Biostatistics
Week X

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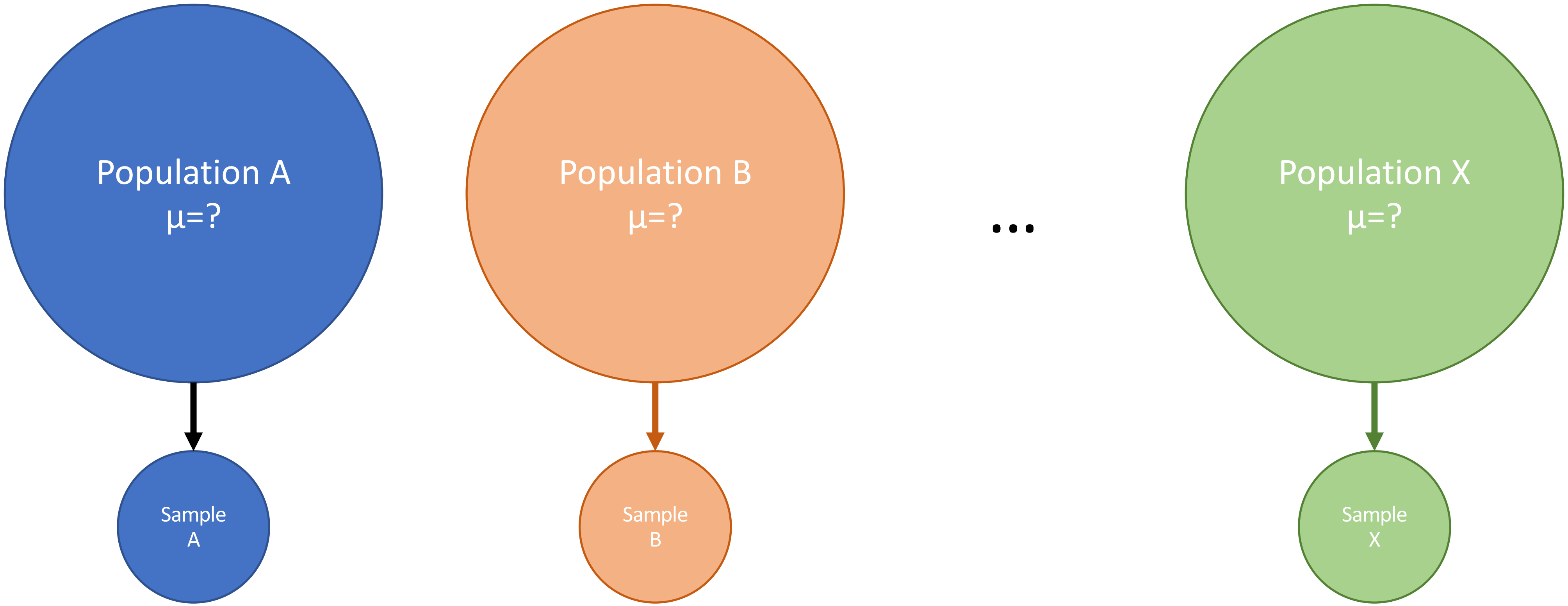
ACIBADEM
MEHMET ALİ AYDINLAR
ÜNİVERSİTESİ

Hypothesis Testing - Steps

1. **Check assumptions, determine H_0 and H_a, choose α**
    - Assumptions differ based on the test
    - The null hypothesis always contains equality (=)
2. **Calculate the appropriate test statistic**
    - z, t, χ^2, …
3. **Calculate critical values/p value**
    - With the aid of precalculated tables/software
4. **Decide whether to reject/fail to reject H_0**
    - Reject if the statistic is within the critical region/$p \leq \alpha$

Analysis of Variance (ANOVA)

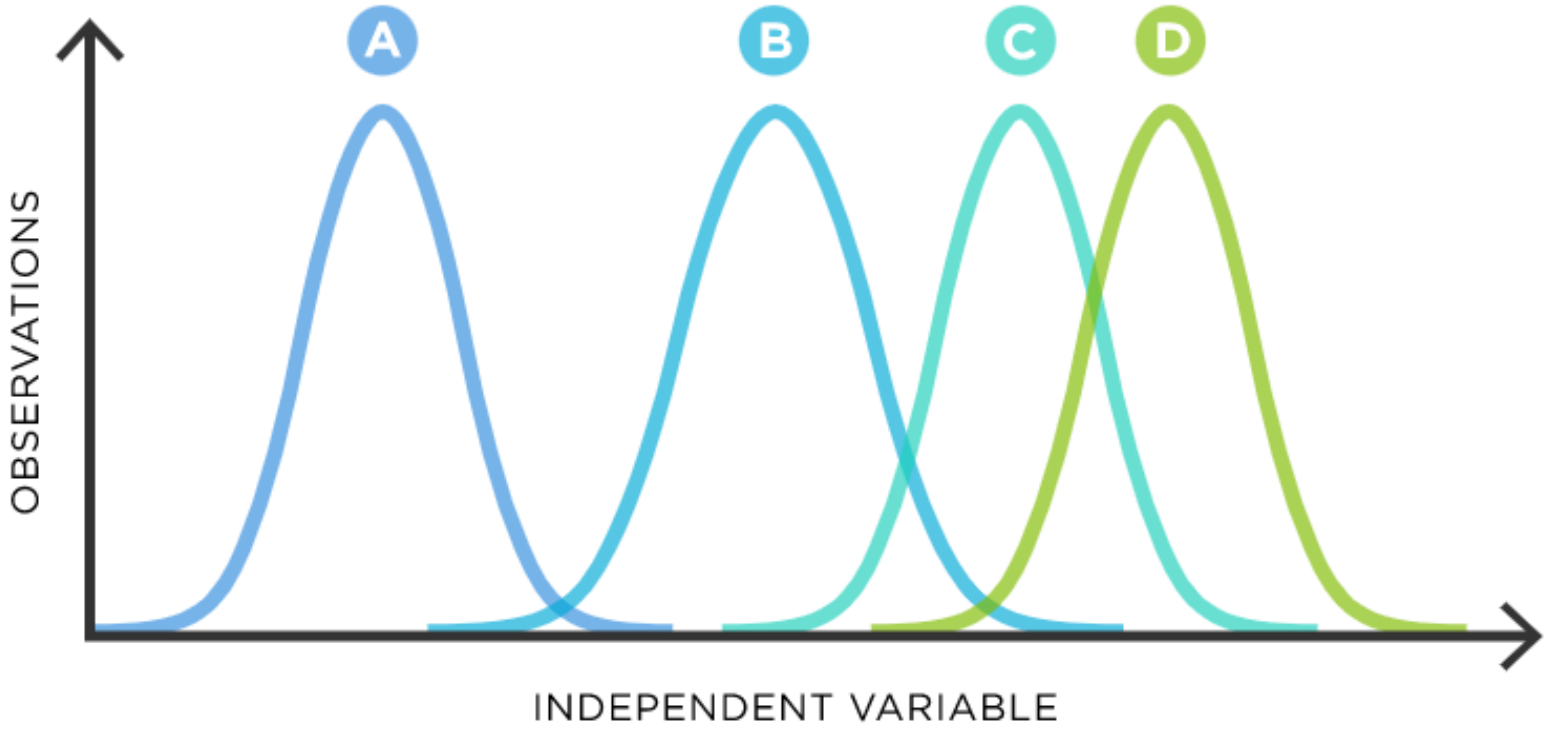
- Analysis of variance (ANOVA) is a statistical technique that is used to check if the means of **two or more groups** are significantly different from each other



One-way ANOVA

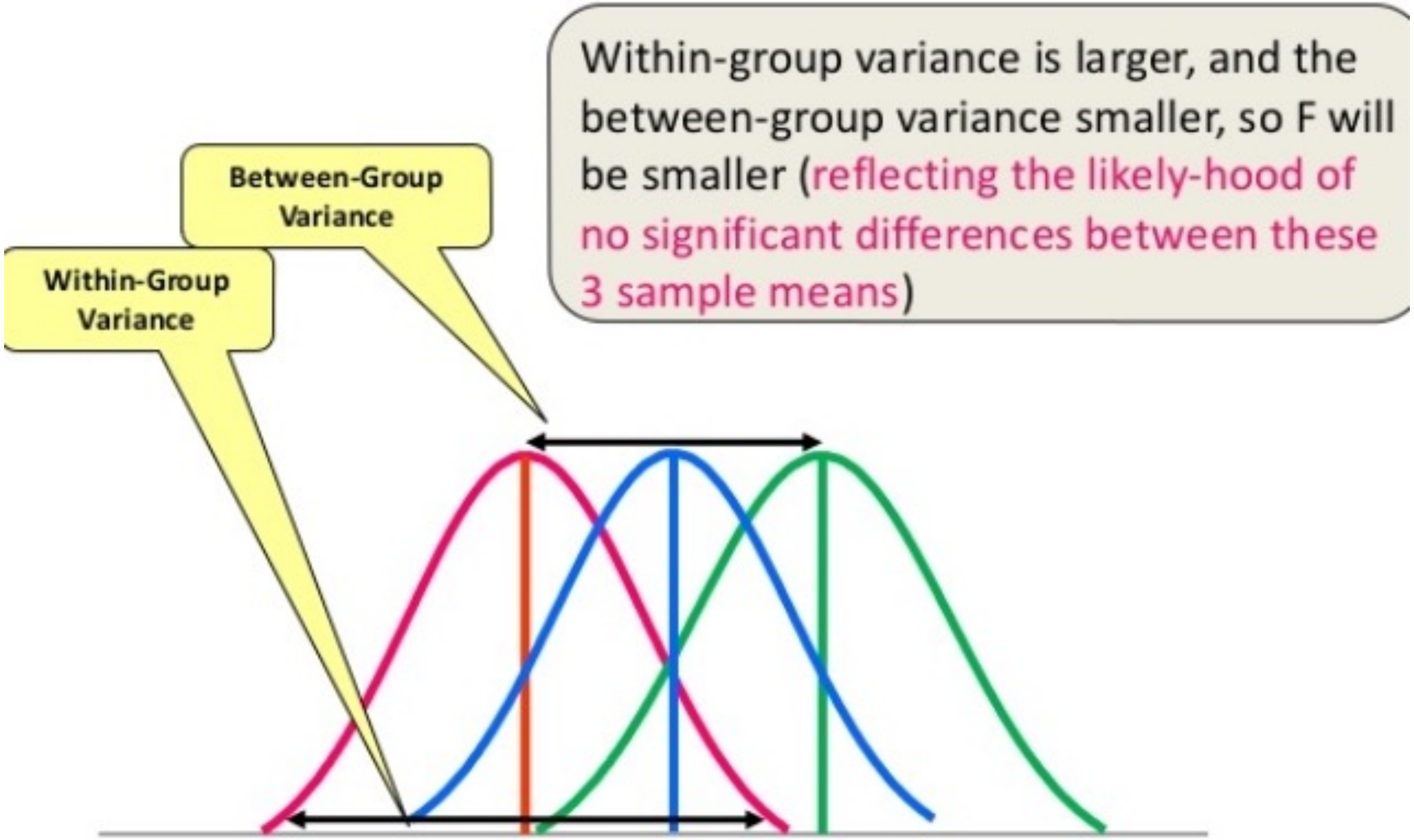
H_0: $\mu_1 = \mu_2 = \ldots = \mu_n$

H_a: at least one μ_i is different



Analysis of variance (Anova) | introduction, types & techniques [Internet]. Analytics Vidhya. 2018 [cited 2021 Oct 6]. Available from: https://www.analyticsvidhya.com/blog/2018/01/anova-analysis-of-variance/

ANOVA



One-way ANOVA

k: number of groups
n: total number of samples
n_i: number of samples in group i

Analysis of Variance(ANOVA)

Source of Variation	Sum of Squares	Degrees of Freedom	Mean Squares (MS)	F
Between	$\sum n_i(\bar{X}_i - \bar{X})^2$	k - 1	SS_b/df_b	$\boxed{F = \frac{MS_b}{MS_w}}$
Within	SS_T - SS_b	n - k	SS_w/df_w	
Total	$\sum (X_j - \bar{X})^2$	n - 1		

How to perform analysis of variance (Anova) - step by step procedure [Internet]. The Genius Blog. 2018 [cited 2021 Oct 6]. Available from: https://kindsonthegenius.com/blog/how-to-perform-analysis-of-variance-anova-step-by-step-procedure/

One-way ANOVA – Example I

Table 1: Percentage benefits for 5 patients from each treatment groups.

Treatment 1	Treatment 2	Treatment 3	Treatment 4
-7.2	-13.0	-3.8	7.0
2.5	-0.4	-2.7	1.5
1.4	-1.6	5.3	9.4
-0.7	4.9	-5.9	9.5
-0.9	-0.7	3.7	9.9

The hypothesis of interest is

$H_0 : \mu_1 = \mu_2 = \mu_3 = \mu_4$
$H_1 :$ at least one is different from the others

One-way ANOVA – Example I (cont.)

1. Check assumptions, determine H_0 and H_a, choose α
    - Check that data is normally distributed
    - H_0: $\mu_1 = \mu_2 = \mu_3 = \mu_4$ H_a: at least one mean is different
    - $\alpha = 0.05$

One-way ANOVA – Example I (cont.)

2. Calculate the appropriate test statistic

Sources of variation	Sum of squares	degrees-of-freedom	Mean squared error	F	p-value
Between treatment					
Within treatment					
Total					

One-way ANOVA – Example I (cont.)

2. Calculate the appropriate test statistic

Step 1: Calculate the treatment means and grand mean:

$$\bar{x}_1 = \frac{-7.2+2.5+1.4+(-0.7)+(-0.9)}{5} = -0.98$$
$$\bar{x}_2 = \frac{-13.0+(-0.4)+(-1.6)+4.9+(-0.7)}{5} = -2.16$$
$$\bar{x}_3 = \frac{-3.8+(-2.7)+(5.3)+(-5.9)+3.7}{5} = 0.68$$
$$\bar{x}_4 = \frac{7.0+1.5+9.4+9.5+9.9}{5} = 7.46$$
$$\bar{x} = \frac{-7.2+...+(-0.9)+(-13.0)+...(-0.7)+(-3.8)+...+3.7+7.0+...+9.9}{20} = 0.91$$

One-way ANOVA – Example I (cont.)

2. Calculate the appropriate test statistic

Step 3: Calculate between treatment sum of squared error:

$$5\ (-0.98 - 0.91\ )^2 + 5(-2.16 - 0.91)^2 + 5(0.68 - 0.91)^2 + 5(7.46 - 0.91)^2 = 292.138$$

Step 4: Calculate the total sum of squared error:

$$(-7.2 - 0.91)^2 + \ldots + (-0.9 - 0.91)^2 + (-13.0 - 0.91)^2 + \ldots (-0.7 - 0.91)^2 + (-3.8 - 0.91)^2 + \ldots + (3.7 - 0.91)^2 + (7.0 - 0.91)^2 + \ldots + (9.9 - 0.91)^2 = 667.198$$

Step 5: Calculate the within-group sum of squared error as $667.198 - 292.138 = 375.06$

One-way ANOVA – Example I (cont.)

2. Calculate the appropriate test statistic

Step 6: Total d.o.f.: 20 - 1, 19; between treatment d.o.f: 4-1=3; within treatment d.o.f.: 19-3=16

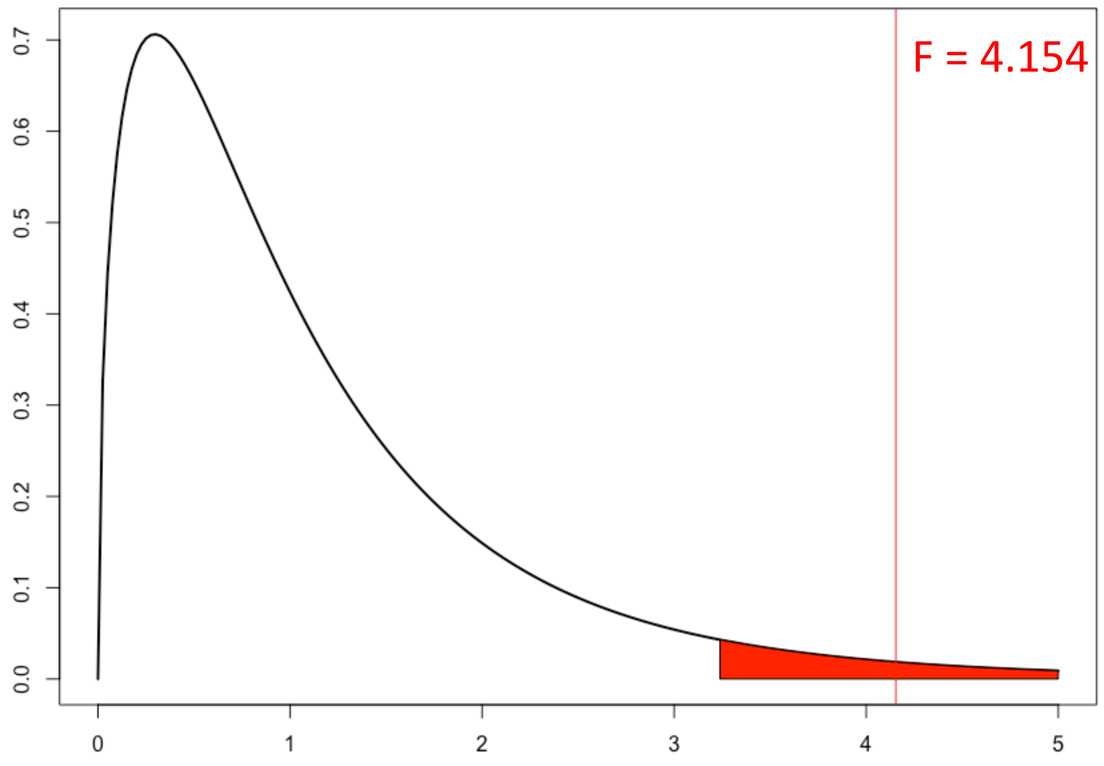
Step 7: Calculate mean suqared error for between treatment as 292.138/3=97.38

Step 8: Calculate mean squared error for within treatment as 375.06.198/16=23.44

Step 9: Calculate F value as 97.38/23.44=4.154

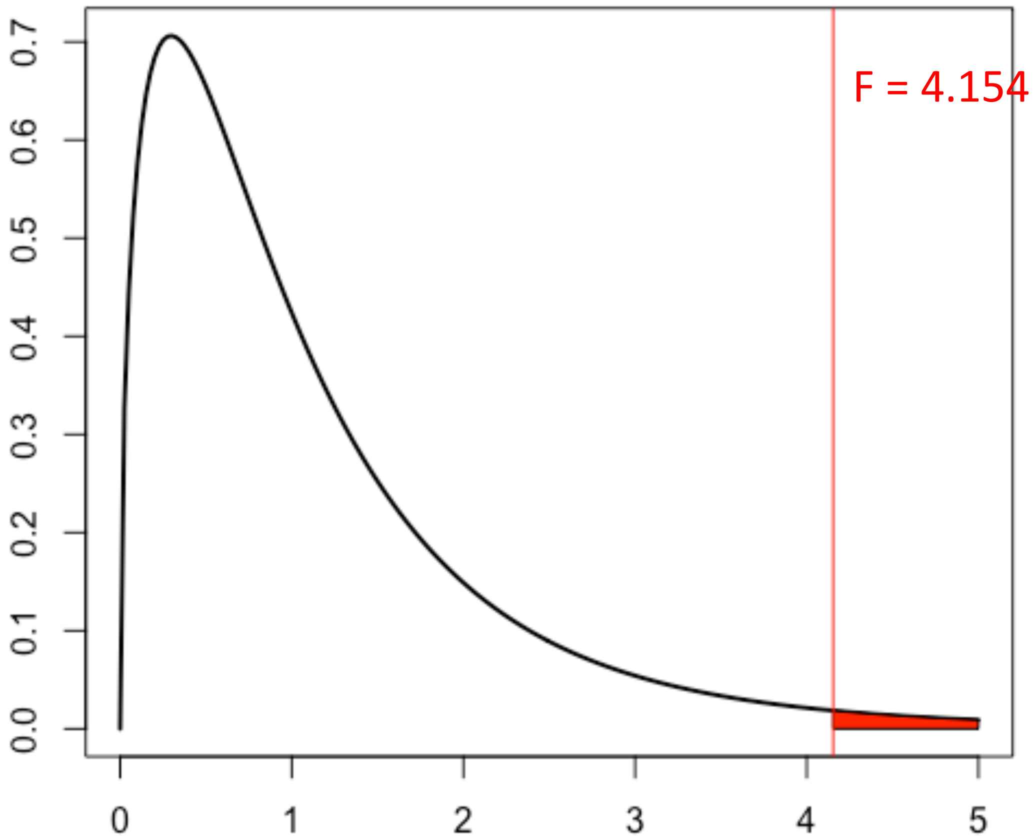
One-way ANOVA – Example II (cont.)

3. Calculate **rejection zone**/p value
4. Decide whether to reject/fail to reject H_0



One-way ANOVA – Example II (cont.)

3. Calculate rejection zone/**p value**
4. Decide whether to reject/fail to reject H_0



p=0.023516

One-way ANOVA – Example II

THE LANCET, AUGUST 12, 1978

MEGALOBLASTIC HÆMOPOIESIS IN PATIENTS RECEIVING NITROUS OXIDE

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- 22 patients who underwent coronary artery bypass graft surgery (CABG) are separated into 3 different treatment groups (different ventilation strategies)
- Is there a difference in red blood cell folic acid measurements at 24 hours between the 3 treatment groups?

One-way ANOVA – Example II (cont.)

Group I.—8 patients received approximately 50% nitrous oxide and 50% oxygen mixture continuously for 24 h. 1 patient received 2000 μg of hydroxocobalamin intramuscularly immediately before and after the operation.

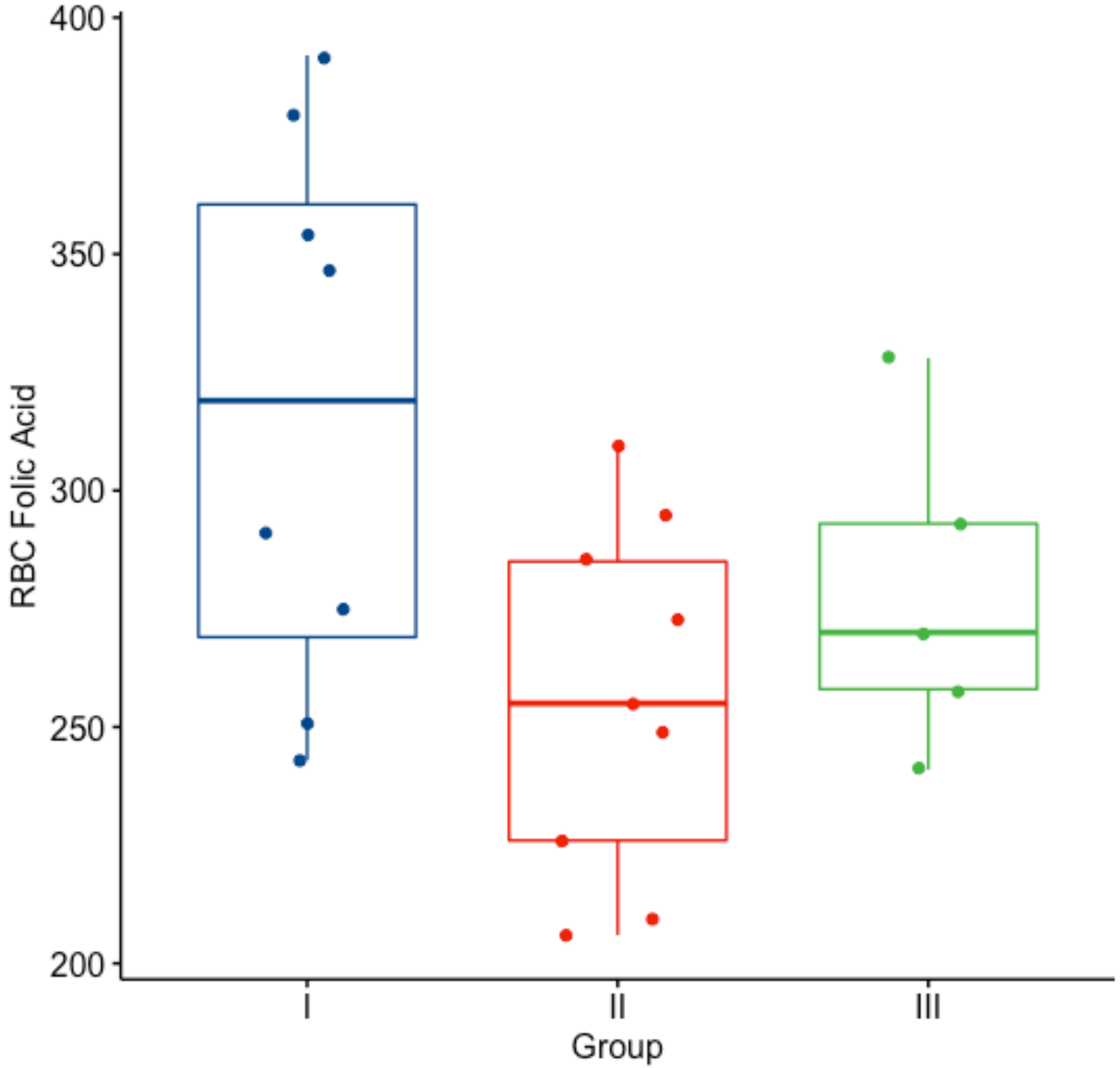
Group II.—9 patients received approximately 50% nitrous oxide and 50% oxygen mixture only during the operation (5–12 h) and thereafter 35–50% oxygen for the remainder of the 24 h period.

Group III.—5 patients received no nitrous oxide but were ventilated with 35–50% oxygen for 24 h.

Group I	Group II	Group III
243	206	241
251	210	258
275	226	270
291	249	293
347	255	328
354	273	
380	285	
392	295	
	309	

Amess JA, Burman JF, Rees GM, Nancekievill DG, Mollin DL. Megaloblastic haemopoiesis in patients receiving nitrous oxide. Lancet. 1978 Aug 12;2(8085):339–42.

One-way ANOVA – Example II (cont.)



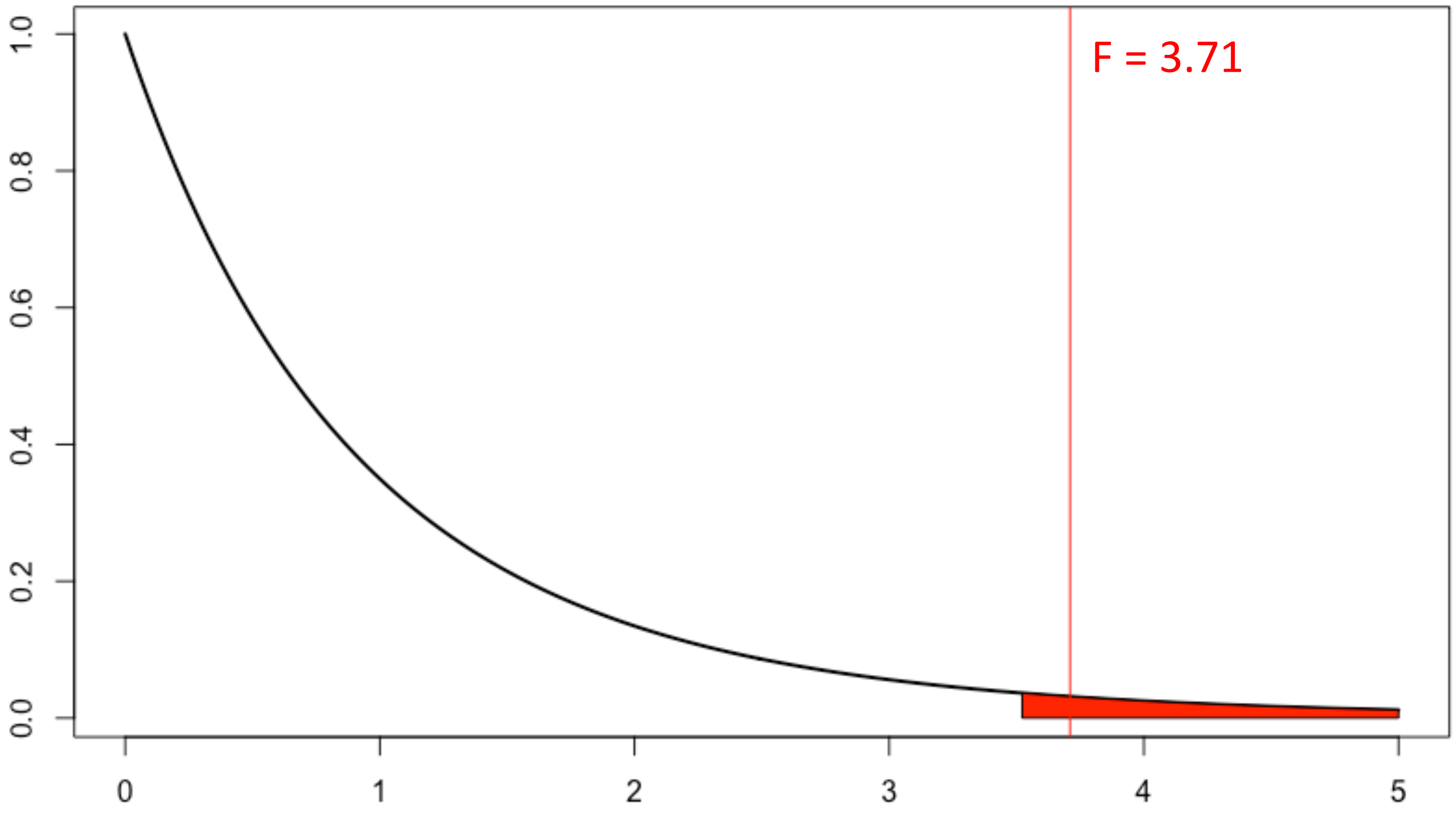
One-way ANOVA – Example II (cont.)

