

# Introduction to R/Bioconductor

**NGS analysis for gene regulation and epigenomics**

Physalia 2021

# R main classes

- Vectors

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  - Defined with `c()` function

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- Vectors
  - Defined with `c()` function
  - All the elements must be from the same class

```
> c(1, 3, 5)
[1] 1 3 5
> c('a', 'b', 'd')
[1] "a" "b" "d"
> c('a', 'b', 'd', 1, 3, 5)
[1] "a" "b" "d" "1" "3" "5"
```

# R main classes

- Vectors
  - Defined with `c()` function
  - All the elements must be from the same class
  - Can be subset with `[...]`

```
> vec <- c(1, 3, 5)
> vec[2]
[1] 3
> vec[3]
[1] 5
> vec[4]
[1] NA
```

# R main classes

- `data.frame`

# R main classes

- data.frame
  - Tabular shape
  - Defined with data.frame()

```
> dat <- data.frame(  
  "vec1" = c(4, 1, 23, 59),  
  "vec2" = c('a', 'b', 'dd', 'fgf')  
)  
> dat  
  vec1 vec2  
1    4    a  
2    1    b  
3   23   dd  
4   59  fgf  
> summary(dat)  
      vec1      vec2  
Min.   : 1.00  Length:4  
1st Qu.: 3.25  Class :character  
Median :13.50  Mode  :character  
Mean   :21.75  
3rd Qu.:32.00  
Max.   :59.00
```

# R main classes

- data.frame
  - Tabular shape
  - Defined with data.frame()
  - Subset with [ ..., ...]

```
> dat[1, ]  
  vec1 vec2  
1    4    a  
> dat[, 2]  
[1] "a"  "b"  "dd" "fgf"  
> dat[2, 2]  
[1] "b"
```



# R main classes

- data.frame
  - Tabular shape
  - Defined with data.frame()
  - Subset with [ ..., ...]
  - Columns can be accessed to with [...]] or \$

```
> dat[["vec2"]]
[1] "a"    "b"    "dd"   "fgf"
> dat$vec2
[1] "a"    "b"    "dd"   "fgf"
```

# R main classes

- Data frames can be created from tabular files using `read.table()`

```
> dat <- read.table('Share/day03/ChIP-seq_design.csv', header = TRUE, sep = ',')
> dat
```

	group	replicate	fastq_1	fastq_2	antibody	control
1	Reb1	1	Share/day03/data/SRR6453183^SLIM-ChIP^Reb1^rep1^r1.fastq.gz	Share/day03/data/SRR6453183^SLIM-ChIP^Reb1^rep1^r2.fastq.gz	Reb1	control_ChIP-Exo
2	Abf1	1	Share/day03/data/SRR6453184^SLIM-ChIP^Abf1^rep1^r1.fastq.gz	Share/day03/data/SRR6453184^SLIM-ChIP^Abf1^rep1^r2.fastq.gz	Abf1	control_ChIP-Exo
3	Rap1	1	Share/day03/data/SRR6453185^SLIM-ChIP^Rap1^rep1^r1.fastq.gz	Share/day03/data/SRR6453185^SLIM-ChIP^Rap1^rep1^r2.fastq.gz	Rap1	control_ChIP-Exo
4	Spt3-TAP	1	Share/day03/data/SRR5511881^ChIP-Exo^Spt3-TAP^rep1^r1.fastq.gz	Share/day03/data/SRR5511881^ChIP-Exo^Spt3-TAP^rep1^r2.fastq.gz	Spt3-TAP	control_ChIP-Exo
5	Spt16-TAP	1	Share/day03/data/SRR5511951^ChIP-Exo^Spt16-TAP^rep1^r1.fastq.gz	Share/day03/data/SRR5511951^ChIP-Exo^Spt16-TAP^rep1^r2.fastq.gz	Spt16-TAP	control_ChIP-Exo
6	Hsf1-TAP	1	Share/day03/data/SRR5511958^ChIP-Exo^Hsf1-TAP^rep1^r1.fastq.gz	Share/day03/data/SRR5511958^ChIP-Exo^Hsf1-TAP^rep1^r2.fastq.gz	Hsf1-TAP	control_ChIP-Exo
7	Hsf1-TAP	2	Share/day03/data/SRR5511959^ChIP-Exo^Hsf1-TAP^rep2^r1.fastq.gz	Share/day03/data/SRR5511959^ChIP-Exo^Hsf1-TAP^rep2^r2.fastq.gz	Hsf1-TAP	control_ChIP-Exo

