Advanced R

(with Tidyverse)

Simon Andrews, Laura Biggins

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Babraham Bioinformatics

Course Content

- Expanding knowledge
  - More functions and operators
- Improving efficiency
  - More options for elegant code
- Awkward cases
  - Dealing with real data
- Tidyverse operations
  - Data Import
  - Filtering, selecting and sorting
  - Restructuring data
  - Grouping and Summarising
  - Extending and Merging
- Custom functions

RStudio Projects

- Associated with a working directory
- Keep workspace, history, source documents in one location
- Avoids the need for setwd() commands
- Works with version control (git or subversion)

https://support.posit.co/hc/en-us/articles/200526207-Using-RStudio-Projects

Tidyverse Packages

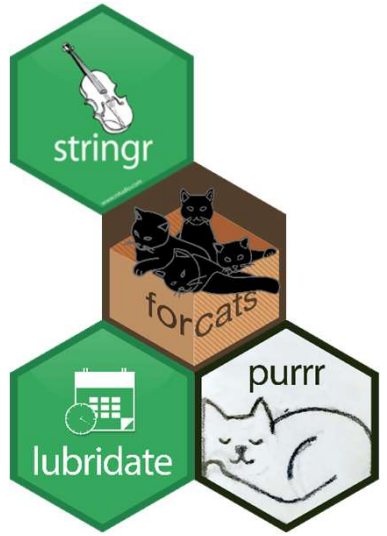
- tibble - data storage

- readR - reading data from files

- tidyR - Model data correctly

- dplyR - Manipulate and filter data

- ggplot2 - Draw figures and graphs



Reading Files with readr

- Tidyverse functions for reading text files into tibbles
  - `read_delim("file.txt")`
    `read_csv("file.csv")`
    `read_tsv("file.tsv")`
  - `read_fwf("file.txt",col_positions=c(1,3,6))`
  - `read_excel("file.xlsx", sheet="Sheet1")`

The `read_excel` function isn't in core tidyverse. You need to install the `readxl` package to get it

Reading files with readr


```
> trumpton <- read_delim("trumpton.txt")
Rows: 7 Columns: 5

-- Column specification --------------------------------
Delimiter: "\t"
chr (2): LastName, FirstName
dbl (3): Age, Weight, Height

i Use `spec()` to retrieve the full column specification for this data.
i Specify the column types or set `show_col_types = FALSE` to quiet this message.
```

```
> trumpton
  # A tibble: 7 x 5
    LastName FirstName   Age Weight Height
    <chr>    <chr>     <dbl>  <dbl>  <dbl>
  1 Hugh     Chris        26     90    175
  2 Pew      Adam         32    102    183
```

Reading files with readr


```
chr (2): LastName, FirstName
dbl (3): Age, Weight, Height
```

```
> spec(trumpton)
cols(
  LastName = col_character(),
  FirstName = col_character(),
  Age = col_double(),
  Weight = col_double(),
  Height = col_double()
)
```

Import Problems


```
> problem_data <- read_delim("import_problems.txt")
Rows: 1042 Columns: 4
-- Column specification --------------------------------------------------
```

Delimiter: "\t"
chr (2): Gene, Significance
dbl (2): Chr, Expression

i Use `spec()` to retrieve the full column specification for this data.
i Specify the column types or set `show_col_types = FALSE` to quiet this message.
Warning message:
One or more parsing issues, call `problems()` on your data frame for details, e.g.:
dat <- vroom(...)
problems(dat)

> head(problem_data)
A tibble: 6 × 4

	Chr	Gene	Expression	Significance
	<dbl>	<chr>	<dbl>	<chr>
1	1	Depdc2	9.19	NS
2	1	Sulf1	9.66	NS
3	1	Rpl7	8.75	0.050626416

Import Problems


```
> problems(problem_data)

# A tibble: 1 x 5
    row   col expected actual file
  <int> <int> <chr>    <chr>  <chr>
1  1042     1 a double Y      import_problems.txt
```

- Data not matching the guessed type
- Wrong number of fields

Fixing guessed columns


```
# A tibble: 1,174 x 4
     Chr Gene     Expression Significance
   <dbl> <chr>         <dbl> <chr>
 1     1 Depdc2         9.19 NS
 2     1 Sulf1          9.66 NS
 3     1 Rpl7           8.75 0.050626416
 4     1 Phf3           8.43 NS
 5     1 Khdrbs2        8.94 NS
 6     1 Prim2          9.64 NS
 7     1 Hs6st1         9.60 0.03441748
 8     1 BC050210       8.74 NS
 9     1 Tmem131        8.99 NS
10     1 Aff3          10.8  NS
```

- Chr should be character
- Significance should be numeric

Fixing guessed columns


```
read_delim(
  "import_problems.txt",
   col_types=cols(Chr=col_character(), Significance=col_double())
)

One or more parsing issues, see `problems()` for details
> problems()
# A tibble: 874 x 5
     row   col expected actual file
   <int> <int> <chr>    <chr>  <chr>
 1     2     4 a double NS     import_problems.txt
 2     3     4 a double NS     import_problems.txt
 3     5     4 a double NS     import_problems.txt
 4     6     4 a double NS     import_problems.txt
 5     7     4 a double NS     import_problems.txt
 6     9     4 a double NS     import_problems.txt
 7    10     4 a double NS     import_problems.txt
 8    11     4 a double NS     import_problems.txt
 9    13     4 a double NS     import_problems.txt
10    14     4 a double NS     import_problems.txt
# ... with 864 more rows
```

```
# A tibble: 1,174 x 4
   Chr   Gene     Expression Significance
   <chr> <chr>         <dbl>        <dbl>
 1 1     Depdc2         9.19       NA
 2 1     Sulf1          9.66       NA
 3 1     Rpl7           8.75        0.0506
 4 1     Phf3           8.43       NA
 5 1     Khdrbs2        8.94       NA
 6 1     Prim2          9.64       NA
 7 1     Hs6st1         9.60        0.0344
 8 1     BC050210       8.74       NA
 9 1     Tmem131        8.99       NA
10 1     Aff3          10.8        NA
# ... with 1,164 more rows
```

