Bioconductor for high-throughput genetic data

分析处理高通量基因数据：*Bioconductor*

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29 May 2016

Bioconductor
OPEN SOURCE SOFTWARE FOR BIOINFORMATICS

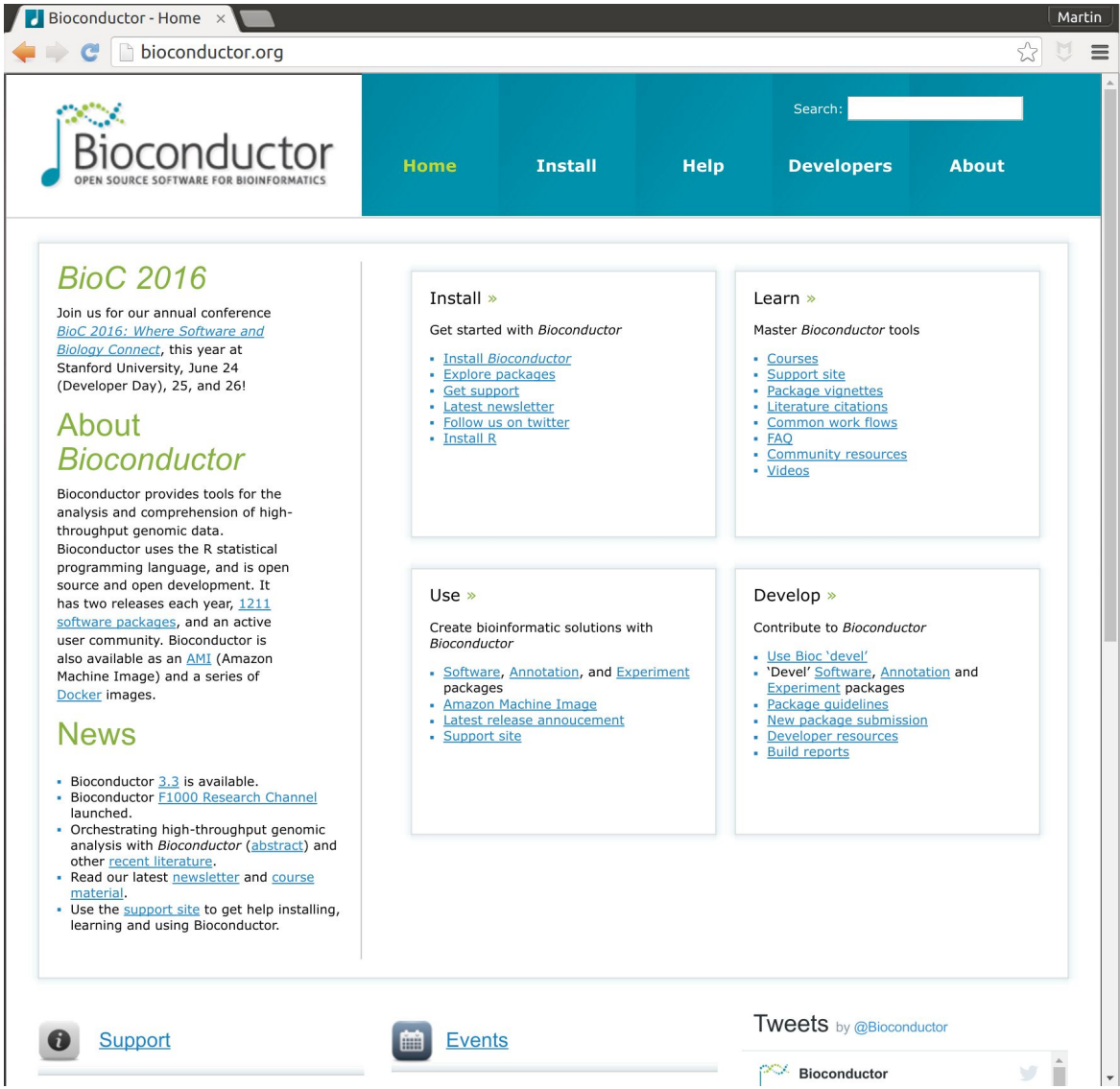
ROSWELL PARK CANCER INSTITUTE

Bioconductor

Statistical analysis and comprehension of high-throughput genomic data
统计学方法分析理解高通量基因 组数据

- Sequence (RNA-seq, ChIP-seq, Variants, …)
- Microarrays (expression, methylation, SNP, copy number, …)
- Flow cytometry
- Proteomics
- Image analysis
- …

https://bioconductor.org/
https://support.bioconductor.org



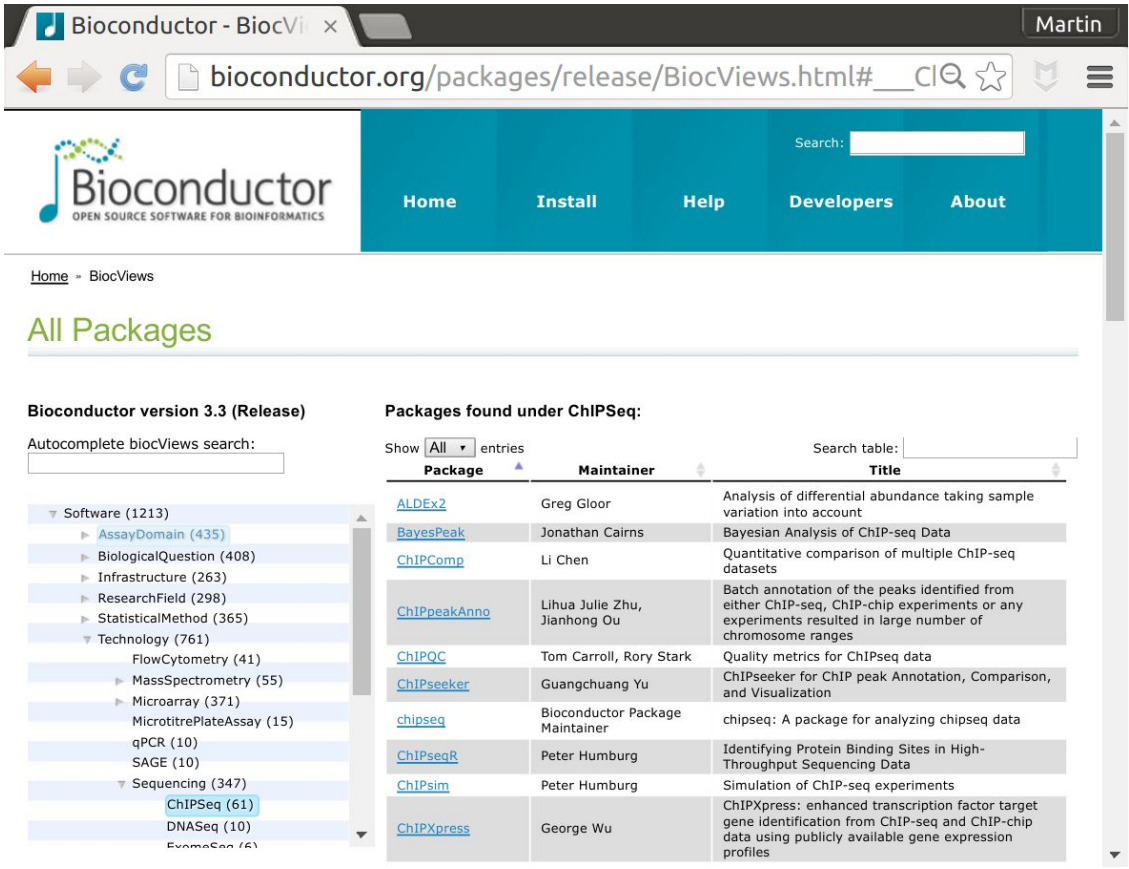
Bioconductor

1211 Software Packages　1211个软件包

- **All packages** https://bioconductor.org/packages
- Example package: GenomicRanges

Features　特征

- Vignettes and support site for learning
- Stable ‘release’ branch for users; reproducible research
- ‘Devel’ branch for new features & packages
- Classes for inter-operability between packages




```
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biocLite(“GenomicRanges”)
```

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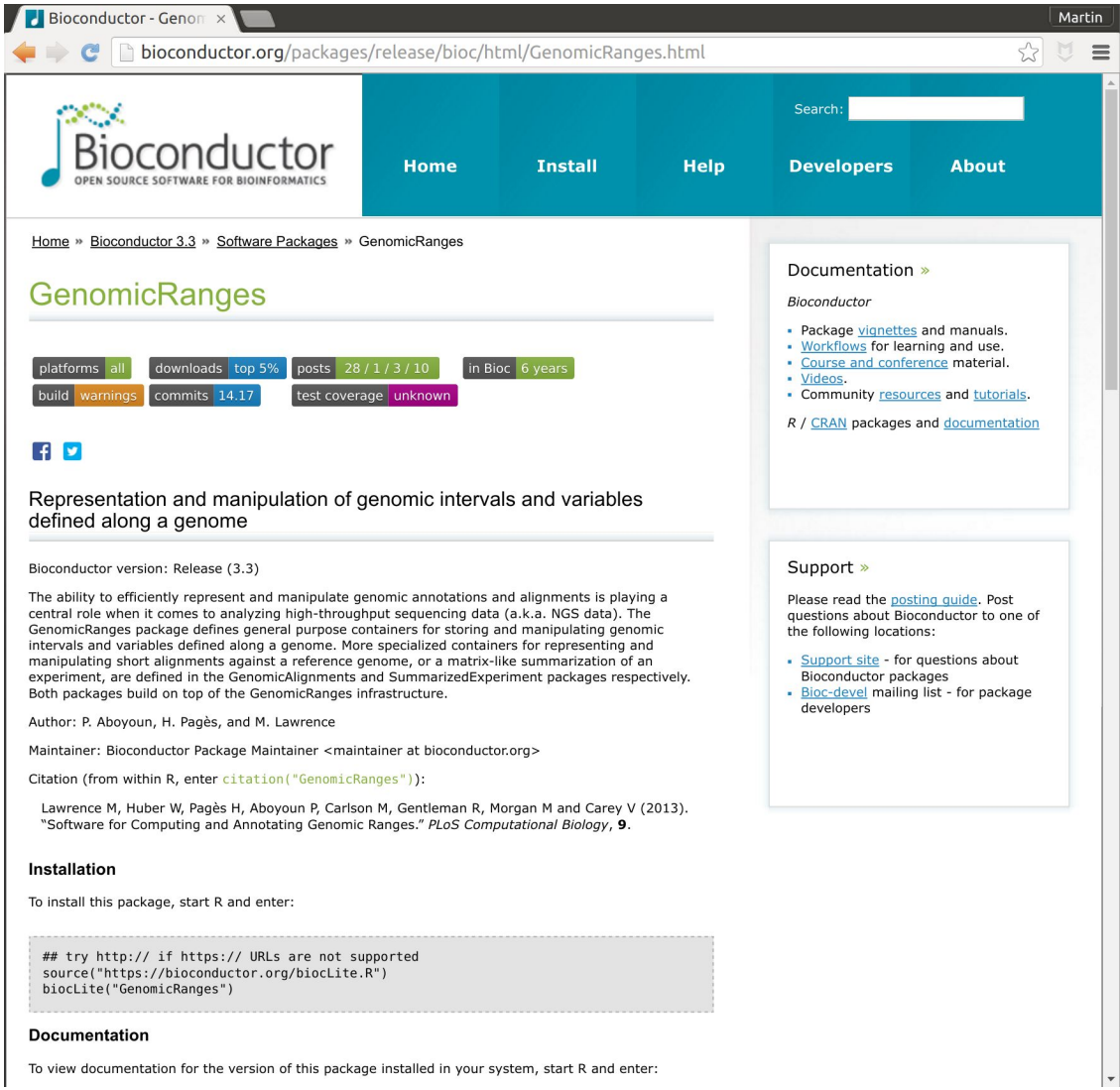


