# EPSE 592: Design \& Analysis of Experiments 

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## Last Time

- Assumptions of simple fixed effects ANOVA models
- ANOVA model diagnostics
- Two-way (one factor) fixed effects ANOVA model


## Today

- More on two-way (one factor) fixed effects ANOVA model
- Interpreting two-way interactions
- Generic n-way ANOVA models


## Two-way (two factor) ANOVA, with interaction

|  | Female | Male |
| :--- | :---: | :---: |
| Bachelor's | $6.2,6.6,6.2$ | $5.8,6.0,5.9$ |
| Master's | $6.9,7.7,7.9$ | $6.2,6.2,6.8$ |
| PhD | $6.9,7.7,8.3$ | $9.0,9.1,8.3$ |

Table: Anxiety data (10 point scale) vs. highest education attained cross sex.

- Two-way, fixed effect ANOVA model, with interaction:

$$
Y_{a n x}=\mu+\tau_{e d u}+\tau_{\text {sex }}+\tau_{e d u \times s e x}+\varepsilon
$$

- This model will allow us to separate the marginal effects of Education and Sex from any potential interaction effect of the two.


## Two-way (two factor) ANOVA, with interaction

Two-way, fixed effect ANOVA model, with interaction:

$$
Y_{a n x}=\mu+\tau_{e d u}+\tau_{\text {sex }}+\tau_{e d u \times s e x}+\varepsilon
$$

ANOVA

|  | Sum of Squares | df | Mean Square | F | p |
| :--- | :---: | ---: | :---: | ---: | :---: |
| Education | 12.754 | 2 | 6.377 | 28.842 | $<.001$ |
| Sex | 0.109 | 1 | 0.109 | 0.492 | 0.496 |
| Education * Sex | 3.694 | 2 | 1.847 | 8.354 | 0.005 |
| Residuals | 2.653 | 12 | 0.221 |  |  |

- Can uncover marginal and interaction effects simultaneously.
- Notice: same main effect SSs as in one-way ANOVAs, and as in two-way ANOVA without interaction (consult notes from previous class).


## Two-way (two factor) ANOVA, with interaction

Two-way, fixed effect ANOVA model, with interaction:

$$
Y_{\text {anx }}=\mu+\tau_{e d u}+\tau_{\text {sex }}+\tau_{\text {edu } \times \text { sex }}+\varepsilon
$$

ANOVA

|  | Sum of Squares | df | Mean Square | F | p |
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- Again, each $F$-statistic corresponds to a different test of hypothesis:
- $F_{e d u}=M S_{e d u} / M S_{\text {res }}$ tests $H_{0}: \tau_{e d u}=0$
- $F_{\text {sex }}=M S_{\text {sex }} / M S_{\text {res }}$ tests $H_{0}: \tau_{\text {sex }}=0$
- $F_{\text {edu } \times \text { sex }}=M S_{\text {edu } \times \text { sex }} / M S_{\text {res }}$ tests $H_{0}: \tau_{\text {edu } \times \text { sex }}=0$


## Types of sums of squares

- Actually though, for ANOVAs with at least two factors, there is more than one way to partition a total sum of squares and to define a reasonable $F$-test on marginal and interaction effects.
- We will not get into the math behind this.
- Generally, always default to the Type 3 sum of squares (Jamovi defaults to this).
- Type 1: some nice mathematical properties, but order of variables matters (bad)
- Type 2: more powerful when interactions not present (unlikely in practice)
- Type 3: good mathematical properties
- Type 4: preferable in certain types of experimental designs


## Two-way (two factor) ANOVA, with interaction



Notice the apparent interaction effect: both sexes report higher anxiety levels with higher education, but the rate of increase seems to be higher for males.

## Two-way (two factor) ANOVA, with interaction



- Plotting the group means and connecting them with a line for each level of one category produces an "interaction plot."
- Very useful for visualizing interaction effects.
- But note: whether or not the lines cross is irrelevant! We are only assessing if the apparent trends are dissimilar.


