

# A Joint Regression model

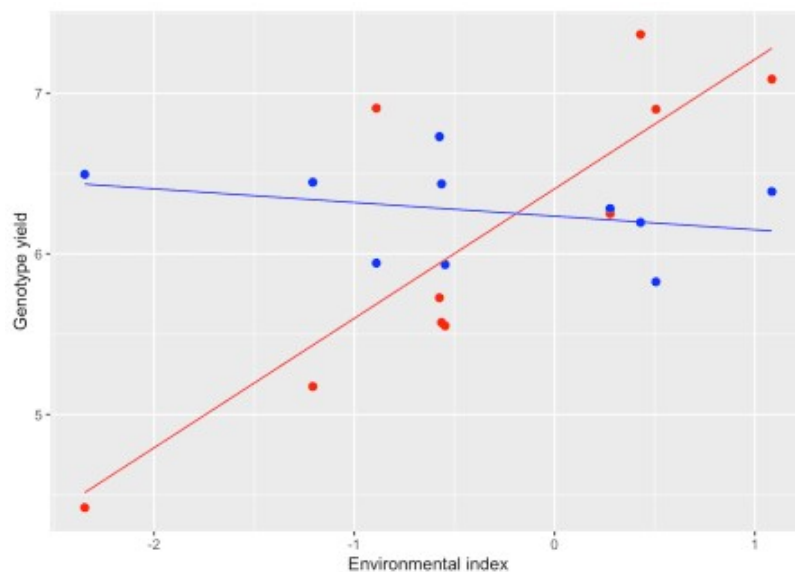
Let's talk about a very old, but, nonetheless, useful technique. It is widely known that the yield of a genotype in different environments depends on environmental covariates, such as the amount of rainfall in some critical periods of time. Apart from rain, also temperature, wind, solar radiation, air humidity and soil characteristics may concur to characterise a certain environment as good or bad and, ultimately, to determine yield potential.

Early in the 60s, several authors proposed that the yield of genotypes is expressed as a function of an environmental index  $(e_j)$ , measuring the yield potential of each environment  $(j)$  (Finlay and Wilkinson, 1963; Eberhart and Russel, 1966; Perkins and Jinks, 1968). For example, for a genotype  $(i)$ , we could write:

$$y_{ij} = \mu_i + \beta_i e_j$$

where the yield  $(y_{ij})$  in a certain environment  $(j)$  is expressed as a linear function of the environmental index  $(e_j)$ ;  $(\mu_i)$  is the intercept and  $(\beta_i)$  is the slope, which expresses how the genotype  $(i)$  responds to the environment.

A graphical example may be useful; in the figure below we have two genotypes tested in 10 environments. The yield of the first genotype (red) increases as the environmental index increases, with slope  $(\beta_1 = 0.81)$ . On the other hand, the yield of the second genotype (blue) does not change much with the environment  $(\beta_2 = -0.08)$ . Clearly, a high value of  $(\beta)$  demonstrates that the genotype is responsive to the environment and makes profit of favorable conditions. Otherwise, a low  $(\beta)$  value (close to 0) demonstrates that the genotype is not responsive and tends to give more or less the same yield in all environments (static stability; Wood, 1976).



By now, it should be clear that  $(\beta)$  is a relevant measure of stability. Now, the problem is: how do we determine such value from a multi-environment genotype experiment? As usual, let's start from a meaningful example.

## A multi-environment experiment

Let's take the data in Sharma (2006; Statistical And Biometrical Techniques In Plant Breeding, New Age International Ltd. New Delhi, India). They refer to a multi-environment experiment with 7 genotypes, 6 environments and 3 blocks; let's load the data in the dataframe 'dataFull'.

```
rm(list=ls())
library(nlme)
library(emmeans)
## Welcome to emmeans.
```

```
## NOTE -- Important change from versions <= 1.41:
##      Indicator predictors are now treated as 2-level factors by default.
##      To revert to old behavior, use emm_options(cov.keep = character(0))
Block <- factor(rep(c(1:3), 42))
Var <- factor(rep(LETTERS[1:7], each=18))
Loc <- factor(rep(rep(letters[1:6], each=3), 7))
Pl <- factor(Loc:Block)
Yield <- c(60,65,60,80,65,75,70,75,70,72,82,90,48,45,50,50,40,40,
          80,90,83,70,60,60,85,90,90,70,85,80,40,40,40,38,40,50,
          25,28,30,40,35,35,35,30,30,40,35,35,35,25,20,35,30,30,
          50,65,50,40,40,40,48,50,52,45,45,50,50,50,45,40,48,40,
          52,50,55,55,54,50,40,40,60,48,38,45,38,30,40,35,40,35,
          22,25,25,30,28,32,28,25,30,26,28,28,45,50,45,50,50,50,
          30,30,25,28,34,35,40,45,35,30,32,35,45,35,38,44,45,40)

dataFull <- data.frame(Block, Var, Loc, Yield)
rm(Block, Var, Loc, Pl, Yield)
head(dataFull)
##      Block Var Loc Yield
## 1      1   A   a    60
## 2      2   A   a    65
## 3      3   A   a    60
## 4      1   A   b    80
## 5      2   A   b    65
## 6      3   A   b    75
```

## What is an environmental index?

First of all, we need to define an environmental index, which can describe the yield potential in each of the seven environments. Yates and Cochran (1937) proposed that we use the mean of all observations in each environment, expressed as the difference between the environmental mean yield ( $\mu_j$ ) and the overall mean ( $\mu$ ) (i.e.  $e_j = \mu_j - \mu$ ). Let's do it; in the box below we use the package 'dplyr' to augment the dataset with a new variable, representing the environmental indices.

```
library(dplyr)
dataFull <- dataFull %>%
  group_by(Loc) %>%
  mutate(ej = mean(Yield) - mean(dataFull$Yield))
head(dataFull)
## # A tibble: 6 x 5
## # Groups:   Loc [2]
##   Block Var   Loc   Yield    ej
##
## 1 1      A     a      60  1.45
## 2 2      A     a      65  1.45
## 3 3      A     a      60  1.45
## 4 1      A     b      80  0.786
## 5 2      A     b      65  0.786
## 6 3      A     b      75  0.786
```

This step is ok with balanced data and it is clear that a high environmental index identifies the favorable environments, while a low (negative) environmental index identifies unfavorable environments. It is necessary to keep in mind that we have unwillingly put a constraint on  $e_j$  values, that have to sum up to zero.

## Full model definition (Equation 1)

Now, it is possible to regress the yield data for each genotype against the environmental indices, according to the following joint regression model:

$$y_{ijk} = \gamma_j + \mu_i + \beta_i e_j + d_{ij} + \varepsilon_{ijk} \quad \text{Equation 1}$$

where:  $y_{ijk}$  is the yield for the genotype  $i$  in the environment  $j$  and block  $k$ ,  $\gamma_j$  is the effect of blocks within environments and  $\mu_i$  is the average yield for the genotype  $i$ . As we have seen in the figure above, the average yield of a genotype in each environment cannot be exactly described by the regression against the environmental indices (in other words: the observed means do not lie along the regression line). As the consequence, we need the random term  $d_{ij}$  to represent the deviation from the regression line for the genotype  $i$  in the environment  $j$ . Finally, the random elements  $\varepsilon_{ijk}$  represent the deviations between the replicates for the genotype  $i$  in the environment  $j$  (within-trial errors). As I said,  $d_{ij}$  and  $\varepsilon_{ijk}$  are random, with variances equal to  $\sigma^2_d$  and  $\sigma^2_e$ , respectively.

According to Finlay-Wilkinson,  $\sigma^2_d$  is assumed to be equal for all genotypes. Otherwise, according to Eberhart-Russell,  $\sigma^2_{d(i)}$  may assume a different value for each genotype ( $\sigma^2_{d(i)}$ ) and may become a further measure of stability: if this is small, a genotype does not show relevant variability of yield, apart from that due to the regression against the environmental indices.

## Model fitting

We can start the analyses by fitting a traditional ANOVA model, to keep as a reference.

```
mod.aov <- lm(Yield ~ Loc/Block + Var*Loc, data = dataFull)
anova(mod.aov)
## Analysis of Variance Table
##
## Response: Yield
##          Df Sum Sq Mean Sq  F value    Pr(>F)
## Loc         5  1856.0    371.2   17.9749 1.575e-11 ***
## Var         6 20599.2   3433.2  166.2504 < 2.2e-16 ***
## Loc:Block   12   309.8     25.8    1.2502  0.2673
## Loc:Var     30 12063.6    402.1   19.4724 < 2.2e-16 ***
## Residuals  72  1486.9     20.7
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

As we said, Equation 1 is a mixed model, which calls for the use of the 'lme()' function. For better understanding, it is useful to start by augmenting the previous ANOVA model with the regression term ('Var/ej'). We use the nesting operator, to have different regression lines for each level of 'Var'.

