Mixed-effects models Solutions to Exercises

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Contents

Mixed-effects models	2
1.10 Exercises	2
1.10.1 PREF Canopy data	2
1.10.2 Mouse metabolism	8
1.10.3 Litter decomposition data	17
1.10.4 Repeated measures in leaf photosynthesis	23
1.10.5 EucFACE ground cover data	29
1.10.6 Logistic regression	34

Chapter 1

Mixed-effects models

1.10 Exercises

In these exercises, we use the following colour codes:

- **Easy**: make sure you complete some of these before moving on. These exercises will follow examples in the text very closely.
- ♦ **Intermediate**: a bit harder. You will often have to combine functions to solve the exercise in two steps.
- ▲ **Hard**: difficult exercises! These exercises will require multiple steps, and significant departure from examples in the text.

We suggest you complete these exercises in an **R** markdown file. This will allow you to combine code chunks, graphical output, and written answers in a single, easy-to-read file.

1.10.1 PREF Canopy data

1. Fit and compare mixed-effects models testing the fixed effects of species and dfromtop on narea (nitrogen concentration in grams per square metre). Evaluate whether the model fit is improved when including a random slope for each individual. Evaluate the significance of the fixed effects and their interaction.

```
library(lme4)
library(visreg)
library(car)

# read in data
pref <- read.csv("prefdata.csv")

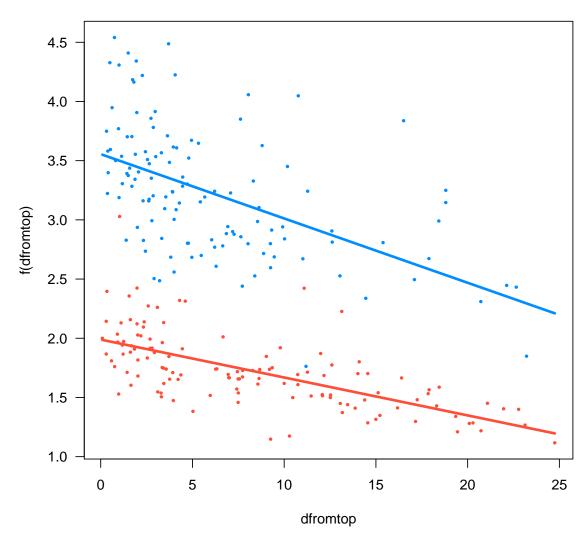
# Random intercept only
lmer1 <- lmer(narea ~ species + dfromtop + species:dfromtop + (1|ID), data=pref)

# Random intercept and slope
lmer2 <- lmer(narea ~ species + dfromtop + species:dfromtop + (dfromtop|ID), data=pref)

# significance of random slope? Not significant</pre>
```

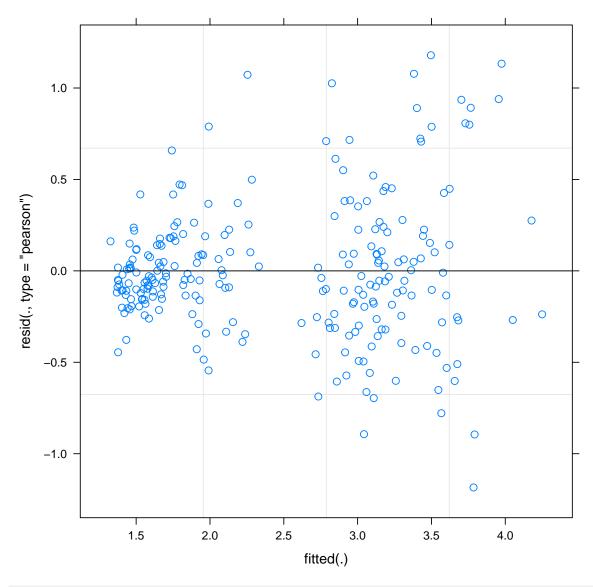
```
anova(lmer1, lmer2)
## Data: pref
## Models:
## lmer1: narea \sim species + dfromtop + species:dfromtop + (1 | ID)
## lmer2: narea ~ species + dfromtop + species:dfromtop + (dfromtop | ID)
## Df AIC BIC logLik deviance Chisq Chi Df Pr(>Chisq)
## lmer1 6 321.72 342.82 -154.86 309.72
## lmer2 8 325.36 353.49 -154.68 309.36 0.3652 2
                                                         0.8331
# significance of fixed effects?
Anova(lmer1, test='F')
## Analysis of Deviance Table (Type II Wald F tests with Kenward-Roger df)
##
## Response: narea
                         F Df Df.res
##
                                      Pr(>F)
## dfromtop
                 108.9832 1 33.061 5.336e-12 ***
                  57.6841 1 238.204 6.983e-13 ***
## species:dfromtop 4.1127 1 241.119 0.04366 *
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
# visualise model prediction
visreg(lmer1, xvar='dfromtop', by='species', overlay=T)
```



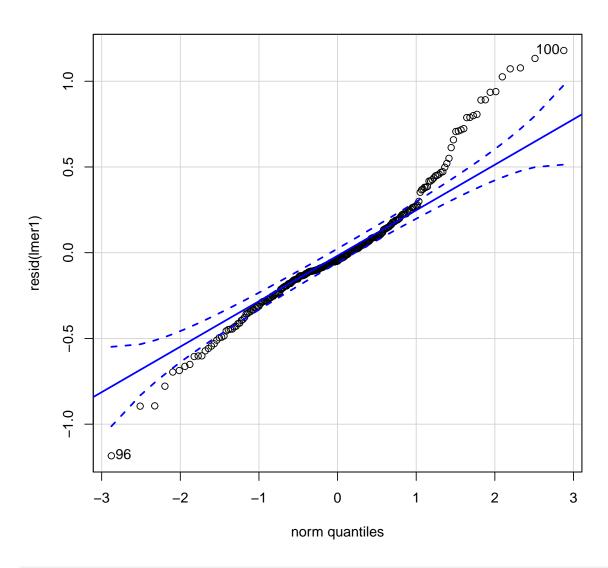


2. Generate diagnostic plots for the output of the previous analysis to evaluate whether assumptions of normality and heteroscedasticity are violated. Does a sqrt- or log-transformation improve model fit? Repeat the analysis on the transformed data, using the most appropriate transformation.

