#### R-Adelaide 2016

# Day 3: Mixed effects models in R

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#### Linear mixed effects models

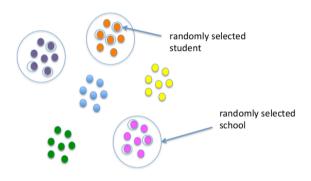
- Linear mixed-effects models are used when you have random effects
  - Occurs when your subjects or units are grouped
  - Groups are assumed to be randomly sampled from a "population" of groups
- Example situations include:
  - when you divide up plots and apply separate treatments to the parts (plot is the random group)
  - when your sampling design is nested, such as quadrats within transects; transects within woodlots; woodlots within districts (transects, woodlots, and districts are the random groups)
  - when you have measurements on related individuals (family is the random group)
  - when you measure subjects or units repeatedly (subject or unit is the random group)

# What are we actually talking about?

#### Alternative names for mixed effcts models

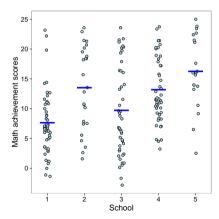
- Mixed model, LMM/GLMM
- Random effects model, mixed eects model
- Longitudinal regression, repeated measures model
- Hierarchical model, multilevel model
- Covariance pattern model

# Example of clustered data



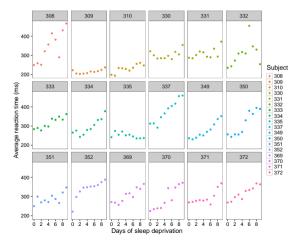
## Example of clustered data

• Record students' math achievement scores where students were randomly selected from schools in a state



## Example of longitudinal data

- 18 study participants are sleep deprived for several days
- Each day, measure that subject's reaction time



#### Definition of LME Models

In matrix notation a linear model can be represented as

$$\hat{y}_i = \beta_0 + \beta_1 x_1 + \dots + \epsilon_i$$

$$y = X\beta + \epsilon$$

#### Definition of LME Models

In matrix notation an LME model can be represented as

$$y = X\beta + Zb + \epsilon$$

#### where

- y is a vector of observations, with mean  $E(y) = X\beta$
- $\bullet$   $\beta$  is a vector of fixed effects
- $m{b}$  is a vector of random effects with mean E(b)=0 and variance-covariance matrix  ${
  m var}(b)=G$
- ullet  $\epsilon$  is a vector of IID random error terms with mean  $E(\epsilon)=0$  and variance  $\mathrm{var}(\epsilon)=R$
- ullet X and Z are matrices of regressors relating the observations y to eta and b, respectively.
- Fixed effects have coefficients
- Random effects have estimates of variation

#### Mixed models in R

- Maximum likelihood or restricted maximum likelihood (REML) estimates of the parameters in linear mixed-effects
- Historically, nlme was main package for mixed models in R
  - Includes linear and nonlinear fixed effects
  - Variety of variance functions and correlation structures
- Development then shifted to package lme4
- The term "mixed effects" denotes model with both fixed- and random-effects terms in a linear predictor

#### Mixed model formulas - 1mer

- As for linear models, first two arguments to lmer
  - Formula specifying the model
  - Data
- Formula for 1m:

```
1 response - expr
```

• Formula for lmer:

```
1 response ~ FEexpr + (REexpr1 | factor1) + (REexpr2 | factor2) + ...
```

#### where,

- ullet FEexpr is fixed-effects model matrix, X
- ullet Random-effects terms (REexpr1 | factor1) and (REexpr2 | factor2) determine random-effects model matrix, Z