```
# Augmented ANOVA model
mod.aov2 <- lm(Yield ~ Loc/Block + Var/ej + Loc:Var, data=dataFull)
anova(mod.aov2)
## Analysis of Variance Table
##
## Response: Yield
##          Df Sum Sq Mean Sq  F value    Pr(>F)
## Loc         5  1856.0    371.2   17.9749 1.575e-11 ***
## Var         6 20599.2   3433.2  166.2504 < 2.2e-16 ***
## Loc:Block   12   309.8     25.8    1.2502  0.2673
## Var:ej       6  9181.2   1530.2   74.0985 < 2.2e-16 ***
## Loc:Var     24  2882.5    120.1    5.8159 2.960e-09 ***
## Residuals  72  1486.9     20.7
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

We see that the GE interaction in the ANOVA model has been decomposed into two parts: the regression term ('Var/ej') and the deviation from regression ('Loc:Var'), with 6 and 24 degrees of freedom, respectively. This second term corresponds to  $d_{ij}$  in Equation 1 (please, note that the two terms 'Var/ej' and 'Loc:Var'

are partly confounded).

The above analysis is only useful for teaching purposes, but it is unsatisfactory, because the  $\sigma_{ij}^2$  terms have been regarded as fixed, which is pretty illogical. Therefore, we change the fixed effect model into a mixed model, where we include the random 'genotype by environment' interaction. We also change the fixed block effect into a random effect and remove the intercept, to more strictly adhere to the parameterisation of Equation 1. The two random effects 'Loc:Block' and 'Loc:Var' are not nested into each other and we need to code them by using 'pdMat' constructs, which are not straightforward. You can use the code in the box below as a guidance to fit a Finlay-Wilkinson model.

```
# Finlay-Wilkinson model
modFull1 <- lme(Yield ~ Var/ej - 1,
               random = list(Loc = pdIdent(~ Var - 1),
                             Loc = pdIdent(~ Block - 1)),
               data=dataFull)

summary(modFull1)$tTable
##              Value Std.Error   DF    t-value    p-value
## VarA      63.1666667 2.4017164 107  26.3006350 1.624334e-48
## VarB      66.1666667 2.4017164 107  27.5497417 2.135264e-50
## VarC      31.8333333 2.4017164 107  13.2544097 2.599693e-24
## VarD      47.1111111 2.4017164 107  19.6156012 3.170228e-37
## VarE      44.7222222 2.4017164 107  18.6209421 2.378452e-35
## VarF      34.2777778 2.4017164 107  14.2722004 1.614127e-26
## VarG      35.8888889 2.4017164 107  14.9430169 6.028635e-28
## VarA:ej    3.2249875 0.6257787 107   5.1535588 1.176645e-06
## VarB:ej    4.7936139 0.6257787 107   7.6602379 8.827229e-12
## VarC:ej    0.4771074 0.6257787 107   0.7624219 4.474857e-01
## VarD:ej    0.3653064 0.6257787 107   0.5837629 5.606084e-01
## VarE:ej    1.2369950 0.6257787 107   1.9767291 5.064533e-02
## VarF:ej   -2.4316943 0.6257787 107  -3.8858692 1.770611e-04
## VarG:ej   -0.6663160 0.6257787 107  -1.0647790 2.893729e-01
VarCorr(modFull1)
##              Variance      StdDev
## Loc =      pdIdent(Var - 1)
## VarA      27.5007919      5.2441197
## VarB      27.5007919      5.2441197
## VarC      27.5007919      5.2441197
## VarD      27.5007919      5.2441197
## VarE      27.5007919      5.2441197
## VarF      27.5007919      5.2441197
## VarG      27.5007919      5.2441197
## Loc =      pdIdent(Block - 1)
## Block1     0.4478291      0.6692003
## Block2     0.4478291      0.6692003
## Block3     0.4478291      0.6692003
## Residual  20.8781458      4.5692610
```

