# Analysis of variance

# **Packages**

```
library(tidyverse)
library(smmr)
library(PMCMRplus)
```

## Jumping rats

- Link between exercise and healthy bones (many studies).
- Exercise stresses bones and causes them to get stronger.
- Study (Purdue): effect of jumping on bone density of growing rats.
- ▶ 30 rats, randomly assigned to 1 of 3 treatments:
  - ► No jumping (control)
    - Low-jump treatment (30 cm)
  - ► High-jump treatment (60 cm)
- ▶ 8 weeks, 10 jumps/day, 5 days/week.
- ▶ Bone density of rats (mg/cm³) measured at end.

# Jumping rats 2/2

- See whether larger amount of exercise (jumping) went with higher bone density.
- Random assignment: rats in each group similar in all important ways.
- So entitled to draw conclusions about cause and effect.

## Reading the data

#### Values separated by spaces:

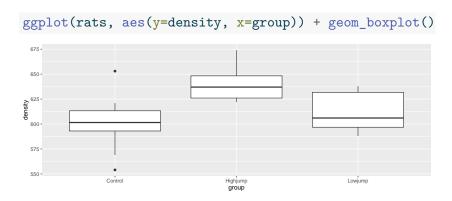
```
my_url <- "http://ritsokiguess.site/datafiles/jumping.txt"
rats <- read_delim(my_url," ")</pre>
```

# The data (some random rows)

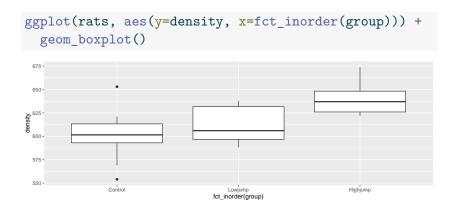
```
rats %>% slice_sample(n=12)
```

```
# A tibble: 12 x 2
            density
   group
   <chr>
              <dbl>
 1 Highjump
                643
 2 Lowjump
               599
 3 Control
               611
 4 Lowjump
               605
                635
 5 Lowjump
 6 Lowjump
                588
 7 Control
                621
                632
 8 Lowjump
 9 Lowjump
                596
10 Highjump
                650
11 Control
                554
                631
12 Lowjump
```

## Boxplots



# Or, arranging groups in data (logical) order



# Analysis of Variance

- ➤ Comparing > 2 groups of independent observations (each rat only does one amount of jumping).
- ► Standard procedure: analysis of variance (ANOVA).
- Null hypothesis: all groups have same mean.
- Alternative: "not all means the same", at least one is different from others.

## Testing: ANOVA in R

```
rats.aov <- aov(density~group,data=rats)
summary(rats.aov)</pre>
```

```
group 2 7434 3717 7.978 0.0019 **
Residuals 27 12579 466
---
Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 '
```

Df Sum Sq Mean Sq F value Pr(>F)

- Usual ANOVA table, small P-value: significant result.
- Conclude that the mean bone densities are not all equal.
- Reject null, but not very useful finding.

# Which groups are different from which?

- ▶ ANOVA really only answers half our questions: it says "there are differences", but doesn't tell us which groups different.
- One possibility (not the best): compare all possible pairs of groups, via two-sample t.
- First pick out each group:

```
rats %>% filter(group=="Control") -> controls
rats %>% filter(group=="Lowjump") -> lows
rats %>% filter(group=="Highjump") -> highs
```

#### Control vs. low

```
t.test(controls$density, lows$density)
```

```
Welch Two Sample t-test
```

```
data: controls$density and lows$density
t = -1.0761, df = 16.191, p-value = 0.2977
alternative hypothesis: true difference in means is not equ
95 percent confidence interval:
   -33.83725   11.03725
sample estimates:
mean of x mean of y
```

No sig. difference here.

601.1 612.5

## Control vs. high

```
t.test(controls$density, highs$density)
```

Welch Two Sample t-test

```
data: controls$density and highs$density
t = -3.7155, df = 14.831, p-value = 0.002109
alternative hypothesis: true difference in means is not equ
95 percent confidence interval:
   -59.19139 -16.00861
sample estimates:
mean of x mean of y
```

These are different.

601.1 638.7

## Low vs. high

```
t.test(lows$density, highs$density)
```

Welch Two Sample t-test

```
data: lows$density and highs$density
t = -3.2523, df = 17.597, p-value = 0.004525
alternative hypothesis: true difference in means is not equ
95 percent confidence interval:
  -43.15242  -9.24758
sample estimates:
mean of x mean of y
```

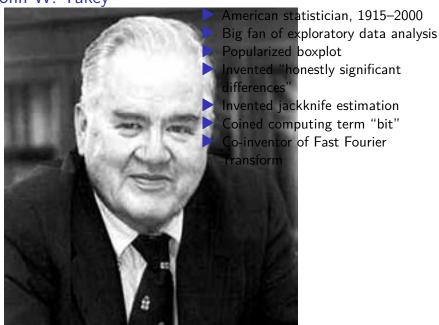
These are different too.

