BMEG 802 – Advanced Biomedical Experimental Design and Analysis

Repeated Measures (Within) Analysis of Variance (ANOVA)

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Recap

- Two-Way ANOVA
  - linear model approach
  - interpret main effects and interactions
  - follow up mean comparisons
- n-Way ANOVA
  - general concepts
  - limitations

Today

- 1-Way Repeated Measures ("within" subjects) Design
  - Repeated Meaures ANOVA
    - Greenhouse-Geisser Corrections
  - Multivariate Approach
  - Friedman Test
- 2-Way Repeated Measures
  - Repeated Meaures ANOVA
    - Greenhouse-Geisser Corrections
  - Multivariate Approach

Repeated Measures (Within) ANOVA

- each individual (or culture, etc) contributes a score for **each level of a factor**
- each individual contribures multiple scores (e.g., pre vs post tests)
    - paired differences between time points
- individuals serve as their own control
- variance between different conditions is no longer due to [effect + between-group sampling variance] multiple tests on the same individual, cell culture, etc.
- it's the same group of subjects! there is no "between- group" sampling variance
- variance only due to the effect

Examples of a Within Design

1. effects of placebo, drug A and drug B can be studied in the same subjects; each subject can serve as their own control
2. behaviour of a culture studied over time; a measurement can be taken from the same culture at multiple time points

Advantages of Repeated Measures Designs

1. more information is obtained from each subject than in a between-subjects design

- within-subjects design: each subject contributes a scores (a is the number of conditions tested)
- between-subjects design: each subject contributes only one score
- # of subjects needed to reach a given level of statistical power is often much lower with within-subjects designs

2. variability in individual differences between subjects is totally removed from the error term
3. each subject serves as his/her own control
4. error term is reduced
5. statistical power increases (few participants needed)

General Linear Model (GLM)

Let's develop a full and restricted model.

Full Model: $Y_{ij} = \mu + \alpha_j + \pi_i + \epsilon_{i,j}$

Restricted Model: $Y_{ij} = \mu + \pi_i + \epsilon_{i,j}$

Effect of Factor

Effect of Subjects

- restricted model only includes effect of subjects (effect of factor is zero)
- so the difference here compared to regular "between- subjects" models is simply the inclusion of terms accounting for the effects of subjects
- remember: the more variance you can account for, the smaller the error term, the higher the F value, and the more powerful the statistical test

Defining Error and DF For the General Case

just as always, we can compute an F statistic based on Error for the full model and Error for the restricted model:

$$F = \frac{(E_{restricted} - E_{full})/(df_{restricted} - df_{full})}{(E_{full}/df_{full})}$$

$$df_{full} = (n-1)(a-1)$$

$$df_{restricted} = n(a-1)$$

$$df_{restricted} - df_{full} = (a-1)$$

see Chapter 11 of Maxwell, Delaney, and Kelley for (the complicated) calculations of $E_{restricted}$ and E_{full}

Assumptions of Repeated Measures ANOVA

- random sampling from population
- independence of subjects
- normality
- homogeneity of treatment-difference variances
  - variance of difference scores between any two levels of a factor must be equal to variance of differences scores between all other pairs of levels of the factor
  - equivalent to showing that the population covariance matrix has a certain form, that is, it displays the property of sphericity
  - this is all very mathematical and we don't need to know the details
  - fortunately there is (1) a test to see if we have violated the assumption, and (2) a method to correct for violations

Homogeneity of Treatment-Difference Variances

- Within ANOVA very sensitive and often violates sphericity
- We will see how to perform a test of sphericity in R
- R will report a number of corrected versions of the F test assuming sphericity is violated
- "Greenhouse-Geisser" adjustment adjusts the degrees of freedom (reducing them) so that Fcrit is larger (more conservative test)
- many people use G-G
- others like Huynh-Feldt because it's slightly less conservative

