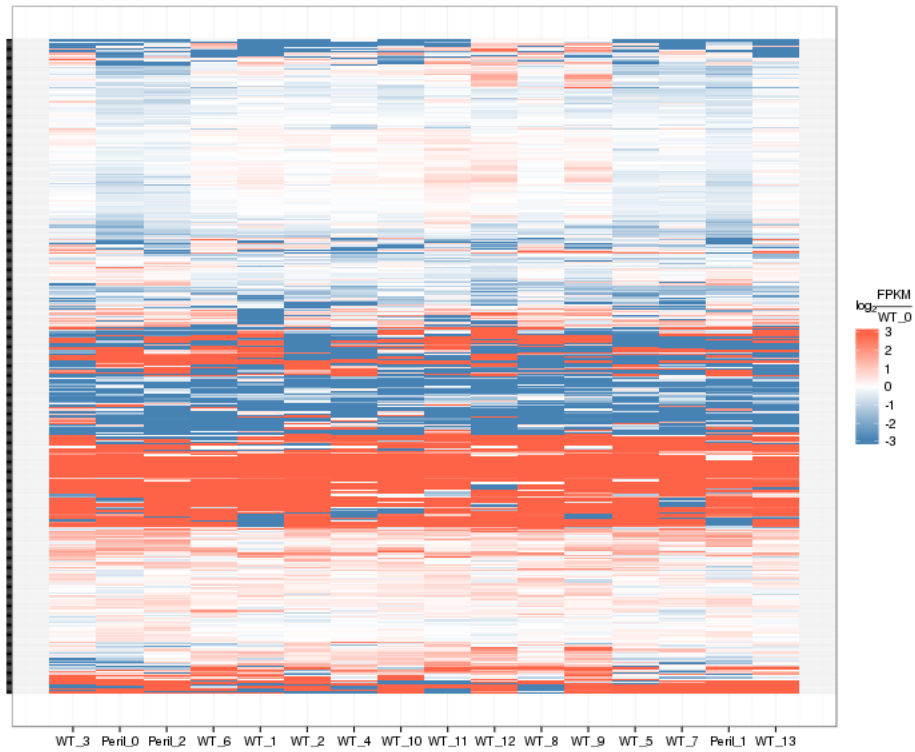
120 Stbd1
121 Hes5
122 Mex3d
123 Rasd1
124 Opalin
125 Ppp1r3g
126 Plekho2

127 Klf13
128 Junb
129 Tnfrsf6
130 Irf2
131 Mat2a
132 1810037117Rik
133 Mettl7a1
134 Per2
135 Kdm5d
136 Mex3b
137 Tpm3-rs7
138 Gm5506
139 Zbtb40
140 Rps7
141 Cox5b
142 Kdr
143 Aldoat2
144 Zbtb16
145 Ddx3y
146 Ahnak
147 Jund
148 Egr4
149 Gm7292
150 Plekhf1
151 B3galt5
152 Fjx1
153 Rpm
154 Ly6a
155 Xist
156 Slc5a3
157 A930033H14Rik
158 Apold1
159 Hspa1b

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkm}_{\text{KO}}/\text{fpkm}_{\text{WT}}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

Biocarta enrichment:

```
## [1] "Not enough significant categories to print a heatmap!"
```

Biocarta zscore:

```
## [1] "Not enough significant categories to print a heatmap!"
```

Reactome enrichment:

```
## Error: argument is of length zero
```

Reactome zscore:

```
## Error: incorrect number of dimensions
```

```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Kegg enrichment:

```
## Error: argument is of length zero
```

Kegg zscore:

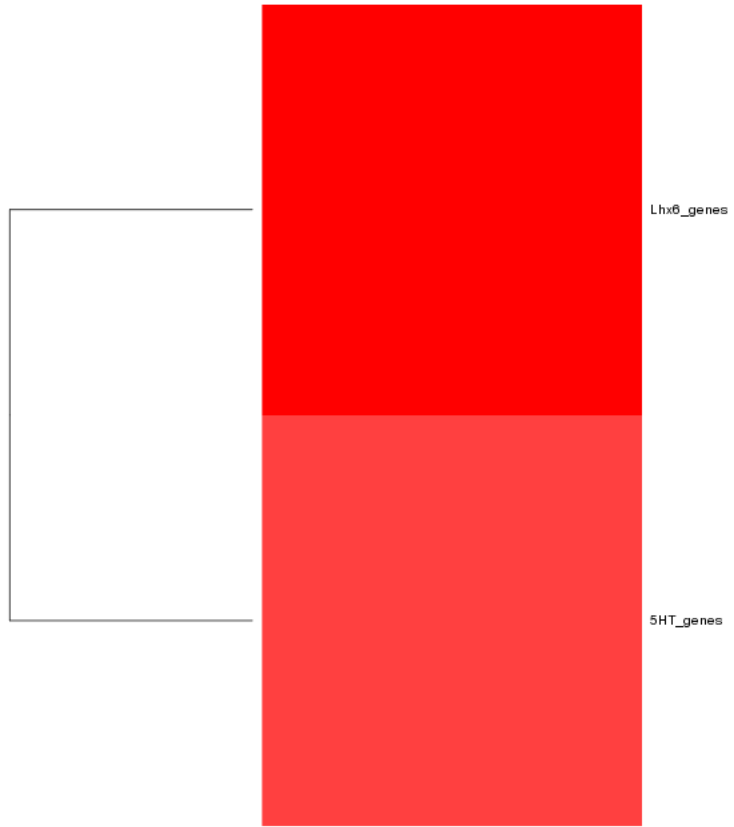
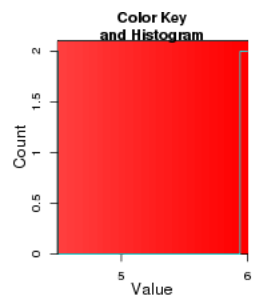
```
## Error: incorrect number of dimensions
```

```
## Error: object 'x_ordered' not found
```

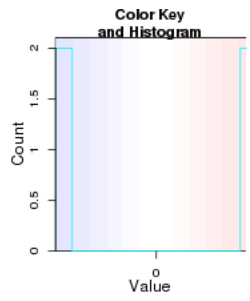
```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

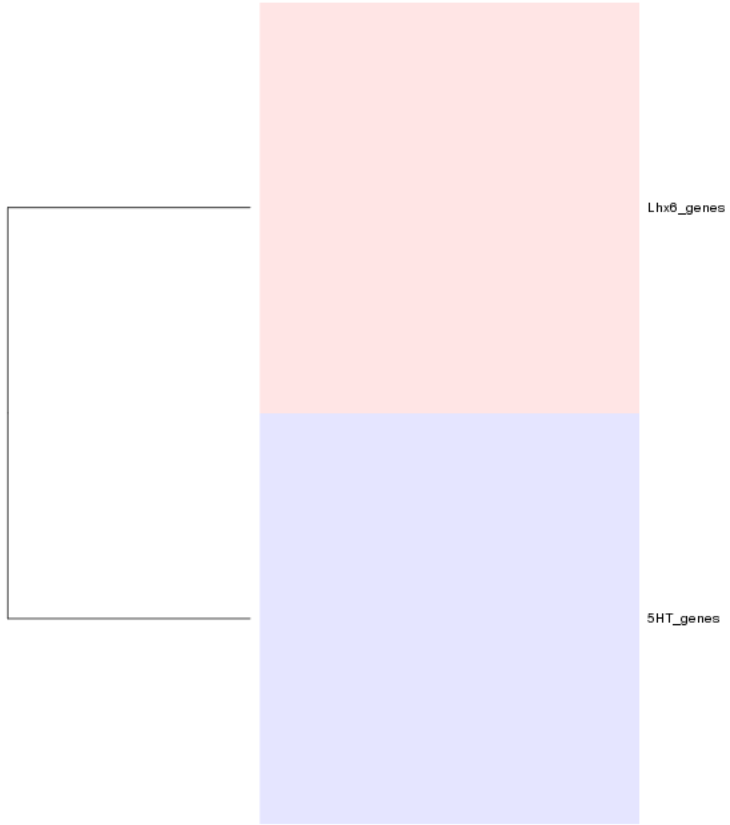
```
## Error: object 'x_ordered' not found
```

Interneuron enrichment:

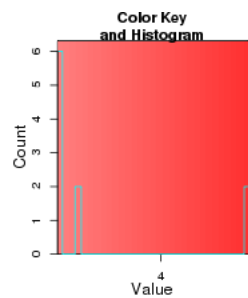


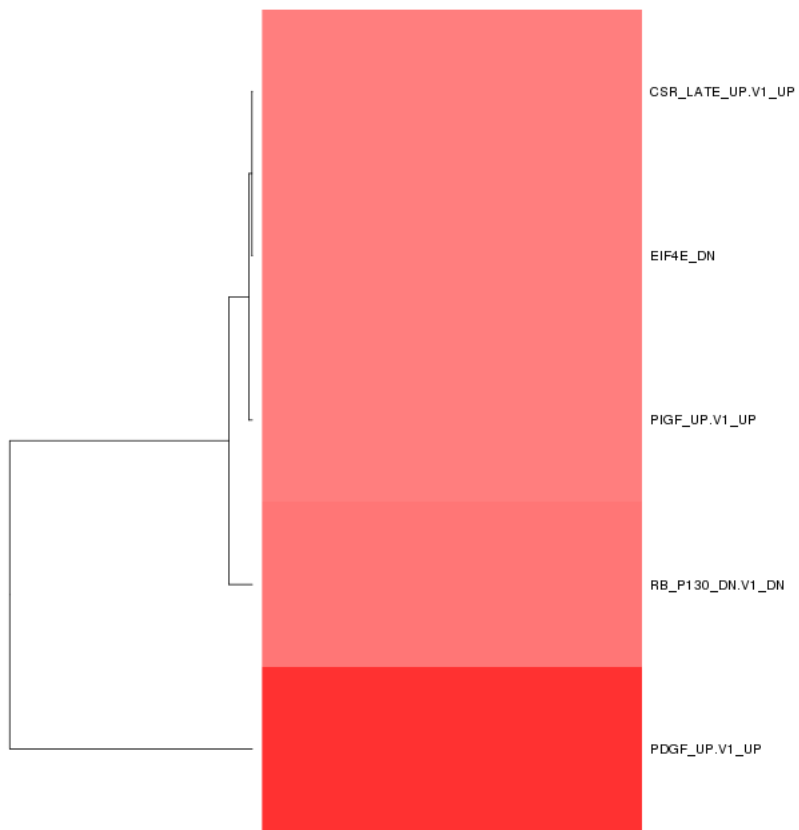
Interneuron zscore:



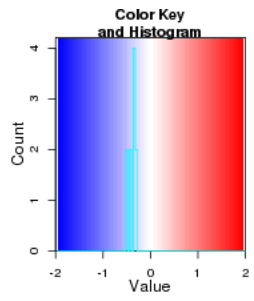


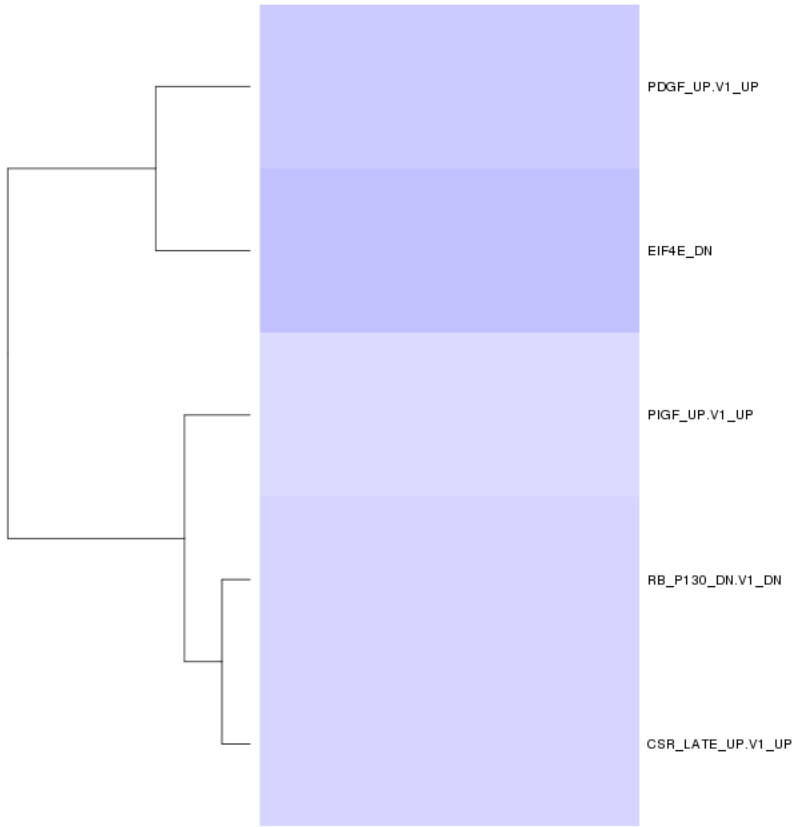
Oncogene enrichment:





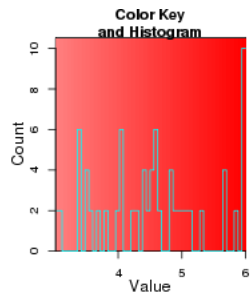
Oncogene zscore:

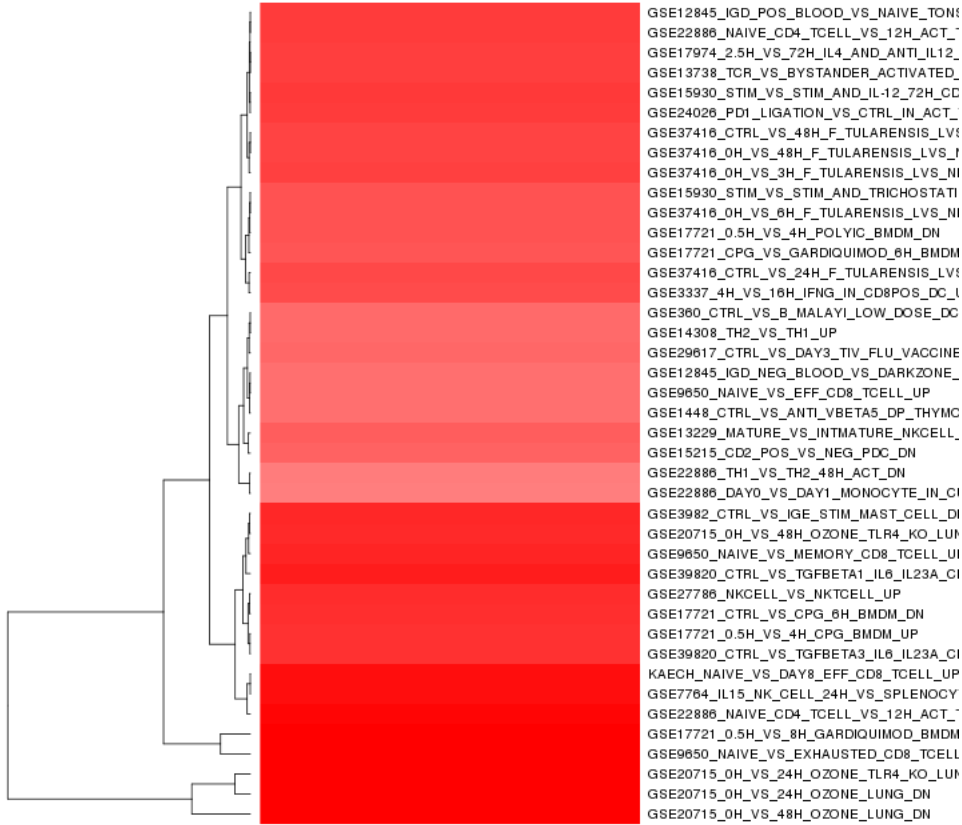




Immuno enrichment:

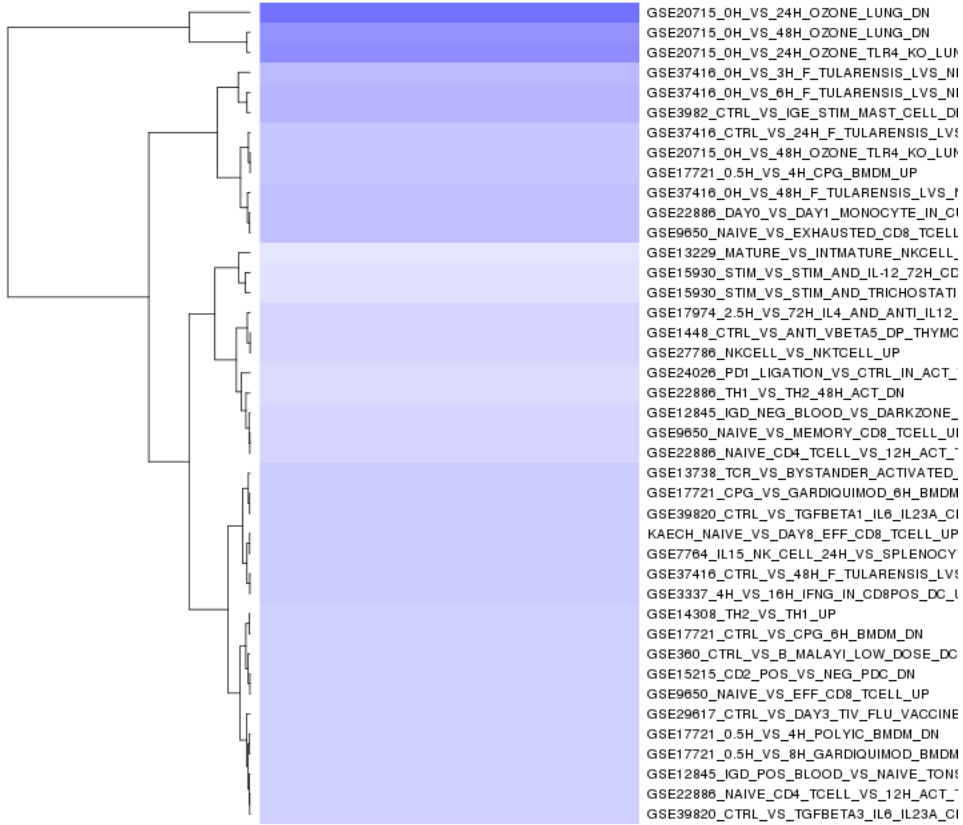
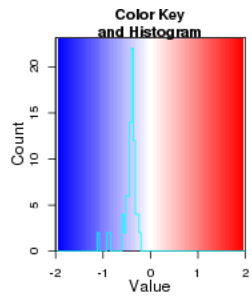
Error: subscript out of bounds

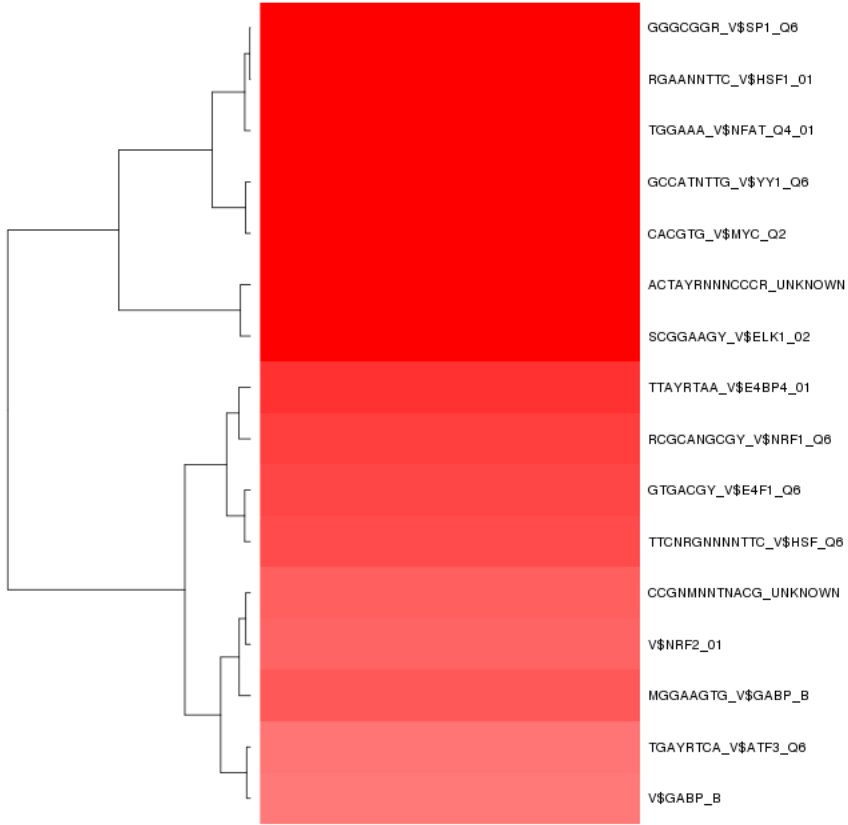
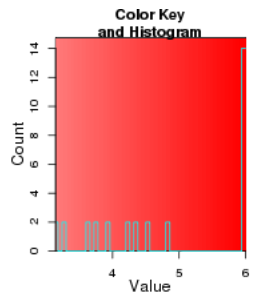




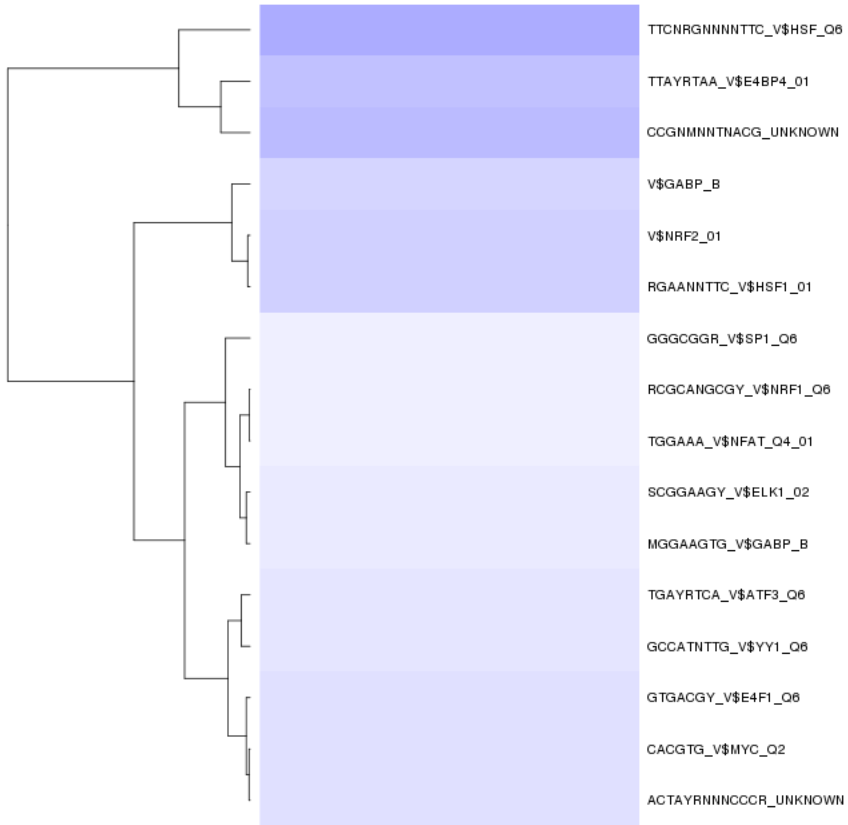
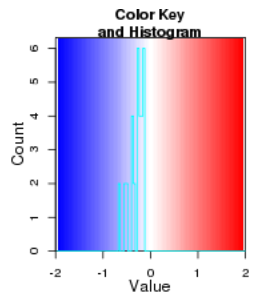
Immuno zscore:

Error: subscript out of bounds



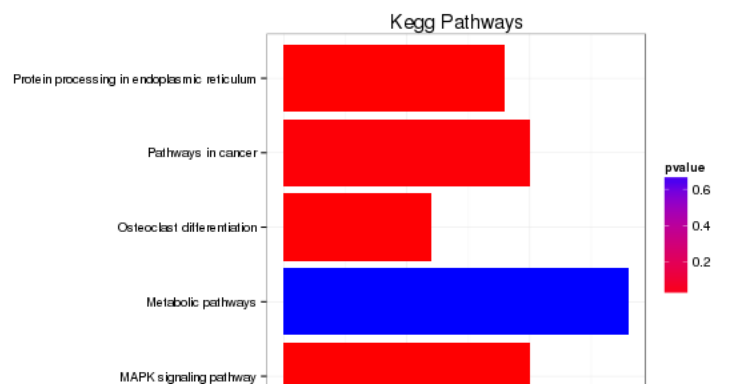
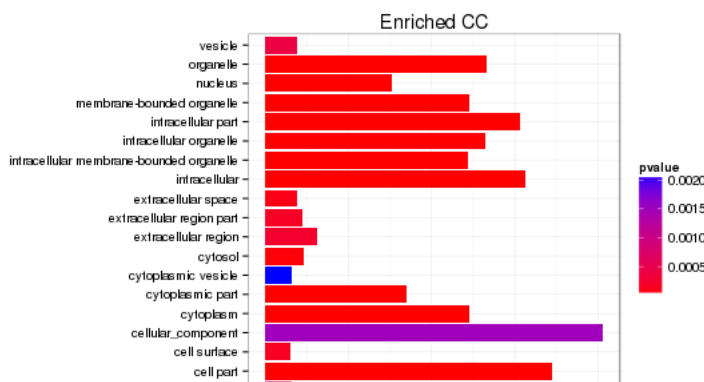
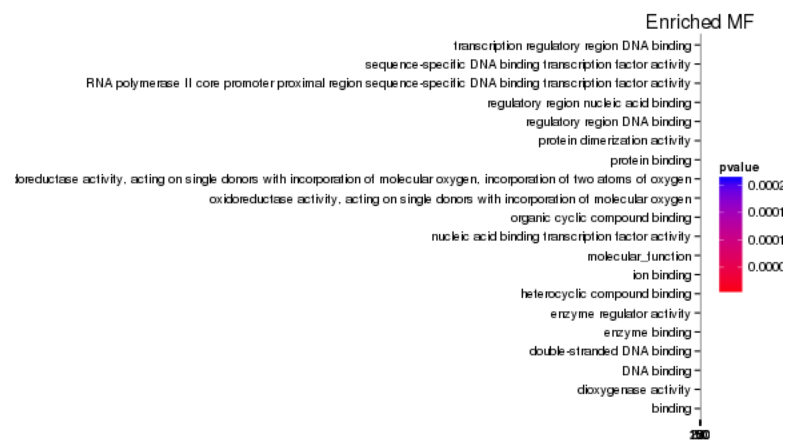
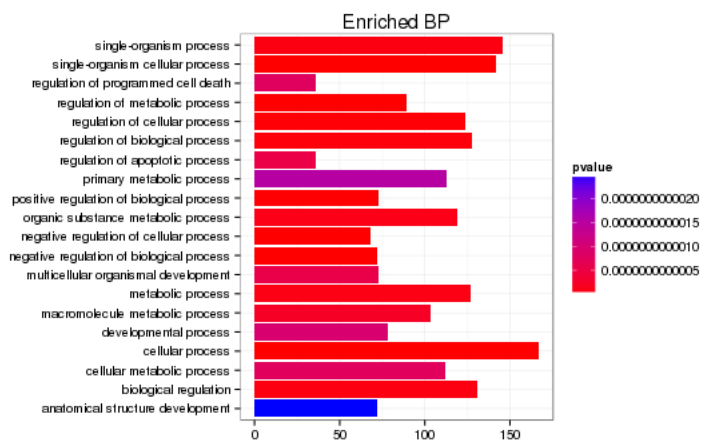


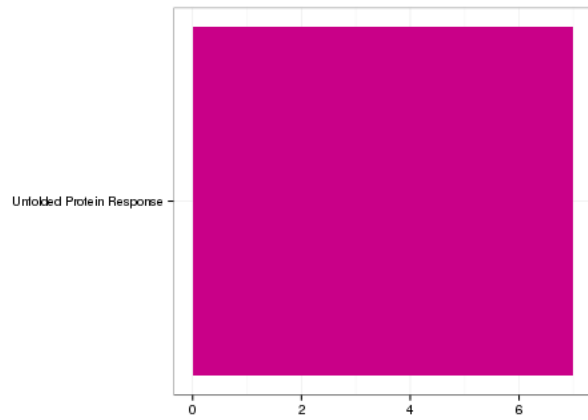
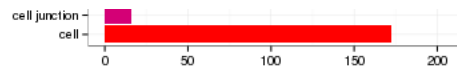
TF zscore:



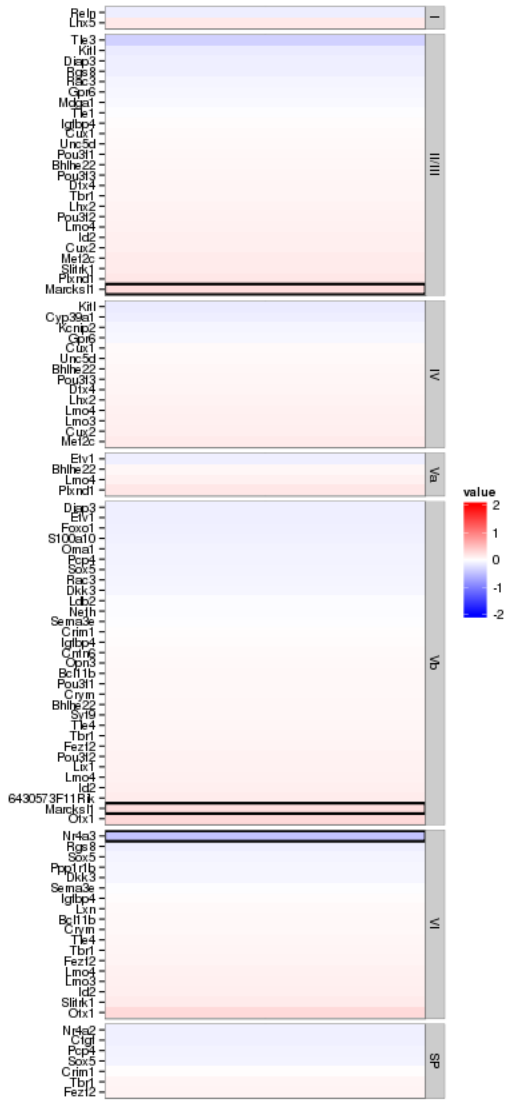
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

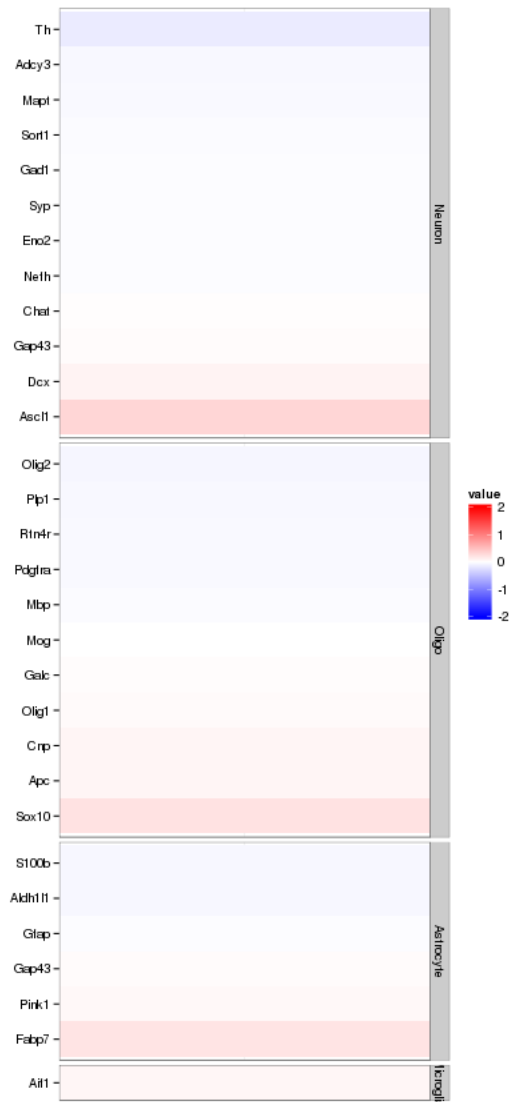




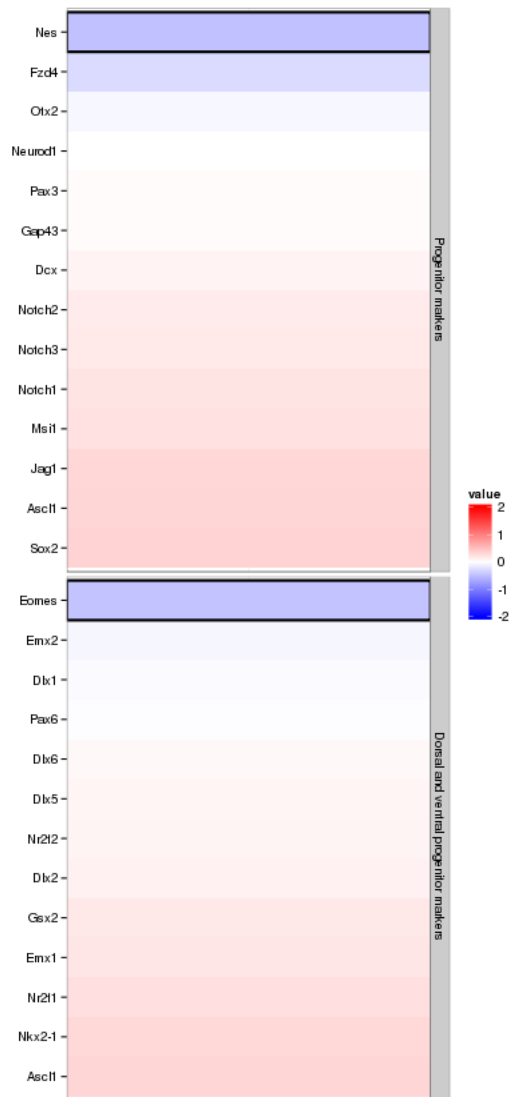
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



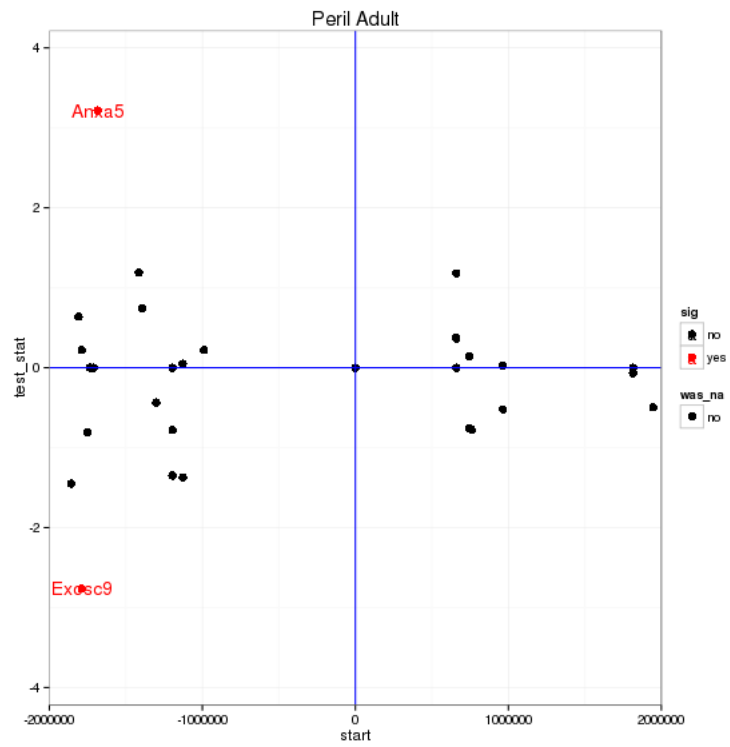
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

log2 Foldchange and test statistic are calculated with the ratio of fpkm(KO)/fpkm(WT), thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 2 genes significantly regulated in a region this size is: 0.1516



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR806
16 JR744
17 JR822

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34714900.00	0.76	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34714900.00	0.58	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34714900.00	0.98	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34714900.00	0.80	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34714900.00	1.03	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34714900.00	1.07	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34714900.00	0.80	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34714900.00	1.15	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34714900.00	1.01	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34714900.00	1.30	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34714900.00	1.50	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34714900.00	1.01	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34714900.00	0.97	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34714900.00	1.20	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR806/abundances.cxb	Peril	0	Peril_0	40528700.00	34714900.00	1.17	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR744/abundances.cxb	Peril	1	Peril_1	38767500.00	34714900.00	1.13	1.00
17 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR822/abundances.cxb	Peril	2	Peril_2	33329500.00	34714900.00	0.97	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igrph_0.7.1         KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1       zlibbioc_1.8.0

```


Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Peril -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Peril_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/ar
## 2
## 3
## 4
## 5
```

Peril KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Peril.

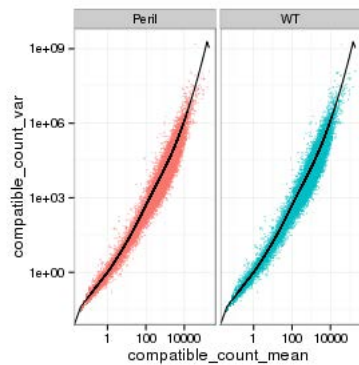
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

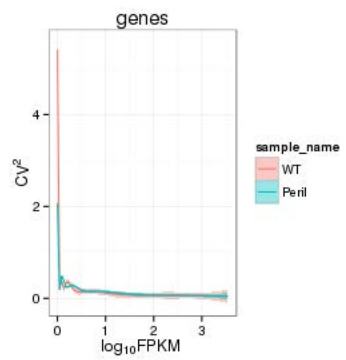
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



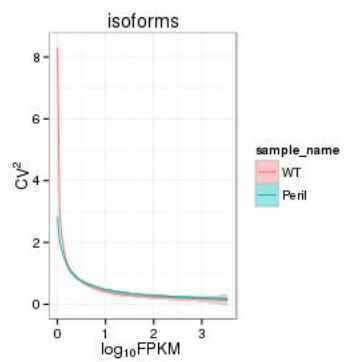
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

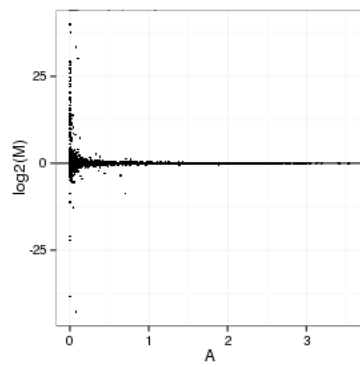
Genes:



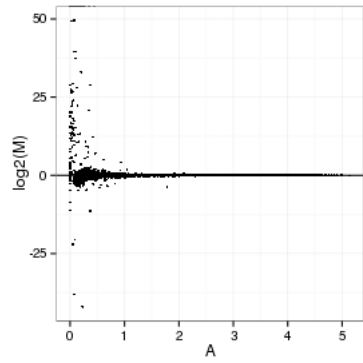
Isoforms:



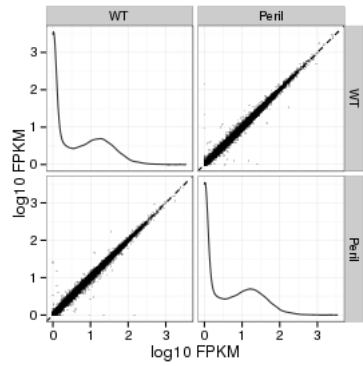
MvA plot



MvA plot counts



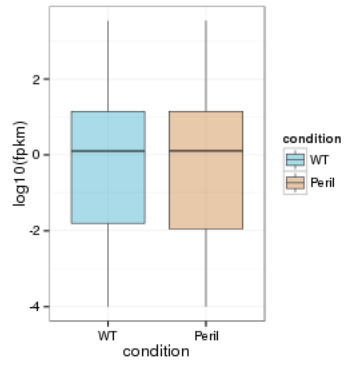
Scatterplot matrix



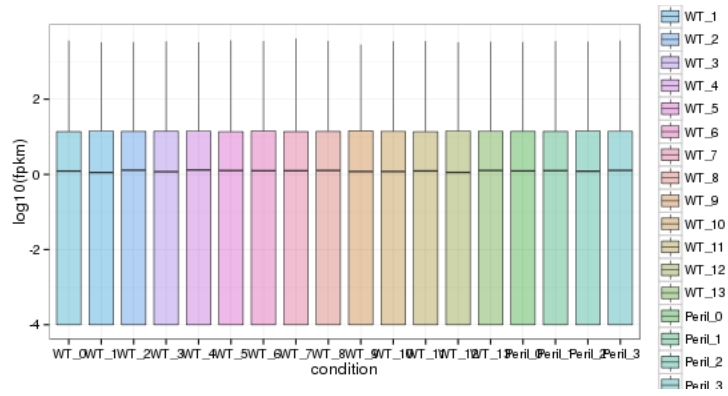
Distributions

Boxplots

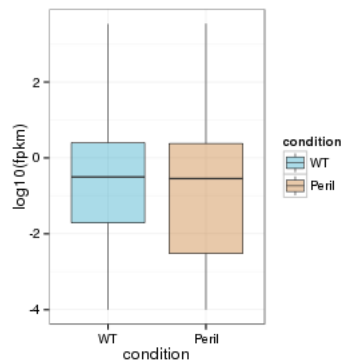
Boxplot (genes)



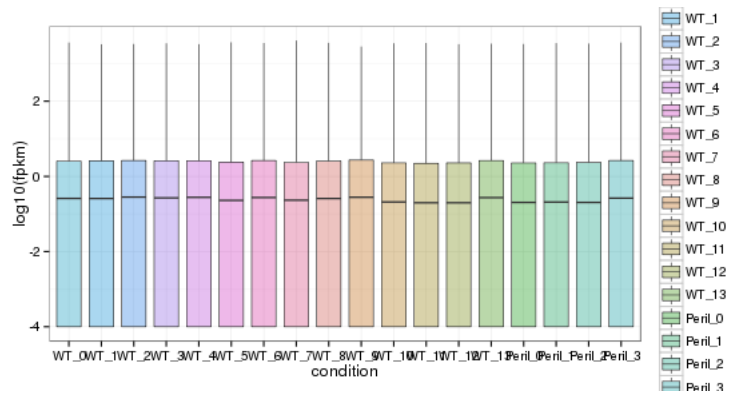
Boxplot (genes, replicates)



Boxplot (isoforms)

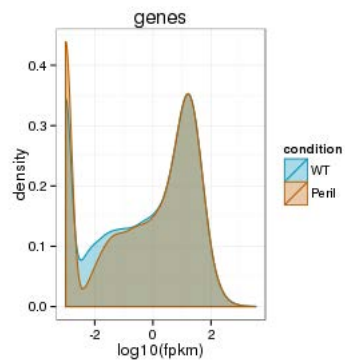


Boxplot (isoforms, replicates)

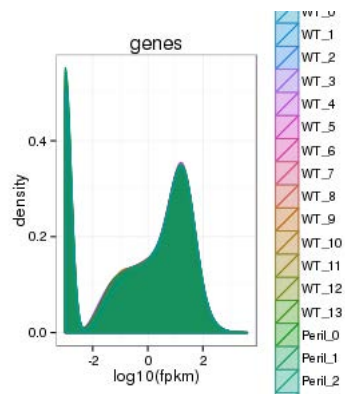


Density

Density (genes)

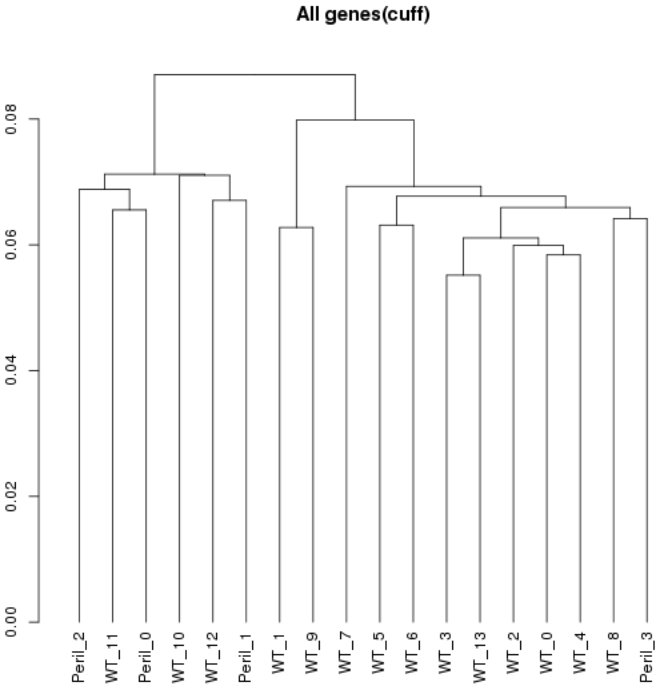


Density (genes, replicates)



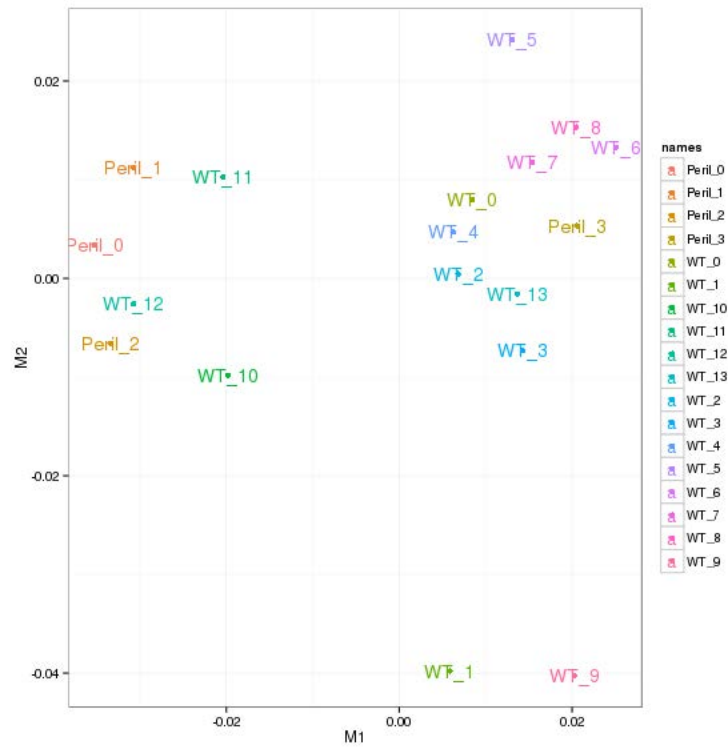
Clustering

Replicate Clusters



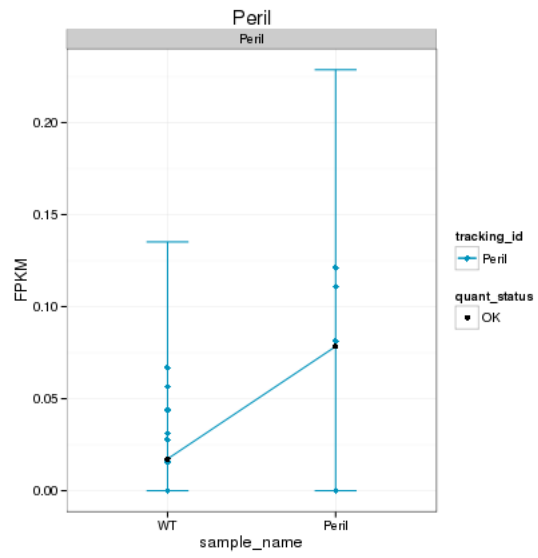
NULL

PCA (genes)

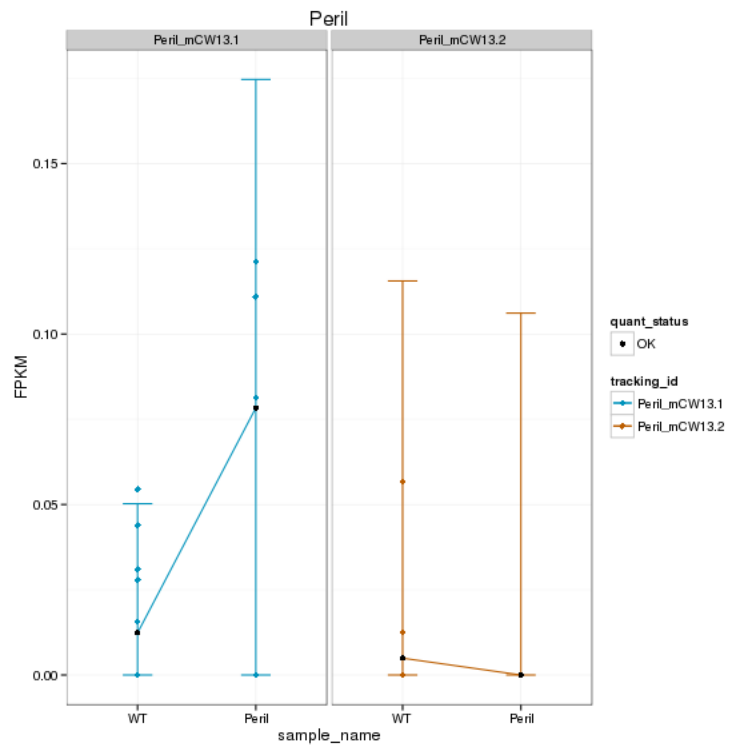


KO assessment

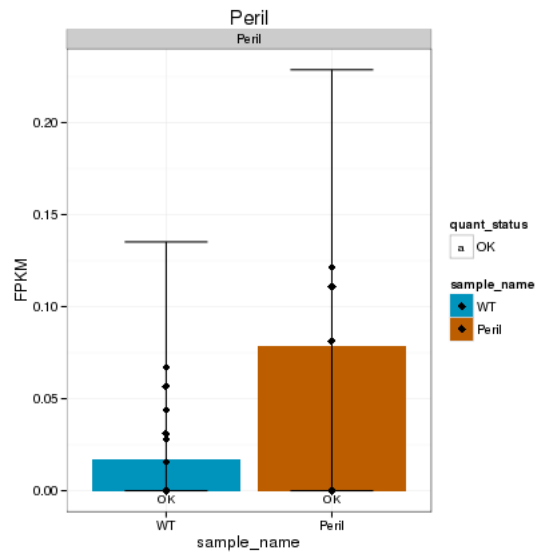
Endogenous IncRNA expression



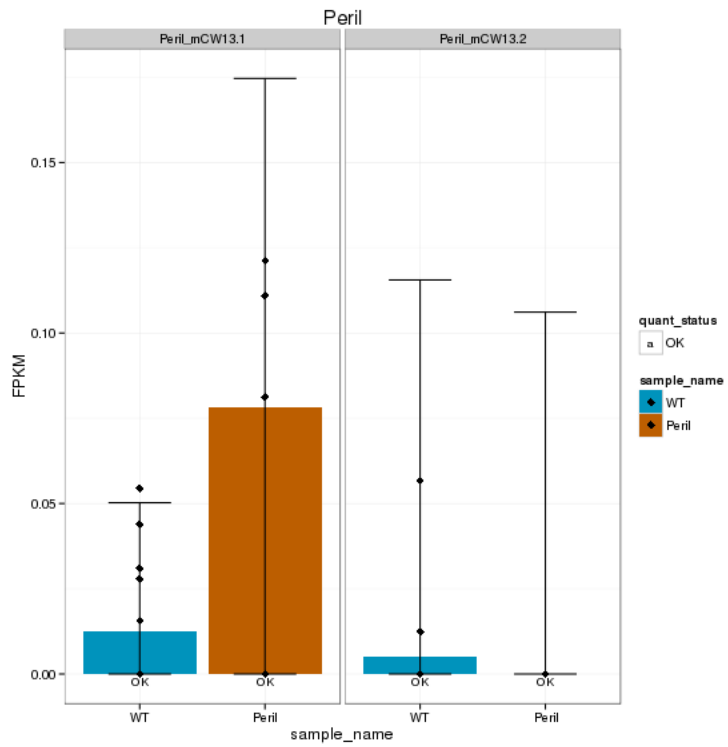
Endogenous expression of Peril isoforms:



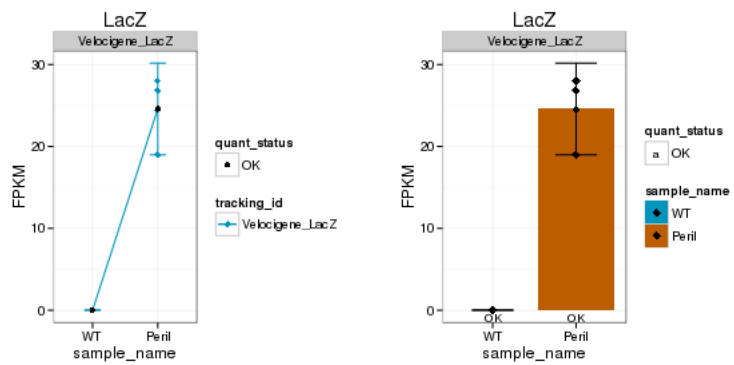
Barplot of gene expression:



Barplot of isoform expression:

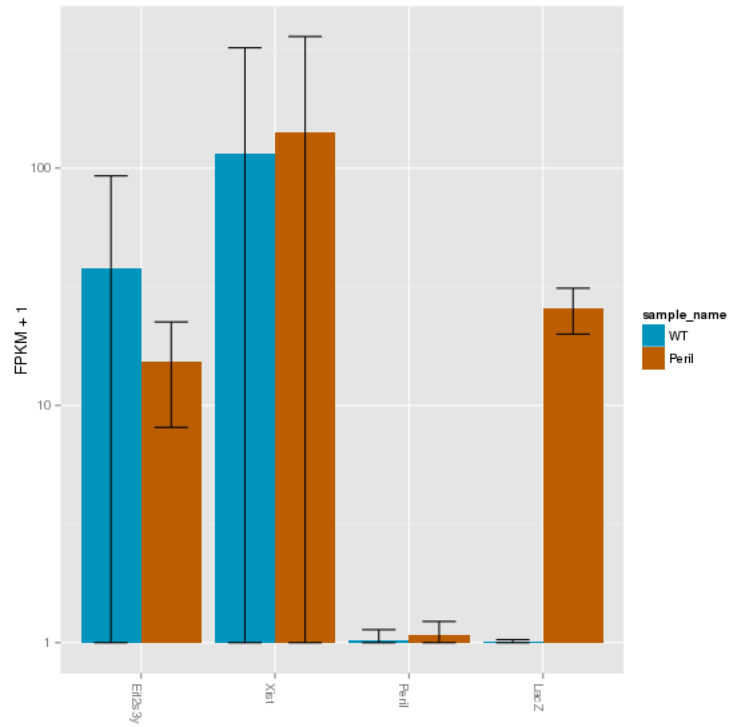


LacZ expression

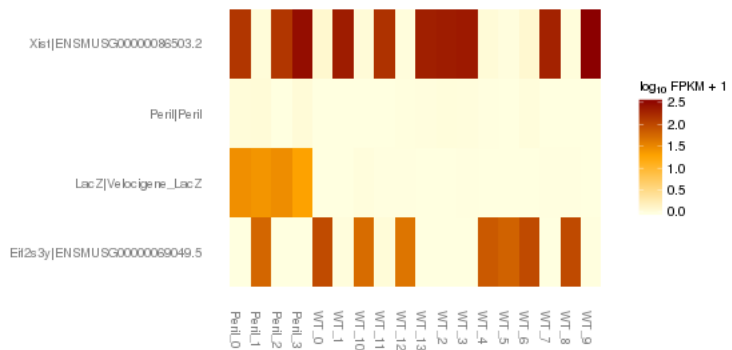


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

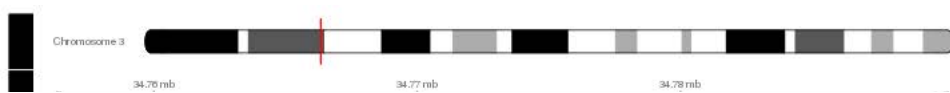
Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



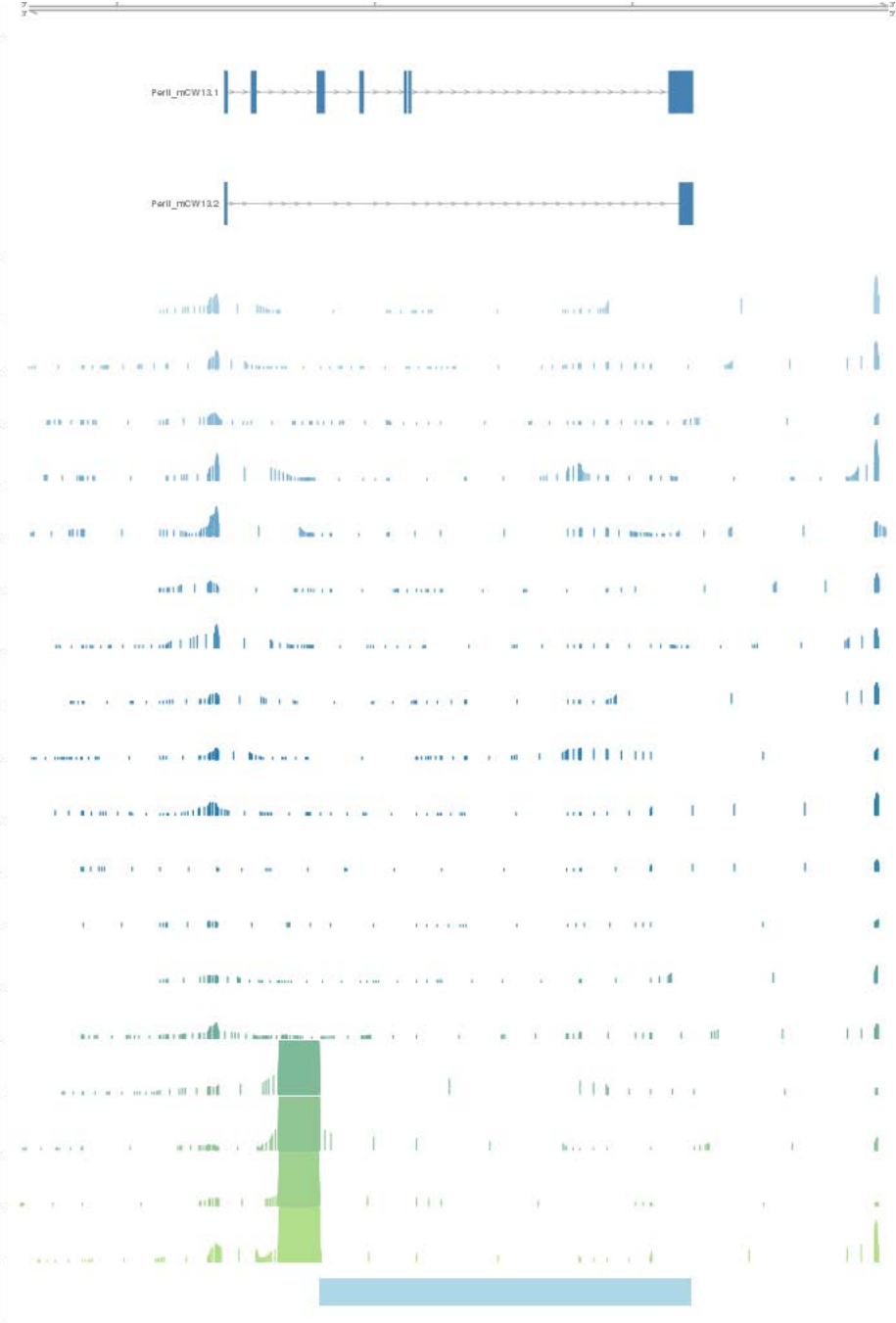
Expression heatmap:



Track visualization



NO Region | Peri0 | Peri1 | Peri2 | Peri3 | Peri4 | Peri5 | Peri6 | Peri7 | Peri8 | Peri9 | Peri10 | Peri11 | Peri12 | Peri13 | Peri14 | Peri15 | Peri16 | Peri17 | Peri18 | Peri19 | Peri20 | Peri21 | Peri22 | Peri23 | Peri24 | Peri25 | Peri26 | Peri27 | Peri28 | Peri29 | Peri30 | Peri31 | Peri32 | Peri33 | Peri34 | Peri35 | Peri36 | Peri37 | Peri38 | Peri39 | Peri40 | Peri41 | Peri42 | Peri43 | Peri44 | Peri45 | Peri46 | Peri47 | Peri48 | Peri49 | Peri50 | Peri51 | Peri52 | Peri53 | Peri54 | Peri55 | Peri56 | Peri57 | Peri58 | Peri59 | Peri60 | Peri61 | Peri62 | Peri63 | Peri64 | Peri65 | Peri66 | Peri67 | Peri68 | Peri69 | Peri70 | Peri71 | Peri72 | Peri73 | Peri74 | Peri75 | Peri76 | Peri77 | Peri78 | Peri79 | Peri80 | Peri81 | Peri82 | Peri83 | Peri84 | Peri85 | Peri86 | Peri87 | Peri88 | Peri89 | Peri90 | Peri91 | Peri92 | Peri93 | Peri94 | Peri95 | Peri96 | Peri97 | Peri98 | Peri99



Differential Analysis

Differential Genes

There are 254 significantly differentially expressed genes. They are:

	geneAnnot\$gene_short_name
1	Ccnd2
2	Itga5
3	Gabra2
4	Col18a1
5	Col1a1
6	Phyhip
7	Ddr1
8	Aqp1
9	Pax3
10	Nes
11	Nid1
12	Hmox1
13	Osgep
14	Rplp1
15	Wnt3a
16	Odc1
17	Aldh1a2
18	Dennd6b
19	Celsr1
20	Sulf1
21	Anxa6
22	Slc16a10
23	Lrr1q1
24	Lama2
25	Ndufa12
26	Rfx4
27	Col6a2
28	Aebp1
29	Pomc
30	Galk1
31	Nxn
32	Ntn1
33	Amn
34	Nid2
35	Gdf10
36	Esd
37	Tnfrsf10b

38 Psmb5
39 Npr3
40 Col14a1
41 Wnt7b
42 Myh9
43 Plec

44 Arhgap31
45 Clic6
46 Wnt1
47 Krt18
48 Dlx2
49 Igf2r
50 Gfra3
51 H2-M5
52 Pou4f3
53 Lox
54 Megf10
55 Fbn2
56 Pdgfrb
57 Glc3c
58 Cpt1a
59 Slc18a2
60 Rps24
61 Podxl
62 Col5a2
63 Col3a1
64 Igfbp5
65 Fn1
66 Ptma
67 Lamc1
68 Ptpn14
69 Ddr2
70 Lmx1a

71 Gad2
72 Col5a1
73 Lamc3
74 Hspa5
75 Lhx6
76 Notch1
77 Lrp2
78 Fbn1
79 Cd82
80 Tspan18

81 Syt13
82 Mccc1
83 Slc7a11
84 Hsd3b6
85 Notch2
86 Vcam1
87 Hadh
88 Wls
89 Tnc
90 Svep1
91 Stmn1
92 Trp73
93 Pdgfra
94 Gm996
95 Col1a2
96 Ephb4
97 Dlx5
98 Lrig1
99 Aldh111
100 Plxnd1
101 Emp1
102 Dbx1
103 Trpm1
104 Hddc3
105 Zkscan2
106 Lyve1
107 Heph
108 Col4a5
109 Plat
110 Slit2
111 Hsd11b2
112 Itga11
113 Gsta4
114 Car12
115 Smad3
116 Lama1
117 Tlr13
118 Gria2
119 Foxj1
120 Tmem132c
121 Scn10a
122 Ptpn13
123 Tdg

124 Prr7
125 Fras1
126 Arl4d
127 Arx
128 Ror1
129 Ccl2
130 Ccdc11
131 Tgfb
132 Cox8a
133 Colec12
134 Lum
135 C1ql2
136 Wnt8b
137 Frem2
138 Atp11b
139 Vstm2l
140 Notch3
141 Spon1
142 Satb2
143 Akap12
144 Atp5l
145 Itpkb
146 Anpep
147 Rbpj
148 Pcsk1n
149 Igfbp2
150 Kcne2
151 Bmf

152 Thbs1
153 Abcb1a

154 Ildr2
155 C1qtnf4
156 Abhd4
157 Npnt
158 Fzd7
159 Ccdc8
160 Arhgap11a
161 Htr2c
162 Prepl
163 Nlrp5-ps
164 Dlx1
165 Rassf4
166 Dmrt3

167 Lor
168 A030009H04Rik
169 Tmem145
170 Wfikkn2
171 Snhg11
172 Gm9493
173 Acp1
174 Cnpy1
175 Basp1
176 Olig1
177 Scand1
178 Fat4
179 Tmem164
180 Adamts12
181 Rps8
182 Pirt
183 Shisa3
184 Pla2g4e
185 Gja1
186 Rspo2
187 Pfdn4
188 Hbb-y
189 Hbb-bh1
190 Dnah6
191 Lamb2
192 Yap1
193 Sh3pxd2a
194 Hba-x
195 Dach1
196 Myh8
197 Col8a2
198 Pgpep1
199 Slit3
200 Cd248
201 Kdm5d
202 Gulp1
203 Hist3h2ba
204 Gm5148
205 Capn11
206 Sclt1
207 Hydin
208 Dcc
209 Cdh3

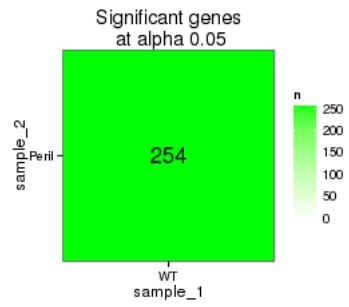
210 Trank1
211 Otof
212 Ldha
213 Cyp26b1
214 Col23a1
215 Scn1a
216 Cldn9
217 Lgals1
218 Slc4a5
219 Flnc
220 Sp9
221 Ddx3y
222 Ahnak
223 Hba-a2
224 Fat1
225 Ccnd1
226 Serpinh1
227 Gm10282
228 Gm1673
229 Gad1
230 Jund
231 Ppia
232 Fam84b
233 Gm7292
234 Drc1
235 2410018L13Rik
236 Wdfy1
237 Hbb-bt
238 Nynrin
239 Ywhaq
240 Fzd10
241 Pisd-ps1
242 Gm14204
243 Ier5l
244 Dlx6os1
245 Pou3f1
246 Ccdc85b
247 Gm21781
248 Gm6472
249 Dynl1f
250 Lhx8
251 A330076H08Rik
252 4930480K23Rik

253 A930011O12Rik

254 Gm27000

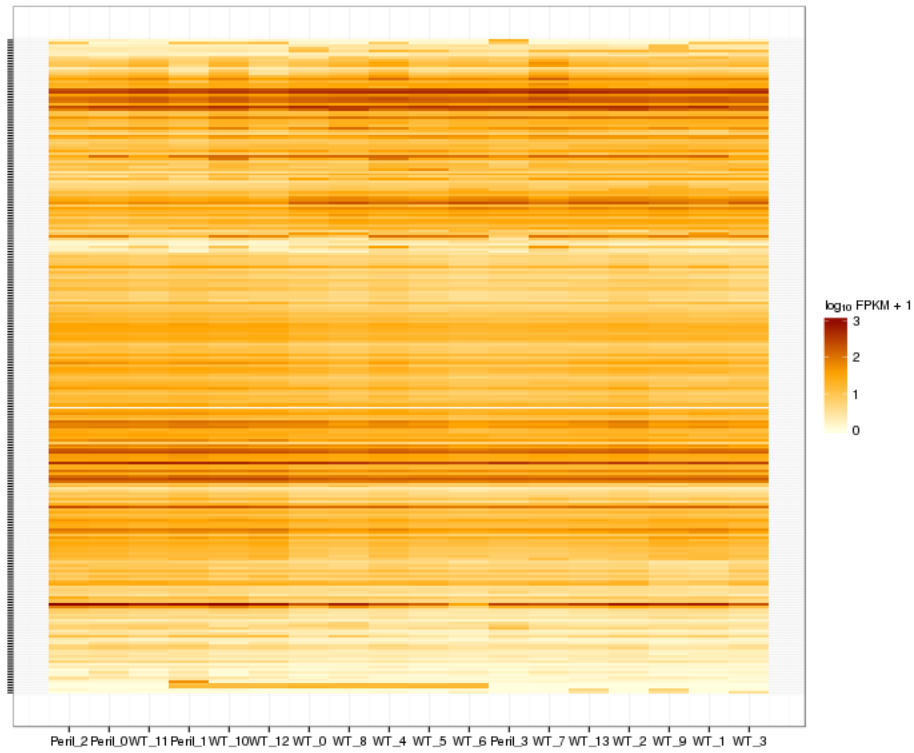
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

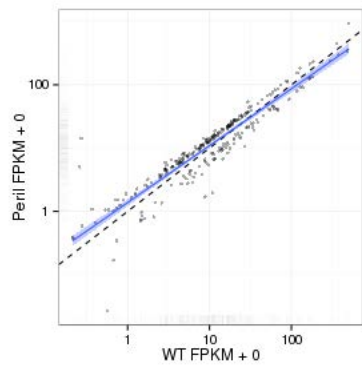


Significant genes with expression >50fpm (any condition):(turned off)

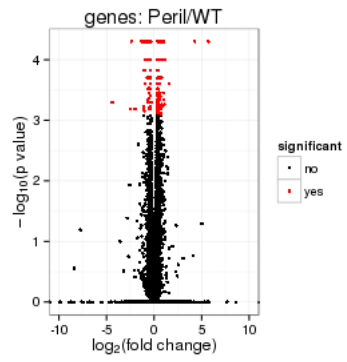
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

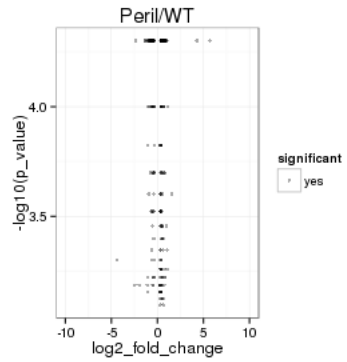
Scatter plot of significant genes only:



Volcano Plot



Volcano plot with significant genes only:



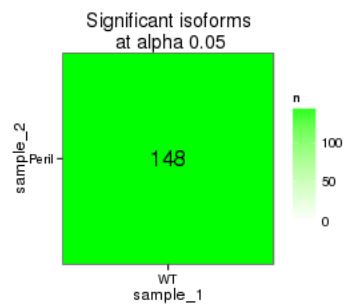
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

isoAnnot\$gene_short_name

- 1 Ccnd2
- 2 Gabra2
- 3 Col18a1
- 4 Col1a1
- 5 Aqp1
- 6 Nes
- 7 Nid1
- 8 Hmox1
- 9 Rplp1

- 10 Odc1
- 11 Cad
- 12 Lama5
- 13 Sulf1
- 14 Slc16a10
- 15 Lrriq1
- 16 Lama2
- 17 Gm872
- 18 Ndufa12
- 19 Pomc
- 20 Mrc2
- 21 Galk1
- 22 Nxn
- 23 Ntn1
- 24 Tnfrsf10b
- 25 Psm5
- 26 Npr3
- 27 Col14a1
- 28 Zbtb20
- 29 Arhgap31
- 30 Adamts1
- 31 Clic6
- 32 Dlx2
- 33 Igf2r
- 34 Svil
- 35 H2-M5
- 36 Pou4f3

- 37 Fbn2
- 38 Pdgfra
- 39 Gldc
- 40 Slc18a2

41 Podxl
42 Col5a2
43 Col3a1
44 Igfbp5
45 Fn1
46 Ptma
47 Lamc1
48 Lmx1a
49 Gad2
50 Col5a1
51 Notch1
52 Lrp2
53 Fbn1
54 Tspan18
55 Syt13
56 Rrbp1
57 Mccc1
58 Notch2
59 Vcam1
60 Svep1
61 Hspg2
62 Rspo1
63 Trp73
64 Col1a2

65 Dlx5
66 Aldh1l1
67 Smad3
68 Lama1
69 Foxj1
70 Tmem132c
71 Ptpn13
72 Prr7
73 Fras1
74 Atr4d
75 Arx
76 Cox8a
77 Colec12
78 Lum
79 Wnt8b
80 Frem2
81 D4Wsu53e
82 Notch3
83 Spon1

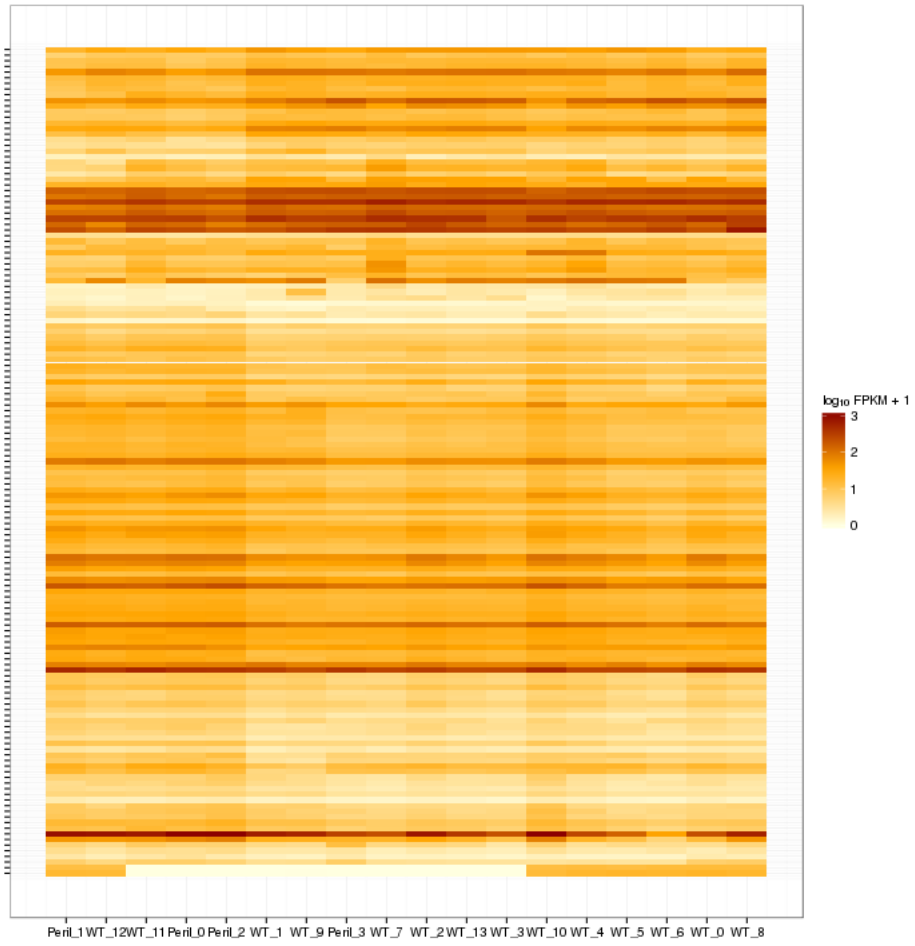
84 Akap12
85 Pcsk1n
86 Kcne2
87 Thbs1
88 C1qtnf4
89 Ccdc8
90 Arhgap11a
91 Prelp
92 Nlrp5-ps
93 Dix1
94 Rassf4
95 Dmrt3
96 Reln
97 Lor
98 Wfikkn2
99 Snhg11
100 Gm9493
101 Basp1
102 Olig1
103 Fat4
104 Adamts12
105 Rps8
106 Col6a3
107 Pla2g4e
108 Rspo2
109 Hbb-y
110 Hbb-bs
111 Ezr
112 Lamb2
113 Glis3
114 Yap1
115 Zfp771
116 Hba-x
117 Myh8

118 Col8a2
119 Pgpep1

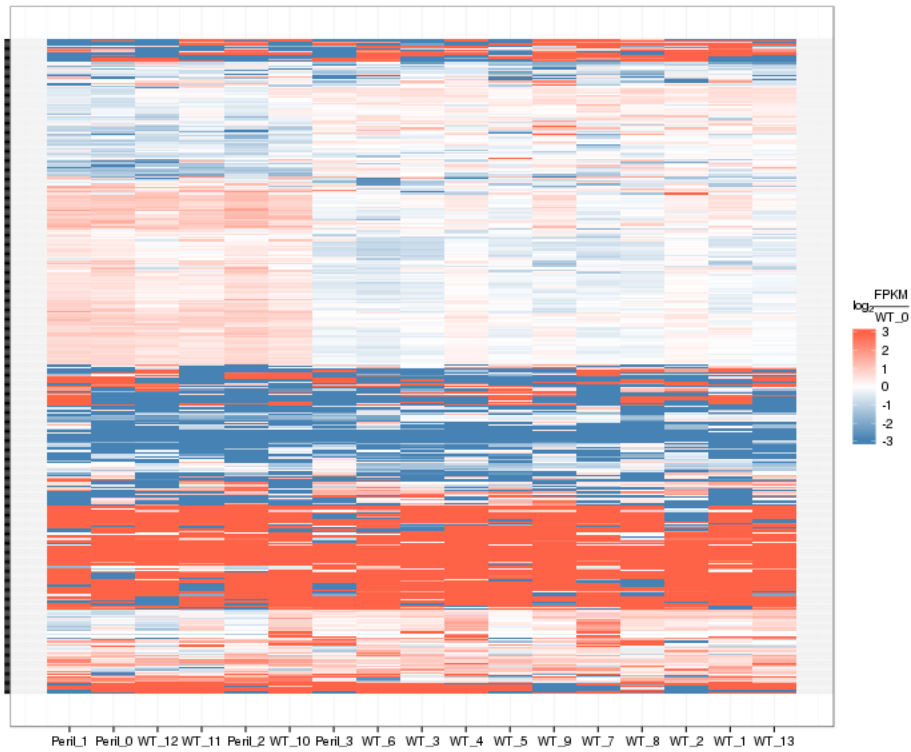
120 Ncoa4
121 Kdm5d
122 Hist3h2ba
123 Hydin
124 Trank1
125 Finc
126 Sp9

127 Ddx3y
128 Gpr98
129 Ahnak
130 Fat1
131 Serpinh1
132 Gm10282
133 Jund
134 Fam84b
135 Drc1
136 Hbb-bt
137 Col22a1
138 Fzd10
139 Ier5l
140 Dlx6os1
141 Pou3f1
142 Ccdc85b
143 Gm6472
144 4930480K23Rik
145 Rmst
146 A930011O12Rik
147 Gm27000

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

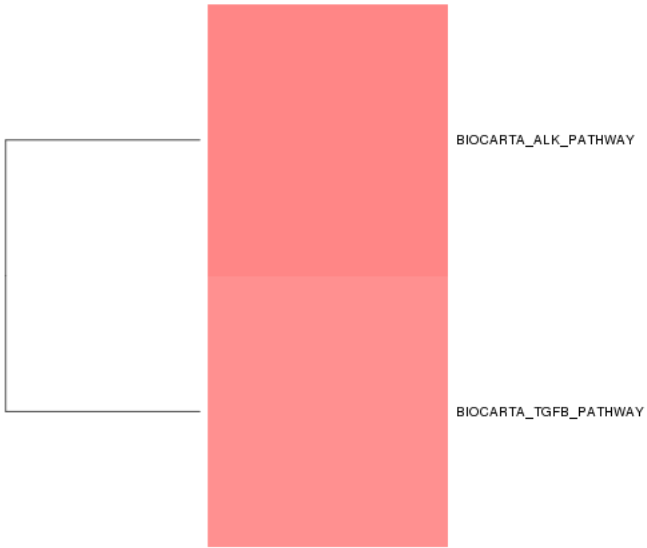
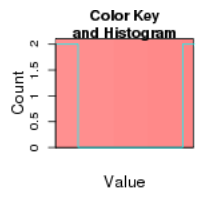
Gene/Pathway Analysis

GSEA

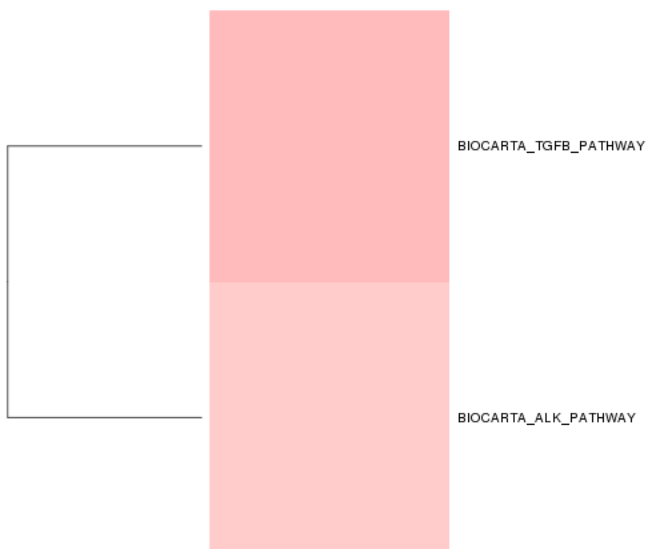
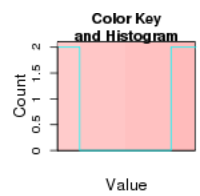
Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

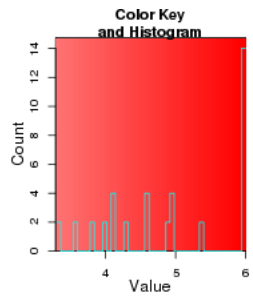
Biocarta enrichment:

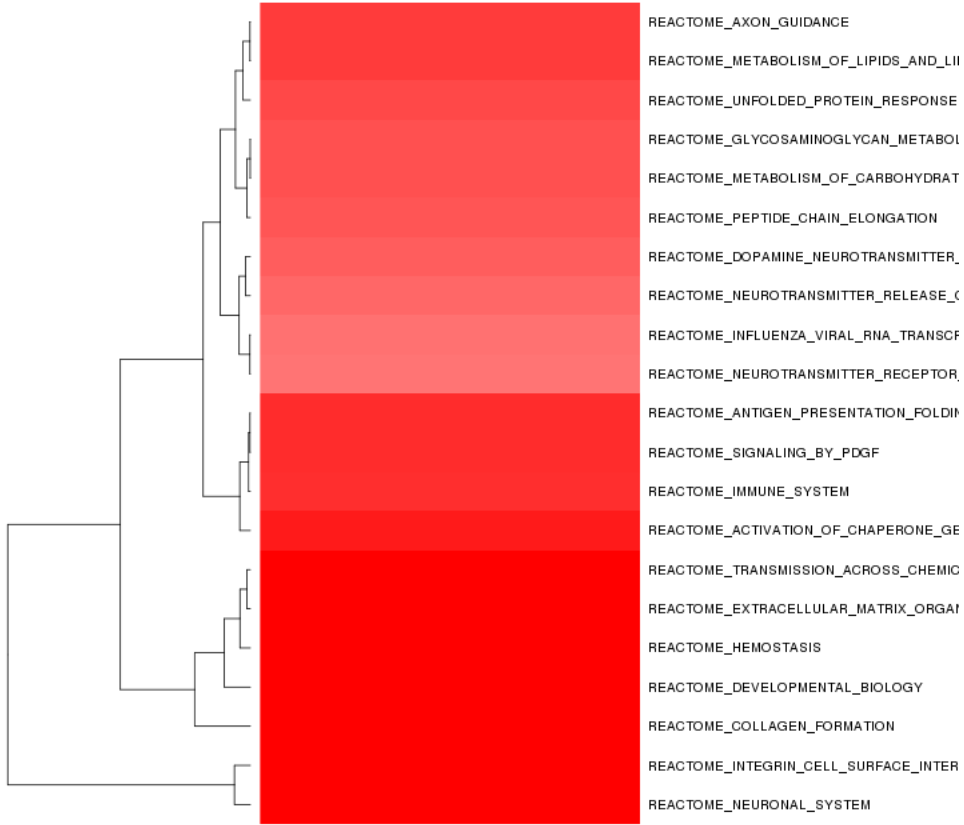


Biocarta zscore:

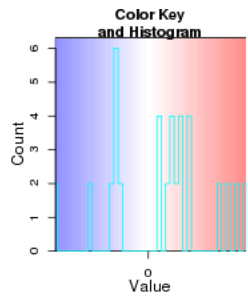


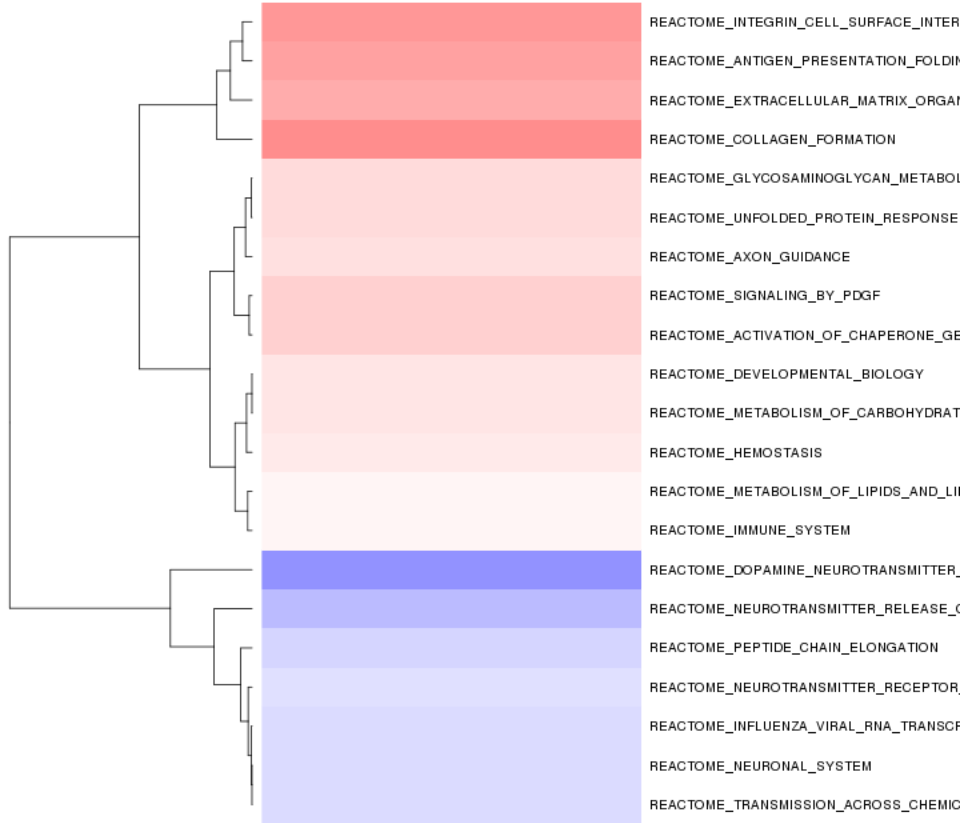
Reactome enrichment:



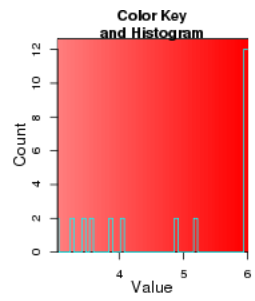


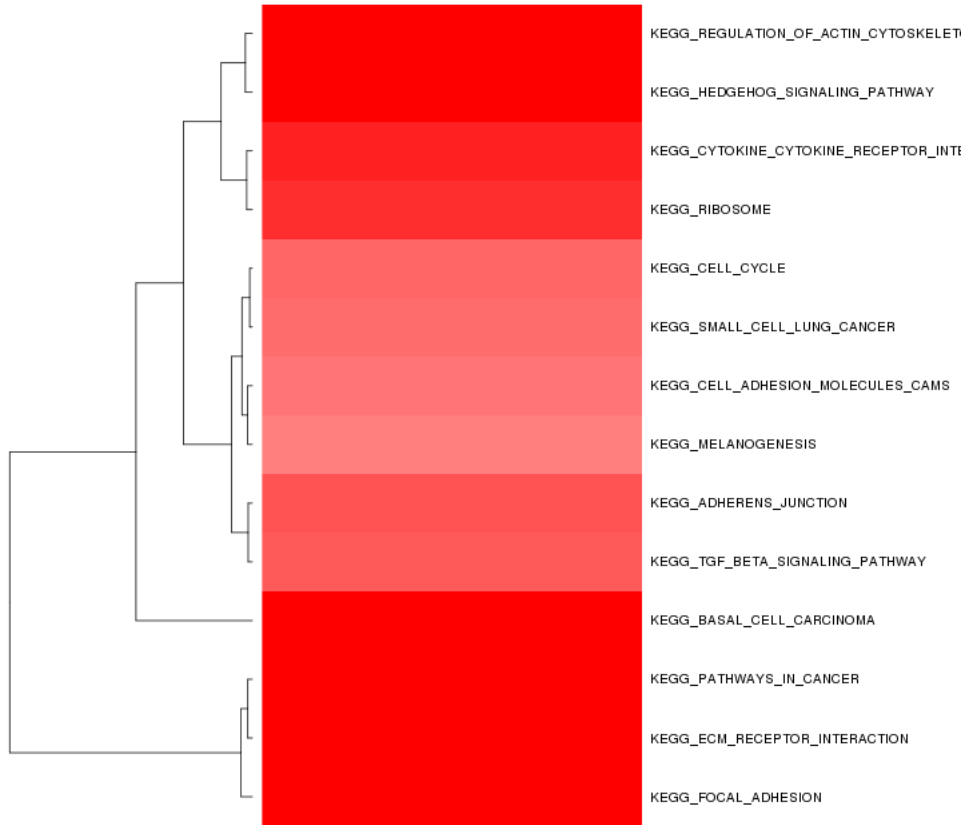
Reactome zscore:



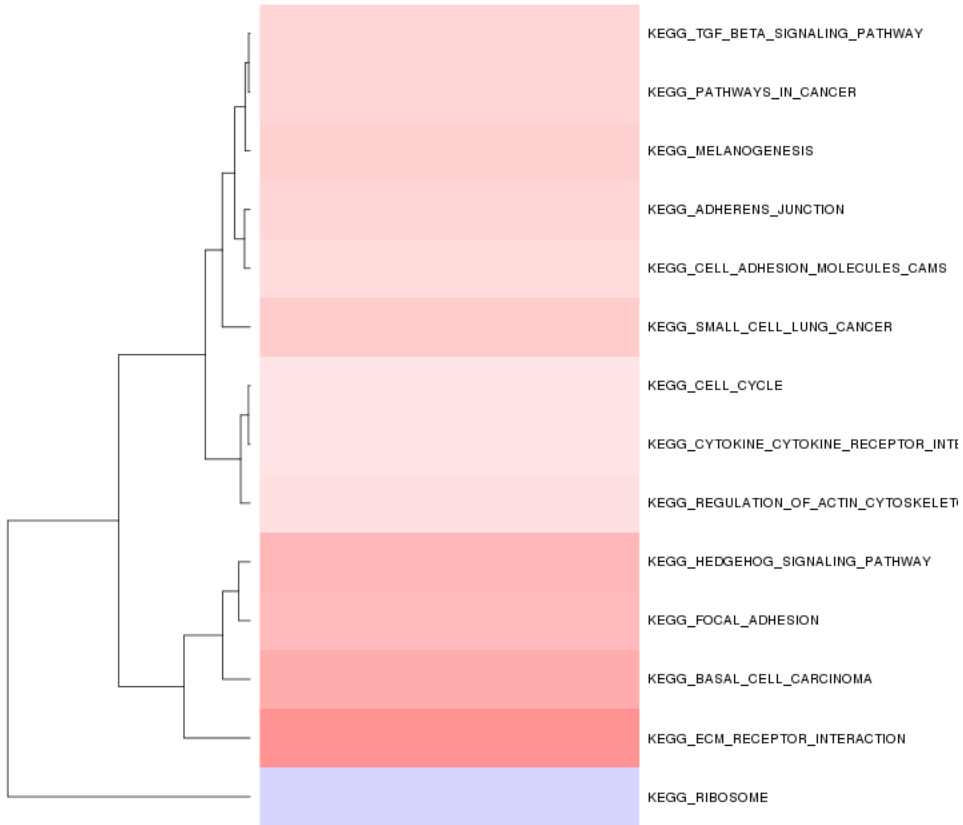
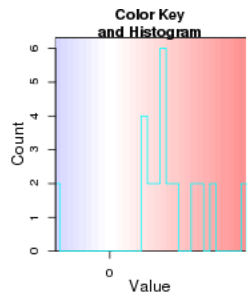


Kegg enrichment:





Kegg zscore:



Interneuron enrichment:

Error: argument is of length zero

Interneuron zscore:

Error: incorrect number of dimensions

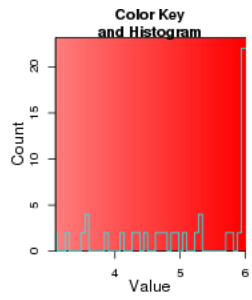
```
## Error: object 'x_ordered' not found
```

