

1 Examining ANOVA results

Comparisons of means

This lecture will concern itself with what happens if you would like to further investigate the results of a significant model I anova.

Because Model I anova focuses upon differences in means, that is the focus of comparisons after a significant model I effect.

Comparison of means in two sample case

In the two sample case, comparing means is easy because a significant result shows that the means really are significantly different. So significant result == different group means

Planned versus unplanned

In situations where there are more than two means to compare (df of groups >1), things become more complicated.

- *Planned comparisons*: occur when you have an understanding of the experimental system and hypothesize that there should be two or more classes of means before the experiment, or there are only some treatments or treatment combinations you wish to compare.
- *Unplanned comparisons* occur when you compare the means based upon results after the fact, without a specific expectation, it is known as an unplanned comparison.

Planned comparisons are always better, but unplanned comparisons can also be useful in many instances.

Planned comparisons: Treatment contrasts

Suppose you ran an experiment where you looked at the response of germination rate to various seed treatments:

- agitated with water
- agitated with water and sand
- water
- water + sand

One possibility is to contrast treatments with sand versus those with no sand This can be performed by a treatment contrast.

Treatment contrasts

For this test, the hypothesis is:

$$(\bar{X}_{\text{ag+water}} + \bar{X}_{\text{water}})/2 - (\bar{X}_{\text{ag+water+sand}} + \bar{X}_{\text{water+sand}})/2 = 0$$

Orthogonality constraint

Treatment contrasts require orthogonal comparisons. The main requirement is that the coefficients for each mean sum to 0.

In the example on the previous slide, the coefficients for each mean is 1/2. They do sum to zero. In general, 1/the number of means in each portion of the contrast determine the coefficients

Inference on treatment contrasts

To construct a test of the significance of contrasts, you must calculate the quantity L

$$L = \sum_i \lambda_i \bar{X}_i$$

The standard error of L is:

$$SE_L = \sqrt{(\sum_i \lambda_i^2)} \times \sqrt{(\sigma^2/n)}$$

Since MS_E estimates σ^2 , this equation can be written as

$$SE_L \approx \sqrt{(\sum_i \lambda_i^2)} \times \sqrt{(MS_E/n)}$$

Inference on treatment contrasts: cont

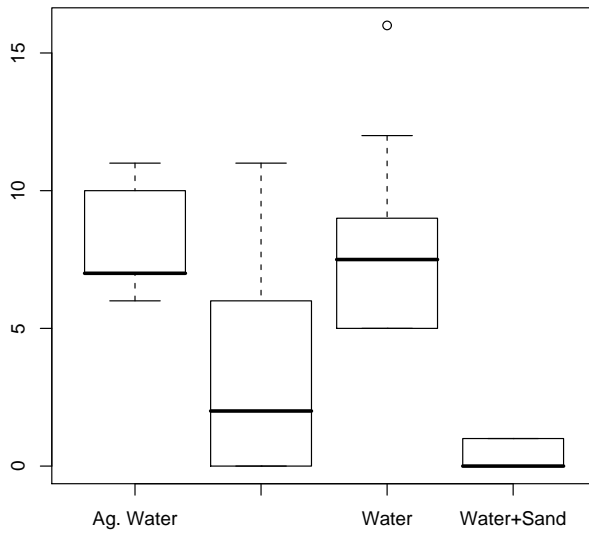
The test can be expressed as a t test where the test statistic t can be calculated:

$$t = \frac{L - 0}{SE_L}$$

with $df = N - k$

Example

These data may not meet the anova assumptions, but are instructive



Example, cont

```
> summary(aov(Cumgerm ~ treattxt, data = germ))
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
treattxt	3	426.50	142.17	18.720	1.739e-07 ***
Residuals	36	273.40	7.59		

```
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

> L <- 0.5 * mean(germ$Cumgerm[germ$treat == 0]) +
+ 0.5 * mean(germ$Cumgerm[germ$treat == 3]) -
+ 0.5 * mean(germ$Cumgerm[germ$treat == 1]) -
+ 0.5 * mean(germ$Cumgerm[germ$treat == 2])
> L
```

```
[1] -6.2
```

t-test

```
> t <- L/sqrt(7.59/10)
> t
```

```
[1] -7.116571
```

```
> pt(t, 36)
```

```
[1] 1.154945e-08
```

Unplanned comparisons

There are a number of methods to perform unplanned comparisons. I will present three (only discuss two).

- Pairwise
 - pairwise t -tests
 - Tukey's Honestly significant differences
- Many to one
 - Dunnett's test

The R package 'multcomp' implements a general mechanism for unplanned comparisons, including comparisons of interaction effects, etc.

pairwise t -tests

Probably the easiest unplanned comparison method to understand is the pairwise t -test. This analysis just does t -tests among all treatments. Note that the experiment-wide α needs to be adjusted

Bonferroni correction

The most conservative method to adjust an experiment-wide error rate is through the use of the Bonferroni correction.

If the experiment-wide α equals 0.05, the Bonferroni correction adjusts this rate for each test by dividing experiment-wide α by the number of tests

If there are 10 tests, and the experimentwide α equals 0.05, the alpha for each test is equal to 0.005.

example: `pairwise.t.test()`