Disadvantages of Repeated Measures Designs

- Position Effect
  - When an observed response differs due to order of treatment
    - Must randomize the sequence of treatments or counterbalance
- Carry-over effect:
  - When an observed response is actually due to residual effect of a previous treatment
    - Must allow sufficient time ("washout period") and a controlled amount of time between treatments
    - Can't always control with counterbalancing since some effects are permanent (e.g., learning, memory, etc.)
    - Some scientific questions are better suited to between-subjects designs

Experimental Design Considerations

Counterbalancing

- counterbalance the order in which treatments are administered
- e.g., Drug A then Drug B to half the participants. Drug B then Drug A to the other half
- Participants are randomly assigned to each group
- Known as a "crossover design"
- Counterbalance an experiment with more than two levels? (e.g. 4)
- there are actually 24 different orderings of 4 conditions
- we would need 24 subjects to represent each order only once (4! = 4x3x2x1)

Experimental Design Considerations

Two alternatives:

- randomize the order for each subject; order effects will be controlled for "in the long run"
- Latin Square Designs: an arrangement of conditions so that each condition appears exactly once in each possible order

Order				
Ss	1	2	3	4
1	A	B	C	D
2	B	C	D	A
3	C	D	A	B
4	D	A	B	C

Three Group Example in R

Let's determine if there is a main effect of physical therapy treatment over time. Lower values represent pain level out of 10.

Subject	Treatment 1	Treatment 2	Treatment 3	Treatment 4
S1	8	10	7	5
S2	9	9	8	6
S3	7	5	8	4
S4	9	6	5	7
S5	8	7	7	6
S6	5	4	4	3
S7	7	6	5	4
S8	8	8	6	6
S9	9	8	6	5
S10	7	7	4	5

$H_0 : \alpha_1 = \alpha_2 = \alpha_3 = \alpha_4$

Restricted Model: $Y_{ij} = \mu + \pi_i + \epsilon_{i,j}$

$H_1 : \alpha_1 \neq \alpha_2 \neq \alpha_3 \neq \alpha_4$

Full Model: $Y_{ij} = \mu + \alpha_j + \pi_i + \epsilon_{i,j}$

Four Treatment Group Example

```
dv <- c(8,10,7,5,9,9,8,6,7,5,8,4,9,6,
        5,7,8,7,7,6,5,4,4,3,7,6,5,4,8,
        8,6,6,9,8,6,5,7,7,4,5)
treatment <- factor(rep(c(1,2,3,4),10))
subject <- factor(c(rep(1,4),rep(2,4),rep(3,4),
                    rep(4,4),rep(5,4),rep(6,4),
                    rep(7,4),rep(8,4),rep(9,4),rep(10,4)))
mydata <- data.frame(dv, treatment, subject)
```

Four Treatment Group Example

```
mydata
```

```
##    dv treatment subject
## 1   8         1       1
## 2  10         2       1
## 3   7         3       1
## 4   5         4       1
## 5   9         1       2
## 6   9         2       2
## 7   8         3       2
## 8   6         4       2
## 9   7         1       3
## 10  5         2       3
## 11  8         3       3
## 12  4         4       3
## 13  9         1       4
## 14  6         2       4
## 15  5         3       4
## 16  7         4       4
## 17  8         1       5
## 18  7         2       5
## 19  7         3       5
## 20  6         4       5
## 21  5         1       6
## 22  4         2       6
## 23  4         3       6
## 24  3         4       6
## 25  7         1       7
```
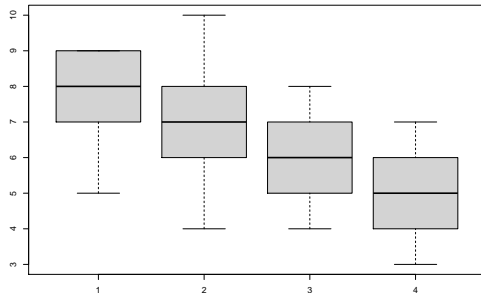
Visualize Data

