

Dirichlet-multinomial framework for differential splicing and SQTl analysis.

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1 Dirichlet-multinomial workflow to detect differential splicing (DS) in RNA-seq data

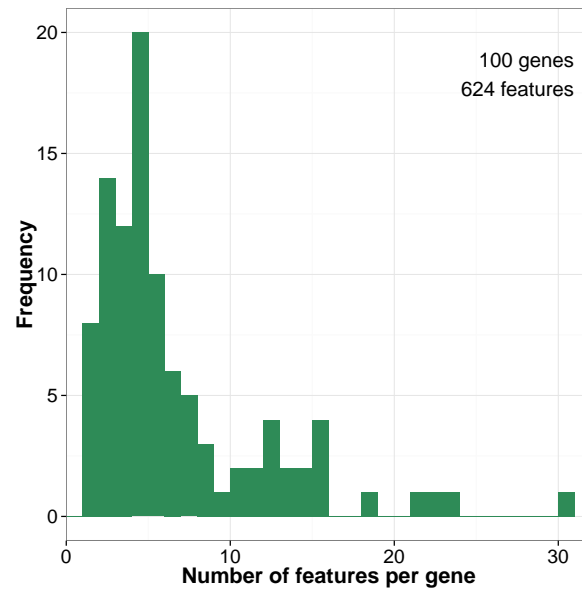
Load *DM* package.

```
suppressPackageStartupMessages(library(DM))
```

Create a `dmDSdata` object that contains counts.

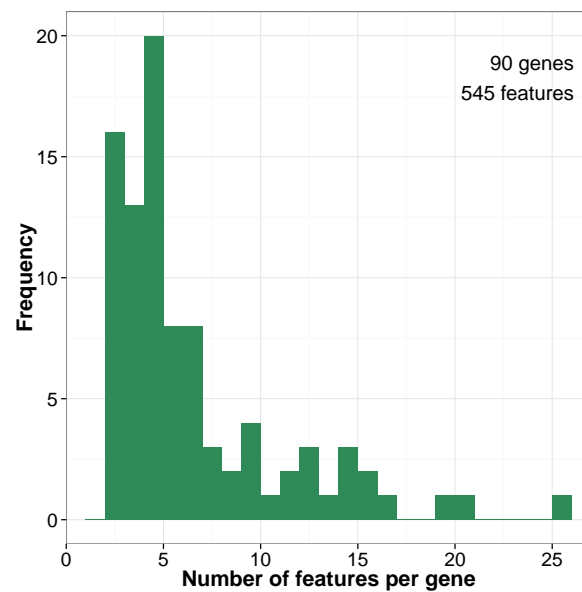
```
counts <- dataDS_counts[, -1]
group_id <- dataDS_counts[, 1]
group_split <- limma::strsplit2(group_id, ":")
gene_id <- group_split[, 1]
feature_id <- group_split[, 2]
sample_id = dataDS_metadata$sample_id
group = dataDS_metadata$group
d <- dmDSdata(counts = counts, gene_id = gene_id, feature_id = feature_id,
  sample_id = sample_id, group = group)
plotData(d)
```

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Filter genes and transcripts with low expression.

```
d <- dmFilter(d)
plotData(d)
```



Estimate dispersion.

