Sushi: An R/Bioconductor package for visualizing genomic data

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Contents

1 Introduction

Sushi is an R package for plotting genomic data stored in multiple common genomic formats including bed, bedpe, bedgraph format. The package was designed to be very flexible to allow for combinations of plots into multipanel figures that can include plots made by Sushi, R basecode, or other R packages. Sushi allows for simple flexible plotting of gene structures, transcript structures, sequencing tracks, ChIP-seq peaks, chromatin interactions, GWAS results and other commen genomic data types. This vignette shows some examples of the functions included in Sushi to get you started with plotting these diverse data types.

2 Data

2.1 Data types

Sushi accepts 4 types of genomic data as input. These include:

- bed format: 3-6 columns (chromosome, start, stop, name, score, strand)
- bedpe format: 6-10 columns (chromosome1, start1, stop1, chromosome2, start2, stop2,name, score, strand1, strand2)
- bedgraph format: 4 columns (chromosome, start, stop, score)
- interaction matrix: This is matrix in which row and column names are genomic coordiates and matrix values are some tye of interaction score.

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** strands can be represented as 1 or -1 or "+" and "-".

** Some functions may require additional information depending on the plot and features desired.

2.2 Example datasets

To illustrate how Sushi works, we have included several publicaly available data sets in the package Sushi. The data types include RNA-seq, ChIP-seq, ChIA-PET, and HiC data:

Sushi_5C.bedpe	?]
Sushi_ChIAPET_pol2.bedpe	?]
Sushi_ChIPExo_CTCF.bedgraph	?]
Sushi_ChIPSeq_CTCF.bedgraph	?]
Sushi_ChIPSeq_pol2.bed	?]
Sushi_ChIPSeq_pol2.bedgraph	?]
Sushi_ChIPSeq_severalfactors.bed	?]
Sushi_DNaseI.bedgraph	?]
Sushi_GWAS.bed	?]
Sushi_HiC.matrix	?]
Sushi_RNASeq_K562.bedgraph	?]
Sushi_genes.bed	?]
Sushi_hg18_genome	?]
Sushi_transcripts.bed	?]

These data sets can be loaded using the following commands:

```
> library('Sushi')
> Sushi_data = data(package = 'Sushi')
> data(list = Sushi_data$results[,3])
```

To see which data sets are loaded

```
> Sushi_data$results[,3]
```

```
 [1] "Sushi_5C.bedpe"                    "Sushi_ChIAPET_pol2.bedpe"
 [3] "Sushi_ChIPExo_CTCF.bedgraph"       "Sushi_ChIPSeq_CTCF.bedgraph"
 [5] "Sushi_ChIPSeq_pol2.bed"            "Sushi_ChIPSeq_pol2.bedgraph"
 [7] "Sushi_ChIPSeq_severalfactors.bed" "Sushi_DNaseI.bedgraph"
 [9] "Sushi_GWAS.bed"                    "Sushi_HiC.matrix"
[11] "Sushi_RNASeq_K562.bedgraph"        "Sushi_genes.bed"
[13] "Sushi_hg18_genome"                 "Sushi_transcripts.bed"
```

3 Functions

3.1 Functions overview

Sushi functions can be broken down into 3 categories: plotting, annotating, zooming, and coloring. Plotting functions generate a basic plot object using the data. Annotating functions add information to the plots such as an x-axis labeling the genomic region or a legend describing the values represented by different colors. Zooming functions allow for highlighting and zooming of genomic regions, which are of particular use for multipanel plots generated with base R functions `mfrow()` or `layout()`. The coloring functions provide simple tools for generating R colors and palettes.

- Plotting functions: `plotBed()`, `plotBedgraph()`, `plotBedpe()`, `plotGenes()`, `plotHiC()`, and `plotManhattan()`
- Annotating functions: `labelgenome()` and `addlegend()`
- Zooming functions: `zoomsregion()` and `zoombox()`
- Coloring functions: `maptocolors()`, `SushiColors()`, and `opaque()`

3.2 Non-Sushi Functions

An important characteristic of Sushi plots is their compatibility with all base R functions and their ability to be combined into complex multipanel figures. Two of the most usefule base R functions for creating multipanel figures are `layout()` and `mfrow()`. Basic R plotting functions such as `axis()`, `mtext()`, and `legend()` are also particularly well suited to combine with Sushi plots. A familiarity with these functions will greatly improve your ability to create Sushi plots.

3.3 plotBedgraph

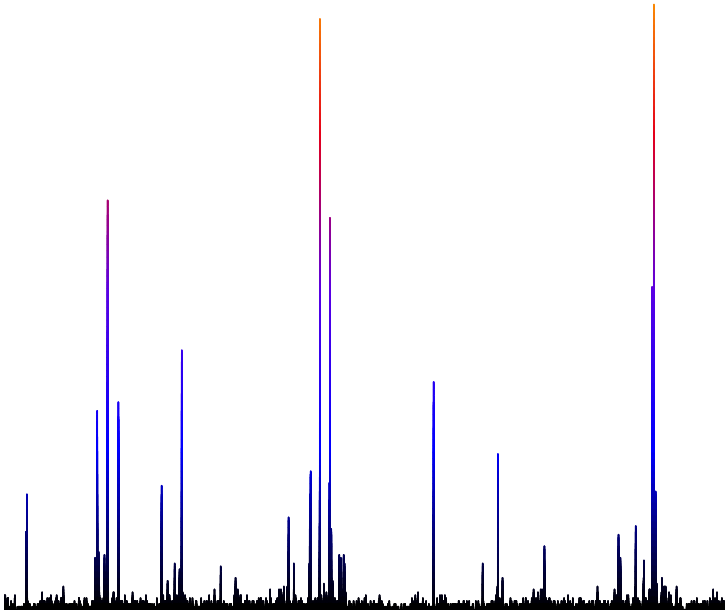
Signal tracks can be plotted using `plotBedgraph()`. The input requires data in bedgraph format. We will demonstrate this using bedgraph data representing a DNaseI hypersensitivity experiment in K562 cells.

```
>   head(Sushi_DNaseI.bedgraph)
```

```
  chrom   start     end value
1 chr11 1640504 1640664     1
2 chr11 1640904 1641004     1
3 chr11 1641004 1641064     2
4 chr11 1641064 1641164     1
5 chr11 1645224 1645384     1
6 chr11 1645504 1645664     1
```

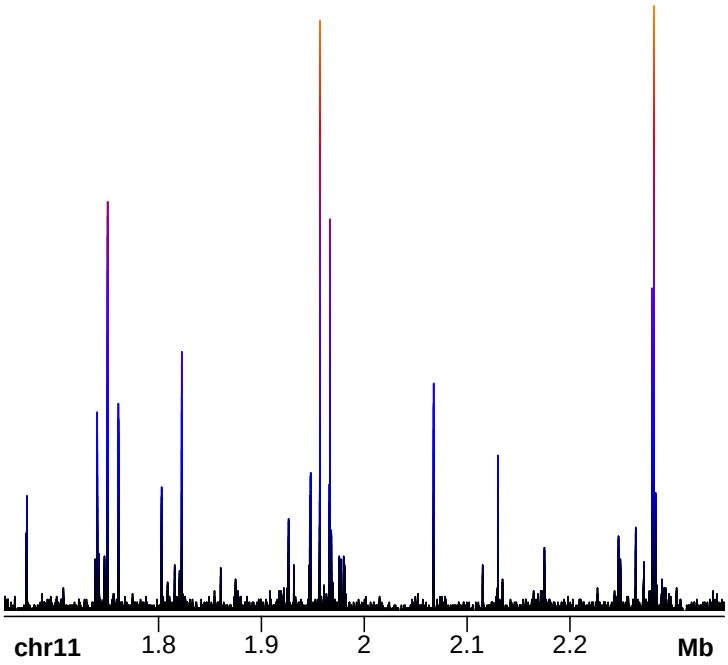
The `plotBedgraph()` function is used to plot the data. As with most Sushi functions the basic required arguments include the data to be plotted, the chromosome, and a start and stop position.

```
> chrom            = "chr11"
> chromstart       = 1650000
> chromend         = 2350000
> plotBedgraph(Sushi_DNaseI.bedgraph,chrom,chromstart,chromend,colorbycol= SushiColors(5))
```



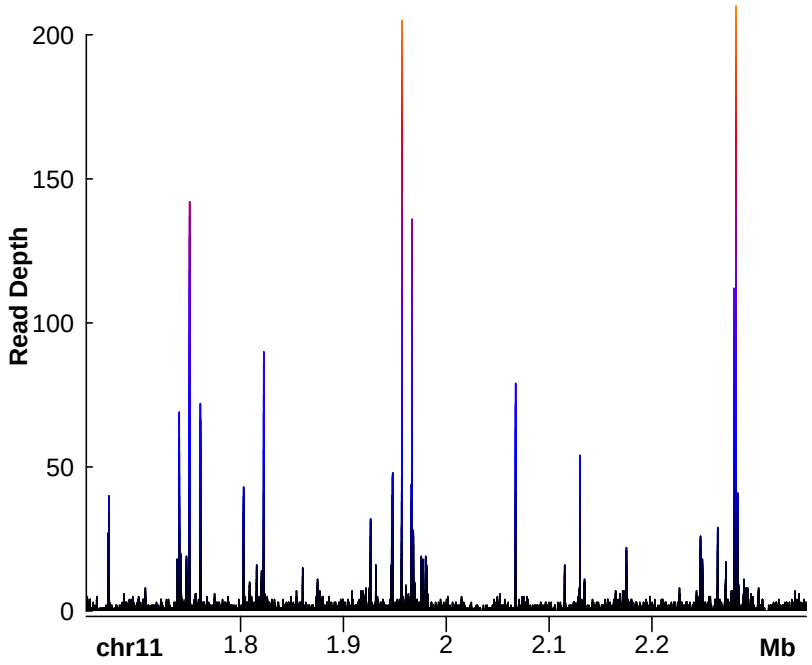
To annotate the genome postion we use the `labelgenome()` function. We us `n = 4` to specify the desired number of tickmarks. The scale is set to `Mb` (other options are `Kb` or `bp`).

```
> labelgenome(chrom,chromstart,chromend,n=4,scale="Mb")
```



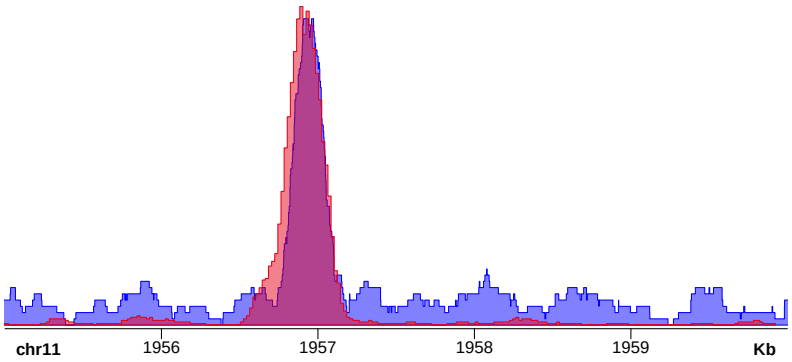
The y-axis can be added using basic R functions `mtext()` and `axis()`.

```
> mtext("Read Depth",side=2,line=1.75,cex=1,font=2)
> axis(side=2,las=2,tcl=.2)
```