Many ways to visualize data. Lets try out a box and whisker plot this time.

```
boxplot(split(mydata$dv,mydata$treatment))
```

Repeated Measures ANOVA - Univariate Approach

```r
m1 <- aov(dv ~ treatment + Error(subject/treatment), data=mydata)
summary(m1)
```

```
##
## Error: subject
##           Df Sum Sq Mean Sq F value Pr(>F)
## Residuals  9   48.4   5.378
##
## Error: subject:treatment
##           Df Sum Sq Mean Sq F value   Pr(>F)
## treatment  3   38.9  12.967   12.24 3.06e-05 ***
## Residuals 27   28.6   1.059
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Here we see a significant influence of treatment ($p < 0.001$)

note: The Error() term tells R to slice out of the model an additional error term corresponding to the variance accounted for by subjects (MS = 5.378).

Univariate: a single dependent variables (we have been doing this throughout the course)

Repeated Measures ANOVA - Sphercity Violations

Repeated Measures are very sensitive to Sphericity Violations

1. Greenhouse-Geisser Corrections

2. Multivariate Approach

- less sensitive to sphericity violations
- assumes normality
- Can also consider one or more dependent measures (e.g., MANOVA).
- see Maxwell, Delaney, Kelley text for details.

3. Friedman Test

- nonparamtric
- 1-way designs only

Let's do the GG, and we'll come back to Multivariate & Friedman afterwords.

Repeated Measures ANOVA - Greenhouse Geisser

First, we must reorganize the data into a format in which each row represents a single subject, and columns represent levels of the treatment factor.

```
response <- with(mydata,cbind(dv[treatment==1], dv[treatment==2], dv[treatment==3], dv[treatment==4]))
response
```

```
##       [,1] [,2] [,3] [,4]
## [1,]    8   10    7    5
## [2,]    9    9    8    6
## [3,]    7    5    8    4
## [4,]    9    6    5    7
## [5,]    8    7    7    6
## [6,]    5    4    4    3
## [7,]    7    6    5    4
## [8,]    8    8    6    6
## [9,]    9    8    6    5
## [10,]   7    7    4    5
```

Repeated Measures ANOVA - Greenhouse Geisser

Next, for the linear model.

```r
mlm1 <- lm(response ~ 1)
mlm1
```

```
##
## Call:
## lm(formula = response ~ 1)
##
## Coefficients:
##                [,1]  [,2]  [,3]  [,4]
## (Intercept)    7.7   7.0   6.0   5.1
```

The ~1 notation simply tells R that there are no between-subjects factors here

- in other words, only fit the model using intercepts.

Repeated Measures ANOVA - Greenhouse Geisser

Now we must set up a variable that defines the design of our study, which is a single factor with four levels:

```
rfactor <- factor(c("r1", "r2", "r3", "r4"))
design = data.frame(rfactor)
```

Repeated Measures ANOVA - Greenhouse Geisser

Now we must set up a variable that defines the design of our study, which is a single factor with four levels:

install.packages("car")

```
library(car)
```

```
## Loading required package: carData
```

```
mlm1.aov <- Anova(mlm1, idata=design,
                  idesign = ~rfactor, type="III")
```

- mlm1, is our multivariate model defined above.
- idata=design defines the number of levels
- idesign=~rfactor defines repeated-measures variable.
- type="III", instructs Anova() to calculate the "Type-III" sums of squares (there are different types of sums of squares (see Maxwell, Delaney, Kelley)

Repeated Measures ANOVA - Greenhouse Geisser

See next slide for full output

```r
summary(mlm1.aov, multivariate=FALSE)
```

```
##
## Univariate Type III Repeated-Measures ANOVA Assuming Sphericity
##
##              Sum Sq num Df Error SS den Df F value    Pr(>F)
## (Intercept) 1664.1      1     48.4      9 309.440 2.808e-08 ***
## rfactor       38.9      3     28.6     27  12.241 3.060e-05 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
##
## Mauchly Tests for Sphericity
```

Repeated Measures ANOVA – Greenhouse Geisser

