

Vignette Title

Vignette Author

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Package

About

Function `test_diff`

Expression

What is it?

About dataset

In this vignette, we will work with the data set `BRCA_mRNAseq_chr17`, which contains information about gene expression. This data set contains per-gene read counts computed for genes from 17 chromosome for 100 patients with breast cancer.

```
load("BRCA_mRNAseq_chr17.rda")
```

```
BRCA_mRNAseq_chr17[1:5,1:5]
```

##		SUBTYPE	AANAT	AARSD1	AATF	AATK
##	TCGA-A1-AOSB-01A-11R-A144-07	Normal	9	2354	2870	317
##	TCGA-A1-AOSD-01A-11R-A115-07	LumA	2	1846	5656	312
##	TCGA-A1-AOSE-01A-11R-A084-07	LumA	11	3391	9522	736
##	TCGA-A1-AOSF-01A-11R-A144-07	LumA	0	2169	4625	169
##	TCGA-A1-AOSG-01A-11R-A144-07	LumA	1	2273	3473	92

Nbinom test

Testa for differences between the base means of two conditions. Beside a data set containing read counts, we need a description of the samples, which we keep in a factor, whose elements correspond to different groups.

In our example we will test for differential expression between groups with LumA breast cancer subtype and other subtypes of that cancer. We will use factor `conditions`, which consist of two values corresponds to subtype of breast cancer: LumA and other.

```
condition<-ifelse(BRCA_mRNAseq_chr17$SUBTYPE=="LumA","LumA","other")
head(condition,8)
```

```
## [1] "other" "LumA" "LumA" "LumA" "LumA" "LumA" "other" "LumA"
```

Evaluation of `expr_nbinom` may take a few minutes

```
test.expr <- test_diff(BRCA_mRNAseq_chr17[,1], condition, "nbinom")
```

As a result we obtain a data frame contains among others: base mean, means for each group and p-value of test.

```
head(test.expr)
```

```
##      id    exp.mean exp.mean.gr1 exp.mean.g2  exp.fold exp.log.fold
## 1  AANAT    3.455436    3.901537    2.952385  0.7567235 -0.40216187
## 2  AARSD1  2779.448414  2712.105777  2855.387983  1.0528306  0.07427334
## 3   AATF  6750.269650  7042.041468  6421.250366  0.9118450 -0.13313947
## 4   AATK   352.805108   355.415829   349.861103  0.9843712 -0.02272566
## 5  ABCA5  1933.257431  1888.913529  1983.262257  1.0499487  0.07031881
## 6  ABCA6   689.547294   589.046234   802.878277  1.3630140  0.44680041
##   exp.pval exp.padj
## 1 0.9453634 0.9920531
## 2 0.6627296 0.8992428
## 3 0.6864598 0.9111193
## 4 0.8051408 0.9585881
## 5 0.5837682 0.8489059
## 6 0.1289441 0.3857753
```

Metylation

What is it?

About dataset

Data set BRCA_methylation_chr17 contains information about methylation of CpG islands located on 17 chromosome for patient with breast cancer.

```
load("BRCA_methylation_chr17.rda")
head(BRCA_methylation_chr17)[1:5,1:5]
```

```
##                                SUBTYPE cg00021527 cg00031162 cg00032227
## TCGA-A1-A0SD-01A-11D-A112-05      LumA 0.03781858 0.7910348 0.006391233
## TCGA-A2-A04N-01A-11D-A112-05      LumA 0.01437552 0.7359370 0.008752293
## TCGA-A2-A04P-01A-31D-A032-05      Basal 0.01360124 0.6967802 0.009442039
## TCGA-A2-A04Q-01A-21D-A032-05      Basal 0.01525656 0.5341244 0.014674247
## TCGA-A2-A04T-01A-21D-A032-05      Basal 0.01167384 0.7378100 0.012251559
##                                cg00050312
## TCGA-A1-A0SD-01A-11D-A112-05 0.02356901
## TCGA-A2-A04N-01A-11D-A112-05 0.02770303
## TCGA-A2-A04P-01A-31D-A032-05 0.01402589
## TCGA-A2-A04Q-01A-21D-A032-05 0.01510194
## TCGA-A2-A04T-01A-21D-A032-05 0.01174021
```

