

15. ANOVA for Balanced Split-Plot Experiments

A Traditional Split-Plot Experiment

Field

*Whole Plot
or Main Plot*

↓

Block 1	Genotype C				Genotype A				Genotype B			
	0	100	150	50	50	100	150	0	150	100	50	0
Block 2	Genotype B				Genotype A				Genotype C			
	150	100	50	0	0	50	150	100	100	50	150	0
Block 3	Genotype A				Genotype B				Genotype C			
	100	50	0	150	0	100	150	50	50	100	150	0
Block 4	Genotype B				Genotype C				Genotype A			
	0	50	100	150	150	100	50	0	50	150	100	0

*Split Plot
or
Sub Plot*

A Model for Data from the Traditional Split-Plot Experiment

Genotype $i = 1, 2, 3$, Fertilizer $j = 1, 2, 3, 4$, Block $k = 1, 2, 3, 4$

$$y_{ijk} = \mu_{ij} + b_k + w_{ik} + e_{ijk}$$

μ_{ij} = mean for Genotype i , Fertilizer j

b_k = random block effect

w_{ik} = random whole-plot exp. unit effect

e_{ijk} = random error = random split-plot exp. unit effect

Table of Means and Marginal Means

Genotype	Fertilizer Amount				
	0	50	100	150	
A	μ_{11}	μ_{12}	μ_{13}	μ_{14}	$\bar{\mu}_{1.}$
B	μ_{21}	μ_{22}	μ_{23}	μ_{24}	$\bar{\mu}_{2.}$
C	μ_{31}	μ_{32}	μ_{33}	μ_{34}	$\bar{\mu}_{3.}$
	$\bar{\mu}_{.1}$	$\bar{\mu}_{.2}$	$\bar{\mu}_{.3}$	$\bar{\mu}_{.4}$	$\bar{\mu}_{..}$

Best Linear Unbiased Estimators

Because the experiment is balanced, the GLS estimator is equal to the OLS estimator for any estimable $C\beta$:

$$C\hat{\beta}_{\Sigma} = C(X'\Sigma^{-1}X)^{-1}X'\Sigma^{-1}y = C(X'X)^{-1}X'y = C\hat{\beta}.$$

Because the elements of $E(y)$ are $\{\mu_{ij} : i = 1, 2, 3; j = 1, 2, 3, 4\}$, the estimable quantities are all linear combinations of the cell means $\{\mu_{ij} : i = 1, 2, 3; j = 1, 2, 3, 4\}$.

The BLUE of $\sum_{i=1}^3 \sum_{j=1}^4 c_{ij}\mu_{ij}$ is $\sum_{i=1}^3 \sum_{j=1}^4 c_{ij}\bar{y}_{ij}$.

Table of Best Linear Unbiased Estimates

Genotype	Fertilizer Amount				
	0	50	100	150	
A	$\bar{y}_{11\cdot}$	$\bar{y}_{12\cdot}$	$\bar{y}_{13\cdot}$	$\bar{y}_{14\cdot}$	$\bar{y}_{1\cdot\cdot}$
B	$\bar{y}_{21\cdot}$	$\bar{y}_{22\cdot}$	$\bar{y}_{23\cdot}$	$\bar{y}_{24\cdot}$	$\bar{y}_{2\cdot\cdot}$
C	$\bar{y}_{31\cdot}$	$\bar{y}_{32\cdot}$	$\bar{y}_{33\cdot}$	$\bar{y}_{34\cdot}$	$\bar{y}_{3\cdot\cdot}$
	$\bar{y}_{\cdot 1}$	$\bar{y}_{\cdot 2}$	$\bar{y}_{\cdot 3}$	$\bar{y}_{\cdot 4}$	$\bar{y}_{\cdot\cdot}$

ANOVA Table for the Traditional Split-Plot Design

Source	DF	
<i>Blocks</i>	$4 - 1$	$= 3$
<i>Genotypes</i>	$3 - 1$	$= 2$
<i>Blocks</i> \times <i>Geno</i>	$(4 - 1)(3 - 1)$	$= 6$
<i>Fert</i>	$4 - 1$	$= 3$
<i>Geno</i> \times <i>Fert</i>	$(3 - 1)(4 - 1)$	$= 6$
<i>Blocks</i> \times <i>Fert</i>	$(4 - 1)(4 - 1)$	
$+Blocks$ \times <i>Geno</i> \times <i>Fert</i>	$+(4 - 1)(3 - 1)(4 - 1)$	$= 27$
<i>C.Total</i>	$48 - 1$	$= 47$

ANOVA Table for the Traditional Split-Plot Design

Source	DF		
<i>Blocks</i>	$4 - 1$	$=$	3
<i>Genotypes</i>	$3 - 1$	$=$	2
<i>Blocks</i> \times <i>Geno</i>	$(4 - 1)(3 - 1)$	$=$	6
<i>Fert</i>	$4 - 1$	$=$	3
<i>Geno</i> \times <i>Fert</i>	$(3 - 1)(4 - 1)$	$=$	6
<i>Error</i>	$3(4 - 1)(4 - 1)$	$=$	27
<i>C.Total</i>	$48 - 1$	$=$	47

Why does $SS_{Blocks \times Fert} + SS_{Blocks \times Geno \times Fert} = SS_{Error}$?