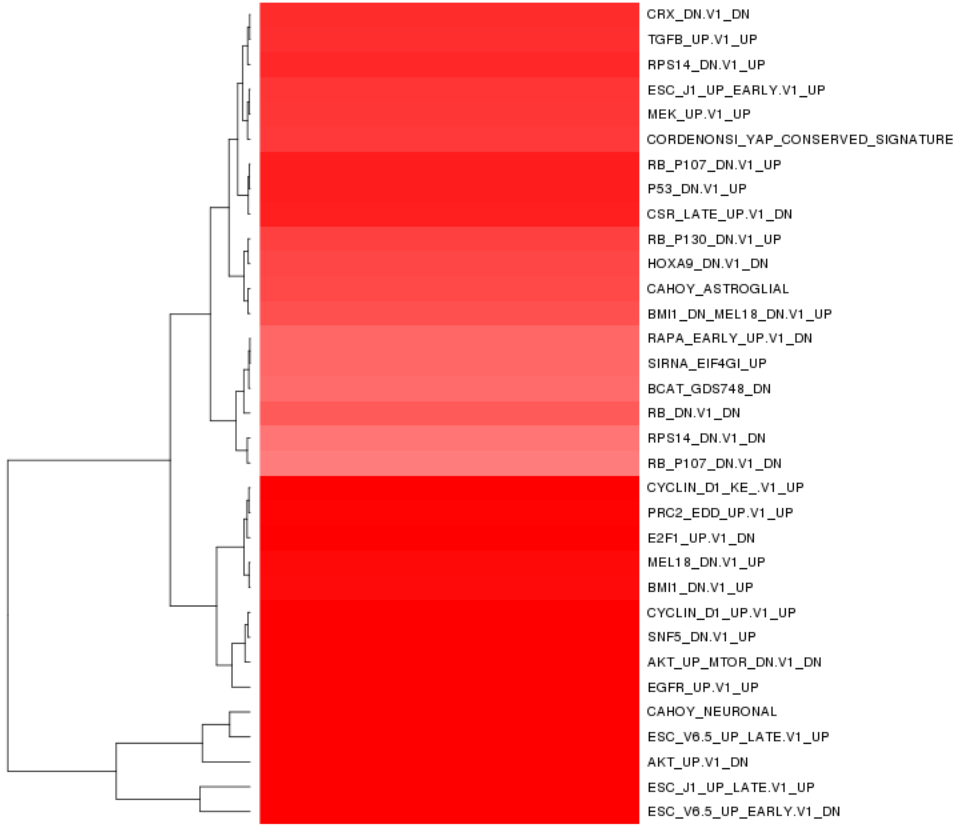
```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Oncogene enrichment:

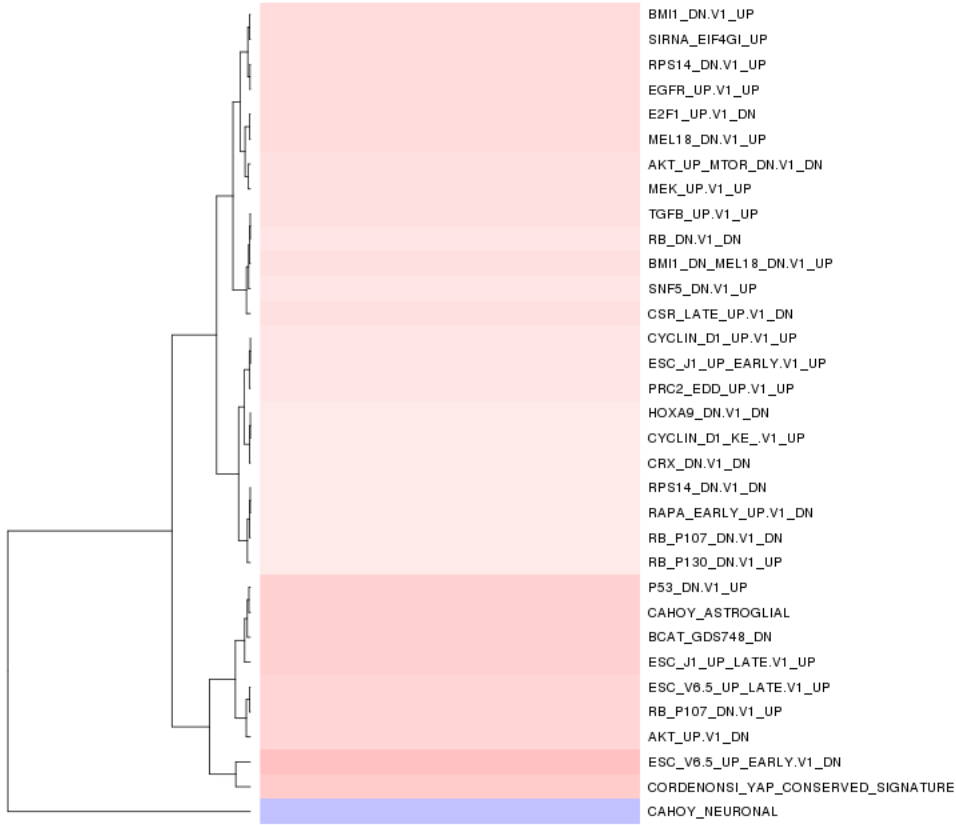
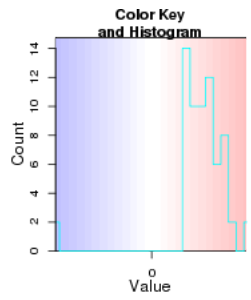
```
## Error: subscript out of bounds
```



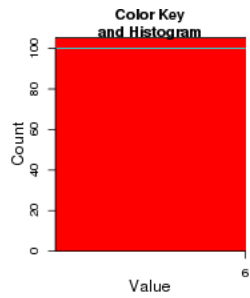


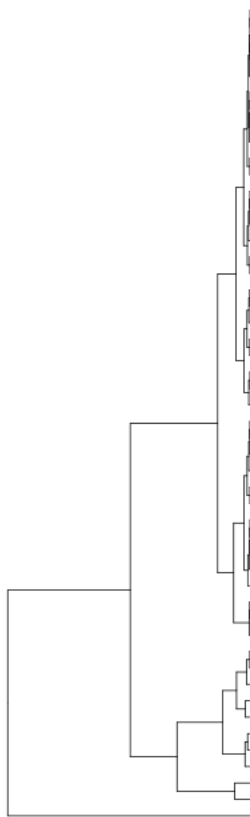
Oncogene zscore:

Error: subscript out of bounds



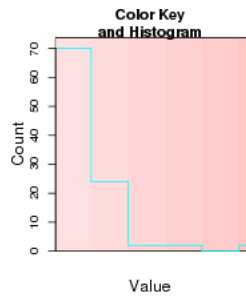
Immuno enrichment:

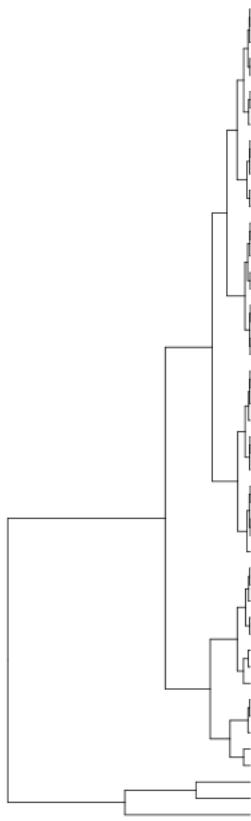




GSE27786_BCELL_VS_NKCELL_DN
 GSE20715_0H_VS_24H_OZONE_TLR4_KO_LUN
 GSE24142_DN2_VS_DN3_THYMOCYTE_FETAL
 GSE24142_EARLY_THYMIC_PROGENITOR_VS_
 GSE3337_4H_VS_16H_IFNG_IN_CD8POS_DC_I
 GSE20715_0H_VS_48H_OZONE_LUNG_DN
 KAECH_NAIVE_VS_DAY8_EFF_CD8_TCELL_DN
 GSE6269_E_COLI_VS_STREP_AUREUS_INF_P
 GSE2706_2H_VS_8H_R848_STIM_DC_UP
 GSE22886_NAIVE_TCELL_VS_DC_DN
 GSE300_L_DONOVANI_VS_B_MALAYI_HIGH_D
 GSE30962_PRIMARY_VS_SECONDARY_ACUTE
 GSE17721_CPG_VS_GARDIQUIMOD_8H_BMDM
 GSE1460_INTRATHYMIC_T_PROGENITOR_VS_
 GSE15930_NAIVE_VS_72H_IN_VITRO_STIM_IL
 GSE29618_MONOCYTE_VS_PDC_UP
 GSE24634_TREG_VS_TCONV_POST_DAY10_IL
 GSE3982_DC_VS_EFF_MEMORY_CD4_TCELL_
 GSE1460_CD4_THYMOCYTE_VS_THYMIC_STR
 GSE24026_PDI_LIGATION_VS_CTRL_IN_ACT_
 GSE22886_NAIVE_BCELL_VS_MONOCYTE_DN
 KAECH_NAIVE_VS_MEMORY_CD8_TCELL_DN
 GSE22886_NAIVE_BCELL_VS_BLOOD_PLASM
 GSE9988_LPS_VS_VEHICLE_TREATED_MONO
 GSE14000_4H_VS_16H_LPS_DC_TRANSLATEC
 GSE17721_CTRL_VS_LPS_12H_BMDM_UP
 GSE24634_IL4_VS_CTRL_TREATED_NAIVE_CF
 GSE9988_ANTI_TREM1_VS_VEHICLE_TREATEI
 GSE300_HIGH_DOSE_B_MALAYI_VS_M_TUBEI
 GSE9650_EFFECTOR_VS_MEMORY_CD8_TCEL
 GSE10325_CD4_TCELL_VS_MYELOID_DN
 GOLDRATH_NAIVE_VS_EFF_CD8_TCELL_DN
 GSE9650_NAIVE_VS_EFF_CD8_TCELL_DN
 GSE24142_EARLY_THYMIC_PROGENITOR_VS_
 GSE6269_HEALTHY_VS_STREP_PNEUMO_INF
 GSE29618_BCELL_VS_MONOCYTE_DAY7_FLU
 GSE15750_DAY8_VS_DAY10_TRAF6KO_EFF_C
 GSE24634_IL4_VS_CTRL_TREATED_NAIVE_CF
 GSE24634_TREG_VS_TCONV_POST_DAY7_IL4
 GSE24142_DN2_VS_DN3_THYMOCYTE_UP
 GSE10325_LUPUS_CD4_TCELL_VS_LUPUS_M
 GSE15767_MED_VS_SCS_MAC_LN_UP
 GSE9988_ANTI_TREM1_AND_LPS_VS_VEHICLI
 GSE24142_EARLY_THYMIC_PROGENITOR_VS_
 GSE24634_TEFF_VS_TCONV_DAY7_IN_CULTU
 GSE29614_CTRL_VS_DAY7_TIV_FLU_VACCINE
 GSE24142_EARLY_THYMIC_PROGENITOR_VS_
 GSE10325_LUPUS_BCELL_VS_LUPUS_MYELO
 GSE24634_TEFF_VS_TCONV_DAY10_IN_CULTI
 GSE24634_TREG_VS_TCONV_POST_DAY10_IL

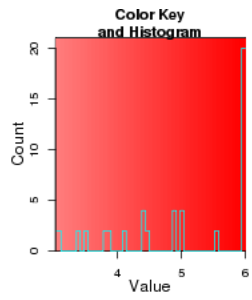
Immuno zscore:

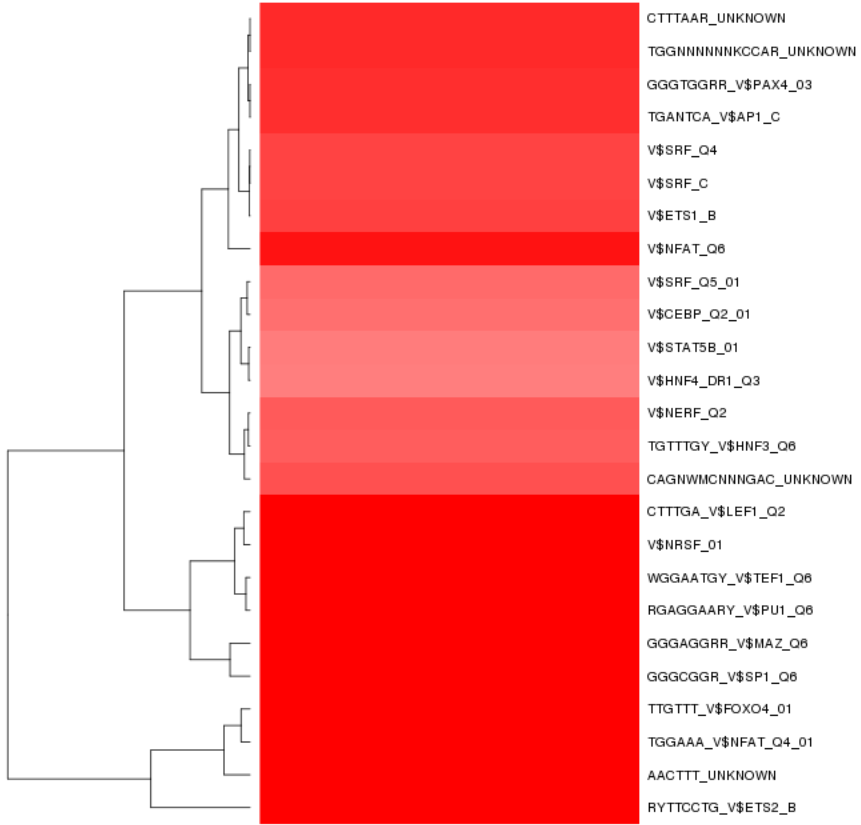




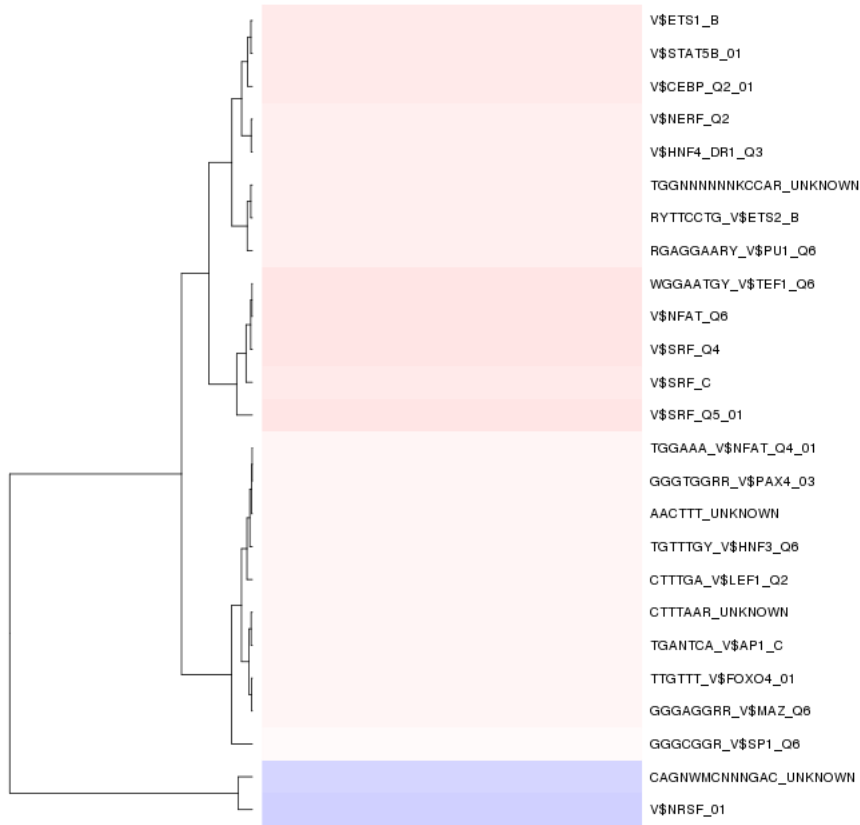
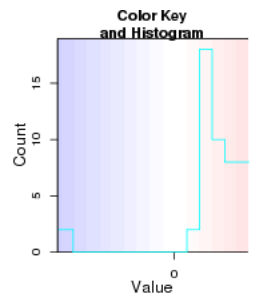
GSE13485_DAY7_VS_DAY21_YF17D_VACCINE
 GSE20715_WT_VS_TLR4_KO_24H_OZONE_LUI
 GSE24034_IL4_VS_CTRL_TREATED_NAIVE_CC
 GSE24142_DN2_VS_DN3_THYMOCYTE_UP
 GSE22880_DC_VS_MONOCYTE_UP
 GSE2706_UNSTIM_VS_8H_R848_DC_UP
 GSE0209_HEALTY_VS_STREP_PNEUMO_INF
 GSE24142_EARLY_THYMIC_PROGENITOR_VS_
 GSE3982_DC_VS_BCELL_UP
 GSE7852_THYMUS_VS_FAT_TCONV_DN
 GSE24034_IL4_VS_CTRL_TREATED_NAIVE_CC
 GSE3337_4H_VS_16H_IFNG_IN_CD8POS_DC_I
 GSE20715_0H_VS_24H_OZONE_TLR4_KO_LUI
 GSE22880_NAIVE_BCELL_VS_MONOCYTE_DN
 GSE29018_MONOCYTE_VS_MDC_DAY7_FLU_V
 GSE29018_BCELL_VS_MONOCYTE_DAY7_FLU
 GSE24034_TREG_VS_TCONV_POST_DAY7_IL4
 GSE10325_BCELL_VS_MYELOID_DN
 GSE9050_NAIVE_VS_EFF_CD8_TCELL_DN
 GSE3982_DC_VS_CENT_MEMORY_CD4_TCELL
 GSE29014_CTRL_VS_DAY7_TIV_FLU_VACCINE
 GSE15767_MED_VS_SCS_MAC_LN_UP
 GSE9988_ANTI_TREM1_VS_VEHICLE_TREATEI
 GSE24142_ADULT_VS_FETAL_DN2_THYMOCY
 GSE360_L_DONOVANL_VS_B_MALAYL_HIGH_D
 GSE20715_0H_VS_48H_OZONE_LUNG_DN
 GSE17721_0.5H_VS_24H_CPG_BMDM_DN
 KAECH_NAIVE_VS_DAY8_EFF_CD8_TCELL_DN
 GSE24034_IL4_VS_CTRL_TREATED_NAIVE_CC
 GOLDRATH_NAIVE_VS_MEMORY_CD8_TCELL
 KAECH_NAIVE_VS_DAY15_EFF_CD8_TCELL_D
 GSE24142_EARLY_THYMIC_PROGENITOR_VS_
 GSE1480_DP_THYMOCYTE_VS_THYMIC_STRO
 GSE9988_ANTI_TREM1_AND_LPS_VS_VEHICLI
 GSE29018_MONOCYTE_VS_MDC_UP
 GSE2706_2H_VS_8H_R848_STIM_DC_UP
 GSE3982_DC_VS_EFF_MEMORY_CD4_TCELL
 GSE10325_LUPUS_CD4_TCELL_VS_LUPUS_M
 GSE1480_CD4_THYMOCYTE_VS_THYMIC_STR
 GSE29018_MONOCYTE_VS_PDC_DAY7_FLU_V
 GSE0209_E_COLI_VS_STREP_AUREUS_INF_P
 GSE3337_CTRL_VS_16H_IFNG_IN_CD8POS_DX
 GSE1480_INTRATHYMIC_T_PROGENITOR_VS_
 GSE24142_EARLY_THYMIC_PROGENITOR_VS_
 GSE24034_TEFF_VS_TCONV_DAY10_IN_CULTI
 GSE29018_MONOCYTE_VS_PDC_UP
 GSE14000_4H_VS_16H_LPS_DC_TRANSLATEE
 GSE10325_LUPUS_BCELL_VS_LUPUS_MYELO
 GSE24034_TEFF_VS_TCONV_DAY7_IN_CULTU
 GSE24034_TREG_VS_TCONV_POST_DAY10_IL

TF enrichment:



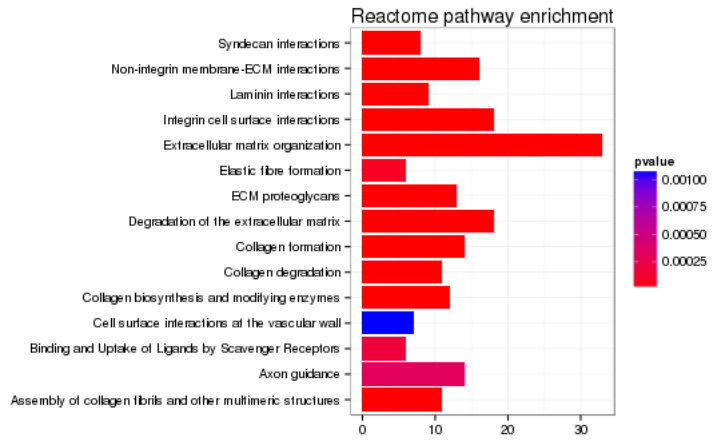
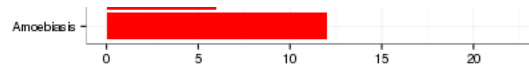
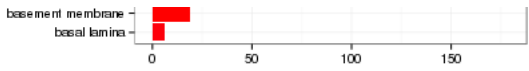


TF zscore:

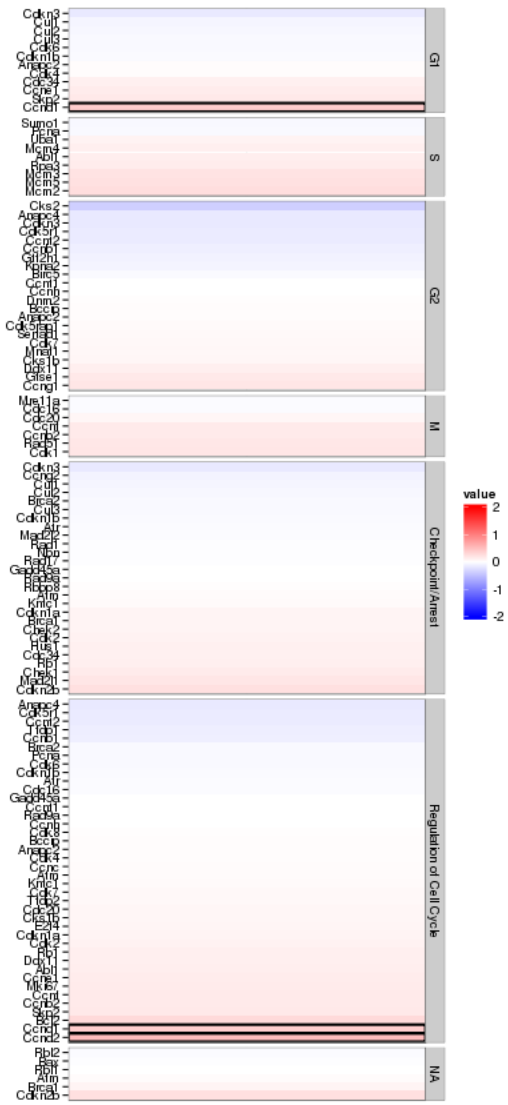


GO enrichment

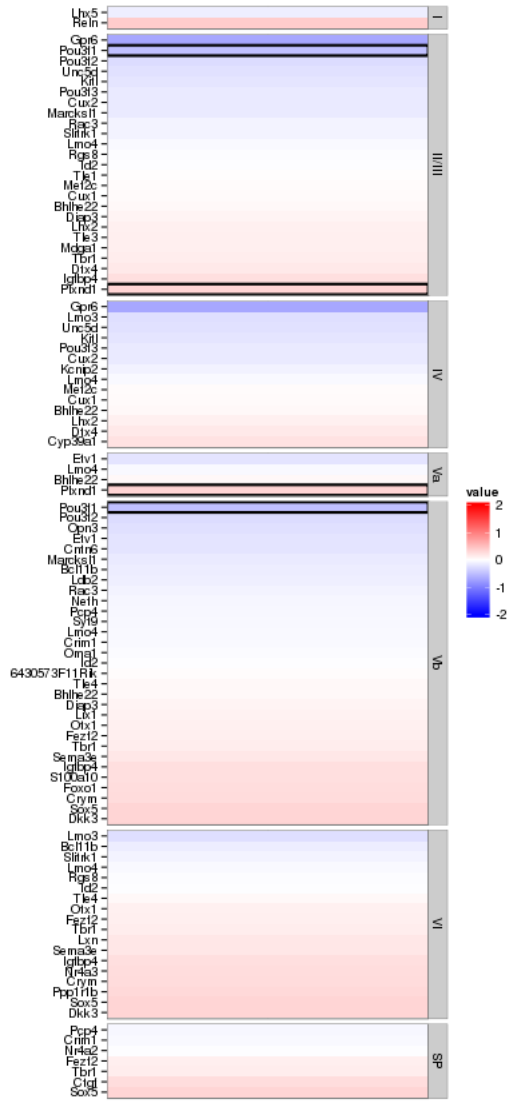
Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.



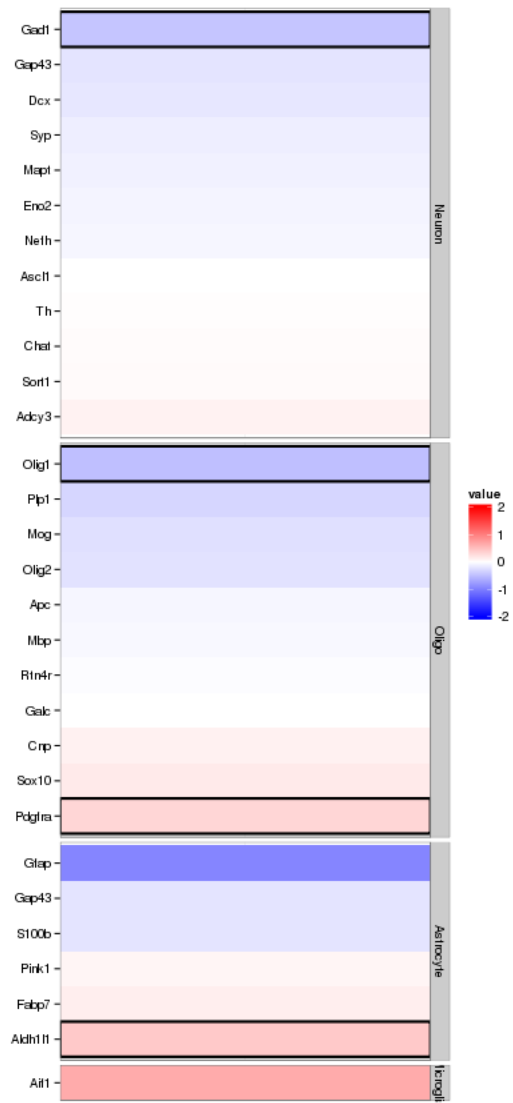
Enrichment or depletion for stage-specific cell cycle markers



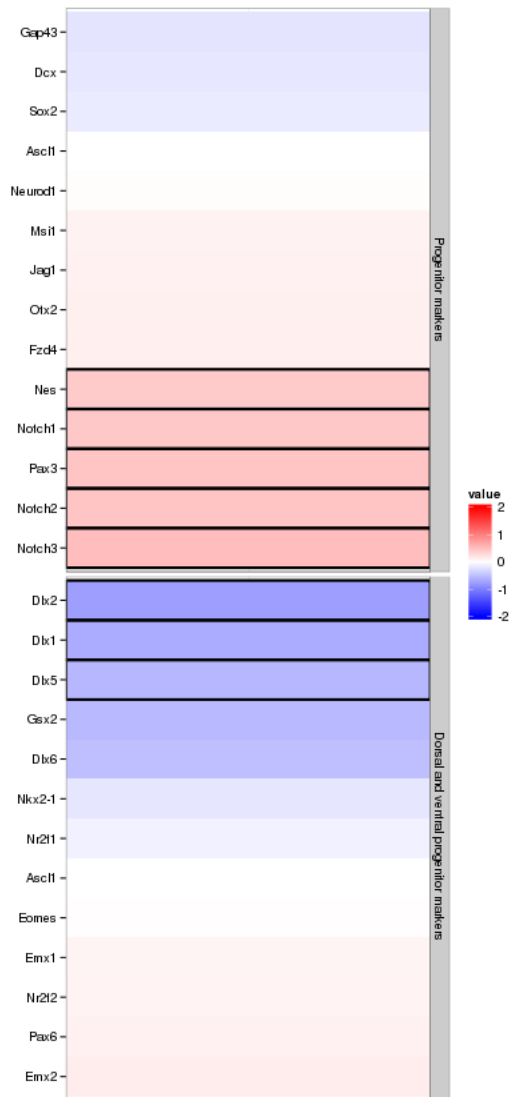
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



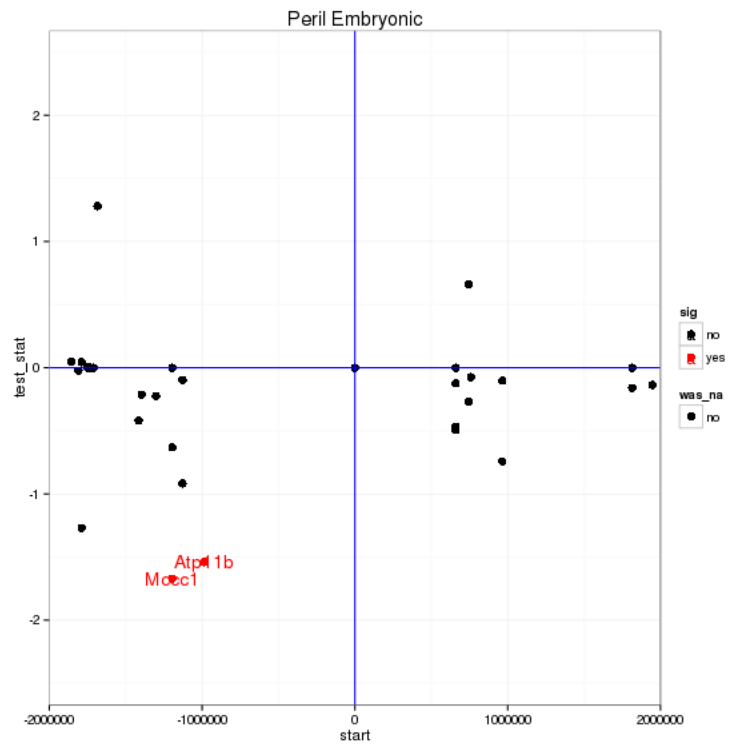
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 2 genes significantly regulated in a region this size is: 0.1906



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR721
16 JR718
17 JR720
18 JR792

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	30202700.00	1.35	1.00
2 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	30202700.00	1.18	1.00
3 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	30202700.00	1.31	1.00
4 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	30202700.00	1.38	1.00
5 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	30202700.00	1.28	1.00
6 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	30202700.00	0.95	1.00
7 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	30202700.00	1.04	1.00
8 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	30202700.00	0.98	1.00
9 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	30202700.00	0.92	1.00
10 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	30202700.00	1.03	1.00
11 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	30202700.00	0.87	1.00
12 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	30202700.00	0.85	1.00
13 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	30202700.00	0.73	1.00
14 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	30202700.00	1.30	1.00
15 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR721/abundances.cxb	Peril	0	Peril_0	22593200.00	30202700.00	0.75	1.00
16 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR718/abundances.cxb	Peril	1	Peril_1	27635500.00	30202700.00	0.92	1.00
17 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR720/abundances.cxb	Peril	2	Peril_2	19589600.00	30202700.00	0.64	1.00
18 /n/rinn_data1/seq/lgooff/Projects/BrainMap/data/quants/JR792/abundances.cxb	Peril	3	Peril_3	29654500.00	30202700.00	0.99	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
```

```

## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] ggplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2     colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4       DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10       Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSeqSim_1.20.3    graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1       Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29    latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3       munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10       qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4       splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7    tcltk_3.0.2

```

```
## [40] tools_3.0.2
```

```
XML_3.98-1.1
```

```
zlibbioc_1.8.0
```

Run Info

```
##      param
## 1    cmd_line
## 2      version
## 3  SVN_revision
## 4 boost_version
## 5      genome
##
## 1 cuffdiff -p 10 -L WT,Peril -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Peril_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/BrainMap/dat
## 2
## 3
## 4
## 5
```

Trp53cor1 KO vs WT (Adult)

This file shows the wt-v-ko comparison for Trp53cor1.

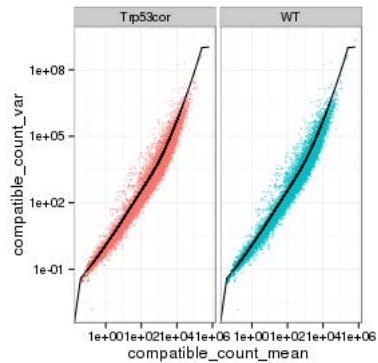
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

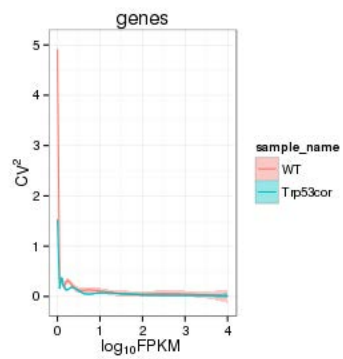
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



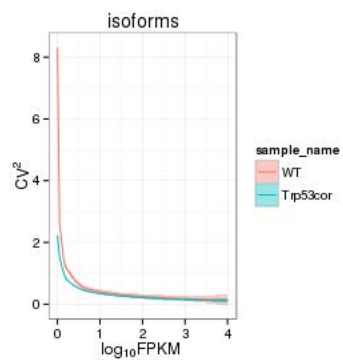
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

Genes:



Isoforms:



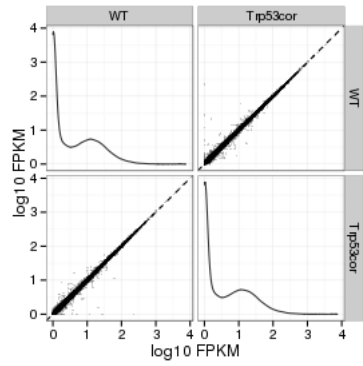
MvA plot

```
## Error: non-numeric argument to binary operator
```

MvA plot counts

```
## Error: non-numeric argument to binary operator
```

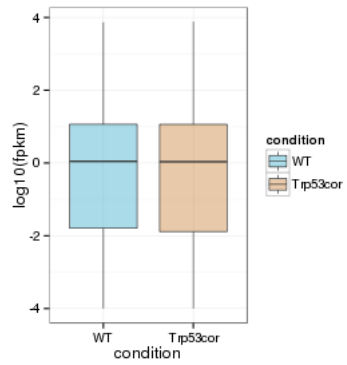
Scatterplot matrix



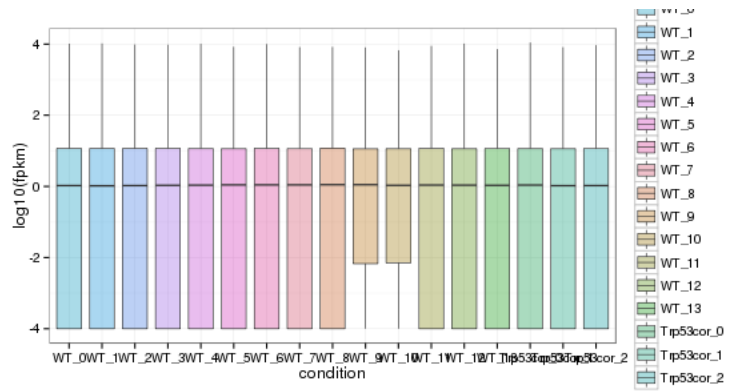
Distributions

Boxplots

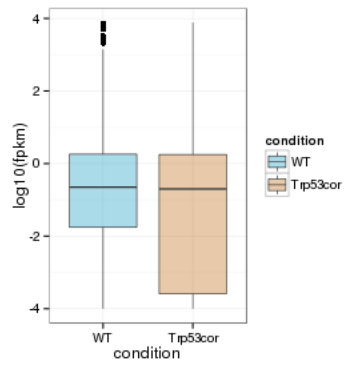
Boxplot (genes)



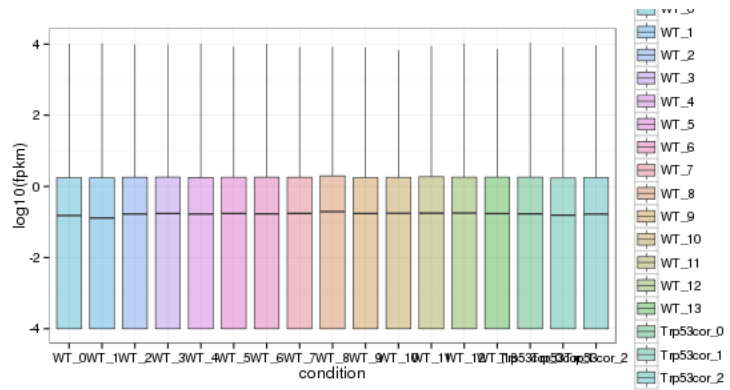
Boxplot (genes, replicates)



Boxplot (isoforms)

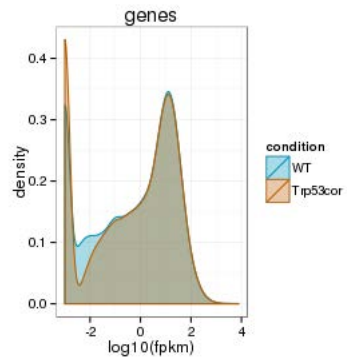


Boxplot (isoforms, replicates)

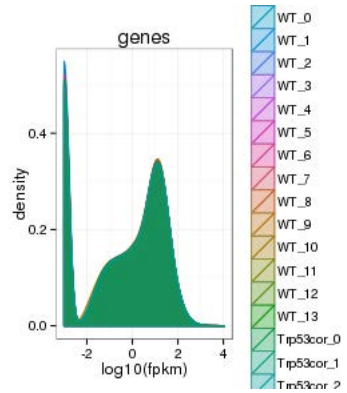


Density

Density (genes)



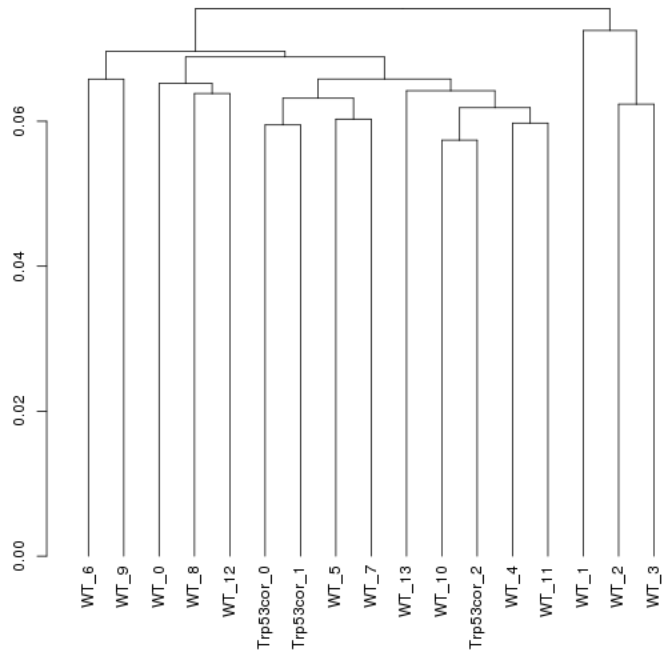
Density (genes, replicates)



Clustering

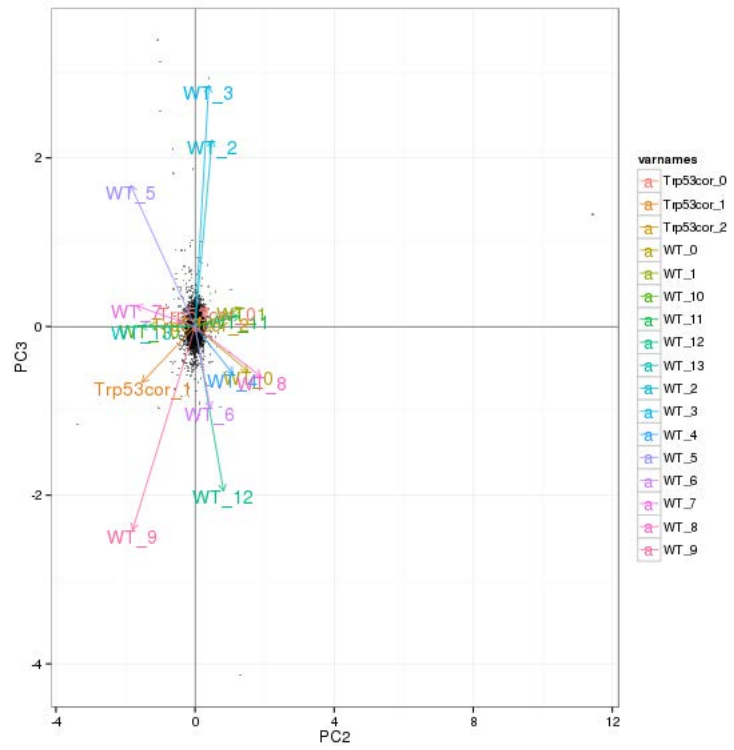
Replicate Clusters

All genes(cuff)

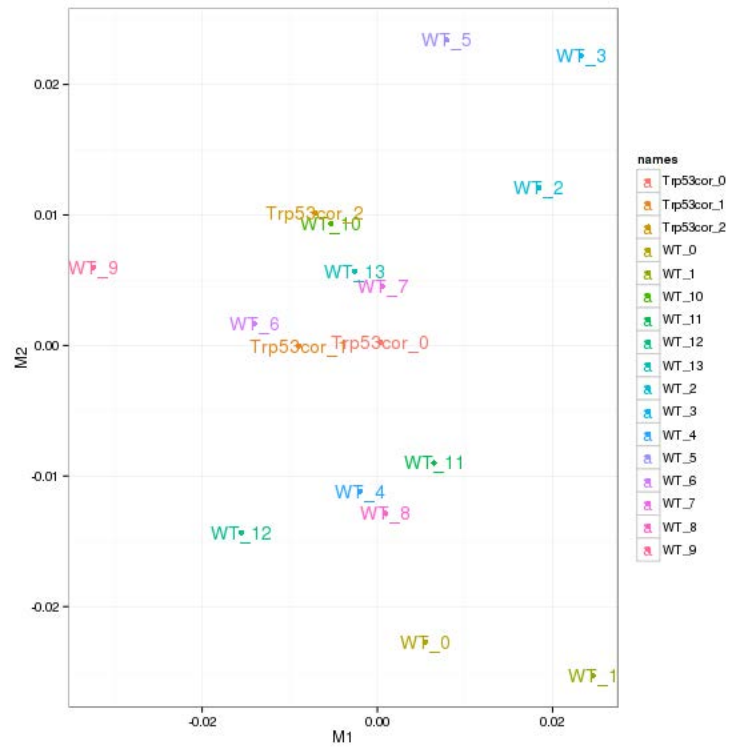


NULL

PCA (genes)

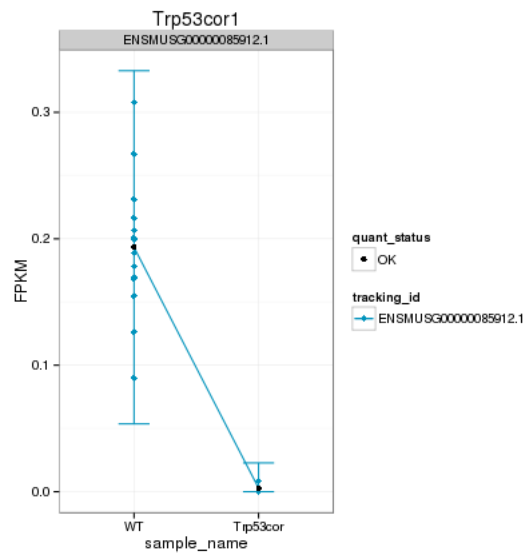


MDS (genes)

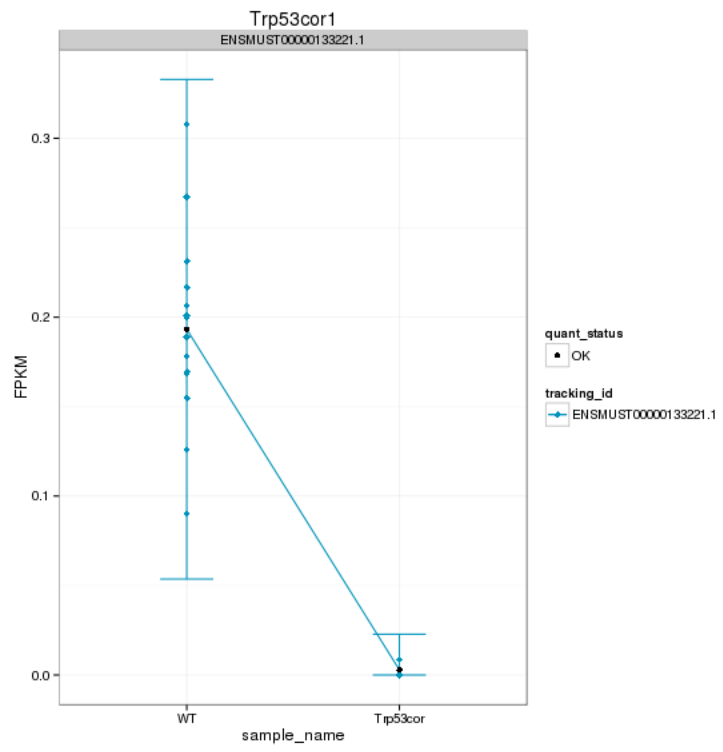


KO assessment

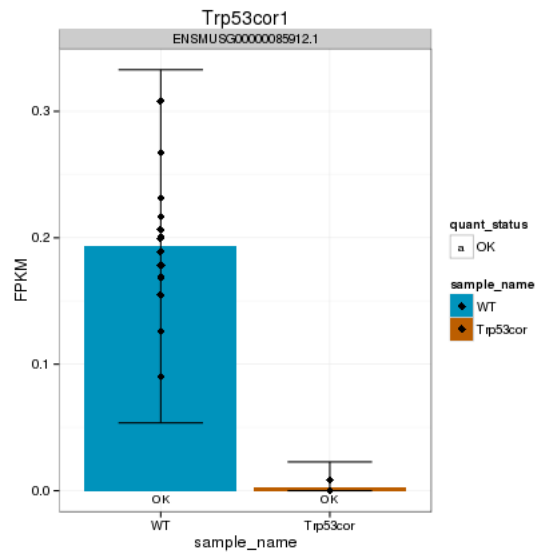
Endogenous lncRNA expression



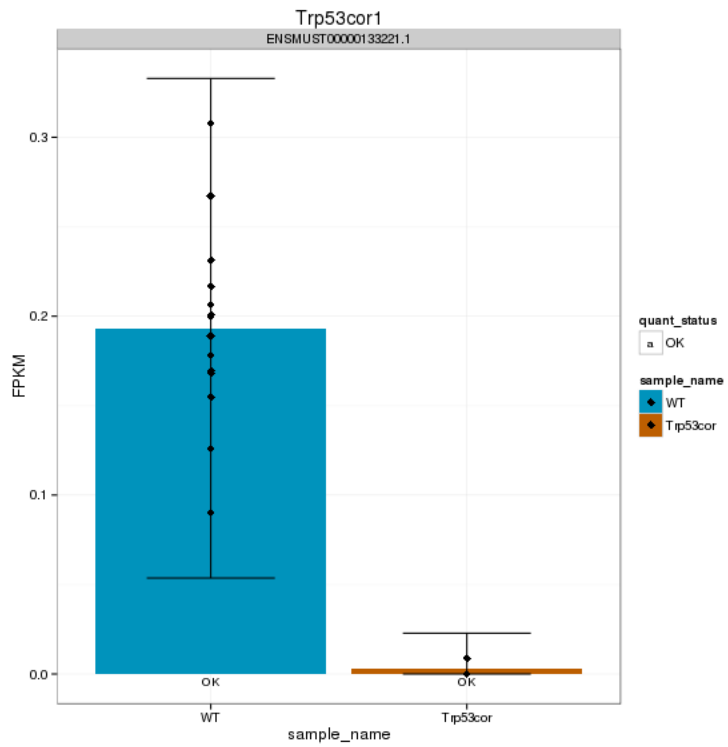
Endogenous expression of Trp53cor1 isoforms:



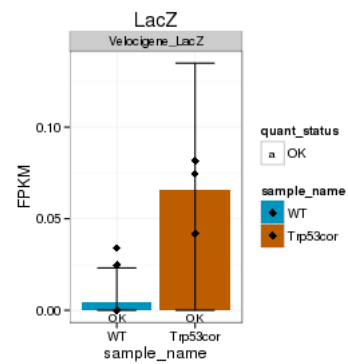
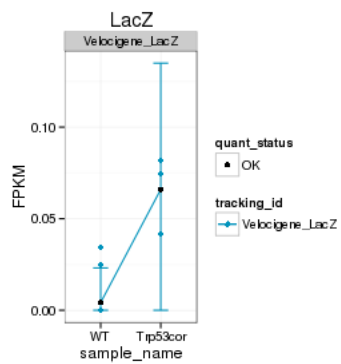
Barplot of gene expression:



Barplot of isoform expression:

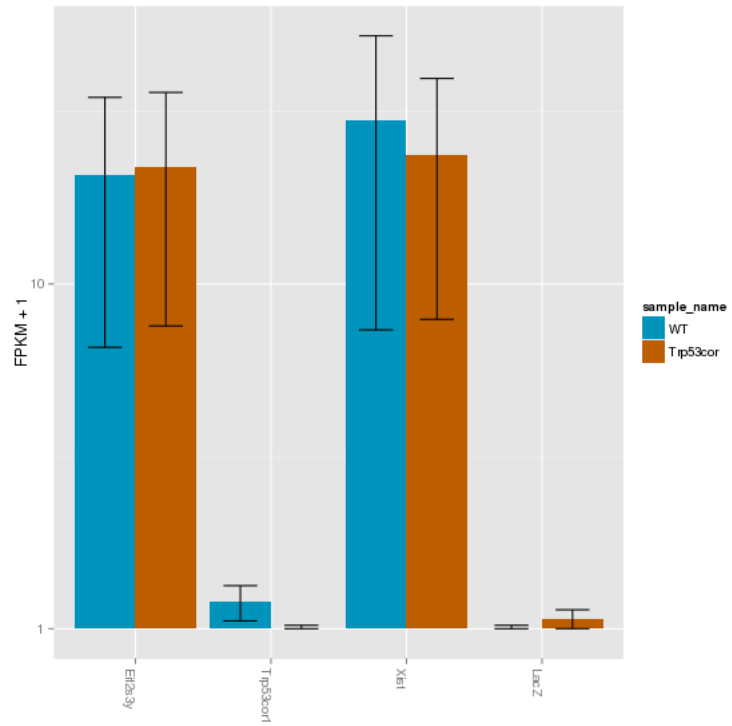


LacZ expression

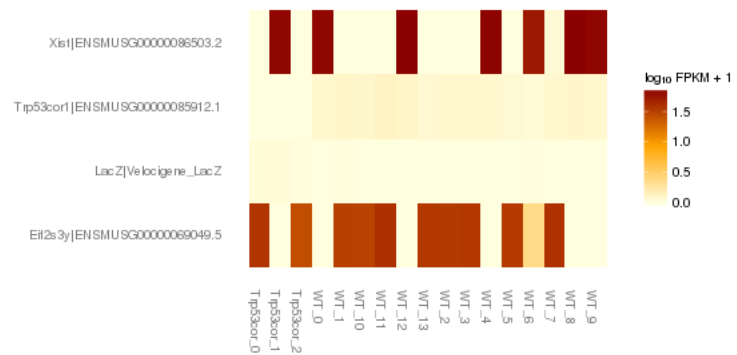


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

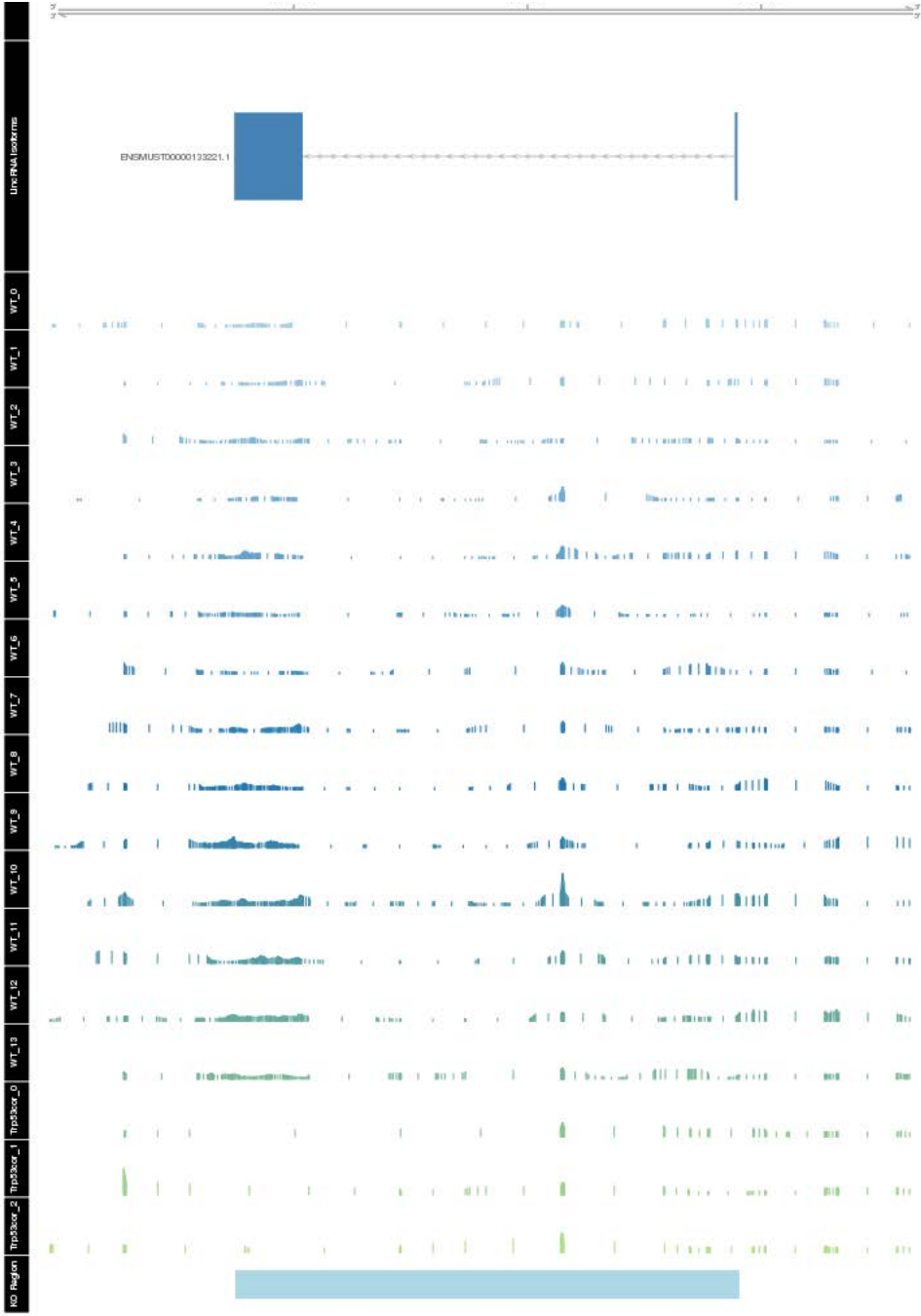


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

There are 69 significantly differentially expressed genes. They are:

geneAnnot\$gene_short_name

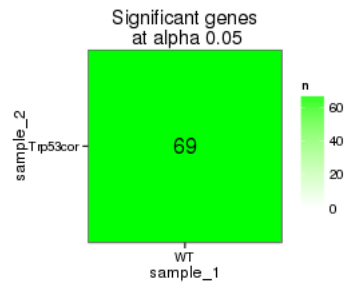
- 1 Gabra2
- 2 Arrdc2
- 3 Nes
- 4 Epha2
- 5 Khgrp
- 6 Odc1
- 7 Tjap1
- 8 Etnppl
- 9 Fabp7
- 10 Sgk1
- 11 Adi1
- 12 Serpina3n
- 13 Kcnv1
- 14 Cdkn1a
- 15 Park2
- 16 Mpc1
- 17 Flywch2
- 18 Mut
- 19 Enpp4
- 20 Ppil1
- 21 Cpne5
- 22 Pi16
- 23 Glo1
- 24 Prss41
- 25 Atp6v0c
- 26 Spsb3
- 27 Adamts10
- 28 Rps24
- 29 Rps6
- 30 Adamts9
- 31 Gkn3
- 32 A2m
- 33 Hddc3
- 34 Tlr13
- 35 Arhgap27
- 36 Rnps1
- 37 Lars2

38 3110052M02Rik
39 Decr2
40 Cramp1l
41 Gm129
42 Akap12
43 Itpkb

44 Lyp1a1
45 Fmo2
46 Rps19
47 Maff
48 Gm9493
49 Zfp758
50 Acp1
51 Hmga1
52 Tmem252
53 Pla2g4e
54 Tagap1
55 Amtl
56 Per2
57 H2-T22
58 Ide
59 Cox5b
60 H2-T23
61 Zfp760
62 H2-BI
63 9030025P20Rik
64 Hmga1-rs1
65 Apold1
66 AA465934
67 Rpl41
68 Gm9825
69 Tmem181b-ps

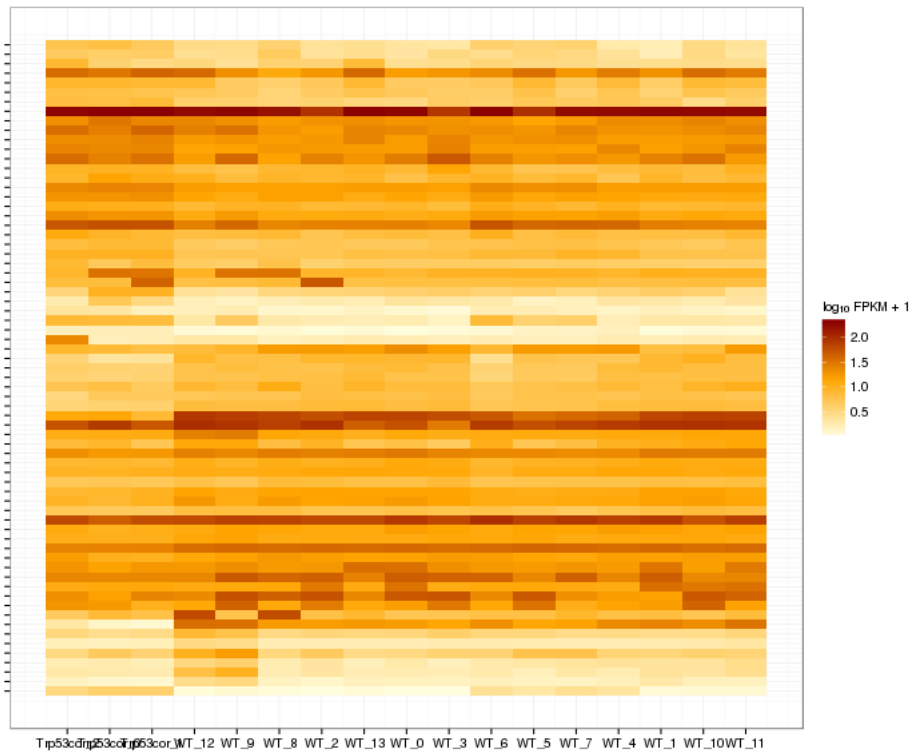
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)



Significant genes with expression >50fpkm (any condition):(turned off)

An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

Scatter plot of significant genes only:

```
## Error: One or more values of 'x' or 'y' are not valid sample names!
```

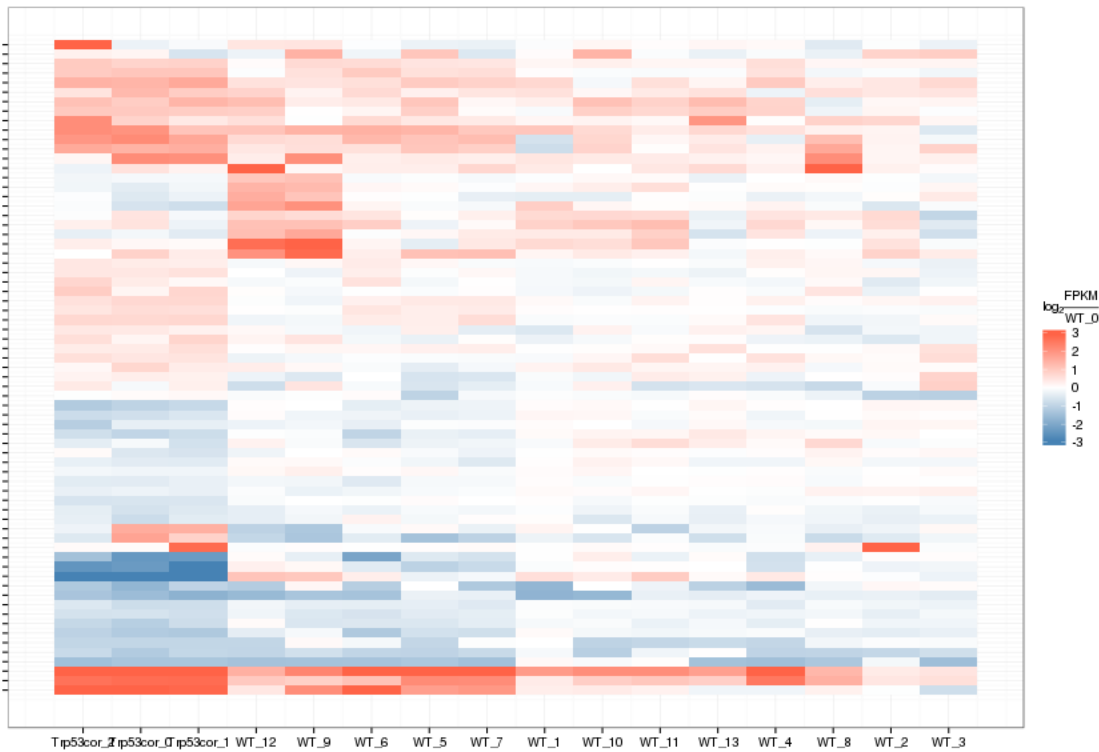
Volcano Plot

```
## Error: One or more values of 'x' or 'y' are not valid sample names!
```

Volcano plot with significant genes only:

```
## Error: One or more values of 'x' or 'y' are not valid sample names!
```

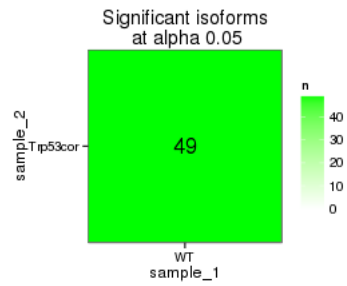
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



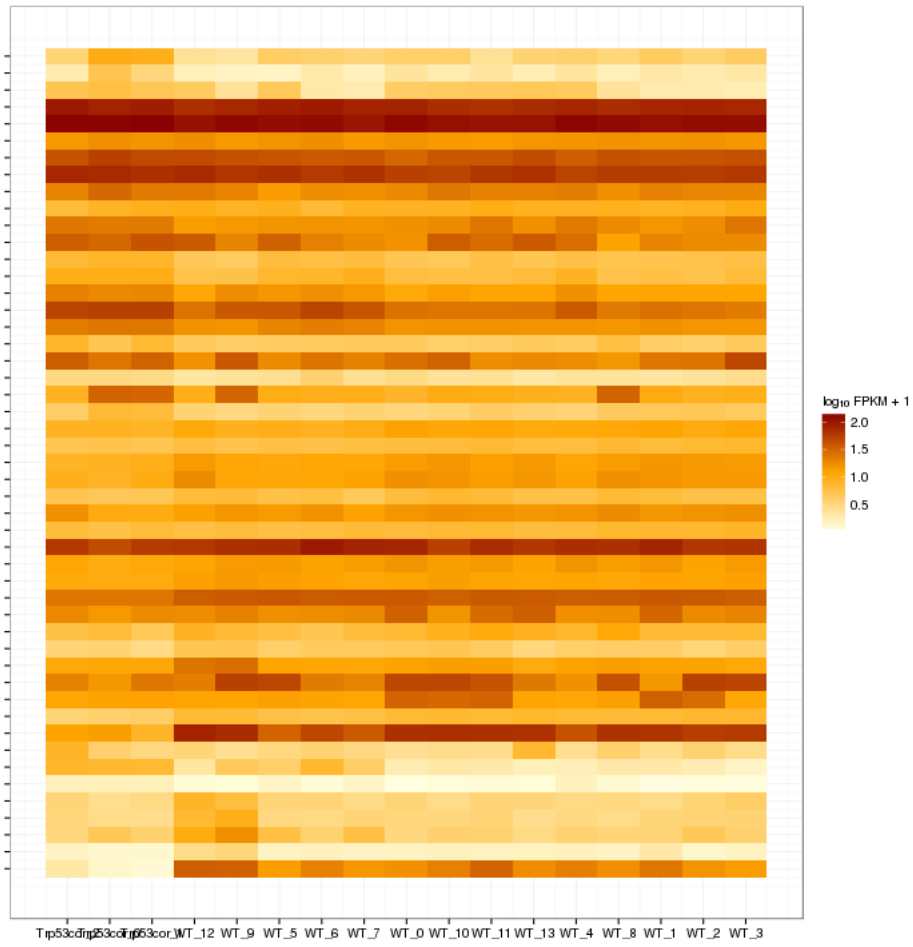
These isoforms are:

isoAnnot	gene_short_name
1	Gabra2
2	Rec8
3	Klf4
4	Nes
5	Prlr
6	Epha2
7	Khsrp
8	Etnppl
9	Fabp7
10	Serpina3n
11	Tfrc
12	Cdkn1a
13	Hcfc1r1
14	Mut
15	Ppil1
16	Cpne5
17	Glo1
18	Atp6v0c
19	Adamts10
20	Rps24
21	Gng13
22	Gkn3
23	A2m
24	Hddc3
25	Vps8
26	Rnps1

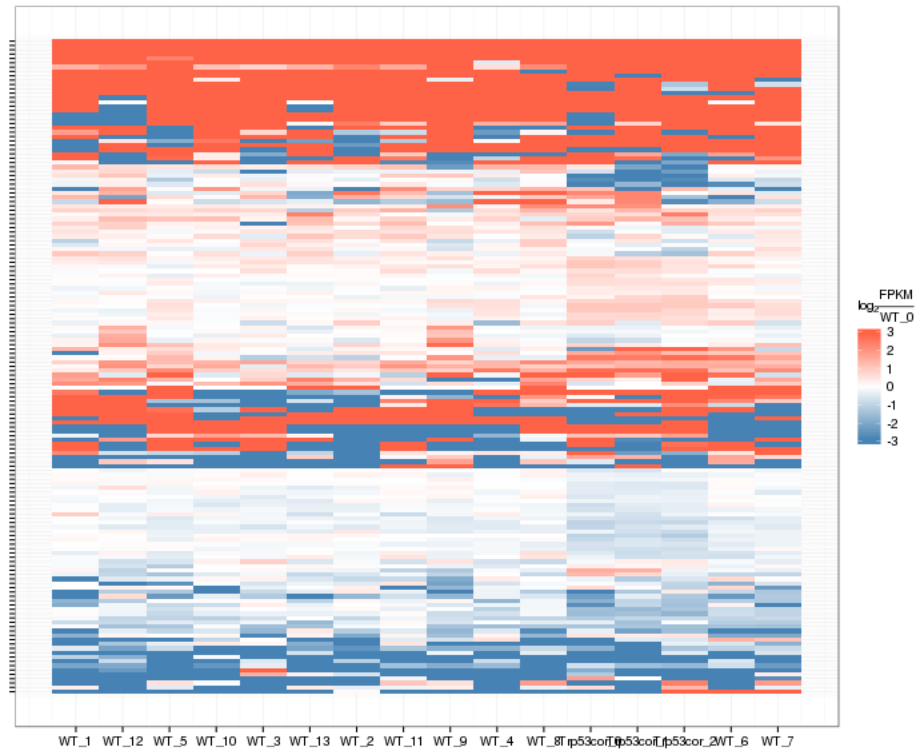
27 Lars2
28 3110052M02Rik
29 Decr2
30 Cramp11
31 Ano2
32 Akap12
33 Itpkb
34 Lyplal1
35 Fmo2
36 Bcor