Unwanted header lines


```
read_delim("unwanted_headers.txt")
Error: Could not guess the delimiter.
```

```
read_csv("unwanted_headers.txt")

Delimiter: ","
chr (1): # Format version 1.0

> problems()
# A tibble: 4 x 5
    row   col expected  actual    file
  <int> <int> <chr>     <chr>     <chr>
1     3     5 1 columns 5 columns unwanted_headers.txt
2     4     5 1 columns 5 columns unwanted_headers.txt
3     5     5 1 columns 5 columns unwanted_headers.txt
4     6     5 1 columns 5 columns unwanted_headers.txt
```

```
# Format version 1.0
# Created 20/05/2020
Gene,Strand,Group_A,Group_B,Group_C
ABC1,+,5.30,4.69,4.84
DEF1,-,14.97,15.66,15.92
HIJ1,-,2.17,3.14,1.94
```

```
# A tibble: 5 x 1
  `# Format version 1.0`
  <chr>
1 # Created 20/05/2020
2 Gene
3 ABC1
4 DEF1
5 HIJ1
```

Unwanted header lines


```
read_delim(
  "unwanted_headers.txt",
  lazy=FALSE,
  skip=2
)
```

```
read_delim(
  "unwanted_headers.txt",
  lazy=FALSE,
  comment="#"
)
```

```
chr (2): Gene, Strand
dbl (3): Group_A, Group_B, Group_C
```

```
# Format version 1.0
# Created 20/05/2020
Gene,Strand,Group_A,Group_B,Group_C
ABC1,+,5.30,4.69,4.84
DEF1,-,14.97,15.66,15.92
HIJ1,-,2.17,3.14,1.94
```

```
# A tibble: 3 x 5
  Gene  Strand Group_A Group_B Group_C
  <chr> <chr>    <dbl>   <dbl>   <dbl>
1 ABC1  +         5.3     4.69    4.84
2 DEF1  -        15.0    15.7    15.9
3 HIJ1  -         2.17    3.14    1.94
```

Exercise 1
Reading Data into Tibbles

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Filtering, Selecting, Sorting etc.

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Subsetting and Filtering

- `select` pick columns by name/position
- `filter` pick rows based on the data

- `slice` pick rows by position
- `arrange` sort rows
- `distinct` deduplicate rows

Trumpton


```
# A tibble: 7 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Hugh     Chris        26     90    175
2 Pew      Adam         32    102    183
3 Barney   Daniel       18     88    168
4 McGrew   Chris        48     97    155
5 Cuthbert Carl         28     91    188
6 Dibble   Liam         35     94    145
7 Grub     Doug         31     89    164
```

Using `select` or `slice`


```
select(data,cols)
slice(data,rows)
```

```
trumpton %>%
    select(LastName,Age,Height)
```

```
# A tibble: 7 x 3
  LastName   Age Height
  <chr>    <dbl>  <dbl>
1 Hugh        26    175
2 Pew         32    183
3 Barney      18    168
4 McGrew      48    155
5 Cuthbert    28    188
6 Dibble      35    145
7 Grub        31    164
```

```
trumpton %>%
    slice(1,4,7)
```

```
# A tibble: 3 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Hugh     Chris        26     90    175
2 McGrew   Chris        48     97    155
3 Grub     Doug         31     89    164
```

Using `select` and `slice`


```
trumpton %>%
  select(LastName, Age, Height) %>%
  slice(1,4,7)
```

```
# A tibble: 3 x 3
  LastName   Age Height
  <chr>    <dbl>  <dbl>
1 Hugh        26    175
2 McGrew      48    155
3 Grub        31    164
```

Functional row selection using `filter`


```
trumpton %>%
  filter(Height>170)
```

```
# A tibble: 3 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Hugh     Chris        26     90    175
2 Pew      Adam         32    102    183
3 Cuthbert Carl         28     91    188
```

Defining Selected Columns

- Single definitions (name, position or function)

  Positive `weight, height, length, 1, 2, 3, last_col(), everything()`

  Negative `-chromosome, -start, -end, -1, -2, -3`

- Range selections

  `3:5` `-(3:5)` `height:length` `-(height:length)`

- Functional selections (positive or negative)

  `starts_with()` `-starts_with()`
  `ends_with()` `-ends_with()`
  `contains()` `-contains()`
  `matches()` `-matches()`

Using `select` helpers

```
colnames(child.variants)
```
```
CHR POS dbSNP REF ALT QUAL GENE ENST  MutantReads COVERAGE MutantReadPercent
```

```
child.variants %>%
 select(REF,COVERAGE)
```
```
REF COVERAGE
```

```
 select(REF,everything())
```
```
REF CHR POS dbSNP ALT QUAL GENE ENST  MutantReads COVERAGE MutantReadPercent
```

```
 select(-CHR, -ENST)
```
```
POS dbSNP REF ALT QUAL GENE MutantReads COVERAGE MutantReadPercent
```

```
 select(-REF,everything())
```
```
CHR POS dbSNP ALT QUAL GENE ENST  MutantReads COVERAGE MutantReadPercent REF
```

```
 select(5:last_col())
```
```
ALT QUAL GENE ENST MutantReads COVERAGE MutantReadPercent
```

```
 select(POS:GENE)
```
```
POS dbSNP REF ALT QUAL GENE
```

```
 select(-(POS:GENE))
```
```
CHR ENST MutantReads COVERAGE MutantReadPercent
```

```
 select(starts_with("Mut"))
```
```
MutantReads MutantReadPercent
```

```
 select(-ends_with("t",ignore.case = FALSE))
```
```
CHR POS dbSNP REF ALT QUAL GENE ENST MutantReads COVERAGE
```

```
 select(contains("Read"))
```
```
MutantReads MutantReadPercent
```

arrange (sorting)
distinct (deduplication)

```
trumpton %>%
  arrange(Height) %>%
  distinct(FirstName, .keep_all = TRUE)
```

```
# A tibble: 6 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Dibble   Liam         35     94    145
2 McGrew   Chris        48     97    155
3 Grub     Doug         31     89    164
4 Barney   Daniel       18     88    168
5 Pew      Adam         32    102    183
6 Cuthbert Carl         28     91    188
```

You need `.keep_all=TRUE` if you want to see more than the distinct column. "**keep_all**" has a dot before it

