

## Lecture 16

### Topic 12: The split-plot design and its relatives (Part I)

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#### Definition

A split plot design results from a **two-stage randomization process of a factorial treatment structure**. Because of this two-stage process, one loses sensitivity in detecting differences among main plot treatments (the first level of randomization) but gains sensitivity in detecting differences among subplot treatments (the second level), as well as the significance of the MainPlot:Subplot interaction.

The basic split-plot design involves assigning the levels of one factor (A) to **main plots** arranged in a CRD, RCBD, Latin Square, etc., and then assigning the levels of a second factor (B) to **subplots** within each main plot.

1. Two-stage randomization process
2. Two distinct error terms (Error for main plot effects > Error for subplot effects)

#### Uses of the split-plot design

1. Physical constraints to randomization  
*e.g.* one factor requires a larger amount of experimental material than another
2. To increase precision of some effects  
*e.g.* split-plots often improve precision for detecting subplot and interaction effects
3. To increase the scope of an experiment.  
*e.g.* seed protectants and varieties.

#### In general, the subplot (Factor B):

- ...requires smaller amounts of experimental material
- ...is of primary importance
- ...is expected to exhibit smaller differences

## Split-plots *within* various designs

Factor A is the main plot factor, with 3 levels

Factor B is the subplot factor, with 2 levels.

There are 4 replications per main plot.

### Simple 3x2 factorial (no split), arranged as a CRD

a1b1	a2b2	a2b1	a1b2	a3b2	a1b1	a2b2	a2b1	a1b2	a3b2	a1b1	a3b2
a2b2	a3b1	a1b2	a3b1	a1b2	a3b2	a2b1	a1b1	a2b2	a3b1	a2b1	a3b1

### Split-plot, with main plots arranged as a CRD

Stage 1: Randomize the levels of factor A over the main plots:

a2	a3	a2	a1	a2	a3	a2	a3	a1	a3	a1	a1
a2	a3	a2	a1	a2	a3	a2	a3	a1	a3	a1	a1

Stage 2: Randomize the levels of factor B over the subplots:

a2b2	a3b2	a2b1	a1b1	a2b1	a3b2	a2b1	a3b2	a1b1	a3b1	a1b1	a1b2
a2b1	a3b1	a2b2	a1b2	a2b2	a3b1	a2b2	a3b1	a1b2	a3b2	a1b2	a1b1

### Split-plot, with main plots arranged as an RCBD

Stage 1: Randomize the levels of factor A within each block.

a2	a1	a3	a1	a2	a3	a1	a3	a2	a3	a2	a1
a2	a1	a3	a1	a2	a3	a1	a3	a2	a3	a2	a1

Stage 2: Randomize the levels of factor B over the subplots.

a2b1	a1b1	a3b2	a1b2	a2b1	a3b2	a1b2	a3b1	a2b2	a3b1	a2b1	a1b1
a2b2	a1b2	a3b1	a1b1	a2b2	a3b1	a1b1	a3b2	a2b1	a3b2	a2b2	a1b2

## Linear models for the split-plot

The linear model for the split-plot, with main plots arranged as an RCBD:

$$Y_{ijk} = \mu + \tau_{Ai} + \beta_j + (\tau_A:\beta)_{ij} + \tau_{Bk} + (\tau_A:\tau_B)_{ik} + \varepsilon_{ijk}$$

where

$i = 1, \dots, a$	indexes the main plot levels
$j = 1, \dots, b$	indexes the blocks
$k = 1, \dots, r$	indexes the subplot levels

The variance associated with  $(\tau_A\tau_B)_{ij}$  (i.e.  $\sigma_{AB}^2$ ) is used to test the main plot effects.

The variance associated with  $\varepsilon_{ijk}$  (i.e.  $\sigma_\varepsilon^2$ ) is used to test the subplot and interaction effects.

Usually,  $\sigma_{AB}^2 > \sigma_\varepsilon^2$ .

The linear model for the split-plot, with main plots arranged as a CRD:

$$Y_{ijk} = \mu + \tau_{Ai} + (\tau_A:\rho)_{ij} + \tau_{Bk} + (\tau_A:\tau_B)_{ik} + \varepsilon_{ijk}$$

where now

$j = 1, \dots, r$	indexes the replications ( $\rho = \text{"rho"} = \text{rep}$ )
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<p><b>CRD Main plot error = <i>Main plot : Replication</i></b> <b>RCBD Main plot error = <i>Main plot : Block</i></b></p>
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<p><b>CRD Subplot error = <i>Subplot : Replication + Main plot : Subplot : Replication</i></b> <b>RCBD Subplot error = <i>Subplot : Block + Main plot : Subplot : Block</i></b></p>
---

## Split-plot ANOVA

The general ANOVA table for the split-plot CRD:

Source	df	SS	MS	F
<b>Total (subplots)</b>	$rab - 1$	TSS		
<b>Main plots total</b>	$ra - 1$	SS(MP)		
<b>Factor A</b>	$a - 1$	SSA	MSA	MSA/MS(MPE)
<i>Main plot error</i>	$a(r - 1)$	SS(MPE)	MS(MPE)	
<b>Factor B</b>	$b - 1$	SSB	MSB	MSB/MS(SPE)
<b>A x B</b>	$(a - 1)(b - 1)$	SS(AxB)	MS(AxB)	MS(AxB)/MS(SPE)
<i>Subplot error</i>	$a(r - 1)(b - 1)$	SS(SPE)	MS(SPE)	

CRD		RCBD		Latin Square	
<b>Total</b>	$ra - 1$	<b>Total</b>	$ra - 1$	<b>Total</b>	$ra - 1$
		<b>Blocks</b>	$r - 1$	<b>Rows</b>	$a - 1$
<b>A</b>	$a - 1$	<b>A</b>	$a - 1$	<b>Columns</b>	$a - 1$
<b>Error A</b>	$a(r - 1)$	<b>Error A</b>	$(r - 1)(a - 1)$	<b>A</b>	$a - 1$
Factor B	$b - 1$	Factor B	$b - 1$	<b>Error A</b>	$(a - 1)(a - 2)$
A x B	$(a - 1)(b - 1)$	A x B	$(a - 1)(b - 1)$	Factor B	$b - 1$
Error B	$a(r - 1)(b - 1)$	Error B	$a(r - 1)(b - 1)$	A x B	$(a - 1)(b - 1)$
Total	$rab - 1$	Total	$rab - 1$	Error B	$a(r - 1)(b - 1)$
				Total	$rab - 1$

### CRD

```
lm( Y ~ A + Rep:A + B + A:B )
```

### RCBD

```
lm( Y ~ Block + A + Block:A + B + A:B )
```

### Latin Square

```
lm( Y ~ Row + Col + A + Row:Col:A + B + A:B )
```

### Replicated Latin Square (shared rows and columns)

```
lm( Y ~ Square + Row + Col + A + Square:Row:Col:A + B + A:B )
```

## Example of a split-plot with main plots arranged as an RCBD

(*Phytopathology* 71: 605-608) Experiment conducted to determine the effect of bacterial vascular necrosis on the root yield of sugar beets planted at different in-row spacings.

**Main plot A:** Inoculation (inoculated vs. not inoculated with *Erwinia carotovora*)

**Subplot B:** In-row spacing between plants (4, 6, 12, and 18 inches)

### Field Layout

Block									
VI	No inoculation	4	12	18	6	6	12	4	18
		21.0	22.9	23.1	22.0	17.6	16.1	16.8	13.1
V	Inoculation	18	6	4	12	6	4	12	18
		12.9	19.8	17.2	16.8	21.2	17.9	22.3	22.0
IV	No inoculation	6	18	4	12	12	18	6	4
		21.1	21.4	18.4	22.8	16.1	14.7	16.3	16.8
III	No inoculation	18	12	4	6	18	6	12	4
		19.3	18.6	18.2	20.8	12.5	19.1	16.6	16.5
II	Inoculation	12	6	18	4	4	12	18	6
		14.9	17.0	12.1	16.4	17.9	21.1	20.1	19.6
I	Inoculation	4	12	18	6	18	12	6	4
		17.4	16.3	12.5	17.3	20.0	21.8	20.2	20.1

## The R code

```
sugar_bad_mod<-lm(yield ~ A_inoc + block + A_inoc:block + B_space +  
  A_inoc:B_space, sugar_dat)  
anova(sugar_bad_mod)
```

The above command follows the linear model specified on the previous page and therefore seems to be a reasonable approach to analyzing this particular dataset. If you run this code, you obtain the following output:

### Analysis of Variance Table

Response: yield

	Df	Sum Sq	Mean Sq	F value	Pr(>F)		
A_inoc	1	256.687	256.687	327.6165	< 2.2e-16	***	<--WRONG!
block	5	16.250	3.250	4.1481	0.005541	**	<--WRONG!
A_inoc:block	5	11.535	2.307	2.9445	0.028023	*	<--MP error
B_space	3	39.638	13.213	16.8634	1.320e-06	***	
A_inoc:B_space	3	64.438	21.479	27.4144	9.838e-09	***	
Residuals	30	23.505	0.783				

Programmed in this way, all the effects in the model are being tested with the residual error (0.783, df = 30). Unfortunately, this is the incorrect error term for testing the effect of the main plot (A\_inoc) and the Block factors, as can be seen in the following table of expected mean squares:

Source	Expected Mean Square
Block	$\text{Var}(\text{Error}) + 4 \text{ Var}(\text{Block:A\_Inoc}) + 8 \text{ Var}(\text{Block})$
A_Inoc	$\text{Var}(\text{Error}) + 4 \text{ Var}(\text{Block:A\_Inoc}) + \text{Fixed\_effect}(\text{A\_Inoc})$
Block:A_Inoc	$\text{Var}(\text{Error}) + 4 \text{ Var}(\text{Block:A\_Inoc})$
B_Space	$\text{Var}(\text{Error}) + \text{Fixed\_effect}(\text{B\_Space})$
A_Inoc:B_Space	$\text{Var}(\text{Error}) + \text{Fixed\_effect}(\text{A\_Inoc:B\_Space})$

The appropriate error term for A is **Block:A**

The appropriate error term for B and A:B is the residual error

To obtain the correct F and p-values for the main plot factor and the blocks, you need to tell R to test those factors with the appropriate error term, namely A:Block. One way to do this is to run a manual F test. Another way is to use the `aov()` function like this:

```
sugar_good_mod<-aov(yield ~ A_inoc + block + Error(A_inoc:block) + B_space +
  A_inoc:B_space, sugar_dat)
summary(sugar_good_mod)
```

The result of this line of code is:

```
Error: A_inoc:block
      Df Sum Sq Mean Sq F value    Pr(>F)
A_inoc   1 256.69  256.69 111.265 0.000132 ***
block     5  16.25    3.25   1.409 0.358023
Residuals 5  11.54    2.31
```

```
Error: Within
      Df Sum Sq Mean Sq F value    Pr(>F)
B_space   3  39.64  13.213   16.86 1.32e-06 ***
A_inoc:B_space 3  64.44  21.479   27.41 9.84e-09 ***
Residuals 30  23.50   0.783
```

Using the wrong error term before, we concluded that there were significant yield differences among blocks and highly a significant effect of inoculation (Main Plot  $p < 2.2e-16$ ). Now, using the correct error term for these effects, we see that the differences among blocks are not significant and the effect of inoculation is twelve orders of magnitude smaller than we thought ( $p = 1.32e-4$ )!

Putting everything together, we arrive at this final ANOVA table for the sugar beet root rot study:

Source	df	SS	MS	F
<b>Total (subplots)</b>	47	412.06		
<b>Block</b>	5	16.26	3.25	1.41 NS
<b>Inoculation (A)</b>	1	256.69	256.69	111.26 ***
<b>Error A (Block:A)</b>	5	11.54	2.31	
<b>Spacing (B)</b>	3	39.64	13.21	16.86 ***
<b>Interaction (A:B)</b>	3	64.44	21.48	27.41 ***
<b>Error B</b>	30	23.50	0.78	

**Notice:  $MSE_A (2.31) > MSE_B (0.78)$**

## Mean comparisons

As with all factorial treatment structures, subsequent analysis of the data depends first and foremost on whether or not a significant interaction exists among the factors. In this particular case, the interaction A\_inoc:B\_space was found to be highly significant ( $p = 9.84e-09$ ).

When the **interaction** between main plot and subplot is **significant**, we can make three different kinds of comparisons:

1. Comparisons among subplot levels *within* a main plot level
2. Comparisons among main plot levels *within* a subplot level
3. Comparisons between subplot levels across different main plot levels

Block									
VI		4	12	18	6	6	12	4	18
		21.0	22.9	23.1	22.0	17.6	16.1	16.8	13.1
		No inoculation				Inoculation			
V		18	6	4	12	6	4	12	18
		12.9	19.8	17.2	16.8	21.2	17.9	22.3	22.0
		Inoculation				No inoculation			
IV		6	18	4	12	12	18	6	4
		21.1	21.4	18.4	22.8	16.1	14.7	16.3	16.8
		No inoculation				Inoculation			
III		18	12	4	6	18	6	12	4
		19.3	18.6	18.2	20.8	12.5	19.1	16.6	16.5
		No inoculation				Inoculation			
II		12	6	18	4	4	12	18	6
		14.9	17.0	12.1	16.4	17.9	21.1	20.1	19.6
		Inoculation				No inoculation			
I		4	12	18	6	18	12	6	4
		17.4	16.3	12.5	17.3	20.0	21.8	20.2	20.1
		Inoculation				No inoculation			



## 1. Comparisons among subplot levels *within* a common main plot level

```
no_inoc_dat<-subset(sugar_dat, A_inoc == 0)
no_inoc_mod<-lm(yield ~ block + B_space, no_inoc_dat)
MP_comp1<-LSD.test(no_inoc_mod, "B_space")
MP_comp1

inoc_dat<-subset(sugar_dat, A_inoc == 1)
inoc_mod<-lm(yield ~ block + B_space, inoc_dat)
MP_comp2<-LSD.test(inoc_mod, "B_space")
MP_comp2
```

### The output:

#### **A\_inoc = 0**

```
alpha: 0.05 ; Df Error: 15
Mean Square Error: 0.8465556
Critical Value of t: 2.13145
Least Significant Difference 1.13225
```

#### Groups, Treatments and means

a	12	21.58
a	18	20.98
a	6	20.82
b	4	18.92

#### **A\_inoc = 1**

```
alpha: 0.05 ; Df Error: 15
Mean Square Error: 0.7204444
Critical Value of t: 2.13145
Least Significant Difference 1.044515
```

#### Groups, Treatments and means

a	6	17.85
ab	4	16.85
b	12	16.13
c	18	12.97

$1/2(0.846556 + 0.720444) = 0.7835$ , the subplot error used in the original analysis!

## 2. Comparisons among main plot levels *within* a common subplot level

```
sp_4_dat<-subset(sugar_dat, B_space == 4)
sp_4_mod<-lm(yield ~ block + A_inoc, sp_4_dat)
SP_comp1<-LSD.test(sp_4_mod, "A_inoc")
SP_comp1

sp_6_dat<-subset(sugar_dat, B_space == 6)
sp_6_mod<-lm(yield ~ block + A_inoc, sp_6_dat)
SP_comp2<-LSD.test(sp_6_mod, "A_inoc")
SP_comp2

sp_12_dat<-subset(sugar_dat, B_space == 12)
sp_12_mod<-lm(yield ~ block + A_inoc, sp_12_dat)
SP_comp3<-LSD.test(sp_12_mod, "A_inoc")
SP_comp3

sp_18_dat<-subset(sugar_dat, B_space == 18)
sp_18_mod<-lm(yield ~ block + A_inoc, sp_18_dat)
SP_comp4<-LSD.test(sp_18_mod, "A_inoc")
SP_comp4
```

The residual error here is automatically the mean square of the Block:A interaction!

### A summary of the output:

	B_space			
	4	6	12	18
MSE	0.74933	0.96133	1.5855	0.85883
LSD	1.28472	1.45515	1.86876	1.37539
No Inoc	18.92 a	20.82 a	21.58 a	20.98 a
Inoc	16.85 b	17.85 b	16.13 b	12.97 b

### Mixed Comparisons: Comparisons between subplot levels *across* different main plot levels

The appropriate error ( $MSE_{Mix}$ ) is a weighted average of  $MSE_A$  (i.e. the appropriate error term for testing factor A, the main plot factor) and  $MSE_B$  (i.e. the appropriate error term for testing factor B, the subplot factor), with emphasis on  $MSE_B$ . Such comparisons require hand computations.

The appropriate weighted error is:

$$MSE_{Mix} = \frac{(b-1) * MSE_B + MSE_A}{b} = \frac{(4-1) * 0.783 + 2.307}{4} = 1.164375$$

where  $b$  is the number of levels in Factor B. Similarly, the appropriate critical value is an intermediate value between the critical value for the main plot ( $t_{A, 5 \text{ df}} = 2.571$ ) and that for the subplot ( $t_{B, 30 \text{ df}} = 2.042$ ). In each case, the degrees of freedom used when finding these critical values are the df's associated with the *error* terms for each factor. In this example, the error term for Factor A is Block:A, and the df associated with Block:A is 5. The error term for Factor B is the residual error, and the df associated with the residual error is 30. Refer to the ANOVA table on the bottom of Page 7.

The formula:

$$t_{Mix} = \frac{(b-1) * t_B * MSE_B + t_A * MSE_A}{(b-1) * MSE_B + MSE_A} = \frac{3 * 2.042 * 0.783 + 2.571 * 2.307}{3 * 0.783 + 2.307} = 2.304$$

Sticking with the LSD method:

$$LSD_{\alpha=0.05} = t_{Mix} \sqrt{\frac{2MSE_{Mix}}{r}} = 2.304 * \sqrt{\frac{2(1.164375)}{6}} = 1.435$$

**Example:** To compare the mean of inoculated / spacing 4 = 16.85 with the mean of not inoculated / spacing 6 = 20.82:

$$|20.82 - 16.85| = 4.32$$

Since  $4.32 > 1.435 \Rightarrow$  This difference is significant

If the (main plot : subplot) **interaction** is **not significant**, there are two possibilities for subsequent analysis:

1. Main effects comparisons among main plot levels
2. Main effects comparisons among subplot levels

### 1. Main plot comparisons in the absence of an interaction

A valid comparison among the means of the main plot levels requires the appropriate error variance ( $MS_{\text{Block:A}} = 2.307$ ).

```
MP_comparison<-LSD.test(sugar_dat$yield, sugar_dat$A_inoc, DFerror = 5,
  MSerror = 2.307)
MP_comparison
```

**The output:**

	Mean	CV	MSerror	LSD
	18.2625	8.31694	2.307	1.127106

  

	trt	means	M
1	0	20.575	a
2	1	15.950	b

Incidentally, just like the means comparison tests, any orthogonal contrasts must also specify the correct error term! Example:

```
# Contrast 'No_Inoc vs. Inoc'          1,-1
contrastmatrix<-cbind(c(1,-1))

contrasts(sugar_dat$A_inoc)<-contrastmatrix

sugar_contrast_mod<-aov(yield ~ A_inoc + block + Error(A_inoc:block) +
  B_space + A_inoc:B_space, sugar_dat)
summary(sugar_contrast_mod, split = list(A_inoc = list("No_inoc vs. Inoc" =
  1)))
```

## 2. Subplot comparisons in the absence of an interaction

For comparisons among subplots, the residual error is the correct error term to use:

```
SP_comparison<-LSD.test(sugar_dat$yield, sugar_dat$B_space, DFerror = 30,  
  MSerror = 0.7835)  
SP_comparison
```

The output:

Mean	CV	MSerror	LSD
18.2625	4.846847	0.7835	0.738002

trt	means	M
1 6	19.33333	a
2 12	18.85833	a
3 4	17.88333	b
4 18	16.97500	c

## Split-split plot design

The concept of the split-plot design extends logically from two to three factors:

### 1. Split-plot with factorial main plot:

*Combinations of levels of Factors A and B* are assigned to main plots, levels of Factor C to subplots within each mainplot.

### 2. Split-plot with factorial subplot:

Levels of Factor A are assigned to main plots, *combinations of levels of Factors B and C* are assigned to subplots.

### 3. Split-split plot:

Levels of Factor A are assigned to main plots, levels of Factor B to subplots within each mainplot, and levels of Factor C to sub-subplots within each subplot.

In the last case, the additional stage of randomization introduces an additional (third) error term, required for testing the main effects of Factor C and all interactions involving Factor C.

**Example:** (Little & Hills) A split-split plot to evaluate the effects of date of planting (A), aphid control (B), and date of harvest (C) on the control of an aphid-borne sugar beet virus.

Block I			Block II								
			A <sub>1</sub>			A <sub>3</sub>			A <sub>2</sub>		
Block III			Block IV								
A <sub>3</sub> B <sub>1</sub>	A <sub>1</sub> B <sub>1</sub>	A <sub>2</sub> B <sub>2</sub>	A <sub>2</sub> B <sub>1</sub> C <sub>1</sub>	A <sub>2</sub> B <sub>1</sub> C <sub>3</sub>	A <sub>2</sub> B <sub>1</sub> C <sub>2</sub>	A <sub>1</sub> B <sub>2</sub> C <sub>3</sub>	A <sub>1</sub> B <sub>2</sub> C <sub>1</sub>	A <sub>1</sub> B <sub>2</sub> C <sub>2</sub>	A <sub>3</sub> B <sub>2</sub> C <sub>1</sub>	A <sub>3</sub> B <sub>2</sub> C <sub>3</sub>	A <sub>3</sub> B <sub>2</sub> C <sub>2</sub>
A <sub>3</sub> B <sub>2</sub>	A <sub>1</sub> B <sub>2</sub>	A <sub>2</sub> B <sub>1</sub>	A <sub>2</sub> B <sub>2</sub> C <sub>3</sub>	A <sub>2</sub> B <sub>2</sub> C <sub>2</sub>	A <sub>2</sub> B <sub>2</sub> C <sub>1</sub>	A <sub>1</sub> B <sub>1</sub> C <sub>1</sub>	A <sub>1</sub> B <sub>1</sub> C <sub>3</sub>	A <sub>1</sub> B <sub>1</sub> C <sub>2</sub>	A <sub>3</sub> B <sub>1</sub> C <sub>3</sub>	A <sub>3</sub> B <sub>1</sub> C <sub>1</sub>	A <sub>3</sub> B <sub>1</sub> C <sub>2</sub>

**ANOVA for the split-split plot design** is an extension of the split-plot case. The various error terms are constructed by extracting different sources of variation from the residual error and pooling them together:

---

**Level One:**

**Block**

**A**

Tested using (**Block:A**) = **Error A**

**Level Two:**

**B**

**A:B**

Tested using (**Block:B + Block:A:B**) = **Error B**

**Level Three:**

**C**

**A:C**

**B:C**

**A:B:C**

Tested using (**Block:C + Block:A:C + Block:B:C + Block:A:B:C**) = resid error  
= **Error C**

---

What this means is that a full analysis requires specifying *TWO* special error terms for custom F tests, in addition to the model's residual error. Unfortunately, the `aov()` function only permits the specification of *ONE* custom error term. There are several options as to how to proceed. One strategy is to run a couple of different `aov()` models and combine the results into a final ANOVA table:

## Model 1: Testing the main plot effects

```
split_split_MP_mod<-aov(Y ~ A + Block +  
                          Error(A:Block) +  
                          B + A:B +  
                          C + A:C + B:C + A:B:C,  
                          split_split_dat)  
summary(split_split_MP_mod)
```

### The resulting ANOVA table:

#### Error: A:Block

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
A	3	251.49	83.83	12.50	0.00543	**
Block	2	275.43	137.72	20.54	0.00207	**
Residuals	6	40.23	6.70			

#### Error: Within

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
<del>B</del>	<del>1</del>	<del>200.3</del>	<del>200.28</del>	<del>52.369</del>	<del>8.80e-09</del>	<del>***</del> ← WRONG!!
<del>C</del>	<del>2</del>	<del>388.1</del>	<del>194.05</del>	<del>50.742</del>	<del>1.06e-11</del>	<del>***</del> ← WRONG!!
<del>A:B</del>	<del>3</del>	<del>2.5</del>	<del>0.83</del>	<del>0.216</del>	<del>0.885</del>	<del>← WRONG!!</del>
<del>A:C</del>	<del>6</del>	<del>29.4</del>	<del>4.89</del>	<del>1.280</del>	<del>0.288</del>	<del>← WRONG!!</del>
<del>B:C</del>	<del>2</del>	<del>2.1</del>	<del>1.05</del>	<del>0.275</del>	<del>0.761</del>	<del>← WRONG!!</del>
<del>A:B:C</del>	<del>6</del>	<del>16.6</del>	<del>2.76</del>	<del>0.722</del>	<del>0.634</del>	<del>← WRONG!!</del>
Residuals	40	153.0	3.82			



## Model 2: Testing the subplot effects

```
split_split_SP_mod<-aov(Y ~ A + Block +
  A:Block +
  B + A:B +
  Error(A:B:Block) +
  C + A:C + B:C + A:B:C,
  split_split_dat)
summary(split_split_SP_mod)
```

### The resulting ANOVA table:

#### Error: A:B:Block

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
A	3	251.49	83.83	51.133	1.46e-05 ***	← WRONG!!
Block	2	275.43	137.72	84.002	4.27e-06 ***	← WRONG!!
B	1	200.28	200.28	122.162	4.00e-06 ***	
A:Block	6	40.23	6.70	4.090	0.0355 *	← WRONG!!
A:B	3	2.48	0.83	0.503	0.6906	
Residuals	8	13.12	1.64			

#### Error: Within

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
C	2	388.1	194.05	44.400	5.88e-10 ***
A:C	6	29.4	4.89	1.120	0.373
B:C	2	2.1	1.05	0.241	0.787
A:B:C	6	16.6	2.76	0.632	0.704
Residuals	32	139.9	4.37		

### Combining the results into a complete table:

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	3	251.49	83.83	12.50	0.00543 **
Block	2	275.43	137.72	20.54	0.00207 **
A:Block	6	40.23	6.70		.
B	1	200.28	200.28	122.162	4.00e-06 ***
A:B	3	2.48	0.83	0.503	0.6906
A:B:Block	8	13.12	1.64		.
C	2	388.1	194.05	44.400	5.88e-10 ***
A:C	6	29.4	4.89	1.120	0.373
B:C	2	2.1	1.05	0.241	0.787
A:B:C	6	16.6	2.76	0.632	0.704
Error	32	139.9	4.37		

Another way to do it is to obtain the SS for all the factors in the linear model and then carry out the custom F-tests manually. For this, one would specify the full model:

```
split_split_mod<-aov(Y ~ A + Block +
                     A:Block +
                     B + A:B +
                     B:Block + A:Block:B +
                     C + A:C + B:C + A:B:C,
                     split_split_dat)
summary(split_split_mod)
```

**The resulting ANOVA table:**

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	3	251.5	83.83	<del>19.180</del>	<del>2.66e-07</del> ***
Block	2	275.4	137.72	<del>31.510</del>	<del>2.74e-08</del> ***
B	1	200.3	200.28	<del>45.824</del>	<del>1.19e-07</del> ***
C	2	388.1	194.05	44.400	5.88e-10 ***
A:Block	6	40.2	6.70	1.534	0.199
A:B	3	2.5	0.83	<del>0.189</del>	<del>0.903</del>
Block:B	2	0.7	0.35	0.079	0.924
A:C	6	29.4	4.89	1.120	0.373
B:C	2	2.1	1.05	0.241	0.787
A:Block:B	6	12.4	2.07	0.474	0.823
A:B:C	6	16.6	2.76	0.632	0.704
Residuals	32	139.9	4.37		

To test the main plot effect (A):

$$F = 83.83/6.70 = 12.50$$

$$p(12.50, 3, 6) = 0.00543$$

To test the subplot effect (B):

$$F = 200.28/[(0.7+12.4)/(2+6)] = 122.162$$

$$p(122.162, 1, 8) = 4.00e-06$$

How would you test the A:B interaction?

## Strip-plot (or split-block) design

In the strip-plot or split-block design, the subunit treatments are applied in strips across a complete set (replication) of main plot levels.

A comparison of the layouts for a 5x4 split-plot design and a 5x4 strip-plot design (only one replication, or block, is shown).

**Split-plot**

A3	A2	A1	A5	A4
B2	B1	B2	B3	B4
B1	B3	B1	B2	B3
B3	B2	B4	B4	B1
B4	B4	B3	B1	B2

**Strip-plot (or split-block)**

A3	A2	A1	A5	A4
B2	B2	B2	B2	B2
B4	B4	B4	B4	B4
B1	B1	B1	B1	B1
B3	B3	B3	B3	B3

In the strip-plot, we retain the terms "Main Plot" and "Subplot." From a theoretical perspective, however, there is no longer any difference between the two.

## Reasons for arranging an experiment as a strip-plot design

1. Physical operations (e.g. tractor manipulation, irrigation, harvesting) may be easier.
2. The design tends to reduce precision in testing the main effects but **improves precision in detecting interaction effects**.

## Linear model for the strip-plot design (RCBD case)

$$Y_{ijk} = \mu + \tau_{Ai} + \tau_{Bj} + \beta_k + (\tau_A:\beta)_{ik} + (\tau_B:\beta)_{jk} + (\tau_A:\tau_B)_{ij} + \varepsilon_{ijk}$$

where

$i = 1, \dots, a$  indexes the main plot levels

$j = 1, \dots, b$  indexes the subplot levels

$k = 1, \dots, r$  indexes the blocks

The extra term  $(\tau_B:\beta)_{jk}$  represents the interaction effect of blocks with subplot levels.

## ANOVA for the split-block design

**CRD:** To test the main effects of Factor B, the proper error term is the strip-plot error  $MS(StPE) = B:Rep$

**RCBD:** To test the main effects of Factor B, the proper error term is the strip-plot error  $MS(StPE) = B:Block$

The general ANOVA table for the RCBD strip-plot design:

Source	df	SS	MS	F
<b>Total</b>	$rab - 1$	TSS		
<b>Block</b>	$r - 1$	SS(Block)		
<b>Factor A</b>	$a - 1$	SSA	MSA	MSA / MS(MPE)
<b><math>MSE_A = A * Block</math></b>	$(a - 1)(r - 1)$	SS(MPE)	MS(MPE)	
<b>Factor B</b>	$b - 1$	SSB	MSB	MSB / MS(StPE)
<b><math>MSE_B = B * Block</math></b>	$(b - 1)(r - 1)$	SS(StPE)	MS(StPE)	
<b>A x B</b>	$(a - 1)(b - 1)$	SS(AxB)	MS(AxB)	MS(AxB) / MS(SPE)
<b><math>MSE_{AB} = A * B * Block</math></b>	$(a-1)(r-1)(b-1)$	SS(SPE)	MS(SPE)	

Note the improved precision in the tests for interaction effects.

## Example of a split-block

(modified from Little and Hills, Chapter 10)

The root yield in tons per acre for each subplot are given in the diagram below.

**Main plot A:** Nitrogen rates, levels N0, 80, 160, and 320

**Subplot B:** Harvest times, levels 1 – 5

		Block I							Block II				
		H4	H5	H1	H3	H2			H4	H2	H3	H5	H1
N80		26.4	29.3	10.1	23.1	18.2	N160		34.2	18.5	22.4	30.3	10.8
N320		31.2	34.2	10.3	25.9	19.2	N0		21.3	12.5	16.7	19.1	5.2
N160		28.0	31.2	10.2	22.3	16.9	N80		29.5	16.9	20.4	26.6	9.5
N0		10.1	11.4	2.3	9.8	8.8	N320		31.9	17.8	22.8	29.2	7.4

Again, we are in situation where the results from two separate models must be combined to produce a complete and correct ANOVA table.

#The ANOVA to test A

```
split_blockA_mod<-aov(yield ~ A_nitrogen + block +
  Error(A_nitrogen:block) +
  B_harvest +
  B_harvest:block +
  A_nitrogen:B_harvest,
  split_block_dat)
```

#The ANOVA to test B

```
split_blockB_mod<-aov(yield ~ A_nitrogen + block +
  A_nitrogen:block +
  B_harvest +
  Error(B_harvest:block) +
  A_nitrogen:B_harvest,
  split_block_dat)
```

**The resulting ANOVA tables:**

Error: A\_nitrogen:block

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A_nitrogen	3	838.3	279.43	7.506	0.066 .
block	1	14.5	14.52	0.390	0.577
Residuals	3	111.7	37.23		

Error: Within

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
B_harvest	4	1898.9	474.7	375.434	1.73e-12 ***
block:B_harvest	4	42.8	10.7	8.459	0.001748 **
A_nitrogen:B_harvest	12	121.0	10.1	7.976	0.000536 ***
Residuals	12	15.2	1.3		

Error: B\_harvest:block

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
block	1	14.5	14.5	1.357	0.30872
B_harvest	4	1898.9	474.7	44.382	0.00144 **
Residuals	4	42.8	10.7		

Error: Within

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A_nitrogen	3	838.3	279.43	220.983	9.19e-11 ***
A_nitrogen:block	3	111.7	37.23	29.441	8.14e-06 ***
A_nitrogen:B_harvest	12	121.0	10.09	7.976	0.000536 ***
Residuals	12	15.2	1.26		

**Combining results into a final ANOVA table:**

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A_nitrogen	3	838.3	279.43	7.506	0.066 .
A:Block	3	111.7	37.23		
B_harvest	4	1898.9	474.7	44.382	0.00144 **
B:Block	4	42.8	10.7		
A:B	12	121.0	10.1	7.976	0.000536 ***
Error	12	15.2	1.3		

**What is the next step in this analysis?  
How would you do it?**

## Trend analysis of the main effects of Nitrogen

For the sake of covering one more concept, let's assume that the A:B interaction was found to be non-significant, thereby justifying *an analysis of the main effects*. In this scenario, notice that the previous *F* test for A\_nitrogen was *almost* significant ( $p = 0.0660$ ).

