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/cm^3) measured at end.

▶ 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," ")
```

The data (some random rows)

```
# rats %>% slice_sample(n=10)
rats
```

```
# A tibble: 30 x 2
   group   density
   <chr>     <dbl>
 1 Control     611
 2 Control     621
 3 Control     614
 4 Control     593
 5 Control     593
 6 Control     653
 7 Control     600
 8 Control     554
 9 Control     603
10 Control     569
# i 20 more rows
```
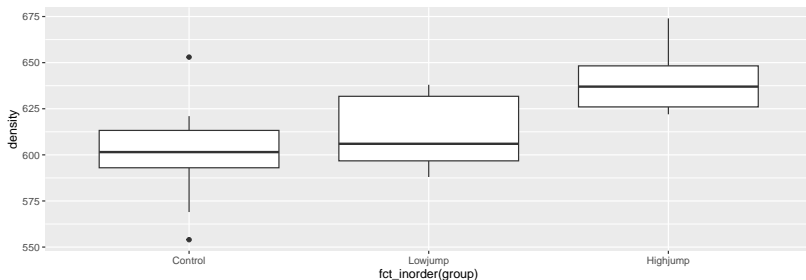
Boxplots

```
ggplot(rats, aes(y=density, x=group)) + geom_boxplot()
```
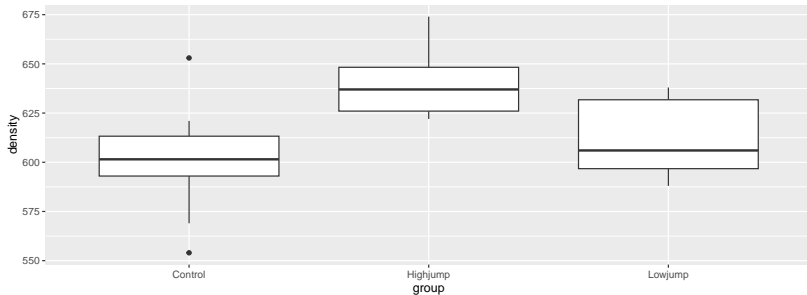
Or, arranging groups in data (logical) order

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

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)


           Df Sum Sq Mean Sq F value Pr(>F)
group       2   7434    3717   7.978 0.0019 **
Residuals  27  12579     466
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 '
```

▶ 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
    601.1     612.5
```

No sig. difference here.

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
    601.1     638.7
```

These are different.

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
    612.5     638.7
```

These are different too.

But…

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

John W. Tukey

- American statistician, 1915–2000
- Big fan of exploratory data analysis
- Popularized boxplot
- Invented "honestly significant differences"
- Invented jackknife estimation
- Coined computing term "bit"
- Co-inventor of Fast Fourier Transform

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)
summary(rats.aov)
```

```
            Df Sum Sq Mean Sq F value Pr(>F)
group        2   7434    3717   7.978 0.0019 **
Residuals   27  12579     466
---
Signif. codes:
0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
TukeyHSD(rats.aov)
```

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

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

$group
```

# 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

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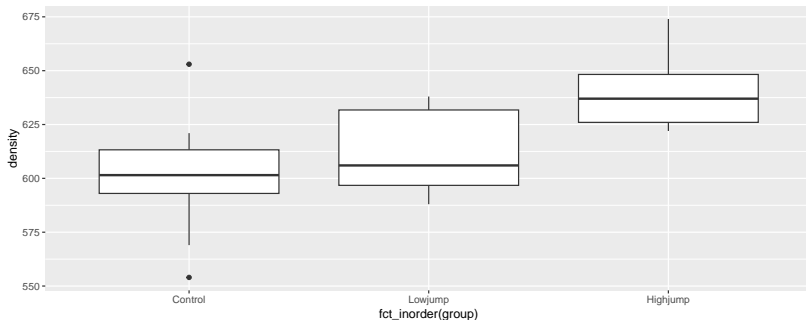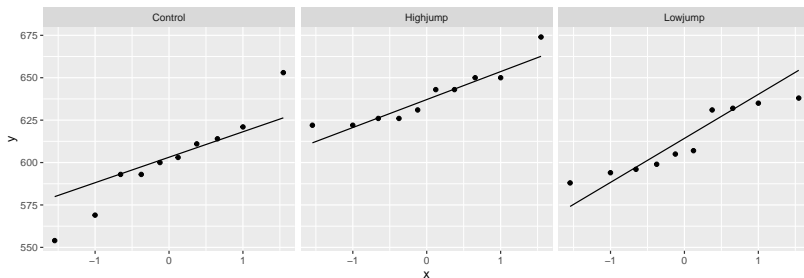

Assumptions:

▶ 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

$table
          above
group    above below
  Control     1     9
  Highjump   10     0
  Lowjump     4     6
```

Comments

▶ No doubt that medians differ between groups (not all same).
▶ 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)
```

```
# A tibble: 3 x 4
  g1       g2       p_value adj_p_value
  <chr>    <chr>      <dbl>       <dbl>
1 Control  Highjump 0.000148    0.000443
2 Control  Lowjump  0.371       1
3 Highjump Lowjump  0.371       1
```

▶ 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 ~ group, data = rats)
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: