

Experimentation and Statistics in Tropical Agriculture: Planning Experiments

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Summary

- General considerations.
- Some history.
- The experimental method.
- Three techniques of experimental designs.
- Standard types of designs.
- Experimental validity

Variability



Variability is a characteristic of biological material.



General considerations

• Experiments

- involve researchers manipulating situations, by applying treatments, in an effort to draw conclusions about what their manipulations have caused;
- represent a very important technique in the acquisition of scientific knowledge.

- **Surveys and observational studies** merely observe some aspect(s) of the world as is.



From the left: unknown, W.G. Cochran, unknown, C.R. Rao, Irvin, G. Rasch, S.C. Pearce, R.A. Fisher, visiting the Campinas Agronomic Institute, 1955.

Some history



The **Hoosfield long-term experiment** on spring barley at Rothamsted Research started in 1852. Spring barley has been grown continuously since then. It tests nitrogen, minerals, farmyard manure (FYM) and sodium silicate.

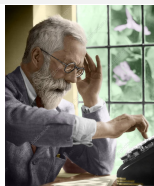
Some history



The **Park Grass (Broadbalk) experiment** was established in 1856 at Rothamsted Research. Designed to measure the effects of fertilisers on yields of permanent grass cut for hay. It is now regarded as the foremost long-term ecological experiment in the world.

Some history - Fisher

- Fisher introduced the subdivision of sums of squares now known as an **analysis of variance (anova) table** (1923),
- derived the exact distribution of the (log of the) **ratio of two independent chi-squared variates** (1924),
- introduced the **principles of randomization and blocking**: the completely randomized (CRD) the randomized complete block (RCBD), Latin square (LSD), and split-plot (SPLD) experiments (1925),
- promoted **factorial experiments**, and the notion of confounding (1926).
- He quickly transformed **agricultural experimentation** in Great Britain and more widely.
- These ideas have remained the statistical basis of agricultural experimentation.



Basic purposes of experimentation

- To provide valid comparison of the effects of treatments.
- To provide valid information about the relationship between variables of interest.

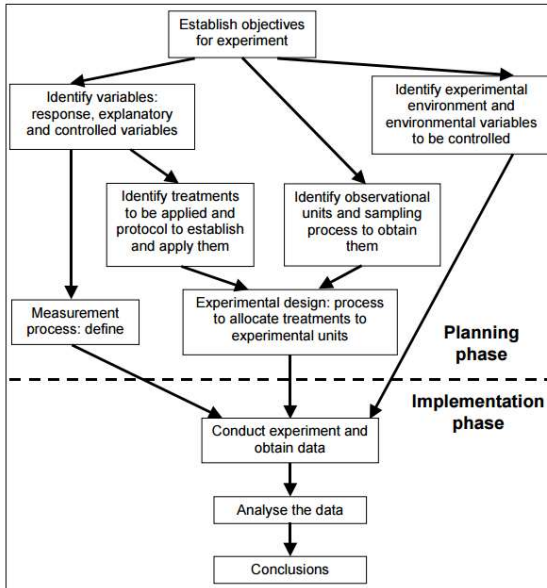
Famous phrases

- It is easy to conduct an experiment in such a way that no useful inference can be made. **William Cochran & Gertrude Cox**
- And so it that . . . borne upon me that very often, when the most elaborate statistics refinements possible could increase the precision by only a few percent, yet a different design involving little or no additional experimental labour might increase the precision two-fold, or five-fold, or even more. **Ronald Fisher**

Basic requirements

- The experimental conditions should represent the situational conditions of the **problem of interest**.
- The comparison of treatments should be free from other possible explanations due to the presence of other variables. (**confounding**)
- The treatment comparison should be made with as little influence of **random variation** as possible.
- The **level of uncertainty** in the conclusions should be assessable.
- The experiment should be as **simple** as we can make it.
- The **data analysis** should follow the planning.

The experimental method (Brien, 2010)



The experimental method (cont.)

Planning

- Needs to be a result of a **collaboration** between a Scientist and a Statistician.
- Ideally, this happens in plenty of time before the experiment.
- The Scientist does not come with statistically precise questions and the Statistician has to ask lots of questions.
- A **clear understanding of the problem** being studied and the **objectives** for the experiment. Why is the experiment being done?
- To answer specific questions:
 - **to estimate something** (how much heavier are pigs on diet A than on diet B?) – we want **unbiased estimators** with small variance;
 - **to test a hypothesis** (that there is no difference between organic and inorganic fertilizers) – we want high **power**.