Because the nitrogen levels are not evenly spaced, it is not very convenient to perform a trend analysis using contrasts. Instead, we rely on multiple regression, using a very simple model to obtain the correct sums of squares:

```
#To perform a trend analysis of A_nitrogen using a multiple regression
#approach, first re-load the dataset and maintain A_nitrogen as a numeric
#regression variable (not a factor); then:
A_nit<-split_block_dat$A_nitrogen
A_nit2<-A_nit^2
A_nit3<-A_nit^3
A_nit4<-A_nit^4
anova(lm(yield ~ A_nit + A_nit2 + A_nit3 + A_nit4, split_block_dat))
```

### The output:

```
Response: yield
```

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A_nit	1	508.21	508.21	<del>8.3005</del>	<del>0.00664</del> **
A_nit2	1	290.19	290.19	<del>4.7396</del>	<del>0.03611</del> *
A_nit3	1	39.90	39.90	<del>0.6517</del>	<del>0.42481</del>
Residuals	36	2204.14	61.23		

While these are the correct sums of squares for the linear, quadratic, and cubic components of the trend analysis (notice they sum to 838.80), the F-tests are incorrect because they use the wrong error term. As we saw from the previous analysis, the correct error term for the main plot effect A\_nitrogen is the A\_nitrogen:Block interaction ( $SS = 111.7$ ,  $df = 3$ ). The correct F and p-values can be obtained using the `customF()` function shown on the next page:

```
#Linear A:
customF(c(508.21,1,111.7,3))
[1] 0.03440893 *
#Quadratic A:
customF(c(290.19,1,111.7,3))
[1] 0.06831985 NS
#Cubic A:
customF(c(39.90,1,111.7,3))
[1] 0.3767054 NS
```

The following function makes custom F tests relatively easy:

```
customF <- function(x) {  
  SS_num=x[1]  
  df_num=x[2]  
  SS_den=x[3]  
  df_den=x[4]  
  Fvalue<-(SS_num/df_num)/(SS_den/df_den)  
  pFvalue<-pf(Fvalue,df_num,df_den,lower.tail=FALSE)  
  print(pFvalue)  
}
```

#To use:

```
#customF(c(SS_num, df_num, SS_err, df_err))
```

The manually-adjusted ANOVA table, featuring the appropriate F tests, shows a significant ( $p < 0.05$ ) linear effect:

```
Response: yield  
      Df  Sum Sq Mean Sq  Pr(>F)  
A_nit   1  508.21  508.21  0.034 *  
A_nit2   1  290.19  290.19  0.068 .  
A_nit3   1   39.90   39.90  0.377 NS  
Block:A   3   111.7   37.23
```

As mentioned above, the study of the simple effects of Nitrogen at each Harvest date and the simple effects of Harvest date at each Nitrogen level would be the appropriate continuation of this study. This same partitioning of the Nitrogen SS could be conducted within that simple effects analysis.



## What about the correct error term for Block?

Consider the following table of expected mean squares for this split-block RCBD:

Source	Expected Mean Square
Block	$\text{Var}(\text{Err}) + 4 \text{ Var}(\text{Block:B}) + 5 \text{ Var}(\text{Block:A}) + 20 \text{ Var}(\text{Block})$
Nitrogen_A	$\text{Var}(\text{Err}) + 5 \text{ Var}(\text{Block:A}) + \text{Fixed\_effect}(A)$
Block:Nitrogen_A	$\text{Var}(\text{Err}) + 5 \text{ Var}(\text{Block:A})$
Harvest_B	$\text{Var}(\text{Err}) + 4 \text{ Var}(\text{Block:B}) + \text{Fixed\_effect}(B)$
Block:Harvest_B	$\text{Var}(\text{Err}) + 4 \text{ Var}(\text{Block:B})$
Nitrogen_A:Harvest_B	$\text{Var}(\text{Err}) + \text{Fixed\_effect}(A:B)$

Block:A is the correct error for A

Block:B is the correct error for B

A synthetic F test is required for Block

**Summary:** Extracting terms from the Error SS for use as appropriate denominators for F tests in split-plot, split-split-plot, and strip-plot designs (the RCBD case):

### Two-way factorial

RCBD	RCBD Split-plot	RCBD Strip-plot
$\text{lm}(Y \sim \text{Block} + A + B + A:B)$	$\text{lm}(Y \sim \text{Block} + A + \text{Block}:A + B + A:B)$	$\text{lm}(Y \sim \text{Block} + A + \text{Block}:A + B + \text{Block}:B + A:B)$
Block A B A:B	Block A Block:A = <b>Error A</b> B A:B	Block A Block:A = <b>Error A</b> B Block:B = <b>Error B</b> A:B
<b>Residual Error</b>	<b>Residual = Error B, A:B</b>	<b>Residual = Error A:B</b>
Block:A Block:B Block:A:B	Block:B Block:A:B	Block:A:B

### Three-way factorial

RCBD	RCBD Split-split plot
$\text{lm}(Y \sim \text{Block} + A + B + C + A:B + A:C + B:C + A:B:C)$	$\text{lm}(Y \sim \text{Block} + A + \text{Block}:A + B + A:B + \text{Block}:A:B + C + A:C + B:C + A:B:C)$
Block A B C A:B A:C B:C A:B:C	Block A Block:A = <b>Error A</b> B A:B Block:A:B = <b>Error B, A:B</b> C A:C B:C A:B:C
<b>Residual = Error A*B*C</b>	<b>Residual = Error C, A:C, B:C, A:B:C</b>
Block:A Block:B Block:A:B } Block:C Block:A:C Block:B:C Block:A:B:C	Block:C Block:A:C Block:B:C Block:A:B:C