# R main classes

```
> dat <- read.table('Share/day03/ChIP-seq_design.csv', header = TRUE, sep = ',')
> dat
```

	group	replicate	fastq_1	fastq_2	antibody	control
1	Reb1	1	Share/day03/data/SRR6453183^SLIM-ChIP^Reb1^rep1^r1.fastq.gz	Share/day03/data/SRR6453183^SLIM-ChIP^Reb1^rep1^r2.fastq.gz	Reb1	control_ChIP-Exo
2	Abf1	1	Share/day03/data/SRR6453184^SLIM-ChIP^Abf1^rep1^r1.fastq.gz	Share/day03/data/SRR6453184^SLIM-ChIP^Abf1^rep1^r2.fastq.gz	Abf1	control_ChIP-Exo
3	Rap1	1	Share/day03/data/SRR6453185^SLIM-ChIP^Rap1^rep1^r1.fastq.gz	Share/day03/data/SRR6453185^SLIM-ChIP^Rap1^rep1^r2.fastq.gz	Rap1	control_ChIP-Exo
4	Spt3-TAP	1	Share/day03/data/SRR5511881^ChIP-Exo^Spt3-TAP^rep1^r1.fastq.gz	Share/day03/data/SRR5511881^ChIP-Exo^Spt3-TAP^rep1^r2.fastq.gz	Spt3-TAP	control_ChIP-Exo
5	Spt16-TAP	1	Share/day03/data/SRR5511951^ChIP-Exo^Spt16-TAP^rep1^r1.fastq.gz	Share/day03/data/SRR5511951^ChIP-Exo^Spt16-TAP^rep1^r2.fastq.gz	Spt16-TAP	control_ChIP-Exo
6	Hsf1-TAP	1	Share/day03/data/SRR5511958^ChIP-Exo^Hsf1-TAP^rep1^r1.fastq.gz	Share/day03/data/SRR5511958^ChIP-Exo^Hsf1-TAP^rep1^r2.fastq.gz	Hsf1-TAP	control_ChIP-Exo
7	Hsf1-TAP	2	Share/day03/data/SRR5511959^ChIP-Exo^Hsf1-TAP^rep2^r1.fastq.gz	Share/day03/data/SRR5511959^ChIP-Exo^Hsf1-TAP^rep2^r2.fastq.gz	Hsf1-TAP	control_ChIP-Exo
8	Rsc9-TAP	1	Share/day03/data/SRR5511984^ChIP-Exo^Rsc9-TAP^rep1^r1.fastq.gz	Share/day03/data/SRR5511984^ChIP-Exo^Rsc9-TAP^rep1^r2.fastq.gz	Rsc9-TAP	control_ChIP-Exo
9	control_ChIP-Exo	1	Share/day03/data/SRR5512036^ChIP-Exo^Neg^rep1^r1.fastq.gz	Share/day03/data/SRR5512036^ChIP-Exo^Neg^rep1^r2.fastq.gz		
10	control_ChIP-Exo	2	Share/day03/data/SRR5512037^ChIP-Exo^Neg^rep2^r1.fastq.gz	Share/day03/data/SRR5512037^ChIP-Exo^Neg^rep2^r2.fastq.gz		
11	Cse4	1	Share/day03/data/SRR5071542^CUTnRUN^Cse4^rep1^r1.fastq.gz	Share/day03/data/SRR5071542^CUTnRUN^Cse4^rep1^r2.fastq.gz	Cse4	control_CUTnRUN
12	H2A	1	Share/day03/data/SRR5071555^CUTnRUN^H2A^rep1^r1.fastq.gz	Share/day03/data/SRR5071555^CUTnRUN^H2A^rep1^r2.fastq.gz	H2A	control_CUTnRUN
13	Mot1	1	Share/day03/data/SRR5071567^CUTnRUN^Mot1^rep1^r1.fastq.gz	Share/day03/data/SRR5071567^CUTnRUN^Mot1^rep1^r2.fastq.gz	Mot1	control_CUTnRUN
14	Sth1	1	Share/day03/data/SRR5071576^CUTnRUN^Sth1^rep1^r1.fastq.gz	Share/day03/data/SRR5071576^CUTnRUN^Sth1^rep1^r2.fastq.gz	Sth1	control_CUTnRUN
15	Sth1	2	Share/day03/data/SRR5071577^CUTnRUN^Sth1^rep2^r1.fastq.gz	Share/day03/data/SRR5071577^CUTnRUN^Sth1^rep2^r2.fastq.gz	Sth1	control_CUTnRUN
16	control_CUTnRUN	1	Share/day03/data/SRR5071586^CUTnRUN^Neg^rep1^r1.fastq.gz	Share/day03/data/SRR5071586^CUTnRUN^Neg^rep1^r2.fastq.gz		
17	control_CUTnRUN	2	Share/day03/data/SRR5071587^CUTnRUN^Neg^rep2^r1.fastq.gz	Share/day03/data/SRR5071587^CUTnRUN^Neg^rep2^r2.fastq.gz		
18	Sua7	1	Share/day03/data/SRR1951347^ChIP-Exo^Sua7^rep1^r1.fastq.gz	Share/day03/data/SRR1951347^ChIP-Exo^Sua7^rep1^r2.fastq.gz	Sua7	control_ChIP-Exo
19	Sua7	2	Share/day03/data/SRR1951348^ChIP-Exo^Sua7^rep2^r1.fastq.gz	Share/day03/data/SRR1951348^ChIP-Exo^Sua7^rep2^r2.fastq.gz	Sua7	control_ChIP-Exo
20	Rpb3	1	Share/day03/data/SRR1951349^ChIP-Exo^Rpb3^rep1^r1.fastq.gz	Share/day03/data/SRR1951349^ChIP-Exo^Rpb3^rep1^r2.fastq.gz	Rpb3	control_ChIP-Exo
21	Rpb3	2	Share/day03/data/SRR1951350^ChIP-Exo^Rpb3^rep2^r1.fastq.gz	Share/day03/data/SRR1951350^ChIP-Exo^Rpb3^rep2^r2.fastq.gz	Rpb3	control_ChIP-Exo
22	Fhl1	1	Share/day03/data/SRR1951353^ChIP-Exo^Fhl1^rep1^r1.fastq.gz	Share/day03/data/SRR1951353^ChIP-Exo^Fhl1^rep1^r2.fastq.gz	Fhl1	control_ChIP-Exo
23	Fhl1	2	Share/day03/data/SRR1951354^ChIP-Exo^Fhl1^rep2^r1.fastq.gz	Share/day03/data/SRR1951354^ChIP-Exo^Fhl1^rep2^r2.fastq.gz	Fhl1	control_ChIP-Exo
24	Ifh1	1	Share/day03/data/SRR1951359^ChIP-Exo^Ifh1^rep1^r1.fastq.gz	Share/day03/data/SRR1951359^ChIP-Exo^Ifh1^rep1^r2.fastq.gz	Ifh1	control_ChIP-Exo
25	Ifh1	2	Share/day03/data/SRR1951360^ChIP-Exo^Ifh1^rep2^r1.fastq.gz	Share/day03/data/SRR1951360^ChIP-Exo^Ifh1^rep2^r2.fastq.gz	Ifh1	control_ChIP-Exo

```
> dat[c(1,4, 6), ]
  group replicate fastq_1 fastq_2 antibody control
1  Reb1         1 Share/day03/data/SRR6453183^SLIM-ChIP^Reb1^rep1^r1.fastq.gz Share/day03/data/SRR6453183^SLIM-ChIP^Reb1^rep1^r2.fastq.gz Reb1 control_ChIP-Exo
4 Spt3-TAP      1 Share/day03/data/SRR5511881^ChIP-Exo^Spt3-TAP^rep1^r1.fastq.gz Share/day03/data/SRR5511881^ChIP-Exo^Spt3-TAP^rep1^r2.fastq.gz Spt3-TAP control_ChIP-Exo
6 Hsf1-TAP      1 Share/day03/data/SRR5511958^ChIP-Exo^Hsf1-TAP^rep1^r1.fastq.gz Share/day03/data/SRR5511958^ChIP-Exo^Hsf1-TAP^rep1^r2.fastq.gz Hsf1-TAP control_ChIP-Exo

> dat[dat$antibody == 'Fhl1', ]
  group replicate fastq_1 fastq_2 antibody control
22 Fhl1         1 Share/day03/data/SRR1951353^ChIP-Exo^Fhl1^rep1^r1.fastq.gz Share/day03/data/SRR1951353^ChIP-Exo^Fhl1^rep1^r2.fastq.gz Fhl1 control_ChIP-Exo
23 Fhl1         2 Share/day03/data/SRR1951354^ChIP-Exo^Fhl1^rep2^r1.fastq.gz Share/day03/data/SRR1951354^ChIP-Exo^Fhl1^rep2^r2.fastq.gz Fhl1 control_ChIP-Exo

> dat[dat$antibody == 'Fhl1', "fastq_1"]
[1] "Share/day03/data/SRR1951353^ChIP-Exo^Fhl1^rep1^r1.fastq.gz" "Share/day03/data/SRR1951354^ChIP-Exo^Fhl1^rep2^r1.fastq.gz"
```