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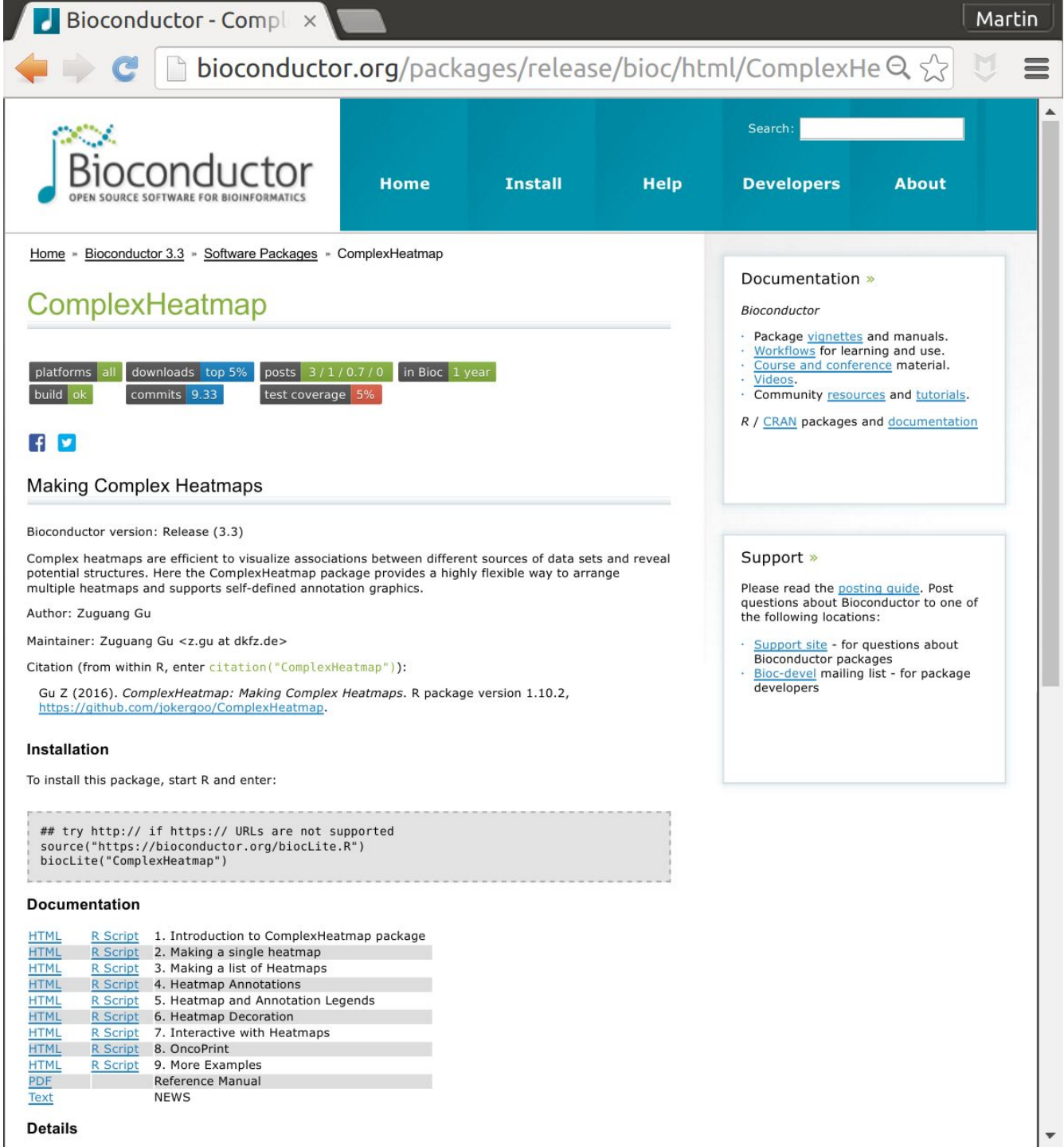
Learn

Vignettes 文档说明

- Each package!

Courses 相关课程

Support site



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Support site 支持网站

Differential analysis of count data – the DESeq2 package 2

Contents

Differential analysis of count data – the DESeq2 package

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[2] Institute for Molecular Medicine Finland (FIMM), Helsinki, Finland;
[3] European Molecular Biology Laboratory (EMBL), Heidelberg, Germany

May 15, 2016

Abstract

A basic task in the analysis of count data from RNA-seq is the detection of differentially expressed genes. The count data are presented as a table which reports, for each sample, the number of sequence fragments that have been assigned to each gene. Analogous data also arise for other assay types, including comparative ChIP-Seq, HiC, shRNA screening, mass spectrometry. An important analysis question is the quantification and statistical inference of systematic changes between conditions, as compared to within-condition variability. The package *DESeq2* provides methods to test for differential expression by use of negative binomial generalized linear models; the estimates of dispersion and logarithmic fold changes incorporate data-driven prior distributions[1]. This vignette explains the use of the package and demonstrates typical workflows. An RN
vignette but at a slower pa

DESeq2 version: 1.12.

