Proučavanje povezanosti između kontinuiranih varijabli

19.01.2024.

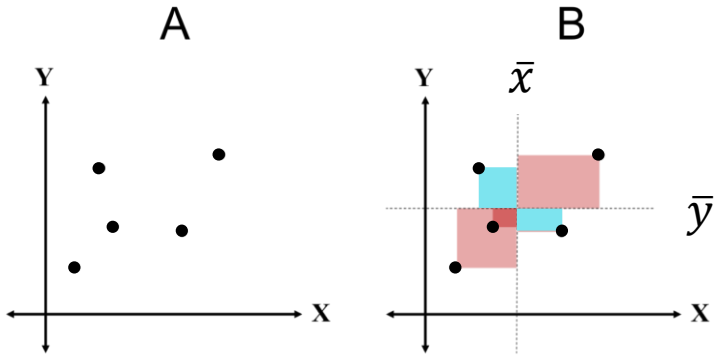
Rosa Karlić

Dva moguća cilja

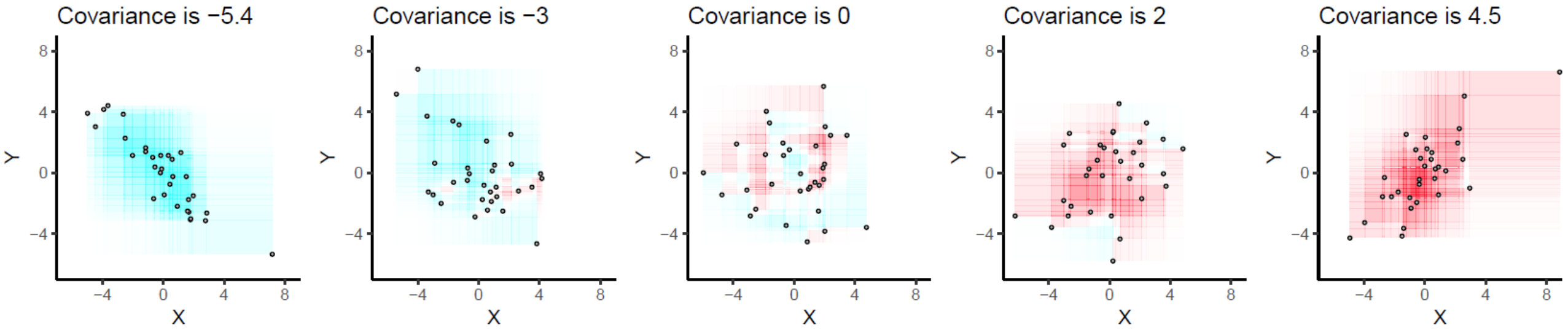
- Opisati odnose između dvije ili više kontinuiranih varijabli
- Koristiti navedene odnose za predviđanje vrijednosti varijabli

Kovarijanca

- Kovarijanca – koliko se jedna varijabla mijenja kad se druga varijabla mijenja σ_{XY}
- Kovarijanca uzorka: $s_{XY} = \frac{\sum_{i=1}^{n}(x_i - \bar{x})(y_i - \bar{y})}{n-1}$



Kovarijanca

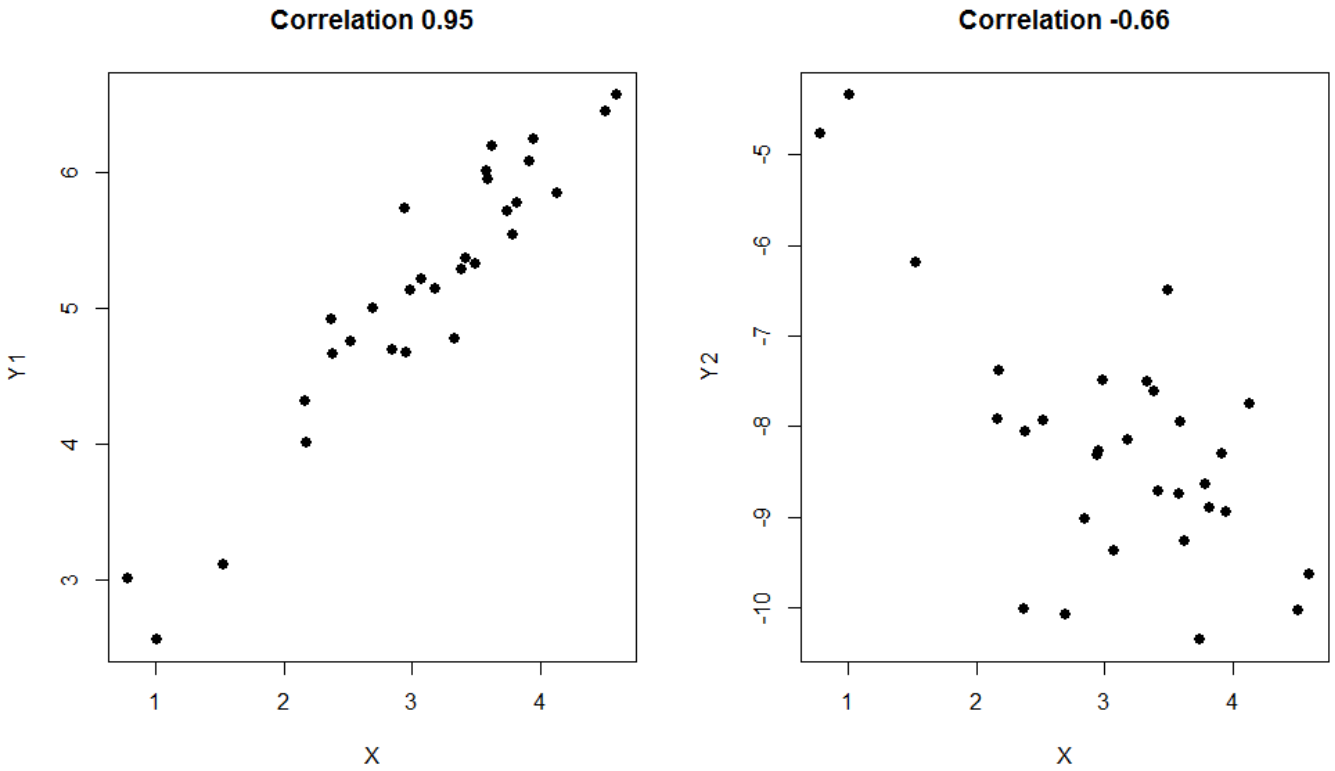


Izvor: https://stats.stackexchange.com/questions/18058/how-would-you-explain-covariance-to-someone-who-understands-only-the-mean

- Proporcionalna skali na kojoj su mjereni X i Y
- Osjetljiva na *outliere* (netipične vrijednosti)

Korelacija

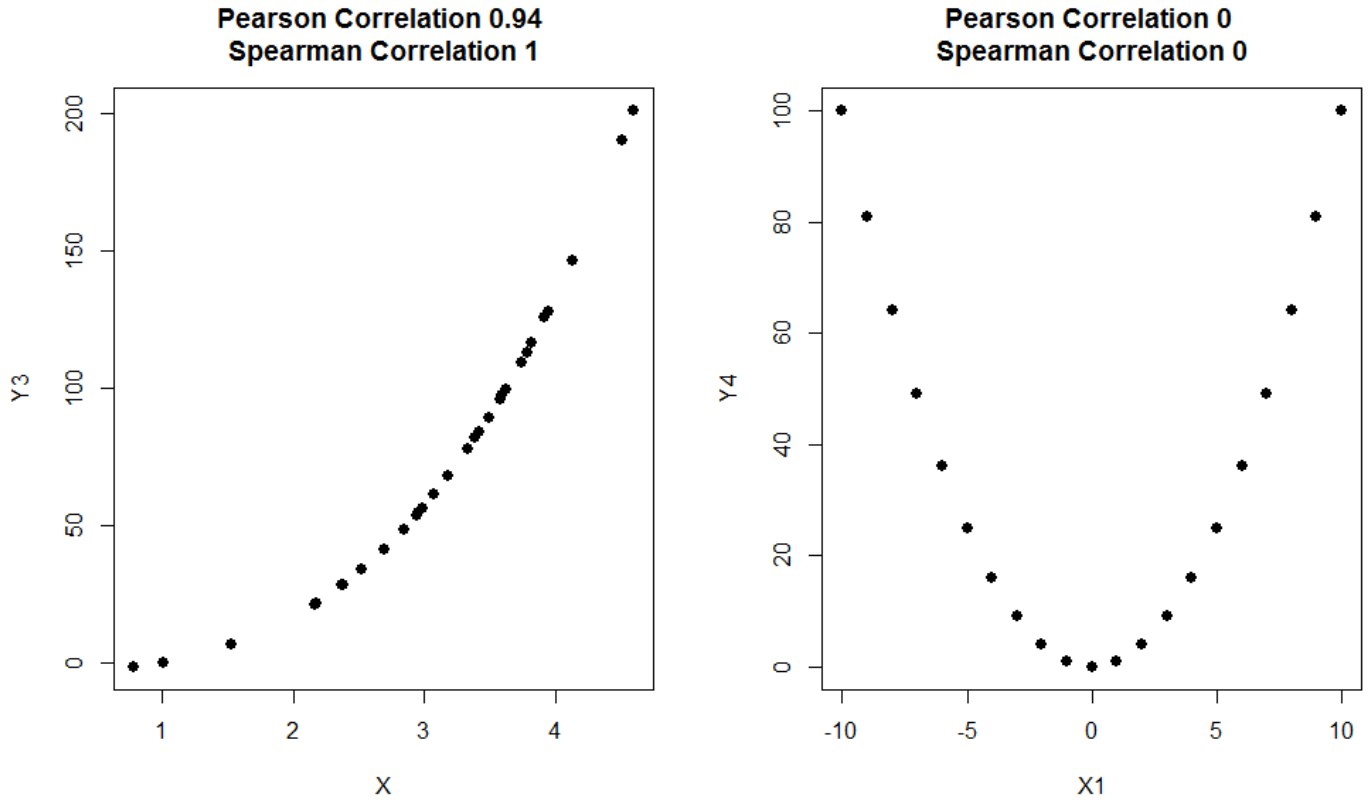
- Korelacija – kovarijanca normalizirana standardnom devijacijom
- Koeficijent korelacije – mjeri snagu odnosa između dviju varijabli
- Pearsonov koeficijent korelacije – linearni odnosi