# R main classes

- Lists
  - Defined with `list()` function
  - Each element can be whatever object you want
  - Each element can be named

# R main classes

```
> l <- list(  
  "first" = c(1, 2, 3, 4, 5),  
  "second" = NA,  
  "third" = seq(100, 200),  
  "fourth" = "bonjour",  
  "fifth" = lm(y ~ x, data = data.frame(x = 1:5, y = 4:8))  
)
```

- Lists

# R main classes

- Lists

```
> l <- list(
  "first" = c(1, 2, 3, 4, 5),
  "second" = NA,
  "third" = seq(100, 200),
  "fourth" = "bonjour",
  "fifth" = lm(y ~ x, data = data.frame(x = 1:5, y = 4:8))
)
> l
$first
[1] 1 2 3 4 5

$second
[1] NA

$third
 [1] 100 101 102 103 104 105 106 107 108 109 110 111 112 113 114 115 116 117 118 119 120 121 122 123 124 125 126 127
[29] 128 129 130 131 132 133 134 135 136 137 138 139 140 141 142 143 144 145 146 147 148 149 150 151 152 153 154 155
[57] 156 157 158 159 160 161 162 163 164 165 166 167 168 169 170 171 172 173 174 175 176 177 178 179 180 181 182 183
[85] 184 185 186 187 188 189 190 191 192 193 194 195 196 197 198 199 200

$fourth
[1] "bonjour"

$fifth

Call:
lm(formula = y ~ x, data = data.frame(x = 1:5, y = 4:8))

Coefficients:
(Intercept)          x
              3          1
```

# R main classes

- Lists

```
> l <- list(
  "first" = c(1, 2, 3, 4, 5),
  "second" = NA,
  "third" = seq(100, 200),
  "fourth" = "bonjour",
  "fifth" = lm(y ~ x, data = data.frame(x = 1:5, y = 4:8))
)
> l
$first
[1] 1 2 3 4 5

$second
[1] NA

$third
 [1] 100 101 102 103 104 105 106 107 108 109 110 111 112 113 114 115 116 117 118 119 120 121 122 123 124 125 126 127
[29] 128 129 130 131 132 133 134 135 136 137 138 139 140 141 142 143 144 145 146 147 148 149 150 151 152 153 154 155
[57] 156 157 158 159 160 161 162 163 164 165 166 167 168 169 170 171 172 173 174 175 176 177 178 179 180 181 182 183
[85] 184 185 186 187 188 189 190 191 192 193 194 195 196 197 198 199 200

$fourth
[1] "bonjour"

$fifth

Call:
lm(formula = y ~ x, data = data.frame(x = 1:5, y = 4:8))

Coefficients:
(Intercept)          x
              3          1

> l[[1]]
[1] 1 2 3 4 5
> l[["first"]]
[1] 1 2 3 4 5
> l$first
[1] 1 2 3 4 5
```

# R main classes

- Lists

```
> l <- list(
  "first" = c(1, 2, 3, 4, 5),
  "second" = NA,
  "third" = seq(100, 200),
  "fourth" = "bonjour",
  "fifth" = lm(y ~ x, data = data.frame(x = 1:5, y = 4:8))
)
> l
$first
[1] 1 2 3 4 5

$second
[1] NA

$third
 [1] 100 101 102 103 104 105 106 107 108 109 110 111 112 113 114 115 116 117 118 119 120 121 122 123 124 125 126 127
[29] 128 129 130 131 132 133 134 135 136 137 138 139 140 141 142 143 144 145 146 147 148 149 150 151 152 153 154 155
[57] 156 157 158 159 160 161 162 163 164 165 166 167 168 169 170 171 172 173 174 175 176 177 178 179 180 181 182 183
[85] 184 185 186 187 188 189 190 191 192 193 194 195 196 197 198 199 200

$fourth
[1] "bonjour"

$fifth

Call:
lm(formula = y ~ x, data = data.frame(x = 1:5, y = 4:8))

Coefficients:
(Intercept)          x
              3          1

> l[[1]]
[1] 1 2 3 4 5
> l[["first"]]
[1] 1 2 3 4 5
> l$first
[1] 1 2 3 4 5
> summary(l)
      Length Class  Mode
first     5  -none-  numeric
second    1  -none-  logical
third   101  -none-  numeric
fourth    1  -none-  character
fifth    12    lm     list
```



# R main classes

- You can iterate a function over each element of a list using `lapply(list, function(element) {...} )`

```
> lapply(l, function(x) class(x))
$first
[1] "numeric"

$second
[1] "logical"

$third
[1] "integer"

$fourth
[1] "character"

$fifth
[1] "lm"
```

# R main classes

- You can iterate a function over each element of a list using `lapply(list, function(element) {...} )`
  - This is the equivalent of a for loop, but:
    - Can be significantly faster
    - Parallelizable
    - Does not modify your environment
    - Returns anything you want in a list
- If you are writing a for loop, there are good chances you can switch to `lapply()`

# Vectors vs. list vs. data.frames

- Vectors are series of single elements
- Lists are nested vectors
- Data.frames are a special case of lists where all elements are of the same length

# R essential packages

- Tidyverse (<https://rstudio-education.github.io/tidyverse-cookbook/program.html>)
  - Verb-based ecosystem
  - dplyr::filter
  - dplyr::arrange
  - dplyr::mutate
  - tidyr::gather
  - ggplot2 plotting functions

Everything documented/summarized here:  
<https://www.r-bloggers.com/2020/12/the-tidyverse-in-a-table/>

# R essential packages

- Magrittr's pipe `%>%`
  - Just like a pipe in bash, for R
  - Very useful in combination with tidyverse's dplyr for data wrangling

# R essential packages

```
> mtcars
```

	mpg	cyl	displacement	horsepower	drat	weight	qsec	vs	am	gear	carb
Mazda RX4	21.0	6	160.0	110	3.90	2.620	16.46	0	1	4	4
Mazda RX4 Wag	21.0	6	160.0	110	3.90	2.875	17.02	0	1	4	4
Datsun 710	22.8	4	108.0	93	3.85	2.320	18.61	1	1	4	1
Hornet 4 Drive	21.4	6	258.0	110	3.08	3.215	19.44	1	0	3	1
Hornet Sportabout	18.7	8	360.0	175	3.15	3.440	17.02	0	0	3	2
Valiant	18.1	6	225.0	105	2.76	3.460	20.22	1	0	3	1
Duster 360	14.3	8	360.0	245	3.21	3.570	15.84	0	0	3	4
Merc 240D	24.4	4	146.7	62	3.69	3.190	20.00	1	0	4	2
Merc 230	22.8	4	140.8	95	3.92	3.150	22.90	1	0	4	2
Merc 280	19.2	6	167.6	123	3.92	3.440	18.30	1	0	4	4
Merc 280C	17.8	6	167.6	123	3.92	3.440	18.90	1	0	4	4
Merc 450SE	16.4	8	275.8	180	3.07	4.070	17.40	0	0	3	3
Merc 450SL	17.3	8	275.8	180	3.07	3.730	17.60	0	0	3	3
Merc 450SLC	15.2	8	275.8	180	3.07	3.780	18.00	0	0	3	3
Cadillac Fleetwood	10.4	8	472.0	205	2.93	5.250	17.98	0	0	3	4
Lincoln Continental	10.4	8	460.0	215	3.00	5.424	17.82	0	0	3	4
Chrysler Imperial	14.7	8	440.0	230	3.23	5.345	17.42	0	0	3	4
Fiat 128	32.4	4	78.7	66	4.08	2.200	19.47	1	1	4	1
Honda Civic	30.4	4	75.7	52	4.93	1.615	18.52	1	1	4	2
Toyota Corolla	33.9	4	71.1	65	4.22	1.835	19.90	1	1	4	1
Toyota Corona	21.5	4	120.1	97	3.70	2.465	20.01	1	0	3	1
Dodge Challenger	15.5	8	318.0	150	2.76	3.520	16.87	0	0	3	2
AMC Javelin	15.2	8	304.0	150	3.15	3.435	17.30	0	0	3	2
Camaro Z28	13.3	8	350.0	245	3.73	3.840	15.41	0	0	3	4
Pontiac Firebird	19.2	8	400.0	175	3.08	3.845	17.05	0	0	3	2
Fiat X1-9	27.3	4	79.0	66	4.08	1.935	18.90	1	1	4	1
Porsche 914-2	26.0	4	120.3	91	4.43	2.140	16.70	0	1	5	2
Lotus Europa	30.4	4	95.1	113	3.77	1.513	16.90	1	1	5	2
Ford Pantera L	15.8	8	351.0	264	4.22	3.170	14.50	0	1	5	4
Ferrari Dino	19.7	6	145.0	175	3.62	2.770	15.50	0	1	5	6
Maserati Bora	15.0	8	301.0	335	3.54	3.570	14.60	0	1	5	8
Volvo 142E	21.4	4	121.0	109	4.11	2.780	18.60	1	1	4	2

```
> mtcars %>%
+   rownames_to_column('car') %>%
+   separate(car, c('brand', 'type'), ' ', extra = "merge") %>%
+   group_by(brand) %>%
+   summarize(n = n(), ave_mpg = mean(mpg), type = list(type)) %>%
+   knitr::kable()
`summarise()` ungrouping output (override with `.groups` argument)
```

brand	n	type	ave_mpg
AMC	1	Javelin	15.20000
Cadillac	1	Fleetwood	10.40000
Camaro	1	Z28	13.30000
Chrysler	1	Imperial	14.70000
Datsun	1	710	22.80000
Dodge	1	Challenger	15.50000
Duster	1	360	14.30000
Ferrari	1	Dino	19.70000
Fiat	2	128 , X1-9	29.85000
Ford	1	Pantera L	15.80000
Honda	1	Civic	30.40000
Hornet	2	4 Drive , Sportabout	20.05000
Lincoln	1	Continental	10.40000
Lotus	1	Europa	30.40000
Maserati	1	Bora	15.00000
Mazda	2	RX4 , RX4 Wag	21.00000
Merc	7	240D , 230 , 280 , 280C , 450SE , 450SL , 450SLC	19.01429
Pontiac	1	Firebird	19.20000
Porsche	1	914-2	26.00000
Toyota	2	Corolla, Corona	27.70000
Valiant	1	NA	18.10000
Volvo	1	142E	21.40000

# R essential packages

- R per-se is useful for statistical analyses.
- How do we unlock the power of R-stats in genomics?