612.5 638.7

But...

- We just did 3 tests instead of 1.
- $\blacktriangleright$  So we have given ourselves 3 chances to reject  $H_0$  : all means equal, instead of 1.
- Thus  $\alpha$  for this combined test is not 0.05.

John W. Tukey



# Honestly Significant Differences

- Compare several groups with one test, telling you which groups differ from which.
- ▶ Idea: if all population means equal, find distribution of highest sample mean minus lowest sample mean.
- Any means unusually different compared to that declared significantly different.

#### Tukey on rat data

```
rats.aov <- aov(density~group, data = rats)
TukeyHSD(rats.aov)

Tukey multiple comparisons of means
   95% family-wise confidence level

Fit: aov(formula = density ~ group, data = rats)

$group

diff lwr upr p adj</pre>
```

Again conclude that bone density for highjump group significantly higher than for other two groups.

Highjump-Control 37.6 13.66604 61.533957 0.0016388 Lowjump-Control 11.4 -12.53396 35.333957 0.4744032 Lowjump-Highjump -26.2 -50.13396 -2.266043 0.0297843

# Why Tukey's procedure better than all t-tests

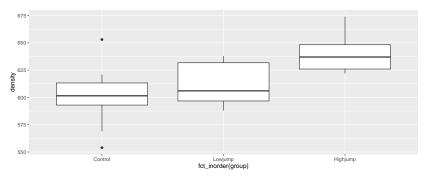
#### Look at P-values for the two tests:

Comparison	Tukey	t-tests
Highjump-Control	0.0016	0.0021
Lowjump-Control	0.4744	0.2977
Lowjump-Highjump	0.0298	0.0045

- ► Tukey P-values (mostly) higher.
- Proper adjustment for doing three t-tests at once, not just one in isolation.

# Checking assumptions

```
ggplot(rats,aes(y = density, x = fct_inorder(group)))+
  geom_boxplot()
```

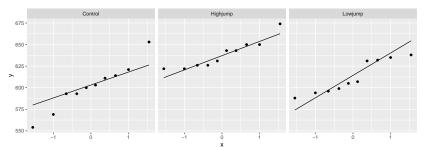


#### Assumptions:

- Normally distributed data within each group
- with equal group SDs.

## Normal quantile plots by group

```
ggplot(rats, aes(sample = density)) + stat_qq() +
   stat_qq_line() + facet_wrap( ~ group)
```



#### The assumptions

- Normally-distributed data within each group
- ► Equal group SDs.
- These are shaky here because:
  - control group has outliers
  - highjump group appears to have less spread than others.
- Possible remedies (in general):
  - Transformation of response (usually works best when SD increases with mean)
  - If normality OK but equal spreads not, can use Welch ANOVA. (Regular ANOVA like pooled t-test; Welch ANOVA like Welch-Satterthwaite t-test.)
  - Can also use Mood's Median Test (see over). This works for any number of groups.

#### Mood's median test here

- Find median of all bone densities, regardless of group
- Count up how many observations in each group above or below overall median
- Test association between group and being above/below overall median, using chi-squared test.
- Actually do this using median\_test:

```
median_test(rats, density, group)
```

```
$grand_median [1] 621.5
```

Highjump

\$table
above
group above below
Control 1 9

10 0

#### Comments

- No doubt that medians differ between groups (not all same).
- ightharpoonup This test is equivalent of F-test, not of Tukey.
- ➤ To determine which groups differ from which, can compare all possible pairs of groups via (2-sample) Mood's median tests, then adjust P-values by multiplying by number of 2-sample Mood tests done (Bonferroni):

```
pairwise_median_test(rats, density, group)
```

Now, lowjump-highjump difference no longer significant.

#### Welch ANOVA

- For these data, Mood's median test probably best because we doubt both normality and equal spreads.
- When normality OK but spreads differ, Welch ANOVA way to go.
- Welch ANOVA done by oneway.test as shown (for illustration):

```
oneway.test(density~group, data=rats)
```

One-way analysis of means (not assuming equal variances

```
data: density and group
```

F = 8.8164, num df = 2.000, denom df = 17.405, p-value = 0

- P-value very similar, as expected.
- Appropriate Tukey-equivalent here called Games-Howell.

#### Games-Howell

Lives in package PMCMRplus. Install first.

gamesHowellTest(density~factor(group),data=rats)

Control Highjump

Highjump 0.0056

Lowjump 0.5417 0.0120

# Deciding which test to do

#### For two or more samples:

