STAT 516 Lec 11

Analysis of covariance (ANCOVA)

Karl Gregory

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VO_2 max data from Kuehl (2000)

Change in VO_2 max of 12 males (after-minus-before) randomly assigned to two exercise programs (running, step aerobics). Ages recorded.

Group	*Age*	*Change*	*Group*	*Age*	*Change*
Aerobic	31	17.05	Running	23	− 0.87
	23	4.96		22	− 10.74
	27	10.40		22	− 3.27
	28	11.05		25	− 1.97
	22	0.26		27	7.50
	24	2.51		20	− 7.25
Mean	25.83	7.71		23.17	− 2.77
Std. Err.	1.40	2.55		1.01	2.54

Source: D. Allen, Exercise Physiology, University of Arizona.

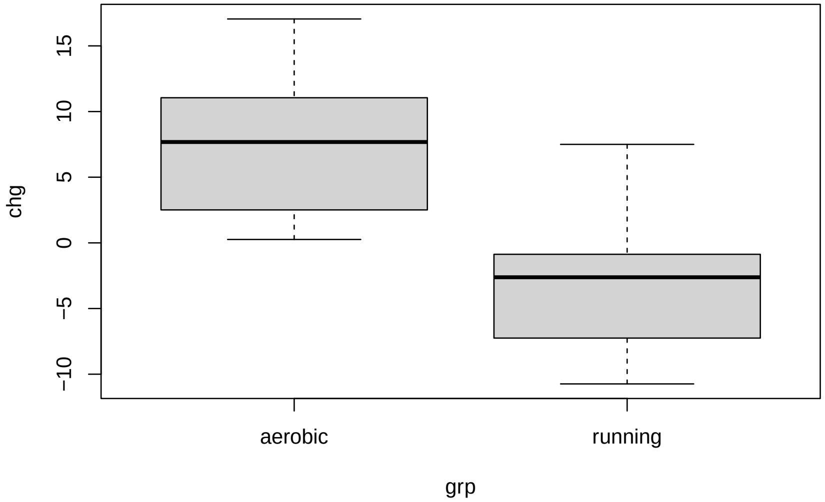
Which exercise programlead to a greater average change in VO_2 max?

What role does age play?

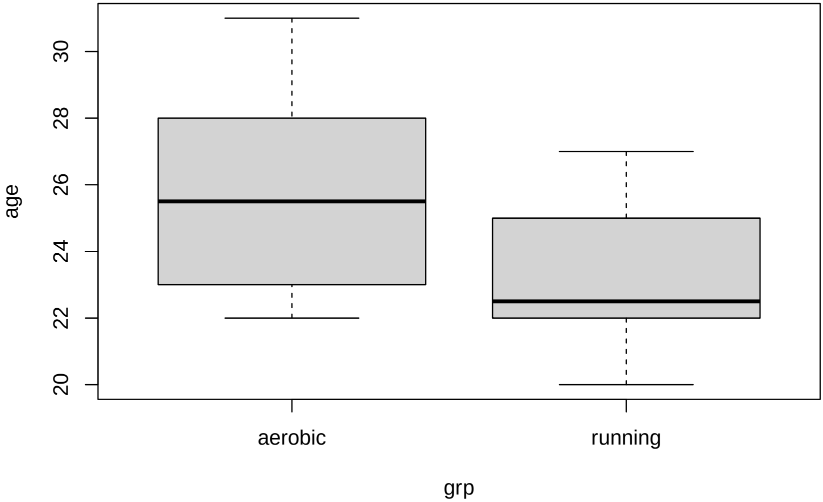
```r
vo2max <- data.frame(chg = c(17.05,4.96,10.40,11.05,0.26,2.51,
                             -0.87,-10.74,-3.27,-1.97,7.50,-7.25),
                     grp = as.factor(c(rep("aerobic",6),rep("running",6))),
                     age = c(31,23,27,28,22,24,23,22,22,25,27,20))

vo2max
```

```
      chg     grp age
1   17.05 aerobic  31
2    4.96 aerobic  23
3   10.40 aerobic  27
4   11.05 aerobic  28
5    0.26 aerobic  22
6    2.51 aerobic  24
7   -0.87 running  23
8  -10.74 running  22
9   -3.27 running  22
10  -1.97 running  25
11   7.50 running  27
12  -7.25 running  20
```