1. Check assumptions, determine H_0 and H_a, choose α
   - Check that data is normally distributed
   - H_0: $\mu_1 = \mu_2 = \mu_3$ H_a: at least one mean is different
   - $\alpha = 0.05$

2. Calculate the appropriate test statistic
   - F = 3.71 $\sim F_{2,19}$

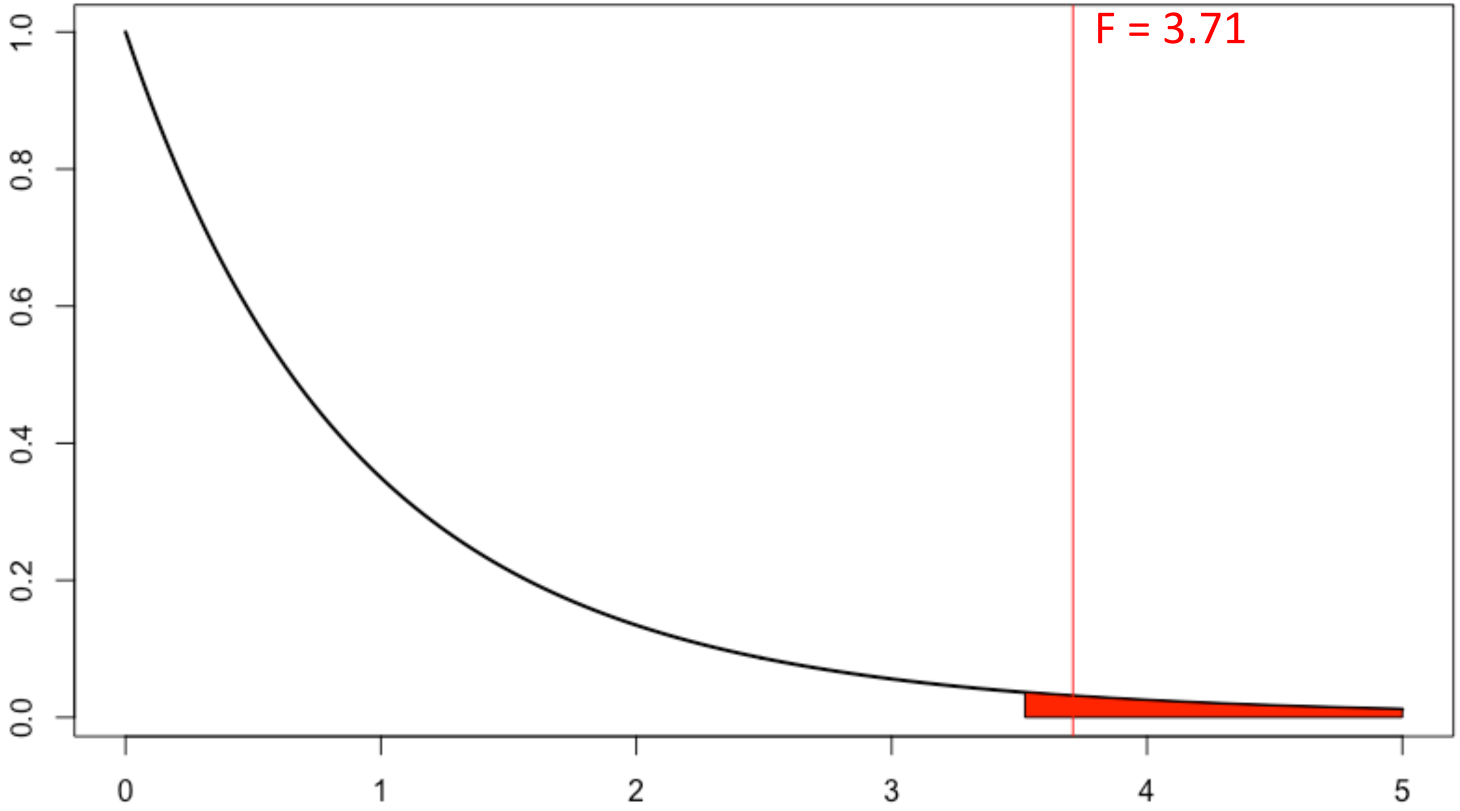
One-way ANOVA – Example II (cont.)

3. Calculate **critical values**/p value
4. Decide whether to reject/fail to reject H_0



One-way ANOVA – Example II (cont.)

