A Manual of Crop Experimentation

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Preface

For some time the Applied Statistics Research Unit of the University of Kent at Canterbury has been arranging courses in statistics for scientists engaged in agricultural research. The first of these was a three-week course run on-site for the staff of the Bangladesh Agricultural Research Council twice in 1984. After further development this course was next given at the International Institute of Tropical Agriculture, Nigeria, in 1985. The course was then developed again and expanded, resulting in a three-month course, given in Canterbury in 1985 and 1986, and hopefully to be repeated on an annual basis from now on. This manual now makes the material covered in these courses generally available. The four authors are the lecturers who participated in the earlier courses overseas and who form part of the larger lecturing team for the Canterbury-based course.

A central feature of these courses has been their thoroughly practical nature, and there has been a deliberate policy of mixing agronomists and biometricians together. The philosophy here has been that it is instructive for each group to be aware of the problems that the other faces, and this interaction has been found to work well in practice. The text reflects the content of the courses: it can either be used as a manual to complement such courses, or can be used as a text in its own right. All stages of the manual are regarded as parts which go together to make up a whole.

The sections of the manual have been developed out of the handouts used to complement the course lectures. Each section is therefore fairly complete in itself, though some have been expanded for this text to emphasize the inter-sectional relationships as much as possible. The sections have been placed in a natural order so that the reader may go through the material in logical sequence, from start to finish, if the intention is to do this rather than use the manual as a work of reference.

A number of people, in addition to the authors, have contributed in one way or another to this manual. In particular thanks go to Dennis Cooke and Michael Kenward for helpful comments from reading earlier versions of the text, and also, in the case of Michael Kenward, for help with the production of solutions to exercises. Byron Jones, Byron Morgan and Andrew Rutherford are also to be thanked for their advice and comments. Thanks go as well to the secretarial staff of the Applied Statistics Research Unit for their patient and painstaking typing of the text throughout its

various versions, and especially to Margaret Wells for her organizational work in addition in connection with the courses.

It is hoped that this will be the first of a series of texts arising from courses in Applied Statistics developed at the University of Kent. Further information on these and on the Applied Statistics Research Unit can be supplied on request.

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Chapter 1

How experiments are conducted

'... and a most curious country it was.... The ground ... was divided up into squares....'

Lewis Carroll, *The Adventures of Alice Through the Looking Glass*, Chapter 2

1.1 Introduction

Anyone who visits a crop research institute—and even more, anyone who flies over one—will see, like Alice, that the ground is divided into squares or, at least, into rectangles. That is because the comparison of different treatments (whether they are different forms of fertilization or anything else) requires that they be grown as close together as possible. It is true that an experimenter could apply each treatment in a different field. He could then reasonably claim that they were being applied as a farmer would apply them, but if there were only one field for each treatment there would be a major difficulty in interpretation. Fields differ for many reasons. There would be no way of knowing whether the differences observed between the fields of an experiment would have been there anyway or were due in part to the various treatments applied.

To avoid that fault another experimenter might go to the opposite extreme. He could take a small area that appeared to be uniform and then divide it into smaller areas, the basic units known as 'plots'. Then each of his treatments could be applied to a selection of plots, chosen so as to be a fair sample of the whole of the experimental area. He might reasonably claim to have made a sound comparison of the various treatments. Nevertheless, he would be open to another criticism, namely, that his plots were so small that his treatments were not realistically applied. Deep ploughing, for example, could have become double digging and the knapsack sprayer could have replaced the spraying machine. A good experimenter should avoid both extremes. In principle there is no objection to using complete fields as plots, but it does lead to an experiment of immense size with plots that are basically very different one from another. If the plots are too small, on the other hand, they may indeed be

comparable but it is not possible to apply treatments to them in the same way that a farmer would apply them in practice. Also, data collected from very small plots cannot properly be used to represent performance in a larger area. (It is not enough to multiply by the ratio of the areas because there will be edge-effects, which may be important in a small plot but negligible in a large one.) Some compromise between the extremes is called for.

In Chapter 2 we shall examine practicalities in the field, how to choose plot size, for example, how to apply treatments and how to record data. Then in Chapter 3 we shall look at the statistical ideas needed to deal with the sort of data that will result. Thereafter we shall look at some of the special problems that will arise. Most of the ideas and problems arise from the need to specify precisely the objectives of a study before an experiment is started in the field. We shall consider that in Chapter 5.

1.2 Replication, randomization and local control

Replication

There are several characteristics that are regarded as essentials of good experimentation. One has already been mentioned, namely, the use of plots of reasonable size. Another is 'replication', i.e. the use of several plots for each treatment. If there is only one for each, there will be no way of deciding how accurately the experiment has been conducted. If there are several, the differences between them will show how much uncontrolled natural variation there is in the measurements. In an agricultural context that is important. The chemist in his laboratory can forecast with some certainty how accurate his determinations will be, but in the field nothing is certain until it has happened. In a dry year some plots will suffer worse than others. That will introduce differences between plots, quite apart from those brought about by the treatments. In a wet year, or a windy one, the pattern of good or bad land could be very different. Also in some years, damage from birds, insects, fungi, etc. will be an additional source of variation, the extent of which could not have been estimated and whose pattern could not have been foreseen. For such reasons the precision of each new experiment has to be assessed separately. An experienced person learns to make rough forecasts of precision, but whether those forecasts will be right or not must depend upon the weather and other disturbing influences.

Randomization

That leads to another important characteristic of a good experiment, namely 'randomization'. It is in any case needed for reasons of objectivity.

If the experimenter can declare that plots were allocated to treatments by the use of random numbers or by the shuffling of cards, we shall be reassured that the best plots were not deliberately selected for some favoured treatment. There is however a more important reason for randomizing, namely, the determination of precision. Where there is replication, i.e. there are several plots for each treatment, a comparison of those plots will show how much variation has been introduced by differences in the soil, differences in the plant material, inaccuracies of measurement, etc. At this point care is needed. Confronted with the task of allocating four treatments to twelve plots, most people would disperse those for each treatment, i.e. they would opt for something like this:

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

However, that will not do. It is true that dispersal leads to the four samples of plots, three for A, three for B, etc., being on average as similar as possible, but it maximizes the differences between them. In fact, although dispersal leads to the area being sampled better for the comparison of treatments, by a paradox it leads to a high estimate of the sampling error. The contrary fault arises when plots for a treatment are kept together, e.g.

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

Here each treatment samples the total area in a most ineffective manner, but because the plots of any one treatment are close together, they will give more uniform results than plots in general. The paradox is resolved when plots are allocated to treatments at random. (It is true that each of the systematic allocations just considered has a a small chance of turning up by a chance randomization, but that possibility can be accepted.) Ways of randomizing will be discussed in Section 2.6.

Local control

One last point concerns the control of local variation. If the area allotted to the experiment shows marked systematic differences, e.g. it slopes or there is a water-course down one side of it, most people would rightly be suspicious of any scheme in which those differences were ignored. (The matter will be examined further in Section 1.8.) The favoured device is to divide the land into 'blocks', not necessarily all of the

same size, each block uniform within itself, or, at least, as uniform as can be contrived. That is called a 'block design'. If no systematic differences are apparent and if none are known from past experience with the land, the plots can be allocated at random within the total area. There is then only one block, not several. In that case the design is said to be 'completely randomized'.

The analysis of data

We shall now direct our thoughts to one topic that underlies many others. Having allocated our plots to the treatments and having measured yield or some other important quantity on each plot, what are we going to do with the data? Our answer is going to determine much else. We shall have to record data bearing in mind what is to be done with them, and we shall have to interpret data bearing in mind what was recorded in the field. The method we shall describe is called the 'analysis of variance'.

It should be emphasized that what follows in the next two sections concerns arithmetic and nothing more. The real skill lies not in the calculation of the analysis of variance, a task that is essentially mechanical and best left to computers, but in the interpretation, which requires careful thought. The approach to data based on the analysis of variance is one of great flexibility and subtlety and one that illuminates a wide range of problems. We cannot begin to use it until we can perform the calculations, but that is only a step on the way. The arithmetic is not an end in itself, necessary though it is.

1.3 The analysis of variance for an orthogonal block design using sweeping

The measurement of variability

Before illustrating the approach it may be helpful if we explain how statisticians measure variability. Always there must be a 'hypothesis' to say what is expected. We will take a very simple case. Someone has declared that if we were to weigh a lot of similar objects we should find that they all weighed 30 grams. Now that we have a clear-cut statement of what to expect we can set to work and see if it is true. We will suppose that we start and get values of 30, 30, 30 (excellent) and then we get 31 (no need to bother much) then 30, 30 and then 29 (again, no need to worry) followed by 30, 29, 30 but then we get 33. At that point we do begin to wonder whether the hypothesis really holds, or perhaps needs to be more complicated. Let us examine our reactions and see if they were justified. First of all, we regarded 29 and 31 in much the same light; we disregarded the signs. Then, we took a deviation of 3 as being much more serious than

three deviations of 1. How far was that reasonable? At this point the mathematicians come to our aid by telling us that we should really be looking at the squares of our deviations. We were therefore quite right to equate deviations of +1 and −1 and to regard a single value of 3 as much more important than three of 1. The position will be examined further in Chapter 3, beginning at (3.3.1).

In most statistical work experimenters are continually setting up hypotheses, taking some data and then asking how far the data deviate from what the hypothesis says they should be. The next step is to aggregate those deviations by squaring each one and adding the squares. In the analysis of variance we first work out deviations assuming that there is no effect of treatments. Then we allow for the treatments and get a new set of deviations. They are of special importance, both in practice and in the theory of the subject, so they have a special name. They are called the 'residuals'—but they are deviations just the same. Finally we ask if the sum of squared deviations ignoring the treatments is so very different from the sum of squared deviations when we do take treatments into account i.e., the sum of squares of the residuals.

Orthogonality

A block design is said to be orthogonal if all the blocks are made up in the same way with respect to treatments. (Treatments do not have to be equally replicated.) To take an example, the following design is orthogonal:

Block	I	A	A	B	C	D	E
	II	A	A	B	C	D	E
	III	A	A	B	C	D	E

(Note that the plots would of course have to be allocated at random within each block and not disposed systematically as here.)

A completely randomized design has only one block. Its data can be dealt with as if the design were orthogonal.

The method of sweeping

A body of data is 'swept' when each value is reduced by an appropriate mean. For example, in the following data

I	A	22	B	17	C	21	
II	C	26	A	17	B	23	(1.3.1)
III	C	20	B	15	A	22	

there are three causes of variation to be considered: two of these are blocks and treatments. The third comes from a lot of small sources, such as, for example, the effects of variable land, variable plant material, the inevitable inaccuracies of measurement and much else. Those combined sources are often described collectively as the 'error' of the experiment, but the term is rather harsh, implying as it does that someone has been at fault. Some prefer to call the last component the 'residual variation', i.e. what is left when block and treatment differences have been allowed for, and that name has its critics also. Here we shall use 'error', the quotation marks implying an inexact use of the word. Such a design as that at (1.3.1), in which each treatment occurs once in each block, is said to be in 'randomized blocks', or more correctly 'randomized complete blocks'.

Additivity

To amplify what has been said, the data of a block design are thought of as made up of three components. First, there is the inherent fertility of the block in which the plot occurs. (It will be recalled that blocks were chosen to be as uniform as possible in that respect.) We shall write that component, b_i, where i is the number of the block. Then there is the effect, t_j, brought about by the plot receiving Treatment j. (That is really what we want to know about.) Finally, there is the residual, e, which sums up the various sources of variation comprised in 'error'. Each plot has its own e and it should not be much affected by the treatment applied to the plot, which is a matter of chance. (That assumption will have to be examined later, especially in Chapter 9, but for the moment we merely state it.) In the analysis of variance we assume that the three components can just be added, i.e., a datum equals

$$b_i + t_j + e \quad \text{for the plot}$$

The assumption of 'additivity', as it is called, serves well enough in most cases, any deviations from it being absorbed in e. Provided those deviations are small, little harm results.

First we will note the block means, i.e., I, 20; II, 22; III, 19. Sweeping by them gives:

I	A, +2	B, −3	C, +1
II	C, +4	A, −5	B, +1
III	C, +1	B, −4	A, +3

These quantities are called 'deviations'. It will be seen that in any block they sum to zero. The effect of blocks has now been removed, so they

represent the action of treatments and 'error'. Squaring the deviations and adding gives 82, which will be called the 'stratum total sum of squares'. (Stratum in this instance means the variation of plots within blocks. The term will be explained further in Section 1.9.) In all this it is necessary to carry enough decimal places, though not so many as to become a burden. The best number will be considered in Section 1.10.

Treatment means are now A, 0; B, −2; C, +2. Sweeping by them gives:

I	A, +2	B, −1	C, −1	
II	C, +2	A, −5	B, +3	(1.3.2)
III	C, −1	B, −2	A, +3	

These quantities are called 'residuals'. They sum to zero over any block or any treatment and represent only the action of 'error'. The sum of their squares is 58, called the 'error sum of squares'.

Degrees of freedom

We have now found two sums of squares, one for the stratum total and one for the 'error', but how would we expect them to be related? Since the residuals were derived using a more adaptable hypothesis than that which led to the deviations (i.e., allowance was made for a possible effect of treatments), we should expect them to give a better fit, but better by how much? Here a note on degrees of freedom may be helpful. Originally there were nine data with no constraints upon them. They were then swept by three quantities (the block means), also unconstrained, leaving six degrees of freedom for treatments and 'error' combined. When they were swept the second time, however, the three treatment means necessarily summed to zero. Consequently only two of them can be regarded as independent; once two are known, the third follows. That reduced the degrees of freedom for 'error' alone to four.

The position is this: if p quantities are under study and there are no constraints upon them, there are p degrees of freedom. If however there are h constraints, there are only $(p - h)$ degrees of freedom because only that number of values can be assigned at will. The other h have to be chosen to satisfy the constraints. If Tom, Dick and Harry are each asked to give a number, those numbers collectively have three degrees of freedom, provided there is no collusion between them and provided they are free to choose whatever number they like. Suppose though that their choice of numbers is part of a trick by an entertainer and Harry, who comes last, is an accomplice told always to choose a number that will make the total come to 21. Tom says 6, Dick says 9 so Harry says 6.

Despite appearances there are only two degrees of freedom. So it is with the treatment means. That for A is 0 and that for B is -2. Since all three must sum to zero that for C must be $+2$, but that is a forced move and contributes nothing to the degrees of freedom.

In a block design with b blocks, v treatments and n plots, the stratum total, derived as it is from sweeping n data by b block means, will have $(n-b)$ degrees of freedom because the block means are unconstrained. Sweeping by treatments takes out another $(v-1)$, leaving $(n-b-v+1)$ for the 'error'. If $n = bv$, which corresponds to the case of randomized blocks, there are $(b-1)(v-1)$ degrees of freedom for 'error'.

The analysis of variance table

The analysis of variance is presented below. The treatment line was found as the difference between that for the stratum total, in which treatments have been ignored, and that for the 'error', in which they have been allowed for.

Source	d.f.	s.s.	m.s.	F
Treatments	2	24	12.0	0.83
'Error'	4	58	14.5	
Stratum total	6	82		

(1.3.3)

In each line the mean square (m.s.) is found by dividing the sum of squares (s.s.) by the degrees of freedom (d.f.). The F-value is the ratio of the treatments and 'error' mean squares. It is sometimes called the 'variance ratio' or VR.

To clarify nomenclature, the 'error' mean square is more usually called the 'error' variance and will be written as s^2. Its square root is called 'the standard error of an observation'. The standard error expressed as a percentage of the grand mean is called 'the coefficient of variation'.

1.4 The analysis of variance for an orthogonal design using summation terms

An alternative method is to use 'summation terms'. First the data are written out, together with the block totals, treatment totals and the grand total, i.e.

	A	B	C	
I	22	17	21	60
II	17	23	26	66
III	22	15	20	57
	61	55	67	183

Four summation terms, S, S_b, S_t and S_o, are formed thus:

S, the total term, equals the sum of squares of the data, i.e. $22^2 + 17^2 + \ldots + 20^2 = 3817$

S_b, the block term, equals the sum of squares of the block totals, divided by the block size, i.e. $(60^2 + 66^2 + 57^2)/3 = 3735$

S_t, the treatment term, equals the sum of squares of the treatment totals, divided by the treatment replications, i.e. $(61^2 + 55^2 + 67^2)/3 = 3745$

S_o, the grand total squared, divided by the number of plots, i.e. $183^2/9 = 3721$ (the 'correction term').

Then the treatment sum of squares equals $S_t - S_o = 24$, the 'error' sum of squares equals $S - S_b - S_t + S_o = 58$ and the stratum sum of squares equals $S - S_b = 82$.

In the example the calculations are simplified because all blocks contain the same number of plots and all treatments are equally replicated. If those conditions do not hold, each total on being squared should be divided by the number of data it contains. Strictly speaking,

$$\begin{aligned} S_b &= 60^2/3 + 66^2/3 + 57^2/3 = 3735 \\ S_t &= 61^2/3 + 55^2/3 + 61^2/3 = 3745 \end{aligned} \qquad (1.4.1)$$

1.5 Residuals

The method of sweeping has two main advantages. It is essentially simple and therefore adapted to computers. Also, it isolates the residuals and therefore enables an experimenter to see which plots are making large contributions to 'error'. It also has a serious disadvantage in requiring a lot of arithmetic for what it achieves. For that reason it is not generally used except for special reasons. With a computer, simplicity of arithmetic is more important than brevity, so many computer packages proceed by sweeping, and an understanding of the method will sometimes help in reading the output.

Residuals are very important. They represent that part of the variation between plots that cannot otherwise be explained. If the 'error' mean square of an experiment has been unusually large, it is helpful to go over the experiment plot by plot and see where the large residuals lie. Clearly something has happened that requires consideration. Large residuals can result from poor technique, untried and little-understood equipment, inexperienced observers, source of material and much else. Also, things can go wrong. If the large residuals are all in a group, that suggests a good or bad patch in the field that was not allowed for in the choice of blocks. Another possibility is that they all occur in one treatment, perhaps one with a large mean. In that case, it might be better to use some other scale of measurement (see Chapter 9). Various other possibilities exist. In general an experimenter just wants to know about the 'error' mean square, but the figures in (1.3.2) are more informative because they show where it came from.

As has been said, an advantage of the sweeping method is that it gives estimates of the residuals. If residuals are required when summation terms have been used, they can still be obtained, though by a laborious method. The analysis of variance may have been calculated more easily but the whole will have been more difficult.

In an orthogonal block design, the error sum of squares is given by $S - S_b - S_t + S_o$. That will serve as a mnemonic of a residual, which equals

$$\text{datum} - \text{block mean} - \text{treatment mean} + \text{grand mean}.$$

Thus, for the plot in Block II with Treatment C, the residual is

$$26 - 66/3 - 67/3 + 183/9 = +2$$

which is the value found at (1.3.2).

1.6 Row-and-column designs

There are occasions when it is not possible to associate all the differences in the field with a single system of blocks. For example, there could be a slope and that would call for blocks along the contours to allow for the many effects associated with altitude, e.g. depth of soil, exposure, and much else. In addition, there might be a stream down one side of the experimental area and that would call for blocks across the contours, i.e. at right angles to those already used, to allow for water effects. In that case, it might be best to have two sets of blocks crossing one another, known as 'rows' and 'columns'.

The commonest row-and-column design is the Latin square, in which each treatment occurs once and once only in each row and each column, e.g.

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

A row-and-column design is said to be orthogonal if each row is made up in the same way with respect to treatments and each column likewise. An obvious example is a Latin square, but the definition includes a design like this one:

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

or one like this:

A	B	B	A
B	A	B	A
A	B	A	B
B	A	A	B

Whatever row-and-column design is chosen, before use the rows should be permuted at random and then the columns likewise.

In an orthogonal row-and-column design much the same procedure applies as for an orthogonal block design in sweeping. The steps are as follows:

(1) Sweep by rows.
(2) Sweep the resulting values by columns to obtain deviations.
(3) Sum the squares of the deviations to give the stratum total. (We recall that the stratum is that of plots within both rows and columns.)
(4) Sweep by treatments to obtain residuals.
(5) Sum the squares of the residuals to give the 'error' sum of squares.

Applying the method of summation terms to a row-and-column design, five are required, i.e.

S, the total term derived from the data
S_r, the row term derived from row totals
S_c, the column term derived from column totals
S_t, the treatment term
and S_o, the correction term.

The treatment sum of squares equals $S_t - S_o$, as before. The stratum total sum of squares is $S - S_r - S_c + S_o$, i.e. $(S - S_o) - (S_r - S_o) - (S_c - S_o)$. That leaves $S - S_r - S_c - S_t + 2S_o$ for 'error'.

If there are r rows, c columns and v treatments, there will be rc plots. Sweeping by rows leaves $(rc - r)$ degrees of freedom. Sweeping further by columns leaves $rc - r - (c - 1) = (r - 1)(c - 1)$ for the stratum total. Since there will be $(v - 1)$ degrees of freedom for treatments, there will be $(rc - r - c - v + 2)$ for 'error'. If, as in a Latin square, $r = c = v = k$, that equals $(k - 1)(k - 2)$.

For a row-and-column design a residual equals

Datum − row mean − column mean − treatment mean + 2(grand mean) (1.6.1)

If only a few residuals are needed, perhaps after the analysis has been worked out, (1.6.1) is convenient. If all are needed, it would have been easier to have swept in the first place.

1.7 An examination of assumptions

The method of sweeping shows clearly some of the assumptions being made in the analysis of variance. First, it implies that the effect of a treatment is much the same wherever it is applied. That can scarcely be strictly true. For example, if a range of new strains is being tested, the hope being to find some that are drought-resistant, and if the land is of varying moisture, it is not to be expected that the assumption will hold precisely. In fact, small departures are absorbed into 'error'. Nevertheless, fields vary in different ways and it would be better to allocate them so that the variability any one exhibits is not of the same kind as the treatments to be applied. Thus one with variable water would be a particularly bad site for a trial of drought-resistant strains.

To take the matter further, it is assumed that the effects of treatments are much the same whatever the block. That is the assumption of additivity already mentioned in Section 1.3. If blocks have been formed with the intention of keeping dry and wet plots together, the difficulty of the last paragraph has merely been transferred. Also important is the assumption that the effect of rows is the same whatever the column. Given a diagonal streak of good or bad land across the area, the assumption fails disastrously and a large 'error' will arise for no apparent reason.

Another assumption, though a much less obvious one, concerns the residuals, which are combined to give a common 'error' mean square as if all were subject to the same sources of variation. Clearly, that would not be so if the area contained a very variable patch. (It is a good practice to avoid hazards of that kind, even if it does mean leaving gaps between blocks so that the patch is not used. It may be sown and harvested, though not as part of the experiment.) The assumption also implies that the residuals are normally distributed (see Section 3.1). That can be important. For example, only with normally distributed quantities is the estimate of the 'error' mean square independent of the mean. For many of the tests that will be described in Chapter 3, the assumption is essential for complete validity.

One way of dealing with all these difficulties is to use 'transformations' (see Chapter 9). For example, the total weight of plants commonly fails the assumptions, but not if it is transformed to its logarithm.

1.8 Blocks, rows and columns

The traditional way of controlling local variation in a site is to divide the land into blocks such that the main differences lie between them. Then each block is fairly uniform within itself. Sometimes that is easier said than done, though there are times when it can be accomplished without difficulty. To use examples already given, the site may slope, or there may be a stream down one side. More often there is no particular reason to form blocks one way rather than the other and that can be awkward. If there are marked differences in the land but it is not clear where they lie, blocks could even do harm, because their boundaries could cut across contours of fertility and encapsulate in each block a wide range of good and bad areas. General experience suggests that in most cases square blocks are beneficial, but even that cannot always be relied upon either. Another difficulty is that some sources of local variation, e.g. bird damage, wind damage, areas of impeded drainage, may appear in a certain season but not in others, so they cannot readily be allowed for in the design of the experiment.

With a small experiment, i.e. one with about 16 plots or fewer, it is usually not a good idea to use blocks at all unless there are obvious sources of local variation that need to be controlled. Using blocks diminishes the number of degrees of freedom for 'error'. (There should be at the very least six or eight, though 20 are desirable.) In a small area there is probably not much local variation to control and the loss of error degrees of freedom could be serious.

In a large experiment the situation is different, even if only because blocks are needed for purposes of administration. Where operations like

the application of treatments, harvesting and data recording are likely to take more than one day, block boundaries are needed to provide a stopping place. If the district is one subject to unpredictable rain, even more block boundaries are needed in case someone should start an operation in the morning and be driven off by rain later in the day. With a large experiment there is not the same need for conserving 'error' degrees of freedom. Also, there is likely to be more local variation to control. There is therefore greater advantage in having blocks, but it is still desirable to know the fertility patterns before deciding how they should be formed.

It should be emphasized that blocks have to fit the local variation or they are no good. For that reason they may have to be of different shapes. Also, they do not have to be of the right size to take each treatment once. Sometimes the site is such that that is not possible. In that case it may be that a non-orthogonal design should be preferred (see Chapter 4). Its data will be less easy to analyse, but the important consideration is to have blocks that will control the local variation. If blocks are ineffective the experiment may be of little value anyway. If such is the case, simplicity or complexity of statistical analysis are scarcely a relevant issue.

As has been said, it is sometimes necessary to use two sets of blocks at right angles, as in a Latin square. Although row-and-column designs have their place, they should not be used unless there is a good reason for doing so. In general row-and-column designs do control local variation better than block designs, but when they fail the result can be disastrous. Also, just as block designs leave fewer degrees of freedom than those that are completely randomized, two blocking systems leave even fewer. That could be serious.

Row-and-column designs have one advantage that can be relied upon. Sometimes there is fear that the outside plots will be different from the rest. Those on one side catch the wind while those on the other are sheltered. All outside plots may do rather better than inside ones on account of having more space. In such circumstances the first and last row and the first and last column can pick up a lot of variation that in a block design would become part of the 'error'. Even so, a better solution would be to provide an adequate discard area round the experimental area.

1.9 Strata

When we have got our plant material ready and we look at the land, we have to make plots that can be compared one with another. We shall not get a good experiment if we compare two treatments, one on good land and the other on bad, or if we plant well-growing bushes for one and stunted bushes for the other. We have to find groups within which

comparisons can fairly be made with the added requirement of randomization to avoid favouritism. In statistical parlance, we are seeing our material in 'strata' within which we can work. So far we have looked at one approach. We assumed that our plants (or seeds or cuttings) were fairly similar and the chief lack of uniformity lay in the land. We therefore divided it into blocks and we were proposing to make our comparisons between plots within blocks. That is in fact a good way of going to work; we can show what we are doing by writing

Total area → Blocks → Plots

That means that there are two 'strata':

Blocks/Total area Plots/Blocks

Usually the treatments would be applied to plots, and comparisons would be made within blocks; that is, to work in the second stratum, but there would be nothing wrong in applying some treatments to complete blocks and comparing them within the area as a whole. To do that would be to work in the first stratum.

In the first stratum, i.e. Blocks/Total area, a deviation equals

Block mean − Grand mean

In the second, i.e. Plots/Blocks, it equals

Plot value − Block mean

If there had been only one block, i.e. if the design had been completely randomized, the second of these would have been

Plot value − Grand mean

and the first would not exist.

Nevertheless there are other strata possible, as the rest of this section will show. The need is to find a unit to which the treatments can be applied at random but in a practical way, and to group those units so that good comparisons can be made between them. For example, each plot could be divided further into sub-plots, giving this scheme:

Total area → Blocks → Plots → Sub-plots

In the third of the strata so formed, i.e. Sub-plots/Plots, a deviati

Sub-plot value − Plot mean.

