

15. ANOVA for Balanced Split-Plot Experiments

A Traditional Split-Plot Experiment

Block x fert

| | | Field | | | | | | | | Whole Plot or Main Plot | | | |
|-------|------|------------|-----|-----|-----|------------|-----|-----|-----|----------------------------|-----|-----|---|
| | | Genotype C | | | | Genotype A | | | | Genotype B | | | |
| Block | Plot | 0 | 100 | 150 | 50 | 50 | 100 | 150 | 0 | 150 | 100 | 50 | 0 |
| | | 150 | 100 | 50 | 0 | 0 | 50 | 150 | 100 | 100 | 50 | 150 | 0 |
| Block | Plot | Genotype A | | | | Genotype B | | | | Genotype C | | | |
| | | 100 | 50 | 0 | 150 | 0 | 100 | 150 | 50 | 50 | 100 | 150 | 0 |
| Block | Plot | 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 $k = 1, 2, 3, 4$

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

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 | | | | $\bar{\mu}_{..}$ |
|----------|-------------------|------------------|------------------|------------------|-------------------|
| | 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} | $\bar{\mu}_{3..}$ |
| | $\bar{\mu}_{.1}$ | $\bar{\mu}_{.2}$ | $\bar{\mu}_{.3}$ | $\bar{\mu}_{.4}$ | |

Best Linear Unbiased Estimators

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

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

Because the elements of $E(\mathbf{y})$ 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 | | | | $\bar{y}_{1..}$ |
|----------|-------------------|------------------|------------------|------------------|-----------------|
| | 0 | 50 | 100 | 150 | |
| A | $\bar{y}_{11..}$ | $\bar{y}_{12..}$ | $\bar{y}_{13..}$ | $\bar{y}_{14..}$ | $\bar{y}_{1..}$ |
| B | $\bar{y}_{21..}$ | $\bar{y}_{22..}$ | $\bar{y}_{23..}$ | $\bar{y}_{24..}$ | $\bar{y}_{2..}$ |
| C | $\bar{y}_{31..}$ | $\bar{y}_{32..}$ | $\bar{y}_{33..}$ | $\bar{y}_{34..}$ | $\bar{y}_{3..}$ |
| | $\bar{y}_{.1..}$ | $\bar{y}_{.2..}$ | $\bar{y}_{.3..}$ | $\bar{y}_{.4..}$ | $\bar{y}_{...}$ |

due to the 4
blocks we have
4 replications
for each of the 12
treatments

ANOVA Table for the Traditional Split-Plot Design

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

| Source | random | DF | looks like an RCBID |
|---|--------|------------------------|---------------------|
| Blocks | fixed | 4 - 1 | = 3 |
| Genotypes | | 3 - 1 | = 2 |
| Blocks × Geno | random | (4 - 1)(3 - 1) | = 6 |
| Fert | fixed | 4 - 1 | = 3 |
| Geno × Fert | | (3 - 1)(4 - 1) | = 6 |
| Blocks × Fert | | (4 - 1)(4 - 1) | |
| +Blocks × Geno × Fert | | +(4 - 1)(3 - 1)(4 - 1) | = 27 |
| C.Total | | 48 - 1 | = 47 |
| not part of the model \Rightarrow SSE | | | |

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

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- 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

is needed to calculate the weights in the weight average

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

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

$$\frac{\text{Var}^{-1}(MS_1)MS_1 + \text{Var}^{-1}(MS_2)MS_2}{\text{Var}^{-1}(MS_1) + \text{Var}^{-1}(MS_2)} = \frac{\frac{df_1}{2\sigma_e^4} MS_1 + \frac{df_2}{2\sigma_e^4} MS_2}{\frac{df_1}{2\sigma_e^4} + \frac{df_2}{2\sigma_e^4}}$$

$$= \frac{df_1 MS_1 + df_2 MS_2}{df_1 + df_2}$$

$$= \frac{SS_1 + SS_2}{df_1 + df_2}$$

MSE

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

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

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

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

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

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

For greater generality, let

here $w = 3$

Genotype : A, B, C

- 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. $b = 4$

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> | $bws - 1$ |

combine

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

Block : $k=1, \dots, b$

$i = 1, \dots, w$

i : genotype

y_{ijk}

$j = \text{fertilizer}$ $j = 1, \dots, s$

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

Simplified ANOVA Table Sums of Squares

$$\frac{\text{Sum of squares}}{df} = MS$$

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

$$E(MS_{Geno}) = \frac{sb}{w-1} \sum_{i=1}^w E(\bar{y}_{i..} - \bar{y}...)^2$$

~~if~~

$$\bar{y}_{i..} = \bar{\mu}_{i.} + \bar{\omega}_{i.} + \bar{e}_{i..}$$

same $\bar{y}...$

note that b_k will

cancel itself

$$= \frac{sb}{w-1} \sum_{i=1}^w E(\bar{\mu}_{i.} - \bar{\mu}.. + \bar{w}_{i.} - \bar{w}.. + \bar{e}_{i..} - \bar{e}...)^2$$