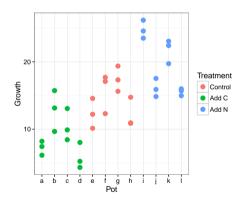
## Mixed model formulas - 1mer

Formula	Alternative	Meaning
(1   g)	$1 + (1 \mid g)$	Random intercept
		with fixed mean
$0 + offset(o) + (1 \mid g)$	$-1 + offset(o) + (1 \mid g)$	Random intercept
(1   1 / 0)	(1   1) (1   1   0)	with a priori means
$(1\mid \mathrm{g1/g2})$	$(1 \mid g1) + (1 \mid g1:g2)$	Intercept varying
		among g1 and g2
(1   -1)   (1   -2)	1 + (1   -1) + (1   -2)	within g1
$(1\mid \texttt{g1}) + (1\mid \texttt{g2})$	$1 + (1 \mid g1) + (1 \mid g2)$	Intercept varying
v + (v   g)	$1 + x + (1 + x \mid g)$	among g1 and g2 Correlated random
$x + (x \mid g)$	$1 + x + (1 + x \mid g)$	intercept and slope
x + (x    g)	1 + x + (1   g) + (0 + x   g)	Uncorrelated random
^   (^    6)	1 / / (1 / 8) / (0 / / 8)	intercept and slope

## A Greenhouse Experiment testing C:N Ratios

- Does changing the C:N Ratio of soil affects plant leaf growth?
  - 3 treatments: control, C addition, and N addition
  - 4 pots per treatment
  - 3 leaves per plant

```
1 X Treatment Pot Growth
2 1 1 Add C a 6.13
3 2 2 Add C a 7.42
4 3 3 Add C a 8.20
5 4 4 Add C b 15.73
6 5 5 Add C b 9.65
7 6 6 Add C b 13.14
```



## Simplifying analysis - averaging

• If design is balanced (and no interest in within pot variance) average within each pot

```
Anova Table (Type II tests)

Response: Growth

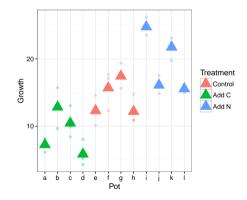
Sum Sq Df F value Pr(>F)

Treatment 217.67771 2 8.91584 0.0073312 **

Residuals 108.86628 9

---

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



### Classical ANOVA Error Decomposition with Expected Mean Squares

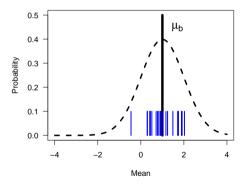
•  $SS_{Total} = SS_{Treatment} + SS_{PotError} + SS_{WithinPotError}$ 

```
plantAOV <- aov(Growth - Treatment + Error(Pot), data = plants)
summary(plantAOV)</pre>
```

```
1
2 Error: Pot
3 Df Sum Sq Mean Sq F value Pr(>F)
4 Treatment 2 653.0331 326.5166 8.91584 0.0073312 **
8 Residuals 9 329.598 36.6221
6 ---
7 Signif. codes: 0 '*** 0.001 '** 0.05 '.' 0.1 ' ' 1
8 Error: Within
Df Sum Sq Mean Sq F value Pr(>F)
11 Residuals 24 97.68967 4.070403
```

# Multilevel/Clustered/Hierarchical/Mixed Model

- $i = \text{treatment}, \ j = \text{pot}, \ k = \text{subsample}$   $\hat{y}_{ijk} = b_{j[i]} + \beta_i X + \epsilon_{ijk}$   $b_{j[i]} \sim \mathcal{N}(\mu_b, \sigma_b^2)$   $\epsilon_{ijk} \sim \mathcal{N}(0, \sigma^2)$
- What is a random effect?
  - Each pot has different parameter value
  - Unlike in linear model, intercept values are constrained around Normal distribution



#### Some Points about Multilevel Models

- Flexible. Can accomodate varying slope, intercept, intercept-slope models
- Linear mixed models are solved using Restricted Maximum Likelihood (REML)
  - ML estimation produces downward biased estimates of random effect variances
- As group level effects are drawn from the same distribution, Best Linear Unbiased Predictors (BLUPs) are shrunk towards grand mean - basically, we use information from all groups to inform within group means - useful for unbalanced designs

# Many R Packages for Multilevel Models

- nlme from Pinhero and Bates 2009
- Imer bleeding edge by Doug Bates
- MCMCglmm uses Bayesian techniques & MCMC (similar syntax to nlme)
- glmmADMB interface for AD Model Builder
- etc

```
plantLMM <- lmer(Growth - Treatment + (1 | Pot), data=plants)
summary(plantLMM)</pre>
```