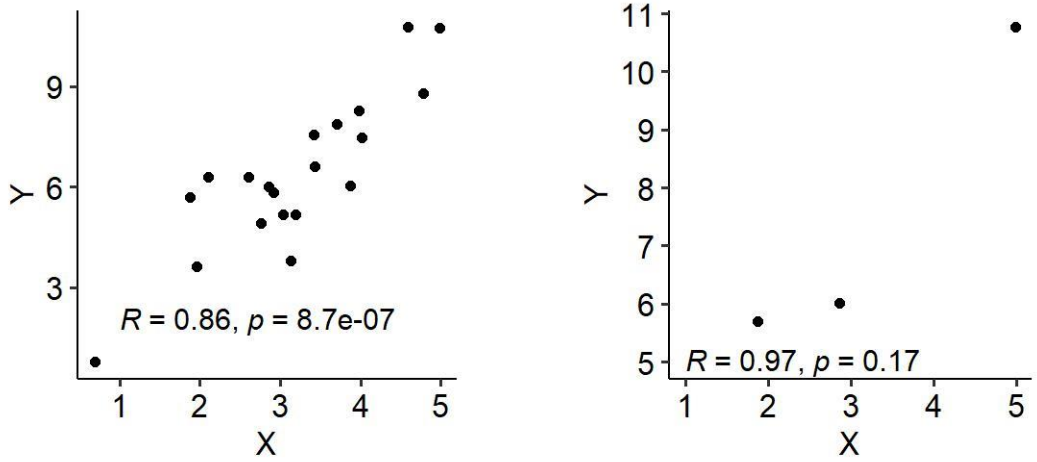
$$\rho = \frac{Cov(X,Y)}{\sigma_X \sigma_Y}$$

Spearmanov koeficijent korelacije

- Pearsonov koeficijent korelacije nakon što su vrijednosti varijabli pretvorene u rangove
- Nulta hipoteza: nema monotonog odnosa između dvije varijable



Značajna povezanost među varijablama?

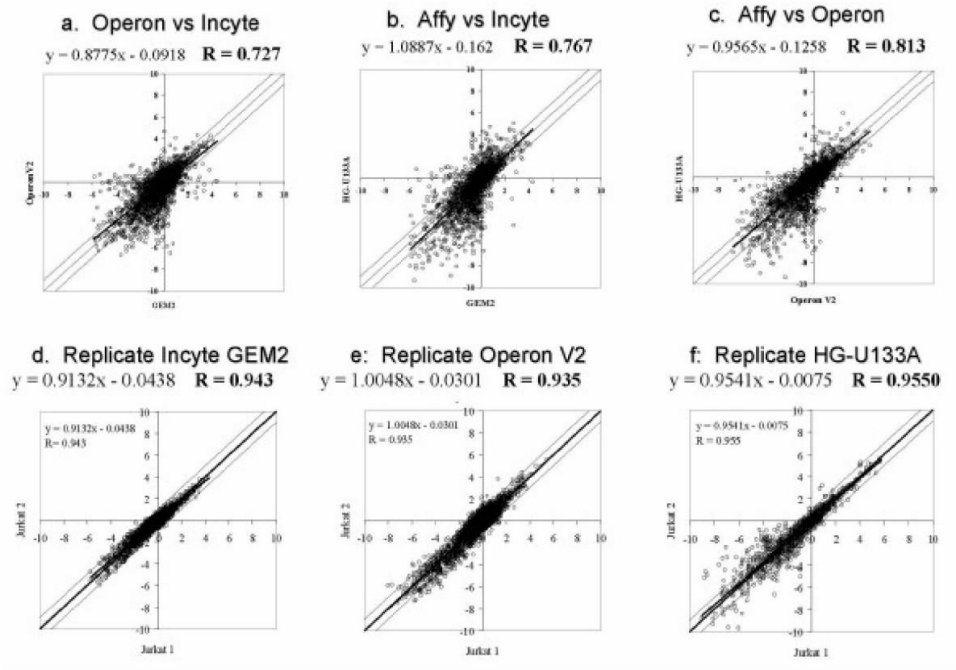


ABSOLUTE VALUE OF R	INTERPRETATION
< 0.19	Slight; almost no relationship
0.20–0.39	Low correlation; definite but small relationship
0.40–0.69	Moderate correlation; substantial relationship
0.70–0.89	High correlation; strong relationship
0.90–1.00	Very high correlation; very dependable relationship
≥ 0.30	Practically significant relationship

Izvor: https://doi.org/10.4102/sajhrm.v7i1.175

- Koeficijent determinacije = r^2 jačina povezanosti

Korelacija u biološkim eksperimentima



- **a-f**. Scatter plot analysis to determine correlation coefficients between and within platforms using Jurkat RNA as an example. Correlations for all cell lines are given in Table 4. (a) Operon versus Incyte (b) Affymetrix versus Incyte (c) Affymetrix versus Operon (d) GEM2 versus GEM2 replicate correlation (e) Operon versus Operon (f) HG-U133A versus HG-U133A

Petersen, D., Chandramouli, G., Geoghegan, J. *et al.* Three microarray platforms: an analysis of their concordance in profiling gene expression. *BMC Genomics* **6**, 63 (2005). https://doi.org/10.1186/1471-2164-6-63

Korelacija u biološkim eksperimentima

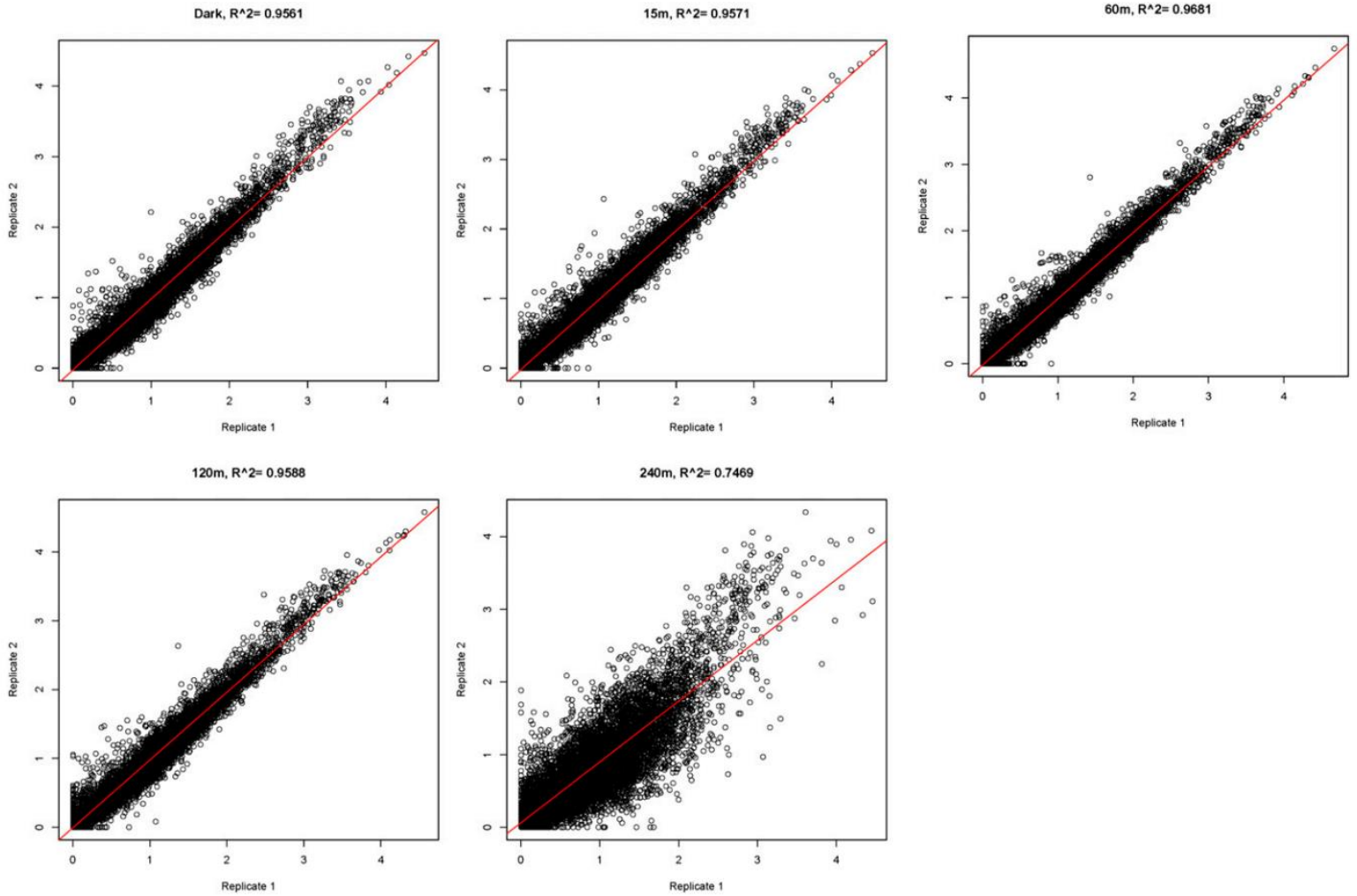


Figure 1 Comparison of RNA-seq replicate experiments. The FPKM for biological replicate 1 is plotted against biological replicate 2 for each gene, demonstrating strong correlation between replicate experiments at each time point. The correlation coefficient, R, is shown for each time point.