In the second, i.e. Plots/Blocks, it equals

Plot mean − Block mean

and in the first, i.e. Blocks/Total area, it equals

Block mean − Grand mean. (1.9.1)

Note that it is now the sub-plots that are being measured, the value for a plot being the mean of its sub-plot values. What we have done is to add an additional stratum, namely, Sub-plots/Plots, to the two of a block design.

It would be permissible to apply some treatments to sub-plots and to make comparisons within plots. Other treatments could be applied to plots within blocks. That might be a useful device. For example, if it were a matter of studying different depths of ploughing in conjunction with different levels of fertilization, the ploughing treatments could only be applied in long narrow plots that ran across the field, but it would be possible to divide those strips into sub-plots, which could receive the fertilization treatments. Such a design is said to be in 'split-plots'. That possibility will be examined in Sections 7.1 to 7.5. With a row-and-column design there are basically three strata:

Rows/Total area
Columns/Total area
Plots/(Rows and columns)

Usually only the last is used, but it is permissible to apply treatments to, say, the rows and to make comparisons within the total area. It is not usually done because there are not enough rows, but occasionally there are enough for them to be treated differently. Such a design can be written

Total area → (Rows × columns)

The deviations for the three strata are respectively

Row mean − Grand mean
Column mean − Grand mean
Plot value − Row mean − Column mean + Grand mean.

The matter can be taken further. For example it would be possible to

divide the plots into sub-plots, as was done with the block design. That gives

Total area → (Rows × columns) → Sub-plots

and again an additional stratum has been added at the end, namely Sub-plots/Plots, with deviations equal to

Sub-plot value − Plot mean. (1.9.2)

Also, it would be possible to have a number of row-and-column designs, say, Latin squares, all with the same treatments but at different sites, the whole being regarded as a single experiment. In that case it would be possible to apply different treatments to the squares and to make comparisons within the whole, always assuming that there were enough sites to make that useful. Such a scheme could be written:

The whole → Sites → (Rows × columns).

The strata are now: Sites/The Whole, Rows/Sites, Columns/Sites, Plots/(Rows × columns), with deviations respectively.

Site mean − Grand mean
Row mean − Site mean
Column mean − Site mean
Plot value − Row mean − Column mean + Site mean.

The point being made is simply this: so long as local control is effected either by 'nesting' areas of different size, e.g.

Plots/Blocks

or by crossing them, e.g.

(Rows × Columns)/Total area

it is always possible to break down the total deviations, i.e.

Plot value/Total area

into component deviations that add up to that total. It is then possible to intervene with treatments in any stratum. Hence, the choice of design is not restricted to those in blocks or to those in rows and columns, but

extends to any stratum. A fuller description will be given in Section 7.5. In all cases, however, the general procedure is the same. First the deviations are calculated for each plot (or sub-plot if that is the smallest area). The deviations are squared and added to give the stratum total. They are then swept by treatments to give the residuals, which are themselves squared and added to give the 'Error'.

Change of strata

Once made, strata should not be altered. Here a distinction should be made between the analysis that is to be used for the interpretation of the data and other alternative analyses calculated as a guide to future blocking. As far as the first is concerned it is important to use the blocks of the original design. There is an old adage, 'As the randomization is, so should the analysis be'. The randomization was appropriate to the blocks originally chosen and the true analysis is determined by that. When the experiment has been completed the residuals may suggest that the blocks had been formed in an unfortunate manner. It is quite legitimate to insert alternative blocks and analyse again to see if that would have given a smaller 'error' mean square. (That may well require the computational method set out in Section 4.2 because the design with modified blocks is unlikely to be orthogonal.) If a reduced error is indeed found that could be a guide for the future; the fact must not alter the interpretation of the data as they will be presented in a report. The reason, as has been said, lies in the randomization. It was made so that the 'error' mean square for the original design should be estimated correctly, i.e., without bias. The same randomization will not do the same for some other blocking system. Consequently the determination of error for the alternative design will not be strictly correct, though it should be good enough to show if there is indeed a marked superiority in the second way of blocking.

A distinction is here being made between the interpretation of data for the purposes of the experiment, i.e., to clarify the doubts that led to its inception, and studies to see if it could have been designed better, i.e., a 'post-mortem'. As to the latter, we strongly recommend that past data should be used to provide information for future designs. At the least the 'error' mean square should be noted to help decide the size of later experiments of the same kind. Also, if there was argument at the design stage as to how the experiment should have been designed, it is wise to look at the data using the rejected blocking system to see if it would have made any difference. All that is desirable and sound practice. What is wrong is trying this and trying that until pleasing results are obtained. In any case, the alternative analyses are not strictly correct, so it would not be right to present conclusions based upon them.

The matter chiefly arises when blocks have proved to be very similar and someone wants to omit them. The randomization was within blocks and the analysis should allow for them. A similar question arises when the columns of a row-and-column design prove ineffective and someone wants to use only one blocking system, that of rows. Again the analysis must follow the randomization. That is not to say that hints for future designs should be ignored; a lot can be learnt by looking at past analyses in a constructive way, but that is a different matter.

1.10 Precision in calculations

In writing down the calculations needed in an analysis of variance, it is important to strike a balance between giving too many decimal places, which is wasteful, and not giving enough, which leads to rounding error.

It is also advisable to write everything down neatly with the decimal points one below the other and successive digits all on the same spacing. Sometimes one sees two quantities to be added written like this:

```
14.88888889
1 2 . 3 7  5
```

That is to invite mistakes. First a decision should be made as to the number of decimal places and then everything should be written consistently. If four places are needed, the figures should be written like this:

```
14.8889
12.3750
```

Then everyone can see what is intended.

Also, in a statistical context one should beware of the bias introduced by rounding all final digits of 5 in the same direction. If the figures are:

13.25 14.50 12.75 12.25 13.00

their true sum is 65.75. If only one decimal place is needed, they should not become

13.3 14.5 12.8 12.3 13.0

or 13.2 14.5 12.7 12.2 13.0

because those sets sum respectively to 65.9 and 65.6. It is better always to round to a number that ends in an even digit, i.e.

13.2 14.5 12.8 12.2 13.0

The total is now 65.7, which is nearer the true value. (Some people always conclude with an odd digit. One way is as good as the other; it is just a matter of convention.)

We can now pass to the number of decimal places needed in the calculations. The first task is to decide the number of decimal places really needed in the data. That will be written as e and rules for finding it appear below. Once that is decided, everything else follows. Deviations and residuals should be worked out to $(e + 2)$ places, sums of squares, mean squares and summation terms to $(2e + 2)$, treatment means in a report to either e or $(e + 1)$, and standard errors to $(e + 1)$ or $(e + 2)$ respectively [(1.10.1)]. In the case of the 'error' mean square it is desirable to have at least four significant digits. The value of F should be given to two decimal places unless the value is very large.

The rule works well but it depends upon the correct determination of e, i.e. the number of decimal places required in the data themselves. (Note that e is the number of places needed, not the number available. Sometimes recorders give too many or too few, but that does not affect the argument.)

In general the data should be given to a precision that allows for at least 20 possible values or preferably 30. For example, if we are given data to one decimal place that lie between 7.2 and 9.6 they allow for 25 possible values, which is rather low but acceptable, so we can put e equal to 1. (It would have been the same if the range had been 107.2 to 109.6 or, for that matter, 1797.2 to 1799.6.)

If the data had in fact been given only to the nearest unit, all would have been 7, 8, 9 or 10, and we should have to point out that that is not good enough. (With visual scores we might have to accept such coarse grading, but we should avoid it when we can.) If, on the other hand, they had been given more precisely—if, for example, they had ranged from 7.21 to 9.58, giving 238 possible values—we would still have e equal to 1 for the calculated figures, but we could retain the additional decimal place in the data themselves. (The same comment applies when we get data presented to the nearest half or quarter, e.g. data that range from $47\frac{1}{2}$ to 74. Here it would have been good enough to have measured to the nearest unit, so we put $e = 0$, but we do not have to discard the fractions if they are there.)

It is possible for e to be negative. If the range went from 7932 to 8316, it would have been good enough if they had been measured to the nearest ten, so we put $e = -1$.

A difficult case arises when we seem to be part way between two values of e; for example, if we are confronted with data that lie between 49.2 and 65.1. If we take e to be 1. i.e. if we accept the decimal place, we are

allowing for 160 possible values, which is more than enough. If we take $e = 0$ that allows for only 16, which is too few. When the higher value of e gives more than 75 possible values, it is permissible, though not essential, to give e a value half-way between the two values. It should however be understood that doubtful cases will be decided in favour of the higher precision. In the above case e could be taken as $\frac{1}{2}$. That makes $e + 2 = 3$, $2e + 3$, $e + 1 = 2$, and so on.

To take an example, if the data of a randomized block design are correctly written with $e = 1$, as here

	Block	I	II	III	IV
Treatment	A	14.6	15.3	14.0	16.6
	B	15.3	16.4	15.4	17.1
	C	13.9	14.0	14.6	15.2

the deviations should be written with three decimal places $(e + 2 = 3)$

0.000	+ 0.067	− 0.667	+ 0.300
− 0.700	+ 1.167	+ 0.733	+ 0.800
+ 0.700	− 1.233	− 0.067	− 1.100

which gives a sum of the squared deviations of 6.7933 with four places $(2e + 2 = 4)$.

The residuals, also to three places, are

+ 0.075	+ 0.142	− 0.592	+ 0.375
− 0.150	+ 0.317	− 0.117	− 0.050
+ 0.075	− 0.458	+ 0.708	− 0.325

The sum of their squared values is 1.4783, also with four places; that is the 'error' sum of squares. Those values give a treatment sum of squares of 5.3150. The mean squares are 2.6575 for treatments and 0.2464 for 'error'. Since the 'error' mean square has four significant places (i.e. 2, 4, 6 and 4) it can be left as it is. If the number of significant places falls below four, an extra decimal place is called for. The value of F should be written 10.79.

Using summation terms, they should be written with four decimal places, $(2e + 4)$, i.e.

Total	2784.8400
Block	2778.0467

Treatment	2777.7950
Correction	2772.4800

Adding and subtracting them will be easier and less liable to error if the corresponding figures range neatly one above the other. Also long summation terms are better written in groups of three or four digits on either side of the decimal point to make them easier to read, like this:

149 7412.3907

or 1 497 412.390 7

and not as an unbroken line, like this

1497412.3907

The treatment means should be written as 15.1, 16.0, 14.4 ($e = 1$), or perhaps 15.12, 16.05, 14.42 ($e + 1 = 2$) if they are needed with special precision. The standard error of a treatment mean should have two decimal places, i.e. as 0.25.

1.11 The role of the statistician in crop experimentation

A field experiment involves a number of stages. At each there are statistical considerations.

Stage 1 Inception Any good experiment has clearly defined objectives. They must be specified right at the start in the form of 'contrasts of interest', which will be explained in Sections 5.1 and 5.2. Thereafter everything must be subordinated to obtaining good estimates of those contrasts. (At this stage the treatments are chosen and the relationships between them clarified.)

Stage 2 Resources Once objectives have been decided, it is necessary to examine the resources available. First there is the land, whether it is variable or not, and whether it is suitable for the task in hand. If it is variable, is there any system of local control (blocks, etc.) that will make it more uniform? Other enquiries should concern plant material (seed quality, etc.) including plant variability, the skill of the workers involved, and the adequacy of the equipment available to them.

Stage 3 Design The design adopted should pay respect to what has been learnt in Stages 1 and 2, so that the experiment shall be both relevant and practicable. The design should find expression in a plan which makes everything clear to those who will have to work on the experiment, whatever their capacity. The scheme should lead to no difficulty in applying the treatments, obtaining the data and carrying out ordinary

farm operations. Also, the design must be such that the statistician can see his way to analysing the data.

Stage 4 Implementation The plan having been handed over, there could well be other preliminary documents, such as the results of soil samples, notes on the source and quality of the plant material and on the weather at sowing or transplanting. Now is the time to start a diary of the experiment in which such matters will be noted in the future. Other operations are the surveying and laying out of the experiment in the field, the application of differential treatments, the recording of what can be measured and the scoring or grading of other features, like leaf colour, which can be observed but do not lend themselves to measurement. It is essential that the statisticians know about all these operations. If they do not, they may design experiments that no one can implement, or analyse data with no idea of what they really represent.

Stage 5 Data analysis The calculation of analyses of variance can be a highly technical process, but the statistician's skills must not stop there. Above all it is necessary to assess well those contrasts that were defined at Stage 1 and were written into the design at Stage 3. Further, it is often useful to look back at Stage 2 and enquire how well the design has allowed for the sources of variation expected and how far other more important sources have been ignored. Not least, the data can be examined to pick up any faults of implementation at Stage 4.

Exercise 1A

Four cultivars, A, B, C, D of dwarf beans were arranged in a completely randomized design with four replicates each. The layout of the design was as indicated below and the leaf areas (cm^2) were recorded, as shown, three days after germination.

Five of the plots suffered damage from a windborne insectide spray from a neighbouring field and these plants are indicated as missing (m).

D 7	A17	B10	C17	A17	D 6	B14	A15
C15	D 8	B14	D10	C16	A19	B13	D 5
A18	D 7	D 6	B12	B10	A10	C m	D m
C15	B 9	A16	C12	B 6	C m	A m	C m

Work out the analysis of variance and write out the residuals on the field plan. Do the residuals suggest anything to you?

Exercise 1B

An experiment on six bean varieties, A to F, was carried out in four randomized blocks. Yields were as follows:

I	F	9.0	D	14.6	C	18.3	E	14.2	F	14.1	C	17.4	II
	E	14.1	B	21.9	A	22.4	B	25.6	A	23.9	D	19.2	
III	C	12.7	D	15.8	E	11.5	E	12.1	D	16.1	C	15.9	IV
	A	21.1	B	23.7	F	6.4	A	19.6	F	12.3	B	18.3	

Data represent fresh weight of crop in kilograms per plot of 36 m^2 and are presented according to the field plan. Write the residuals on a copy of the plan and work out the analysis of variance both from the deviations and the residuals and by the use of summation terms.

[Data from P. Dagnelie, *Principes d'Expérimentation* (1981), p. 94. Sub-plots have been ignored.]

Exercise 1C

An experiment was conducted on ten strains of carrot, here called A–J. It had four randomized blocks, each consisting of a line of plots. The four lines were side by side. Yields were as follows:

Block I		Block II		Block III		Block IV	
J	27.7	I	35.5	G	30.2	A	31.8
I	36.7	E	33.0	C	31.2	F	31.8
G	32.6	D	25.2	B	31.9	D	22.3
F	30.6	A	28.0	E	30.1	I	32.4
B	33.4	J	34.3	I	35.7	B	29.8
D	22.2	F	30.0	D	24.4	H	29.5
E	30.2	B	29.5	A	28.3	C	25.8
A	30.0	C	29.0	H	27.6	E	27.8
H	30.1	G	31.7	J	31.7	G	30.8
C	32.9	H	29.7	F	28.5	J	27.7

Work out residuals and the analysis of variance using two methods.

Note: The residuals are of interest because the blocks are very long and narrow, so trends along them are to be expected. The position will be examined in more detail in the Exercises of Chapter 8.

[Data from A. A. Rayner, *Biometry for Agriculture Students* (1969), pp. 267–8.]

Exercise 1D

One early example of a design in randomized blocks compared sixteen fertilizer treatments, A–Q (I omitted), in four blocks. The data from Blocks III and IV are given below. Work out residuals and consider whether the block boundaries had been formed to best advantage

Block III								Block IV							
A	351.5	L	495.5	J	443.0	C	383.5	P	559.0	Q	550.0	B	359.0	E	395.5
K	472.5	B	367.5	G	455.5	O	502.5	C	328.5	H	390.5	J	483.0	O	512.0
E	357.5	F	381.5	Q	531.0	D	316.0	N	522.0	M	444.0	A	325.O	D	259.0
N	385.5	H	354.0	P	496.5	M	474.5	F	410.5	G	351.5	K	430.0	L	394.5

Data represent the yields of potatoes in pounds per plot of unstated area (1 pound = 454 grams).

[Data from T. Eden and R. A. Fisher, *J. of Agricultural Science*, **19** (1929), p. 207.]

Exercise 1E

An experimenter had three promising new varieties of maize, which we will call X, Y and Z. He wished to compare them with his existing recommended variety, which we will call R. To discover how the new varieties compared, he tested all four in a Latin square. The following figures give yields in cavans per hectare. (A cavan is a measure of volume used in the Philippines: 1 cavan = 77.5 litres.)

Y 32.8	Z 24.2	R 28.5	X 26.9
R 29.5	X 23.7	Z 28.0	Y 25.8
X 33.4	R 14.2	Y 33.3	Z 23.6
Z 31.3	Y 25.8	X 33.1	R 13.2

Work out the analysis of variance using two methods.

[Data from K. A. Gomez and A. A. Gomez, *Statistical Procedures for Agricultural Research with Emphasis on Rice* (1976), p. 27.]

Exercise 1F

An experiment was conducted on six strains (A–F) of maize using a Latin square. The following data represent bushels per acre. (In general, it

is better to record crops by weight, but it is admittedly easier on occasion to measure by volume.)

B 20.3	C 14.9	A 13.7	D 14.7	E 16.7	F 18.6
E 18.2	A 15.3	C 10.1	F 15.3	B 10.8	D 11.3
A 12.2	E 16.7	F 12.2	C 11.2	D 11.4	B 10.5
D 18.9	F 17.9	E 16.8	B 16.0	A 14.3	C 16.4
F 17.7	B 20.1	D 17.0	A 15.3	C 15.5	E 17.0
C 18.7	D 17.9	B 19.9	E 17.7	F 18.4	A 12.1

Work out the analysis of variance using both methods.
1 bushel = 36.4 litres 1 acre = 0.405 hectare

[Data from C. I. Bliss, *Statistics in Biology*, Vol. I (1967), p. 309.]

Exercise 1G

An experiment was conducted to compare the cropping of four new raspberry varieties, B, C, D and E, with a standard variety. A. The following data came from the row-and-column design used:

B 88	A 246	E 174	A 236	C 61	D 83
C 122	A 165	A 194	B 97	D 118	E 145
D 132	E 124	A 221	A 127	B 105	C 100
A 149	C 76	D 96	E 122	A 145	B 68
A 253	D 106	B 94	C 98	E 151	A 145
E 195	B 106	C 130	D 93	A 180	A 128

The data represent ounces of fruit per plot. The area of each plot is not stated. (One ounce = 28.4 grams)
Calculate the analysis of variance.

Note: If the treatment summation term is needed, it should be calculated using the method of (1.4.1).

[Data from S. C. Pearce, *Field Experimentation with Fruit Trees and Other Perennial Plants*, p. 92 of first edition (1953).]

Chapter 2

Practical matters of experimentation

Note: Many of the topics of this chapter are dealt with more fully in the book by Dyke (1987).

2.1 Introduction

If an agricultural field experiment is to be successful, careful attention must be given to all stages of its existence, from the choice of the site and the drawing of the plan to the harvesting of the crop and the recording of the yields of the plots. In this chapter we discuss some of the points that commonly arise in settling the correct procedure at all these stages.

Agricultural crops, like most biological material, have an intrinsic variability which, at least with present knowledge, cannot be eliminated. If, at some particular stage of the conduct of an experiment, there is a choice between an easy, slightly inaccurate procedure and a more arduous or more lengthy but more accurate procedure, it may be tempting to choose the easier option. For example, if a fertilizer is measured by volume instead of by weight, the variation of quantity from plot to plot will be slightly increased. In many circumstances, the effect on variability of the crop yields will be very small in comparison with the irreducible variability likely in the experiment. The experimenter should, however, think twice (at least) before settling for the second best; any noticeable departure from the maximum accuracy that can reasonably be achieved tends to encourage a careless attitude to the experiment at all stages. Most agronomists visiting a research station for the first time have confidence in the experimentation in proportion to the tidiness of the plot-work. They take that as showing the keenness and discipline of the field workers and scientists; it is difficult to put much trust in an experiment that has unnecessary defects, even small ones. If a less good method is adopted, it should be for a considered reason that can be argued convincingly. In general the pursuit of excellence should dominate all else.

2.2 Choice of site

Assuming that a particular field or plantation has been allocated for an experiment, the exact site should be chosen so as to minimize likely variation, at least within blocks. Such variation may be caused by changes of soil, for example between the top and bottom of a slope, by past farming operations—for example, ploughing on different dates—by different degrees of shelter, or perhaps by nearby trees. Edges of fields are conspicuously different from the interior parts, sometimes because of cross-ploughing, spraying overlaps, sometimes because of incursions by trespassers. It may be wise to modify the design to allow for features of the site, for example, to change from randomized blocks to a row-and-column design, or to change the direction of the blocks.

In the analysis of variance it is assumed that the differential effects of treatments will be the same anywhere. For that reason it is wrong to accept a site for which that assumption will not hold. For example, the effect of fertilizers will be different on poor soil from what it would have been on rich. It would therefore be foolish to allocate a field with a wide fertility range to a study of level of fertilization, though it would be less objectionable for a trial of insecticides.

2.3 Size and shape of plots

Anyone who proclaims the optimum plot size for a particular crop, regardless of circumstances and treatments, should be disbelieved; no such optimum can exist. He is oversimplifying a problem that has many aspects. For a given crop on a given site there will be an optimum size for plots testing fertilizers, another for comparisons of cultivations, another for tests of sprayed fungicides. On another site the pattern of variability of the soil will indicate, at least for some of these types of experiment, different plot sizes. A season of heavy storms may cause patchy lodging or local waterlogging and indicate, perhaps too late, that unusually large plots should have been used. For those reasons calculations of optimum size based on analyses of uniformity trials are of little value in the determination of plot size for experiments, except for those of a single type in closely similar conditions. The analysis of a uniformity trial ignores the fact that, at least on some sites, treatment-effects in real experiments will vary with changes of conditions within the site. It is true that the assumption of additivity (Section 1.3) requires that such variation should be minimal, but it does sometimes arise with block designs and similarly with row-and-column designs, which differ only in having two sets of blocks. That casts doubt on calculations of optimum plot-size from uniformity data. We urge rather that practical considerations are most

important in settling plot-size and we list below some of the commoner ones.

(1) The nature of the treatments may necessitate discard areas (which receive the respective plot-treatments) or guard areas (treated uniformly throughout the experiment).
(2) Crop-sampling may be best done in 'sacrifice' areas adjacent to the areas taken for yield. (See Section 2.8).
(3) Machinery used for sowing, spraying, harvesting, etc., may influence one or both of the dimensions of the plot.
(4) Will the produce be weighed at the site or carted to a central station for weighing? Either way careful planning is needed and the plot size may have to be chosen to fit in.
(5) The area available for the experiment, leaving out any obviously anomalous parts for the land allocated, may place restrictions on the individual plots, both as to size and shape. (It may be wise to extend the area excluded beyond the minimum if the remaining plots are to be comparable. This could call for a change of site.)

In short, the plots must be thoroughly practical in both size and shape. Once a tractor has started to cross the land, unless special arrangements have been made it must continue to the other side, but long, narrow plots could be most suitable for the application of fertilizer. Treatments have to be applied properly; also, crops have to be harvested and measurements made. All that has to be considered in a choice of plots.

2.4 Marking out

Marking out the plots of an experiment usually involves setting out right angles and distances, most of the latter determined in advance by the dimensions of each plot, and of paths or guard areas or headlands between plots. Some of the distances may depend on hazards of the chosen site; for example a gap may be left in a row of plots to avoid soil contaminated by the ashes of a bonfire.

Occasionally a field of a particular shape may suggest plots of parallelogram shape, with angles not equal to 90°, in order to make best use of the available area. If wide machines are to be used, that causes complications. In some experimentation (for example on curved contour strips) other layouts of a more flexible type will be needed. In the remainder of this section we consider only a conventional experiment with rectangular or square plots, all of equal size.

The first step is to mark out the rectangle (rarely a square) that will contain the whole experiment, including any gaps left to avoid dubious patches. Lengths are usually set out by use of measuring tapes—though

the surveyors' chains used in the past were also suitable—and right angles by means of optical square, crosshead, or a Pythagorean triangle with sides in the ratio 3:4:5. In general the rectangle will not close; that is, if we mark out the four sides successively the final point will not coincide with the starting-point. Mostly that is on account of the angles not being exactly 90°. For that reason, corrections must be made to angles, not to lengths. That also ensures that all plots will be of equal dimensions as well as of the same shape. Once the sides have been measured, it is necessary to divide them equally to determine the corners of individual plots. Here again, discrepancies can arise. If so, it is important to adjust all plots equally or some will be wider than others.

Although for most purposes the metric system is best, there are circumstances which suggest the use of feet and inches, for example, the availability of a seed-drill of many spouts with unit spacing of 7 inches. A mixture of units (e.g. plot-lengths in metres, width in feet and inches) presents no great difficulty, provided a doubly-marked tape is available. The only task made more difficult is the calculation of plot-area, and that has only to be done once and in conditions of comfort indoors. Excessive accuracy, e.g. measuring to the nearest millimetre, is absurd in connection with crop-plant experimentation; the width of a plot, however, should be written down with enough significant figures to reconcile the width of many plots with the total width across the experiment. Measuring tapes have been known to stretch with much use and should be checked from time to time.

2.5 The plan

The plan of an experiment is the chief means of conveying information between the various people involved in the field work. Often those performing the analysis and interpretation of the results will need it too. (All these jobs may be done by the same person, but even so people can forget details so a record is needed.) In most experimentation the plan should be in such a form as to make the field worker's task as easy as possible; he (or she) may be tired, cold, wet or hot or may be hurrying to finish a job. Perhaps a difficult task is involved like sowing different batches of seed on the plots of a variety trial, before dark, before a thunderstorm, or before the farmer wants his seed-drill back. A mistake made by the field worker at such a stage, even if it is detected and recorded, may seriously damage the experiment. Other users of the plan are almost always able to study it in better conditions and with less urgency. The guiding principle, then, is to produce a plan that serves the field worker as well as possible. Sometimes, e.g. if the experiment involves the application of treatments at different dates, like the sowing of different

varieties in autumn and the application of different rates of fertilizer in the following spring, it may be helpful to provide, in addition to the full plan, simplified 'overlays'. They will include only symbols needed at a certain stage, for example one overlay might show varieties but not rates of fertilizer. A plan or overlay should be carefully checked, preferably not by the person who drew it.

One great help is a generally agreed system of plot numbering. It should be decided in the first place with the interests of the field workers in mind. Thereafter, everyone else should use it, up to those responsible for data input to the computer. If that is done, two important sources of error are minimized. (1) Direct copying, which is a potent source of error, because it appears to be so simple; everyone supposes him or herself able to do it, but minds wander. (2) Indirect copying, in which figures are de-randomized in the process of copying. For a preview of the conclusions, people will work out treatment means from the field sheets, and that is all right provided nothing final is built on them. They should be only interim indications.

We conclude this section by listing some of the things that are required on the plans of most experiments:

(1) Alignments of plots should be correctly indicated, in one or two directions as appropriate.
(2) Dimensions of each plot, and of the whole experiment, together with clear indications of the positions and widths of all paths, gaps, etc.
(3) Orientation. Usually a North point is shown.
(4) Plot-numbers, treatment-symbols.
(5) Block-boundaries.
(6) Name, location and date of experiment. Also its reference number if it had one.
(7) Crop, variety, seed-rate or plant spacing.
(8) Full description of treatments under test, and of 'basal' ('blanket') applications.
(9) Any normal farming operations that must be omitted.

The exact location should also be made clear by reference to nearby features that can be relied upon not to alter.

Field labels

Related to the plan is the system of plot labels. The one should complement the other. If abbreviations are used on the plan, they can be used on the labels also. At no point should there be any conflict between the two.