37 Gm9493
38 Pla2g4e
39 Tagap1
40 Amtl
41 Ide
42 Zfp945
43 Zfp760
44 Fndc1
45 H2-BI
46 9030025P20Rik
47 Mog
48 Apold1
49 Gm9825

Gene-level DE isoform heatmap



Isoform foldchange heatmap by isoform:



Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

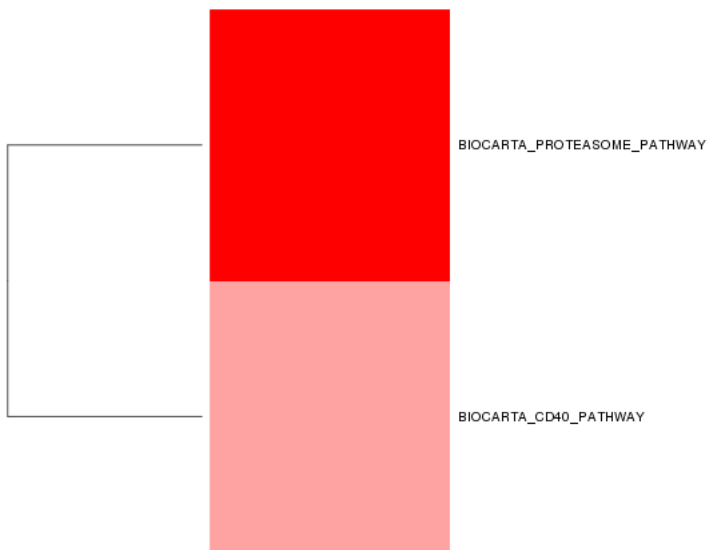
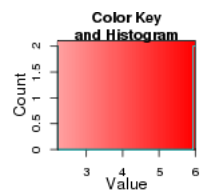
Gene/Pathway Analysis

GSEA

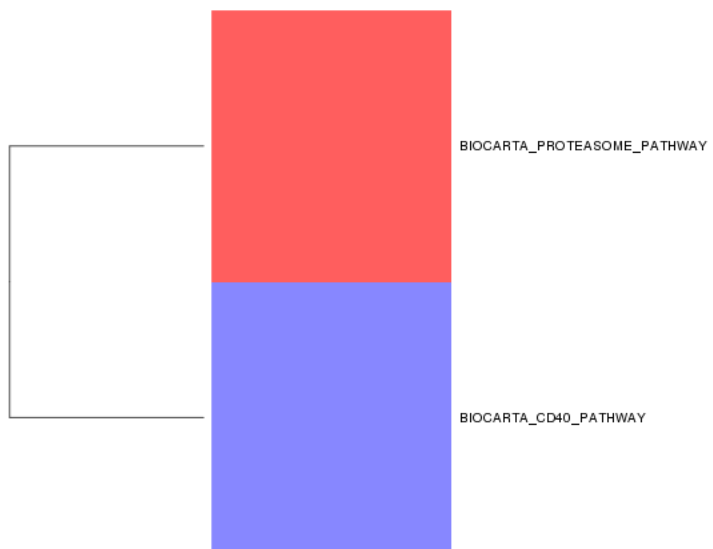
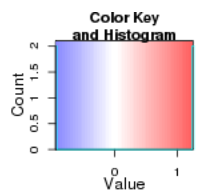
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

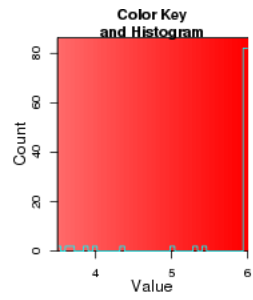
Biocarta enrichment:

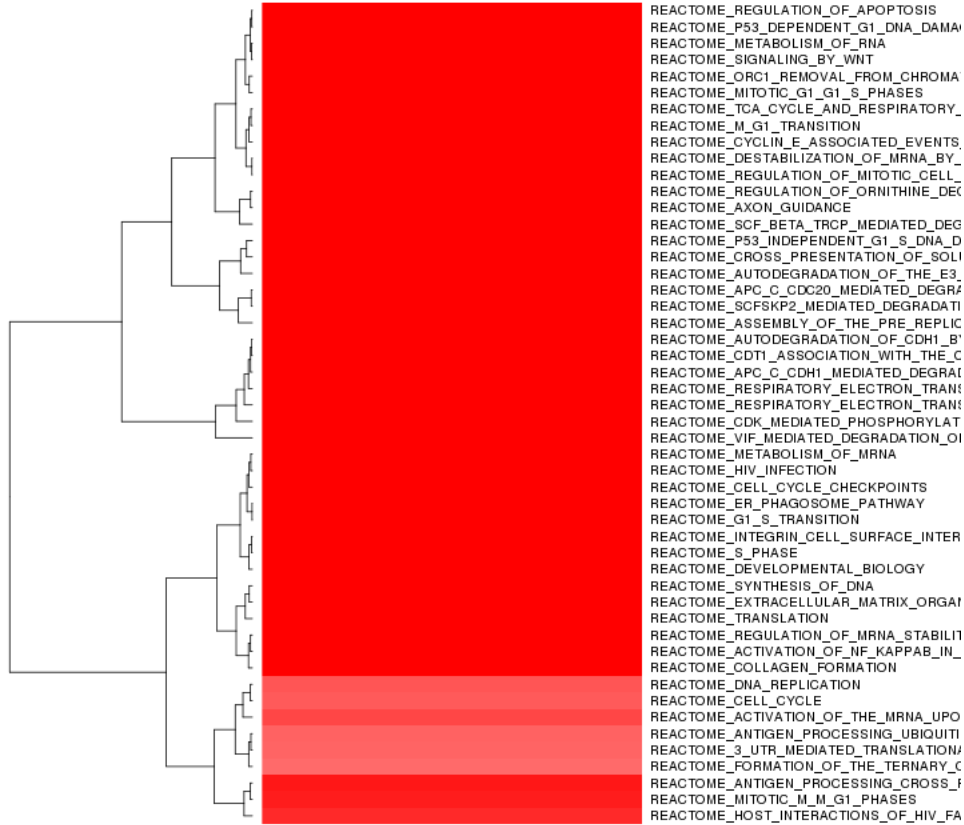


Biocarta zscore:

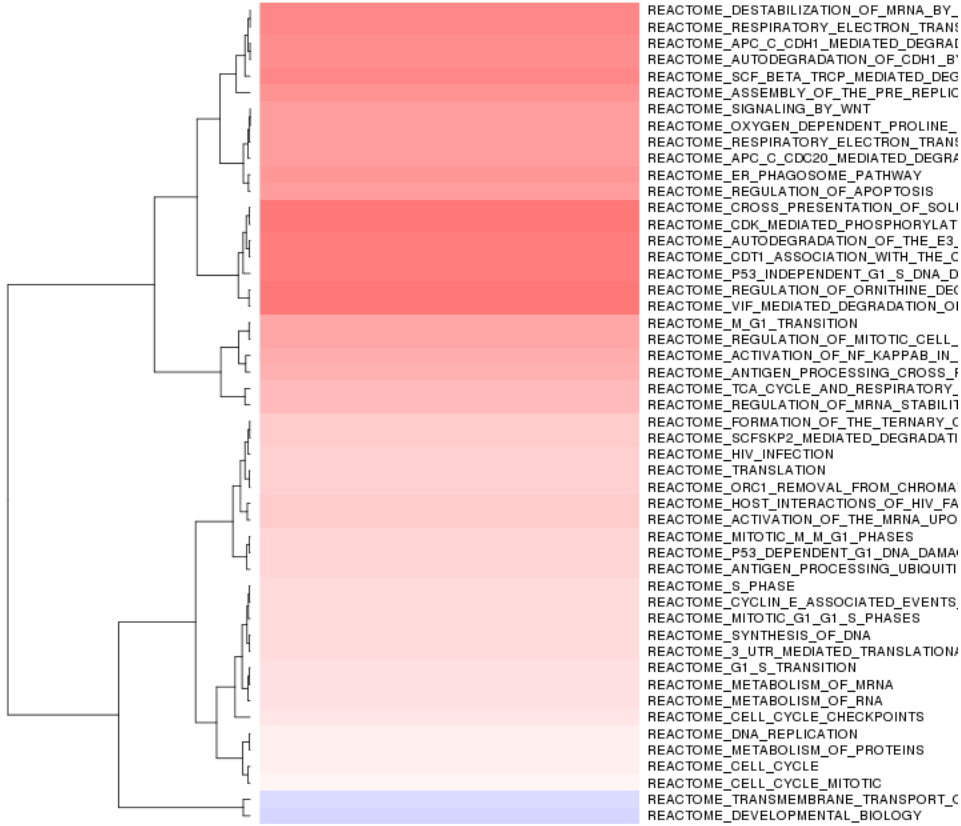
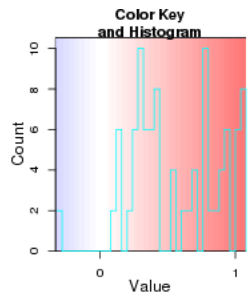


Reactome enrichment:

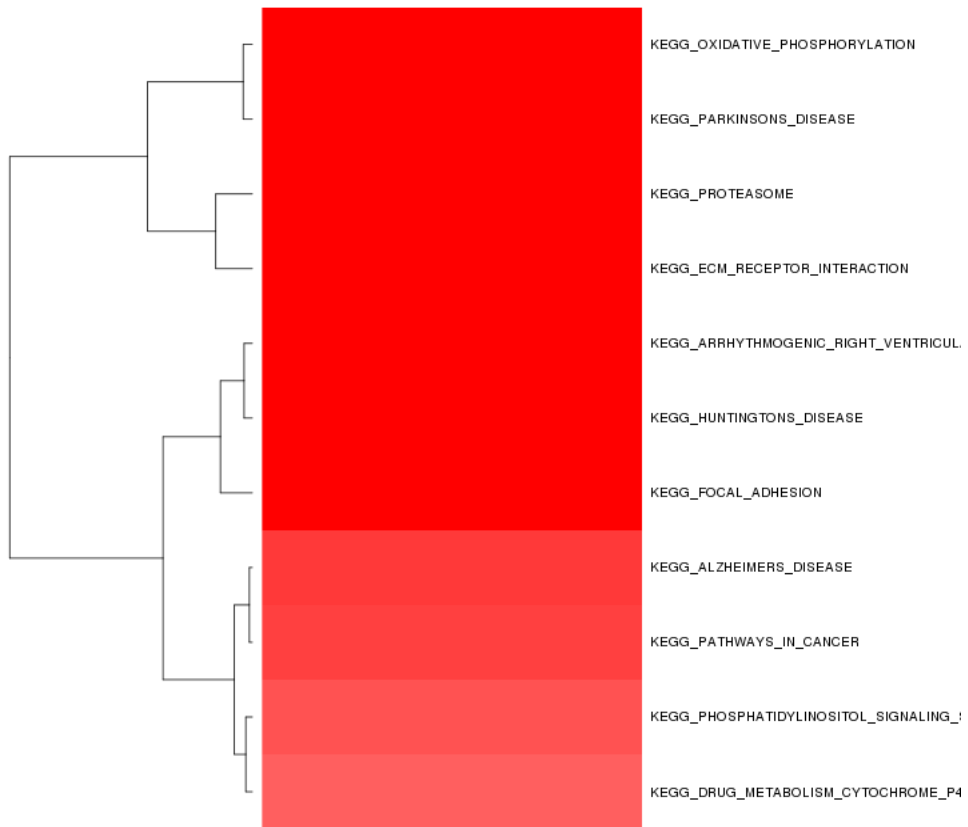
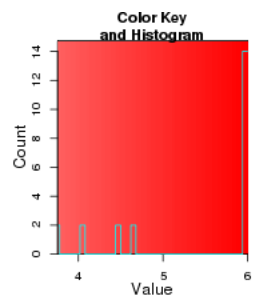




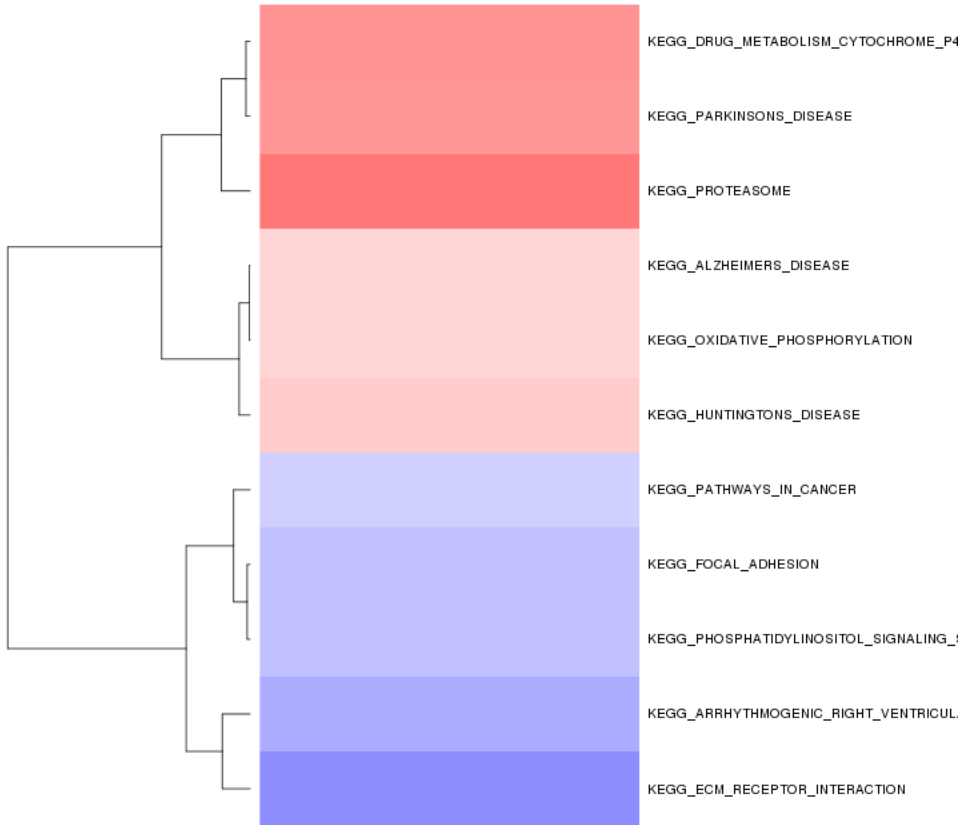
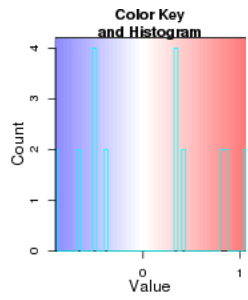
Reactome zscore:



Kegg enrichment:



Kegg zscore:



Interneuron enrichment:

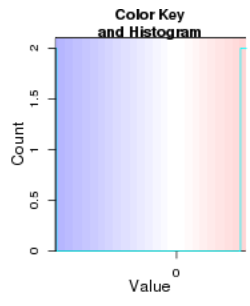
Error: no locations are finite

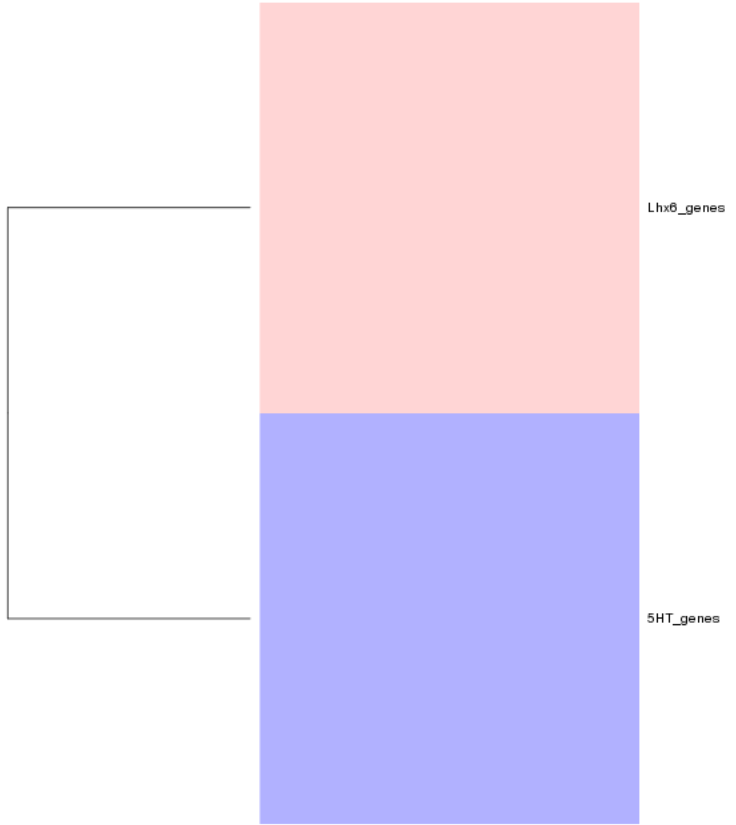


5HT_genes

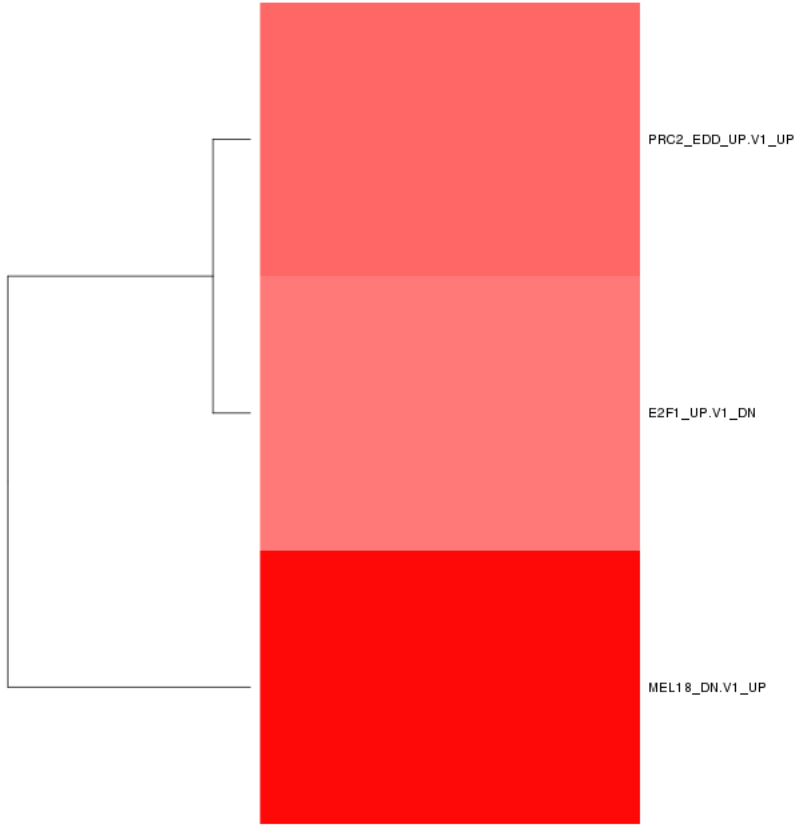
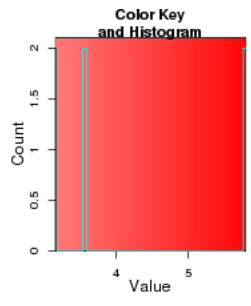
Lhx0_genes

Interneuron zscore:

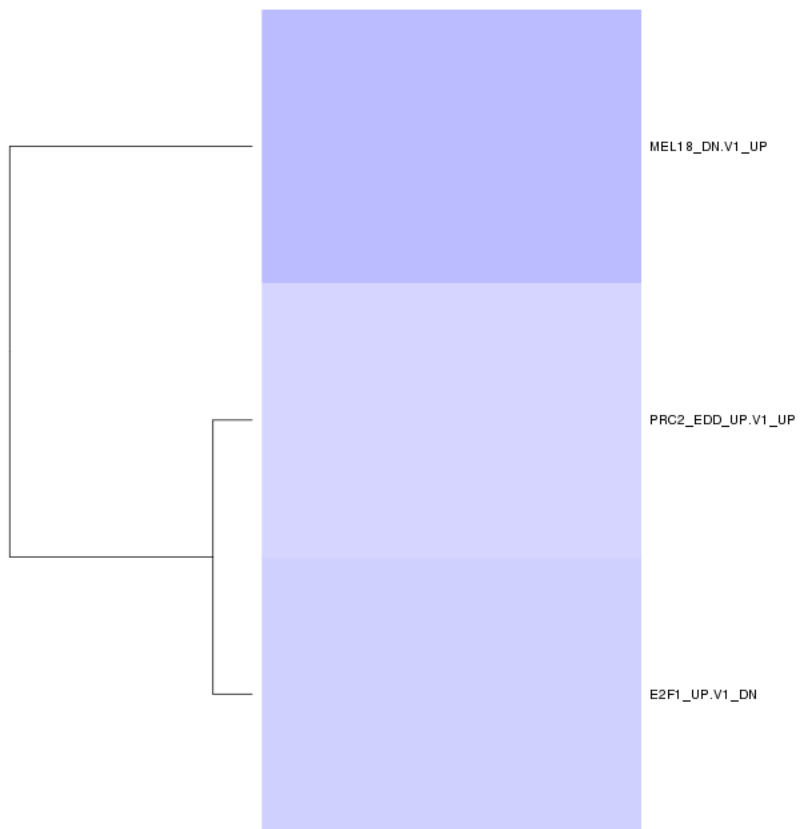
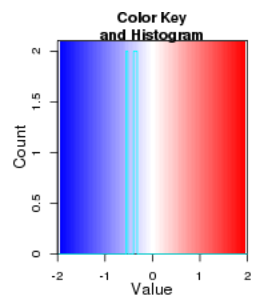




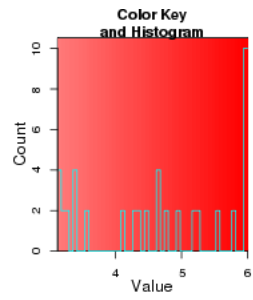
Oncogene enrichment:

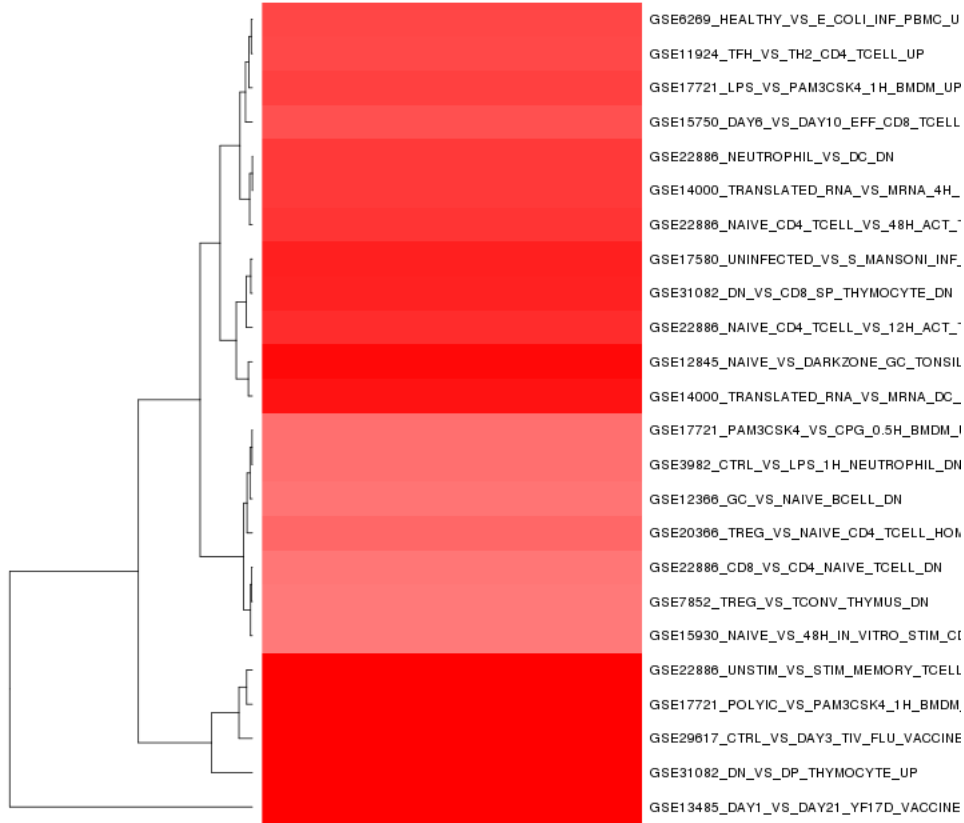


Oncogene zscore:

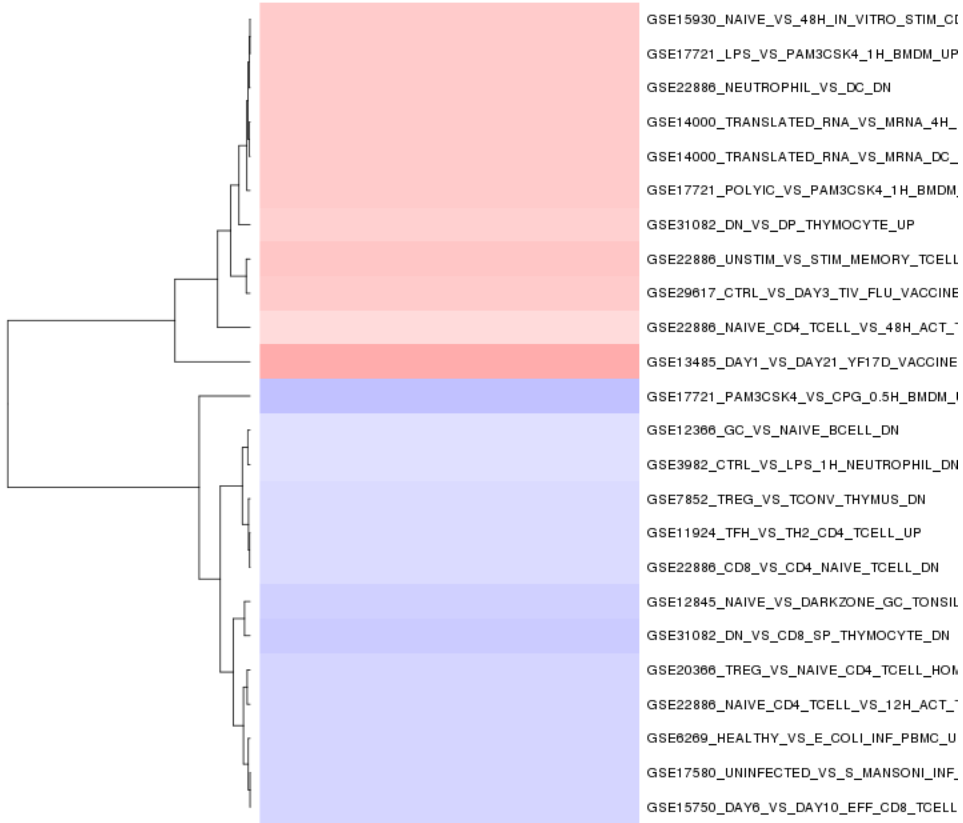
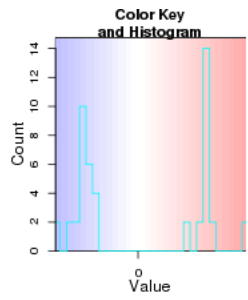


Immuno enrichment:



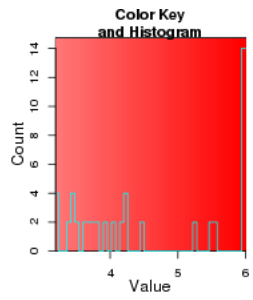


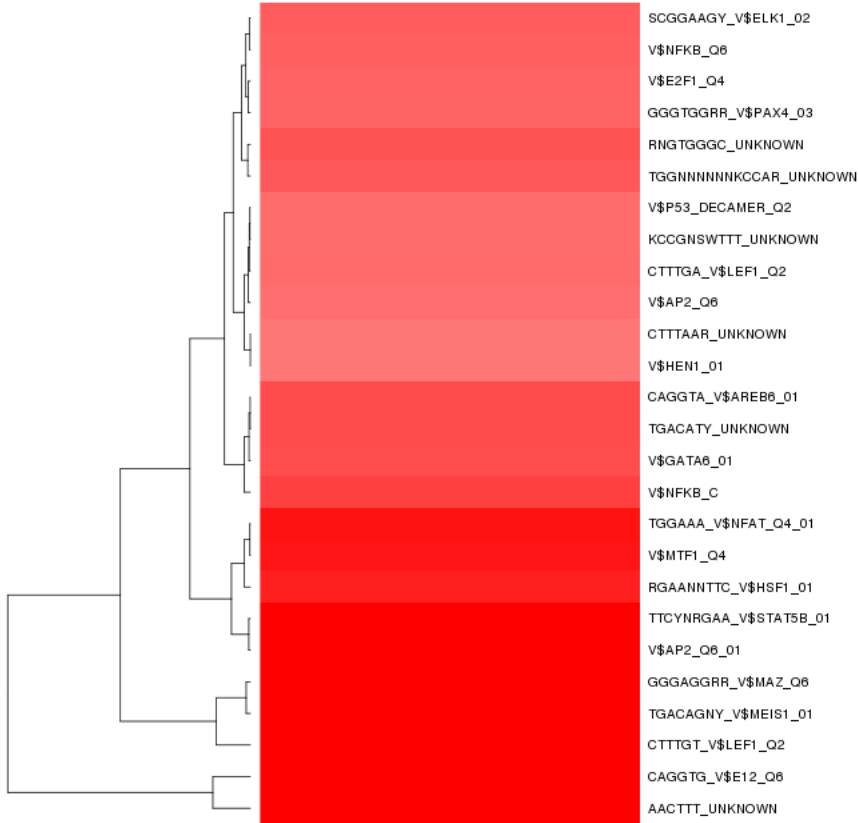
Immuno zscore:



TF enrichment:

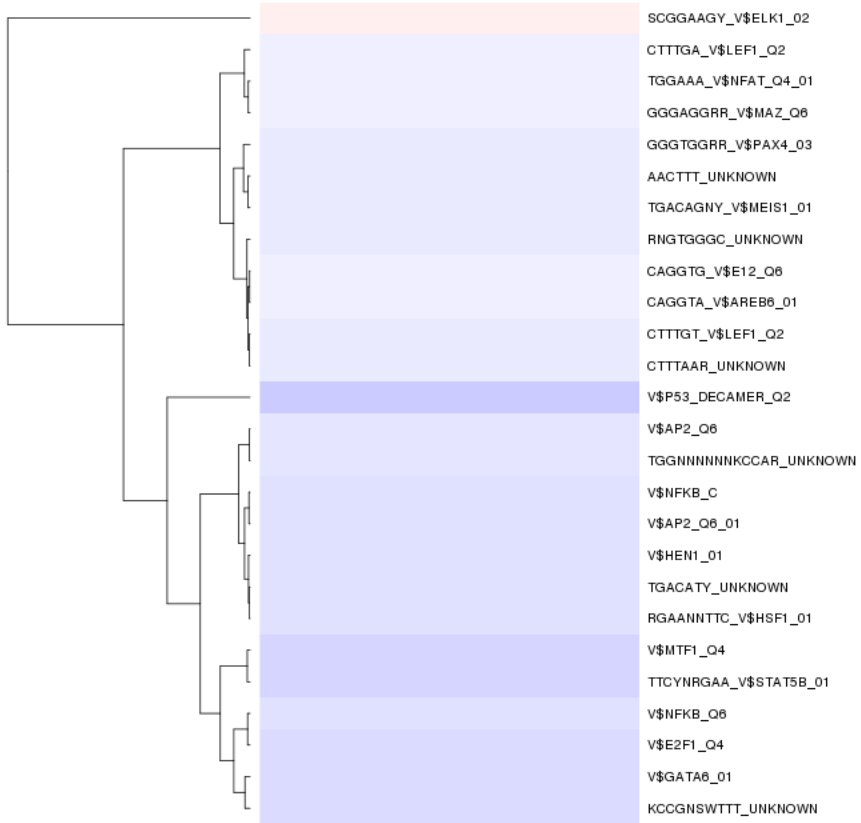
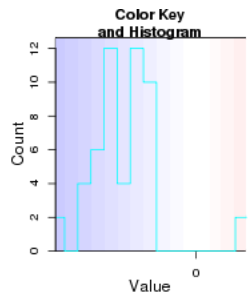
Error: subscript out of bounds





TF zscore:

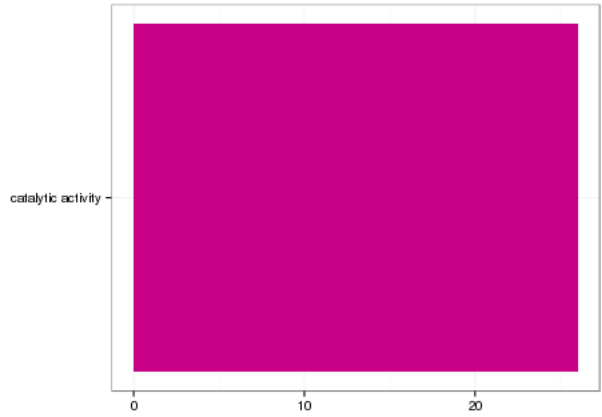
Error: subscript out of bounds



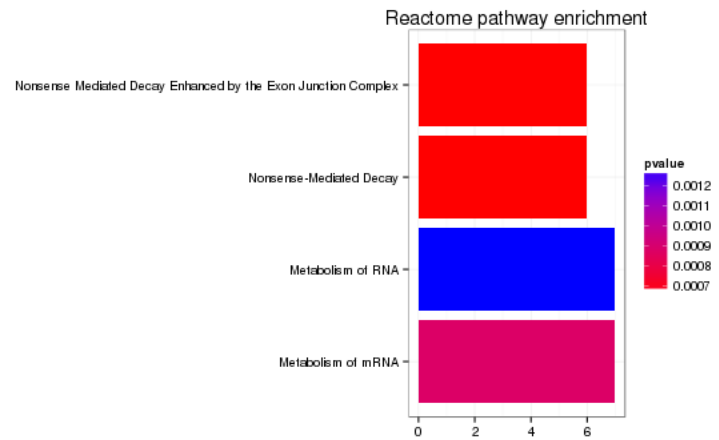
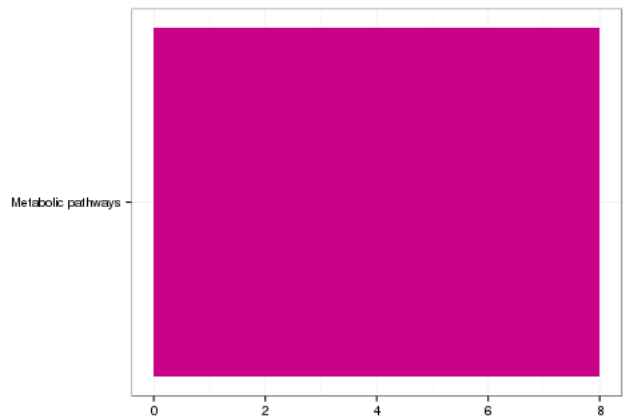
GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

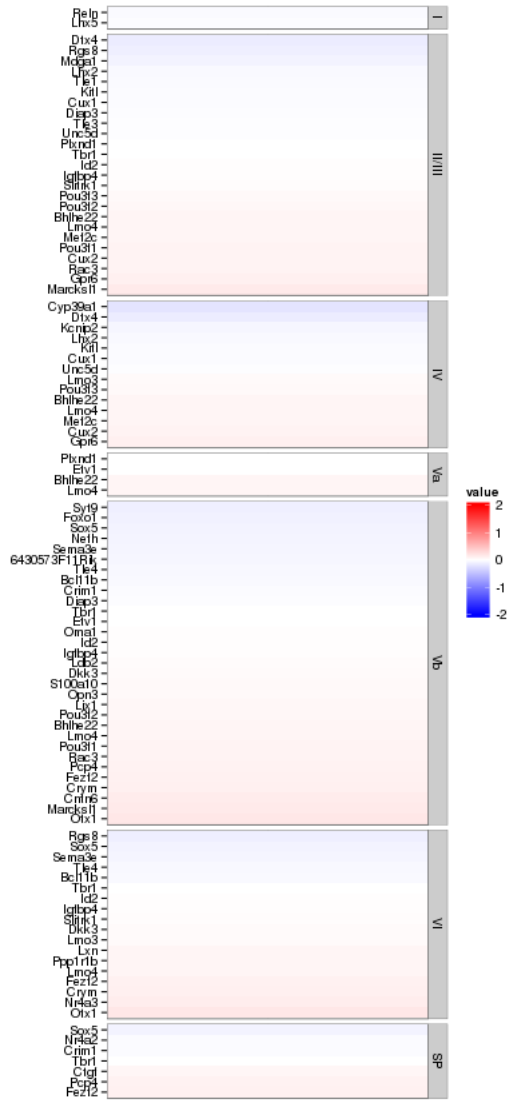
```
## Error: 'x' and 'units' must have length > 0
```



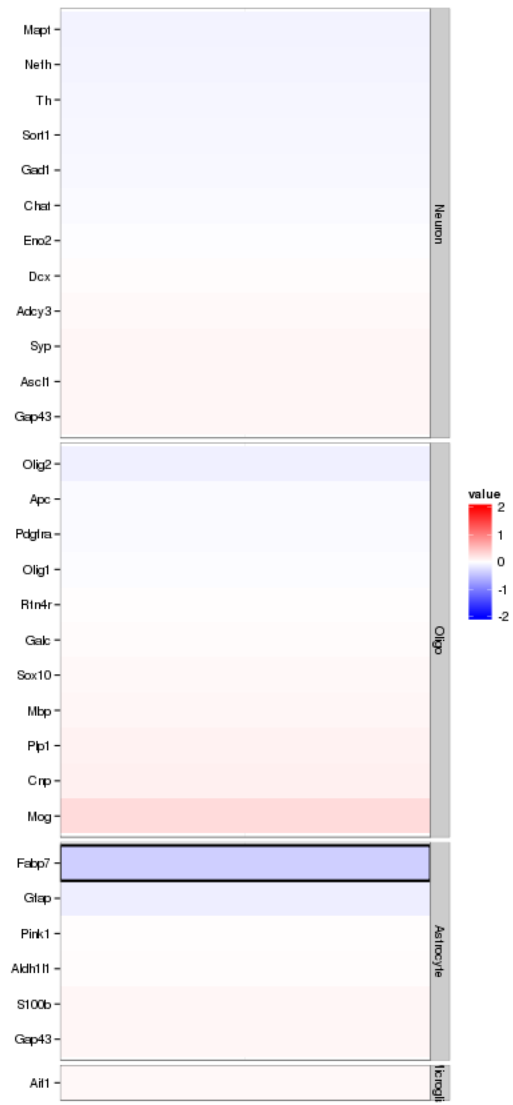
```
## Error: 'x' and 'units' must have length > 0
```



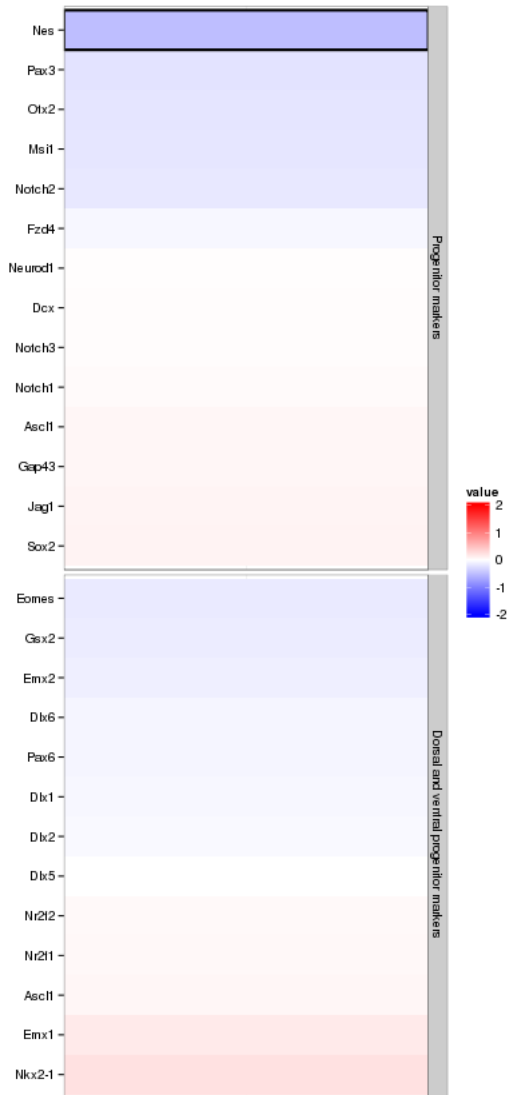
Enrichment or depletion for stage-specific cell cycle markers



Enrichment or depletion for specific neural cell types



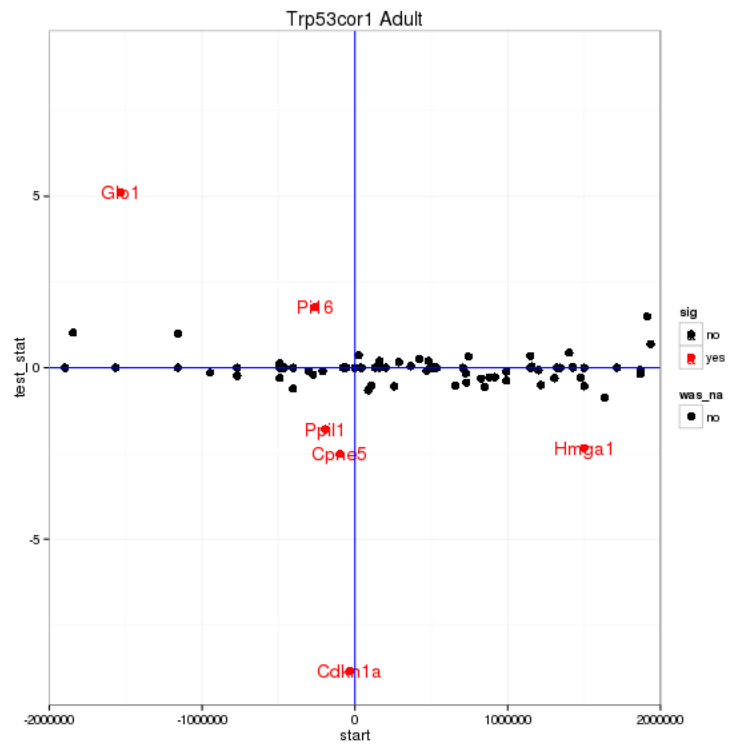
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpm(KO)}/\text{fpm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 6 genes significantly regulated in a region this size is: 0.0038



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR743
16 JR786
17 JR805

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR729/abundances.cxb	WT	0	WT_0	26334400.00	34671700.00	0.76	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR728/abundances.cxb	WT	1	WT_1	20329200.00	34671700.00	0.58	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR796/abundances.cxb	WT	2	WT_2	34089000.00	34671700.00	0.98	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR797/abundances.cxb	WT	3	WT_3	28103300.00	34671700.00	0.80	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR740/abundances.cxb	WT	4	WT_4	35808200.00	34671700.00	1.03	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR800/abundances.cxb	WT	5	WT_5	37012200.00	34671700.00	1.07	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR827/abundances.cxb	WT	6	WT_6	27786600.00	34671700.00	0.81	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR778/abundances.cxb	WT	7	WT_7	39541900.00	34671700.00	1.15	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR734/abundances.cxb	WT	8	WT_8	34480600.00	34671700.00	1.01	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR802/abundances.cxb	WT	9	WT_9	45467400.00	34671700.00	1.30	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR803/abundances.cxb	WT	10	WT_10	52130400.00	34671700.00	1.50	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR735/abundances.cxb	WT	11	WT_11	34994400.00	34671700.00	1.02	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR785/abundances.cxb	WT	12	WT_12	34173600.00	34671700.00	0.97	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR781/abundances.cxb	WT	13	WT_13	41538900.00	34671700.00	1.21	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR743/abundances.cxb	Trp53cor	0	Trp53cor_0	32358500.00	34671700.00	0.94	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR786/abundances.cxb	Trp53cor	1	Trp53cor_1	38185400.00	34671700.00	1.10	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR805/abundances.cxb	Trp53cor	2	Trp53cor_2	41355100.00	34671700.00	1.19	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```

```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1      KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1      reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1        zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Trp53cor -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Trp53cor_vs_WT_Adult /n/rinn_data1/seq/lgoff/Projects/BrainMap/c
## 2
## 3
## 4
## 5
```

Trp53cor1 KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Trp53cor1.