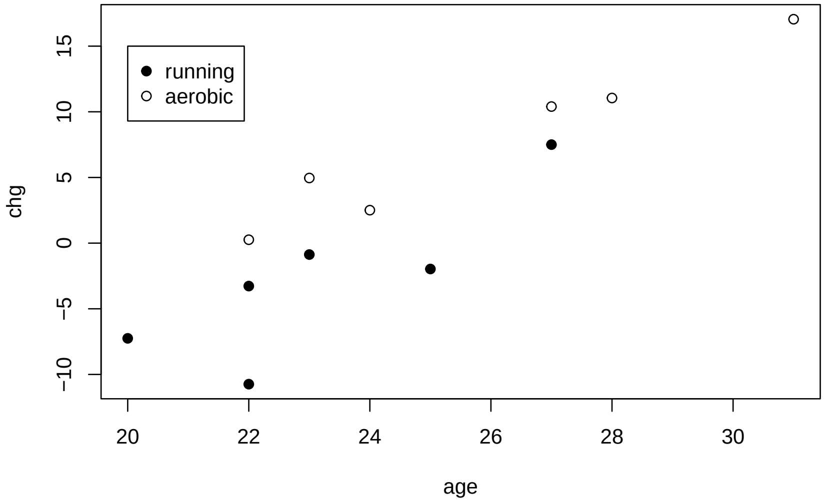
```r
boxplot(chg ~ grp, data = vo2max)
```



```r
boxplot(age ~ grp, data = vo2max)
```



```r
plot(chg ~ age, pch = ifelse(grp == "running",19,1), data = vo2max)
legend(x = 20,y = 15,legend = c("running","aerobic"), pch = c(19,1))
```



Analysis of covariance

- Useful when EUs are not homogeneous.
- Measurement capturing EU inhomogeneity is called a <u>covariate</u>.
- Can isolate treatment effects even when EUs differ across treatment groups.

Single-slope analysis of covariance (ANCOVA) model

Assume

$$Y_{ij} = \mu + \tau_i + \beta x_{ij} + \varepsilon_{ij}$$

for $i = 1, \dots, a$, $j = 1, \dots, n_i$, where

- Y_{ij} is the response of EU j in treatment group i.
- μ is a baseline or overall mean.
- the τ_i are treatment effects.
- the x_{ij} are <u>covariate</u> values measured on the EUs.
- β is a slope coefficient expressing the effect of the covariate.
- the ε_{ij} are independent Normal$(0, \sigma_\varepsilon^2)$ error terms.
- $n_1 + \cdots + n_a = N$. Unbalancedness not an issue.

Set $\mu_i = \mu + \tau_i$ for $i = 1, \dots, a$.

Goals in analysis of covariance

1. Estimate the parameters μ, $\tau_1, \ldots, \tau_a$, and β.
2. Visualize the data.
3. Fit full and reduced models and collect error sums of squares.
4. Test whether the covariate has any effect.
5. Test for a treatment effect.
6. Adjust treatment group means for the inhomogeneity of the EUs.
7. Compare the adjusted group means.

Parameter constraints

- There are a treatment groups and one covariate slope.
- The model has $a + 1$ parameters, which is one too-many.
- R will set $\tau_1 = 0$ so that it can estimate all the parameters.

VO_2 max data (cont)