The experimental method (cont.)

- **Identification of the variables** of interest including the response (dependent) variable, the explanatory (independent) variable (or factor) to be studied and the other variables to be controlled.
- **Identification of the experimental environment** and how this is to be controlled (representative farmer's fields or a well-controlled station?).
- **Identification of the experimental/observational units** (number, shape, ...) to be used and the sampling that is to be done to obtain the observational units taking into account the need to control natural variation (in many experiments the observational and experimental units are the same).
- **Identification of the treatments** to be applied and the protocol for establishing and applying them (Which treatments? What quantities? What combinations?)

The experimental method (cont.)

- **Allocating the treatments to the experimental units** through an experimental design, using blocking to overcome any major variation in the experimental units..
- **Identification of the measurement process** to be used and controlling the key sources of variation that might affect the measurement process.

Constraints

- **Costs.**
- **Availability of treatments** (for example, not many seeds of a new variety).
- **Availability of experimental units** (e.g. plots in fields, people to test equipment, time ...).
- **Natural “blocks”** or divisions among the experimental units.
- Other constraints imposed by **management**.

The experimental method (cont.)

Implementation

- **Carrying out the experiment.**
- **Collecting the data**, implementing any control protocols recognized as necessary for this step and paying attention to any unusual aspects of this step.
- In **collaboration** with the Scientist, design a paper for collecting data (or an electronic equivalent, data-logger).

Pig	Farm	Feed	...	Weigth at ...	in kg	...	empty columns
1	1	A	
2	1	B	
3	1	C	refused to eat
4	2	A	

- Encourage the Scientist to record all the relevant data as soon as possible, with **NO copying**, **NO changing** the order, **NO intermediate calculations**, **NO leaving to juniors**.

The experimental method (cont.)

- **Analysing the data**

- Look over the data sheets for obvious anomalies and ask the Scientist to explain any dubious data, before he forgets.
- should be planned at the design stage, but can be modified in the light of unforeseen circumstances.
- in principle, we should know how to do this by hand (using a calculator) but in practice we use suitable software.

- **Drawing conclusions** and acting on them in relation to the original problem.

- ANOVA tables, tables of means and standard errors, p-values may not mean much to the Scientist.
- the Statistician must explain what this means in the Scientist's context.

Soya experiment

A randomized block design experiment with 3 blocks was conducted to investigate the effects of two factors (3 sources of phosphorus fertilizer and 3 methods to fertilize) on the yield of soya. The plots were areas of 5.4m^2 (6 lines of 1.8m separated by 0.5m).



- **Objective:** to investigate the effects of two factors (sources of phosphorus and methods of application of fertilizers) on the yield of soya measured by weight of grains ($\text{dag}/5.4\text{m}^2$).
- **Observational/Experimental units:** areas of 5.4m^2 (6 lines of 1.8m separated by 0.5m)
- **Response variable:** weight of soya grains ($\text{dag}/5.4\text{m}^2$).
- **Variable for local control:** 3 blocks (different countours of the land).
- **Treatments:** All possible combinations of the levels of the factors leading to nine treatments.

Soya experiment (cont.)

- **Explanatory variables (factors):** There were two treatment factors in this experiment and each factor was tested at three different levels. The treatment factors and their levels were:
 - sources of phosphorus: Superfosfato (90kg/ha of P_2O_5), Fosfato de Olinda (90kg/ha of P_2O_5), Superfosfato + Cloreto de Potássio (90kg/ha of P_2O_5 + 90kg/ha of K_2O);
 - methods to fertilize: spreading the fertilizer, in the row by the seed, in the row near the seed.
- **Experimental design:** A randomized block design with 3 blocks and 9 treatments.
- **Measurement process:** yield of soya measured by weight of grains (dag/5.4m²).
- **Analysis:** by graphical analysis and analysis of variance.
- **Conclusions:** A number of conclusions are drawn about the effect of the sources of phosphorus, the methods to fertilize and their interaction on the yield of soya.

Definition of some key terms

- **Factor:** the explanatory variable(s) manipulated or set by the experimenter; can be quantitative and qualitative.
- **Levels of a Factor:** the values that an individual factor takes.
- **Treatment:** the complete description of what will be applied to an experimental unit; a combination of one of the levels from each of the factors and this combination is applied to particular experimental units.