Sometimes there are conflicting ideas about what to put on the labels. Put too much and they look confusing; put too little and an opportunity

will have been missed. One particularly difficult matter concerns the treatments. If they are not shown, there could be difficulties when they come to be applied; if they are, the information could prejudice the taking of some records, especially if plots are to be scored. (Field recorders can be told not to read labels except to check the plot number, but they can hardly fail to see what is clearly displayed. Further, if they are also the people who had to apply the treatments and they remember how the plan went, they can scarcely be expected to put the information out of their minds.) To take another consideration, sometimes the treatments will have been applied before the labels were set up. In that case there is little point in displaying them. (The argument that visitors will be interested need carry no weight; they should be conducted round by someone with a plan, not left to wander.)

Whatever else the labels show, the plot number is essential. Ideally plots should have a standard order that is used for all purposes, the application of treatments, the recording of data and the presentation of data to the computer. Further it should be decided with the interests of the field staff in mind. Even then, there may be occasions when they will want to deal with plots in a different order. If that does happen, it is a courtesy on their part to write boldly on the sheets 'Plots in non-standard order' to help people further down the line, who otherwise might not notice the change. How is the standard order to be chosen? Most people when they have finished one row will want to work back along the next, like this:

1	2	3	4	5	6	7	8
16	15	14	13	12	11	10	9
17	18	etc.					

There is in fact a word to describe such a progression; it is *boustrophedon*, meaning the way in which an ox would plough. Field workers do it in the same way and for much the same reason as the ox.

The alternative is to work along each row in the same direction, like this:

1	2	3	4	5	6	7	8
9	10	11	12	13	14	15	16
17	18	etc.					

Its chief feature is that all plots are approached from the same direction. This is perhaps not much of an advantage except in scoring, because the sunny side of a plot can look very different from the shady side. In any case good field recorders know that, and they avoid letting it interfere with their scoring. Whatever method is chosen it should ensure that all plots of

a block are kept together. Further, it is good practice to distinguish the last plot of a block with a label of a different colour or a different shape. In that way the field staff know where they can safely stop if they have to do so. In the above example, if there were blocks, four plots wide and two plots deep, the following might be a good system of plot numbering:

4	3	2	1	16	15	14	13
5	6	7	8	9	10	11	12
		18	17				

If it is found that the field staff do not follow the standard order once it has been decided, that suggests that it was badly chosen in the first place.

Once a suitable system of numbering has been agreed, try to use it always. The numbering of plots should never be changed during the life of an experiment. If necessary in a long-term experiment if plots are subsequently split, plot 1 may become sub-plots 11, 12, ..., or 1a, 1b, ...

2.6 Methods of randomizing

We list below some of the many methods of allocating treatments at random to plots within blocks, or sub-plots within whole plots, together with notes on necessary precautions. Much of this applies to the randomization of row-and-column designs, but we do not deal with these in detail.

(1) By use of published tables of 'random numbers'. (They are really tables of numbers, generated in some complicated but systematic way, that have passed various tests. They are therefore known to be free from undesirable sequences likely to lead to bias.) For 2–10 treatments, it is possible to use digits one at a time provided all treatments are to be equally replicated. For 11–100 there is another method which can well be adopted as standard and used even when the treatments are few enough to be represented by single digits. If the number of treatments exceeds 100, it is readily extended. The method is this: If the next plot can have A treatments, a two-digit number is taken at random and divided by A. The remainder indicates the treatment to be chosen. Thus, if $A = 12$ and the random number is 87 ($= 12 \times 7 + 3$), the third treatment should be chosen for that plot. There are three points to be noted. (a) The double zero, 00, should be read as 100. (b) If the random number is an exact multiple of A, that indicates the last treatment on the list. (c) If 100 is not a multiple of A there are some two-digit numbers that should be avoided. For example, if $A = 12$, no use should be made of 97, 98, 99 or 00; to do so would be to give Treatments 1, 2, 3 and 4 nine chances of being selected as compared with eight for the rest. That is to say, the range of random numbers must

terminate at the highest multiple of A less than 100 in the case where 100 is not itself a multiple.

The method can be illustrated by supposing that a block with six plots is to contain treatments A, A, B, C, D and E. It will further be supposed that the first two-digit random numbers given by tables are 22, 81, 68, 00, 53, 16, 45, 51, 34. The stages are:

(i) The first random number is 22. Since $A = 6$ and $22 = 6 \times 3 + 4$, the fourth treatment is indicated. That leaves AABDE.

(ii) The second number is 81. Since $A = 5$, that indicates A for the second plot, which leaves ABDE.

(iii) The next number 68, is an exact multiple of A ($= 4$). Accordingly E should be chosen. That leaves ABD.

(iv) The next number is 100, which is not itself a multiple of A, so it must be ignored, though all numbers from 01 to 99 would have been acceptable.

(v) The next number is 53, which indicates the second out of the three treatments still available, i.e., B. The choice now lies between A and D.

(vi) The final number is 16. It is even, so D is chosen for the next plot and A for the last. The result of the process has been to assign the treatments in the order CAEBDA. The other random numbers can be disregarded or used to start another block.

(2) By use of a computer to generate random numbers. Some machines, however, have quite crude generators, so, if many experiments are being designed, there may be a risk of repeating similar patterns.

(3) By shuffling packs of cards, either made up specially for the purpose, or normal playing cards.

(4) By dice, for example, a 10-sided die (a treasured possession of one of the authors).

(5) By tossing a coin (which we hope is unbiased). Add imaginary treatments to make the total number a power of 2 (e.g. 32) and use each toss to decide whether to discard the first or second half of the treatments remaining at each stage; some runs of tosses indicate imaginary treatments and are ignored.

In the case of a row-and-column design it is necessary to find a design that has the right allocation of treatments to rows and columns and then to permute both rows and columns at random.

2.7 The application of treatments

Once the treatments that are to be compared have been settled it is necessary to plan how they should be applied. If they are varieties or rates of fertilizer the matter is fairly simple, but even then problems can arise. Should varieties be sown at the same seed-rate measured by weight or by number of seeds per unit area? Should allowance be made if the seed of one variety is of poor germination? Surely fertilizers should be applied evenly? But, in experiments designed to be the basis of recommendations to farmers, who are likely to apply them unevenly, perhaps they should be applied about as they would be on farms.

Often the treatments to be compared are really different systems of farming. An exceptionally early-ripening variety may require harvesting before the others in the experiment—with some awkward consequences. A new system of cultivation may permit earlier sowing than is traditional, but that raises a difficult question. Should plots be sown at different dates, or should the new system be penalized by delay until all plots can be sown on the same day?

Machines used to apply treatments often need to be calibrated to assess their performance; for example a seed-drill may need adjustment if it is to deliver the same amounts of seed of different varieties per unit area. Calibration should be done in conditions similar to those on the experiment. Nevertheless, the measurement of amounts used on the plots of an experiment is not necessarily satisfactory because machines must start and stop work beyond plot boundaries. Finally, experiments should not be used only to test cultural practices, varieties, etc.; sometimes they are needed to test experimental techniques. For example, there might be different opinions on how to record the crop, and some plots might be recorded using both methods to see if they led to different results.

It need hardly be said that treatments should as far as possible be applied to plots following the plot order described in Section 2.5. Sometimes, indeed often, that is difficult and cannot be insisted upon, but some bad practices should be avoided. For example, in a spraying experiment it is decidedly wrong to make up Spray X today and to apply it; then, on the next fine day to make up Spray Y and so on until all treatments have been applied but over a period of time. Good experimenters do better than that.

2.8 Sampling in a field experiment

Sampling of a field experiment may be needed before the plots are marked out, after the crop has been harvested, or at any stage between those extremes. Soil may be sampled for determination of nutrients, pH, organic matter, salinity, soil-borne pests or diseases. The crop may be sampled during growth for chemical analysis, for estimation of leaf-area, for diseases or pests; it may be sampled just before or during harvest for

measures of quality, for dry matter or for nutrient content. The plots may be sampled after harvest to assess losses incurred before or during harvest. This tiresome list by no means exhausts the possibilities.

Sampling a field experiment is different from sampling a field crop for the purpose of a survey; the remainder of this section is not a guide to the conduct of sample surveys but of experiments. Once treatments have been applied, all sampling should be done by plots. Occasionally, e.g. when deciding what 'basal' (or 'blanket') operation to use before the crop is grown, other methods may be used. (That might be done when deciding whether or not to lime a possibly acid site.)

The two main objectives are:

(1) to assess differences between treatments so that the knowledge of their effects on yield can be supplemented by understanding of their probable mode of action;
(2) to provide calibration data which can be used to make more accurate estimates of the effects of treatments on yield (or other primary observation), typically by use of analysis of covariance (see Sections 8.5, 8.6 and 8.7).

For both these objectives the great need is to establish differences between plots, not absolute values. Here experiments differ from surveys. If two sampling schemes are practicable and if one gives more accurate estimates of plot-differences than the other, even though it may have a degree of bias, it is the more accurate that should be adopted.

There are three main ways of planning the sampling of plots (we are thinking mainly of sampling the growing crop):

(1) Random sampling
(2) Stratified random sampling
(3) Fixed-pattern sampling

Random sampling implies a separate pair of random numbers (chosen from suitable ranges) to define the position of each sample within each plot.

Stratified sampling is appropriate when there is important variation within each plot of a known pattern, for example, between land marked by tractor wheels and the remainder, or between rows sown by different spouts of a seed-drill. The variation of plot-means is lessened if the same proportion of samples is taken from each type of land within each plot. Also, plots may be divided into strata in some purely geometric manner, e.g. by subdividing the plot into four sub-plots.

Fixed-pattern sampling implies the location of samples in the same positions relative to the boundaries of all plots. It minimizes the effects of patterned variation, known or unknown, related in any way or at any stage to operations on the plots. It provides no estimate of within-plot sampling 'error', whereas random sampling of whole plots or of strata within plots does provide estimates of sampling error, provided there are at least two samples in each plot or stratum. Such estimates are needed if alternative sampling schemes, e.g. taking more or less samples per plot, are under consideration for future work. For all other purposes the relevant estimate of error is the one based on differences between plot-means. This is equally true whether sampling is done for either, or both, of the purposes indicated previously.

If substantial destructive sampling is planned it may be advisable to increase the plot-size so that a part of each plot, after excluding discard areas near edges, may be given over entirely to sampling, leaving a separate area for estimation of yield. Sometimes sampling may be confined to discard areas, provided they are not unduly unrepresentative. Samples may be taken solely from yield areas, or partly from discard, partly yield areas. Sampling from yield areas gives the best chance of detecting a relationship between yield and the sample observation; if sampling is from 'sacrifice' or discard areas the relationship is blurred by an unknown degree of within-plot variation.

Sampling part of an experiment, e.g. from certain treatments only, may be necessary occasionally, some treatments being of no interest to the sampler. If sampling affects yield areas, that will bias the estimate of some treatment contrasts. Sometimes, on the other hand, a lower degree of replication will suffice. Sampling all plots on a selection of blocks is less objectionable. Nevertheless it may increase 'error' variance if treatments behave differently in different blocks. As explained in Section 2.3, that usually does happen to some extent, though not to such a degree as to call for a change of site (Section 2.2).

Adjusting recorded yields to allow for the effect of crop-sampling is arbitrary unless the sampling can be done just before harvest and without damage to the adjacent crop. Some thickly-sown crops may make good almost all the 'lost' yield by more vigorous growth of plants adjacent to small sample areas. On the other hand, sampling apple shoots, to measure growth soon after planting, will almost certainly leave permanent effects. Hence sampling all plots equally is much preferable to sampling some only.

Samples are sometimes bulked before examination; bulking samples taken from one plot at one time is usually acceptable. Bulking samples from different plots is a more risky practice and usually better avoided. For example, if fresh yield and percentage dry matter are positively correlated between plots, bulking equal weights of produce from replicates will cause underestimation of the mean yield of dry matter.

2.9 Scoring the plots of an experiment

A perceptive, conscientious agronomist who regularly examines the plots of an experiment can estimate colour, growth, population of plants, weeds, some diseases, some pests, and no doubt many other things that may be important. The estimates may not be quantitative, but they should put the treatments into their correct order and distinguish between large and small effects of treatments. Many of the characters he or she can estimate would be difficult or impossible to measure by use of instruments, at least without doing substantial damage to the crop. An industrious person can record scores for one or two characters on each of 100 plots in less than an hour without setting a foot in a plot. So, within its limitations, scoring is an efficient means of gathering subsidiary information.

Regular scoring can reveal mistakes in the application of treatments and isolated mishaps, such as failures of a seed-drill. If the experimenter pays some attention to nearby non-experimental crops during a scoring visit, indications may be found that explain a trend in fertility on the site. General notes about the state of the crop in relation to other areas of the same crop at the same time can be useful when an experiment is reviewed perhaps months later.

When setting out to make scores the experimenter should first look quickly at all the plots and establish the appearances of extreme conditions; the palest and the darkest, the least weedy and the most weedy, or whatever is appropriate. Scores can then be allotted to these extremes; perhaps 0 and 4, or 0 and 6. The plots are then studied in order, one block or row or column at a time. As far as possible, even numbers should be used for scores 0, 2, 4, 6 (say) but the intervening odd numbers can be used when necessary. (This procedure is a little simpler than starting with 0, 1, 2, 3 and then finding it necessary to resort to 0.5, 1.5 etc.; the actual numbers used make no difference to the final result.) The experimenter should score without knowledge of the plot-treatments and, if possible, should look at all plots from the same direction. Anything exceptional about any individual plot should be noted. Finally, a description of the scale of scoring should be written on the record sheet ('0 = 5 cm tall, 6 = 15 cm', for example) together with a note of the general state of the crop.

Most people can usefully distinguish about 2 to 10 different grades; an example of the simplest grading is to score plots '0' for light-green leaves and dark ones '1' in an experiment testing nitrogen fertilizer on a cereal crop. In the same experiment, some weeks later, one might score for lodging, using grades 0, 1, 2 ... 10 corresponding to 0, 10, 20 ... 100 percent of area lodged. The difference lies in the second having a numerical base which the first lacks.

In principle, scores may be analysed statistically like any other quantity. In practice they can cause problems on account of discontinuity. As was explained in Section 1.10, it is desirable that a variate should be able to take at least 20 values, but it would be unwise to attempt so many grades, even in the best case when they have a quantitative basis. (Without one, no observer can hope to keep so many standards clearly in mind.) One possibility is to use more than one observer and to add their scores. If more than one practised person is available that is a good plan, but it would be a mistake to supplement the team with an inexperienced novice, whose scores might be little better than a sequence of random numbers. If only one good observer is available and discontinuity is a problem, the analysis of variance will only be an approximation.

The fact is that some people are not good at scoring. Partly it is a matter of temperament. The injudicious put down extreme values with little reason. At the other extreme are the timorous who will not put them down at all. In between are the judicious who make wise use of them. Further, in a team the injudicious will outweigh the more cautious when scores come to be added. People reveal a lot about themselves in the way they score. The person in charge should now and again look at the grades returned by his staff to detect both those who think everything either superb or terrible and those who will never commit themselves even when faced by the remarkable.

There are also differences in training. People have positively to be taught to look at all plots from the same direction and to take precautions of that kind. They also have to be taught to distinguish between related phenomena. For example, large blossoms are not the same as numerous blossoms, but at first glance they create much the same impression. Again, no one can score the incidence of a disease without knowing where to find it. Two varieties of different habit may be equally infected, but the damage may be more obvious on one than on the other.

Finally, the best way to learn is to have one's scores checked, either by a more experienced observer or by a proper weighing. Too many scientists think that their scoring must be good because they understand the underlying botany, but that is often a distraction rather than a help. If it is a matter of observing leaf colour, for example, the first requirement is that the observer shall not be colour-blind. Botany has nothing to do with it. One of the writers was impressed by the confidence with which a group of horticulturists scored the crops of apple trees. When they had departed he noticed that the pickers, who claimed no scientific skills but whose ease of working depended upon their putting the right number of boxes under each tree, were much better. No one will learn to score well without the feed-back that comes from checking their figures against other people's or, even better, against objective data.

2.10 Harvesting and recording

Yields are usually the most important observations made on an experiment. Other quantities are often calculated from them, for example uptakes of nutrients. The area taken for yield should be the same for every plot, except where an accident has reduced the good area of a plot and in some spacing experiments. If there is any doubt about including plants near the edges of the harvested area, for example in wheat sown broadcast, the same person must make the decision on all plots of a block (or row, or column). Harvest should be done by blocks (or rows or columns) to equalize conditions (weather, fatigue or workers) as much as possible. If there are several workers or gangs, each one should be given a block or group of blocks and told to harvest all the plots so defined. The object is to confound all 'extraneous' variation with block-differences.

Harvest produce should be labelled with plot-number, not treatment; partly to lessen the risk of bias, conscious or unconscious, partly to deal with designs in which there are duplicated treatments within a block. Write down the area harvested exactly as it is measured; do not calculate in the field. '100 feet by 15 rows at 10 cm' is fine. It can be worked out and checked at leisure later. Do not record yields to an unnecessary degree of accuracy; that increases the risk of gross error in recording, or in later manual copying of results. As a guide: guess the smallest likely standard error per plot, halve it, round yields to the next convenient unit smaller than or equal to this. Avoid manual copying like the plague; if it has to be done, it must be checked. But yield records should be copied, perhaps by xerography, or the use of carbon-backed paper, and the two copies separated as soon as possible. At all costs avoid losing both copies!

Produce may be weighed on the site, or 'at home'. For weighing on the site a spring balance or steelyard on a tripod, or better, a 'tipping jib', may be used. Spring balances should be checked occasionally. At base a fixed balance will probably be used; direct links to a microcomputer, or to a mainframe, are sometimes available. Avoid parallax errors in reading balance-dials; have a second worker to verify the readings. Tare weights may be constant. If so, some balances can be set to read net weights directly. If tares are variable, they must be recorded as a separate variate; working out the net weight in the field is too risky. If samples are needed for the determination of the percentage of dry matter, they should be taken from well-mixed produce at the time of weighing.

Procedures for recording need to be carefully worked out and not left to improvisation when the time comes. It is important to have a good system of labelling containers as well as plots. If someone can go round before harvest and note the numbers of the containers assigned to each plot, there will be less difficulty later in deciding the plot to which a lost

container belongs. It is also important to have well-established procedures, understood by all, so that the yield of a plot is put in the right place until collected and so that no one harvests across plot boundaries. Where experimenters appreciate the difficulties, such matters are attended to and clear procedures are laid down. Sometimes, unfortunately, no one realizes that there are problems to be resolved and that can lead to disaster.

Exercise 2A

You are asked to lay out a block in the freshly ploughed field. It is to be 30 metres long and 16 metres wide so as to contain five plots, each 6 metres by 16 metres.

Start by setting up a base-line AB 30 metres long. At B set up a line at right-angles to AB and mark a point C 16 metres from B. (Turn left at each corner). Set up a right angle at C and mark D at 30 metres from C. At D set up another right angle and mark point E at 16 metres.

What is the distance between A and E? If they do not coincide adjust the angles at C and D to bring them together.

Now mark P, Q, R, S in AB so that AP, PQ, QR, RS and SB are all 6 metres; if there is any discrepancy move P, Q, R, S until the 5 distances are equal. Similarly locate W, X, Y and Z in CD. How long is it since your tape (or chain) was checked?

Exercise 2B

An experiment is to be designed in four randomized blocks with six treatments, A, B, C, D, E and F. Reference to tables of random numbers gives the following four sequences of two-digit numbers:

71	46	30	49
52	85	01	50
27	99	41	28
61	62	42	29
96	83	23	56
83	07	55	07
52	83	51	14
62	80	03	42

Using the four columns respectively for Blocks I, II, III and IV, allot the treatments at random within each block.

Exercise 2C

Before an experiment is harvested, score its plots for crop weight. When the data are available, check your scores against them. We suggest that you do it more than once.

Exercise 2D

Find an experiment on cereals (or some similar crop) in which the treatments are affecting the growth of plants. Score each plot for height of plants with some quantitative base in mind, i.e., one grade equals 5 cm, or something like that. Then go round again with a measuring scale and see how good your scores were. (How do you determine the height of a plot anyway? The plants in it will vary.) Again the exercise can be done more than once.

Exercise 2E

The 96 plots of an experiment fall into eight blocks as follows:

1	1	1	1	2	2	2	2	3	3	3	3	4	4	4	4
1	1	1	1	2	2	2	2	3	3	3	3	4	4	4	4
1	1	1	1	2	2	2	2	3	3	3	3	4	4	4	4
5	5	5	5	6	6	6	6	7	7	7	7	8	8	8	8
5	5	5	5	6	6	6	6	7	7	7	7	8	8	8	8
5	5	5	5	6	6	6	6	7	7	7	7	8	8	8	8

Suggest a scheme for numbering the plots.

Exercise 2F

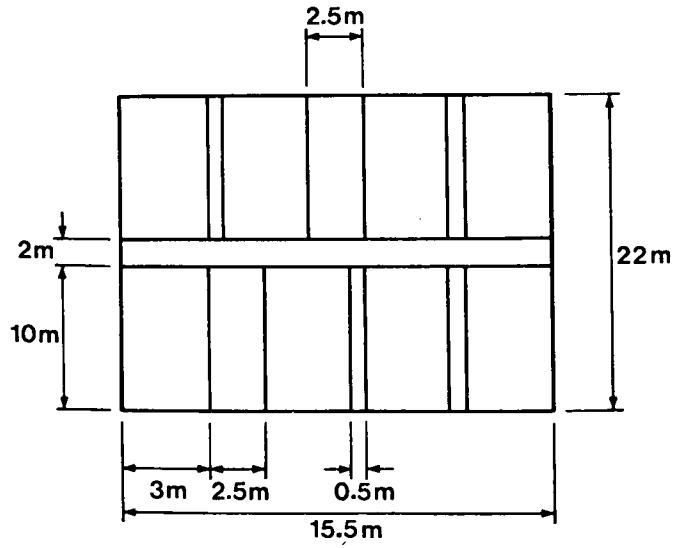
Draw the plan of an experiment that you might have to deal with in your normal work; put in all necessary details. To save time we suggest a limit of 16 plots.

Exercise 2G

The crop growing on the experiment of Exercise 2F is to be sampled during growth for some secondary character (e.g. total dry matter, or plant population, or an insect pest). Decide which character is to be studied and write full instructions to the field worker for the sampling of the plots.

Exercise 2H

Working as in Exercise 2A, mark out all the points needed to define the plan below, which is not to scale.



Chapter 3

Basic statistics

Note: Sections 3.1 to 3.6 set out the basic statistical knowledge assumed in the rest of this manual. A reader who finds the material completely unfamiliar is advised to do some preliminary reading first. We suggest the book by Clarke (1980), but there are others.

3.1 The Normal distribution

In order to carry out statistical inferences from experimental data, we need to assume that data follow some particular known distribution. Most analyses commonly used in agricultural statistics assume that data are normally distributed. The conditions for a normal distribution to apply are

(i) there is a strong tendency to take a central 'average' value;
(ii) deviations from this 'average' are equally likely to be positive or negative, i.e. the distribution is symmetrical;
(iii) the frequency distribution drops off quickly from the 'average', large deviations being very unlikely.

There is a mathematical definition that is more precise, but those are the main characteristics. The normal distribution is sometimes called the 'Gaussian' and sometimes the 'Laplacian', but we have preferred 'normal' as being the most usual term, at least among English-speaking workers.

We can sometimes argue that these conditions (i), (ii) and (iii), are likely to hold theoretically. With a new crop, uniformity data are useful; plot a histogram of a large amount of data under standard growing conditions and see whether it is symmetrical. If it appears to conform to the conditions, the normal distribution may not hold exactly, but it should be close enough to the truth to be acceptable.

Specifying the mean, μ, and the variance, σ^2, fixes completely which member of the normal family of distributions is used: larger σ indicates a more scattered, or spread-out, set of measurements. We commonly assume that two different treatments applied to a crop will alter only its

mean value of yield, growth, etc., not its scatter or variability; often this appears satisfactory, but it may not always be so.

If a measurement X is normally distributed with mean μ and variance σ^2, then $Z = (X - \mu)/\sigma$ follows the *standard normal* with mean 0 and variance 1. Tables give us those values of Z *outside* which p percent of this distribution lies ($\frac{1}{2}p$ percent at the top and $\frac{1}{2}p$ percent at the bottom). A few useful values are:

p%	10	5	2	1	0.2	0.1
Value of Z	± 1.645	± 1.960	± 2.326	± 2.576	± 3.090	± 3.291

When a random sample of r observations is drawn from a normally distributed population, their mean $\bar{X}$ will also follow a normal distribution (in repeated samplings), with theoretical mean μ and variance σ^2/r. Hence $Z = \dfrac{\bar{X} - \mu}{\sigma/\sqrt{r}}$ is again standard normal.

We should note here a certain inconsistency in nomenclature. Although people commonly talk about 'the 5 percent level', they more usually write it as '$P = 0.05$'. So long as it is understood that both expressions mean the same thing, no harm is done. In what follows we shall use either terminology as may be convenient.

Much statistical theory is built on the assumption that quantities are distributed normally. In fact, if we had enough data to detect divergences every time, we should probably find that most measurements do *not* follow the distribution exactly, though for most of them the assumption holds closely enough. After a while the observant practitioner comes to notice occasions when one or other of the characteristics (i), (ii), and (iii) above, is lacking. That is the time to consider the position and think about possible transformations (see Chapter 9).

Note: The 'shorthand' expression $N(\mu,\sigma^2)$ is often used to stand for a normally distributed variable whose mean is μ and variance σ^2.

3.2 Use of samples for estimation and hypothesis testing

At the outset of a programme of research, an estimate of μ will be needed. The sample mean, $\bar{x}$, is a good estimator of μ, but it is much more informative to have a 'confidence interval' which will cover the true (but unknown) value of μ in the population with a stated probability. This gives an idea of how precisely μ has been estimated: a very wide interval gives very little information about μ.

If σ^2 is known, the variance of $\bar{x}$ is known also, i.e., σ^2/r, so

$$Z = \frac{\bar{x} - \mu}{\sigma/\sqrt{r}}$$

will be standard normal and will lie within the range ± 1.96 with probability 0.95. Hence

$$Prob\ (-1.96 < \frac{\bar{x} - \mu}{\sigma/\sqrt{r}} < +1.96) = 0.95, \quad \text{so that}$$

$$Prob\ (\bar{x} - 1.96\sigma/\sqrt{r} < \mu < \bar{x} + 1.96\sigma/\sqrt{r}) = 0.95 \qquad (3.2.1)$$

which is a 95 percent confidence interval for μ based on the known σ^2 and the sample size. Increasing r leads to a narrower interval.

If an interval with higher probability of covering μ is required, it will be wider: a 99 percent confidence interval uses 2.576 instead of 1.96 in (3.2.1).

Later in a research programme, with enough knowledge or experience of a crop to propose a specific value μ_o for its mean, we shall wish to test this 'hypothesis' (see Section 1.3).

Using a sample of r observations, and assuming it is drawn from a normal distribution with known variance σ^2, test the hypothesis 'true mean $= \mu_o$' by calculating $Z = \frac{\bar{x} - \mu_o}{\sigma/\sqrt{r}}$ in which $\bar{x}$ is the mean of the sample of r. If Z lies between -1.96 and $+1.96$, within the central 95 percent of the standard normal distribution, then μ_o is an acceptable value for the true mean; otherwise reject this hypothesis. A value of Z outside ± 1.96 is called 'significant at the level $P = 0.05$', meaning that it gives significant evidence against the hypothesis being tested, which is usually called the 'null hypothesis'.

