

Linear mixed models, part 1

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Objectives

- Apply a linear mixed model to grouped data.
- Explain how a mixed model is a compromise between a model ignoring the group effect and a model with fixed group effects.
- Determine the situations where it is most beneficial to use a mixed model.

Motivation

Example: Growth of white spruce on several sites

Suppose you need to model the growth of white spruce at multiple sites within a region. You measured the following numeric variables on a total sample of 100 trees, spread across multiple sites:

- annual growth (response variable);
 - diameter (DBH);
 - age;
 - competition index (CI, based on the number and size of neighboring trees).
- (a) What would be the equation of a linear model (e.g. $y \sim x$) representing the relationship between annual growth and the three predictors?

- (b) If the mean growth varies by site, how can you include this effect in your model?
- (c) If the effects of diameter, age and competition index on growth vary by site, how do you account for this in your model?
- (d) Do the methods proposed in (b) and (c) work best with few sites (e.g. 100 trees across 3 sites) or many sites (e.g. 100 trees across 20 sites)? Do they work better if the number of trees sampled by site is similar (e.g. 5 trees at each of the 20 sites) or variable (e.g. 2 to 30 trees depending on the site)?
- (e) Can you use this model to predict the growth of white spruce at a new site?

Suppose now that you know some characteristics of each site that could explain the differences in growth between sites, for example:

- the drainage class (categorical variable);
- the slope of the site (numerical variable).

- (a) How can you include these variables in your model?
- (b) Does the model proposed in (a) work better with few sites or many sites?
- (c) Can you use that model to predict the growth of white spruce at a new site?

Finally, suppose you measured growth and other individual predictors on the same 100 white spruce trees for three consecutive years.

- (a) How could you add year-to-year growth variation in your model?
- (b) If there is a weather station in the region, would it be useful to add the effect of weather variables (e.g. mean temperature) to explain this interannual variation?
- (c) From a statistical point of view, does measuring the same trees each year influence the accuracy of the regression estimates?

Example: Benthic fauna in the Netherlands

The rikz.csv dataset, taken from the textbook by Zuur et al. (see references at the bottom of this page), presents data on the benthic communities of 9 beaches in the Netherlands. Species richness was measured for 5 sites on each of the 9 beaches for a total of 45 observations. The NAP variable measures the vertical position of each site relative to mean sea level, while the exposure index (**Exposure**) is measured at the beach scale.

```
rikz <- read.csv("../donnees/rikz.csv")
# Correct the representation of categorical variables
rikz <- mutate(rikz, Beach = as.factor(Beach),
               Exposure = as.factor(Exposure))
head(rikz)
```

##	Sample	Richness	Exposure	NAP	Beach
## 1	1	11	10	0.045	1
## 2	2	10	10	-1.036	1
## 3	3	13	10	-1.336	1
## 4	4	11	10	0.616	1
## 5	5	10	10	-0.684	1
## 6	6	8	8	1.190	2

Since the response is a number of species, a Poisson regression might be more appropriate. However, we will not cover generalized linear mixed models in this class. Therefore, we apply a square root transformation to the response to use linear regression. This transformation generally improves the homogeneity of variance for count data.

```
rikz <- mutate(rikz, srich = sqrt(Richness))
```

From these data, we will illustrate different strategies to account for the grouped nature of the data (9 beaches) and the group-level predictor (exposure index).

Model 1: Ignore groups

The first model includes only the site-level predictor (NAP), with an adjusted $R^2 = 0.46$. The effect of this predictor is significant and indicates that the higher sites have a lower species richness.

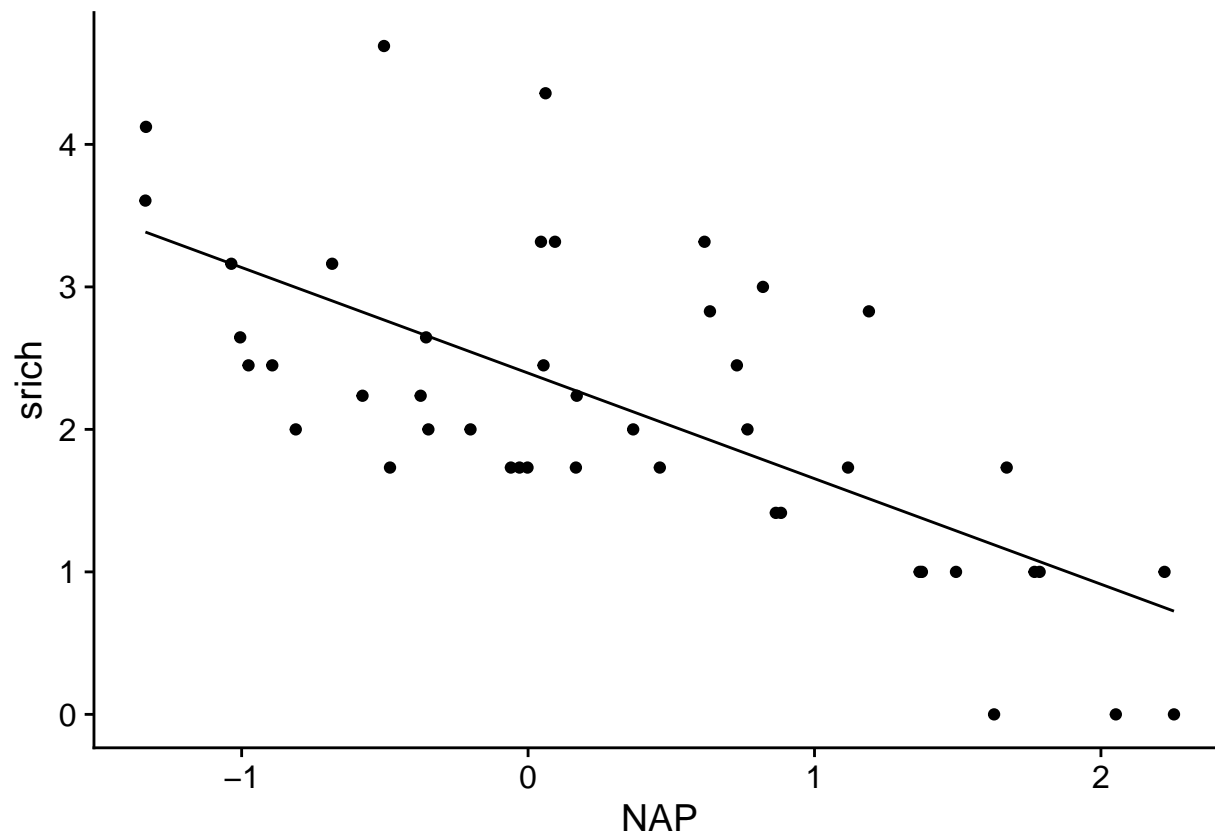
```
mod1 <- lm(srich ~ NAP, rikz)
summary(mod1)

##
## Call:
## lm(formula = srich ~ NAP, data = rikz)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -1.1898 -0.5874 -0.1233  0.2603  2.0089
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)   2.3952     0.1241  19.295 < 2e-16 ***
## NAP          -0.7409     0.1190  -6.224 1.72e-07 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.7851 on 43 degrees of freedom
## Multiple R-squared:  0.474, Adjusted R-squared:  0.4617
## F-statistic: 38.74 on 1 and 43 DF, p-value: 1.724e-07
```

Here is the line representing model predictions as a function of NAP.

