

# Experimental Design

## Note 4

### Randomized complete block design (RCBD)

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# Nuisance Factor

Nuisance Factor (may be present in experiment)

- Has effect on response but its effect is not of interest
- if unknown → Protecting experiment through randomization
- If known (measurable) but uncontrollable → Analysis of Covariance (Chapter 15 Section 3)
- If known and controllable → Blocking

## Example: Penicillin Experiment I

- In this experiment, four penicillin manufacturing processes ( $A$ ,  $B$ ,  $C$ , and  $D$ ) were being investigated. Yield was the response. It was known that an important raw material, corn steep liquor, was quite variable. The experiment and its results were given below:

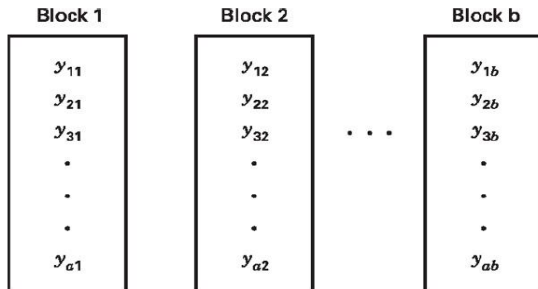
	blend 1	blend 2	blend 3	blend 4	blend 5
$A$	89 <sub>1</sub>	84 <sub>4</sub>	81 <sub>2</sub>	87 <sub>1</sub>	79 <sub>3</sub>
$B$	88 <sub>3</sub>	77 <sub>2</sub>	87 <sub>1</sub>	92 <sub>3</sub>	81 <sub>4</sub>
$C$	97 <sub>2</sub>	92 <sub>3</sub>	87 <sub>4</sub>	89 <sub>2</sub>	80 <sub>1</sub>
$D$	94 <sub>4</sub>	79 <sub>1</sub>	85 <sub>3</sub>	84 <sub>4</sub>	88 <sub>2</sub>

- Blend is a nuisance factor, treated as a block factor.

## Example: Penicillin Experiment II

- (Complete) Blocking: all the treatments are applied within each block, and they are compared within blocks.
- Advantage: Eliminate blend-to-blend (between-block) variation from experimental error variance when comparing treatments.
- Cost: degree of freedom.

# Randomized Complete Block Design (RCBD) I



- $b$  blocks each consisting of (partitioned into)  $a$  experimental units.

## Randomized Complete Block Design (RCBD) II

- $a$  treatments are randomly assigned to the experimental units within each block.
- Typically after the runs in one block have been conducted, then move to another block.
- Typical blocking factors: day, batch of raw material etc.
- Results in restriction on randomization because randomization is only within blocks.
- Data within a block are related to each other, When  $a = 2$ , randomized complete block design becomes paired two sample case.

# Statistical Model: two-way ANOVA I

- $b$  blocks and  $a$  treatments
- Statistical model is

$$y_{ij} = \mu + \tau_i + \beta_j + \epsilon_{ij}, \text{ for } i = 1, 2, \dots, a; j = 1, 2, \dots, b$$

where  $\mu$  is the grand mean,  $\tau_i$  is the  $i$ th treatment effect,  $\beta_j$  is the  $j$ th block effect, and  $\epsilon_{ij} \sim^{iid} N(0, \sigma^2)$ .

- The model is additive because within a fixed block, the block effect is fixed; for a fixed treatment, the treatment effect is fixed across blocks. In other words, blocks and treatments do not interact.
- parameter constraints:  $\sum_{i=1}^a \tau_i = 0$ ;  $\sum_{j=1}^b \beta_j = 0$

## Estimates for Parameters

- Rewrite observation  $y_{ij}$  as:

$$y_{ij} = \bar{y}_{..} + (\bar{y}_{i.} - \bar{y}_{..}) + (\bar{y}_{.j} - \bar{y}_{..}) + (y_{ij} - \bar{y}_{i.} - \bar{y}_{.j} + \bar{y}_{..})$$

- Compared with the model

$$y_{ij} = \mu + \tau_i + \beta_j + \epsilon_{ij}.$$

- The estimates for the parameters are

$$\hat{\mu} = \bar{y}_{..},$$

$$\hat{\tau}_i = \bar{y}_{i.} - \bar{y}_{..},$$

$$\hat{\beta}_j = \bar{y}_{.j} - \bar{y}_{..},$$

$$\hat{\epsilon}_{ij} = y_{ij} - \bar{y}_{i.} - \bar{y}_{.j} + \bar{y}_{..}$$



# Sum of Squares (SS) I

- Can partition  $SS_T = \sum_i \sum_j (y_{ij} - \bar{y}_{..})^2$  into

$$b \sum_i (\bar{y}_{i.} - \bar{y}_{..})^2 + a \sum_j (\bar{y}_{.j} - \bar{y}_{..})^2 + \sum_i \sum_j (y_{ij} - \bar{y}_{i.} - \bar{y}_{.j} + \bar{y}_{..})^2,$$

$$SS_{Treatment} = b \sum_i (\bar{y}_{i.} - \bar{y}_{..})^2 = b \sum_i \hat{\tau}_i^2, \quad df = a - 1,$$

$$SS_{Block} = a \sum_j (\bar{y}_{.j} - \bar{y}_{..})^2 = a \sum_j \hat{\beta}_j^2, \quad df = b - 1,$$

$$SSE = \sum_i \sum_j (y_{ij} - \bar{y}_{i.} - \bar{y}_{.j} + \bar{y}_{..})^2 = \sum_i \sum_j \hat{\epsilon}_{ij}^2, \quad df = (a - 1)(b - 1).$$

Hence,

$$SST = SS_{Treatment} + SS_{Block} + SSE$$

- The Mean Squares are  $MS_{Treatment} = SS_{Treatment}/(a - 1)$ ,  
 $MS_{Block} = SS_{Block}/(b - 1)$ , and  $MSE = SSE/((a - 1)(b - 1))$ .

# Testing Basic Hypotheses I

- $H_0 : \tau_1 = \tau_2 = \cdots = \tau_a = 0$  vs  $H_1$  : at least one is not
- Can show:

$$E(MSE) = \sigma^2$$

$$E(MS_{Treatment}) = \sigma^2 + b \sum_i \tau_i^2 / (a - 1)$$

$$E(MS_{Block}) = \sigma^2 + a \sum_j \beta_j^2 / (b - 1)$$

- Use  $F$ -test to test  $H_0$  :

$$F_0 = \frac{MS_{Treatment}}{MSE} = \frac{SS_{Treatment} / (a - 1)}{SSE / ((a - 1)(b - 1))}$$

# Testing Basic Hypotheses II

- Caution testing block effects
  - Usually not of interest.
  - Randomization is restricted: Differing opinions on  $F$ -test for testing blocking effect.
  - Can use ratio  $MS_{Block}/MSE$  to check if blocking successful.
  - Block effects can be random effects (considered fixed effects in this chapter).

# Analysis of Variance (ANOVA) Table I

Source of Variation	Sum of Squares	Degrees of Freedom	Mean Square	$F_0$
Blocks	$SS_{Block}$	$b - 1$	$MS_{Block}$	$MS_{Treatment} / MSE$
Treatment	$SS_{Treatment}$	$a - 1$	$MS_{Treatment}$	
Error	$SSE$	$(a - 1)(b - 1)$	$MSE$	
Total	$SS_T$	$ab - 1$		

$$SS_T = \sum_i \sum_j y_{ij}^2 - y_{..}^2 / N,$$

$$SS_{Treatment} = \frac{1}{b} \sum_i y_{i.}^2 - y_{..}^2 / N,$$

$$SS_{Block} = \frac{1}{a} \sum_j y_{.j}^2 - y_{..}^2 / N,$$

$$SSE = SS_T - SS_{Treatment} - SS_{Block}.$$

Decision rule: If  $F_0 > F_{\alpha, a-1, (a-1)(b-1)}$ , then reject  $H_0$ .

## Another example I

An experiment was designed to study the performance of our different detergents in cleaning clothes. The following “cleanness” readings (higher=cleaner) were obtained specially designed equipment for three different types of common stains. Is there a difference between the detergents?

	Stain 1	Stain 2	Stain 3
Detergent 1	45	43	51
Detergent 2	47	46	52
Detergent 3	48	50	55
Detergent 4	42	39	49

## Another example II

$$\sum_i \sum_j y_{ij} = 565 \text{ and } \sum_i \sum_j y_{ij}^2 = 26867$$

$$y_{1\cdot} = 139, y_{2\cdot} = 145, y_{3\cdot} = 153, \text{ and } y_{4\cdot} = 128;$$

$$y_{\cdot 1} = 182, y_{\cdot 2} = 176, \text{ and } y_{\cdot 3} = 207$$

$$SS_T = 26867 - 565^2/12 = 265$$

$$SS_{Trt} = (139^2 + 145^2 + 153^2 + 128^2)/3 - 565^2/12 = 111$$

$$SS_{Block} = (182^2 + 176^2 + 207^2)/4 - 565^2/12 = 135$$

$$SSE = 265 - 111 - 135 = 19$$

$$F_0 = (111/3)/(19/6) = 11.6, \text{ P-value} < 0.01$$

# Checking Assumptions (Diagnostics) I

- Assumptions
  - Model is additive (no interaction between treatment effects and block effects) (additivity assumption).
  - Errors are independent and normally distributed.
  - Constant variance.
- Checking normality:
  - Histogram, QQ plot of residuals, Shapiro-Wilk Test.
- Checking constant variance
  - Residual Plot: Residuals vs  $\hat{y}_{ij}$
  - Residuals vs blocks
  - Residuals vs treatments
- Additivity
  - Residual plot: residuals vs  $\hat{y}_{ij}$

## Checking Assumptions (Diagnostics) II

- If residual plot shows curvilinear pattern, interaction between treatment and block likely exists.
- Interaction: block effects can be different for different treatments.
- Formal test: Tukey's One-degree Freedom Test of Non-additivity.
- If interaction exists, usually try transformation to eliminate interaction.

See block-design.SAS.



# Model adequacy checking: additivity

- We used a linear statistical model for RCBD:

$$y_{ij} = \mu + \tau_i + \beta_j + \epsilon_{ij}$$

- In other word, it is additive.
- The linear model is very useful, but in some situations it may be inadequate.
  - i.e., there may be an interaction between the treatment and block.

# Tukey's Test for Non-additivity I

- Additivity assumption (or no interaction assumption) is crucial for block designs or experiments.
- To check the interaction between block and treatment fully needs  $(a - 1)(b - 1)$  degree of freedom. It is not affordable when without replicates.
- Instead consider a special type of interaction. Assume following model

$$y_{ij} = \mu + \tau_i + \beta_j + \gamma\tau_i\beta_j + \epsilon_{ij}$$

## Tukey's Test for Non-additivity II

- $H_0 : \gamma = 0$  vs  $H_1 : \gamma \neq 0$

Sum of Squares caused by possible interaction:

$$SS_N = \frac{\left[ \sum_i \sum_j y_{ij} y_{i\cdot} y_{\cdot j} - y_{\cdot\cdot} (SS_{Trt} + SS_{Block} + y_{\cdot\cdot}^2 / ab) \right]^2}{ab SS_{Trt} SS_{Block}}, \quad df = 1.$$

Remaining error SS:  $SSE' = SSE - SS_N$ ,

$$df = (a-1)(b-1) - 1$$

Test Statistic:

$$F_0 = \frac{SS_N / 1}{SSE' / [(a-1)(b-1) - 1]} \sim F_{1, (a-1)(b-1) - 1}$$

- Decision rule: Reject  $H_0$  if  $F_0 > F_{\alpha, 1, (a-1)(b-1) - 1}$ .

# Tukey's Test for Non-additivity III

- A Convenient Procedure to Calculate  $SS_N$ ,  $SSE'$  and  $F_0$ 
  - Fit additive model  $y_{ij} = \mu + \tau_i + \beta_j + \epsilon_{ij}$
  - Obtain  $\hat{y}_{ij}$  and  $q_{ij} = \hat{y}_{ij}^2$
  - Fit the model  $y_{ij} = \mu + \tau_i + \beta_j + q_{ij} + \epsilon_{ij}$ 

Use the test for  $q_{ij}$  in the ANOVA table with type III SS and ignore the test for the treatment and block factors.

See block-design-additivity.SAS.

## Type I, III sum of squares

$$Y_i = \beta_0 + \beta_1 X_{i1} + \beta_2 X_{i2} + \beta_3 X_{i3} + \epsilon_i$$

- Type I (sequential) Sums of Squares

- The Type I Sums of Squares for  $\beta_1$  are the Sums of Squares obtained from fitting  $\beta_1$  over and above the mean;
- The Type I Sums of Squares for  $\beta_2$  are the Sums of Squares obtained from fitting  $\beta_2$  after  $\beta_1$ .
- etc

- Type III (marginal) Sums of Squares

- The Sums of Squares obtained by fitting each effect after all the other terms in the model.
- The Type III (marginal) Sums of Squares do not depend upon the order in which effects are specified in the model.