1 Standard workflow

1.1 Quick start

Here we show the most basic steps for a differential expression analysis. These steps require you have a *RangedSummarizedExperiment* object `se` which contains the counts and information about samples. The `design` indicates that we want to measure the effect of condition, controlling for batch differences. The two factor variables `batch` and `condition` should be columns of `colData(se)`.

```
dds <- DESeqDataSet(se, design = ~ batch + condition)
dds <- DESeq(dds)
res <- results(dds, contrast=c("condition","trt","con"))
```

If you have a count matrix and sample information table, the first line would use `DESeqDataSetFromMatrix` instead of `DESeqDataSet`, as shown in Section 1.3.3.

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Vignettes 文档说明

- Each package!

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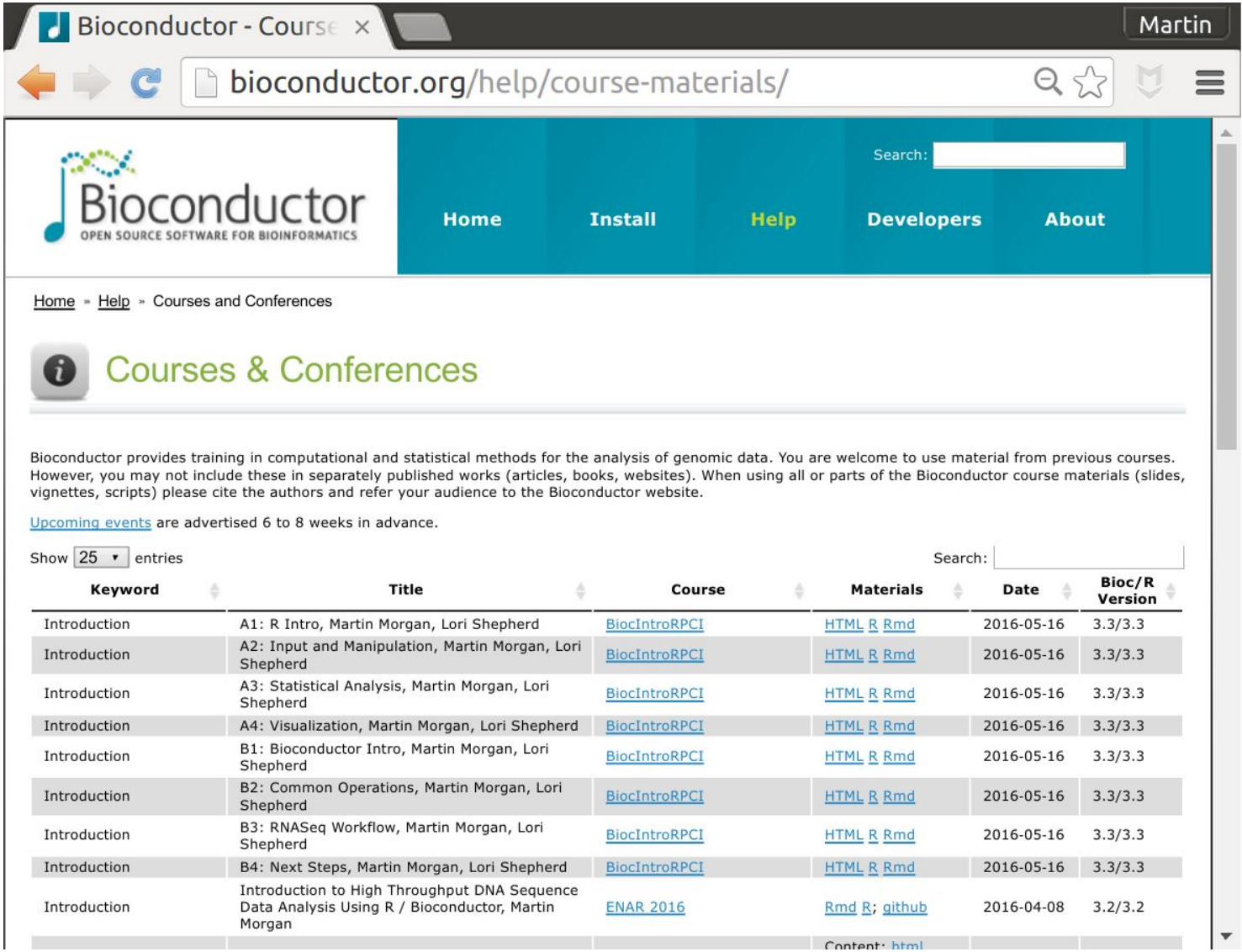
Support site 支持网站



Keyword	Title	Course	Materials	Date	Bioc/R Version
Introduction	A1: R Intro, Martin Morgan, Lori Shepherd	BiocIntroRPCI	HTML R Rmd	2016-05-16	3.3/3.3
Introduction	A2: Input and Manipulation, Martin Morgan, Lori Shepherd	BiocIntroRPCI	HTML R Rmd	2016-05-16	3.3/3.3
Introduction	A3: Statistical Analysis, Martin Morgan, Lori Shepherd	BiocIntroRPCI	HTML R Rmd	2016-05-16	3.3/3.3
Introduction	A4: Visualization, Martin Morgan, Lori Shepherd	BiocIntroRPCI	HTML R Rmd	2016-05-16	3.3/3.3
Introduction	B1: Bioconductor Intro, Martin Morgan, Lori Shepherd	BiocIntroRPCI	HTML R Rmd	2016-05-16	3.3/3.3
Introduction	B2: Common Operations, Martin Morgan, Lori Shepherd	BiocIntroRPCI	HTML R Rmd	2016-05-16	3.3/3.3
Introduction	B3: RNASeq Workflow, Martin Morgan, Lori Shepherd	BiocIntroRPCI	HTML R Rmd	2016-05-16	3.3/3.3
Introduction	B4: Next Steps, Martin Morgan, Lori Shepherd	BiocIntroRPCI	HTML R Rmd	2016-05-16	3.3/3.3
Introduction	Introduction to High Throughput DNA Sequence Data Analysis Using R / Bioconductor, Martin Morgan	ENAR 2016	Rmd R; github	2016-04-08	3.2/3.2