3. Calculate critical values/**p value**
4. Decide whether to reject/fail to reject H_0

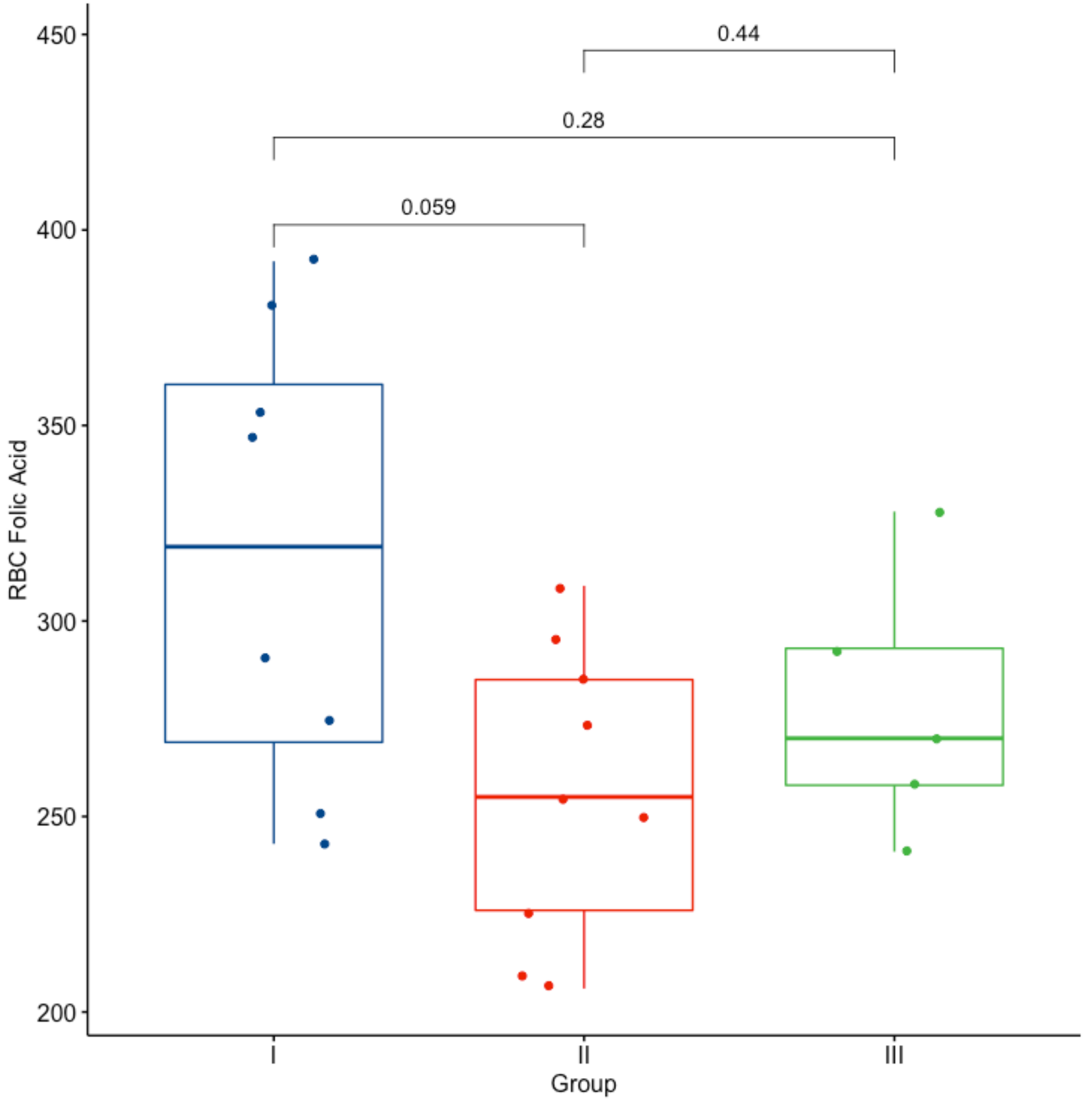


p = 0.043631

One-way ANOVA – Example II (cont.)

- With 95% confidence, we can conclude that the mean RBC folic acid level of at least one group is significantly different than the others
- Next, we perform 2-sample t-tests between all pairs of groups

One-way ANOVA – Example II (cont.)



Variations of ANOVA

- Two-way ANOVA – effect of 2 independent variables on one dependent variable
- Multivariate ANOVA (MANOVA) – effect of independent variable(s) on multiple dependent variables

- Analysis of Covariance (ANCOVA) - compares a dependent variable by both a factor and a continuous independent variable
- MANCOVA
- …

χ^2 Test for Independence

- Used to assess the association between two categorical variables
- More generally, used to investigate the significance of the difference between expected and observed values

- Are the 2 categorical variables **independent**?

χ^2 Test – Test Statistic

$$\chi^2 = \sum \frac{(observed\ - expected)^2}{expected}$$

Cupples ME, McKnight A. Randomised controlled trial of health promotion in general practice for patients at high cardiovascular risk. BMJ. 1994 Oct 15;309(6960):993–6.

χ^2 Test – Example

TABLE III—*Changes in frequency of physical exercise in patients with angina between baseline and review at two years*

	No (%) of patients	
	Intervention group	Control group
Increased	108 (34)	63 (21)
No change	120 (38)	74 (25)
Decreased	89 (28)	163 (54)

χ^2 Test – Example

	Intervention Group	Control Group	Total
Increased	108	63	**171**
No change	120	74	**194**
Decreased	89	163	**252**
Total	**317**	**300**	**617**

$$expected_{1,1} = 317 \times \frac{171}{617} \qquad expected_{1,2} = 300 \times \frac{171}{617}$$

$$expected_{2,1} = 317 \times \frac{194}{617} \qquad expected_{2,2} = 300 \times \frac{194}{617}$$

$$expected_{3,1} = 317 \times \frac{252}{617} \qquad expected_{3,2} = 300 \times \frac{252}{617}$$

χ^2 Test – Example

OBSERVED	Intervention Group	Control Group
Increased	108	63
No change	120	74
Decreased	89	163

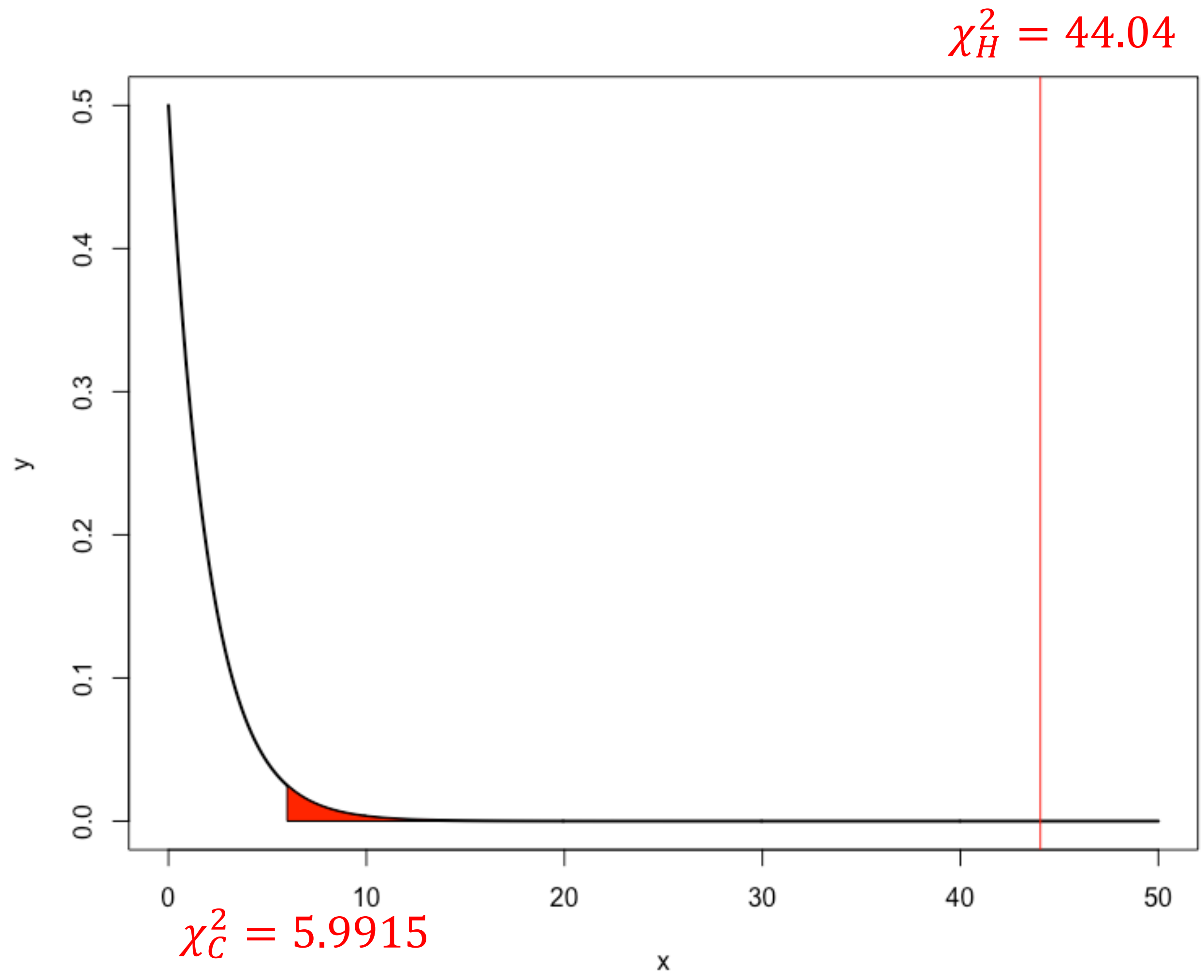
EXPECTED	Intervention Group	Control Group
Increased	87.86	83.14
No change	99.67	94.33
Decreased	139.47	122.53