`arrange` (sorting)
`distinct` (deduplication)

```
trumpton %>%
  arrange(desc(Height)) %>%
  distinct(FirstName, .keep_all = TRUE)
```

```
# A tibble: 6 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Cuthbert Carl         28     91    188
2 Pew      Adam         32    102    183
3 Hugh     Chris        26     90    175
4 Barney   Daniel       18     88    168
5 Grub     Doug         31     89    164
6 Dibble   Liam         35     94    145
```

Exercise 2
Filtering and selecting

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More clever filtering

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Multi-condition `filter`


```
trumpton %>%
  filter(Height > 170) %>%
  filter(Age > 30)
```

```
# A tibble: 1 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Pew      Adam         32    102    183
```

Multi-condition `filter`


```
trumpton %>%
  filter(Height > 170 & Age > 30)
```

```
# A tibble: 1 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Pew      Adam         32    102    183
```

`&` = logical AND

`|` = logical OR

`!` = logical NOT

Multi-condition `filter`


```
trumpton %>%
  filter(Height > 170 | Age > 30)
```

&	= logical AND
|	= logical OR
!	= logical NOT

```
# A tibble: 6 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Hugh     Chris        26     90    175
2 Pew      Adam         32    102    183
3 McGrew   Chris        48     97    155
4 Cuthbert Carl         28     91    188
5 Dibble   Liam         35     94    145
6 Grub     Doug         31     89    164
```

Multi-condition `filter`


```
trumpton %>%
  filter(!(Height > 170 | Age > 30))
```

```
# A tibble: 1 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Barney   Daniel       18     88    168
```

& = logical AND

| = logical OR

! = logical NOT

Using `filter` with `%in%`


```
> hits
 [1] "FGFR1"  "RASAL1" "GLB1L2" "DNAH1"  "PTH1R"

child.variants %>%
  filter(GENE %in% hits)
```

```
# A tibble: 5 x 11
  CHR         POS dbSNP      REF   ALT    QUAL GENE   ENST          MutantReads COVERAGE MutantReadPerce~
  <chr>     <dbl> <chr>      <chr> <chr> <dbl> <chr>  <chr>               <dbl>    <dbl>            <dbl>
1 11    134226278 rs3802928  C     T       200 GLB1L2 ENST03898~             13       43               30
2 12    113539822 rs1674101  A     G       200 RASAL1 ENST05465~             19       22               86
3 3      46944274 rs1138518  T     C       200 PTH1R  ENST04495~             32       75               42
4 3      52430526 rs12163565 G     A       200 DNAH1  ENST04203~             38       50               76
5 8      38271182 .          TG    T       200 FGFR1  ENST04259~              9       31               29
```

Using `filter` with `str_detect`


```
child.variants %>%
  filter(str_detect(GENE,"ZFP"))
```

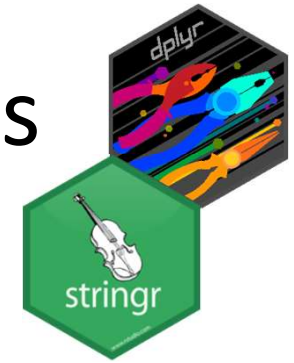
```
# A tibble: 9 x 11
  CHR         POS dbSNP     REF   ALT    QUAL GENE   ENST            MutantReads COVERAGE MutantReadPerce~
  <chr>     <dbl> <chr>     <chr> <chr> <dbl> <chr>  <chr>                 <dbl>    <dbl>            <dbl>
1 16     68598007 rs1177648 A     G       200 ZFP90  ENST0398253              43      100               43
2 16     88552370 rs3751673 A     G        53 ZFPM1  ENST0319555               4       23               17
3 18      5292030 rs620652  A     G       200 ZFP161 ENST0357006              28       71               39
4 19     57065189 rs145011  T     C       200 ZFP28  ENST0301318              59      137               43
5 20     50768672 .         GT    G       200 ZFP64  ENST0216923              36       41               87
6 5     180276402 rs168726  C     T       200 ZFP62  ENST0502412              74       83               89
7 8     106814656 rs2920048 G     C       200 ZFPM2  ENST0407775              33       79               41
8 8     144332012 rs6558339 T     C       200 ZFP41  ENST0330701              32       37               86
9 9     115818949 rs2282076 A     T       200 ZFP37  ENST0374227              18       43               41
```

Using `filter` with `str_detect`


```
child.variants %>%
  filter(str_detect(GENE,"Z.P"))
```

```
# A tibble: 15 x 11
   CHR         POS dbSNP      REF   ALT    QUAL GENE   ENST        MutantReads COVERAGE MutantReadPercent
   <chr>     <dbl> <chr>      <chr> <chr> <dbl> <chr>  <chr>             <dbl>    <dbl>              <dbl>
 1 7      99569394 rs17295356 G     A       200 AZGP1  ENST0292401           9       34                 26
 2 12     51636259 rs1049467  C     T       200 DAZAP2 ENST0549555          62       68                 91
 3 3     137786442 rs442800   T     C       200 DZIP1L ENST0327532           9       32                 28
 4 3     108403086 rs9856097  T     C       200 DZIP3  ENST0361582          26       30                 86
 5 20     56179586 rs6123710  G     A       200 ZBP1   ENST0371173          15       44                 34
 6 18      5292030 rs620652   A     G       200 ZFP161 ENST0357006          28       71                 39
 7 19     57065189 rs145011   T     C       200 ZFP28  ENST0301318          59      137                 43
 8 9     115818949 rs2282076  A     T       200 ZFP37  ENST0374227          18       43                 41
```

Using `filter` with other string operations




```
child %>%
  select(REF,ALT) %>%
  filter(startsWith(REF,"GAT"))
```

```
# A tibble: 3 x 2
  REF   ALT
  <chr> <chr>
1 GATA  G
2 GATAT GAT
3 GAT   G
```

```
child %>%
  select(GENE,ENST) %>%
  filter(endsWith(ENST,"878"))
```

```
# A tibble: 4 x 2
  GENE    ENST
  <chr>   <chr>
1 CIB3    ENST0269878
2 KCTD18  ENST0359878
3 KIAA1407 ENST0295878
4 RBM33   ENST0401878
```

These are different to the select helpers **starts_with** and **ends_with** which are used for picking columns

Using `filter` with `is` functions


```
> data.with.na
# A tibble: 8 x 2
  sample value
  <chr>  <dbl>
1 A       9.98
2 A       8.58
3 A      10.4
4 A      11.4
5 B       9.75
6 B      11.2
7 B      NA
8 B      NA
```

```
data.with.na %>%
  filter(!is.na(value))
```

```
# A tibble: 6 x 2
  sample value
  <chr>  <dbl>
1 A       9.98
2 A       8.58
3 A      10.4
4 A      11.4
5 B       9.75
6 B      11.2
```

`is_finite`
`is_infinite`
`is.nan`

Note that some functions have dots whilst others have an underscore.