```
## Univariate Type III Repeated-Measures ANOVA Assuming Sphericity
##
##              Sum Sq num Df Error SS den Df F value    Pr(>F)
## (Intercept) 1664.1      1     48.4      9 309.440 2.808e-08 ***
## rfactor       38.9      3     28.6     27  12.241 3.060e-05 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
##
## Mauchly Tests for Sphericity
##
##         Test statistic p-value
## rfactor        0.34613 0.14884
##
##
## Greenhouse-Geisser and Huynh-Feldt Corrections
##  for Departure from Sphericity
##
##         GG eps Pr(>F[GG])
## rfactor 0.7426  0.0002388 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
##            HF eps   Pr(>F[HF])
## rfactor 0.9981017 3.106252e-05
```

Mauchly Test for Sphericity not violated (p = 0.149) [i.e., p > 0.05]

Look at GG correction if violated (p = 0.0002)

Organize Data by Treatment

Let's quickly split our data into each treatment

```
treat1 = response[1:10]
treat2 = response[11:20]
treat3 = response[21:30]
treat4 = response[31:40]
```

Testing normalility

Performed on the PAIRED differences

```
shapiro.test(treat2-treat1)$p.value #
```

```
## [1] 0.4208624
```
```
shapiro.test(treat3-treat1)$p.value #
```

```
## [1] 0.5195284
```
```
shapiro.test(treat4-treat1)$p.value #
```

```
## [1] 0.008488973
```
```
shapiro.test(treat3-treat2)$p.value #
```

```
## [1] 0.1526917
```
```
shapiro.test(treat4-treat2)$p.value #
```

```
## [1] 0.6088367
```
```
shapiro.test(treat4-treat3)$p.value #
```

```
## [1] 0.6420479
```

normality violated—consider using Friedman Test, Wilcoxon signed-rank, CLES

Follow up Mean Comparisons

```r
p1v2 = t.test(treat1, treat2, paired = TRUE, alternative = "two.sided")$p.value #
p1v3 = t.test(treat1, treat3, paired = TRUE, alternative = "two.sided")$p.value #
p1v4 = t.test(treat1, treat4, paired = TRUE, alternative = "two.sided")$p.value #
p2v3 = t.test(treat2, treat3, paired = TRUE, alternative = "two.sided")$p.value #
p2v4 = t.test(treat2, treat4, paired = TRUE, alternative = "two.sided")$p.value #
p3v4 = t.test(treat3, treat4, paired = TRUE, alternative = "two.sided")$p.value #
pvals = c(p1v2, p1v3, p1v4, p2v3, p2v4,p3v4)
pvals_holm = p.adjust(pvals, method = "holm", n = length(pvals))
sprintf("%.5f", pvals_holm) #outputs in decimal (not scientific notation)
```

```
## [1] "0.31977" "0.02139" "0.00001" "0.31977" "0.02139" "0.31977"
```

There is a reduction in pain between treatments 1 and 3 ($p = 0.021$), treatments 1 and 4 ($p < 0.001$), and treatment 2 and 4 ($p = 0.021$)

Paired Sample Effect Sizes

```r
install.packages("effsize")
library(effsize)
cohen.d(treat1, treat2, paired = TRUE)$estimate #
```

```
## [1] 0.4182333
```
```r
cohen.d(treat1, treat3, paired = TRUE)$estimate #
```

```
## [1] 1.226729
```
```r
cohen.d(treat1, treat4, paired = TRUE)$estimate #
```

```
## [1] 2.117492
```
```r
cohen.d(treat2, treat3, paired = TRUE)$estimate #
```

```
## [1] 0.5951196
```
```r
cohen.d(treat2, treat4, paired = TRUE)$estimate #
```

```
## [1] 1.181162
```
```r
cohen.d(treat3, treat4, paired = TRUE)$estimate #
```

