

"99 percent of all statistics only tell 49 percent of the story" Ron DeLegge II (Economist)

## Statistics in its best revealing unexpected effects

# THE WOW Factor!

#### **General linear models (not Generalized linear model)**

- Traditionally, authors tend to separate t-tests, ANOVA, Regression and Analysis of Covariance (ANCOVAs). However, because they share the same calculations (and theories and assumptions), we often classify these methods under the category of *General Linear Models*.

- *General Linear Models* (unlike Generalized linear models) assume that response variables are normally distributed.

#### **General linear models**

	Linear Model	Common name
	$Y = \mu + X$	Simple linear regression
<ul> <li>Image: A start of the start of</li></ul>	$Y = \mu + A_1$	One-factorial (one-way) ANOVA
	$Y = \mu + A_1 + A_2 + A_1 \times A_2$	Two-factorial (two-way) ANOVA
	$Y = \mu + A_1 + X + (A_1 \times X)$	Analysis of Covariance (ANCOVA)
	$Y = \mu + X_1 + X_2 + X_3$	Multiple regression
	$Y = \mu + A_1 + g + A_1 \times g$	Mixed model ANOVA
	$Y_1 + Y_2$ = $\mu + A_1 + A_2 + A_1 \times A_2$	Multivariate ANOVA (MANOVA)

Y (response) is a continuous variableX (predictor) is a continuous variableA represents categorical predictors (factors)g represents groups of data (more on this later)

#### $Y = \mu + A_1 + X_1 + (A_1 \times X_1)$ Analysis of Covariance (ANCOVA)

- Test for differences in slopes among groups (treatments).

- Adjust for the effects of a covariate  $X_1$  (continuous) in an ANOVA design (response variable Y and a categorical variable  $A_1$ ).

-  $\mu$  the grand mean (i.e., mean of the response across all observations independent of their groups).



<u>Oecologia</u>

June 1992, Volume 90, <u>Issue 3</u>, pp 435–444 | <u>Cite as</u>

# The effects of grazers on the performance of individuals and populations of scarlet gilia, *Ipomopsis aggregata*

Authors

Authors and affiliations

Joy Bergelson, Michael J. Crawley

*I. aggregata* exhibits considerable powers of regrowth following removal of its primary shoot by herbivores, but we found no evidence of overcompensation (i.e. of significantly higher plant performance where plants were exposed to ungulate herbivory) in a comparison between individuals on grazed and ungrazed sides of exclosure fences





<u>Oecologia</u>

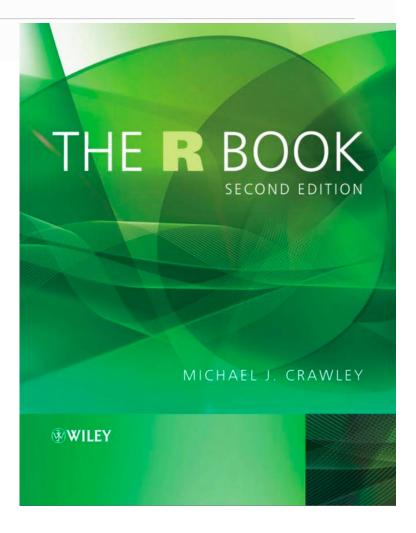
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# The effects of grazers on the performance of individuals and populations of scarlet gilia, *Ipomopsis aggregata*

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#### Study design and data

- 40 plants (*Ipomopsis*) were allocated to a grazing factor (two levels: grazed or ungrazed):
- 1) Grazed plants were exposed to grazers for the first two weeks of stem elongation (initial plant size measured as diameter of the rootstock top).
- 2) After two weeks, fence was built to prevent grazing.
- 3) At the end of the growing season, fruit production (dry weight in *mg* was recorded for each plant.
- 4) Initial plant size (diameter of the *rootstock* top, i.e., root size) was thought to influence fruit production and it was also measured.



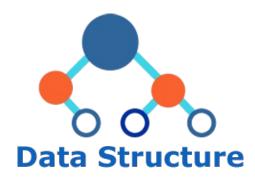
#### **Data structure**

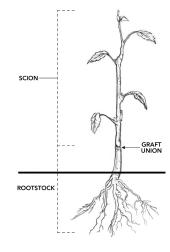


Fruit production (dry weight in mg).

Grazing (Yes / No).

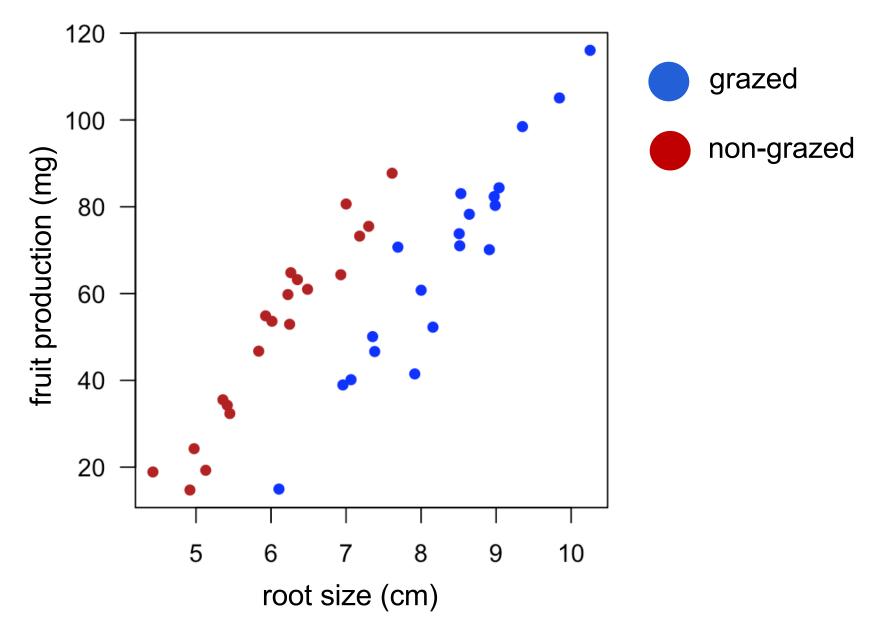
Initial plant size (diameter of the rootstock top).