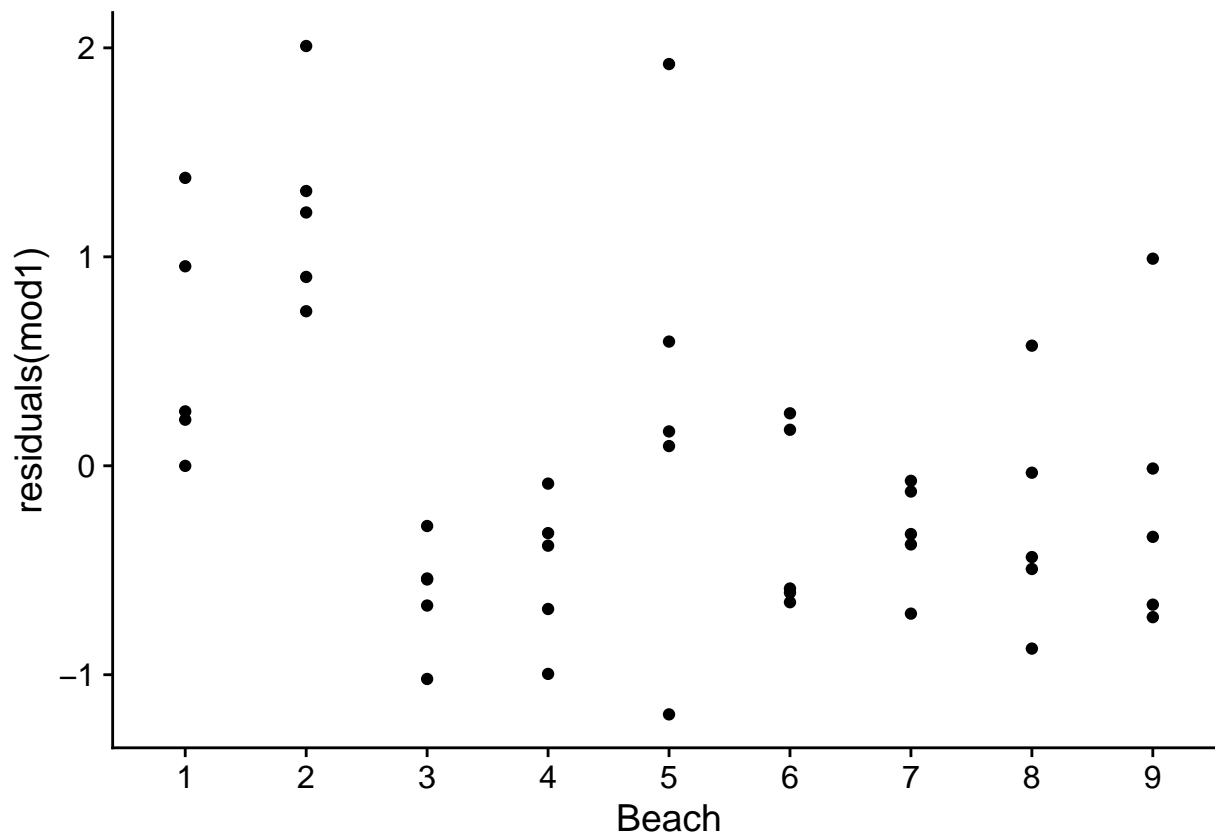
```
rikz$fit1 <- fitted(mod1)

ggplot(rikz, aes(x = NAP, y = srich)) +
  geom_point() +
  geom_line(aes(y = fit1))
```



Linear regression assumes that residuals are independent from one observation to another. This is not the case here, since the 5 sites of the same beach are more similar compared to the sites of different beaches.

```
ggplot(rikz, aes(x = Beach, y = residuals(mod1))) +  
  geom_point()
```



Model 2: Estimation of coefficients for each group

By adding the Beach factor, this model can estimate systematic differences in species richness between beaches.

```
mod2 <- lm(srich ~ NAP + Beach, rikz)
summary(mod2)
```

```
##
## Call:
## lm(formula = srich ~ NAP + Beach, data = rikz)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -1.58544 -0.28653 -0.06544  0.23657  1.69043
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)   2.99457    0.26711  11.211 3.92e-13 ***
## NAP          -0.66410    0.09655  -6.878 5.49e-08 ***
## Beach2         0.61544    0.37909   1.623  0.11346
## Beach3        -1.21158    0.37491  -3.232  0.00268 **
## Beach4        -1.13596    0.38510  -2.950  0.00564 **
## Beach5        -0.32863    0.38648  -0.850  0.40093
## Beach6        -0.90219    0.37835  -2.385  0.02265 *
## Beach7        -0.98741    0.39419  -2.505  0.01705 *
## Beach8        -0.89080    0.38392  -2.320  0.02628 *
```

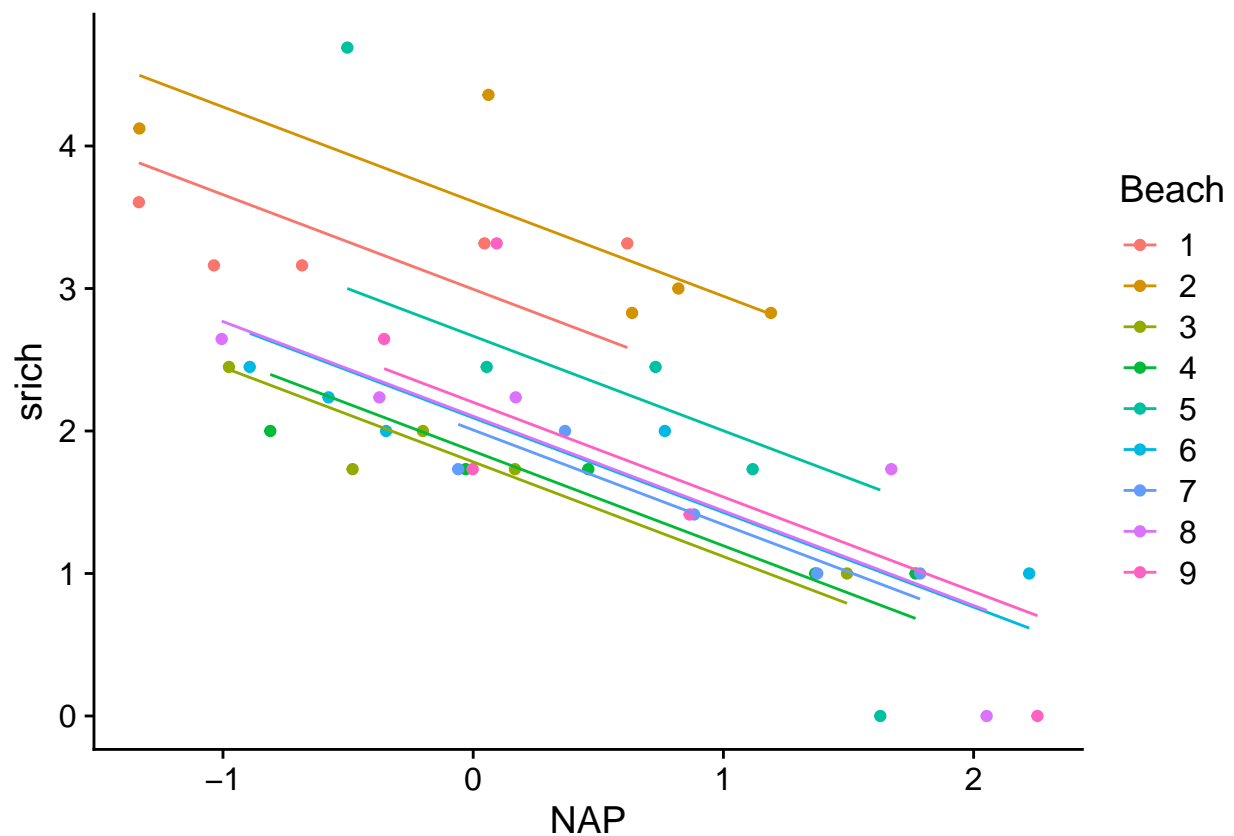
```
## Beach9      -0.79350    0.38561  -2.058  0.04712 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.5882 on 35 degrees of freedom
## Multiple R-squared:  0.7596, Adjusted R-squared:  0.6978
## F-statistic: 12.29 on 9 and 35 DF,  p-value: 1.744e-08
```

This model amounts to estimating a different intercept for each group: the **Intercept** is the mean of **srich** for Beach 1 when NAP = 0 (at mean sea level), the value of **Beach2** is the difference in means between beaches 2 and 1, etc.