```
## [1] 0.6631177
```

ANOVA Effect Size in R

Bakeman, R. (2005). Recommended effect size statistics for repeated measures designs. Behavior research methods, 37(3), 379-384.

install.packages("effectsize")

```
library(effectsize)
omega_squared(mlm1.aov)
```

```
## Parameter | Omega2 (partial) |       90% CI
## -------------------------------------------
## rfactor   |             0.92 | [0.75, 0.96]
```

Summary of One-Way Repeated Measures ANOVA

We found a significant main effect of Treatment [$F(3,27) = 12.24$, $p < 0.001$ (GG-corrected), $\omega^2 = 0.92$]. There was a reduction in pain between treatments 1 and 3 ($p = 0.021$, $d = 1.23$), treatments 1 and 4 ($p < 0.001$, $d = 2.12$), and treatment 2 and 4 ($p = 0.021$, $d = 1.18$).

Power Analysis on ANOVA

install.packages("WebPower")

```
library(WebPower)
```

```
## Loading required package: MASS

## Loading required package: lme4

## Loading required package: Matrix

## Registered S3 methods overwritten by 'lme4':
##   method                         from
##   cooks.distance.influence.merMod car
##   influence.merMod               car
##   dfbeta.influence.merMod        car
##   dfbetas.influence.merMod       car

## Loading required package: lavaan
```

This is lavaan 0.6-7

Power Analysis on ANOVA

install.packages("WebPower")

```r
library(WebPower)
# n=sub, ng=#ofgroups, nm=#ofmeasurements, nscor=sphericity(1=perfect)
#type "0" between-effect; "1" within-effect; and "2" interaction effect
wp.rmanova(n = NULL, ng = 1, nm = 4, f = .4, nscor = 1,
           alpha = 0.05, power = 0.8, type = 1)
```

```
## Repeated-measures ANOVA analysis
##
##            n   f ng nm nscor alpha power
##     69.45101 0.4  1  4     1  0.05   0.8
##
## NOTE: Power analysis for within-effect test
## URL: http://psychstat.org/rmanova
```

We need 70 participants per group for a sufficiently powered.

Repeated Measures ANOVA - Multivariate Approach

Multivariate approach does not require the assumption of sphericity

- Good news, we can use the same linear model and set Multivariate = TRUE
- See next slide for multivariate output

```r
summary(mlm1.aov, multivariate=TRUE)
```

```
## 
## Type III Repeated Measures MANOVA Tests:
## 
## ------------------------------------------
## 
## Term: (Intercept)
## 
##  Response transformation matrix:
##       (Intercept)
```

Repeated Measures ANOVA – Multivariate Approach

```
Term: rfactor

  Response transformation matrix:
      rfactor1 rfactor2 rfactor3
[1,]        1        0        0
[2,]        0        1        0
[3,]        0        0        1
[4,]       -1       -1       -1

  Sum of squares and products for the hypothesis:
          rfactor1 rfactor2 rfactor3
rfactor1      67.6     49.4     23.4
rfactor2      49.4     36.1     17.1
rfactor3      23.4     17.1      8.1

Multivariate Tests: rfactor
                  Df test stat approx F num Df den Df    Pr(>F)
Pillai             1   0.95080 45.09192      3      7 6.0316e-05 ***
Wilks              1   0.04920 45.09192      3      7 6.0316e-05 ***
Hotelling-Lawley   1  19.32511 45.09192      3      7 6.0316e-05 ***
Roy                1  19.32511 45.09192      3      7 6.0316e-05 ***
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

There is a significant main effect of group ($p < 0.001$).

I suggest using Pillai's Trace because it is the most robust. Can use the same mean comparisons and effect size estimates we did with univariate approach.