χ^2 Test – Test Statistic

$$\chi^2 = \sum \frac{(observed \ - expected)^2}{expected}$$

$$\chi_H^2 = 44.04 \ \ \sim\chi_{(3\ -1)(2\ -1)=2}^2$$

χ^2 Test – Test Statistic



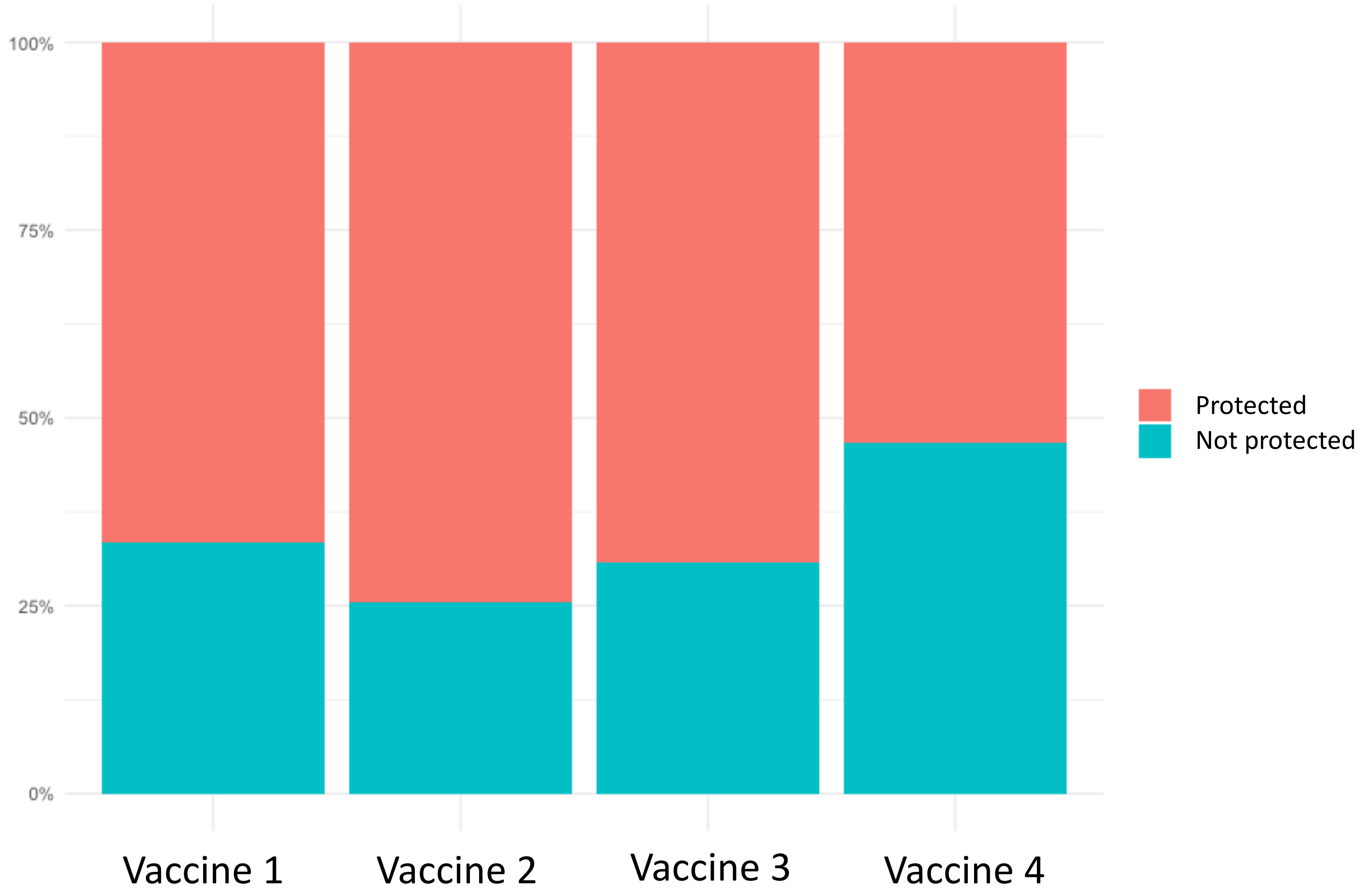
$p < 0.001$

χ^2 Test – Example

- Is there an association between vaccine type and protection status?

	Protected	Not protected
Vaccine 1	82	41
Vaccine 2	70	24
Vaccine 3	45	20
Vaccine 4	48	42

χ^2 Test – Example



χ^2 Test – Example

1. Check assumptions, determine H_0 and H_a, choose α
   - H_0: there is **no difference** in efficacy
     H_a: there is a difference in efficacy
   - $\alpha = 0.05$

2. Calculate the appropriate test statistic

$$\chi_H^2 = 9.297 \sim\chi_3^2$$

χ² Test – Example

	Proctected	Not protected	Total
Vaccine 1	82	41	**123**
Vaccine 2	70	24	**94**
Vaccine 3	45	20	**65**
Vaccine 4	48	42	**90**
Total	**245**	**127**	**372**