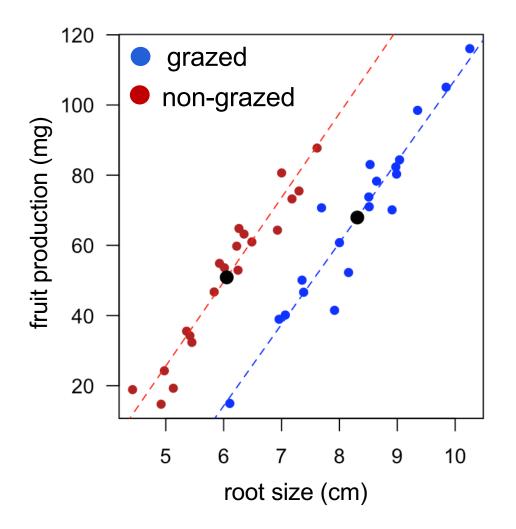


В	С	D	
Fruit	Grazing	Root	
59.77	Ungrazed	6.225	
60.98	Ungrazed	6.487	
14.73	Ungrazed	4.919	
19.28	Ungrazed	5.13	
34.25	Ungrazed	5.417	
35.53	Ungrazed	5.359	
87.73	Ungrazed	7.614	
63.21	Ungrazed	6.352	
24.25	Ungrazed	4.975	
64.34	Ungrazed	6.93	
52.92	Ungrazed	6.248	
32.35	Ungrazed	5.451	
53.61	Ungrazed	6.013	
54.86	Ungrazed	5.928	
64.81	Ungrazed	6.264	
73.24	Ungrazed	7.181	
80.64	Ungrazed	7.001	
18.89	Ungrazed	4.426	
75.49	Ungrazed	7.302	
46.73	Ungrazed	5.836	
116.05	Grazed	10.253	
38.94	Grazed	6.958	
60.77	Grazed	8.001	
84.37	Grazed	9.039	
70.11	Grazed	8.91	
14.95	Grazed	6.106	
70.7	Grazed	7.691	
80.31	Grazed	8.988	
82.35	Grazed	8.975	
105.07	Grazed	9.844	
73.79	Grazed	8.508	
50.08	Grazed	7.354	
78.28	Grazed	8.643	
41.48	Grazed	7.916	
98.47	Grazed	9.351	
40.15	Grazed	7.066	
52.26	Grazed	8.158	
46.64	Grazed	7.382	
71.01	Grazed	8.515	
83.03	Grazed	8.53	

The data: initial thoughts?



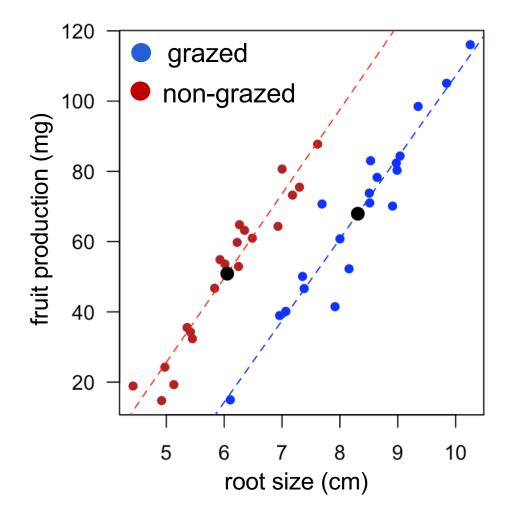
#### Key initial observations about the grazing experiment data



- 1) Plants with different initial sizes were allocated to the two treatments, i.e., The regression line for the non-grazed plants is above the line for the grazed plants.
- 2) Grazed plants produced more fruits.
- 3) The two regression lines are almost parallel.

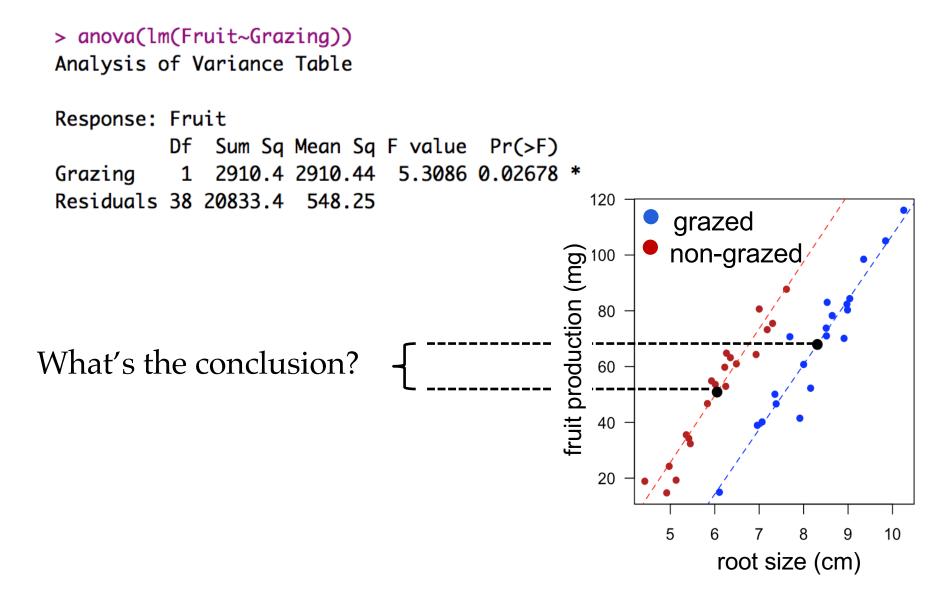
#### Two major statistical potential biases tackled by ANCOVA:

Were plants assigned to treatments randomly according to initial size?
 How could the assignment influence the interpretation of results?