Korelacija – uzorkovanje iz više različitih populacija

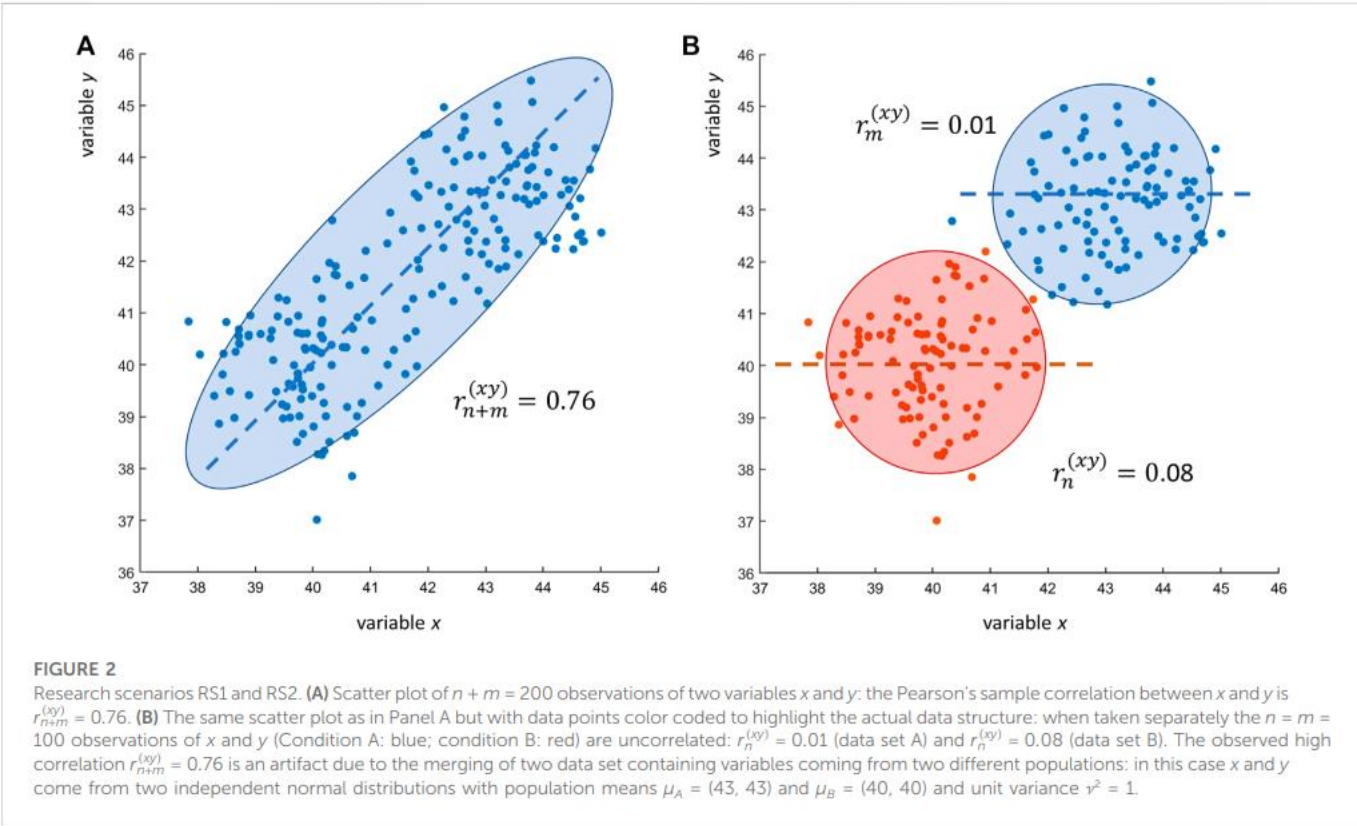


FIGURE 2
Research scenarios RS1 and RS2. **(A)** Scatter plot of $n + m = 200$ observations of two variables x and y: the Pearson's sample correlation between x and y is $r_{n+m}^{(xy)} = 0.76$. **(B)** The same scatter plot as in Panel A but with data points color coded to highlight the actual data structure: when taken separately the $n = m = 100$ observations of x and y (Condition A: blue; condition B: red) are uncorrelated: $r_n^{(xy)} = 0.01$ (data set A) and $r_n^{(xy)} = 0.08$ (data set B). The observed high correlation $r_{n+m}^{(xy)} = 0.76$ is an artifact due to the merging of two data set containing variables coming from two different populations: in this case x and y come from two independent normal distributions with population means $\mu_A = (43, 43)$ and $\mu_B = (40, 40)$ and unit variance $\nu^2 = 1$.

https://www.frontiersin.org/articles/10.3389/fsysb.2023.1042156/full

Korelacija – uzorkovanje iz više različitih populacija

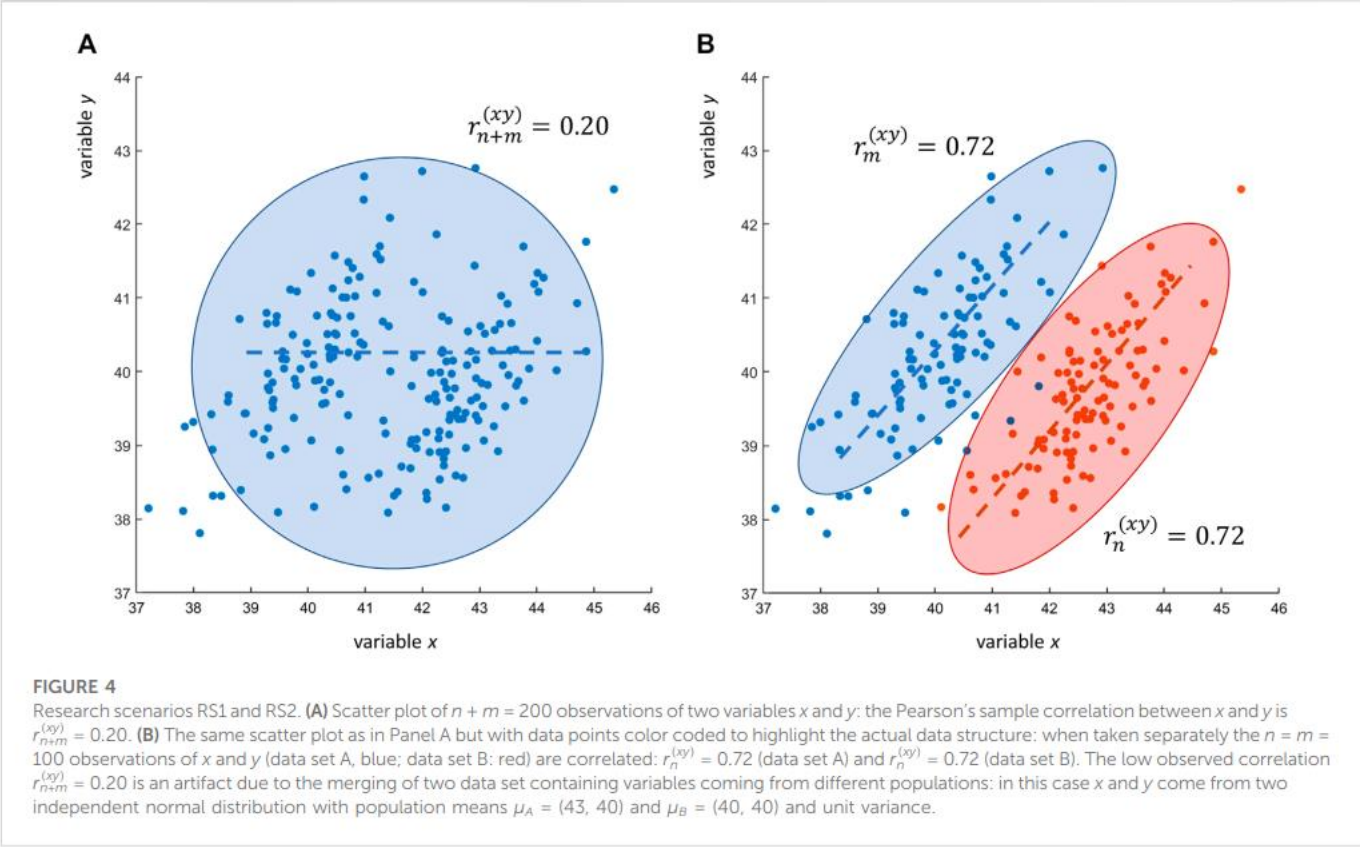


FIGURE 4
Research scenarios RS1 and RS2. **(A)** Scatter plot of $n + m = 200$ observations of two variables x and y: the Pearson's sample correlation between x and y is $r_{n+m}^{(xy)} = 0.20$. **(B)** The same scatter plot as in Panel A but with data points color coded to highlight the actual data structure: when taken separately the $n = m = 100$ observations of x and y (data set A, blue; data set B: red) are correlated: $r_n^{(xy)} = 0.72$ (data set A) and $r_n^{(xy)} = 0.72$ (data set B). The low observed correlation $r_{n+m}^{(xy)} = 0.20$ is an artifact due to the merging of two data set containing variables coming from different populations: in this case x and y come from two independent normal distribution with population means $\mu_A = (43, 40)$ and $\mu_B = (40, 40)$ and unit variance.