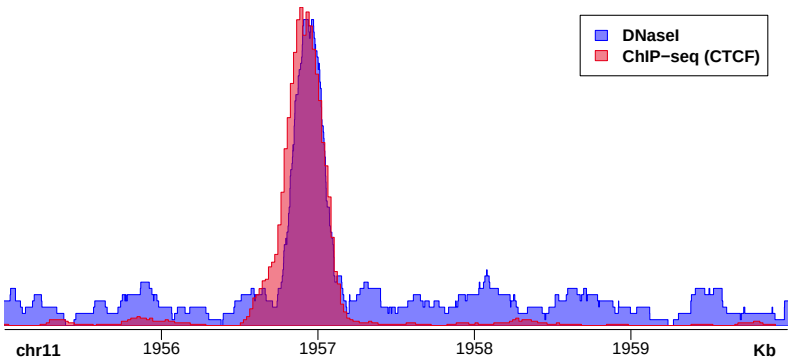
Multiple bedgraph tracks can be plotted on the same plot by setting `overlay=TRUE`. Transparencies can be added for easier viewing by adjusting the transcparency value. The second plot can be rescaled to the maximum of the first plot by setting `rescaleoverlay=TRUE`.

```
> chrom            = "chr11"
> chromstart       = 1955000
> chromend         = 1960000
> plotBedgraph(Sushi_ChIPSeq_CTCF.bedgraph,chrom,chromstart,chromend,
              transparency=.50,color=SushiColors(2)(2)[1])
> plotBedgraph(Sushi_DNaseI.bedgraph,chrom,chromstart,chromend,
              transparency=.50,color=SushiColors(2)(2)[2],overlay=TRUE,
              rescaleoverlay=TRUE)
> labelgenome(chrom,chromstart,chromend,n=3,scale="Kb")
```



Then we can use the base R function `legend()` to add a legend to the plot. First we need to use the rgb function to add transparency to the colors in order to match out plot.

```
> legend("topright",inset=0.025,legend=c("DNaseI","ChIP-seq (CTCF)"),
        fill=opaque(SushiColors(2)(2)),border=SushiColors(2)(2),text.font=2,
        cex=1.0)
```

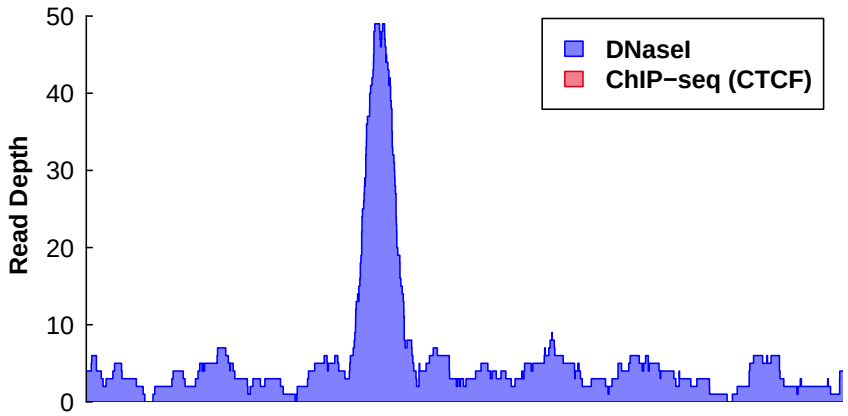


Setting `flip=TRUE` is another method that can be used to compare tracks. First, we will use mfrow to divided the plotting divice into two vertically stacked regions.

```
> par(mfrow=c(2,1),mar=c(1,4,1,1))
```

Next, we plot the first plot. We set the transparency of the plot to 0.5. We will also add the legend.

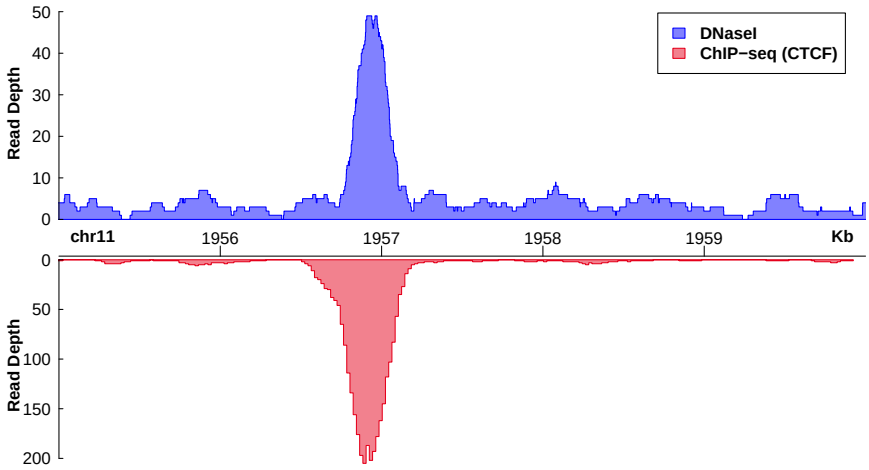
```
> plotBedgraph(Sushi_ChIPSeq_CTCF.bedgraph,chrom,chromstart,chromend,transparency=.50,
              color=SushiColors(2)(2)[1])
> axis(side=2,las=2,tcl=.2)
> mtext("Read Depth",side=2,line=1.75,cex=1,font=2)
> legend("topright",inset=0.025,legend=c("DNaseI","ChIP-seq (CTCF)"),
         fill=opaque(SushiColors(2)(2)),border=SushiColors(2)(2),text.font=2,
         cex=1.0)
```



Finally, we add the second plot with `flip=TRUE`. We will also label the x-axis using `labelgenome()` and label the y-axis using `mtext()` and `axis()`.

```
> plotBedgraph(Sushi_DNaseI.bedgraph, chrom, chromstart, chromend,
              transparency=.50, flip=TRUE, color=SushiColors(2)(2)[2])
> labelgenome(chrom,chromstart,chromend,side=3,n=3,scale="Kb")
```

```
> axis(side=2,las=2,tcl=.2,at=pretty(par("yaxp")[c(1,2)]),
              labels=-1*pretty(par("yaxp")[c(1,2)]))
> mtext("Read Depth",side=2,line=1.75,cex=1,font=2)
```



3.4 plotHic

HiC interaction plots can be plotted given an interaction matrix in which row and column names are genomic coordiates and matrix values are some tye of interaction score.

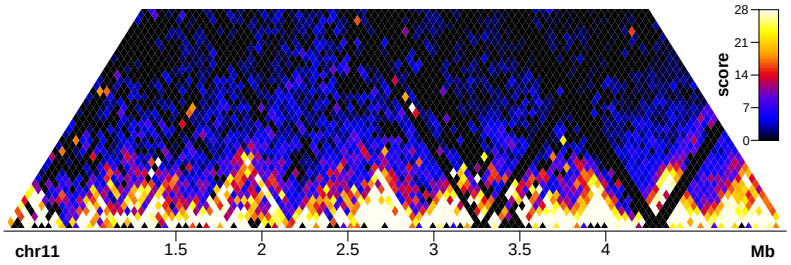
```
>   Sushi_HiC.matrix[100:105,100:105]
```

```
          4460000  4500000  4540000  4580000   4620000  4660000
4460000 60.758775 18.84723 33.31506 22.56641  7.926361 10.69235
4500000 18.847231 32.56282 36.31212 29.04343 13.375643 12.67360
4540000 33.315060 36.31212 17.97024 43.43753 20.411952 16.98875
4580000 22.566409 29.04343 43.43753 38.93754 25.206417 23.87764
4620000  7.926361 13.37564 20.41195 25.20642  9.201501 38.33665
4660000 10.692351 12.67360 16.98875 23.87764 38.336646 22.55054
```

The `plotHic()` function is used to plot the data while the `labelgenome()` function is used to add the genome labels to the x-axis. `plotHic()` returns an object indicating the color palette and data range that can be fed into `addlegend()` to create a legend.

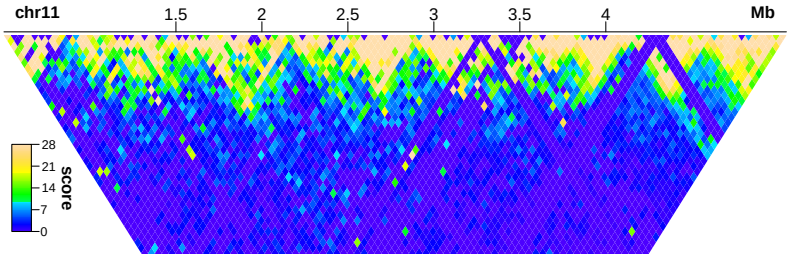
```
> chrom            = "chr11"
> chromstart       = 500000
> chromend         = 5050000
> phic = plotHic(Sushi_HiC.matrix, chrom,chromstart, chromend, max_y = 20,
                 zrange=c(0,28), palette=SushiColors(7))
```

```
> addlegend(phic[[1]], palette=phic[[2]], title="score", side="right",
           bottominset=0.4, topinset=0, xoffset=-.035, labelside="left",
           width=0.025, title.offset=0.035)
> labelgenome(chrom, chromstart, chromend, n=4, scale="Mb",
              edgeblankfraction=0.20)
```



`plotHic()` has a number of customizable options. The plot can be flipped over the x-axis by setting `flip = TRUE`. The color palette can be changed by the palette argument.
`addlegend()` also has customizable features. The legend can be moved to the left side of the plot by setting `side = "left"` and the labeling can be moved to the right side of the lenged buy setting `labelside = "right"`. The vertical position of the legend can be adjusted by changing the `topinset` and `bottominset`.
Finally, the x-axis label can be moved to the top of the plot by setting `side = 3` in the `labelgenome()` function.

```
> chrom            = "chr11"
> chromstart       = 500000
> chromend         = 5050000
> phic = plotHic(Sushi_HiC.matrix,chrom,chromstart,chromend,max_y = 20,
                 zrange=c(0,28),flip=TRUE,palette=topo.colors)
> addlegend(phic[[1]],palette=phic[[2]],title="score",side="left",bottominset=0.1,
           topinset=0.5,xoffset=-.035,labelside="right",width=0.025,title.offset=0.035)
> labelgenome(chrom,chromstart,chromend,side=3,n=4,scale="Mb",edgeblankfraction=0.20)
```



3.5 plotBedpe

`plotBedpe()` allows for data in bedpe format to be plotted in multiple fashions. To illustrate this we will use 5C data formatted in the following way.

