Mouse Infinium methylation data processing with RnBeads

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Introduction

The goal is to display some key data frame structures and visuals that you can expect to get from running the code in RnBeads_with_RepeatMasker.R

Using RnBeads dev version (Müller et al. 2019).

Setup[1][2]

What should RepeatMasker table and $SAMPLE.csv file look like? `sample.table` was created solely for demonstration purposes; it's not needed to run the code.

```
head(tab.rmsk)
```

```
head(sample.table,14)
```

Interesting but confusing functions

`get.table()` returns a useful, unsorted data frame of statistics for each $ANNOTATION probe(rows) from `diffmeth.\$ANNOTATION` S4 object. `annotation()` returns row-matching annotation for each probe in tab.$ANNOTATION.

```
diffmeth.rmsk <- rnb.execute.computeDiffMeth(rnb.set, cmp.cols, region.types="rmsk")
tab.rmsk <- get.table(diffmeth.rmsk,comparison, region.type="rmsk",return.data.frame=T)
aa.rmsk <- annotation(rnb.set,type="rmsk")
```

```
head(tab.rmsk)
```

```
head(aa.rmsk)
```

These two data frames are bound together.

```
annotated.tab.rmsk <- data.frame(tab.rmsk,aa.rmsk, row.names=NULL)
head(annotated.tab.rmsk)
```

Order the rows by "combinedRank". The rownames of the `annotated.tab.\$ANNOTATION.order` data frame refers to the original row numbers of the annotated.tab.sites data frame, which is not ordered.

[1] Run rmarkdown::render("RnBeads_with_RepeatMasker.Rmd") in the same workspace as .R to avoid worrying about global environment.

[2] rnb.set object is not stored well in the global environment. If there are errors, re-run the script from top to bottom (remove output folders "result" and "resultFull"). There is a way to write theses objects to disk instead of RAM, but I didn't want to lose any disk space.

```
annotated.tab.rmsk.order <- annotated.tab.rmsk[order(annotated.tab.rmsk[,"combinedRank"]),]
head(annotated.tab.rmsk.order,3)
```

```
rownames(annotated.tab.rmsk.order)[1:10]
```

```
##  [1] "12842" "2839"  "22365" "24294" "6831"  "20269" "17342"
##  [8] "17343" "7604"  "8467"
```

`annotated.tab.\$ANNOTATION.order` data frame does not contain any information about beta values. These are extracted from rnb.set using `meth()`, annotation name, and row index from `annotated.tab.\$ANNOTATION.order`.

```
topDiffBeta.rmsk.full <- meth(rnb.set.full, type="rmsk")[as.integer(rownames(annotated.tab.rmsk.order)),]
```

Heatmap(s)

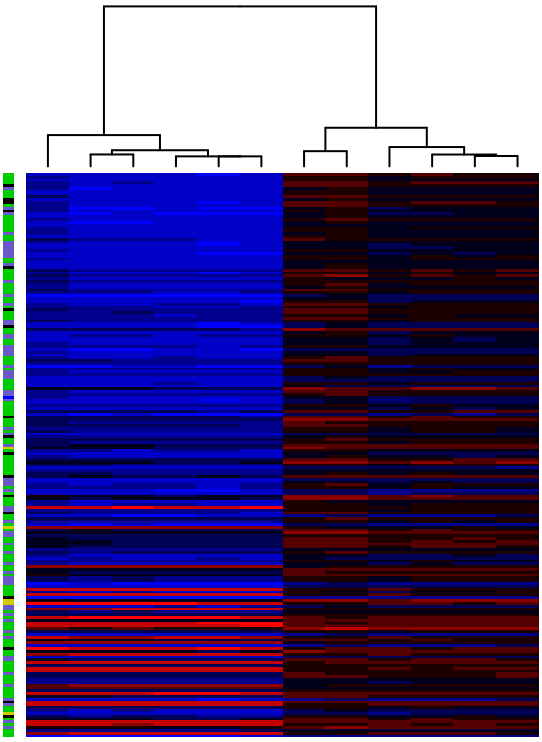
Differentially methylated probes annotated with RepeatMasker

```
colSide <- annotated.tab.rmsk.order$name

colSide <-gsub("LTR","slateblue3",colSide)
colSide <- gsub("LINE","green3",colSide)
colSide <- gsub("SINE","green3",colSide)
colSide <- gsub("DNA","black",colSide)
colSide <- gsub("Simple_repeat","yellow3",colSide)
colSide <- gsub("Other","blue",colSide)
colSide <- gsub("Unknown","blue",colSide)

par(font=2, font.axis=2, font.lab=2,cex.lab=1.5, cex.axis=1.5, lty=1)
heatmap.2(topDiffBeta.rmsk.full[1:200,c(1,5,9,2,6,10,3,7,11,4,8,12)],
          col=colfunc(10), scale="none", Rowv=F, Colv=T,
          cexCol=1, labCol=NA, labRow=NA,srtCol=0, adjCol=c(0.5,0),
          density.info = "none",trace="none", dendrogram = "column",
          symkey=FALSE,symbreaks=F,revC = FALSE, RowSideColors = colSide[1:200],
          breaks=c(0.0,0.1,0.2,0.3,0.4,0.5,0.6,0.7,0.8,0.9,1),
          lmat=rbind(c(5,0,4),c(3,1,2)),
          lhei=c(1,4),
          lwid=c(1.75,0.1, 3.5),margins=c(5,12),cexRow=1.2)
```

```
## Error in plot.new(): figure margins too large
```



Müller, Fabian, Michael Scherer, Yassen Assenov, Pavlo Lutsik, Jörn Walter, Thomas Lengauer, and Christoph Bock. 2019. "RnBeads 2.0: Comprehensive Analysis of DNA Methylation Data." *Genome Biology* 20 (1): 55. https://doi.org/10.1186/s13059-019-1664-9.