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Korelacija – utjecaj netipičnih točaka (outliers)

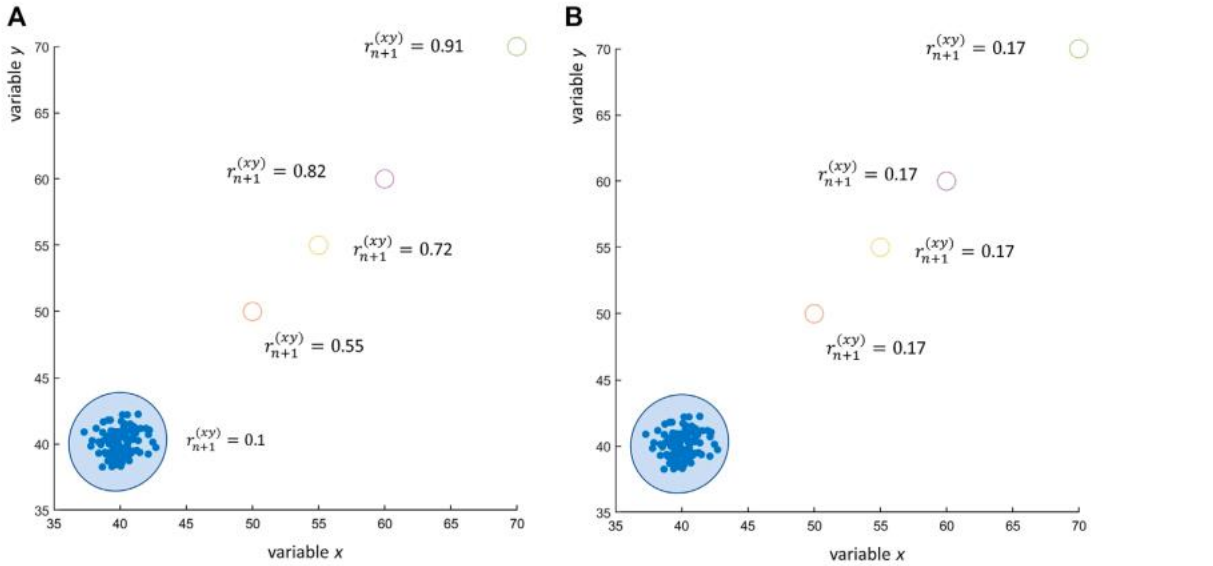

FIGURE 5
Effect of outliers on the correlation coefficient: 100 observations of two uncorrelated variables x and $y \sim N$ (40, 1) (observed Pearson's correlation $r_n^{(xy)} = 0.1$) to which an outlier observation x_{n+1}, y_{n+1}, with increasing distance from the sample mean (open dots ○), is added. **(A)** The Pearson's correlation $r_{n+1}^{(xy)}$ calculated on the $n + 1$ observations (including the outlier x_{n+1}, y_{n+1}) increases with the distance of the outlier x_{n+1}, y_{n+1} from the sample mean, consistently with Eq. 21. **(B)** The Spearman's correlation coefficient is not sensitive to the distance of the outlier from the sample mean. However, this is not true if $m \geq$ 2 outliers are present, as shown in Figure 6.

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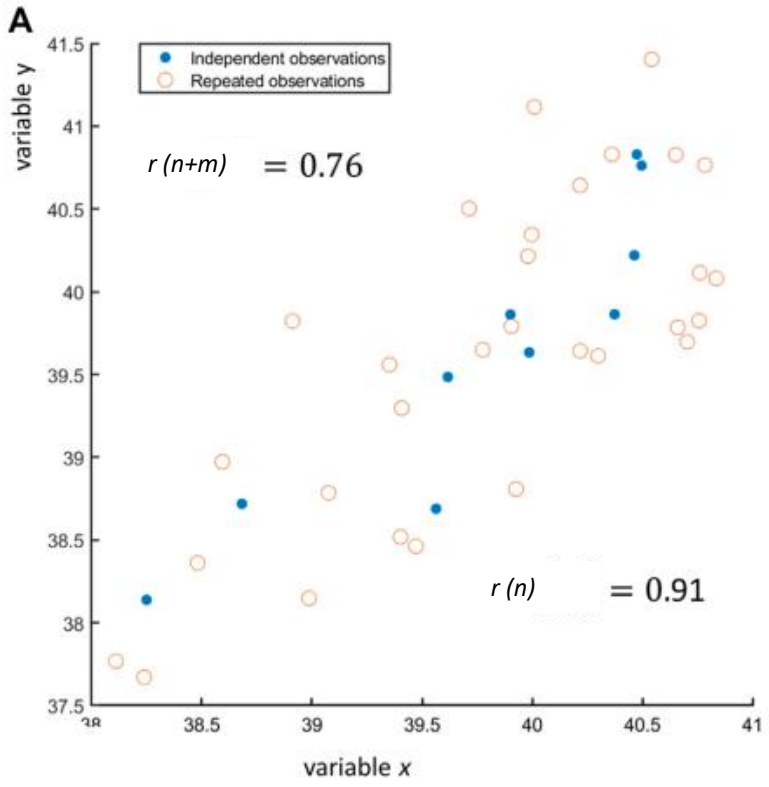
Korelacija – uzorci nisu međusobno neovisni



Tumačenje i značajnost koeficijenta korelacije

Pitanje: U istraživanju povezanosti raspoloženja i količine tekućine unesene pijenjem tijekom dana dobivena je povezanost r = 0,12; P = 0,003. Je li ispravno zaključiti kako postoji značajna povezanost raspoloženja i količine popijene tekućine?

Udovičić, Martina ; Baždarić Ksenija ; Bilić-Zulle Lidija ; Petrovečki Mladen Što treba znati kada izračunavamo koeficijent korelacije? // Biochemia medica, 17 (2007), 1; 10-15

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Odgovor: Nije ispravno.
Tumačenje: Nakon izračuna koeficijenta korelacije važno je znati kako rezultat protumačiti, odnosno objasniti što vrijednosti koeficijenta korelacije zaista znače. U prikazu rezultata korelacija obvezno se navode koeficijent povezanosti (korelacije) "r" i to brojem s dva decimalna mjesta, te značajnost koeficijenta korelacije "P" brojem s tri decimalna mjesta (4). Ukoliko je koeficijent korelacije značajan s obzirom na postavljenu granicu značajnosti (uobičajeno $P < 0,05$), zaključujemo da je koeficijent korelacije značajan i da se smije tumačiti. Ukoliko je vrijednost $P > 0,05$ zaključujemo da koeficijent korelacije nije značajan i tada se bez obzira na njegovu vrijednost ne smije tuma-

Udovičić, Martina ; Baždarić Ksenija ; Bilić-Zulle Lidija ; Petrovečki Mladen Što treba znati kada izračunavamo koeficijent korelacije? // Biochemia medica, 17 (2007), 1; 10-15

Tumačenje i značajnost koeficijenta korelacije

Pitanje: U istraživanju povezanosti koncentracije alkohola u krvi i prometnih nesreća utvrđeni su r = 0,78 i P=0,002. Možemo li zaključiti kako uzimanje alkohola uzrokuje prometne nesreće, tj. promatrane prometne nesreće su posljedica uzimanja alkohola?

Udovičić, Martina ; Baždarić Ksenija ; Bilić-Zulle Lidija ; Petrovečki Mladen Što treba znati kada izračunavamo koeficijent korelacije? // Biochemia medica, 17 (2007), 1; 10-15

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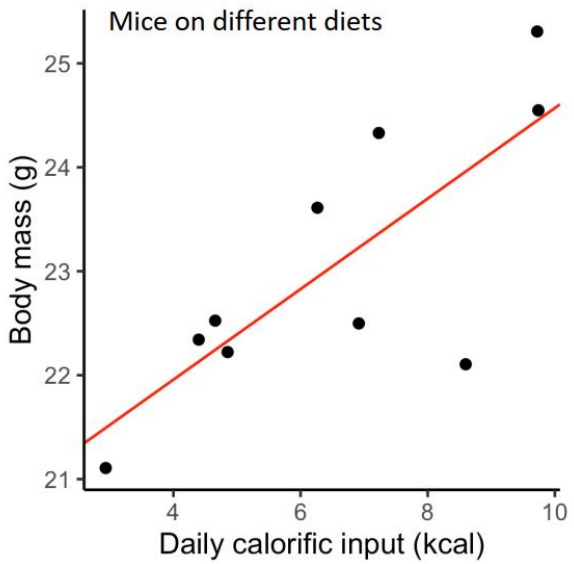
Odgovor: Ne, ne možemo.
Tumačenje: Korelacija govori o povezanosti, a ne o uzročno posljedičnoj vezi među varijablama. Dakle, ukoliko postoji visoka povezanost između uzimanja alkohola i prometnih nesreća ne možemo zaključiti da jedna varijabla utječe na drugu, odnosno da uzimanje alkohola uzrokuje nesreće u prometu. Moguće je da veća količina alkohola uzorkuje više prometnih nesreća, no postoji mogućnost značajnog utjecaja ostalih neispitivanih čimbenika ili rijetkih događaja (7,8). U opisanom primjeru to bi moglo biti stanje na cesti, ispravnost vozila, moguća bolest vozača nevezana za alkohol, djelovanje drugih farmakološki aktivnih tvari i sl.