# What do you need in bioinformatics to study genomics?

- Most common genomic files:
  - BED format: essentially a set of chromosomal ranges



# What do you need in bioinformatics to study genomics?

- Most common genomic files:
  - BED format: essentially a set of chromosomal ranges
  - BigWig format: essentially veeeeeeeeee...eeeeery long numerical vectors

# What do you need in bioinformatics to study genomics?

- Most common genomic files:
  - BED format: essentially a set of chromosomal ranges
  - BigWig format: essentially veeeeeeeeee...eeeeery long numerical vectors
  - Fasta format: letters, letters, letters

# What do you need in bioinformatics to study genomics?

- Most common genomic files:
  - BED format: essentially a set of chromosomal ranges
  - BigWig format: essentially veeeeeeeeee...eeeeery long numerical vectors
  - Fasta format: letters, letters, letters
  - Others (bam, GFF, ...): can usually be described/built on as one of the two options above

# Bioconductor

<https://www.bioconductor.org/>

- Free, open-source repository
- Open development software project
- Based primarily on the statistical R programming language
- Bioconductor's releases are bi-annual
- **Provides tools for the analysis and comprehension of genomic data**

# Bioconductor installation

- Through BiocManager
- As easy as: `install.packages("BiocManager")`

# Bioconductor

- Integrated in R

```
jacquesserizay@LOCAL[23:08:21]:~$ R

R version 4.0.3 (2020-10-10) -- "Bunny-Wunnies Freak Out"
Copyright (C) 2020 The R Foundation for Statistical Computing
Platform: x86_64-apple-darwin17.0 (64-bit)

R is free software and comes with ABSOLUTELY NO WARRANTY.
You are welcome to redistribute it under certain conditions.
Type 'license()' or 'licence()' for distribution details.

Natural language support but running in an English locale

R is a collaborative project with many contributors.
Type 'contributors()' for more information and
'citation()' on how to cite R or R packages in publications.

Type 'demo()' for some demos, 'help()' for on-line help, or
'help.start()' for an HTML browser interface to help.
Type 'q()' to quit R.

> library(BiocManager)
Bioconductor version 3.12 (BiocManager 1.30.10), ?BiocManager::install for help
```

# Bioconductor

- Integrated in R
- Bioconductor's version depends on your R version

```
jacquesserizay@LOCAL [23:08:21]:  
R version 4.0.3 (2020-10-10) -- "Bunny-Wunnies Freak Out"  
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Type 'q()' to quit R.  
  
> library(BiocManager)  
Bioconductor version 3.12 (BiocManager 1.30.10), ?BiocManager::install for help
```

# Bioconductor

- Some Bioc packages are restricted to a certain version!

```
jacquesserizay@LOCAL [23:08:21]:  
R version 4.0.3 [2020-10-10] -- "Bunny-Wunnies Freak Out"  
Copyright (C) 2020 The R Foundation for Statistical Computing  
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Type 'q()' to quit R.  
  
> library(BiocManager)  
Bioconductor version 3.12 (BiocManager 1.30.10), ?BiocManager::install for help
```



# Installation of Bioconductor packages

- Bioconductor packages are on Bioconductor, not CRAN
- So you install them using Bioconductor's BiocManager!

```
BiocManager::install("Biostrings")
```

```
BiocManager::install(c("GenomicRanges", "rtracklayer"))
```

# Bioconductor essentials

- GRanges (through GenomicRanges package)
- XNABiostrings (through Biostrings)
- Import/export from/to common genomic files with import() and export() (through rtracklayer package)

# GRanges

- Workhorse class of Bioconductor
- Used to describe genomic intervals or ranges

```
> peaks <- rtracklayer::import('Share/day03/results/bwa/mergedLibrary/mac3/narrowPeak/Reb1_R1_peaks.narrowPeak')
> peaks
GRanges object with 369 ranges and 6 metadata columns:
      seqnames      ranges strand |          name      score signalValue    pValue    qValue      peak
      <Rle>      <IRanges> <Rle> | <character> <numeric> <numeric> <numeric> <numeric> <integer>
[1]      I      102-492      * | Reb1_R1_peak_1       81      6.32656    10.6179    8.16077       28
[2]      I    92560-92807      * | Reb1_R1_peak_2      118      8.90459    14.3647   11.80700      105
[3]     II     5846-6092      * | Reb1_R1_peak_3       63      6.16472     8.7750    6.36486       51
[4]     II  111226-111389      * | Reb1_R1_peak_4     1714     63.70210   175.6310  171.48900       76
[5]     II  124859-125004      * | Reb1_R1_peak_5      397     20.54910    42.7364   39.77400       99
...     ...      ...      ... | ...      ...      ...      ...      ...      ...
[365]   XVI  840491-840698      * | Reb1_R1_peak_365      63      6.13093     8.80750    6.39684       98
[366]   XVI  844287-844438      * | Reb1_R1_peak_366      17      3.42484     3.93128    1.76562       88
[367]   XVI  870371-870586      * | Reb1_R1_peak_367     292     16.43930   32.08000   29.23800      161
[368]   XVI  899847-900090      * | Reb1_R1_peak_368    1279     45.43150  131.80100  127.92500      175
[369]   XVI  942584-942868      * | Reb1_R1_peak_369     162     10.95950   18.90550   16.25210      111
-----
seqinfo: 17 sequences from an unspecified genome; no seqlengths
```