Although P is usually taken to be 0.05, other values may be used, for example 0.01 or 0.001 if we want stronger evidence. If the null hypothesis is true, there is a probability of only P that the data would have fallen out as they actually did. As P becomes smaller there will be a growing suspicion that the null hypothesis must be false, but the point at which this suspicion turns into conviction will depend upon the reasonableness of the null hypothesis itself. It may be so speculative and come at so early a stage in the research programme that we reject it at quite a high value of P, say, 0.20. On the other hand, it may be so well established that a low value of P will lead first to questions whether the experiment itself has been properly conducted rather than to doubts about the hypothesis. The value $P = 0.05$ has generally been felt to be convenient as a working level by applied statisticians but it has no greater status than that. It is not derived from any law of Nature.

Sometimes the experimenter may declare that, if a treatment is going to have an effect, it will be in a certain direction. For example, he may be applying a spray in the belief that it can only reduce the infestation of an insect pest. There are some questions to be asked here. Is he quite sure? Could the spray not kill some important predators and so bring about the opposite effect from what is intended? (Also, if he is really going to declare that the spray can have an effect in only one direction, which sounds rather dogmatic, he had better say so right at the beginning. If he says it later, he may find himself accused of changing the rules in the middle of the game.) However, he is quite certain. The spray may possibly decrease the level of infestation; it cannot increase it. That firm belief will change the nature of the test to be used.

It follows from the declaration that any increase in infestation is to be disregarded; it must have arisen from chance. If limits like those at (3.2.1) are set at ± 1.96 times the standard error, there is a 2½ percent chance of Z falling below the lower limit and so discrediting the null hypothesis. There is also a 2½ percent chance of its lying above the upper limit, but that will be ignored if it occurs. In fact, we are really working to 2½ percent when we wanted 5 percent. The correct approach is to use ± 1.645 times the standard error. That is the usual value for a 10 percent level of chance, but if only half the extreme deviations are to be regarded as meaningful, it gives the desired 5 percent level.

In saying that the treatment could not possibly increase the infestation the experimenter was setting up an 'alternative hypothesis' that, in this example, the true value may be less than μ_o but cannot be more. In that case positive values of Z support the null hypothesis rather than the alternative and should be seen as doing so.

The process just described is called a 'one-tail' test. Where there is no good reason to say which way a change will go, the 'two-tail' test, rejecting both extremes (large and small) of Z, should be carried out.

3.3 Estimating and testing μ when σ^2 is not known

In practice, if μ is not known in a population then neither is σ^2 known. The estimate of σ^2 from the sample is

$$s^2 = \frac{1}{(r-1)} \sum_{i=1}^{r} (x_i - \bar{x})^2. \qquad (3.3.1)$$

This estimate has to be used instead of σ^2; but $\frac{\bar{X} - \mu}{s/\sqrt{r}}$ is not normally distributed and (3.2.1) needs modification. The necessary distribution is that studied by 'Student' (W. S. Gosset), the t-distribution. Its shape is similar to the standard normal, but its spread depends on the sample size,

being broader the smaller r is; tables therefore have to give percentage points of t (as in Section 3.1) for each separate value of degrees of freedom. Degrees of freedom (abbreviated d.f.) number $(r-1)$ when a sample of r values is available to estimate σ^2 (see Section 1.3). For many purposes the variance is taken as the 'error' mean square from the analysis of variance and has the 'error' degrees of freedom.

Confidence intervals follow from the result that, when a sample is drawn from a normal distribution of unknown variance, $t = \dfrac{\bar{X} - \mu}{s/\sqrt{r}}$ is distributed as t with $(r-1)$ d.f. If $\pm t_{(5\%)}$ denotes those values of t with $(r-1)$ d.f. between which the central 95 percent of the distribution lies, then

$$Prob\,(-t_{(5\%)} < \frac{\bar{x} - \mu}{s/\sqrt{r}} < +t_{(5\%)}) = 0.95.$$

That is perhaps more conveniently written as:

$$Prob\,(\bar{x} - t_{(5\%)}s/\sqrt{r} < \mu < \bar{x} + t_{(5\%)}s/\sqrt{r}) = 0.95 \qquad (3.3.2)$$

which is a 95 percent confidence interval for the true value of the mean, based on a sample whose mean is $\bar{x}$ with estimated variance s^2.

For example, $t_{(5\%)}$ with 15 d.f. is ± 2.131 (instead of ± 1.96). For samples larger than 30, the $t_{(5\%)}$ values become very close to standard normal, so t need only be used in 'small' samples even if the population variance σ^2 is not known.

Also, for testing a hypothesis that the true mean is μ_o, using a sample of n observations whose mean is $\bar{x}$ and from which the variance has been estimated as s^2, $t = \dfrac{\bar{x} - \mu_o}{s/\sqrt{r}}$ is tested against the tables of t with $(r-1)$ d.f. (instead of the standard normal tables). Otherwise the procedure exactly follows that described in Section 3.2.

It is a weakness of much statistical practice that so much attention is given to testing and so little to estimation, although latterly the balance has perhaps moved a little more towards estimation. Mostly an experimenter has quite a good idea of the sort of response he can expect from his treatments, but he may have doubts how large it will be. For example, there may be little doubt that a fertilizer application will increase yield, but there could be serious reservations whether the gain in crop will pay for the cost. Again, in a trial of varieties known to be different, a test is absurd; the only sensible thing is to estimate the differences known to exist.

3.4 Difference between two means

In the case where independent random samples are drawn from two normal distributions, r_1 observations from the first whose variance is σ_1^2, and r_2 from the second whose variance is σ_2^2, the difference between the two sample means $\bar{x}_1 - \bar{x}_2$ is again normally distributed. The mean of this distribution of differences is $\mu_1 - \mu_2$, these being the true means in the two distributions, and its variance is

$$\frac{\sigma_1^2}{r_1} + \frac{\sigma_2^2}{r_2}$$

We shall write that as V, the variance of the difference. Therefore the quantity

$$Z = \frac{(\bar{x}_1 - \bar{x}_2) - (\mu_1 - \mu_2)}{\sqrt{V}}$$

once again follows a standard normal distribution.

A 95 percent confidence interval for the true value of $(\mu_1 - \mu_2)$ based on the sample and using the known σ_1^2, σ_2^2 is found by exactly the same argument as (3.2.1):

$$Prob\{(\bar{x}_1 - \bar{x}_2) - 1.96\sqrt{V} < (\mu_1 - \mu_2) < (\bar{x}_1 - \bar{x}_2) + 1.96\sqrt{V}\} = 0.95. \qquad (3.4.1)$$

Often in agricultural trials we require a test of the hypothesis that the true difference $(\mu_1 - \mu_2)$ is equal to D; frequently D is 0 but it does not have to be so. This is carried out by examining the value of

$$Z = \frac{(\bar{x}_1 - \bar{x}_2) - D}{\sqrt{V}} \qquad (3.4.2)$$

which follows a standard normal distribution if the hypothesis is true; with probability 0.95, we therefore expect it to lie between -1.96 and $+1.96$. The hypothesis is rejected only if Z lies outside this range.

In the common practical situation where σ_1^2 and σ_2^2 are not known, calculations can best proceed if they are assumed to be equal, i.e. $\sigma_1^2 = \sigma_2^2 = \sigma^2$. Therefore the first step is always to test whether the sample variances s_1^2 and s_2^2 calculated separately, in the usual way, for each sample differ significantly (section 3.5). If they do, and the sample sizes r_1, r_2 are less than about 15 each, no further calculations should be attempted. For r_1, r_2 between 15 and 30, provided one sample is not very much larger than

the other, results obtained from the following calculations will be reasonably reliable even if s_1^2 and s_2^2 do differ, while samples larger than 30 can be treated as 'large' and the values of s_1^2, s_2^2 can be used instead of σ_1^2, σ_2^2 in the formulae already given above, i.e. (3.4.1) and (3.4.2).

Provided s_1^2 and s_2^2 are not significantly different, a 'pooled' estimate, s^2, of the common σ^2 is found as

$$s^2 = \frac{(r_1 - 1)s_1^2 + (r_2 - 1)s_2^2}{(r_1 + r_2 - 2)}$$

Then the quantity

$$t = \frac{(\bar{x}_1 - \bar{x}_2) - (\mu_1 - \mu_2)}{\sqrt{s^2\left(\frac{1}{r_1} + \frac{1}{r_2}\right)}}$$

follows Student's t-distribution with $(r_1 + r_2 - 2)$ d.f. for any values of r_1 and r_2. Just as we wrote V for the known variance of the difference of means, i.e.

$$V = (1/r_1 + 1/r_2)\sigma^2$$

so we will write

$$\hat{V} = (1/r_1 + 1r_2)s^2.$$

Here $\hat{V}$ is an estimate of V found from using s^2 as an estimate of σ^2. In using it we shall have to depart from the normal distribution and go to the t-distribution instead. Hence a 95 percent confidence interval for the true value of the difference $(\mu_1 - \mu_2)$ is given by

$$(\bar{x}_1 - \bar{x}_2) - t_{(5\%)}s\sqrt{V} < (\mu_1 - \mu_2) < (\bar{x}_1 - \bar{x}_2) + t_{(5\%)}s\sqrt{\hat{V}} \qquad (3.4.3)$$

instead of (3.4.1). Here s^2 is the pooled estimate of variance and $t_{(5\%)}$ is the 5 percent point of t (i.e. $P = 0.05$) with $(r_1 + r_2 - 2)$ d.f. In the same way the test of the hypothesis that $(\mu_1 - \mu_2)$ equals D is to compare

$$t = \frac{(\bar{x}_1 - \bar{x}_2) - D}{\sqrt{\hat{V}}} \qquad (3.4.4)$$

with table values of Student's t with $(r_1 + r_2 - 2)$ d.f. Two-tail or one-tail

tests, depending on the alternative hypothesis, are carried out as described in Section 3.2.

With the appearance of (3.4.3) and (3.4.4) we enter the realm of comparative experiments. Means are not being compared with constants but with one another. That is especially relevant in an agricultural context, where no one expects the yield of a crop to be the same in all locations and at all times, but there is nevertheless a reasonable expectation that differences will be reliable. In other words, if we compare two treatments and observe an advantage for one of them at one place, we shall expect to find a similar advantage elsewhere and in different seasons, provided that conditions are broadly similar.

3.5 Variance in samples from normal distributions

The unbiased estimate of σ^2 is found from a sample $\{x_1, x_2, \ldots, x_r\}$ of r observations as

$$s^2 = \frac{1}{(r-1)}\sum_{i=1}^{r}(x_i - \bar{x})^2.$$

For use in estimation and testing, the result needed is that $(r-1)s^2/\sigma^2$ follows the chi-squared distribution with $(r-1)$ d.f., $\chi^2(r-1)$. [In the unusual case where the population mean is known, the best estimator of the variance is

$$s_o^2 = \frac{1}{r}\sum_{i=1}^{r}(x_i - \mu)^2, \quad \text{and} \quad \frac{rs_o^2}{\sigma^2} \quad \text{follows } \chi^2_{(r)}.]$$

Tables of χ^2 contain a row for each number of degrees of freedom, and they give the values above which p percent of the distribution lies, for a selection of P-values such as 99 percent, 97½ percent, 95 percent, 5 percent, 2½ percent, 1 percent. To find a 95 percent confidence interval for σ^2, when μ is not known, use

$$Prob\left(\chi^2_{(L)} < \frac{(r-1)s^2}{\sigma^2} < \chi^2_{(U)}\right) = 0.95$$

in which $\chi^2_{(L)}$ and $\chi^2_{(U)}$ are the 97½ percent and 2½ percent points in the tables for $(r-1)$ d.f. This gives

$$Prob\left(\frac{(r-1)s^2}{\chi^2_{(U)}} < \sigma^2 < \frac{(r-1)s^2}{\chi^2_{(L)}}\right) = 0.95 \qquad (3.5.1)$$

as the 95 percent interval for σ^2 based on the sample.

In order to test a hypothesis that the true value of the variance in a population is σ^2, when we have a random sample of r observations from it whose estimated variance is s^2, the value $\frac{(r-1)s^2}{\sigma^2}$ is calculated. If this lies between $\chi^2_{(L)}$ and $\chi^2_{(U)}$ (as defined above) we can accept that σ^2 is a reasonable value for the true variance.

When comparing two estimated variances, to test the hypothesis that the samples giving them were drawn from populations with the same true value of variance, it is their ratio that is required. If the estimates are s_1^2 and s_2^2, from samples of r_1 and r_2 observations respectively, s_1^2/s_2^2 follows the F-distribution with $(r_1 - 1)$ and $(r_2 - 1)$ d.f. It is important to keep d.f. in the right order. Because of the way in which F tables are usually printed it is also necessary to write the ratio so that s_1^2 is the larger estimate, and the ratio is greater than 1. When the calculated ratio is significantly greater than 1, i.e. greater than the value in the table at the chosen probability level ($P = 5$ percent, 1 percent, etc.), we should not accept a hypothesis that says the two population variances were equal.

We have here reached a test that has special relevance to the analysis of variance. In Chapter 1 we obtained some simple examples of such analyses. Their common feature is the obtaining of two variances, one for treatments and the other for 'error', and we did in fact work out F as their ratio. We are now in a position to say whether these F-values are significant or not. If there were no differences due to treatments, the two mean squares would be measuring the same σ^2 and F would, within statistical limits, be 1.00. If F so far exceeds that value that the probability of its doing so by chance is small ($P < 0.05$, say, or some other low value) then we are forced to consider whether there may indeed be effects of treatments. If there are, they will inflate the treatment mean-square but not that for 'error'. (Note by the way, that the F-test uses only one tail. No provision is made for the case where the mean square for treatments is less than that for 'error'.)

At this point someone may be surprised that we raise the question at all. Of course, he may say, there are treatment differences. Why, do we suppose, is the experiment being done at all if none are expected? The question is a fair one; in many instances an F-test is quite unnecessary. The treatments have been included in the full knowledge that they must have an effect. Sometimes, however, there can be doubts, e.g. a strange scorch has appeared on the leaves and someone suggests that it indicates a deficiency of magnesium. In that case, a test is called for to see if there is a difference between plots that have received a dressing of magnesium sulphate and those that have not. As with all these questions, the important thing is to see clearly what the experiment is intended to do. If

the general effects of the treatments can be foreseen and the enquiry concerns only the magnitudes of those effects, no test is needed (least of all an F-test), only estimates. If, on the other hand, a 'Yes or No' answer is required to the question whether there are any treatment differences, a test is in order. Even then there may be no place for an F-test but rather for a series of t-tests of the kind considered in (3.4.3), mostly with $D = 0$.

3.6 More general contrasts among means assuming orthogonality

In Chapter 5 we shall examine in some detail contrasts between the treatments, that are more complicated than simple ones like $\bar{x}_1 - \bar{x}_2$, but we will make a start now. Suppose that one treatment, A, is a standard method of cultivating a crop, while two other methods, B and C, both involve using additional fertilizer but in different ways. An interesting comparison, or contrast, among the means is to look at the difference between A and the average of B and C: i.e. to consider $\bar{x}_A - \frac{1}{2}(\bar{x}_B + \bar{x}_C)$. We shall now examine the position for orthogonal designs.

This is an example of a general contrast

$$l\bar{x}_A + m\bar{x}_B + p\bar{x}_C,$$

where l, m, p are numbers ('constants'), here 1, $-\frac{1}{2}$, $-\frac{1}{2}$, respectively. They must sum to zero. Provided the samples of experimental plots used for each treatment were chosen independently (in a properly randomized experiment) the variance of the contrast will be

$$l^2\text{var}(\bar{x}_A) + m^2\text{var}(\bar{x}_B) + p^2\text{var}(\bar{x}_C).$$

Also this contrast will follow a normal distribution, so long as the distribution of each x is normal. Often each treatment will have the same replication, but provided a design is orthogonal, the treatment means will still be estimated independently of each other (as for example in Section 5.2) without having to be equally replicated. If A, B, C are replicated r_A, r_B, r_C times respectively, then the variance of $l\bar{x}_A + m\bar{x}_B + p\bar{x}_C$ is

$$l^2\frac{\sigma_A^2}{r_A} + m^2\frac{\sigma_B^2}{r_B} + p^2\frac{\sigma_C^2}{r_C}. \qquad (3.6.1)$$

In experimental work we can usually assume $\sigma_A^2 = \sigma_B^2 = \sigma_C^2 = \sigma^2$ say, and an estimate of σ^2 is found from the residual 'error' mean square in analysis of variance. This estimate has the same degrees of freedom as the 'error' mean square, i.e. f. The estimated variance of the contrast is then

$$s^2\left(\frac{l^2}{r_A}+\frac{m^2}{r_B}+\frac{p^2}{r_C}\right) \tag{3.6.2}$$

and it has f d.f.

In the example above, $\bar{x}_A - \frac{1}{2}(\bar{x}_B + \bar{x}_C)$ $(= d)$ is an estimate of $\mu_A - \frac{1}{2}(\mu_B + \mu_C)$, and it has variance

$$s^2\left(\frac{1}{r_A}+\frac{1}{4r_B}+\frac{1}{4r_C}\right) = V, \text{ say,}$$

with f d.f. Then a 95 percent confidence interval ($P = 0.05$) for the true value $\mu_A - \frac{1}{2}(\mu_B + \mu_C)$ $(= \delta)$ is

$$d - t\sqrt{V} < \delta < d + t\sqrt{V} \tag{3.6.3}$$

in which t stands for the upper and lower 5 percent points of Student's t with f d.f.

Although significance tests for contrasts are usually done in an analysis of variance as F-tests, there is an equivalent t-test of the hypothesis that the true value of the contrast is zero. In the example above, if

$$\mu_A = \tfrac{1}{2}(\mu_B + \mu_C) \tag{3.6.4}$$

then

$$t = \frac{\bar{x}_A - \frac{1}{2}(\bar{x}_B + \bar{x}_C)}{\sqrt{V}}$$

follows the t-distribution with f d.f. and can be tested in the same way as any other t-variable.

3.7 The Binomial distribution and proportions

When counting the number of items of a special type among a random sample of n members from a population, let us denote the number of the special type found in the sample by r. Suppose the following conditions hold:

(i) the sample size n is fixed before selection begins;
(ii) the proportion of special type in the whole population is p, and this remains unchanged throughout the sampling, as each fresh member is selected: the population being sampled must therefore be large, or at least capable of regenerating according to the same rules, as in genetic inheritance.

Then r follows the biomial distribution, given by

$$Prob\ (r) = \frac{n!}{r!(n-r)!}p^r(1-p)^{n-r} \quad \text{for } r = 0, 1, 2, \ldots, n$$

where $n!$ stands for n-factorial, i.e.

$$n! = n \times (n-1) \times (n-2) \times \ldots \times 3 \times 2 \times 1.$$

After repeated sampling, each sample being of size n, the distribution of r has mean np and variance $np(1-p)$.

Often the sample proportion $\hat{p} = r/n$ is of interest; this has a distribution whose mean is p and variance $p(1-p)/n$. It follows that the distribution is not going to be exactly normal. For one thing there is no constant variance, σ^2; for another, the distribution is not symmetrical unless $p = 0.5$. However, when n is large (in practice, greater than 100 is usually satisfactory) and p is not too near 0 or 1 (say in the range 0.1 to 0.9) the distribution of a sample proportion can be taken as approximately normal, with mean p and variance $p(1-p)/n$. Confidence intervals and significance tests can therefore be calculated as though we were dealing with a normal distribution, though the results obtained are approximate rather than exact theoretically.

A 95 percent confidence interval for the true value of p, based on a sample of n giving a sample proportion $\hat{p}$, is given by

$$\hat{p} - 1.96\sqrt{\frac{\hat{p}(1-\hat{p})}{n}} < p < \hat{p} + 1.96\sqrt{\frac{\hat{p}(1-\hat{p})}{n}} \tag{3.7.1}$$

The hypothesis that p is the true proportion is tested by calculating

$$Z = \frac{\hat{p} - p}{\sqrt{p(1-p)/n}}$$

and testing it as a standard normal variable. We note that (3.7.1) is an analogue of (3.4.1) with $V = p(1-p)/n$.

If two samples of sizes n_1 and n_2 are available, giving sample proportions $\hat{p}_1 = r_1/n_1$ and $\hat{p}_2 = r_2/n_2$ of the same special type of member, it is often required to test the hypothesis that the true proportions are the same, i.e. $p_1 = p_2 = p$. If this is true then $\hat{p}_1$ has variance $p(1-p)/n_1$ and $\hat{p}_2$ has variance $p(1-p)/n_2$; the best estimate of p using all the data from both samples is $\hat{p} = (r_1 + r_2)/(n_1 + n_2)$ and this is what must be used in the test. The variance of $(\hat{p}_1 - \hat{p}_2)$ is

$$\left[\frac{\hat{p}(1-\hat{p})}{n_1}+\frac{\hat{p}(1-\hat{p})}{n_2}\right]$$

which we write S^2 for short. If $p_1 = p_2$ then $Z = (\hat{p}_1 - \hat{p}_2)/S$ is approximately a standard normal variable and can be treated as in Section 3.2.

An approximate 95 percent confidence interval for the true difference $p_1 - p_2$ does not of course make the assumption that they have common value p, and therefore the expression for variance is different from S^2: it is instead

$$\left(\frac{\hat{p}_1(1-\hat{p}_1)}{n_1}+\frac{\hat{p}_2(1-\hat{p}_2)}{n_2}\right) = V, \text{ say.}$$

Then a 95 per cent confidence interval for $(p_1 - p_2)$ follows from

$$Prob\left((\hat{p}_1-\hat{p}_2)-1.96\sqrt{V}<(p_1-p_2)<(\hat{p}_1-\hat{p}_2)+1.96\sqrt{V}\right)=0.95 \quad (3.7.2)$$

This is a further analogue of (3.4.1).

The methods described above are satisfactory for simple tests and inferences about binomially distributed variables and proportions. But observations of $\hat{p}$ can only be subjected to analysis of variance after transformation (see Chapter 9).

3.8 Measurements following the Poisson distribution

When counting items that arise independently of one another, at random in space (or time), the Poisson distribution may be appropriate. If the items (for example, insects, lesions on a leaf, or weeds) occur at a constant average rate of m per unit area, and if a large number of unit areas are counted, the actual number of items in each unit being c, then the distribution of c follows the Poisson. If the rate per unit area, m, does not remain constant over a complete population of units being studied then the Poisson distribution will not be a suitable model. Another case where it does not work is when the items being counted are not fully independent of one another but tend to arise in groups.

The mean and the variance are both equal to m in a Poisson distribution, and if m is greater than about 5, the Poisson variable c can be well approximated by a normal distribution whose mean and variance are both m. These results lead to approximate confidence intervals and to methods of testing which are similar to those already described. If a single Poisson variable c has variance m, then the mean of n observations $\bar{c}$ will have variance m/n. Hence an approximate 95% confidence interval for the true value of m, using its sample estimate $\bar{c}$, is given by

$$Prob\left(\bar{c}-1.96\sqrt{\frac{\bar{c}}{n}}<m<\bar{c}+1.96\sqrt{\frac{\bar{c}}{n}}\right)=0.95. \quad (3.8.1)$$

Also, the sample may be used to test a hypothesis that the true mean is M, by calculating $Z = (c - M)/\sqrt{M/n}$ and treating Z as a standard normal variable (as described in Section 3.2).

Poisson distributions often form suitable models also for counts taken over time, such as numbers of radioactive particles emitted per minute from a source, and for counts per unit volume such as the number of organisms per ml of liquid. The same conditions must hold, namely that items arise individually (not in groups) at random at a constant average rate (per minute or per ml).

As in the binomial case, a transformation (see Chapter 9) is needed if data following Poisson distributions are to be used in an analysis of variance.

3.9 Uses of tests and estimates

As was remarked in Section 3.3, skill in data analyses lies largely in knowing what to test and what to estimate. Several methods for doing what may be appropriate have been set out in this chapter, but when should each be used?

The answer in any instance must depend upon the questions asked of an experiment. Unless they are clearly understood, the answers may well be irrelevant to the experimenter's needs. The quality chiefly needed in planning an experiment is clear-headedness. Once the experimental problem is perceived with sufficient clarity, it is to be hoped that someone has the statistical skill to solve it. If no one has the skill, it can perhaps be acquired by reading, whether in this book or another. There is nothing to be said for by-passing the whole consideration and using some test because it looks impressive or because the computer package makes it available.

As long as there are only two treatments, all tests come to the same conclusion. As will be explained in the next Section, the t-test at (3.4.4) with $D = 0$ is the same as the F-test in Section 3.5. The difficulties come when there are more than two treatments, because then there are several contrasts that could be examined. Which are the important ones and how are they related?

We will suppose that there are four treatments, A, B, C, D, and we will suppose that they give means of 25, 29, 28 and 32 units respectively. If we are going to rely upon t-tests, we have to look at all six differences, i.e.

(A − B), − 4 (B − C), + 1

(A − C), − 3	(B − D), − 3
(A − D), − 7	(C − D), − 4

We might find that only (A − D) was significant at $P = 0.05$. We can scarcely declare in general that there are treatment differences when only one out of six has shown up at the level of one-in-twenty. On the other hand, an *F*-test, which averages the six contrasts, may declare them collectively not to be significant. A procedure that allows a genuine effect to be missed by diluting it with a lot of others is equally unsatisfactory. What then is to be done?

One much-used technique is that of multiple comparisons based on one of the various multiple range tests. We strongly urge you not to use them. What they do is to search out the largest difference, in this example (A − D), and to examine its significance, bearing in mind that it has been selected from six possible candidates. As a piece of probability theory that is all right, but it does not relate to practicalities. The treatments are not nameless, their characters being of no importance, but a set, carefully selected to answer the questions under study, and we must know what those questions are. A procedure that ignores purpose and would give the same answer whatever the nature of A, B, C and D is clearly inadequate, if not downright wrong. The correct procedure is that presented in Chapter 5, i.e. the purpose of the experiment should be expressed as contrasts of interest, which are then tested or estimated. When we can write down the questions in a form that relates to the analysis of variance, we can try to use our analysis to find the answers.

Is there then no need for the *F*-test, which is really a test of a composite hypothesis? It can in fact be useful in answering general questions, especially in conditional situations. To return to the example with four treatments, A, B, C and D, we might learn that the treatments represented increasing applications of fertilizer. Further enquiry to elucidate the questions might reveal that there were two, the second conditional upon the first:

(i) Do the figures suggest a departure from a straight-line relationship of yield on fertilizer application?
(ii) *If so*, what is the shape of the response curve?
If not, what is the ratio of increased yield to increased fertilizer?

The general question at (i) calls for a test to decide what to do at (ii).

However, as has been said, estimation is more usually appropriate than testing, because mostly people know more or less what will happen and they want it quantified. That in no way diminishes the need to find out exactly the purpose of the investigation. When we give a figure for the quantity being estimated, we must append a variance or a standard error

so that a judgement can be made as to the precision of our estimate. We have mentioned this in Section 3.6 and we shall return to it in Section 5.2.

3.10 Computational procedures

Before leaving the subject some words are needed on computation. From what has been said it will be clear that the difference between estimation and testing is one of logic. The aim is to provide answers relevant to the questions being asked. When it comes to the calculations, however, there is little difference. For most of the procedures already presented and for most of those that will come later, the main arithmetical operations are the same.

(i) Data are collected and it is believed that they are all subject to the same sources of variance, the effect of which is represented by σ^2. (Note that the Binomial and Poisson distributions negate that assumption from the start.)

(ii) Methods are sought of finding a quantity, s^2, that will provide an estimate of σ^2. Once s^2 is found, we call s 'the standard error of an observation'. Sometimes the variance is found as in (3.3.1), but in the analysis of variance, s^2 is taken to be the 'error' mean-square as at (1.3.3). Sometimes other methods are used.