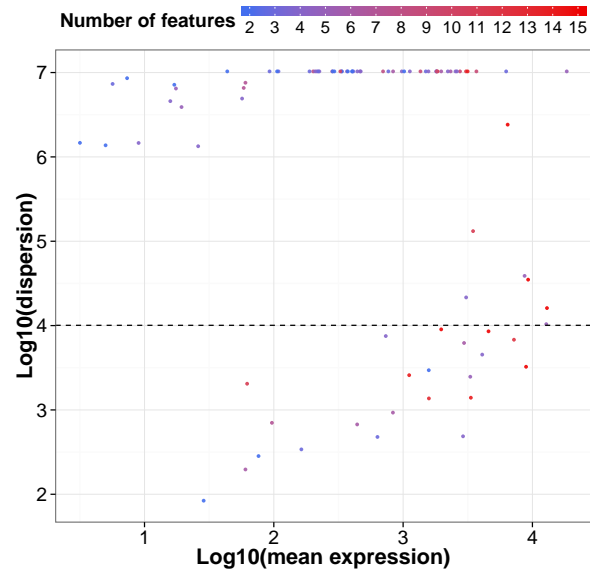
```
d <- dmDispersion(d, BPPARAM = BiocParallel::MulticoreParam(workers = 5))

## * Calculating mean gene expression..
## Took 0.459 seconds.
## * Estimating common dispersion..
## Took 16.767 seconds.

## ! Using common_dispersion = 10077.27 as disp_init !

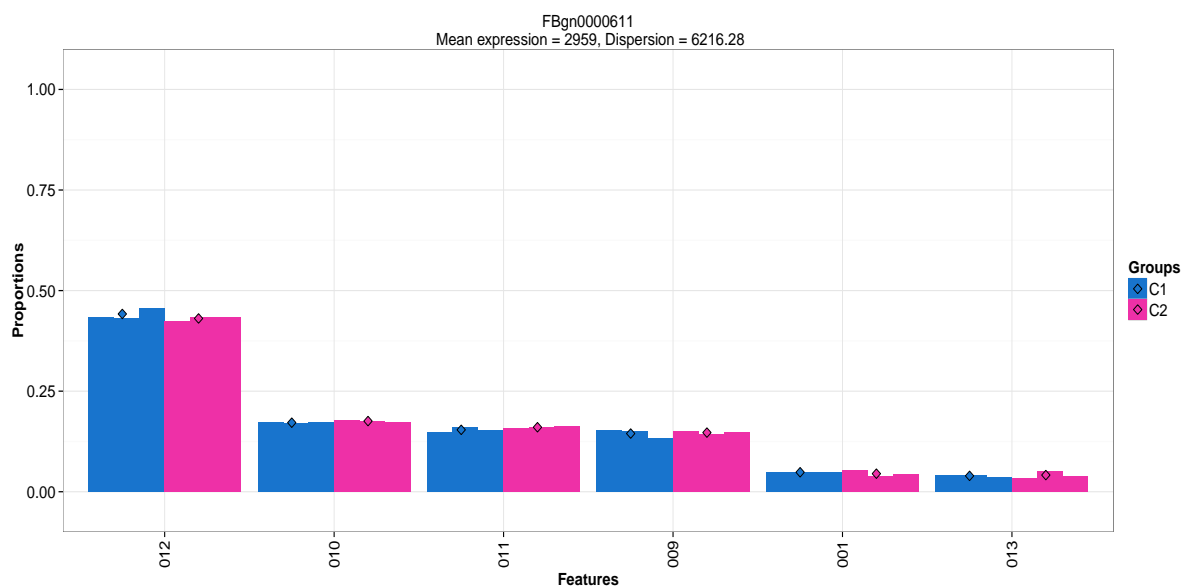
## * Estimating genewise dispersion..
## Took 4.43 seconds.
```

```
plotDispersion(d)
```



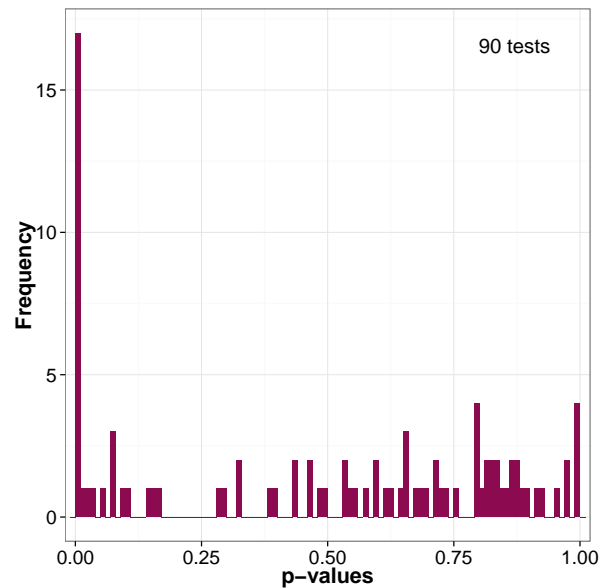
Estimate proportions.

```
d <- dmFit(d)
## * Fitting full model..
## Took 10.777 seconds.
gene_id <- names(d)[1]
plotFit(d, gene_id = gene_id, plot_type = "barplot")
## Plot gene 1: FBgn0000611
```



Use likelihood ratio to test for differential splicing.

```
d <- dmTest(d)
## Running comparison between groups: C1, C2
## * Fitting null model..
## Took 10.853 seconds.
## * Calculating likelihood ratio statistics..
## Took 0.001286983 seconds.
plotTest(d)
```



```
res <- results(d)
gene_id <- res$gene_id[1]
plotFit(d, gene_id = gene_id)
## Plot gene 1: FBgn0000611
```

