

# **Categorical associations**

Experimental and Statistical Methods in Biological Sciences I

Heini Saarimäki, BECS 16/10/2014

# **Outline for today**

- Categorical associations
- Nonparametric tests
- Exercises part I
- Summary of the demo sessions
- Exercises part II



#### What we know after today

- 1. Preparing your data
- 2. Describing your data
- 3. T-tests and ANOVA
- 4. Correlations and regression

#### 5. Categorical associations and nonparametric tests



### **Contingency tables**

Contingency tables (from demo2)

- Frequency of observations per category

```
> table(sex, agegroup)
> round(prop.table(table(sex, agegroup), margin=1)*100,1)
```



#### **Chi-square test**

The chi-square test tests the probability that samples of observations come from the same distributions.

We explore two possibilities:

- Goodness-of-fit test: test whether the sample distribution is same as known population distribution
- Test for independence: test whether two independent samples come from the same distributions



#### **Chi-square test**

Pearson's chi-squared goodness-of-fit test

```
> chisq.test(table(sex), p=c(.5,.5))
```

```
> chisq.test(table(sex), p=c(.5,.5))
```

Chi-squared test for given probabilities

```
data: table(sex)
X-squared = 0, df = 1, p-value = 1
```



#### **Chi-square test**

Chi-squared test for independence

> chisq.test(sex, agegroup)

Sex distribution is different in different age groups ( $\chi^2(2) = 6.02, p < .05$ ).



#### **Non-parametric tests**

Mann-Whitney-Wilcoxon test (U-test)

- Independent samples t-test
- > wilcox.test(headache\_on ~ sex)

```
> wilcox.test(headache on ~ sex)
```

Wilcoxon rank sum test with continuity correction

```
data: headache_on by sex
W = 285.5, p-value = 0.963
alternative hypothesis: true location shift is not equal to 0
```



#### **Non-parametric tests**

Wilcoxon's signed ranks test

- Repeated measures t-test
- > wilcox.test(headache1, headache\_on, paired=T)
- > wilcox.test(headache1, headache\_on, paired=T)

Wilcoxon signed rank test with continuity correction

```
data: headache1 and headache_on
V = 36, p-value = 0.2999
alternative hypothesis: true location shift is not equal to 0
```



#### **Non-parametric tests**

Kruskall-Wallis test

- One-way ANOVA
- > kruskal.test(headache\_on ~ ageggroup)
- > kruskal.test(headache\_on ~ ageggroup)

Kruskal-Wallis rank sum test

```
data: headache_on by ageggroup
Kruskal-Wallis chi-squared = 1.7418, df = 2, p-value = 0.4186
```



#### Which test should I use?

Lots of cook book solutions available online – but aim for understanding the differences between each method yourself!

http://www.ats.ucla.edu/STAT/mult\_pkg/whatstat/default.htm



### **Data for part I**

Experience of subjective symptoms caused by cellphone

- baseline, cellphone on, cellphone off

Variable	Type Description			
sex	categorical	male or female		
age group	categorical	three age groups		
rfjarj	categorical	order of the experiment		
dizziness	continuous	measured three times		
headache	continuous	measured three times		
tiredness	continuous	measured three times		
itchiness	continuous	measured three times		
blushing	continuous	measured three times		
warmth	continuous	measured three times		





# Summary of the course

Experimental and Statistical Methods in Biological Sciences I

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#### Take home

- The most important tool for research is you!
  - Starting from planning the experiment...
  - ... to stating the hypotheses...
  - ... to evaluating and interpreting the results...
  - ... to drawing conclusions.
- Statistics is a tool that helps you to draw conclusions based on your data.
- And R is just a tool that runs the statistics for you.



# What you should be able to do based on demos

- Formulate the hypothesis you are testing
- Choose a relevant test to test your hypothesis
- Interpret the R output to get results
- Report the results verbally, numerically (and graphically)
  - "Reading times of regular words are faster than those of exceptional words (t(997) = -68.8, p < .001)".



# If you need help in how to report the results...

Use APA format:

- Consult lecture slides
- Consult Jeffrey Kahn's summary:

http://my.ilstu.edu/~jhkahn/apastats.html



### **Recap of the course**

For each topic:

- Basic concepts
- Useful functions
- Important outputs



#### **Structure**

- 1. Preparing your data
- 2. Describing your data
- 3. Basics in statistical testing
- 4. Correlations
- 5. Group comparisons using T-tests
- 6. Group comparisons using ANOVA
- 7. Regression



#### 1. Preparing your data: Before R

Standard format: columns = variables rows = observations (e.g., participants)

- Collect your data into one table using your favorite software (Excel, Matlab, R, ...)
- Save preferably as .csv or .txt



#### 1. Preparing your data: Load in data

```
# Load in data from .csv file:
> data <- read.csv('naming.csv')
# Load in data where first line is header information:
> data <- read.csv('naming.csv', header=T)
# Load in data from .txt file:
> data <- read.table('naming', header=T, sep="")
# Load in online data - here, .csv file:
> data <- read.csv('http://becs.aalto.fi/~heikkih3/naming_wide.csv', header=T)</pre>
```



#### **1. Preparing your data: Checking data**

- # Check your data:
  > summary(data)
- # Correct for factors:
- > data\$variable1 <- factor(data\$variable1)</pre>
- # Identify funny coding of missing values:
- > summary(data)
- > which(data\$variable1 == 999)
- # Recode missing values as NAs:
- > data\$variable1[which(data\$variable1 == 999)] <- NA</pre>



#### 1. Preparing your data: adding/removing variables

# Adding new variables in the data frame
> data\$new\_variable <- 0</pre>

# Removing variables from the data frame
> data\$new\_variable <- NULL</pre>



#### 1. Preparing your data: Saving changes into a new data frame

```
# Copy the old data frame to a new data frame:
> data2 <- data</pre>
```

```
# Now make changes to the new data frame data2.
> data2$variable1 <- NULL</pre>
```

```
# E.g., remove a row from data frame and save to a new dataframe at
the same time:
> data2 <- data[-10,] # row 10 removed from data2</pre>
```



# **1. Preparing your data:** attach/detach data

```
# When the data frame is ready,
> attach(data)
```

# When attaching a new data frame, detach the previous one
> detach(data)



# **1. Preparing your data:** saving your data

# Save your data frame in .csv format:

> write.csv(data, "my\_data.csv")



### 1. Preparing your data

Important outputs: summary()





# 2. Describing your data

- Descriptive statistics
- Frequencies
- Plotting



# 2. Describing your data: descriptive statistics

```
# Take a basic summary:
> summary(data)
```

```
# Take all descriptive statistics:
> library(psych)
> describe(data)
```

# load package first

```
# Get single descriptive statistics:
> mean(iq)
> sd(iq)
```



# 2. Describing your data: frequencies

# Take a frequency table:
> table(sex)

# Take a contingency table:
> table(sex, reading\_group)



### 2. Describing your data

Important outputs: describe()

> describe() is included in package 'psych'

look up ?describe if you are unsure what the displayed statistics are

factors are marked					
with asterisks					

> library(psyc	ch)						
> describe(dat	ta)						
	var	n	n mean	n sd	median	trimmed	mad
iq	1	998	100.00	15.00	100.02	100.10	14.50
hrs	2	998	4.00	0.50	4.00	4.00	0.52
sex*	3	998	1.54	0.50	2.00	1.55	0.00
ms.regular	4	998	1000.00	75.00	998.69	999.14	72.56
ms.exception	5	998	1200.00	80.00	1198.70	1199.75	78.61
<pre>reading_time*</pre>	6	998	2.02	0.85	2.00	2.02	1.48
	I	nin	max	range	skew k	urtosis	se
iq	45	.13	163.41	118.29	-0.05	0.53	0.47
hrs	2	.42	5.47	3.05	-0.01	-0.18	0.02
sex*	1	.00	2.00	1.00	-0.17	-1.97	0.02
ms.regular	700	.97	1263.12	562.14	0.07	0.30	2.37
ms.exception	941	.96	1452.20	510.23	0.06	0.16	2.53
reading_time*	1	.00	3.00	2.00	-0.03	-1.60	0.03

### 2. Describing your data

How to save your table?

```
> desc <- round(describe(data[c(1:2,4:5)]),2)
> write.csv(desc, "descriptives.csv")
```

Open in Excel. Choose first column, then Data  $\rightarrow$  Text to columns.



#### 2. Describing your data: Structure of plotting functions

# 1. Define your basic format:
> plot(variable1, variable2)
> hist(variable1)



#### 2. Describing your data: Plotting distribution of one variable

```
# Histograms:
> hist(iq)
```

```
# Adding a normal curve to the histogram:
> h<- hist(iq, freq=F)
> x <- min(h$breaks):max(h$breaks)
> y <- dnorm(x, mean=mean(iq), sd=sd(iq))
> lines(x, y, col="red", lty="dashed", lwd=2)
```

# Boxplots: boxplot(iq)



#### 2. Describing your data Plotting associations between variables

# Scatterplots:
> plot(iq ~ hrs)

# Add regression line to a scatterplot:
> plot(iq ~ hrs)
> abline(lm(iq ~ hrs))



#### 2. Describing your data: Plotting categories separately

```
# Boxplots for categories separately:
> boxplot(ig ~ sex)
```

```
# Scatterplots for categories separately:
```

- > coplot(iq ~ hrs | sex)
- > library(lattice)
- > xyplot(iq ~ hrs | sex) # from package 'lattice'



### 2. Describing your data: Saving your plots

- 1. Switch the device to a file (the command you use depends on the graphics format you want: pdf is good, or you can find a whole list with ?Devices);
  - > png(filename="my\_plot.png")
- 2. Run the commands which create the graphics;

> plot(iq ~ hrs)

Switch the file device off (which switches back to the screen by default)
 > dev.off()


### 3. Basics in statistical testing

Idea behind all statistical testing:

You are testing a null hypothesis. If the test statistic is significant (usually p < . 05) you have enough confidence to *reject* the null hypothesis.



#### **3. Basics in statistical testing Underlying assumptions**

- T test assumes:
  - Normality, sample size > 20
- ANOVA assumes:
  - Normality, sample size > 15, equality of variances
- Regression assumes:
  - Normality, linearity, sample size > 50, multicollinearity
- What if I cannot meet the assumptions?
   → consider data transformations or non-parametric tests!



### **3. Basics in statistical testing Testing for normality**

- # Descriptive statistics:
  > describe(iq)
- # Histogram
  > bist(ic)
- > hist(iq)
- # Quantile-quantile plot
- > qqnorm(iq)
- > qqplot(iq)

```
# Normality tests
> ks.test(iq, "pnorm", mean=mean(iq), sd=sd(iq))
> shapiro.test(iq)
```



Important outputs: describe()

#### mean close to median suggests normality

> describe(data[c(1:2,4:5)])

	var	n	mean		$\mathbf{sd}$	med	lian	tri	immed	I	nad
iq	1	998	100	15	5.0	100	.02	10	00.10	14.	.50
hrs	2	998	4	0	).5	4	.00		4.00	0.	. 52
ms.regular	3	998	1000	75	5.0	998	.69	99	99.14	72.	.56
ms.exception	4	998	1200	80	0.0	1198	.70	119	99.75	78.	61
	n	nin '	m	ax	ra	ange	ske	ew l	kurtos	sis	se
iq	45.	.13	163.	41	118	3.29	-0.0	)5	0.	53	0.47
hrs	2.	42	5.	47	3	3.05	-0.0	01	-0.	18	0.02
ms.regular	700.	.97 :	1263.	12	562	2.14	0.0	07	Ο.	30	2.37
ms.exception	941.	.96 :	1452.3	20	510	0.23	0.0	06	Ο.	16	2.53

skewness and kurtosis close to 0 suggest normality







Important outputs: qqnorm() qqline()







Normal Q-Q Plot

Important outputs: ks.test() or shapiro.test()

tests the null hypothesis that the distributions for the first variable (here, "iq") and for the second variable (here, "pnorm", i.e. normal distribution with mean and sd based on "iq") come from the same distribution

```
> ks.test(iq, "pnorm", mean=mean(iq), sd=sd(iq)) > shapiro.test(iq)

One-sample Kolmogorov-Smirnov test

data: iq
D = 0.0196, p-value = 0.8386
alternative hypothesis: two-sided

p values > .05 suggest that the null hypothesis cannot be rejected,
i.e. in beth tests that the test suggest resumption
```

i.e., in both tests that the test suggest normality

tests the null hypothesis that the sample

(here, "iq") comes from a normally

distributed population

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- Diagnostic plots (regression, ANOVA):
  - Equality of variances
  - Influential cases
  - Normality (in ANOVA)
  - Linearity (in regression)
- Other assumptions:
  - Multicollinearity (in regression)
    - $\rightarrow$  correlation matrix (no really high correlations)







#### 4. Correlations

Tests for associations between numeric variables: how much do the variables covary.

Null hypothesis:

The true correlation between the variables is equal to 0.



#### 4. Correlations: Pearson product-moment correlation coefficient

```
# Correlation between two variables:
> cor.test(iq, hrs)
```

```
# Correlation matrix for all numeric variables:
> corr.test(data[c(1:2,4:5)])
```



#### 4. Correlations: Spearman rank-correlation coefficient

# Correlation between two variables:
> cor.test(iq, hrs, method="spearman")

# Correlation matrix for all numeric variables:
> corr.test(data[c(1:2,4:5)], method="spearman")



#### 4. Correlations

> cor.test(iq, hrs)

Important outputs: cor.test()

#### p value tells you whether you can safely reject the null hypothesis

Pearson's product-moment correlation



## 4. Correlations

### Important outputs: corr.test()





### 5. Comparing means with t tests

Null hypothesis: The true differences in means is equal to 0.

Compare the means between

- Sample and population  $\rightarrow$  one-sample t-test, Mann-Whitney-Wilcoxon test
  - Independent samples
    - $\rightarrow$  independent samples t-test, Mann-Whitney-Wilcoxon test
  - Repeated measures
    - $\rightarrow$  repeated measures t-test, Wilcoxon signed-rank test



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- # One-sample t-test
- > t.test(iq, mu=100)

```
# Independent samples t-test
> t.test(iq ~ sex)
```

```
# Repeated-measures t-test
> t.test(ms.regular, ms.exception, paired=T)
```



Important outputs: one-sample t-test

values you need to report: t value, degrees of freedom, p





Important outputs: independent samples t-test

values you need to report: t value, degrees of freedom, p



Important outputs: repeated measures t-test





# One sample: Mann-Whitney-Wilcoxon Test

```
> wilcox.test(iq, mu=100)
```

# Independent samples: Mann-Whitney-Wilcoxon Test
> wilcox.test(iq ~ sex)

# Repeated-measures: Wilcoxon Signed-Rank Test
> wilcox.test(ms.regular, ms.exception, paired=T)



Important outputs: Mann-Whitney-Wilcoxon test

(similar principle for Wilcoxon signed-rank test)





Null hypothesis: The means in all groups are equal.

One group-level variable  $\rightarrow$  one-way between-subjects ANOVA

Two or more group-level variables  $\rightarrow$  two-way between-subjects ANOVA



- Which pairwise differences between groups cause the significant group differences?
  - [Planned comparisons: contrasts (preferred)]
  - Post hoc comparisons



# 6. Comparing groups with ANOVA: post-hoc comparisons

- If ANOVA shows a surprising significant group difference, and you want to know where the pairwise differences are
- Several options for post-hoc comparisons
  - Pairwise t-test with correction for multiple comparisons



# 6. Comparing groups with ANOVA: one-way ANOVA

# Save and run anova model
> model.anova <- aov(ms.exception ~ reading\_time)</pre>

# Get sums of squares
> summary(model.anova)

# Get coefficients if needed
> summary.lm(model.anova)

# Take the means for each group
> tapply(ms.exception, reading\_time, mean)



# 6. Comparing group with ANOVA: post-hoc comparisons

# Bonferroni-corrected:

> pairwise.t.test(ms.exception, reading\_time,

p.adj="bonf")