due to $E(\omega_{ik}) = E(e_{ijk}) = 0$ all crossproducts

$$= sb \left\{ \frac{\sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}..)^2}{w-1} + E \left[\frac{\sum_{i=1}^w (\bar{w}_{i.} - \bar{w}..)^2}{w-1} \right] + E \left[\frac{\sum_{i=1}^w (\bar{e}_{i..} - \bar{e}...)^2}{w-1} \right] \right\}$$

$$= \sigma_w^2 / b$$

$$= sb \frac{\sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}..)^2}{w-1} + sb \frac{\sigma_w^2}{b} + sb \frac{\sigma_e^2}{sb}$$

involving either
 $\bar{\omega}_{i.}$, $\bar{\omega}..$, $\bar{e}_{i..}$, $\bar{e}...$
 will disappear

$$= sb \frac{\sum_{i=1}^w (\bar{\mu}_{i.} - \bar{\mu}..)^2}{w-1} + s\sigma_w^2 + \sigma_e^2$$

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

It can be shown that

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

Putting it all together yields

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

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



The Test for Whole-Plot Factor Main Effects

~~end lecture 30
4-9-25~~

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

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

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 $\bar{y}_{1..} - \bar{y}_{2..}$

(write out $\bar{y}_{1..}$ & $\bar{y}_{2..}$
in terms of the model)

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

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

$$\text{Var}(\bar{w}_{i..}) = \frac{\sigma_w^2}{b}$$

$$\text{Var}(\bar{e}_{i..}) = \frac{\sigma_e^2}{sb}$$

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

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

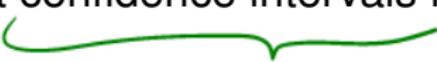
from the
output

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
 $\bar{\mu}_{1.} - \bar{\mu}_{2..}$

Practical vs.
statistical
significance


give us an estimate
of the effect size

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

$$\text{Var} \left(C \begin{bmatrix} \bar{y}_{1..} \\ \vdots \\ \bar{y}_{w..} \end{bmatrix} \right) = \text{Var} \left(C \begin{bmatrix} \bar{b}_{..} + \bar{w}_{1..} + \bar{e}_{1..} \\ \vdots \\ \bar{b}_{..} + \bar{w}_{w..} + \bar{e}_{w..} \end{bmatrix} \right)$$

$$= \text{Var} \left(\boxed{C\bar{b}_{..}} + C \begin{bmatrix} \bar{w}_{1..} + \bar{e}_{1..} \\ \vdots \\ \bar{w}_{w..} + \bar{e}_{w..} \end{bmatrix} \right) = C \text{Var} \left(\begin{bmatrix} \bar{w}_{1..} + \bar{e}_{1..} \\ \vdots \\ \bar{w}_{w..} + \bar{e}_{w..} \end{bmatrix} \right) C^\top$$

$$= C \left(\left(\frac{\sigma_w^2}{b} \right) + \left(\frac{\sigma_e^2}{sb} \right) \right) I C^\top = \boxed{\left(\frac{\sigma_w^2}{b} + \frac{\sigma_e^2}{sb} \right) C C^\top} = \frac{\text{E}(MS_{Block \times Geno})}{sb} C C^\top$$

output

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

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

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

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

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

Likewise, it can be shown that

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

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

The Test for Split-Plot Factor Main Effects

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

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

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 $\bar{y}_{.1.} - \bar{y}_{.2.}$

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

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

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

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

interpretation: averaging over
blocks and genotype,

We can use

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

the average difference
between fertilizer level 1

to get tests of $H_0 : \bar{\mu}_{\cdot 1} = \bar{\mu}_{\cdot 2}$ or to construct confidence intervals
for $\bar{\mu}_{\cdot 1} - \bar{\mu}_{\cdot 2}$.

and 2 is . . .

Furthermore, suppose C is a matrix with rows that are contrast vectors so that $\underline{C1} = \underline{0}$. Then

$$\begin{aligned} \text{Var} \left(\mathbf{C} \begin{bmatrix} \bar{y}_{.1.} \\ \vdots \\ \bar{y}_{.s.} \end{bmatrix} \right) &= \text{Var} \left(\mathbf{C} \begin{bmatrix} \bar{b}_{.} + \bar{w}_{..} + \bar{e}_{.1.} \\ \vdots \\ \bar{b}_{.} + \bar{w}_{..} + \bar{e}_{.s.} \end{bmatrix} \right) \\ &= \text{Var} \left(\mathbf{C1}\bar{b}_{.} + \mathbf{C1}\bar{w}_{..} + \mathbf{C} \begin{bmatrix} \bar{e}_{.1.} \\ \vdots \\ \bar{e}_{.s.} \end{bmatrix} \right) = \mathbf{C} \text{Var} \begin{pmatrix} \bar{e}_{.1.} \\ \vdots \\ \bar{e}_{.s.} \end{pmatrix} \mathbf{C}^{\top} \\ &= \mathbf{C} \left(\frac{\sigma_e^2}{wb} \right) \mathbf{I} \mathbf{C}^{\top} = \frac{\text{E}(MS_{Error})}{wb} \mathbf{C} \mathbf{C}^{\top} \end{aligned}$$

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

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

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

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

Inference for Interactions

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

It can be shown that

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

is equivalent to

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

Thus,

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

is equivalent to no interactions between genotypes and fertilizers.