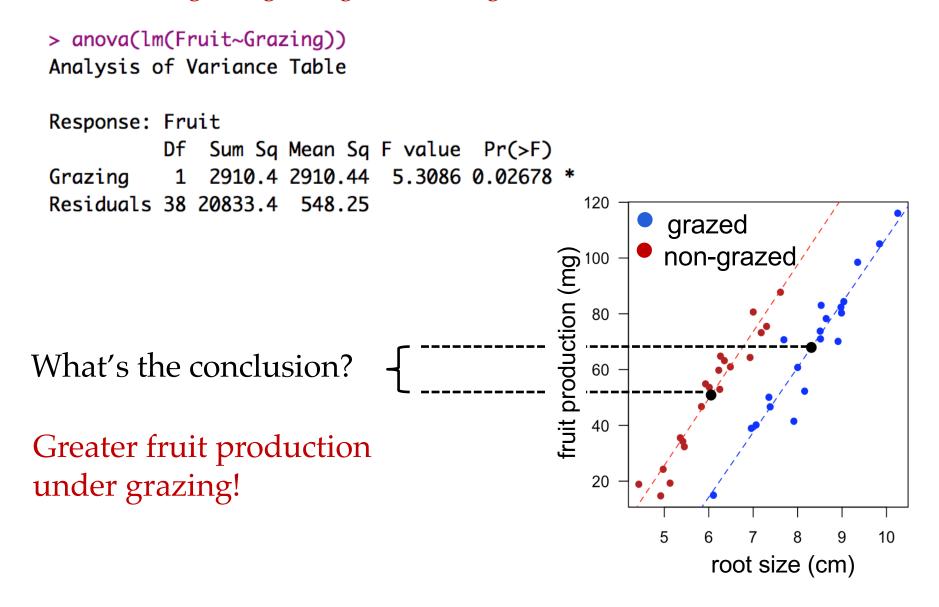


- 1) Plants with different initial sizes were allocated to the two treatments, i.e., The regression line for the non-grazed plants is above the line for the grazed plants.
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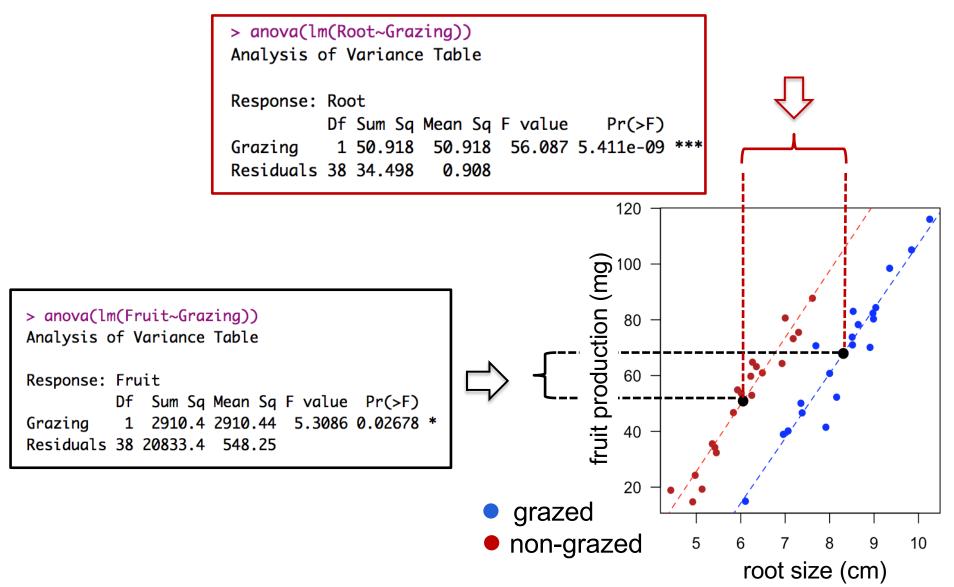
Let's start with a simple ANOVA (a two-sample *t* test could had been used as well;  $t^2 = F$ ) comparing the fruit production as a function of grazing (i.e., grazed, non-grazed).