Transforming data in a `filter`


```
trumpton %>%
  filter(log(Height)>5)
```

```
# A tibble: 6 x 5
  LastName FirstName   Age Weight Height
  <chr>    <chr>     <dbl>  <dbl>  <dbl>
1 Hugh     Chris        26     90    175
2 Pew      Adam         32    102    183
3 Barney   Daniel       18     88    168
4 McGrew   Chris        48     97    155
5 Cuthbert Carl         28     91    188
6 Grub     Doug         31     89    164
```

log
abs
sqrt
nchar
substr
tolower
toupper
etc.

Transforming `filter` examples


```
trumpton %>%
  filter(str_detect(tolower(LastName),"h"))

trumpton %>%
  filter(Weight*0.16 > 15)

trumpton %>%
  filter(nchar(LastName) == nchar(FirstName))
```

log
abs
nchar
str_sub
tolower
toupper
etc.

Exercise 3
More clever filtering

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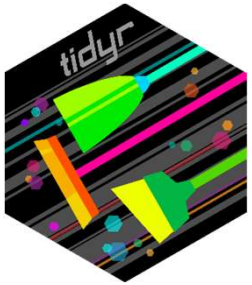

Restructuring Data

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'Tidy' Data Format

- Tibbles give you a 2D data structure where each column must be of a fixed data type
- Often data can be put into this sort of structure in more than one way
- Is there a right / wrong way to structure your data?

Journal of Statistical Software
August 2014, Volume 59, Issue 10. http://www.jstatsoft.org/

- Tidyverse has an opinion!

Tidy Data

Hadley Wickham
RStudio

Wide Format

Gene	WT_1	WT_2	WT_3	KO_1	KO_2	KO_3
ABC1	8.86	4.18	8.90	4.00	14.52	13.39
DEF1	29.60	41.22	36.15	11.18	16.68	1.64

- Compact
- Easy to read
- Shows linkage for genes

- No explicit genotype or replicate
- Values spread out over multiple rows and columns
- Not extensible to more metadata

Long Format

Gene	Genotype	Replicate	Value
ABC1	WT	1	8.86
ABC1	WT	2	4.18
ABC1	WT	3	8.90
ABC1	KO	1	4.00
ABC1	KO	2	14.52
ABC1	KO	3	13.39
DEF1	WT	1	29.60
DEF1	WT	2	41.22
DEF1	WT	3	36.15
DEF1	KO	1	11.18
DEF1	KO	2	16.68
DEF1	KO	3	1.64

- More verbose (repeated values)
- Explicit genotype and replicate
- All values in a single column
- Extensible to more metadata

Converting to "Tidy" format


```
# A tibble: 3 x 8
  Gene    Chr   Start     End  WT_1  WT_2  KO_1  KO_2
  <chr> <dbl>   <dbl>   <dbl> <dbl> <dbl> <dbl> <dbl>
1 Gnai3     2  163898  167465  9.39  10.9  33.5  81.9
2 Pbsn      5 4888573 4891351 91.7   59.6  45.3  82.3
3 Cdc45     7 1250084 1262669 69.2   36.1  54.4  38.1
```

- Put all measures into a single column
- Add a 'genotype' and 'replicate' column
- Duplicate the gene information as required
  - Or separate it into a different table

Converting to "Tidy" format


```
# A tibble: 3 x 8
  Gene    Chr   Start     End  WT_1  WT_2  KO_1  KO_2
  <chr> <dbl>   <dbl>   <dbl> <dbl> <dbl> <dbl> <dbl>
1 Gnai3     2  163898  167465  9.39  10.9  33.5  81.9
2 Pbsn      5 4888573 4891351 91.7   59.6  45.3  82.3
3 Cdc45     7 1250084 1262669 69.2   36.1  54.4  38.1
```

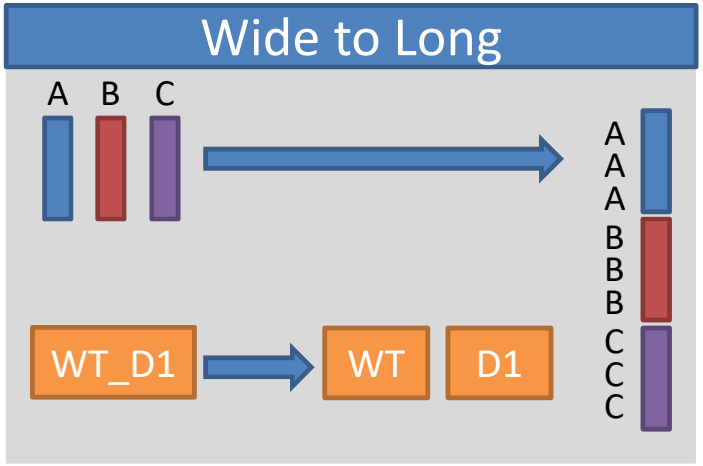
```
non.normalised %>%
 pivot_longer(cols=WT_1:KO_2, names_to="sample", values_to="value") %>%
 separate(sample,into=c("genotype","replicate"),convert = TRUE,sep="_")
```