Friedman Test

Nonparametric omnibus test for a 1-way repeated measures design. Let's perform this test on the previous example

install.packages("rstatix")

```
library(rstatix)
```

```
##
## Attaching package: 'rstatix'

## The following object is masked from 'package:MASS':
##
##     select

## The following objects are masked from 'package:effectsize':
##
##     cohens_d, eta_squared
```

Friedman Test

```
library(rstatix)
friedman_test(dv ~ treatment | subject, data=mydata)

## # A tibble: 1 x 6
##   .y.       n statistic    df        p method
## * <chr> <int>     <dbl> <dbl>    <dbl> <chr>
## 1 dv       10      19.5     3 0.000219 Friedman test
```

There is a significant main effect of group ($p < 0.001$). Can follow up with tests for Wilcoxon signed-rank and common language effect size.

2-Way Repeated Measures

2-Way Repeated Measures

Factor A	A1			A2		
Factor B	B1	B2	B3	B1	B2	B3
Subject 1	420	420	480	480	600	780
Subject 2	480	480	540	660	780	780
Subject 3	540	660	540	480	660	720
Subject 4	480	480	600	360	720	840
Subject 5	540	600	540	540	720	780

e.g., Is there an Influence of Sound Volume (Factor A) and Light Luminance (Factor B) cues on Reaction Time

2-Way - Univariate Approach

```
dv1 <- c(420,420,480,480,600,780,480,480,540,660,780,780,540,660,
         540,480,660,720,480,480,600,360,720,840,540,600,540,540,720,
         780)
factorA <- factor(rep(c(rep(1,3),rep(2,3)),5))
factorB <- factor(rep(c(1,2,3),10))
subject1 <- factor(c(rep(1,6),rep(2,6),rep(3,6),
                     rep(4,6),rep(5,6)))
mydata1 <- data.frame(dv1, factorA, factorB, subject1)
```

2-Way - Univariate Approach

see next slide for full output

```
m1 <- aov(dv1 ~ factorA*factorB + Error(subject1/(factorA*factorB)),
          data=mydata1)
summary(m1)
```

```
##
## Error: subject1
##            Df Sum Sq Mean Sq F value Pr(>F)
## Residuals  4  33600    8400
##
## Error: subject1:factorA
##            Df Sum Sq Mean Sq F value Pr(>F)
## factorA     1 147000  147000   17.5 0.0139 *
## Residuals  4  33600    8400
```

2-Way - Univariate Approach

```
Error: subject1
          Df Sum Sq Mean Sq F value Pr(>F)
Residuals  4  33600    8400

Error: subject1:factorA
          Df Sum Sq Mean Sq F value Pr(>F)
factorA    1 147000  147000   17.5 0.0139 *
Residuals  4  33600    8400
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Error: subject1:factorB
          Df Sum Sq Mean Sq F value  Pr(>F)
factorB    2 138480   69240   14.16 0.00235 **
Residuals  8  39120    4890
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Error: subject1:factorA:factorB
                Df Sum Sq Mean Sq F value  Pr(>F)
factorA:factorB  2  67920   33960   11.67 0.00425 **
Residuals        8  23280    2910
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Significant Interaction between A and B (p = 0.004)

- Since there is a significant interaction, don't look at main effects (SAME RULES AS A 2-way between ANOVA!)

2-Way - Greenhouse Geisser Corrections

Reorganizing data

```
DV1 <- with(mydata1,cbind(dv1[(factorA==1 & factorB == 1)],
                                 dv1[(factorA==1 & factorB == 2)],
                                 dv1[(factorA==1 & factorB == 3)],
                                 dv1[(factorA==2 & factorB == 1)],
                                 dv1[(factorA==2 & factorB == 2)],
                                 dv1[(factorA==2 & factorB == 3)]))
DV1
```

```
##      [,1] [,2] [,3] [,4] [,5] [,6]
## [1,]  420  420  480  480  600  780
## [2,]  480  480  540  660  780  780
## [3,]  540  660  540  480  660  720
## [4,]  480  480  600  360  720  840
## [5,]  540  600  540  540  720  780
```

2-Way - Greenhouse Geisser Corrections

Defining the Design

```
FACTOR_A <- factor(c(rep(1,3),rep(2,3)))
FACTOR_B <- factor(rep(c(1,2,3),2))
design1 <- data.frame(FACTOR_A, FACTOR_B)
design1
```

```
##   FACTOR_A FACTOR_B
## 1        1        1
## 2        1        2
## 3        1        3
## 4        2        1
## 5        2        2
## 6        2        3
```

2-Way - Greenhouse Geisser Corrections

Multivariate, linear model.

