

One and Two-way Analysis of Variance

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Monday Statistical inference, and the t-test

Tuesday Simple and Multiple regression

Wednesday ANOVA, ANCOVA, and linear models

Thursday Categorical data, statistical report writing,
logistic Regression

Friday Introduction to repeated measures , Principal
Component Analysis

Lecture on one-way analysis of variance

Exercise

Recap of exercise and discussion

Lecture on two-way analysis of variance and the general linear model

Exercise

Recap of exercises and discussion

Outline, 1 and 2 Way ANOVA

05_ANOVA

1 One-way Analysis of Variance

- Descriptive
- ANOVA Model
- Data Example
- Model Control
- Multiple Testing
- Exercise

2 Two-way Analysis of Variance

- Interaction
- Interaction estimates
- Hypotheses
- Exercise

Overview

05_ANOVA

1 One-way Analysis of Variance

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One-way Analysis of Variance

05_ANOVA

- Comparing the means in more than two groups.
- One-way ANOVA gives joint test for any differences between the groups.
- **One-way** because only one grouping.
- **Analysis of variance** because we are comparing the variance between groups to the variance within groups.

Example: Birth Weight (Hosmer & Lemeshow data 2000)

05_ANOVA

- * Birth weight of 189 babies.
- * Race is grouped in three groups: white, black and other.
- * We want to compare the birth weight for these three groups.
- * The first step is to describe the data using simple tables and plots.

Example: Birth Weight

05_ANOVA

```
lbw <- read.delim("lbw.txt")
by(lbw$bwt, lbw$race, summary)
```

```
## lbw$race: black
```

##	Min.	1st Qu.	Median	Mean	3rd Qu.	Max.
##	1140	2370	2850	2720	3060	3860

```
## -----
```

```
## lbw$race: other
```

##	Min.	1st Qu.	Median	Mean	3rd Qu.	Max.
##	709	2310	2840	2800	3270	4050

```
## -----
```

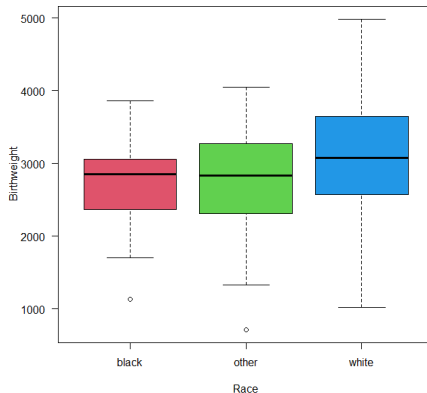
```
## lbw$race: white
```

##	Min.	1st Qu.	Median	Mean	3rd Qu.	Max.
##	1020	2580	3080	3100	3650	4990

Example: Birth Weight, Boxplot

05_ANOVA

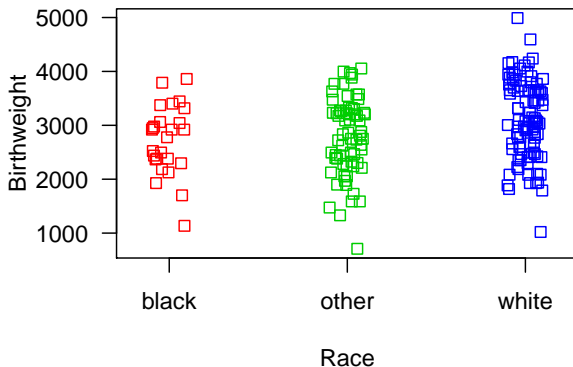
```
boxplot(bwt ~ race, data = lbw, xlab = 'Race', ylab = 'Birthweight',  
las = 1, col = 2:4)
```



Example: Birth Weight, Stripchart

05_ANOVA

```
stripchart(bwt ~ race, data = lbw, vertical = TRUE, xlab = "Race",  
ylab = "Birthweight", method = "jitter", las = 1, col = 2:4)
```



One-way Analysis of Variance Model

05_ANOVA

Let Y_{gi} be birth weight for child i in group g , $i \in \{1, \dots, n_g\}$ and $g \in \{1, \dots, k\}$.

We assume:

$$\underbrace{Y_{gi}}_{\text{Observation } i \text{ in group } g} = \underbrace{\mu_g}_{\text{Mean in group } g} + \underbrace{\epsilon_{gi}}_{\text{Individual variation}}$$

The observations are assumed to follow a normal distribution within each group, with a common variance σ^2

$$\epsilon_{gi} \sim N(0, \sigma^2), \quad Y_{gi} \sim N(\mu_g, \sigma^2)$$

These model assumptions need to be checked before interpreting the results.

Test of Hypothesis

05_ANOVA

In a one-way analysis of variance there is only one hypothesis: **Are the group means the same?**

$$H_0 : \mu_1 = \mu_2 = \dots = \mu_k$$

If the hypothesis is accepted it means that we can assume the simpler model:

$$Y_{gi} = \mu + \epsilon_{gi}, \quad i \in \{1, \dots, n_g\}, \quad g \in \{1, \dots, k\}$$

ANOVA math: Sums of Squares

05_ANOVA

Decomposition of deviation from grand mean:

$$y_{gi} - \bar{y}_{..} = (y_{gi} - \bar{y}_{g.}) + (\bar{y}_{g.} - \bar{y}_{..})$$

Decomposition of variation (sum of squares, SS).

$$\underbrace{\sum_{gi} (y_{gi} - \bar{y}_{..})^2}_{\text{Total variation}} = \underbrace{\sum_{gi} (y_{gi} - \bar{y}_{g.})^2}_{\text{Within groups}} + \underbrace{\sum_{gi} (\bar{y}_{g.} - \bar{y}_{..})^2}_{\text{Between groups}}$$

y_{gi} i 'th observation in group g .

$\bar{y}_{g.}$ Average in group g .

$\bar{y}_{..}$ Overall average or "grand mean".

Decomposition of Variation

05_ANOVA

Total = Between + Within

$$\begin{aligned}SS_{Total} &= SS_{Between} + SS_{Within} \\ N - 1 &= (k - 1) + (N - k)\end{aligned}$$

F-test statistic

$$F = \frac{MS_{Between}}{MS_{Within}} = \frac{SS_{Between}/(k - 1)}{SS_{Within}/(N - k)}$$

Decomposition of Variation

05_ANOVA

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$$F = \frac{MS_{Between}}{MS_{Within}} = \frac{SS_{Between}/(k - 1)}{SS_{Within}/(N - k)}$$

Hypothesis test

Reject null hypothesis if F large, i.e. if the variation *between* groups is too large compared to the variation *within* groups.

Decomposition of Variation

05_ANOVA

Total = Between + Within

$$\begin{aligned}SS_{Total} &= SS_{Between} + SS_{Within} \\ N - 1 &= (k - 1) + (N - k)\end{aligned}$$

F-test statistic

$$F = \frac{MS_{Between}}{MS_{Within}} = \frac{SS_{Between}/(k - 1)}{SS_{Within}/(N - k)}$$

Hypothesis test

Reject null hypothesis if F large, i.e. if the variation *between* groups is too large compared to the variation *within* groups.

F-test statistic

The F-test statistic follows F-dist. with $(k - 1)$ and $(N - k)$ df.

Reject if F is large in F-distribution.

Test of Hypothesis in Example

05_ANOVA

We want to test whether the birth weight of the children are the same irrespective of race. This corresponds to the hypothesis:

$$H_0 : \mu_1 = \mu_2 = \mu_3$$

If the hypothesis is accepted it means that we can assume the simpler model:

$$Y_{gi} = \mu + \epsilon_{gi}, \quad i \in \{1, \dots, n_g\}, \quad g \in \{1, 2, 3\}$$

ANOVA IN R (Analysis of Variance Table)

05_ANOVA

```
model1<-lm(bwt ~ race, data = lbw)
anova(model1)

## Analysis of Variance Table
##
## Response: bwt
##              Df    Sum Sq Mean Sq  F value    Pr(>F)
## race           2   5048361 2524181    4.949 0.008052 **
## Residuals    186  94866938  510037
```

The F test

05_ANOVA

The test statistic F is calculated as:

$$F = \frac{\text{Mean Sq race}}{\text{Mean Sq residual}} = \frac{2524181}{510037} = 4.949$$

- Nominator (Mean Sq race) is the variation between races.
- Denominator (Mean Sq residual) is the residual variation (or within group).

We reject the null hypothesis of equal means if F is large, i.e. when large variation between groups compared to variation within groups.

The F test

05_ANOVA

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- Nominator (Mean Sq race) is the variation between races.
- Denominator (Mean Sq residual) is the residual variation (or within group).

We reject the null hypothesis of equal means if F is large, i.e. when large variation between groups compared to variation within groups.

Here $F \sim F(2, 186)$ and the test probability is found to be

$p = P(F \geq 4.495) = 0.008 < 0.05$ so the null hypothesis is rejected

ANOVA IN R (estimates)

05_ANOVA

```
summary(model1)

##
## Call:
## lm(formula = bwt ~ race, data = lbw)
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)   2719.7      140.1    19.42  <2e-16 ***
## raceother      84.3       165.0     0.51   0.610
## racewhite     383.3       157.9     2.43   0.016 *
##
## Residual standard error: 714 on 186 degrees of freedom
## Multiple R-squared:  0.0505, Adjusted R-squared:  0.0403
## F-statistic: 4.95 on 2 and 186 DF,  p-value: 0.00805
```

ANOVA IN R (estimates)

05_ANOVA

```
confint(model1)
```

```
##              2.5 %   97.5 %  
## (Intercept) 2443.38 2996.00  
## raceother   -241.22  409.86  
## racewhite    71.83   694.81
```

ANOVA IN R (estimates) 05_ANOVA

```
confint(model1)
```

```
##              2.5 %   97.5 %  
## (Intercept) 2443.38 2996.00  
## raceother   -241.22  409.86  
## racewhite    71.83   694.81
```

- Intercept is the level of the **reference group**. R has taken the first group alphabetically or numerically. In our example **raceblack**.
- **racewhite** this is the **difference** in birth weight between white and black babies.
- **raceother** this is the **difference** in birth weight between other and black babies.

Model Assumptions

05_ANOVA

There are some assumptions connected to the model that should be checked:

Model Assumptions

05_ANOVA

There are some assumptions connected to the model that should be checked:

- Independent observations.

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05_ANOVA

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- Independent observations.
- Variance homogeneity.

Model Assumptions

05_ANOVA

There are some assumptions connected to the model that should be checked:

- Independent observations.
- Variance homogeneity.
- Normally distributed observations.

Model Control 1: Independent Observations

05_ANOVA

Based on knowledge about the sample.

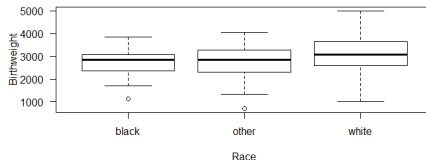
- * No twins, siblings,...
- * Only one observation from each person.

Model Control 2: Same Variance in Groups

05_ANOVA

Also called variance homogeneity (just one σ^2). Can be checked:

- Boxplots

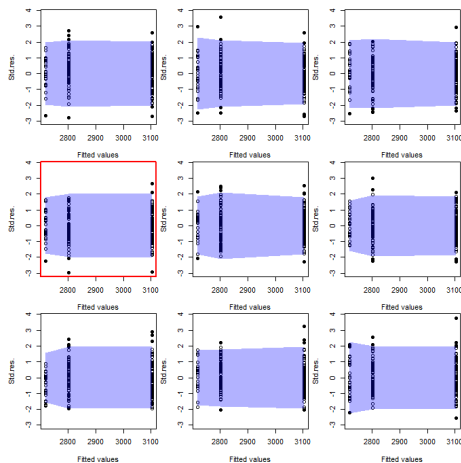


- Residuals versus fitted values, look for trumpet shape.
- Test the hypothesis of equal variances (Levene's test)

Model Control 2: Residual plot

05_ANOVA

```
library(MESS)
wallyplot(model1)
```



Model Control 2: Levene's Test

05_ANOVA

Test the hypothesis that

$$H_0 : \sigma_1^2 = \sigma_2^2 = \sigma_3^2$$

```
library(car)
leveneTest(model1)

## Levene's Test for Homogeneity of Variance (center = median)
##           Df    F value    Pr(>F)
## group      2      0.4666    0.6277
##           186
```

Model Control 2: Levene's Test

05_ANOVA

Test the hypothesis that

$$H_0 : \sigma_1^2 = \sigma_2^2 = \sigma_3^2$$

```
library(car)
leveneTest(model1)

## Levene's Test for Homogeneity of Variance (center = median)
##           Df    F value    Pr(>F)
## group      2      0.4666    0.6277
##           186
```

As $p = 0.6277 > 0.05$ then H_0 is accepted.

Model Control 3: Normal Distribution

05_ANOVA

We have assumed that the observations follow a normal distribution within each group. This can be checked:

- Draw qq-plots for each group (only if many observations)
- Draw qq-plots for the residuals

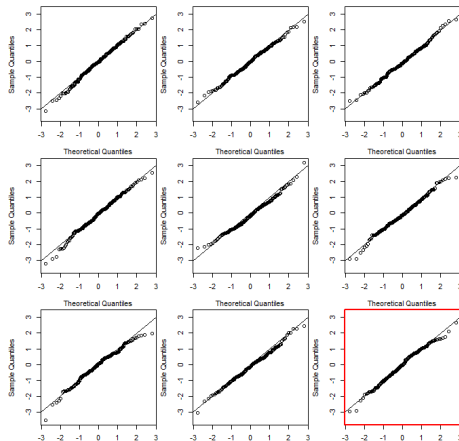
$$r_{gi} = Y_{gi} - \hat{\mu}_g = Y_{gi} - \bar{Y}_g$$

- Not advisable to do formal tests for normality
 - If the data set is large then nearly always reject normality.
 - If the data set is small then nearly always accept.

Model Control 3: QQ-plots

05_ANOVA

```
qqwrap <- function(x, y, ...) {qqnorm(y, main="", ...)
  abline(a=0, b=1)}
wallyplot(model1, FUN=qqwrap)
```



Conclusion for the Birthweight Data

05_ANOVA

- The model check showed that the assumptions were fulfilled.
- The F-test showed that there is a significant difference in birthweight for different races.
- But not where the differences were to be found.

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05_ANOVA

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- But not where the differences were to be found.

Pairwise Comparisons

Give problems with multiple testing. We have $m = k(k - 1)/2$ possible tests so true level of significance (probability of rejecting a true H_0) $1 - (1 - \alpha)^m$. For $k=3$ we get 0.14.

Multiple Testing

05_ANOVA

No completely satisfactory solution to the multiple testing problem:

Multiple Testing

05_ANOVA

No completely satisfactory solution to the multiple testing problem:

- Try to avoid the problem (focus the project).

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- Try to avoid the problem (focus the project).
- Select a small number of comparisons in the *planning* stage.

Multiple Testing

05_ANOVA

No completely satisfactory solution to the multiple testing problem:

- Try to avoid the problem (focus the project).
- Select a small number of comparisons in the *planning* stage.
- Make a graph of the averages $\pm 2 \times SEM$ and judge visually.

Multiple Testing

05_ANOVA

No completely satisfactory solution to the multiple testing problem:

- Try to avoid the problem (focus the project).
- Select a small number of comparisons in the *planning* stage.
- Make a graph of the averages $\pm 2 \times SEM$ and judge visually.
- Modify the t-test.
 - Bonferroni test at level α/m .
 - Conservative (accepts in general too often).

Pairwise Tests in R, no correction

05_ANOVA

```
pairwise.t.test(lbw$bwt, lbw$race, p.adj = "none")

##
##  Pairwise comparisons using t tests with pooled SD
##
## data:  lbw$bwt and lbw$race
##
##          black  other
## other 0.6100  -
## white 0.0161 0.0093
##
## P value adjustment method: none
```

Pairwise Tests in R, bonferroni

05_ANOVA

```
pairwise.t.test(lbw$bwt, lbw$race, p.adj = "bonferroni")
```

Pairwise comparisons using t tests with pooled SD

```
data: lbw$bwt and lbw$race
```

	black	other
other	1.000	-
white	0.048	0.028

Exercise: One-way ANOVA

05_ANOVA

- Exercise 1 Birth Weight and Mother's Weight.

Overview

05_ANOVA

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Two-way analysis of variance

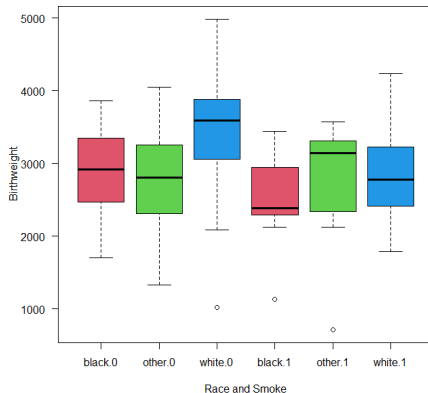
05_ANOVA

- * Before we had one factor (race), now we have two (race and smoke).
- * We want to study how race and smoking during pregnancy are associated with birth weight.
- * Our outcome is still birth weight, factor 1: race (3 groups), factor 2: smoke (2 groups).

Example: Birth weight, Boxplot

05_ANOVA

```
boxplot(bwt ~ race*smoke, data=lbw, xlab = 'Race and Smoke',
        ylab = 'Birthweight', las = 1, col = 2:4)
```



Two-way analysis of variance, additive model

05_ANOVA

We have an effect of race (r) and smoke (s):

$$Y_{rsi} = \mu + \alpha_r + \beta_s + \epsilon_{rsi}, \quad \epsilon_{rsi} \sim N(0, \sigma^2)$$

and the effects are **additive**.

Here Y_{rsi} is the birth weight for baby i of race r and mother's smoking status s .

α_r is the effect of race.

β_s is the effect of smoking.

Interaction

05_ANOVA

- * Or effect modification.
- * How do the factors work together?
- * Maybe the effect of smoking is not the same for all white, black and other.

Model with Interaction

05_ANOVA

$$Y_{rsi} = \mu + \alpha_r + \beta_s + \alpha\beta_{rs} + \epsilon_{rsi}, \quad \epsilon_{rsi} \sim N(0, \sigma^2)$$

Here Y_{rsi} is the birth weight for baby i of race r and mother's smoking status s .

α and β are called main effects and $\alpha\beta$ the interaction.

The effect of smoking can depend on race and the effect of race can depend on smoking status.

Model with Interaction

05_ANOVA

$$Y_{rsi} = \mu + \alpha_r + \beta_s + \alpha\beta_{rs} + \epsilon_{rsi}, \quad \epsilon_{rsi} \sim N(0, \sigma^2)$$

Here Y_{rsi} is the birth weight for baby i of race r and mother's smoking status s .

α and β are called main effects and $\alpha\beta$ the interaction.

The effect of smoking can depend on race and the effect of race can depend on smoking status.

We would like to simplify the model to the additive ($\alpha\beta_{rs}$):

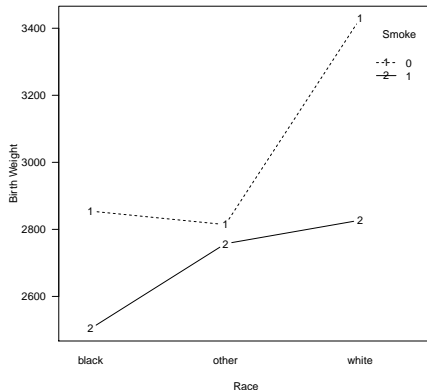
$$Y_{rsi} = \mu + \alpha_r + \beta_s + \epsilon_{rsi}$$

Here both smoking and race have an effect on birth weight. But the effect of smoking does not depend on race and vice versa.