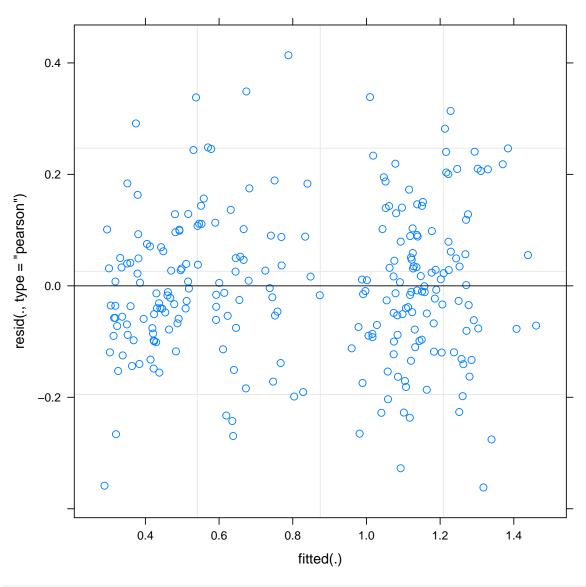
residuals versus fitted plot
plot(lmer1)



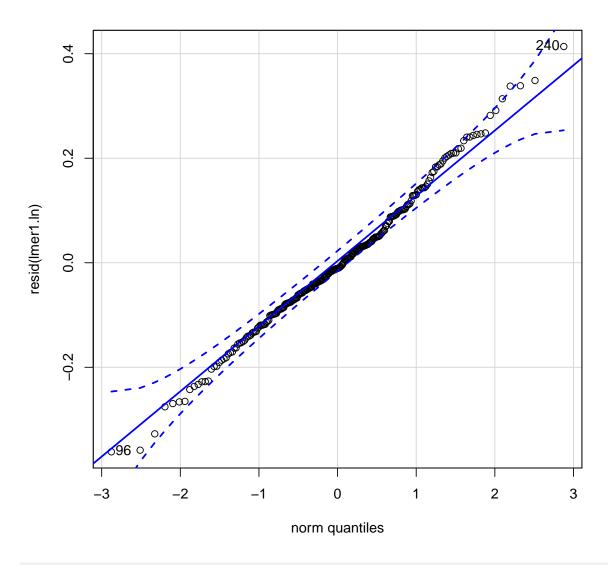
qqplot
qqPlot(resid(lmer1))



```
## [1] 96 100
# example with log-transformation
lmer1.ln <- lmer(log(narea) ~ species + dfromtop + species:dfromtop + (1|ID), data=pref)
# residuals versus fitted plot
plot(lmer1.ln)</pre>
```



qqplot
qqPlot(resid(lmer1.ln))



[1] 240 96

1.10.2 Mouse metabolism

In this exercise you will practice with the mouse metabolism data ('wildmousemetabolism.csv').

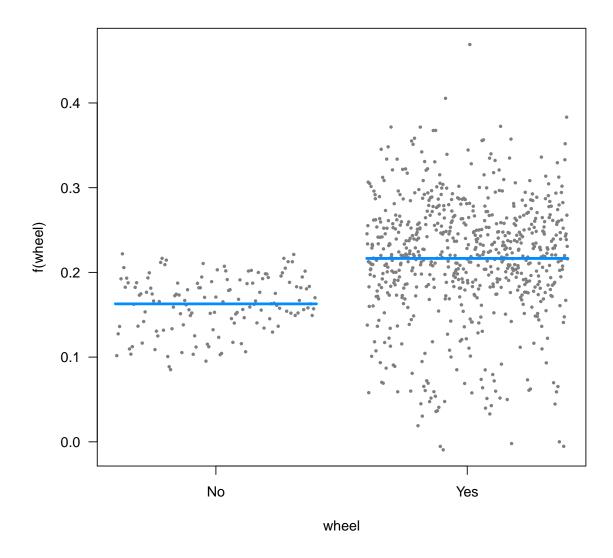
1. ■ For the mouse_m5 model, it was determined that wheel (whether or not the mice were using the wheel for exercise) was a significant predictor. What was the effect size? Use both summary and visreg.

```
mouse <- read.csv("wildmousemetabolism.csv")

# Make sure the individual label ('id') is a factor variable
mouse$id <- as.factor(mouse$id)

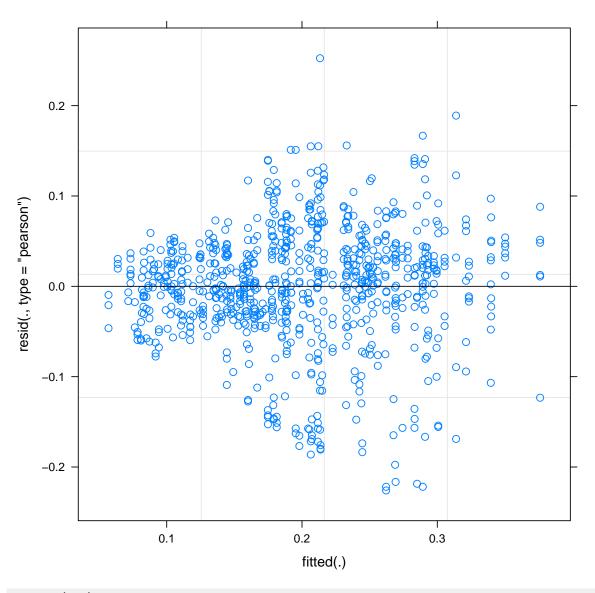
# recode intercept for temperature</pre>
```

```
mouse$temp31 <- mouse$temp - 31
# fit model
mouse_m5 <- lmer(rmr ~ bm*temp31 + wheel + (temp31|id/run), data=mouse)</pre>
# model summary
summary(mouse_m5)
## Linear mixed model fit by REML ['lmerMod']
## Formula: rmr ~ bm * temp31 + wheel + (temp31 | id/run)
     Data: mouse
##
## REML criterion at convergence: -1992.5
##
## Scaled residuals:
## Min 1Q Median
                           3Q
## -3.3436 -0.4558 0.0601 0.5774 3.7363
##
## Random effects:
## Groups Name
                       Variance Std.Dev. Corr
## run:id (Intercept) 2.015e-04 0.014195
           temp31 3.216e-06 0.001793 -1.00
## id
            (Intercept) 0.000e+00 0.000000
##
                        1.454e-06 0.001206 NaN
            temp31
## Residual
                        4.566e-03 0.067575
## Number of obs: 834, groups: run:id, 48; id, 16
## Fixed effects:
               Estimate Std. Error t value
##
## (Intercept) 0.0032002 0.0296136 0.108
              0.0064455 0.0019549 3.297
## bm
## temp31
             0.0013840 0.0033899 0.408
## wheelYes
              0.0536174 0.0078597 6.822
## bm:temp31 -0.0005032 0.0002252 -2.235
## Correlation of Fixed Effects:
##
          (Intr) bm temp31 whelYs
## bm
            -0.979
            0.232 -0.242
## temp31
## wheelYes -0.140 0.001 0.078
## bm:temp31 -0.241 0.246 -0.985 0.001
## convergence code: 0
## unable to evaluate scaled gradient
## Model failed to converge: degenerate Hessian with 1 negative eigenvalues
# visualise effect size
visreg(mouse_m5, xvar='wheel')
## Conditions used in construction of plot
## bm: 14.6
## temp31: -11
## id: 99
## run: 2
```