Soya experiment: two factors with three levels each, resulting in nine treatments

- Sources of phosphorus: Superfosfato (90kg/ha of P_2O_5), Fosfato de Olinda (90kg/ha of P_2O_5), Superfosfato + Cloreto de Potássio (90kg/ha of P_2O_5 + 90kg/ha of K_2O).
- Methods to fertilize: spreading the fertilizer, in the row by the seed, in the row near the seed.

Definition of some key terms (cont.)

- **Experimental unit:** the smallest unit to which a single treatment could be randomly allocated.
- **Observational unit:** the native physical entity that yields a single value of the response variable (the smallest unit on which a response will be measured). In our case it is a plot since it produces a single value of each of the response variable – the total weight of grains.
- **Note 1:** In many experiments, the observational and experimental units are the same.
- **Note 2:** An **Experimental unit** can give rise to many **Observational units**.

Soya experiment:

- **Observational/Experimental units:** areas of 5.4m^2 (6 lines of 1.8m separated by 0.5m).

Examples of Experimental units

- a plot in a land;
- a vase with one plant or three plants, or a group of vases;
- a Petri dish with ten seeds;
- a patient in a hospital (called a subject);
- a lump of dough;
- an animal or a group of animals in a pen;
- a specific run on a machine with given conditions;
- a batch of raw material.



Foto: Embrapa

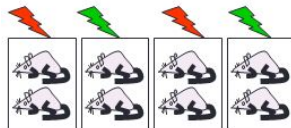


Teratology experiment

- In a teratology experiment the pregnant female is treated with the test compound (red) or a placebo (green), in a CRD.
- The pregnant females are killed at about mid-gestation and the pups are weighed, measured and studied for abnormalities.



- In case 1, the animals are all housed in **one cage** and the treatment is given by injection.
- Any two animals can receive different treatments, the animal is the experimental unit and N , the total number of subjects is 8.



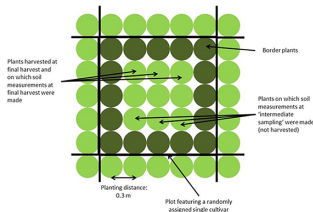
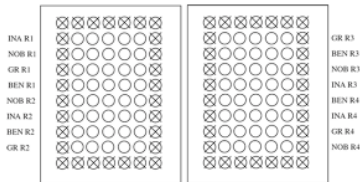
- In case 2, the animals are housed **two per cage** and the treatment is given in the food or water.
- What do you think is N , the total number of experimental units in this case?

Border and core-plot or net plot

- In some types of experiments it is necessary to have **borders** or 'guard areas'.
- If **borders** are necessary the plot consists of a **core-plot** (surrounded by the border) and the total-plot, which includes the border.
- There may be additional uncropped pathways between the plots for access.
- The plot yield can be measured as the sum of the individual plants in the core-plot or as the total yield of the core-plot without separating individual plants.
- In some circumstances measuring the yield of the total-plot may also be useful to assess the effect the border had on the research design.



Border and core-plot or net plot (cont.)



- Graphical representation of the experimental design.
- Crossed drops were left out because of the border effect.
- R1-4 indicate the four replicates per cultivar.

- The experimental set up was a RCBD, each block with 150 plots.
- Two plots per block were left empty for measurements in bare soil.
- Note bare plots were planted to 25 (5x5) plants of the same cultivar.
- Plants at the beginning and end of each row were considered guard plants.
- Nine (3x3 inner) plants of the net plot, measured.

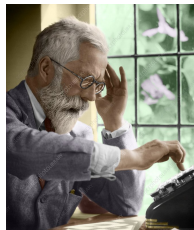
Definition of some key terms (cont.)

- **Replications**: the number of times that each treatment is tested.
 - **Soya experiment**: three replicates (three blocks).
 - **Teratology experiment**: four (case 1) and two (case 2) replicates.
- **Local control (blocking)**: dividing up the experimental units into blocks of alike units.
 - **Soya experiment**: three blocks (different countours of the land).
 - **Teratology experiment**: no local control (CRD).
- **Response variable**: what is measured, can be quantitative (continuous, discrete), qualitative or semi-quantitative.
 - **Soya experiment**: weight of soya grains (dag/5.4m²).
 - **Teratology experiment**: weight, measures for abnormality.