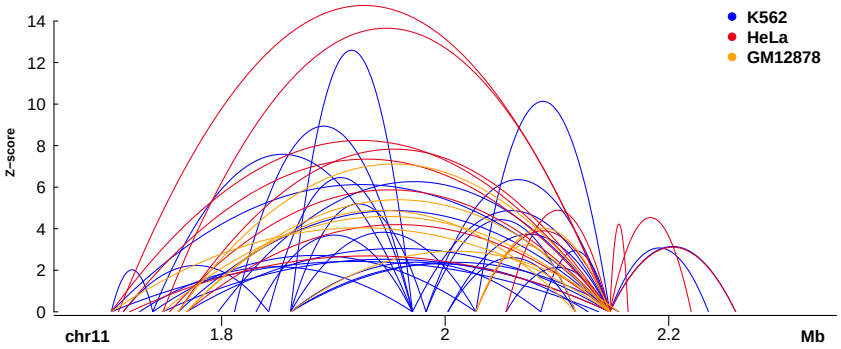
```
>   head(Sushi_5C.bedpe)
```

```
  chrom1    start1      end1 chrom2    start2      end2 name    score strand1
1   chr2 234208447 234223064   chr2 234156762 234159135   NA 44.39862       .
2  chr15  41711734  41718116  chr15  41802421  41808201   NA 20.62534       .
3  chr11  64172456  64183193  chr11  64068878  64079209   NA 16.91630       .
4   chr2 234208447 234223064   chr2 234163674 234170252   NA 12.34501       .
5   chr6  41755186  41769245   chr6  41435903  41452283   NA 11.63480       .
6  chr11  64159283  64172456  chr11  64068878  64079209   NA 11.13098       .
  strand2 samplenumber
1       .            1
2       .            1
3       .            1
4       .            1
5       .            1
6       .            1
```

`plotBedpe()` can plot bedpe as arches. The height, linewidth, and color of each arch can be scaled to represent different aspects of the data. Here the height of the arches represents the Z-score of the 5C interaction, the color represents the cell line each interaction was detected in, and the line widths are kept constant (default lwd = 1).

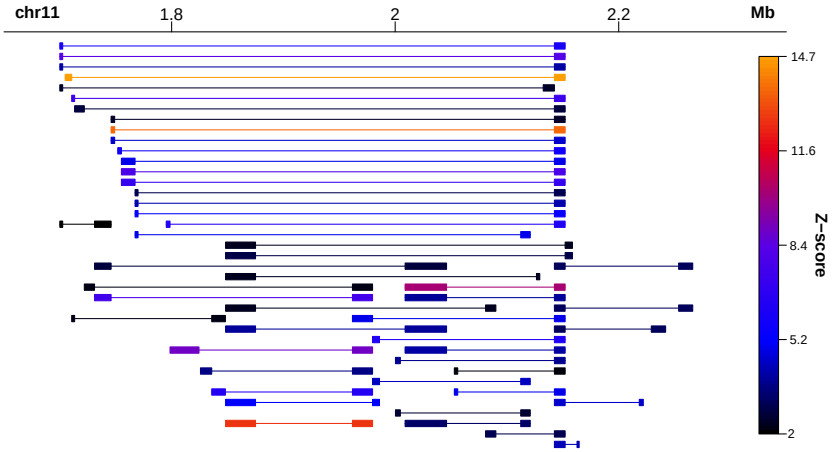
```
> chrom            = "chr11"
> chromstart       = 1650000
> chromend         = 2350000
> pbpe = plotBedpe(Sushi_5C.bedpe,chrom,chromstart,chromend,
                   heights = Sushi_5C.bedpe$score,plottype="loops",
```

```
                    colorby=Sushi_5C.bedpe$samplenumber,
                    colorbycol=SushiColors(3))
> labelgenome(chrom, chromstart,chromend,n=3,scale="Mb")
> legend("topright",inset =0.01,legend=c("K562","HeLa","GM12878"),
         col=SushiColors(3)(3),pch=19,bty='n',text.font=2)
> axis(side=2,las=2,tcl=.2)
> mtext("Z-score",side=2,line=1.75,cex=.75,font=2)
```



The plot can be flipped over the x-axis by setting `flip = TRUE`, Bedpe elements can be represented by boxes and straight lines by setting `plottype = "lines"`. And colors can be used to represent Z-scores by setting `colorby = "Sushi_5C.bedpe$score"`.

```
> chrom            = "chr11"
> chromstart       = 1650000
> chromend         = 2350000
> pbpe = plotBedpe(Sushi_5C.bedpe,chrom,chromstart,chromend,flip=TRUE,
                   plottype="lines",colorby=Sushi_5C.bedpe$score,
                   colorbycol=SushiColors(5))
> labelgenome(chrom, chromstart,chromend,side=3,n=3,scale="Mb")
> addlegend(pbpe[[1]],palette=pbpe[[2]],title="Z-score",side="right",bottominset=0.05,
           topinset=0.05,xoffset=-.035,labelside="right",width=0.025,title.offset=0.045)
```



3.6 plotBed

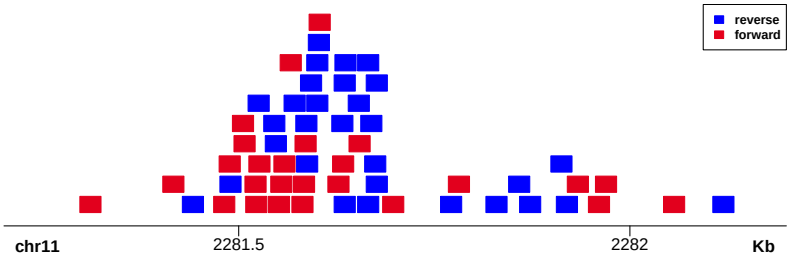
`plotBed` provides multiple different ways to represent genomic data stored in bed format. Below are the first six lines of a bed file detailing reads from Pol2 ChIP-Seq analysis of K562 cells.

```
>   head(Sushi_ChIPSeq_pol2.bed)
```

```
  chrom   start     end                          name score strand
1 chr11 2280543 2280570 GGGCTCTCTCCGGCTTCCCTGTCCCGT    63      -1
2 chr11 2288946 2288973 CCTTCCCATCCGCAGGGGCACCACATG  1000      -1
3 chr11 2272471 2272498 TGGGCATCAGTCAGGCTCCTTCCCCAG  1000      -1
4 chr11 2288939 2288966 ATCCGCAGGGGCACCACATGAGTCACC  1000      -1
5 chr11 2281534 2281561 TGTCCTAGTGACAAGTGGCCGGACTTG   250      -1
6 chr11 2286805 2286832 GGTGAGGGCCAGCAGCTCCCTGGGGGG   250       1
```

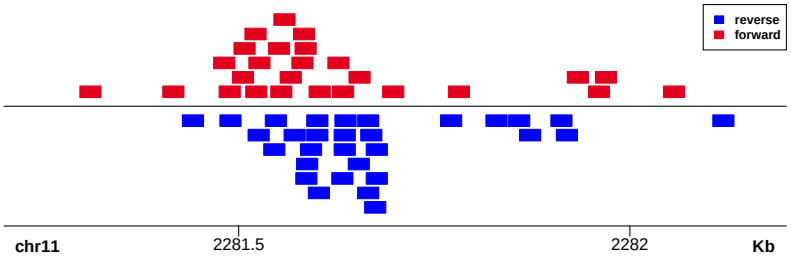
Leaving row set to `auto` provides a pile-sup style plot. Here the `colorby` argument is used to color the bed elements by the strand.

```
> chrom            = "chr11"
> chromstart       = 2281200
> chromend         = 2282200
> plotBed(beddata    = Sushi_ChIPSeq_pol2.bed,chrom = chrom,chromstart = chromstart,
         chromend =chromend,colorby    = Sushi_ChIPSeq_pol2.bed$strand,
         colorbycol = SushiColors(2),row  = "auto",wiggle=0.001)
> labelgenome(chrom,chromstart,chromend,n=2,scale="Kb")
> legend("topright",inset=0,legend=c("reverse","forward"),fill=SushiColors(2)(2),
        border=SushiColors(2)(2),text.font=2,cex=0.75)
```



Setting `splitstrand = TRUE` plots reads from different strands in two separate vertical regions.

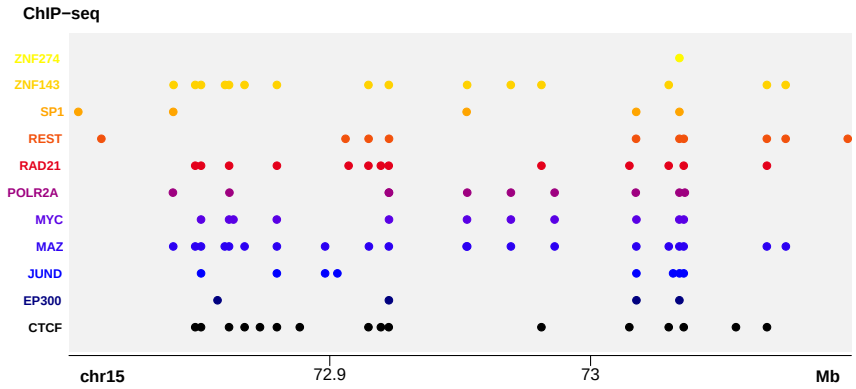
```
> chrom            = "chr11"
> chromstart       = 2281200
> chromend         = 2282200
> plotBed(beddata    = Sushi_ChIPSeq_pol2.bed,chrom = chrom,chromstart = chromstart,
         chromend =chromend,colorby    = Sushi_ChIPSeq_pol2.bed$strand,
         colorbycol = SushiColors(2),row  = "auto",wiggle=0.001,splitstrand=TRUE)
> labelgenome(chrom,chromstart,chromend,n=2,scale="Kb")
> legend("topright",inset=0,legend=c("reverse","forward"),fill=SushiColors(2)(2),
        border=SushiColors(2)(2),text.font=2,cex=0.75)
```



plotBed can also plot bed elements on different rows as specified by the user. First, we will use the Sushi function `maptocolors()` to assign a different color to each row.

```
> Sushi_ChIPSeq_severalfactors.bed$color =
         maptocolors(Sushi_ChIPSeq_severalfactors.bed$row,
         col=SushiColors(6))
```

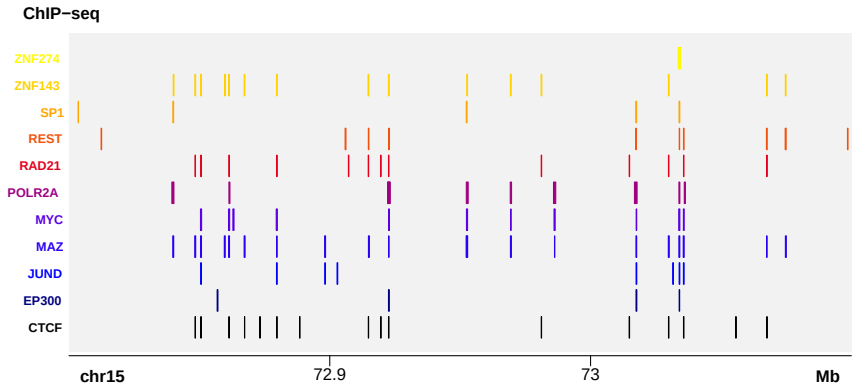
By providing row and color information `plotBed()` can be used to compare bed elements from different samples by plotting them on different rows.




```
> chrom            = "chr15"
> chromstart      = 72800000
> chromend         = 73100000
> plotBed(beddata    = Sushi_ChIPSeq_severalfactors.bed,chrom = chrom,
         chromstart = chromstart,chromend =chromend,
         rownumber  = Sushi_ChIPSeq_severalfactors.bed$row, type = "circles",
         color=Sushi_ChIPSeq_severalfactors.bed$color,row="given",
         plotbg="grey95",rowlabels=unique(Sushi_ChIPSeq_severalfactors.bed$name),
         rowlabelcol=unique(Sushi_ChIPSeq_severalfactors.bed$color),rowlabelcex=0.75)
> labelgenome(chrom,chromstart,chromend,n=3,scale="Mb")
> mtext("ChIP-seq",side=3, adj=-0.065,line=0.5,font=2)
```

That same data can be represented by rectangles that depict the actual width of each bed element.

```
> plotBed(beddata    = Sushi_ChIPSeq_severalfactors.bed,chrom = chrom,
         chromstart = chromstart,chromend =chromend,
         rownumber  = Sushi_ChIPSeq_severalfactors.bed$row, type = "region",
         color=Sushi_ChIPSeq_severalfactors.bed$color,row="given",
         plotbg="grey95",rowlabels=unique(Sushi_ChIPSeq_severalfactors.bed$name),
         rowlabelcol=unique(Sushi_ChIPSeq_severalfactors.bed$color),rowlabelcex=0.75)
> labelgenome(chrom,chromstart,chromend,n=3,scale="Mb")
> mtext("ChIP-seq",side=3, adj=-0.065,line=0.5,font=2)
```



`plotBed()` can also be used to plot heatmaps representing the density of bed elements. First, we will use the biomaRt function `getBM()` to get the gene information we require.

```
> chrom            = "chr15"
> chromstart       = 60000000
> chromend         = 80000000
> chrom_biomart    = gsub("chr","",chrom)
> mart=useMart(host='may2009.archive.ensembl.org', biomart='ENSEMBL_MART_ENSEMBL',
              dataset='hsapiens_gene_ensembl')
> geneinfobed = getBM(attributes = c("chromosome_name","start_position","end_position"),
                      filters= c("chromosome_name","start","end"),
                      values=list(chrom_biomart,chromstart,chromend),mart=mart)
> geneinfobed[,1] = paste("chr",geneinfobed[,1],sep="")
```

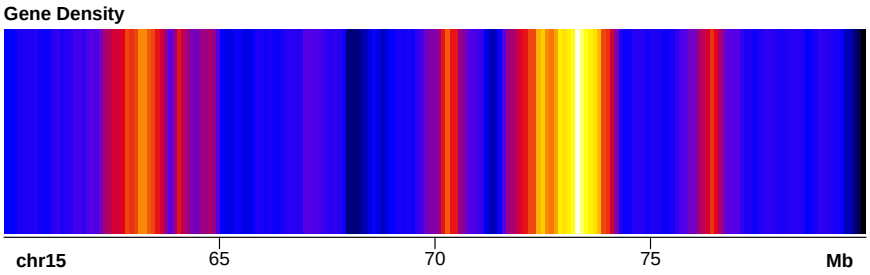
The data is in simple bed format with just three columns representing chromosome, start, and stop.

```
> head (geneinfobed)
```

```
  chromosome_name start_position end_position
1           chr15       73372069     73372334
2           chr15       64580642     64580710
3           chr15       63375442     63375557
4           chr15       72570353     72570422
5           chr15       60903209     60903293
6           chr15       70130646     70130724
```

Now we can make a gene density plot using the plotBed function.

```
> plotBed(beddata = geneinfobed[!duplicated(geneinfobed),],chrom = chrom,
        chromstart = chromstart,chromend =chromend,row='supplied',
        palettes = list(SushiColors(7)), type = "density")
> labelgenome(chrom, chromstart, chromend,  n=4,scale="Mb",edgeblankfraction=0.10)
> mtext("Gene Density",side=3, adj=0,line=0.20,font=2)
```



3.7 plotManhattan

`plotManhattan()` differs from most other Sushi functions in that it can plot multiple chromosomes in a single plot. Because of this plotManhattan requires some additional inputs. It requires an object in bed format describing the location of data points as well as vector of p-values (typically one of the columns of the bed file). But it also requires an genome object that describes which chromosomes to plot and their sizes (in bp). The genome object is very similar to the genome files used for bedtools.

The bed data should look something like this:

```
>   head(Sushi_GWAS.bed)

  chr.hg18 pos.hg18 pos.hg18.1       rsid pval.GC.DBP V6
1     chr1  1695996    1695996  rs6603811    0.003110  .
2     chr1  1696020    1696020  rs7531583    0.000824  .
3     chr1  1698661    1698661 rs12044597    0.001280  .
4     chr1  1711339    1711339  rs2272908    0.001510  .
5     chr1  1712792    1712792  rs3737628    0.001490  .
6     chr1  1736016    1736016 rs12408690    0.004000  .
```

And the genome file should look like this:

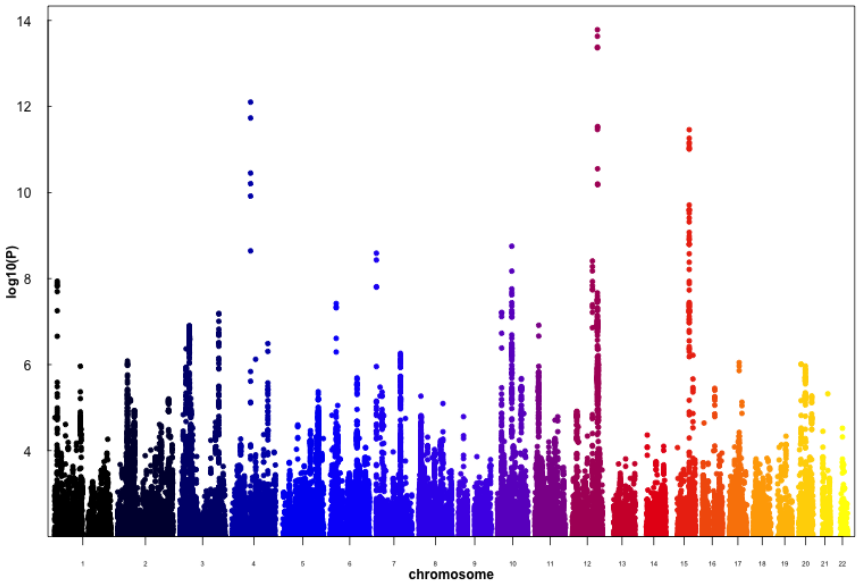
```
>   head(Sushi_hg18_genome)

     V1        V2
1  chr1 247249719
2 chr10 135374737
```

```
3 chr11 134452384
4 chr12 132349534
5 chr13 114142980
6 chr14 106368585
```

The `plotManhattan()` function is used to plot the data while the `labelgenome()` function is used to add the genome labels to the x-axis. The `labelgenome()` function also requires a genome object.

```
> plotManhattan(bedfile=Sushi_GWAS.bed,pvalues=Sushi_GWAS.bed[,5],
                col=SushiColors(6),genome=Sushi_hg18_genome,cex=0.75)
> labelgenome(genome=Sushi_hg18_genome,n=4,scale="Mb",
                edgeblankfraction=0.20,cex.axis=.5)
> axis(side=2,las=2,tcl=.2)
> mtext("log10(P)",side=2,line=1.75,cex=1,font=2)
> mtext("chromosome",side=1,line=1.75,cex=1,font=2)
```

```
pdf
  2
```



3.8 plotGenes

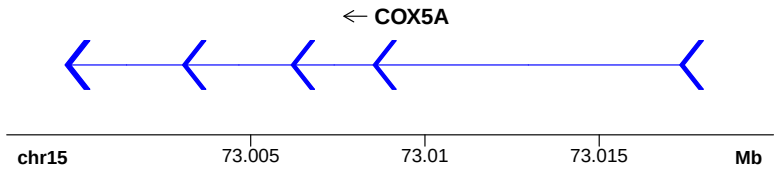
`plotGenes()` can be used to plot gene structures that are stored in bed format. If no `geneinfo` object is provided genes are looked up in the region using biomart with biomart='ensembl' and dataset='hsapiens_gene_ensembl'.

```
> head(Sushi_genes.bed)
```

```
  chrom    start     stop  gene score strand
1 chr15 73017309 73017438 COX5A     .     -1
2 chr15 72999672 72999836 COX5A     .     -1
3 chr15 73003042 73003164 COX5A     .     -1
4 chr15 73006160 73006281 COX5A     .     -1
5 chr15 73008510 73008626 COX5A     .     -1
```

Using `plotGenes()` with arguments `bentline=FALSE` and `plotgenetype="arrow"` produces arrow and line gene structures.

```
> chrom            = "chr15"
> chromstart       = 72998000
> chromend         = 73020000
> pg = plotGenes(Sushi_genes.bed,chrom,chromstart,chromend ,
                 types=Sushi_genes.bed$type,maxrows=1,bheight=0.2,
                 plotgenetype="arrow",bentline=FALSE,
           labeloffset=.4,fontsize=1.2,arrowlength = 0.025,
                 labeltext=TRUE)
> labelgenome( chrom, chromstart,chromend,n=3,scale="Mb")
```



This function can also be used to plot transcript structures. The first 20 lines of a data frame describing RNA seq data are shown below.

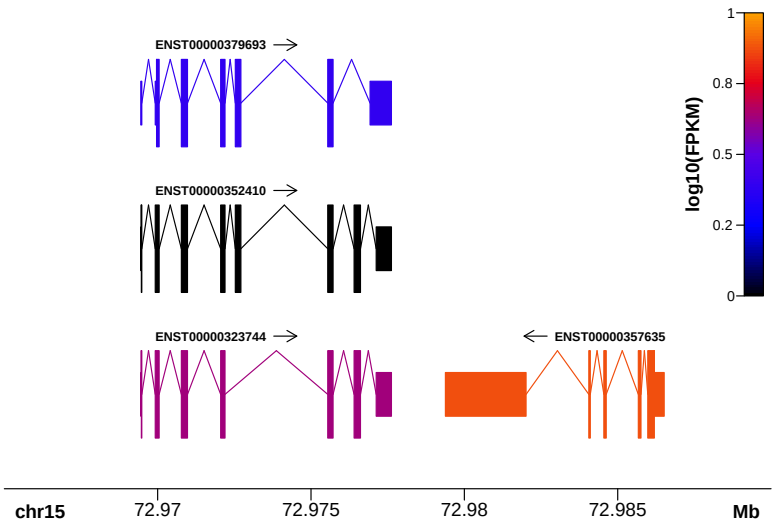
```
> Sushi_transcripts.bed[1:20,]
```

```
   chrom    start     stop            gene     score strand type
1  chr15 73062668 73062770 ENST00000362710  0.000000     -1 exon
2  chr15 73097788 73097929 ENST00000361900  0.000000      1 exon
3  chr15 73097264 73097365 ENST00000361900  0.000000      1 exon
4  chr15 73095987 73096143 ENST00000361900  0.000000      1 exon
5  chr15 73092071 73092199 ENST00000361900  0.000000      1 exon
6  chr15 73091234 73091240 ENST00000361900  0.000000      1 exon
7  chr15 73017309 73017408 ENST00000322347 31.488695     -1 exon
8  chr15 73006160 73006281 ENST00000322347 31.488695     -1 exon
```

```
9  chr15 73008510 73008626 ENST00000322347 31.488695     -1 exon
10 chr15 72984058 72984106 ENST00000357635  7.473977     -1 exon
11 chr15 72984548 72984625 ENST00000357635  7.473977     -1 exon
12 chr15 72985672 72985759 ENST00000357635  7.473977     -1 exon
13 chr15 72985981 72986194 ENST00000357635  7.473977     -1 exon
14 chr15 72975546 72975719 ENST00000379693  2.422616      1 exon
15 chr15 72972532 72972714 ENST00000379693  2.422616      1 exon
16 chr15 72972055 72972196 ENST00000379693  2.422616      1 exon
17 chr15 72970773 72970973 ENST00000379693  2.422616      1 exon
18 chr15 72969965 72970048 ENST00000379693  2.422616      1 exon
19 chr15 72972532 72972714 ENST00000352410  0.917141      1 exon
20 chr15 72972055 72972196 ENST00000352410  0.917141      1 exon
```