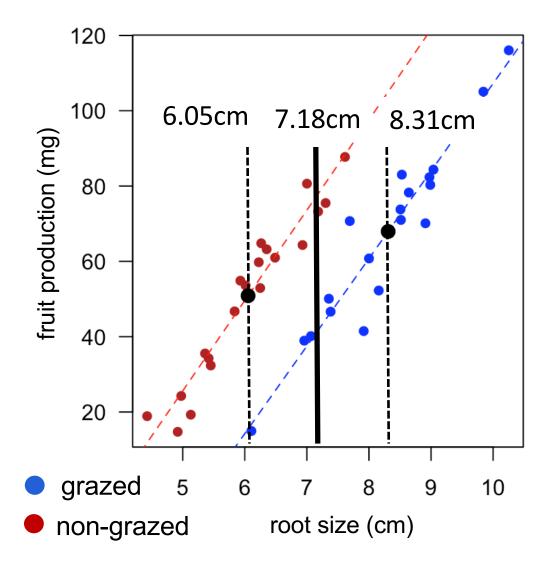
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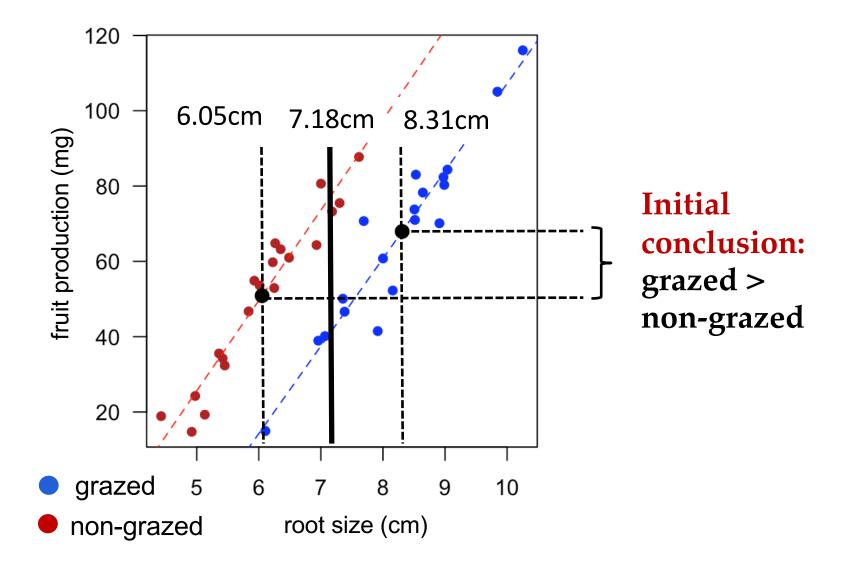
Greater fruit production under grazing! **Can this conclusion be** justified given that the initial root sizes in grazed plants are larger than non-grazed plants?



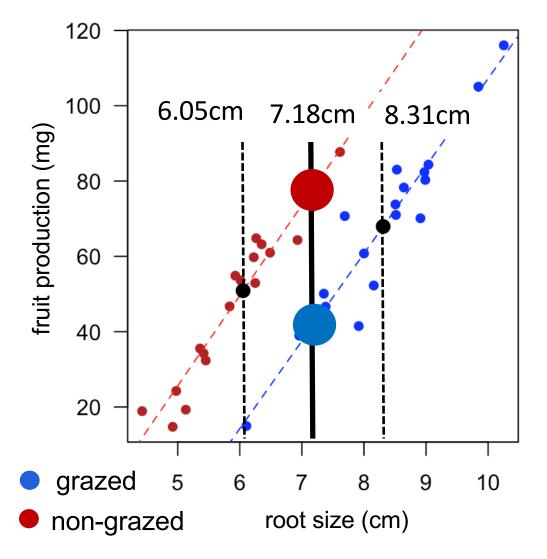
Greater fruit production under grazing! **Can this conclusion be** justified given that the initial root sizes in grazed plants are larger than non-grazed plants?



What would the effect of grazing be if all plants had started with the same initial (root) size? Say the average value for the two treatments = (6.05 + 8.31)/2 = 7.18cm Assuming a common initial mean size of root size (7.18cm), we can adjust (predict) the fruit production for each treatment (grazed / non-grazed), i.e., as if root size would had been the same in the beginning of the experiment.

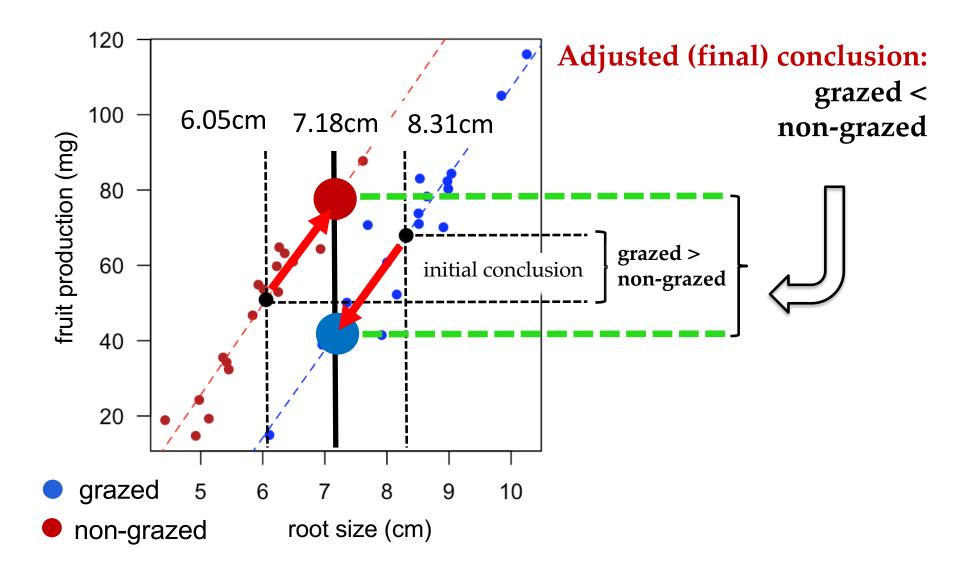


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What would the effect of grazing be if all plants had started with the same initial (root) size? Say the average value for the two treatments = (6.05 + 8.31)/2 = 7.18cm

Let's adjust each group's average by predicting their expected values. Assuming a common initial mean size of root size (7.18cm), we can adjust (predict) the fruit production for each treatment (grazed / non-grazed), i.e., as if root size would had been the same in the beginning of the experiment.



Statistics in its best revealing "unexpected" effects Initial conclusion: grazed > nongrazed.

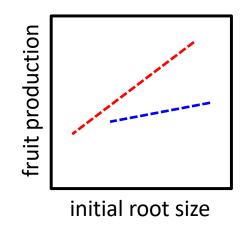
Adjusted (final) conclusion: grazed < non-grazed.

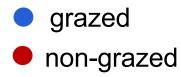
WOW Factor!