Three key techniques of experimental design

- **The three key basic principles**, introduced by Fisher in 1935, are:

- **replication**,
- **randomization**,
- **blocking**.



- Why use statistical principles in the design of experiments?
 - The short answer is: because of the **uncontrolled variation**.

Uncontrolled variation is the variation between units treated as similarly as possible that arises from all the minor differences which we are unable to control.

Uncontrolled variation

Uncontrolled variation is the variation between units treated as similarly as possible that arises from all the minor differences which we are unable to control.

Soya experiment: The differences can likely be caused by a large number of small uncontrollable differences, viz. slight differences in

- environment - ambient temperature, soil conditions (fertility, acidity, humidity etc), pests, diseases etc;
- raw materials - slight differences in seed (seedling) condition;
- management regimes.

Teratology experiment: slight differences in initial weight, genetic background, health condition of the animals.

Confounding

Two effects are said to be **confounded** when it is not possible to separately estimate them.

- The problem of the confounding of treatment effects with uncontrolled variation is widespread in the biological, physical and social sciences.
- How does one overcome it?

Teratology experiment: the Treatment effect is confounded with animal effect.

Answer: Use statistical principles: *Replication*, *Randomization* and *Blocking*, in the design of the experiment.

Note: We do not eliminate uncontrolled variation, rather we adopt strategies that enable us to live with it.

Replication

Replication

- provides a measure of uncontrolled variation;
- it is the application of each treatment several times, i.e., to several experimental units.

Note:

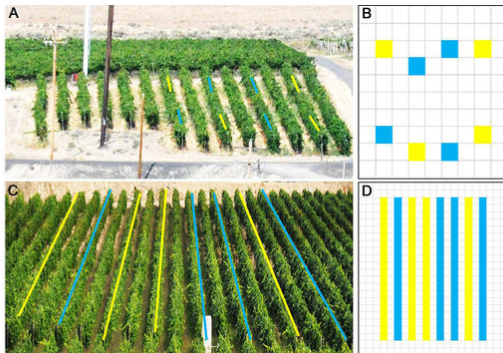
- The spread in the replicate observations provides a measure of uncontrolled variation.
- Compare total spread with spread from uncontrolled variation alone to decide if treatment has an effect.

Randomization

- **Problem:** While a measure of uncontrolled variability can be obtained, the observed treatment difference might be caused by systematic effects.
 - To overcome systematic effects - **Randomization**.
- **Randomization**
 - is the assignment of treatments to experimental units so that every unit has the same probability of receiving each treatment.
 - results in an arrangement with no particular pattern.
 - The simplest statistical design involving randomization is the **Completely Randomized Design (CRD)**.

CRD Vineyard experiment

- A CRD with four replicates of a single treatment (yellow lines) and a control (blue lines) in (A) a small and (C) a mid-size vineyard block.
- Note: Both examples show buffer rows between treatments.
- Sometimes the same treatment may end up in adjacent rows.
- Schematic diagrams of the trials shown in A and C are shown in B and D, respectively.



Why to randomize?

- **to overcome systematic effects** – e.g. an engineering PhD student tested all replicates of Machine 1 in January (Winter!), then he tested all replicates of Machine 2 in March (Spring!);
- **to avoid selection bias** – e.g. a doctor selects the most healthy patients to receive his favorite treatment;
- **to avoid accidental bias** – e.g. the technician takes rats from the cage one by one and gives the first treatment to the first six rats;
- **to stop experimenter cheating** (for good or bad) – sometimes an experimenter wants to make easier for the technician or the statistician;
- **to force the experimenter to tell the truth** – statistician and experimenter agree on a design; statistician randomizes, gives to the experimenter; experimenter says “I cannot do that because ...”

Completely randomized design (CRD)

- A **completely randomized design** is one in which each treatment occurs a specified, possibly unequal, number of times.
- Suppose that there are t treatments and that treatment i is applied to r_i plots (is replicated r_i times). Then $\sum_{i=1}^t r_i = n$.
- If there is no need to group the plots into blocks the treatments should be applied to the plots at random.
 - (i) Number the plots $1, 2, \dots, n$
 - (ii) Write down a systematic design:
put treatment 1 on plots $1, 2, \dots, r_1$
put treatment 2 on plots $r_1 + 1, r_1 + r_2 + 1, \dots, r_1 + r_2, \dots$
 - (iii) Choose a random permutation of $1, 2, \dots, n$ and apply it to the design.