mapping to genes using function map_to_gene

```
BRCA_methylation_chr17_gen <- map_to_gene(BRCA_methylation_chr17[, -1])
head(BRCA_methylation_chr17_gen)[1:5,1:5]
```

```
##                                AANAT    AARSD1    AATF    AATK
## TCGA-A1-A0SD-01A-11D-A112-05 0.7148533 0.8625816 0.24294092 0.7835302
## TCGA-A2-A04N-01A-11D-A112-05 0.5850106 0.8355825 0.21367129 0.8466190
## TCGA-A2-A04P-01A-31D-A032-05 0.4495537 0.8786166 0.03277413 0.3417919
## TCGA-A2-A04Q-01A-21D-A032-05 0.7120650 0.8819490 0.03460160 0.7264985
## TCGA-A2-A04T-01A-21D-A032-05 0.6010397 0.7739978 0.02501599 0.6276399
##                                ABC1
## TCGA-A1-A0SD-01A-11D-A112-05 0.01401806
```

```
## TCGA-A2-A04N-01A-11D-A112-05 0.01275865
## TCGA-A2-A04P-01A-31D-A032-05 0.01455092
## TCGA-A2-A04Q-01A-21D-A032-05 0.01283647
## TCGA-A2-A04T-01A-21D-A032-05 0.01278928
```

ttest

```
condition <- ifelse(BRCA_methylation_chr17$SUBTYPE=="LumA", "LumA", "other")
test.mety <- test_diff(BRCA_methylation_chr17_gen, condition)
```

As a result we obtain a data frame ...

```
head(test.mety)
```

```
##      met.log.fold met.mean met.t.stat   met.pval   met.padj
## ICAM2   -0.15151320 0.3330801 -8.916763 3.754116e-17 3.063359e-14
## RILP    -0.05073691 0.3341447 -7.638110 2.575168e-13 1.050668e-10
## PIPOX    0.11505558 0.3647812  7.165992 5.360053e-12 1.196885e-09
## TNFSF12 -0.13412855 0.2485025 -7.151613 5.867083e-12 1.196885e-09
## CD7      0.09822690 0.8127112  6.986460 1.641919e-11 2.679612e-09
## KSR1     0.19973400 0.5549808  6.950127 2.054467e-11 2.794075e-09
##      met.B      id
## ICAM2   27.37828  ICAM2
## RILP    18.64868  RILP
## PIPOX   15.65860  PIPOX
## TNFSF12 15.56967  TNFSF12
## CD7     14.55763   CD7
## KSR1    14.33731   KSR1
```

Combining data

We can combine results for both tests, for expression and methylation, to easily use functions designed to visualize those data.

```
test.both <- full_data(test.expr, test.mety)
head(test.both)
```