## Two-way (two factor) ANOVA, with interaction

- To make the previous interaction plot in Jamovi:
- Use the "Estimated marginal means" tab in the "ANOVA" window
- Drag variables you want to plot together over to the "Marginal means" window
- Can also click option to produce tables of fitted values


## Two-Way ANOVA, post-hoc pairwise comparisons

Two-Way fixed effects ANOVA model:

$$
Y=\mu+\tau_{A}+\tau_{B}+\tau_{A \times B}+\varepsilon .
$$

- Can perform post-hoc tests, taking care to adjust for the multiple comparisons problem: recall Scheffé and Tukey's HSD.


## Post Hoc Comparisons in Two-way, Fixed Effects ANOVA Model

Post Hoc Comparisons - Education * Sex

| Comparison |  |  |  |  | Mean Difference | SE | df | t | $\mathrm{p}_{\text {tukey }}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Education | Sex |  | Education | Sex |  |  |  |  |  |
| Bachelor's | F | - | Bachelor's | M | 0.433 | 0.384 | 12.000 | 1.129 | 0.860 |
|  |  | - | Master's | F | -1.167 | 0.384 | 12.000 | -3.039 | 0.085 |
|  |  | - | Master's | M | -0.067 | 0.384 | 12.000 | -0.174 | 1.000 |
|  |  | - | PhD | F | -1.300 | 0.384 | 12.000 | -3.386 | 0.048 |
|  |  | - | PhD | M | -2.367 | 0.384 | 12.000 | -6.164 | <. 001 |
|  | M | - | Master's | F | -1.600 | 0.384 | 12.000 | -4.167 | 0.013 |
|  |  | - | Master's | M | -0.500 | 0.384 | 12.000 | -1.302 | 0.779 |
|  |  | - | PhD | F | -1.733 | 0.384 | 12.000 | -4.515 | 0.007 |
|  |  | - | PhD | M | -2.800 | 0.384 | 12.000 | -7.293 | <. 001 |
| Master's | F | - | Master's | M | 1.100 | 0.384 | 12.000 | 2.865 | 0.113 |
|  |  | - | PhD | F | -0.133 | 0.384 | 12.000 | -0.347 | 0.999 |
|  |  | - | PhD | M | -1.200 | 0.384 | 12.000 | -3.126 | 0.074 |
|  | M | - | PhD | F | -1.233 | 0.384 | 12.000 | -3.212 | 0.064 |
|  |  | - | PhD | M | -2.300 | 0.384 | 12.000 | -5.991 | <. 001 |
| PhD | F | - | PhD | M | -1.067 | 0.384 | 12.000 | -2.778 | 0.129 |

## Assumptions of Two-way, Fixed Effects ANOVA Model

Same assumptions as one-way model!

- Independence of observations $\Leftrightarrow$ independence of errors
- Equal variances across factor levels (homoskedasticity)
- Errors should be normally distributed, $\varepsilon \sim N\left(0, \sigma^{2}\right)$

Check assumptions same way as one-way model.

## Assumptions 1: independence

Recall that this is the most important assumption and the most difficult to check.

- In general, good study design should ensure independence of observations (errors).
- Did any of our sample individuals know each other?
- Were the sampled individuals assessed for the anxiety measure jointly (as in a focus group setting), or independently of each other?


## Assumptions 2: homoskedasticity (equal variances across groups)

Many ways to check this assumption (use more than one!)

- Levene's test ( $F$-test)
- Compare boxplots
- Compare plots of raw data


# Assumptions 2: homoskedasticity (equal variances across groups) 

## Test for Homogeneity of Variances (Levene's)

| $F$ | df1 | df2 | $p$ |
| :---: | :---: | :---: | :---: |
| 2.142 | 5 | 12 | 0.130 |

- Levene's test not significant; thus, gives no evidence of heteroskedasticity.
- Lack of power? See plots...


## Assumptions 2: homoskedasticity (equal variances across groups)



- Some visual evidence of heteroskedasticity, but...


## Assumptions 2: homoskedasticity (equal variances across groups)



- ...only based on three data points per group. Not really enough data per group to adequately assess the homoskedasticity assumption.


## Assumptions 3: normality of errors



- Looks pretty good! But again, only 18 data points total....


