

# Introduction to *R* and *Bioconductor*

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June 23 – 28, 2013

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# Overview

*R*

1. Vectors, data frames, arrays
2. Statistical concepts
3. Functions
4. Packages
5. From script to collaboration
6. HELP!

*Bioconductor*

1. A short history
2. Sequence analysis
3. Classes, generics and methods
4. HELP!

## Vectors, data frames, arrays

```
> x <- c(1, 2, 3, 4, 5)
> x
[1] 1 2 3 4 5
> log(x)
[1] 0.0000000 0.6931472 1.0986123 1.3862944 1.6094379
> x[c(3, 2)]
[1] 3 2
> x[3:2]
[1] 3 2
> x[c(TRUE, FALSE)]
[1] 1 3 5
```

## Vectors, data frames, arrays

```
> df <- data.frame(  
+     age = c(17, 23, 32, 37),  
+     sex = c("Male", "Female", "Female", "Male"))  
> df  
  
  age   sex  
1 17 Male  
2 23 Female  
3 32 Female  
4 37 Male  
  
> df[df$age < 30 & df$sex == "Male", ]  
  
  age   sex  
1 17 Male
```

## Vectors, data frames, arrays

```
> m <- matrix(1:8, 2, 4)
> m
      [,1] [,2] [,3] [,4]
[1,]    1    3    5    7
[2,]    2    4    6    8
> log(m)
      [,1]      [,2]      [,3]      [,4]
[1,] 0.0000000 1.098612 1.609438 1.945910
[2,] 0.6931472 1.386294 1.791759 2.079442
> rowSums(m)
[1] 16 20
```

## Statistical concepts

```
> df$sex  
[1] Male   Female Female Male  
Levels: Female Male  
  
> df$height <- c(180, 172, NA, 177)  
> df  
  
  age    sex height  
1 17   Male    180  
2 23 Female   172  
3 32 Female    NA  
4 37   Male    177
```

# Functions

- `dir, read.table (and friends), scan` List files, input data.
- `c, factor, data.frame, matrix` Create objects.
- `summary, table, xtabs` Summarize, cross-tabulate variables.
- `t.test, aov, lm, anova, chisq.test` Compare groups.
- `dist, hclust` Cluster data.
- `plot` Visualize data.
- `lapply, sapply, mapply, aggregate` Apply a function to each element of a list or vector.
- `ls, str` List objects and their structure.
- `library, search` Attach to or describe the library search path.

# Packages

Base *base, stats, graphics, ...*

Recommended *lattice, ...*

Contributed *data.table, XML, biglm, ...*

- ▶ Install a contributed package

```
> source("http://bioconductor.org/biocLite.R")
> biocLite("data.table")
```

- ▶ Use during a session

```
> library(lattice)
```

# From script to collaboration

Increasingly complicated tasks

1. Commands entered at the prompt
2. Scripts and functions saved in .R files
3. .R files and documentation ordered in packages

```
% dir MyPackage  
data/ NAMESPACE DESCRIPTION man/ R/ vignettes/
```

4. Packages shared with colleagues

Net result: sophisticated, highly reproducible research

# HELP!

```
> help.start()  
> ?t.test  
> vignette("datatable-faq")
```

- ▶ Books and training resources
- ▶ R web site and mailing list<sup>2</sup>
- ▶ StackOverflow<sup>3</sup>

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<sup>2</sup><http://r-project.org>

<sup>3</sup><http://stackoverflow.com/questions/tagged/r> 

# *Bioconductor: A short history*

## Then

- ▶ Founded 2001
- ▶ Analysis and *comprehension* of high throughput genomic data
- ▶ Initial focus: microarrays
- ▶ Reproducibility, statistical analysis, computation

## Now

- ▶ > 670 software packages
- ▶ World-wide contributions
- ▶ Sequence analysis, microarrays, systems biology, flow cytometry, . . .

Bioconductor - Home x bioconductor.org

# Bioconductor

OPEN SOURCE SOFTWARE FOR BIOINFORMATICS

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## About Bioconductor

Bioconductor provides tools for the analysis and comprehension of high-throughput genomic data. Bioconductor uses the R statistical programming language, and is open source and open development. It has two releases each year, [671 software packages](#), and an active user community. Bioconductor is also available as an [Amazon Machine Image \(AMI\)](#).

### Use Bioconductor for...

- Microarrays**  
Import Affymetrix, Illumina, Nimblegen, Agilent, and other platforms. Perform quality assessment, normalization, differential expression, clustering, classification, gene set enrichment, genetical genomics and other workflows for expression, exon, copy number, SNP, methylation and other assays. Access GEO, ArrayExpress, Biomart, UCSC, and other community resources.
- Variants**  
Read and write VCF files. Identify structural location of variants and compute amino acid coding changes for non-synonymous variants. Use SIFT and PolyPhen database packages to predict consequence of amino acid coding changes.
- Transcription Factors**  
Find candidate binding sites for known transcription factors via sequence matching.
- Counting Reads for Differential Expression**  
The [parathyroidSE](#) ExperimentData package and vignette illustrates how to count reads and perform other common operations required for differential expression analysis.