Udovičić, Martina ; Baždarić Ksenija ; Bilić-Zulle Lidija ; Petrovečki Mladen Što treba znati kada izračunavamo koeficijent korelacije? // Biochemia medica, 17 (2007), 1; 10-15

Linearna regresija

- Jednostavan kvantitativni model

	Daily calorific input (kcal)	Body mass (g)
1	8.6	22.1
2	2.9	21.1
3	4.4	22.3
4	4.9	22.2
5	9.7	25.3
6	9.7	24.5
7	6.3	23.6
8	4.7	22.5
9	7.2	24.3
10	6.9	22.5



Best-fitting linear model

$$y_i = \beta_0 + \beta_1 x_i + \epsilon_i$$

Adapted from https://dag.compbio.dundee.ac.uk/workshops/statistics_lectures/13_Linear_models.pdf

Linearna regresija

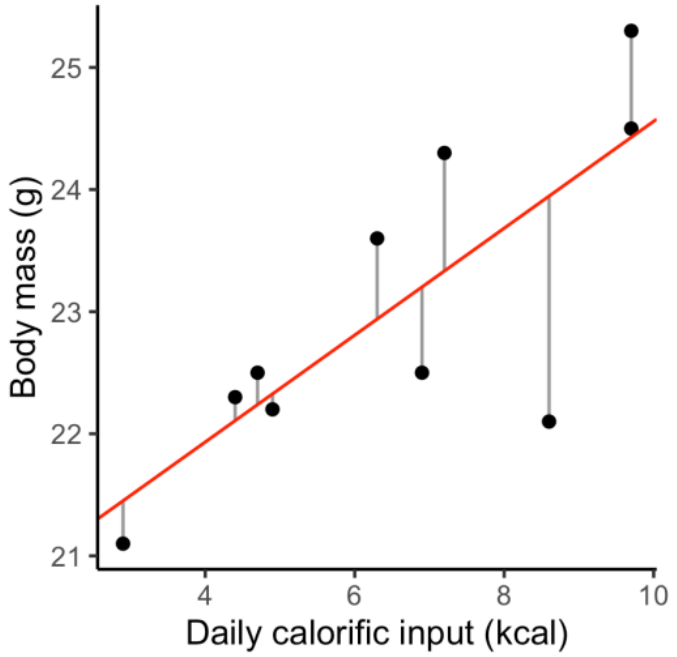
```
> f <- lm(mass ~ kcal, data=ms)
> summary(f)

Call:
lm(formula = mass ~ kcal, data = ms)

Residuals:
    Min      1Q  Median      3Q     Max
-1.8462 -0.2947  0.1323  0.5608  0.9667

Coefficients:
            Estimate Std. Error t value Pr(>|t|)
(Intercept)  20.1813     0.8750  23.065 1.33e-08 ***
kcal          0.4378     0.1269   3.449  0.00871 **
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.8862 on 8 degrees of freedom
Multiple R-squared:  0.5979,         Adjusted R-squared:  0.5476
F-statistic:  11.9 on 1 and 8 DF,  p-value: 0.008709
```



- β_0 odsječak na osi Y (i.e. prosječna vrijednost Y ako su svi X jednaki 0)
- β_j prosječno povećanje Y kad se X_j poveća za 1 jedinicu i svi ostali **X su konstantni**

Adapted from https://dag.compbio.dundee.ac.uk/workshops/statistics_lectures/13_Linear_models.pdf

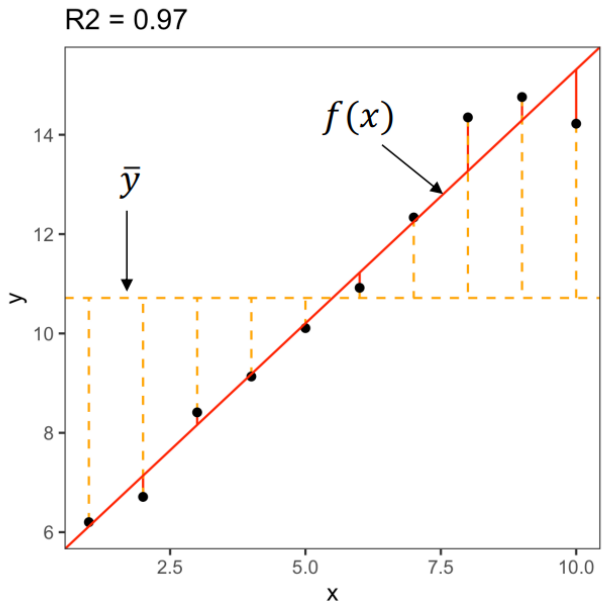
Linearna regresija

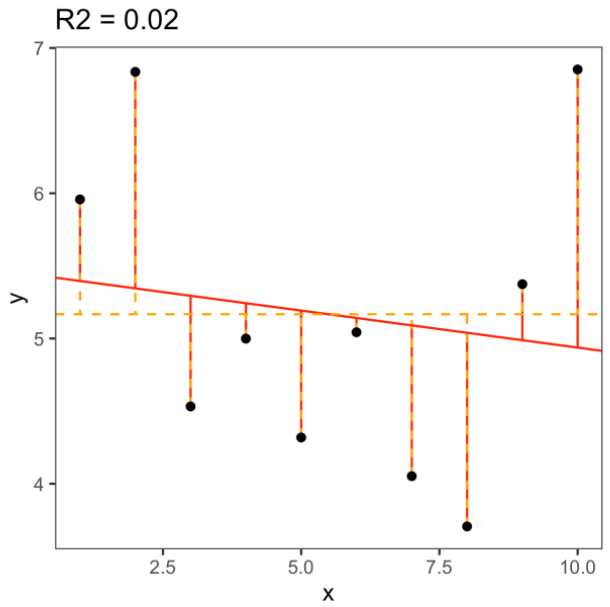
$$R^2 = 1 - \frac{\text{variance explained by the model}}{\text{total variance}}$$

$$R^2 = 1 - \frac{\sum(y_i - f(x_i))^2}{\sum(y_i - \bar{y})^2}$$

It is a measure of fit quality.
The higher R^2, the better fit.

Adjusted R^2 takes into account number of model parameters.

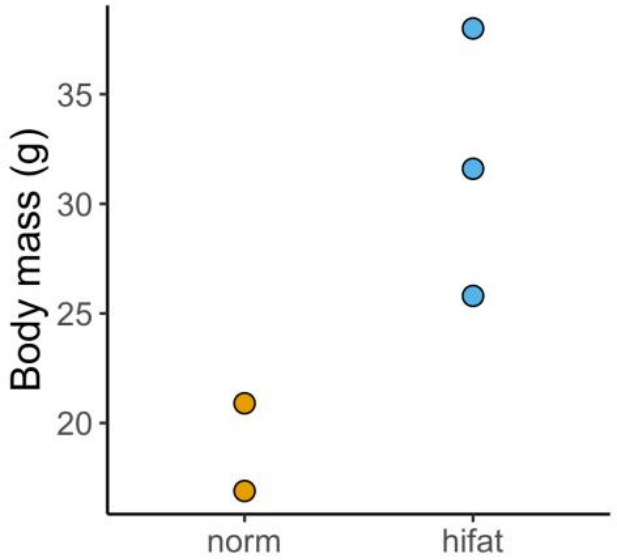




Adapted from https://dag.compbio.dundee.ac.uk/workshops/statistics_lectures/13_Linear_models.pdf

Kategorički prediktori

	Body mass (g)	Diet
1	16.8	norm
2	20.9	norm
3	25.8	hifat
4	38.0	hifat
5	31.6	hifat



$$\begin{pmatrix} y_1 \\ y_2 \\ y_3 \\ y_4 \\ y_5 \end{pmatrix} = \begin{pmatrix} 1 & 0 \\ 1 & 0 \\ 0 & 1 \\ 0 & 1 \\ 0 & 1 \end{pmatrix} \begin{pmatrix} \mu_1 \\ \mu_2 \end{pmatrix} + \begin{pmatrix} \epsilon_1 \\ \epsilon_2 \\ \epsilon_3 \\ \epsilon_4 \\ \epsilon_5 \end{pmatrix}$$

```
> f1 <- lm(mass ~ diet, data = mdat)
> summary(f1)
             Estimate Std. Error t value Pr(>|t|)
(Intercept)    18.900      3.708   5.098   0.0146 *
diethifat      12.900      4.787   2.695   0.0741 .
```

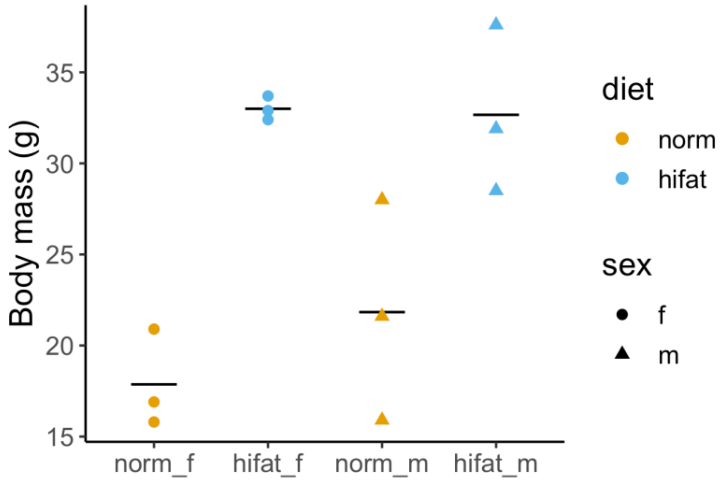
H_0: effect size is equal zero

dietfat = difference between normal and high-fat diet

Adapted from https://dag.compbio.dundee.ac.uk/workshops/statistics_lectures/13_Linear_models.pdf

Više od jednog prediktora

	Body mass (g)	Diet	Sex
1	16.9	norm	f
2	20.9	norm	f
3	15.8	norm	f
4	28.0	norm	m
5	21.6	norm	m
6	15.9	norm	m
7	32.4	hifat	f
8	33.7	hifat	f
9	32.9	hifat	f
10	28.5	hifat	m
11	37.6	hifat	m
12	31.9	hifat	m



Adapted from https://dag.compbio.dundee.ac.uk/workshops/statistics_lectures/13_Linear_models.pdf

Linearna regresija s interakcijama

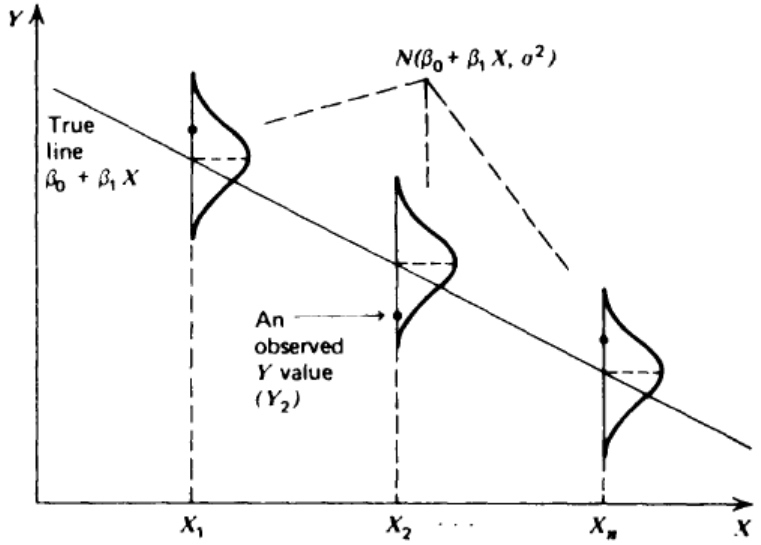
```
> f <- lm(mass ~ diet + sex + diet:sex, data = mds)
> summary(f)$coefficients
               Estimate Std. Error t value Pr(>|t|)
(Intercept)      17.867      2.335   7.652 6.01e-05 ***
diethifat        15.133      3.302   4.583  0.00179 **
sexm              3.967      3.302   1.201  0.26400
diethifat:sexm   -4.300      4.670  -0.921  0.38407
                    .
```