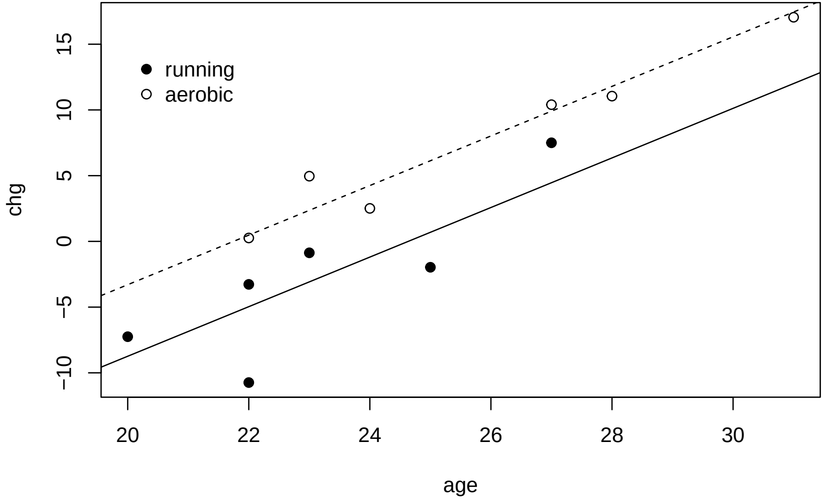
```r
lm_out <- lm(chg ~ grp + age, data = vo2max)
summary(lm_out)
```

```

Call:
lm(formula = chg ~ grp + age, data = vo2max)

Residuals:
    Min      1Q  Median      3Q     Max 
-5.7731 -0.9902  0.1395  1.8254  3.0374 

Coefficients:
            Estimate Std. Error t value Pr(>|t|)    
(Intercept) -41.0139     7.7144  -5.317 0.000483 ***
grprunning   -5.4426     1.7965  -3.030 0.014255 *  
age           1.8859     0.2953   6.386 0.000127 ***
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 2.797 on 9 degrees of freedom
Multiple R-squared:  0.902,	Adjusted R-squared:  0.8802 
F-statistic: 41.42 on 2 and 9 DF,  p-value: 2.887e-05
```

```r
parms <- coef(lm_out)
parms
```

```
(Intercept)  grprunning         age 
 -41.013882   -5.442621    1.885892 
```

```r
plot(chg ~ age, pch = ifelse(grp == "running",19,1), data = vo2max)
abline(parms[1] + parms[2],parms[3])
abline(parms[1],parms[3],lty = 2)
legend(x = 20,y = 15,legend = c("running","aerobic"), pch = c(19,1), bty = "n")
```



Full and reduced models in ANCOVA

We construct test statistics by fitting full and reduced models:

- Full model: $Y_{ij} = \mu + \tau_i + \beta x_{ij} + \varepsilon_{ij}$
- Reduced model (no covariate effect): $Y_{ij} = \mu + \tau_i + \varepsilon_{ij}$
- Reduced model (no treatment effect): $Y_{ij} = \mu + \beta x_{ij} + \varepsilon_{ij}$

Full- and reduced-model error sums of squares

SS	Formula
$\mathrm{SS}_{\mathrm{Error}}$(Full)	$\sum_{i=1}^{a} \sum_{j=1}^{n_{ij}} (Y_{ij} - (\hat{\mu} + \hat{\tau}_i + \hat{\beta} x_{ij}))^2$
$\mathrm{SS}_{\mathrm{Error}}$(No covariate)	$\sum_{i=1}^{a} \sum_{j=1}^{n_{ij}} (Y_{ij} - (\hat{\mu} + \hat{\tau}_i))^2$
$\mathrm{SS}_{\mathrm{Error}}$(No treatment)	$\sum_{i=1}^{a} \sum_{j=1}^{n_{ij}} (Y_{ij} - (\hat{\mu} + \hat{\beta} x_{ij}))^2$

Now define these difference in error sums of squares:

- $\mathrm{SS}_{\mathrm{Cov}} = \mathrm{SS}_{\mathrm{Error}}$(No covariate) $- \mathrm{SS}_{\mathrm{Error}}$(Full)
- $\mathrm{SS}_{\mathrm{Trt}} = \mathrm{SS}_{\mathrm{Error}}$(No treatment) $- \mathrm{SS}_{\mathrm{Error}}$(Full)

ANCOVA table

Source	Df	SS	MS	F value
Covariate	1	$\mathrm{SS}_{\mathrm{Cov}}$	$\mathrm{MS}_{\mathrm{Cov}}$	$\mathrm{F}_{\mathrm{Cov}} = \mathrm{MS}_{\mathrm{Cov}} \,/\, \mathrm{MS}_{\mathrm{Error}}$
Treatment	$a-1$	$\mathrm{SS}_{\mathrm{Trt}}$	$\mathrm{MS}_{\mathrm{Trt}}$	$\mathrm{F}_{\mathrm{Trt}} = \mathrm{MS}_{\mathrm{Trt}} \,/\, \mathrm{MS}_{\mathrm{Error}}$
Error	$N-a-1$	$\mathrm{SS}_{\mathrm{Error}}$	$\mathrm{MS}_{\mathrm{Error}}$	

1. Reject H_0: $\beta = 0$ at α if $\mathrm{F}_{\mathrm{Cov}} > F_{1,N-a-1,\alpha}$.
2. Reject H_0: $\mu_1 = \cdots = \mu_a$ at α if $\mathrm{F}_{\mathrm{Trt}} > F_{a-1,N-a-1,\alpha}$.

VO_2 max data (cont)

Using `anova()` on the `lm()` output gives the wrong SS (gives sequential).

Use `Anova()` from R package car on the `lm()` output.

```
library(car) # first time run install.packages("car")
# Use type = "II" or type = "III"
Anova(lm_out,type = "III")
```

```
Anova Table (Type III tests)

Response: chg
            Sum Sq Df F value    Pr(>F)
(Intercept) 221.06  1 28.2653 0.0004832 ***
grp          71.79  1  9.1788 0.0142548 *
age         318.91  1 40.7759 0.0001274 ***
Residuals    70.39  9
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
lm_nocov <- lm(chg ~ grp, data = vo2max) # reduced model with no covariate
lm_notrt <- lm(chg ~ age, data = vo2max) # reduced model with no treatment
lm_full <- lm(chg ~ age + grp, data = vo2max) # full model

SSE_nocov <- sum(lm_nocov$resid^2)
SSE_notrt <- sum(lm_notrt$resid^2)
SSE_full <- sum(lm_full$resid^2)

SSCov <- SSE_nocov - SSE_full
SSTrt <- SSE_notrt - SSE_full
SSE <- SSE_full

a <- 2
N <- nrow(vo2max)

MSCov <- SSCov / 1
MSTrt <- SSTrt / (a-1)
MSE <- SSE / (N - a - 1)

FCov <- MSCov / MSE
FTrt <- MSTrt / MSE

pCov <- 1 - pf(FCov,1,N - a - 1)
pTrt <- 1 - pf(FTrt,a-1,N - a - 1)
```

Treatment means in terms of parameter estimates

- We can write the treatment group means as

$$\bar{Y}_{i.} = \hat{\mu} + \hat{\tau}_i + \hat{\beta}\bar{x}_{i.} \quad \text{for } i = 1, \ldots, a,$$

- We also define the covariate-adjusted means

$$\bar{Y}_{i.}^{\text{adj}} = \hat{\mu} + \hat{\tau}_i + \hat{\beta}\bar{x}_{..} \quad \text{for } i = 1, \ldots, a,$$

- We can equivalently write covariate-adjusted means as

$$\bar{Y}_{i.}^{\text{adj}} = \bar{Y}_{i.} - \hat{\beta}(\bar{x}_{i.} - \bar{x}_{..}) \quad \text{for } i = 1, \ldots, a.$$

Variances of covariate-adjusted means and their differences

Contrast	Variance
$\bar{Y}_{i.}^{\text{adj}}$	$\sigma^2 \left[\frac{1}{n_i} + \frac{(\bar{x}_{i.} - \bar{x}_{..})^2}{E_{xx}} \right]$
$\bar{Y}_{i.}^{\text{adj}} - \bar{Y}_{i'.}^{\text{adj}}$	$\sigma^2 \left[\frac{1}{n_i} + \frac{1}{n_{i'}} + \frac{(\bar{x}_{i.} - \bar{x}_{i'.})^2}{E_{xx}} \right]$

where $E_{xx} = \sum_{i=1}^{a} \sum_{j=1}^{n_i} (x_{ij} - \bar{x}_{i.})^2$.

Some (unadjusted) CIs in the ANCOVA model

Define the true or population-level covariate-adjusted means as

$$\mu_i^{\text{adj}} = \mu + \tau_i + \beta\bar{x}_{..} \quad \text{for } i = 1, \ldots, a.$$

Target	$(1-\alpha)100\%$ confidence interval
μ_i^{adj}	$\bar{Y}_{i.}^{\text{adj}} \pm t_{N-a-1,\alpha/2}\sqrt{\text{MS}_{\text{Error}}}\sqrt{\dfrac{1}{n_i} + \dfrac{(\bar{x}_{i.} - \bar{x}_{..})^2}{E_{xx}}}$
$\mu_i^{\text{adj}} - \mu_{i'}^{\text{adj}}$	$\bar{Y}_{i.}^{\text{adj}} - \bar{Y}_{i'.}^{\text{adj}} \pm t_{N-a-1,\alpha/2}\sqrt{\text{MS}_{\text{Error}}}\sqrt{\dfrac{1}{n_i} + \dfrac{1}{n_{i'}} + \dfrac{(\bar{x}_{i.} - \bar{x}_{i'.})^2}{E_{xx}}}$

VO_2 max data (cont)

```
y1. <- mean(vo2max$chg[vo2max$grp == "aerobic"])
y2. <- mean(vo2max$chg[vo2max$grp == "running"])

x1. <- mean(vo2max$age[vo2max$grp == "aerobic"])
x2. <- mean(vo2max$age[vo2max$grp == "running"])
x.. <- mean(vo2max$age)

bhat <- coef(lm_out)[3]

y1.adj <- y1. - bhat * (x1. - x..)
y2.adj <- y2. - bhat * (x2. - x..)
```

```r
n1 <- 6
n2 <- 6
alpha <- 0.05

tval <- qt(1-alpha/2,N-a-1)
age1 <- vo2max$age[vo2max$grp == "aerobic"]
age2 <- vo2max$age[vo2max$grp == "running"]
Ex1 <- sum((age1 - mean(age1))^2)
Ex2 <- sum((age2 - mean(age2))^2)
Exx <- Ex1 + Ex2

se1 <- sqrt(MSE) * sqrt(1/n1 + (x1. - x..)^2 / Exx)
lo1 <- y1.adj - tval * se1
up1 <- y1.adj + tval * se1

se2 <- sqrt(MSE) * sqrt(1/n2 + (x2. - x..)^2 / Exx)
lo2 <- y2.adj - tval * se2
up2 <- y2.adj + tval * se2

se12 <- sqrt(MSE) * sqrt(1/n1 + 1/n2 + (x1. - x2.)^2 / Exx)
lo12 <- y1.adj - y2.adj - tval * se12
up12 <- y1.adj - y2.adj + tval * se12
```

Table 5: Mean change in VO_2 max in aerobic and running groups

	Unadjusted	Age-adjusted(CI)
Aerobic	7.71	5.19 (2.46,7.92)
Running	-2.77	-0.25 (-2.98,2.48)
Aerobic - Running	10.47	5.44 (1.38,9.51)

Soybean data from Dr. Longnecker's notes

Soybean plants assigned to three greenhouse conditions: Supplemental lighting (SL), partial shading (PS), and control (C). The response was seed yield. The pre-treatment height of each plant was also recorded.

Yield	Height	TRT	Yield	Height	TRT	Yield	Height	TRT	Yield	Height	TRT	Yield	Height	TRT
12.2	45	C	12.4	52	C	11.9	42	C	11.3	35	C	11.8	40	C
12.1	48	C	13.1	60	C	12.7	61	C	12.4	50	C	11.4	33	C
12.3	48	C	12.2	51	C	12.6	56	C	13.2	65	C	12.3	51	C
16.6	63	SL	15.8	50	SL	16.5	63	SL	15.0	33	SL	15.4	38	SL
15.6	45	SL	15.8	50	SL	15.8	48	SL	16.0	50	SL	15.8	49	SL
15.0	35	SL	16.2	50	SL	16.7	62	SL	15.8	49	SL	15.9	52	SL
9.5	52	PS	9.5	54	PS	9.6	58	PS	8.8	45	PS	9.5	57	PS
9.8	62	PS	9.1	52	PS	10.3	67	PS	9.5	55	PS	8.5	40	PS
8.6	41	PS	10.4	67	PS	9.4	55	PS	10.2	66	PS	9.3	56	PS

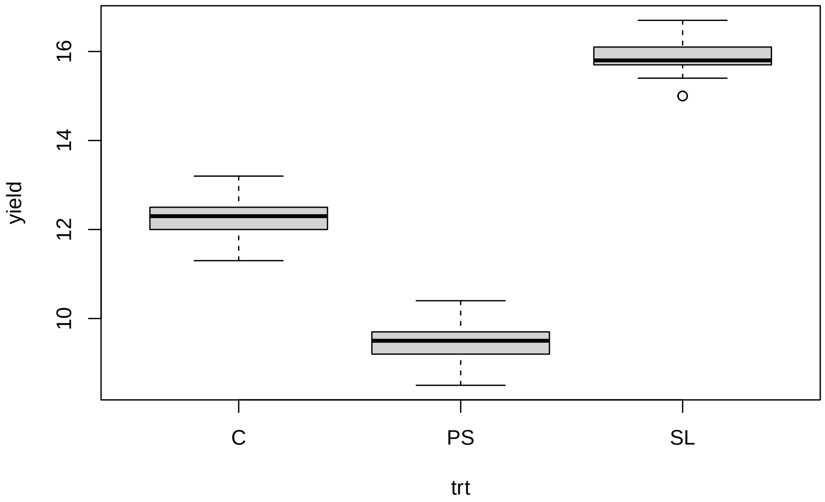
Do the greenhouse conditions effect the seed yield?

What is the role of plant height (proxy for plant vigor)?

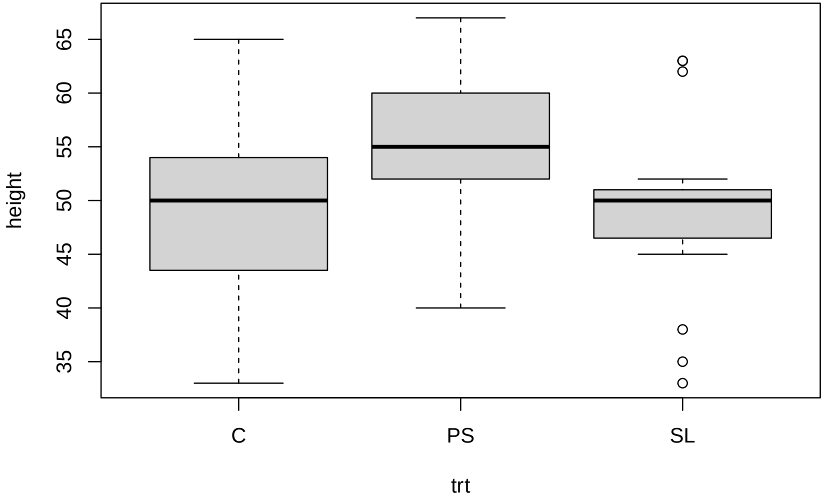
```r
soybean <- data.frame(yield = c(12.2,12.1,12.3,16.6,15.6,15.0,9.5,9.8,8.6,
                                12.4,13.1,12.3,15.8,15.8,16.2,9.5,9.1,10.4,
                                11.9,12.7,12.6,16.5,15.8,16.7,9.6,10.3,9.4,
                                11.3,12.4,13.2,15.0,16.0,15.8,8.8,9.5,10.2,
                                11.8,11.4,12.3,15.4,15.8,15.9,9.5,8.5,9.3),
                      trt = as.factor(rep(c(rep("C",3),rep("SL",3),rep("PS",3)),5)),
                      height = c(45,48,48,63,45,35,52,62,41,
                                 52,60,51,50,50,50,54,52,67,
                                 42,61,56,63,48,62,58,67,55,
                                 35,50,65,33,50,49,45,55,66,
                                 40,33,51,38,49,52,57,40,56))
head(soybean,n = 12)
```

```
   yield trt height
1   12.2   C     45
2   12.1   C     48
3   12.3   C     48
4   16.6  SL     63
5   15.6  SL     45
6   15.0  SL     35
7    9.5  PS     52
8    9.8  PS     62
9    8.6  PS     41
10  12.4   C     52
11  13.1   C     60
12  12.3   C     51
```

