## **Distribution of m6A sites (MetaTX)**

The <u>MetaTX</u> is designed to visualize the transcriptomic distribution of RNA-related genomic features [12]. We are going to use this tool to display the distribution of reads along the transcriptome.

## Install MetaTX

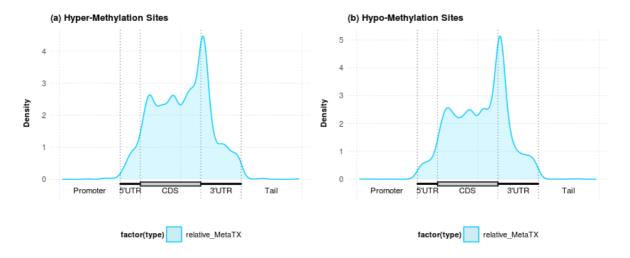
```
$ git clone https://github.com/yue-wang-biomath/MetaTX.1.0
$ unzip master.zip
$ cd MetaTX.1.0-master
$ R CMD INSTALL MetaTX_1.0.tar.gz
```

## **Visualization of the Distribution of Peaks**

The following code gives separate figures of the distribution of hyper-methylation sites and hypomethylation sites.

```
# Load libraries
library(MetaTX)
library(rtracklayer)
library(readr)
library(bedr)
library(genomation)
library(GenomicRanges)
# Import BED file from exomePeak2
file <- "exomePeak2_output_peakcalling_1strand/Mod.bed"</pre>
gr_obj = import(file)
# Separate by hyper and hypo methylation sites
data1 \leftarrow data[strand(gr_obj) == "+",]
data2 <- data[strand(gr_obj) == "-",]</pre>
df1 <- data.frame(segnames=segnames(data1),</pre>
                   starts=start(data1),
                   ends=end(data1),
                   names=elementMetadata(data1)$name,
                   scores=elementMetadata(data1)$score,
                   strands=strand(data1))
df2 <- data.frame(segnames=segnames(data2),</pre>
                   starts=start(data2),
                   ends=end(data2),
                   names=elementMetadata(data2)$name,
                   scores=elementMetadata(data2)$score,
                   strands=strand(data2))
write.table(df1, file="Mod_metaTX_pos.bed", quote=F, sep="\t", row.names=F,
col.names=F)
write.table(df2, file="Mod_metaTX_neg.bed", quote=F, sep="\t", row.names=F,
col.names=F)
```

```
# Import separated bed files
file_pos <- "Mod_metaTX_pos.bed"</pre>
file_neg <- "Mod_metaTX_neg.bed"</pre>
gr_obj_pos <- import(file_pos)</pre>
gr_obj_neg <- import(file_neg)</pre>
gr_obj_pos <- resize(gr_obj_pos, width = 1, fix = "center")</pre>
gr_obj_neg <- resize(gr_obj_neg, width = 1, fix = "center")</pre>
# Download information about mRNA components
txdb <- TxDb.Hsapiens.UCSC.hg19.knownGene
cds_by_tx0_1 <- cdsBy(txdb, "tx")</pre>
fiveUTR_tx0_1 <- fiveUTRsByTranscript(txdb,use.names=FALSE)</pre>
threeUTR_tx0_1 <- threeUTRsByTranscript(txdb,use.names=FALSE)</pre>
# Map peaks to the RNA model
remap_results_m6A_1 <- remapCoord(features = gr_obj_pos, txdb = txdb, num_bin =
10, includeNeighborDNA = TRUE, cds_by_tx0 = cds_by_tx0_1, fiveUTR_tx0 =
fiveUTR_tx0_1,
                                   threeUTR_tx0 = threeUTR_tx0_1
# Plot 1
p1 <- metaTXplot(remap_results_m6A_1,</pre>
                  num_bin
                                        = 10,
                   includeNeighborDNA = TRUE,
                   relativeProportion = c(1, 3, 2, 3),
                  title = '(a) Hyper-Methylation Sites',
                  legend = 'relative',
                  type = 'relative'
)
# Map peaks to the RNA model
remap_results_m6A_2 <- remapCoord(features = gr_obj_neg, txdb = txdb, num_bin =
10, includeNeighborDNA = TRUE, cds_by_tx0 = cds_by_tx0_1, fiveUTR_tx0 =
fiveUTR_tx0_1,
                                   threeUTR_tx0 = threeUTR_tx0_1
# Plot 1
p2 <- metaTXplot(remap_results_m6A_2,</pre>
                   num_bin
                                        = 10,
                   includeNeighborDNA = TRUE,
                   relativeProportion = c(1, 3, 2, 3),
                   title = '(b) Hypo-Methylation Sites',
                  legend = 'relative',
                   type = 'relative'
)
# Plot all
ggdraw() +
  draw_plot(p1, 0, 0, .5, 1) +
  draw_plot(p2, .5, 0, .5, 1)
```



## **Report Isoform Probabilities**

MetaTX also provides a function for computing the probabilities of a particular feature being located on different isoforms.

```
isoform_probs <- isoformProb(remap_results_m6A_1, num_bin = 10,
includeNeighborDNA = TRUE, lambda = 2)
write.csv(isoform_probs, "isoform_probs.csv")</pre>
```

Here are the first few rows of the outputs.

index_trans (double)	index_methyl (double)	seqnames (character)	methyl_pos (double)	strand (character)	trans_ID (double)	isoform_prob (double)
1	1	chr18	25532104	-	65378	0.4363063
2	1	chr18	25532104	-	65379	0.5636937
3	2	chr3	101396098	+	14200	1.0000000
4	3	chr3	101396173	+	14200	1.0000000
5	4	chr2	162087846	+	9525	0.5121048
6	4	chr2	162087846	+	9526	0.4878952
7	5	chr7	75052219	-	30728	1.0000000
8	6	chr7	75047839	-	30728	1.0000000
9	7	chr18	32858834	-	65418	0.0000000
10	7	chr18	32858834	-	65419	0.0000000