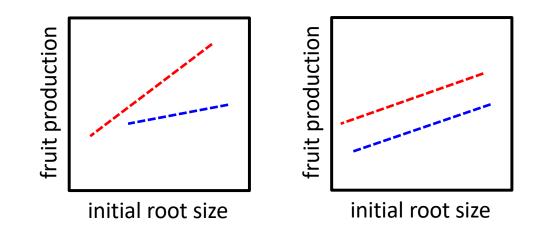
### When can we use ANCOVA to adjust for a continuous predictor (here initial plant size)?

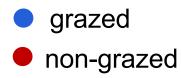


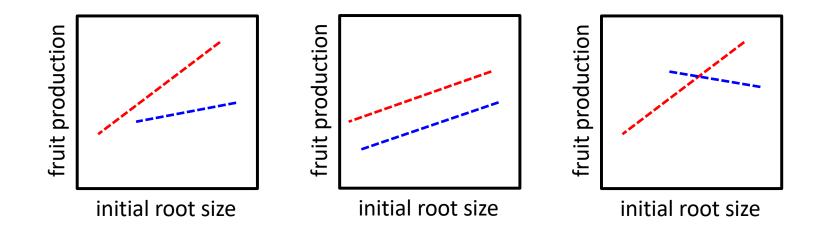
- It "combines" ANOVA and regression into one analysis (we know they are the same models).
- As such, it includes at least one categorical predictor (factor, e.g., Grazing) and one continuous predictor (e.g., initial root size).
- The goal of an ANCOVA (in general) is to test for the effect of a categorical predictor while adjusting (controlling) for the effect of a continuous predictor.
- The continuous predictor is called covariate.

