Introduction to *R* and *Bioconductor*

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R: Statistical Computing Environment

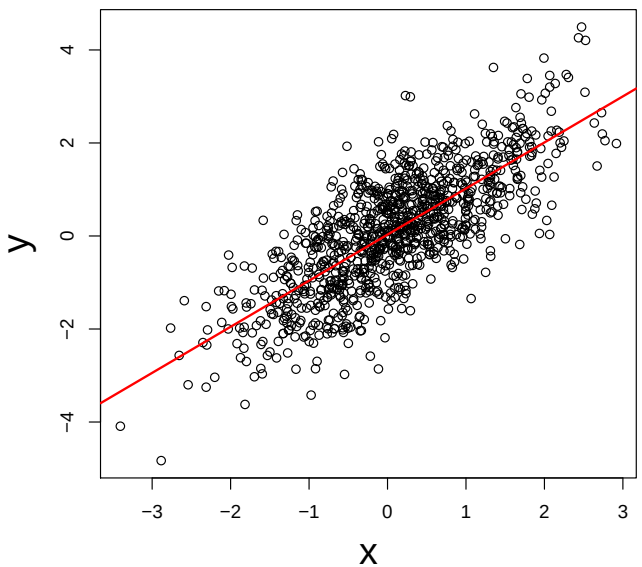
- Vectors – logical, integer, numeric, character, ...
  - `list()` – contains other vectors (recursive)
  - `factor()`, `NA` – statistical concepts
  - Can be *named* – `c(Germany=1, Argentina=0)`
- `matrix()`, `array()` – a vector with a 'dim' attribute.
- `data.frame()` – like spreadsheets; list of equal length vectors.
  - Homogenous types within a column, heterogenous types across columns.
  - An example of an *R class*.
- Other classes – more complicated arrangement of vectors.
  - Examples: the value returned by `lm()`; the *DNAStringSet* class used to hold DNA sequences.
  - plain, 'accessor', 'generic', and 'method' functions
- Packages – base, recommended, contributed.

R: Statistical Computing Environment

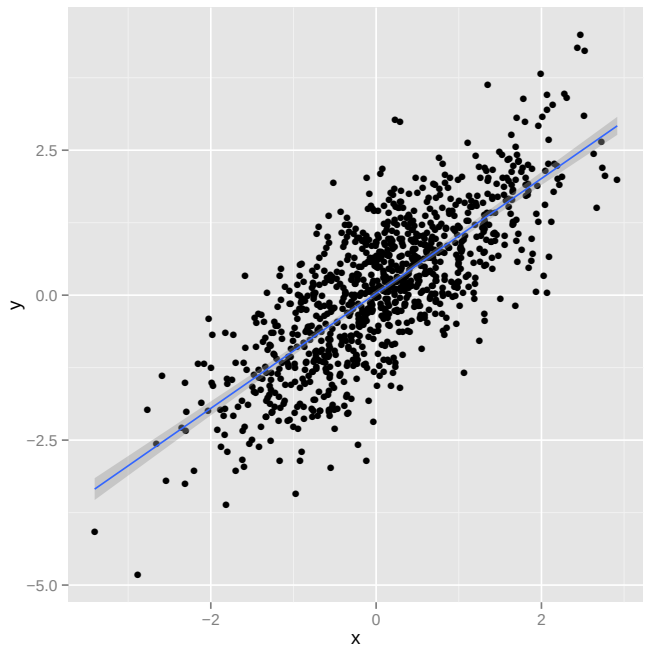
```
> 1 + 2                                  # calculator

[1] 3

> x <- rnorm(1000)                       # vectors, statistical
> y <- x + rnorm(1000, sd=.8)            # vectorized calculation
> df <- data.frame(x=x, y=y)             # object construction
> fit <- lm(y ~ x, df)                   # linear model, formula
> class(fit)                             # discovery

[1] "lm"
```

R: Statistical Computing Environment

```
> plot(y ~ x, df, cex.lab=2)
> abline(fit, col="red",
+        lwd=2)
```




```
> library(ggplot2)
> ggplot(df, aes(x, y)) +
+     geom_point() +
+     stat_smooth(method="lm")
```



R: programming concepts

- Functions – built-in (e.g., `rnorm()`); user-defined
- Subsetting – logical, numeric, character; `df[df$x > 0,]`;
- Iteration – over vector elements, `lapply()`, `mapply()`, `apply()`, `...`; e.g., `lapply(df, mean)`

R: help!

- `?data.frame`
- `methods(lm)`, `methods(class=class(fit))`
- `?"plot<tab>"`
- `help(package="Biostrings")`
- `vignette(package="GenomicRanges")`
- StackOverflow; R-help mailing list

"Hey, can you help me with this? I tried..."