The adjusted R^2 increased to 0.70 for this model. Here are its prediction lines superimposed to the data.

```
rikz$fit2 <- fitted(mod2)
```

```
ggplot(rikz, aes(x = NAP, y = srich, color = Beach)) +
  geom_point() +
  geom_line(aes(y = fit2))
```



A more flexible model would allow the effect of the NAP (the slope of the line) to vary from one beach to another, by adding the interaction between NAP and Beach.

```
mod2_inter <- lm(srich ~ NAP * Beach, rikz)
summary(mod2_inter)
```

```
##
## Call:
## lm(formula = srich ~ NAP * Beach, data = rikz)
```

```
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -0.84831 -0.16080 -0.03091  0.14909  0.98737
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)   3.28835    0.24259  13.555 1.45e-13 ***
## NAP          -0.05077    0.28172  -0.180 0.858319
## Beach2         0.30239    0.32158   0.940 0.355394
## Beach3        -1.50542    0.31542  -4.773 5.61e-05 ***
## Beach4        -1.56073    0.33715  -4.629 8.25e-05 ***
## Beach5         0.11078    0.35432   0.313 0.756947
## Beach6        -1.25466    0.31812  -3.944 0.000513 ***
## Beach7        -1.39537    0.41116  -3.394 0.002144 **
## Beach8        -1.17907    0.32697  -3.606 0.001242 **
## Beach9        -0.85912    0.33879  -2.536 0.017314 *
## NAP:Beach2    -0.54313    0.36261  -1.498 0.145780
## NAP:Beach3    -0.47943    0.37104  -1.292 0.207267
## NAP:Beach4    -0.37552    0.35511  -1.057 0.299666
## NAP:Beach5    -1.82561    0.38805  -4.705 6.74e-05 ***
## NAP:Beach6    -0.36229    0.33258  -1.089 0.285636
## NAP:Beach7    -0.48212    0.41379  -1.165 0.254155
## NAP:Beach8    -0.62429    0.32975  -1.893 0.069089 .
## NAP:Beach9    -1.01278    0.35527  -2.851 0.008256 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.4508 on 27 degrees of freedom
## Multiple R-squared:  0.8911, Adjusted R-squared:  0.8225
## F-statistic:    13 on 17 and 27 DF,  p-value: 7.079e-09
```

The adjusted R^2 increased to 0.82, but this model has 17 adjustable parameters for only 45 observations. According to the AICc, the model without interaction is the best compromise between complexity and good fit.

```
library(AICcmodavg)
aictab(list(mod1 = mod1, mod2 = mod2, mod2_inter = mod2_inter))
```

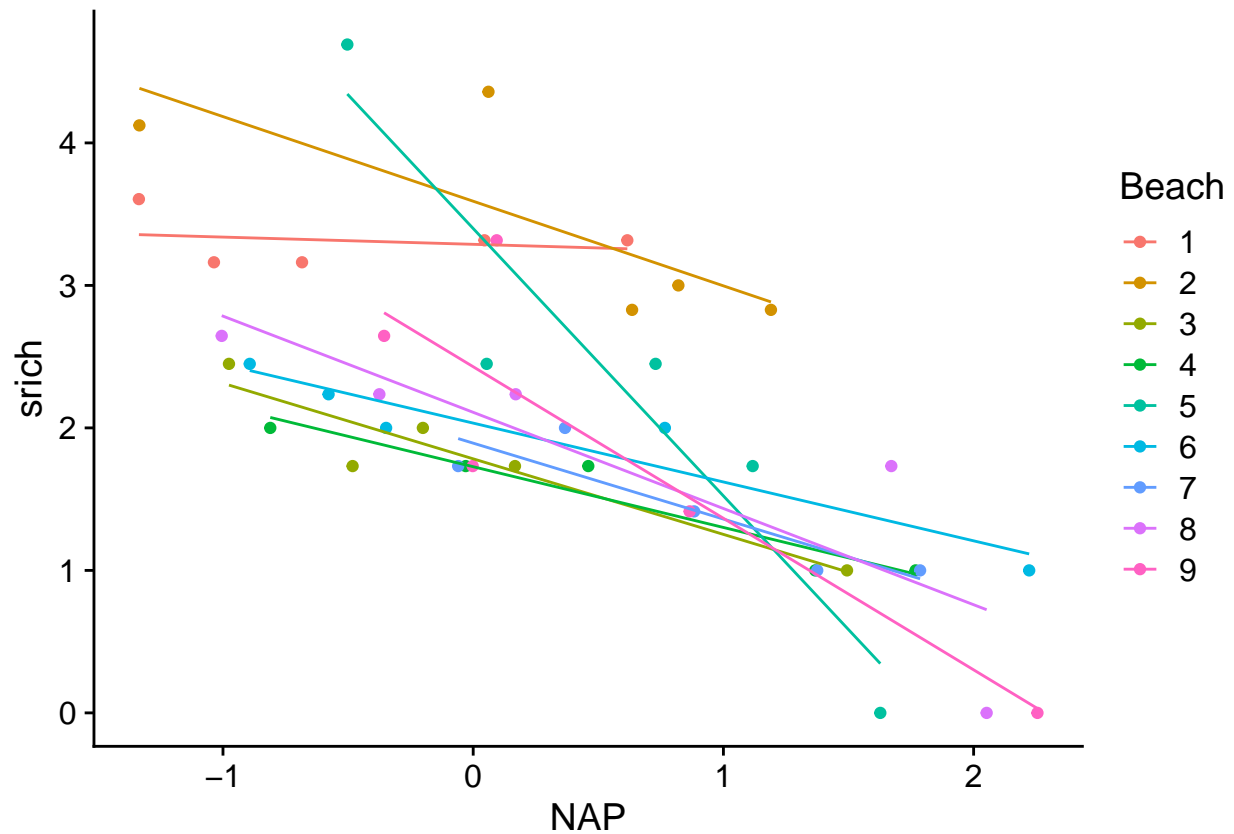
```
##
## Model selection based on AICc:
##
##           K   AICc Delta_AICc AICcWt Cum.Wt    LL
## mod2       11  98.64      0.00    0.8   0.8 -34.32
## mod2_inter 19 101.41      2.77    0.2   1.0 -16.50
## mod1        3 110.47     11.82    0.0   1.0 -51.94
```

Here are the prediction lines of the model with interaction. This model is almost equivalent to performing a separate linear regression for each group, except that the residual variance must be the same for all groups. Each line is estimated from only 5 observations, so it is likely that the model exaggerates the differences between beaches (overfitting).

```
rikz$fit2i <- fitted(mod2_inter)

ggplot(rikz, aes(x = NAP, y = srich, color = Beach)) +
  geom_point() +
```

```
geom_line(aes(y = fit2i))
```



Model 3: Group-level predictor

Rather than estimating mean species richness and/or the relationship between richness and NAP separately for each beach (as in the previous model), we could instead try to explain these differences based on predictors measured at the beach level.

The following model assumes that the (transformed) richness is a function of the interaction between NAP (defined by site) and the exposure index defined by beach.

```
mod3 <- lm(srich ~ NAP * Exposure, rikz)
summary(mod3)
```

```
##
## Call:
## lm(formula = srich ~ NAP * Exposure, data = rikz)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -1.01561 -0.26022 -0.07632  0.18031  1.62592
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)    3.5907     0.2641  13.597 < 2e-16 ***
## NAP           -0.5939     0.2856  -2.079  0.04420 *
## Exposure10     -1.0101     0.2950  -3.424  0.00146 **
```



```
## Exposure11      -1.7375      0.2976   -5.839 8.68e-07 ***
## NAP:Exposure10 -0.3680      0.3113   -1.182 0.24429
## NAP:Exposure11  0.1281      0.3140    0.408 0.68549
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.5639 on 39 degrees of freedom
## Multiple R-squared:  0.7538, Adjusted R-squared:  0.7223
## F-statistic: 23.88 on 5 and 39 DF,  p-value: 6.417e-11
```

Note that it is not enough to check the coefficients to see if the interaction is significant, rather we must look at the ANOVA table.