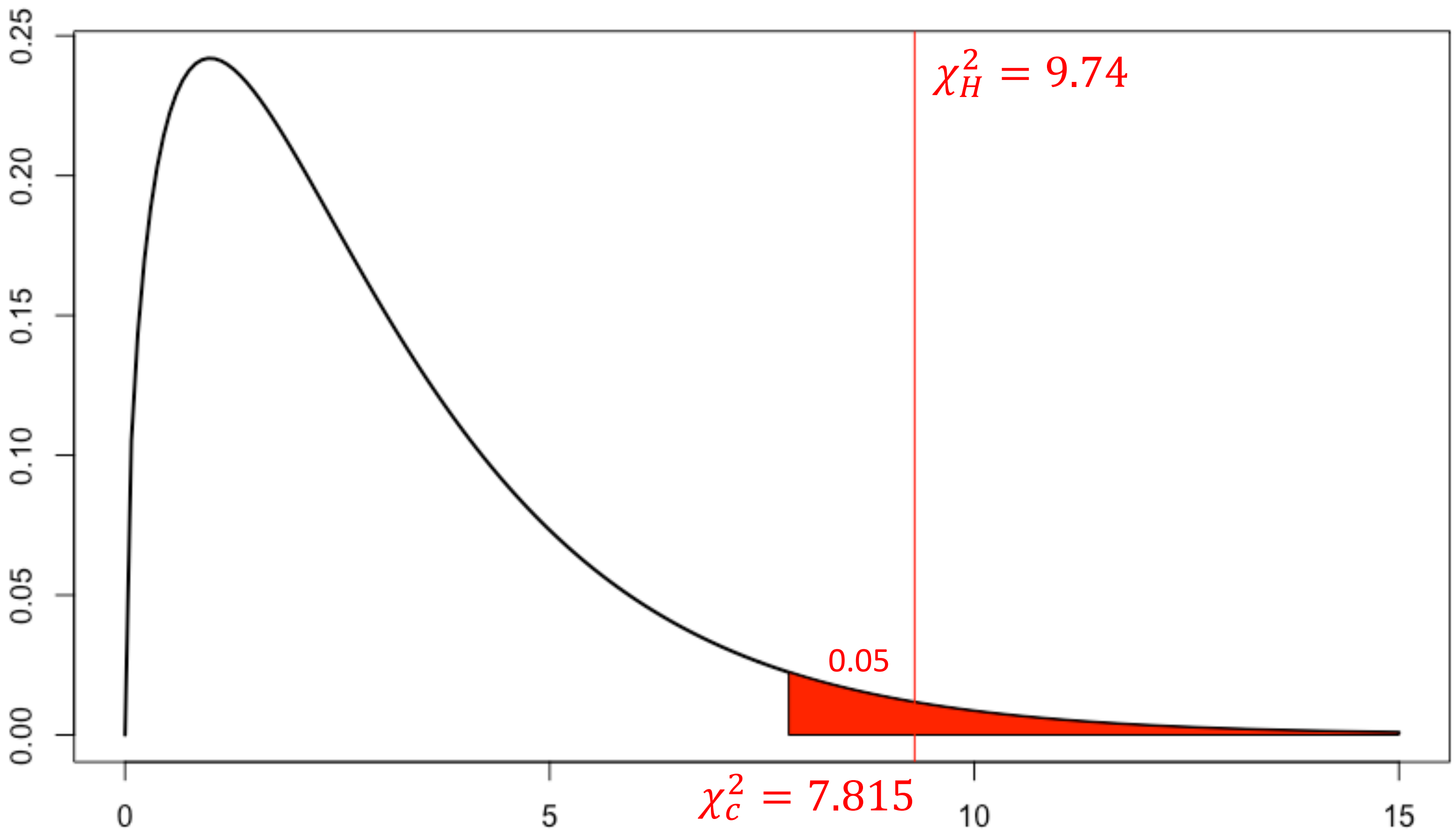
$$expected_{4,1} = 245 \times \frac{90}{372} = 59$$

$$expected_{4,2} = 127 \times \frac{90}{372} = 31$$

$$\chi_H^2 = \sum_{j=1}^{m} \sum_{i=1}^{n} \frac{(observed_{ij} - expected_{ij})^2}{expected_{ij}} \sim \chi^2_{(m-1)(n-1)}$$

$$\boldsymbol{\chi_H^2 = 9.74 \sim \chi_3^2}$$

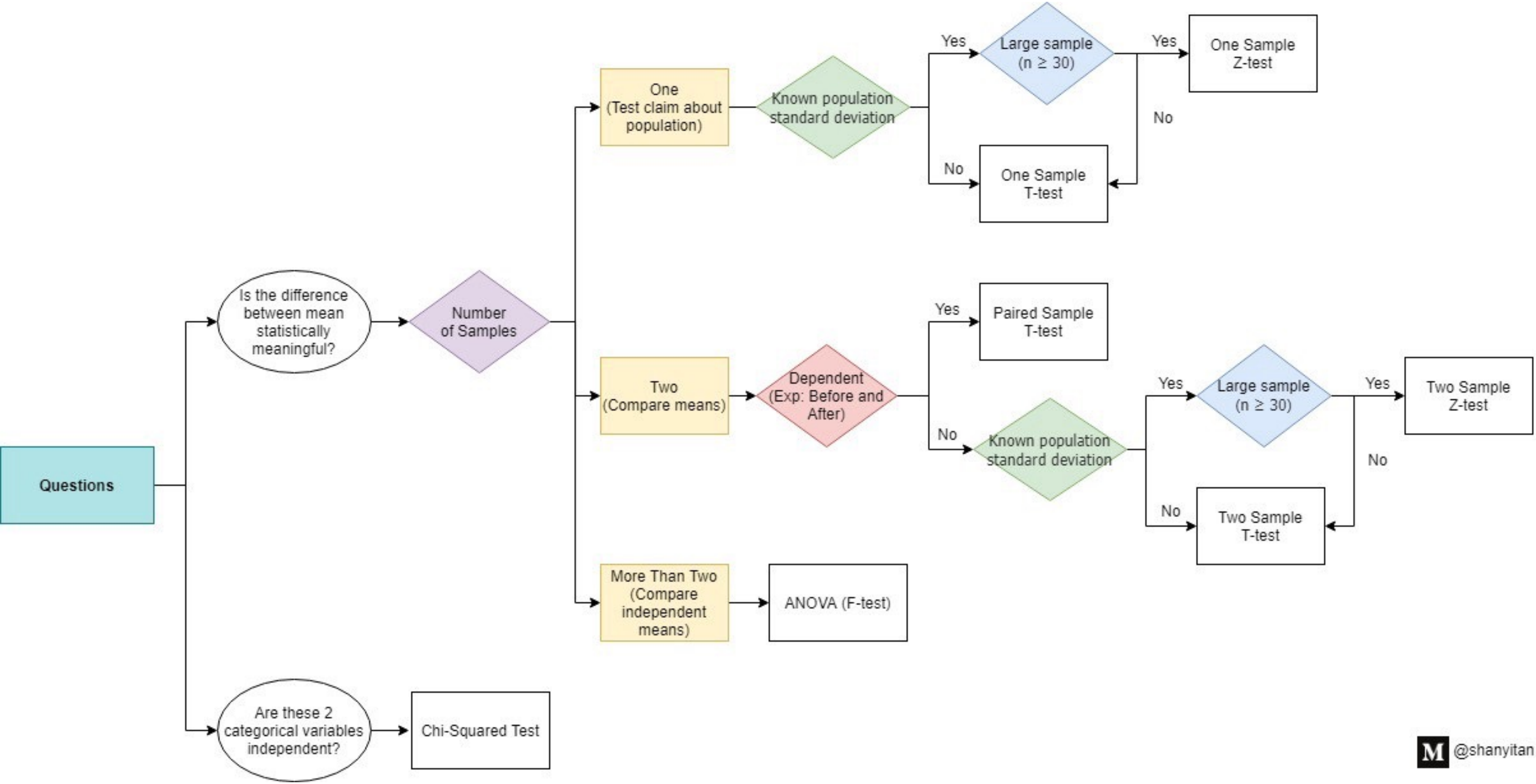
χ^2 Test – Example



p = 0.021

*χ^2 Goodness of Fit Test

- Decide if one variable is likely to come from a given distribution or not


Questions
Is the difference between mean statistically meaningful?
Number of Samples
One (Test claim about population)
Known population standard deviation
Yes
Large sample (n ≥ 30)
Yes
One Sample Z-test
No
No
One Sample T-test
Two (Compare means)
Dependent (Exp: Before and After)
Yes
Paired Sample T-test
No
Known population standard deviation
Yes
Large sample (n ≥ 30)
Yes
Two Sample Z-test
No
No
Two Sample T-test
More Than Two (Compare independent means)
ANOVA (F-test)
Are these 2 categorical variables independent?
Chi-Squared Test
@shanyitan