## Two-Way ANOVA, in practice

Two-way fixed effects ANOVA (full) model:

$$
Y=\mu+\tau_{A}+\tau_{B}+\tau_{A \times B}+\varepsilon
$$

- \# of obs. in each category can be different. If all the same, then the design is said to be "balanced".
- Balanced analyses have higher power and are more robust to unequal variances across categories (i.e. violations of Assumption 2). They are very robust to moderate departures from normality; i.e. skewness not a big problem, but multiple modes or outliers can be.
- The interaction term, $\tau_{A \times B}$, is often of the greatest interest.
- However, need lots of data to detect meaningful interaction effects.


## Generic $n$-way ANOVAs

Nothing special about two factors; can write models with as many explanatory factors as we like.

- For example, three-way fixed effects ANOVA (full) model:

$$
Y=\mu+\tau_{A}+\tau_{B}+\tau_{A \times B}+\tau_{C}+\tau_{A \times C}+\tau_{B \times C}+\tau_{A \times B \times C}+\varepsilon
$$

- Or, for example, a four-way ANOVA with two pairwise interactions:

$$
Y=\mu+\tau_{A}+\tau_{B}+\tau_{C}+\tau_{D}+\tau_{A \times C}+\tau_{B \times D}+\varepsilon
$$

- Theoretically, the possibilities are endless.


## Generic n-way ANOVAs

- However, in practice, the more complicated your model:
(1) the more data you need to detect effects
(2) the better experimental control you need to make sure you are isolating the effects of interest
(3) the harder it is to diagnose your model and check your assumptions (need lots more data!)
(4) the easier it is to fool yourself into thinking "complicated answer" means the same thing as "right answer"


## Two-Way ANOVA: partitioning the variance

Return to the two-way fixed effects ANOVA (full) model:

$$
Y=\mu+\tau_{A}+\tau_{B}+\tau_{A \times B}+\varepsilon
$$

- Recall that we worked out mathematically how a one-way ANOVA model partitions the observed variance in our response variable into two pieces:
- (1) variance explained by the (average) differences between the explanatory (categorical) variable,
- (2) variance leftover (attributable to within-group/individual differences).
- An analagous kind of partitioning happens when we work with a more complicated ANOVA model....


## Extreme examples to clarify partitions of variance: Ex. 1

- Suppose we have these sample data on $Y$ over two categorical variables $X$ and $Z$ with 2 factor levels each:

|  | $X=M$ | $X=F$ |
| :---: | :---: | :---: |
| $Z=$ No | $2.0,2.5,2.3$ | $1.9,2.3,2.6$ |
| $Z=$ Yes | $1.5,1.6,1.1$ | $1.6,1.7,0.9$ |

- Then:

$$
\overline{X_{M}}=1.83, \overline{X_{F}}=1.83, \overline{Z_{N}}=2.27, \overline{Z_{Y}}=1.40
$$

- And

$$
\overline{X_{M} Z_{N}}=2.27, \overline{X_{M} Z_{Y}}=1.40, \overline{X_{F} Z_{N}}=2.27, \overline{X_{F} Z_{Y}}=1.40
$$

- $X$ denotes sex and $Z$ denotes presence of a tumour; response is white blood cell count.


## Extreme examples to clarify partitions of variance: Ex. 1



## Extreme examples to clarify partitions of variance: Ex. 1

| ANOVA |  |  |  |  |  |
| :--- | :---: | :---: | ---: | ---: | ---: |
|  | Sum of Squares | df | Mean Square | F | p |
| Sex | 0.000 | 1 | 0.000 | $4.152 \mathrm{e}-30$ | 1.000 |
| Tumour | 2.253 | 1 | 2.253 | 20.179 | 0.002 |
| Sex * Tumour | 0.000 | 1 | 0.000 | $4.312 \mathrm{e}-30$ | 1.000 |
| Residuals | 0.893 | 8 | 0.112 |  |  |

- No variation explained by averaging over sex
- Clear variation explained by averaging over tumour presence
- No additional variation explained by averaging over sex $\times$ tumour factor levels
- Leftover (residual) variation from individual observations within each fixed factor level