```
mlm2 <- lm(DV1 ~ 1)
mlm2
```

```
##
## Call:
## lm(formula = DV1 ~ 1)
##
## Coefficients:
##               [,1]  [,2]  [,3]  [,4]  [,5]  [,6]
## (Intercept)   492   528   540   504   696   780
```

2-Way - Greenhouse Geisser Corrections

install.packages("car")

```r
library(car)
mlm2.aov <- Anova(mlm2, idata=design1,
                  idesign = ~FACTOR_A*FACTOR_B, type="III")
```

2-Way - Greenhouse Geisser Corrections

See next slide for full output

```r
summary(mlm2.aov, multivariate=FALSE, univariate=TRUE)
```

```
## Warning in summary.Anova.mlm(mlm2.aov, multivariate = FALSE, univariate
## HF eps > 1 treated as 1

##
## Univariate Type III Repeated-Measures ANOVA Assuming Sphericity
##
##                      Sum Sq num Df Error SS den Df  F value     Pr(>F)
## (Intercept)        10443000      1    33600      4 1243.214 3.861e-06 **
## FACTOR_A             147000      1    33600      4   17.500  0.013881 *
## FACTOR_B             138480      2    39120      8   14.159  0.002354 **
## FACTOR_A:FACTOR_B     67920      2    23280      8   11.670  0.004246 4**
##
```

2-Way - Greenhouse Geisser Corrections

```
Univariate Type III Repeated-Measures ANOVA Assuming Sphericity

                 Sum Sq num Df Error SS den Df  F value    Pr(>F)
(Intercept)     10443000     1    33600      4 1243.214 3.861e-06 ***
FACTOR_A          147000     1    33600      4   17.500  0.013881 *
FACTOR_B          138480     2    39120      8   14.159  0.002354 **
FACTOR_A:FACTOR_B  67920     2    23280      8   11.670  0.004246 **
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1


Mauchly Tests for Sphericity

                 Test statistic p-value
FACTOR_B                0.76047 0.66317
FACTOR_A:FACTOR_B       0.96131 0.94254


Greenhouse-Geisser and Huynh-Feldt Corrections
 for Departure from Sphericity

                  GG eps Pr(>F[GG])
FACTOR_B         0.80676    0.005291 **
FACTOR_A:FACTOR_B 0.96275    0.004857 **
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

                  HF eps  Pr(>F[HF])
FACTOR_B         1.271242 0.002354093
FACTOR_A:FACTOR_B 1.838414 0.004245732
```

Sphericity not violated (Mauchly Test, p > 0.05). GG-corrected interaction significant (p = 0.004857)

2-Way - Multivariate Approach

See Partial Output on next page

```r
summary(mlm2.aov, multivariate=TRUE, univariate=FALSE)
```

```
##
## Type III Repeated Measures MANOVA Tests:
##
## -------------------------------------------
##
## Term: (Intercept)
##
##  Response transformation matrix:
##      (Intercept)
## [1,]           1
## [2,]           1
```

2-Way - Multivariate Approach

```
Multivariate Tests: FACTOR_A
                 Df test stat approx F num Df den Df   Pr(>F)
Pillai            1  0.813953     17.5       1      4 0.013881 *
Wilks             1  0.186047     17.5       1      4 0.013881 *
Hotelling-Lawley  1  4.375000     17.5       1      4 0.013881 *
Roy               1  4.375000     17.5       1      4 0.013881 *
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1