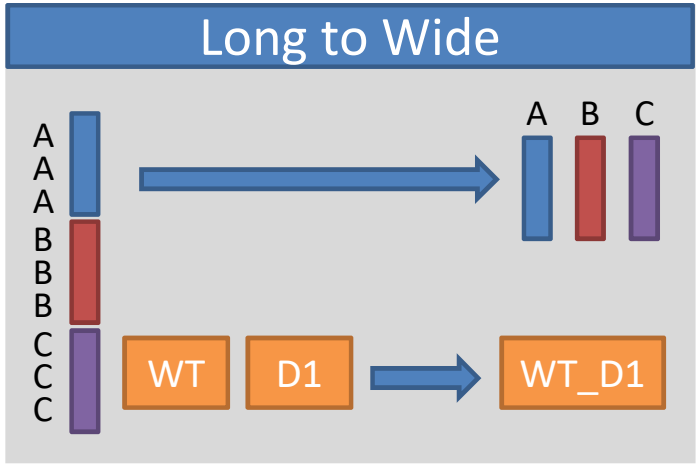
Converting to "Tidy" format


```
# A tibble: 12 x 7
   Gene    Chr   Start     End genotype replicate value
   <chr> <dbl>   <dbl>   <dbl> <chr>         <int> <dbl>
 1 Gnai3     2  163898  167465 WT                1  9.39
 2 Pbsn      5 4888573 4891351 WT                1 91.7
 3 Cdc45     7 1250084 1262669 WT                1 69.2
 4 Gnai3     2  163898  167465 WT                2 10.9
 5 Pbsn      5 4888573 4891351 WT                2 59.6
 6 Cdc45     7 1250084 1262669 WT                2 36.1
 7 Gnai3     2  163898  167465 KO                1 33.5
 8 Pbsn      5 4888573 4891351 KO                1 45.3
 9 Cdc45     7 1250084 1262669 KO                1 54.4
10 Gnai3     2  163898  167465 KO                2 81.9
11 Pbsn      5 4888573 4891351 KO                2 82.3
12 Cdc45     7 1250084 1262669 KO                2 38.1
```

Tidying operations

- `pivot_longer`
  - Takes multiple columns of the same type and puts them into a pair of key-value columns
- `separate`
  - Splits a delimited column into multiple columns
- `pivot_wider`
  - Takes a key-value column pair and spreads them out to multiple columns of the same type
- `unite`
  - Combines multiple columns into one





Converting to "Tidy" format


```
non.normalised %>%

  pivot_longer(
    cols=WT_1:KO_2,
    names_to="sample",
    values_to="value"
  ) %>%

  separate(
    col=sample,
    into=c("genotype","replicate"),
    sep="_",
    convert = TRUE
  )
```

```
# A tibble: 3 x 8
  Gene    Chr   Start     End  WT_1  WT_2  KO_1  KO_2
  <chr> <dbl>   <dbl>   <dbl> <dbl> <dbl> <dbl> <dbl>
1 Gnai3     2  163898  167465  9.39  10.9  33.5  81.9
2 Pbsn      5 4888573 4891351 91.7   59.6  45.3  82.3
3 Cdc45     7 1250084 1262669 69.2   36.1  54.4  38.1
```

`convert=TRUE` makes separate re-detect the type of the column, so replicate becomes a numeric value

Pivoting Examples


```
> pivot.data
# A tibble: 4 x 3
  gene     WT    KO
  <chr> <dbl> <dbl>
1 ABC1  18608  7831
2 DEF1  31988 55502
3 GHI1   7647 93299
4 JKL1  96002 47945
```

- Log transform all of the values
- Pivot longer
  - Which columns are we pivoting?
  - What do we want to call the new column of names?
  - What do we want to call the new column of values?

```
pivot.data %>%
  pivot_longer(
    cols=WT:KO,
    names_to = "Condition",
    values_to = "Count"
  ) -> pivot.long
```

```
# A tibble: 8 x 3
  gene  Condition Count
  <chr> <chr>     <dbl>
1 ABC1  WT        18608
2 ABC1  KO         7831
3 DEF1  WT        31988
4 DEF1  KO        55502
5 GHI1  WT         7647
6 GHI1  KO        93299
7 JKL1  WT        96002
8 JKL1  KO        47945
```

Pivoting Examples


```
> pivot.long
# A tibble: 8 x 3
  gene  Condition Count
  <chr> <chr>     <dbl>
1 ABC1  WT         14.2
2 ABC1  KO         12.9
3 DEF1  WT         15.0
4 DEF1  KO         15.8
5 GHI1  WT         12.9
6 GHI1  KO         16.5
7 JKL1  WT         16.6
8 JKL1  KO         15.5
```

- Plot WT vs KO
- Pivot wider
  - Which column of names?
  - Which column of values?

```
pivot.long %>%
  pivot_wider(
    names_from = Condition,
    values_from = Count
  )
```

```
# A tibble: 4 x 3
  gene     WT    KO
  <chr> <dbl> <dbl>
1 ABC1   14.2  12.9
2 DEF1   15.0  15.8
3 GHI1   12.9  16.5
4 JKL1   16.6  15.5
```

Splitting into multiple tibbles

Where you have a lot of annotation, you can split this into a separate tibble to reduce the amount of data duplication

```
# A tibble: 3 x 8
  Gene    Chr   Start     End  WT_1  WT_2  KO_1  KO_2
  <chr> <dbl>   <dbl>   <dbl> <dbl> <dbl> <dbl> <dbl>
1 Gnai3     2  163898  167465  9.39  10.9  33.5  81.9
2 Pbsn      5 4888573 4891351 91.7   59.6  45.3  82.3
3 Cdc45     7 1250084 1262669 69.2   36.1  54.4  38.1
```

Splitting into multiple tibbles

Annotation tibble

```
# A tibble: 3 x 4
  Gene    Chr   Start     End
  <chr> <dbl>   <dbl>   <dbl>
1 Gnai3     2  163898  167465
2 Pbsn      5 4888573 4891351
3 Cdc45     7 1250084 1262669
```

These can be recombined later on as needed using a join operation

Data tibble

```
# A tibble: 12 x 4
   Gene  genotype replicate value
   <chr> <chr>         <int> <dbl>
 1 Gnai3 WT                1  9.39
 2 Pbsn  WT                1 91.7
 3 Cdc45 WT                1 69.2
 4 Gnai3 WT                2 10.9
 5 Pbsn  WT                2 59.6
 6 Cdc45 WT                2 36.1
 7 Gnai3 KO                1 33.5
 8 Pbsn  KO                1 45.3
 9 Cdc45 KO                1 54.4
10 Gnai3 KO                2 81.9
11 Pbsn  KO                2 82.3
12 Cdc45 KO                2 38.1
```

Exercise 4
Restructuring data into ‘tidy’ format

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Adding or creating new data

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Adding or creating data

- `add_row` adds a single new row
- `add_column` adds a column of new data
- `mutate` create a new column from existing columns

Adding new rows or columns


```
trumpton %>%
  add_row(
    FirstName="Simon",
    LastName="Andrews",
    Age=39,
    Weight=80,
    Height=185
  )


trumpton %>%
  add_column(
    vegetarian = c(T,F,F,T,F,F,T)
  )
```

Creating columns with `mutate`


```
trumpton %>%
  mutate(
    weight_stones=Weight*0.16,
    height_feet=Height*0.033
  )
```

```
# A tibble: 7 x 7
  LastName FirstName   Age Weight Height weight_stones height_feet
  <chr>    <chr>     <dbl>  <dbl>  <dbl>         <dbl>       <dbl>
1 Hugh     Chris        26     90    175          14.4        5.78
2 Pew      Adam         32    102    183          16.3        6.04
3 Barney   Daniel       18     88    168          14.1        5.54
4 McGrew   Chris        48     97    155          15.5        5.12
5 Cuthbert Carl         28     91    188          14.6        6.20
6 Dibble   Liam         35     94    145          15.0        4.78
7 Grub     Doug         31     89    164          14.2        5.41
```

Tricks with `mutate` – Creating categories


```
trumpton %>%
  mutate(Category=if_else(Height > 180, "Tall", "Short"))
```

```
# A tibble: 7 x 6
  LastName FirstName   Age Weight Height Category
  <chr>    <chr>     <dbl>  <dbl>  <dbl> <chr>
1 Hugh     Chris        26     90    175 Short
2 Pew      Adam         32    102    183 Tall
3 Barney   Daniel       18     88    168 Short
4 McGrew   Chris        48     97    155 Short
5 Cuthbert Carl         28     91    188 Tall
6 Dibble   Liam         35     94    145 Short
7 Grub     Doug         31     89    164 Short
```

More than 2 categories


```
trumpton %>%
  mutate(
    Category = case_when(
      Height > 180 ~ "Tall",
      Height > 160 ~ "Medium",
      TRUE ~ "Short"
    )
  )
```

The first condition to match is used.

Good to have TRUE as the last test to catch anything which hasn't yet matched.

```
# A tibble: 7 × 6
  LastName FirstName   Age Weight Height Category
  <chr>    <chr>     <dbl>  <dbl>  <dbl> <chr>
1 Hugh     Chris        26     90    175 Medium
2 Pew      Adam         32    102    183 Tall
3 Barney   Daniel       18     88    168 Medium
4 McGrew   Chris        48     97    155 Short
5 Cuthbert Carl         28     91    188 Tall
6 Dibble   Liam         35     94    145 Short
7 Grub     Doug         31     89    164 Medium
```

Tricks with `mutate` – replacing values


```
data.with.na %>%
  mutate(value = replace(value,value>10, 10))
```

```
> data.with.na
# A tibble: 8 x 2
  sample value
  <chr>  <dbl>
1 A       9.98
2 A       8.58
3 A      10.4
4 A      11.4
5 B       9.75
6 B      11.2
7 B      NA
8 B      NA
```

```
# A tibble: 8 x 2
  sample value
  <chr>  <dbl>
1 A       9.98
2 A       8.58
3 A      10
4 A      10
5 B       9.75
6 B      10
7 B      NA
8 B      NA
```

```
data.with.na %>%
  mutate(value = replace_na(value,0))
```

```
# A tibble: 8 x 2
  sample value
  <chr>  <dbl>
1 A       9.98
2 A       8.58
3 A      10.4
4 A      11.4
5 B       9.75
6 B      11.2
7 B      0
8 B      0
```

Exercise 5
Adding or creating new data

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Grouping and Summarising

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Grouping and Summarising

- `group_by` sets groups for summarisation
- `ungroup` removes grouping information

- `summarise` collapse grouped variables
- `count` count grouped variables

Grouping and Summarising Workflow

1. Load a tibble with repeated values in one or more columns

2. Use `group_by` to select all of the categorical columns you want to combine to define your groups

3. Run `summarise` saying how you want to combine the quantitative values or `count` to see how many values are in each group

4. Run `ungroup` to remove any remaining group information

Grouping and Summarising Workflow

1. Load a tibble with repeated values in one or more columns

2. Use `group_by` to select all of the categorical columns you want to combine to define your groups

3. Run `summarise` saying how you want to combine the quantitative values or `count` to see how many values are in each group

4. Run `ungroup` to remove any remaining group information

Grouping and Summarising


```
> group.data

# A tibble: 8 x 5
  Sample Genotype Sex   Height Length
   <dbl> <chr>    <chr>  <dbl>  <dbl>
1      1 WT       M         15    200
2      2 WT       F         13    185
3      3 WT       F         14    221
4      4 WT       M         18    265
5      5 KO       M         26    120
6      6 KO       F         22    165
7      7 KO       F         19    143
8      8 KO       M         27    110
```

- Want to get the average Height and Length for each combination of sex and genotype

Grouping and Summarising Workflow

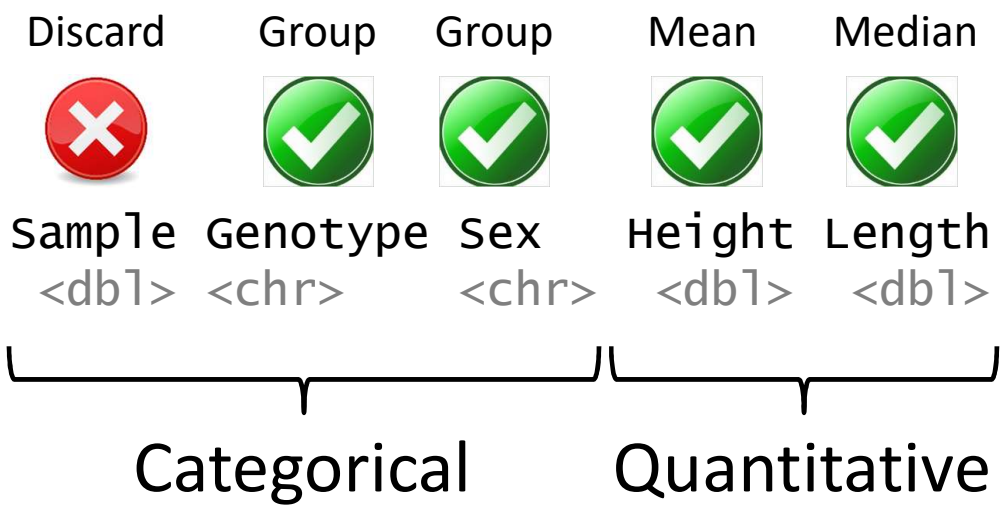
1. Load a tibble with repeated values in one or more columns