Bioconductor

Analysis & comprehension of high-throughput genomic data

- $>$ 12 years old; 1024 packages; widely used
- Sequencing (RNAseq, ChIPseq, variants, copy number, . . .), microarrays, flow cytometery, proteomics, . . .
- `http://bioconductor.org`, `https://support.bioconductor.org`

Themes

- Interoperable – classes to work with genome-scale data, shared (where possible!) across packages
- Usable – package vignettes, man pages, examples, . . .
- Reproducible – 'release' and 'devel' versions, updated every 6 months

Bioconductor: *GenomicRanges*

```
> gr = exons(TxDb.Hsapiens.UCSC.hg19.knownGene); gr
GRanges with 289969 ranges and 1 metadata column:
           seqnames              ranges strand |   exon_id
             <Rle>           <IRanges>  <Rle> | <integer>
       [1]     chr1   [11874, 12227]      + |         1
       [2]     chr1   [12595, 12721]      + |         2
       [3]     chr1   [12613, 12721]      + |         3
       ...      ...               ...    ... .       ...
  [289967]     chrY [59358329, 59359508]      - |    277748
  [289968]     chrY [59360007, 59360115]      - |    277749
  [289969]     chrY [59360501, 59360854]      - |    277750
  ---
  seqinfo: 93 sequences (1 circular) from hg19 genome
```

GRanges

```
length(gr); gr[1:5]
seqnames(gr)
start(gr)
end(gr)
width(gr)
strand(gr)
```

DataFrame

```
mcols(gr)
gr$exon_id
```

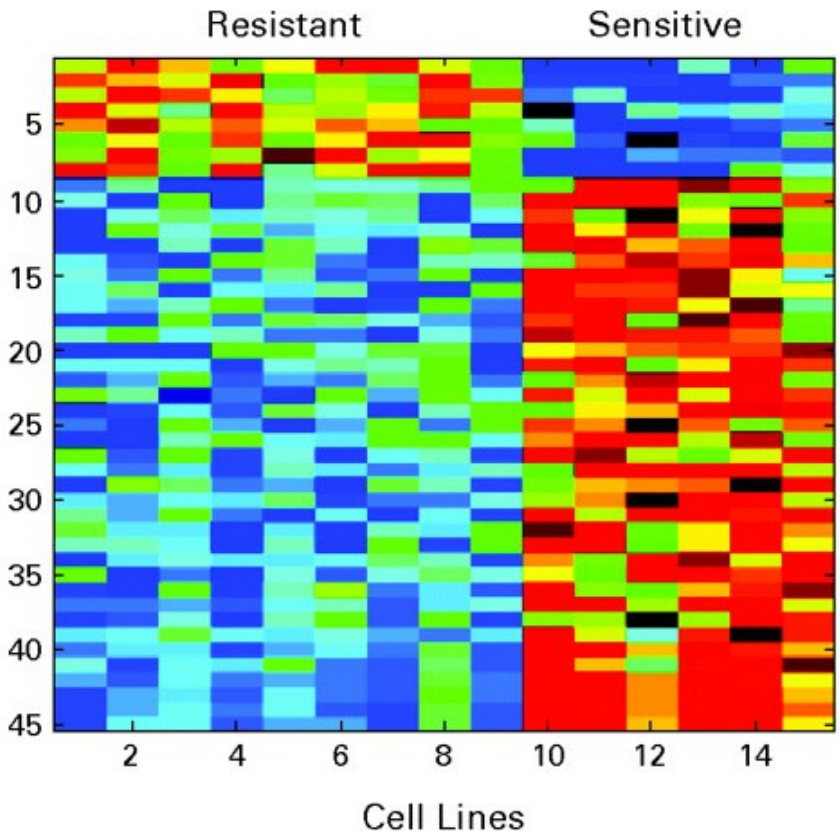
Seqinfo

```
seqlevels(gr)
seqlengths(gr)
genome(gr)
```

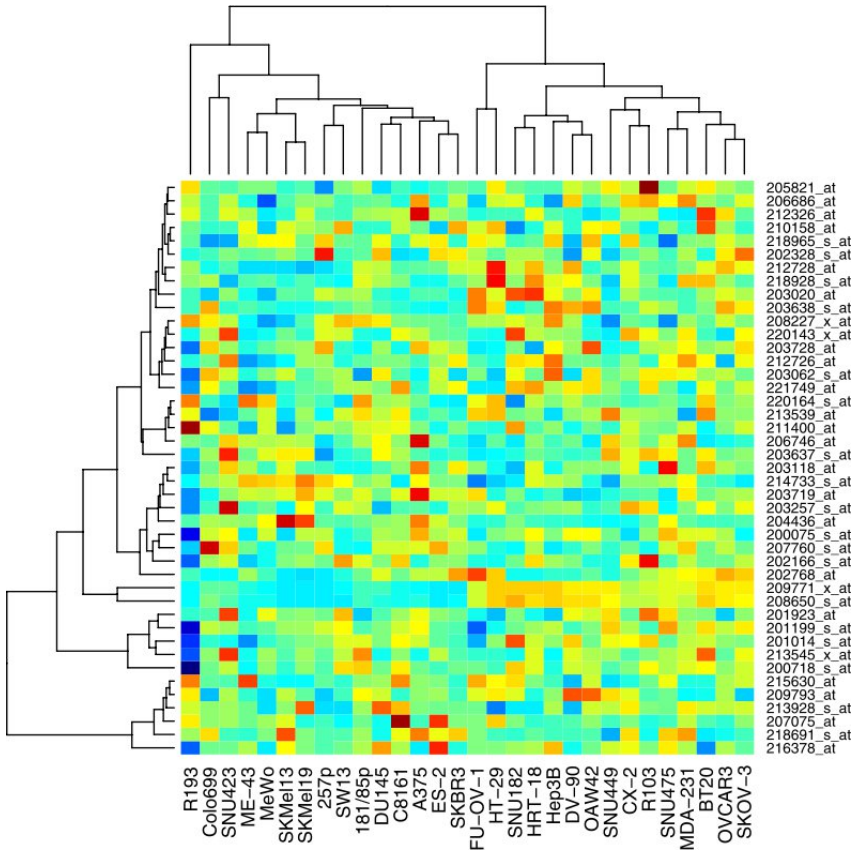
- Data: aligned reads, called peaks, SNP locations, CNVs, ...
- Annotation: gene models, variants, regulatory regions, ...
- `findOverlaps()`, `nearest()`, and many other useful range-based operations.