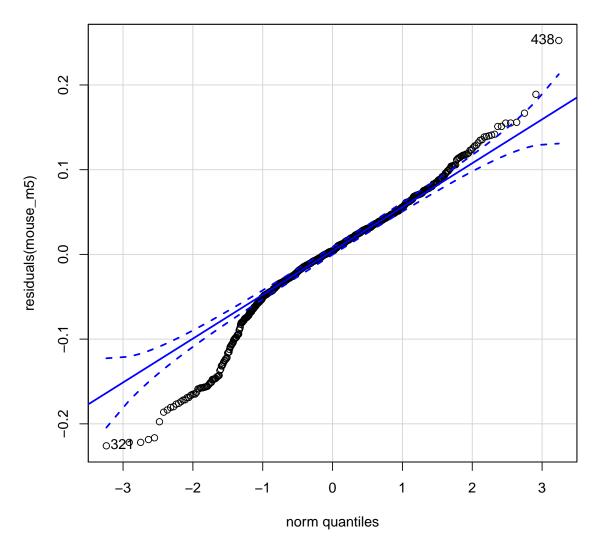


2. Inspect diagnostics for the best of the mouse models (use AIC on all models to choose the best one), including QQ plot, and residuals versus fitted. Does a logarithmic transformation (either predictor or response) improve the diagnostics?

```
mouse_m5 <- lmer(rmr ~ bm*temp31 + wheel + (temp31|id/run), data=mouse)
plot(mouse_m5)</pre>
```



library(car)
qqPlot(residuals(mouse_m5))



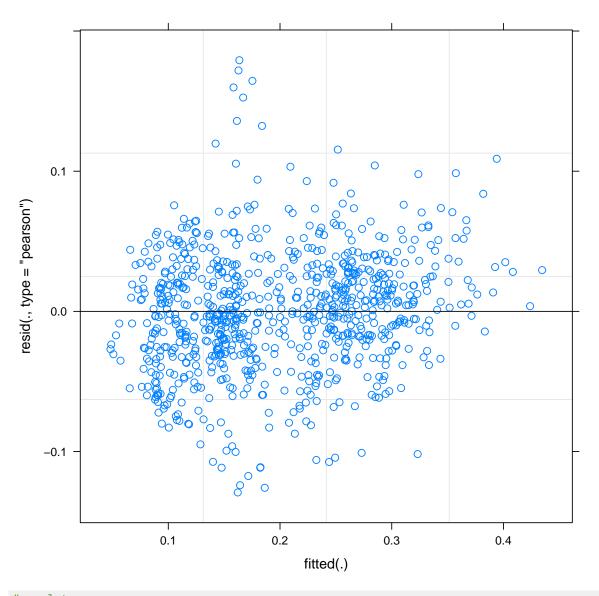
```
## 438 321
## 429 318
```

3. The mouse dataframe also contains a variable indicating the day on which the measurements took place. Each mouse was exposed to each temperature once on each of six sequential days. Include day of measurement as another random effect, check whether the model fit is improved, and evaluate the significance of each of the fixed effects.

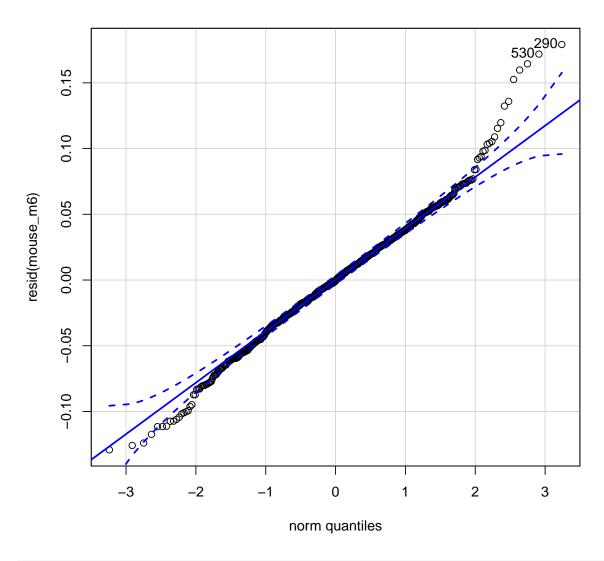
```
mouse_m5 <- lmer(rmr ~ bm*temp31 + wheel + (temp31|id/run), data=mouse)
mouse_m6 <- lmer(rmr ~ bm*temp31 + wheel + (temp31|id/day/run), data=mouse)
anova(mouse_m5, mouse_m6)

## Data: mouse
## Models:
## mouse_m5: rmr ~ bm * temp31 + wheel + (temp31 | id/run)
## mouse_m6: rmr ~ bm * temp31 + wheel + (temp31 | id/day/run)
## Df AIC BIC logLik deviance Chisq Chi Df Pr(>Chisq)
## mouse_m5 12 -2024.8 -1968.1 1024.4 -2048.8
```

```
## mouse_m6 15 -2221.1 -2150.2 1125.5 -2251.1 202.27 3 < 2.2e-16 ***
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
mouse_m7 <- lmer(rmr ~ bm*temp31 + wheel + (temp31|id/day), data=mouse)</pre>
anova(mouse_m6, mouse_m7)
## Data: mouse
## Models:
## mouse_m7: rmr \sim bm * temp31 + wheel + (temp31 | id/day)
## mouse_m6: rmr ~ bm * temp31 + wheel + (temp31 | id/day/run)
        Df AIC BIC logLik deviance Chisq Chi Df Pr(>Chisq)
## mouse_m7 12 -2146.6 -2089.9 1085.3 -2170.6
## mouse_m6 15 -2221.1 -2150.2 1125.5 -2251.1 80.51 3 < 2.2e-16 ***
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
# residuals versus fitted plot
plot(mouse_m6)
```



qqplot
qqPlot(resid(mouse_m6))



```
## 290 530
## 287 512
# fixed effects test
Anova(mouse_m6, test='F')
## Analysis of Deviance Table (Type II Wald F tests with Kenward-Roger df)
##
## Response: rmr
##
                 F Df Df.res
                               Pr(>F)
            18.9720 1 31.873 0.0001286 ***
## bm
            77.6848 1 17.495 7.562e-08 ***
## temp31
## wheel
           ## bm:temp31
             2.6467 1 51.324 0.1098908
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

4. ♦ Take a subset of the mouse data where temperature (temp) is 31. Run a mixed-effects model of resting metabolic rate with only sex (Male or Female) as predictor. Choose appropriate random effects. Does rmr vary with sex of the mouse?