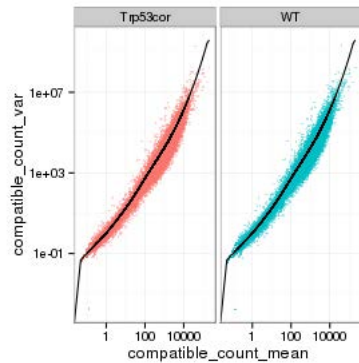
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

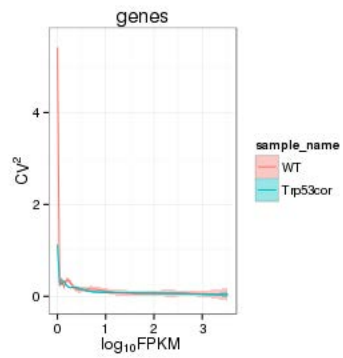
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



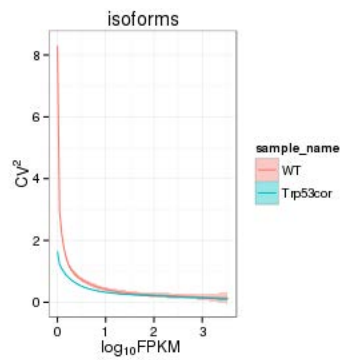
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

Genes:



Isoforms:



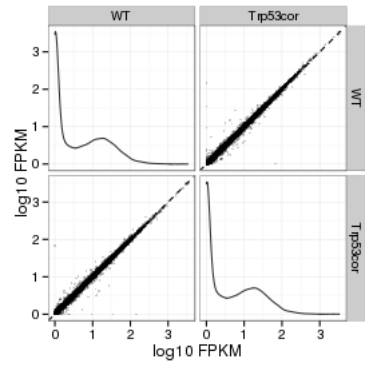
MvA plot

```
## Error: non-numeric argument to binary operator
```

MvA plot counts

```
## Error: non-numeric argument to binary operator
```

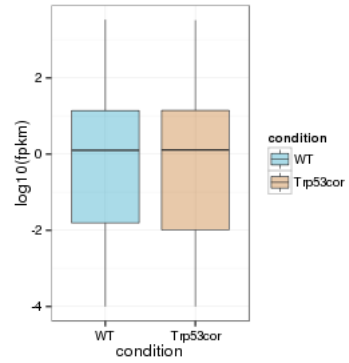
Scatterplot matrix



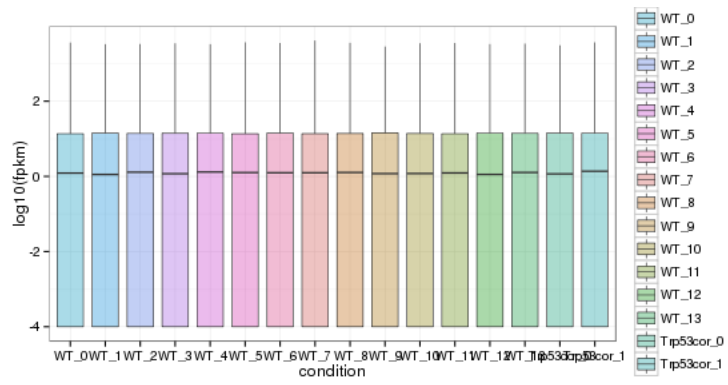
Distributions

Boxplots

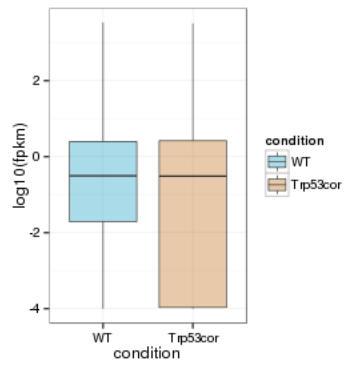
Boxplot (genes)



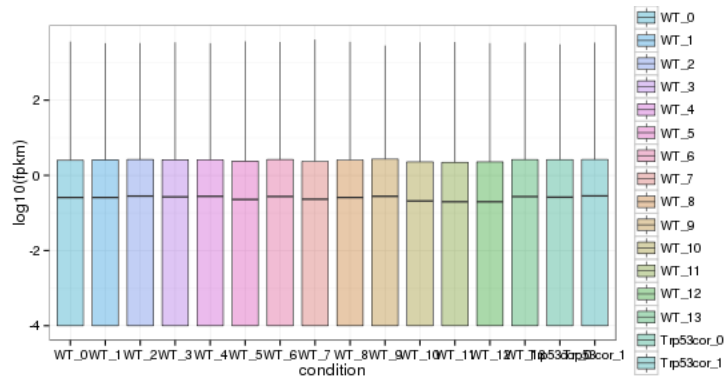
Boxplot (genes, replicates)



Boxplot (isoforms)

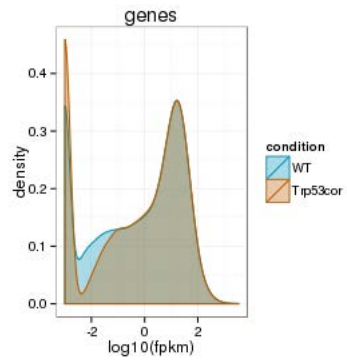


Boxplot (isoforms, replicates)

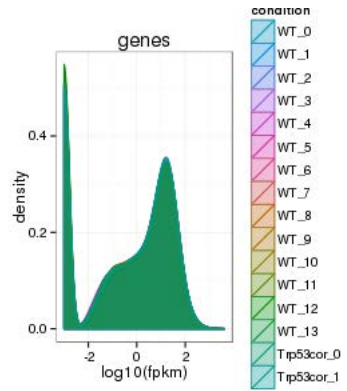


Density

Density (genes)



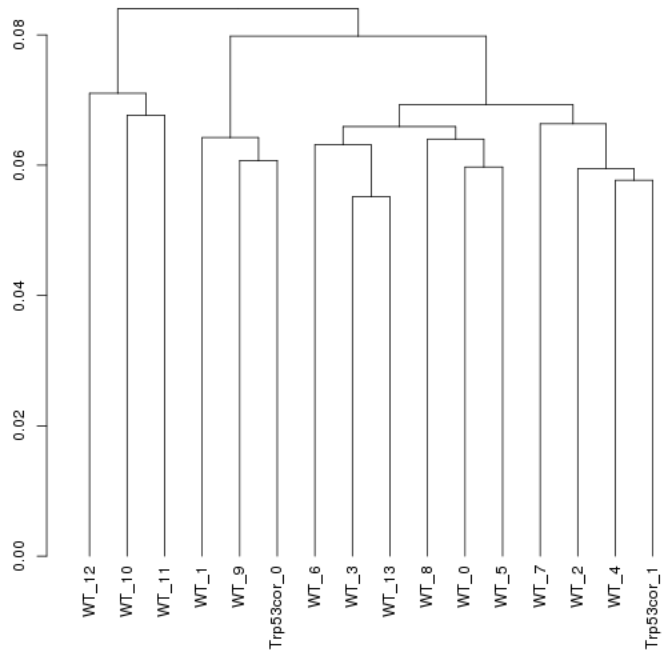
Density (genes, replicates)



Clustering

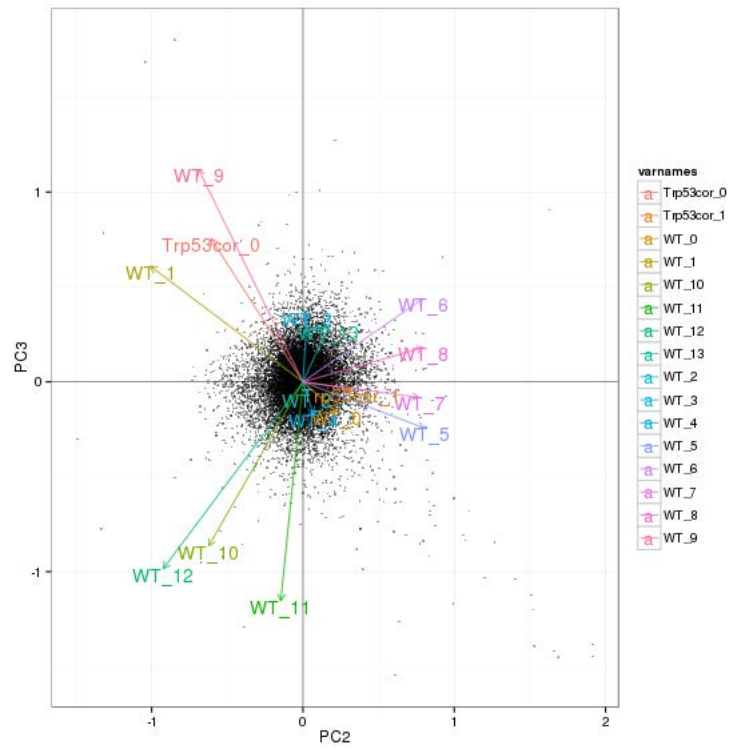
Replicate Clusters

All genes(cuff)

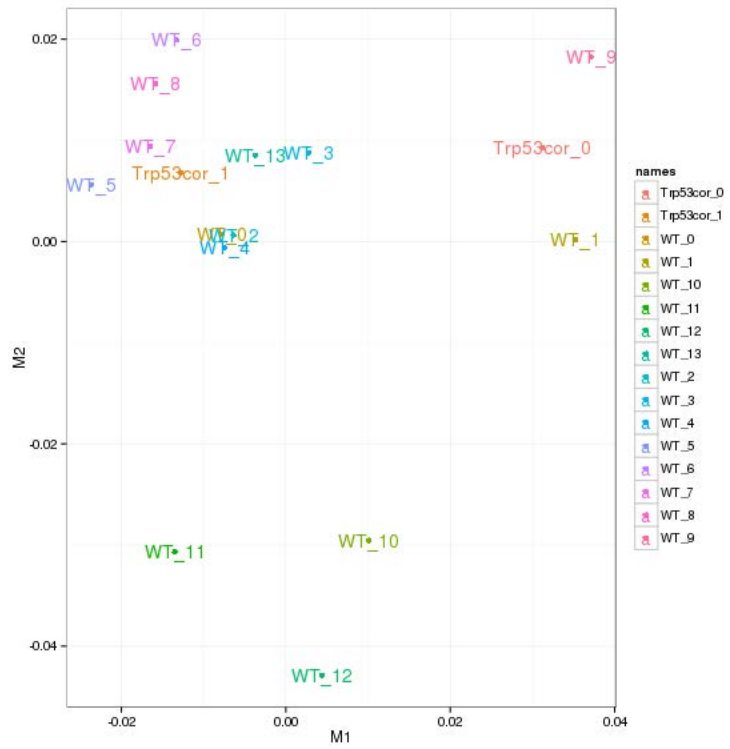


NULL

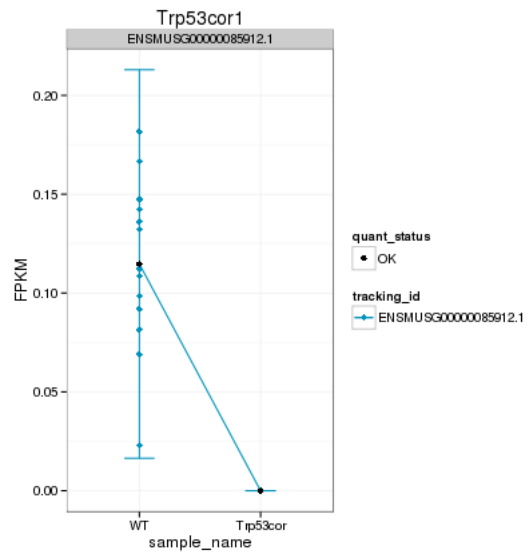
PCA (genes)



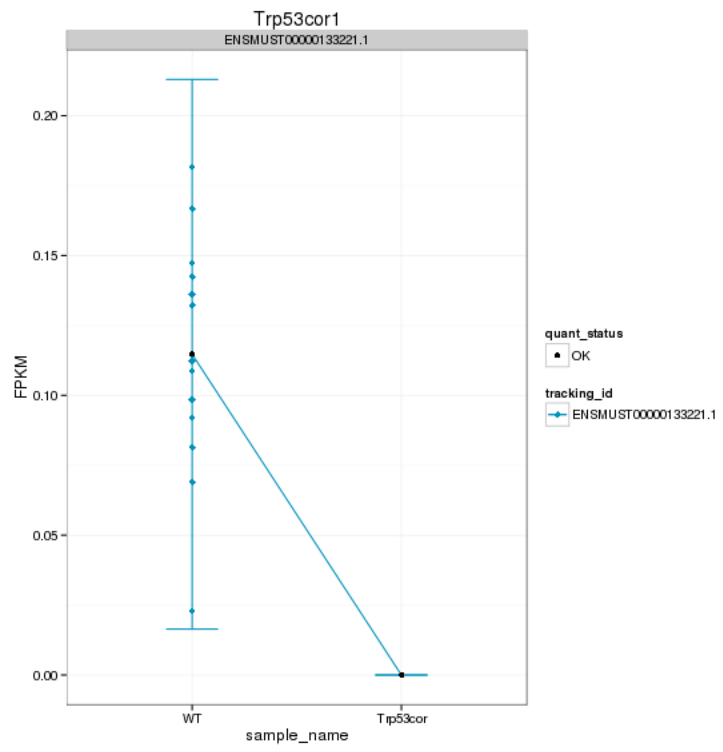
MDS (genes)



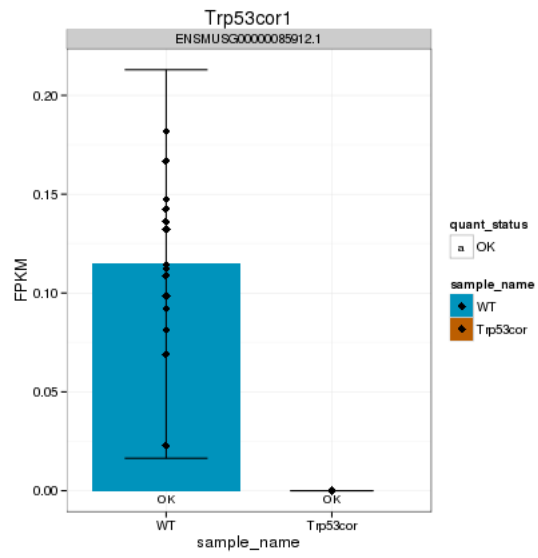
KO assessment
Endogenous IncRNA expression



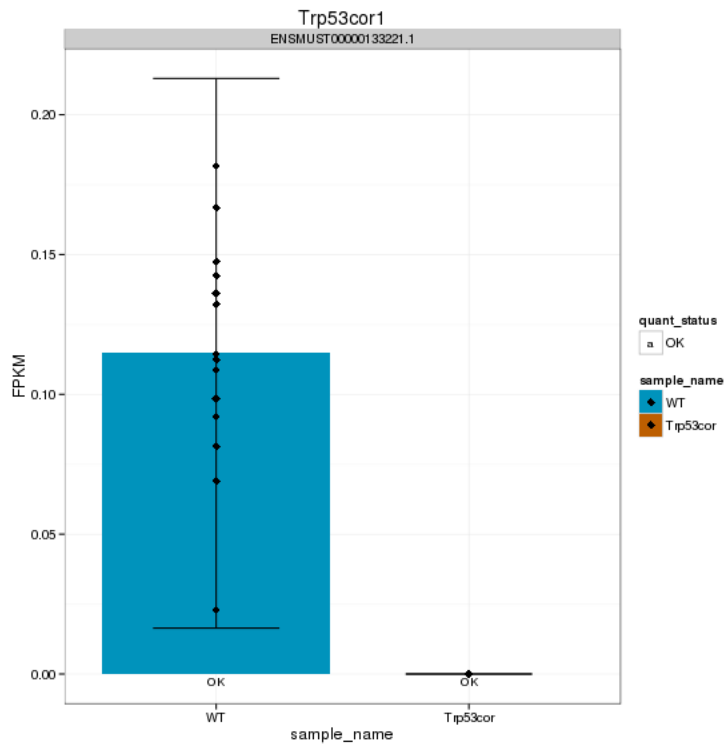
Endogenous expression of Trp53cor1 isoforms:



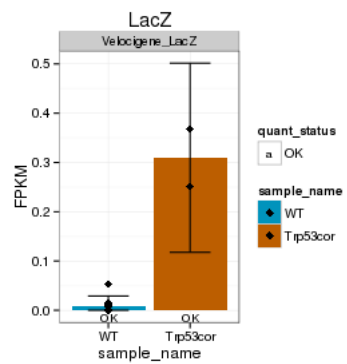
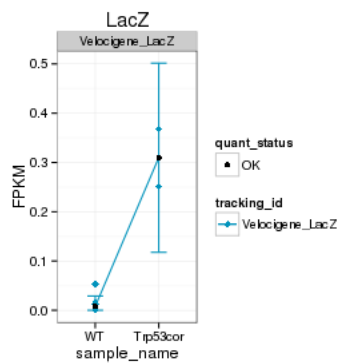
Barplot of gene expression:



Barplot of isoform expression:

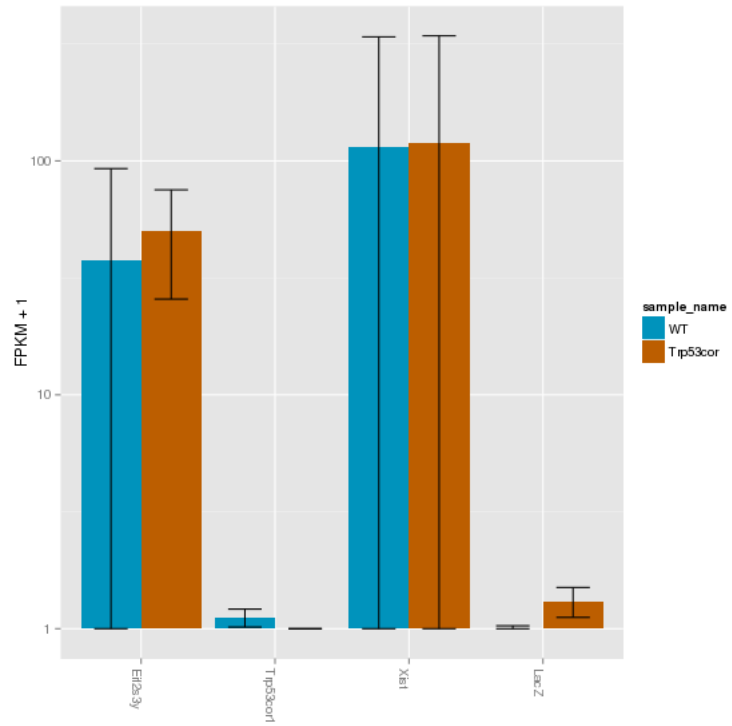


LacZ expression

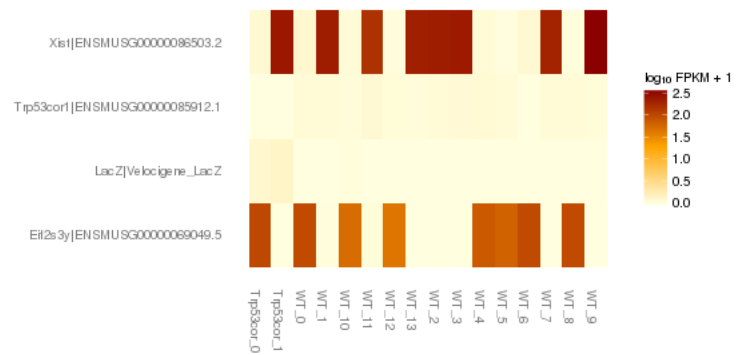


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):

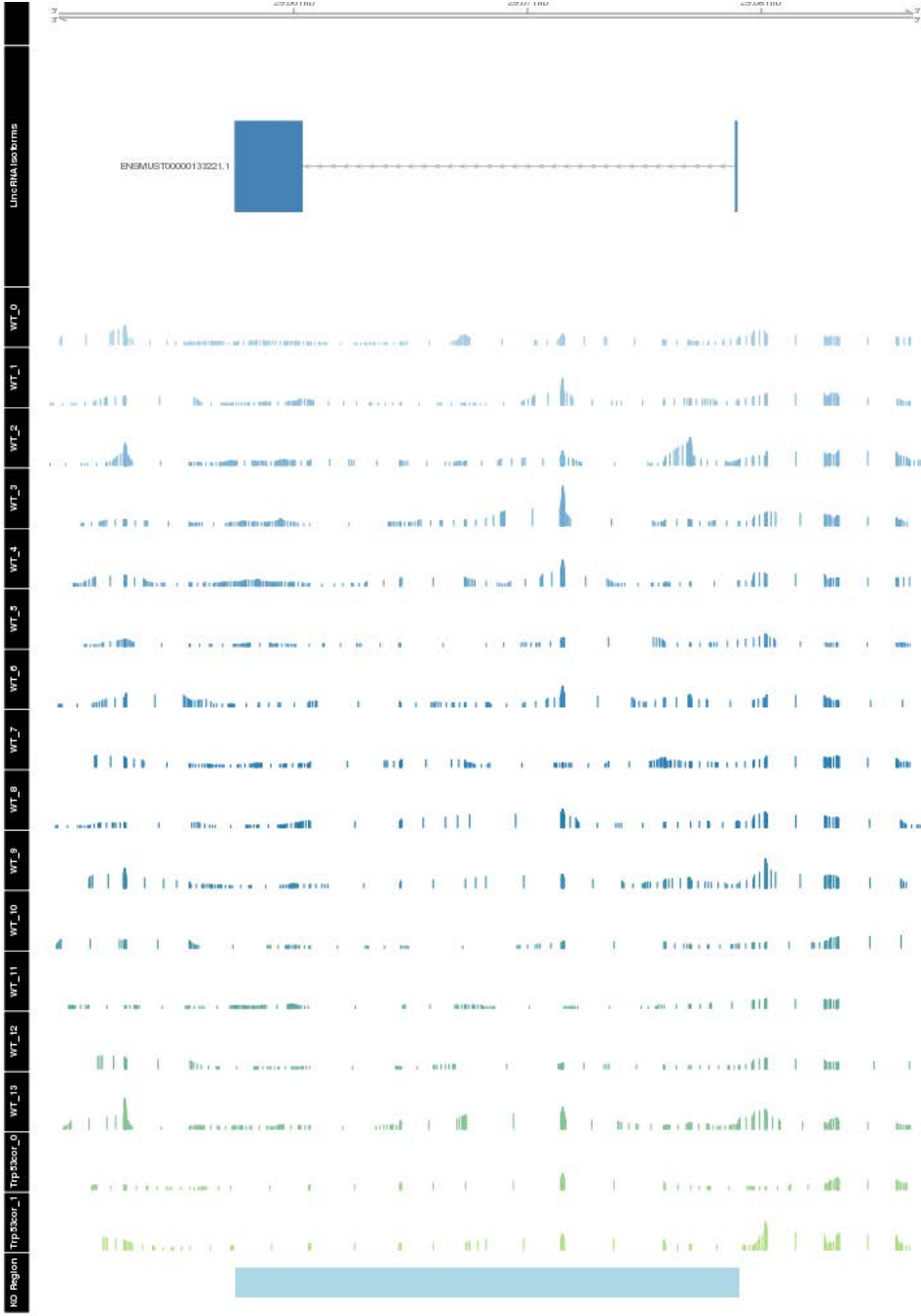


Expression heatmap:



Track visualization





Differential Analysis

Differential Genes

```
## Error: RS-DBI driver: (error in statement: near ")": syntax error)
```

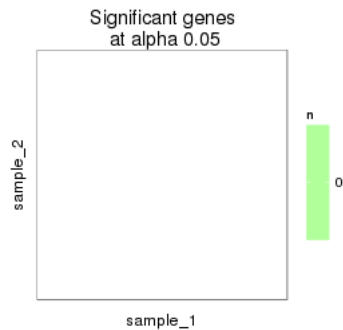
```
## Error: error in evaluating the argument 'object' in selecting a method for function 'annotation': Error: object 'sigGenes' not found
```

There are 0 significantly differentially expressed genes. They are:

```
[1] "No sig genes!"
```

Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

```
## Error: error in evaluating the argument 'object' in selecting a method for function 'csHeatmap': Error: object 'sigGenes' not found
```

Significant genes with expression >50fpkm (any condition):(turned off)

An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

Scatter plot of significant genes only:

```
## Error: error in evaluating the argument 'object' in selecting a method for function 'csScatter': Error: object 'sigGenes' not found
```

Volcano Plot

```
## Error: One or more values of 'x' or 'y' are not valid sample names!
```

Volcano plot with significant genes only:

```
## Error: error in evaluating the argument 'object' in selecting a method for function 'csvolcano': Error: object 'sigGenes' not found
```

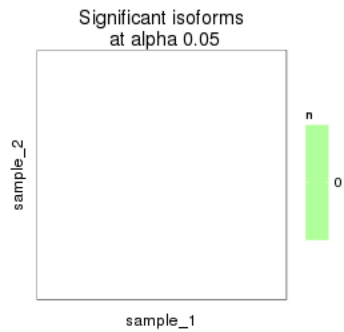
FoldChange Heatmap

```
## Error: error in evaluating the argument 'object' in selecting a method for function 'csFoldChangeHeatmap': Error: object 'sigGenes' not found
```

Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

```
[1] "no sig isoforms"
```

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

Biocarta enrichment:

```
## Error: `x` must have at least 2 rows and 2 columns
```

Biocarta zscore:

```
## Error: incorrect number of dimensions
```

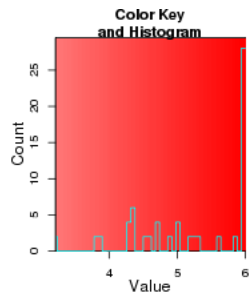
```
## Error: object 'x_ordered' not found
```

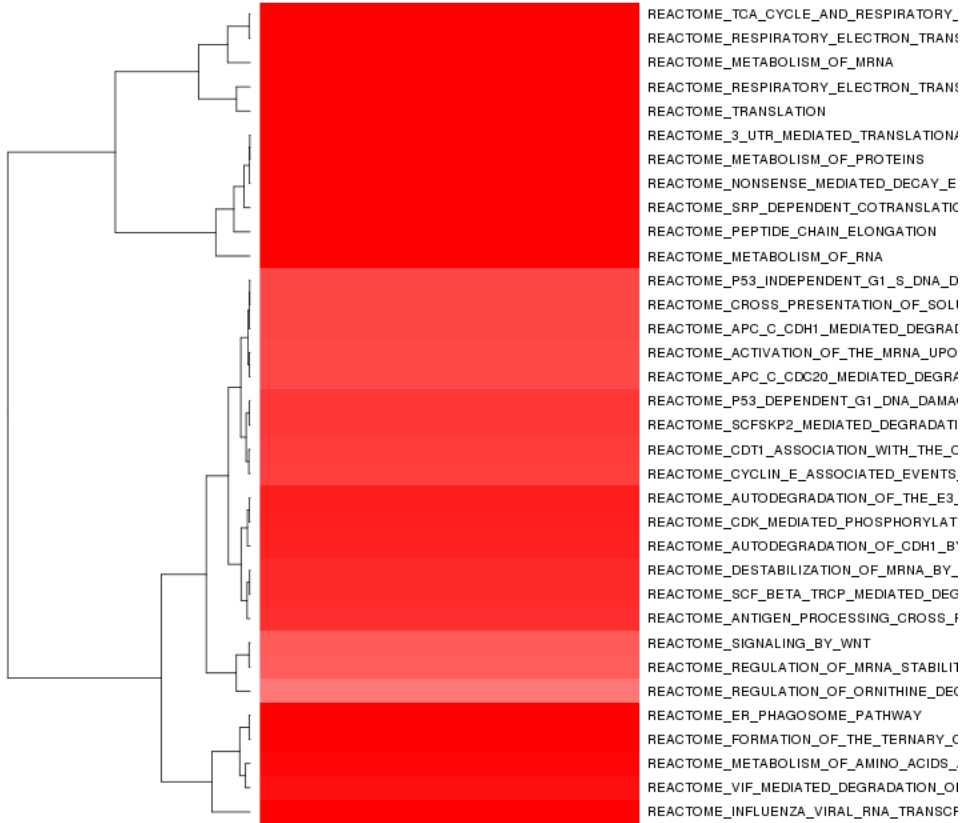
```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

```
## Error: object 'x_ordered' not found
```

Reactome enrichment:

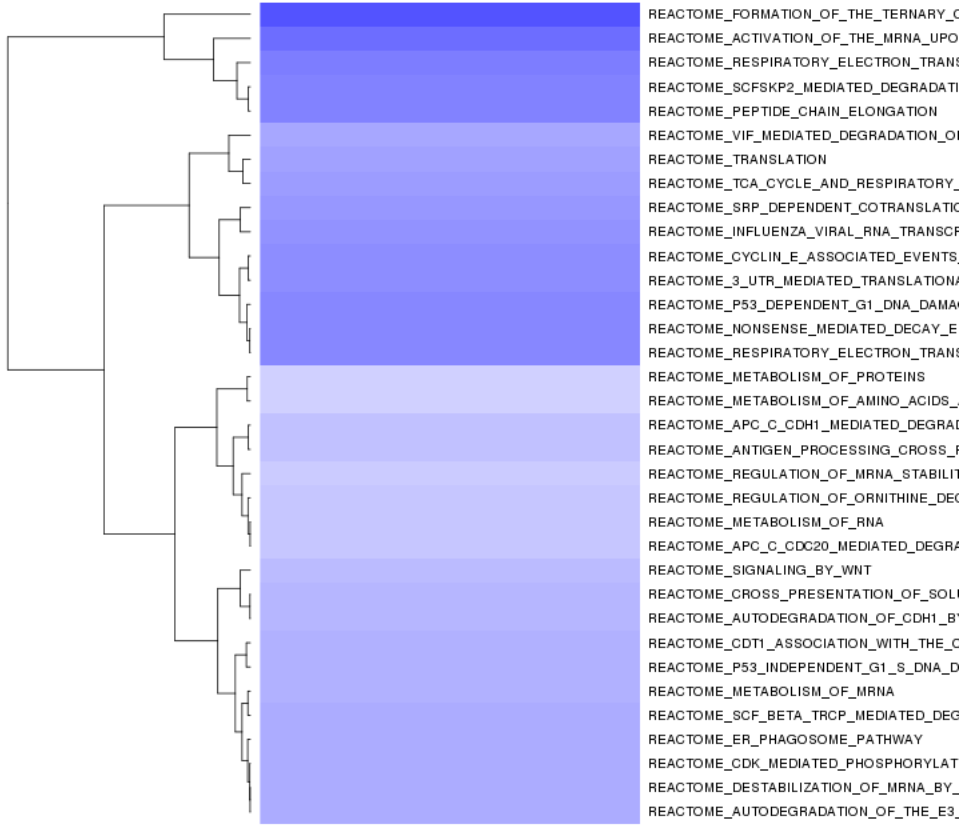
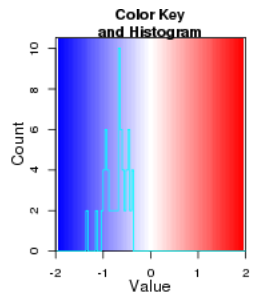
```
## Error: subscript out of bounds
```



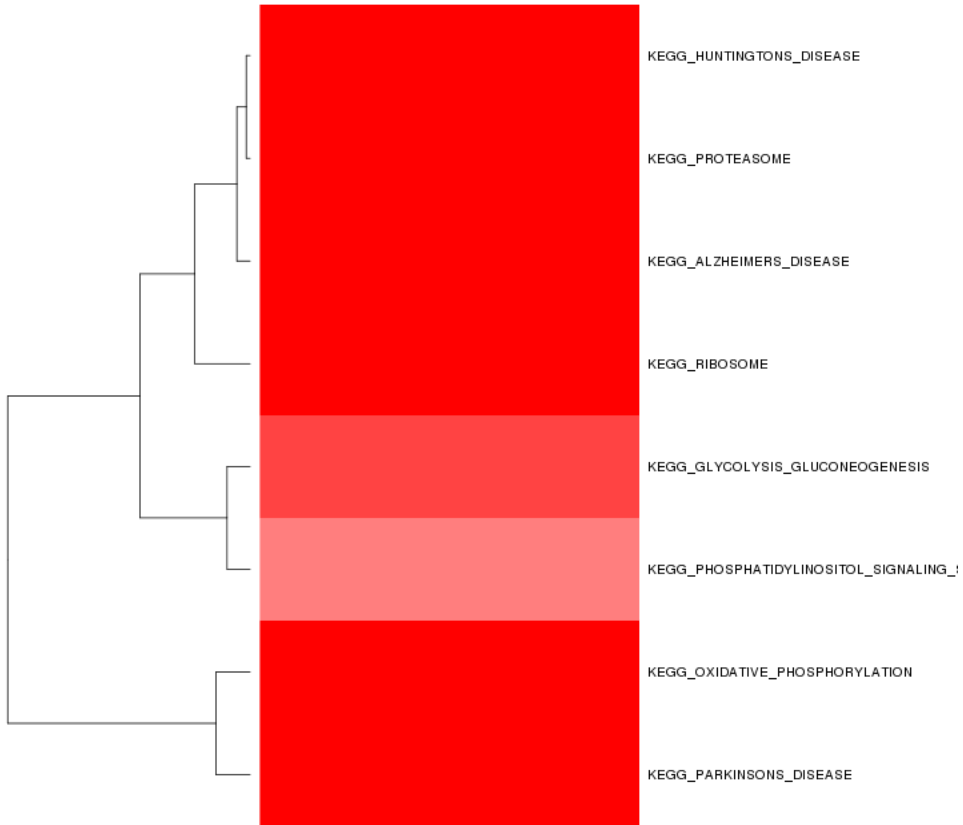
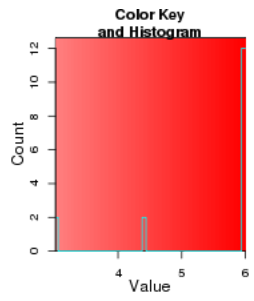


Reactome zscore:

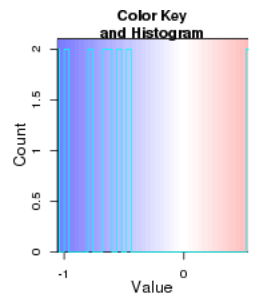
Error: subscript out of bounds

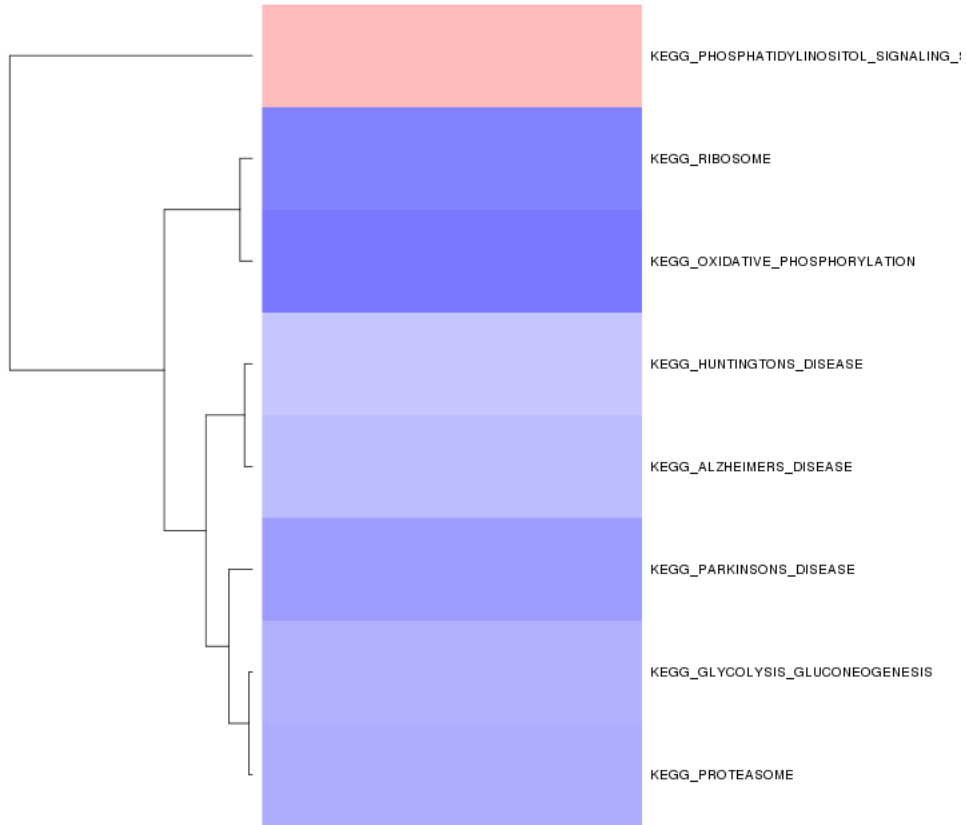


Kegg enrichment:



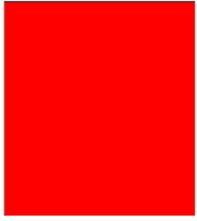
Kegg zscore:





Interneuron enrichment:

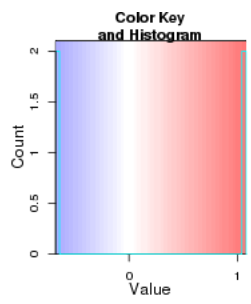
Error: no locations are finite

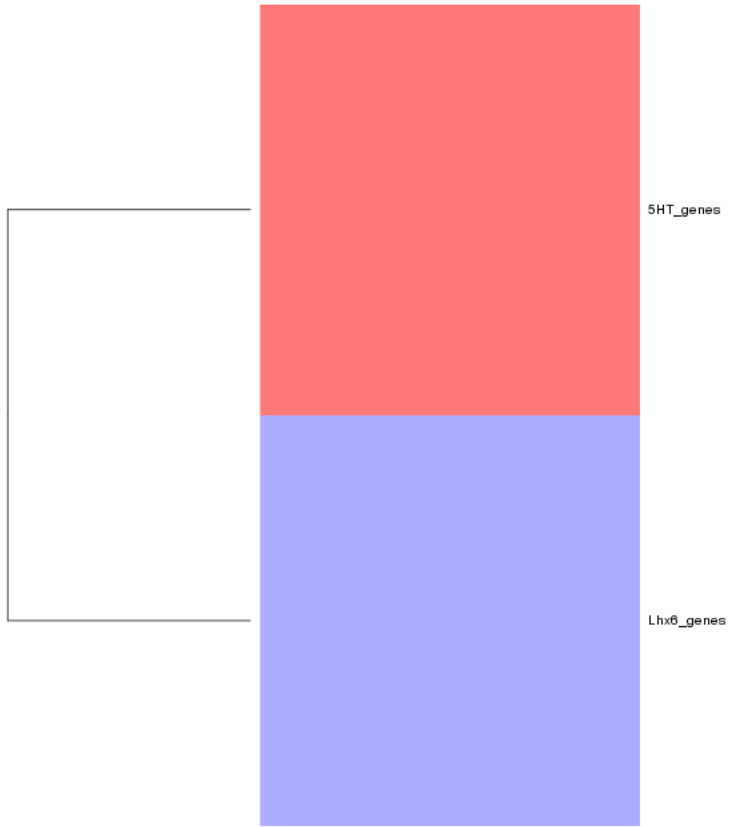


5HT_genes

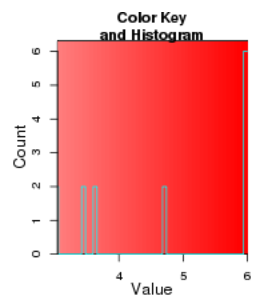
Lhx6_genes

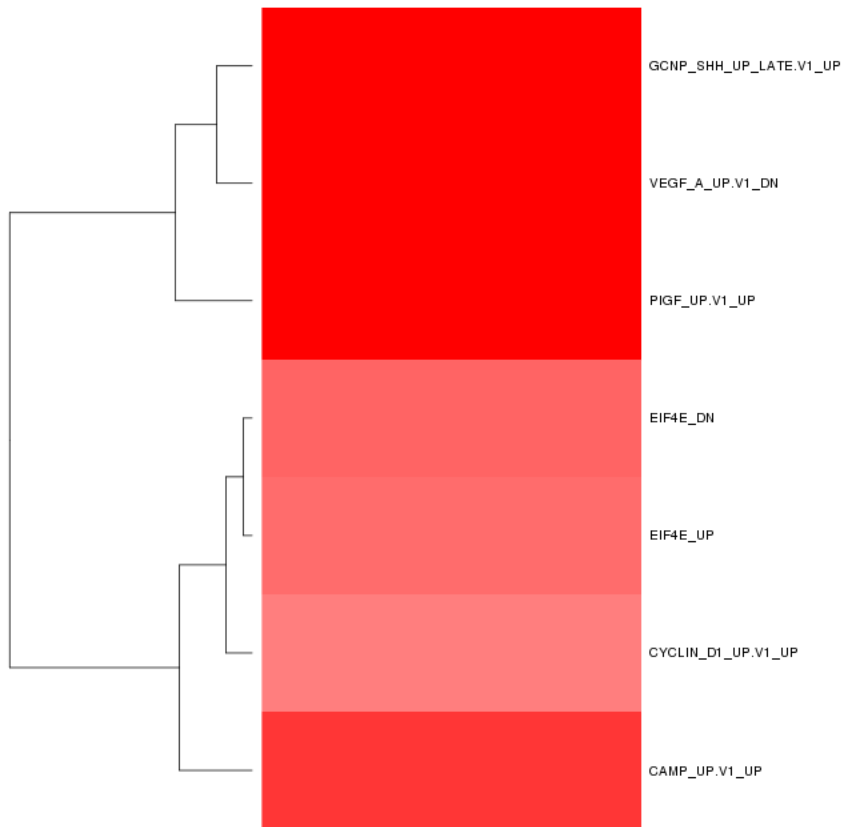
Interneuron zscore:



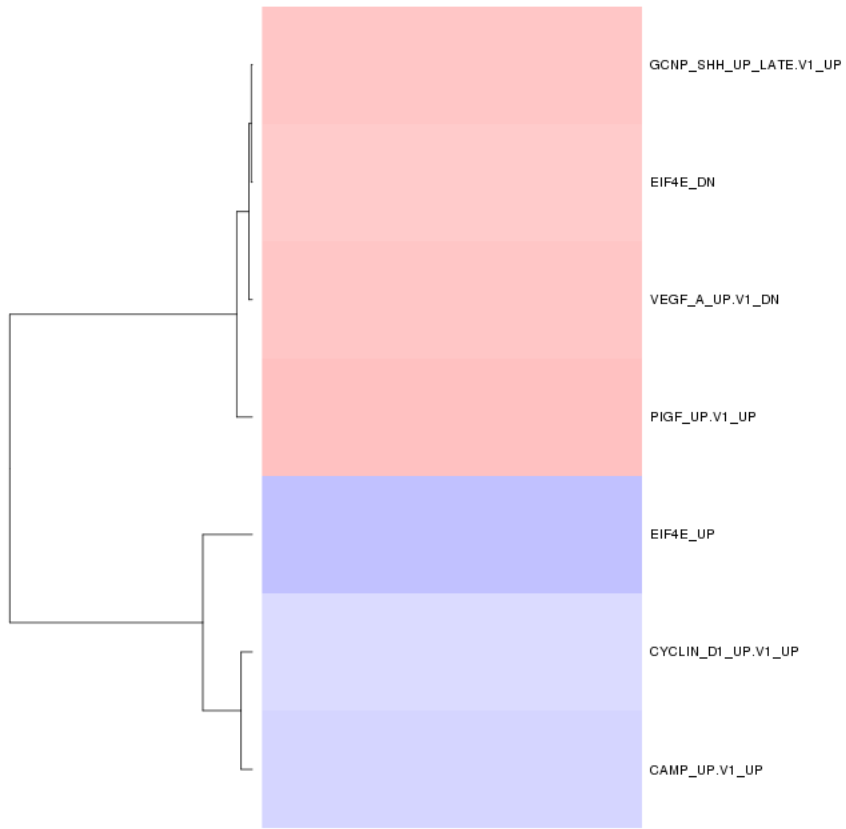
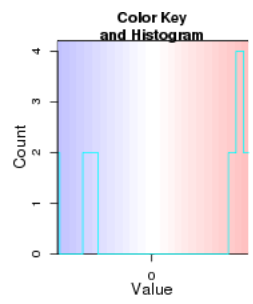


Oncogene enrichment:

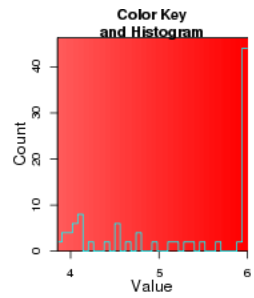


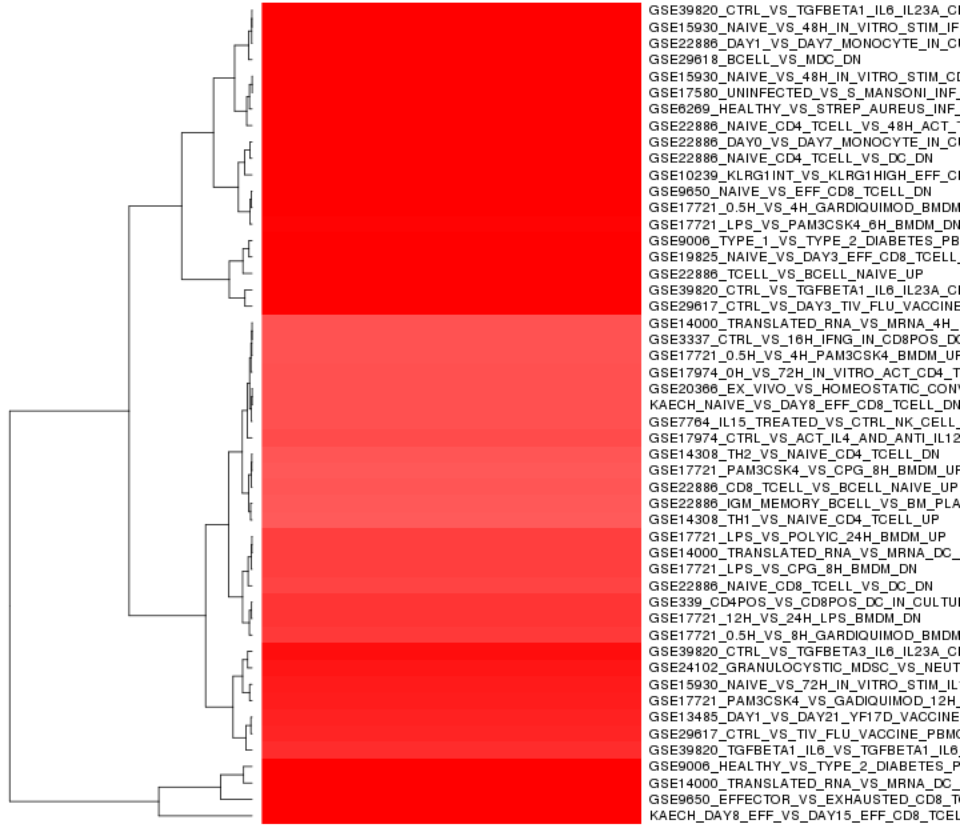


Oncogene zscore:

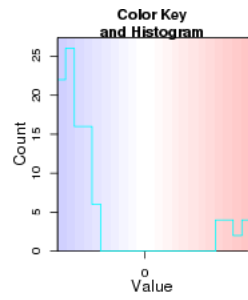


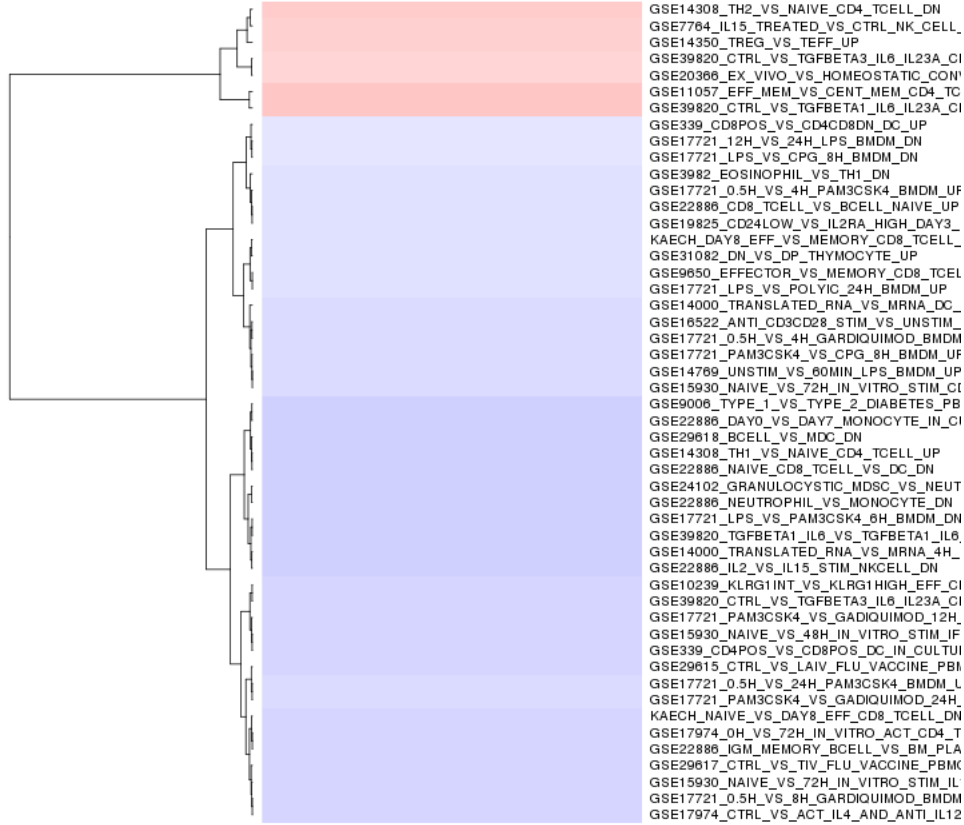
Immuno enrichment:



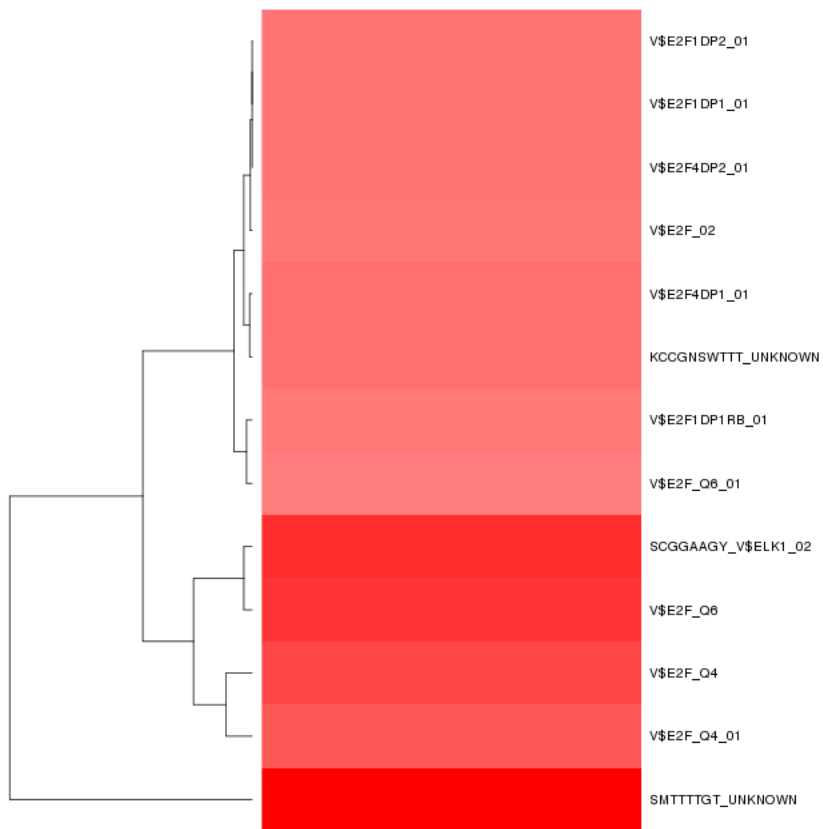
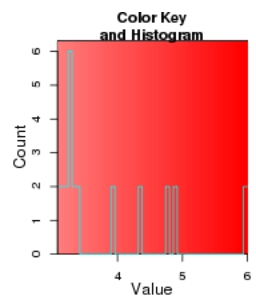


Immuno zscore:

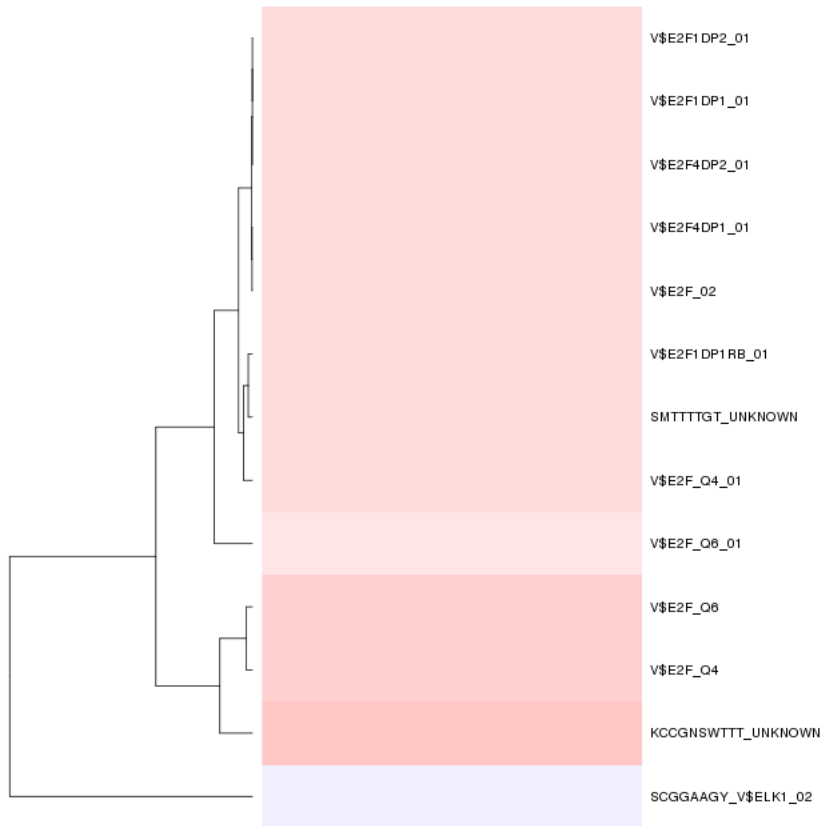
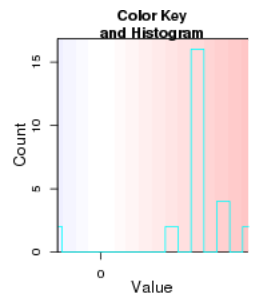




TF enrichment:



TF zscore:



GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

```
## Error: RS-DBI driver: (error in statement: near ")": syntax error)
```


Error: error in evaluating the argument 'object' in selecting a method for function 'annotation': Error: object 'sigGenes' not found

Error: object 'geneAnnot' not found

Error: error in evaluating the argument 'object' in selecting a method for function 'diffData': Error: object 'sigGenes' not found

Error: object 'sigDiff' not found

Error: object 'geneNames' not found

Error: object 'sigEntrez' not found

Error: error in evaluating the argument 'x' in selecting a method for function 'unlist': Error: object 'sigEZ' not found

Error: object 'sigEZ' not found

Error: object 'sigEZ' not found

Error: object 'sigEZ' not found

Error: object 'sigEZ' not found

Error: object 'sigEZ' not found

Error: error in evaluating the argument 'x' in selecting a method for function 'plot': Error: object 'goBP' not found

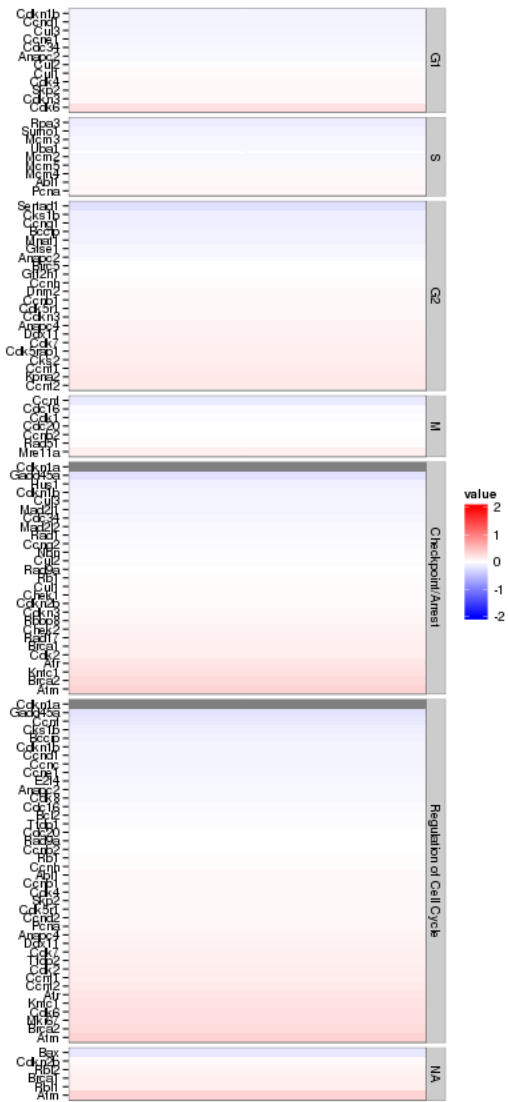
Error: error in evaluating the argument 'x' in selecting a method for function 'plot': Error: object 'goMF' not found

Error: error in evaluating the argument 'x' in selecting a method for function 'plot': Error: object 'goCC' not found

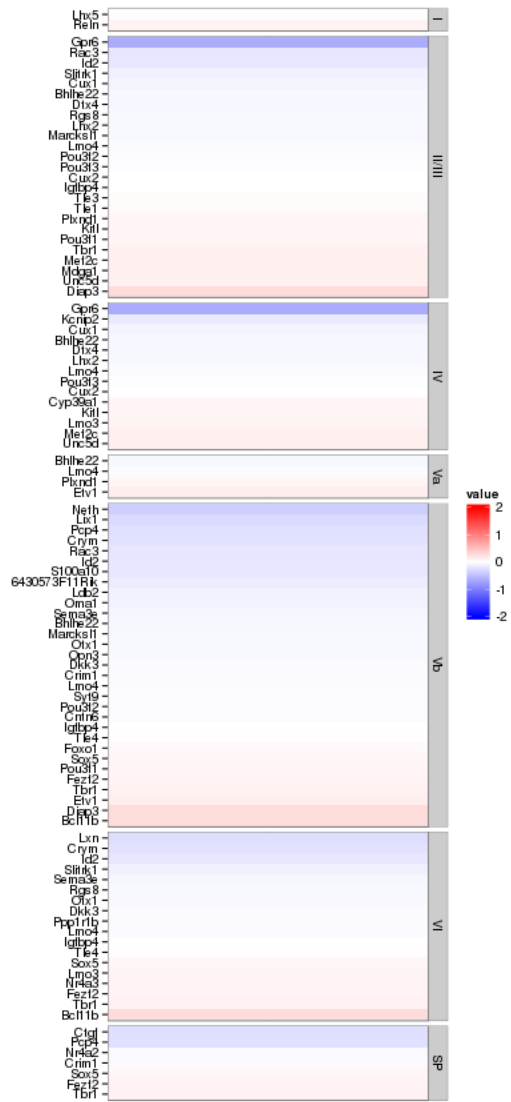
Error: error in evaluating the argument 'x' in selecting a method for function 'plot': Error: object 'kegg' not found

Error: error in evaluating the argument 'x' in selecting a method for function 'plot': Error: object 'pathway' not found

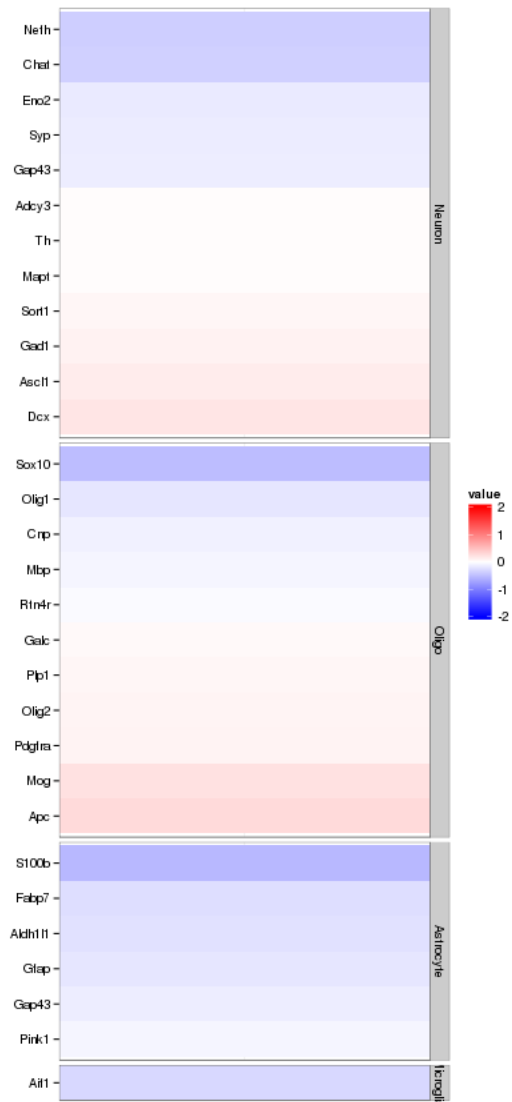
Enrichment or depletion for stage-specific cell cycle markers



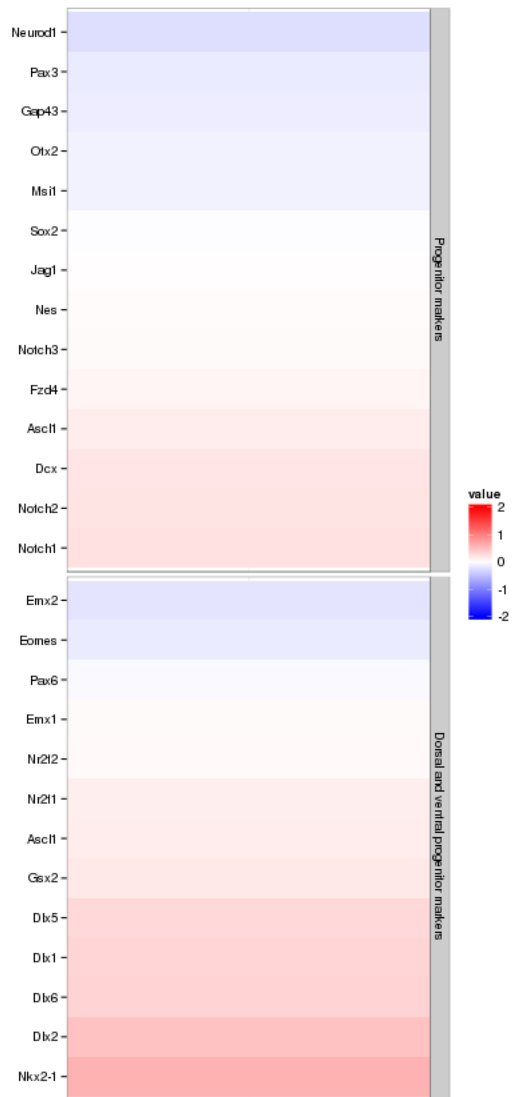
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



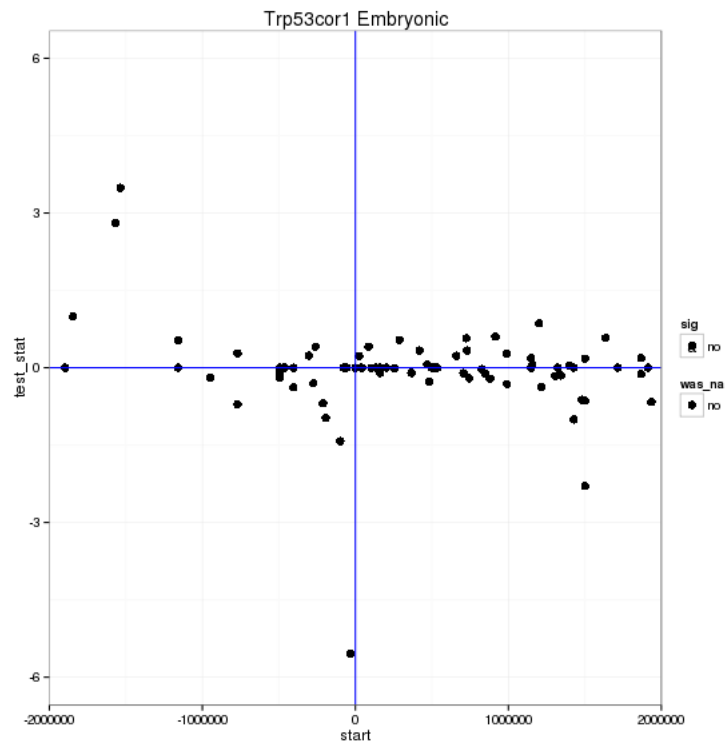
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 0 genes significantly regulated in a region this size is: 1



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR749

16 JR814

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	31980100.00	1.28	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	31980100.00	1.11	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	31980100.00	1.24	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	31980100.00	1.30	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	31980100.00	1.21	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	31980100.00	0.90	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	31980100.00	0.98	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	31980100.00	0.93	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	31980100.00	0.87	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	31980100.00	0.97	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	31980100.00	0.82	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	31980100.00	0.81	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	31980100.00	0.69	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	31980100.00	1.23	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR749/abundances.cxb	Trp53cor	0	Trp53cor_0	31241000.00	31980100.00	0.95	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR814/abundances.cxb	Trp53cor	1	Trp53cor_1	31403400.00	31980100.00	1.00	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
```

```

## [7] gdata_2.13.3
## [8] org.Mm.eg.db_2.10.1
## [9] clusterProfiler_1.13.1
## [10] DOSE_2.0.0
## [11] ReactomePA_1.6.1
## [12] AnnotationDbi_1.24.0
## [13] Biobase_2.22.0
## [14] mgcv_1.7-29
## [15] nlme_3.1-117
## [16] RMySQL_0.9-3
## [17] RColorBrewer_1.0-5
## [18] gridExtra_0.9.1
## [19] gtable_0.1.2
## [20] marray_1.40.0
## [21] gplots_2.13.0
## [22] GSA_1.03
## [23] limma_3.18.13
## [24] xtable_1.7-3
## [25] cummerbund_2.7.2
## [26] Gviz_1.6.0
## [27] rtracklayer_1.22.7
## [28] GenomicRanges_1.14.4
## [29] XVector_0.2.0
## [30] IRanges_1.20.7
## [31] fastcluster_1.1.13
## [32] reshape2_1.4
## [33] ggplot2_1.0.0
## [34] RSQLite_0.11.4
## [35] DBI_0.2-7
## [36] BiocGenerics_0.8.0
## [37] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GO.db_2.10.1        GOsemSim_1.20.3
## [16] graph_1.40.1        graphite_1.8.1      gtools_3.4.1
## [19] Hmisc_3.14-4        igraph_0.7.1        KEGG.db_2.10.1
## [22] kernSmooth_2.23-12 labeling_0.2         lattice_0.20-29
## [25] latticeExtra_0.6-26 MASS_7.3-33         Matrix_1.1-3
## [28] munsell_0.4.2       org.Hs.eg.db_2.10.1 proto_0.3-10
## [31] qvalue_1.36.0       Rcpp_0.11.2         RCurl_1.95-4.1
## [34] reactome.db_1.46.1  Rsamtools_1.14.3    scales_0.2.4
## [37] splines_3.0.2       stats4_3.0.2        survival_2.37-7
## [40] tcltk_3.0.2         tools_3.0.2         XML_3.98-1.1
## [43] zlibbioc_1.8.0

```

Run Info


```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Trp53cor -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Trp53cor_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/BrainM
## 2
## 3
## 4
## 5
```

Tug1 KO vs WT (Adult)

This file shows the wt-v-ko comparison for Tug1.