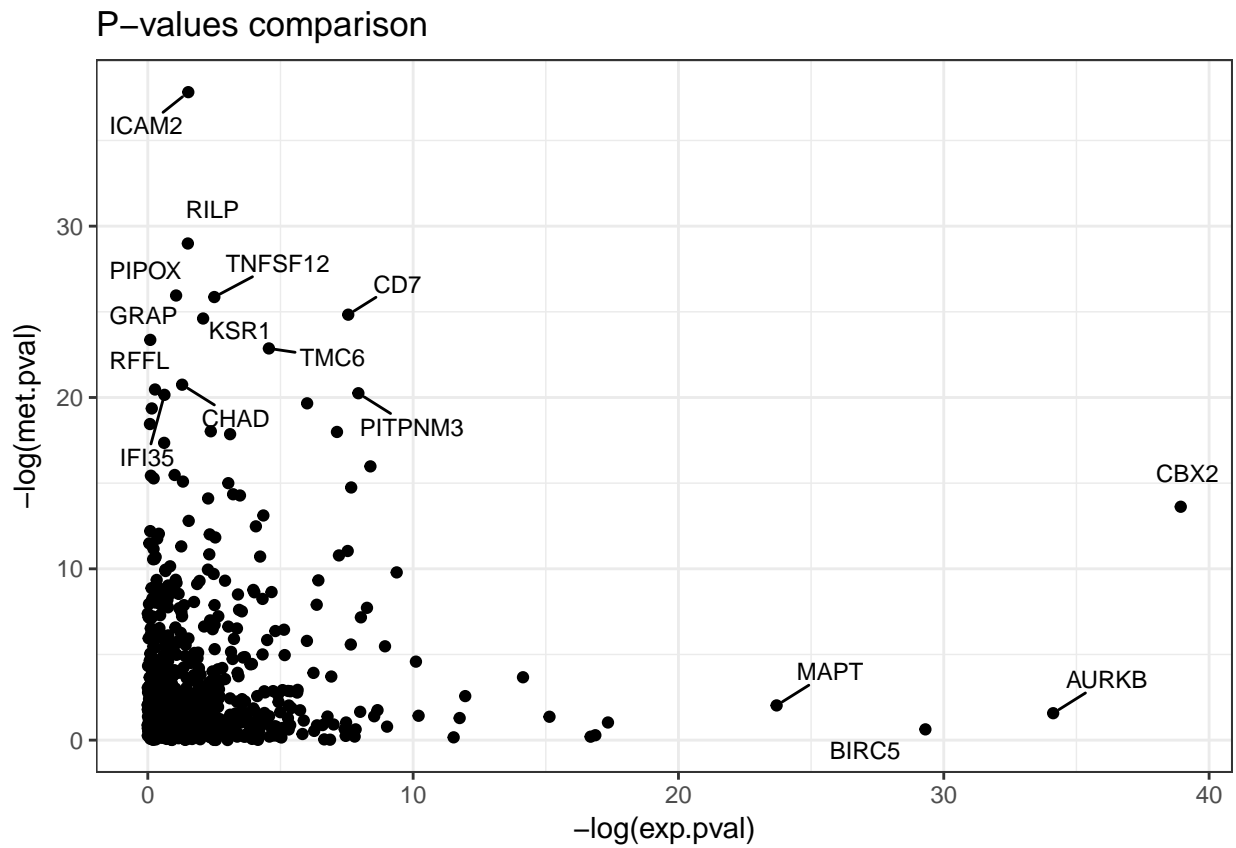
```
##      id    exp.mean exp.mean.gr1 exp.mean.g2 exp.fold exp.log.fold
## 1  AANAT    3.455436    3.901537    2.952385 0.7567235 -0.40216187
## 2  AARSD1 2779.448414 2712.105777 2855.387983 1.0528306  0.07427334
## 3   AATF 6750.269650 7042.041468 6421.250366 0.9118450 -0.13313947
## 4   AATK 352.805108  355.415829 349.861103 0.9843712 -0.02272566
## 5  ABCA5 1933.257431 1888.913529 1983.262257 1.0499487  0.07031881
## 6  ABCA6 689.547294  589.046234 802.878277 1.3630140  0.44680041
##      exp.pval exp.padj met.log.fold met.mean met.t.stat   met.pval
## 1 0.9453634 0.9920531  0.07237890 0.70420013  4.4829647 1.026904e-05
## 2 0.6627296 0.8992428  0.01114861 0.75430861  0.6669291 5.052981e-01
## 3 0.6864598 0.9111193  0.01360521 0.12700442  1.2459842 2.136847e-01
## 4 0.8051408 0.9585881  0.06619897 0.69837465  4.4078014 1.427292e-05
## 5 0.5837682 0.8489059  0.00510806 0.08834311  0.4746493 6.353608e-01
## 6 0.1289441 0.3857753 -0.03402781 0.24872043 -2.0227305 4.393248e-02
##      met.padj met.B
## 1 0.0001862119 1.559446
```

```
## 2 0.6620225949 -7.986421
## 3 0.3944948472 -7.431520
## 4 0.0002283667 1.244016
## 5 0.7590840893 -8.096320
## 6 0.1417004023 -6.171584
```