# GRanges

## Getter functions

- `seqnames()` → chromosome
- `ranges()` → location
- `strand()`
- `start()`
- `end()`
- `width()`
- `mcols(...)$...` → metadata
- `...$...` → metadata

2020/01/13

Jacques Serizay

```
> peaks
GRanges object with 369 ranges and 6 metadata columns:
      seqnames      ranges strand |      name      score signalValue      pValue      qValue      peak
      <Rle>      <IRanges> <Rle> | <character> <numeric> <numeric> <numeric> <numeric> <integer>
[1]      I      102-492      * | Reb1_R1_peak_1      81      6.32656      10.6179      8.16077      28
[2]      I    92560-92807      * | Reb1_R1_peak_2     118      8.90459      14.3647     11.80700     105
[3]     II     5846-6092      * | Reb1_R1_peak_3      63      6.16472      8.7750      6.36486      51
[4]     II 111226-111389      * | Reb1_R1_peak_4    1714     63.70210     175.6310    171.48900      76
[5]     II 124859-125004      * | Reb1_R1_peak_5     397     20.54910     42.7364     39.77400      99
...     ...      ...      ... | ...      ...      ...      ...      ...
[365]    XVI 840491-840698      * | Reb1_R1_peak_365     63      6.13093      8.80750      6.39684      98
[366]    XVI 844287-844438      * | Reb1_R1_peak_366     17      3.42484      3.93128      1.76562      88
[367]    XVI 870371-870586      * | Reb1_R1_peak_367    292     16.43930     32.08000     29.23800     161
[368]    XVI 899847-900090      * | Reb1_R1_peak_368   1279     45.43150    131.80100    127.92500     175
[369]    XVI 942584-942868      * | Reb1_R1_peak_369    162     10.95950     18.90550     16.25210     111
-----
seqinfo: 17 sequences from an unspecified genome; no seqlengths
> seqnames(peaks)
factor-Rle of length 369 with 17 runs
Lengths:  2  19  13  39  13   6  25  12  37   9  17  23  26  35  28  33  32
Values : I  II III IV  IX MT  V   VI  VII VIII X   XI XII XIII XIV XV  XVI
Levels(17): I II III IV IX MT V VI VII VIII X XI XII XIII XIV XV XVI
> ranges(peaks)
IRanges object with 369 ranges and 0 metadata columns:
      start      end      width
      <integer> <integer> <integer>
[1]      102      492      391
[2]    92560    92807      248
[3]     5846     6092      247
[4]    111226    111389      164
[5]    124859    125004      146
...     ...      ...      ...
[365]   840491   840698      208
[366]   844287   844438      152
[367]   870371   870586      216
[368]   899847   900090      244
[369]   942584   942868      285
> strand(peaks)
factor-Rle of length 369 with 1 run
Lengths: 369
Values : *
Levels(3): + - *
> mcols(peaks)$score
[1]  81  118  63 1714  397  452  508  76  554  132  603  13  79  28  701  415  642  90  474  84 2934  53
[48]  51  35  193  309  995  416  36  191  584  519  14  76  366  904  162  193  863  470 1430  826  341  258
[95] 289  690  229  591  147  379 1807  125  73  824  676  262  198  31  356  156  252  741  103  296  514  173
[142] 694  422  434  132  36  255  61  314  49  555  87  623  57  185  367 1537  235  146  130  341  258 1303
[189]1287  672  193  68  77  47 1187  681  140  397  28 1625  26 1026  285  36  820  394  28  701  742  177
[236] 138  118 2258  326  408 2001  605 1138  50  434  120  46  28  238  141  207  370  124  502  931 1131  27
[283] 172  103  147  177  73 1154  367  359  701 2737  545  642  913 1093  177  87  423  155 1231 1200  566  603
[330] 278  292  476  211 1865  884  863  944  718  900  571  128  326  781  209  560  39  28 2640  760  147  802
```

# GRanges

## Genome information

- `seqinfo()`
- `seqlevelsStyle()`

```
Seqinfo object with 17 sequences from an unspecified genome; no seqlengths:
  seqnames seqlengths isCircular genome
I          <NA>       <NA>      <NA>
II         <NA>       <NA>      <NA>
III        <NA>       <NA>      <NA>
IV         <NA>       <NA>      <NA>
IX         <NA>       <NA>      <NA>
...        ...        ...        ...
XII        <NA>       <NA>      <NA>
XIII       <NA>       <NA>      <NA>
XIV        <NA>       <NA>      <NA>
XV         <NA>       <NA>      <NA>
XVI        <NA>       <NA>      <NA>
> seqlevelsStyle(peaks)
[1] "NCBI"
> seqlevelsStyle(peaks) <- "UCSC"
> seqinfo(peaks)
Seqinfo object with 17 sequences from an unspecified genome; no seqlengths:
  seqnames seqlengths isCircular genome
chrI       <NA>       <NA>      <NA>
chrII      <NA>       <NA>      <NA>
chrIII     <NA>       <NA>      <NA>
chrIV      <NA>       <NA>      <NA>
chrIX      <NA>       <NA>      <NA>
...        ...        ...        ...
chrXII     <NA>       <NA>      <NA>
chrXIII    <NA>       <NA>      <NA>
chrXIV     <NA>       <NA>      <NA>
chrXV      <NA>       <NA>      <NA>
chrXVI     <NA>       <NA>      <NA>
```

# GRanges

## Action functions

- ...[...] (to subset)
- shift()
- resize()
- reduce()
- coverage()
- ...