Birth Weight Example with Interaction

05_ANOVA

```
interaction.plot(lbw$race, lbw$smoke, lbw$bwt, type=c("b"),  
ylab = "Birth Weight", las=1, trace.label = "Smoke", xlab = "Race")
```



Birth Weight Example with Interaction

05_ANOVA

```
lbw$smoke <- as.factor(lbw$smoke)
model2 <- lm(bwt ~ race + smoke + race:smoke, data = lbw)
summary(model2)
```

```
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)    2854.50    170.84  16.708  <2e-16 ***
## raceother      -40.26    194.11  -0.207   0.83591
## racewhite      574.25    199.50   2.878   0.00447 **
## smoke1        -350.50    275.47  -1.272   0.20486
## raceother:smoke1  293.43    351.13   0.836   0.40443
## racewhite:smoke1 -250.87    309.00  -0.812   0.41792
##
## Residual standard error: 683.4 on 183 degrees of freedom
## Multiple R-squared:  0.1447, Adjusted R-squared:  0.1213
## F-statistic: 6.191 on 5 and 183 DF,  p-value: 2.505e-05
```

Understanding the Estimates

05_ANOVA

Interpretation of the estimates from the previous slide.

- **Intercept** = 2854.50

The estimated birth weight for the reference group. Here race = "black" and smoke = 0.

- **raceother** = -40.26.

The effect of Other vs. Black for the ref. group (i.e. non-smoker).

- **racewhite** = 574.25

The effect of White vs. Black for the ref. group (i.e. non-smoker).

- **smoke1** = -350.50

The effect of smoker vs non-smoker for the ref. group (i.e. Black).

Understanding the Estimates (contd)

05_ANOVA

- * $\text{raceother:smoke1} = 293.43$

The extra effect of Other and smoker. The total effect of smoker vs nonsmoker for Other $-350.50 + 293.43 = -57.07$

- * $\text{racewhite:smoke1} = -250.87$

The extra effect of White and smoker. The total effect of smoker vs nonsmoker for White $-350.50 - 250.87 = -601.37$

Understanding the Estimates (contd)

05_ANOVA

The estimated birth weight for combinations of race and smoke.

Smoke	Race		
	Black	Other	White
0	2854.50	2854.50	2854.50
		- 40.26	+ 574.25
		= 2814.24	= 3428.75
1	2854.50	2854.50	2854.50
	- 350.50	- 350.50	- 350.50
		- 40.26	+ 574.25
		+ 293.43	- 250.87
	= 2504	= 2757.17	= 2827.38

Hypotheses

05_ANOVA

Model without interaction:

$$M_2 : \mu_{rsi} = \mu + \alpha_r + \beta_s$$

$$H_{20} : (\alpha\beta)_{rs} = 0 \text{ for all } r, s$$

$$H_{2A} : (\alpha\beta)_{rs} \neq 0$$

No effect of race:

$$M_3 : \mu_{rsi} = \mu + \beta_s$$

$$H_{30} : \alpha_r = 0 \text{ for all } r$$

$$H_{3A} : \alpha_r \neq 0$$

No effect of smoking:

$$M_{3*} : \mu_{rsi} = \mu + \alpha_r$$

$$H_{3*0} : \beta_s = 0 \text{ for all } s$$

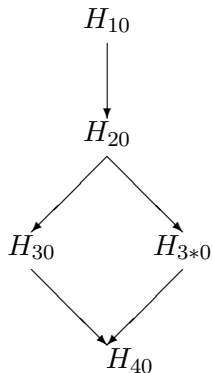
$$H_{3*A} : \beta_s \neq 0$$

No effect of smoking or race:

$$M_{3*} : \mu_{rsi} = \mu$$

Hypotheses in Two-way Analysis of Variance

05_ANOVA



- If H_{10} or H_{20} is rejected then the analysis is finished.
- H_{30} and H_{3*0} are on the same level.
- If reject H_{30} then test H_{3*0} .