From the output, we see that the variance component  $\sigma_d^2$  (27.50) is the same for all genotypes; if we want to let a different value for each genotype (Eberhart-Russel model), we need to change the 'pdMat' construct for the 'Loc:Var' effect, turning from 'pdIdent' to 'pdDiag', as shown in the box below.

```
# Eberhart-Russel model
modFull2 <- lme(Yield ~ Var/ej - 1,
               random = list(Loc = pdDiag(~ Var - 1),
                             Loc = pdIdent(~ Block - 1)),
               data=dataFull)

summary(modFull2)$tTable
##              Value Std.Error   DF    t-value    p-value
## VarA      63.1666667 3.0507629 107  20.7052032 3.221930e-39
```

```
## VarB      66.1666667  2.7818326  107  23.7852798  1.604422e-44
## VarC      31.8333333  1.7240721  107  18.4640387  4.753742e-35
## VarD      47.1111111  2.3526521  107  20.0246824  5.564350e-38
## VarE      44.7222222  2.4054296  107  18.5921974  2.699536e-35
## VarF      34.2777778  1.9814442  107  17.2993906  8.947485e-33
## VarG      35.8888889  2.2617501  107  15.8677515  7.076551e-30
## VarA:ej    3.2249875  0.7948909  107   4.0571447  9.466174e-05
## VarB:ej    4.7936139  0.7248198  107   6.6135249  1.522848e-09
## VarC:ej    0.4771074  0.4492152  107   1.0620909  2.905857e-01
## VarD:ej    0.3653064  0.6129948  107   0.5959372  5.524757e-01
## VarE:ej    1.2369950  0.6267462  107   1.9736777  5.099652e-02
## VarF:ej   -2.4316943  0.5162748  107  -4.7100774  7.473942e-06
## VarG:ej   -0.6663160  0.5893098  107  -1.1306718  2.607213e-01
VarCorr(modFull2)
##          Variance          StdDev
## Loc =    pdDiag(Var - 1)
## VarA      48.7341240          6.9809830
## VarB      39.3227526          6.2707856
## VarC      10.7257438          3.2750181
## VarD      26.1010286          5.1089166
## VarE      27.6077467          5.2543074
## VarF      16.4479246          4.0556041
## VarG      23.5842788          4.8563648
## Loc =    pdIdent(Block - 1)
## Block1     0.4520678          0.6723599
## Block2     0.4520678          0.6723599
## Block3     0.4520678          0.6723599
## Residual  20.8743411          4.5688446
```

From the regression slopes we see that the genotypes A and B are the most responsive to the environment ( $\beta_A = 3.22$ ) and ( $\beta_B = 4.79$ ), respectively), while the genotypes C and D are stable in a static sense, although their average yield is pretty low.

## Fitting a joint regression model in two-steps (Equation 2)

In the previous analyses we used the plot data to fit a joint regression model. In order to reduce the computational burden, it may be useful to split the analyses in two-steps:

1. we analyse the plot data, to retrieve the means for the 'genotype by environment' combinations;
2. we fit the joint regression model to those means.

The results of the two approaches are not necessarily the same, as some information in the first step is lost in the second. Several weighing schemes have been proposed to make two-steps fitting more reliable (Möhring and Piepho, 2009); in this example, I will show an unweighted two-steps analyses, which is simple, but not necessarily the best way to go.

A model for the second step is:

$$y_{ij} = \mu_i + \beta_i e_j + f_{ij} \quad \text{trm} \{ \text{(Equation 2)} \}$$

where the residual random component ( $f_{ij}$ ) is assumed as normally distributed, with mean equal to zero and variance equal to  $\sigma^2_f$ . In general,  $\sigma^2_f > \sigma^2_d$ , as the residual sum of squares from Model 2 also contains a component for within trial errors. Indeed, for a balanced experiment, it is:

$$\sigma^2_f = \sigma^2_d + \frac{\sigma^2_k}{k}$$

where  $\sigma^2$  is the within-trial error, which needs to be obtained from the first step. In the previous analyses we have already fitted an anova model to the whole dataset ('mod.aov'). In the box below, we make use of the 'emmeans' package to retrieve the least squares means for the seven genotypes in all locations.

Subsequently, the environmental means are calculated and centered, by subtracting the overall mean.

```
library(emmeans)
muGE <- as.data.frame( emmeans(mod.aov, ~Var:Loc) )[,1:3]
names(muGE) <- c("Var", "Loc", "Yield")
muGE <- muGE %>%
  group_by(Loc) %>%
  mutate(ej = mean(Yield) - mean(muGE$Yield))
```