2020/01/13

Jacques Serizay

```
> peaks[2:6]
GRanges object with 5 ranges and 6 metadata columns:
      seqnames      ranges strand |           name      score signalValue      pValue      qValue      peak
      <Rle>      <IRanges> <Rle> | <character> <numeric> <numeric> <numeric> <numeric> <integer>
[1]   chrI    92560-92807      * | Reb1_R1_peak_2      118      8.90459      14.3647      11.80700      105
[2]   chrII     5846-6092      * | Reb1_R1_peak_3       63      6.16472       8.7750       6.36486       51
[3]   chrII 111226-111389      * | Reb1_R1_peak_4     1714     63.70210     175.6310     171.48900       76
[4]   chrII 124859-125004      * | Reb1_R1_peak_5       397     20.54910     42.7364     39.77400       99
[5]   chrII 135791-136046      * | Reb1_R1_peak_6       452     22.60400     48.2640     45.24710      132
-----
seqinfo: 17 sequences from an unspecified genome; no seqlengths
> shift(peaks[2:6])
GRanges object with 5 ranges and 6 metadata columns:
      seqnames      ranges strand |           name      score signalValue      pValue      qValue      peak
      <Rle>      <IRanges> <Rle> | <character> <numeric> <numeric> <numeric> <numeric> <integer>
[1]   chrI    92560-92807      * | Reb1_R1_peak_2      118      8.90459      14.3647      11.80700      105
[2]   chrII     5846-6092      * | Reb1_R1_peak_3       63      6.16472       8.7750       6.36486       51
[3]   chrII 111226-111389      * | Reb1_R1_peak_4     1714     63.70210     175.6310     171.48900       76
[4]   chrII 124859-125004      * | Reb1_R1_peak_5       397     20.54910     42.7364     39.77400       99
[5]   chrII 135791-136046      * | Reb1_R1_peak_6       452     22.60400     48.2640     45.24710      132
-----
seqinfo: 17 sequences from an unspecified genome; no seqlengths
> resize(peaks[2:6], 1, fix = 'center')
GRanges object with 5 ranges and 6 metadata columns:
      seqnames      ranges strand |           name      score signalValue      pValue      qValue      peak
      <Rle> <IRanges> <Rle> | <character> <numeric> <numeric> <numeric> <numeric> <integer>
[1]   chrI      92683      * | Reb1_R1_peak_2      118      8.90459      14.3647      11.80700      105
[2]   chrII       5969      * | Reb1_R1_peak_3       63      6.16472       8.7750       6.36486       51
[3]   chrII     111307      * | Reb1_R1_peak_4     1714     63.70210     175.6310     171.48900       76
[4]   chrII     124931      * | Reb1_R1_peak_5       397     20.54910     42.7364     39.77400       99
[5]   chrII     135918      * | Reb1_R1_peak_6       452     22.60400     48.2640     45.24710      132
-----
seqinfo: 17 sequences from an unspecified genome; no seqlengths
> reduce(peaks[2:6])
GRanges object with 5 ranges and 0 metadata columns:
      seqnames      ranges strand
      <Rle>      <IRanges> <Rle>
[1]   chrI    92560-92807      *
[2]   chrII     5846-6092      *
[3]   chrII 111226-111389      *
[4]   chrII 124859-125004      *
[5]   chrII 135791-136046      *
-----
seqinfo: 17 sequences from an unspecified genome; no seqlengths
```

# GRanges

## Comparison functions

- %over%
- distance()
- distanceToNearest()
- findOverlaps()
- subsetByOverlaps()

```
> peaks[1:5] %over% Reb1_hits
[1] TRUE FALSE TRUE TRUE TRUE

> distanceToNearest(peaks[1:5], Reb1_hits)
Hits object with 5 hits and 1 metadata column:
      queryHits subjectHits | distance
      <integer>  <integer> | <integer>
[1]           1         1673 |          0
[2]           2          427 |         5386
[3]           3         2081 |          0
[4]           4           14 |          0
[5]           5         1124 |          0
-----
queryLength: 5 / subjectLength: 3642

> findOverlaps(peaks, Reb1_hits)
Hits object with 446 hits and 0 metadata columns:
      queryHits subjectHits
      <integer>  <integer>
[1]           1          450
[2]           1          895
[3]           1         1620
[4]           1         1673
[5]           3          516
...           ...         ...
[442]         365          216
[443]         365         1803
[444]         367          288
[445]         368          197
[446]         369         2763
-----
queryLength: 369 / subjectLength: 3642
```



# GRanges

## Comparison functions

- %over%
- distance()
- distanceToNearest()
- findOverlaps()
- subsetByOverlaps()

```
> table(peaks %over% Reb1_hits)
```

```
FALSE TRUE  
   36   333
```

```
> subsetByOverlaps(peaks, Reb1_hits)
```

GRanges object with 333 ranges and 6 metadata columns:

	seqnames	ranges	strand	name	score	signalValue	pValue	qValue	peak
	<Rle>	<IRanges>	<Rle>	<character>	<numeric>	<numeric>	<numeric>	<numeric>	<integer>
[1]	I	102-492	*	Reb1_R1_peak_1	81	6.32656	10.6179	8.16077	28
[2]	II	5846-6092	*	Reb1_R1_peak_3	63	6.16472	8.7750	6.36486	51
[3]	II	111226-111389	*	Reb1_R1_peak_4	1714	63.70210	175.6310	171.48900	76
[4]	II	124859-125004	*	Reb1_R1_peak_5	397	20.54910	42.7364	39.77400	99
[5]	II	135791-136046	*	Reb1_R1_peak_6	452	22.60400	48.2640	45.24710	132
...	...	...	...	...	...	...	...	...	...
[329]	XVI	829041-829232	*	Reb1_R1_peak_364	147	10.27450	17.3633	14.74180	129
[330]	XVI	840491-840698	*	Reb1_R1_peak_365	63	6.13093	8.8075	6.39684	98
[331]	XVI	870371-870586	*	Reb1_R1_peak_367	292	16.43930	32.0800	29.23800	161
[332]	XVI	899847-900090	*	Reb1_R1_peak_368	1279	45.43150	131.8010	127.92500	175
[333]	XVI	942584-942868	*	Reb1_R1_peak_369	162	10.95950	18.9055	16.25210	111