(iii) Each standard error, s, when found is associated with a certain number of degrees of freedom, which measure the amount of information used in its estimation. If there were an infinite number of degrees of freedom, Section 3.3 would reduce to Section 3.2, i.e. σ would be known. At the other extreme, with few data, the number of degrees of freedom may be barely sufficient.

(iv) Attention is then concentrated on some quantity A, chosen because it gives the required information. Thus, in Section 3.3 A was the mean of a sample of n data. The mathematicians are now called in to find the standard error of A in terms of s. In the case of the sample mean they reply that it is $s/\sqrt{n}$. In other cases they may give expressions that are more complicated.

At this point estimation and testing diverge though not in arithmetic. In estimation, confidence limits are put round the estimate of A with limits $A \pm t \times$ (the standard error of A), where t is looked up in tables using the appropriate significance level and the number of degrees of freedom. That was done at (3.6.3). If, on the other hand, someone wants to test whether A has its expected value, a, the procedure is to take $(A - a)$, divide by the standard error of A and ask whether the result is less than t, the same value being used as before. That was done at (3.6.4), a being zero. In a sense this comes down to asking whether a lies within the confidence limits of A. The difference lies not in the arithmetic but in the motivation for doing it.

The F-test may appear to be different and it is so because it tests a composite hypothesis. It does not ask if A differs from B, but whether A, B, C, . . ., etc. differ among themselves. That is both its strength and its weakness. It asks a general question, which can also be a confused one. If, however, there are only two quantities, A and B, to be compared, it becomes in effect a t-test. That can be seen easily by looking at tables. If there is only one contrast under study, the F-test has one and f degrees of freedom. Then at any significance level, $F = t^2$ where t has f degrees of freedom. To take an example, for $P = 0.01$ and twelve degrees of freedom, $t = 3.055$. For the same P and (1, 12) degrees of freedom. $F = 9.33 = t^2$. In fact, whenever the two tests are both available, they will always give the same answer.

Note: The general practice in this text will be to cite F-values in the analyses of variance as a matter of course, despite the reserve expressed in Section 3.9 as to their not always being appropriate. We shall do so because an F-value gives a general indication how far there are treatment differences to be sought, though it will be understood that there could be important effects of treatments when the overall F is not significant. Further, we shall adopt the convention of using one asterisk, *, to indicate a significance level of $P = 0.05$, two, **, for $P = 0.01$ and three, ***, for $P = 0.001$.

Exercise 3A

64 observations are selected at random from a normal distribution whose variance is 25. Their mean is calculated and found to be 11.1. Test the hypothesis that the true value of the population mean is 10.

Exercise 3B

200 observations are selected at random from a distribution whose mean is thought to be 5 and variance known to be 8. The mean of the 200 observations is 4.77. Test the hypothesis that the population mean is 5. Repeat this for a sample size of 20 with the same mean instead of 200.

Exercise 3C

A group of 10 strawberry plants is grown in ground treated with a chemical soil-conditioner, and the mean yield per plant is 114 g. Experience has shown that when the same variety of strawberry is grown under similar conditions, but with no soil-conditioner, the mean has been 110 g and the variance 84. Test whether it can reasonably be claimed that the soil-conditioner had a beneficial effect on yield.

Exercise 3D

The weights of a large number of plants taken at random in a region have been measured, and the mean and variance in this population are respectively 102 lb and 49 lb^2. If 100 plants from an adjacent region are weighed in bulk, and their mean weight is 99 lb, is there any evidence that the plants in the second region are of a different size from that in the first? If only five plants had been available, how would you examine their weight records?

Exercise 3E

8 observations from a normal distribution were 1.6, − 0.8, 0.1, − 0.4, 1.2, 0.7, 0.3, 0.5. Test the null hypothesis that the distribution has mean 0.1. Also set 95 percent and 99 percent confidence limits to the true value of this mean.

Exercise 3F

A random sample of 25 seeds of a given variety is planted in pots. The mean time from planting to opening of the first leaflet is 5.8 days. The variance estimated from the data was 4.84. Assuming that this time is normally distributed, test the hypothesis that its mean is 4. Also set 99 percent confidence limits to its true value.

Exercise 3G

Two samples of observations on the diameters of fungal mycelium colonies gave these results: Sample A of 11 observations had mean 6.65

and variance 15.2824, while Sample B of 16 observations had mean 4.28 and variance 8.0275. (Both variances were estimated from the data.) Assume the diameter to be normally distributed. Find a pooled estimate of variance (check that it is valid to 'pool'). Test the null hypothesis that the samples came from distributions with the same mean. Also set, separately for Samples A and B, 95 percent confidence intervals for the true values of their means. Comment on the results.

Exercise 3H

Soya bean seedlings were grown in pairs of adjacent pots, one pot of each pair being watered twice as often as the other (with half the volume each time). The differences in height at the end of a given period of time (expressed as more regular *minus* less regular in each pair) were + 6.0, + 1.3, + 3.1, + 6.8, − 1.5, + 4.2, − 3.3, + 2.7, + 10.2, + 0.1, − 0.4 mm. Test whether regularity of watering made a significant difference to height.

Also set 95 percent confidence limits to the true difference in height due to regularity of watering.

Exercise 3I

Seedlings from a specially bred population have in the past shown normally distributed weight increases, with variance 15 units, when grown in standard conditions for a fixed period. Six new seedlings are selected at random, and grown for the same fixed time at a higher temperature. Their weight increases are 18, 21, 12, 16, 25, 20 units. Are they more variable in growth than those fed on the standard diet?

Exercise 3J

Repeat Exercise 3I, assuming that any change in variability due to a higher temperature will be an increase and cannot be a decrease.

Exercise 3K

The height to which seedlings of two apple varieties (A and B) grow in

standard conditions in a greenhouse is assumed to be normally distributed. From A, 10 seedlings grow 44, 26, 1, 79, 53, 38, 62, 80, 33, 13 cm and from B, 12 seedlings grow 33, 47, 55, 39, 24, 61, 38, 12, 26, 64, 52, 51 cm. Test the null hypothesis that the variance of height is the same in both varieties. Also set 95 percent confidence limits to the true value of variance for each variety.

Exercise 3L

A large normal population has mean 100 and variance 10. Find 95 percent confidence limits to

(a) the value of one single observation drawn from this population.
(b) the mean value of 10 randomly selected observations from this population,
(c) the mean of 100 randomly selected observations from this population.

If the variance had not been known, how could limits be found?

Exercise 3M

A sample of 9 plants of the same variety were grown in one type of soil in a greenhouse and after a fixed time they were removed and dried. Their dry weights were 25.5, 22.3, 24.7, 28.1, 26.5, 19.0, 31.0, 25.3, 29.6 g. A further sample of 11 similar plants were grown in identical conditions but in another type of soil. Their dry weights were 31.8, 30.3, 26.4, 24.2, 27.8, 29.1, 25.5, 28.9, 30.0, 26.9, 29.7 g. Do the two soil types have different effects on the plants?

Exercise 3N

Two formulations of an insecticide are tried out by a number of farmers. Preparation A is given to 250 farmers, of whom 172 claim it is effective; B is given to 200, of whom 158 say it is effective. Is there evidence of a difference between A and B? (It may be assumed that the 450 farmers were fairly chosen at random.) Also find the approximate confidence limits (95 percent) for the proportion of satisfied farmers who had used

each of A, B. For preparation A, how many farmers would be needed in order to estimate the proportion of successes to within ± 3% (with 95 percent confidence)?

Exericse 3O

50 samples, each of unit volume, were drawn at random from a liquid suspension containing cells. Each sample was examined under a microscope, on a slide, and the average number of cells per unit volume for these 50 samples was 4. Set approximate 95 percent confidence limits to the mean number of cells per unit volume in the whole suspension.

Chapter 4

Analysis of variance from non-orthogonal designs

4.1 Why use non-orthogonal designs?

Although there are considerable simplifications resulting from the use of orthogonal designs as defined in Sections 1.3 and 1.5, there are times when the experimenter has to resort to non-orthogonality. For example, the land may be a narrow strip running downhill and only wide enough to take at most three plots across itself. Clearly, blocks should be formed on the basis of altitude, which means that each must contain three plots of minimum size or perhaps two larger ones. If the number of treatments exceeds three, a non-orthogonal design is unavoidable. If there were four, to take the example further, it would be possible to leave out each treatment in turn, like this:

Block						
	I	B	A	D	leaving out	C
	II	C	B	A	leaving out	D
	III	B	D	C	leaving out	A
	IV	C	A	D	leaving out	B

Sometimes too the design was originally orthogonal, but someone made a mistake in executing it and the experimenter is left to analyse the data that have in fact been obtained, though the design is not what was intended. The situation may be quite complicated.

There is no need to be afraid of non-orthogonality. It is better avoided because of the complications it causes, but if no orthogonal design will fit the site or if mistakes have been made, the data can be analysed in a completely valid manner. In Sections 4.2 and 4.5 a method will be given that can be used with any block design, provided it has been randomized. (That is not quite true because the method of finding variances, given in Section 4.3, can cause difficulties with some of the 'confounded' designs to be presented in Chapter 7, but those difficulties can be overcome.) Simpler methods will be described (Sections 4.7 to 4.12) for some of the commoner forms of non-orthogonality that might be introduced deliberately. (Where

it was not intended but arose as the result of an accident, mostly the general method has to be used.)

In these days of computers it may be asked why analytical difficulties should stand in the way of using desirable designs. The question is a good one, but non-orthogonal designs have a further disadvantage. It relates to the variances of contrasts. If the 'error' mean square is written as s^2,

$$\text{the variance of a contrast} = Ks^2, \tag{4.1.1}$$

where the constant, K, depends upon the contrast seen in relation to the design. The expressions (3.6.1) and (3.6.2) give K for orthogonal designs. For non-orthogonal designs K can never be less than the values so obtained and usually it will be more; that is to say, the variances of contrasts may be inflated. (In choosing a non-orthogonal design a large part of the skill lies in arranging that the contrasts of least interest shall be the ones to suffer most.) To understand exactly where the inflation occurs it is necessary to work out an array of figures called the 'covariance matrix'. Its full use will become apparent in Chapter 5; here it will be used only in a limited way.

The mention of matrices may alarm some readers, but there is no need for that. For purposes of reading the present text it is necessary only to know what a matrix is. It is an array of numbers, written in rows and columns, such that meaning can be attached both to the rows and to the columns. It is in fact what most people call a 'table of figures'. It is true that mathematicians have developed an advanced system of algebra to deal with matrices and true also that nowadays the theory of experiment designs depends upon that system. Nevertheless no knowledge of matrix algebra is required for an understanding of this book. To continue with nomenclature, the numbers entered in a matrix are called its 'elements'. There will be one concession to the mathematicians. Where a symbol represents a group of values, not a single value, following the practice in matrix algebra it will be printed in bold face type.

In this chapter the analysis of variance for a non-orthogonal design will be treated in the same way that Sections 1.3 and 1.4 dealt with the analysis for orthogonal designs. That is to say, only arithmetic will be considered. Interpretation can come later.

4.2 The analysis of variance for any block design

The following method ('the Kuiper–Corsten iteration') applies whether the design is orthogonal or not. (If it is orthogonal, simpler methods are available.)

Consider the following data from a non-orthogonal design:

Block	I	A, 14	C, 16	D, 14	E, 17			
	II	B, 12	A, 11	D, 13	C, 16			
	III	E, 16	C, 17	B, 17	A, 14			
	IV	D, 19	B, 15	E, 16	A, 9			

(4.2.1)

The block means are I, 15.25; II, 13.00; III, 16.00; IV, 14.75. Sweeping by them gives

Block	I	A, −1.25	C, +0.75	D, −1.25	E, +1.75			
	II	B, −1.00	A, −2.00	D, 0.00	C, +3.00			
	III	E, 0.00	C, +1.00	B, +1.00	A, −2.00			
	IV	D, +4.25	B, +0.25	E, +1.25	A, −5.75			

(4.2.2)

The deviations correctly add to zero in each block. The treatment means are now A, −2.75; B, +0.08; C, +1.58; D, +1.00; E, +1.00. They will be called collectively, $\mathbf{v}_1$. Sweeping by them gives

Block	I	A, +1.50	C, −0.83	D, −2.25	E, +0.75			
	II	B, −1.08	A, +0.75	D, −1.00	C, +1.42			
	III	E, −1.00	C, −0.58	B, +0.92	A, +0.75			
	IV	D, +3.25	B, +0.17	E, +0.25	A, −3.00			

(4.2.3)

Although these quantities sum to zero over treatments they do not do so over blocks, as residuals should. (If the design had been orthogonal they would have done so.) As it is, they give block means of

$$\text{I, } -0.21; \text{ II, } +0.02; \text{ III, } +0.02; \text{ IV, } +0.17,$$

which quantities will be called $\mathbf{u}_1$. Sweeping by $\mathbf{u}_1$ gives

Block	I	A, +1.71	C, −0.62	D, −2.04	E, +0.96			
	II	B, −1.10	A, +0.73	D, −1.02	C, +1.40			
	III	E, −1.02	C, −0.60	B, +0.90	A, +0.73			
	IV	D, +3.08	B, 0.00	E, +0.08	A, −3.17			

(4.2.4)

In turn (4.2.4) gives rise to treatment means, $\mathbf{v}_2$, namely

$$\text{A, } 0.00; \quad \text{B, } -0.07; \quad \text{C, } +0.06; \quad \text{D, } +0.01; \quad \text{E, } +0.01.$$

Clearly this process can continue, $\mathbf{v}_2$ forming $\mathbf{u}_2$, $\mathbf{u}_2$ forming $\mathbf{v}_3$ and so on until a set of zeros is obtained. The deviations obtained at that stage can fairly be regarded as residuals, since they will sum to zero over both blocks

and treatments. In effect the original data at (4.2.1) will have been swept first by blocks and then by $\mathbf{V} = \Sigma \mathbf{v}_j$ and $\mathbf{U} = \Sigma \mathbf{u}_i$.

These calculations are best set out in the form of the Kuiper–Corsten iteration. There is one further point of nomenclature: the symbol **Q** is used to represent the treatment totals in (4.2.2). Here **Q** equals (− 11.00 + 0.25 + 4.75 + 3.00 + 3.00) for the treatments A to E in order.

The first step is to write down the 'incidence matrix'. It has a row for each treatment and a column for each block. The values in it show the number of times that treatment occurs in that block. To the right of it is written $\mathbf{v}_1$, but space has to be left for $\mathbf{v}_2$, $\mathbf{v}_3$, etc., which will be derived later.

				$\mathbf{v}_1$
1	1	1	1	− 2.75
0	1	1	1	+ 0.08
1	1	1	0	+ 1,58
1	1	0	1	+ 1.00
1	0	1	1	+ 1.00

(4.2.5)

The next step is to work out $\mathbf{u}_1$. It is formed by multiplying out successive columns of the incidence matrix and $\mathbf{v}_1$; the result is then divided by the respective block size, i.e.,

$$\begin{aligned} \mathbf{u}_1 = (-2.75 + 1.58 + 1.00 + 1.00)/4 &= +0.21 \\ (-2.75 + 0.08 + 1.58 + 1.00)/4 &= -0.02 \\ (-2.75 + 0.08 + 1.58 + 1.00)/4 &= -0.02 \\ (-2.75 + 0.08 + 1.00 + 1.00)/4 &= -0.17 \end{aligned}$$

It should be written horizontally below the incidence matrix. Then each row of the incidence matrix is successively multiplied out by the values of $\mathbf{u}_1$ and then the sum divided by the appropriate treatment replication, to form $\mathbf{v}_2$, i.e.

$$\begin{aligned} \mathbf{v}_2 = (\ 0.21 - 0.02 - 0.02 - 0.17)/4 &= \ 0.00 \\ (-0.02 - 0.02 - 0.17)/3 &= -0.07 \\ (\ 0.21 - 0.02 - 0.02)/3 &= +0.06 \\ (\ 0.21 - 0.02 - 0.17)/3 &= +0.01 \\ (\ 0.21 - 0.02 - 0.17)/3 &= +0.01 \end{aligned}$$

Those values are then written to the right of $\mathbf{v}_1$. Next $\mathbf{u}_2$ is formed from $\mathbf{v}_2$ in the same way that $\mathbf{u}_1$ was formed from $\mathbf{v}_1$, i.e.

$$\begin{aligned} \mathbf{u}_2 = (\ 0.00 + 0.06 + 0.01 + 0.01)/4 &= +0.02 \\ (\ 0.00 - 0.07 + 0.06 + 0.01)/4 &= \ 0.00 \end{aligned}$$

$$\begin{aligned} (0.00 - 0.07 + 0.06 + 0.01)/4 &= \ 0.00 \\ (0.00 - 0.07 + 0.01 + 0.01)/4 &= -0.01 \end{aligned} \qquad (4.2.6)$$

The values of $\mathbf{u}_2$ are written horizontally below $\mathbf{u}_1$ and they give rise to $\mathbf{v}_3$. There is no need to go on because all the values are now less than 3% of the precision to which the data were measured, which was to the nearest unit. All values in $\mathbf{v}_3$ lie between ± 0.03 so the iteration can be discontinued. The treatment effects, **V**, can be found by summing $\mathbf{v}_1$, $\mathbf{v}_2$, $\mathbf{v}_3$, etc., i.e.

$$-2.75 \quad +0.01 \quad +1.65 \quad +1.01 \quad +1.01 \qquad (4.2.7)$$

for the five treatments, A to E in that order. However, these changes in the estimates of treatment effects need consequent changes in the estimates of block effects. They are found by calculating **U**, which is the sum of $\mathbf{u}_1$, $\mathbf{u}_2$, etc., i.e. I, + 0.23; II, − 0.02; III, − 0.02; IV, − 0.18.

With these revised figures it becomes possible to calculate the residuals again. Thus, to take the first plot (Treatment A in Block I) its residual is obtained from its deviation (− 1.25) by adding the element of **U** (+ 0.23) and subtracting the treatment effect, i.e. the element of **V** (− 2.75). That gives 1.73. The residuals are therefore:

Block	I	A, + 1.73	C, − 0.67	D, − 2.03	E, + 0.97
	II	B, − 1.03	A, + 0.73	D, − 1.03	C, + 1.33
	III	E, − 1.03	C, − 0.67	B, + 0.97	A, + 0.73
	IV	D, + 3.05	B, + 0.05	E, + 0.05	A, − 3.19

It will be seen that they now correctly sum to zero in all blocks and in all treatments, at least within the limits set by rounding.

The stratum total sum of squares is found in the usual way by summing the squares of the deviations set out in (4.2.2) i.e. $(-1.25)^2 + (0.75)^2 + \ldots + (-5.75)^2 = 79.5000$. It has the usual degrees of freedom, namely 12. (It may be noted that deviations should be calculated ignoring treatments; consequently, the procedure just adopted is correct despite the later adjustment of block means using **U**.) The 'error' sum of squares is likewise found by summing the squares of the residuals set out in (4.2.7) i.e. $(+1.73)^2 + (-0.67)^2 + \ldots + (-3.19)^2 = 35.394\,44$. There is, however, an independent way of obtaining the treatment sum of squares by multiplying out the values of **Q** with the treatment effects, **V**, namely

$$\begin{aligned} (-11.00 \times -2.75) + (0.25 \times 0.01) + (4.75 \times 1.65) + (3.00 \times 1.01) \\ + (3.00 \times 1.01) = 44.1500 \end{aligned} \qquad (4.2.8)$$

with 4 degrees of freedom. It will be seen that rounding errors have prevented exact agreement, but the analysis of variance can be taken to be

Source	d.f.	s.s.	m.s.	F
Treatments	4	44.10	11.03	2.49
'Error'	8	35.39	4.424	
Stratum total	12	79.50		

(4.2.9)

The discrepancy has been resolved by taking the larger of the two estimates of 'error'.

The treatment means need adjustment because some treatments do not occur in all blocks and so receive an advantage or disadvantage according as they occur in blocks that are good or bad. The adjusted means are found by adding the values of **V** to the grand mean, which in this instance is 236/16 = 14.75, i.e.

$$\begin{aligned}
&\text{Adjusted mean of A} = 14.75 - 2.75 = 12.0\\
&\text{Adjusted mean of B} = 14.75 + 0.01 = 14.8\\
&\text{Adjusted mean of C} = 14.75 + 1.65 = 16.4\\
&\text{Adjusted mean of D} = 14.75 + 1.01 = 15.8\\
&\text{Adjusted mean of E} = 14.75 + 1.01 = 15.8
\end{aligned} \qquad (4.2.10)$$

4.3 Variances, etc.

A covariance matrix (see Section 4.1) for the adjusted treatment means can be found by the use of the iteration. To take Treatment A first, it has 4 plots, there being 16 in all. Accordingly, we take $\mathbf{v}_1$ as

$$(1/4 - 1/16, -1/16, -1/16, -1/16, -1/16)$$
$$= (+0.1875, -0.0625, -0.0625, -0.0625, -0.0625)$$

and enter the iteration with it at (4.2.5). The calculation makes **V** equal to $\mathbf{v}_1$, because $\mathbf{u}_1$ proves to be a series of zeros. That provides the first row of the covariance matrix.

Turning now to Treatment B, it has 3 plots, so $\mathbf{v}_1$ is now

$$(-1/16, 1/3 - 1/16, -1/16, -1/16, -1/16)$$
$$(0.0625, +0.2708, -0.0625, 0.0625, -0.0625).$$

Using that as the $\mathbf{v}_1$ at (4.2.5) leads to **V** as

$$(0.0625, +0.2936, -0.0701, -0.0701, -0.0701)$$

which gives the second row of the covariance matrix, and so on. Actually, there is no need to do more, because subsequent rows will be permutations of the second. The matrix is in fact:

$$\begin{pmatrix}
+0.1875 & -0.0625 & -0.0625 & -0.0625 & -0.0625\\
-0.0625 & +0.2936 & -0.0701 & -0.0701 & -0.0701\\
-0.0625 & -0.0701 & +0.2936 & -0.0701 & -0.0701\\
-0.0625 & -0.0701 & -0.0701 & +0.2936 & -0.0701\\
-0.0625 & -0.0701 & -0.0701 & -0.0701 & +0.2936
\end{pmatrix} \qquad (4.3.1)$$

As will appear in Section 5.4, it should really be multiplied throughout by the 'error' mean square i.e. 4.424. The uses of this matrix will be explained in more detail in Section 5.2. Here it suffices to show how it gives the value, K, introduced at (4.1.1). For an orthogonal design it is $(1/r_1 + 1/r_2)$, where r_1 and r_2 are the replications of the two treatments. Thus, here, if the design had been orthogonal, K for the contrast of treatments A and B would have been (1/4 + 1/3) = 0.5833. As it is, we must use the covariance matrix at (4.3.1). We add the diagonal elements that relate to the two treatments, namely, 0.1875 and 0.2936. From the total we subtract the two off-diagonal elements that relate to them, i.e., − 0.0625 (twice). That gives

$$K = 0.1875 + 0.2936 - 2(-0.0625) = 0.6061 \qquad (4.3.2)$$

which is more than 0.5833. The 'information loss', as it is called, is measured by the 'efficiency factor', which is the ratio of the two values of K, i.e. 0.5833/0.6061 = 0.962. (We may note that the off-diagonal elements are necessarily equal because a covariance matrix is always symmetric, so their equality in this instance is no coincidence.)

Turning to the contrast between treatments B and C, if the design had been orthogonal K would have been (1/3 + 1/3) = 0.6667, but for the design actually used it is

$$0.2936 + 0.2936 - 2(-0.0701) = 0.7274$$

giving an efficiency factor of 0.6667/0.7274 = 0.917. It will be noticed that the efficiency factor is not necessarily the same whatever the contrast.

The difference between the adjusted means of treatments A and B can be found from the values at (4.2.10). It is − 2.8 (= 12.0 − 14.8). Its variance found from (4.1.1) is (0.6061 × 4.424), the error mean square in

the analysis of variance being used as the value of s^2. Hence the variance is $2.681 = 1.64^2$, so the standard error is 1.64 and the difference equals 1.71 times its own standard error. That of course is the value of t as defined at (3.4.4), D being zero. Similarly the difference between treatments B and C is -1.6 and its standard error is 1.79, t being therefore 0.89.

4.4 A warning

The whole method depends upon two relationships:

(1) If each value of **u** is multiplied by the corresponding block size, products sum to zero.
(2) If each value of **v** is multiplied by the corresponding treatment replication, the products sum to zero.

Thus, in the first iteration, i.e. for $\mathbf{u}_1$ above (4.2.4),

$$4(+0.21-0.02-0.02+0.17)=0.00$$

and for $\mathbf{v}_2$ below (4.2.4),

$$4(0.00)+3(-0.07+0.06+0.01+0.01)=0.03$$

which is near enough, but such rounding errors can build up. If the check fails badly it is wise to make it work by adding or subtracting a constant from all values. Suppose, for example, that a **v** had read

$$(-1.05+1.06-0.21+0.62-0.12)$$

which gives

$$4(-1.05)+3(1.06-0.21+0.62-0.12)=-0.15.$$

It would be better to amend the **v** to:

$$(-1.04+1.07-0.20+0.63-0.11).$$

Then

$$4(-1.04)+3(1.07-0.20+0.63-0.11)=+0.01,$$

which is much better.

Since differences are preserved, comparisons between treatments are unaltered.

4.5 Two simplifications

There are two main simplifications that should be noted and looked out for. One always arises when the design is orthogonal and sometimes in other instances. It is the case when $\mathbf{u}_1 = \mathbf{O}$. As a result, $\mathbf{V} = \mathbf{V}_1$ and there is no need to continue the iteration. (That occurred in Section 4.3 when finding the first row of the covariance matrix.) The other arises quite often in cases of intended non-orthogonality and occasionally when the design has become non-orthogonal on account of an accident or mistake. It will sometimes be noted that one set of values of **u** bear a constant ratio, p, to the one before. In fact, it happened at (4.2.6) with $p = 1/12$, though more decimal places are needed to see it.

$$\mathbf{u}_1 = (+0.2083, -0.0208, -0.0208, -0.1667)$$
$$\mathbf{u}_2 = (+0.0174, -0.0017, -0.0017, -0.0139)$$

If $\mathbf{u}_2 = \mathbf{u}_1/12$ then $\mathbf{v}_3 = \mathbf{v}_2/12$, $\mathbf{u}_3 = \mathbf{u}_3 = \mathbf{u}_1/12^2$, $\mathbf{v}_4 = \mathbf{v}_2/12^2$ and so on. It is known that

$$a + ap + ap^2 + ap^3 + \ldots = a/(1-p)$$

provided $p^2 < 1$ and the summation continues without end. (Such a series is known as a geometric progression.) It follows, with $p = 1/12$ and $1/(1-p) = 12/11$, that

$$\mathbf{V} = \mathbf{v}_1 + 12/11\ \mathbf{v}_2$$

Here a = an element of $\mathbf{v}_2$ and $1/(1-p) = 12/11$. Working out **V** by this alternative method, the results agree with (4.2.7). In fact, it gives a better answer because the summation has been taken further, not stopping at $\mathbf{v}_3$, but in this instance the **v**'s are diminishing so rapidly that the advantage is small. Also

$$\mathbf{U} = 12/11\ \mathbf{u}_1.$$

In carrying out the iteration on a desk computer it is worthwhile looking out to see whether a geometric progression appears. This can save a lot of work.

4.6 Concurrences

Whenever two treatments come together in the same block, they are said to 'concur'. More generally, if treatment A occurs a times in a block and treatment B occurs b times in that block, they are said to make ab

concurrences. If such figures are summed over all blocks, the result is the number of concurrences of those two treatments in the whole experiment.

In what follows it will be assumed that all blocks are the same size, each containing k plots. The number of treatments will be written v and the number of plots, n.

4.7 Designs with total balance

If all pairs of treatments have the same number of concurrences, λ, in the experiment, the design is said to be in 'total balance'.

Block			Block		Block		
I	A B C	III	A F G	VI	B E F
II	A D E	IV	B D G	VII	C D F
		V	C E G		

Here $k = 3$, $v = 7$, $n = 21$ and $\lambda = 1$.

The above design is in 'balanced incomplete blocks', which is a particular form of total balance. The additional requirements are that all treatments are applied to the same number of plots, r, and that a treatment occurs either once in a block or not at all, never twice or more. Also $k < v$. Here $r = 3$.

Whether the design is in balanced incomplete blocks or not, if it is in total balance the following gives a general method of analysis:

(1) Sweep the data by block means to give deviations.
(2) Sum the deviations over treatments to give the 'adjusted treatment totals', $Q_1, Q_2, \ldots, Q_v$. (4.7.1)
(3) Work out the 'effective replication', R, such that $R = v\lambda/k$. In the example, $R = 7/3$. (4.7.2)
(4) The treatment sum of squares will equal
$$(Q_1^2 + Q_2^2 + \ldots + Q_v^2)/R. \quad (4.7.3)$$
(5) The quantities, Q_A/R, $Q_B/R, \ldots$, etc. are estimates of the so-called 'treatment parameters' which show the relative effects of the various treatments. They have a number of uses. For present purposes it is enough to note that the 'adjusted treatment means'—adjusted, that is to say, for the differences between blocks—are found by adding the grand mean to each treatment parameter, i.e. the adjusted mean for Treatment j is estimated by
$$\text{Grand mean} + Q_j/R. \quad (4.7.4)$$
(6) The variance of the difference between two adjusted treatment means is
$$2(\text{'Error' mean-square})/R. \quad (4.7.5)$$

If an analysis of variance is attempted by use of the Kuiper–Corsten iteration, it will be found that the geometric progression starts at once with a constant ratio of $p = (v - k)/\{k(v - 1)\}$.

4.8 Designs in supplemented balance

Sometimes it is not possible for all the treatments to be equally replicated. In that case it is often possible to designate one of them 'the supplementing treatment' and allow it to be different from the rest. Sometimes that is positively advantageous, as for example in Exercise 1G, where one treatment holds a position of special importance in the scheme of the experiment. Sometimes, on the other hand, a few plots of an untreated control may be introduced into an experiment on insecticides, only to establish the existence of the pest in the area. For a design to be in 'supplemented balance', the following conditions must obtain:

(1) The supplementing treatment has r_o plots: each of the others has r.
(2) The supplementing treatment concurs λ_o times with each of the others, which concur λ times with one another.

The following is an example:

Block					
I	O	B	C	D
II	O	A	C	D
III	O	A	B	D
IV	O	A	B	C

Here $v = 5$, O being the supplementing treatment, $k = 4$, $r_o = 4$, $r = 3$, $\lambda_o = 3$, $\lambda = 2$.

For all such designs the Kuiper–Corsten iteration can be used, but a simpler method is available, as follows:

(1) Sweep by block means to give deviations.
(2) Sum the deviations by treatments to give adjusted treatment totals
$$Q_o, Q_1, Q_2, \ldots, Q_{v-1}. \quad (4.8.1)$$
(3) Find the two effective replications
$$R = [\lambda_o + (v - 1)\lambda]/k \quad (4.8.2)$$
and
$$R_o = R\lambda_o/\lambda.$$
(4) The treatment sum of squares is
$$Q_o^2/R_o + (Q_1^2 + Q_2^2 + \ldots + Q_{v-1})/R. \quad (4.8.3)$$
(5) The treatment parameters are now estimated by
$$Q_o/R_o, Q_A/R, Q_B/R, \ldots \text{ etc.}, \quad (4.8.4)$$
each value of Q being divided by the appropriate effective replication. For most purposes it is enough to find the adjusted treatment means by adding the grand mean to each of the parameters. That is

not quite correct because no allowance is being made for the unequal replication, but the bias is usually so small as not to matter. To be precise, the quantity

$$\frac{r}{R} - \frac{r_o}{R_o}\frac{Q_o}{n} \qquad (4.8.5)$$

should be added to each value. (Here n stands for the total number of plots in the experiment.) Clearly differences between means have not been affected.

(6) The variance of the difference between two adjusted treatment means is

$$(1/R + 1/R_o)s^2 \qquad (4.8.6)$$

if the supplementing treatment is involved or $2s^2/R$ if it is not, where s^2 is the error mean square.

In the two cases $K = 1/R + 1/R_o$ and $1/R + 1/R$ respectively. This means that R and R_o have taken the places of r and r_o. In the example $R = 11/4$, $R_o = 33/8$.

4.9 Simple lattices

If there are a lot of new varieties to be tested it may not be possible to manage more than two replicates. If the number of variates is a perfect square, $v = p^2$, it is possible to use a 'simple lattice'. With such a low degree of replication it would be unreasonable to expect a high degree of precision. Nevertheless, using a simple lattice is a great improvement on having only one plot of each variety. In any case it may give the highest degree of replication possible.

The treatments are written in a square in any convenient order, e.g. for $v = 16$,

A	B	C	D
E	F	G	H
I	J	K	L
M	N	O	P

(4.9.1)

Then eight (or $2p$) blocks are needed. The first p are made up of the rows of the square, i.e.,

Block					
	I	A	B	C	D
	II	E	F	G	H
	III	I	J	K	L
	IV	M	N	O	P

A further p are needed for the columns, i.e.

Block					
	V	A	E	I	M
	VI	B	F	J	N
	VII	C	G	K	O
	VIII	D	H	L	P

It will be seen that each treatment concurs once with $2(p-1)$ others and not at all with the other $(p-1)^2$.

Again the Kuiper–Corsten iteration is available, but it is usually easier to use the following:

(1) Sweep by block means to find deviations.

(2) Sum the deviations over treatments to give adjusted treatment totals, Q_A, Q_B, ..., Q_P and assign them to a table based on the original format at (4.9.1)

				Row totals
Q_A	Q_B	Q_C	Q_D	R_1
Q_E	Q_F	Q_G	Q_H	R_2
Q_I	Q_J	Q_K	Q_L	R_3
Q_M	Q_N	Q_O	Q_P	R_4
C_1	C_2	C_3	C_4	0 = the grand total
Column totals				

From this point it will be convenient to designate each treatment by two suffixes, the first to indicate the row in the format in which it occurs and the second for the column. Thus, Q_G will become Q_{23} and so on.

(3) The values of Q are then summed over rows and columns to give row totals of $R_1, R_2, \ldots, R_p$ and column totals of $C_1, C_2, \ldots, C_p$ as above.

(4) The treatment sum of squares equals

$$\tfrac{1}{2}[\Sigma\Sigma Q_{ij}^2 + (\Sigma R_i^2 + \Sigma C_j^2)/p] \qquad (4.9.2)$$

(5) The parameter for Treatment ij is estimated by

$$\tfrac{1}{2}[Q_{ij} + (R_i + C_j)/p] \qquad (4.9.3)$$

The adjusted treatment means are found by adding the grand mean.

(6) The variance of the difference between two adjusted treatment means is

$$\left[\frac{p+1}{p}\right]s^2 \qquad (4.9.4)$$

if the treatments concur and

$$\left[\frac{p+2}{p}\right]s^2$$

if they do not. As ever, s^2 is the error mean square.

It is usually desirable to include some standard varieties for comparison with the new ones. Further, in order to obtain good comparisons with the standards, each of them can be entered into the lattice more than once. That helps to overcome the difficulty that the total number of treatments, including duplicates, must be a perfect square. Also, there should be no difficulty about obtaining more seed for the standard whereas it is likely to be in short supply for the new ones. Where the same variety is duplicated it is sensible to allot it to two treatments that otherwise would not concur. For example, given the format used at (4.9.1), if there were 14 new strains it would be sensible to allot the standard to A and G or some other non-concurring pair. If that were done, the simpler method of analysis would not be available, but it is always possible to use the Kuiper–Corsten iteration.

There are other designs for comparing numbers of varieties and some of them do not require a restriction to perfect squares. There are, for example, those of Patterson and Williams (1976) and those of Patterson and Hunter (1983). In the absence of special computer programs, it will be recalled that the Kuiper–Corsten iteration is of general application.

4.10 Triple lattices

Occasionally it is possible to find enough material for a third replicate. If that is so, a 'triple lattice' may well provide the best design.

As with a simple lattice, p^2 treatments are assigned to a square format like the one at (4.9.1). The first p blocks are derived from the rows of the format and another p blocks from the columns. Then a Latin square is applied and another p blocks derived from its letters. Thus, if the square were

α	β	γ	δ
δ	γ	α	β
γ	δ	β	α
β	α	δ	γ ,

the Greek letters give four additional blocks, namely

IX	A	G	L	N	(α)
X	B	H	K	M	(β)
XI	C	F	I	P	(γ)
XII	D	E	J	O	(δ)

The method of analysis is very like that for a simple lattice. The data are first swept by block means to give deviations, which are summed according to treatments to give Q_A, Q_B, ... etc. Now comes the first change. With a simple lattice it is enough to find row totals, R_1, R_2, etc. and column totals, C_1, C_2, etc. Here it is necessary to add L_1, L_2, etc., being the Q-totals for the letters of the Latin square. From this point the general method is the same, though the algebraic expressions need modification.

The treatment sum of squares equals

$$\frac{1}{3}\left[\sum_{i}\sum_{j} Q_{ij}^2 + (\sum_{i} R_i^2 + \sum_{j} C_j^2 + \sum_{h} L_h^2)/(2p)\right]. \qquad (4.10.1)$$

The parameter for treatment ij is

$$\frac{1}{3}\left[Q_{ij} + (R_i + C_j + L_h)/(2p)\right]. \qquad (4.10.2)$$

and adjusted treatment means are found in the usual way by adding the grand mean to all the parameters.

The variance of the difference of two adjusted treatment means is

$$\left(\frac{2(p+1)}{3p}\right)s^2 \qquad (4.10.3)$$

if the treatments concur and

$$\left(\frac{2(p+3)}{3p}\right)s^2 \qquad (4.10.4)$$

if they do not. Again, s^2 is the error mean square in the analysis of variance.

4.11 Non-orthogonal row-and-column designs

With row-and-column designs, which have two blocking systems, there can be as much difficulty in fitting everything into the given area as with block designs, perhaps more. For example, with seven treatments, it might

be possible to arrange for seven columns so that the treatments can be orthogonal to rows, but it might not be feasible to divide the area the other way to obtain seven rows. In that case, one possibility is to take the design in balanced incomplete blocks that begins Section 4.7 and rearrange it thus:

F	B	D	A	G	E	C
A	E	G	B	C	D	F
G	F	B	C	E	A	D

It will be seen that the treatments are disposed orthogonally with regard to the rows (the first blocking system) and in total balance to the columns (the second). Such a design is said to be of Type O:OT. The letter before the colon shows how the columns are disposed relative to the rows. (It is a requirement of a row-and-column design that they shall be orthogonal.) The two letters after the colon show respectively how the treatments are disposed relative to rows (here orthogonal) and to columns (here in total balance). (More specifically where, as here, the orthogonal component contains only one plot of each treatment and the non-orthogonal is in balanced incomplete blocks, the design is said to be a 'Youden square').

It will be understood here and throughout that a row-and-column design, once written down, must be randomized before it is used. First a specimen design, like that at (4.11.1), must be written down with the treatments allocated both to rows and columns in the required manner. Then the rows must be permuted at random and lastly the columns.

The analysis of data from such a design presents little difficulty. First, as for any row-and-column design, the data are swept first by row means and then by column means to give deviations. Then the deviations are summed by treatments to give the Q-values. From this point there is no need to bother further with the blocking system, whether rows or columns, to which the treatments are orthogonal. It has been dealt with completely by the sweeping. It is necessary to consider only the non-orthogonal component and that is analysed in whatever way is usual, whether by the Kuiper–Corsten iteration or by one of the special methods set out in Sections 4.7 and 4.8. An example will illustrate how all that is done. We will take a design of Type O:TO with field plan and data like this:

										(4.11.1)
A	16	D	15	B	20	A	15	C	14	
B	19	A	15	D	16	C	17	D	18	
C	14	B	19	A	19	D	16	B	22	
D	16	C	16	C	15	B	17	A	16	

For the non-orthogonal component $v = 4$, $k = 5$, $r = 5$, $n = 20$, $\lambda = 6$, $R = 4.8$.

The first step is to find the deviations. Sweeping by rows gives:

A	0	D	−1	B	+4	A	−1	C	−2
B	+2	A	−2	D	−1	C	0	D	+1
C	−4	B	+1	A	+1	D	−2	B	+4
D	0	C	0	C	−1	B	+1	A	0

A further sweep by columns gives the deviations, namely:

										(4.11.2)
A	+0.50	D	−0.50	B	+3.25	A	−0.50	C	−2.75	
B	+2.50	A	−1.50	D	−1.75	C	+0.50	D	+0.25	
C	−3.50	B	+1.50	A	+0.25	D	−1.50	B	+3.25	
D	+0.50	C	+0.50	C	−1.75	B	+1.50	A	−0.75	

The sum of their squares gives the stratum total sum of squares, which is 64.50 with 12 degrees of freedom.

The columns are orthogonal to both rows and treatments. Their effect has therefore been completely allowed for in the sweeping; it remains to adjust the treatment effects by the rows, to which treatments are non-orthogonal.

Treatment totals of deviations, Q, can be found from (4.11.2), namely,

A	B	C	D	(4.11.3)
−2.00	+12.00	−7.00	−3.00	

The treatment sum of squares is

$$[(-2.00)^2 + (+12.00)^2 + (-7.00)^2 + (-3.00)^2]/R$$

where R was found below (4.11.1). That value is appropriate because the only non-orthogonality is that of treatments relative to rows. The columns are orthogonal both to rows and treatments, so their effect was sufficiently allowed for when sweeping by their means. Once that had been done, it was as if the design were one of treatments in the blocking system given by rows. Hence R is the effective replication both for that design and for the full design in rows and columns.

If the non-orthogonality in rows had taken some less usual form it might have been necessary to have taken treatment means at (4.11.3) to obtain the starting vector, v_1, for the Kuiper–Corsten iteration. The incidence matrix required would have been that of treatments and rows, namely,

2	1	1	1	$v_1 = -0.40$
1	1	2	1	$+2.40$
1	1	1	2	-1.40
1	2	1	1	-0.60

However, none of that is necessary here, where the non-orthogonality has a defined form. We note that R has already been worked out from (4.7.2) and equals 4.8, so the treatment sum of squares from (4.11.3) is 42.92. It has three degrees of freedom, so the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	3	42.92	14.307	5.97
'Error'	9	21.58	2.398	
Stratum total	12	64.50		

As to treatment means, no adjustment is needed on account of the columns, which are orthogonal with respect to the treatments, but it is required for the rows, which are non-orthogonal. In this instance (4.7.4) applies. The grand mean is 16.75 so the adjusted means are:

A	B	C	D
16.33	19.25	15.29	16.12

The variance of the difference of two adjusted means is shown by (4.7.5) to be $0.9992 = (0.9996)^2$.

Other non-orthogonal components may be used. The following design arose when the statisticians were asked for a plan to add treatments, O, A, B, C, and D to an experiment in which there were four blocks of ten rootstocks, each plot containing a single cherry tree. The treatments were (A, B, C, D), the injection of four different viruses, or (O), not injecting anything. The design was like this:

	Rootstocks										
Blocks	a	b	c	d	e	f	g	h	i	j	
I	B	C	B	A	C	D	A	O	O	D	
II	O	D	C	B	O	C	B	A	D	A	(4.11.4)
III	A	A	O	D	B	B	O	D	C	C	
IV	C	O	D	C	A	A	D	B	B	O	

The treatments are orthogonal to rows and in total balance relative to columns, i.e. the design is of Type O:OT.

In general, it is better to avoid designs in which the treatments are non-orthogonal to both rows and columns. For one thing, they are usually very complex; for another, any non-orthogonality loses information. It is bad enough to have the information loss from one blocking system without having it from two.

The last point needs a little explanation. A completely randomized design, i.e. one without blocks, is said to be 'of full efficiency'. If two treatments have replications of r_1 and r_2 plots, the variance of the difference between their means (see Section 3.4) is

$$Ks^2, \quad \text{where } K = 1/r_1 + 1/r_2. \tag{4.11.5}$$

We will first consider block designs. When they are orthogonal, there is no 'information loss', and the expression at (4.11.5) still applies, but not if there is non-orthogonality. With a non-orthogonal row-and-column design the position is worse because there could then be two information losses, one due to the rows and the other to the columns. For that reason, doubly non-orthogonal designs are better avoided as being inefficient, though there are occasions when practical considerations leave no alternative. Also, the better local control and the consequent reduction in s^2 could outweigh the loss of efficiency. One simple form will be described in the next section.

4.12 Supplemented balance in both rows and columns

When the designer of an experiment is in real difficulties with the land and is not able to contrive rows and columns that will permit orthogonality, he is in a most awkward situation. There is, however, one class of doubly non-orthogonal row-and-column design that is quite feasible and not too difficult to evolve. It is the class in which the treatments are in supplemented balance with regard to both rows and columns, i.e. those Type O:SS.

Examples are easily given. First, there is the Latin square in which one row and one column are omitted, the two intersecting in a plot with the supplementing treatment, e.g.

A	B	C*	D	O
C	O	B*	A	D
B*	D*	O*	C*	A*
O	A	D*	B	C
D	C	A*	O	B

giving

A	B	D	O
C	O	A	D
O	A	B	C
D	C	O	B

(4.12.1)

Plots marked with an asterisk are to be omitted

Then there is the Latin square with a row and column added, the additional plot at their intersection being assigned the supplementing treatment, e.g.

A	O	B	B		A	O	B	B	
B	A	O	A	giving	B	A	O	A	(4.12.2)
O	B	A	O		O	B	A	O	
O	A	B	O		O	A	B	O	

Also, there is the Latin square with a column added and a row omitted, the supplementing treatment being at the intersection, e.g.

B	D	O	A	C	B		B	D	O	A	C	B
O	C	D	B	A	C	giving	O	C	D	B	A	C	(4.12.3)
C	A	B	O	D	A		C	A	B	O	D	A
D*	O*	A*	C*	B*	O*		A	B	C	D	O	D
A	B	C	D	O	D							

Again plots marked with an asterisk are to be omitted.

There are many other possibilities. For example, if at (4.11.4) there had been only nine rootstocks, the omission of the last column would have given a design of Type O:SS. Such designs need to be validated in the usual way by a random permutation of rows followed by a random permutation of columns. Also, they are best suited to problems in which one treatment does have a different status from the rest. If that is not so, a difficult choice has to be made. Nevertheless, it will usually be better to proceed with a design that is statistically less than perfect if the alternative is to use a system of rows and columns that makes difficulties in the field. (Another occasion for using such designs will appear in Section 10.8.)

The method of analysis is not difficult. First, as for any other row-and-column design, it is necessary to sweep by rows and then by columns to find the deviations. The treatment totals of deviations will be written **Q**, as before.

It is now necessary to find R and R_o for the row-and-column design. Both follow easily from the 'components', i.e. the designs given by rows and columns separately. To take R first and using the suffixes r and c to indicate, respectively, the designs in which only the rows are considered or only the columns, r being the actual replication,

$$R = R_r + R_c - r. \tag{4.12.4}$$

Thus, for the design at (4.12.3), ignoring columns and using only the

blocking system given by rows, $k_r = 6$, $\lambda_r = 6$, $\lambda_{or} = 5$, $v = 5$, $R_r = 29/6$. Turning now to the design in which only columns are considered, $k_c = 4$, $\lambda_c = 4$, $\lambda_{oc} = 3$, $v = 5$, $R_c = 19/4$. Hence

$$R = 29/6 + 19/4 - 5 = 55/12 = 4.58.$$

The expression for R_o is more complicated:

$$R_0 = \frac{b\lambda_{or} + c\lambda_{oc} - rr_o}{b\lambda_r + c\lambda_c - r^2} R \tag{4.12.5}$$

where b $(= k_c)$ is the number of rows and c $(= k_r)$ is the number of columns. Hence, in this instance,

$$R_o = \frac{20 + 18 - 20}{24 + 24 - 25} \times \frac{55}{12} = 3.59.$$

From this point, with Q, R and R_o known, everything proceeds as in steps (4), (5) and (6) of Section 4.8. Also, the adjustment at (4.8.5) is available if needed.

4.13 Residuals with non-orthogonal designs

When data are analysed using the Kuiper–Corsten iteration, as described in Section 4.2, the residuals are calculated explicitly as at (4.2.7) and there are no difficulties. Nor are there any problems when shorter methods are used for special designs, because the residuals can be estimated from the treatment parameters. In several cases expressions have been given to enable those quantities to be found, i.e. at (4.7.4), (4.8.4), (4.9.3) and (4.10.2). The method is as follows. The deviations are already known. From each one the appropriate treatment parameter is subtracted. Finally, the resulting figures are swept by block means to give the residuals. Row-and-column designs will be considered later.

At this point it may be helpful to return to the data at (4.2.1), the deviations being at (4.2.2). The values for Q are

A, − 11.00; B, + 0.25; C, + 4.75; D, + 3.00; E, + 3.00.

The values of R and R_o are respectively 11/4 and 33/8. Hence the parameters are

A, − 2.67; B, + 0.09; C, + 1.73; D, + .09; E, + 1.09.

Sweeping the deviations by the parameters gives:

Block	I	A,	+1.42	C,	−0.98	D,	−2.34	E, +0.66
	II	B,	−1.09	A,	+0.67	D,	−1.09	C, +1.27
	III	E,	−1.09	C,	−0.73	B,	+0.91	A, +0.67
	IV	D,	+3.16	B,	+0.16	E,	+0.16	A, −3.08

A further sweep by block means gives the residuals already found at (4.2.7). (Actually the figures just found are rather better, the block and treatment totals in the main lying closer to zero.)

The adjustment for non-equality of replication at (4.8.5) equals −0.08. It will be seen that the adjusted treatment mean for A is

$$\text{Grand mean} + \text{parameter for A} + \text{adjustment}$$

$$= 14.75 - 2.67 - 0.08 = 12.0,$$

which is the value at (4.2.9), and so for the other treatments also.

With a simple lattice there is usually little point in finding residuals because there are only two replications. Since residuals necessarily sum to zero over each treatment, each large residual must be balanced by another—of the same magnitude but of opposite sign—on the second plot of the same treatment. There is usually no way of knowing whether the first plot had given an abnormally high value or the second an abnormally low one. For some other purposes, however, e.g. those set out in Chapter 12, residuals need to be known.

With the row-and-column designs described in Sections 4.11 and 4.12 much the same method applies. The singly non-orthogonal designs in Section 4.11 need one final sweep, by either rows or columns, according to which one carries the non-orthogonality. The doubly non-orthogonal designs in Section 4.12 must be swept twice before finishing, once by rows and once by columns, because the treatments are non-orthogonal to both blocking systems.

Exercise 4A

The following data have been made up to illustrate what happens when two plots in a randomized block design are interchanged as the result of an error.

Block	I	A	14.7	A	13.9	C	12.5	D 14.2
	II	B	16.4	B	17.1	C	13.3	D 15.0
	III	A	15.4	B	17.6	C	15.0	D 16.7
	IV	A	16.3	B	18.2	C	16.6	D 17.4
	V	A	16.8	B	19.1	C	17.8	D 18.9

The letters A–D indicate treatments. Note that plots have not been given in field order but rearranged so as to show what has happened.

Analyse the data. Find the standard error of the contrast between the adjusted means of (i) A and B, (ii) A and C, (iii) C and D, both for the achieved design and for the one intended.

[Data from S. C. Pearce, 'Randomized blocks with interchanged and substituted plots', *J. Royal Statist. Soc.* **B10** (1948), pp. 252–6.]

Exercise 4B

The following data have been made up to illustrate the case in which one treatment has been substituted for another in certain blocks of a randomized block design. (In this instance Treatment B has been substituted for A in Blocks I and II).

Block	I	B	1.74	B	1.69	C	1.81	D 1.86
	II	B	1.85	B	1.97	C	1.96	D 1.94
	III	A	2.01	B	1.90	C	2.00	D 1.91
	IV	A	2.14	B	1.99	C	2.11	D 2.06
	V	A	1.96	B	1.96	C	1.95	D 1.90
	VI	A	2.08	B	1.94	C	2.02	D 2.06

The letters A–D indicate treatments. Again plots are not in field order. Analyse the data. Find the standard error of the contrast between the adjusted means of (i) A and B, (ii) A and C, (iii) B and C, (iv) C and D, both for the achieved design and for one in randomized blocks.

[Data from S. C. Pearce, 'Randomized blocks with interchanged and substituted plots', *J. Royal Statist. Soc.*, **B10** (1948), pp. 252–6.]

Exercise 4C

The following data have been made up to illustrate a design in Balanced Incomplete Blocks. In this instance, $b = 10$, $v = 5$, $k = 3$, $r = 6$, $\lambda = 3$. The letters A–E indicate treatments.

Block								
	I	D	34	E	30	A	26
	II	D	34	B	33	E	26
	III	B	30	A	27	D	30
	IV	E	31	C	27	A	26
	V	C	22	D	30	E	26
	VI	B	33	C	22	A	23
	VII	A	27	C	23	D	28
	VIII	C	21	E	25	B	26
	IX	D	29	B	29	C	23
	X	E	27	A	25	B	26

Analyse the data in such a way as to obtain residuals. Having found them, can you suggest a better way in which the experiment could have been designed?

Exercise 4D

An investigator had ten new varieties of maize (A–J) and three standard varieties (X, Y and Z). He compared them using balanced incomplete blocks. The data, which represent crop weight in pounds per plot, were as follows:

Block												
	I	C	25.3	F	19.9	I	29.0	X	24.6
	II	C	23.0	D	19.8	H	33.3	Y	22.7
	III	J	16.2	X	19.3	Y	31.7	Z	26.6
	IV	B	27.3	E	27.0	H	35.6	X	17.4
	V	G	23.4	H	30.5	I	30.8	J	32.4
	VI	D	30.6	E	32.4	F	27.2	J	32.8
	VII	A	34.7	E	31.1	I	25.7	Y	30.5
	VIII	C	34.4	E	32.4	G	33.3	Z	36.9
	IX	A	38.2	B	32.9	C	37.3	J	31.3
	X	B	28.7	D	30.7	I	26.9	Z	35.3
	XI	A	36.6	D	31.1	G	31.1	X	28.4
	XII	A	31.8	F	33.7	H	27.8	Z	41.1
	XIII	B	30.3	F	31.5	G	39.3	Y	26.7

Analyse the data and note any of the new varieties that appears to be significantly better than each of the standards.

[Data from W. G. Cochran and G. M. Cox, *Experimental Designs*, 2nd edn (1957), p. 448. It is not stated in the original presentation which of the thirteen varieties were the standards, so the last three have been assigned that status, as is implied by the text. For purposes of an exercise the doubt does not matter, though it would be very important if anything depended upon the decision.]

Exercise 4E

A study was made of the control of the *Venturia* fungus on apples, using the design presented at (4.8.1). On a scale that measured the size of the lesions, the data were:

Block												
	I	A	2.5	B	1.5	C	1.4	O	2.5
	II	B	4.7	C	4.3	D	10.4	O	6.6
	III	A	3.3	C	1.2	D	7.3	O	4.0
	IV	A	6.4	B	4.9	D	8.8	O	4.8

Is there evidence that any of the spray treatments, A, B, C and D, reduced the incidence of the fungus as compared with doing nothing, i.e. treatment O?

[Data from S. C. Pearce, *Biological Statistics: an Introduction*, (1965), p. 94.