- There are no terms in our model corresponding to $Block \times Fert$ combinations; thus, there is no reason to devote a separate line of our ANOVA table to $Block \times Fert$.
- Also, it can be shown that

$$E(MS_{Blocks \times Fert}) = E(MS_{Blocks \times Geno \times Fert}) = \sigma_e^2$$

Thus, it makes sense to estimate σ_e^2 with an inverse variance weighted average of independent unbiased estimators:

For this slide only, let

1 = *Blocks* × *Fert* and 2 = *Blocks* × *Geno* × *Fert*.

For $\ell = 1, 2$, $MS_\ell \sim \frac{E(MS_\ell)}{df_\ell} \chi_{df_\ell}^2 \implies \text{Var}(MS_\ell) = 2\sigma_e^4/df_\ell$.

$$\begin{aligned} \frac{\text{Var}^{-1}(MS_1)MS_1 + \text{Var}^{-1}(MS_2)MS_2}{\text{Var}^{-1}(MS_1) + \text{Var}^{-1}(MS_2)} &= \frac{\frac{df_1}{2\sigma_e^4}MS_1 + \frac{df_2}{2\sigma_e^4}MS_2}{\frac{df_1}{2\sigma_e^4} + \frac{df_2}{2\sigma_e^4}} \\ &= \frac{df_1MS_1 + df_2MS_2}{df_1 + df_2} \\ &= \frac{SS_1 + SS_2}{df_1 + df_2} \end{aligned}$$

Thus, we combine the $Blocks \times Fert$ and $Blocks \times Geno \times Fert$ lines of the ANOVA table and label the resulting line as $Error$.

$$SS_{Blocks \times Fert} + SS_{Blocks \times Geno \times Fert} = SS_{Error}$$

$$df_{Blocks \times Fert} + df_{Blocks \times Geno \times Fert} = df_{Error}$$

$$MS_{Error} = SS_{Error} / df_{Error}$$

$$E(MS_{Error}) = \sigma_e^2$$

Now let's look at the ANOVA table and the analyses that can be done with it in more detail.

For greater generality, let

- w = the number of levels of the whole-plot treatment factor,
- s = the number of levels of the split-plot treatment factor, and
- b = the number of blocks.

ANOVA Table for the Traditional Split-Plot Design

Source	DF
<i>Blocks</i>	$b - 1$
<i>Genotypes</i>	$w - 1$
<i>Blocks</i> \times <i>Geno</i>	$(b - 1)(w - 1)$
<i>Fert</i>	$s - 1$
<i>Geno</i> \times <i>Fert</i>	$(w - 1)(s - 1)$
<i>Blocks</i> \times <i>Fert</i>	$(b - 1)(s - 1)$
<i>+Blocks</i> \times <i>Geno</i> \times <i>Fert</i>	$+(b - 1)(w - 1)(s - 1)$
<i>C.Total</i>	$bws - 1$

ANOVA Table for the Traditional Split-Plot Design

Source	DF
<i>Blocks</i>	$b - 1$
<i>Genotypes</i>	$w - 1$
<i>Blocks</i> \times <i>Geno</i>	$(b - 1)(w - 1)$
<i>Fert</i>	$s - 1$
<i>Geno</i> \times <i>Fert</i>	$(w - 1)(s - 1)$
<i>Error</i>	$w(b - 1)(s - 1)$
<i>C.Total</i>	$bws - 1$

ANOVA Table Sums of Squares

Source	Sum of Squares
<i>Block</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (\bar{y}_{\cdot\cdot k} - \bar{y}_{\dots})^2$
<i>Geno</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (\bar{y}_{i\cdot\cdot} - \bar{y}_{\dots})^2$
<i>Block</i> \times <i>Geno</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (\bar{y}_{i\cdot k} - \bar{y}_{i\cdot\cdot} - \bar{y}_{\cdot\cdot k} + \bar{y}_{\dots})^2$
<i>Fert</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (\bar{y}_{\cdot j\cdot} - \bar{y}_{\dots})^2$
<i>Geno</i> \times <i>Fert</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (\bar{y}_{ij\cdot} - \bar{y}_{i\cdot\cdot} - \bar{y}_{\cdot j\cdot} + \bar{y}_{\dots})^2$
<i>Error</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (y_{ijk} - \bar{y}_{i\cdot k} - \bar{y}_{\cdot j\cdot} + \bar{y}_{i\cdot\cdot})^2$
<i>C.Total</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (y_{ijk} - \bar{y}_{\dots})^2$

Simplified ANOVA Table Sums of Squares

Source	Sum of Squares
<i>Block</i>	$ws \sum_{k=1}^b (\bar{y}_{\cdot k} - \bar{y}_{\dots})^2$
<i>Geno</i>	$sb \sum_{i=1}^w (\bar{y}_{i \cdot} - \bar{y}_{\dots})^2$
<i>Block</i> \times <i>Geno</i>	$s \sum_{i=1}^w \sum_{k=1}^b (\bar{y}_{i \cdot k} - \bar{y}_{i \cdot} - \bar{y}_{\cdot k} + \bar{y}_{\dots})^2$
<i>Fert</i>	$wb \sum_{j=1}^s (\bar{y}_{\cdot j} - \bar{y}_{\dots})^2$
<i>Geno</i> \times <i>Fert</i>	$b \sum_{i=1}^w \sum_{j=1}^s (\bar{y}_{ij \cdot} - \bar{y}_{i \cdot} - \bar{y}_{\cdot j} + \bar{y}_{\dots})^2$
<i>Error</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (y_{ijk} - \bar{y}_{i \cdot k} - \bar{y}_{\cdot j} + \bar{y}_{\dots})^2$
<i>C.Total</i>	$\sum_{i=1}^w \sum_{j=1}^s \sum_{k=1}^b (y_{ijk} - \bar{y}_{\dots})^2$

$$\begin{aligned}
E(MS_{Geno}) &= \frac{sb}{w-1} \sum_{i=1}^w E(\bar{y}_{i..} - \bar{y}_{...})^2 \\
&= \frac{sb}{w-1} \sum_{i=1}^w E(\bar{\mu}_{i.} - \bar{\mu}_{..} + \bar{w}_{i.} - \bar{w}_{..} + \bar{e}_{i..} - \bar{e}_{...})^2 \\
&= sb \left\{ \frac{\sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}_{..})^2}{w-1} + E \left[\frac{\sum_{i=1}^w (\bar{w}_{i.} - \bar{w}_{..})^2}{w-1} \right] + E \left[\frac{\sum_{i=1}^w (\bar{e}_{i..} - \bar{e}_{...})^2}{w-1} \right] \right\} \\
&= sb \frac{\sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}_{..})^2}{w-1} + sb \frac{\sigma_w^2}{b} + sb \frac{\sigma_e^2}{sb} \\
&= sb \frac{\sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}_{..})^2}{w-1} + s\sigma_w^2 + \sigma_e^2
\end{aligned}$$

$$\begin{aligned}
E(MS_{Block \times Geno}) &= \frac{s}{(w-1)(b-1)} \sum_{i=1}^w \sum_{k=1}^b E(\bar{y}_{i \cdot k} - \bar{y}_{i \cdot} - \bar{y}_{\cdot k} + \bar{y}_{\cdot \cdot})^2 \\
&= \frac{s}{(w-1)(b-1)} \sum_{i=1}^w \sum_{k=1}^b E(w_{ik} - \bar{w}_{i \cdot} - \bar{w}_{\cdot k} + \bar{w}_{\cdot \cdot} + \bar{e}_{i \cdot k} - \bar{e}_{i \cdot} - \bar{e}_{\cdot k} + \bar{e}_{\cdot \cdot})^2 \\
&= \frac{s}{(w-1)(b-1)} E \left[\sum_{i=1}^w \sum_{k=1}^b (w_{ik} - \bar{w}_{i \cdot})^2 - 2 \sum_{i=1}^w \sum_{k=1}^b (w_{ik} - \bar{w}_{i \cdot})(\bar{w}_{\cdot k} - \bar{w}_{\cdot \cdot}) \right. \\
&\quad \left. + \sum_{i=1}^w \sum_{k=1}^b (\bar{w}_{\cdot k} - \bar{w}_{\cdot \cdot})^2 + e^2 \text{ sum} \right] \\
&= \frac{s}{(w-1)(b-1)} E \left[\sum_{i=1}^w \sum_{k=1}^b (w_{ik} - \bar{w}_{i \cdot})^2 - w \sum_{k=1}^b (\bar{w}_{\cdot k} - \bar{w}_{\cdot \cdot})^2 + e^2 \text{ sum} \right] \\
&= \frac{s}{(w-1)(b-1)} [w(b-1)\sigma_w^2 - w(b-1)\sigma_w^2/w + E(e^2 \text{ sum})]
\end{aligned}$$

It can be shown that

$$\begin{aligned} E(e^2 \text{ sum}) &= E \left[\sum_{i=1}^w \sum_{k=1}^b (\bar{e}_{i \cdot k} - \bar{e}_{i \cdot} - \bar{e}_{\cdot k} + \bar{e}_{\dots})^2 \right] \\ &= \frac{(w-1)(b-1)}{s} \sigma_e^2. \end{aligned}$$

Putting it all together yields

$$E(MS_{Block \times Geno}) = s\sigma_w^2 + \sigma_e^2.$$

Source	Expected Mean Squares
<i>Block</i>	
<i>Geno</i>	$s\sigma_w^2 + \sigma_e^2 + \frac{sb}{w-1} \sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}_{..})^2$
<i>Block</i> \times <i>Geno</i>	$s\sigma_w^2 + \sigma_e^2$
<i>Fert</i>	
<i>Geno</i> \times <i>Fert</i>	
<i>Error</i>	

The Test for Whole-Plot Factor Main Effects

To test for genotype main effects, i.e.,

$$H_0 : \bar{\mu}_{1.} = \cdots = \bar{\mu}_{w.} \iff H_0 : \frac{sb}{w-1} \sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}_{..})^2 = 0,$$

compare $\frac{MS_{Geno}}{MS_{Block \times Geno}}$ to a central F distribution with $w - 1$ and $(w - 1)(b - 1)$ degrees of freedom.