```
> attach(germ)
```

```
The following object(s) are masked _by_ .GlobalEnv :
```

```
germ
```

```
> pairwise.t.test(Cumgerm, treattxt)
```

Pairwise comparisons using t tests with pooled SD

data: Cumgerm and treattxt

	Ag. Water	Ag. Water+Sand	Water
Ag. Water+Sand	0.0016	-	-
Water	0.9358	0.0016	-
Water+Sand	2.1e-06	0.0484	1.9e-06

P value adjustment method: holm

```
> detach(germ)
```

Tukey's HSD

Although the pairwise *t*-test procedure in R does use an estimate of the pooled SD, it still performs each test essentially in the exclusion of the others. Furthermore, it focuses upon the distribution of observations within groups as a measure of noise (just like a *t*-test. Tukey's Honest Significant Difference test tries to jointly test whether means are different in a post-hoc test. It uses the distribution of treatment means along with the variance within groups over the entire experiment to find the amount of noise. Tukey's HSD also automatically uses an experiment-wise error rate to calculate the error rates per comparison. In general, Tukey's HSD is preferred over pairwise *t*-tests.

Example

```
> TukeyHSD(aov(Cumgerm ~ as.factor(treattxt), data = germ))
```

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

Fit: aov(formula = Cumgerm ~ as.factor(treattxt), data = germ)

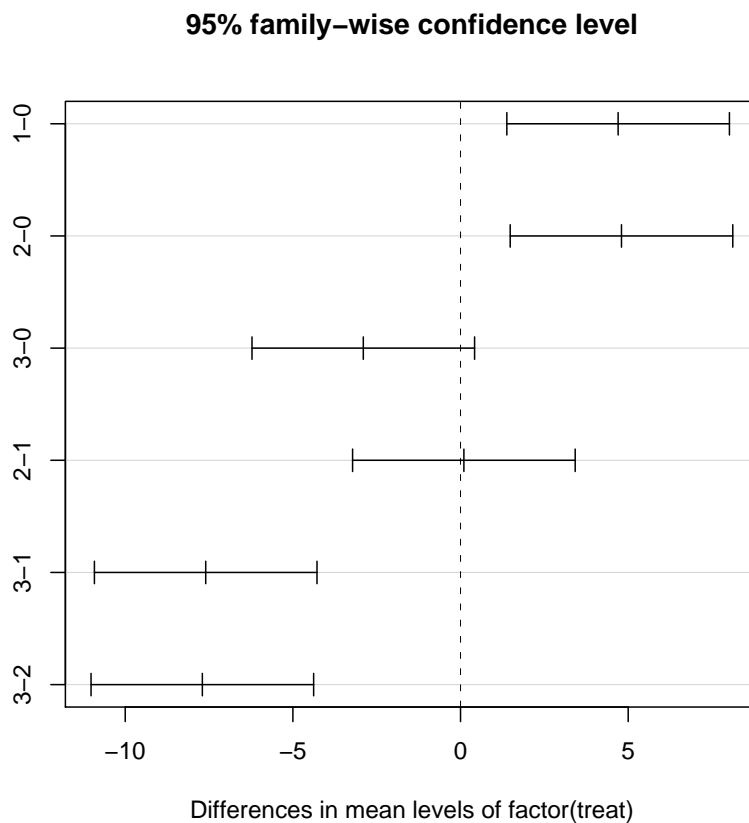
```
$'as.factor(treattxt)'
```

	diff	lwr	upr
Ag. Water+Sand-Ag. Water	-4.7	-8.019220	-1.3807804
Water-Ag. Water	0.1	-3.219220	3.4192196
Water+Sand-Ag. Water	-7.6	-10.919220	-4.2807804
Water-Ag. Water+Sand	4.8	1.480780	8.1192196
Water+Sand-Ag. Water+Sand	-2.9	-6.219220	0.4192196
Water+Sand-Water	-7.7	-11.019220	-4.3807804

	p adj
Ag. Water+Sand-Ag. Water	0.0027886
Water-Ag. Water	0.9998047
Water+Sand-Ag. Water	0.0000024
Water-Ag. Water+Sand	0.0022195
Water+Sand-Ag. Water+Sand	0.1049506
Water+Sand-Water	0.0000019

Example plot

```
> plot(TukeyHSD(aov(Cumgerm ~ factor(treat), data = germ)))
```



Dunnett's test

In some cases, you are not interested in all pairwise combinations of factor levels. Instead, you want to compare a single mean (control) to all the other treatment means. Dunnett's test was designed for this purpose.

We will not discuss Dunnett's test. It is implemented in the R package library 'multcomp' (also requires the package 'mvtnorm') along with a host of other multiple comparison methods (SNK, etc)