A vector `type` can be used to specify if each region is an 'exon' or 'utr' while `plotgenetype="box"` plots regions as a boxes rather than arrows. The data can be plotted using `plotGenes()`. The `colorby` argument is used to color the transcripts by log 10(FPKM). UTR regions are drawn as shorter boxes than exons.

```
> chrom            = "chr15"
> chromstart       = 72965000
> chromend         = 72990000
> pg = plotGenes(Sushi_transcripts.bed,chrom,chromstart,chromend ,
                 types = Sushi_transcripts.bed$type,
                 colorby=log10(Sushi_transcripts.bed$score+0.001),
                 colorbycol= SushiColors(5),colorbyrange=c(0,1.0),
                 labeltext=TRUE,maxrows=50,height=0.4,plotgenetype="box")
> labelgenome( chrom, chromstart,chromend,n=3,scale="Mb")
> addlegend(pg[[1]],palette=pg[[2]],title="log10(FPKM)",side="right",
            bottominset=0.4,topinset=0,xoffset=-.035,labelside="left",
            width=0.025,title.offset=0.055)
```



3.9 Zoom functions

A critical characteristic of the Sushi package is its ability to create highly customizable, publication-ready, multi-panel figures. Here, we will create a basic three panel figure and demonstrate how the zoom functions work (`zoomsregion` and `zoombox`). To illustrate these feature we will use the `plotBedgraph()` function to plot bedgrpah data representing a DNaseI hypersensitivity experiment in K562 cells.

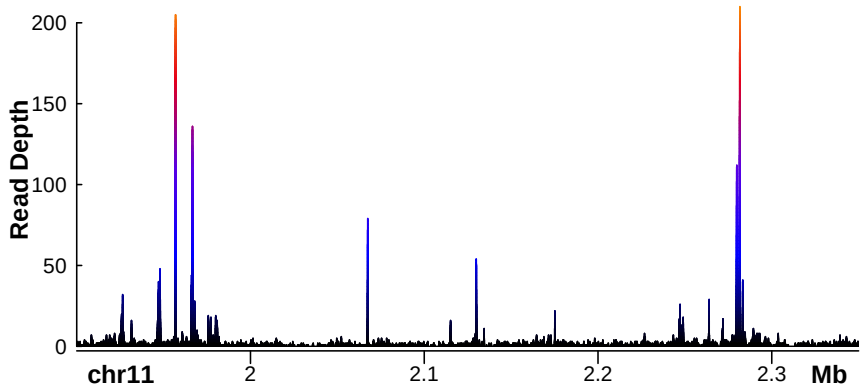
In order to make a multipanel figure we will use the R function layout. Layout divides the device into rows and columns accoriding to a matrix you provide. The matrix also tells it which plots will appear on which parts of the plotting device. Below we make a 2 by 2 matrix. The entire top row will be used to plot the first plot while the bottom row with contain two plots. For more info on layout try `?layout`.

```
> layout(matrix(c(1,1,2,3),2, 2, byrow = TRUE))
> par(mar=c(3,4,1,1))
```

Next we will add the first plot

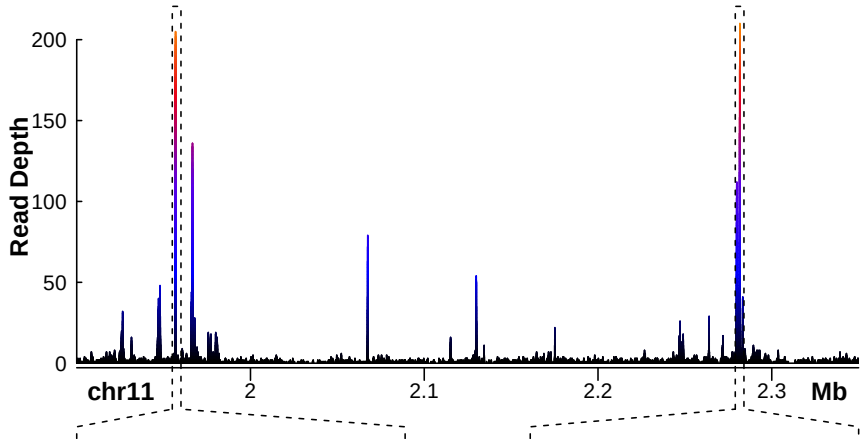
```
> chrom           = "chr11"
> chromstart      = 1900000
> chromend        = 2350000
```

```
> plotBedgraph(Sushi_DNaseI.bedgraph,chrom,chromstart=chromstart,
              chromend=chromend,colorbycol= SushiColors(5))
> labelgenome(chrom,chromstart=chromstart,chromend=chromend,n=4,
             scale="Mb")
> mtext("Read Depth",side=2,line=1.75,cex=1,font=2)
> axis(side=2,las=2,tcl=.2)
```



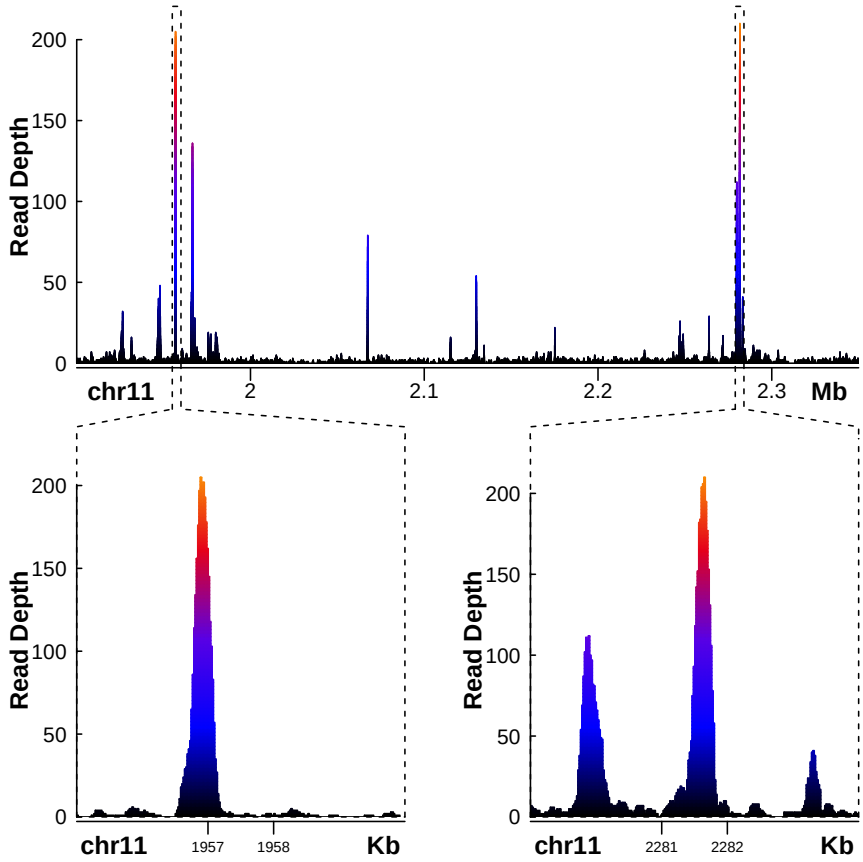
Next we will add the zoom regions using the function `zoomsregion()`. The argument `offsets` is used to precisely position the left and right edges of the widest part of the zoom.

```
> zoomregion1      = c(1955000,1960000)
> zoomregion2      = c(2279000,2284000)
> zoomsregion(zoomregion1,extend=c(0.01,0.13),wideextend=0.05,
             offsets=c(0,0.580))
> zoomsregion(zoomregion2,extend=c(0.01,0.13),wideextend=0.05,
             offsets=c(0.580,0))
```



Then we can add each of the zoomed inset regions. For, each region we need execute the `zoombox` function in order to draw the lines around the new plots.

```
> plotBedgraph(Sushi_DNaseI.bedgraph,chrom,chromstart=zoomregion1[1],
              chromend=zoomregion1[2],colorbycol= SushiColors(5))
> labelgenome(chrom,chromstart=zoomregion1[1],chromend=zoomregion1[2],
             n=4,scale="Kb",edgeblankfraction=0.2,cex.axis=.75)
> zoombox()
> mtext("Read Depth",side=2,line=1.75,cex=1,font=2)
> axis(side=2,las=2,tcl=.2)
> plotBedgraph(Sushi_DNaseI.bedgraph,chrom,chromstart=zoomregion2[1],
              chromend=zoomregion2[2],colorbycol= SushiColors(5))
> labelgenome(chrom,chromstart=zoomregion2[1],chromend=zoomregion2[2],
             n=4,scale="Kb",edgeblankfraction=0.2,cex.axis=.75)
> zoombox()
> mtext("Read Depth",side=2,line=1.75,cex=1,font=2)
> axis(side=2,las=2,tcl=.2)
```



3.10 Color functions

Sushi includes three functions to assist in the generating of R colors and color palettes: `SushiColors()`, `maptocolors()`, `opaque()`.

3.10.1 SushiColors

`SushiColors()` provides default color palettes for the Sushi package.