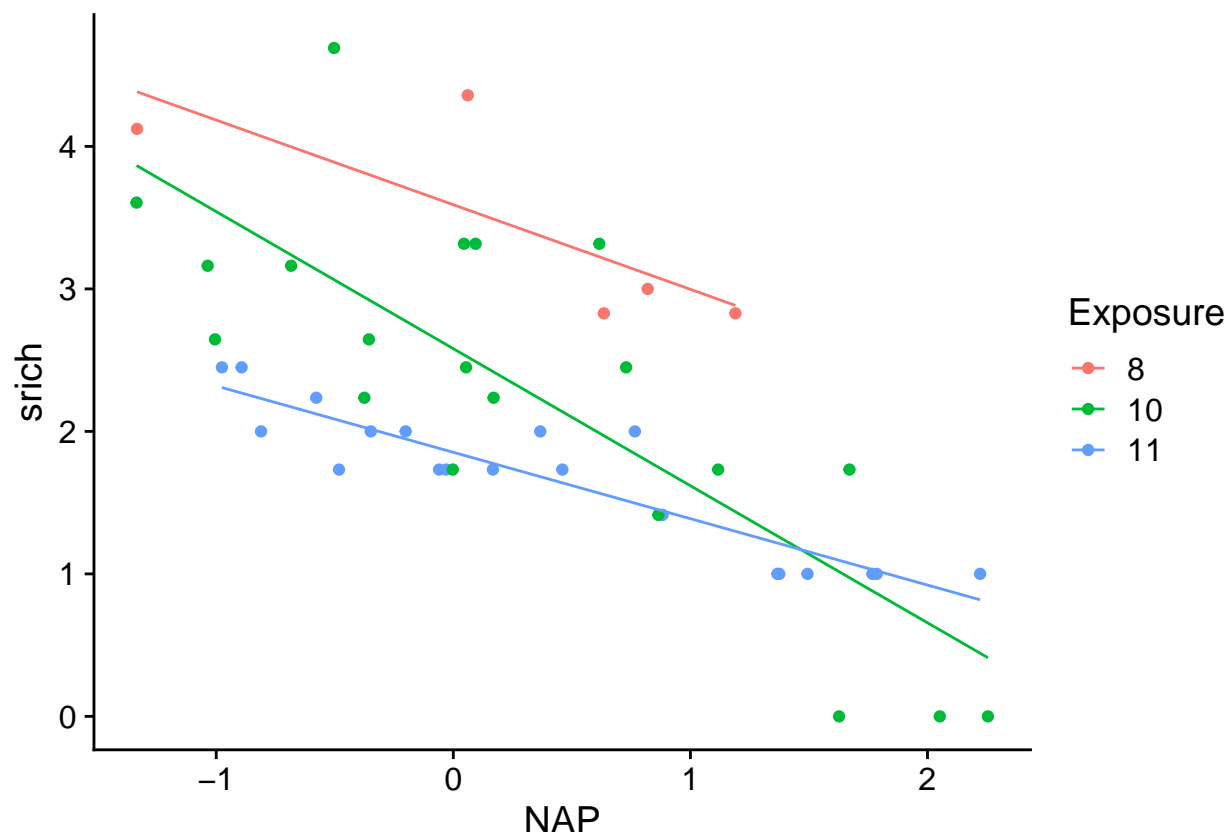
```
anova(mod3)
```

```
## Analysis of Variance Table
##
## Response: srich
##           Df Sum Sq Mean Sq F value    Pr(>F)
## NAP         1 23.8787  23.8787 75.0869 1.255e-10 ***
## Exposure    2 11.6166   5.8083 18.2643 2.527e-06 ***
## NAP:Exposure 2  2.4831   1.2416  3.9041 0.02848 *
## Residuals   39 12.4026   0.3180
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Here, the interaction is significant even if the model's coefficients for `NAP: Exposure10` and `NAP: Exposure11` are not. These coefficients only indicate the difference between the slopes of *srich* vs. *NAP* for exposure levels 10 vs. 8 (in the first case) or 11 vs. 8 (in the second case). It is possible that the most significant difference is between 10 and 11, as suggested by the prediction lines.

```
rikz$fit3 <- fitted(mod3)

ggplot(rikz, aes(x = NAP, y = srich, color = Exposure)) +
  geom_point() +
  geom_line(aes(y = fit3))
```



Compared to the previous model, this approach is not only more parsimonious (fewer parameters to fit), but it also allows us to explain the differences between beaches according to environmental parameters. We could therefore apply the predictions of the model to other beaches as long as we know their exposure index.

In this case, it appears that the exposure index accounts for much of the variation between groups. However, how could we include differences that remain between groups after controlling for the effect of this index?

In a classical linear regression context, it is not possible to include both a categorical variable indicating the group and a group-level predictor. In the following example, since the mean differences in richness between beaches (after controlling for the effect of the NAP) are fully taken into account by the **Beach** factor, there is no variation left for the **Exposure** variable to explain.

```
mod_exp_beach <- lm(srich ~ NAP + Beach + Exposure, rikz)
summary(mod_exp_beach)
```

```
##
## Call:
## lm(formula = srich ~ NAP + Beach + Exposure, data = rikz)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -1.58544 -0.28653 -0.06544  0.23657  1.69043
##
## Coefficients: (2 not defined because of singularities)
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)  2.99457    0.26711  11.211 3.92e-13 ***
## NAP         -0.66410    0.09655  -6.878 5.49e-08 ***
## Beach2       0.61544    0.37909   1.623  0.11346
```

```
## Beach3      -1.21158    0.37491   -3.232  0.00268 **
## Beach4      -1.13596    0.38510   -2.950  0.00564 **
## Beach5      -0.32863    0.38648   -0.850  0.40093
## Beach6      -0.90219    0.37835   -2.385  0.02265 *
## Beach7      -0.98741    0.39419   -2.505  0.01705 *
## Beach8      -0.89080    0.38392   -2.320  0.02628 *
## Beach9      -0.79350    0.38561   -2.058  0.04712 *
## Exposure10      NA          NA      NA      NA
## Exposure11      NA          NA      NA      NA
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.5882 on 35 degrees of freedom
## Multiple R-squared:  0.7596, Adjusted R-squared:  0.6978
## F-statistic: 12.29 on 9 and 35 DF,  p-value: 1.744e-08
```

Thus, we obtain NA values for the coefficients related to the exposure index. Note that the output of this model is identical to that of `mod2`. If we had written the formula with `Exposure` before `Beach`, the model would estimate the effects of the two coefficients of the exposure index, but in return, two of the beach effects would become NA. In terms of predictions, it would be the same model.

An identical problem arises if one tries to model a variation of the slope of `srich ~ NAP` according to both the exposure index and the beach. That model would be equivalent to `mod2_inter` above.

```
mod_exp_beach <- lm(srich ~ NAP * Exposure + NAP * Beach , rikz)
all.equal(fitted(mod_exp_beach), fitted(mod2_inter))
```

```
## [1] TRUE
```

Model 4: Two-step linear model

To solve the problem mentioned at the end of the previous section, we could first estimate the richness differences between beaches from the site-level data, and then model the beach effects according to the exposure index.

For the first step, we use model 2 above (intercept varies by beach), except that we add `- 1` to the model formula to force the intercept to be zero. This trick allows us to obtain coefficients for each beach that correspond to the intercept for that beach, rather than a difference measured from a reference level. Note that this trick only works for the first categorical variable in a regression.

```
mod4_1 <- lm(srich ~ NAP + Beach - 1, rikz)
summary(mod4_1)

##
## Call:
## lm(formula = srich ~ NAP + Beach - 1, data = rikz)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -1.58544 -0.28653 -0.06544  0.23657  1.69043
##
## Coefficients:
##      Estimate Std. Error t value Pr(>|t|)
## NAP      -0.66410    0.09655  -6.878 5.49e-08 ***
## Beach1    2.99457    0.26711  11.211 3.92e-13 ***
## Beach2    3.61000    0.26440  13.653 1.39e-15 ***
## Beach3    1.78298    0.26307   6.778 7.41e-08 ***
```

```
## Beach4  1.85861    0.26839    6.925 4.78e-08 ***
## Beach5  2.66594    0.26948    9.893 1.12e-11 ***
## Beach6  2.09238    0.26404    7.924 2.55e-09 ***
## Beach7  2.00715    0.27616    7.268 1.73e-08 ***
## Beach8  2.10377    0.26751    7.864 3.04e-09 ***
## Beach9  2.20106    0.26879    8.189 1.20e-09 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.5882 on 35 degrees of freedom
## Multiple R-squared:  0.9527, Adjusted R-squared:  0.9392
## F-statistic: 70.48 on 10 and 35 DF,  p-value: < 2.2e-16
```

We then create a second dataset containing only the beaches and their exposure index. We use the `distinct` function of *dplyr*, which extracts the unique combinations the specified variables from a data frame.

```
rikz_beach <- distinct(rikz, Beach, Exposure)
rikz_beach
```

```
##   Beach Exposure
## 1     1         10
## 2     2          8
## 3     3         11
## 4     4         11
## 5     5         10
## 6     6         11
## 7     7         11
## 8     8         10
## 9     9         10
```

We then add to this data frame the beach intercepts obtained at step 1, which is the response we want to model at step 2.