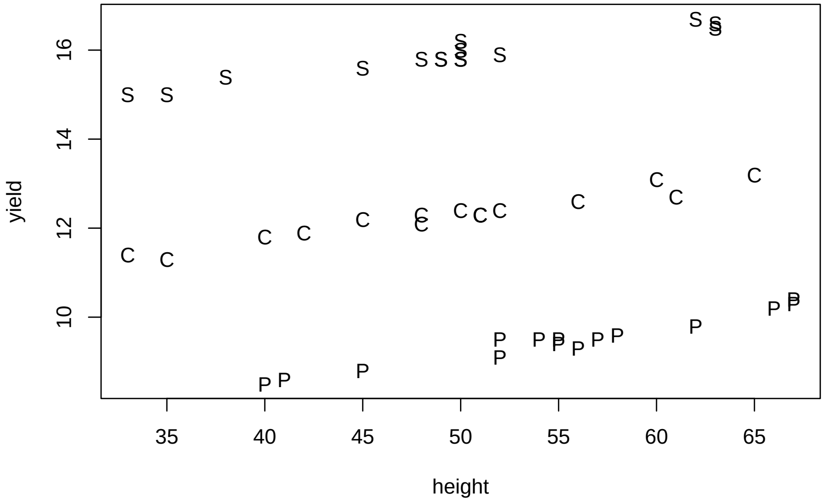
```r
boxplot(yield~trt, data = soybean)
```



```r
boxplot(height~trt, data = soybean)
```



```
plot(yield ~ height, pch = as.character(trt), data = soybean)
```



ANCOVA allowing different slopes in each group

Assume

$$Y_{ij} = \mu + \tau_i + (\beta + (\tau\beta)_i)x_{ij} + \varepsilon_{ij}$$

for $i = 1, \dots, a$, $j = 1, \dots, n_i$, where

- Y_{ij} is the response of EU j in treatment group i.
- μ is a baseline or overall mean.
- the τ_i are treatment effects.
- the x_{ij} are <u>covariate</u> values measured on the EUs.
- β is a slope coefficient expressing the effect of the covariate.
- the $(\tau\beta)_i$ allow interaction between the treatment and the covariate.
- the ε_{ij} are independent Normal$(0, \sigma_\varepsilon^2)$ error terms.
- $n_1 + \cdots + n_a = N$. Unbalancedness not an issue.

Set $\mu_i = \mu + \tau_i$ and $\beta_i = \beta + (\tau\beta)_i$ for $i = 1, \dots, a$.

Parameter constraints in multiple slopes model

- There are a treatment groups and a covariate slopes.
- The model has $2(a+1)$ parameters, which is two too-many.
- R will set $\tau_1 = 0$ and $(\tau\beta)_1 = 0$ to make all parameters estimable.