```
Linear mixed model fit by REML
     t-tests use Satterthwaite approximations to degrees of freedom [']merMod
     Formula: Growth ~ Treatment + (1 | Pot)
        Data: plants
     REML criterion at convergence: 167.2
     Scaled residuals:
             Min
                                 Median
     -1.60894241 -0.73479195 -0.08365836 0.76529269 1.63672837
     Random effects:
      Groups Name
                          Variance Std Dev
      Pot.
               (Intercept) 10.850564 3.294019
      Regidual
                           4.070403 2.017524
     Number of obs: 36, groups: Pot. 12
     Fixed offects:
                     Estimate Std Error
                                               df t value
                                                             Pr(>1+1)
     (Intercept)
                    14.403830 1.746952 8.999998 8.24512 1.7379e-05 ***
     Treatment Add C -5.301297
                              2.470563 8.999998 -2.14579
                                                             0.060450
     TreatmentAdd N 5.130825
                              2.470563 8.999998 2.07678
                                                             0.067617 .
24
     Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' 1
26
     Correlation of Fixed Effects:
                 (Intr) TrtmAC
     TretmntAddC -0 707
     TretmntAddN -0.707 0.500
```

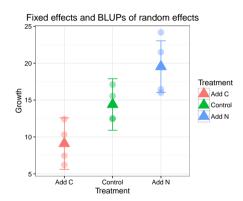
```
\hat{y}_{ijk} = b_{j[i]} + \beta_i X + \epsilon_{ijk}
```

```
(Intercept) TreatmentAdd C TreatmentAdd N BLUP.Random.Intercepts
            12 759
                           -5 301
                                           5 131
                                                                 -1 645
            17.727
                           -5.301
                                           5.131
                                                                  3.323
            15 612
                          -5 301
                                           5.131
                                                                  1 208
            11.518
                          -5.301
                                           5.131
                                                                 -2.886
            12 534
                           -5 301
                                           5 131
                                                                 -1 870
            15 552
                           -5 301
                                           5 131
                                                                  1 149
                          -5 301
            17 100
                                           5 131
                                                                  2 696
Q
            12.429
                           -5.301
                                           5.131
                                                                 -1.975
            19.046
                           -5.301
                                           5.131
                                                                  4.642
                           -5 301
            11 344
                                           5 131
                                                                 -3 060
            16.362
                           -5.301
                                           5 131
                                                                  1 958
            10.864
                           -5.301
                                           5.131
                                                                 -3.540
```

### Visualizing Fixed Effects and BLUPS

#### Note use of altered model for ease of plotting

```
plantLMM2 <- lmer(Growth ~ 0 + Treatment + (1 | Pot), data=plants)
     summary(plantLMM2)
     # Fixed effect estimates
     fixDF <- data.frame(summary(plantLMM2)$coef)</pre>
     fixDF$Treatment <- factor(gsub("Treatment", "", rownames(fixDF)), levels
             \hookrightarrow = c("Add C", "Control", "Add N"))
     # Random effect BLUPs
     ranDF <- data.frame(Estimate = unlist(ranef(plantLMM, condVar = TRUE) $Pot
             \hookrightarrow ) + rep(fixDF$Estimate[c(2, 1, 3)], rep(4, 3)))
     ranDF$Treatment <- factor(as.character(meanPlants$Treatment), levels = c(
             → "Add C", "Control", "Add N"))
     # Plot
     p <- ggplot(fixDF, aes(x=Treatment, v=Estimate, color=Treatment)) +</pre>
       theme_bw() +
       vlab("Growth") +
       geom_errorbar(aes(ymin=Estimate-Std..Error*2, ymax=Estimate+Std..Error*
               \hookrightarrow 2), width = 0.2) +
       geom_point(shape = 17, size = 6) +
14
       ggtitle("Fixed effects and BLUPs of random effects")
     p + geom point(mapping = aes(x=Treatment, v=Estimate), data = ranDF, size
             \hookrightarrow =4, alpha = 0.4)
```

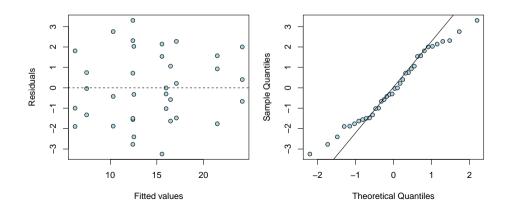


#### What next?

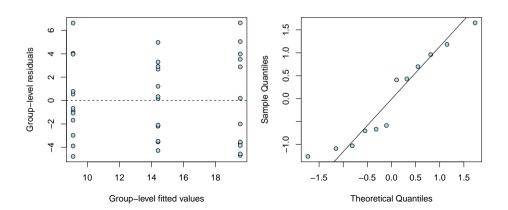
- Diagnostics in Mixed Models
- Assessing hypotheses
- Hierarchical Models
- Variable Slope-Intercept models
- A Procedure for Assessing Mixed Model Structure
- A Taste of Generalized Linear Mixed Models

# Residuals diagnostics for observational units

