

Bound genes and gene regions of MREs

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1 Libraries and settings

```
# -----  
# libraries  
# -----  
library(tidyverse)  
library(GenomicRanges)  
library(colorspace)  
library(eulerr)  
library(gghalves)  
  
# -----  
# settings  
# -----  
  
here <- here::here()  
  
source(paste0(here, "/Supporting_scripts/themes/theme_paper.R"))  
source(paste0(here, "/Supporting_scripts/themes/CustomThemes.R"))  
  
out <- paste0(here, "/Figure4/01_MRE_bound_gene_and_bound_region/")  
  
# farben  
farbe4 <- "#7AA6DCFF"
```

```
farbe6 <- "#003C67FF"
```

2 What was done?

- the genotype and gene region of the mir 181 binding sites (union) are plotted (Figure4)

3 Files

```
# -----
# MREs
# -----

mir181_bs <- readRDS(paste0(here, "/Figure2/01_mir181-enriched_binding_site_definition/mir181_bs.rds"))

names(mir181_bs) <- 1:NROW(mir181_bs)
mir181_bs <- as.data.frame(mir181_bs)

mir181_enriched_set <- mir181_bs %>%
  as.data.frame(.) %>%
  subset(set %in% c("ago_bs_mir181_chi&mir181_enriched", "mir181_enriched"))
```

4 Characterise MRE

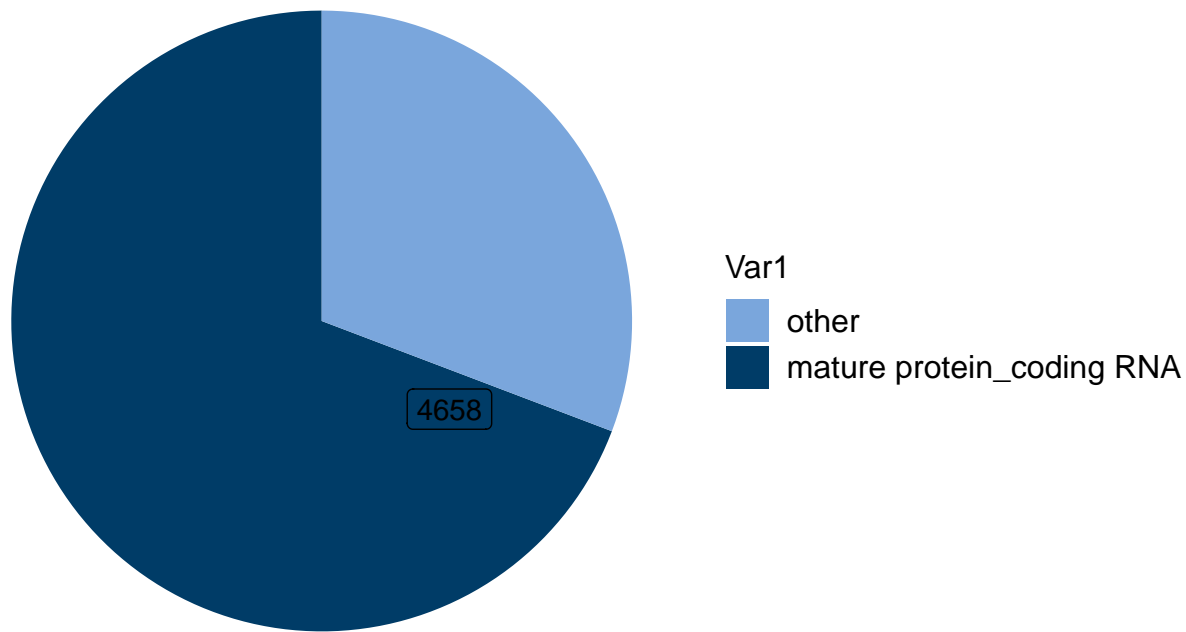
4.1 mir181 bound genes - Figure 4A

```
mir181_enriched_set <- mutate(mir181_enriched_set, geneType =
  case_when(geneType != "protein_coding" ~ "other",
            region == "intron" ~ "other",
            region == "outside" ~ "other",
            is.na(region) ~ "other",
            T ~ "mature protein_coding RNA"))

gene_type_df <- table(mir181_enriched_set$geneType) %>%
  as.data.frame(.)

p <- ggplot(gene_type_df, aes(y=Freq, x="", fill=Var1)) +
  geom_col() +
  coord_polar(theta="y") +
  # xlim(c(2, 4)) +
  geom_label(data = gene_type_df %>% subset(gene_type_df == "mature protein_coding RNA"), aes(y=Freq, x=
    position = position_stack(vjust = 0.5),
    show.legend = FALSE) +
  scale_fill_manual(values = c (farbe6, farbe4)) +
  theme_paper() +
  theme_nice_pie() +
  #theme(legend.position = "none") +
  guides(fill = guide_legend(reverse = TRUE)) +
  labs(y = NULL,
       x = NULL)

p
```



```
ggsave(p, filename = paste0(out, "Figure4A_bound_gene_types_mir181_BS.pdf"), width = unit(8, "cm"), height = unit(6, "cm"))
```