CRD potato experiment

Consider five varieties with three, five, three, three and four replicates, respectively, to be randomized to 18 plots.

Using library `dae` (Brien, 2018) from R, the result might be as follows.

Systematic design and permutation:

St. O.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
Perm.	2	7	17	6	9	11	14	18	4	10	12	3	5	16	1	8	13	15
Var.	A	A	A	B	B	B	B	B	C	C	C	D	D	D	E	E	E	E

Plan or layout (give to the Scientist)

Plots	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
Varieties	E	A	D	C	D	B	A	E	B	C	B	C	E	B	E	D	A	B

A layout in the field

- The resulting arrangement is one with no particular pattern.

E	C	A	C	E	D
A	D	E	B	B	A
D	B	B	C	E	B

CRD

Unrandomized factor: Plots

1	2	3	4
5	6	7	8
9	10	11	12

A layout in the field, after randomization of treatments (randomized factor) to plots

A	C	A	C
B	A	C	A
B	C	B	B

ANOVA Table

Source	df	SSq	MSq	F
Plots	$n - 1$			
Treatments	$t - 1$			
Residual	$n - t$			

Blocking (local control)

- If the plots are not all reasonably similar, we should group them into blocks in such a way that plots within each block are reasonably similar, in order to improve the experiment by reducing amount of variability affecting the treatment differences.
- Not absolutely necessary but very important because it allows some control of uncontrolled variation.

Blocking is the grouping of experimental units into groups called **Blocks**, the units within a group being as similar as possible.

- If possible
 - 1 blocks should have the same size;
 - 2 blocks should be big enough to have each treatment at least once.

Types of blocks

(i) Natural discrete divisions

- young animals – litters;
- people or animal – sex;
- plots in a rice – paddy – irrigation groups;
- industrial process – batches of chemical (of raw material);
- consumer experiment – tester, week;
- lab experiment – technician, day, bench.



(ii) Continuous gradient of change – can choose boundaries for homogeneity

- plots in a field – small compact areas;
- people or animals – age or weight or size;
- clinical trial – severity of a disease;
- long-term trial – time, periods.



Types of blocks (cont.)

(iii) Blocking for managing the experiment

- clinical trial – doctor or a nurse;
- agricultural field trial – long thin area for tractor operation.

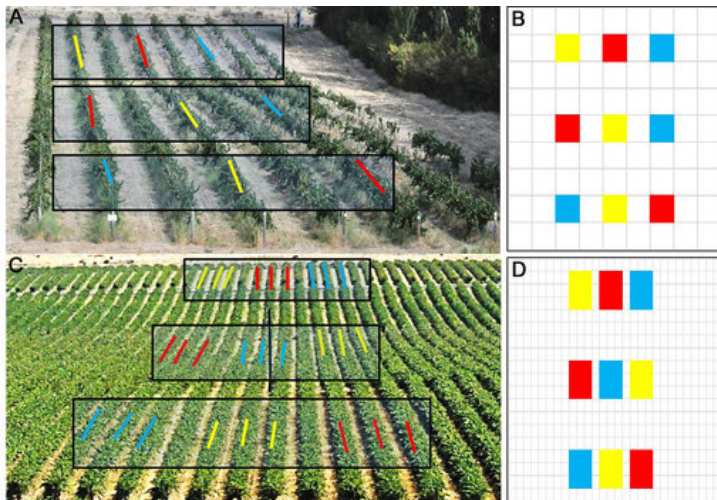
Important:

- Everything (sowing, spraying, sampling, harvesting, measuring, injecting) done during the experiment needs to be done block-by-block, in case of interruptions, replacement of staff, improvements in technique etc.
- Sometimes you may need more than one sort of block.

RCBD Vineyard experiment

- RCBD with three replicates of two treatments and one control.
- The black frames in A and C represent the grouping of different zones that was done to account for variability due to slope.
- In this design, the treatments and control (red, blue, and yellow lines) are randomized within each zone.
- In the smaller field design (A), two to three vines in a single row might be the replicate unit.
- In a larger design (C), three to five vines across multiple rows might serve as the replicate unit.
- Schematic diagrams of the trials shown in A and C are shown in B and D, respectively, where individual cells represent a single vine.
- Photos and illustrations by Hemant Gohil.