```
# Plot residuals vs fitted values
plot(fitted(plantLMM), residuals(plantLMM), xlab="Fitted values", ylab="Residuals", pch=21, bg="lightblue")
abline(a=0, b=0, cole-"black", lty=2)
# Q-Q plot of residuals
qqnorm(residuals(plantLMM, type="pearson"), main="", pch=21, bg="lightblue")
qqline(residuals(plantLMM, type="pearson"))
```

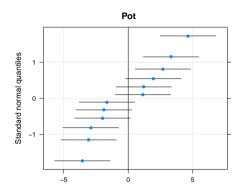


# Residuals diagnostics at group-level



#### Random effects BLUPS

```
library(lattice)
qqmath(ranef(plantLMM, condVar = TRUE), strip = FALSE)
# Test for heterogeneity
library(RLRsim)
exactRLRT(plantLMM)
```



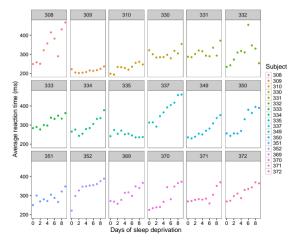
#### Model inference

10

```
Single term deletions
Model:
Growth ~ Treatment + (1 | Pot)
         Df
                  ATC
                          LRT Pr(Chi)
            185,60902
<none>
Treatment 2 194.71732 13.1083 0.0014242 **
Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' ' 1
                        2.5 %
                                   97.5 %
.sig01
                1.06733677269 4.920323813
.sigma
                1.34597084395 2.635024473
(Intercept)
               11.74738290407 17.594991016
TreatmentAdd C -10.70452432258 -1.196672772
TreatmentAdd N 0.01244979266 9.969124979
                       2.5 %
                                   97.5 %
.sig01
                 1.359105917 4.849531572
.sigma
                 1.411892016 2.549262368
TreatmentControl 10.245785828 18.000970897
TreatmentAdd C
                 6.097065684 12.975063104
TreatmentAdd N 15.193178109 24.386497203
```

## Example dataset: sleepstudy

- 18 study participants are sleep deprived for several days
- Each day, measure that subject's reaction time



### A simple linear model

9

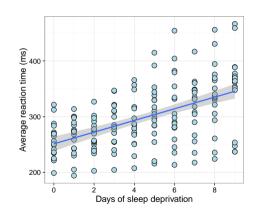
14

16

18

```
##BEGIN_SRC R :results output raw :exports both
lm.simple <- lm(Reaction - Days, data=sleepstudy)
summary(lm.simple)</pre>
```

```
Call:
lm(formula = Reaction ~ Days, data = sleepstudy)
Residuals:
   Min
            1Q Median
                                  Max
-110.85 -27.48 1.55 26.14 139.95
Coefficients:
           Estimate Std. Error t value Pr(>|t|)
(Intercept) 251.41
                         6.61
                                38.03 < 2e-16 ***
Davs
              10.47
                         1.24
                                 8.45 9.9e-15 ***
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ', 1
Residual standard error: 47.7 on 178 degrees of freedom
Multiple R-squared: 0.286, Adjusted R-squared: 0.282
F-statistic: 71.5 on 1 and 178 DF, p-value: 9.89e-15
```



#+REGIN SRC R :results output raw :exports both

```
lm.complete <- lm(Reaction ~ 0 + factor(Subject) + Days, data=sleepstudy)
```