4.1.1 Remove non protein-coding binding sites

For all further analyses we removed binding sites on non protein-coding RNAs.

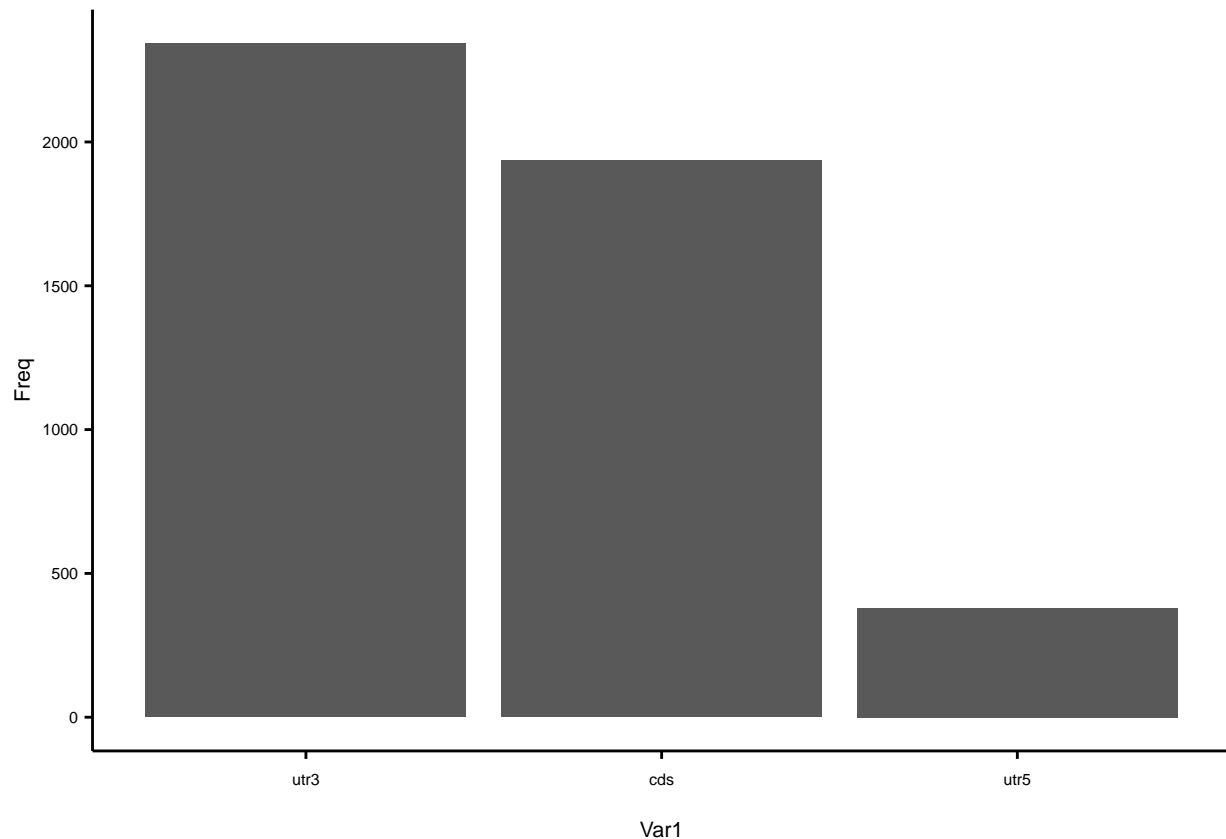
```
mir181_enriched_set <- subset(mir181_enriched_set, geneType == "mature protein_coding RNA")
```