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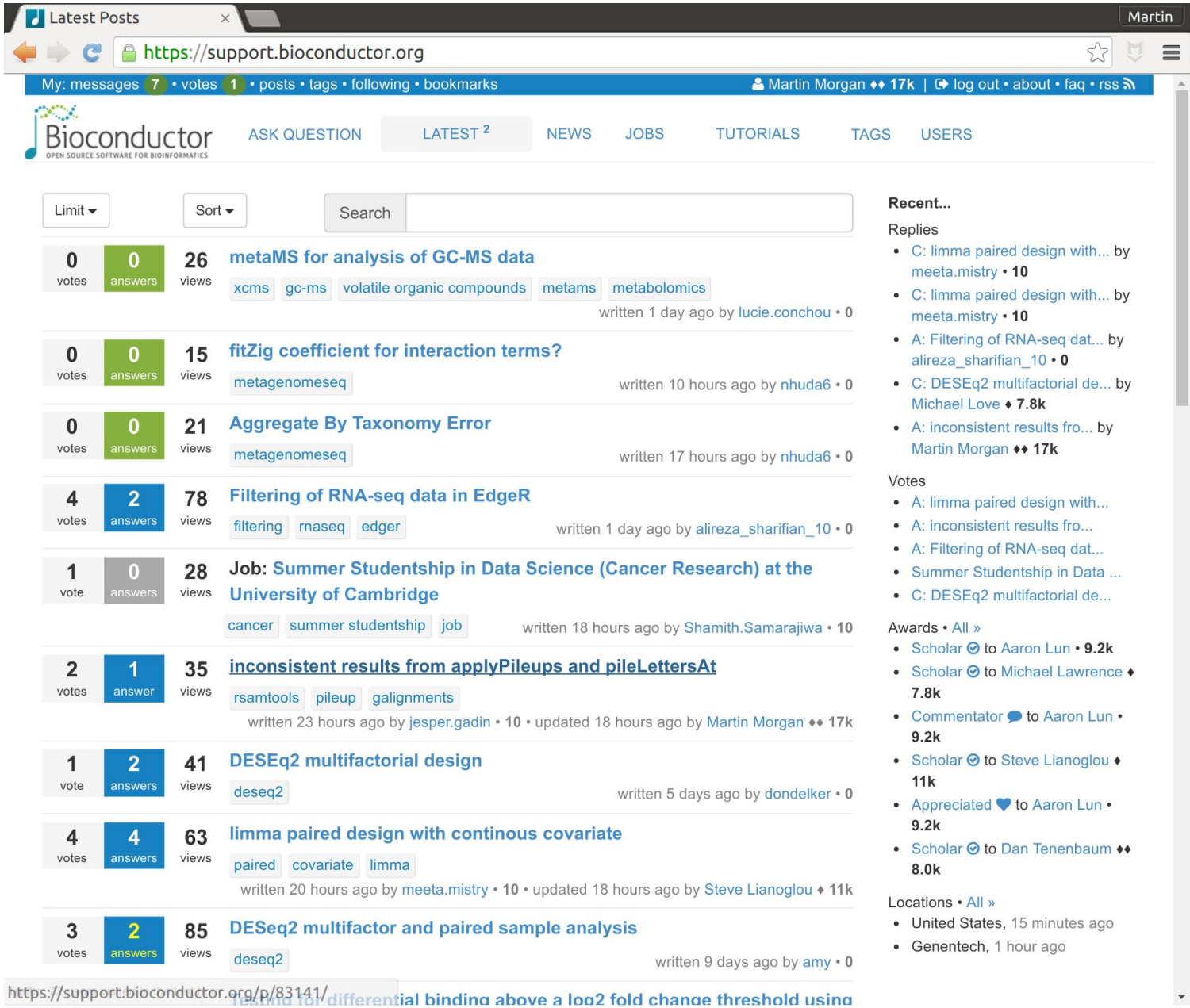


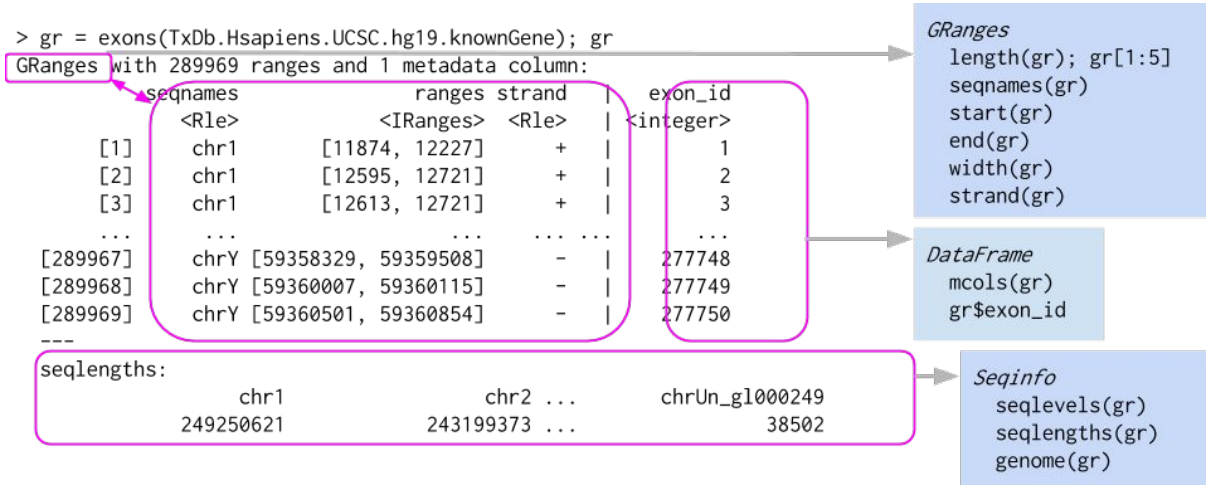
Learn

Essential Software　基础软件包

- *Biostrings*
- ***GenomicRanges***
- *SummarizedExperiment*

Annotation Resources　注释资源

- Packages
  - org.*
  - TxDb.*
  - BSgenome.*
- On-line
  - biomaRt
  - AnnotationHub