```
16
19
24
     factor(Subject)370 244.599
     factor(Subject)371 247.881
     factor(Subject)372 270.783
```

Days

```
summary(lm.complete)
Call:
lm(formula = Reaction ~ 0 + factor(Subject) + Days, data = sleepstudy)
Residuals:
    Min
             10 Median
                                    Max
-100.54 -16.39
                 -0.34
                         15.22 131.16
Coefficients:
                   Estimate Std. Error t value Pr(>|t|)
factor(Subject)308
                   295.031
                                10.447
                                          28.2
                                                 <2e-16 ***
factor(Subject)309
                   168.130
                                10.447
                                          16.1
                                                 <2e-16 ***
factor(Subject)310 183.898
                               10.447
                                          17.6
                                                 <2e-16 ***
factor(Subject)330 256.119
                               10.447
                                          24.5
                                                 <2e-16 ***
factor(Subject)331 262.333
                               10.447
                                          25.1
                                                 <2e-16 ***
factor(Subject)332 260.199
                               10.447
                                          24.9
                                                 <2e-16 ***
factor(Subject)333 269.056
                                10.447
                                          25.8
                                                 <20-16 ***
factor(Subject)334 248.199
                                10.447
                                          23 8
                                                 <20-16 ***
factor(Subject)335 202.967
                               10.447
                                          19.4
                                                 <20-16 ***
factor(Subject)337 328.618
                                10.447
                                          31.5
                                                 <2e-16 ***
factor(Subject)349 228.732
                                10.447
                                          21.9
                                                 <2e-16 ***
factor(Subject)350 266,500
                                10.447
                                          25.5
                                                 <2e-16 ***
factor(Subject)351 242.995
                                10.447
                                          23.3
                                                 <2e-16 ***
factor(Subject)352 290.319
                                10.447
                                          27.8
                                                 <2e-16 ***
factor(Subject)369 258.932
                                10.447
                                          24.8
                                                 <20-16 ***
```

10.447

10.447

10.447

0.804

10.467

23.4

23.7

25.9

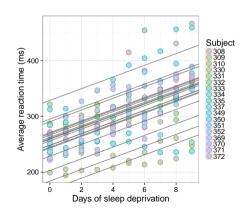
13.0

<2e-16 \*\*\*

<2e-16 \*\*\*

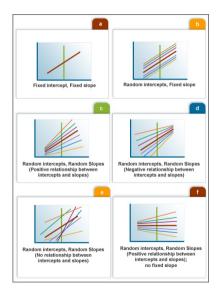
<2e-16 \*\*\*

<2e-16 \*\*\*



## Mixed models enable us to account for variability

- Varying intercepts
- Varying slopes



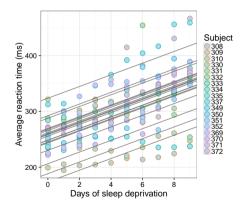
## Hence there's gradient between

- Mixed models estimate varying parameters (intercepts and/or slopes) with pooling among levels (rather than considering them fully independent)
- complete pooling: Single overall intercept
  - lm (Reaction ~ Days)
- no pooling: One independent intercept for each plot
  - lm (Reaction ~ Days + factor(Subject)
- partial pooling: Inter-related intercepts
  - lm (Reaction ~ Days + (1 | Subject))

## Fitting a varying intercepts model with 1mer

```
#+BEGIN_SRC R :results output raw :exports both
fm.rInt <- lmer(Reaction - Days + (1 | Subject), sleepstudy)
summary(fm.rInt)</pre>
```

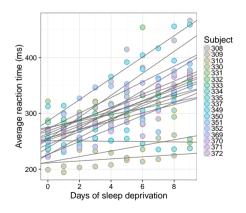
```
Linear mixed model fit by REML
     t-tests use Satterthwaite approximations to degrees of freedom ['lmerMod
     Formula: Reaction ~ Days + (1 | Subject)
        Data: sleepstudy
     REML criterion at convergence: 1786.5
     Scaled residuals:
               10 Median
     -3.226 -0.553 0.011 0.519 4.251
     Random effects:
      Groups Name
                          Variance Std.Dev.
      Subject (Intercept) 1378
                                   37.1
      Regidual
                           960
                                  31.0
     Number of obs: 180, groups: Subject, 18
     Fixed offects:
                Estimate Std. Error
                                        df t value Pr(>|t|)
     (Intercept) 251,405 9,747 22,800
                  10.467 0.804 161.000
     Davs
                                             13.0 <2e-16 ***
     Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '. '0.1 ' 1
24
     Correlation of Fixed Effects:
26
          (Intr)
     Days -0.371
```



### Fitting a varying intercepts and slopes model with 1mer