```
mouse31 <- subset(mouse, temp==31)

m31_1 <- lmer(rmr ~ sex + (1|run/id), data=mouse31)
Anova(m31_1)

## Analysis of Deviance Table (Type II Wald chisquare tests)

##

## Response: rmr

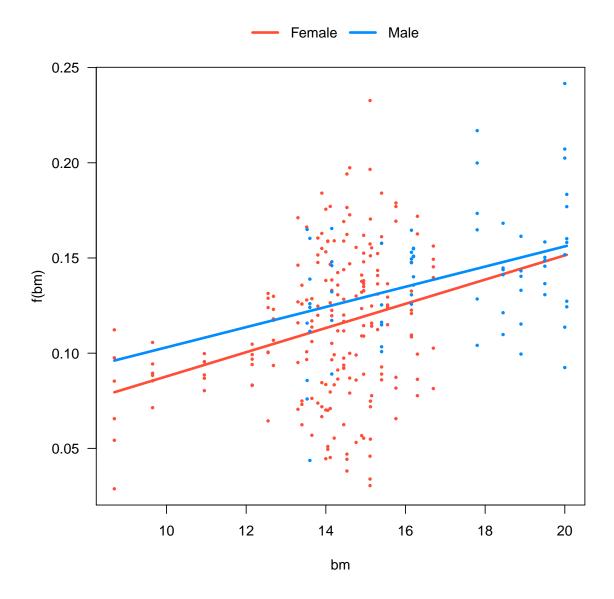
## Chisq Df Pr(>Chisq)

## sex 9.7379 1 0.001805 **

## ---

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

5. Now add body mass (bm) to the model with sex, as a main effect and interaction. Is sex still significant? Use visreg to understand how body mass, sex, and resting metabolic rate are related. If it helps, also test whether body mass varies with sex.



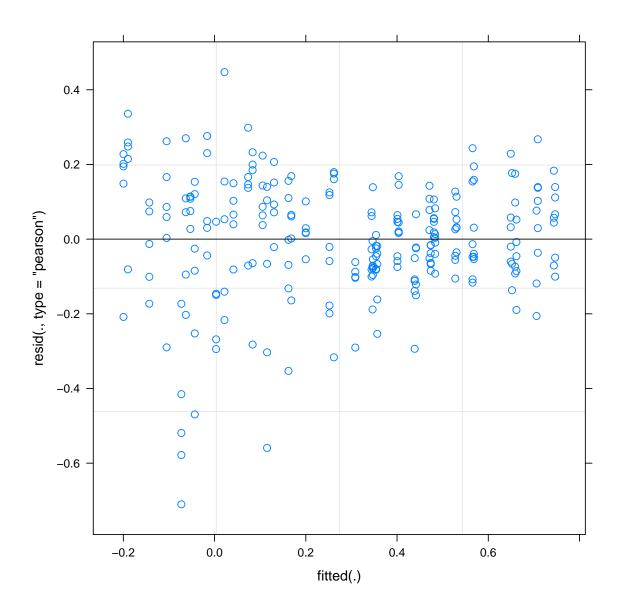
1.10.3 Litter decomposition data

1. ■ Generate some diagnostic plots for the litter_m1 model described in Section ??.

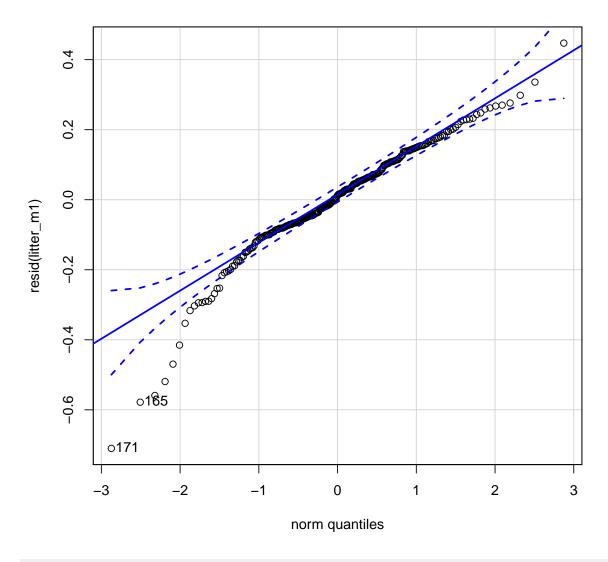
```
# Read data
litter <- read.csv("masslost.csv")

# Make sure the intended random effects (plot and block) are factors
litter$plot <- as.factor(litter$plot)
litter$block <- as.factor(litter$block)

# Represent date as number of days since the start of the experiment
library(lubridate)
##</pre>
```



```
# qqplot
qqPlot(resid(litter_m1))
```



[1] 171 165

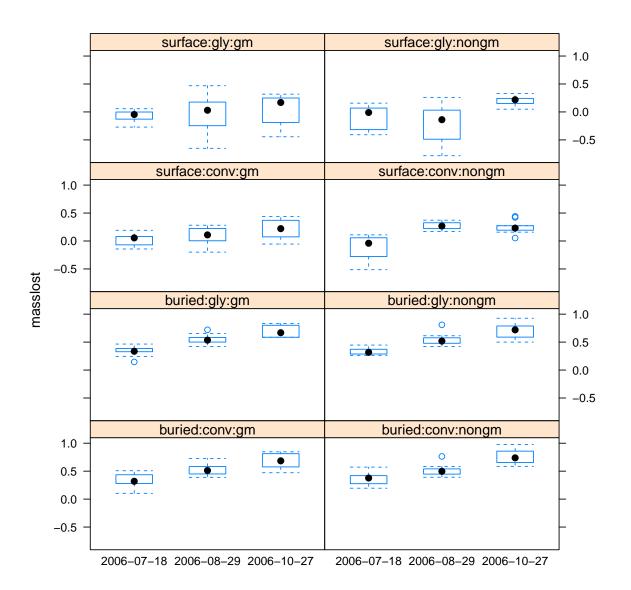
2. The litter data contain a factor (variety) describing whether the litter is derived from a genetically modified (gm) or conventional (nongm) soy variety. Plot the data to observe the effect of variety. Use lmer to test the effect of variety, in addition to the other significant variables, on litter decomposition.

```
# Read data and get summary
litter <- read.csv('masslost.csv')

# Change random effects (plot and block) to factors
litter$plot <- as.factor(litter$plot)
litter$block <- as.factor(litter$block)</pre>
```

```
# Represent date as number of days since the start of the experiment
library(lubridate)
litter$date <- as.Date(mdy(litter$date))
litter$date2 <- litter$date - as.Date('2006-05-23')

# look for treatment effects
library(lattice)
bwplot(masslost ~ factor(date) | profile:herbicide:variety, data=litter, layout=c(2,4))</pre>
```