2. Use `group_by` to select all of the categorical columns you want to combine to define your groups

3. Run `summarise` saying how you want to combine the quantitative values or `count` to see how many values are in each group

4. Run `ungroup` to remove any remaining group information

Grouping and Summarising



- Want to get the average Height and Length for each combination of sex and genotype

Grouping and Summarising


```
group.data %>% group_by(Genotype,Sex)
```

```
# A tibble: 8 x 5
# Groups:   Genotype, Sex [4]
  Sample Genotype Sex   Height Length
   <dbl> <chr>    <chr>  <dbl>  <dbl>
1      1 WT       M         15    200
4      4 WT       M         18    265
2      2 WT       F         13    185
3      3 WT       F         14    221
5      5 KO       M         26    120
8      8 KO       M         27    110
6      6 KO       F         22    165
7      7 KO       F         19    143
```

Grouping and Summarising Workflow

1. Load a tibble with repeated values in one or more columns

2. Use `group_by` to select all of the categorical columns you want to combine to define your groups

3. Run `summarise` saying how you want to combine the quantitative values or `count` to see how many values are in each group

4. Run `ungroup` to remove any remaining group information

Grouping and Summarising


```
group.data %>%
  group_by(Genotype,Sex) %>%
  count()
```

```
# A tibble: 4 x 3
# Groups:   Genotype, Sex [4]
  Genotype Sex       n
  <chr>    <chr> <int>
1 KO       F         2
2 KO       M         2
3 WT       F         2
4 WT       M         2
```

Grouping and Summarising


```
group.data %>%
  group_by(Genotype,Sex) %>%
  summarise(
    Height2=mean(Height),
    Length=median(Length)
  )
```

```
# A tibble: 4 x 4
# Groups:   Genotype [2]
  Genotype Sex   Height2 Length
  <chr>    <chr>   <dbl>  <dbl>
1 KO       F        20.5   154
2 KO       M        26.5   115
3 WT       F        13.5   203
4 WT       M        16.5   232.
```

If you want the count of values as part of a summarised result use the n() function

Grouping and Summarising Workflow

1. Load a tibble with repeated values in one or more columns

2. Use `group_by` to select all of the categorical columns you want to combine to define your groups

3. Run `summarise` saying how you want to combine the quantitative values

4. Run `ungroup` to remove any remaining group information

Ungrouping

- A summarise operation removes the the last level of grouping ("Sex" in our worked example)
- Other levels of grouping ("Genotype") remain annotated on the data, so you could do an additional summarisation if needed
- If you're not going to use them it's a good idea to use `ungroup` to remove remaining groups so they don't interfere with other operations

Grouping affects lots of operations

Find the tallest member of each Sex


```
group.data %>%
  arrange(desc(Height)) %>%
  group_by(Sex) %>%
  slice(1)
```

```
# A tibble: 2 x 5
# Groups:   Sex [2]
  Sample Genotype Sex   Height Length
   <dbl> <chr>    <chr>  <dbl>  <dbl>
1      6 KO       F         22    165
2      8 KO       M         27    110
```

Grouping affects lots of operations

Normalise by mean centring the Length values


```
group.data %>%
  mutate(Diff=Length - mean(Length))
```

```
# A tibble: 8 x 6
  Sample Genotype Sex   Height Length  Diff
   <dbl> <chr>    <chr>  <dbl>  <dbl> <dbl>
1      1 WT       M         15    200  23.9
2      2 WT       F         13    185  8.88
3      3 WT       F         14    221  44.9
4      4 WT       M         18    265  88.9
5      5 KO       M         26    120 -56.1
6      6 KO       F         22    165 -11.1
7      7 KO       F         19    143 -33.1
8      8 KO       M         27    110 -66.1
```

```
group.data %>%
  group_by(Genotype) %>%
  mutate(Diff=Length - mean(Length))
```

```
# A tibble: 8 x 6
# Groups:   Genotype [2]
  Sample Genotype Sex   Height Length   Diff
   <dbl> <chr>    <chr>  <dbl>  <dbl>  <dbl>
1      1 WT       M         15    200 -17.8
2      2 WT       F         13    185 -32.8
3      3 WT       F         14    221   3.25
4      4 WT       M         18    265  47.2
5      5 KO       M         26    120 -14.5
6      6 KO       F         22    165  30.5
7      7 KO       F         19    143   8.5
8      8 KO       M         27    110 -24.5
```

Exercise 6
Grouping and Summarising

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Joining Tibbles

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Simple joins

- `bind_rows` join tibbles by row
- `bind_cols` join tibbles by column
- `rename` rename a column

```
trumpton %>%
  rename(Surname=LastName)
```

Complex joins for tibbles x and y

- `left_join` join matching values from y into x
- `right_join` join matching values of x into y
- `inner_join` join x and y keeping only rows in both
- `full_join` join x and y keeping all values in both

Join types

```
> join1
  name   count
1 Simon      3
2 Laura      6
3 Felix      2
```

```
> join2
  name    percentage
1 Felix           10
2 Anne            25
3 Simon           36
```

left_join(join1,join2)

```
  name   count percentage
1 Simon      3         36
2 Laura      6         NA
3 Felix      2         10
```

right_join(join1,join2)

```
  name   count percentage
1 Felix      2         10
2 Anne      NA         25
3 Simon      3         36
```

inner_join(join1,join2)

```
  name   count percentage
1 Simon      3         36
2 Felix      2         10
```

full_join(join1,join2)

```
  name   count percentage
1 Simon      3         36
2 Laura      6         NA
3 Felix      2         10
4 Anne      NA         25
```

Rejoining split tables

Find the highest value for each genotype


```
> gathered.data
# A tibble: 12 x 4
   Gene  genotype replicate value
   <chr> <chr>        <int> <dbl>
 1 Gnai3 WT               1  9.39
 2 Pbsn  WT               1 91.7
 3 Cdc45 WT               1 69.2
 4 Gnai3 WT               2 10.9
 5 Pbsn  WT               2 59.6
 6 Cdc45 WT               2 36.1
 7 Gnai3 KO               1 33.5
 8 Pbsn  KO               1 45.3
 9 Cdc45 KO               1 54.4
10 Gnai3 KO               2 81.9
11 Pbsn  KO               2 82.3
12 Cdc45 KO               2 38.1
```

```
> gathered.annotation
# A tibble: 3 x 4
  Gene    Chr   Start     End
  <chr> <dbl>   <dbl>   <dbl>
1 Gnai3     2  163898  167465
2 Pbsn      5 4888573 4891351
3 Cdc45     7 1250084 1262669
```