To see a list of available color palettes:

```
> SushiColors(palette='list')

[1] 2 3 4 5 6 7
```

To view the color palettes:

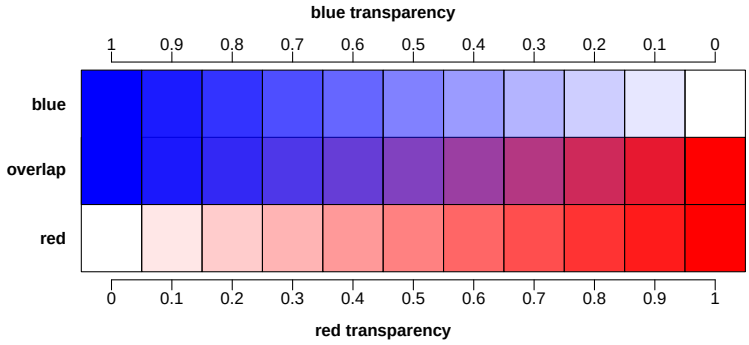
```
> plot(1,xlab='',xaxt='n',ylab='',yaxt='n',xlim=c(0.5,7.5),
      ylim=c(2,7.5),type='n')
> for (i in (2:7))
 {
   for (j in (1:i))
   {
     rect(j-.5,i,j+.5,i+.5,col=SushiColors(i)(i)[j])
   }
 }
> axis(side=2,at=(2:7),labels=(2:7),las=2)
> axis(side=1,at=(1:7),labels=(1:7))
> mtext("SushiColors",side=3,font=2, line=1, cex=1.5)
> mtext("colors",side=1,font=2, line=2)
> mtext("palette",side=2,font=2, line=2)
```

3.10.2 opaque

`opaque()` takes any color or vector of colors and makes themp opaque. The degree of transparency is determined by the argument `transparency` which is a value between 0 and 1.

```
> plot(1,xlab='',xaxt='n',ylab='',yaxt='n',bty='n',type='n',
      xlim=c(-.15,1.05),ylim=c(-1,2))
> for (i in seq(0,1,by=0.1))
 {
     rect(i-.05,-1,i+.05,1,col=opaque("red",transparency=i))
     rect(i-.05,0,i+.05,2,col=opaque("blue",transparency=1-i))
 }
> axis(side=1,at=seq(0,1,by=0.1),labels=seq(0,1,by=0.1))
> mtext("red transparency",side=1,font=2, line=2)
> axis(side=3,at=seq(0,1,by=0.1),labels=seq(1,0,by=-0.1))
> mtext("blue transparency",side=3,font=2, line=2)
> text(-0.075,1.5,labels="blue",font=2,adj=1)
```

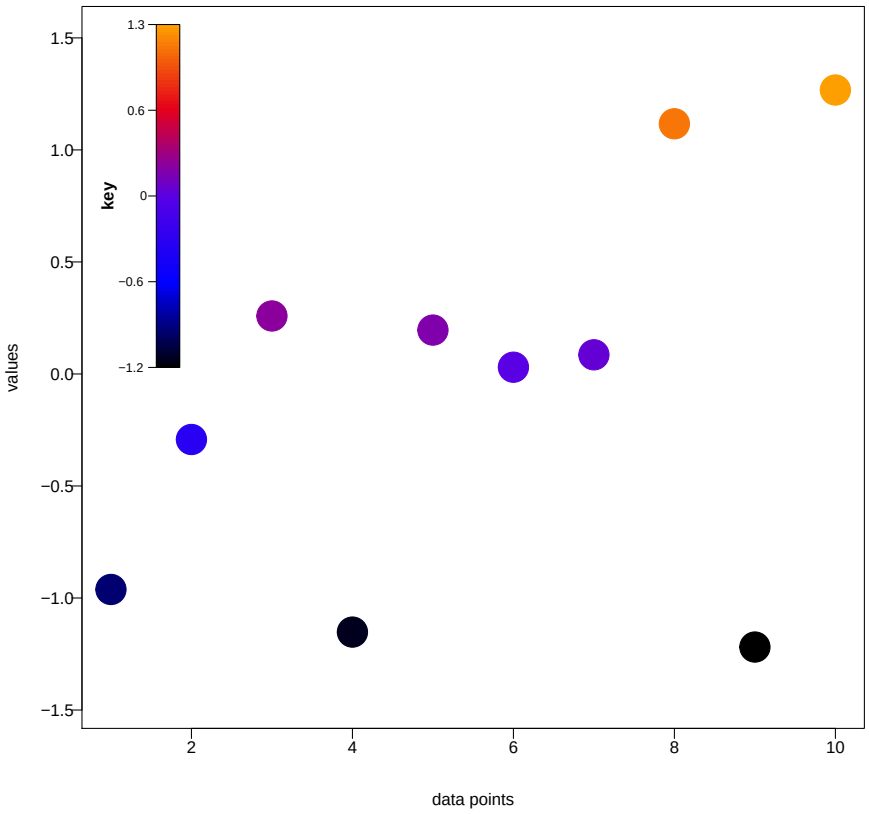
```
> text(-0.075,0.5,labels="overlap",font=2,adj=1)
> text(-0.075,-.5,labels="red",font=2,adj=1)
```



3.10.3 maptocolors

`maptocolors()` takes a vector of values and maps them to a color palette which can be used for plotting.

```
> set.seed(3)
> values = rnorm((1:10))
> colorpalette = SushiColors(5)
> plot(x=(1:10),y=values,col=maptocolors(values,colorpalette),
      pch=19,cex=4,xlab="data points",yaxt='n',ylim=range(values)*1.2)
> addlegend(range(values),title="key",palette=colorpalette,
           side='left',xoffset = -0.125,width=0.03,bottominset = 0.5, topinset = 0.025)
> axis(side=2,las=2)
```

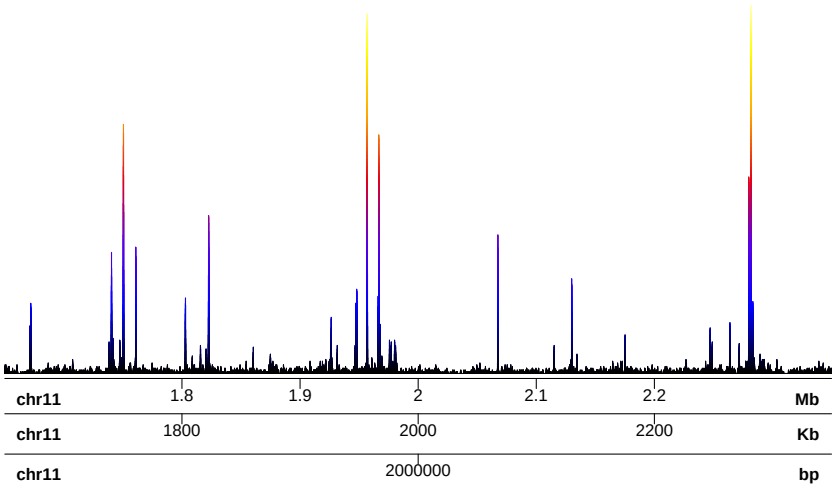


3.11 labeling functions

3.11.1 labelgenome

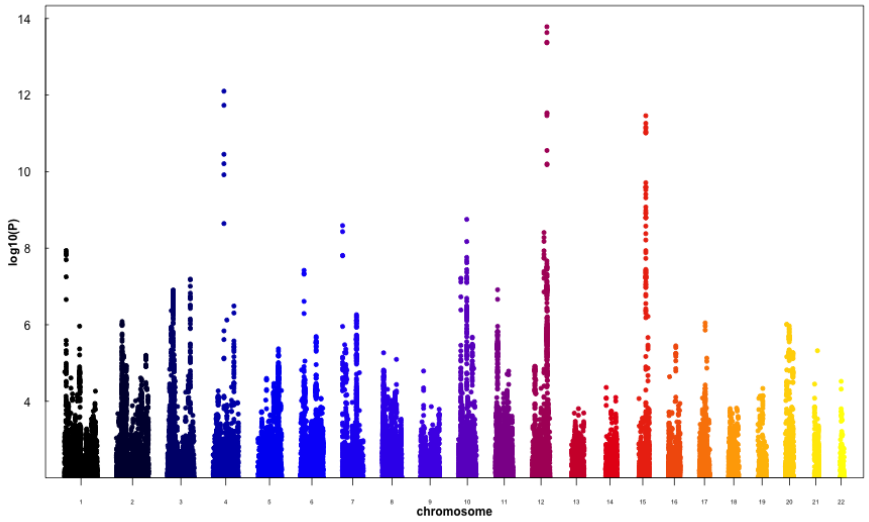
`labelgenome()` Add genome coordinates to the x-axis of a plot. The `line` argument can be used to offset the axis and `n` can be used to determine the desired umber of tick marks.

```
> par(mar=c(8,3,3,1),mgp=c(3, .3, 0))
> plotBedgraph(Sushi_DNaseI.bedgraph,chrom="chr11",chromstart=1650000,
              chromend=2350000,colorbycol=SushiColors(7))
> labelgenome(chrom="chr11",chromstart=1650000,chromend=2350000,
             side=1,n=4,scale="Mb",line=.25)
> labelgenome(chrom="chr11",chromstart=1650000,chromend=2350000,
             side=1,n=3,scale="Kb",line=2)
> labelgenome(chrom="chr11",chromstart=1650000,chromend=2350000,
             side=1,n=1,scale="bp",line=4)
```



Manhattan plots include multiple genomes and labeling the axes of Manhattan plots requires the same `genome` oject and value of `space` that were used to in `plotManhattan()`

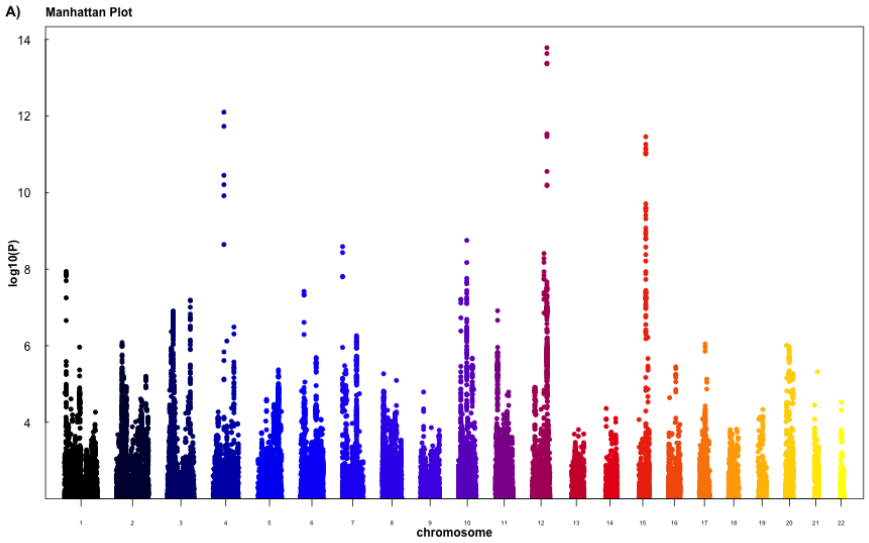
```
> plotManhattan(bedfile=Sushi_GWAS.bed,pvalues=Sushi_GWAS.bed[,5],
                col=SushiColors(6),genome=Sushi_hg18_genome,
                cex=0.75,space=0.05)
> labelgenome(genome=Sushi_hg18_genome,n=4,scale="Mb",
              edgeblankfraction=0.20,cex.axis=.5,space=0.05)
> axis(side=2,las=2,tcl=.2)
> mtext("log10(P)",side=2,line=1.75,cex=1,font=2)
> mtext("chromosome",side=1,line=1.75,cex=1,font=2)

pdf
  2
```



3.11.2 labelplot

Plot labels and titles can be added with the `labelplot()` function.

```
> labelplot("A) ","Manhattan Plot")

> plotManhattan(bedfile=Sushi_GWAS.bed,pvalues=Sushi_GWAS.bed[,5],
                col=SushiColors(6),genome=Sushi_hg18_genome,
                cex=0.75,space=0.05)
> labelgenome(genome=Sushi_hg18_genome,n=4,scale="Mb"
             ,edgeblankfraction=0.20,cex.axis=.5,space=0.05)
> axis(side=2,las=2,tcl=.2)
> mtext("log10(P)",side=2,line=1.75,cex=1,font=2)
> mtext("chromosome",side=1,line=1.75,cex=1,font=2)
> labelplot("A) ","Manhattan Plot")

pdf
  2
```




```
[1] TRUE
```

4 Tips

Other popular file formats such as BAM and GFF are not explicitly supported by Sushi. However, data stored in these formats can be easily converted to BED format using common command line tools such as the `bedtools` software suite available at https://github.com/arq5x/bedtools2. Some examples taken from the `bedtools` are shown below.