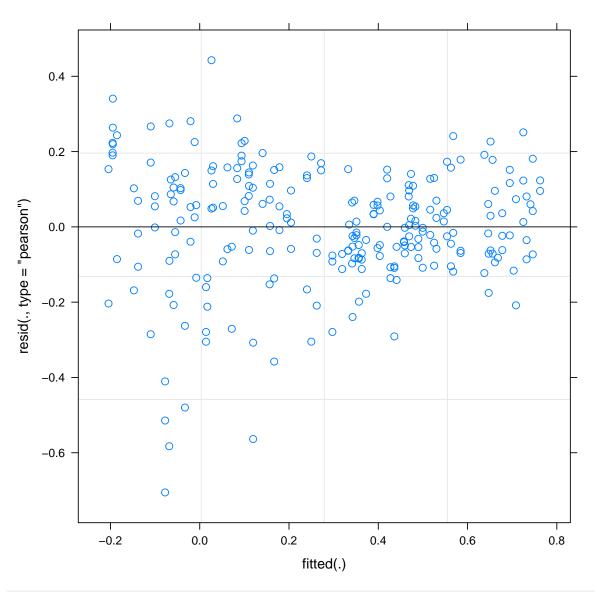


```
# there does not look to be much effect of 'variety' (look across two
# columns - 'layout' argument controls this)

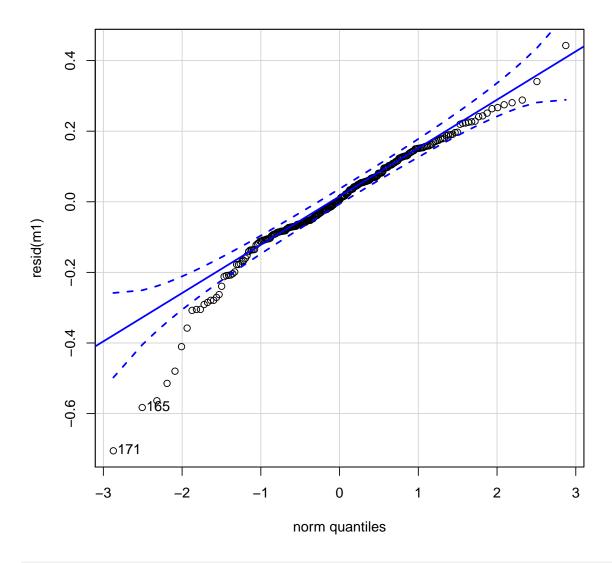
library(lme4)
library(car)
m1 <- lmer(masslost ~ date2 + herbicide * profile * variety + (1|block/plot), data = litter)</pre>
```

```
Anova(m1, test='F')
## Analysis of Deviance Table (Type II Wald F tests with Kenward-Roger df)
## Response: masslost
                                       F Df Df.res Pr(>F)
##
## date2
                              140.0675 1 231.436 < 2.2e-16 ***
## herbicide
                               12.7977 1 3.005 0.0372580 *
## profile
                             519.2389 1 231.498 < 2.2e-16 ***
## variety
                                0.3392 1 231.424 0.5608858
## herbicide:profile 11.7097 1 231.277 0.0007351
## herbicide:variety 0.4590 1 231.820 0.4987471
## profile:variety 0.0758 1 231.795 0.7832914
                               11.7097 1 231.277 0.0007351 ***
## herbicide:profile:variety 0.0051 1 231.304 0.9431549
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
# residuals versus fitted plot
plot(m1)
```

21



qqplot
qqPlot(resid(m1))



[1] 171 165

1.10.4 Repeated measures in leaf photosynthesis

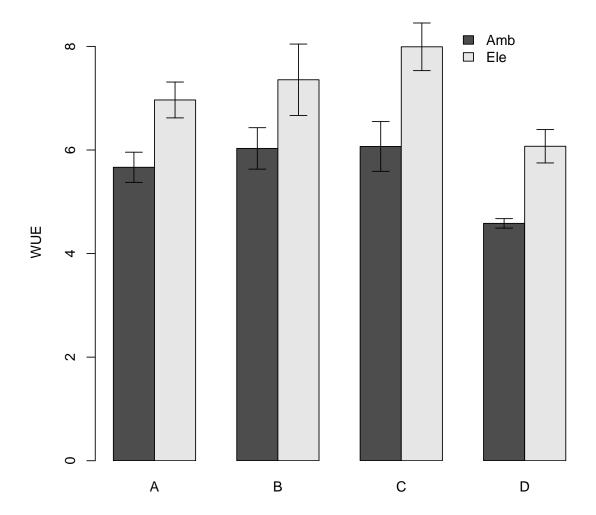
For this exercise you will use the EucFACE leaf gas exchange dataset. First walk through Section ?? before starting this exercise.

1. Read the data, and add a new variable called WUE (water-use efficiency), calculated as Photo divided by Trmmol. This new variable represents the amount of CO2 taken up per unit water lost.

```
eucgas <- read.csv("eucface_gasexchange.csv")
eucgas$WUE <- with(eucgas, Photo / Trmmol)</pre>
```

2. Make a barplot of WUE by Date and CO2 treatment, as shown in Section ??.

```
library(sciplot)
bargraph.CI(Date, WUE, CO2, data=eucgas, legend=TRUE, ylab="WUE")
```



3. Fit a linear mixed-effects model with Tree within Ring as the random effects structure, and Date and CO2 as fixed effect. Fit a second model with additionally the interaction between Date and CO2; is the interaction significant (use both anova on the two models, and Anova on the model with the interaction).

```
wue_m0 <- lmer(WUE ~ Date + CO2 + (1|Ring/Tree), data=eucgas)
wue_m1 <- lmer(WUE ~ Date * CO2 + (1|Ring/Tree), data=eucgas)

# Likelihood ratio test
anova(wue_m0, wue_m1)

## Data: eucgas
## Models:
## wue_m0: WUE ~ Date + CO2 + (1 | Ring/Tree)</pre>
```

```
## wue_m1: WUE ~ Date * CO2 + (1 | Ring/Tree)
        Df
                     BIC logLik deviance Chisq Chi Df Pr(>Chisq)
## wue_m0 8 288.64 307.99 -136.32
                                   272.64
## wue_m1 11 293.79 320.40 -135.89
                                   271.79 0.8477
                                                             0.838
# Chi-square test
Anova(wue m1)
## Analysis of Deviance Table (Type II Wald chisquare tests)
## Response: WUE
##
             Chisq Df Pr(>Chisq)
## Date
           23.0332 3 3.974e-05 ***
## CO2
          18.2081 1 1.980e-05 ***
## Date:CO2 0.7605 3
                          0.8589
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

4. Inspect the standard deviation of the random effects with VarCorr. Notice that the standard deviation for the term Tree:Ring (trees within rings) is very small compared to the Ring random effect. A naive user might remove this random effect since it explains little variation. Why is that a bad idea?

VarCorr(wue_m0)

```
## Groups Name Std.Dev.
## Tree:Ring (Intercept) 0.00000
## Ring (Intercept) 0.28164
## Residual 1.26842
# The trees are the experimental units. If we remove it from the random effects structure, we assum # all measurements within rings are independent, and are thus committing pseudoreplication.
```

5. A Look at the summary statement of the model for WUE that does not contain an interaction. Notice that Date D is significantly lower than the intercept (Date A). Refit the model without an intercept, and use glht to test whether Date D was lower than all of Date A, B and C. Hint: this follows closely the example shown at the end of Section ??.

