

# Split-Plot Experimental Designs

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# Split-Plot Experimental Designs

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

Split Plot or Sub Plot

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# Split-Plot Experimental Designs

- This experiment has two factors: genotype and fertilizer amount.
- Genotype has levels A, B, and C.
- Fertilizer has levels 0, 50, 100, 150 lbs. N / acre.
- Genotype is called the *whole-plot factor* because its levels are randomly assigned to whole plots.
- Fertilizer is called the *split-plot factor* because its levels are randomly assigned to split plots within each whole plot.

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# Definition of Experimental Units in Split-Plot Designs

- Plots are the *whole-plot experimental units* because the levels of the whole-plot factor (genotype) are randomly assigned to plots.
- The split-plots are the *split-plot experimental units* because the levels of the split-plot factor (amount of fertilizer) are randomly assigned to split plots within each whole plot.
- Thus we have two different sizes of experimental units in split-plot experimental designs.

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# Same Treatment Structure in a RCBD

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

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# Same Treatment Structure in a CRD

	Field											
	B	B	A	B	A	C	A	B	A	C	C	C
	50	0	150	100	100	150	50	0	50	100	0	100
	A	A	C	B	B	B	A	C	A	C	B	B
	50	0	50	50	150	50	0	0	100	50	150	0
	C	A	A	A	A	B	A	B	A	B	C	A
	0	0	100	150	0	0	150	150	50	150	100	100
	B	B	B	C	C	C	A	C	C	C	C	B
	50	100	100	150	100	50	150	50	150	0	150	100

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### Which Design to Use?

Consider a CRD if

1. There is equal interest in all treatment comparisons.
2. There is no natural way to group similar experimental units together in blocks.
3. No logistical constraints make complete randomization impractical.

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### Which Design to Use?

Consider a RCBD if

1. There is equal interest in all treatment comparisons.
2. There are potentially important sources of variation that can be controlled by blocking.
3. No logistical constraints make complete randomization within blocks impractical.

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### Which Design to Use?

Consider a split-plot design if

1. Comparisons among the levels of the split-plot factor are of greater interest than comparisons among the levels of the whole-plot factor.
2. Logistical constraints make a CRD or RCBD impractical.

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### Split-Plot Experimental Design

Field

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

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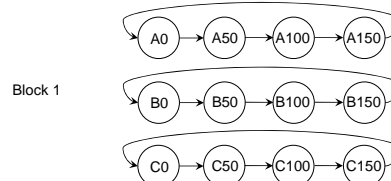
### How would you design the microarray portion of the experiment?

- Suppose the researcher in the split-plot experiment is willing to use 48 two-color microarray slides to measure gene expression in plants from the field.
- The researcher is primarily interested in understanding how gene expression changes in response to fertilizer rate within each genotype.

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### One Possible Design

Use a loop design to compare split-plot experimental units assigned adjacent fertilizer amounts within each whole-plot experimental unit.



Consider the same loop designs for blocks 2, 3, and 4 perhaps reversing loop directions for two of the four blocks.

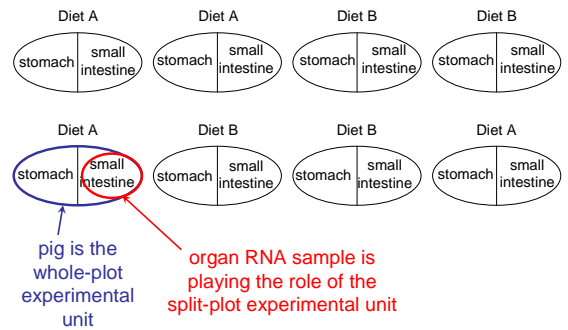
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Split-plot design structures don't necessarily involve plots in the usual sense.

- Suppose a total of 8 pigs are assigned to two diets (A and B) using a completely randomized design with 4 pigs per diet.
- After 4 weeks on the assigned diet, each pig is sacrificed and RNA samples are taken from the stomach and small intestine of each pig.
- Affymetrix GeneChips are used to measure gene expression in each RNA sample with one GeneChip for each of the samples.

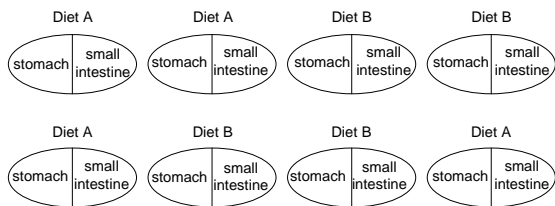
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### Conceptual Picture of the Experiment



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### Conceptual Picture of the Experiment

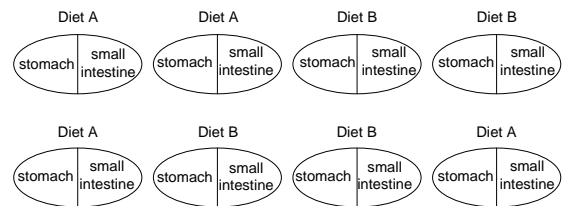


Diet is the whole-plot factor with levels A and B.

Organ type is the split-plot factor with levels stomach and small intestine.

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### Conceptual Picture of the Experiment



Note that the levels of the factor *diet* are randomly assigned to experimental units.

That is not the case for the levels of the factor *organ type*, but we will analyze it and refer to it just as we would for an experimental factors whose levels are randomly assigned.

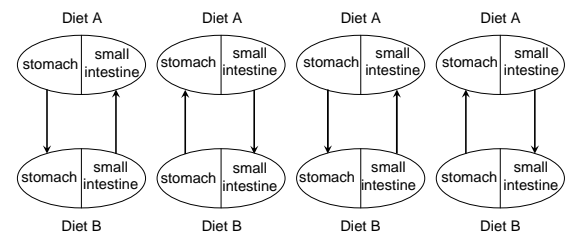
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### How would you design the microarray portion of the experiment?

- Suppose the researcher in this split-plot experiment can afford to use 8 two-color microarray slides to measure gene expression in the RNA samples.
- The researcher is primarily interested in understanding how diets affect gene expression within each organ type.

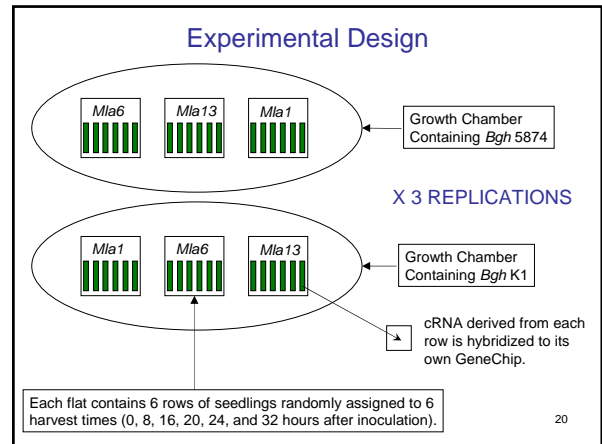
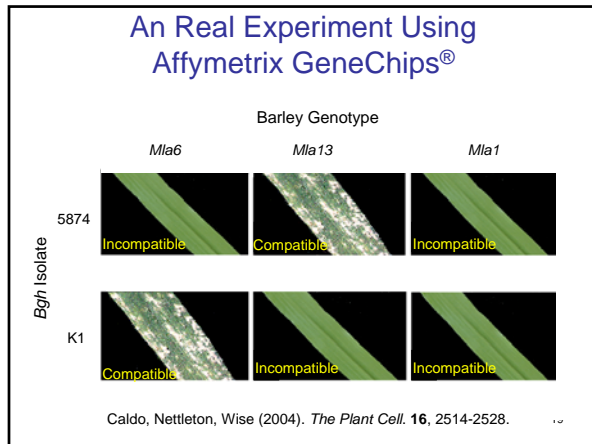
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### One Possible Design



Connect samples of the same organ type between pigs treated with different diets because this will give us the tightest comparison between diets for each organ type (and this is the comparison of primary interest).

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This is a split-split-plot experimental design.

- Whole-plot experimental units are groups of three flats (trays containing plants).
- Split-plot experimental units are flats themselves.
- Split-split-plot experimental units are rows of seedlings within flats.

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This is a split-split-plot experimental design.

- Whole-plot treatment factor is *isolate* with levels 5874 and K1.
- Split-plot treatment factor is *genotype* with levels Mla1, Mla6, and Mla13.
- Split-split-plot treatment factor is *hours after inoculation* with levels 0, 8, 16, 20, 24, and 32.

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