# Look for other ways to correct for multiple comparisons

# using the same function!

# Tukey's test:

> TukeyHSD(model.anova)



# 6. Comparing groups with ANOVA: two-way ANOVA

# Save and run anova model
> model.anova.3 <- aov(ms.exception ~ reading\_time \* sex)</pre>

# Get sums of squares
> summary(model.anova.3)

# Get coefficients if needed
> summary.lm(model.anova.3)

# Plot the results
interaction.plot(model.anova.3)



Important outputs: summary for one-way ANOVA









Important outputs: results for contrast comparison
using summary.lm()

```
contrasts specified using
contrasts()
               > summary.lm(anova.model.2)
               Call:
               aov(formula = ms.exception ~ reading time)
               Residuals:
                                                               p value < .05 tells you the
                              1Q Median
                                                3Q
                    Min
                                                       Max
                                                               contrast is significant
               -231.781 -52.773 -2.025
                                            54.068 246.614
               Coefficients:
                             Estimate Std. Error t value Pr(>|t|)
                             1201.621
                                           2.455 489.442 < 2e-16 ***
                (Intercept)
                                           3.424 -8.142 1.15e-15
               reading time1 -27.879
               reading time2
                                                 4.004 6.68e-05
                               12.205
                                           3.048
                               0 `***' 0.001 `**' 0.01 `*' 0.05 `.' 0.1 ` ' 1
               Signif. codes:
               Residual standard error: 77.12 on 995 degrees of freedom
               Multiple R-squared: 0.07268, Adjusted R-squared: 0.07082
               F-statistic: 38.99 on 2 and 995 DF, p-value: < 2.2e-16
```

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Important outputs: post-hoc comparison using pairwise.t.test()

> pairwise.t.test(ms.exception, reading time, p.adj="bonf")

Pairwise comparisons using t tests with pooled SD

data: ms.exception and reading time



P value adjustment method: bonferroni

p values: p value < .05 tells you the pairwise difference is significant



Important outputs: post-hoc comparison using TukeyHSD()

```
p value < .05 tells you that the
                                           pairwise group difference is
    > TukeyHSD(model.anova)
                                           significant
      Tukey multiple comparisons of means
        95% family-wise confidence level
    Fit: aov(formula = ms.exception ~ reading time)
    $reading time
                     diff
                                lwr
                                                  p adj
                                          upr
    low-high
                 54.02305 39.56251 68.48359 0.0000000
    medium-high
                29.61292
                           16.05155 43.17428 0.0000011
                -24.41013 -38.71825 -10.10202 0.0001971
    medium-low
pairwise comparisons
```



Important outputs: summary for two-way ANOVA





Important outputs: interaction.plot()





#### 7. Regression

Does variance in one or more predictors account for variance in another variable?



### 7. Regression

```
# Model with one predictor:
> model1 <- lm(ms.regular ~ iq)</pre>
```

```
# Summary of the regression results:
> summary(model1)
```

```
# Model with two predictors:
> model2 <- lm(ms.regular ~ iq * hrs)</pre>
```

# Compare models:
> anova(model1, model2)


## 7. Regression

Important outputs: summary(model)





## 7. Regression

Important outputs: anova(model1,model2)





## 8. Extra topics: multiple comparisons

Goal: reduce the number of false positives given by the statistical test

E.g.

- Structural MRI: Testing for group differences in 10 different regions-ofinterest
- Functional MRI: testing for differences between conditions in the whole brain (i.e., running a separate linear model for each voxel)
- Genomics: testing for multiple genes, environmental factors, their interactions...

http://www.biostathandbook.com/multiplecomparisons.html





- Good luck with the exam
- Remember to give feedback (Oodi)
- See you in spring 2015 at Experimental and Statistical Methods in Biological Sciences II



## **Our final dataset: NHEFS**

- Personality variables (continuous):
  - Neuroticism
  - Extraversion
  - Openness
- Depression variable (continuous):
  - CES-D
- Health variables (categorical):
  - Heart disease
  - Cancer
  - Stroke
  - Diabetes
  - Hypertension