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

The Test for Whole \times Split Interaction Effects

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

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

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

compare $\boxed{\frac{MS_{Geno \times Fert}}{MS_{Error}}}$ 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 $\bar{y}_{11\cdot} - \bar{y}_{12\cdot}$.

$$\begin{aligned}\text{Var}(\bar{y}_{11\cdot} - \bar{y}_{12\cdot}) &= \text{Var}(\mu_{11} - \mu_{12} + \cancel{\bar{b}_1} - \cancel{\bar{b}_2} + \cancel{\bar{w}_{11\cdot}} - \cancel{\bar{w}_{12\cdot}} + \cancel{\bar{e}_{11\cdot}} - \cancel{\bar{e}_{12\cdot}}) \\ &= \frac{2}{b} \sigma_e^2\end{aligned}$$

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

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 $\bar{y}_{11\cdot} - \bar{y}_{21\cdot}$.

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

need to identify
an unbiased estimator

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

identify an appropriate
linear combin.
of available MS

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

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

$$= \sigma_w^2 + \sigma_e^2.$$

Thus,

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

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

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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

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

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

Inferences for Cell Mean μ_{ij}

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

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 $\bar{\mu}_i$.

$$\text{Var}(\bar{y}_{i..}) = \text{Var}(\bar{\mu}_{i.} + \bar{b}_{.} + \bar{w}_{i.} + \bar{e}_{i..})$$

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

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 $\bar{\mu}_{\cdot j}$

If block effects are considered random,

$$\text{Var}(\bar{y}_{\cdot j}) = \text{Var}(\bar{\mu}_{\cdot j} + \bar{b}_{\cdot} + \bar{w}_{..} + \bar{e}_{\cdot j})$$

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

than $\text{Var}(\bar{y}_{\cdot j})$
when blocks
are fixed

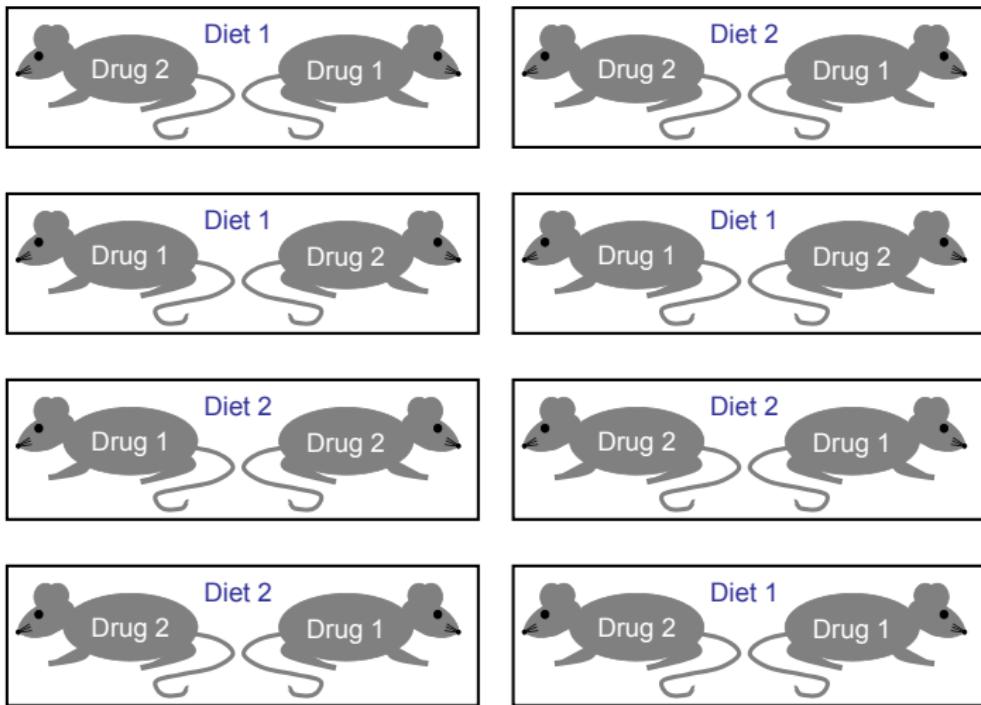
If block effects are considered fixed,

$$\text{Var}(\bar{y}_{\cdot j}) = \text{Var}(\bar{\mu}_{\cdot j} + \bar{b}_{\cdot} + \bar{w}_{..} + \bar{e}_{\cdot j})$$

$$= \frac{\sigma_w^2}{wb} + \frac{\sigma_e^2}{wb}.$$

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)

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

ℓ_{ik} random effects

$\mu + \alpha_i + \beta_j + \gamma_{ij}$ = 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

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

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