Exercise 4F

An experiment was conducted on apples to see the effect of 'thinning', i.e. of removing fruitlets at an early stage of development so that the ones left would be able to grow larger. There were 60 trees. In the absence of any obvious environmental differences they were divided into five blocks each of twelve trees for purposes of administration. There were four treatments:

A Control—no thinning.
B Hand thinning of 'king fruit', i.e. of those from the first bud of each blossom cluster.
C Hand thinning of fruit on laterals.
D Thinning by a chemical spray, which would of course not discriminate between different kinds of fruitlets.

In four of the blocks (not IV) three trees were allocated at random to each treatment, but in Block IV four trees were allocated to each of B, C and D. In that way less emphasis was given to the control, which was considered to be of less importance. In Block III some trees died, so examination of the full data will be left till Chapter 12.

The following data give the crop in pounds (1 pound = 454 grams) per tree. Note that crop, though important, is subsidiary to the main purpose

of the study, which concerned size and colour of fruit. With respect to crop, A is as important as B, C or D, but the design does not allow for that.

Block I	D 122	D 125	C 112
	A 76	B 189	B 119
	D 154	C 125	B 117
	C 121	A 170	A 148
Block II	D 105	B 142	D 143
	C 129	A 180	A 139
	C 130	C 134	D 181
	A 263	B 171	B 184
Block III	Omitted		
Block IV	D 143	D 187	D 181
	B 235	B 202	C 121
	C 148	C 124	D 143
	B 200	B 219	C 190
Block V	D 187	A 259	C 149
	B 217	B 215	A 83
	B 169	D 181	D 194
	C 178	A 190	C 162

Write down concurrences for all pairs of treatments and decide what class of design was used. Analyse the data to find adjusted treatment means and standard errors of contrasts between pairs of treatments.

[Data presented by permission of the Director of the East Malling Research Station, where the experiment was carried out.]

Exercise 4G

The following figures come from an experiment designed in a simple lattice. They represent the number of tillers per square metre of rice.

First replicate

Block	I	A	147	B	152	C	167	D 150
	II	E	127	F	155	G	162	H 172
	III	I	147	J	100	K	192	L 177
	IV	M	155	N	195	O	192	P 205

Second replicate

Block	V	A	140	I	182	E	165	M 152
	VI	J	97	B	155	N	192	F 142
	VII	G	155	O	182	C	192	K 192
	VIII	P	182	H	207	L	232	D 162

In the first replicate blocks correspond to rows of the format

A	B	C	D
E	F	G	H
I	J	K	L
M	N	O	P

In the second they correspond to the columns. Analyse the data.

[*Note*: In the next exercise a third replicate from the same experiment is added.]

[Data from K. A. Gomez and A. A. Gomez, *Statistical Procedures for Agricultural Research with Emphasis on Rice* (1976), p. 36.]

Exercise 4H

There was a third replicate to the last example, which makes the design a triple lattice. The additional data were:

Third replicate

Block	IX	A	155	F	162	K	177	P 152
	X	E	182	B	130	O	177	L 165
	XI	I	137	N	185	C	152	H 152
	XII	M	185	J	122	G	182	D 192

In the first replicate blocks correspond to rows of the format

A	B	C	D
E	F	G	H
I	J	K	L
M	N	O	P

In the second they correspond to the columns and in the third to the letters of the Latin square

α	β	γ	δ
β	α	δ	γ
γ	δ	α	β
δ	γ	β	α

Analyse all the data available using the short method.

[Data from K. A. Gomez and A. A. Gomez, *Statistical Procedures for Agricultural Research with Emphasis on Rice* (1976), p. 36. There were in all five replicates, the full design being in total balance.]

Exercise 41

What are the values of R and R_o for the designs at (4.12.1) and (4.12.2)?

Chapter 5

Contrasts as a means of specifying purpose

5.1 The need to specify objectives

No experiment will be conducted to best advantage unless everyone is agreed about what needs to be done. Some method is required for writing down the sort of information that is to be obtained. More than that, it may be necessary to set out what is being assumed.

The statistical approach is to specify 'contrasts'. For example, there could be four treatments, A, B, C and D. There must have been good reasons for choosing them, but what were those reasons? We can suggest a few of the more likely answers to that question, but any experiment may provide unexpected and unusual cases. We will just indicate some of the more common ones.

(1) It may be that A is the standard method, while B, C and D are new ideas advanced as possible improvements. In that case the immediate need is not to compare B, C and D among themselves but to compare each of them with A. That defines three contrasts, which will be written:

$$\begin{array}{cccc} A & B & C & D \\ (-1 & +1 & 0 & 0) \\ (-1 & 0 & +1 & 0) \\ (-1 & 0 & 0 & +1) \end{array} \qquad (5.1.1)$$

The first contrast represents the mean of B less that of A; the means of C and D do not come into it. The others are similar.

The expressions at (5.1.1) are really a sort of shorthand. The experimenter has values, Y_A, Y_B, Y_C and Y_D, one for each treatment. In the case of an orthogonal design or one that is completely randomized they are the treatment means; in the case of a non-orthogonal design the method has been given at (4.2.10), with simpler expressions for special designs at (4.7.4) and (4.8.4). The contrasts at (5.1.1) indicate that we want to know about

$$(-Y_A + Y_B), \quad (-Y_A + Y_C), \quad (-Y_A + Y_D).$$

(2) It could be that A, B, C and D represent increasing applications of something, e.g. increasing concentrations of a spray substance or increasing dressings of a fertilizer. In that case the need is to establish the shape of the response curve, whether it is straight or not, and perhaps whether it has a maximum within the range under study. Suitable contrasts will be sought in Sections 5.7 and 5.8.

(3) It sometimes happens that the effect of a treatment depends upon the presence or absence of another. For example, Variety A may yield better than Variety B on unfertilized soil, but B may be better where fertilizer is supplied. Similarly, a spray may control fungal infection in ordinary conditions but not where sprinkler irrigation is used. Conditional effects like that are called 'interactions'.

When an interaction is suspected it is better to study the two factors in conjunction; that is to say, each needs to be investigated along with the other. Thus in the example of the two varieties with and without added fertilizer, the four treatments are needed, namely:

AO	Variety A	No fertilization
AF	Variety A	Fertilized
BO	Variety B	No fertilization
BF	Variety B	Fertilized.

There are three degrees of freedom between those four treatments. The first contrast compares plots of Variety A with those of Variety B, i.e.

AO	AF	BO	BF	
(+1	+1	−1	−1).	(5.1.2a)

The second picks up the effect of fertilization, i.e.

(−1	+1	−1	+1)	(5.1.2b)

There are three ways in which four things can be divided into two pairs, and (5.1.2) gives only two of them.

There is a third:

	(−1	+1	+1	−1)

What does it mean?

It is in fact the 'interaction'. Suppose that fertilization had had the same effect on both varieties, i.e. the contrasts

	(0	0	+1	−1)	
and	(+1	−1	0	0)	(5.1.3)

gave the same result. Then their difference would be zero, i.e. the contrast

(−1	+1	+1	−1)	(5.1.4)

would be zero—just as the first two would be zero if there were respectively no effect of varieties or of fertilization. If it is not zero, then the fertilizer is having different effects on the two varieties and the contrast at (5.1.2b) represents the sum of those effects. More meaning attaches to the 'particular effects' at (5.1.3).

In general, the contrast for an interaction is found by multiplying out those for the two 'main effects' from which it is derived. Thus, here

Treatment	*Variety (V)*	*Fertilization (F)*	*Interaction (V × F)*
AO	+1	−1	(+1) × (−1) = −1
AF	+1	+1	(+1) × (+1) = +1
BO	−1	−1	(−1) × (−1) = +1
BF	−1	+1	(−1) × (+1) = −1.

It should be noted that there is a logical order for examining the contrasts. The first thing is to look at the interaction at (5.1.4). If it is small, it is reasonable to conclude that the effect of fertilization is much the same for the two varieties or, to put the same thing in another way, the difference between the two varieties is much the same at both levels of fertilization. It is therefore reasonable to look at the 'main effects' at (5.1.2), but if there is an interaction the next step is to look at the 'particular effects'. Those for fertilization are given at (5.1.3); those for variety are

(+1	0	−1	0)	with no fertilization	
(0	+1	0	−1)	with fertilization	(5.1.5)

It should be noted that if X interacts with Y then Y interacts with X. Thus, the contrast at (5.1.4) can be derived either as a difference of those at (5.1.3) or of those at (5.1.5). Factorial design will be considered in more detail in Chapter 6.

5.2 A look at fundamentals

Before proceeding further it may be helpful to go to fundamentals. The following approach is too long to be used in practice, but it does show what will be happening in the sections that follow. They will set out the methods used in practice, but they are really short cuts of the full procedure.

The point is this. Any contrast, however expressed, whether as a function of means or in any other way, is really a contrast of data. For example, let there be three randomized blocks, each of four treatments, A, B, C and D, then the data may be represented thus:

$$\begin{matrix} y_{1A} & y_{1B} & y_{1C} & y_{1D} \\ y_{2A} & y_{2B} & y_{2C} & y_{2D} \\ y_{3A} & y_{3B} & y_{3C} & y_{3D} \end{matrix} \qquad (5.2.1)$$

The first particular effect at (5.1.3), i.e. the difference between the means of treatments A and B, equals

$$(y_{1A} + y_{2A} + y_{3A})/3 - (y_{1B} + y_{2B} + y_{3B})/3.$$

Taking the data in order of rows at (5.2.1), this may be written

$$(+1 \quad -1 \quad 0 \quad 0 \quad +1 \quad -1 \quad 0 \quad 0 \quad +1 \quad -1 \quad 0 \quad 0).$$

Clearly it has a variance of

$$[(+1)^2 + (-1)^2 + 0^2 + \ldots + (-1)^2 + 0^2 + 0^2]\sigma^2/9 = 2\sigma^2/3.$$

Considering now the main effect of varieties at (5.1.2a), it may be written thus in terms of the data:

$$(+1 \quad +1 \quad -1 \quad -1 \quad +1 \quad +1 \quad -1 \quad -1 \quad +1 \quad +1 \quad -1 \quad -1)/3$$

and its variance is $4\sigma^2/3$. Further the covariance of the two is

$$[(+1)(+1) + (-1)(+1) + 0(-1) + \ldots + (-1)(+1) + 0(-1) + 0(-1)]\sigma^2/9$$

which is zero. In that case they are said, using a technical term already encountered in the context of blocks and treatments, to be 'orthogonal' to one another.

The above method is short enough in the simple case just considered, but it would be impossibly prolix in many instances. It would, for example, be a very lengthy process to work a Kuiper–Corsten iteration algebraically on the data. That is why simpler methods are needed, though they can obscure what is really going on.

5.3 Contrasts of interest written in matrix form

We have seen two ways of expressing a contrast, but we will now show a

third in which it is written as a matrix. (In Section 4.1 we explained that a matrix is only an array of numbers written in rows and columns, both rows and columns having meaning.) In the 'matrix of the contrast' there is a row for each treatment and a column also. We will illustrate how it is formed by using the contrast $(+4 \quad -1 \quad -1 \quad -1 \quad -1)$.

The first row of the matrix is found by multiplying the contrast by its own first coefficient, thus;

$$+16 \quad -4 \quad -4 \quad -4 \quad -4.$$

The second is found similarly by multiplying the contrast by its second coefficient, which gives:

$$-4 \quad +1 \quad +1 \quad +1 \quad +1.$$

The other rows are the same as the second, so the full matrix is:

$$\begin{pmatrix} +16 & -4 & -4 & -4 & -4 \\ -4 & +1 & +1 & +1 & +1 \\ -4 & +1 & +1 & +1 & +1 \\ -4 & +1 & +1 & +1 & +1 \\ -4 & +1 & +1 & +1 & +1 \end{pmatrix} \qquad (5.3.1)$$

It is necessarily square and symmetrical. Any contrast can be written as a matrix in this manner.

There are times when we want to express not a single contrast but the relationship between two different ones. This also can be done by way of a matrix and we will illustrate the method by considering the first two contrasts at (5.1.1), namely,

$$(-1 \quad +1 \quad 0 \quad 0) \quad \text{and} \quad (-1 \quad 0 \quad +1 \quad 0).$$

We take the first contrast and multiply it by the first coefficient of the other. This gives

$$+1 \quad -1 \quad 0 \quad 0.$$

That is the first row of the matrix to be generated. Then we multiply the first contrast by the second coefficient of the other. Since that coefficient is zero, we shall get a second row consisting entirely of zero elements. Proceeding in that way, we form the entire matrix like this:

$$\begin{pmatrix} +1 & -1 & 0 & 0 \\ 0 & 0 & 0 & 0 \\ -1 & +1 & 0 & 0 \\ 0 & 0 & 0 & 0 \end{pmatrix} \qquad (5.3.2)$$

It is square but no longer symmetrical. Its usefulness will be explained in Section 5.6.

5.4 Covariance matrices

To complement the matrices that express the contrasts of interest and the relationships between them, we need others to show the attributes of possible designs. The two, purpose and capability, have to be seen in relation one to the other. Designs are not good or bad in themselves; they are good only in so far as they provide precise estimates of the contrasts declared to be of interest.

The attributes of a design can be expressed by giving its 'covariance matrix'. The matter was touched on in Sections 4.1 and 4.3; we shall now look at it more fully. First of all, a covariance matrix, like the matrix of a contrast, has a row and a column for each treatment. For any design that is orthogonal, whether in blocks or in rows and columns (including those that are completely randomized), an element on the diagonal is equal to the variance of an observation, σ^2, divided by the replication, r, of the treatment to which the element refers, i.e., σ^2/r. Off-diagonal elements equal zero. Hence, if we have five treatments in six randomized blocks, the covariance matrix will be

$$\begin{pmatrix} 1/6 & 0 & 0 & 0 & 0 \\ 0 & 1/6 & 0 & 0 & 0 \\ 0 & 0 & 1/6 & 0 & 0 \\ 0 & 0 & 0 & 1/6 & 0 \\ 0 & 0 & 0 & 0 & 1/6 \end{pmatrix} s^2 \qquad (5.4.1)$$

It will be noticed that we have used s^2 instead of σ^2. If the true variance, σ^2, were known, we would use it, but we are writing in the context of the analysis of variance where σ^2 has to be estimated by s^2, the 'error' mean square with a stated number of degrees of freedom. The matrix at (5.4.1) is therefore not the covariance matrix itself but an estimate of it. It is as near the true matrix as we can get, and for most purposes it serves very well.

That is the way by which to find the covariance matrix of an orthogonal design. The more difficult case, when the design is non-orthogonal, has already been considered in Section 4.3, where it was found by using the Kuiper–Corsten iteration. Sometimes the task can be performed more

easily. Thus, the methods used in Sections 4.7 and 4.8 show that covariance matrices for designs in total and supplemented balance can be found using R or R_o, as may be appropriate, instead of r in generating matrices like the one at (5.4.1).

5.5 The variance of a contrast

To find the variance of a contrast if a certain design is used, it is necessary to know both the matrix of the contrast and the covariance matrix of the design. They will be of the same size, their elements corresponding each to each. It is necessary only to multiply corresponding elements and to add the products to obtain the variance, Ks^2, defined at (4.1.1).

We will suppose that someone is interested in the contrast

$$(+4 \quad -1 \quad -1 \quad -1 \quad -1).$$

Its matrix has been presented at (5.3.1). One possibility is to use six randomized blocks. In that case, the covariance matrix is at (5.4.1), so we can begin. We have

1	product of + 16 and $s^2/6$	$= 16s^2/6$
4	products of + 1 and $s^2/6$	$= 4s^2/6$
8	products of − 4 and 0	$= 0$
12	products of + 1 and 0	$= 0$
25	products with a total of	$3.333s^2$

Hence, the variance of the contrast is 3.333 times the 'error' mean square in the analysis of variance. The square root of that quantity will be the standard error of the contrast.

We can take another example by supposing that someone wanted to study the same contrast using the design that gave rise to the covariance matrix at (4.3.1). It was non-orthogonal, but the method is the same. We now have:

1	product of + 16 and $0.1875s^2$	$= 3.000s^2$
4	products of + 1 and $0.2936s^2$	$= 1.174s^2$
8	products of − 4 and $-0.0625s^2$	$= 2.000s^2$
12	products of + 1 and $-0.0701s^2$	$= -0.841s^2$
25	products giving a total of	$5.333s^2$

In this instance K equals 5.333. We know from (4.2.9) that s^2 is 4.424, so we can go further and say that the variance of the contrast is 23.593 and its standard error is 4.86, the square root of 23.593. From (4.2.10) it appears that the contrast itself equals

$$4(12.0) - 14.8 - 16.4 - 15.8 - 15.8 = -14.8,$$

which is 3.05 times its standard error. Contrasts like

$$(+1 \quad -1 \quad 0 \quad 0 \quad 0) \quad \text{and} \quad (0 \quad +1 \quad -1 \quad 0 \quad 0)$$

give the results already obtained at (4.3.2) and (4.3.3).

This is perhaps the place to mention that there is no need to strive for equality of replication as if it were an end in itself. Indeed, Sections 4.8 and 4.12 have already shown that there are occasions when it can well be abandoned. It is not necessary with orthogonal designs either. For example, in the situation that gave rise to the contrasts at (5.1.1) there is a lot to be said for giving the standard enhanced replication. If the intention is solely to compare each new variety with the standard, the ideal is to give the standard a replication a times that of the others, where a is the square root of the number of new treatments. Thus, if A is the standard and B, C, D and E are new treatments, five blocks of AABCDE would be better than six of ABCDE, always assuming that the increased 'error' mean square, s^2, that may result from using larger blocks will not annul the advantage given by a lower value of K. To examine the position further, the design with unequal replication will give the covariance matrix

$$\begin{pmatrix} 0.1 & 0 & 0 & 0 & 0 \\ 0 & 0.2 & 0 & 0 & 0 \\ 0 & 0 & 0.2 & 0 & 0 \\ 0 & 0 & 0 & 0.2 & 0 \\ 0 & 0 & 0 & 0 & 0.2 \end{pmatrix} s^2$$

In that case the variance of a contrast like $(-1 + 1\ 0\ 0\ 0)$ will be $0.300s^2$. If the matrix at (5.4.1) had been used, the variance would have been $0.333s^2$. The gain is not great but, if it can be achieved at no cost, it is worth having.

5.6 Contrasts in the analysis of variance

If the value of a contrast is squared and divided by the value of K, the result gives the sum of squares corresponding to that contrast in the 'error' line of the analysis of variance. It will have one degree of freedom. For

example, to return to Example (3) of Section 5.1, we will suppose that the four treatments were studied in five randomized blocks. The diagonal elements of the covariance matrix will all equal 0.2. Further, the three contrasts at (5.1.2) and (5.1.4) give respectively the following matrices:

$$\begin{pmatrix} +1 & +1 & -1 & -1 \\ +1 & +1 & -1 & -1 \\ -1 & -1 & +1 & +1 \\ -1 & -1 & +1 & +1 \end{pmatrix}, \begin{pmatrix} +1 & -1 & +1 & -1 \\ -1 & +1 & -1 & +1 \\ +1 & -1 & +1 & -1 \\ -1 & +1 & -1 & +1 \end{pmatrix} \text{ and } \begin{pmatrix} +1 & -1 & -1 & +1 \\ -1 & +1 & +1 & -1 \\ -1 & +1 & +1 & -1 \\ +1 & -1 & -1 & +1 \end{pmatrix} \quad (5.6.1)$$

It emerges that for each of the contrasts relative to the given covariance matrix, $K = 0.8$.

We will now suppose further that the treatment totals were:

AO, 50; AF, 60; BO, 44; BF, 48.

That being so, the treatment sum of squares can be found as a difference between the two summation terms, i.e.,

$$(50^2 + 60^2 + 44^2 + 48^2)/5 - 202^2/20 = 27.80.$$

The next step is to estimate the contrasts:

Main effect of varieties	$(+50 + 60 - 44 - 48)/5 = +3.60$	
Main effect of fertilizer	$(-50 + 60 - 44 + 48)/5 = +2.80$	(5.6.2)
Interaction	$(-50 + 60 + 44 - 48)/5 = +1.20.$	

Consequently the three sums of squares are:

Main effect of varieties	$(3.60)^2/0.8 = 16.20$
Main effect of fertilizer	$(2.80)^2/0.8 = 9.80$
Interaction	$(1.20)^2/0.8 = 1.80.$

It will be seen that the three sums of squares, each with one degree of freedom, sum to 27.80, which has three; that is the total found from the summation terms.

However, this property does not always hold. To take a different example, the contrasts at (5.1.1) give components that do not add up correctly. First, to evaluate the contrasts themselves, they are:

A *v.* B	$(-50 + 60)/5 = +2.00$	
A *v.* C	$(-50 + 44)/5 = -1.20$	(5.6.3)
A *v.* D	$(-50 + 48)/5 = -0.40.$	

Since K is now 0.4, the sums of squares are:

A *v.* B	$(2.00)^2/0.4 = 10.00$
A *v.* C	$(1.20)^2/0.4 = 3.60$
A *v.* D	$(0.40)^2/0.4 = 0.40.$

The sum is now 14.00, not 27.80 as before.

There is clearly some important difference between the two sets of contrasts and it lies in the orthogonality already noted in Section 5.2. The word was used of blocks and treatments in Section 1.3 to indicate that their two effects can be estimated independently of one another. It is used in the same sense here. A knowledge of the value of the contrast (+ 1 + 1 − 1 − 1) tells us nothing about the value of (+ 1 − 1 + 1 − 1), but a knowledge of (− 1 + 1 0 0) does affect our expectation of the value of (− 1 0 + 1 0). The contrasts at (5.6.2) have been estimated independently of one another, but that is not true of those at (5.6.3).

Orthogonality is readily tested. To find out if (+ 1 + 1 − 1 − 1) and (− 1 + 1 − 1 + 1) are orthogonal, it is necessary to write down a matrix like that at (5.3.1), except that the rows are governed by one contrast and the columns by another. This has already been done at (5.3.2) though for a different pair of contrasts. The result here is

$$\begin{pmatrix} -1 & +1 & -1 & +1 \\ -1 & +1 & -1 & +1 \\ +1 & -1 & +1 & -1 \\ +1 & -1 & +1 & -1 \end{pmatrix} \qquad (5.6.4)$$

The matrix so found is then multiplied out by the covariance matrix of the design in the same way that the matrix at (5.3.6) was multiplied out by those at (5.2.1) and (4.3.1), the products being added. If the result is zero, the two contrasts are estimated orthogonally by that design. Here the covariance matrix is

$$\begin{pmatrix} 0.2 & 0 & 0 & 0 \\ 0 & 0.2 & 0 & 0 \\ 0 & 0 & 0.2 & 0 \\ 0 & 0 & 0 & 0.2 \end{pmatrix} s^2 \qquad (5.6.5)$$

which gives:

2	products of − 1 and $0.2s^2$	$= -0.4s^2$
2	products of + 1 and $0.2s^2$	$= +0.4s^2$
6	products of − 1 and 0	$= 0.0$
6	products of + 1 and 0	$= 0.0$
16	products with a total of	0.0

The total being zero, the two contrasts are orthogonal, at least for this design. Turning to the other contrasts, i.e., those at (5.6.4), the total is $+ 0.2s^2$, which is not zero, so the two contrasts are not orthogonal. (They would be so only if the mean of treatment A could be determined perfectly and that is, of course, quite impossible. Any discrepancy in its determination will affect both contrasts equally; for that reason they cannot be estimated independently.)

Where the covariance matrix has the simple form of (5.6.5) considerable simplification becomes possible. In general, if all diagonal elements of the covariance matrix have the same value, α, and all off-diagonal elements similarly equal β, then each treatment can be ascribed an effective replication of

$$R = 1/(\alpha - \beta) \qquad (5.6.6)$$

which will apply whatever the contrast. It leads to the conclusion that, for any contrast,

$$K = U/R \qquad (5.6.7)$$

where U is the sum of squares of the coefficients of the contrast. Thus, for the contrasts at (5.1.2) and (5.1.4), $U = 4$ and $R = r = 5$, so $K = 0.8$, the figure already found. Another example is afforded by the balanced incomplete block design at the beginning of Section 4.7. Here the Kuiper–Corsten iteration shows all α to be 18/49 and all β to be − 3/49. From (5.6.6) R should be 7/3, the figure that was found at (4.7.2).

Further, in this simple but common case, i.e., that of constant α and constant β, orthogonality between two contrasts is established by multiplying out the coefficients, each to each, and adding the products. If they sum to zero the two contrasts are orthogonal for that design; if they do not, the contrasts are mutually dependent. Thus, for the case studied at (5.6.4),

$$(+1)(-1) + (+1)(+1) + (-1)(-1) + (-1)(+1) = 0, \qquad (5.6.8)$$

but for that at (5.3.2), where two contrasts from (5.1.1) were considered,

$$(-1)(-1) + (+1)(0) + (0)(+1) + (0)(0) = +1$$

and the contrasts are not orthogonal.

The matrix at (5.6.5) exemplifies also another special case—and a very common one—that of an orthogonal (or completely randomized) design with all treatments equally replicated. In that case $R = r$ and the rule given in the first paragraph of this section can be expressed in an improved form. To take the calculations at (5.6.2) as an example, the method is to work out the contrast in terms of the treatment totals instead of the means and to use the divisor, rU, instead of K. In that way the sum of squares for the main effect of varieties is found as

$$(50 + 60 - 44 - 48)^2/(5 \times 4) = 16.20. \qquad (5.6.9)$$

The advantage lies in the minimization of rounding errors.

The rest of this chapter will be taken up by a study of a particular sort of contrast, namely, that mentioned in Example (2) of Section 5.1. The treatments now have a clear structure, being increasing quantities of some substance applied to the soil or the plants.

5.7 Quantitative levels of factors

It often happens that the levels of a factor depend upon some quantity like the concentration of an insecticide or the number of times it is applied. Qualitative factors like varieties or the form of nitrogenous fertilizer also give useful information, but it is of a different kind. The most important distinction is that quantitative factors have an in-built structure that needs to be respected. We shall examine them now.

Three levels only

We will first consider the case in which a factor, X, is used at three equally spaced levels, which we will call x_0, x_1 and x_2. Their responses will be called respectively y_0, y_1 and y_2. If the responses lie on a straight line when plotted against the levels of X, i.e., if

$$y_j = a + bx_j \quad \text{for } j = 0, 1, 2,$$

then

$$y_0 = a, \quad y_1 = a + b, \quad y_2 = a + 2b. \qquad (5.7.1)$$

Hence b can be estimated from both $(y_1 - y_0)$ and $(y_2 - y_1)$. Consequently, their difference,

$$y_0 - 2y_1 + y_2,$$

should be zero. We therefore examine the contrast $(1 \ -2 \ 1)$. If it is not significant at an acceptable level, there is no need to regard the graph of the responses against the levels as departing from a straight line. This contrast is called the 'quadratic effect'.

However, we still have not isolated the effect of b, though we have obtained two estimates of it, both of the same precision, namely

$$(y_1 - y_0) \quad \text{and} \quad (y_2 - y_1).$$

Their average, the so-called linear effect, provides a better estimate of b than either of the quantities given above. It is $\frac{1}{2}(y_2 - y_0)$. The sum of y_0, y_1 and y_2 is clearly $3(a + b)$, so now that b is known we know a also. The sum of squares between the three levels can thus be broken into two contrasts,

L, the linear effect, measured by $(-1 \quad 0 \quad +1)$
and Q, the quadratic effect, measured by $(+1 \quad -2 \quad +1)$. (5.7.2)

Where (5.6.6) holds, they are orthogonal, each with one degree of freedom. To take matters a little further, if the response is indeed quadratic, i.e., if

$$y_j = a + bx + cx_j^2, \qquad (5.7.3)$$

then the three treatments give means estimated by

$$a, \quad a + b + c \quad \text{and} \quad a + 2b + 4c.$$

Hence the contrast, L, estimates $2b + 4c$, Q estimates $2c$, while the general mean of data is $(3a + 3b + 5c)/3$, so a, b and c can all be estimated if need arises.