-----

seqinfo: 17 sequences from an unspecified genome; no seqlengths



# Biostrings

## Biostrings in R

```
> seqs <- Biostrings::readDNAStringSet('Share/day03/results/bwa/mergedLibrary/mac/s/narrowPeak/Reb1_R1_peaks.narrowPeak.fa')
> seqs
DNAStringSet object of length 369:
      width seq
[1] 391 CCAACCTGTCTCTCAACTTACCCTCCATTACCCTGCCTCCACTCGTT...ATATACCATCTCAAACTTACCCTACTCTCAGATTCCACTTCACTCCA I:101-492
[2] 248 TACTGCTAAACTTCGAGATATTTTCGAATTTTTCAGTCTTTTCTTTT...CTAACTGTTACCTTTTGAAATAAAATAAGGGGAAGGTCAAAAAGCTA I:92559-92807
[3] 247 ATACCCTAACACTACCCTAACCTACCCTATTTCAACCCTTCCAACC...TTCACTACCACTTACCCTGCCATTACTCTACCATCCACCATCTGCTA II:5845-6092
[4] 164 TTCATCTCTTTGTAAATAGTGTTATACCATAGTAGTAGTTTCAATAA...GAACGGAAGGGGTTTAATAGTTGTATGCTTAACATATTTTCGATTAA II:111225-111389
[5] 146 AATCTCAGCTGAAAGGCTGCCTTTAATTGTTATTCTTTTCCAGGAAA...AATCTATTACCTCGGATTAACCTGAATTAATAAGGACACACAGGTAT II:124858-125004
...
[365] 208 ACTTACTGGTCTTTAGCACACGACGACCGTACTTTGCACGTGGCTGC...TTCAGACCCACACAAAATCCGCGTAGCCGAGATTGCTTATGTATGTT XVI:840490-840698
[366] 152 AAGGGGTATGTTCTCAGCATTATCTGAAGGTACTCCTCTAAATTTT...ATAATATCAGGTAAAGAAATTGTTGGAATAAAAATCCACTATCGTCT XVI:844286-844438
[367] 216 AGGAAAAAAGGAAAAAGCAAAAAATATCGATTTTATGACTTACAAA...TACCCGCATATTATCGGGAAACAGAAGCCATGTTAGAGTGATTTCCA XVI:870370-870586
[368] 244 TAGTCGTCGCAAGCGACAAATCTCAACTGACAGTAAATAACGGTGAT...TTCTTGTTCCACCTCTTTTCCCCAACATATATGAACATGAGATGGTA XVI:899846-900090
[369] 285 TGGGTGAATGGCACAGGGTATAGACCGCTGAGGCAAGTGCCGTGCAT...GAAGCGTGAGGTCTGATACCTAATAAGGAAATGTAATTTATACTTT XVI:942583-942868
```

# Biostrings

```
> seqs[2:5]
DNAStringSet object of length 4:
      width seq
[1] 248 TACTGCTAAACTTCGAGATATTTTCGAATTTTTCAGTCTTTTCTTTT...CCTAACTGTTACCTTTTGAAATAAAATAAGGGGAAGGTCAAAAAGCTA I:92559-92807
[2] 247 ATACCTAACAACCTACCCTAACCCTACCCTATTTCAACCCTTCCAACCT...CTTCACTACCACTTACCCTGCCATTACTCTACCATCCACCATCTGCTA II:5845-6092
[3] 164 TTCATCTCTTTGTAAATAGTGTTATACCATAGTAGTAGTTTCAATAAT...AGAACGGAAGGGGTTTAATAGTTGTATGCTTAACATATTTGATTAA II:111225-111389
[4] 146 AATCTCAGCTGAAAGGCTGCCTTTAATTGTTATTCTTTCCAGGAAAA...TAATCTATTACCTCGGATTAACCTGAATTAATAAGGACACACAGGTAT II:124858-125004

> reverse(seqs[2:4])
DNAStringSet object of length 3:
      width seq
[1] 248 ATCGAAAACTGGAAGGGGAATAAAATAAAGTTTTCCATTGTCAATCC...TTTTTCTTTTCTGACTTTTTTAAGCTTTTATAGAGCTTCAAATCGTCAT I:92559-92807
[2] 247 ATCGTCTACCACCTACCATCTCATTACCGTCCCATTCAACATCACTTC...TCCAACCTTCCCAACTTTATCCCATCCCAATCCCATCACAATCCCATA II:5845-6092
[3] 164 AATTAGCTTTATACAATTGATGTTGATAATTTGGGAAGGCAAGA...TAATAACTTTGATGATGATACCATATTGTGATAAATGTTTCTCTACTT II:111225-111389

> reverseComplement(seqs[2:4])
DNAStringSet object of length 3:
      width seq
[1] 248 TAGCTTTTTGACCTTCCCTTATTTTATTTCAAAGGTAACAGTTAGG...AAAAAGAAAAGACTGAAAAATTGAAAATATCTCGAAGTTTAGCAGTA I:92559-92807
[2] 247 TAGCAGATGGTGGATGGTAGAGTAATGGCAGGGTAAGTGGTAGTGAAG...AGGTTGGAAGGGTTGAAATAGGGTAGGGTTAGGGTAGTGTAGGGTAT II:5845-6092
[3] 164 TTAAATCGAAATATGTTAAGCATACAACCTATTAACCCCTTCCGTTCT...ATTATTGAAACTACTACTATGGTATAACACTATTTACAAAGAGATGAA II:111225-111389

> width(seqs[2:4])
[1] 248 247 164
> names(seqs[2:4])
[1] "I:92559-92807" "II:5845-6092" "II:111225-111389"
```