```
wue_m0_re <- lmer(WUE ~ Date + CO2 -1 + (1|Ring/Tree), data=eucgas)
library(multcomp)

## Loading required package: mutnorm

## Loading required package: survival

## Loading required package: TH.data

## Loading required package: MASS

##

## Attaching package: 'TH.data'

## The following object is masked from 'package:MASS':

##

## geyser

##

## Attaching package: 'multcomp'

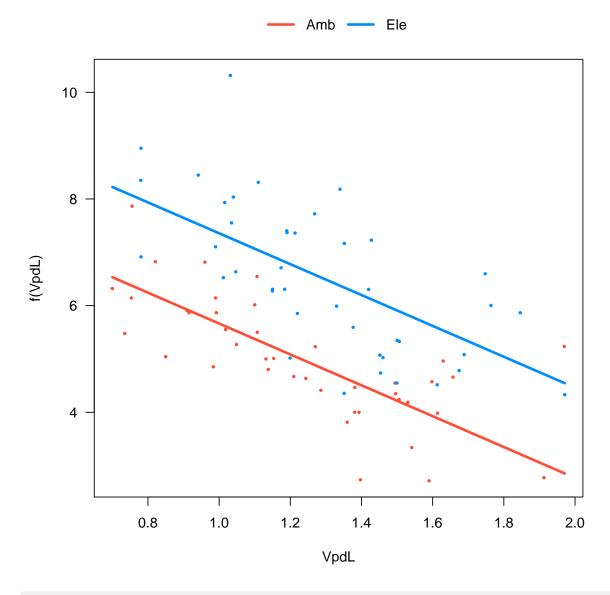
## The following object is masked _by_ '.GlobalEnv':</pre>
```

6. ♦ A colleague tells you that water-use efficiency may be lower on Date D because air temperature was higher, resulting in a lower air humidity. This is indicated by the variable VpdL in the dataset, which represents the 'vapour pressure deficit' - higher values indicate drier air. Refit the model for WUE with the CO2*Date interaction, and now add VpdL as a fixed effect to the model (but no interactions between VpdL and Date or CO2). Is VPD significant (use multiple methods to test this)?

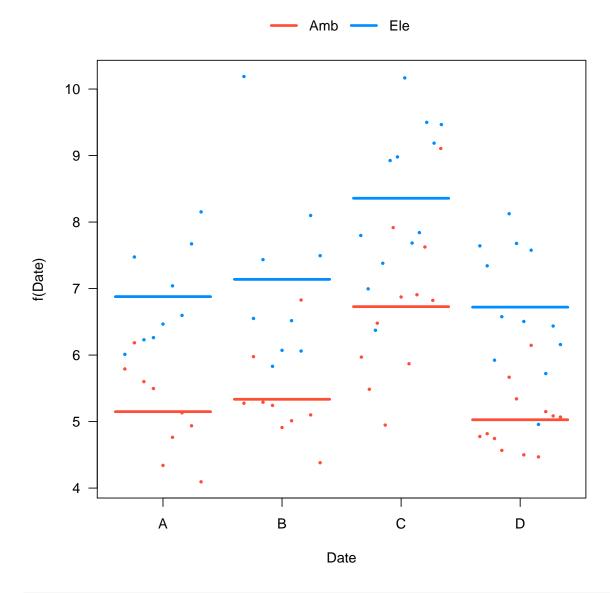
```
wue_m2 <- lmer(WUE ~ VpdL + Date * CO2 + (1|Ring/Tree), data=eucgas)</pre>
Anova(wue_m2)
## Analysis of Deviance Table (Type II Wald chisquare tests)
##
## Response: WUE
##
             Chisq Df Pr(>Chisq)
## VpdL
           36.5358 1 1.499e-09 ***
           36.2087 3 6.765e-08 ***
## Date
## CO2
           20.4753 1 6.040e-06 ***
## Date:CO2 0.0718 3
                           0.995
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

7. A Use visreg to visualize a) the VpdL effect (make a visreg plot with VPD as the predictor, showing predictions for both CO2 treatment), b) The Date effect. For this latter plot, use Date as the xvar argument to visreg, and CO2 as the by argument (with overlay=TRUE). For this last plot, visreg will show the Date effect at a median value of VpdL, that is, the effect of VpdL has been removed first. Is Date D still different from the other Dates? Can you explain this in terms of the VpdL covariate?

```
# VPD effect - visreg will take the first Date for the predictions
visreg(wue_m2, "VpdL", by="CO2", overlay=TRUE)
```



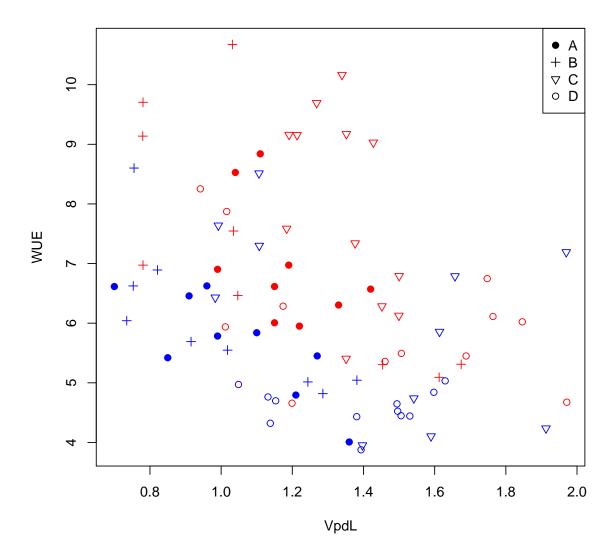
The Date effect. visreg will show the predictions at a median value of VPD.
visreg(wue_m2, "Date", by="CO2", overlay=TRUE)



```
# The Dates are now very similar, and Date D is no longer lower. This means that VPD explained the # WUE on Date D, because in the above model its effect has been accounted for.

# We can visually check this in the figure below. Note that Date D is lower, but also has higher VP # all data for a CO2 treatment are mose or less on one line.

with(eucgas, plot(VpdL, WUE, pch=c(19,3,6,1)[Date], col=c("blue", "red")[CO2]))
legend("topright", levels(eucgas$Date), pch=c(19,3,6,1))
```