Rejoining split tables

Find the highest value for each genotype


```
gathered.data %>%
  arrange(desc(value)) %>%
  group_by(genotype) %>%
  slice(1) %>%
  ungroup()
```

```
# A tibble: 2 x 4
  Gene  genotype replicate value
  <chr> <chr>        <int> <dbl>
1 Pbsn  KO               2  82.3
2 Pbsn  WT               1  91.7
```

```
gathered.data %>%
  arrange(desc(value)) %>%
  group_by(genotype) %>%
  slice(1) %>%
  ungroup() %>%
  left_join(gathered.annotation)
```

```
# A tibble: 2 x 7
  Gene  genotype replicate value   Chr   Start     End
  <chr> <chr>        <int> <dbl> <dbl>   <dbl>   <dbl>
1 Pbsn  KO               2  82.3     5 4888573 4891351
2 Pbsn  WT               1  91.7     5 4888573 4891351
```

Exercise 7
Joining Tibbles

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Custom Functions

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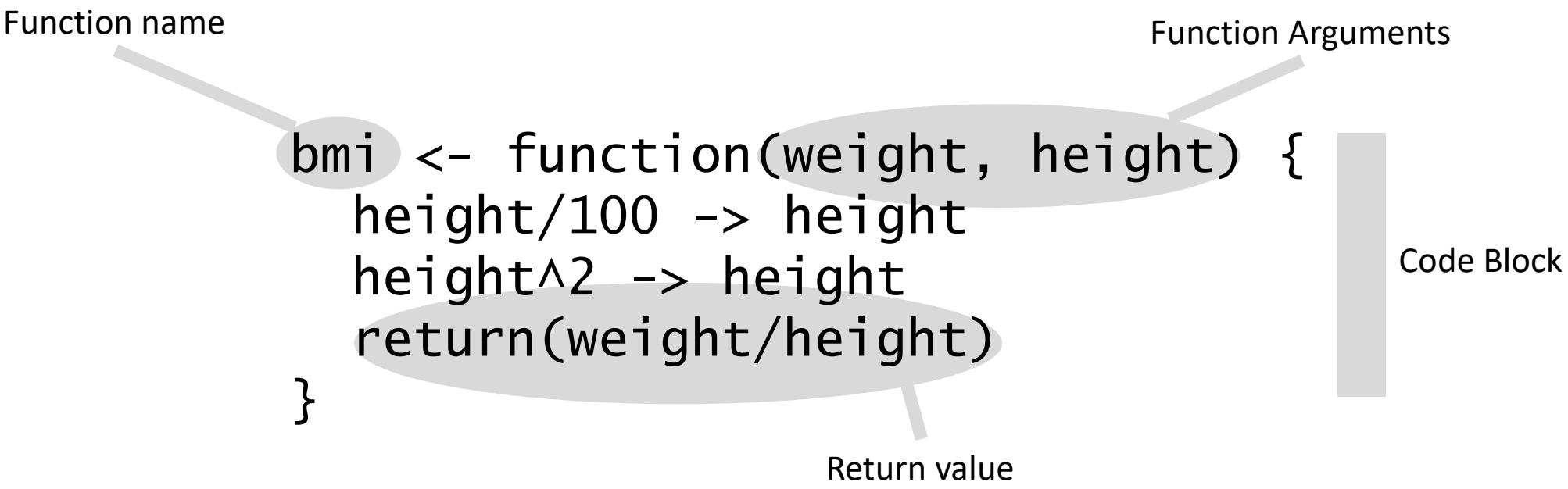
Custom Functions




```
> bmi(90,175)
[1] 29.38776
```

```
> bmi(c(90,102), c(175,183))
[1] 29.38776 30.45776
```

Custom function with mutate

```
trumpton %>% mutate(bmi=Weight/(Height/100)^2)
```

```
trumpton %>%
    mutate(bmi=calc_bmi(Weight,Height))
```

```
calc_bmi <- function(w, h) {
  h <- h/100
  h = h^2
  return(w/h)
}
```

Custom Functions with Tidyverse

```
summarise.gene <- function(tbl, genename="NANOG") {
  tbl %>%
    filter(GENE==genename) %>%
    filter(str_length(REF) == 1, str_length(ALT) == 1) %>%
    group_by(REF, ALT) %>%
    count() %>%
    ungroup() %>%
    return()
}
```

```
child %>%
  summarise.gene("PLEC")
```

```
# A tibble: 6 x 3
  REF   ALT       n
  <chr> <chr> <int>
1 A     C         1
2 A     G         9
3 C     T         6
4 G     A         8
5 T     C         6
6 T     G         1
```

Custom Functions with Tidyverse

```
count_mutations <- function(tbl, col=REF) {
  tbl %>%
    group_by({{ col }}) %>%
    count() %>%
    ungroup() %>%
    arrange(desc(n)) %>%
    return()
}
```

```
child %>%
  count_mutations(col=ALT)
```

```
# A tibble: 257 x 2
   ALT       n
   <chr> <int>
 1 C      6480
 2 G      6404
 3 A      6275
 4 T      6103
 5 GA       43
 6 TA       37
 7 GC       33
 8 AG       24
 9 CT       22
10 CA       20
# ... with 247 more rows
```

Exercise 8
Custom Functions

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Import Problems


```
> problem_data <- read_delim("import_problems.txt")
Rows: 1042 Columns: 4
-- Column specification --------------------------------------------------------
Delimiter: "\t"
chr (2): Gene, Significance
dbl (2): Chr, Expression

> head(problem_data)
    Chr Gene    Expression Significance
  <dbl> <chr>        3     1 Rpl7          8.75 0. <dbl> <chr>
1     1 Depdc2        9.19 NS
050626416

> tail(problem_data)
    Chr Gene          Expression Significance
  <dbl> <chr>               <dbl> <chr>
4     6 2610528E23Rik       9.55 NS
6    NA Huwe1               8.54 NS
Warning message:
One or more parsing issues, see `problems()` for details
```