More info: <http://afni.nimh.nih.gov/sscc/gangc/SS.html>

# Post-ANOVA Treatments Comparison I

- Multiple Comparisons/Contrasts
  - procedures (methods) are similar to those for Completely Randomized Design (CRD)
    - $n$  is replaced by  $b$  in all formulas
    - Degree of freedom for error is  $(b - 1)(a - 1)$
- Example: Comparison of Detergents See block-design-additivity.SAS.

- Tukey's Method ( $\alpha = .05$ )

$$q_{\alpha}(a, df) = q_{\alpha}(4, 6) = 4.896.$$

$$CD = \frac{q_{\alpha}(4,6)}{\sqrt{2}} \sqrt{MSE \left( \frac{1}{b} + \frac{1}{b} \right)} = 4.896 \sqrt{\frac{19}{6 \times 3}} = 5.001$$

## Post- ANOVA Treatments Comparison II

### Comparison of Treatment Means

#### Treatments

4	1	2	3
42.67	46.33	48.33	51.00
A	A		
	B	B	B

# Random Blocks and/or Treatments I

Assuming that the RCBD model is appropriate, if the blocks are random and the treatments are fixed, then

$$y_{ij} = \mu + \tau_i + \beta_j + \epsilon_{ij},$$
$$\beta_j \stackrel{iid}{\sim} N(0, \sigma_\beta^2), \quad \epsilon_{ij} \stackrel{iid}{\sim} N(0, \sigma^2),$$

for  $i = 1, \dots, a$ ;  $j = 1, \dots, b$ .  $\beta_j$  and  $\epsilon_{ij}$  are independent. Then we have

$$\begin{aligned} E(y_{ij}) &= \mu + \tau_i, \quad \text{var}(y_{ij}) = \sigma_\beta^2 + \sigma^2, \\ \text{cov}(y_{ij}, y_{i'j'}) &= 0, \quad j \neq j', \\ \text{cov}(y_{ij}, y_{i'j}) &= \sigma_\beta^2, \quad i \neq i', \end{aligned}$$



## Random Blocks and/or Treatments II

Thus, the variance of the observations is constant, the covariance between any two observations in different blocks is zero, but the covariance between two observations from the same block is  $\sigma_\beta^2$ . The expected mean squares from the usual ANOVA partitioning of the total sum of squares are

$$E(MS_{Treatment}) = \sigma^2 + \frac{b \sum_{i=1}^a \tau_i^2}{a-1},$$

$$E(MS_{Block}) = \sigma^2 + a\sigma_\beta^2,$$

$$E(MSE) = \sigma^2.$$

## Random Blocks and/or Treatments III

The appropriate statistic for testing the null hypothesis of no treatment effects (all  $\tau_i = 0$ ) is

$$F_0 = \frac{MS_{Treatment}}{MSE}$$

which is exactly the same test statistic we used in the case where the blocks were fixed. Based on the expected mean squares, we can obtain an ANOVA-type estimator of the variance component for blocks as

$$\hat{\sigma}_\beta^2 = \frac{MS_{Block} - MSE}{a}.$$

# Missing Values I

- When missing
  - Orthogonality lost
  - Design unbalanced
- Procedures
  - Exact (Regression) approach: Use Type III SS's (general regression significant test)
  - Approximate approach: Estimate missing value  
Choose value to minimize  $SSE$   
Take derivative and set equal to zero

## Missing Values II

$$\begin{aligned}SS_E &= \sum \sum y_{ij}^2 - y_{..}^2/ab - \frac{1}{b} \sum y_{i.}^2 + y_{..}^2/ab - \frac{1}{a} \sum y_{.j}^2 + y_{..}^2/ab \\&= x^2 - \frac{1}{b}(y'_{i.} + x)^2 - \frac{1}{a}(y'_{.j} + x)^2 + \frac{1}{ab}(y'_{..} + x)^2 + R\end{aligned}$$

$$x = \frac{ay'_{i.} + by'_{.j} - y'_{..}}{(a-1)(b-1)}$$

- Example: detergent comparison example
  - Suppose  $y_{4,2} = 37$  is missing

## Missing Values III

- Estimate approach

$$y'_{4.} = 91, \quad y'_{..} = 528, \quad y'_{.2} = 139$$

Estimate is

$$x = \frac{4(91) + 3(139) - 528}{6} = 42.17$$

Do analysis but adjust error degrees of freedom

- Estimate:  $\hat{\sigma}^2 = 1.097$  (must divide by 5 not 6)
- See missing\_block.SAS.
- $F_0 = \frac{71.95/3}{5.49/5} = 21.84$ , p-value= 0.0027
- Remark: Approximate approach produces a biased mean square for treatment.  
Exact analysis approach is preferred.