Convert BAM alignments to BED format.

```
bamToBed -i reads.bam > reads.bed
```

Convert BAM alignments to BED format using edit distance (NM) as the BED `score`.

```
bamToBed -i reads.bam -ed > reads.bed
```

Convert BAM alignments to BEDPE format.

```
bamToBed -i reads.bam -bedpe > reads.bedpe
```

These BED files can easily be read into R for use with Sushi using the following R command:

```
> read.table(file="reads.bed",sep="\t")
```

5 Appendix

For illustrative purposes we include a complex figure as published in the accompanying manuscript (Phanstiel, et al.).

```
library('Sushi')
pdfname = "vignettes/Figure_1.pdf"
Sushi_data = data(package = 'Sushi')
data(list = Sushi_data$results[,3])
makepdf = TRUE

###
### CODE
###

if (makepdf == TRUE)
{
  pdf(pdfname, height=10, width=12)
}

# make a layout for all of the plots
layout(matrix(c(1,1,1,1,
                1,1,1,1,
                2,2,8,8,
                2,2,9,9,
                3,3,10,10,
                3,3,10,10,
                4,4,11,11,
                4,4,11,11,
                5,5,12,12,
                5,5,12,12,
                6,7,13,13,
                6,7,14,14
), 12, 4, byrow=TRUE))
par(mgp=c(3,.3,0))


###
### (A) manhattan plot
###

# set the margins
par(mar=c(3,4,3,2))

# set the genomic regions
chrom1            = "chr11"
chromstart1       = 500000
chromend1         = 5050000

chrom2            = "chr15"
```

```
46 chromstart2        = 73000000
47 chromend2          = 89500000
48
49 # make the manhattan plot
50 plotManhattan(bedfile=Sushi_GWAS.bed, pvalues=Sushi_GWAS.bed
       [,5], genome=Sushi_hg18_genome, cex=0.75)
51
52 # add zoom 1
53 zoomsregion(region=c(chromstart1,chromend1), chrom=chrom1,
       genome=Sushi_hg18_genome, extend=c(0.07,0.2), wideextend
       =0.2, offsets=c(0,.535))
54
55 # add zoom 2
56 zoomsregion(region=c(chromstart2,chromend2), chrom=chrom2,
       genome=Sushi_hg18_genome, extend=c(0.07,0.2), wideextend
       =0.2, offsets=c(.535,0))
57
58 # add labels
59 labelgenome(genome=Sushi_hg18_genome, n=4, scale="Mb",
       edgeblankfraction=0.20)
60
61 # add y-axis
62 axis(side=2, las=2, tcl=.2)
63 mtext("log10(P)", side=2, line=1.75, cex=.75, font=2)
64
65 # Add plot label
66 labelplot("A)", "␣GWAS", letteradj=-.025)
67
68
69 ###
70 ### (B) Hi-C
71 ###
72
73 # set the margins
74 par(mar=c(3,4,2,2))
75
76 # set the genomic regions
77 chrom             = "chr11"
78 chromstart        = 500000
79 chromend          = 5050000
80 zoomregion        = c(1700000,2350000)
81
82 # plot the HiC data
83 phic = plotHic(Sushi_HiC.matrix, chrom, chromstart, chromend,
       max_y = 20, zrange=c(0,28))
84
85 # add labels
86 labelgenome(chrom, chromstart, chromend, n=4, scale="Mb",
       edgeblankfraction=0.20)
87
```

```
88  # add the legend
89  addlegend(phic[[1]], palette=phic[[2]], title="score", side="
        right", bottominset=0.4, topinset=0, xoffset=-.035,
        labelside="left", width=0.025, title.offset=0.035)
90
91  # add zoom
92  zoomsregion(region=zoomregion, extend=c(0.05,0.25))
93
94  # add zoombox
95  zoombox(zoomregion=zoomregion)
96
97  # Add plot label
98  labelplot("B)", "␣HiC")
99
100
101 ###
102 ### (C) 5C
103 ###
104
105 # set the margins
106 par(mar=c(3,4,2,2))
107
108 # set the genomic regions
109 chrom           = "chr11"
110 chromstart      = 1650000
111 chromend        = 2350000
112
113 # plot the loops
114 pbpe = plotBedpe(Sushi_5C.bedpe, chrom, chromstart, chromend,
        heights=Sushi_5C.bedpe$score, offset=0, flip=FALSE, bty='n
        ', lwd=1, plottype="loops", colorby=Sushi_5C.bedpe$
        samplenumber, colorbycol=SushiColors(3))
115
116 # add zoombox
117 zoombox(passthrough=TRUE)
118
119 # add the genome labels
120 labelgenome(chrom, chromstart, chromend, n=3, scale="Mb")
121
122 # add the legend
123 legend("topright", inset=0.01, legend=c("K562","HeLa","GM12878
        "), col=SushiColors(3)(3), pch=19, bty='n', text.font=2)
124
125 # add y-axis
126 axis(side=2, las=2, tcl=.2)
127 mtext("Z-score", side=2, line=1.75, cex=.75, font=2)
128
129 # Add plot label
130 labelplot("C)", "␣5C")
131
```

```
132
133 ###
134 ### (D) ChIA PET (PolII)
135 ###
136
137 # set the margins
138 par(mar=c(3,4,2,2))
139
140 # set the genomic regions
141 chrom             = "chr11"
142 chromstart        = 1650000
143 chromend          = 2350000
144
145 # plot the loops
146 pbpe = plotBedpe(Sushi_ChIAPET_pol2.bedpe, chrom, chromstart,
        chromend, flip=TRUE, bty='n', lwd=1, plottype="lines",
        colorby=abs(Sushi_ChIAPET_pol2.bedpe$start1-Sushi_ChIAPET_
        pol2.bedpe$start2), colorbycol=SushiColors(5))
147
148 # add the genome labels
149 labelgenome(chrom, chromstart, chromend, n=4, scale="Mb")
150
151 # add the legend
152 addlegend(pbpe[[1]], palette=pbpe[[2]], title="distance␣(bp)",
        side="right", bottominset=0.05, topinset=0.35, xoffset
        =-.035, labelside="left", width=0.025, title.offset=0.08,
        labels.digits=0)
153
154 # add zoombox
155 zoombox(passthrough=TRUE)
156
157 # Add plot label
158 labelplot("D)", "␣ChIA-PET␣(Pol2)")
159
160
161 ###
162 ### (E) DNaseI
163 ###
164
165 # set the margins
166 par(mar=c(3,4,2,2))
167
168 # set the genomic regions
169 chrom             = "chr11"
170 chromstart        = 1650000
171 chromend          = 2350000
172 zoomregion1       = c(1860000,1861000)
173 zoomregion2       = c(2281000,2282400)
174
175 # overlapping, transparent, and rescaled
```

```
176 plotBedgraph(Sushi_DNaseI.bedgraph, chrom, chromstart,
      chromend, colorbycol=SushiColors(5))
177
178 # add zoom 1
179 zoomsregion(zoomregion1, extend=c(-0.8,0.18), wideextend=0.10,
      offsets=c(0,.577))
180
181 # add the genome labels
182 labelgenome(chrom, chromstart, chromend, n=4, scale="Mb")
183
184 # add zoombox
185 zoombox(zoomregion=zoomregion)
186
187 # add zoom 2
188 zoomsregion(zoomregion2, extend=c(0.01,0.18), wideextend=0.10,
      offsets=c(.577,0))
189
190 # add y-axis
191 axis(side=2, las=2, tcl=.2)
192 mtext("Read Depth", side=2, line=1.75, cex=.75, font=2)
193
194 # Add plot label
195 labelplot("E)", " DnaseI")
196
197
198 ###
199 ### (F) ChIP-Seq ChIP Exo
200 ###
201
202 # set the genomic regions
203 chrom           = "chr11"
204 chromstart      = 1650000
205 chromend        = 2350000
206 zoomregion1     = c(1860000,1861000)
207 zoomregion2     = c(2281000,2282400)
208
209 # plot chip-seq data
210 plotBedgraph(Sushi_ChIPSeq_CTCF.bedgraph, chrom, zoomregion1
      [1], zoomregion1[2], transparency=.50, color=SushiColors
      (2)(2)[1])
211
212 # plot chip-seq data
213 plotBedgraph(Sushi_ChIPExo_CTCF.bedgraph, chrom, zoomregion1
      [1], zoomregion1[2], transparency=.50, color=SushiColors
      (2)(2)[2], overlay=TRUE, rescaleoverlay=TRUE)
214
215 # Add plot label
216 labelplot("F)", " ChIP-Seq / ChIP-Exo", letteradj=-.125)
217
218 # add the genome labels
```

```
219 labelgenome(chrom, zoomregion1[1], zoomregion1[2], n=3, line
    =.5, scale="Mb", edgeblankfraction=0.2)
220
221 # add zoombox
222 zoombox()
223
224 # add legend
225 legend("topright", inset=0.025, legend=c("ChIP-seq␣(CTCF)","
    ChIP-exo␣(CTCF)"), fill=opaque(SushiColors(2)(2),0.5),
    border=SushiColors(2)(2), text.font=2, cex=0.75)
226
227
228 ###
229 ### (G) Bed Pile up
230 ###
231
232 # set the genomic regions
233 chrom           = "chr11"
234 chromstart      = 1650000
235 chromend        = 2350000
236 zoomregion1     = c(1955000,1965000)
237 zoomregion2     = c(2281000,2282400)
238
239 # plt the chip-seq data as a pile-up
240 plotBed(beddata=Sushi_ChIPSeq_pol2.bed, chrom=chrom,
    chromstart=zoomregion2[1], chromend=zoomregion2[2],
    colorby=Sushi_ChIPSeq_pol2.bed$strand, colorbycol=
    SushiColors(2), wiggle=0.001, height=0.25)
241
242 # add the genome labels
243 labelgenome(chrom, zoomregion2[1], zoomregion2[2], n=2, scale=
    "Mb")
244
245 # add zoombox
246 zoombox()
247
248 # add legend
249 legend("topright", inset=0.025, legend=c("reverse","forward"),
     fill=SushiColors(2)(2), border=SushiColors(2)(2), text.
    font=2, cex=0.75)
250
251 # Add plot label
252 labelplot("G)", "␣ChIP-Seq", letteradj=-.125)
253
254
255 ###
256 ### (H) manhattan plot zoomed
257 ###
258
259 # set the margins
```