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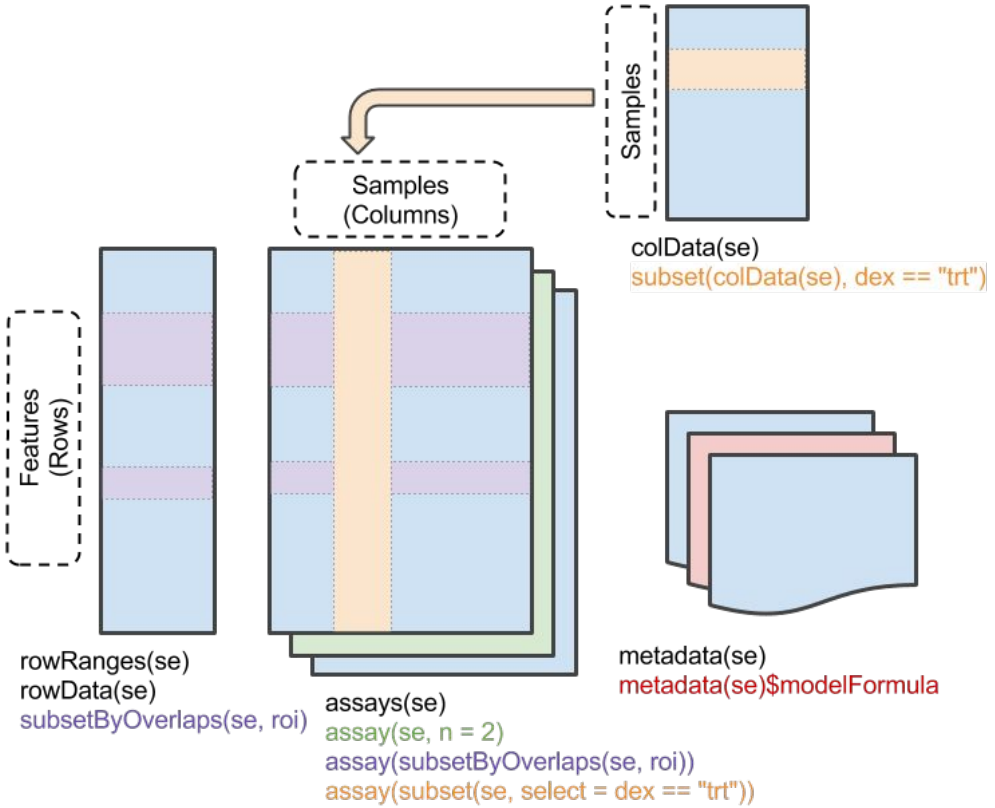


Learn

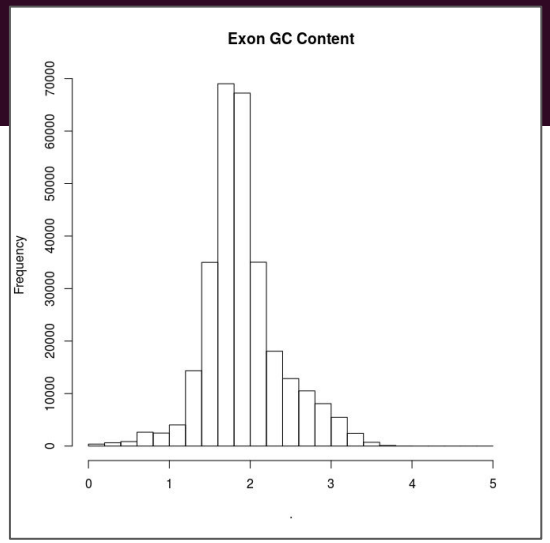
Essential Software　基础软件包

- *Biostrings*
- *GenomicRanges*
- *SummarizedExperiment*

Annotation Resources 注释资源

- Packages
  - Identifier mapping
  - Gene models
  - Genome sequence
- On-line
  - biomaRt
  - AnnotationHub

```
> library(Homo.sapiens)
> head(id <- select(Homo.sapiens, "BRCA1", c("GENENAME", "TXID"), "SYMBOL"), 3)
'select()' returned 1:many mapping between keys and columns
  SYMBOL        GENENAME  TXID
1  BRCA1 breast cancer 1 63594
2  BRCA1 breast cancer 1 63595
3  BRCA1 breast cancer 1 63596
> brca1tx <- exonsBy(Homo.sapiens, "tx")[id$TXID]
> library(BSgenome.Hsapiens.UCSC.hg19)
> head(dna <- getSeq(BSgenome.Hsapiens.UCSC.hg19, brca1tx))
DNAStringSetList of length 6
[["63594"]] TTCATTGGAACAGAAAGAAATGGATTTATCTGCTCTTCGCGTTGAAGAAGTACAAAATGTCATTA...
[["63595"]] AGATAACTGGGCCCCTGCGCTCAGGAGGCCTTCACCCTCTGCTCTGGGTAAAGGTAGTAGAGTCC...
[["63596"]] AGATAACTGGGCCCCTGCGCTCAGGAGGCCTTCACCCTCTGCTCTGGGTAAAGGTAGTAGAGTCC...
[["63597"]] CTTAGCGGTAGCCCCTTGGTTTCCGTGGCAACGGAAAAGCGCGGGAATTACAGATAAATTAAAAC...
[["63598"]] CTTAGCGGTAGCCCCTTGGTTTCCGTGGCAACGGAAAAGCGCGGGAATTACAGATAAATTAAAAC...
[["63599"]] GTACCTTGATTTCGTATTCTGAGAGGCTGCTGCTTAGCGGTAGCCCCTTGGTTTCCGTGGCAACG...
> library(magrittr)
> getSeq(BSgenome.Hsapiens.UCSC.hg19, exons(Homo.sapiens))  %>%
+     letterFrequency("GC") %>% log10 %>% hist(main="Exon GC Content")
>
```