RCBD Vineyard experiment (cont.)



Obtaining a layout for an RCBD in R

To obtain randomized layouts we need to:

- (1) Apply the treatments to the plots in Block 1, and randomize, just as for a completely randomized design.
- (2) Repeat for each block, using a fresh randomization every time.

Potato RCBD experiment: The effects of three varieties (A, B and C) of potato are to be investigated. The experiment will be conducted using 4 blocks (different areas) with 3 plots and randomizing the 3 varieties to the 3 plots in each block.

A layout in the field, using package `dae` (Brien, 2018)

- The blocks are columns.

C	B	C	B
A	A	B	A
B	C	A	C

RCBD

Unrandomized factors:
Blocks, Plots (Plots nested
within Blocks)

Block 1	1	2	3
---------	---	---	---

Block 2	1	2	3
---------	---	---	---

Block 3	1	2	3
---------	---	---	---

A layout in the field, after random-
ization of treatments (randomized
factor) to plots within blocks

Block 1	B	C	A
---------	---	---	---

Block 2	C	A	B
---------	---	---	---

Block 3	C	B	A
---------	---	---	---

ANOVA Table

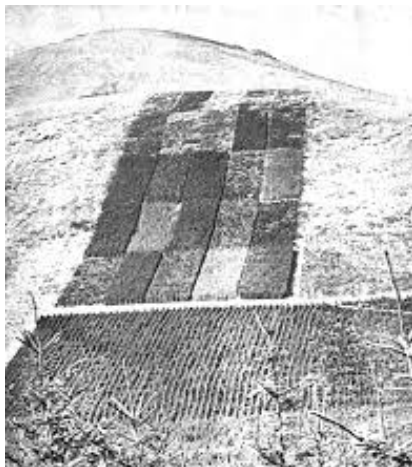
Source	df	SSq	MSq	F	Prob
Blocks	$(r - 1)$				
Plots[Blocks]	$r(t - 1)$				
Treat	$t - 1$				
Residual	$(r-1)(t-1)$				

Latin Square

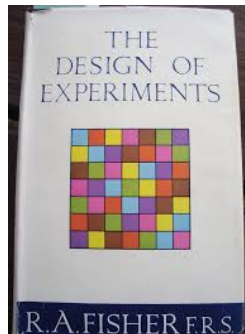
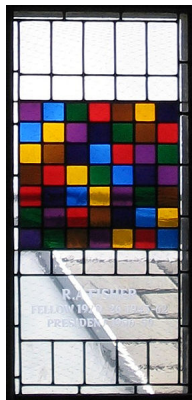
- Sometimes we need more than one type of blocks. In general call one sort of blocks “rows” and the other sort “columns”.
- A **Latin square design (LSD)** is one in which
 - each treatment occurs once and only once in each row and each column;
 - so that the numbers of rows, columns and treatments are all equal.
- Note that:
 - Clearly, the total number of observations is $n = t^2$.
- For example, suppose in a field trial moisture is varying across the field and the stoniness down the field.
 - A **Latin square** can eliminate both sources of variability.

LS tree species experiment

The first field experiment in the world to use a randomized experimental design established by the Forestry Commission in March 1929, on a hillside near Beddgelert Forest, designed by Fisher. (© The Forestry Commission)



Latin Square and Fisher



- A stained glass window in Caius College, Cambridge;
- photograph by J. P. Morgan.
- This Latin square was on the cover of the first edition of The Design of Experiments.
- Why this one?
- It does not appear in the book.

Obtaining a layout for an LS in R

- randomly select one of the systematic LS designs for a value of t ;
- randomly permute the rows and then the columns;
- randomly assign letters to treatments.

A layout in the field, using
package `dae` (Brien, 2018)

B	E	D	A	C
D	B	A	C	E
A	D	C	E	B
C	A	E	B	D
E	C	B	D	A

LS

Unrandomized factors: Rows, Columns (Rows, Columns crossed)

5 × 5 Latin Square

		Column				
		1	2	3	4	5
Row	I	I,1	I,2	I,3	I,4	I,5
	II	II,1	II,2	II,3	II,4	II,5
	III	III,1	III,2	III,3	III,4	III,5
	IV	IV,1	IV,2	IV,3	IV,4	IV,5
	V	V,1	V,2	V,3	V,4	V,5