Interaction not significant, we are overfitting

Adapted from https://dag.compbio.dundee.ac.uk/workshops/statistics_lectures/13_Linear_models.pdf

Pregled reziduala

- Reziduali sadrže informacije o tome zašto model možda ne odgovara podacima
- Reziduali – uočene pogreške ako je model točan
- Pretpostavke o pogreškama:
  - Pogreške su neovisne
  - Slijede normalnu distribuciju sa srednjom vrijednošću 0 i konstantnom varijancom σ2
  - Nakon ispitivanja reziduala možemo zaključiti vrijede li ove pretpostavke ili ne



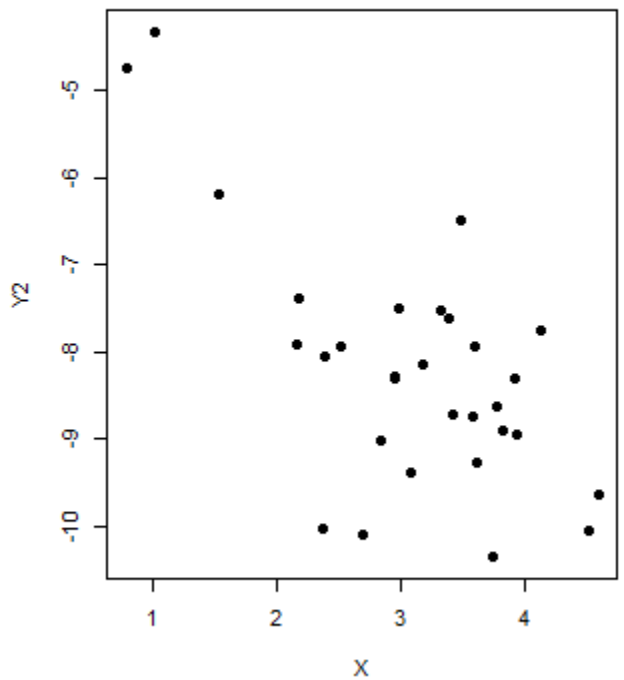
$$\varepsilon_i \sim N(0, \sigma^2)$$

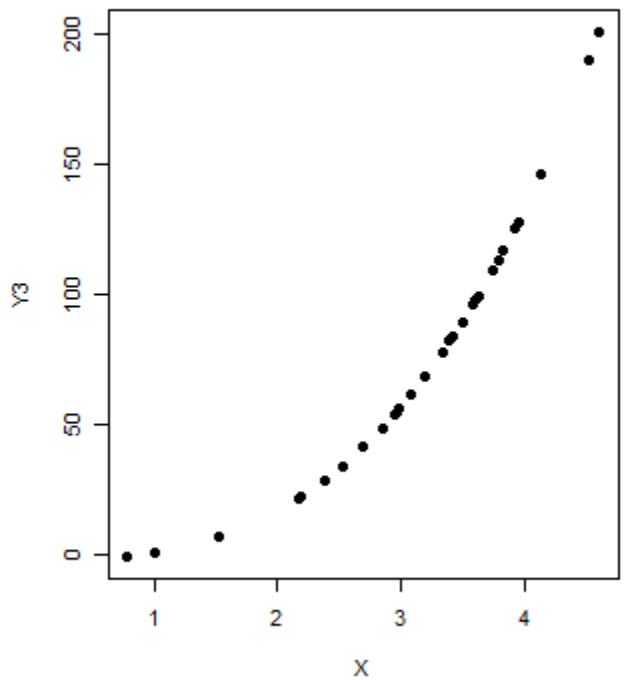
Testiranje normalnosti reziduala

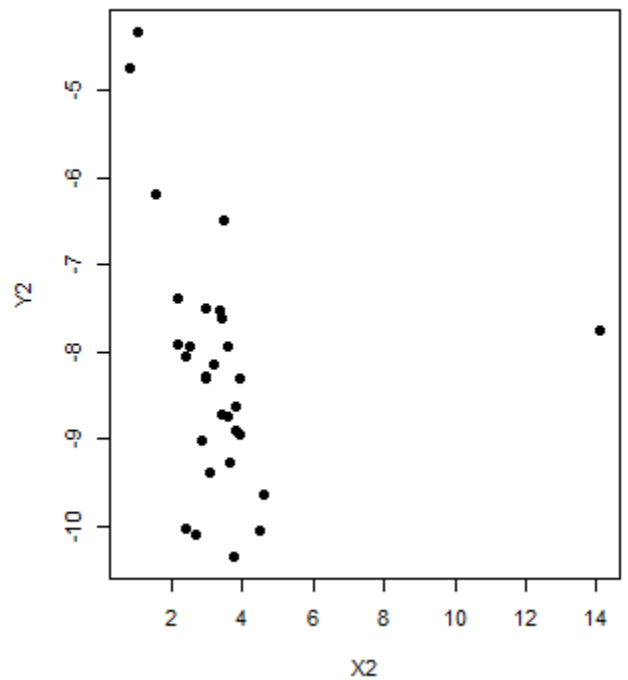
- Grafičke metode:
  - Histogram
  - Kvantil-kvantil prikaz s obzirom na standardnu normalnu distribuciju

- Testovi za normalnost
  - Anderson-Darling test, Shapiro-Wilk test, Lilliefors test (adaptacija Kolmogorov-Smirnoff testa), D'Agostino-Pearson test…

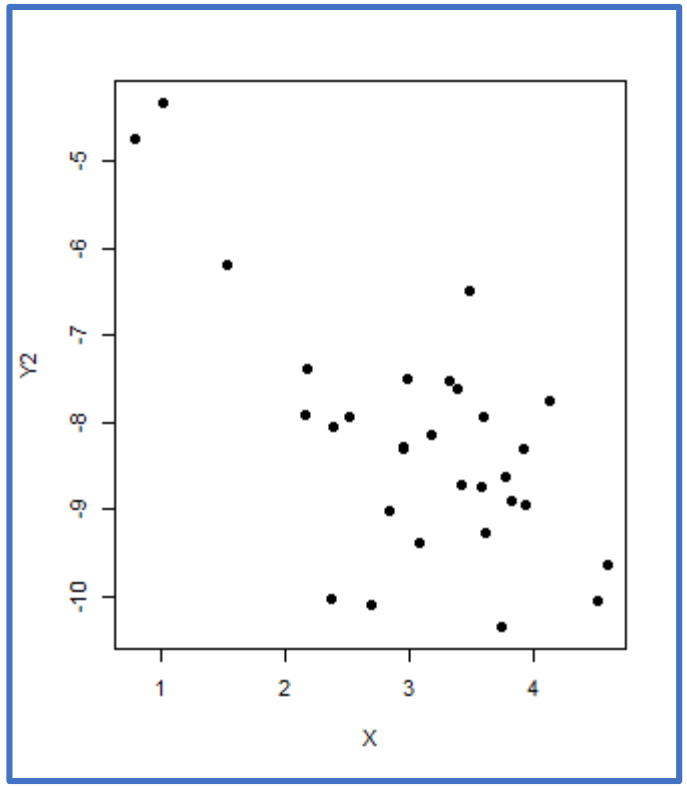
Je li linearan model prikladan za sljedeće analize?