```
rikz_beach$coef <- coef(mod4_1)[-1] # Coefficient 1 is NAP
```

```
mod4_2 <- lm(coef ~ Exposure, rikz_beach)
summary(mod4_2)
```

```
##
## Call:
## lm(formula = coef ~ Exposure, data = rikz_beach)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -0.3876 -0.1523  0.0000  0.1571  0.5032
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)   3.6100     0.3101  11.640 2.42e-05 ***
## Exposure10    -1.1187     0.3467  -3.226  0.01800 *
## Exposure11    -1.6747     0.3467  -4.830  0.00291 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.3101 on 6 degrees of freedom
## Multiple R-squared:  0.803, Adjusted R-squared:  0.7373
```

F-statistic: 12.23 on 2 and 6 DF, p-value: 0.007645

One problem with this approach is that each beach is considered a single point at the second step. If the number of samples differed from one beach to another, this model would under-represent the better-sampled beaches. For this case, each beach has the same number of points and the estimated coefficients for the exposure index are very close to those obtained by model 3.

Mixed linear models

Mixed models are conceptually similar to the two-step regression performed above, except that both steps are performed simultaneously to account for the uncertainty of the group effects. This type of model is particularly useful when one or more of the following conditions apply:

- the data are grouped or have a hierarchical structure at two or more levels (e.g.: plots grouped by sites grouped by region);
- the explanatory variables are also defined at several levels;
- the number of groups is too large, or the number of observations in some groups is too small, to estimate a separate effect for each group;
- there is more interest in variation between groups than in the effect of particular groups;
- we want to apply the model to new groups not represented in the data.

Mathematical representation of a linear mixed model

A **linear mixed model** is a linear regression where one or more of the coefficients vary from one group of observations to another, *and where this variation is modeled by a statistical distribution*. They are also known as *hierarchical* or *multilevel* models because they model the variation at least at two levels (individual and group of observations).

We will start by dealing with mixed models where only the intercept varies by group. For that type of model, the mean value of the response for observation k (\hat{y}_k) is the linear combination of the predictors x_1 , x_2 , and so on, to which we add an intercept specific to the group j containing observation k : $\alpha_{j[k]}$. (We use α rather than β_0 for the intercept to simplify the notation a bit.)

$$\hat{y}_k = \alpha_{j[k]} + \beta_1 x_{1k} + \beta_2 x_{2k} + \dots$$

Just like in the linear regression model, the observed y_k follows a normal distribution around its mean. The suffix y has been added to σ to specify that it is the standard deviation of the y .

$$y_k \sim N(\hat{y}_k, \sigma_y)$$

So far, this model is identical to a linear regression with a categorical variable, if α_j was the intercept for category j . The particular feature of mixed models is that the α_j are random values from a normal distribution with a mean μ_α and a standard deviation σ_α . In other words, the observed groups are part of a “population” of possible groups and we wish to estimate the mean and the variance of α in that population.

$$\alpha_j \sim N(\mu_\alpha, \sigma_\alpha)$$

The group effect α_j is called a *random* effect, as opposed to *fixed* effects that are independently estimated without originating from a common distribution (for example, the $m - 1$ coefficients of a factor of m categories in a classical linear regression). This model is called *mixed* because it contains both fixed effects (the β associated with predictors x_1 , x_2 , etc.) and random effects.

There is no particular reason for choosing a normal distribution for α_j . However, if all that is known of a variable is its mean and its standard deviation, the normal distribution is in some sense the most likely, that is, the one that brings the least additional assumptions.

Linear mixed model with R

In this example, we will fit a mixed model of the type shown above to the same data in `rikz`. The model includes the NAP predictor and an intercept that varies by beach.

To estimate the parameters of a mixed model, we use the `lmer` function of the *lme4* package. The formula for a mixed model resembles those seen before, except for the last term: `(1 | Beach)`. This term means that the intercept (referred to as 1) varies between the levels of the `Beach` factor.

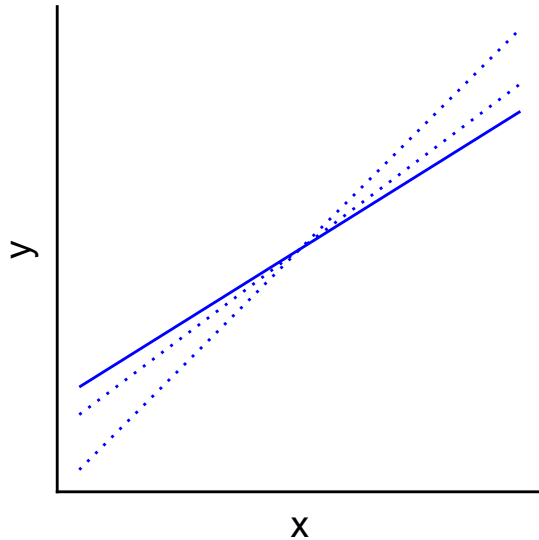
```
library(lme4)

mmod <- lmer(srlich ~ NAP + (1 | Beach), rikz)
summary(mmod)

## Linear mixed model fit by REML ['lmerMod']
## Formula: srlich ~ NAP + (1 | Beach)
## Data: rikz
##
## REML criterion at convergence: 97.1
##
## Scaled residuals:
##      Min       1Q   Median       3Q      Max
## -2.5693 -0.4286 -0.1869  0.3230  2.9399
##
## Random effects:
## Groups   Name                Variance Std.Dev.
## Beach    (Intercept)  0.2957     0.5438
## Residual                    0.3460     0.5882
## Number of obs: 45, groups: Beach, 9
##
## Fixed effects:
##              Estimate Std. Error t value
## (Intercept)  2.37424    0.20405  11.635
## NAP         -0.68063    0.09501  -7.163
##
## Correlation of Fixed Effects:
##      (Intr)
## NAP -0.162
```

Let's look at the different components of this result. The first line indicates that the model has been fit with the restricted maximum likelihood (REML) method. Without going into too much detail, the maximum likelihood produces a bias when estimating variances (random effects), and the REML serves to correct that bias.

The random effects block contains the estimates for the variance and standard deviation of the intercept (σ_α), as well as the residual variance and standard deviation (σ_y). The fixed effects block shows the estimate for the mean intercept (μ_α) and the effect of the NAP. The summary also indicates the estimated correlation between the two fixed effects. A negative correlation is to be expected when the mean value of the predictor is greater than zero: if the line is rotated around the mean so as to increase the slope, the position of the intercept moves down the y axis (see diagram below).



Note that `lmer` does not provide a p -value for the estimated effects, because unlike the case of a linear model, the exact distribution of these estimates is not known. We can nevertheless obtain approximate confidence intervals with `confint`. The argument `oldNames = FALSE` ensures that the random effects are clearly identified in the table.

```
confint(mmod, oldNames = FALSE)
```

```
## Computing profile confidence intervals ...
##                2.5 %    97.5 %
## sd_(Intercept)|Beach 0.2664346 0.9469885
## sigma              0.4682062 0.7455375
## (Intercept)        1.9537581 2.7916024
## NAP                -0.8729868 -0.4948024
```

The effect of the NAP is therefore significant with a threshold of $\alpha = 0.05$. The first two rows in the table correspond to σ_α and σ_y , respectively. Standard deviations must be positive. A value less than or equal to zero for a standard deviation or its confidence interval indicates that the model is incorrectly specified.

We can separately extract the fixed and random effects of the model with the `fixef` and `ranef` functions.

```
fixef(mmod)
```

```
## (Intercept)      NAP
##    2.3742423  -0.6806307
```

```
ranef(mmod)
```

```
## $Beach
## (Intercept)
## 1    0.4962684
## 2    1.0050819
## 3   -0.4791252
## 4   -0.4104708
## 5    0.2444787
## 6   -0.2252788
## 7   -0.2858167
## 8   -0.2124511
## 9   -0.1326862
```

```
##
## with conditional variances for "Beach"
```

The random effects are the estimates of the difference between the intercept of each beach (1 to 9) and their mean given by (Intercept). The estimated intercept for beach 1 is $2.37 + 0.50 = 2.87$ and so on. Instead of calculating these values for each beach, we can get them with the `coef` function.

```
coef(mmod)

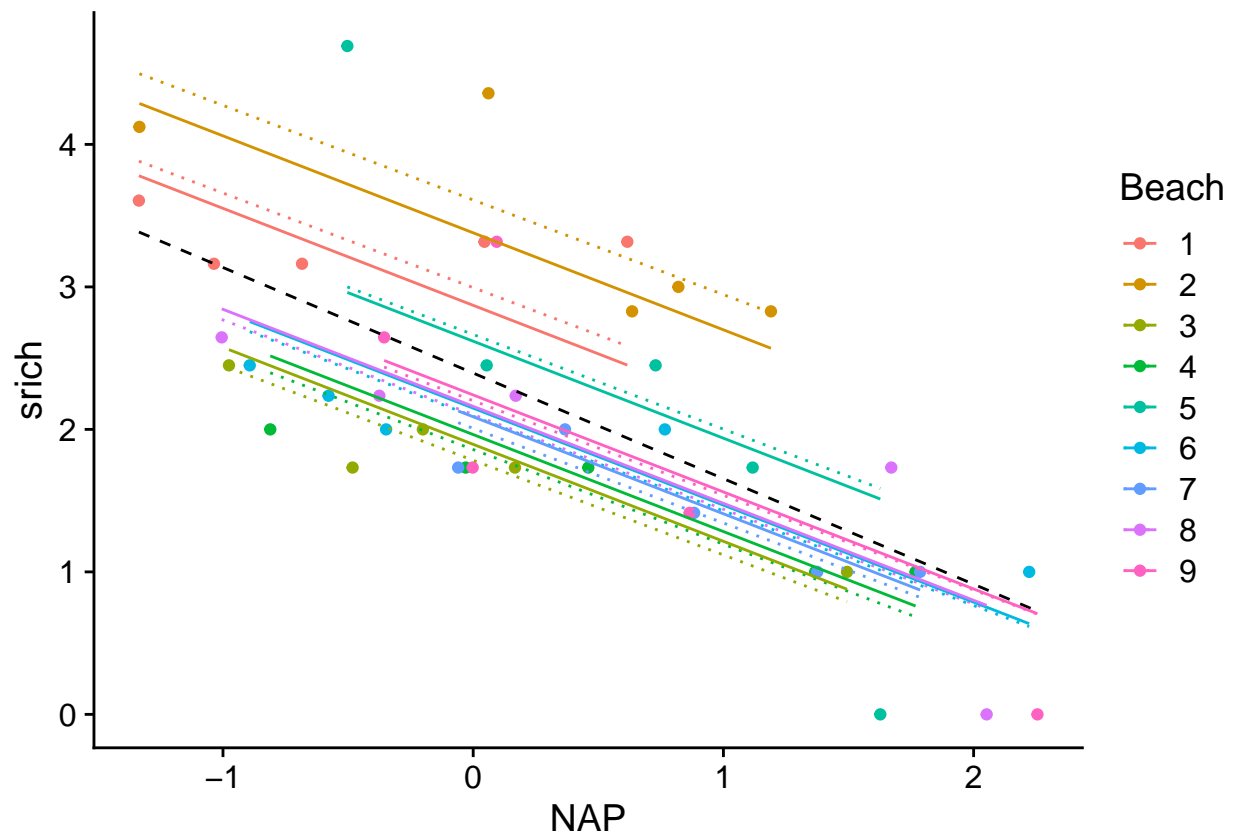
## $Beach
##      (Intercept)      NAP
## 1      2.870511 -0.6806307
## 2      3.379324 -0.6806307
## 3      1.895117 -0.6806307
## 4      1.963771 -0.6806307
## 5      2.618721 -0.6806307
## 6      2.148964 -0.6806307
## 7      2.088426 -0.6806307
## 8      2.161791 -0.6806307
## 9      2.241556 -0.6806307
##
## attr(,"class")
## [1] "coef.mer"
```

The result of this function gives us the coefficients of the linear regression by group. Our model does not include random effects on the coefficient of NAP, so it is constant for all groups.

What is the difference between these estimated effects for each beach and those of a classical linear model where the `Beach` categorical variable was specified as a fixed effect? Let's compare the prediction lines of the mixed model (solid lines) with those of model 2 seen above (`lm(srich ~ NAP + Beach)`, dotted lines). We also add the line corresponding to model 1 which ignores the differences between beaches (dashed black line).

```
rikz$fitmm <- fitted(mmod)

ggplot(rikz, aes(x = NAP, y = srich, color = Beach)) +
  geom_point() +
  geom_line(aes(y = fit1, color = "black", linetype = "dashed") +
  geom_line(aes(y = fit2, linetype = "dotted") +
  geom_line(aes(y = fitmm))
```

Predictions of the mixed model for each beach are shifted towards the overall mean. This effect is more pronounced as one moves away from the mean (see for example beach 2 at the top of the graph). In statistics, this is called a *shrinkage* of the group effects.

Other useful packages for mixed models

The *lmerTest* package adds approximate p -values to a *lmer* model, whereas the *MuMIn* package contains a `r.squaredGLMM` function that calculates an equivalent of the R^2 for mixed models.

```
library(lmerTest)
mmod <- lmer(srich ~ NAP + (1 | Beach), rikz)
summary(mmod)
```

```
## Linear mixed model fit by REML. t-tests use Satterthwaite's method [
## lmerModLmerTest]
## Formula: srich ~ NAP + (1 | Beach)
## Data: rikz
##
## REML criterion at convergence: 97.1
##
## Scaled residuals:
##    Min     1Q  Median     3Q      Max
## -2.5693 -0.4286 -0.1869  0.3230  2.9399
##
## Random effects:
## Groups   Name                Variance Std.Dev.
## Beach    (Intercept)  0.2957     0.5438
```

```
## Residual          0.3460    0.5882
## Number of obs: 45, groups: Beach, 9
##
## Fixed effects:
##           Estimate Std. Error      df t value Pr(>|t|)
## (Intercept)  2.37424    0.20405   8.36034  11.635 1.88e-06 ***
## NAP         -0.68063    0.09501  37.15971  -7.163 1.68e-08 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Correlation of Fixed Effects:
##      (Intr)
## NAP -0.162

library(MuMIn)
r.squaredGLMM(mmod)

##           R2m          R2c
## [1,] 0.4164714 0.6853641
```

In the output of `r.squaredGLMM`, the `R2m` value (marginal R^2) estimates the fraction of the variance explained by the fixed effects in the model, whereas the `R2c` value (conditional R^2) estimates the fraction explained by the fixed and random effects. Therefore, the conditional R^2 is always greater than the marginal R^2 .

Mixed models as a compromise between underfitting and overfitting

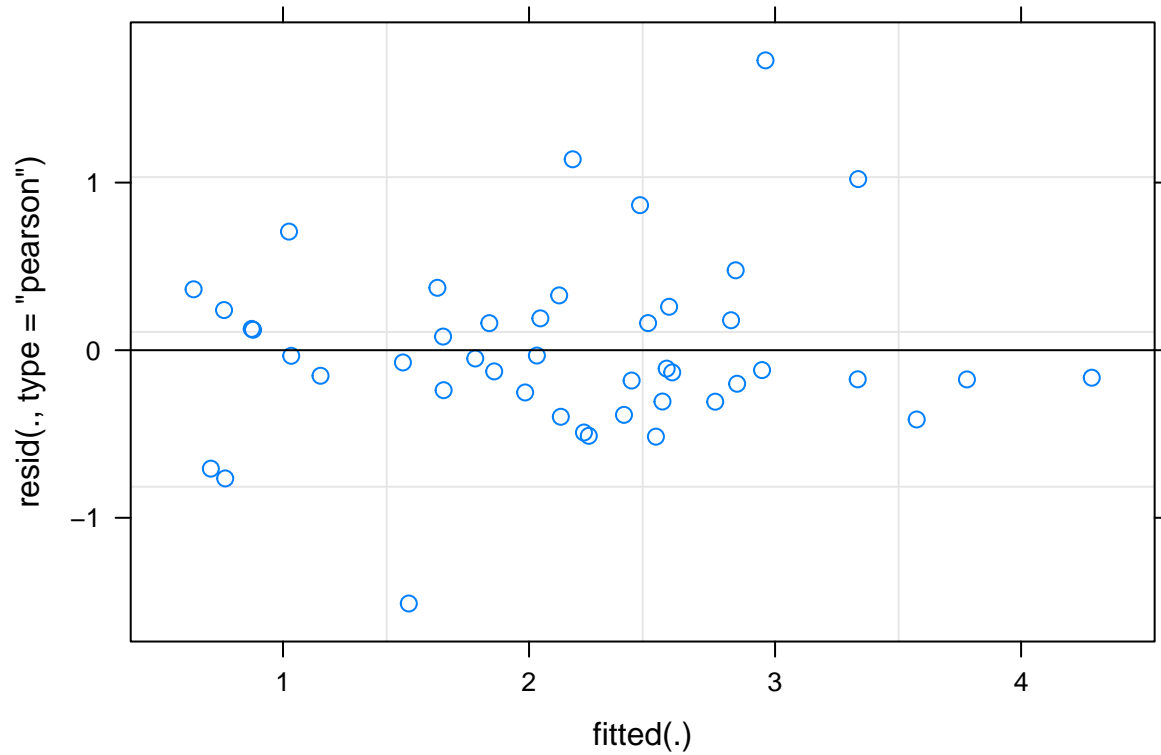
What is happening here? Assuming that the effects of each beach come from a common distribution, the mixed model estimates the intercept of each beach by taking into account not only the values measured at that beach, but also those of the other beaches. It is therefore a compromise between underfitting (e.g. model 1, which totally ignores differences between beaches) and overfitting (e.g. model 2, which independently estimates the mean intercept for each beach based on its 5 sites).

In other words, the differences between the mean values measured by group can be due either to real differences in the response between the groups, or to random sampling. The ANOVA we saw earlier in this semester would ask whether these differences between groups of observations are too large to be due to chance. The mixed model attempts instead to estimate the portion of observed differences that is due to a real group effect and the portion that is due to random sampling in each group.

Diagnostic plots

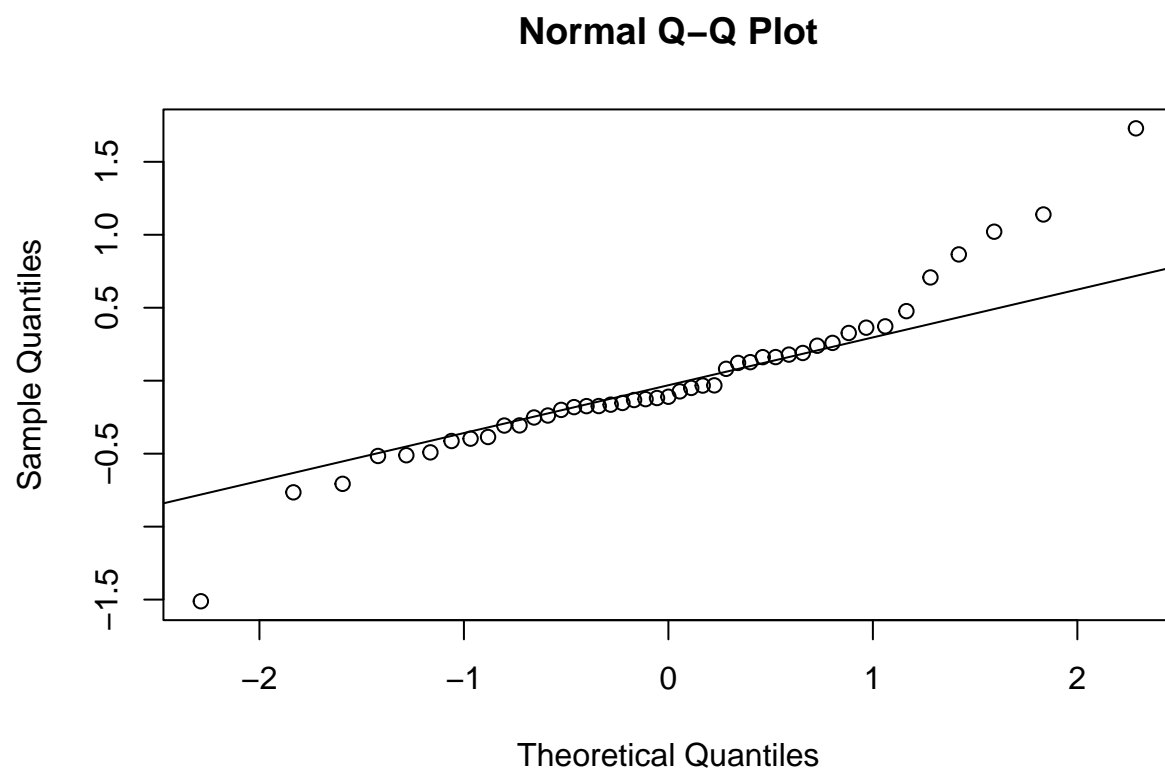
The `plot` function applied to a mixed model output only produces one graph, that of residuals vs. fitted values.

```
plot(mmod)
```



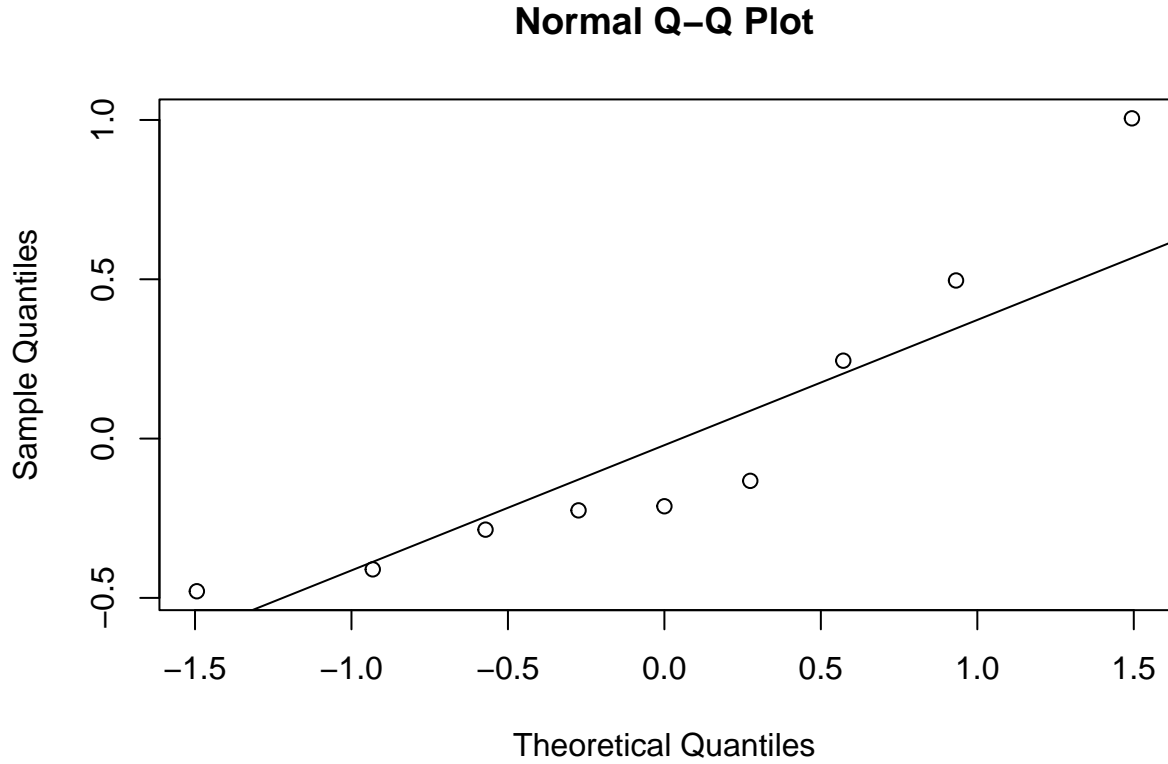
To reproduce the quantile-quantile plot of residuals, we can call the `qqnorm` and `qqline` functions. This graph shows that the most extreme residuals are further from the mean than those predicted by the normal distribution.

```
qqnorm(residuals(mmod))  
qqline(residuals(mmod))
```



In a mixed model, group effects are also assumed to follow a normal distribution. We test this assumption with a quantile-quantile plot of random effects (`ranef`).