Use

File Edit Code View Plots Session Build Debug Tools Help

Console | R Markdown

~/a/bioC/Docs/Talks/CI4CC/

```
> dds = DESeq(DESeqDataSet(airway, ~ cell + dex))
estimating size factors
estimating dispersions
gene-wise dispersion estimates
mean-dispersion relationship
final dispersion estimates
fitting model and testing
> plotMA(dds, alpha=.01)
```

Report.Rmd

Knit HTML | Run | Chunks

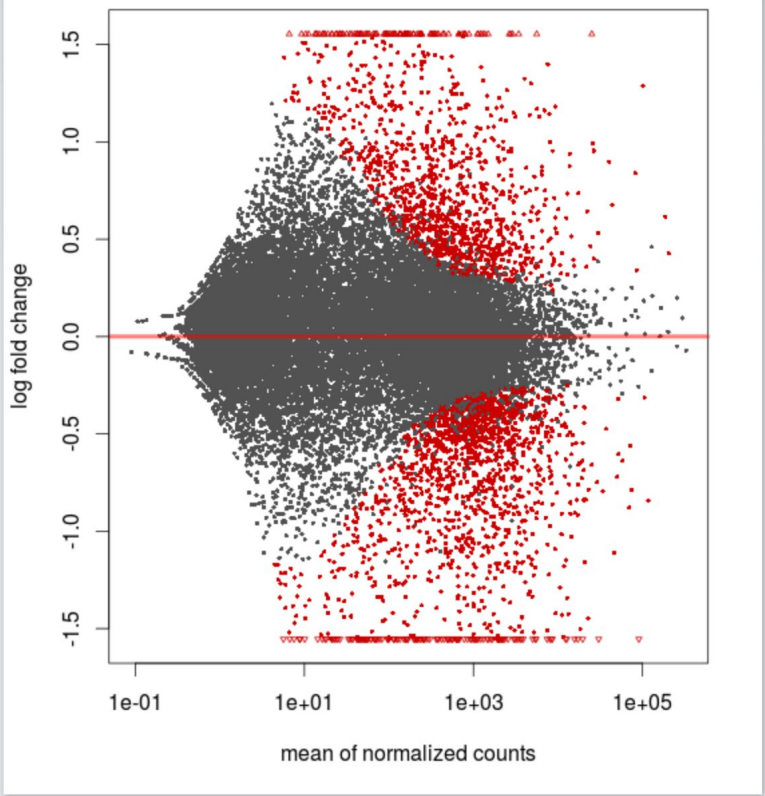
```
1  ---
2  title: "Bioinformatics Report"
3  author: "Martin Morgan"
4  date: "March 12, 2016"
5  output: html_document
6  ---
7
8  # Introduction
9
10 It is very easy to create advanced reports in a fully
   reproducible way. Reports can be distributed as PDF, HTML, ...
11
12 Reports can include figures, tables, analytic results,
   interactive applets., etc.
13
14 ```{r, warning=FALSE, message=FALSE, echo=FALSE}
15 ## R code evaluated when report produced
16 library(DESeq2)
17 library(airway)      # example data set
18 data(airway)
19 dds = DESeq(DESeqDataSet(airway, ~ cell + dex))
20 plotMA(dds, alpha=.01)
21 ```
```

17:21 | Chunk 1 | R Markdown

Environment | History

Files | Plots | Packages | Help | Viewer

Zoom | Export | Publish



Use

Gene expression　基因表达

- RNA-seq: *DESeq2*, *edgeR, scde, …*
- Microarray: *limma*

Gene regulation　基因调控

- ChIP-seq: *csaw, DiffBind*
- Methylation arrays: *minfi, missMethyl*
- Gene set enrichment: *topGO, limma*

Variants　变异

- *VariantAnnotation*
- *VariantFiltering*

Flow cytometry　流式细胞仪

- *flowCore*

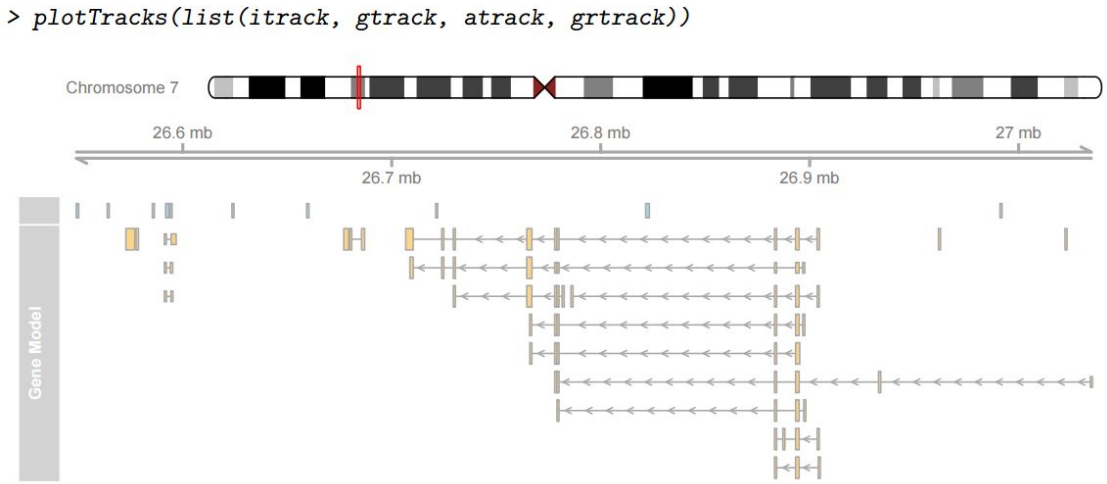
Data access　数据访问

- *biomaRt*
- *GEOquery / SRAdb*
- *TCGAbiolinks*
- *AnnotationHub / ExperimentHub*

Visualization　可视化

- *Gviz, ComplexHeatmap, ggbio, ggtree, ...*

Many other packages!