Plots

log-log p-value

```
p_values_plot(test.both)
```

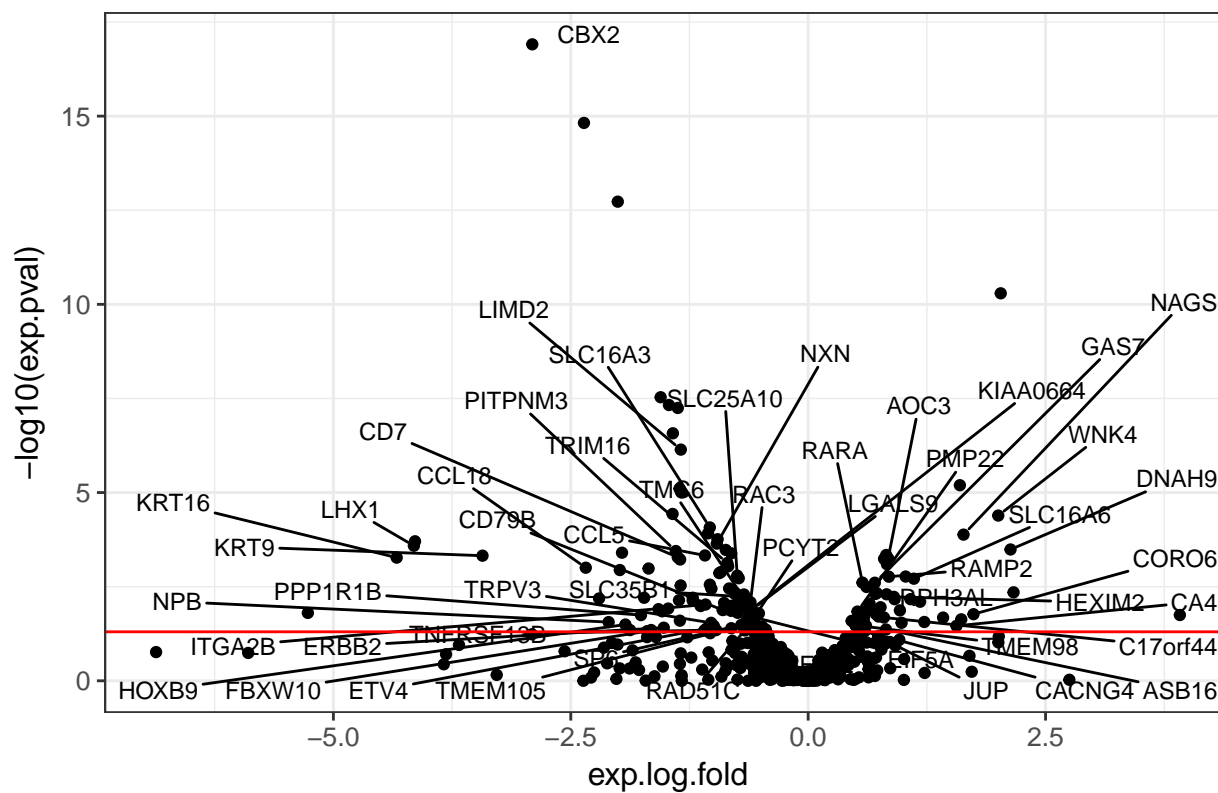


volcano

Volcan plot for expression

```
volcano_plot(test.both, type="expression")
```

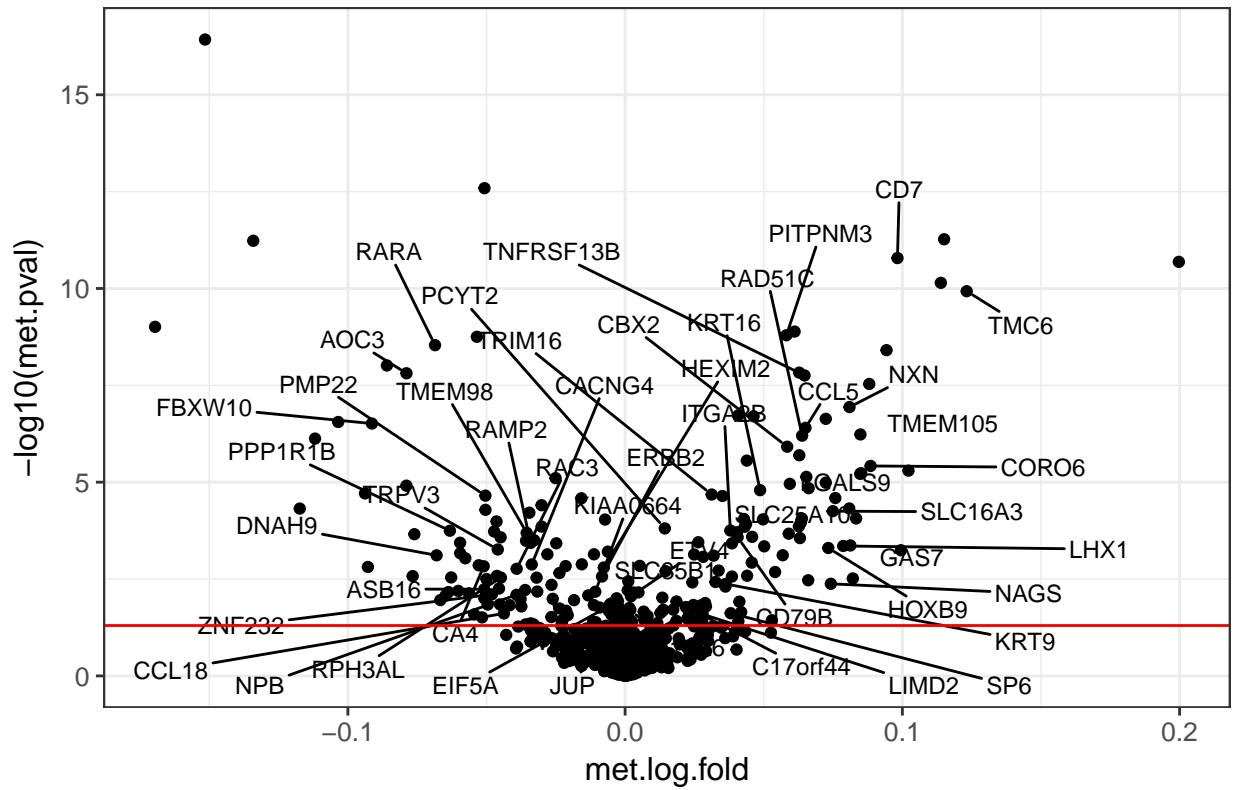
Volcano plot of expression



volcano plot for methylation

```
volcano_plot(test.both, type="methylation")
```

Volcano plot of methylation



metyl “road” + boxplots for expr