```
fm.rIntSlp <- lmer(Reaction - Days + (Days | Subject), sleepstudy)
summary(fm.rIntSlp)</pre>
```

```
Linear mixed model fit by REML
     t-tests use Satterthwaite approximations to degrees of freedom [']merMod
     Formula: Reaction ~ Days + (Days | Subject)
        Data: sleepstudy
     REML criterion at convergence: 1743.6
     Scaled residuals:
                10 Median
     -3.954 -0.463 0.023 0.463 5.179
     Random effects:
                          Variance Std.Dev. Corr
      Groups Name
      Subject (Intercept) 612.1
                                   24.74
                                    5 92
               Davs
                           35.1
                                            0.07
      Regidual
                          654 9
                                   25.59
     Number of obs: 180, groups: Subject. 18
     Fixed offects:
20
                Estimate Std. Error
                                        df t value Pr(>|t|)
     (Intercept) 251.41
                                6.82 17.00
                               1.55 17.00
     Davs
                    10.47
                                              6.77 3.3e-06 ***
24
     Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '. '0.1 ' 1
25
26
     Correlation of Fixed Effects:
          (Intr)
     Days -0.138
```



# Why no p-values in the 1me4 summaries?

- Recall that p-values in the coefficient summary of fitted linear models are the probability of getting a test statistic as large (or larger) if the coefficient was indeed 0.
- Also recall that p-values are determined using null reference distributions
  - Under the null hypothesis, the test statistic has a known distribution
- In LME models, the null reference distribution is technically not known, at least not for unbalanced data
  - Thus the lme4 authors elected to not output p-values based on a distribution that is not
    actually the distribution of the test statistic

## How do I know if a coefficient is "significant"?

- Quick, common-sense way: look at the t value
- If the t value is greater than 2, then the coefficient is likely significant
  - In other words, the coefficient estimate is more than 2 standard errors away from 0
- Note:
  - t value = Estimate / Std. Error
  - Of course ask yourself if the result is practically significant as well

# How do I know if a coefficient is "significant"?

- Another way is to compute 95% confidence intervals using the confint() function
  - If the interval contains 0, it is not significant
  - Further, confidence intervals give an indication of coefficient size and variability
- Say your fitted model object is lme1 Two ways to compute confidence intervals are as follows:
  - confint(lme1) computes a likelihood profile and finds the appropriate cutoffs based on the likelihood ratio test
  - $\bullet$  confint(lme1, method="boot") parametric bootstrapping (B = 500) with confidence intervals computed from the bootstrap distribution

### Confidence intervals for the sleepstudy data

```
fm1 <- lmer(Reaction - Days + (Days|Subject), sleepstudy)
fm1.ci <- confint(fm1, method="boot")
fm1.ci</pre>
```

```
1 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 97.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5 % 2.5
```

```
Linear mixed model fit by REML ['lmerMod']
Formula: Reaction ~ Days + (Days | Subject)
  Data: sleepstudy
REML criterion at convergence: 1743.6
Scaled residuals:
          10 Median
-3.954 -0.463 0.023 0.463 5.179
Random effects:
Groups Name
                    Variance Std. Dev. Corr
Subject (Intercept) 612.1
                             24.74
         Davs
                     35.1 5.92
                                      0.07
                     654.9
                            25.59
 Residual
Number of obs: 180, groups: Subject, 18
Fixed effects:
           Estimate Std Error t value
(Intercept)
             251.41
                         6.82
                                 36.8
              10.47
                         1.55
                                6.8
Davs
Correlation of Fixed Effects:
     (Intr)
Days -0.138
```

### But I want p-values!

- The lmerTest package provides the kind of p-values SAS provides (based on Satterthwaite's approximations)
- Just load it library(lmerTest), run lmer as usual and call summary on the object

