RmirR

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Outline

Introduction I

An analysis with RmiR start giving to the function read.mir the list of miRNAs and the gene's list with the right annotation With the default values the function searches integerScan and Pictar and prints only the couples of gene and miRNA present in bout databases.

Downloading the package I

source("http://bioconductor.org/biocLite.R")
biocLite("RmiR")
And we are going to download and annotation (could be a microarray data) biocLite("hgug4112a.db")
biocLite("org.Hs.eg.db")

Packages for this session I

```
> library(RmiR)
> library(hgug4112a.db)
> library(org.Hs.eg.db)
```

Starting to use the package

An analisys with RmiR start giving to the function read.mir the list of miRNAs and the gene's list with the right anotation
With the default values the function searches integerScan and Pictar and prints only the coupels of gene and miRNA present in bouth databases.

Datasets to work with I


```
> genes <- data.frame(genes = c("A_23_P171258",
+     "A_23_P150053", "A_23_P150053",
+     "A_23_P150053", "A_23_P202435",
+     "A_24_P90097", "A_23_P127948"))
> genes$expr <- c(1.21, -1.5, -1.34,
+     -1.45, -2.41, -2.32, -3.03)
> mirna <- data.frame(mirna = c("hsa-miR-148b",
+     "hsa-miR-27b", "hsa-miR-25",
+     "hsa-miR-181a", "hsa-miR-27a",
+     "hsa-miR-7", "hsa-miR-32",
+     "hsa-miR-32", "hsa-miR-7"))
> mirna$expr <- c(1.23, 3.52, -2.42,
+     5.2, 2.2, -1.42, -1.23, -1.2,
+     -1.37)
```

The dataset I


```
> genes

          genes  expr
1 A_23_P171258  1.21
2 A_23_P150053 -1.50
3 A_23_P150053 -1.34
```

The basic function read.mir I

With read.mir we search genes and miRNAs on bout of the databases earlier mentioned, the function gives the average of the expression of different probes of the microarray identifying the same gene and it also computes the coefficient of variation (CV).If there is just one probe identifying a gene, no average cantbe done, so CV will be NA.

```
> read.mir(genes = genes, mirna = mirna,
+     annotation = "hgug4112a.db")
```

```
  gene_id mature_miRNA mirExpr
1      22 hsa-miR-148b   1.230

       mirCV symbol geneExpr geneCV
1         NA  ABCB7     1.21     NA
```

And what about if you have another identification than the probe id? I

If the result is annotated by another identification than the platform probe id we can specify the annotation identifiers with other parameter id, the possible values for this id's "probes","genes", "alias", "ensemble", "unigene".

Some examples I

An example with entrez gene identifiers.

```
> genes.e <- genes
> genes.e$gene_id <- c(22, 59, 59,
+     59, 120, 120, 133)
> genes.e <- genes.e[, c("gene_id",
+     "expr")]
> genes.e
```

```
  gene_id  expr
1      22  1.21
2      59 -1.50
```

```
> read.mir(genes = genes.e, mirna = mirna,
+     annotation = "hgug4112a.db",
+     id = "genes")
```

Some examples II


```
  gene_id mature_miRNA mirExpr
1      22 hsa-miR-148b   1.230
4     133 hsa-miR-181a   5.200

      mirCV symbol geneExpr geneCV
1        NA  ABCB7     1.21     NA
4        NA    ADM    -3.03     NA
```

More examples I


```
> genes.a <- genes
> genes.a$alias <- c("ABCB7", "ADD3",
+     "ADDL", "ADD3", "AAT6", "ACTA2",
+     "ADM")
> genes.a <- genes.a[, c("alias",
+     "expr")]
> genes.a
```

```
  alias  expr
1 ABCB7  1.21
2  ADD3 -1.50
```

```
> read.mir(genes = genes.a, mirna = mirna,
+     annotation = "hgug4112a.db",
+     id = "alias")
```