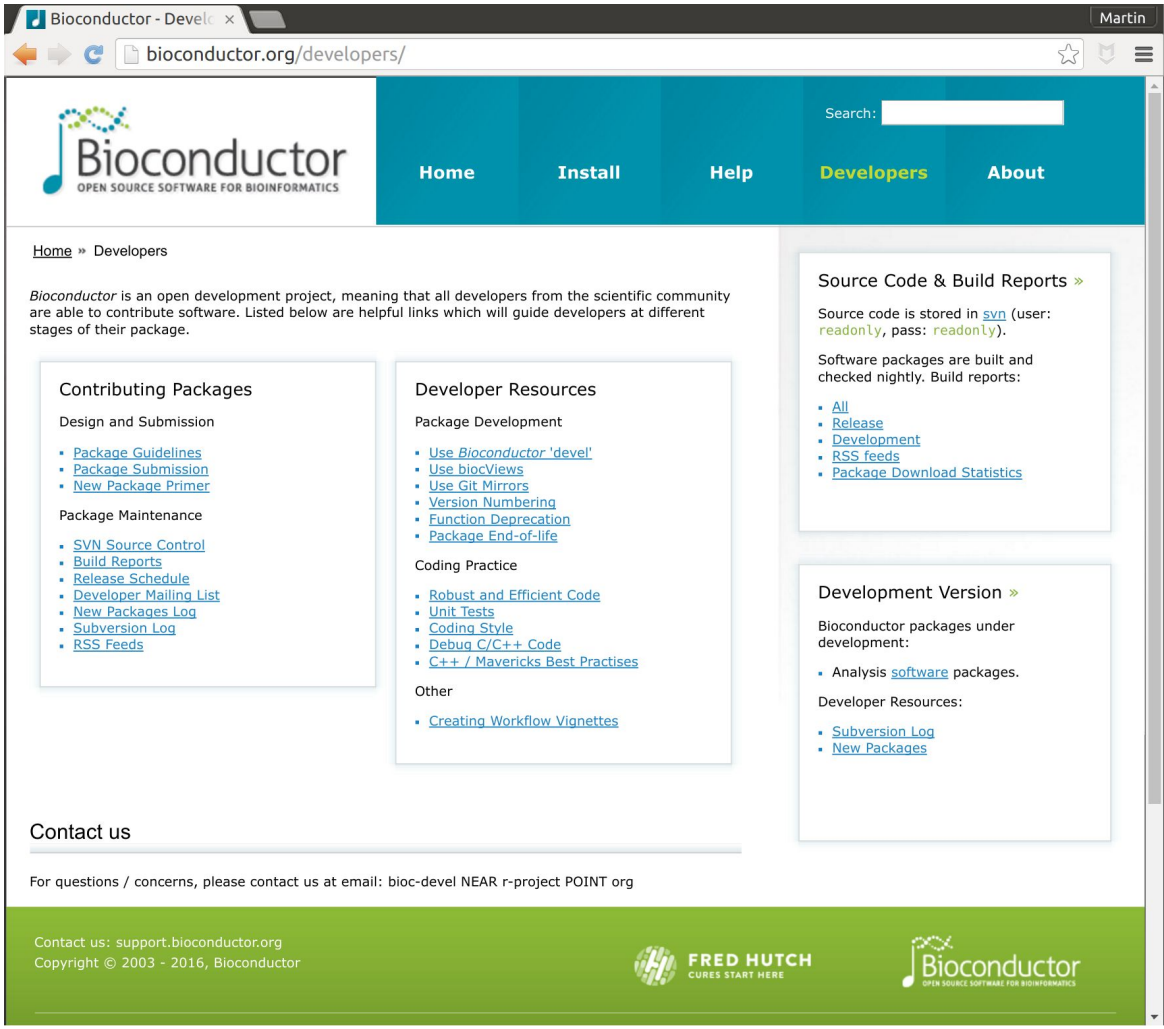
Develop

Create a package　创建包

- Use existing classes -- *DNAStringSet, GRanges, SummarizedExperiment*
- Use existing packages -- *rtracklayer, Rsamtools, Biostrings, …*
- Full vignette and examples
- Unit tests and other best practices

Submit to *Bioconductor*　提交

- Technical review
- Long-term support & maintenance
- Introduced to ‘devel’ branch, new release every April and October



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Core team (current & recent)

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- Vincent Carey, Wolfgang Huber, Robert Gentleman, Rafael Irizzary, Levi Waldron, Michael Lawrence, Sean Davis, Aedin Culhane

Scientific advisory board

- Simon Tavare (CRUK), Paul Flicek (EMBL/EBI), Simon Urbanek (AT&T), Vincent Carey (Brigham & Women's), Wolfgang Huber (EBI), Raphael Irizzary, Robert Gentleman (23andMe)

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Resources

Web sites

- https://bioconductor.org
- https://support.bioconductor.org

Events

- Annual Conference, Stanford, CA, USA, 24-26 June
- *Bioconductor* Asia-Pacific Conference, Brisbane, Australia 3-4 November
- *Bioconductor* European Developer Workshop, Basel, Switzerland, 6-7 December