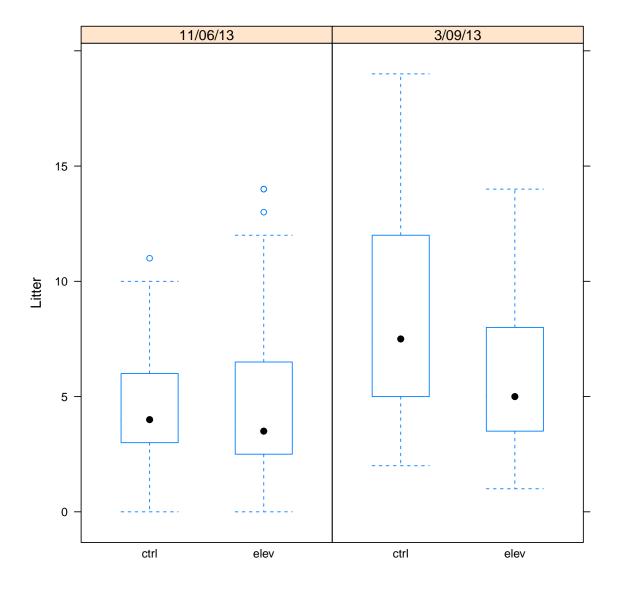
1.10.5 EucFACE ground cover data

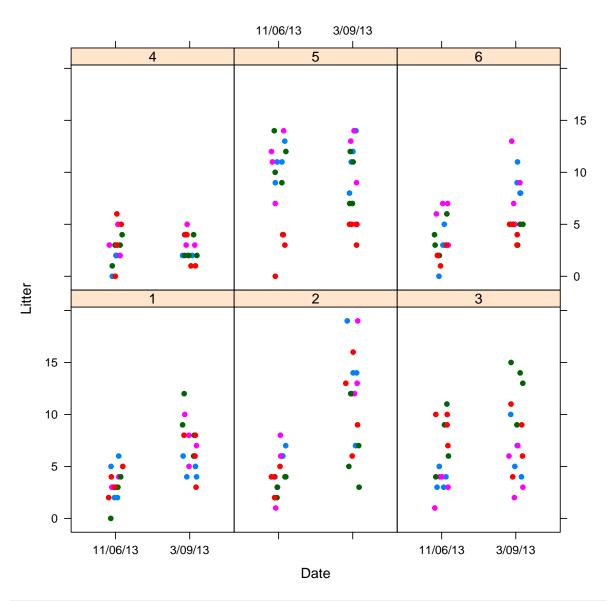
The file eucfaceGC.csv contains estimates of plant and litter cover within the rings of the EucFACE experiment, evaluating forest ecosystem responses to elevated CO2, on two dates; the data description can be found in Section ?? (p. ??).

1. ♦ Convert the variables indicating the nested sampling design to factors, then use glmer in lme4 to test for an interaction between Trt and Date on Litter cover. Litter cover represents counts (use the poisson family).

```
# read data
gc <- read.csv('eucfaceGC.csv')
str(gc)
## 'data.frame': 192 obs. of 8 variables:
## $ Date : Factor w/ 2 levels "11/06/13", "3/09/13": 1 1 1 1 1 1 1 1 1 1 1 ...</pre>
```

```
## $ Ring : int 1 1 1 1 1 1 1 1 1 1 1 ...
## $ Plot : int 1 1 1 1 1 1 1 1 1 1 ...
## $ Sub : int 1 2 3 4 1 2 3 4 1 2 ...
## $ Forbes: int 0 2 0 0 2 5 4 0 1 3 ...
## $ Grass : int 16 14 16 16 15 16 16 16 13 ...
## $ Litter: int 5 6 2 2 3 3 4 3 0 4 ...
## $ Trt : Factor w/ 2 levels "ctrl","elev": 2 2 2 2 2 2 2 2 2 2 2 2 2 ...
## convert random variables to factors
gc$Ring <- factor(gc$Ring)
gc$Plot <- factor(gc$Plot)
gc$Sub <- factor(gc$Plot)
## Plot variation associated with 'Trt' and date
## bwplot(Litter ~ Trt | Date, data = gc)</pre>
```



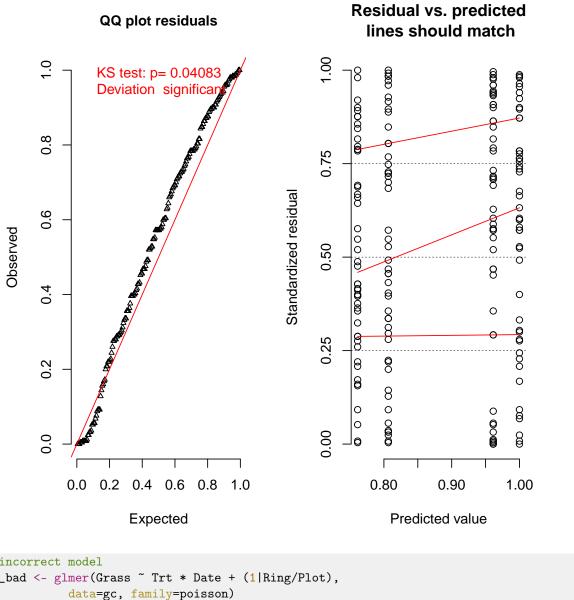


```
# fit model and test for effects
m2 <- glmer(Litter ~ Trt * Date + (1|Ring/Plot), data=gc, family=poisson)</pre>
summary(m2)
## Generalized linear mixed model fit by maximum likelihood (Laplace
     Approximation) [glmerMod]
## Family: poisson ( log )
## Formula: Litter ~ Trt * Date + (1 | Ring/Plot)
##
      Data: gc
##
##
        AIC
                 BIC
                       logLik deviance df.resid
                       -455.6
##
      923.2
               942.8
                                 911.2
                                             186
##
```