## Extreme examples to clarify partitions of variance: Ex. 2

- Now response is measure of blood pressure; experimental design assigns people to no drug, Drug A, Drug B, or both.

|  | Drug A No | Drug A Yes |
| :---: | :---: | :---: |
| Drug B No | $2.0,2.5,2.3$ | $1.6,1.7,0.9$ |
| Drug B Yes | $1.5,1.6,1.1$ | $1.9,2.3,2.6$ |

- Then:

$$
\overline{A_{N}}=1.83, \overline{A_{Y}}=1.83, \overline{B_{N}}=1.83, \overline{B_{Y}}=1.83
$$

- And

$$
\overline{A_{N} B_{N}}=2.27, \overline{A_{N} B_{Y}}=1.40, \overline{A_{Y} B_{N}}=1.40, \overline{A_{Y} B_{Y}}=2.27
$$

## Extreme examples to clarify partitions of variance: Ex. 2



## Extreme examples to clarify partitions of variance: Ex. 2

ANOVA

|  | Sum of Squares | df | Mean Square | F | p |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Drug A | 0.000 | 1 | 0.000 | $4.760 \mathrm{e}-30$ | 1.000 |
| Drug B | 0.000 | 1 | 0.000 | $3.140 \mathrm{e}-30$ | 1.000 |
| Drug A * Drug B | 2.253 | 1 | 2.253 | 20.179 | 0.002 |
| Residuals | 0.893 | 8 | 0.112 |  |  |

- No variation explained by taking Drug A (marginally)
- No variation explained by taking Drug B (marginally)
- Clear variation explained by taking both Drug A and Drug B
- Leftover (residual) variation from individual observations within each fixed factor level


## Extreme examples to clarify partitions of variance: Ex. 2



- Those who took Drug A only saw blood pressure go down.
- Those who took Drug B only saw blood pressure go down.
- But those who took both drugs (or neither) have high blood pressure; drugs seem to be interacting to negate effects of treatment.


## Extreme examples to clarify partitions of variance: Ex. 3

- Now suppose we have these sample data on $Y$ over two categorical variables $X$ and $Z$ with 2 factor levels each:

|  | $X=A$ | $X=B$ |
| :---: | :---: | :---: |
| $Z=1$ | $2.0,2.5,2.3$ | $-1.2,4.9,3.1$ |
| $Z=2$ | $0.7,3.0,3.1$ | $1.9,2.3,2.6$ |

- Then:

$$
\overline{X_{A}}=2.27, \overline{X_{B}}=2.27, \overline{Z_{1}}=2.27, \overline{Z_{2}}=2.27
$$

- And

$$
\overline{X_{A} Z_{1}}=2.27, \overline{X_{A} Z_{2}}=2.27, \overline{X_{B} Z_{1}}=2.27, \overline{X_{B} Z_{2}}=2.27
$$

## Extreme examples to clarify partitions of variance: Ex. 3



## Extreme examples to clarify partitions of variance: Ex. 3

ANOVA

|  | Sum of Squares | df | Mean Square | F | p |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Factor X | 0.000 | 1 | 0.000 | $1.664 \mathrm{e}-32$ | 1.000 |
| Factor $Z$ | 0.000 | 1 | 0.000 | $9.359 \mathrm{e}-33$ | 1.000 |
| Factor $\mathrm{X} *$ Factor $Z$ | 0.000 | 1 | 0.000 | $1.664 \mathrm{e}-32$ | 1.000 |
| Residuals | 23.707 | 8 | 2.963 |  |  |

- No variation explained by averaging over $X$ factor levels
- No variation explained by averaging over $Z$ factor levels
- No variation explained by averaging over $X \times Z$ factor levels
- All variation is residual variation from individual observations within each fixed factor level


## Extreme examples to clarify partitions of variance: Ex. 3



Notice: obviously there are differences between the $X \times Z$ groups, but not average differences.

- ANOVAs are only able to detect average differences between groups.
- But there are many ways groups can be different, e.g. different variance, skewness, kurtosis, etc.
- This is why it is always important to look at your data; don't just rely on statistical tests of hypotheses.