Bioconductor: *SummarizedExperiment* motivation

Cisplatin-resistant non-small-cell lung cancer gene sets



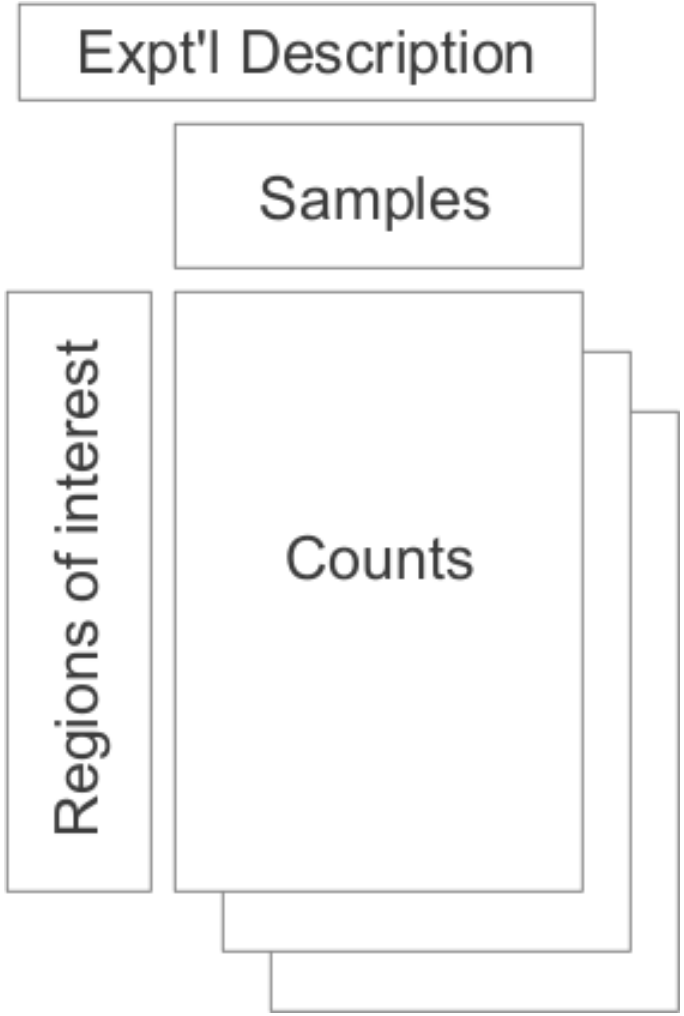
Hsu *et al.* 2007 J Clin Oncol 25: 4350-4357 (retracted)



Baggerly & Coombes 2009 Ann Appl Stat 3: 1309-1334

Coordinated, programmatic manipulation of feature, sample, and assay data

Bioconductor: *SummarizedExperiment*



Regions of interest × samples

- `assay()` – `matrix`, e.g., counts of reads overlapping regions of interest.
- `rowData()` – regions of interest as `GRanges` or `GRangesList`
- `colData()` – `DataFrame` describing samples.

```
> assay(se)[,se$Treatment == "Control"]  # Control counts
```

Bioconductor: a fun demo of *GRanges* interoperability

GenomicFeatures And 'annotation' packages to represent gene models as `GRanges`.

GenomicAlignments To input aligned reads as `GRanges`.

Gviz For visualization.

shiny For interactivity.

Bioconductor: Resources

http://bioconductor.org

- Packages – biocViews, landing pages (e.g., *AnnotationHub*)
- Course & conference material; work flows; publications
- Developer resources

https://support.bioconductor.org

- Queston & answer forum for users; usually fast, expert, friendly responses
- Contributed tutorials, news

Citations

- Huber et al. (2015) Orchestrating high-throughput genomic analysis with *Bioconductor*. Nature Methods 12:115-121.
- Lawrence et al. (2013) Software for Computing and Annotating Genomic Ranges. PLoS Comput Biol 9(8): e1003118.

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