

ChromHeatMap

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1 Introduction

The **ChromHeatMap** package provides functions for visualising expression data in a genomic context, by generating heat map images in which data is plotted along a given chromosome for all the samples in a data matrix.

These functions rely on the existence of a suitable **AnnotationDbi** package which provides chromosome location information for the probe- or gene-level identifiers used in your data set. The data themselves must be in either an `ExpressionSet`, or a data matrix with row names corresponding to probe or gene identifiers and columns corresponding to samples. While the **ChromHeatMap** package was originally designed for use with microarray data, given an appropriate **AnnotationDbi** package it can also be used to visualise data from next-generation sequencing experiments.

The output heatmap can include sample clustering, and data can either be plotted for each strand separately, or both strands combined onto a single heat map. An idiogram showing the cytogenetic banding pattern of the chromosome will be plotted for supported organisms (at the time of writing: *Homo sapiens*, *Mus musculus* and *Rattus norvegicus*; please contact the maintainer to request additions).

Once a heat map has been plotted, probes or genes of interest can be identified interactively. These identifiers may then be mapped back to gene symbols and other annotation via the **AnnotationDbi** package.

2 Data preparation

Expression data in the form of a data matrix must initially be mapped onto its corresponding chromosome coordinates. This is done using the `makeChrStrandData`:

```
> library("ALL")
> data("ALL")
> selSamples <- ALL$mol.biol %in% c("ALL1/AF4", "E2A/PBX1")
> ALLs <- ALL[, selSamples]
> library("ChromHeatMap")
> chrdata <- makeChrStrandData(exprs(ALLs), lib = "hgu95av2")
```

The output *chrdata* object here contains the expression data indexed by coordinate. Note that the `makeChrStrandData` function is based on the `Makesense` function in the **geneplotter** package, removing the internal call to `lowess` to avoid smoothing the data (which is undesirable in this case). The `makeChrStrandData` function is used specifically because it incorporates information on both the start and end chromosome coordinates for each locus. This allows the `plotChrMap` function to accurately represent target widths on the chromosome plot.

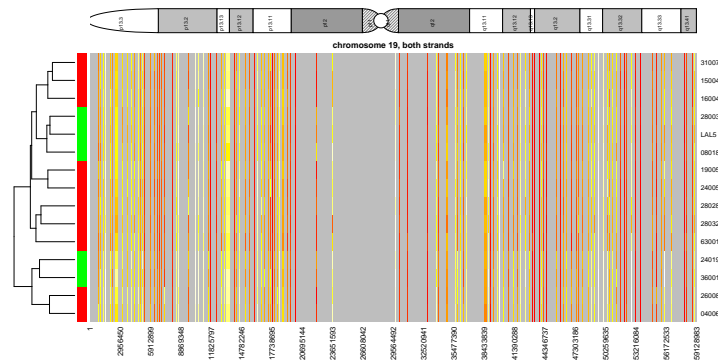
3 Plotting the heat map

Once the data has been prepared, a single call to `plotChrMap` will generate the chromosome heat map. There are many options available for this plot, and only a couple of them are illustrated here. Here we generate a whole-chromosome plot (chromosome 19), with both strands combined into a single heat map:

```
> groupcol <- ifelse(ALLs$mol.biol == "ALL1/AF4", "red", "green")
> plotChrMap(chrdata, 19, strands = "both", RowSideColors = groupcol)
```

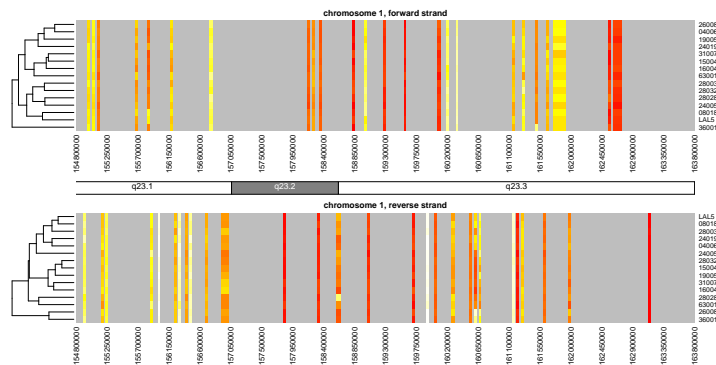
ChrMapPlot

Number of features plotted: 199



Chromosomes can be subsetted by cytoband or start/end coordinates along the chromosome. The following illustrates how one might plot the strands separately (this is the default behavior):

```
> plotmap <- plotChrMap(chrdata, 1, cytoband = "q23", interval = 50000,
+   srtCyto = 0, cexCyto = 1.2)
```



Other options include subsetting of samples, adding a color key to indicate sample subsets, deactivating the sample-based clustering and so on. See the help pages for `plotChrMap` and `drawMapDendro` for details.

Note that the default colors provided by the `heat.colors` function are not especially attractive or informative; consider using custom-defined colors, for example by using the **RColorBrewer** package.

The output of the `plotChrMap` function can be subsequently used with the `grabChrMapProbes` function which enables the user to identify the probes or genes responsible for heatmap bands of interest.

Note that the `layout` and `par` options for the current graphics device are *not* reset following generation of the image. This is so that the `grabChrMapProbes` function can accurately identify the region of interest when the user interactively clicks on the diagram.

4 Interactive probe/gene identification

Often it will be of interest to determine exactly which probes or genes are shown to be up- or down-regulated by the `plotChrMap` heat map. This can be done using the `grabChrMapProbes` function. This takes the output of the `plotChrMap` function, asks the user to mouse-click the heatmap on either side of the bands of interest and returns a character vector of the locus identifiers in that region. These can then be passed to the **AnnotationDbi** function `mget` to identify which genes are being differentially expressed.

```
> probes <- grabChrMapProbes(plotmap)
> genes <- unlist(mget(probes, envir = hgu95av2SYMBOL, ifnotfound = NA))
```

Note that due to the way the expression values are plotted, genes which lie very close to each other on the chromosome may have been averaged to give a signal that could be usefully plotted at screen resolution. In such cases the locus identifiers will be returned concatenated, separated by semicolons (e.g. “37687_i_at;37688_f_at;37689_s_at”). Typically this is easily solved by zooming in on a region of interest, using either the “cytoband” or “start” and “end” options to `plotChrMap`. See also the “interval” option for another approach to this problem.

5 Session information

The version number of R and packages loaded for generating the vignette were:

R version 2.12.0 RC (2010-10-11 r53293)

Platform: i386-pc-mingw32/i386 (32-bit)

locale:

```
[1] LC_COLLATE=C
[2] LC_CTYPE=English_United States.1252
[3] LC_MONETARY=English_United States.1252
[4] LC_NUMERIC=C
[5] LC_TIME=English_United States.1252
```

attached base packages:

```
[1] stats      graphics  grDevices  utils      datasets  methods    base
```

other attached packages:

```
[1] ChromHeatMap_1.4.0  hgu95av2.db_2.4.5    org.Hs.eg.db_2.4.6
[4] RSQLite_0.9-2       DBI_0.2-5             annotate_1.28.0
[7] AnnotationDbi_1.12.0 ALL_1.4.7             Biobase_2.10.0
```

loaded via a namespace (and not attached):

```
[1] BSgenome_1.18.0     Biostrings_2.18.0    GenomicRanges_1.2.0
[4] IRanges_1.8.0       RCurl_1.4-4.1        XML_3.2-0.1
[7] rtracklayer_1.10.0  tools_2.12.0         xtable_1.5-6
```