Less
stony of
field



Stonier
end of
field

Less
moisture



More
moisture

A layout in the field, after randomization

5 × 5 Latin Square

		Column				
		1	2	3	4	5
Row	I	A	D	C	E	B
	II	C	A	E	B	D
	III	B	E	D	A	C
	IV	E	C	B	D	A
	V	D	B	A	C	E

Less
stony of
field



Stonier
end of
field

Less
moisture



More
moisture

ANOVA Table

Source	df	SSq	MSq	F
Rows	$t - 1$			
Columns	$t - 1$			
Rows:Columns	$(t - 1)^2$			
Treatments	$t - 1$			
Residual	$(t - 1)(t - 2)$			
Total	$t^2 - 1$			

6x6 Latin Square at Rothamsted



Design of factorial experiments

- Often, the experimenter is interested in more than one factor.
- Experiments that involve more than one randomized or treatment factor are called **factorial experiments**.
- In general, the number of treatments in a factorial experiment is the product of the numbers of levels of the treatment factors.
- The disadvantage of this is that the number of treatments increases very quickly.
- Given the number of treatments, the experiment could be laid out as
 - a Completely Randomized Design,
 - a Randomized Complete Block Design or
 - a Latin Square with that number of treatments.
- The incomplete block designs, such as BIBDs or Youden Squares are not suitable for factorial experiments.

Advantages of factorial experiments

Relative to **one-factor-at-a-time experiments**, **factorial experiments** have the advantages that:

- if the factors interact, factorial experiments allow this to be detected and estimates of the **interaction effect** can be obtained, and
- if the factors are independent, factorial experiments result in the estimation of the main effects with greater precision.

Design of split-plot experiments (SPLD)

- Designs in which main effects confounded with more variable units such as large plots.
- Their defining attribute is that there is randomization to two different physical entities such that some main effects are randomized to the more variable entities.
- The **standard split-plot design** is one in which two factors, say A and B with a and b levels, respectively are assigned as follows:
 - one of the factors, A say, is randomized according to a RCBD with say r blocks and
 - each of its ra plots, called the **main plots**, is split into b **subplots** (or split-plots) and levels of B randomized independently in each subplot.
 - Altogether the experiment involves $n = rab$ subplots.
- That is, the generic factor names for this design are Blocks, MainPlots, SubPlots, A and B.

Split-plot principle

- Very flexible principle that can be used to generate a large number of different types of experiments.
- For example, the main plots could be arranged in any of a CRD, RCBD, Latin square, balanced incomplete block design (BIBD), Youden Square
 - each plot of the design is subdivided into subplots.
- The subplots may utilize more complicated designs as well.
 - For example, the main plots may be arranged in a RCBD each of which are subdivided in such a way as to allow a Latin Square to be placed in each main plot.
- Also, subplots can be split into subsubplots and subsubplots into ...
- Nor is one restricted to applying just one factor to each type of unit.
 - More than one factor can be randomized to main plots, more than one to subplots and so on.
- The standard split-plot design is nearly the simplest possibility; only a CRD in the main plots would be simpler.

When to use a split-plot design

1. When the physical attributes of a factor require the use of larger units of experimental material than other factors.
 - For example, land preparation treatments usually require to be performed on larger areas of land than do the sowing of different varieties (due to the different pieces of equipment).
 - Temperature control for storage purposes involves the use of relatively large chambers in which several samples can usually be stored.
 - Different processing runs are often of a minimum size such that their produce can be readily subdivided for the application of further treatments.
 - Also, some factors are relatively hard to change. For example, the temperature of a production operation is often difficult to change so that it might be better to change it less often by making it a main-plot factor.

When to use a split-plot design (cont.)

2. When it is desired to incorporate an additional factor into an experiment.
3. When it is expected that differences amongst the levels of certain factors are larger than amongst those of other factors.
 - The levels of the factors with larger differences are randomized to main plots.
 - One effect of this may be to increase the precision of comparisons between the levels of the other factors.
4. When it is desired to ensure greater precision between some factors than others.
 - Irrespective of the size of the differences between the main plot treatment factors, it is desired to increase the precision of some factors by assigning them to subplots.
 - One may be less interested in main effects of some factors. A particular example of such factors is “noise” factors.

Designing a standard split-plot experiment

- In the standard split-plot, the main-plot treatment factor, A, is randomized to main plots and the subplot treatment factor, B, is randomized to subplots.