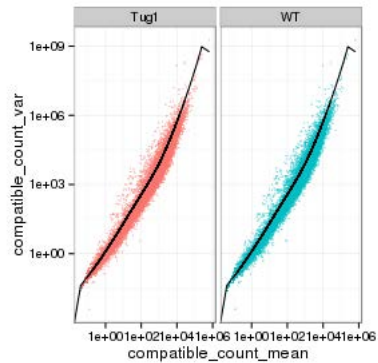
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

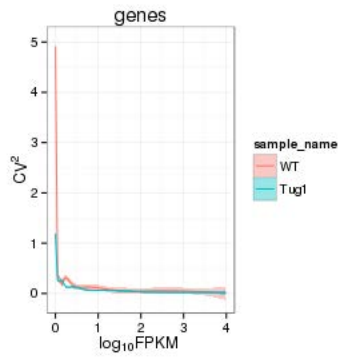
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



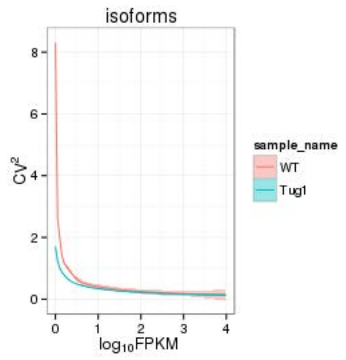
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

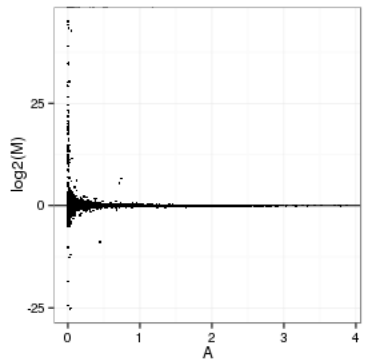
Genes:



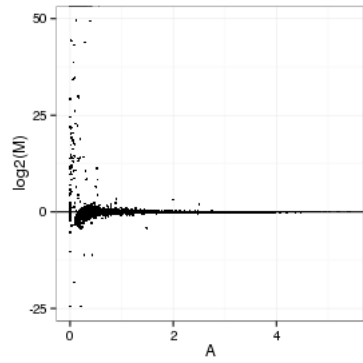
Isoforms:



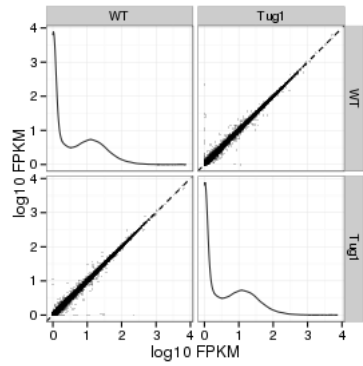
MvA plot



MvA plot counts



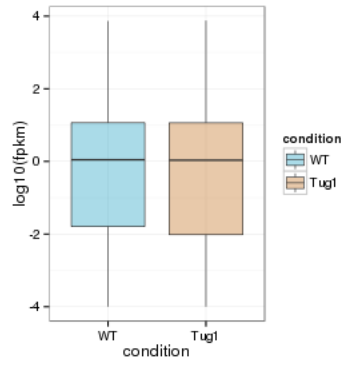
Scatterplot matrix



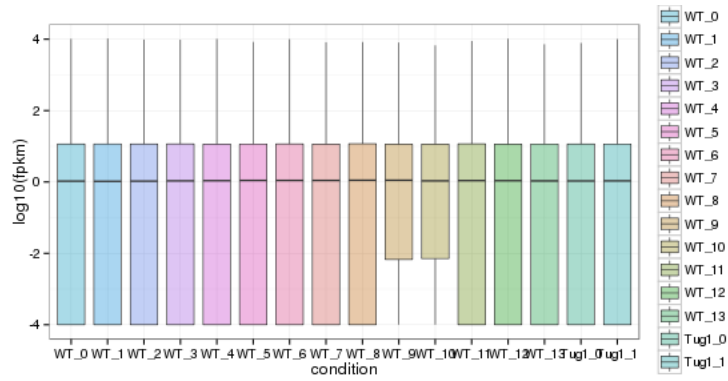
Distributions

Boxplots

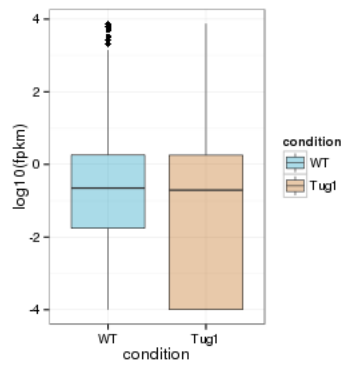
Boxplot (genes)



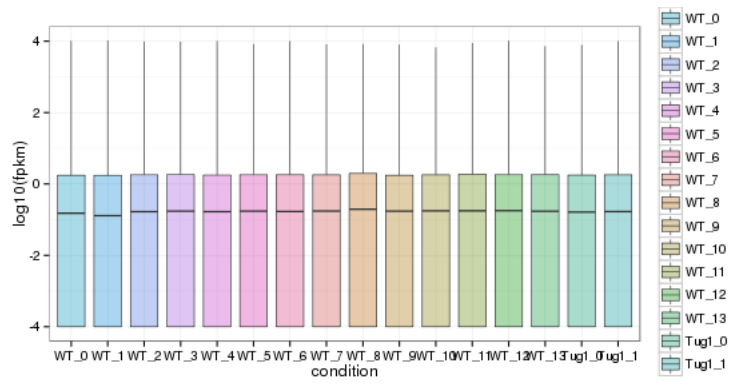
Boxplot (genes, replicates)



Boxplot (isoforms)

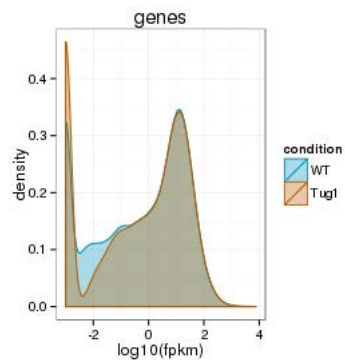


Boxplot (isoforms, replicates)

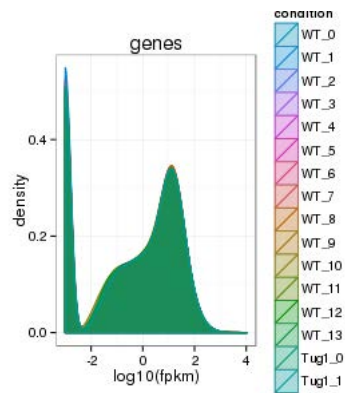


Density

Density (genes)

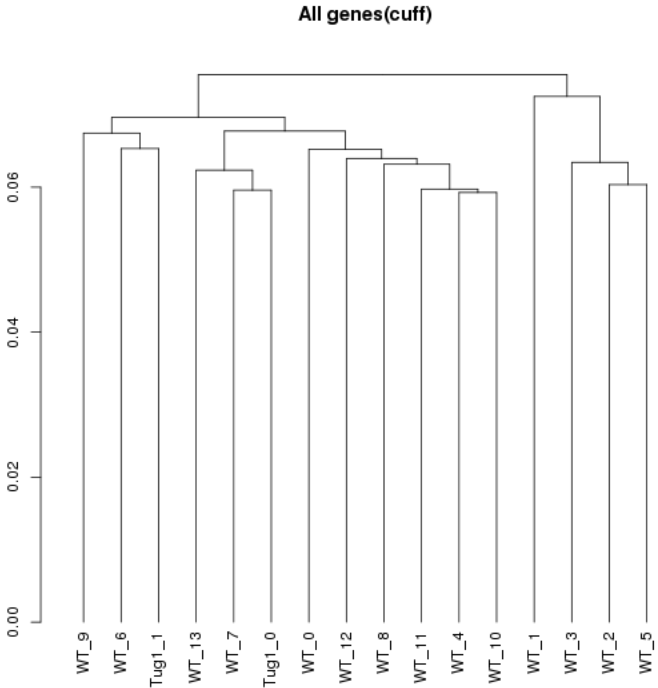


Density (genes, replicates)



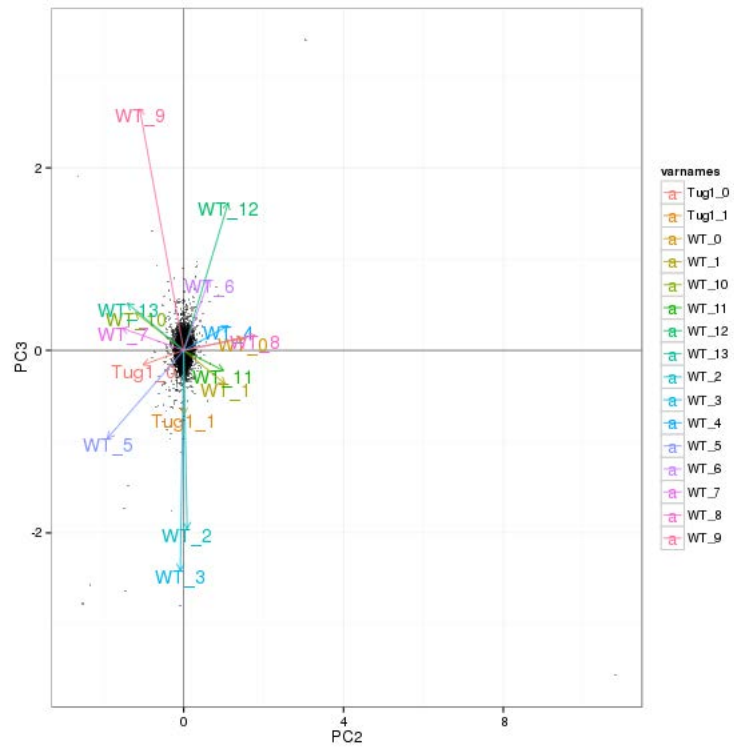
Clustering

Replicate Clusters

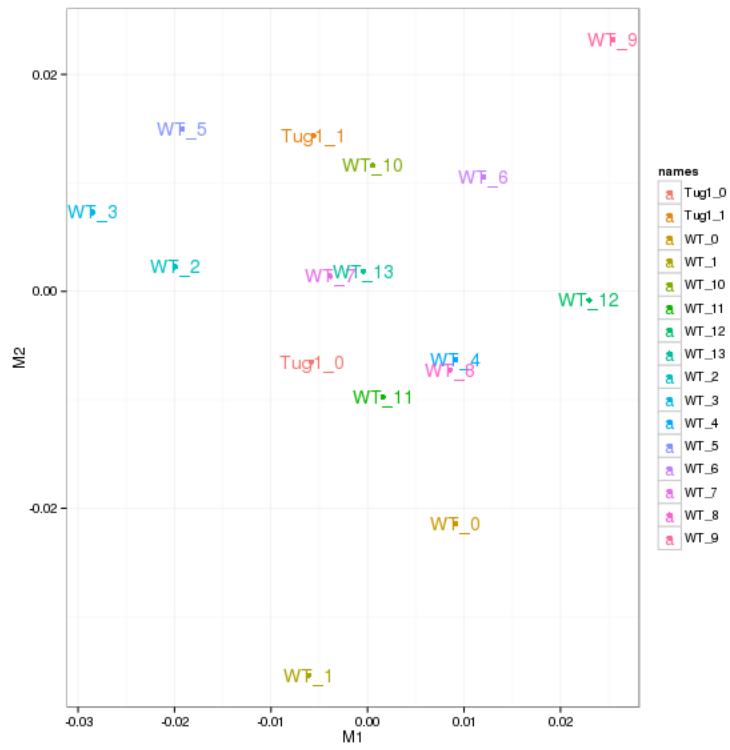


NULL

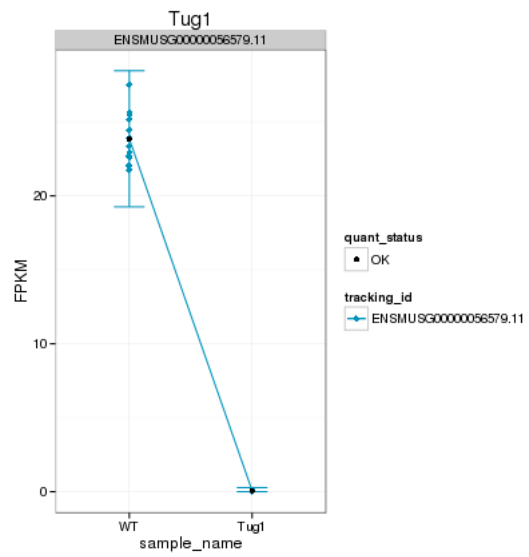
PCA (genes)



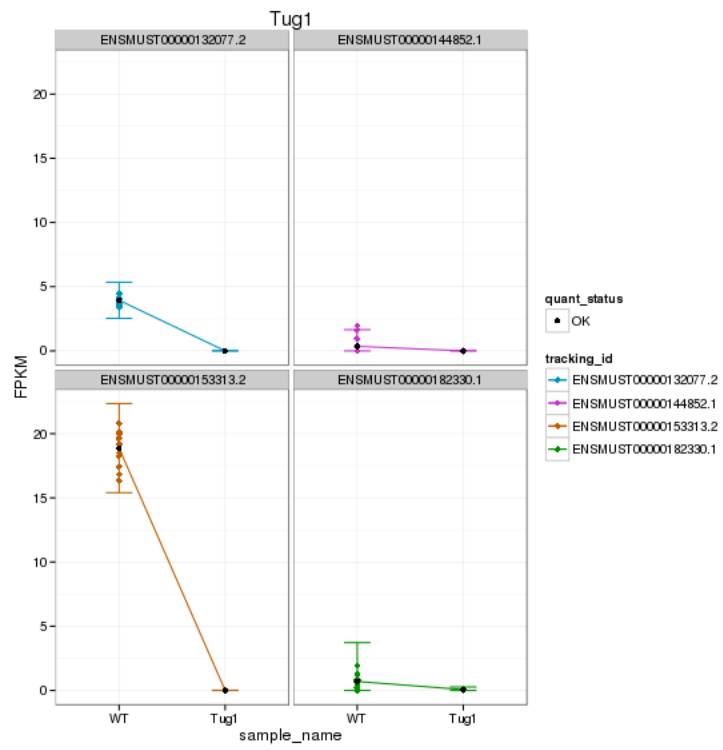
MDS (genes)



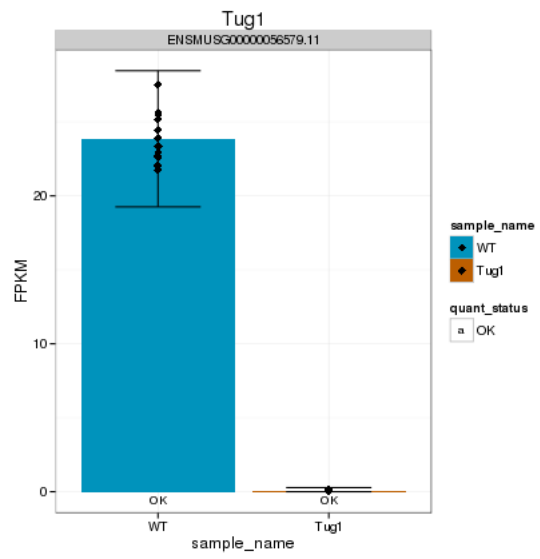
KO assessment
Endogenous IncRNA expression



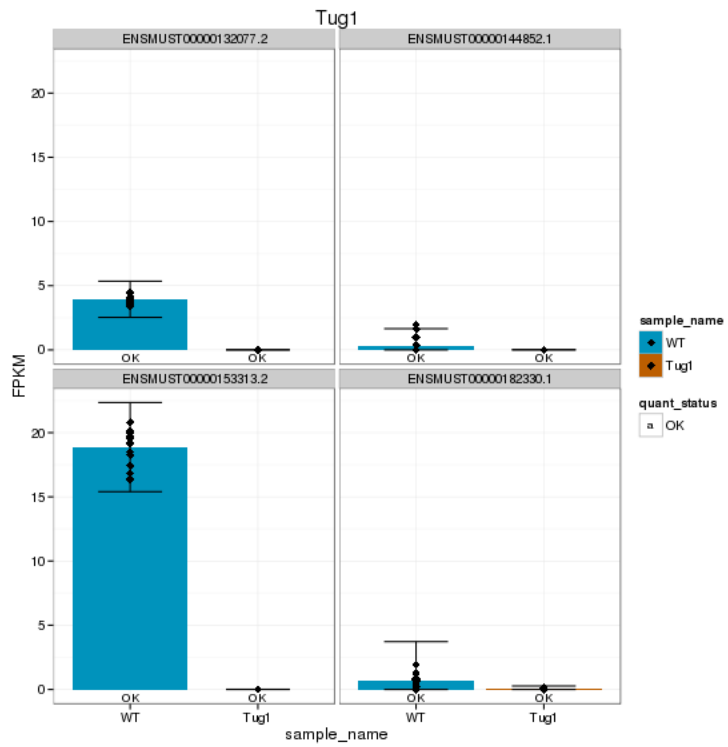
Endogenous expression of Tug1 isoforms:



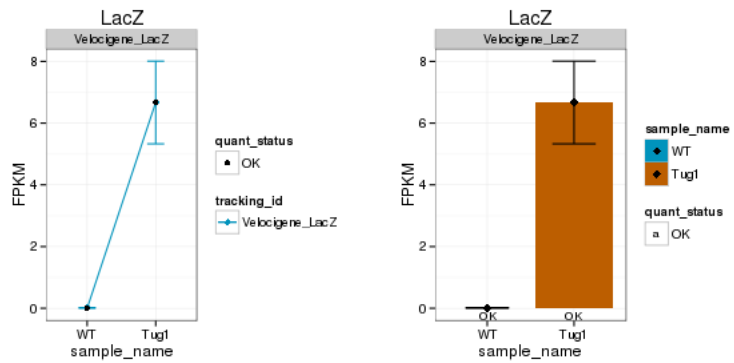
Barplot of gene expression:



Barplot of isoform expression:

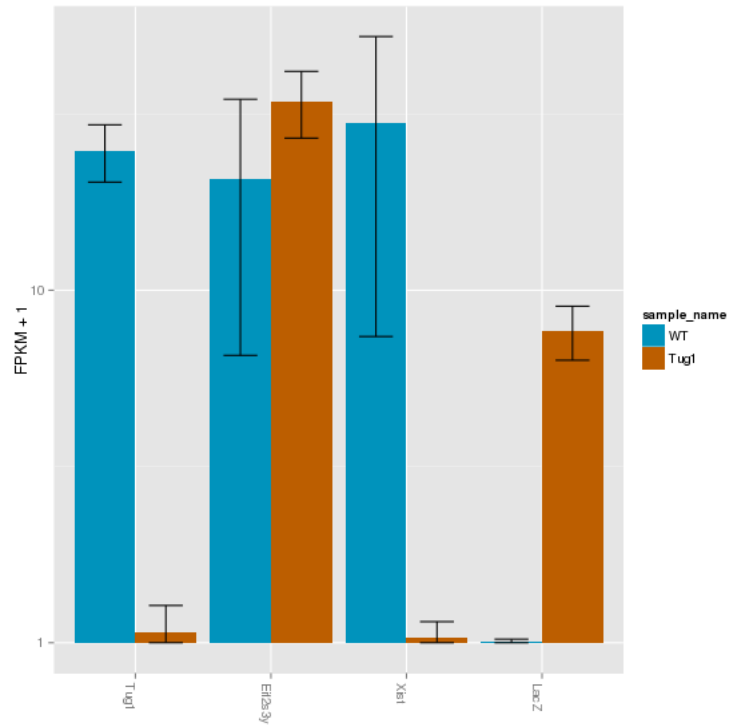


LacZ expression

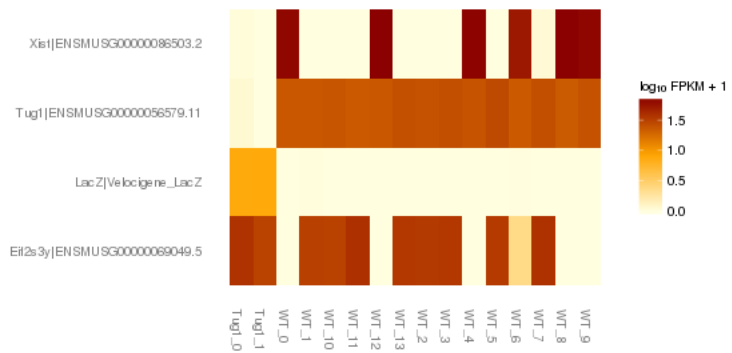


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



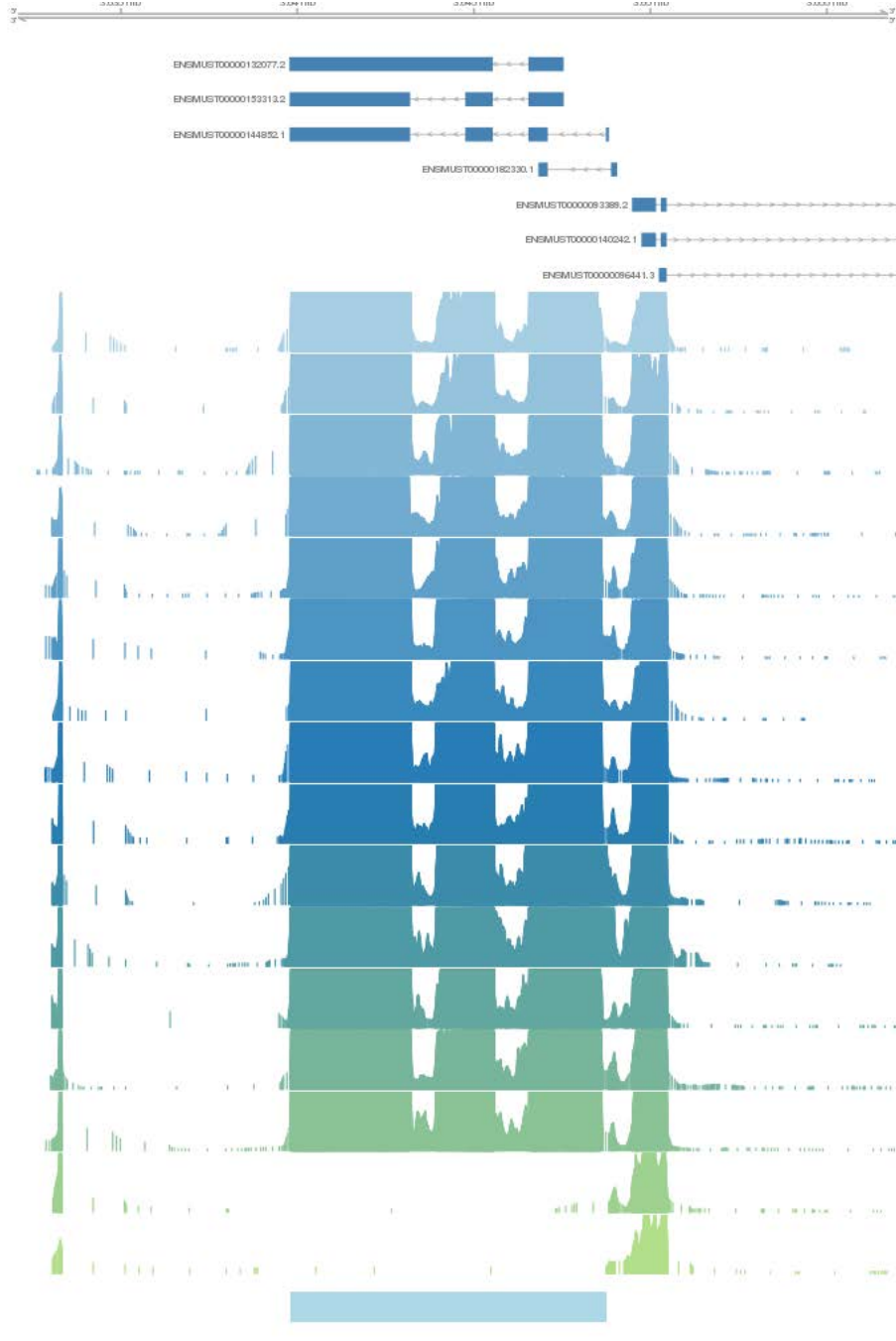
Expression heatmap:



Track visualization



KO Region Tug_1 Tug_0 WT_13 WT_12 WT_11 WT_10 WT_9 WT_8 WT_7 WT_6 WT_5 WT_4 WT_3 WT_2 WT_1 WT_0 LncRNA isoforms



Differential Analysis

Differential Genes

There are 39 significantly differentially expressed genes. They are:

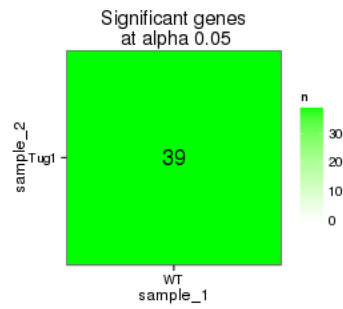
```
geneAnnot$gene_short_name
1 Gabra2
2 Pde1c
3 Nes
4 Plxdc1
5 Ubb
6 Pdk4
7 Mybpc1
8 Rnf185
9 Adi1
10 Serpina3n
11 Esco1
12 Rps24
13 Myoc
14 Herc6
15 Hddc3
16 Dis3l
17 Ndn
18 Arhgap27
19 Morc2a
20 Inpp5j
21 8430429K09Rik
22 Anln
23 Akap12
24 Pyurf
25 Nhirc1
26 Fat2
27 Kdm5d
28 Cox5b
29 Zbtb16
30 Ddx3y
31 Egr4
32 Gm9008
33 3222401L13Rik
34 Selm
35 Gm14418
36 Xist
37 AA465934
```


38 Gm14403

39 Gm26924

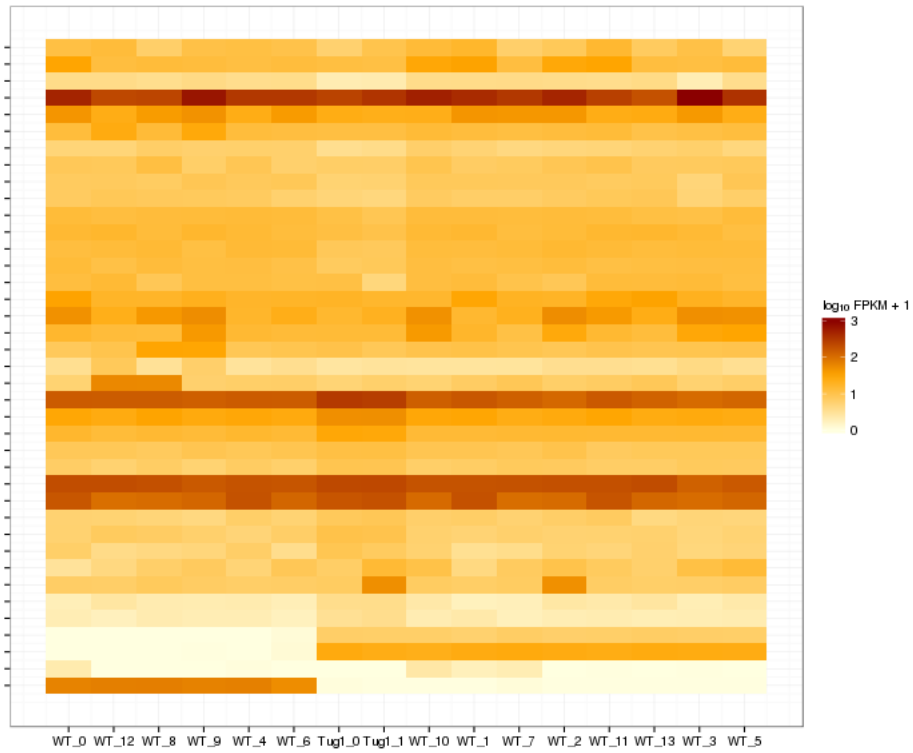
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

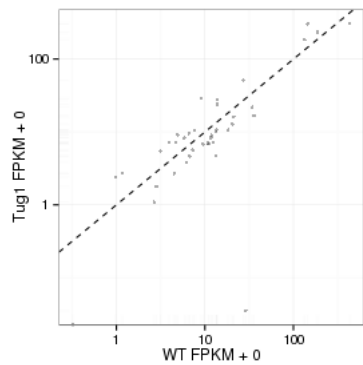


Significant genes with expression >50fpkm (any condition):(turned off)

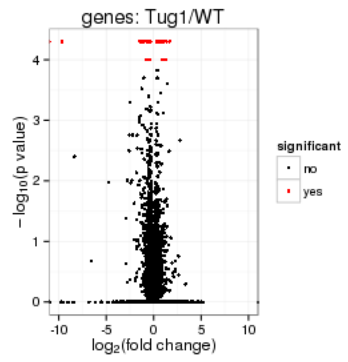
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

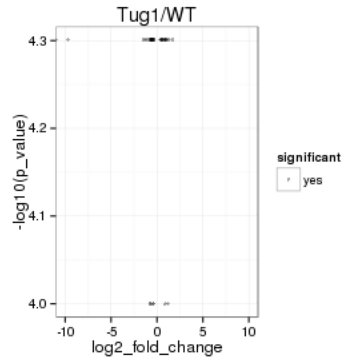
Scatter plot of significant genes only:



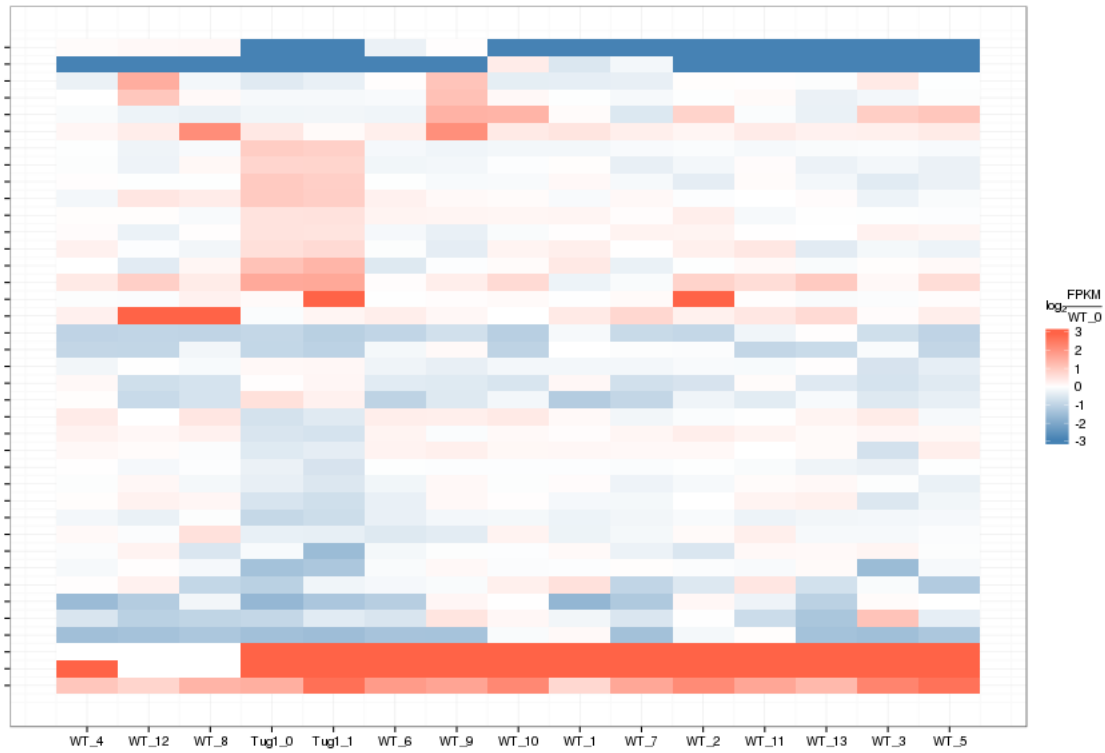
Volcano Plot



Volcano plot with significant genes only:



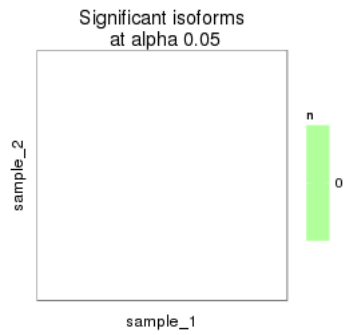
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

[1] "no sig isoforms"

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

Gene/Pathway Analysis

GSEA

Enrichment and zscores are calculated based on expression in KO vs WT (fpkmKO/fpkmWT), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KOWT Blue = down in KO Red = Up in KO

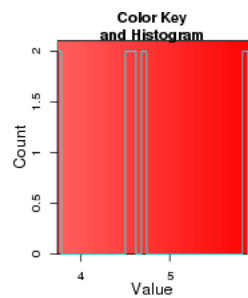
Biocarta enrichment:

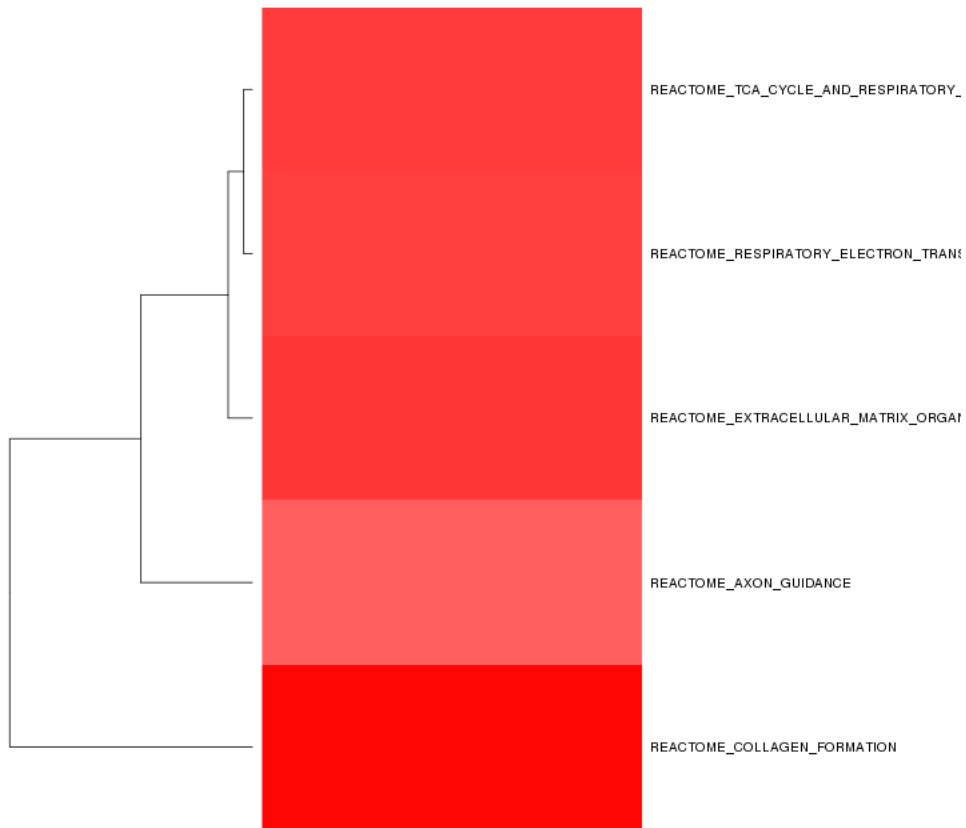
```
## [1] "Not enough significant categories to print a heatmap!"
```

Biocarta zscore:

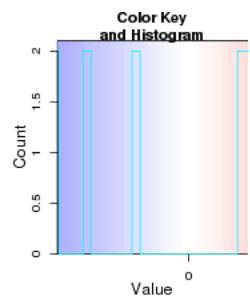
```
## [1] "Not enough significant categories to print a heatmap!"
```

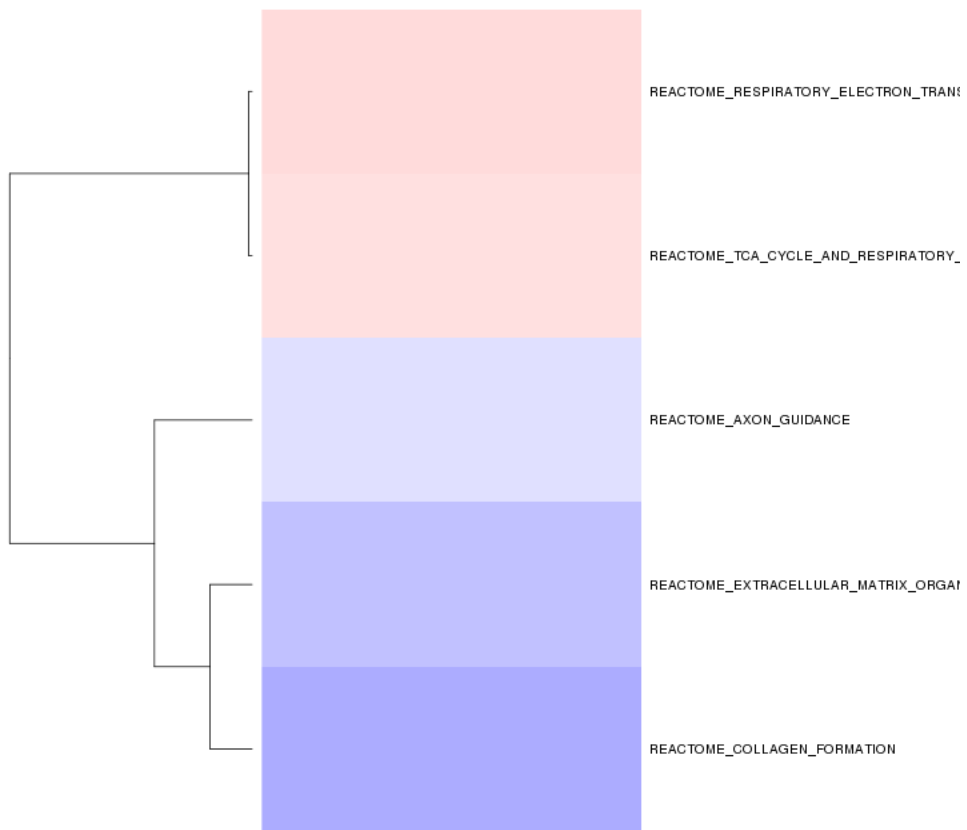
Reactome enrichment:



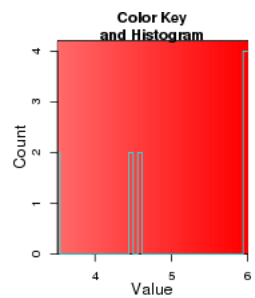


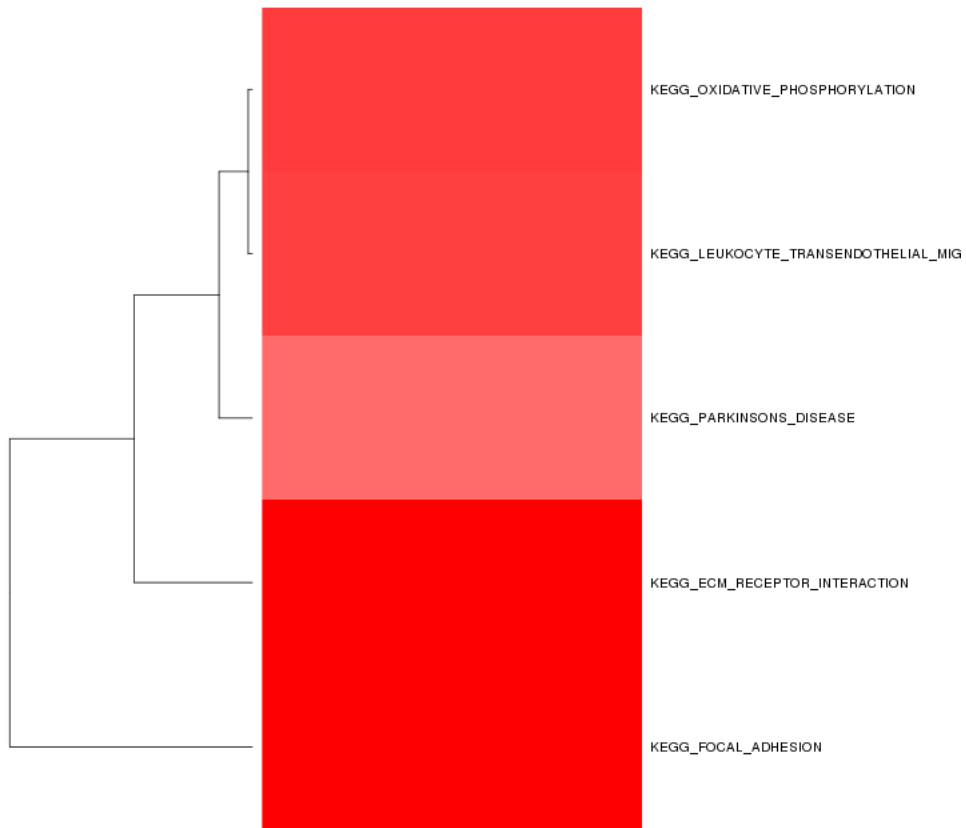
Reactome zscore:



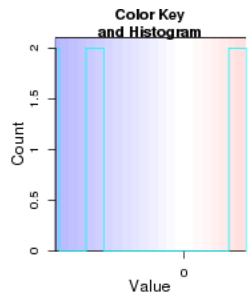


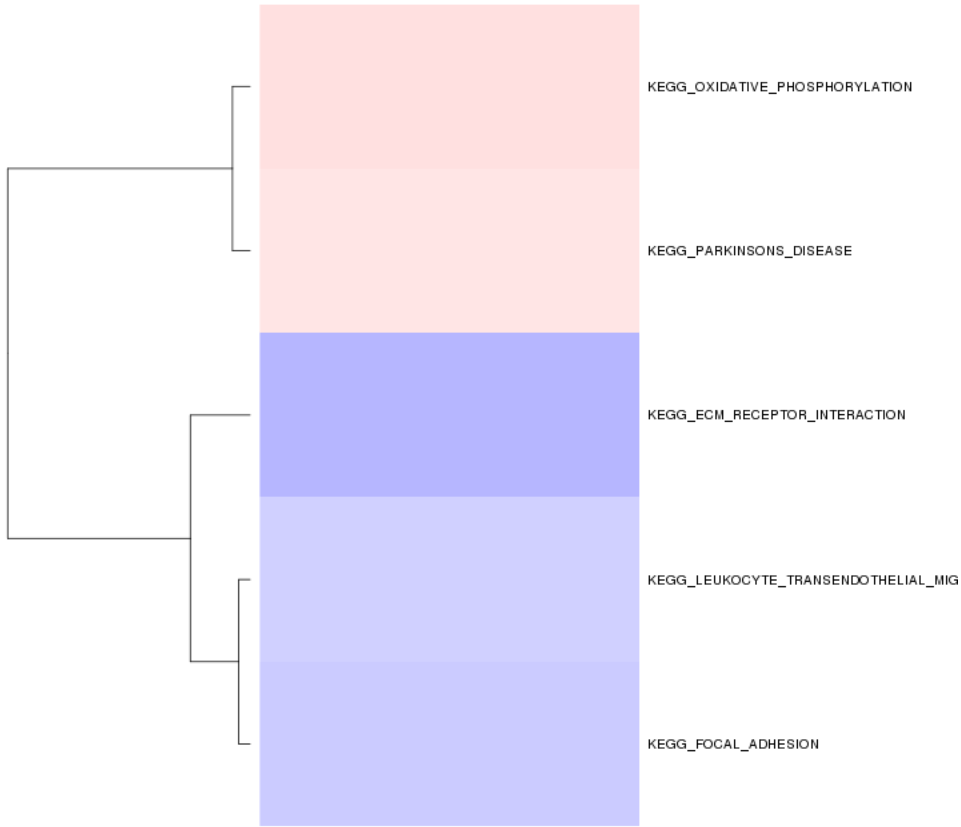
Kegg enrichment:





Kegg zscore:





Interneuron enrichment:

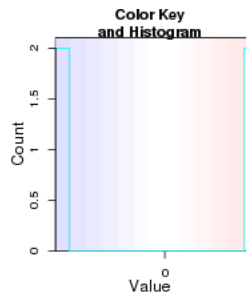
Error: no locations are finite

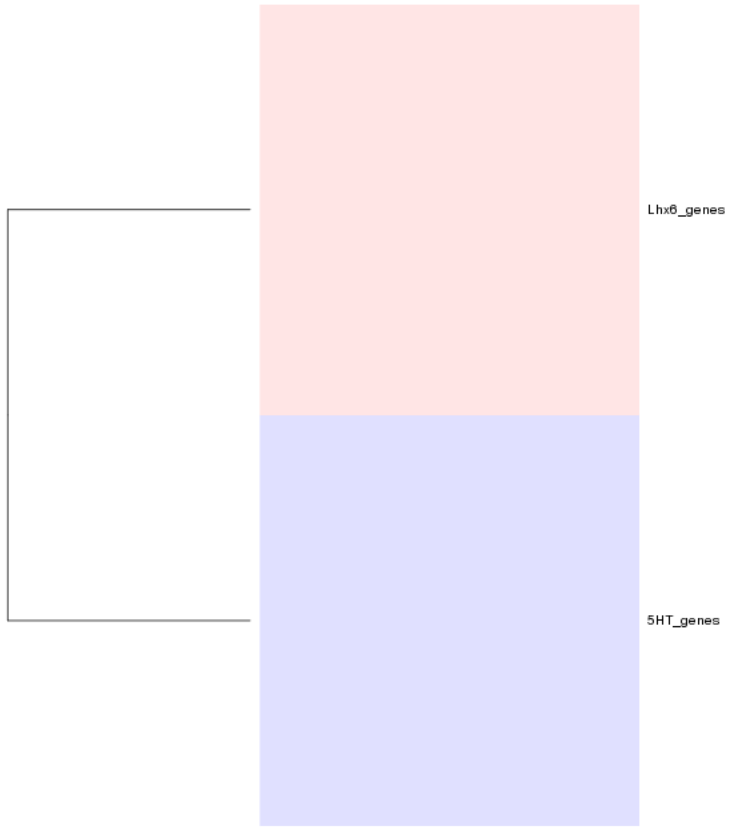


5HT_genes

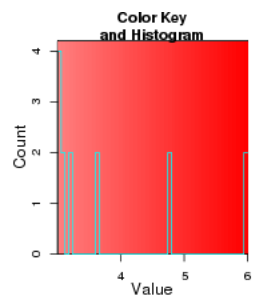
Lhx0_genes

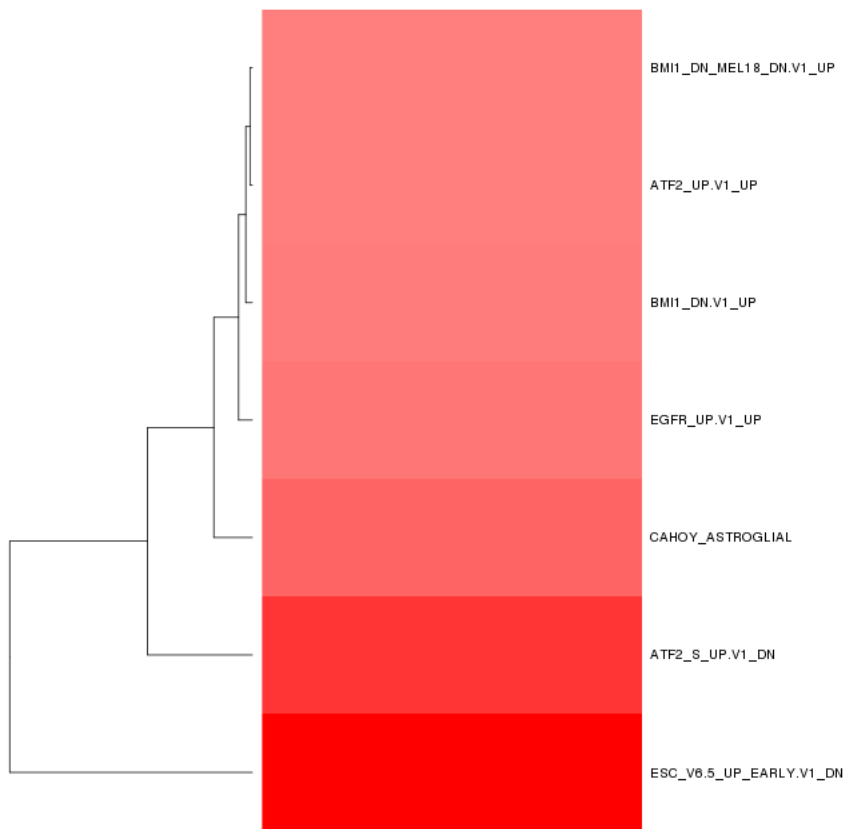
Interneuron zscore:



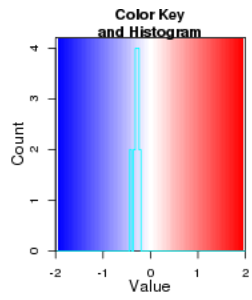


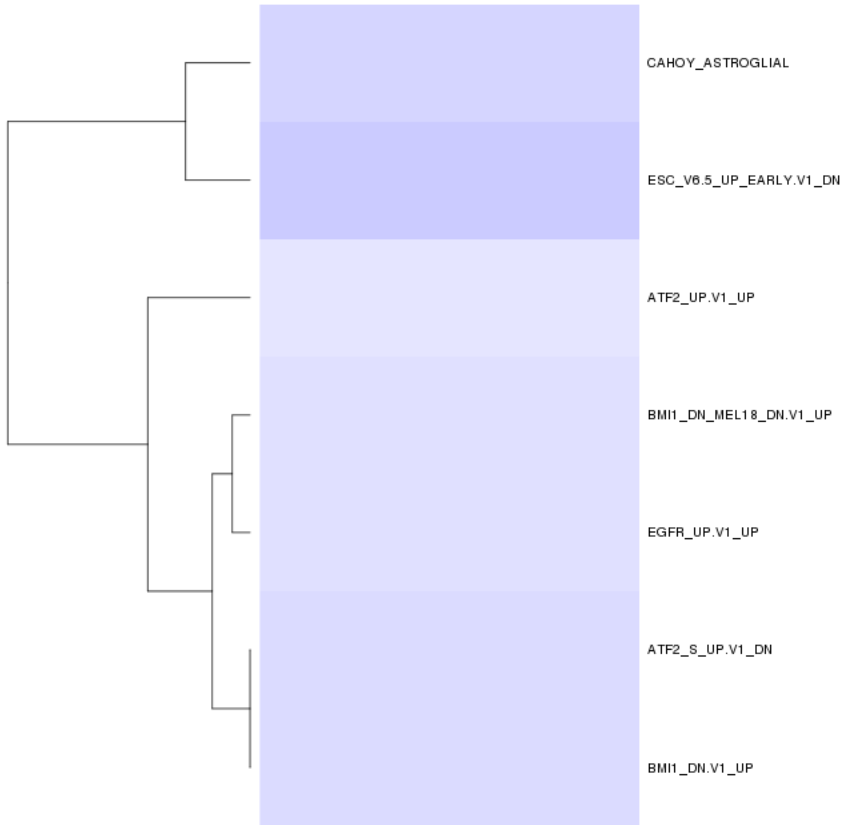
Oncogene enrichment:





Oncogene zscore:





Immuno enrichment:

```
## Error: argument is of length zero
```

Immuno zscore:

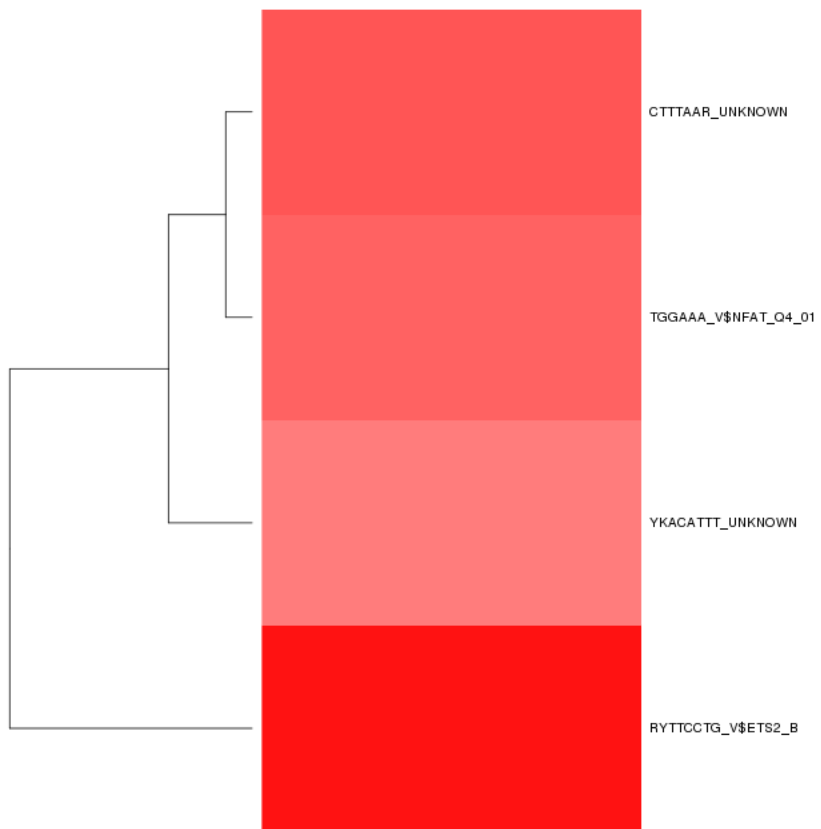
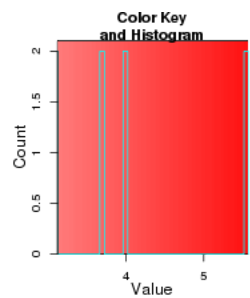
```
## Error: incorrect number of dimensions
```

```
## Error: object 'x_ordered' not found
```

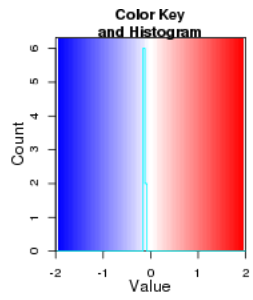
```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

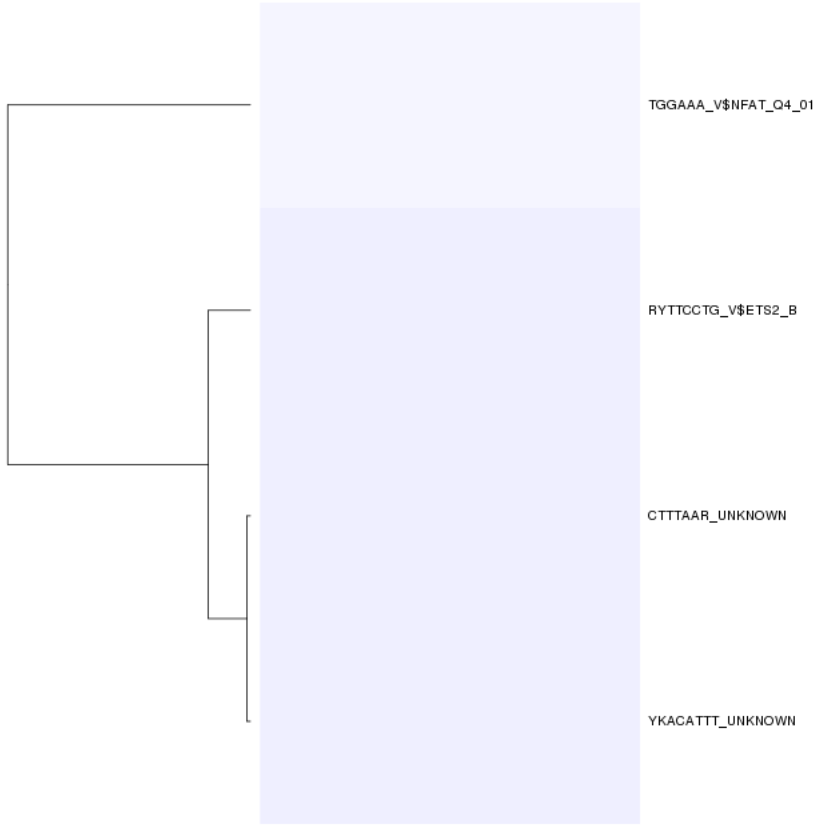
```
## Error: object 'x_ordered' not found
```

TF enrichment:



TF zscore:

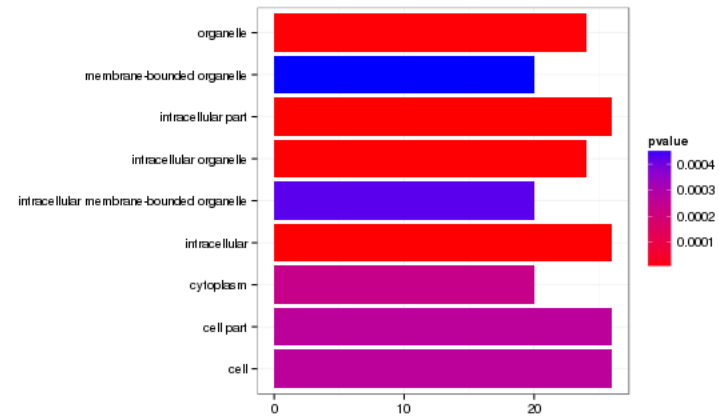
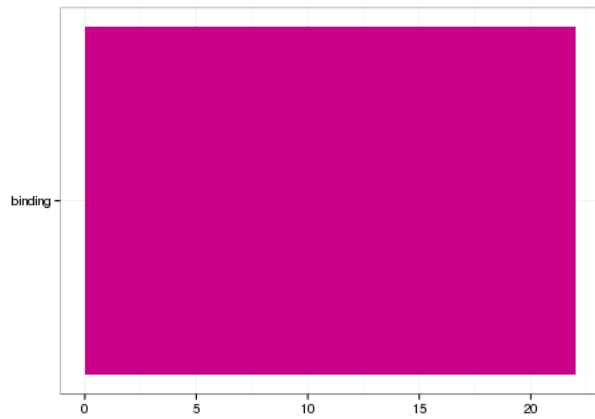




GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

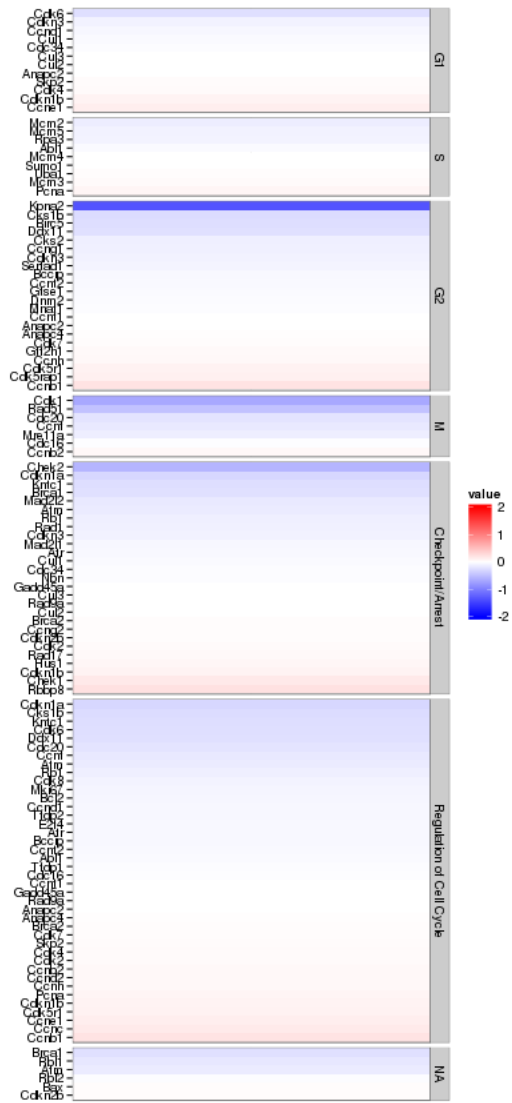
```
## Error: 'x' and 'units' must have length > 0
```

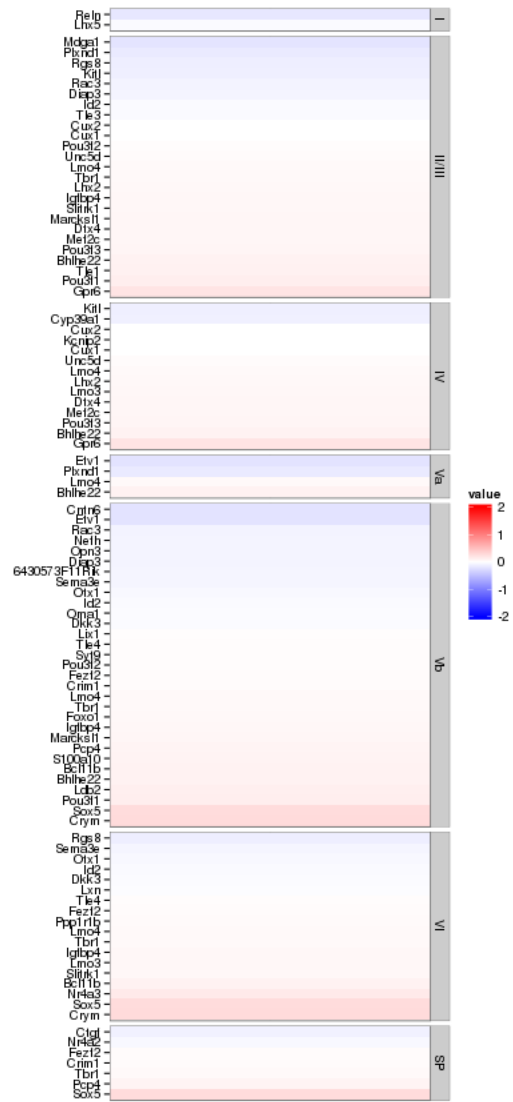
```
## Error: need finite 'xlim' values
```

```
## Error: need finite 'xlim' values
```

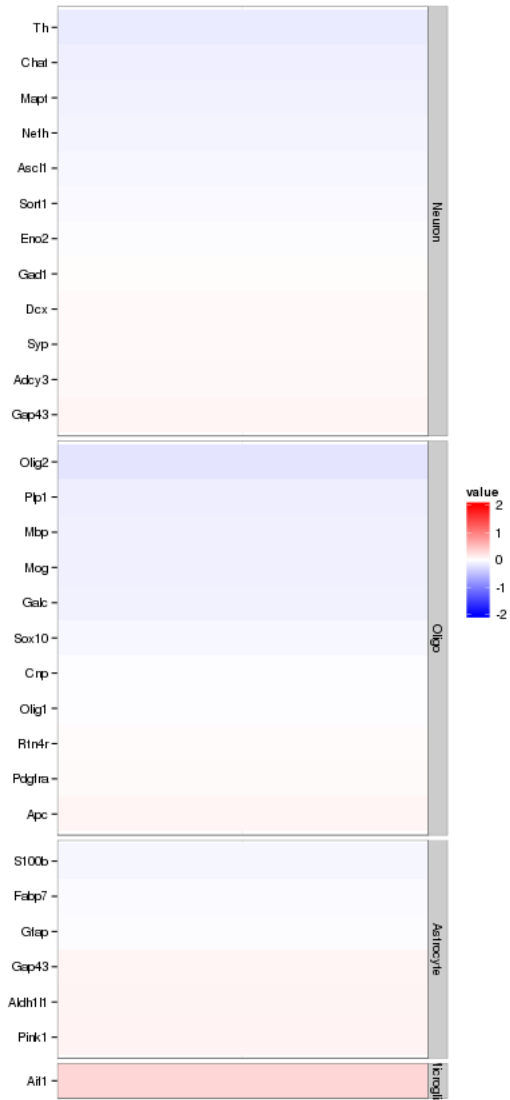
Enrichment or depletion for stage-specific cell cycle markers



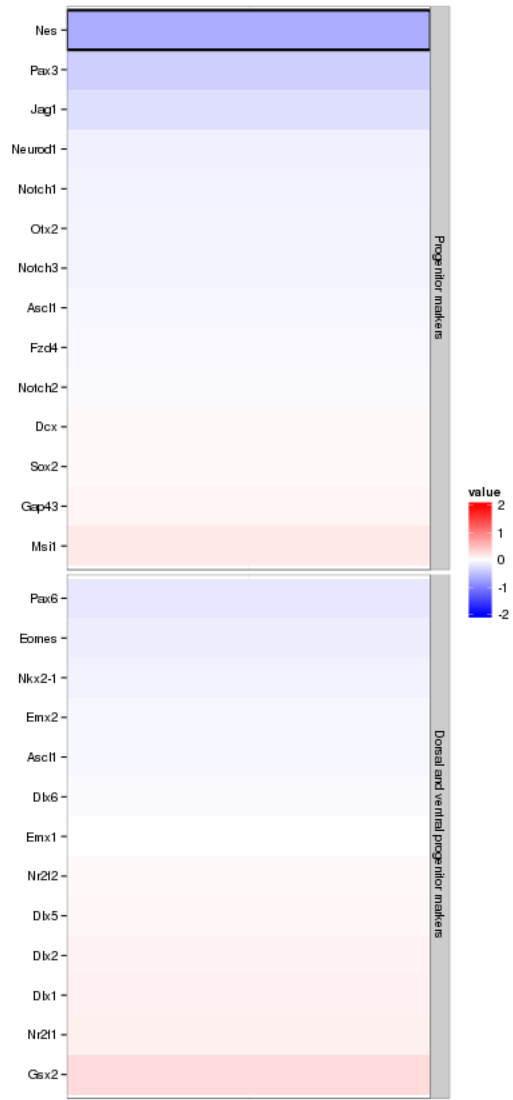
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



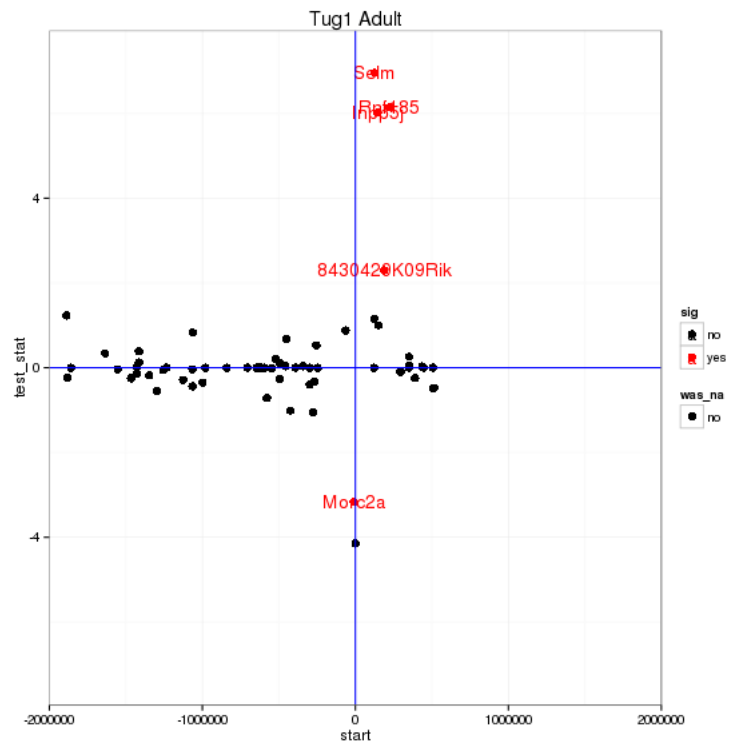
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

\log_2 Foldchange and test statistic are calculated with the ratio of $\text{fpkm(KO)}/\text{fpkm(WT)}$, thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 5 genes significantly regulated in a region this size is: 0.0023



Notes

Samples used are:

10

- 1 JR729
- 2 JR728
- 3 JR796
- 4 JR797
- 5 JR740
- 6 JR800
- 7 JR827
- 8 JR778
- 9 JR734
- 10 JR802
- 11 JR803
- 12 JR735
- 13 JR785
- 14 JR781

15 JR745

16 JR807

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR729/abundances.cxb WT	0	WT_0		26334400.00	34125900.00	0.77	1.00
2 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR728/abundances.cxb WT	1	WT_1		20329200.00	34125900.00	0.59	1.00
3 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR796/abundances.cxb WT	2	WT_2		34089000.00	34125900.00	1.00	1.00
4 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR797/abundances.cxb WT	3	WT_3		28103300.00	34125900.00	0.81	1.00
5 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR740/abundances.cxb WT	4	WT_4		35808200.00	34125900.00	1.05	1.00
6 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR800/abundances.cxb WT	5	WT_5		37012200.00	34125900.00	1.09	1.00
7 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR827/abundances.cxb WT	6	WT_6		27786600.00	34125900.00	0.82	1.00
8 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR778/abundances.cxb WT	7	WT_7		39541900.00	34125900.00	1.17	1.00
9 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR734/abundances.cxb WT	8	WT_8		34480600.00	34125900.00	1.03	1.00
10 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR802/abundances.cxb WT	9	WT_9		45467400.00	34125900.00	1.32	1.00
11 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR803/abundances.cxb WT	10	WT_10		52130400.00	34125900.00	1.53	1.00
12 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR735/abundances.cxb WT	11	WT_11		34994400.00	34125900.00	1.03	1.00
13 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR785/abundances.cxb WT	12	WT_12		34173600.00	34125900.00	0.99	1.00
14 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR781/abundances.cxb WT	13	WT_13		41538900.00	34125900.00	1.23	1.00
15 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR745/abundances.cxb Tug1	0	Tug1_0		38091200.00	34125900.00	1.12	1.00
16 /n/rinn_data1/seq/Igoff/Projects/BrainMap/data/quants/JR807/abundances.cxb Tug1	1	Tug1_1		29979900.00	34125900.00	0.87	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats     graphics grDevices utils
## [8] datasets base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
```

```

## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8  bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2     colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4       DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10       Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOsemSim_1.20.3    graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1       Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1     KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29    latticeExtra_0.6-26
## [25] MASS_7.3-33        Matrix_1.1-3       munSELL_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10       qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1     reactome.db_1.46.1
## [34] Rsamtools_1.14.3   scales_0.2.4       splines_3.0.2
## [37] stats4_3.0.2       survival_2.37-7    tcltk_3.0.2
## [40] tools_3.0.2        XML_3.98-1.1       zlibbioc_1.8.0

```

Run Info


```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Tug1 -o /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/diffs/Tug1_vs_WT_Adult /n/rinn_data1/seq/lgoft/Projects/BrainMap/data/ann
## 2
## 3
## 4
## 5
```

Tug1 KO vs WT (Embryonic)

This file shows the wt-v-ko comparison for Tug1.

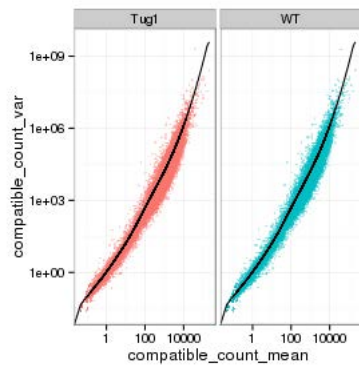
Cuff overview:

```
## CuffSet instance with:  
## 2 samples  
## 26754 genes  
## 77524 isoforms  
## 0 TSS  
## 23066 CDS  
## 0 promoters  
## 0 splicing  
## 19954 relCDS
```

QC

Dispersion

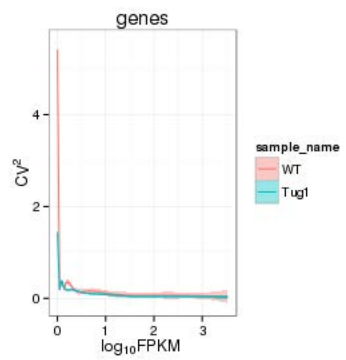
Dispersion plot for genes in cuff: (Overdispersion can lead to innacurate quants)



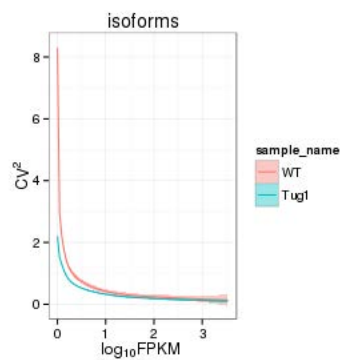
Cross-replicate variability (fpkmSCVplot)

Differences in CV 2 can result in lower numbers of differentially expressed genes due to a higher degree of variability between replicate fpkm estimates.

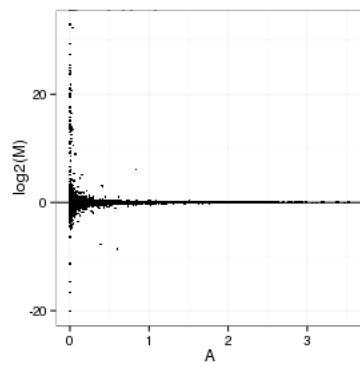
Genes:



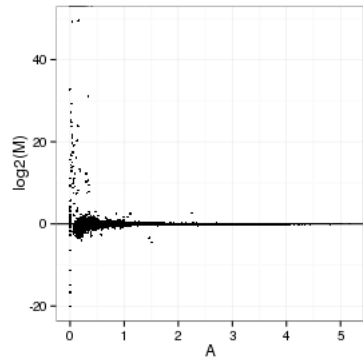
Isoforms:



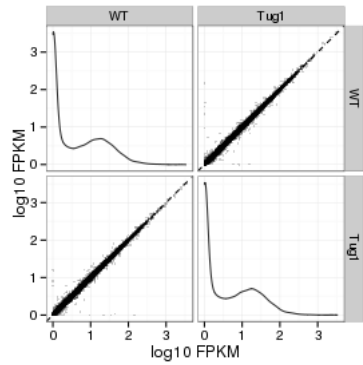
MvA plot



MvA plot counts



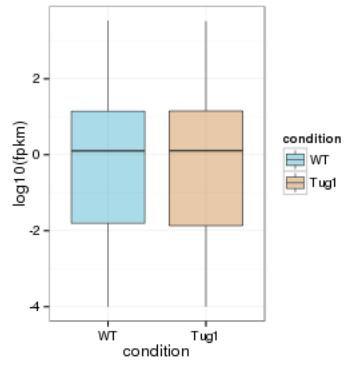
Scatterplot matrix



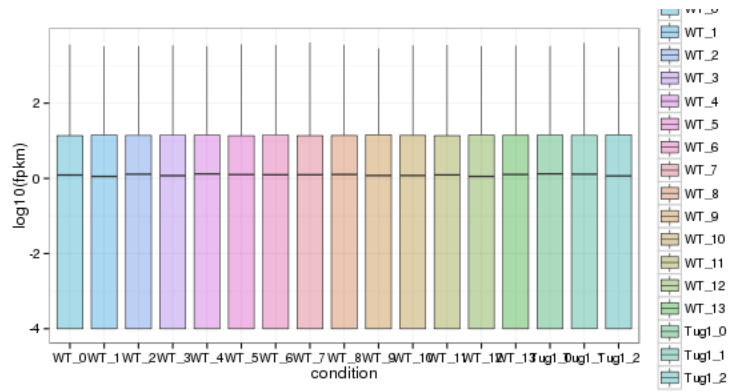
Distributions

Boxplots

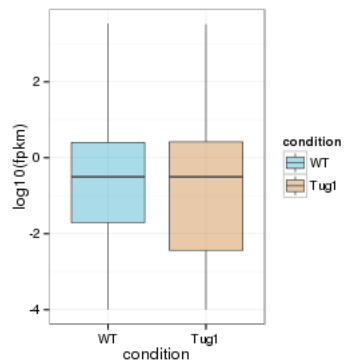
Boxplot (genes)



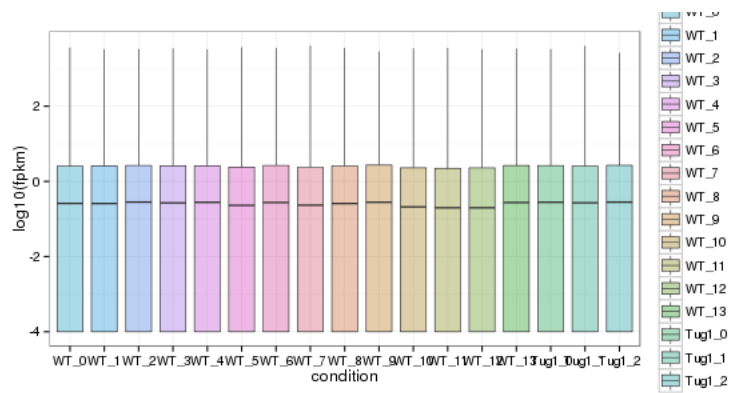
Boxplot (genes, replicates)



Boxplot (isoforms)

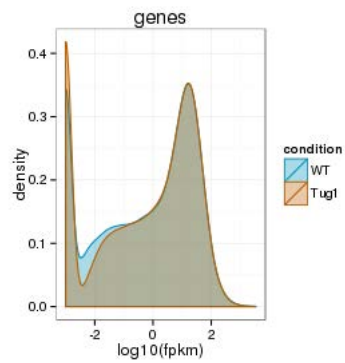


Boxplot (isoforms, replicates)

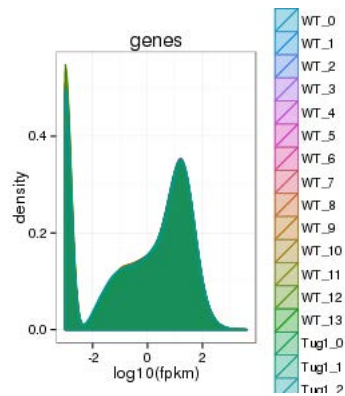


Density

Density (genes)

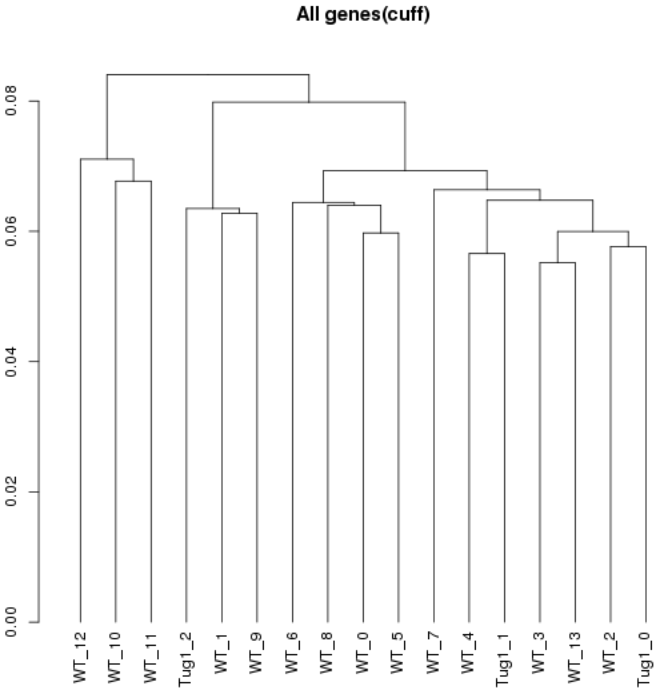


Density (genes, replicates)



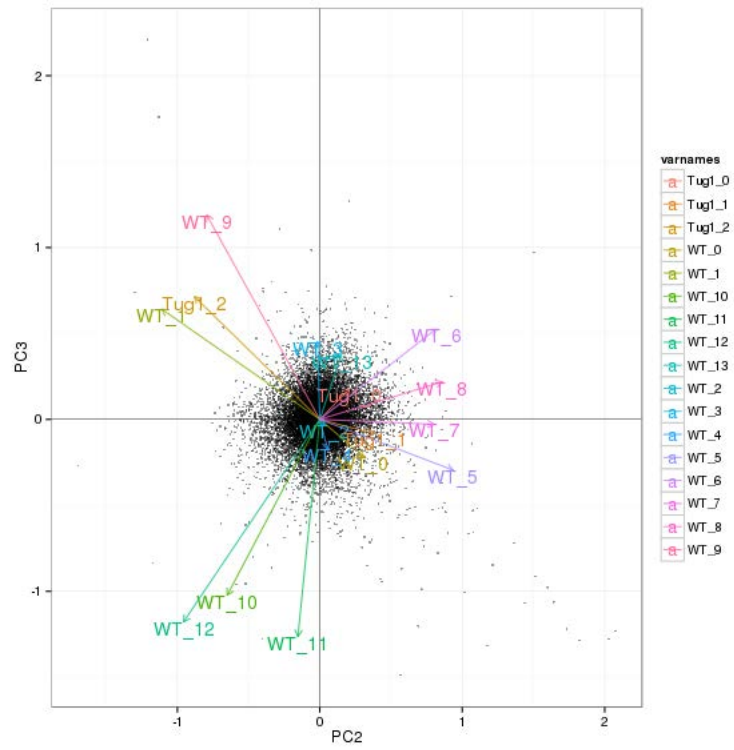
Clustering

Replicate Clusters

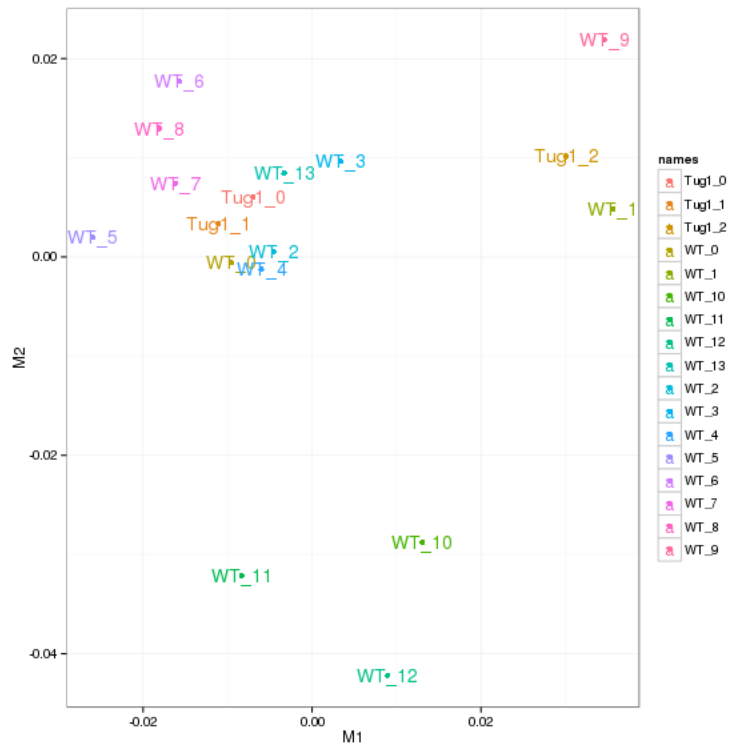


NULL

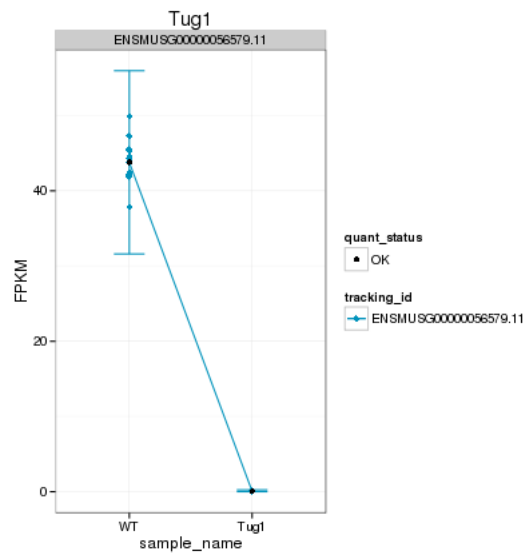
PCA (genes)



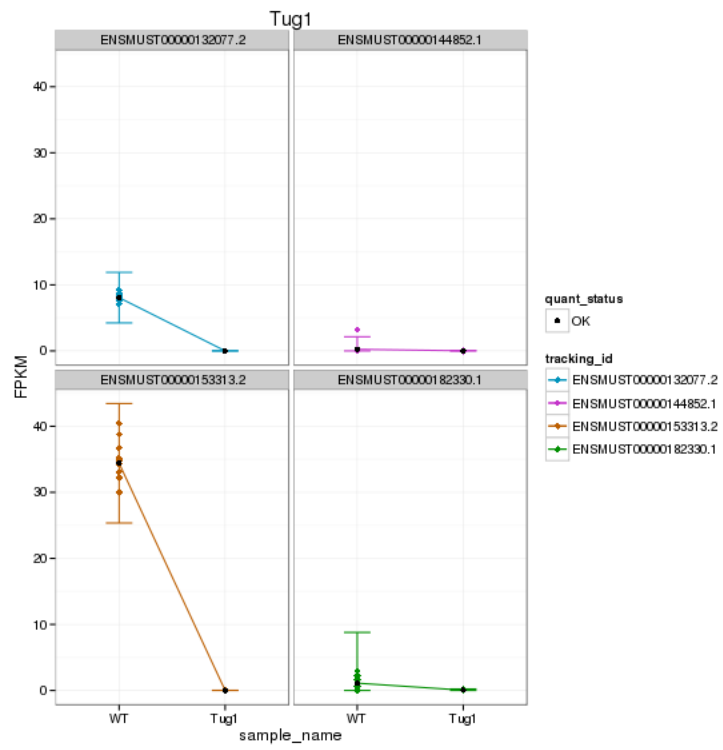
MDS (genes)



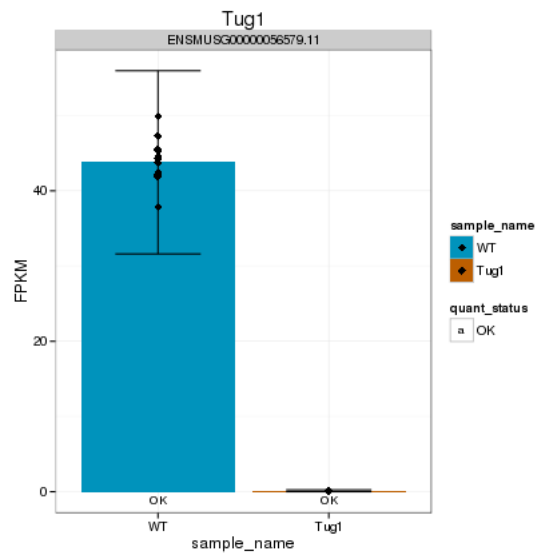
KO assessment
Endogenous IncRNA expression



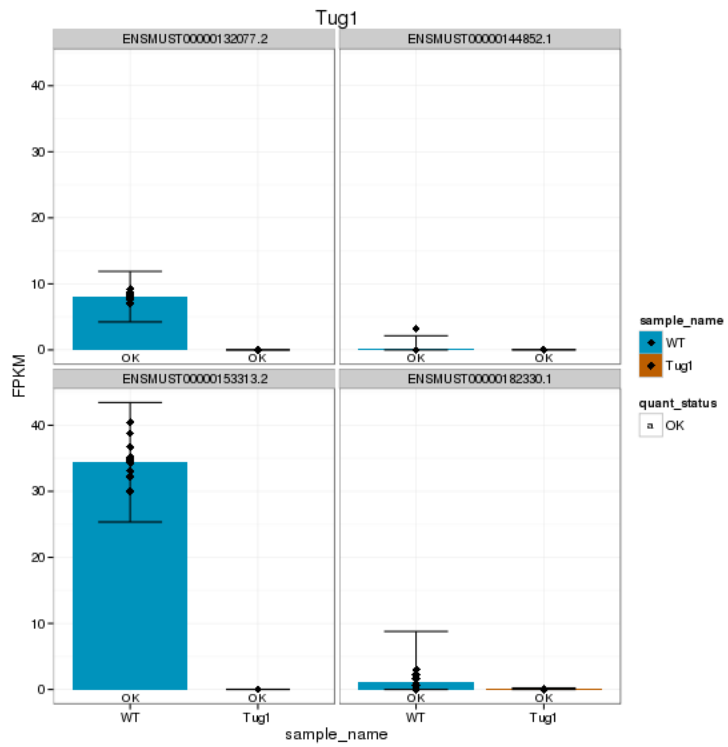
Endogenous expression of Tug1 isoforms:



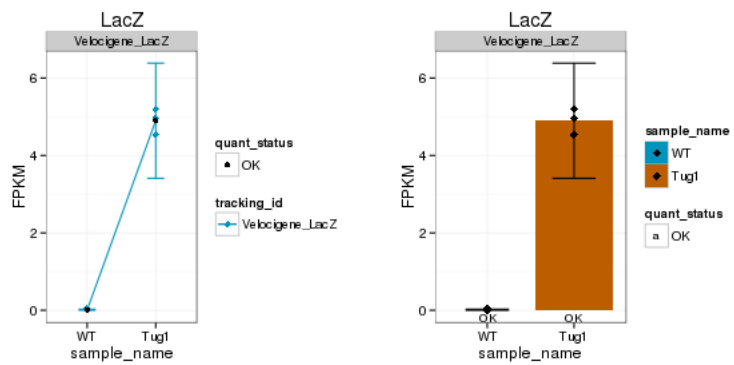
Barplot of gene expression:



Barplot of isoform expression:

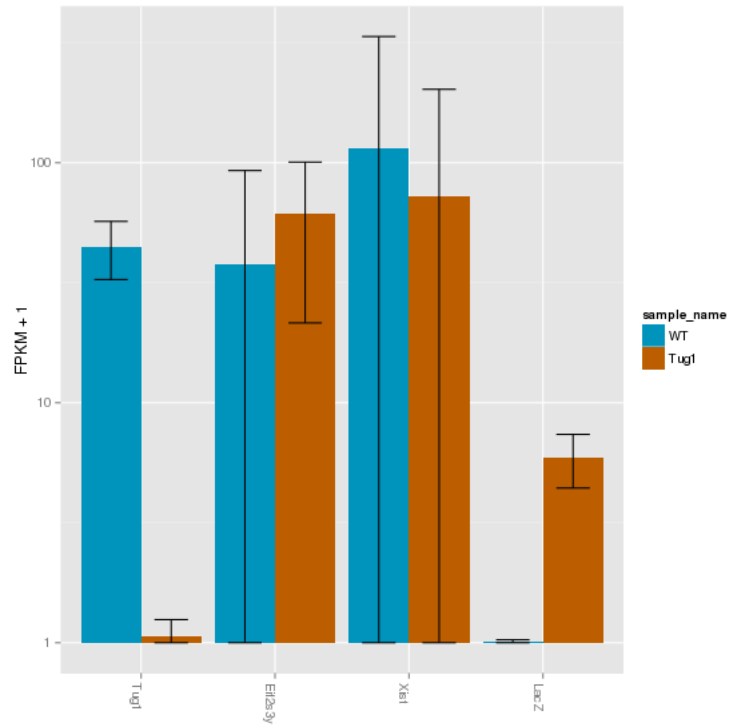


LacZ expression

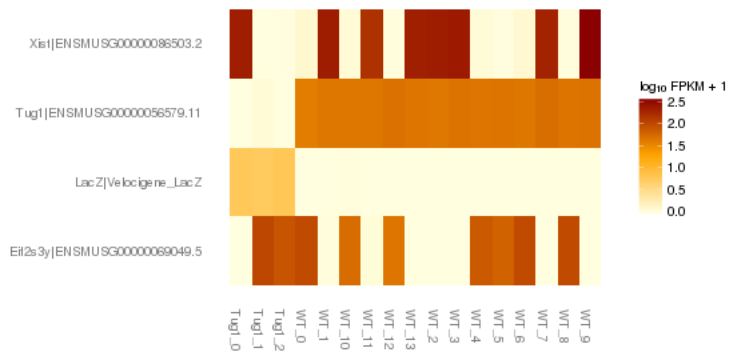


Digital Genotyping (LacZ vs Endogenous lincRNA and Sex)

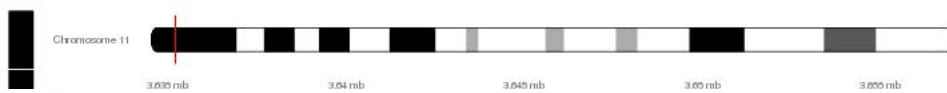
Eif2s3y is a y-expressed gene, Xist is an x-expressed gene Expression plot (endogenous linc, lacZ, Y-expressed gene):



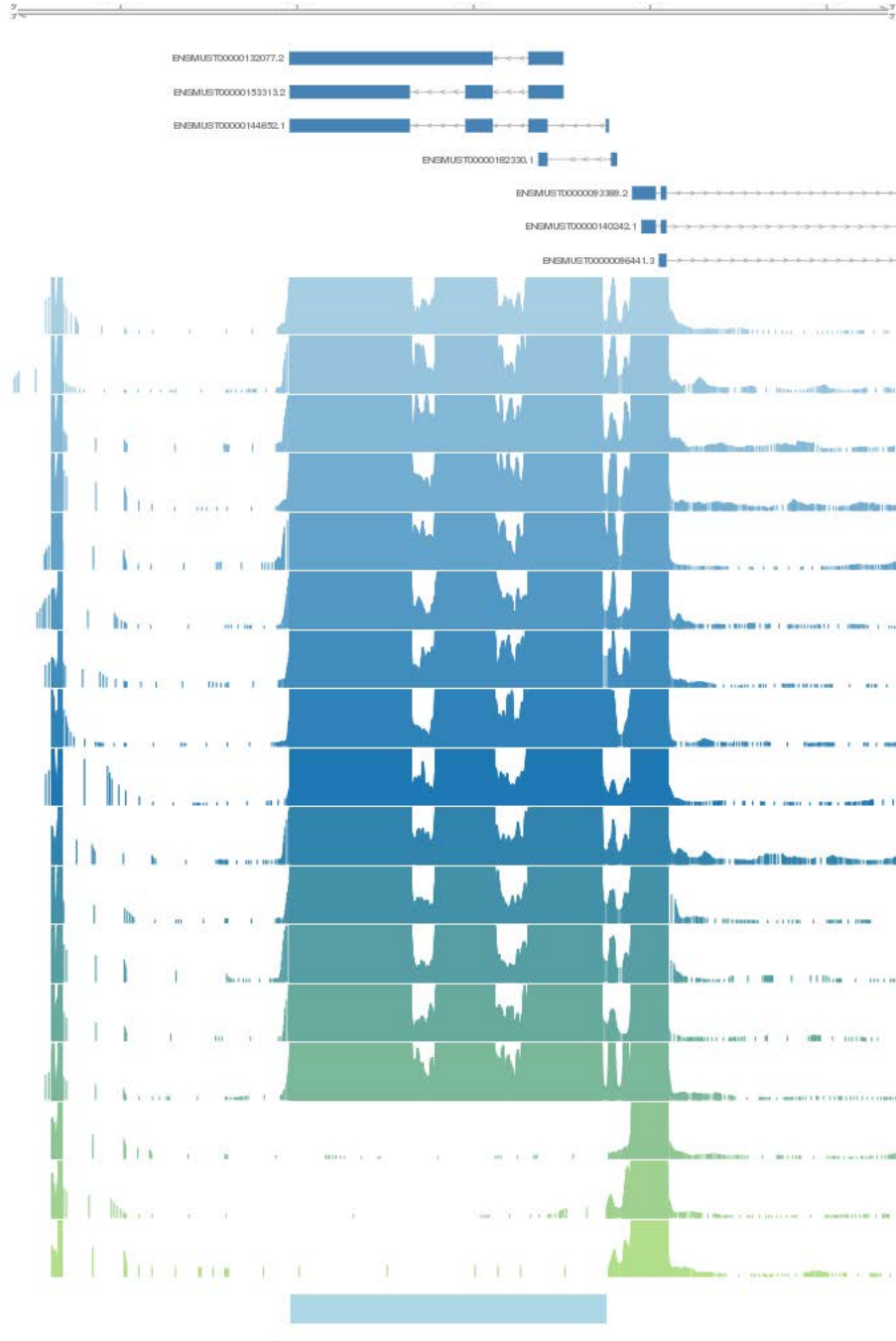
Expression heatmap:



Track visualization



KO Region WT_0 WT_1 WT_2 WT_3 WT_4 WT_5 WT_6 WT_7 WT_8 WT_9 WT_10 WT_11 WT_12 WT_13 Tug_0 Tug_1 Tug_2 LncRNA isoforms



Differential Analysis

Differential Genes

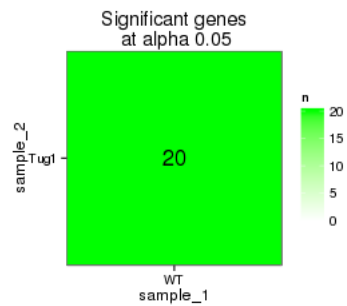
There are 20 significantly differentially expressed genes. They are:

geneAnnot\$gene_short_name

- 1 Rec8
- 2 Rnf185
- 3 Rps14
- 4 Morc2a
- 5 Inpp5j
- 6 8430429K09Rik
- 7 Fmod
- 8 Map10
- 9 Hbb-bs
- 10 Tug1
- 11 Kdm5d
- 12 Cox5b
- 13 Rps4y2
- 14 Eno1
- 15 Lyz2
- 16 Slfn9
- 17 3222401L13Rik
- 18 Hbb-bt
- 19 Selm
- 20 Gm26809

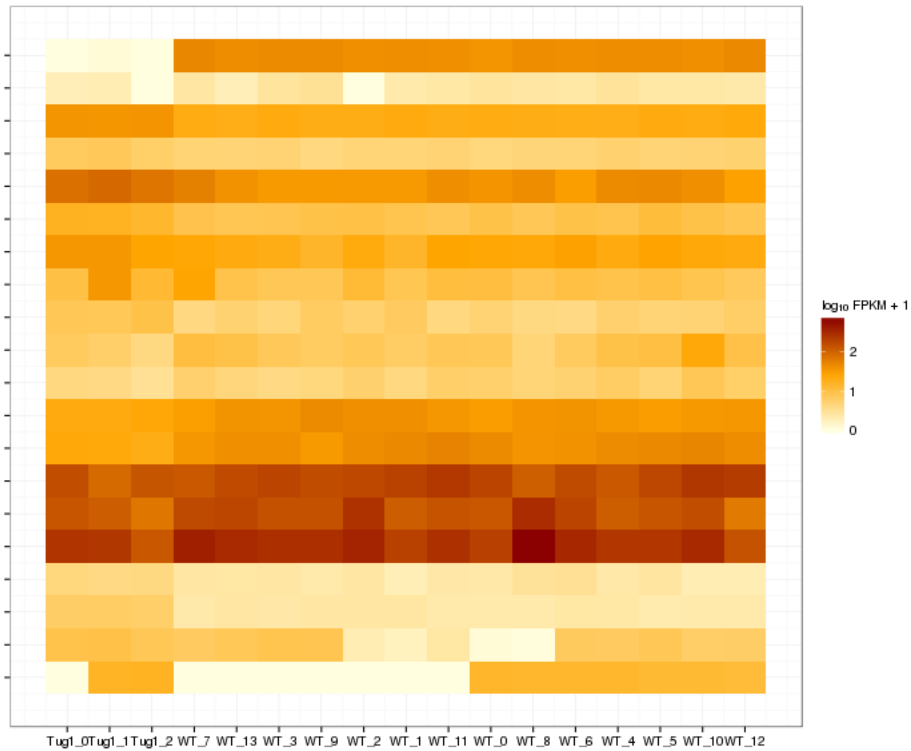
Matrix of gene significant differences between conditions

(skip for Brainmap wt-v-ko comparisons)



Significant gene expression differences between conditions

Expression plot (genes):(turned off)

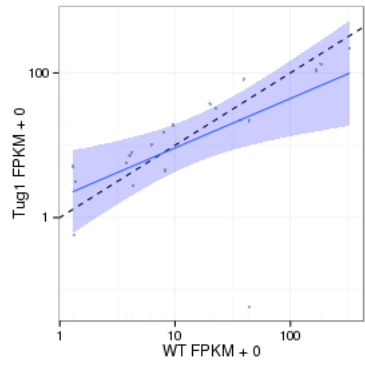


Significant genes with expression >50fpm (any condition):(turned off)

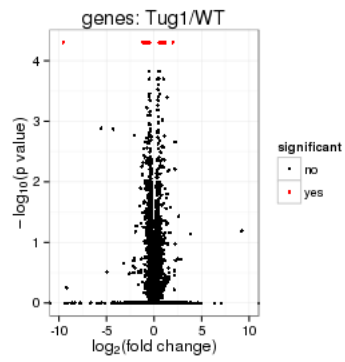
An individual look at each of the highly expressed significantly differentially regulated genes: (eval=false for first pass)

Expression-level/significance relationship

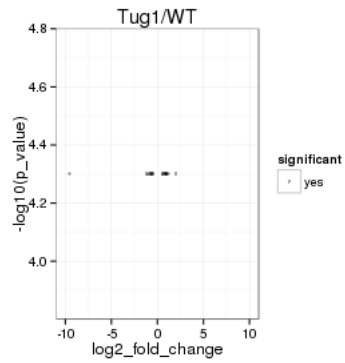
Scatter plot of significant genes only:



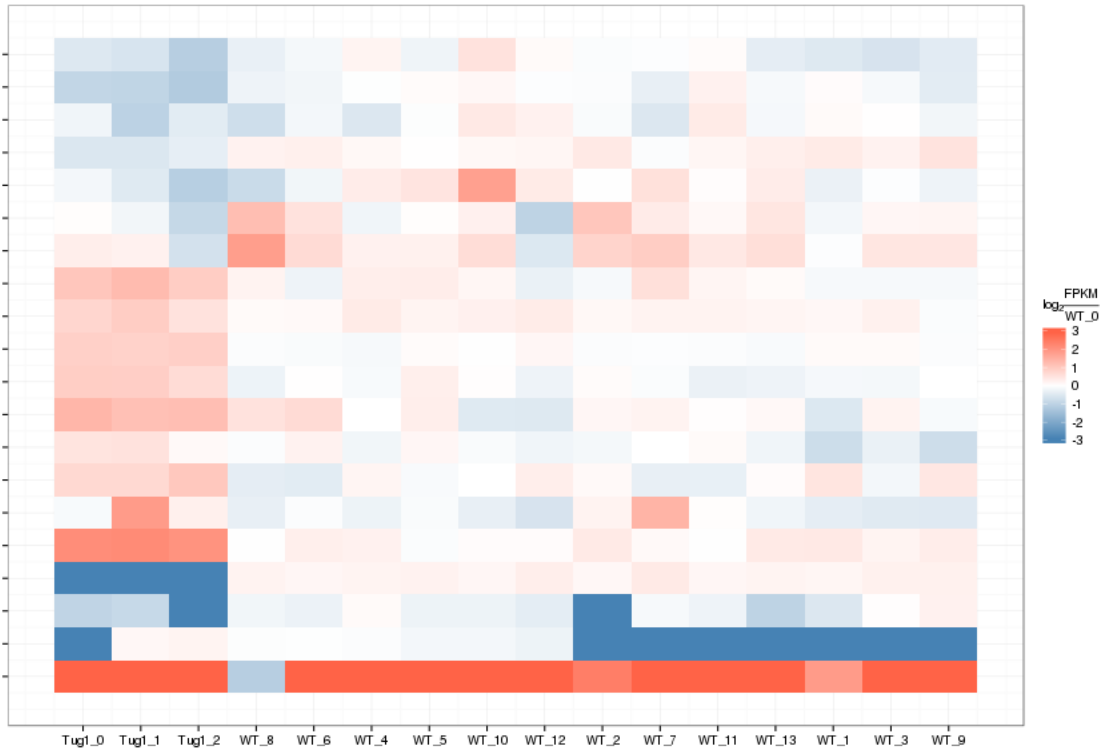
Volcano Plot



Volcano plot with significant genes only:



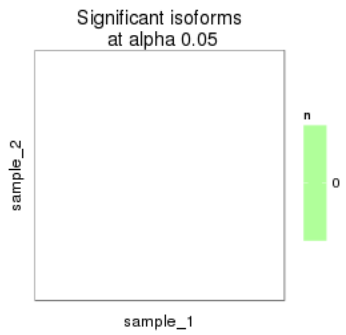
FoldChange Heatmap



Differential Splicing

Differential Isoforms between conditions

Per isoform difference between conditions:



These isoforms are:

[1] "no sig isoforms"

Gene-level DE isoform heatmap

```
## [1] "no sig isoforms"
```

Isoform foldchange heatmap by isoform:

```
## [1] "no sig isoforms"
```

Differential Splicing between conditions

(eval false for first pass)

Per condition differences in isoforms (Does gene have diff piechart between conditions?)