Hypotheses in Birth Weight Example

05_ANOVA

```
anova(model2)

## Analysis of Variance Table
##
## Response: bwt
##              Df    Sum Sq Mean Sq  F value    Pr(>F)
## race           2   5048361 2524181    5.4052 0.005229 **
## smoke          1   7298537 7298537   15.6288 0.000110 ***
## race:smoke     2   2108643 1054321    2.2577 0.107499
## Residuals    183  85459758  466993
```

Note:

$$Mean Sq = \frac{Sum Sq}{Df}$$

Test for No Interaction, Birth Weight Example

05_ANOVA

We will try to simplify the model by testing the hypothesis of no interaction:

$$F = \frac{\text{Mean Sq race : smoke}}{\text{Mean Sq Residual}} = \frac{1054321}{466993} = 2.26$$

Under the hypothesis of no interaction we have $F \sim F(2, 183)$ and the test probability is found as:

$$p = P(F \geq f) = P(F \geq 2.26) = 0.107 > 0.05$$

Test for No Interaction, Birth Weight Example

05_ANOVA

We will try to simplify the model by testing the hypothesis of no interaction:

$$F = \frac{\text{Mean Sq } \textit{race : smoke}}{\text{Mean Sq Residual}} = \frac{1054321}{466993} = 2.26$$

Under the hypothesis of no interaction we have $F \sim F(2, 183)$ and the test probability is found as:

$$p = P(F \geq f) = P(F \geq 2.26) = 0.107 > 0.05$$

The hypothesis of no interaction is accepted.

Simplifying the model, Birth Weight Example

05_ANOVA

```
model3 <- lm(bwt ~ race + smoke, data = lbw)
anova(model3)
```

Analysis of Variance Table

##

Response: bwt

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
## race	2	5048361	2524181	5.3327	0.0056021	**
## smoke	1	7298537	7298537	15.4191	0.0001214	***
## Residuals	185	87568401	473343			

Simplifying the model, Birth Weight Example

05_ANOVA

```

model3 <- lm(bwt ~ race + smoke, data = lbw)
anova(model3)

## Analysis of Variance Table
##
## Response: bwt
##              Df    Sum Sq Mean Sq    F value    Pr(>F)
## race           2   5048361 2524181     5.3327 0.0056021 **
## smoke          1   7298537 7298537    15.4191 0.0001214 ***
## Residuals    185   87568401  473343

```

Test for no effect of race H_{30} :

$$F = \frac{\text{Mean Sq race}}{\text{Mean Sq Residual}} = \frac{2524181}{473343} = 5.3327, p = 0.006 < 0.05$$

Simplifying the model, Birth Weight Example

05_ANOVA

```
model3 <- lm(bwt ~ race + smoke, data = lbw)
anova(model3)

## Analysis of Variance Table
##
## Response: bwt
##              Df    Sum Sq Mean Sq    F value    Pr(>F)
## race           2   5048361 2524181     5.3327 0.0056021 **
## smoke          1   7298537 7298537    15.4191 0.0001214 ***
## Residuals    185   87568401  473343
```

Test for no effect of race H_{30} :

$$F = \frac{\text{Mean Sq race}}{\text{Mean Sq Residual}} = \frac{2524181}{473343} = 5.3327, p = 0.006 < 0.05$$

Test for no effect of smoke H_{3*0} :

$$F = \frac{\text{Mean Sq smoke}}{\text{Mean Sq Residual}} = \frac{7298537}{473343} = 15.4191, p = 0.0001 < 0.05$$

Final model, Birth Weight Example

05_ANOVA

```
confint(model3)
```

##		2.5 %	97.5 %
##	(Intercept)	2605.5688	3163.0662
##	raceother	-320.3599	313.0773
##	racewhite	148.5607	752.5194
##	smoke1	-643.0746	-212.9761

Interpretation of estimates:

For two mothers of the **same race** where one is a smoker and the other nonsmoker, then the birth weight of the smoker's baby will be 428g less than for the nonsmoker. The difference could be as much as 643g or as little as 213g based on the 95% CI.

Exercise: Two-way ANOVA

05_ANOVA

- Exercise: Beer Tasting
- Exercise: Fish