Multivariate Tests: FACTOR_B
                 Df test stat approx F num Df den Df   Pr(>F)
Pillai            1  0.868521 9.908686       2      3 0.047674 *
Wilks             1  0.131479 9.908686       2      3 0.047674 *
Hotelling-Lawley  1  6.605791 9.908686       2      3 0.047674 *
Roy               1  6.605791 9.908686       2      3 0.047674 *
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Multivariate Tests: FACTOR_A:FACTOR_B
                 Df test stat approx F num Df den Df   Pr(>F)
Pillai            1  0.849438 8.462687       2      3 0.058421 .
Wilks             1  0.150562 8.462687       2      3 0.058421 .
Hotelling-Lawley  1  5.641791 8.462687       2      3 0.058421 .
Roy               1  5.641791 8.462687       2      3 0.058421 .
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

The multivariate approach suggests that there are only main effects of A ($p = 0.013$) and B ($p = 0.048$) on our dependent measure [Interaction ($p = 0.058$)]

2-Way - Multivariate Approach

Notes on Main Effect of A.

- We see sphericity tests for the main effect of factorB, and the factorA:factorB interaction effect.
- We don't see a test of the main effect of factorA
  - Because in this case, factorA has only two levels
    - and so there is no variances of differences-between-groups.
    - since there are only two levels, there is only a single variance of differences (between the two levels)

Different Results, What do you do?

The 2-way RM ANOVA (with and without GG corrections) suggested there was a significant interaction between factor A and B (as well as main effects), but the multivariate approach suggests there were ONLY main effects.

- Can justify that GG corrections were made and look at interactions
- Alternatively, go with the more conservative test and look at main effects (recommended)
  - replication crisis
- Ultimately you will have to justify your decisions in your paper and to reviewers.

Let's say we went with the conservative approach. Let's look at the mean comparisons for the main effects of A (sound) and B (light) on the DV (reaction time).

Main Effects of A (Sound)

Group all data sound 1 or sound 2 (ignore effect of light)

```
# sound1 vs sound2 --- group all data sound 1 or sound 2 (ignore effect of light)
sound1 = c(dv1[1:3],dv1[7:9],dv1[13:15], dv1[19:21], dv1[25:27])
sound2 = c(dv1[4:6],dv1[10:12],dv1[16:18], dv1[22:24], dv1[28:30])
t.test(sound1, sound2, paired = TRUE, alternative = "two.sided")$p.value
```

[1] 0.001168222

There is a significant difference in reaction time between sound 1 and 2

Main Effects of B (Light)

Group all data light 1, 2, or 3 (ignore effect of sound)

```
# light1 vs light2 --- group all data light 1 or light 2 (ignore effect of sound)
light1 = c(dv1[1],dv1[4],dv1[7], dv1[10], dv1[13], dv1[16], dv1[19], dv1[22], dv1[25],dv1[28])
light2 = c(dv1[2],dv1[5],dv1[8], dv1[11], dv1[14], dv1[17], dv1[20], dv1[23], dv1[26],dv1[29])
light3 = c(dv1[3],dv1[6],dv1[9], dv1[12], dv1[15], dv1[18], dv1[21], dv1[24], dv1[27],dv1[20])
pval_l1vl2 = t.test(light1, light2, paired = TRUE, alternative = "two.sided")$p.value
pval_l1vl3 = t.test(light1, light3, paired = TRUE, alternative = "two.sided")$p.value
pval_l2vl3 = t.test(light2, light3, paired = TRUE, alternative = "two.sided")$p.value
pvals = c(pval_l1vl2, pval_l1vl3, pval_l2vl3)
p.adjust(pvals, method = "holm", n = length(pvals))
```

```
## [1] 0.03034858 0.06358186 0.66374348
```

There is a significant difference in reaction time between light 1 and 2

Note: You would also have to find effect sizes, test assumption, etc., as in the univariate example, but we'll stop here for today lecture.

Next Week

- Factorial (Between and Within) ANOVA