Comparison of Whole-Plot Factor Marginal Means

The BLUE of $\bar{\mu}_{1.} - \bar{\mu}_{2.}$ is $\bar{y}_{1..} - \bar{y}_{2..}$.

$$\text{Var}(\bar{y}_{1..} - \bar{y}_{2..}) = \text{Var}(\bar{\mu}_{1.} - \bar{\mu}_{2.} + \bar{w}_{1.} - \bar{w}_{2.} + \bar{e}_{1..} - \bar{e}_{2..})$$

$$= \frac{2\sigma_w^2}{b} + \frac{2\sigma_e^2}{sb}$$

$$= \frac{2}{sb} (s\sigma_w^2 + \sigma_e^2) = \frac{2}{sb} E(MS_{Block \times Geno})$$

$$\widehat{\text{Var}}(\bar{y}_{1..} - \bar{y}_{2..}) = \frac{2}{sb} MS_{Block \times Geno}$$

We can use

$$t = \frac{\bar{y}_{1..} - \bar{y}_{2..} - (\bar{\mu}_{1.} - \bar{\mu}_{2.})}{\sqrt{\frac{2}{sb} MS_{Block \times Geno}}} \sim t_{(w-1)(b-1)}$$

to get tests of $H_0 : \bar{\mu}_{1.} = \bar{\mu}_{2.}$

or construct confidence intervals for $\bar{\mu}_{1.} - \bar{\mu}_{2.}$.

Furthermore, suppose C is a matrix whose rows are contrast vectors so that $C\mathbf{1} = \mathbf{0}$. Then

$$\begin{aligned} \text{Var} \left(\mathbf{C} \begin{bmatrix} \bar{y}_{1..} \\ \vdots \\ \bar{y}_{w..} \end{bmatrix} \right) &= \text{Var} \left(\mathbf{C} \begin{bmatrix} \bar{b}_{.} + \bar{w}_{1.} + \bar{e}_{1..} \\ \vdots \\ \bar{b}_{.} + \bar{w}_{w.} + \bar{e}_{w..} \end{bmatrix} \right) \\ &= \text{Var} \left(\mathbf{C}\mathbf{1}\bar{b}_{.} + \mathbf{C} \begin{bmatrix} \bar{w}_{1.} + \bar{e}_{1..} \\ \vdots \\ \bar{w}_{w.} + \bar{e}_{w..} \end{bmatrix} \right) = \mathbf{C} \text{Var} \left(\begin{bmatrix} \bar{w}_{1.} + \bar{e}_{1..} \\ \vdots \\ \bar{w}_{w.} + \bar{e}_{w..} \end{bmatrix} \right) \mathbf{C}' \\ &= \mathbf{C} \left(\frac{\sigma_w^2}{b} + \frac{\sigma_e^2}{sb} \right) \mathbf{I}\mathbf{C}' = \left(\frac{\sigma_w^2}{b} + \frac{\sigma_e^2}{sb} \right) \mathbf{C}\mathbf{C}' = \frac{E(MS_{Block \times Geno})}{sb} \mathbf{C}\mathbf{C}' \end{aligned}$$

An F statistic, with q and $(w - 1)(b - 1)$ degrees of freedom, for testing

$$H_0 : \mathbf{C} \begin{bmatrix} \bar{\mu}_{1\cdot} \\ \vdots \\ \bar{\mu}_{w\cdot} \end{bmatrix} = \mathbf{0}, \text{ is}$$

$$F = \frac{\left(\mathbf{C} \begin{bmatrix} \bar{y}_{1\cdot} \\ \vdots \\ \bar{y}_{w\cdot} \end{bmatrix} \right)' \left[\frac{MS_{Block \times Geno}}{sb} \mathbf{C} \mathbf{C}' \right]^{-1} \left(\mathbf{C} \begin{bmatrix} \bar{y}_{1\cdot\cdot} \\ \vdots \\ \bar{y}_{w\cdot\cdot} \end{bmatrix} \right)}{q},$$

where q is the number of rows of \mathbf{C} (which must have full row rank to ensure that the hypothesis is testable).