Production rate experiment

- Suppose that one is interested in comparing 3 methods of work organization and 3 sources of raw material on the production rate of a certain product.
- Decided that four factories are to be used in the experiment and that each factory is to be divided into three areas.
- The methods of work organization are to be assigned at random to areas.
- Each area is to be subdivided into 3 parts and the source of raw material for each part is obtained by randomizing the three sources to the three parts.

A layout for RCBD Production data

A layout in the field, using package dae (Brien, 2018)

Randomization of the three Methods to the three Areas (Mainplots) within Factories (Blocks)

		Subplots		
		1	2	3
Blocks, MainPlots	1,1	1	1	1
	1,2	3	3	3
	1,3	2	2	2
	2,1	3	3	3
	2,2	2	2	2
	2,3	1	1	1
	3,1	1	1	1
	3,2	2	2	2
	3,3	3	3	3
	4,1	2	2	2
	4,2	3	3	3
	4,3	1	1	1

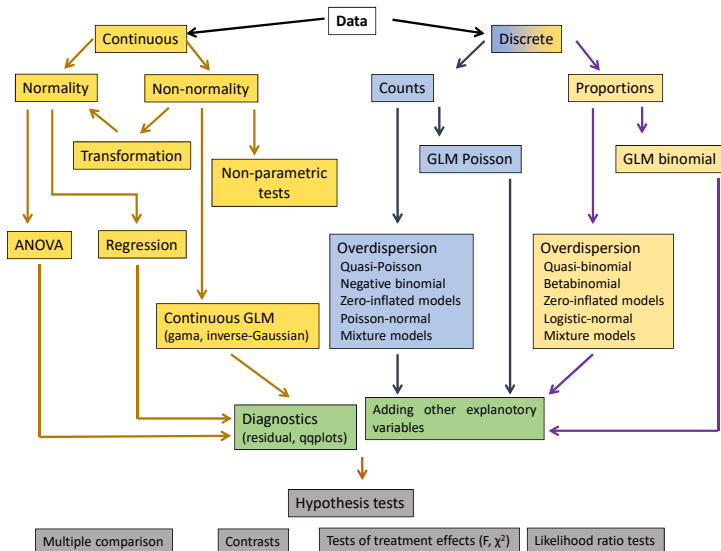
Randomization of the three sources of raw material to the three Parts (Subplots)

		Subplots		
		1	2	3
Blocks, MainPlots	1,1	3	2	1
	1,2	1	2	3
	1,3	1	3	2
	2,1	2	3	1
	2,2	1	2	3
	2,3	2	1	3
	3,1	2	3	1
	3,2	1	3	2
	3,3	3	2	1
	4,1	3	2	1
	4,2	3	1	2
	4,3	3	2	1

Experimental validity

- The assessment of the **quality of an experimental design** requires knowledge of the factors that influence or cause variation in the measured outcomes.
- With the potato experiment, what factors affect yield and what can we do about these factors in the design of the experiment?
- Two concepts help with this:
 - **Internal validity**: conclusions can be appropriately drawn from within this experiment about the relationship between the independent and dependent variable.
 - **External validity**: conclusions from the experiment can be appropriately generalised to a wider situation of interest.

Data analysis



Final remarks

- Get statistical thinking involved early when you are preparing to design an experiment!
- Getting well into an experiment before you have considered these implications can be disastrous.
- Think and experiment sequentially.
- Experimentation is a process where what you know informs the design of the next experiment, and what you learn from it becomes the knowledge base to design the next.

To call in the statistician after the experiment is done may be no more than asking him to perform a post-mortem examination: he may be able to say what the experiment died of!

'Presidential Address by Professor R. A. Fisher, Sc.D., F.R.S. Sankhya: The Indian Journal of Statistics (1933-1960), 4: 14-17, 1938'.

References

Brien, C.J. (2010) Design and randomization-based analysis of experiments in R. URL: <http://chris.brien.name/ee2>.

Brien, C.J. (2018). `dae`: Functions Useful in the Design and ANOVA of Experiments. R package version 3.0-23.
<https://CRAN.R-project.org/package=dae>

Fisher, R.A. (1926) The Arrangement of Field Experiments. J. Minist. Agric. 33: 53-513.

Fisher, R.A. (1935). The Design of Experiments. Oliver and Boyd, Edinburgh.