More examples II


```
  gene_id mature_miRNA mirExpr
1      22 hsa-miR-148b   1.230
4     133 hsa-miR-181a   5.200

      mirCV symbol geneExpr geneCV
1        NA  ABCB7     1.21     NA
4        NA    ADM    -3.03     NA
```

Results by individual probes I

Some times we do not need the average of the results, for example when we would like to test this probe separately, in this case we prefer the results as they are, it is to say that with each probe annotated individually.

```
> read.mir(genes = genes, mirna = mirna,
+     annotation = "hgug4112a.db",
+     at.least = 1, id.out = "probes")

    gene_id mature_miRNA mirExpr
1        22 hsa-miR-148b   1.230
2        59  hsa-miR-27a   2.200

        mirCV     probe_id geneExpr
1          NA A_23_P171258     1.21
2          NA A_23_P150053    -1.50
```

A bit of biology I

The regulation of target genes by miRNAs occure usually at three diferent ways.

- The miRNAs could promote the expression of genes
- They could repress expression
- May be we cannot see any difference

Correlation or anti-correlation I

Looking at the correlation between different miRNAs targets couples we can obtain the correlated and the anti correlated couples.
This does not mean that there is short biological relevance but it could be some hints for farther investigation.

The function RmiRtc I

To use the function RmiRtc we need two or more objects of the class read.mir

```
> data(RmiR)
> res1 <- read.mir(gene = gene1,
+      mirna = mir1, annotation = "hgug4112a.db",
+      verbose = TRUE)

In targetscan database there are 13 genes and 35
          ------------------------------------


In pictar database there are 7 genes and 27 micro
          ---------------------------------------

> res2 <- read.mir(gene = gene2,
+      mirna = mir2, annotation = "hgug4112a.db",
+      verbose = TRUE)
```

The function RmiRtc II

```
In targetscan database there are 12 genes and 23 mi
           ------------------------------------

In pictar database there are 6 genes and 24 microRNA
           ------------------------------------

> res3 <- read.mir(gene = gene3,
+      mirna = mir3, annotation = "hgug4112a.db",
+      verbose = TRUE)

In targetscan database there are 13 genes and 35 micr
           ------------------------------------

In pictar database there are 7 genes and 27 microRNA
           ------------------------------------

> res_tc <- RmiRtc(timeline = c("res1",
+      "res2", "res3"), timevalue = c(12,
+      24, 48))
```

The function RmiRtc I

And now we use the function and look for the correlation. We can decide to filter the object by a correlation andor a gene expression threshold

```
> data(RmiR)
> res_fil <- readRmiRtc(res_tc, correlation = -0.9,
+     exprLev = 1, annotation = "hgug4112a.db")
> res_fil$reps
```

```
  symbol miRNAs gene_id
2    APP      3     351
3  VLDLR      3    7436
1  CENPV      1  201161
```

```
> res_fil
```

The function RmiRtc II

```
An object of class miRtcList
[[1]]
   gene_id mature_miRNA
22     351  hsa-miR-20a
17     351  hsa-miR-20b



[[2]]
      12    24    48
5   0.32  1.73  2.12
7   0.06  1.10  1.61


[[3]]
      12    24    48
5   0.71 -0.95 -1.67
```

The function RmiRtc III

```
7    0.71 -0.95 -1.67
```

The function readRmiRtc filter the genes ad returns a list ranked by the number of the miRNA satisfying the thresholds.