Inference for the Split-Plot Factor

$$\begin{aligned} E(MS_{Fert}) &= \frac{wb}{s-1} \sum_{j=1}^s E(\bar{y}_{.j} - \bar{y}_{...})^2 \\ &= \frac{wb}{s-1} \sum_{j=1}^s E(\bar{\mu}_{.j} - \bar{\mu}_{..} + \bar{e}_{.j} - \bar{e}_{...})^2 \\ &= \frac{wb}{s-1} \sum_{j=1}^s (\bar{\mu}_{.j} - \bar{\mu}_{..})^2 + \sigma_e^2. \end{aligned}$$

Likewise, it can be shown that

$$E(MS_{Error}) = \sigma_e^2.$$

Source	Expected Mean Squares
<i>Block</i>	
<i>Geno</i>	$s\sigma_w^2 + \sigma_e^2 + \frac{sb}{w-1} \sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}_{..})^2$
<i>Block</i> \times <i>Geno</i>	$s\sigma_w^2 + \sigma_e^2$
<i>Fert</i>	$\sigma_e^2 + \frac{wb}{s-1} \sum_{j=1}^s (\bar{\mu}_{.j} - \bar{\mu}_{..})^2$
<i>Geno</i> \times <i>Fert</i>	
<i>Error</i>	σ_e^2

The Test for Split-Plot Factor Main Effects

To test for fertilizer main effects, i.e.,

$$H_0 : \bar{\mu}_{.1} = \cdots = \bar{\mu}_{.s} \iff H_0 : \frac{wb}{s-1} \sum_{j=1}^s (\bar{\mu}_{.j} - \bar{\mu}_{..})^2 = 0,$$

compare $\frac{MS_{Fert}}{MS_{Error}}$ to a central F distribution with $s - 1$ and $w(s - 1)(b - 1)$ degrees of freedom.

Comparison of Split-Plot Factor Marginal Means

The BLUE of $\bar{\mu}_{.1} - \bar{\mu}_{.2}$ is $\bar{y}_{.1.} - \bar{y}_{.2.}$.

$$\bar{y}_{.1.} - \bar{y}_{.2.} = (\bar{\mu}_{.1} + \bar{b}_{.} + \bar{w}_{..} + \bar{e}_{.1.}) - (\bar{\mu}_{.2} + \bar{b}_{.} + \bar{w}_{..} + \bar{e}_{.2.})$$

$$\text{Var}(\bar{y}_{.1.} - \bar{y}_{.2.}) = \text{Var}(\bar{\mu}_{.1} - \bar{\mu}_{.2} + \bar{e}_{.1.} - \bar{e}_{.2.})$$

$$= \frac{2}{wb} \sigma_e^2 = \frac{2}{wb} E(MS_{Error})$$

$$\widehat{\text{Var}}(\bar{y}_{.1.} - \bar{y}_{.2.}) = \frac{2}{wb} MS_{Error}$$

We can use

$$t = \frac{\bar{y}_{\cdot 1} - \bar{y}_{\cdot 2} - (\bar{\mu}_{\cdot 1} - \bar{\mu}_{\cdot 2})}{\sqrt{\frac{2}{wb}MS_{Error}}} \sim t_{w(s-1)(b-1)}$$

to get tests of $H_0 : \bar{\mu}_{\cdot 1} = \bar{\mu}_{\cdot 2}$

or to construct confidence intervals for $\bar{\mu}_{\cdot 1} - \bar{\mu}_{\cdot 2}$.

Furthermore, suppose \mathbf{C} is a matrix with rows that are contrast vectors so that $\mathbf{C}\mathbf{1} = \mathbf{0}$. Then

$$\begin{aligned} \text{Var} \left(\mathbf{C} \begin{bmatrix} \bar{y}_{.1.} \\ \vdots \\ \bar{y}_{.s.} \end{bmatrix} \right) &= \text{Var} \left(\mathbf{C} \begin{bmatrix} \bar{b}_{.} + \bar{w}_{..} + \bar{e}_{.1.} \\ \vdots \\ \bar{b}_{.} + \bar{w}_{..} + \bar{e}_{.s.} \end{bmatrix} \right) \\ &= \text{Var} \left(\mathbf{C}\mathbf{1}\bar{b}_{.} + \mathbf{C}\mathbf{1}\bar{w}_{..} + \mathbf{C} \begin{bmatrix} \bar{e}_{.1.} \\ \vdots \\ \bar{e}_{.s.} \end{bmatrix} \right) = \mathbf{C} \text{Var} \left(\begin{bmatrix} \bar{e}_{.1.} \\ \vdots \\ \bar{e}_{.s.} \end{bmatrix} \right) \mathbf{C}' \\ &= \mathbf{C} \left(\frac{\sigma_e^2}{wb} \right) \mathbf{I} \mathbf{C}' = \frac{E(MS_{Error})}{wb} \mathbf{C} \mathbf{C}' \end{aligned}$$

An F statistic, with q and $w(s - 1)(b - 1)$ degrees of freedom, for testing

$$H_0 : \mathbf{C} \begin{bmatrix} \bar{\mu}_{.1} \\ \vdots \\ \bar{\mu}_{.s} \end{bmatrix} = \mathbf{0}, \text{ is}$$

$$F = \frac{\left(\mathbf{C} \begin{bmatrix} \bar{y}_{.1.} \\ \vdots \\ \bar{y}_{.s.} \end{bmatrix} \right)' \left[\frac{MS_{Error}}{wb} \mathbf{C}\mathbf{C}' \right]^{-1} \left(\mathbf{C} \begin{bmatrix} \bar{y}_{.1.} \\ \vdots \\ \bar{y}_{.s.} \end{bmatrix} \right)}{q}$$

where q is the number of rows of \mathbf{C} (which must have full row rank to ensure that the hypothesis is testable).