```
beach_coef <- ranef(mmod)$Beach  
qqnorm(beach_coef$`(Intercept)`)  
qqline(beach_coef$`(Intercept)`)
```



Even with just 9 points, it is obvious that differences between beaches are not normally distributed.

Note that the influence measures of observations such as Cook distance do not apply to mixed models.

Mixed model with group-related predictor

At the end of the previous section, we saw that the differences between each beach and the overall mean do not follow a normal distribution. Could some of that variation be explained by the exposure index measured at the beach level?

To include the effect of a group-level predictor, we need to modify the model from the previous section. The group-level intercept was defined by a normal distribution:

$$\alpha_j \sim N(\mu_\alpha, \sigma_\alpha)$$

In this case, μ_α is not a constant, but varies linearly with the predictor u_1 .

$$\mu_\alpha = \gamma_0 + \gamma_1 u_1$$

It is therefore a two-level regression model, where u_1 is a group-level predictor and γ_1 is the coefficient of that predictor. From a conceptual point of view, it is useful to differentiate group-level and individual-level predictors. However, we could also rewrite the model to separate the fixed effects (at both levels) and the random effects (which have a mean of 0), for example:

$$y_k = \gamma_0 + \gamma_1 u_{1j[k]} + \beta_1 x_{1k} + \beta_2 x_{2k} + \nu_{j[k]} + \epsilon_k$$

In this representation, the value y_k of observation k depends on the intercept γ_0 , a group-level predictor u_1 , two individual-level predictors (x_1, x_2). The last two terms respectively represent the random variation for group j and observation k :

$$\begin{aligned}\nu_j &\sim N(0, \sigma_\alpha) \\ \epsilon_k &\sim N(0, \sigma_y)\end{aligned}$$

We can think of ν_j as the part of the residual that is shared by all the observations of group j , while ϵ is an independent residual from one observation to another. To fit this model in R, we simply add the predictor to the formula in `lmer`. The estimate works without the need to explicitly tell R that the value of this variable is the same for all observations in a group.

```
mmod_exp <- lmer(srich ~ NAP + Exposure + (1 | Beach), rikz)
summary(mmod_exp)
```

```
## Linear mixed model fit by REML. t-tests use Satterthwaite's method [
## lmerModLmerTest]
## Formula: srich ~ NAP + Exposure + (1 | Beach)
## Data: rikz
##
## REML criterion at convergence: 86
##
## Scaled residuals:
##      Min       1Q   Median       3Q      Max
## -2.38707 -0.48281 -0.07465  0.47962  3.03495
##
## Random effects:
## Groups Name Variance Std.Dev.
## Beach (Intercept) 0.02315  0.1521
## Residual          0.34658  0.5887
## Number of obs: 45, groups: Beach, 9
##
## Fixed effects:
##              Estimate Std. Error      df t value Pr(>|t|)
## (Intercept)  3.62081    0.30510   5.76006  11.867 2.89e-05 ***
## NAP          -0.70349    0.09107  40.64879  -7.724 1.69e-09 ***
## Exposure10   -1.11766    0.33998   5.69172  -3.287  0.0180 *
## Exposure11   -1.66923    0.34021   5.70544  -4.907  0.0031 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Correlation of Fixed Effects:
##              (Intr) NAP      Exps10
## NAP          -0.082
## Exposure10   -0.891 -0.007
## Exposure11   -0.888 -0.037  0.800
```

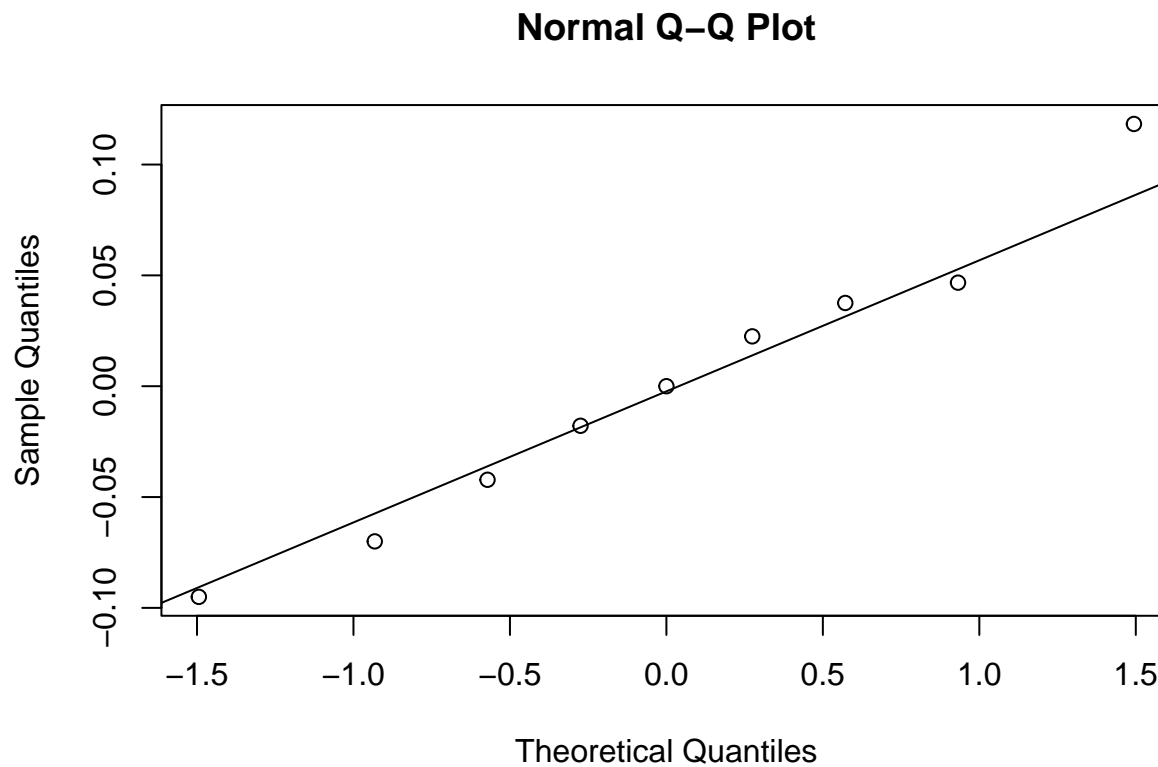
Some notes on this result:

- The estimate of σ_α (0.15) has decreased compared to its value in the previous model (0.54), showing that the predictor explains a good amount of the variation observed between the groups. The standard deviation of individual observations (0.59) has not changed, which is expected because a group-level predictor cannot explain the differences between observations of the same group.
- Correlations between the fixed effects (Intercept), Exposure10, and Exposure11 are to be expected. With the default contrasts in R, (Intercept) is the intercept for the reference level of the variable

(Exposure = 8), **Exposure10** is the difference between levels 10 and 8, and **Exposure11** is the difference between levels 11 and 8. So, if the estimate of the mean for level 8 increases, the two differences (10 - 8 and 11 - 8) decrease.

Note that after taking into account the effect of the exposure index, the random group effects are closer to a normal distribution.

```
beach_coef <- ranef(mmod_exp)$`Beach`
qqnorm(beach_coef$`(Intercept)`)
qqline(beach_coef$`(Intercept)`)
```



Intra-class correlation

In the mixed models presented above, σ_α represents the standard deviation of the mean response between groups while σ_y represents the standard deviation of the response between observations of the same group.

The *intra-class correlation* is defined as the ratio:

$$\frac{\sigma_\alpha^2}{\sigma_\alpha^2 + \sigma_y^2}$$

This ratio approaches 0 if $\sigma_y \gg \sigma_\alpha$ (the variation between groups is negligible compared to the variation between individuals of the same group) and approaches 1 if $\sigma_\alpha \gg \sigma_y$ (almost all the variation is due to differences between groups).

The intra-class correlation is about 0.45 for the first model (**mmod**) and decreases to 0.06 taking into account the exposure index (model **mmod_exp**).

From a practical point of view, the intra-class correlation indicates which scale contributes more to the variation of the response, so at which scale we could sample more to reduce the uncertainty of the model. In our example, if this correlation was close to 1, it would be better to sample multiple beaches with few samples per beach; if not, we could get a larger sample over a limited number of beaches. Of course, this assumes that the beaches observed so far are representative of the “population” of beaches that interest us.

Summary

- A linear mixed model (also called a multilevel model) is an extension of linear regression for grouped data, where some of the coefficients vary randomly from one group to another.
- In this class, we have seen examples where intercept includes a random effect. In the next class, we’ll see how to add random effects to other coefficients.
- A mixed model is a compromise between a linear regression that ignores the group structure and a regression that estimates a separate fixed effect for each group.
- To do this, the mixed model corrects the estimation of the effects of each group to bring them closer to the general trend (shrinkage).
- In classical linear regression, one cannot include both a fixed effect per group and a group-level predictor. A mixed model can include both the random effect of groups and an explanatory variable defined at the group level.

References

Here are two great textbooks to consult for more details on mixed or multi-level models.

- Gelman, A. and Hill, J. (2006) *Data Analysis Using Regression and Multilevel/Hierarchical Models*. Cambridge, Cambridge University Press.
- Zuur, A.F., Ieno, E.N., Walker, N.J., Saveliev, A.A., Smith, G.M. (2009) *Mixed Effects Models and Extensions in Ecology with R*. New York, Springer-Verlag.