```
library(lmerTest)
m <- lmer(dv - x1 + (x1 | g), data=df)
summary(m)</pre>
```

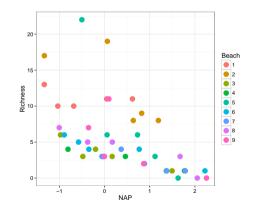
A column of p-values is included in the summary output. Again, they're approximate.

## Exercise: Random Effects on Richness using glmer

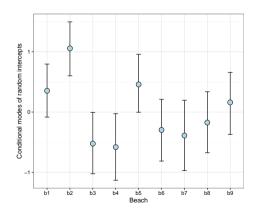
- Poisson distribution is widely used for the description of count data
- Invertebrates species richness in 45 sites on Dutch coast beaches versus height above average sea level
- We will:
  - Fit data from RIKZ survey
  - Random Effect of Beach ONLY
  - Visualize Random Effects

### Fitting and summarising a mixed model

```
Generalized linear mixed model fit by maximum likelihood (Laplace
            → Approximation) ['glmerMod']
      Family: poisson (log)
     Formula: Richness ~ NAP * angleRad + (1 | Beach)
        Data: rikz
                       logLik deviance df.resid
        217.2
                226 2 =103 6
                                  207 2
9
     Scaled residuals:
        Min
               10 Median
                             30
                                   Max
     -1 747 -0 629 -0 212 0 273 2 624
     Random effects:
14
                        Variance Std.Dev.
      Groups Name
      Beach (Intercept) 0.307
                                 0.554
16
     Number of obs: 45, groups: Beach, 9
     Fixed effects:
19
                 Estimate Std. Error z value Pr(>|z|)
     (Intercept)
                     1.850
                               0.216
                                        8.57 < 2e-16 ***
                    -0.767
     NAP
                               0.135
                                      -5.70 1.2e-08 ***
                   -0.228
     angleRad
                               0.105
                                      -2.18
                                               0.030 *
     NAP:angleRad 0.184
                                       2.22
                               0.083
                                               0.026 *
```



### Parametric bootstrap to estimate variation in random effects

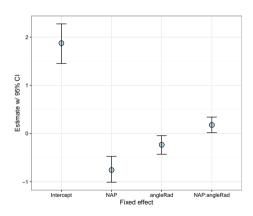


# Parametric bootstrap confidence intervals

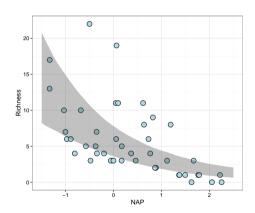
```
# modi_glmer_ci <- confint(mod1_glmer, method="boot")
mod1_glmer_ci</pre>
```

```
1 2.5 % 97.5 % 2 .8ig01 0.1635 0.80056 3 (Intercept) 1.4111 2.27656 4 MAP -1.0454 -0.56724 5 angleRad -0.5659 -0.00781 6 NAP:angleRad -0.0415 0.40827
```

## Parametric bootstrap fixed effects estimates



### Parametric bootstrap model predictions



#### Differences between 1mer and 1me

- Package nlme contains function lme
- Package 1me4 contains function 1mer
- 1me has more flexibility in specifying the covariance structure
- lmer allows you to fit GLMMs (using glmer)
  - Includes glmer.nb for negative binomial variance models
- Different specification of random effects in call

```
# lmer
fm.rIntSlp <- lmer(Reaction ~ Days + (Days | Subject), sleepstudy)
summary(fm.rIntSlp)
# lne
fm.rIntSlp <- lme(Reaction ~ Days, random = ~ Days | Subject, sleepstudy)
summary(fm.rIntSlp)</pre>
```

### Challenge

- Growth and maternal effects in mice (mouse.csv)
  - In a laboratory experiment, 20 female mice each gave birth to a litter of pups
  - Each litter was reared in a single cage, and each pup was weighed at several timepoints
- Some biological questions of interest are:
  - Do males and females differ with respect to their average growth rates?
  - Is there a maternal effect? (i.e., do individuals from the same litter tend to be similar?)
- See R script for data and code!