### Mailing Lists

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**Re: Unexpected results of differentia...**  
about 2 hours ago

### Events

**BioC2013**  
18 - 19 July 2013 — Seattle, WA, USA

**CSAMA 2013 (Computational Statistics for Genome Biology)**  
24 - 28 June 2013 — Brixen-Bressanone, Italy

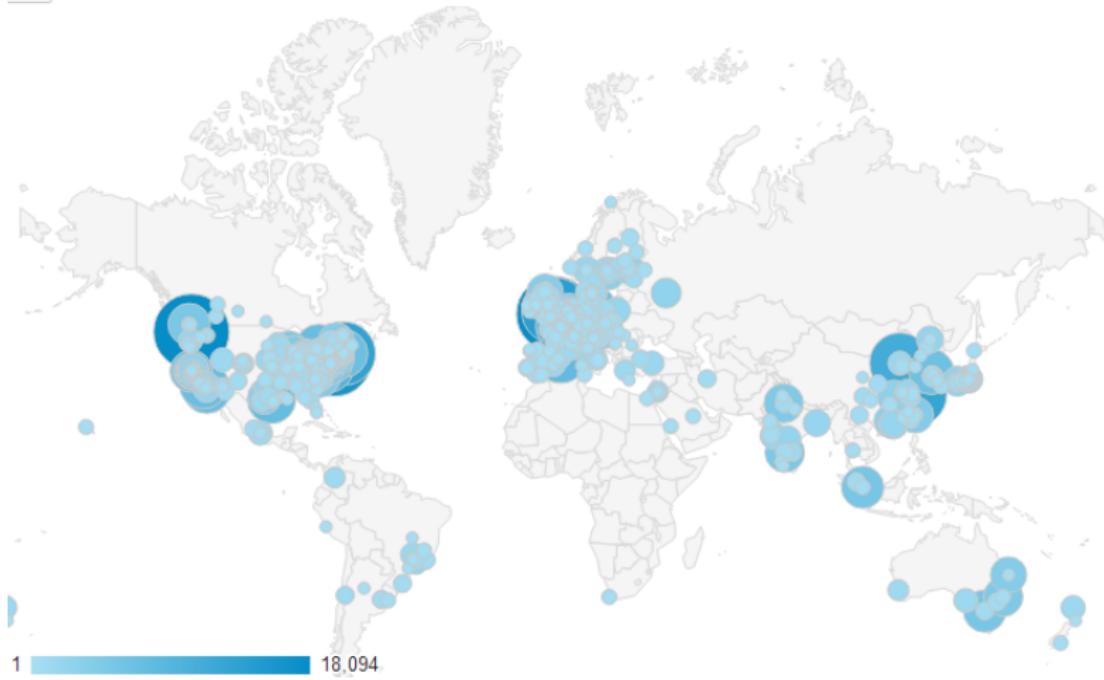
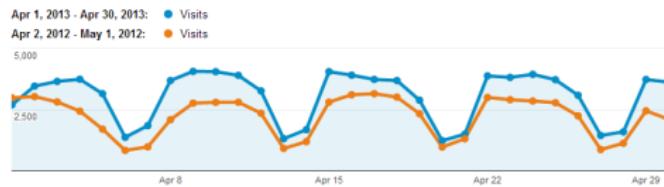
### Tweets

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Classification of alternative splicing and...

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Classification of alternative splicing and...

# Bioconductor: A short history



# Sequence Analysis (many packages!)

I/O	<i>ShortRead</i> (FASTQ), <i>Rsamtools</i> (BAM), <i>rtracklayer</i> (GFF, WIG, BED), <i>VariantAnnotation</i> (VCF), ...
Representation	<i>IRanges</i> , <i>GenomicRanges</i> , <i>GenomicFeatures</i> , <i>Biostrings</i> , <i>BSgenome</i> , ...
Annotation	<i>GenomicFeatures</i> , <i>biomaRt</i> , <i>AnnotationHub</i> , ...
Alignment	<i>gmapR</i> , <i>Rsubread</i> , <i>Biostrings</i> , ...
Visualization	<i>ggbio</i> , <i>Gviz</i> , ...
RNA-seq	<i>DESeq</i> , <i>edgeR</i> , <i>DEXSeq</i> , <i>cummeRbund</i> , ...
ChIP-seq	<i>DiffBind</i> , <i>ChIPpeakAnno</i> , ...
Variants	<i>VariantAnnotation</i> , <i>VariantTools</i> , ...
Motifs	<i>MotifDb</i> , <i>seqLogo</i> , ...
Work flows	<i>QuasR</i> , ...
...	...

## Classes, generics and methods

```
> library(ShortRead)
> fq <- 
+   readFastq("~/BigData/fastq/SRR031724_1_subset.fastq")
> fq           # 'S4' class

class: ShortReadQ
length: 1000000 reads; width: 37 cycles

> sread(fq)    # 'generic' and 'method'; another S4 class

A DNAStringSet instance of length 1000000
      width seq
[1]     37 GTTTGTCCAAGTTCTGGTAGCTGAATCCTGGGGCGC
[2]     37 GTTGTGCGATTCCCTTACTCTCATTCGGGAATTCTGTT
[3]     37 GAATTTTGAGAGCGAAATGATAGCCGATGCCCTGA
...
[1000000] 37 GAAGTCGGTACCCCTCGAACAGAGAGTCGATCTCAATG
```

# HELP!

- ▶ Use tab completion to find help on *generics* and *methods*

```
> ?sread  
> ?"reverseComplement<tab>" # tab key for completions!  
> ?"reverseComplement,DNAStringSet-method"
```

- ▶ Discover available functions and their definition.

```
> showMethods("reverseComplement")  
> showMethods(class="DNAStringSet", where=search())  
> selectMethod("reverseComplement", "DNAStringSet")
```

# Acknowledgements

## *Bioconductor* core

- ▶ Vince Carey
- ▶ Wolfgang Huber
- ▶ Robert Gentleman
- ▶ Rafael Irizarry
- ▶ Sean Davis
- ▶ Kasper Hansen

## *Bioconductor* team

- ▶ Marc Carlson (annotation)
- ▶ Valerie Obenchain (variants, ranges)
- ▶ Hervé Pagès (ranges, strings)
- ▶ Paul Shannon (systems biology)
- ▶ Dan Tenenbaum (web, build)

And the *Bioconductor* community!