Now, we fit Equation 2 to the means. In the code below we assume homoscedasticity and, thus, we are fitting the Finlay-Wilkinson model. The variance component  $\sigma^2_d$  is obtained by subtracting a fraction of the residual variance from the first step.

```
# Finlay-Wilkinson model
modFinlay <- lm(Yield ~ Var/ej - 1, data=muGE)
summary(modFinlay)
##
## Call:
## lm(formula = Yield ~ Var/ej - 1, data = muGE)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -7.3981 -3.5314 -0.8864  3.7791 11.2045
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## VarA          63.1667     2.3915  26.413 < 2e-16 ***
## VarB          66.1667     2.3915  27.668 < 2e-16 ***
## VarC          31.8333     2.3915  13.311 1.24e-13 ***
## VarD          47.1111     2.3915  19.699 < 2e-16 ***
## VarE          44.7222     2.3915  18.701 < 2e-16 ***
## VarF          34.2778     2.3915  14.333 2.02e-14 ***
## VarG          35.8889     2.3915  15.007 6.45e-15 ***
## VarA:ej         3.2250     0.6231   5.176 1.72e-05 ***
## VarB:ej         4.7936     0.6231   7.693 2.22e-08 ***
## VarC:ej         0.4771     0.6231   0.766 0.450272
## VarD:ej         0.3653     0.6231   0.586 0.562398
## VarE:ej         1.2370     0.6231   1.985 0.056998 .
## VarF:ej        -2.4317     0.6231  -3.902 0.000545 ***
## VarG:ej        -0.6663     0.6231  -1.069 0.294052
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 5.858 on 28 degrees of freedom
## Multiple R-squared:  0.9905, Adjusted R-squared:  0.9857
## F-statistic: 208.3 on 14 and 28 DF,  p-value: < 2.2e-16
sigmaf <- summary(modFinlay)$sigma^2
sigma2 <- summary(mod.aov)$sigma^2
sigmaf - sigma2/3 #sigma2_d
## [1] 27.43169
```

In the box below, we allow for different variances for each genotype and, therefore, we fit the Eberarth-Russel model. As before, we can retrieve the variance components  $\sigma^2_{d(i)}$  from the fitted model object, by subtracting the within-trial error obtained in the first step.

```
# Eberarth-Russel model
modEberarth <- gls(Yield ~ Var/ej - 1,
  weights=varIdent(form=~1|Var), data=muGE)
coefs <- summary(modEberarth)$tTable
```

```

coefs
##              Value Std.Error    t-value      p-value
## VarA      63.1666667  3.0434527  20.7549360  1.531581e-18
## VarB      66.1666667  2.7653537  23.9270177  3.508778e-20
## VarC      31.8333333  1.7165377  18.5450822  2.912238e-17
## VarD      47.1111111  2.3344802  20.1805574  3.204306e-18
## VarE      44.7222222  2.3899219  18.7128381  2.304763e-17
## VarF      34.2777778  1.9783684  17.3262868  1.685683e-16
## VarG      35.8888889  2.2589244  15.8876005  1.537133e-15
## VarA:ej    3.2249875  0.7929862   4.0668898  3.511248e-04
## VarB:ej    4.7936139  0.7205262   6.6529352  3.218756e-07
## VarC:ej    0.4771074  0.4472521   1.0667527  2.951955e-01
## VarD:ej    0.3653064  0.6082600   0.6005761  5.529531e-01
## VarE:ej    1.2369950  0.6227056   1.9864844  5.684599e-02
## VarF:ej   -2.4316943  0.5154734  -4.7174004  6.004832e-05
## VarG:ej   -0.6663160  0.5885736  -1.1320862  2.672006e-01
sigma <- summary(modEberarth)$sigma
sigma2fi <- (c(1, coef(modEberarth$modelStruct$varStruct, uncons = FALSE)) *
sigma)^2
names(sigma2fi)[1] <- "A"
sigma2fi - sigma2/3 #sigma2_di
##           A           B           C           D           E           F           G
## 48.69203 38.99949 10.79541 25.81519 27.38676 16.60005 23.73284

```

Fitting in two steps we obtain the very same result as with fitting in one step, but it ain't necessarily so.

I would like to conclude by saying that a joint regression model, the way I have fitted it here, is simple and intuitively appealing, although it has been criticized for a number of reasons. In particular, it has been noted that the environmental indices  $\{e_j\}$  are estimated from the observed data and, therefore, they are not precisely known. On the contrary, linear regression makes the assumption that the levels of explanatory variables are precisely known and not sampled. As the consequence, our estimates of the slopes  $\{\beta_i\}$  may be biased. Furthermore, in our construction we have put some arbitrary constraints on the environmental indices ( $\sum e_j = 0$ ) and on the regression slopes ( $\sum \beta_i / G = 1$ ); where G is the number of genotypes), which are not necessarily reasonable.