



INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Tento projekt je spolufinancován Evropským sociálním fondem a Státním rozpočtem ČR  
InoBio – CZ.1.07/2.2.00/28.0018

Statistical Analysis in Ecology using R

# ANOVA, Linear Regression, ANCOVA

Juliette Chamagne  
Institute of Evolutionary Biology and  
Environmental Sciences  
University of Zurich

November 12<sup>th</sup>, 2013



european  
social fund in the  
czech republic



EUROPEAN UNION



MINISTRY OF EDUCATION,  
YOUTH AND SPORTS



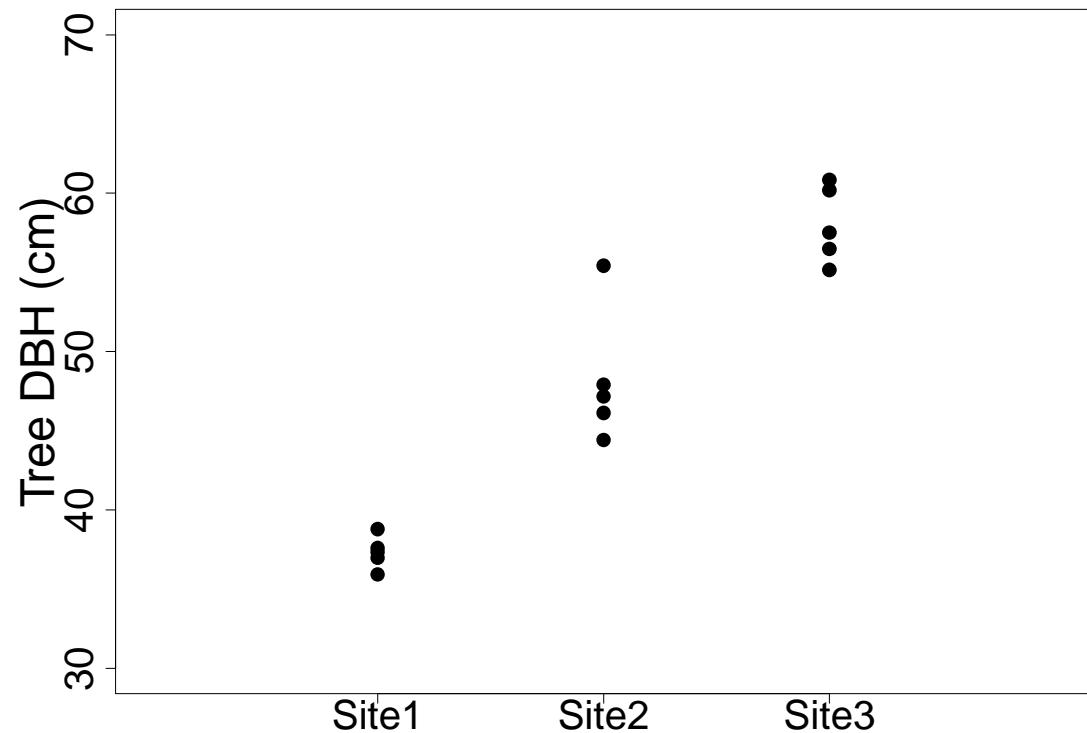
INVESTMENTS IN EDUCATION DEVELOPMENT

# Recap

- Type of data
- We want to **explain Y**
  - Central tendency
  - Dispersion around the central tendency
- Correlations
- Visuals (graphs)

# ANalysis Of VAriance (ANOVA)

- Used to analyze how **mean values** of continuous variable (Y) **differ among groups** of elements.
- Y (size)
- Groups (sites)
- Elements (trees)
- $k = 3$  sites
- $n = 15$  trees



# ANOVA: Assumptions

- **Independence:** The elements have been sampled randomly and independently from each group.
- **Normality:** For each group the values of Y follow a normal distribution.
- **Homoscedasticity:** the variance should be the same for each group.

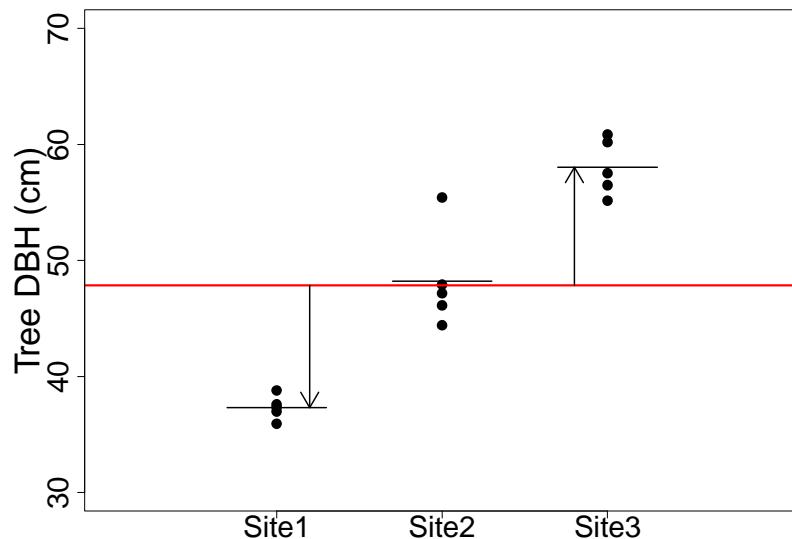
# ANOVA: Hypotheses

- $H_0$ : The **null hypothesis**. The mean value of Y is the same for all groups. The observed variability is only within groups and is due to random sampling. **Nothing interesting is happening.**
- $H_A$ : The **alternative hypothesis**. The mean value of Y differs among populations. There is variability within groups AND among groups. **Something might be of interest.**
- **We test  $H_0$**

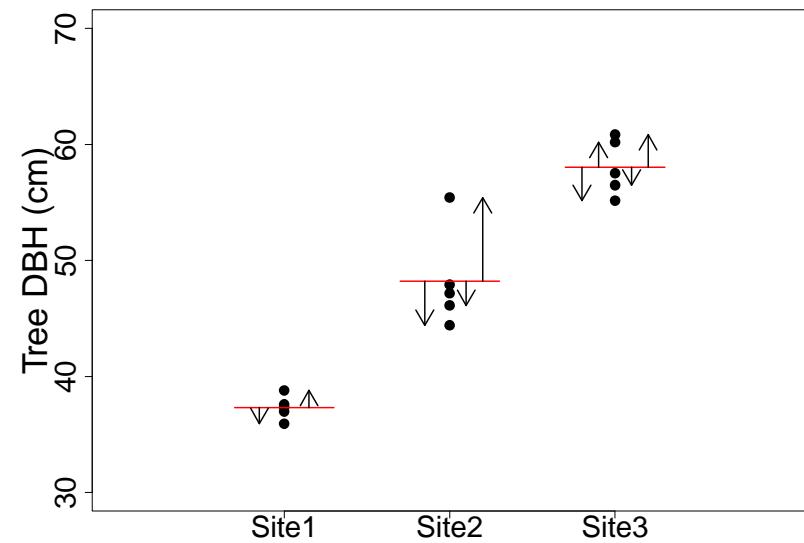
# ANOVA: F ratio: Variation among groups/Variation within groups

$$SS_{among} = \sum_{ij} (\bar{y}_j - \bar{y})^2 = \sum_j n_j (\bar{y}_j - \bar{y})^2 \quad SS_{within} = \sum_{ij} (y_{ij} - \bar{y}_j)^2$$

- “**Group effects**”



- “**Residuals**”



SS = Sum of Squares

$n_j$  = number of elements in group j

$y_{ij}$  = value of element i in group j

$\bar{y}_j$  = mean of  $y_{ij}$  for group j

$\bar{y}$  = overall mean

# ANOVA: F ratio:

## Variation among groups/Variation within groups

- **F-ratio = MS among groups/MS within groups**
  - $MS = SS/df$
  - $MS = \text{Mean Square}$
  - $df = \text{degree of freedom}$
  - $df_{\text{total}} = n-1$
  - $df_{\text{among}} = k-1$
  - $df_{\text{within}} = n-k$
  - **Total = residual + group**
- $k = \text{number of groups } (j = 1 \text{ to } k)$   
 $n = \text{total sample size } (\text{sum of } n_j)$
- $$MS_{\text{total}} = MS_{\text{residual}} + MS_{\text{group}}$$
- $$SS_{\text{total}} = SS_{\text{residual}} + SS_{\text{group}}$$
- $$df_{\text{total}} = df_{\text{residual}} + df_{\text{group}}$$

# ANOVA: F ratio:

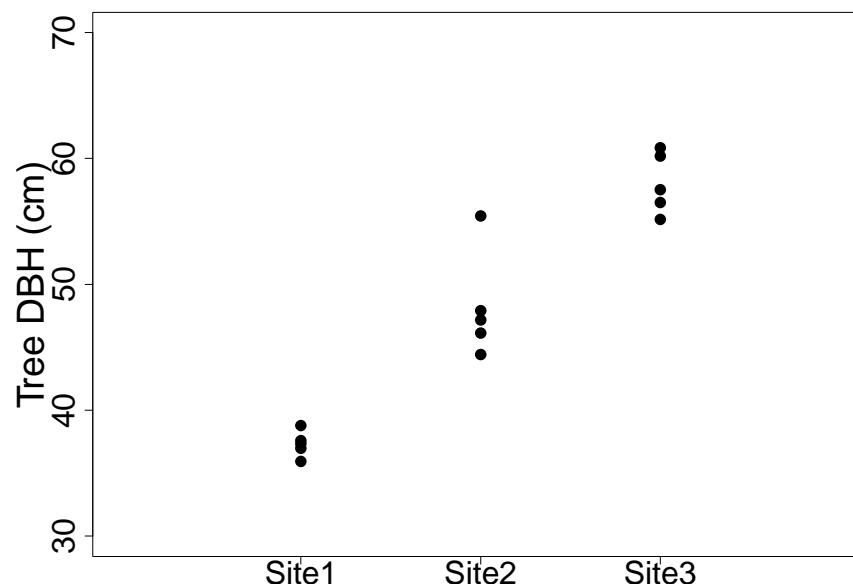
## Variation among groups/Variation within groups

- If F-ratio = 1:  $MS_{\text{residual}} = MS_{\text{group}}$
  - $H_0$  is true: no difference between groups
  - The bigger the F-ratio is, the more likely  $H_0$  to be rejected
- 
- R squared indicates the fraction of the total variation that is due to differences among groups.
  - **R squared = SS of group effects / SS total**
  - $0 < R \text{ squared} < 1$

# ANOVA in R: aov()

```
> model.aov <- aov(size ~ site, data = data)  
> summary(model.aov)
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
site	2	1073.4	536.7	64.54	3.79e-07 ***
Residuals	12	99.8	8.3		



# ANOVA in R: aov()

```
> model.aov <- aov(size ~ site, data = data)
```

```
> summary(model.aov)
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
site	2	1073.4	536.7	64.54	3.79e-07 ***
Residuals	12	99.8	8.3		

$$n = 15 \text{ trees} \rightarrow df_{\text{total}} = n-1 = 14$$

$$k = 3 \text{ sites} \rightarrow df_{\text{groups}} = k-1 = 2$$

$$\rightarrow df_{\text{residuals}} = 14 - 2 = 12$$

# ANOVA in R: aov()

```
> model.aov <- aov(size ~ site, data = data)
```

```
> summary(model.aov)
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
site	2	1073.4	536.7	64.54	3.79e-07 ***
Residuals	12	99.8	8.3		

$$MS_{\text{among}} = MS_{\text{groups}} = SS_{\text{groups}} / df_{\text{groups}} = 1073.4 / 2$$

$$MS_{\text{within}} = MS_{\text{residuals}} = SS_{\text{residuals}} / df_{\text{residuals}} = 99.8 / 12$$

# ANOVA in R: aov()

```
> model.aov <- aov(size ~ site, data = data)
```

```
> summary(model.aov)
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
site	2	1073.4	536.7	64.54	3.79e-07 ***
Residuals	12	99.8	8.3		

$$F \text{ value} = MS_{\text{groups}} / MS_{\text{residuals}} = 536.7 / 8.3$$

Much bigger than 1

# ANOVA in R: aov()

```
> model.aov <- aov(size ~ site, data = data)  
> summary(model.aov)
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
site	2	1073.4	536.7	64.54	3.79e-07 ***
Residuals	12	99.8	8.3		

**P value = Probability of observing data as extreme as this if the null hypothesis were true.**

Here, the P value is almost 0, so it is very unlikely that  $H_0$  is true.

Therefore, we accept  $H_A$  : "There is a difference among groups".

# ANOVA

$H_A$ :

Tree size = Mean size + site effect + effects of age, genes, species...

$Y$  = Fitted value + residual (error)

$H_0$ :

Tree size = Mean size + effects of age, genes, species...

Data = Fitted value + residual (error)

**SS site effect** = SS residual (model without site effect)

- SS residual (model with site effect)

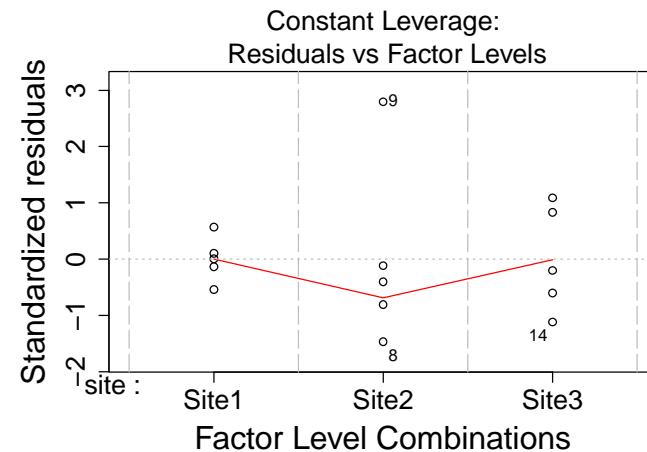
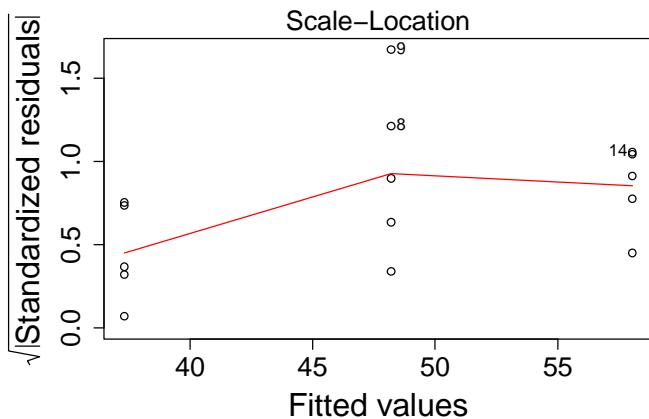
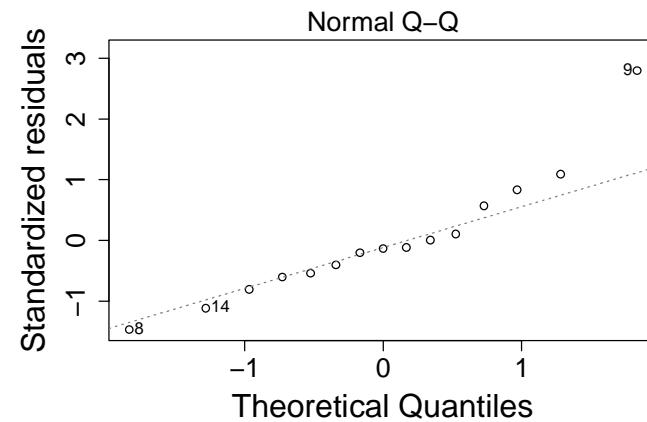
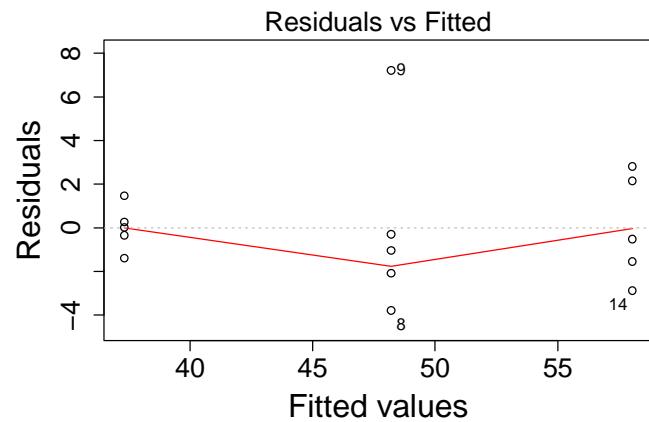
# ANOVA

- Answers 3 questions:
  - Do mean values of Y differ among the sites from which samples were taken?
  - Is the variability of group means larger than expected from the variability within groups?
  - Does a model with site effects describe the data better than a model without site effects?

# ANOVA in R: checking assumptions

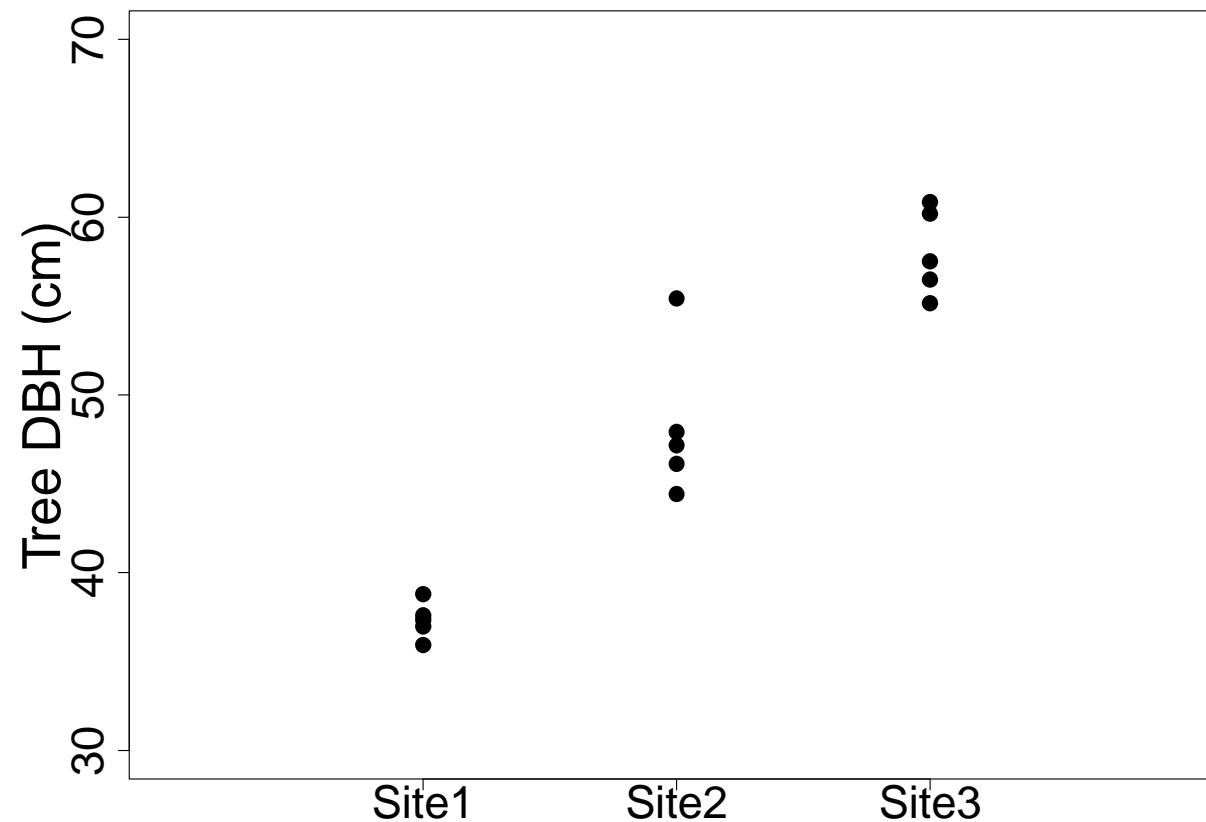
```
> par(mfrow=c(2,2))
```

```
> plot(model)
```



# One Way-ANOVA: one factor

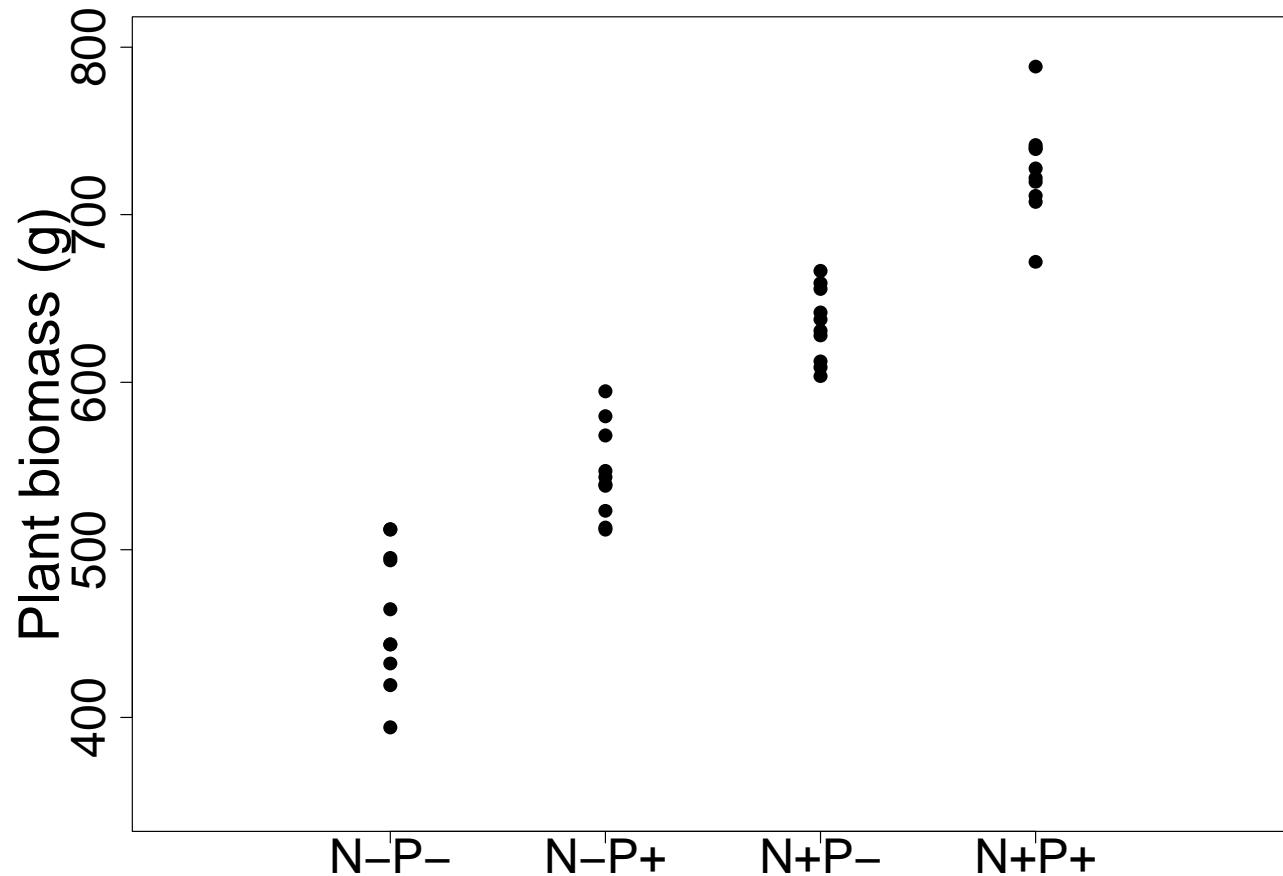
```
model <- aov(Y~X, data=data)
```



# Two Way-ANOVA: two factors

- $Y \sim X_1 * X_2$
- $Y$  = Plant biomass (g)
- N+/N- : Nitrogen addition / control
- P+/P- : Phosphate addition / control

# Two Way-ANOVA: two factors



# Two Way-ANOVA: two factors

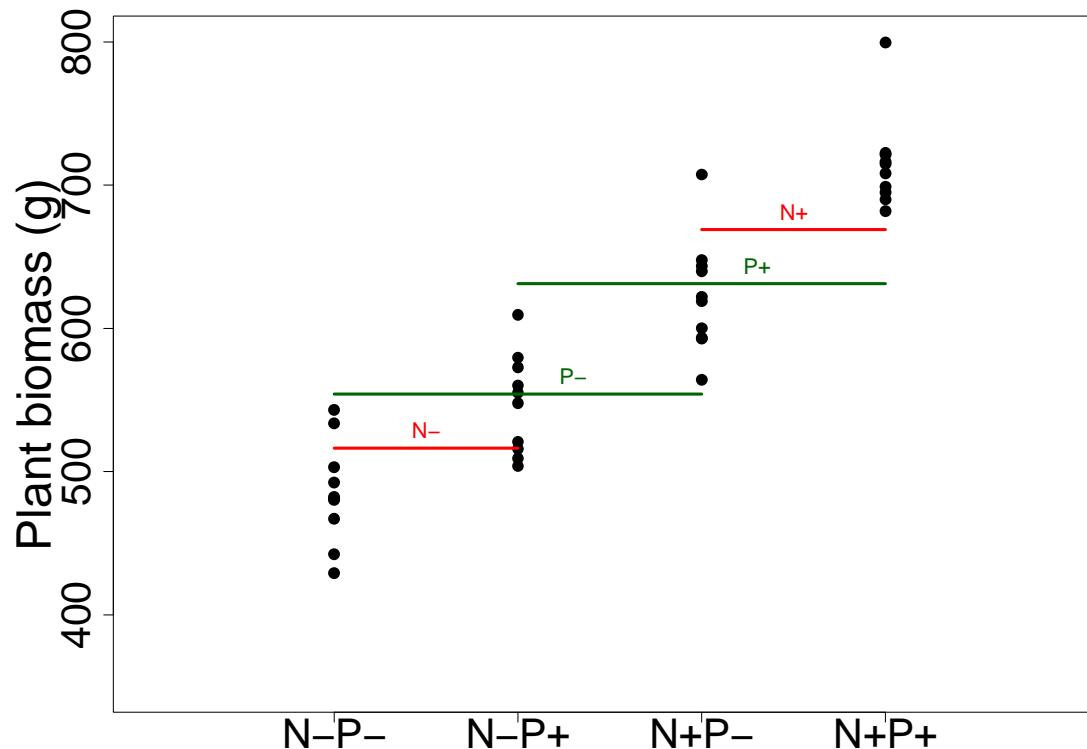
```
> model.aov <- aov(biomass ~ nitrogen * phosphate, data = fert)
> summary(model.aov)

          Df Sum Sq Mean Sq F value    Pr(>F)
nitrogen          1 191576  191576 232.983 < 2e-16 ***
phosphate        1   78295   78295  95.217 1.19e-11 ***
nitrogen:phosphate 1       1       1   0.001    0.976
Residuals         36  29602     822
```

# Two Way-ANOVA: two factors

```
> summary(model.aov)
```

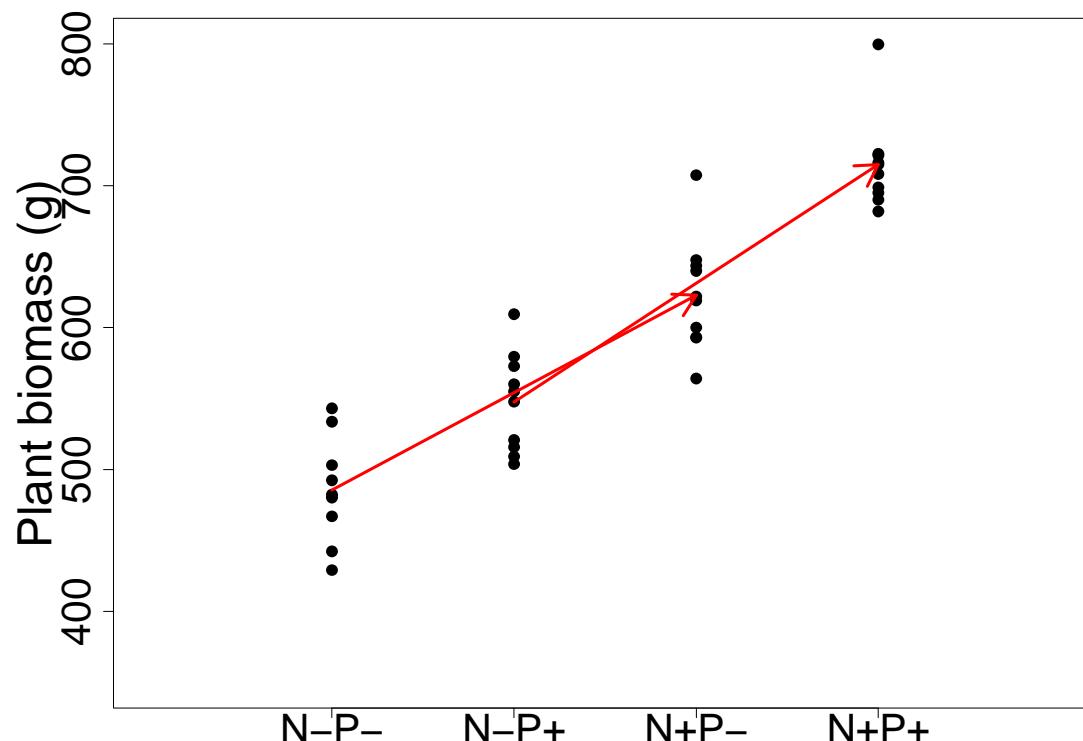
	Df	Sum Sq	Mean Sq	F value	Pr (>F)	
nitrogen	1	191576	191576	232.983	< 2e-16	***
phosphate	1	78295	78295	95.217	1.19e-11	***
nitrogen:phosphate	1	1	1	0.001	0.976	
Residuals	36	29602	822			



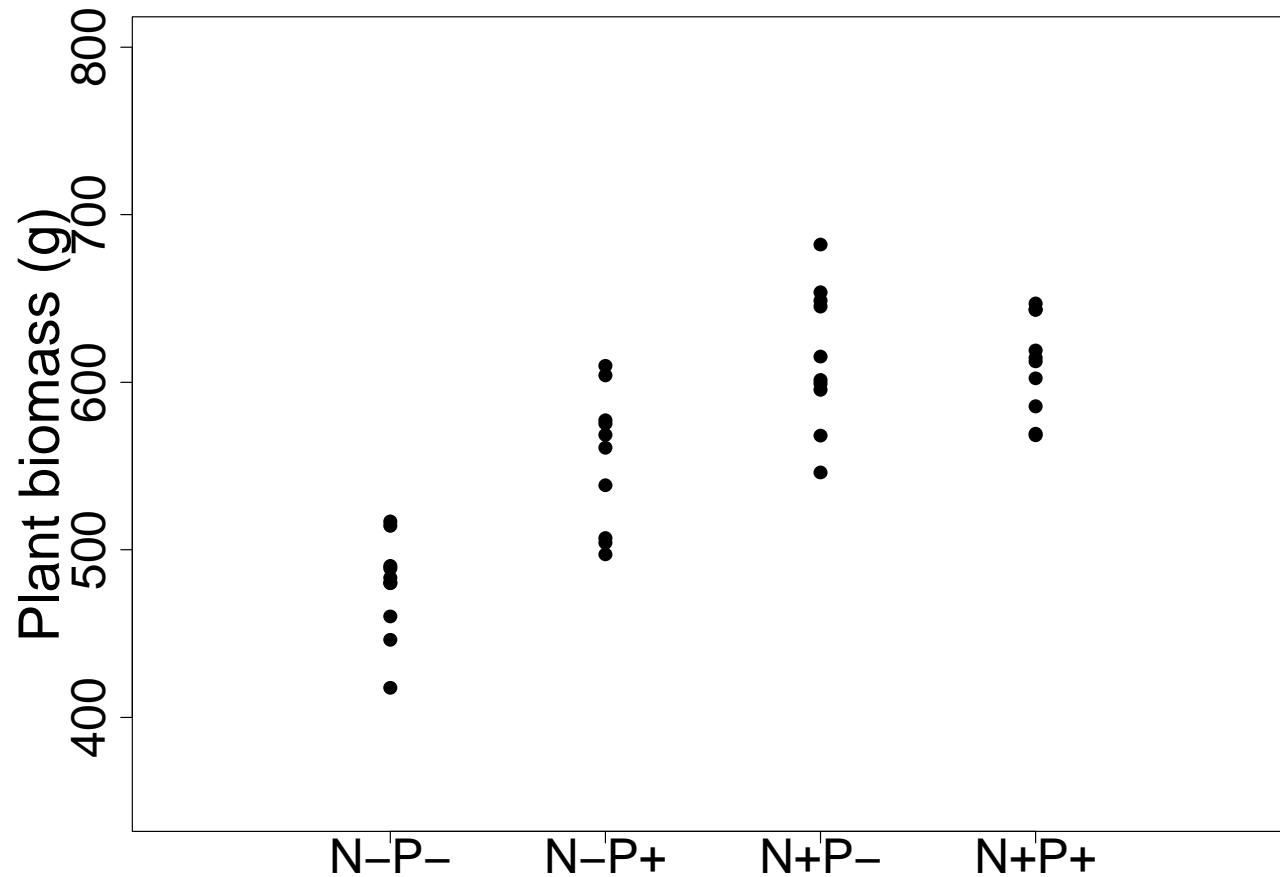
# Two Way-ANOVA: two factors

```
> summary(model.aov)
```

	Df	Sum Sq	Mean Sq	F value	Pr (>F)	
nitrogen	1	191576	191576	232.983	< 2e-16	***
phosphate	1	78295	78295	95.217	1.19e-11	***
nitrogen:phosphate	1	1	1	0.001	0.976	
Residuals	36	29602	822			



# Two Way-ANOVA: two factors



# Two Way-ANOVA: two factors

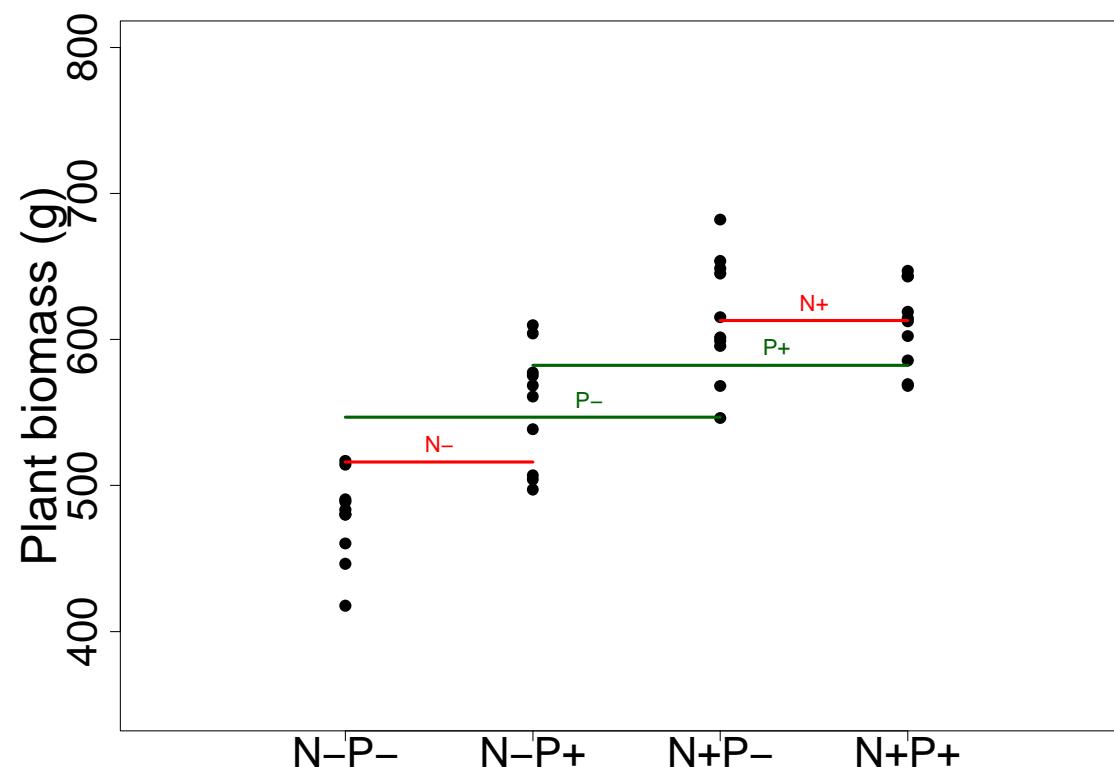
```
> model.aov <- aov(biomass ~ nitrogen * phosphate, data =  
fert)  
> summary(model.aov)
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
nitrogen	1	93956	93956	72.499	3.79e-10	***
phosphate	1	12747	12747	9.836	0.00340	**
nitrogen:phosphate	1	16586	16586	12.798	0.00101	**
Residuals	36	46655	1296			

# Two Way-ANOVA: two factors

```
> summary(model.aov)
```

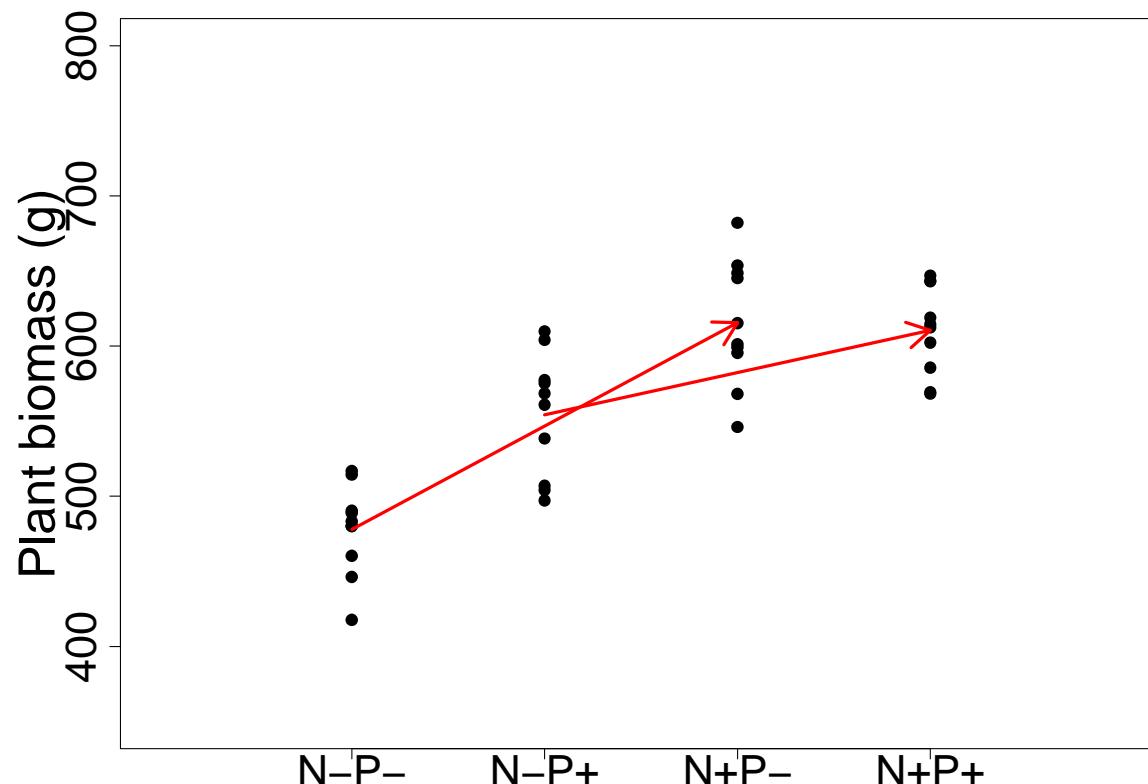
	Df	Sum Sq	Mean Sq	F value	Pr (>F)	
nitrogen	1	93956	93956	72.499	3.79e-10	***
phosphate	1	12747	12747	9.836	0.00340	**
nitrogen:phosphate	1	16586	16586	12.798	0.00101	**
Residuals	36	46655	1296			



# Two Way-ANOVA: two factors

```
> summary(model.aov)
```

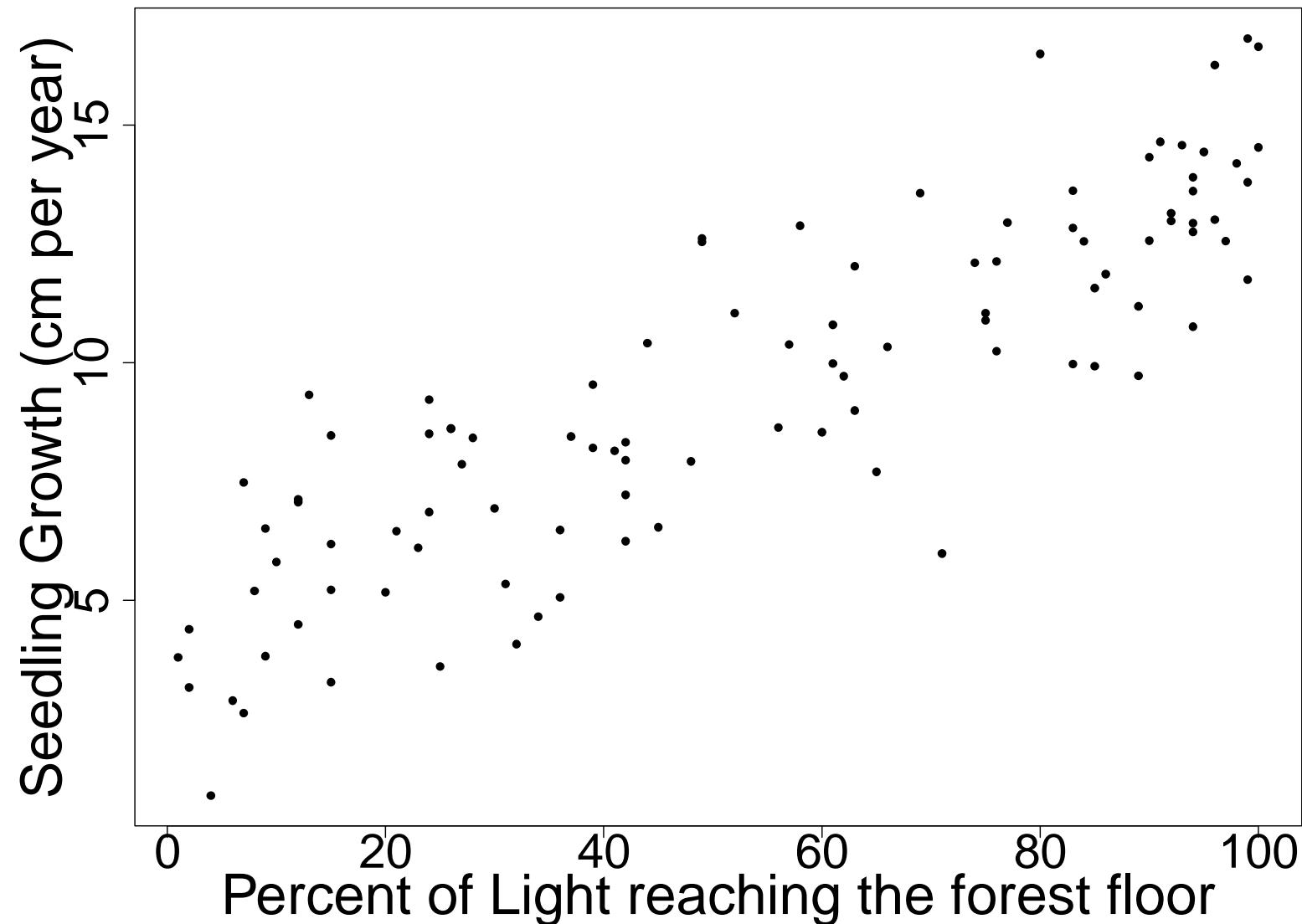
	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
nitrogen	1	93956	93956	72.499	3.79e-10	***
phosphate	1	12747	12747	9.836	0.00340	**
nitrogen:phosphate	1	16586	16586	12.798	0.00101	**
Residuals	36	46655	1296			



# Linear regression

- Same assumptions as ANOVA:
  - Independence
  - Normality
  - Homoscedasticity
- Here the explanatory variable **X is continuous**
- Instead of difference among groups, we want to model the **intercept** ( $a$ = value of  $Y$  when  $X=0$ ) and the **slope** ( $b$ ) of the regression.
- $Y \sim a + b*X$

# Linear regression



# Linear regression in R: lm()

```
> model.lm <- lm(growth ~ light, data = data)
> summary(model.lm)
```

Call:

```
lm(formula = growth ~ light, data = data)
```

Residuals:

Min	1Q	Median	3Q	Max
-5.1620	-1.1587	-0.0605	1.2966	4.4653

Coefficients:

	Estimate	Std. Error	t value	Pr(> t )		
(Intercept)	4.168827	0.362514	11.50	<2e-16 ***		
light	0.098287	0.005811	16.91	<2e-16 ***		
---						
Signif. codes:	0 '***'	0.001 '**'	0.01 '*'	0.05 '.'	0.1 ' '	1

Residual standard error: 1.854 on 98 degrees of freedom

Multiple R-squared: 0.7449, Adjusted R-squared: 0.7423

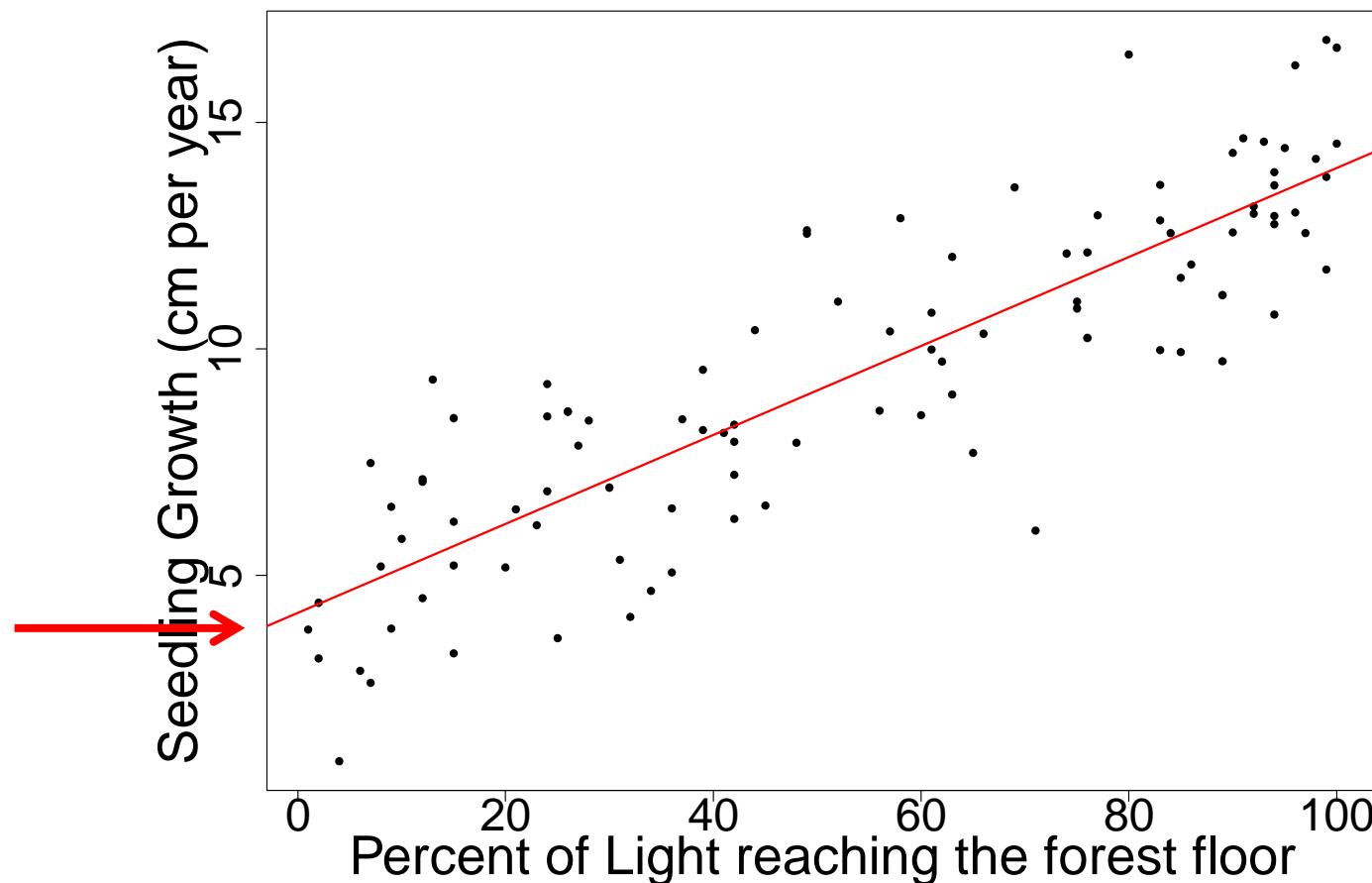
F-statistic: 286.1 on 1 and 98 DF, p-value: < 2.2e-16

# Linear regression in R: lm()

```
> summary(model.lm)
```

Coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	4.168827	0.362514	11.50	<2e-16 ***
light	0.098287	0.005811	16.91	<2e-16 ***

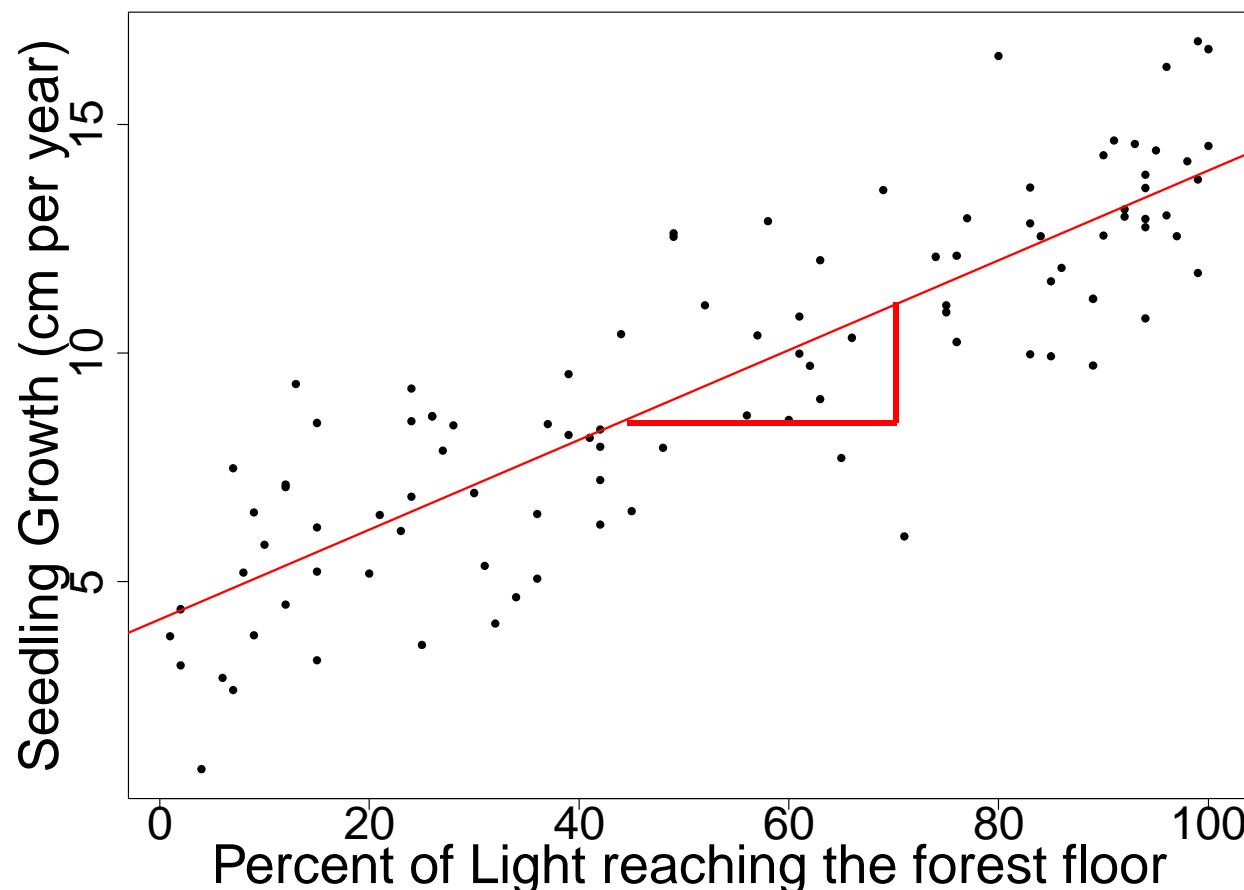


# Linear regression in R: lm()

```
> summary(model.lm)
```

Coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	4.168827	0.362514	11.50	<2e-16 ***
light	0.098287	0.005811	16.91	<2e-16 ***



# Linear regression in R: lm()

```
> model.lm <- lm(growth ~ light, data = data)
> summary(model.lm)
```

Call:

```
lm(formula = growth ~ light, data = data)
```

Residuals:

Min	1Q	Median	3Q	Max
-5.1620	-1.1587	-0.0605	1.2966	4.4653

Coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	4.168827	0.362514	11.50	<2e-16 ***
light	0.098287	0.005811	16.91	<2e-16 ***
---				

Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 1.854 on 98 degrees of freedom

Multiple R-squared: 0.7449, Adjusted R-squared: 0.7423

F-statistic: 286.1 on 1 and 98 DF, p-value: < 2.2e-16

SE of the mean:

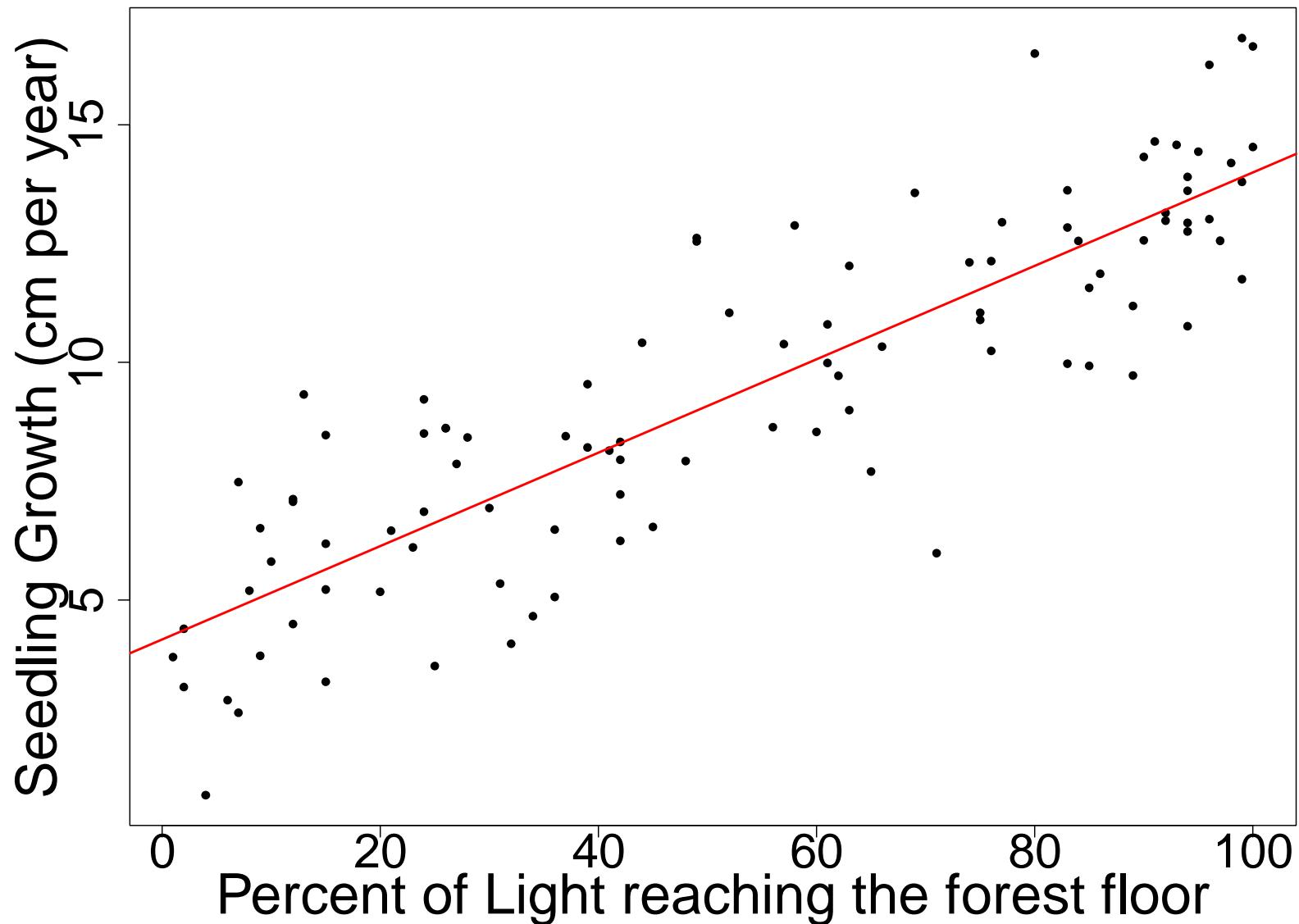
SD/ $\sqrt{n}$

SE: Standard Error

SD: Standard Deviation

n: population size

# Linear regression in R: lm()



# Linear regression in R: lm()

```
> model.lm <- lm(growth ~ light, data = data)
> summary(model.lm)
```

Call:

```
lm(formula = growth ~ light, data = data)
```

Residuals:

Min	1Q	Median	3Q	Max
-5.1620	-1.1587	-0.0605	1.2966	4.4653

Coefficients:

	Estimate	Std. Error	t value	Pr(> t )		
(Intercept)	4.168827	0.362514	11.50	<2e-16 ***		
light	0.098287	0.005811	16.91	<2e-16 ***		
---						
Signif. codes:	0 '***'	0.001 '**'	0.01 '*'	0.05 '.'	0.1 ' '	1

Residual standard error: 1.854 on 98 degrees of freedom  
Multiple R-squared: 0.7449, Adjusted R-squared: 0.7423  
F-statistic: 286.1 on 1 and 98 DF, p-value: < 2.2e-16

# Linear regression in R: lm()

```
> anova(model.lm)
```

```
Analysis of Variance Table
```

```
Response: growth
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
light	1	983.50	983.50	286.11	< 2.2e-16 ***
Residuals	98	336.88	3.44		

```
---
```

```
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

- Gives the same output as summary(aov(model.aov))

# ANOVA is a special case of LM

- Confusing because we can use the function `anova()` to get a table of analysis of variance whatever the model was made with (`aov()` or `lm()`)
- ANOVA = special case of LM when X:factors
- `lm()` in R works for `anova`, but the way it's shown is a bit different.
- Same assumptions

# ANOVA is a special case of LM

- In the end, you can always use `lm()` :
  - As long as your Y is continuous, and all the assumptions are met;
  - You can use the function `anova(model)` to get the degrees of freedom, SS, MS, F-test and p-value of every X variable in your model
  - You can use the function `summary(model)` to get the estimated means and SE of each levels, and the differences between levels

# Exercise: Forest: ANCOVA (ANalysis of COVAriance)

- Productivity: continuous Y variable that we want to explain
- Forest type: factorial X variable
- Species Diversity: continuous X variable
- ANCOVA:  $Y \sim \text{fact.X} * \text{cont.X}$