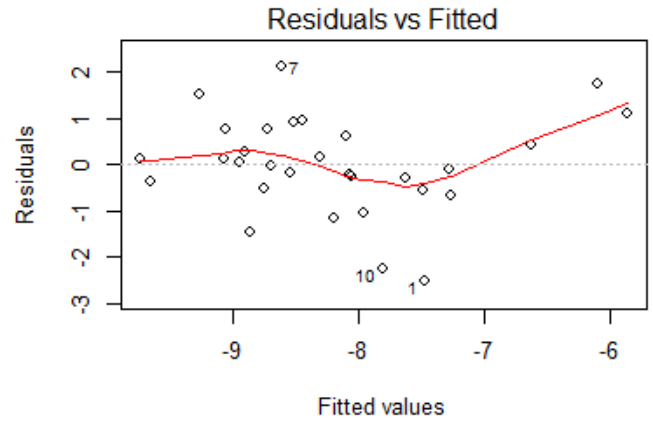


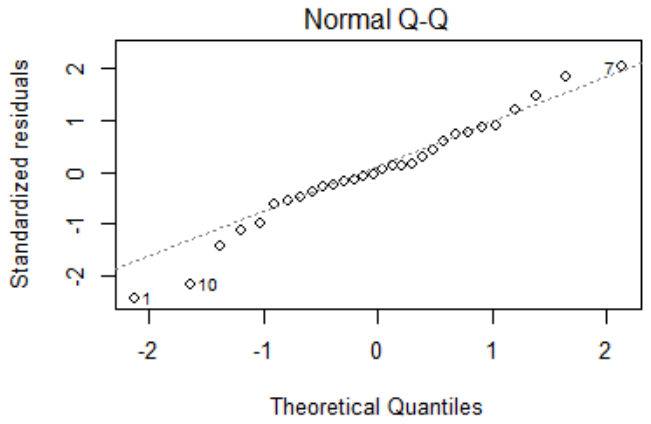


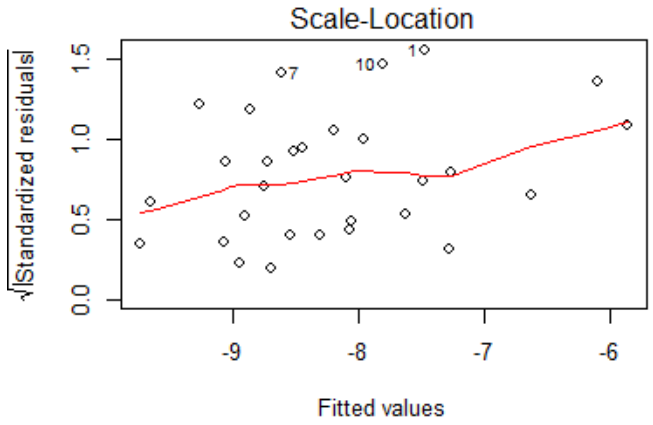


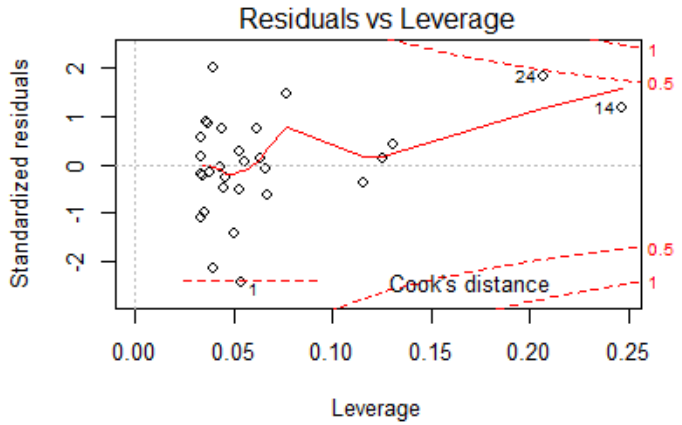
Grafički prikaz reziduala



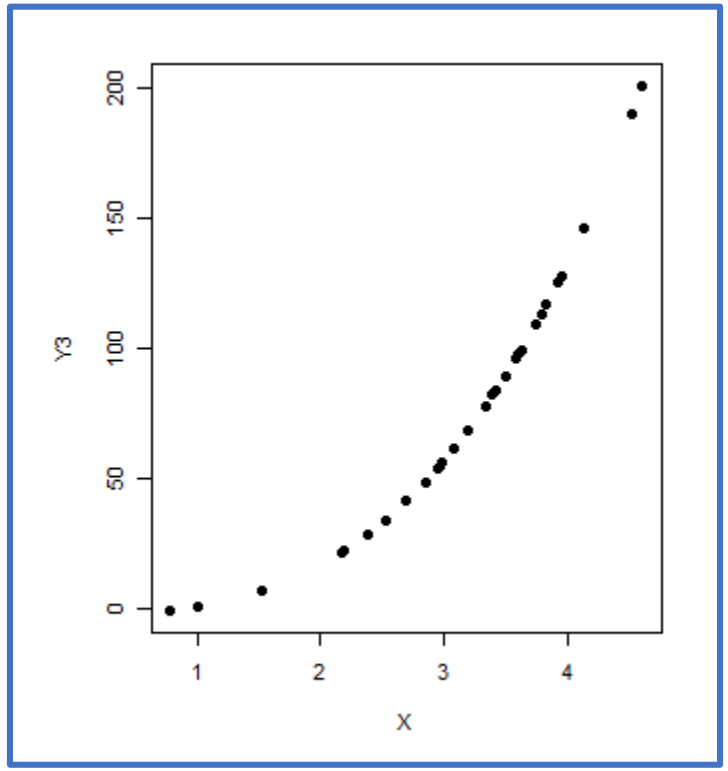


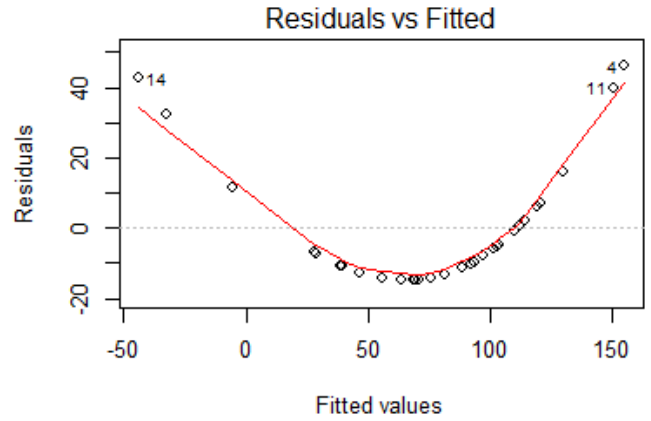


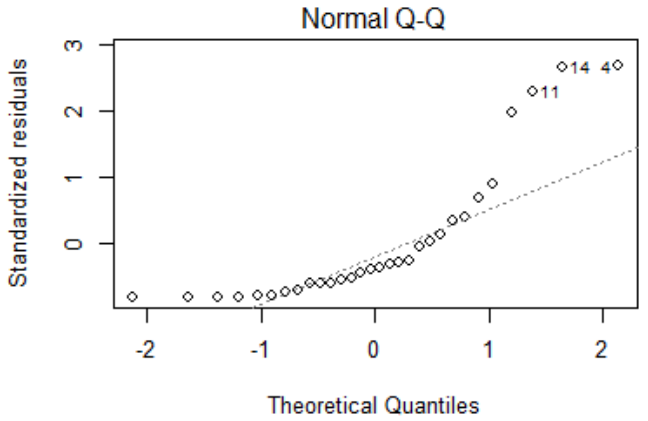


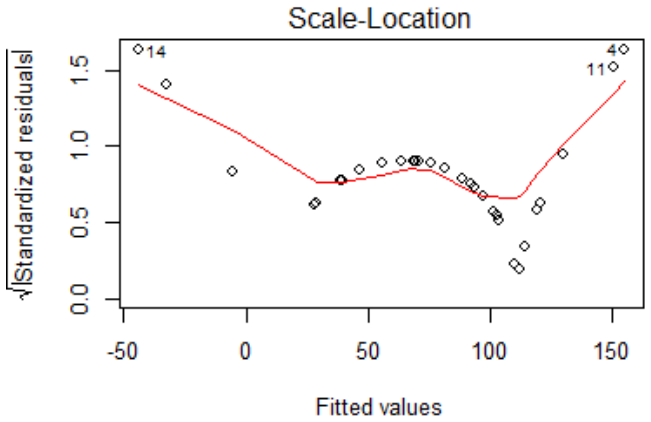


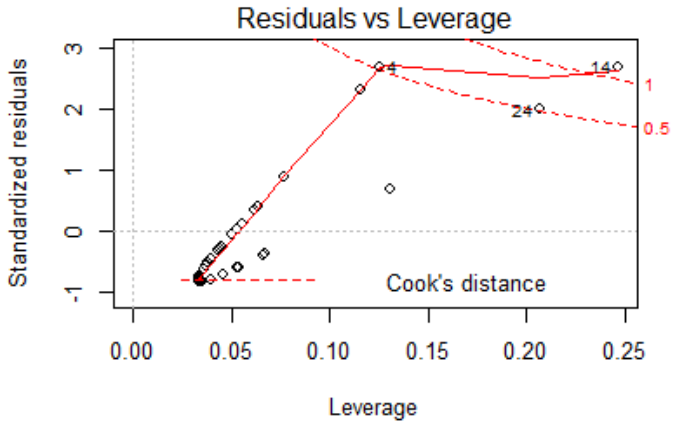
Ne-linearni odnosi



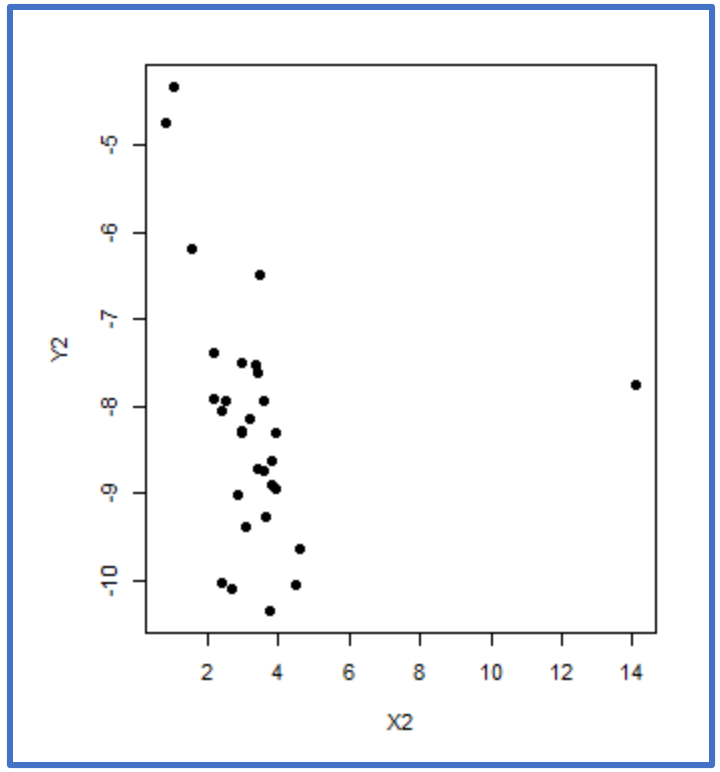


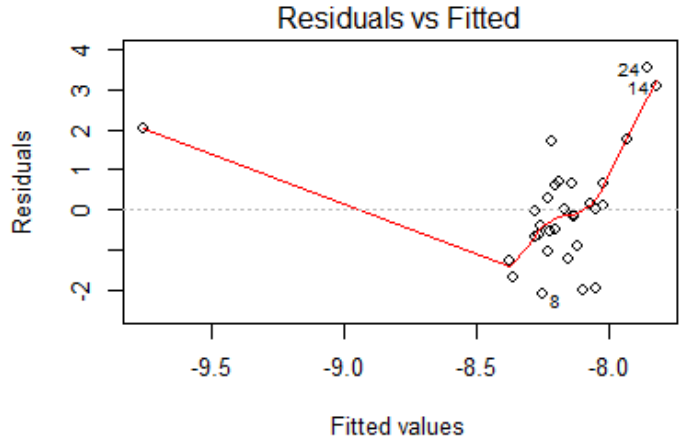


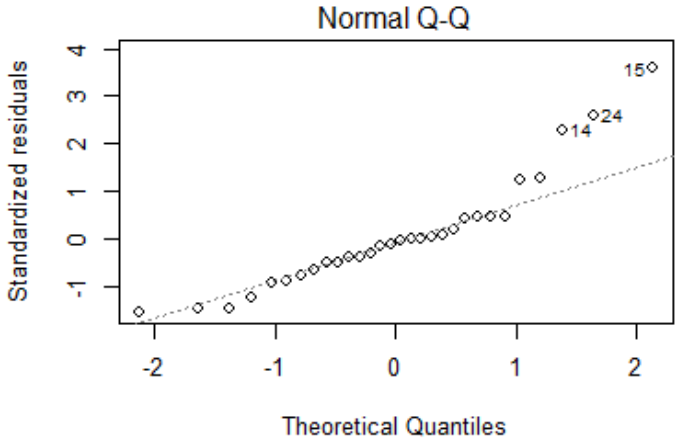


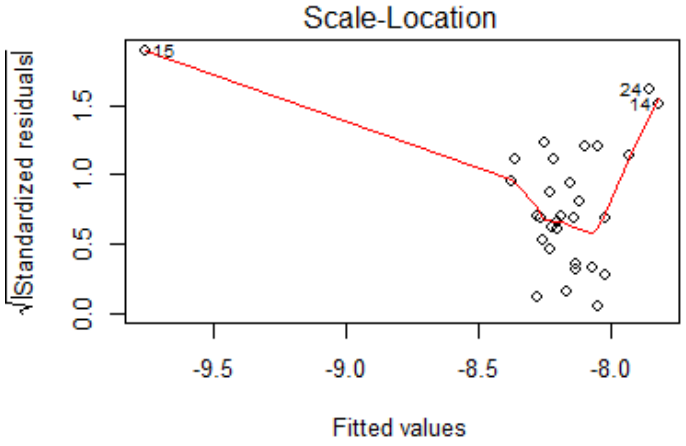


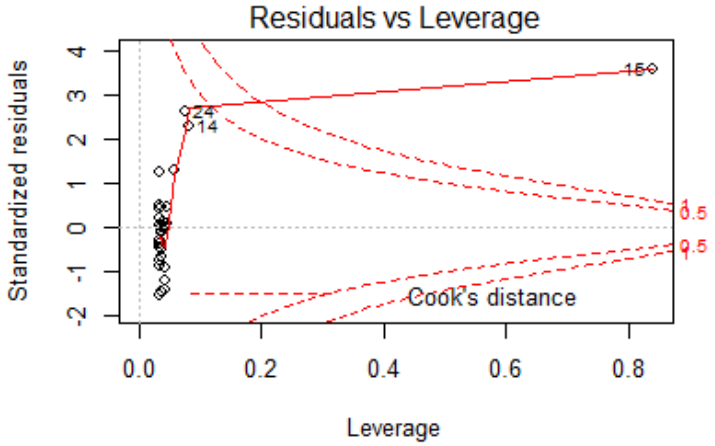
High leverage points (točke visokog utjecaja)





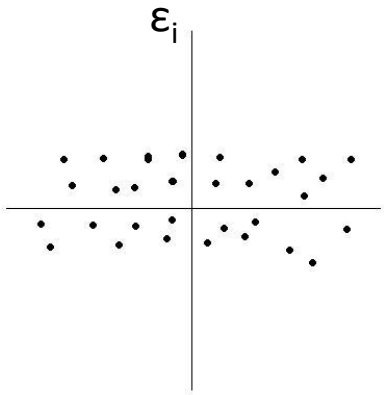




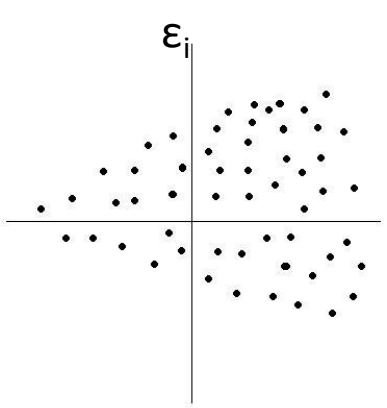


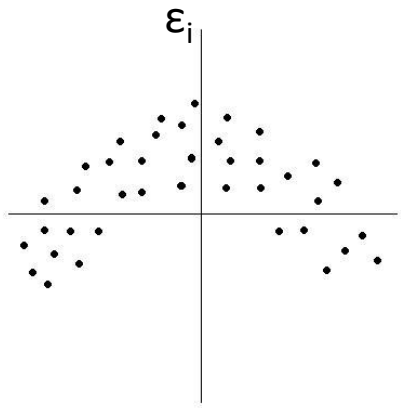