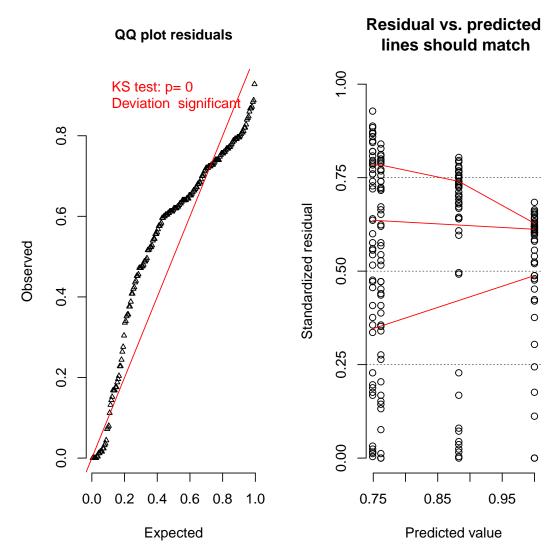
```
## Scaled residuals:
       Min 1Q
                     Median
                                  30
                                          Max
## -2.22194 -0.66709 0.02852 0.54770 2.40787
##
## Random effects:
## Groups
             Name
                         Variance Std.Dev.
## Plot:Ring (Intercept) 0.1023 0.3199
             (Intercept) 0.0867
                                  0.2945
## Number of obs: 192, groups: Plot:Ring, 24; Ring, 6
##
## Fixed effects:
                      Estimate Std. Error z value Pr(>|z|)
##
## (Intercept)
                       1.45991
                                  0.20529 7.111 1.15e-12 ***
                                  0.29009
                                            0.062 0.950914
## Trtelev
                       0.01786
                                  0.08334 7.554 4.22e-14 ***
## Date3/09/13
                       0.62952
## Trtelev:Date3/09/13 -0.42318
                                  0.12015 -3.522 0.000428 ***
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Correlation of Fixed Effects:
##
              (Intr) Trtelv D3/09/
## Trtelev
              -0.707
## Date3/09/13 -0.265 0.187
## Tr:D3/09/13 0.184 -0.248 -0.694
library(car)
Anova(m2)
## Analysis of Deviance Table (Type II Wald chisquare tests)
##
## Response: Litter
##
             Chisq Df Pr(>Chisq)
## Trt
            0.7057 1 0.400889
## Date
           50.3421 1 1.291e-12 ***
## Trt:Date 12.4057 1 0.000428 ***
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

2. Make model diagnostics plot for the model of Grass with treatment and Date as fixed effects. Use the DHARMa package as shown in Fig. ??. Now refit this model with the poisson family - which is a wrong model specification because Grass count is clearly bounded by a maximum (16 quadrants in each plot). Make the model diagnostic plot again, and note how it has changed.

DHARMa scaled residual plots



DHARMa scaled residual plots



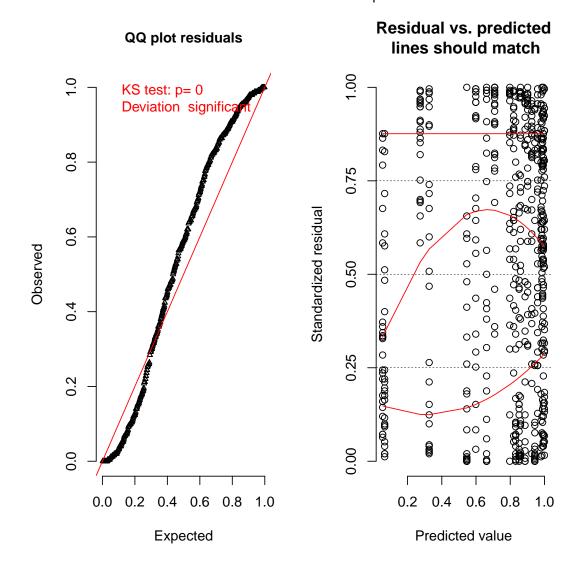
1.10.6 Logistic regression

For this exercise, we will use the seed germination data (fire experiment). First walk through Section ??.

For the seedfire dataset, the examples in the text use temp as a factor variable. Attempt to instead
use it as an (untransformed) continuous variable in the model. Compare the model with temp as
a factor and temp as a factor, in terms of AIC. Now plot diagnostic plots for both models (see
Section ??). Ignore the warning.

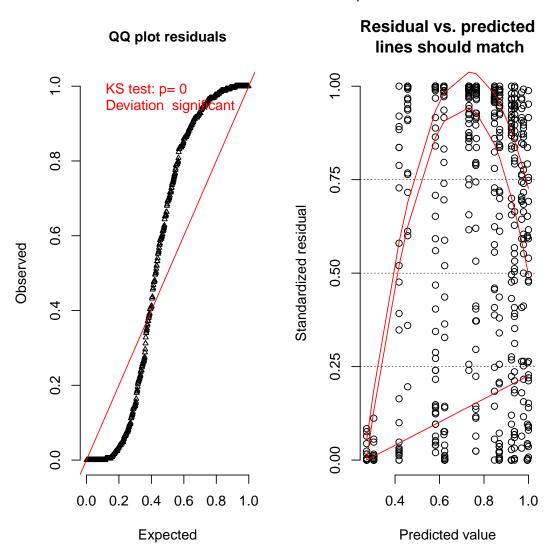
```
# Make sure temperature treatment is a factor
seedfire$temp <- as.factor(seedfire$temp)</pre>
# temp as factor
```

DHARMa scaled residual plots



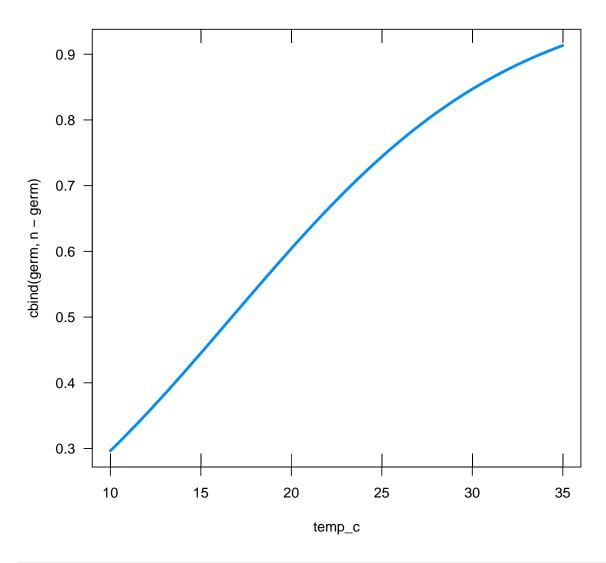
plotSimulatedResiduals(simulateResiduals(firefitc1))

DHARMa scaled residual plots

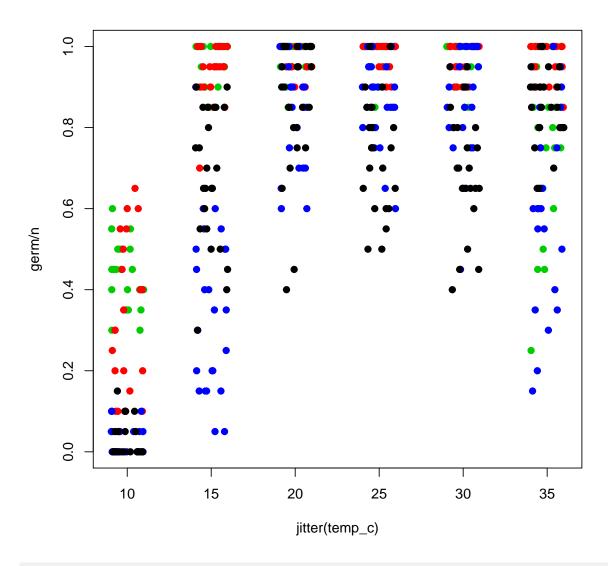


2. Try to understand why temp as a continuous variable does so badly, using visreg (set temp as the xvar, and also use scale="response"). Compare the fitted response to the data (i.e., also make a plot of germination success versus temperature, and compare to the visreg plot).

```
visreg(firefitc1, "temp_c", scale="response")
```



```
# Compare to:
palette("default")
with(seedfire, plot(jitter(temp_c), germ/n, col=species, pch=19))
```



The data show an abrupt increase in germination success with temperature, but the predictor # was added as a linear term, which is much too gradual.