4.2 mir181 bound regions - Figure 4B

```
gene_region_df <- table(mir181_enriched_set$region) %>%
  as.data.frame(.) %>%
  arrange(desc(Freq))

gene_region_df$Var1 <- factor(gene_region_df$Var1, levels = gene_region_df$Var1)

p <- ggplot(gene_region_df %>% subset(Var1 != "outside"), aes(y=Freq, x=Var1)) +
  geom_col()+
  theme_paper()
p
```



```
ggsave(p, filename = paste0(out, "Figure2B_bound_gene_regions_miR181_BS.pdf"), width = unit(6, "cm"), h
sum(gene_region_df$Freq)

## [1] 4658
```

4.2.1 Remove binding sites in introns or outside of any gene region

For all further analyses we removed binding sites in introns or in a gene regions with no region annotation.

```
mir181_bs <- mutate(mir181_bs, geneType =
  case_when(geneType != "protein_coding" ~ "other",
            region == "intron" ~ "other",
            region == "outside" ~ "other",
            is.na(region) ~ "other",
            T ~ "mature protein_coding RNA")) %>%
subset(., geneType == "mature protein_coding RNA")
```

5 Save filtered BS

```
saveRDS(mir181_bs, paste0(out, "mir181_bs_afterFigure4B.rds"))
```

6 Session Info

```
sessionInfo()
```

```

## R version 4.2.2 (2022-10-31)
## Platform: x86_64-apple-darwin17.0 (64-bit)
## Running under: macOS Big Sur ... 10.16
##
## Matrix products: default
## BLAS: /Library/Frameworks/R.framework/Versions/4.2/Resources/lib/libRblas.0.dylib
## LAPACK: /Library/Frameworks/R.framework/Versions/4.2/Resources/lib/libRlapack.dylib
##
## locale:
## [1] en_US.UTF-8/en_US.UTF-8/en_US.UTF-8/C/en_US.UTF-8/en_US.UTF-8
##
## attached base packages:
## [1] stats4      stats      graphics  grDevices  utils      datasets  methods
## [8] base
##
## other attached packages:
## [1] gghalves_0.1.4      eulerr_7.0.0        colorspace_2.1-0
## [4] GenomicRanges_1.50.2 GenomeInfoDb_1.34.9 IRanges_2.32.0
## [7] S4Vectors_0.36.2    BiocGenerics_0.44.0 lubridate_1.9.2
## [10] forcats_1.0.0       stringr_1.5.0       dplyr_1.1.2
## [13] purrr_1.0.1         readr_2.1.4         tidyr_1.3.0
## [16] tibble_3.2.1        ggplot2_3.4.2       tidyverse_2.0.0
## [19] knitr_1.43
##
## loaded via a namespace (and not attached):
## [1] Rcpp_1.0.11         here_1.0.1          rprojroot_2.0.3
## [4] digest_0.6.33       utf8_1.2.3          R6_2.5.1
## [7] backports_1.4.1     evaluate_0.21       highr_0.10
## [10] pillar_1.9.0        zlibbioc_1.44.0     rlang_1.1.1
## [13] rstudioapi_0.15.0   car_3.1-2           rmarkdown_2.23
## [16] textshaping_0.3.6   labeling_0.4.2      RCurl_1.98-1.12
## [19] munsell_0.5.0       broom_1.0.5         compiler_4.2.2
## [22] xfun_0.39           pkgconfig_2.0.3     systemfonts_1.0.4
## [25] htmltools_0.5.5     tidyselect_1.2.0    GenomeInfoDbData_1.2.9
## [28] fansi_1.0.4         tzdb_0.4.0          withr_2.5.0
## [31] ggpubr_0.6.0        bitops_1.0-7        grid_4.2.2
## [34] gtable_0.3.3        lifecycle_1.0.3     magrittr_2.0.3
## [37] scales_1.2.1        cli_3.6.1           stringi_1.7.12
## [40] carData_3.0-5       farver_2.1.1        XVector_0.38.0
## [43] ggsignif_0.6.4      ragg_1.2.5          generics_0.1.3
## [46] vctrs_0.6.3         tools_4.2.2         glue_1.6.2
## [49] hms_1.1.3           abind_1.4-5         fastmap_1.1.1
## [52] yaml_2.3.7          timechange_0.2.0    rstatix_0.7.2

```