These genes are:

Splicing heatmap by isoform:

Splicing heatmap by gene

The following are significantly differentially spliced genes (relative portion of isoform per condition):

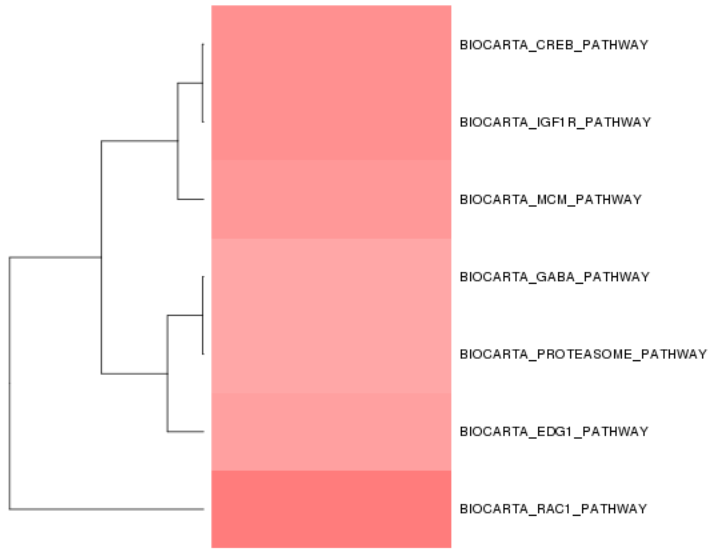
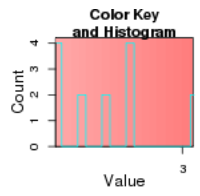
Gene/Pathway Analysis

GSEA

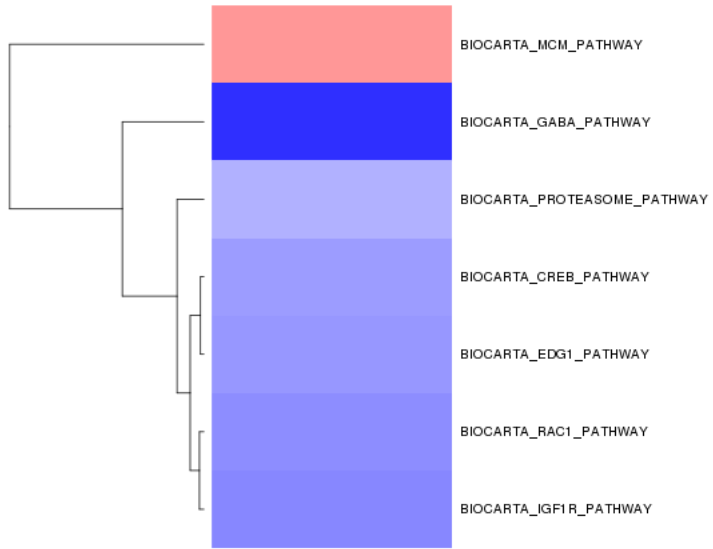
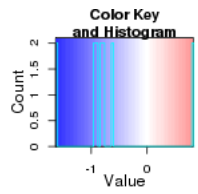
Enrichment and zscores are calculated based on expression in KO vs WT ($\text{fpkmKO}/\text{fpkmWT}$), so genes that are down regulated in KO are shown in blue, while upregulation is shown in red.

KO/WT Blue = down in KO Red = Up in KO

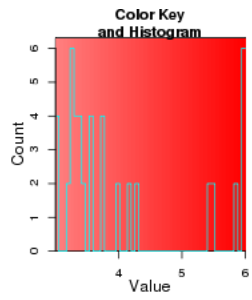
Biocarta enrichment:

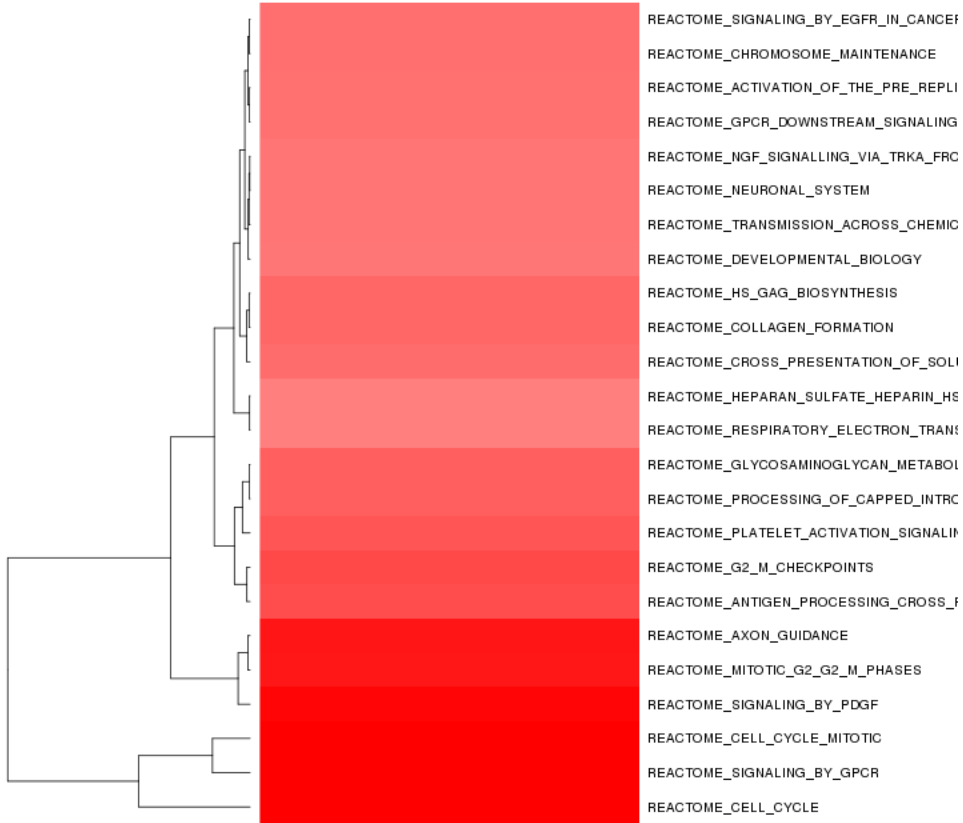


Biocarta zscore:

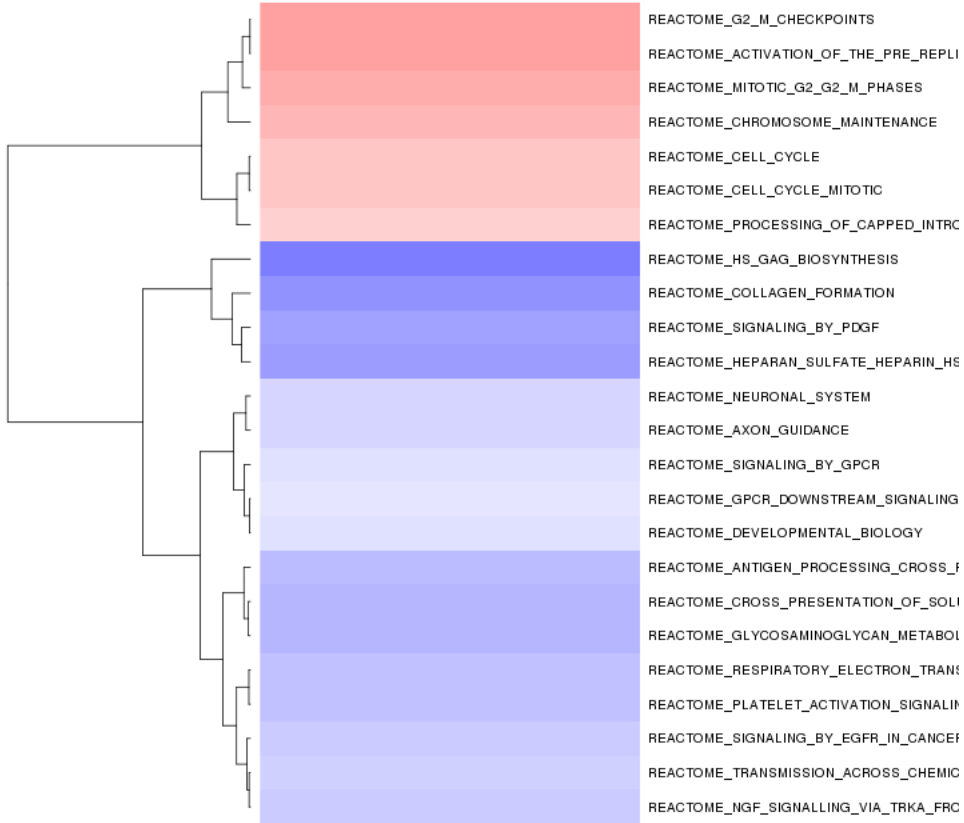
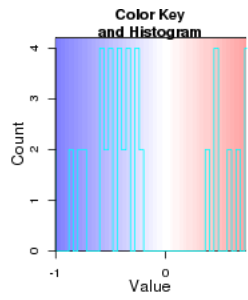


Reactome enrichment:

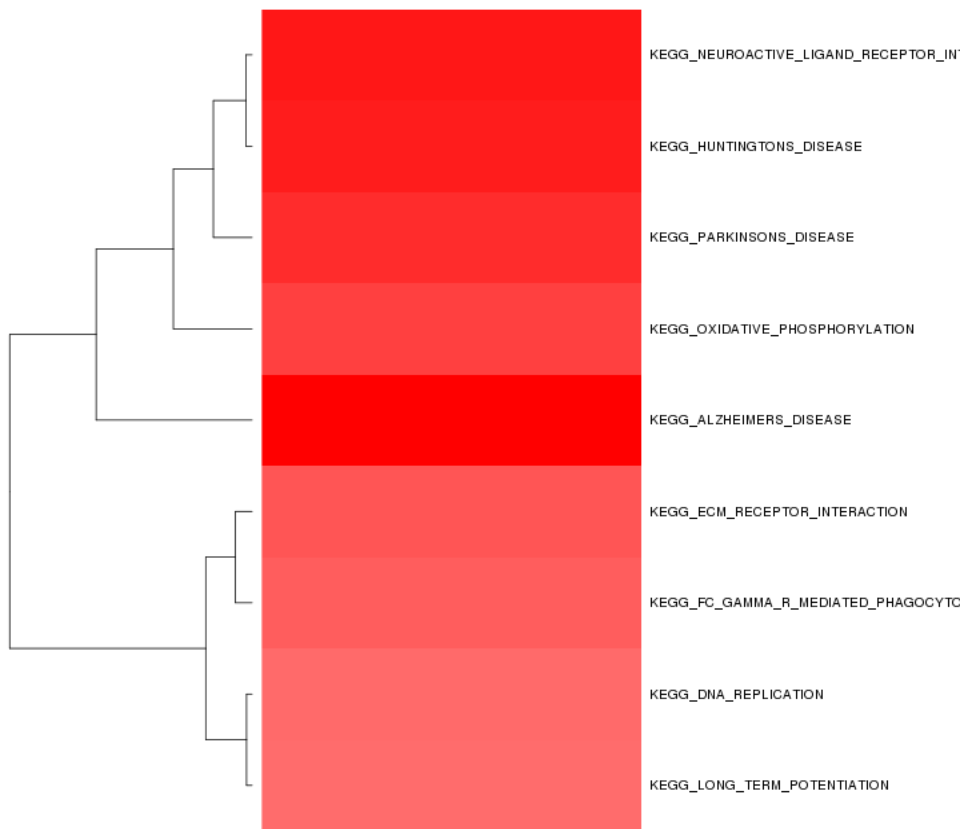
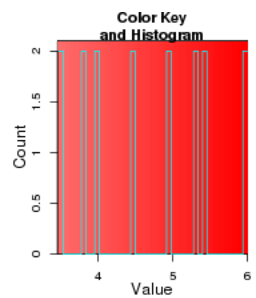




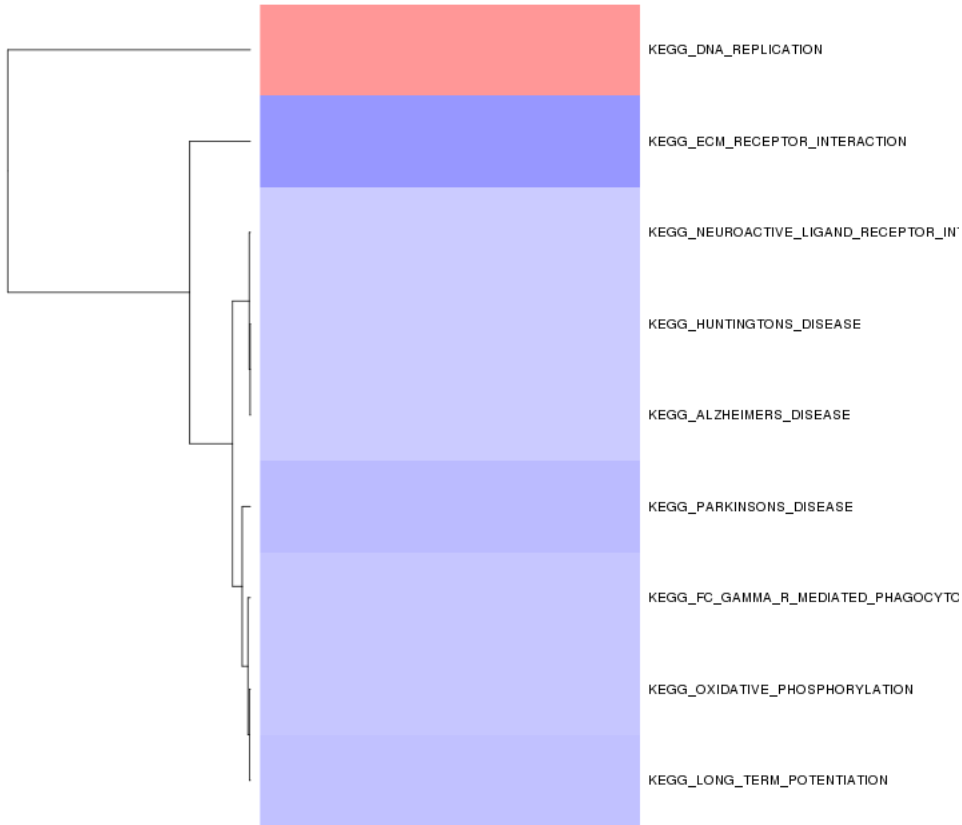
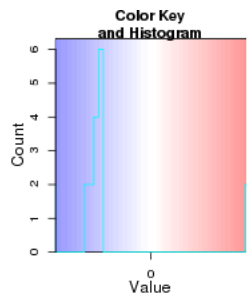
Reactome zscore:



Kegg enrichment:



Kegg zscore:



Interneuron enrichment:

```
## Error: argument is of length zero
```

Interneuron zscore:

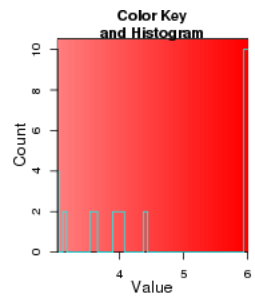
```
## Error: incorrect number of dimensions
```

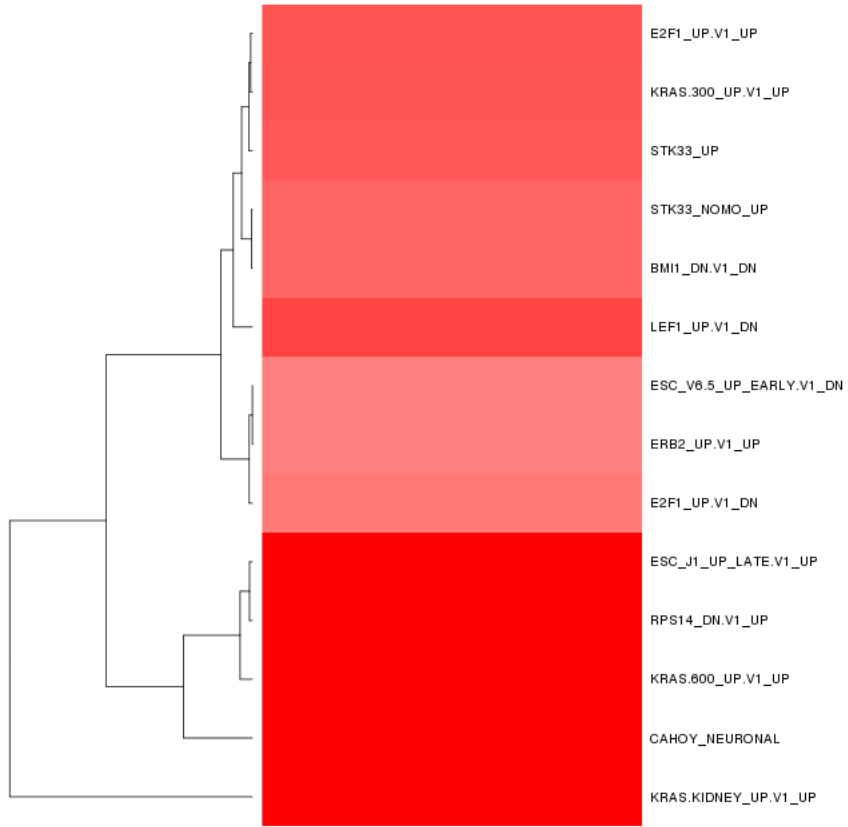
```
## Error: object 'x_ordered' not found
```

```
## Error: error in evaluating the argument 'x' in selecting a method for function 'as.matrix': Error: object 'x_ordered' not found
```

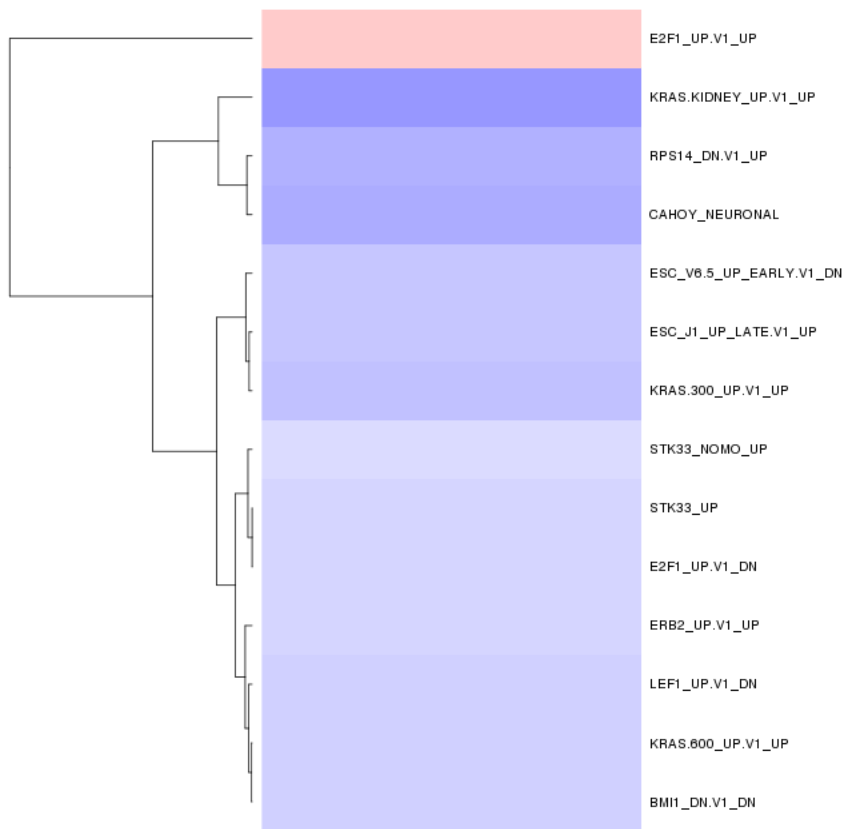
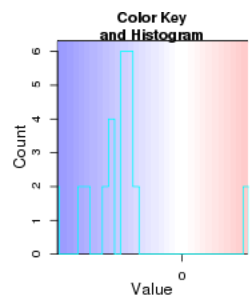
```
## Error: object 'x_ordered' not found
```

Oncogene enrichment:

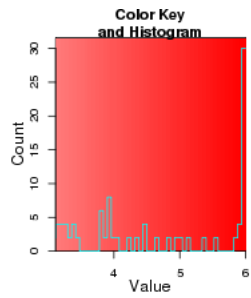




Oncogene zscore:



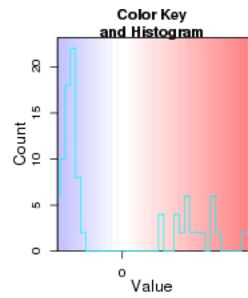
Immuno enrichment:

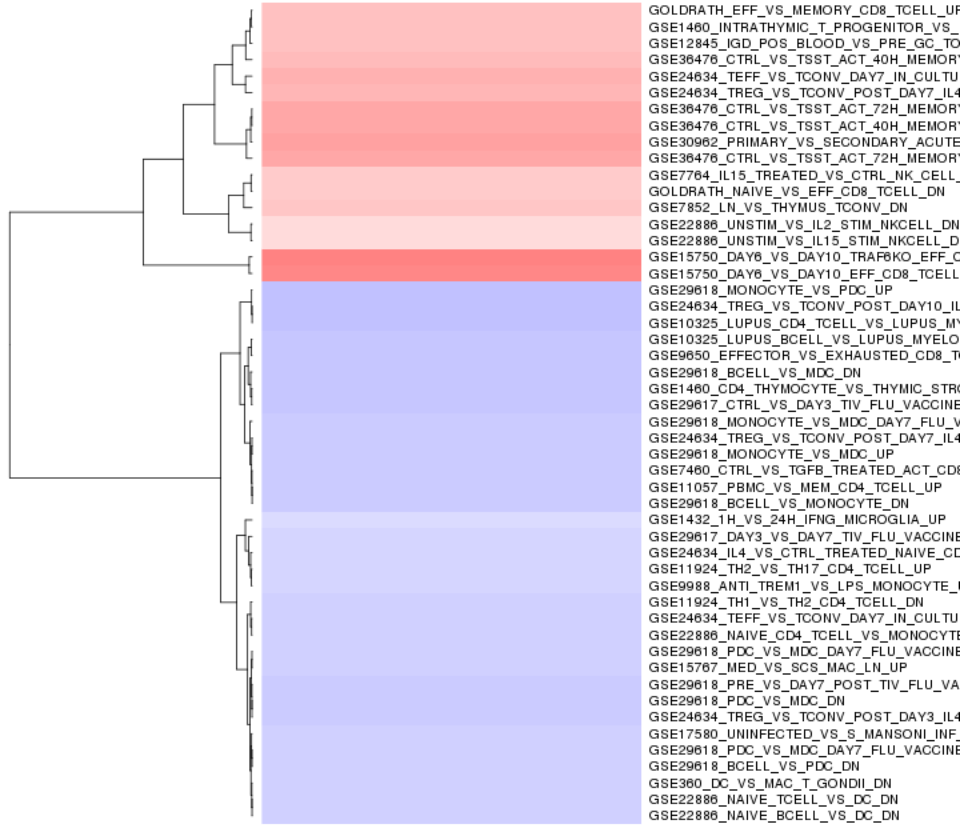




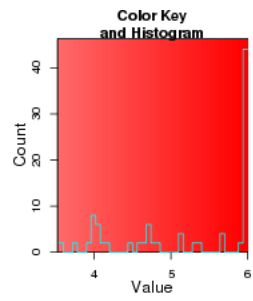
GSE22886_UNSTIM_VS_IL15_STIM_NKCELL_D
 GSE22886_NAIVE_CD4_TCELL_VS_MONOCYTE
 GSE29618_PDC_VS_MDC_DAY7_FLU_VACCINE
 GSE29618_PRE_VS_DAY7_POST_TIV_FLU_VA
 GSE9650_EFFECTOR_VS_EXHAUSTED_CD8_T
 GSE11057_PBMC_VS_MEM_CD4_TCELL_UP
 GSE22886_UNSTIM_VS_IL2_STIM_NKCELL_DN
 GSE29617_CTRL_VS_DAY3_TIV_FLU_VACCINE
 GSE24634_TREG_VS_TCONV_POST_DAY3_IL4
 GSE29618_BCELL_VS_PDC_DN
 GSE11924_TH2_VS_TH17_CD4_TCELL_UP
 GOLDRATH_EFF_VS_MEMORY_CD8_TCELL_UF
 GSE15767_MED_VS_SCS_MAC_LN_UP
 GSE17580_UNINFECTED_VS_S_MANSONI_INF
 GSE7852_LN_VS_THYMUS_TCONV_DN
 GSE1460_INTRATHYMIC_T_PROGENITOR_VS_
 GSE29618_BCELL_VS_MONOCYTE_DN
 GSE11924_TH1_VS_TH2_CD4_TCELL_DN
 GSE24634_TREG_VS_TCONV_POST_DAY7_IL4
 GOLDRATH_NAIVE_VS_EFF_CD8_TCELL_DN
 GSE7460_CTRL_VS_TGFB_TREATED_ACT_CD4
 GSE22886_NAIVE_TCELL_VS_DC_DN
 GSE29618_PDC_VS_MDC_DN
 GSE360_DC_VS_MAC_T_GONDII_DN
 GSE9988_ANTI_TREM1_VS_LPS_MONOCYTE_I
 GSE22886_NAIVE_BCELL_VS_DC_DN
 GSE1432_IH_VS_24H_IFNG_MICROGLIA_UP
 GSE29618_PDC_VS_MDC_DAY7_FLU_VACCINE
 GSE24634_IL4_VS_CTRL_TREATED_NAIVE_CI
 GSE29617_DAY3_VS_DAY7_TIV_FLU_VACCINE
 GSE10239_NAIVE_VS_MEMORY_CD8_TCELL_I
 GSE7764_IL15_TREATED_VS_CTRL_NK_CELL
 GSE1460_CD4_THYMOCYTE_VS_THYMIC_STR
 GSE29618_BCELL_VS_MDC_DN
 GSE29618_MONOCYTE_VS_MDC_DAY7_FLU_V
 GSE10325_LUPUS_BCELL_VS_LUPUS_MYELO
 GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR
 GSE10325_LUPUS_CD4_TCELL_VS_LUPUS_M
 GSE29618_MONOCYTE_VS_MDC_UP
 GSE12845_IGD_POS_BLOOD_VS_PRE_GC_TO
 GSE24634_TREG_VS_TCONV_POST_DAY7_IL4
 GSE30962_PRIMARY_VS_SECONDARY_ACUTE
 GSE36476_CTRL_VS_TSST_ACT_40H_MEMOR
 GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR
 GSE36476_CTRL_VS_TSST_ACT_72H_MEMOR
 GSE24634_TEFF_VS_TCONV_DAY7_IN_CULTU
 GSE24634_TREG_VS_TCONV_POST_DAY10_IL
 GSE29618_MONOCYTE_VS_PDC_UP
 GSE15750_DAY6_VS_DAY10_EFF_CD8_TCELL
 GSE15750_DAY6_VS_DAY10_TRAF6KO_EFF_C

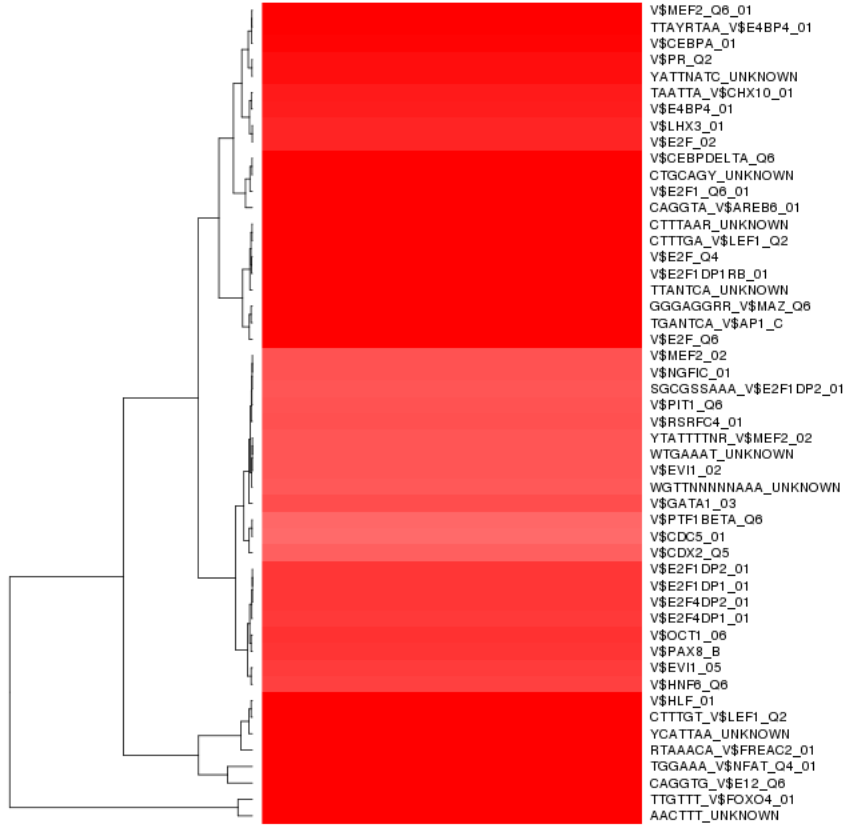
Immuno zscore:





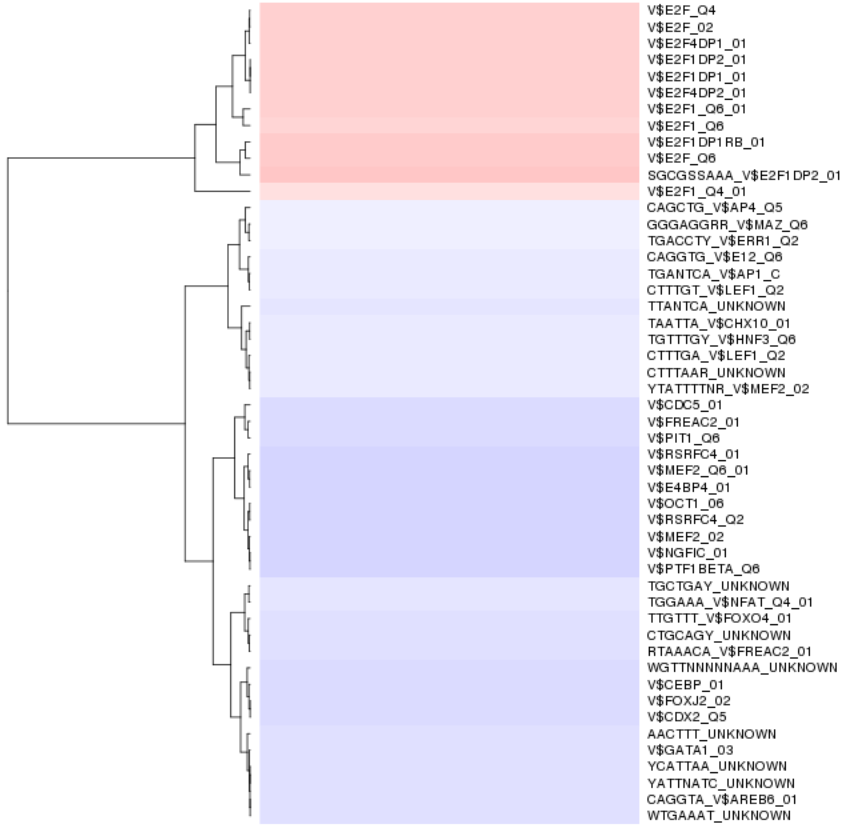
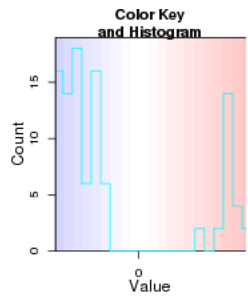
TF enrichment:





V\$MEF2_Q6_01
 TTAYRTAA_V\$E4BP4_01
 V\$CEBPA_01
 V\$PR_Q2
 YATTNATC_UNKNOWN
 TAATTA_V\$CHX10_01
 V\$E4BP4_01
 V\$LHX3_01
 V\$E2F_02
 V\$CEBPDELTA_Q6
 CTGCAGY_UNKNOWN
 V\$E2F1_Q6_01
 CAGGTA_V\$AREB6_01
 CTTTAAR_UNKNOWN
 CTTTGA_V\$LEF1_Q2
 V\$E2F_Q4
 V\$E2F1DP1RB_01
 TTANTCA_UNKNOWN
 GGGAGGRR_V\$MAZ_Q6
 TGANTCA_V\$AP1_C
 V\$E2F_Q6
 V\$MEF2_Q2
 V\$NGFIC_01
 SGGSSAAA_V\$E2F1DP2_01
 V\$PIT1_Q6
 V\$RSRRC4_01
 YATTTTTNR_V\$MEF2_Q2
 WTGAAAT_UNKNOWN
 V\$EV11_Q2
 WGTNNNNHAAA_UNKNOWN
 V\$GATA1_Q3
 V\$PTF1BETA_Q6
 V\$CDC5_Q1
 V\$CDX2_Q5
 V\$E2F1DP2_01
 V\$E2F1DP1_01
 V\$E2F4DP2_01
 V\$E2F4DP1_01
 V\$OCT1_Q6
 V\$PAX8_B
 V\$EV11_Q5
 V\$HNF8_Q6
 V\$HLF_Q1
 CTTTGT_V\$LEF1_Q2
 YCAATTA_UNKNOWN
 RTAAACA_V\$FREAC2_01
 TGGAAA_V\$NFAT_Q4_01
 CAGGTG_V\$E12_Q6
 TTGTTT_V\$FOXO4_01
 AACTTT_UNKNOWN

TF zscore:



GO enrichment

Cluster profiler used to call enrichments of significantly differentially regulated genes that map to Entrez IDs.

```
## Error: 'x' and 'units' must have length > 0
```

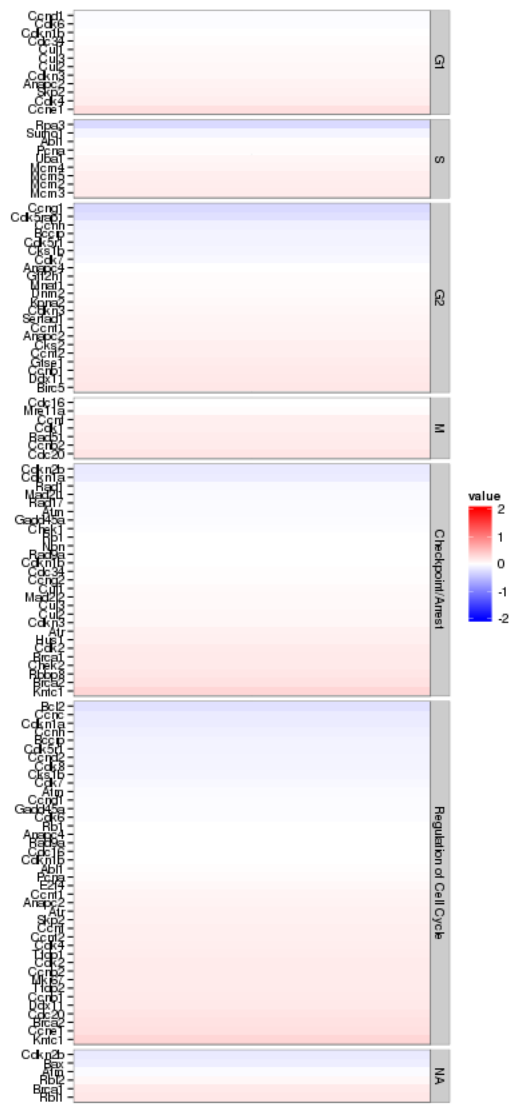
```
## Error: 'x' and 'units' must have length > 0
```

```
## Error: 'x' and 'units' must have length > 0
```

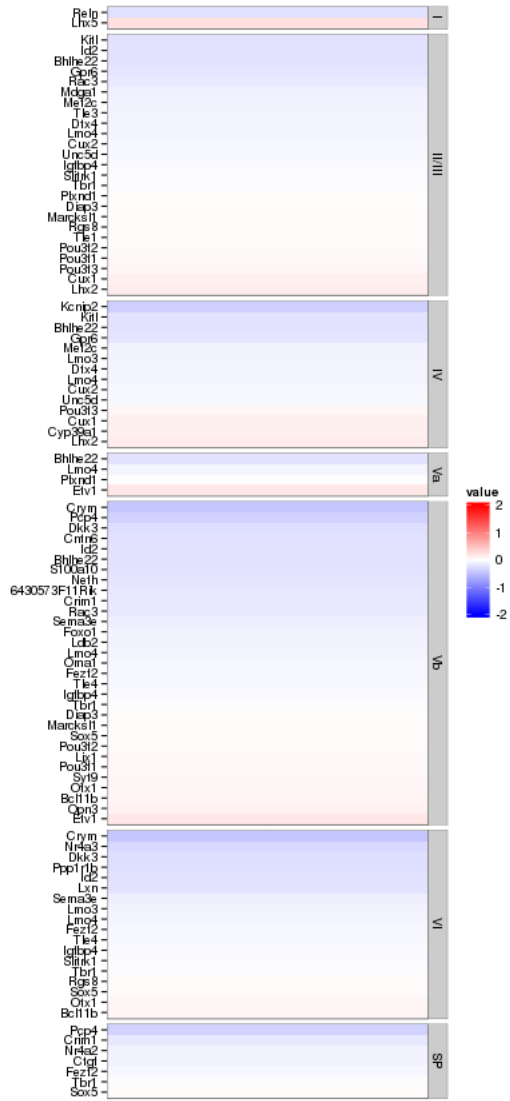
```
## Error: need finite 'xlim' values
```

```
## Error: need finite 'xlim' values
```

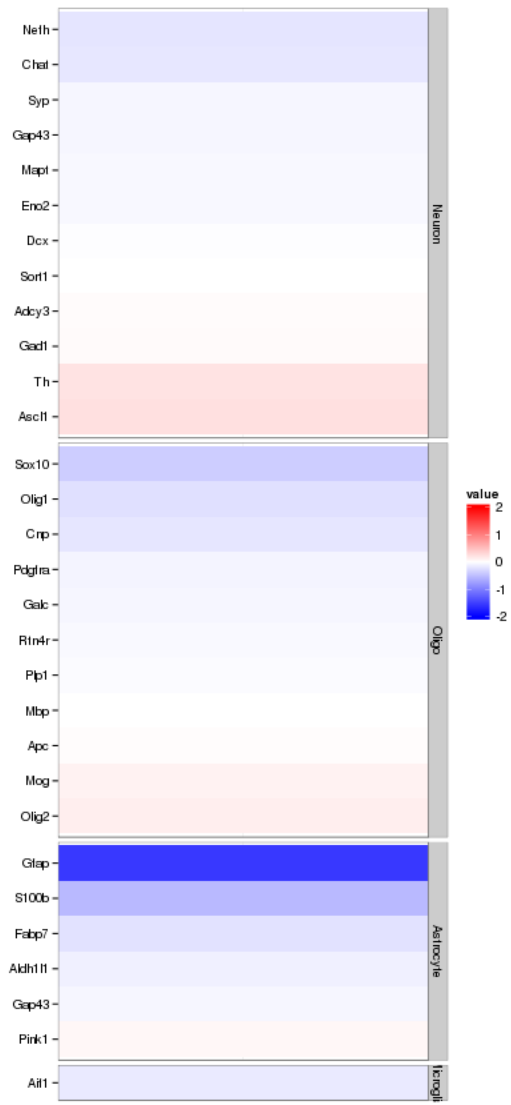
Enrichment or depletion for stage-specific cell cycle markers



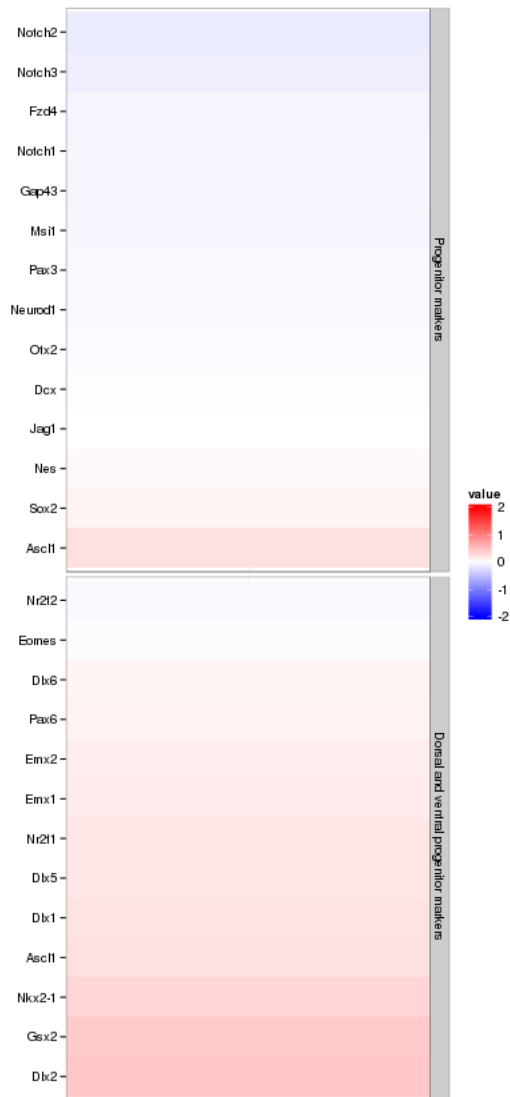
Enrichment or depletion for markers of specific cortical layers



Enrichment or depletion for specific neural cell types



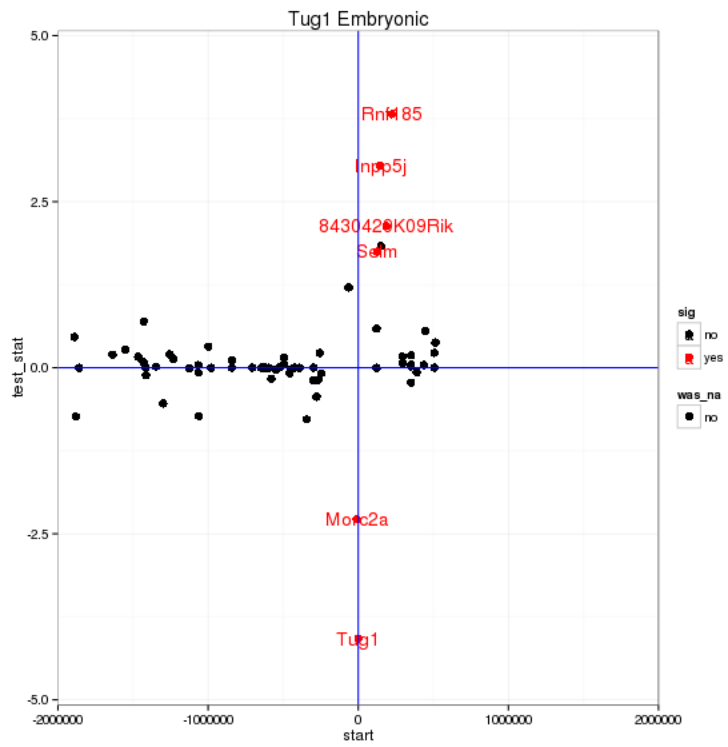
Enrichment or Depletion of neural differentiation markers



Cis vs Trans (locally)

log2 Foldchange and test statistic are calculated with the ratio of fpkm(KO)/fpkm(WT), thus the test_stat is positive if a gene is higher in the KO and negative if a gene has lower expression in the KO

The pvalue for 5 genes significantly regulated in a region this size is: 0.0023



Notes

Samples used are:

10

- 1 JR753
- 2 JR750
- 3 JR771
- 4 JR755
- 5 JR811
- 6 JR768
- 7 JR761
- 8 JR815
- 9 JR789
- 10 JR748
- 11 JR716
- 12 JR717
- 13 JR719
- 14 JR756

15 JR757
16 JR857
17 JR861

Replicates

file	sample_name	replicate	rep_name	total_mass	norm_mass	internal_scale	external_scale
1 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR753/abundances.cxb	WT	0	WT_0	39331900.00	33091500.00	1.23	1.00
2 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR750/abundances.cxb	WT	1	WT_1	36031100.00	33091500.00	1.07	1.00
3 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR771/abundances.cxb	WT	2	WT_2	38857100.00	33091500.00	1.20	1.00
4 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR755/abundances.cxb	WT	3	WT_3	41811300.00	33091500.00	1.25	1.00
5 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR811/abundances.cxb	WT	4	WT_4	38172800.00	33091500.00	1.17	1.00
6 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR768/abundances.cxb	WT	5	WT_5	28375200.00	33091500.00	0.87	1.00
7 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR761/abundances.cxb	WT	6	WT_6	31706200.00	33091500.00	0.95	1.00
8 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR815/abundances.cxb	WT	7	WT_7	29733100.00	33091500.00	0.90	1.00
9 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR789/abundances.cxb	WT	8	WT_8	27681500.00	33091500.00	0.84	1.00
10 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR748/abundances.cxb	WT	9	WT_9	32367000.00	33091500.00	0.94	1.00
11 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR716/abundances.cxb	WT	10	WT_10	26144700.00	33091500.00	0.79	1.00
12 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR717/abundances.cxb	WT	11	WT_11	25565400.00	33091500.00	0.78	1.00
13 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR719/abundances.cxb	WT	12	WT_12	22697800.00	33091500.00	0.67	1.00
14 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR756/abundances.cxb	WT	13	WT_13	38948900.00	33091500.00	1.18	1.00
15 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR757/abundances.cxb	Tug1	0	Tug1_0	33398300.00	33091500.00	1.02	1.00
16 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR857/abundances.cxb	Tug1	1	Tug1_1	39157600.00	33091500.00	1.22	1.00
17 /n/rinn_data1/seq/fgoff/Projects/BrainMap/data/quants/JR861/abundances.cxb	Tug1	2	Tug1_2	43124000.00	33091500.00	1.27	1.00

Session Info

```
## R version 3.0.2 (2013-09-25)
## Platform: x86_64-unknown-linux-gnu (64-bit)
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
## [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
## [9] LC_ADDRESS=C             LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] grid      parallel  methods  stats    graphics  grDevices  utils
## [8] datasets  base
##
## other attached packages:
## [1] plyr_1.8.1
## [2] stringr_0.6.2
## [3] seqbias_1.10.0
```



```

## [4] BSgenome.Mmusculus.UCSC.mm10_1.3.19
## [5] BSgenome_1.30.0
## [6] Biostrings_2.30.1
## [7] gdata_2.13.3
## [8] GO.db_2.10.1
## [9] org.Mm.eg.db_2.10.1
## [10] clusterProfiler_1.13.1
## [11] DOSE_2.0.0
## [12] ReactomePA_1.6.1
## [13] AnnotationDbi_1.24.0
## [14] Biobase_2.22.0
## [15] mgcv_1.7-29
## [16] nlme_3.1-117
## [17] RMySQL_0.9-3
## [18] RColorBrewer_1.0-5
## [19] gridExtra_0.9.1
## [20] gtable_0.1.2
## [21] marray_1.40.0
## [22] gplots_2.13.0
## [23] GSA_1.03
## [24] limma_3.18.13
## [25] xtable_1.7-3
## [26] cummeRbund_2.7.2
## [27] Gviz_1.6.0
## [28] rtracklayer_1.22.7
## [29] GenomicRanges_1.14.4
## [30] XVector_0.2.0
## [31] IRanges_1.20.7
## [32] fastcluster_1.1.13
## [33] reshape2_1.4
## [34] ggplot2_1.0.0
## [35] RSQLite_0.11.4
## [36] DBI_0.2-7
## [37] BiocGenerics_0.8.0
## [38] knitr_1.6
##
## loaded via a namespace (and not attached):
## [1] biomaRt_2.18.0      biovizBase_1.10.8   bitops_1.0-6
## [4] caTools_1.17        cluster_1.15.2      colorspace_1.2-4
## [7] dichromat_2.0-0     digest_0.6.4        DO.db_2.7
## [10] evaluate_0.5.5      formatR_0.10        Formula_1.1-1
## [13] GenomicFeatures_1.14.5 GOSemSim_1.20.3     graph_1.40.1
## [16] graphite_1.8.1      gtools_3.4.1        Hmisc_3.14-4
## [19] igraph_0.7.1        KEGG.db_2.10.1      KernSmooth_2.23-12
## [22] labeling_0.2        lattice_0.20-29     latticeExtra_0.6-26
## [25] MASS_7.3-33         Matrix_1.1-3        munsell_0.4.2
## [28] org.Hs.eg.db_2.10.1 proto_0.3-10        qvalue_1.36.0
## [31] Rcpp_0.11.2         RCurl_1.95-4.1      reactome.db_1.46.1
## [34] Rsamtools_1.14.3    scales_0.2.4        splines_3.0.2
## [37] stats4_3.0.2        survival_2.37-7     tcltk_3.0.2
## [40] tools_3.0.2         XML_3.98-1.1        zlibbioc_1.8.0

```

Run Info

```
##          param
## 1      cmd_line
## 2          version
## 3  SVN_revision
## 4  boost_version
## 5          genome
##
## 1 cuffdiff -p 10 -L WT,Tug1 -o /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/diffs/Tug1_vs_WT_Embryonic /n/rinn_data1/seq/lgoff/Projects/BrainMap/data/
## 2
## 3
## 4
## 5
```