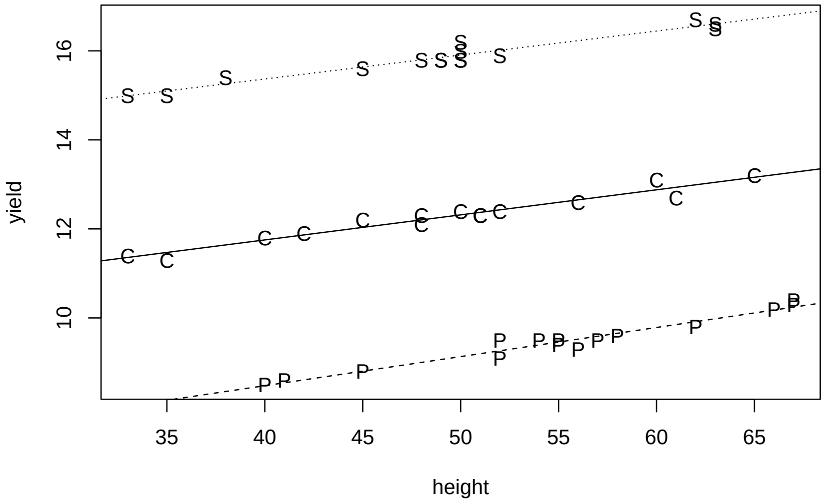
```r
lm_out <- lm(yield ~ trt + height + trt:height, data = soybean)
summary(lm_out)
```

```
Call:
lm(formula = yield ~ trt + height + trt:height, data = soybean)

Residuals:
      Min        1Q    Median        3Q       Max 
-0.234733 -0.088745 -0.003954  0.057644  0.293320 

Coefficients:
              Estimate Std. Error t value Pr(>|t|)    
(Intercept)   9.500573   0.181934  52.220  < 2e-16 ***
trtPS        -3.639588   0.285527 -12.747 1.74e-15 ***
trtSL         3.713050   0.258575  14.360  < 2e-16 ***
height        0.056298   0.003644  15.451  < 2e-16 ***
trtPS:height  0.009102   0.005372   1.694   0.0982 .  
trtSL:height -0.002437   0.005179  -0.470   0.6407    
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 0.1256 on 39 degrees of freedom
Multiple R-squared:  0.9981,	Adjusted R-squared:  0.9978 
F-statistic:  4054 on 5 and 39 DF,  p-value: < 2.2e-16
```

```r
parms <- coef(lm_out)
parms
```

```
 (Intercept)        trtPS        trtSL       height trtPS:height trtSL:height
 9.500572519 -3.639588070  3.713050304  0.056297710  0.009101605 -0.002436573
```

```
plot(yield ~ height, pch = as.character(trt), data = soybean)
abline(parms[1],parms[4],lty = 1)
abline(parms[1] + parms[2],parms[4] + parms[5],lty = 2)
abline(parms[1] + parms[3],parms[4] + parms[6],lty = 3)
```



An F-test for equal slopes

Define the error sums of squares:

SS	Formula
$\mathrm{SS}_{\mathrm{Error}}$(Full)	$\sum_{i=1}^{a}\sum_{j=1}^{n_{ij}}(Y_{ij}-(\hat{\mu}+\hat{\tau}_i+(\hat{\beta}+(\widehat{\tau\beta})_i)x_{ij}))^2$
$\mathrm{SS}_{\mathrm{Error}}$(Equal slopes)	$\sum_{i=1}^{a}\sum_{j=1}^{n_{ij}}(Y_{ij}-(\hat{\mu}+\hat{\tau}_i+\hat{\beta}x_{ij}))^2$

Now set

$$\mathrm{F}_{\mathrm{T\times C}}=\frac{[\mathrm{SS}_{\mathrm{Error}}(\text{Equal slopes})-\mathrm{SS}_{\mathrm{Error}}(\text{Full})]/(a-1)}{\mathrm{SS}_{\mathrm{Error}}(\text{Full})/(N-2a)}$$

Reject H_0: *Equal slopes* at α if $\mathrm{F}_{\mathrm{T\times C}}>F_{a-1,N-2a,\alpha}$.

Soybean data (cont)

```
lm_eqslp <- lm(yield ~ trt + height, data = soybean) # equal slopes model
lm_full <- lm(yield ~ trt + height + trt:height, data = soybean) # full model

SSE_eqslp <- sum(lm_eqslp$resid^2)
SSE_full <- sum(lm_full$resid^2)

a <- 3
N <- nrow(soybean)

FTrtCov <- ((SSE_eqslp - SSE_full) / (a-1)) / (SSE_full / (N - 2*a))
pTrtCov <- 1 - pf(FTrtCov,a-1,N - 2*a)
```

Same as interaction p value from `anova()` on the `lm()` output.

```r
anova(lm_out)
```

```
Analysis of Variance Table

Response: yield
            Df  Sum Sq Mean Sq   F value  Pr(>F)
trt          2 308.134 154.067 9770.7982 < 2e-16 ***
height       1  11.389  11.389  722.2854 < 2e-16 ***
trt:height   2   0.079   0.039    2.4938 0.09568 .
Residuals   39   0.615   0.016
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

When slopes are unequal

If we reject H_0: *Equal slopes*, then

- We can compute covariate-adjusted means with the different slopes.
- We can compare the covariate adjusted means—but the CI formulas are different from the ones above (details omitted).

References

Kuehl, R. O. 2000. *Design of Experiments: Statistical Principles of Research Design and Analysis*. Duxbury/Thomson Learning.