Inference for Interactions

$$\begin{aligned} E(MS_{Geno \times Fert}) &= \frac{b}{(w-1)(s-1)} \sum_{i=1}^w \sum_{j=1}^s E(\bar{y}_{ij.} - \bar{y}_{i..} - \bar{y}_{.j.} + \bar{y}_{...})^2 \\ &= \frac{b}{(w-1)(s-1)} \sum_{i=1}^w \sum_{j=1}^s E(\mu_{ij} - \bar{\mu}_{i.} - \bar{\mu}_{.j} + \bar{\mu}_{..} + \bar{e}_{ij.} - \bar{e}_{i..} - \bar{e}_{.j.} + \bar{e}_{...})^2 \\ &= \dots \\ &= \frac{b}{(w-1)(s-1)} \sum_{i=1}^w \sum_{j=1}^s (\mu_{ij} - \bar{\mu}_{i.} - \bar{\mu}_{.j} + \bar{\mu}_{..})^2 + \sigma_e^2. \end{aligned}$$

It can be shown that

$$\mu_{ij} - \bar{\mu}_{i.} - \bar{\mu}_{.j} + \bar{\mu}_{..} = 0 \quad \forall i, j$$

is equivalent to

$$\mu_{ij} - \mu_{ij^*} - \mu_{i^*j} + \mu_{i^*j^*} = 0 \quad \forall i \neq i^*, j \neq j^*.$$

Thus,

$$\frac{b}{(w-1)(s-1)} \sum_{i=1}^w \sum_{j=1}^s (\mu_{ij} - \bar{\mu}_{i.} - \bar{\mu}_{.j} + \bar{\mu}_{..})^2 = 0$$

is equivalent to no interactions between genotypes and fertilizers.

Source	Expected Mean Squares
<i>Block</i>	
<i>Geno</i>	$s\sigma_w^2 + \sigma_e^2 + \frac{sb}{w-1} \sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}_{..})^2$
<i>Block</i> \times <i>Geno</i>	$s\sigma_w^2 + \sigma_e^2$
<i>Fert</i>	$\sigma_e^2 + \frac{wb}{s-1} \sum_{j=1}^s (\bar{\mu}_{.j} - \bar{\mu}_{..})^2$
<i>Geno</i> \times <i>Fert</i>	$\sigma_e^2 + \frac{b}{(w-1)(s-1)} \sum_{i=1}^w \sum_{j=1}^s (\mu_{ij} - \bar{\mu}_{i.} - \bar{\mu}_{.j} + \bar{\mu}_{..})^2$
<i>Error</i>	σ_e^2

The Test for Whole \times Split Interaction Effects

To test for genotype \times fertilizer interaction effects, i.e.,

$$H_0 : \mu_{ij} - \bar{\mu}_{i.} - \bar{\mu}_{.j} + \bar{\mu}_{..} = 0 \quad \forall i, j \iff$$

$$H_0 : \frac{b}{(w-1)(s-1)} \sum_{i=1}^w \sum_{j=1}^s (\mu_{ij} - \bar{\mu}_{i.} - \bar{\mu}_{.j} + \bar{\mu}_{..})^2 = 0,$$

compare $\frac{MS_{Geno \times Fert}}{MS_{Error}}$ to a central F distribution with $(w-1)(s-1)$ and $w(s-1)(b-1)$ degrees of freedom.

Inference for Simple Effects

Consider the difference between two fertilizer means within a genotype, e.g., $\mu_{11} - \mu_{12}$ whose BLUE is $\bar{y}_{11\cdot} - \bar{y}_{12\cdot}$.

$$\begin{aligned}\text{Var}(\bar{y}_{11\cdot} - \bar{y}_{12\cdot}) &= \text{Var}(\mu_{11} - \mu_{12} + \bar{b}\cdot - \bar{b}\cdot + \bar{w}_{1\cdot} - \bar{w}_{1\cdot} + \bar{e}_{11\cdot} - \bar{e}_{12\cdot}) \\ &= \frac{2}{b}\sigma_e^2\end{aligned}$$

$$\widehat{\text{Var}}(\bar{y}_{11\cdot} - \bar{y}_{12\cdot}) = \frac{2}{b}MS_{Error}$$

We can use

$$t = \frac{\bar{y}_{11\cdot} - \bar{y}_{12\cdot} - (\mu_{11} - \mu_{12})}{\sqrt{\frac{2}{b}MS_{Error}}} \sim t_{w(s-1)(b-1)}$$

to get tests of $H_0 : \mu_{11} = \mu_{12}$

or construct confidence intervals for $\mu_{11} - \mu_{12}$.

Now consider the difference between two genotype means within a fertilizer, e.g., $\mu_{11} - \mu_{21}$ whose BLUE is $\bar{y}_{11\cdot} - \bar{y}_{21\cdot}$.

$$\begin{aligned}\text{Var}(\bar{y}_{11\cdot} - \bar{y}_{21\cdot}) &= \text{Var}(\mu_{11} - \mu_{21} + \bar{w}_{1\cdot} - \bar{w}_{2\cdot} + \bar{e}_{11\cdot} - \bar{e}_{21\cdot}) \\ &= \frac{2\sigma_w^2}{b} + \frac{2\sigma_e^2}{b} \\ &= \frac{2}{b}(\sigma_w^2 + \sigma_e^2).\end{aligned}$$

This variance is not a constant times any expected mean square from our ANOVA table.

We need an estimator of $\sigma_w^2 + \sigma_e^2$. We have

$$E(MS_{Block \times Geno}) = s\sigma_w^2 + \sigma_e^2, \quad E(MS_{Error}) = \sigma_e^2, \quad \text{and}$$

$$\begin{aligned} E\left(\frac{1}{s}MS_{Block \times Geno} + \frac{s-1}{s}MS_{Error}\right) &= \sigma_w^2 + \frac{\sigma_e^2}{s} + \frac{(s-1)\sigma_e^2}{s} \\ &= \sigma_w^2 + \sigma_e^2. \end{aligned}$$

Thus,

$$\frac{1}{s}MS_{Block \times Geno} + \frac{s-1}{s}MS_{Error}$$

is an unbiased estimator of $\sigma_w^2 + \sigma_e^2$.

It follows that

$$\widehat{\text{Var}}(\bar{y}_{11\cdot} - \bar{y}_{21\cdot}) \equiv \frac{2}{sb} MS_{Block \times Geno} + \frac{2(s-1)}{sb} MS_{Error}$$

is an unbiased estimator of $\text{Var}(\bar{y}_{11\cdot} - \bar{y}_{21\cdot})$.

We can use

$$\frac{\bar{y}_{11\cdot} - \bar{y}_{21\cdot} - (\mu_{11} - \mu_{21})}{\sqrt{\widehat{\text{Var}}(\bar{y}_{11\cdot} - \bar{y}_{21\cdot})}} \sim t_d, \text{ with } d \text{ degrees of freedom}$$

computed by Cochran-Satterthwaite to get approximate tests of $H_0 : \mu_{11} = \mu_{21}$ or to construct approximate confidence intervals for $\mu_{11} - \mu_{21}$.

Full Table of Expected Mean Squares

Source	Expected Mean Squares
<i>Block</i>	$ws\sigma_b^2 + s\sigma_w^2 + \sigma_e^2$
<i>Geno</i>	$s\sigma_w^2 + \sigma_e^2 + \frac{sb}{w-1} \sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}_{..})^2$
<i>Block</i> \times <i>Geno</i>	$s\sigma_w^2 + \sigma_e^2$
<i>Fert</i>	$\sigma_e^2 + \frac{wb}{s-1} \sum_{j=1}^s (\bar{\mu}_{.j} - \bar{\mu}_{..})^2$
<i>Geno</i> \times <i>Fert</i>	$\sigma_e^2 + \frac{b}{(w-1)(s-1)} \sum_{i=1}^w \sum_{j=1}^s (\mu_{ij} - \bar{\mu}_{i.} - \bar{\mu}_{.j} + \bar{\mu}_{..})^2$
<i>Error</i>	σ_e^2

Inferences for Cell Mean μ_{ij}

$$\begin{aligned}\text{Var}(\bar{y}_{ij.}) &= \text{Var}(\mu_{ij} + \bar{b}. + \bar{w}_i. + \bar{e}_{ij.}) \\ &= \frac{\sigma_b^2}{b} + \frac{\sigma_w^2}{b} + \frac{\sigma_e^2}{b}\end{aligned}$$

We can construct the unbiased estimator

$$\widehat{\text{Var}}(\bar{y}_{ij.}) = \frac{1}{wbs} [MS_{Block} + (w - 1) MS_{Block \times Geno} + w(s - 1) MS_{Error}]$$

with approximate degrees of freedom from Cochran-Satterthwaite.

Inferences for Whole-Plot-Factor Means $\bar{\mu}_i$.

$$\begin{aligned}\text{Var}(\bar{y}_{i..}) &= \text{Var}(\bar{\mu}_i + \bar{b} + \bar{w}_i + \bar{e}_{i..}) \\ &= \frac{\sigma_b^2}{b} + \frac{\sigma_w^2}{b} + \frac{\sigma_e^2}{sb}\end{aligned}$$

This can be estimated with a linear combination of mean squares.

If block effects are considered fixed rather than random,

$$\begin{aligned}\text{Var}(\bar{y}_{i..}) &= \text{Var}(\bar{\mu}_{i.} + \bar{b}. + \bar{w}_{i.} + \bar{e}_{i..}) \\ &= \frac{\sigma_w^2}{b} + \frac{\sigma_e^2}{sb} \\ &= \frac{1}{sb} (s\sigma_w^2 + \sigma_e^2)\end{aligned}$$

We can estimate this variance by $\frac{1}{sb}MS_{Block \times Geno}$ with $(w - 1)(b - 1)$ degrees of freedom.

Inferences for Split-Plot-Factor Means $\bar{\mu}_{.j}$

If block effects are considered random,

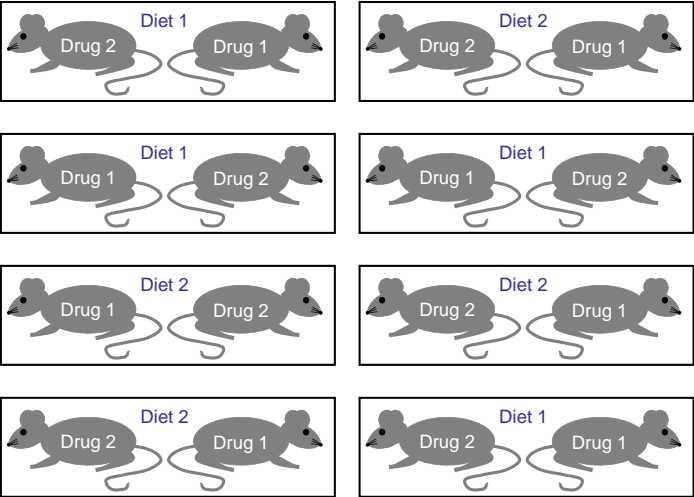
$$\begin{aligned}\text{Var}(\bar{y}_{.j}) &= \text{Var}(\bar{\mu}_{.j} + \bar{b}_{.} + \bar{w}_{..} + \bar{e}_{.j}) \\ &= \frac{\sigma_b^2}{b} + \frac{\sigma_w^2}{wb} + \frac{\sigma_e^2}{wb}\end{aligned}$$

If block effects are considered fixed,

$$\begin{aligned}\text{Var}(\bar{y}_{.j}) &= \text{Var}(\bar{\mu}_{.j} + \bar{b}_{.} + \bar{w}_{..} + \bar{e}_{.j}) \\ &= \frac{\sigma_w^2}{wb} + \frac{\sigma_e^2}{wb}.\end{aligned}$$

Both can be estimated by linear combinations of mean squares.

Diet and Drug Split-Plot Experiment



Linear Mixed-Effects Model

Diet $i = 1, 2$, Drug $j = 1, 2$, Litter $k = 1, 2, 3, 4$ (within each Diet i)

$$y_{ijk} = \mu + \alpha_i + \beta_j + \gamma_{ij} + \ell_{ik} + e_{ijk} \quad (i = 1, 2; j = 1, 2; k = 1, \dots, 4)$$

$\mu + \alpha_i + \beta_j + \gamma_{ij}$ = mean for Diet i and Drug j

ℓ_{ik} = random litter effect = whole-plot exp. unit random effect

e_{ijk} = random error effect = split-plot exp. unit random effect

$\ell_{ik} \stackrel{iid}{\sim} N(0, \sigma_\ell^2)$ independent of $e_{ijk} \stackrel{iid}{\sim} N(0, \sigma_e^2)$

Start of the ANOVA Table

With $v = \#$ of whole-plot experimental units per level of the whole-plot factor...

Source	DF
<i>Diet</i>	$w - 1$
<i>Litter(Diet)</i>	$(v - 1)w$
<i>Drug</i>	$s - 1$
<i>Diet</i> \times <i>Drug</i>	$(w - 1)(s - 1)$
<i>Drug</i> \times <i>Litter(Diet)</i>	$(s - 1)(v - 1)w$
<i>C.Total</i>	$wvs - 1$

Start of the ANOVA Table

With $v = \#$ of whole-plot experimental units per level of the whole-plot factor...

Source	DF
<i>Diet</i>	$w - 1$
<i>Whole Plot Error</i>	$(v - 1)w$
<i>Drug</i>	$s - 1$
<i>Diet \times Drug</i>	$(w - 1)(s - 1)$
<i>Split Plot Error</i>	$(s - 1)(v - 1)w$
<i>C.Total</i>	$wvs - 1$

Completing the ANOVA Table and Additional Analyses

- The techniques used in the previous slides can be applied to find expressions for the sums of squares, mean squares, expected mean squares, and F statistics.
- Because of balance, \bar{y}_{ij} is the BLUE of $\mu + \alpha_i + \beta_j + \gamma_{ij}$.
- The reasoning in the previous slides can be used to determine appropriate standard errors, tests, and confidence intervals.

Summary of ANOVA for a Balanced Split-Plot

- Use whole-plot-error mean square for inferences on
 - contrasts among whole-plot-factor marginal means
- Use split-plot-error mean square for inferences on
 - contrasts among split-plot-factor marginal means
 - whole \times split interactions
 - comparison of split-plot levels within a whole-plot level
- Construct a linear combination of mean squares for inferences on
 - comparison of whole-plot levels within a split-plot level
 - a comparison within neither whole-plot nor split-plot levels (e.g.,
 $\mu_{11} - \mu_{22}$)
 - most means