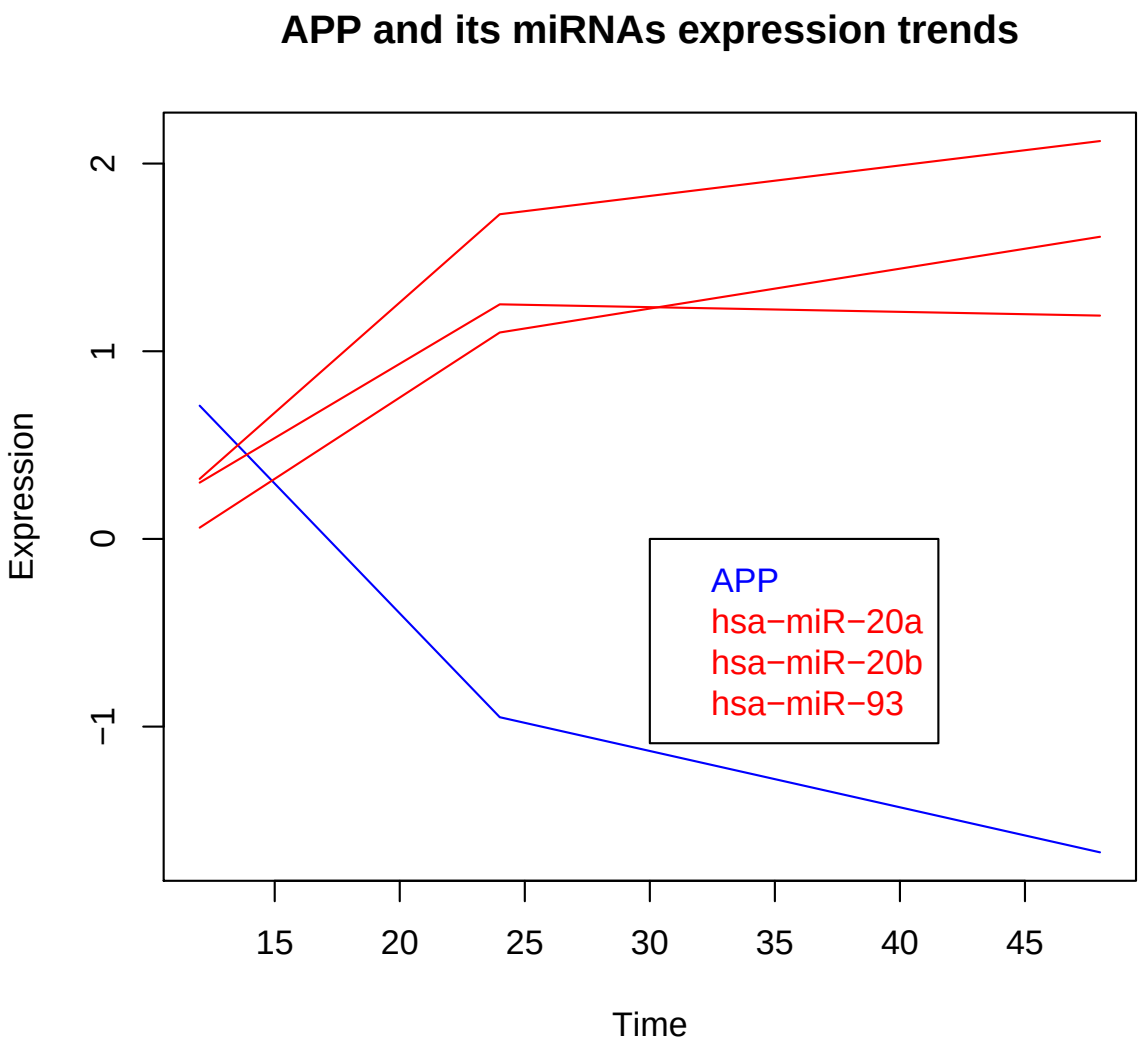
```
> cbind(res_fil$couples, res_fil$geneExpr,
+     res_fil$mirExpr)[res_fil$couples$gene_id ==
+     351 & res_fil$cor <= -0.9,
+     ]
```

```
   gene_id mature_miRNA   12    24
22     351  hsa-miR-20a 0.71 -0.95
17     351  hsa-miR-20b 0.71 -0.95
19     351   hsa-miR-93 0.71 -0.95
      48   12   24   48
22 -1.67 0.32 1.73 2.12
17 -1.67 0.06 1.10 1.61
19 -1.67 0.30 1.25 1.19
```

Plotting a time course experiment I

```
> plotRmiRtc(res_fil, gene_id = 351,
+     legend.y = 0, legend.x = 30)
```

Plotting a time course experiment II



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