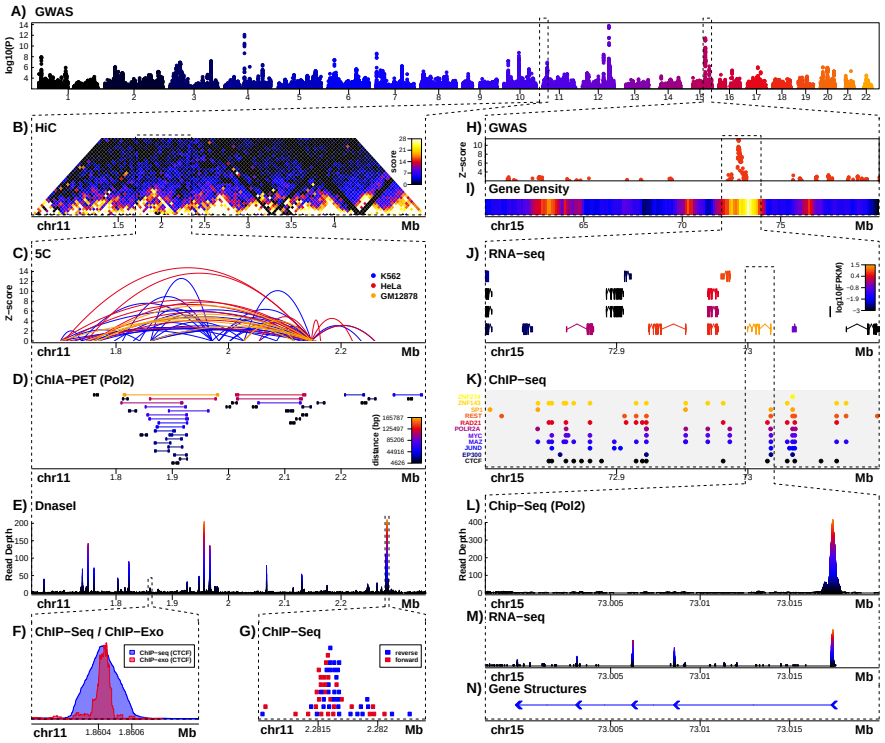
```
260 par(mar=c(0.1,4,2,2))
261
262 # set the genomic regions
263 chrom           = "chr15"
264 chromstart      = 60000000
265 chromend        = 80000000
266 chromstart2     = 72000000
267 chromend2       = 74000000
268
269 # make the manhattan plot
270 plotManhattan(bedfile=Sushi_GWAS.bed, chrom=chrom2, chromstart
        =chromstart, chromend=chromend, pvalues=Sushi_GWAS.bed$
        pval.GC.DBP, col=SushiColors(6)(nrow(Sushi_hg18_genome))
        [15], cex=0.75)
271
272 # add zoom in
273 zoomsregion(region=c(chromstart2,chromend2), chrom=chrom2,
        genome=NULL, extend=c(0.075,1), offsets=c(0.0,0))
274
275 # add zoom box
276 zoombox(passthrough=TRUE, topextend=5)
277
278 # add y-axis
279 axis(side=2, las=2, tcl=.2)
280 mtext("Z-score", side=2, line=1.75, cex=.75, font=2)
281
282 # Add plot label
283 labelplot("H)", "␣GWAS")
284
285
286 ###
287 ### (I) Gene density
288 ###
289
290 # set the margins
291 par(mar=c(3,4,1.8,2))
292
293 # set the genomic regions
294 chrom           = "chr15"
295 chromstart      = 60000000
296 chromend        = 80000000
297 chrom_biomart   = gsub("chr","",chrom)
298
299 # set the mart (since we want hg18 coordinates)
300 mart=useMart(host='may2009.archive.ensembl.org', biomart='
        ENSEMBL_MART_ENSEMBL', dataset='hsapiens_gene_ensembl')
301
302 # get just gene info
303 geneinfobed = getBM(attributes=c("chromosome_name","start_
        position","end_position"), filters=c("chromosome_name","
```

```
        start","end"), values=list(chrom_biomart,chromstart,
        chromend), mart=mart)
304
305 # add "chr" to the chrom column
306 geneinfobed[,1] = paste("chr",geneinfobed[,1],sep="")
307
308 # plot gene density
309 plotBed(beddata=geneinfobed[!duplicated(geneinfobed),], chrom=
        chrom, chromstart=chromstart, row='supplied', chromend=
        chromend, palettes=list(SushiColors(7)), type="density")
310
311 #label genome
312 labelgenome(chrom=chrom, chromstart, chromend, n=4, scale="Mb"
        , edgeblankfraction=0.10)
313
314 # add zoom in
315 zoomsregion(region=c(chromstart2,chromend2), chrom=chrom2,
        genome=NULL, extend=c(2,1.0), wideextend=.75, offsets=c
        (0.0,0))
316
317 # add zoombox
318 zoombox(zoomregion=c(chromstart2,chromend2), topextend=5)
319
320 # Add plot label
321 labelplot("I)", "␣Gene␣Density")
322
323
324 ###
325 ### (J) RNA seq
326 ###
327
328 # set the margins
329 par(mar=c(3,4,2,2))
330
331 # set the genomic regions
332 chrom2              = "chr15"
333 chromstart2         = 72800000
334 chromend2           = 73100000
335 zoomregion          = c(72998000,73020000)
336 chrom2_biomart      = 15
337
338 # plot transcripts
339 pg = plotGenes(Sushi_transcripts.bed, chrom2, chromstart2,
        chromend2, types=Sushi_transcripts.bed$type, colorby=log10
        (Sushi_transcripts.bed$score+0.001), colorbycol=
        SushiColors(5), labeltext=FALSE, maxrows=50, height=0.4,
        plotgenetype="box")
340
341 # label genome
342 labelgenome(chrom2, chromstart2, chromend2, n=3, scale="Mb")
```

```
343
344 # add the legend
345 addlegend(pg[[1]], palette=pg[[2]], title="log10(FPKM)", side=
        "right", bottominset=0.4, topinset=0, xoffset=-.035,
        labelside="left", width=0.025, title.offset=0.055)
346
347 # add zoombox
348 zoombox(passthrough=TRUE)
349
350 # add zoom
351 zoomsregion(region=zoomregion, extend=c(-.025,1))
352
353 # Add plot label
354 labelplot("J)", "␣RNA-seq")
355
356
357 ###
358 ### (K) ChIP Seq peaks
359 ###
360
361 # set the margins
362 par(mar=c(3,4,2,2))
363
364 # set the genomic regions
365 chrom           = "chr15"
366 chromstart      = 72800000
367 chromend        = 73100000
368 zoomregion      = c(72998000,73020000)
369
370 Sushi_ChIPSeq_severalfactors.bed$color = maptocolors(Sushi_
        ChIPSeq_severalfactors.bed$row, col=SushiColors(6))
371
372 # plot it
373 plotBed(beddata=Sushi_ChIPSeq_severalfactors.bed, chrom=chrom,
        chromstart=chromstart, chromend=chromend, rownumber=Sushi
        _ChIPSeq_severalfactors.bed$row, type="circles", color=
        Sushi_ChIPSeq_severalfactors.bed$color, row="given",
        plotbg="grey95", rowlabels=unique(Sushi_ChIPSeq_
        severalfactors.bed$name), rowlabelcol=unique(Sushi_ChIPSeq
        _severalfactors.bed$color), rowlabelcex=0.75)
374
375 # label genome
376 labelgenome(chrom, chromstart, chromend, n=3, scale="Mb")
377
378 # add zoom
379 zoombox(zoomregion = zoomregion)
380
381 # add zoom in
382 zoomsregion(region=zoomregion, chrom=chrom, extend=c(0.5,.22),
        wideextend=0.15, offsets=c(0.0,0))
```

```
383
384 # Add plot label
385 labelplot("K", " ChIP-seq")
386
387
388 ###
389 ### (L) Pol2 bedgrpah
390 ###
391
392 # set the margins
393 par(mar=c(3,4,2,2))
394
395 # set the genomic regions
396 chrom           = "chr15"
397 chromstart      = 72998000
398 chromend        = 73020000
399
400 # plot the Pol2 bedgraph data
401 plotBedgraph(Sushi_ChIPSeq_pol2.bedgraph, chrom, chromstart,
        chromend, colorbycol=SushiColors(5))
402
403 # label genome
404 labelgenome(chrom, chromstart, chromend, n=3, scale="Mb")
405
406 # add zoombox
407 zoombox(passthrough=TRUE)
408
409 # add y-axis
410 axis(side=2, las=2, tcl=.2)
411 mtext("Read Depth", side=2, line=1.75, cex=.75, font=2)
412
413 # Add plot label
414 labelplot("L", " Chip-Seq (Pol2)")
415
416
417 ###
418 ### (M) RNA-seq bedgraph
419 ###
420
421 # set the margins
422 par(mar=c(2,4,.5,2))
423
424 # set the genomic regions
425 chrom           = "chr15"
426 chromstart      = 72998000
427 chromend        = 73020000
428
429 # plot the K562 RNAseq bedgraph data
430 plotBedgraph(Sushi_RNASeq_K562.bedgraph, chrom, chromstart,
        chromend, colorbycol=SushiColors(5))
```

```
431
432 # label genome
433 labelgenome(chrom, chromstart, chromend, n=3, scale="Mb")
434
435 # add zoombox
436 zoombox(passthrough=TRUE)
437
438 # Add plot label
439 labelplot("M)", " RNA-seq")
440
441
442 ###
443 ### (N) Gene Structures
444 ###
445
446 # set the margins
447 par(mar=c(3,4,.5,2))
448
449 # set the genomic region
450 chrom           = "chr15"
451 chromstart      = 72998000
452 chromend        = 73020000
453
454 # plot gene structures
455 plotGenes(Sushi_genes.bed, chrom, chromstart, chromend,
      maxrows=1, bheight=0.15, plotgenetype="arrow", bentline=
      FALSE, labeloffset=1, fontsize=1.2,arrowlength = 0.01)
456
457 # label genome
458 labelgenome(chrom, chromstart, chromend, n=3, scale="Mb")
459
460 # add zoombox
461 zoombox()
462
463 # Add plot label
464 labelplot("N)", " Gene Structures", letterline=-0.4, titleline
      =-0.4)
465
466 if (makepdf == TRUE)
467 {
468   dev.off()
469 }
```


A) GWAS
log10(P)
B) HiC
score
chr11
Mb
C) 5C
Z−score
K562
HeLa
GM12878
D) ChIA−PET (Pol2)
distance (bp)
E) DnaseI
Read Depth
F) ChIP−Seq / ChIP−Exo
ChIP−seq (CTCF)
ChIP−exo (CTCF)
G) ChIP−Seq
reverse
forward
H) GWAS
Z−score
I) Gene Density
chr15
J) RNA−seq
log10(FPKM)
K) ChIP−seq
ZNF274
ZNF263
CEBPB
RAD21
POLR2A
MYC
MAZ
JUND
EP300
CTCF
L) Chip−Seq (Pol2)
M) RNA−seq
N) Gene Structures