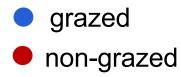


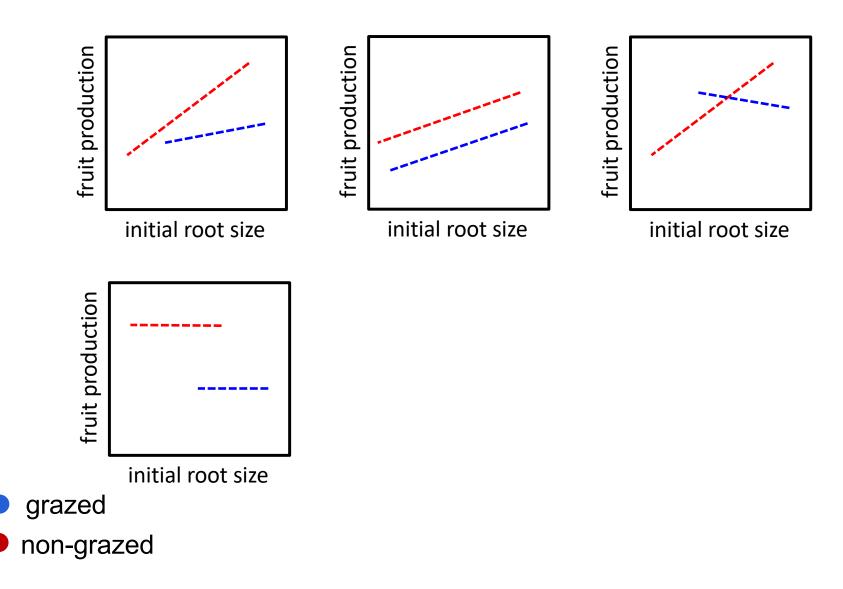


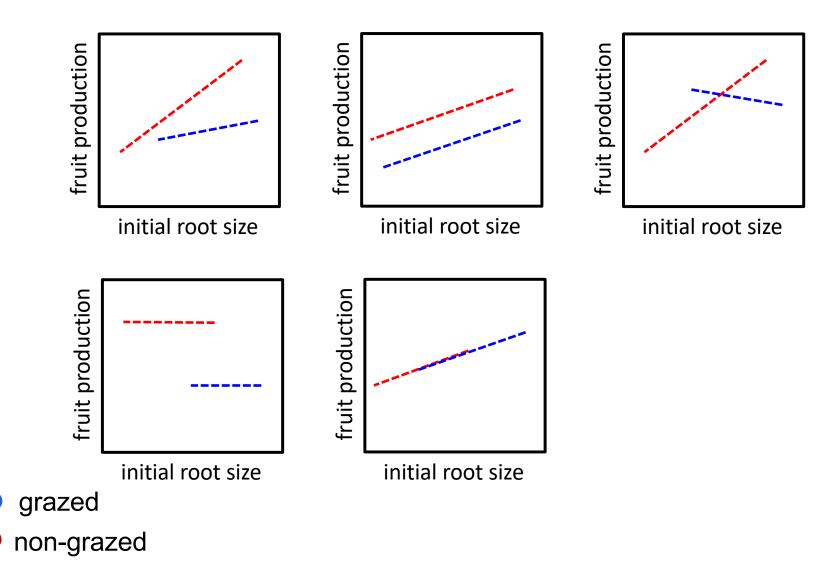


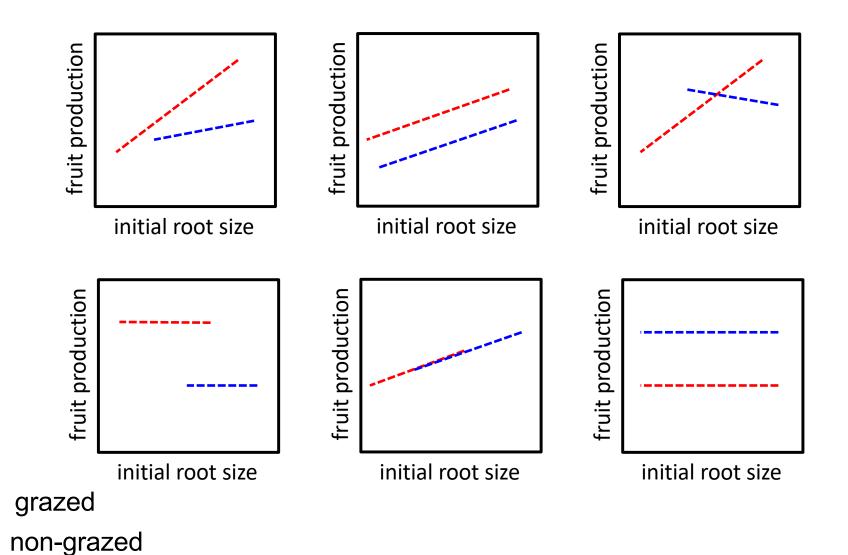


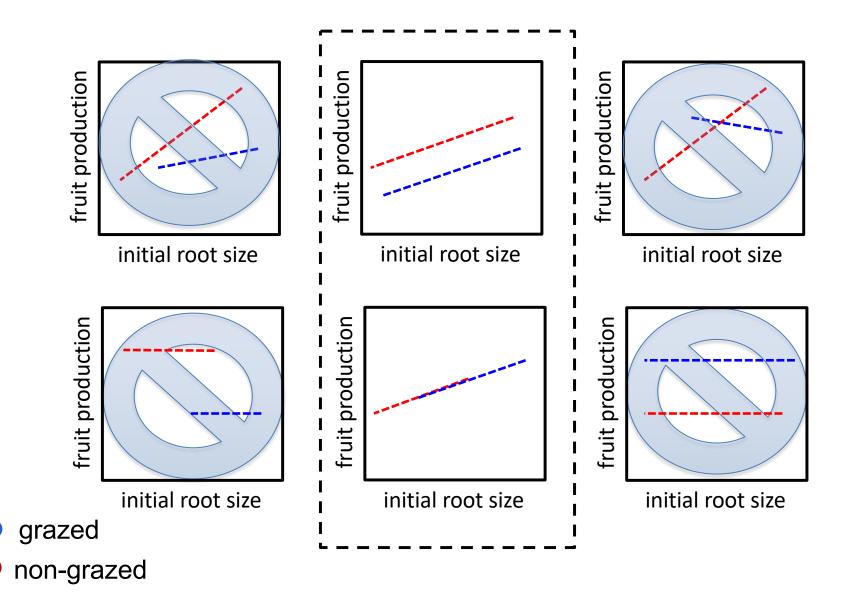




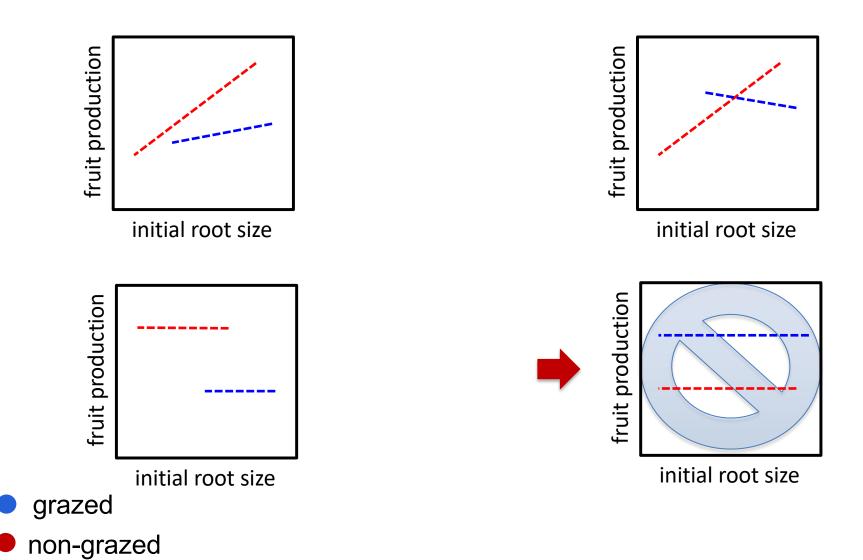




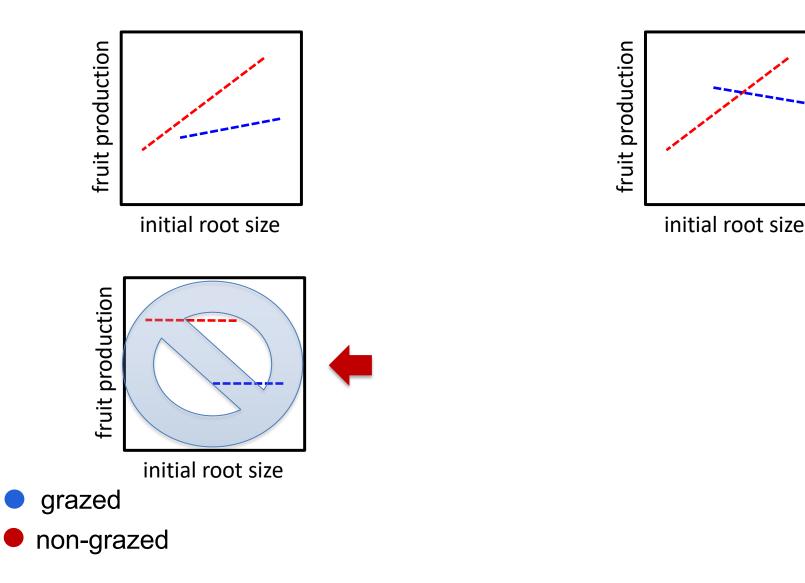




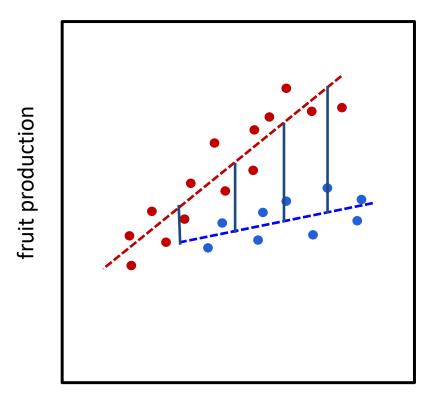
No need for adjustments as initial root size do not differ between grazed and non-grazed treatments



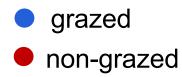
Can't be adjusted because the fruit production does not change as a function of initial root size, so root size can't be used to predict fruit production on the basis of a common mean



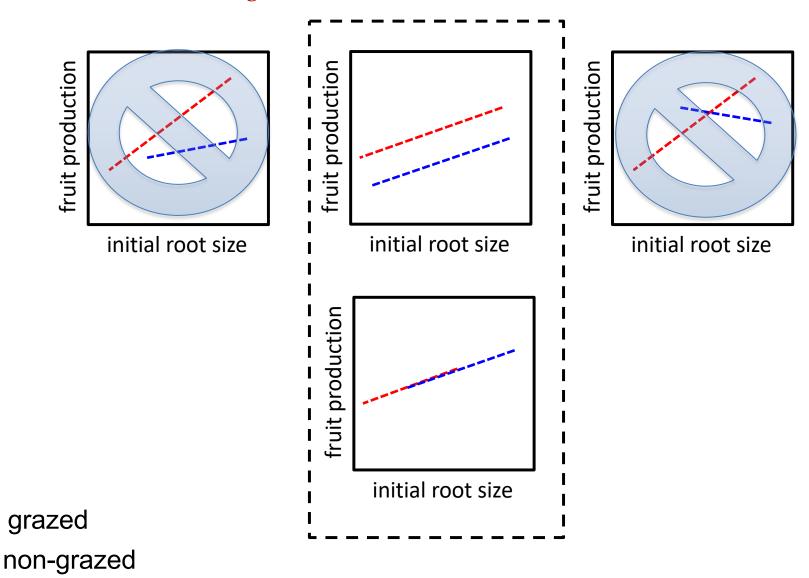
Can't be adjusted because an interaction between initial root size and grazing treatment. When there is an interaction, the differences in mean values between treatments vary as a function of the covariate, so we can't generalize to all initial root sizes.



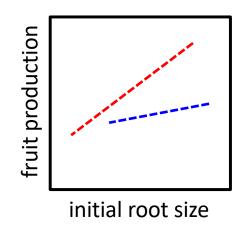


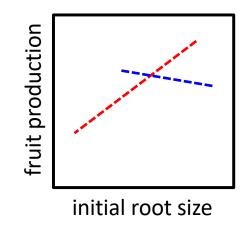


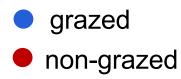
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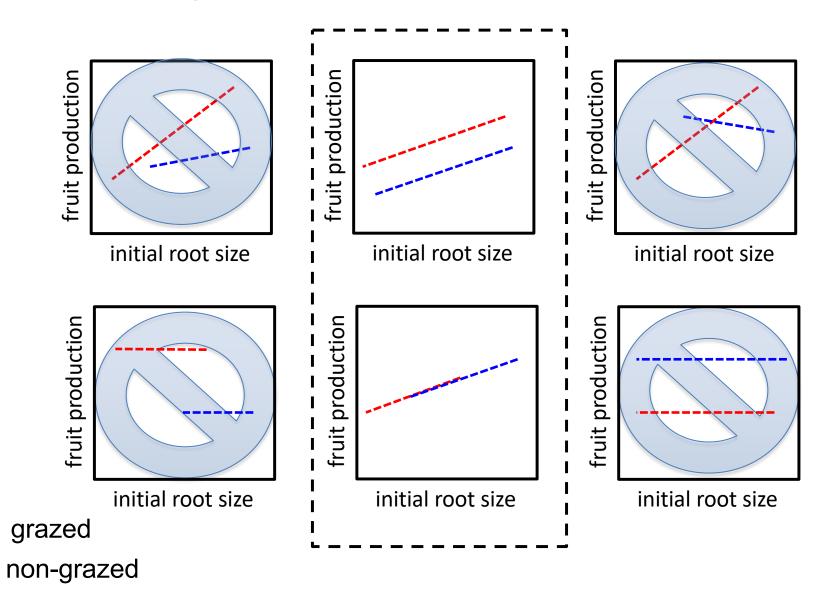
There are approaches for the more complex cases when slopes differ between groups or when the response (Y) does not depend on the covariate (initial root size) – next lecture.







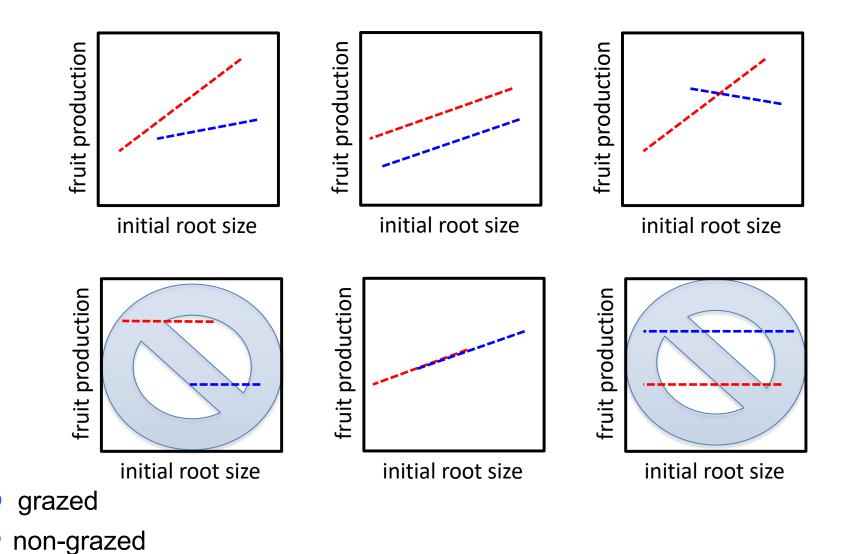
A common (equal) slope (parallel curves) between groups implies that mean differences between groups in their response (fruit production) are the same regardless of the value of the covariate (initial root size).



# When can ANCOVA adjustments be used? Statistical assessments



#### First assessment – Can the covariate predict the response?



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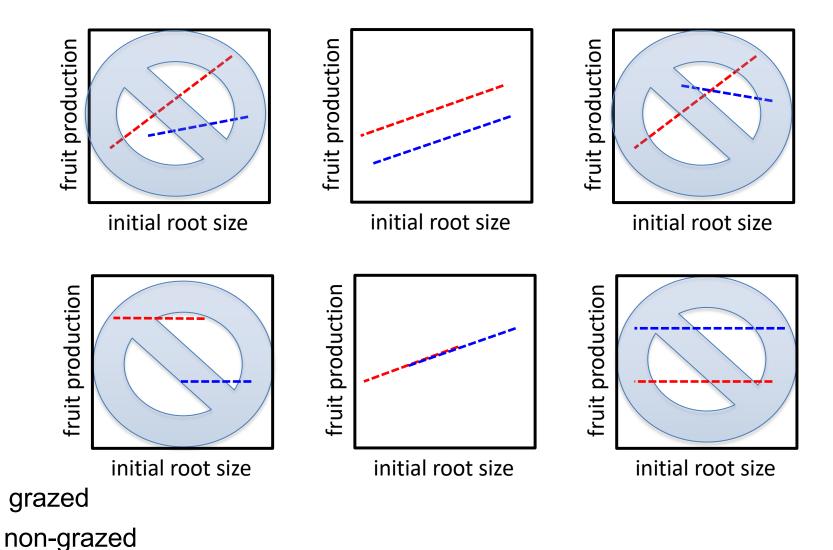
**H**<sub>0</sub>: The slope of the regression of fruit production on initial root size is zero ( $\beta = 0$ ).

**H**<sub>A</sub>: The slope of the regression of fruit production on initial root size is not zero ( $\beta \neq 0$ ).

> anova(lm(Fruit ~ Root))
Analysis of Variance Table

Response:	Fru	Jit					
_	Df	Sum Sq	Mean Sq	F value	Pr(>F)		_
Root	1	16795.0	16795.0	91.844	1.099e-11	***	]
Residuals	38	6948.8	182.9				

Second assessment – Do groups share a common slope? Test whether the interaction between covariate (Initial root size) and factor (grazing) is significant; if significant, they don't share a common slope and initial root size can't be used for adjusting fruit production.



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**H**<sub>0</sub>**:** There is no interaction between grazing treatment and initial root size (i.e., grazing/no-grazing (groups) do not differ in their slopes).

**H**<sub>A</sub>**:** There is an interaction between grazing treatment and initial root size (i.e., grazing/no-grazing differ in their slopes).

```
> anova(lm(Fruit ~ Root*Grazing))
Analysis of Variance Table
```

Response:	Fruit						
	Df	Sum Sq	Mean Sq	F value	Pr(>F)		
Root	1	16795.0	16795.0	359.9681	< 2.2e-16	***	
Grazing	1	5264.4	5264.4	112.8316	1.209e-12		
Root:Grazi	ng 1	4.8	4.8	0.1031	0.75		
Residuals	36	1679.6	46.7				

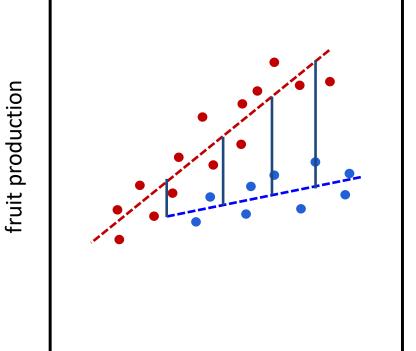
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Note that testing for differences in slopes (Y on X) between groups (e.g., grazed versus non-grazed), i.e., testing the interaction between the categorical (groups) and X, is interesting in itself.

In the problem analysed here we don't want to have them different but in other cases we may (e.g., allometric differences). Second assessment – Do groups share a common slope? Test whether the interaction between covariate (Initial root size) and factor (grazing) is significant.



covariate, so we can't generalize to all initial root sizes (more on this later).

Remember: when

there is an interaction,

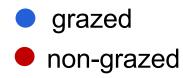
then the differences in

mean values between

treatments vary as a

function of the

initial root size



#### So far, we have:

Covariate can predict the response

```
> anova(lm(Fruit ~ Root))
Analysis of Variance Table

Response: Fruit
    Df Sum Sq Mean Sq F value Pr(>F)
Root 1 16795.0 16795.0 91.844 1.099e-11 ***
Residuals 38 6948.8 182.9
```

Groups share a common slope

```
> anova(lm(Fruit ~ Root*Grazing))
Analysis of Variance Table
```

Response: Fruit								
	Df	Sum Sq	Mean Sq	F value	Pr(>F)			
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Residuals	36	1679.6	46.7					

Now we can test for differences in adjusted means; but before that:

Critical statistical issues underlying General Linear Models (including ANCOVAs)

Lecture 10 a pedagogical guide (Type I and III sum-of-square)