Provjeriti vremenske učinke, nestalnu varijancu, potrebu za transformacijom i zakrivljenost

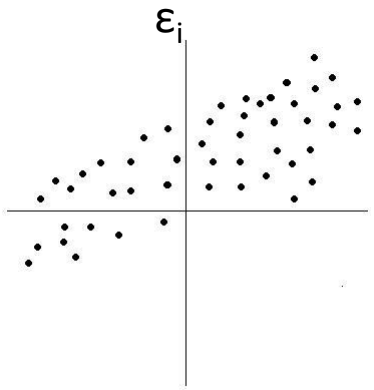
- Zadovoljavajući dijagram reziduala trebao bi pokazivati slučajni uzorak



- Nezadovoljavajući prikazi reziduala:



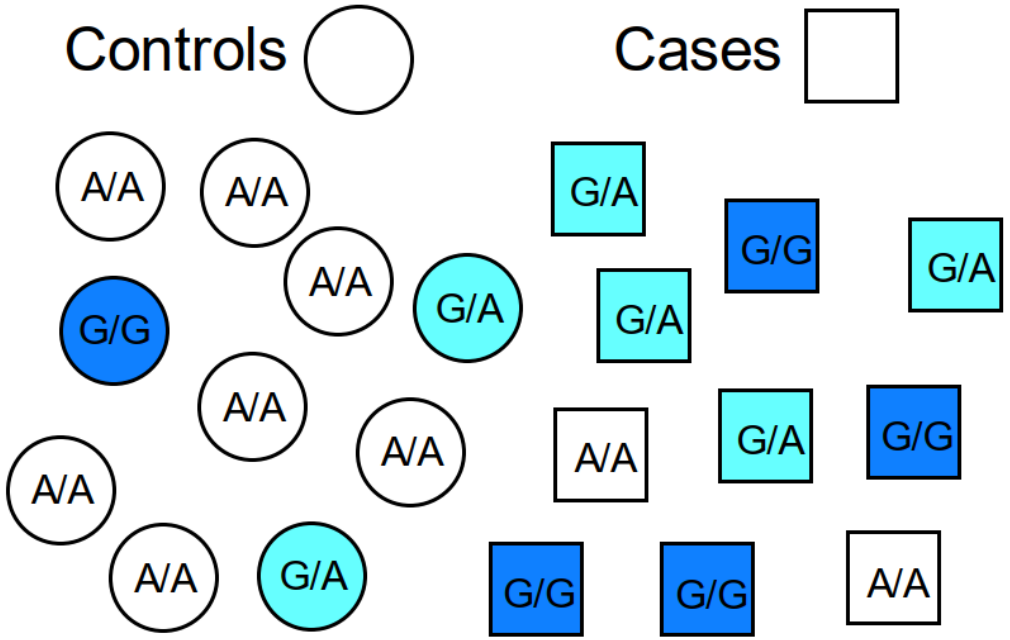




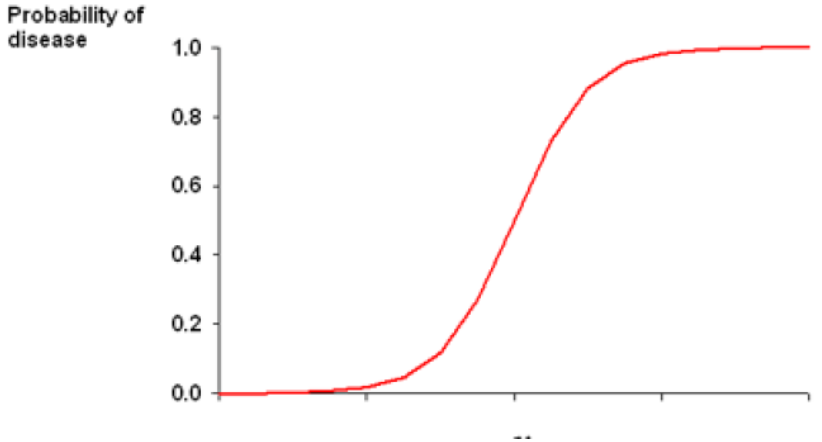
Logistička regresija

$$p(X) = \frac{e^{\beta_o + \beta_1 X}}{1 + e^{\beta_o + \beta_1 X}}$$

Case/control



The G allele is associated with disease



$\ln(P(X)/1-P(X)) = \alpha + \beta X + \varepsilon$

β = difference in log odds for cases vs. controls

$e^{(\beta)}$ = difference in odds

= Odd Ratio (OR)

Allelic effect is an OR:

OR > 1 increased risk

OR < 1 decreased risk

Adapted from https://www.colorado.edu/ibg/isg-workshop/isgw-online-resources/b1-introduction-gwas-part-1