More than three levels

If there are more levels, more contrasts arise. If there are four, a 'cubic' term, C, has to be added to those already considered, the contrasts being

L, the linear effect, measured by $(-3 \quad -1 \quad +1 \quad +3)$
Q, the quadratic effect, measured by $(+1 \quad -1 \quad -1 \quad +1)$
and C, the cubic effect, measured by $(-1 \quad +3 \quad -3 \quad +1)$ (5.7.4)

Those contrasts repay study. If the response is really linear, as at (5.7.1), it will be found that Q and C will be zero. (On account of 'error' they will not be exactly so, but they should not depart much from it.) If the response is really quadratic, as at (5.7.2), C will be zero, but not L or Q. Only if there is a cubic term in the response curve, i.e., only if the curve is given by

$$y_j = a + bx_j + cx_j^2 + dx_j^3 \qquad (5.7.5)$$

will C depart from zero. Any pair of L, Q and C will be orthogonal if (5.6.6) holds.

Response curves do not usually follow polynomial expressions of the kind we have been using, so there is little point in going further. The three we have found are useful, because L indicates a general rise or fall as x increases, Q indicates a general curvature, and C picks up an inflexion, e.g. the case when a curve starts off rising at an increasing rate but ceases to rise so quickly as it nears a maximum. If the only reason for having more levels is to obtain a better fit of the response curve, restraint is called for.

Sometimes five equally spaced levels are used. In that case a 'quartic effect' can be found in addition to the others, the contrasts being

L,	the linear effect, measured by	(−2	−1	0	+1	+2)	(5.7.6)
Q,	the quadratic effect, measured by	(+2	−1	−2	−1	+2)	
C,	the cubic effect, measured by	(−1	+2	0	−2	+1)	
Qu,	the quartic effect, measured by	(+1	−4	+6	−4	+1)	

The cubic and quartic effects cannot do much more than indicate a departure from the simple curvature measured by Q. There are, however, occasions when many levels are needed, the purpose usually being to fit some other sort of response curve or to detect the point at which a discontinuity occurs. (For example, if the growth of a plant is measured at regular intervals, there may come a time when the setting of blossoms or some similar phenomenon will inaugurate a new phase in which the growth rate will be different. Such matters, however, lie outside the scope of the present text.)

It should be noted that the contrasts at (5.7.2), (5.7.4) and (5.7.6) have a logical order of testing. If there is an effect of C or Qu, then the response curve is not quadratic in form, so it would be a mistake to discuss a and b as if it were. Likewise, if there is an effect of Q, C or Qu, the response is not a straight line, so it would be wrong to cite b as the slope, except as an average, because there is no constant slope. The matter will be exemplified at the end of this section.

An example

To take an example, Campbell (*Statistics in Biology*, 1967) has presented some fictitious data from an experiment with five randomized blocks of five treatments, namely, maize at relative densities of 20, 25, 30, 35 and 40 plants per unit area. The data represent yield of dry matter per plot in kilograms. The 'error' sum of squares with 16 degrees of freedom was 42.170. Treatment totals were:

20, 87.9; 25, 118.2; 30, 135.9; 35, 141.0; 40, 135.7

so the sum of squares for treatments was 380.802 with four degrees of freedom. It will be convenient to set out the means also, i.e.,

20, 17.582; 25, 23.64; 30, 27.18; 35, 28.20; 40, 27.14.

From (5.7.6) the value of the linear effect is

$$[-2(87.9) - 1(118.2) + 0(135.9) + 1(141.0) + 2(135.7)]/5 = +23.68.$$

From (5.6.6) the K-value is

$$[(-2)^2 + (-1)^2 + (0)^2 + (+1)^2 + (+2)^2]/5 = 2,$$

so the contribution of the linear effect to the treatment sum of squares is $(23.68)^2/2 = 280.371$. Similarly the value of the quadratic effect, Q, is -16.76 with K equal to 2.8, so its contribution is 100.321. For the cubic effect, C, and the quartic, Qu, the contributions are respectively 0.097 and 0.014, a total of 0.111 with two degrees of freedom. They will be taken together as indicating deviations from the quadratic form. The analysis of variance therefore reads:

Source	d.f.	s.s.	m.s.	F
Linear effect, L	1	280.371	280.371	106.36***
Quadratic effect, Q	1	100.321	100.321	38.06***
Deviations, C and Qu	2	0.111	0.056	0.02
'Error'	16	42.170	2.636	
Stratum total	20	422.973		

The very low value of F for the deviations from the quadratic is rather

remarkable. (It is perhaps the result of the data having been simulated on a computer. They are not genuine.) At least it dispels any suspicion that the cubic and quartic effects may be important. From the mean squares for L and Q it is clear that density had a marked effect which was not linear. Indeed the curvature is such that there is a maximum at a density of 30 or so. Since the relationship appears to be quadratic in form, the successive values for the five densities can be written in the form of (5.7.3) as

$$a,\ a+b+c,\ a+2b+4c,\ a+3b+9c,\ a+4b+16c$$

The situation can be summarized thus:

(1) The mean of those quantities is
$(a + 2b + 6c) = (17.58 + 23.64 + 27.18 + 28.20 + 27.14)/5 = 24.75.$
(2) The contrast, L, has the value of $(10b + 40c) = 23.68$.
(3) The contrast, Q, which equals -16.76, may be represented as $14c$.

This makes the response curve, corresponding to (5.7.3), to be

$$y = 17.62 + 7.16x - 1.197x^2.$$

It has a maximum at $x = 2.99$, corresponding to a density of 34.9. At that point the yield is 28.33.

A warning has already been given about assuming a quadratic relationship when there is evidence of a cubic or a quartic effect, but that does not apply here.

5.8 General method of deriving contrasts for quantitative levels

The levels, $x_1, x_2, x_3, \ldots$ that specify the treatments need not be equally spaced, though it is more convenient if they are. If they are not, the linear, quadratic effects, etc. can still be found. The method is more easily illustrated than described.

We shall suppose that the levels are 0, 2, 3, 4 and 6. For the linear effect we need a contrast with coefficients, p_0, p_2, p_3, etc. that themselves exhibit linearity, i.e.,

$$p_j = a + bx_j. \qquad (5.8.1)$$

That is to say,

$$p_0 = a,\quad p_2 = a + 2b,\quad p_3 = a + 3b,$$
$$p_4 = a + 4b,\quad p_6 = a + 6b.$$

Since those values are the coefficients of a contrast they must sum to zero, i.e., $5a + 15b = 0$. Only the ratio of a and b matters, so we will write $a = -3b = +1$. Using (5.8.1), this makes the contrast of the linear effect, L, to be $(-3\ -1\ 0\ +1\ +3)$.

The quadratic effect, Q, similarly needs a contrast with coefficients that exhibit a quadratic relationship. That is to say, a coefficient q_j should have the form,

$$q_j = f + gx_j + hx_j^2. \qquad (5.8.2)$$

The required coefficients are accordingly

$$f,\quad f+2g+4h,\quad f+3g+9h,\quad f+4g+16h,\quad f+6g+36h.$$

It is required that L and Q shall be mutually orthogonal. In the case of some more complicated designs that could be difficult to achieve, but if the conditions are such that (5.6.6) holds, it is required only that

$$-3f-(f+2g+4h)+(f+4g+16h)+3(f+6g+36h)$$

shall equal zero, i.e., $20g + 120h = 0$. That suggests making g equal to -6 and h equal to $+1$.

It is also required that the coefficients shall indicate a contrast, i.e., they must sum to zero. Hence,

$$5f + 15g + 65h = 0.$$

With the values of g and h already adopted, this makes f equal to $+5$. The contrast of the quadratic effect, Q, is thus shown by (5.8.2) to be

$$(+5\ -3\ -4\ -3\ +5).$$

If a cubic component is required, it should have coefficients of the form,

$$c_j = s + tx_j + ux_j^2 + vx_j^3.$$

The values of c_j should sum to zero. They should also do so when multiplied by the respective values of p_j and by those of q_j. In the example the contrast of the cubic effect is $(-1\ +3\ \ 0\ -3\ +1)$.

Exercise 5A

The following data represent a design that has been completely randomized. There are four treatments

the untreated control;
involve treatment by a substance,
X, obtained from two different
manufacturers;
involves treatment by substance Y.

ta are:

A	34	37	40	29	29
B	38	44	36	40	47
C	48	51	48	56	52
D	31	35	36	36	32.

Work out an analysis of variance, the treatment sum of squares being partitioned thus:

(1) Control *v.* treatments
(2) Substance X *v.* substance Y
(3) One manufacturer of X *v.* the 'other'.

Check that the three contrasts you use are orthogonal.

Exercise 5B

In Exercise 1C the ten strains are not all of one variety. Strains A–E come from one variety and F–J from another. Modify the analysis accordingly and interpret the result.

Exercise 5C

The data given below, which come from a Latin square, show yields in pounds of wheat from plots, each with an area of 0.025 acre. Treatment O is lack of fertilization. In Treatment S sulphate of ammonia was applied as a single dressing in March, whereas in M the same amount was applied in regular monthly dressings over a six-month period. In Treatment C an equivalent amount of nitrogen was applied as cyanamide in a single dressing in October, whereas in D half was applied as cyanamide and half as dicyanadiomide, also in one dressing in October.

D 72.2	M 55.4	O 36.6	C 67.9	S 73.0
O 36.4	C 46.9	M 46.8	S 54.9	D 68.5
M 71.5	S 55.6	D 71.6	O 67.5	C 78.4
S 68.9	O 53.2	C 69.8	D 79.6	M 77.2
C 82.0	D 81.0	S 76.0	M 87.9	O 70.9

Suggest a set of contrasts that appears to you to do justice to the treatment structure and analyse the data accordingly.

1 acre = 4047 m² 1 pound = 454 grams.

[Data from A. A. Rayner, *Biometry for Agriculture Students* (1969), p. 273.]

Exercise 5D

An experiment was carried out on the fertilization of potatoes. Treatment A represents lack of fertilization, while B–F represent different fertilization schedules. The experiment was laid out in four randomized blocks, the plots each having an area of 1/85 acre. The following table shows both the field plan and the yields in pounds per plot.

	F 331	B 286	E 312	D 292	F 323	A 185		
I	C 311	D 280	A 177	C 294	B 278	E 322		II
	F 313	C 266	A 182	C 291	B 253	E 328		
III	E 319	D 284	B 258	A 193	D 233	F 319		IV

The treatments were as follows:

A No fertilization
B Nitrogen alone
C Nitrogen + phosphorus
D Nitrogen + lime
E Nitrogen + phosphorus + potassium
F Special mixture of the three elements (nitrogen, phosphorus and potassium)

Consider what treatment contrasts are of greatest interest and find the sum of squares for each. Complete the analysis of variance and present results in kg/ha.

1 acre = 4047 m² 1 pound = 454 grams

[Data from A. A. Rayner, *Biometry for Agriculture Students* (1969), p. 247.]

Exercise 5E

The following figures represent yields of sugar beet from an experiment intended to compare five substances containing nitrogen.

$A = (NH_4)_2SO_4$; $B = NH_4NO_3$; $C = CO(NH_4)_2$; $D = Ca(NO_3)_2$; $E = NaNO_3$

All were applied so as to give 100 units of nitrogen per acre. In the sixth treatment (O) no nitrogen was applied. The design was a Latin square:

O	D	A	B	E	C
28.2	29.1	32.1	33.1	31.1	32.4
E	B	C	O	D	A
31.0	29.5	29.4	24.8	33.0	30.6
D	E	O	C	A	B
30.6	28.8	21.7	30.8	31.9	30.1
C	A	B	D	O	E
33.1	30.4	28.8	31.4	26.7	31.9
B	O	E	A	C	D
29.9	25.8	30.3	30.3	33.5	32.3
A	C	D	E	B	O
30.8	29.7	27.4	29.1	30.7	21.4

Data are in tons per acre (1 acre = 4047 m^2, 1 ton = 1.016 tonnes)

Consider what contrasts between the treatments are of greatest interest to yourself and find the sum of squares for each. Complete the analysis of variance.

[Data from T. M. Little and F. J. Hills, *Agricultural Experimentation: Design and Analysis* (1978), p. 79, John Wiley & Sons, New York.]

Exercise 5F

In Exercise 1E write down the contrasts of interest. Confirm that for each contrast the *t*-test gives the same result as the *F*-test. (*Note*: in a variety trial there is no need of a test to show that differences exist, but it would be unwise for a research institute to recommend a new variety as giving a higher crop, if it might well be no better, or even worse, than the standard.)

Exercise 5G

An experiment was carried out to find the best rate of seeding for a given variety of rice, using six rates set out in four randomized blocks. Grain yields in kg/ha were:

Seeding rate	Block I	II	III	IV	Total
25	5113	5398	5307	4678	20 496
50	5346	5952	4719	4264	20 281
75	5272	5713	5483	4749	21 217
100	5164	4831	4986	4410	19 391
125	4804	4848	4432	4748	18 832
150	5254	4542	4919	4098	18 813

They gave an analysis of variance, as follows:

Source	d.f.	s.s.	m.s.	*F*
Treatments	5	119 8330	23 9666	2.17
'Error'	15	165 8375	11 0558	
Stratum total	20	285 7705		

Satisfy yourself that the coefficients for the linear and quadratic contrasts are

L, $(-5 \quad -3 \quad -1 \quad +1 \quad +3 \quad +5)$;
Q, $(+5 \quad -1 \quad -4 \quad -4 \quad -1 \quad +5)$.

Partition the sum of squares for treatments, separating out the linear and quadratic effects. If the departure from a parabola looks interesting, further contrasts are:

Cubic, $(-5 \quad +7 \quad +4 \quad -4 \quad -7 \quad +5)$
Quartic, $(+1 \quad -3 \quad +2 \quad +2 \quad -3 \quad +1)$
Quintic, $(-1 \quad +5 \quad -10 \quad +10 \quad -5 \quad +1)$.

Write a short report on what you conclude about the yield response to

different seeding rates, together with any recommendation you think should be made.

[Data from K. A. Gomez and A. A. Gomez, *Statistical Procedures for Agricultural Research with Emphasis on Rice* (1976), page 20.]

Exercise 5H

Experimenters sometimes use the sequence, 0, 1, 2, 4, for applications of fertilizer or sprays. (It annoys the statisticians because the progression is neither arithmetic nor geometric.) Find the contrasts that correspond to the linear, quadratic and cubic effects.

Exercise 5I

In Exercise 6C it will be necessary to deal with nitrogen at levels, 0, 40, 70, 100, 130. Anticipate that problem by working out the linear and quadratic effects now.

Chapter 6

Factorial designs

6.1 Summation terms in a factorial design

The main effects and interaction introduced at (5.1.2) and (5.1.4) can be approached in another way using summation terms (see Section 1.4). For the moment a method will be given for use with designs that are orthogonal and such that all treatments have r plots, i.e. for the case considered at (5.6.8). Let the treatments described in Example (3) of Section 5.1 have totals T_{AO}, T_{AF}, T_{BO}, and T_{BF}; then the treatment sum of squares is

$$\frac{(T_{AO}^2 + T_{AF}^2 + T_{BO}^2 + T_{BF}^2)}{r} - \frac{(T_{AO} + T_{AF} + T_{BO} + T_{BF})^2}{4r}$$

The two summation terms will be called

$$S_{VF} = (T_{AO}^2 + T_{AF}^2 + T_{BO}^2 + T_{BF}^2)/r$$

and

$$S_O = (T_{AO} + T_{AF} + T_{BO} + T_{BF})^2/4r$$

because in the first the data have been classified by both variety (V) and fertilization (F), whereas in S_O they have not been classified at all. (Previously S_{VF} has been called S_r.)

If now the main effects are studied separately, two more summation terms will be needed, namely

$$S_V = [(T_{AO} + T_{AF})^2 + (T_{BO} + T_{BF})^2]/2r$$

and

$$S_F = [(T_{AO} + T_{BO})^2 + (T_{AF} + T_{BF})^2]/2r$$

to give sums of squares of respectively $(S_V - S_O)$ and $(S_F - S_O)$, the first for varieties and the second for fertilizations.

It appears then that the treatment sum of squares amounts to $(S_{VF} - S_O)$, of which $(S_V - S_O)$ is due to varieties acting alone and

$(S_F - S_O)$ is due to fertilizer acting alone. That leaves $(S_{VF} - S_V - S_F + S_O)$ for the interaction in which the two work together.

As to degrees of freedom, there are three between four treatments; of those one is for the difference between the varieties and one for the difference between the two levels of fertilization. That leaves one for the interaction.

6.2 The $m \times n$ factorial design

In the examples examined so far, each factor has had two levels, but there is no need to accept that restriction. There could be many varieties and many fertilizer programs. It would still be sensible to test them together in all combinations to see how far the choice of fertilizer should depend upon the variety, or the choice of variety depend upon the level of fertilization that it is proposed to use. The same goes for any other two factors that need to be studied together.

An example is afforded by the data in Table 6*a*, where there were five varieties (A–E) with three methods of cultivation. Using the methods set out in Section 1.4, the following summation terms arise:

$$S = 61^2 + 56^2 + \ldots + 50^2 = 198\,184.00$$
$$S_b = (929^2 + 869^2 + 807^2 + 815^2)/15 = 195\,578.40$$
$$S_t = (190^2 + 223^3 + \ldots + 257^2)/4 = 197\,013.50 = S_{vc}$$
$$S_O = 3420^2/60 = 194\,940.00$$

The basis for these figures is the fact that the block totals are respectively 929, 869, 807 and 815 and that the treatment totals are those set out in Table 6β.

Accordingly the analysis of variance reads:

Source	d.f.	s.s.	m.s.	*F*
Treatments	14	2073.50	148.11	11.69***
'Error'	42	532.10	12.67	
Stratum total	56	2605.60		

Clearly there are large effects of treatments, but it is not yet clear where they lie. That will become more apparent when the treatment line is partitioned according to the factorial structure. Two more summation terms are required:

TABLE 6*a*

The data in the following table were recorded on a cowpea experiment in which there were four randomized blocks. The fifteen treatments were derived from all combinations of five varieties (A–E) with three methods of cultivation (1, 2, 3). The figures represent yields per plot in pounds. Each plot had an area of 0.01 morgen.

	$B_1$61	$A_1$56	$E_3$62	$C_1$63	$A_2$66
I	$D_2$53	$B_2$59	$D_1$65	$D_3$60	$B_3$60
	$E_1$60	$A_3$60	$C_3$65	$C_2$66	$E_2$73
	$A_3$50	$C_1$53	$E_2$77	$A_2$57	$B_1$58
II	$D_1$61	$D_2$53	$C_2$58	$E_3$68	$D_3$58
	$C_3$56	$B_2$55	$E_1$61	$B_3$59	$1_1$45
	$E_3$67	$C_3$50	$C_1$49	$A_2$50	$A_3$45
III	$B_2$51	$B_3$54	$E_2$77	$D_3$56	$B_1$55
	$E_1$50	$C_2$52	$D_2$48	$A_1$43	$D_1$60
	$E_1$53	$D_3$60	$E_3$60	$B_2$52	$B_1$56
IV	$E_2$65	$D_1$63	$D_2$55	$A_3$48	$A_1$46
	$B_3$54	$C_2$55	$C_1$48	$C_3$50	$A_2$50

1 morgen = 8565 m² 1 pound = 454 grams
Note that $e = 0$ (see Section 1.10)

[Data from A. A. Rayner, *Biometry for Agricultural Students* (1969), pp. 439–40.]

Table 6*β* **Treatment totals for the data set out in Table 6*α* (Each total is based on four data)**

		Varieties					
		A	B	C	D	E	Cultivation totals
Methods of cultivation	1	190	230	213	249	224	1106
	2	223	217	231	209	292	1172
	3	203	227	221	234	257	1142
Variety totals		616	674	665	692	773	3420

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$$c - S_O) - (S_V - S_O) - (S_C - S_O)$$
$$= S_{VC} - S_V - S_C + S_O$$

$$(vc - 1) - (v - 1) - (c - 1) = (v - 1)(c - 1)$$

of freedom for the interaction. Hence the full analysis is

Source	d.f.	s.s.	m.s.	F
Varieties (V)	4	1089.17	272.29	21.49***
Cultivations (C)	2	109.20	54.60	4.31*
Interaction (V × C)	8	875.13	109.39	8.63***
'Error'	42	532.10	12.67	
Stratum total	56	2605.60		

The important feature here is the large interaction, and we need to see where it comes from. At this point it will be easier if we convert the values in Table 6β to means, namely:

	A	B	C	D	E
1	47.5	57.5	53.2	62.2	56.0
2	55.8	54.2	57.8	52.2	73.0
3	50.8	56.8	55.2	58.5	64.2

Each of those values is the mean of four data, each of which has a variance of 12.67 ($= s^2$). Accordingly, a difference between any two of them has a

variance of $(\frac{1}{4} + \frac{1}{4})$ 12.67 = 6.335 = 2.52^2, so the standard error of a difference is 2.52.

What happens now should depend upon the questions being asked. It could be that someone wants to know if it matters much what cultivation is used. The main effect of cultivations is significant ($P = 0.05$), but the large interaction warns us that the effect of cultivation depends on the variety. Hence we should ask the question of each variety separately. The following sums of squares, each with two degrees of freedom, will enable us to do so. Each is the difference of two summation terms derived from the data for a single variety (see Table 6β).

A $(190^2 + 223^2 + 203^2)/4 - 616^2/12 = 138.17$
B $(230^2 + 217^2 + 227^2)/4 - 674^2/12 = 23.17$
C $(213^2 + 231^2 + 221^2)/4 - 665^2/12 = 40.67$
D $(249^2 + 209^2 + 234^2)/4 - 692^2/12 = 204.17$
E $(224^2 + 292^2 + 257^2)/4 - 773^2/12 = 578.17$

The five sums of squares represent the combination of the main effect of cultivations and the interaction of cultivations and varieties (i.e. 984.33 with 10 d.f.). The individual values of F are A, 5.45**; B, 0.91; C, 1.60; D, 8.06**; E, 22.82**, each with two and 42 d.f. It certainly appears that the varieties have very different responses to a change in cultivation method. Without knowing more about the exact nature of those methods and those varieties, it is not possible to pursue that line of enquiry much further.

There is no one way of interpreting an analysis of variance. Everything depends upon the questions being asked, but we do emphasize that the first thing to look at in an $m \times n$ factorial design is the interaction. If it is appreciable there is no point in looking at the main effects.

6.3 The $m \times n \times p$ factorial design

It sometimes happens that there are several factors and not just two. An obvious example is afforded by the classical NPK fertilizer trial in which all three elements, nitrogen, phosphorus and potassium, are applied at a range of levels. There could be more factors than that. For example, with some species large dressings of potassium may lead to a scorch on the leaves that can be cured by magnesium, so someone might want a fourth factor. That would make the scheme NPKMg. There could be even more.

However, the immediate object of study is the experiment with three factors, called the $m \times n \times p$ factorial. That means that there are m levels of the first factor M, n levels of the second factor N, and p levels of the third factor P.

That gives mnp treatment totals. The sum of their squares divided by the

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$$S_{MP} - S_M - S_P + S_0 \text{ with } (m-1)(p-1)\text{df}$$
$$= S_{MN} - S_M - S_N + S_0 \text{ with } (m-1)(n-1)\text{df}$$

mething left over, namely

$$_{.P} - S_{NP} - S_{MP} - S_{MN} + S_M + S_N + S_P - S_0$$

as the remaining degrees of freedom, i.e.

$$(mnp-1) - (m-1) - (n-1) - (p-1) - (mp-n-p+1)$$
$$-(mp-m-p+1) - (mn-m-n+1) = (m-1)(n-1)(p-1)$$

This 'three-factor interaction', as it is called, can be looked at in three ways. It can be taken to indicate:

(1) that the interaction, N × P, depends upon the level of M.
(2) that the interaction, M × P, depends upon the level of N.
(3) that the interaction, M × N, depends upon the level of P.

In fact, all three interpretations mean the same thing, though biologically one may be the easiest to explain.

The method can be illustrated thus: Let $m = 3$, the levels being M_1, M_2 and M_3. Let $n = 2$, the levels being N_1 and N_2. Let $p = 4$, the levels being P_1, P_2, P_3 and P_4. Suppose that the 24 treatment totals, each based on four data, were

	P_1	P_2	P_3	P_4	Total
M_1N_1	20	24	25	27	96
M_1N_2	18	21	22	23	84
M_2N_1	26	28	28	30	112
M_2N_2	22	28	27	27	104
M_3N_1	24	29	31	32	116
M_3N_2	22	26	29	35	112
Total	132	156	162	174	624

The vertical margin to the right gives the MN-totals and the horizontal margin below gives the P-totals. Before calculating summation terms, however, these values will need to be checked: it can be done like this.

The first step is to form M × P and N × P totals, and then to form further margins, i.e.

	P_1	P_2	P_3	P_4	Totals
M_1	38	45	47	50	180
M_2	48	56	55	57	216
M_3	46	55	60	67	228
N_1	70	81	84	89	324
N_2	62	75	78	85	300
Total	132	156	162	174	624

The vertical margin gives the M– and N– totals, and they should be checked from the vertical margin of the table above ($96 + 84 = 180$, $112 + 104 = 216$, $116 + 112 = 228$, $96 + 112 + 116 = 324$, $84 + 104 + 112 = 300$).

Similarly the horizontal margin should be obtained from both parts of the table ($38 + 48 + 46 = 132 = 70 + 62$, $45 + 56 + 55 = 156 = 81 + 75$, etc.). Finally, the grand total should be obtained from the three main classifications:

$(132 + 156 + 162 + 174 = 624 = 180 + 216 + 228 = 324 + 300)$.

If everything agrees it is time to calculate the summation terms. They are:

$$S_{MNP} = (20^2 + 24^2 + \ldots + 35^2)/4 = 4151.50$$
$$S_{NP} = (70^2 + 81^2 + \ldots + 85^2)/12 = 4101.33$$
$$S_{MP} = (38^2 + 45^2 + \ldots + 67^2)/8 = 4140.25$$
$$S_{MN} = (96^2 + 84^2 + \ldots + 112^2)/16 = 4102.00$$
$$S_M = (180^2 + 216^2 + 228^2)/32 = 4095.00$$

$$S_N = (324^2 + 300^2)/48 = 4062.00$$
$$S_P = (132^2 + 156^2 + \ldots + 174^2)/24 = 4095.00$$
$$S_0 = 624^2/96 = 4056.00$$

It will be recalled that each of the MNP-totals contains four data and this explains the divisors. It follows that the treatment sum of squares ($S_{MNP} - S_0 = 95.50$) with $23[= (3 \times 2 \times 4) - 1]$ degrees of freedom is partitioned thus:

Source	d.f.	s.s.	m.s.
M	2	39.00	19.500
N	1	6.00	6.000
P	3	39.00	13.000
N × P	3	0.33	0.111
M × P	6	6.25	1.042
M × N	2	1.00	0.500
M × N × P	6	3.92	0.653
Treatments	23	95.50	

As with any other factorial design, it is best to start with the highest-order interaction. Here it is M × N × P. If that is significant there is little point in looking further because the operation of any factor depends upon the levels of the other two, so the most reasonable course is to set out the 24 treatment means without attempting to generalize about them.

If M × N × P appears not to exist, the next step is to look at the two-factor interactions—here they are N × P, M × P and M × N. If any of them are significant, a two-way table is indicated. If none of them shows up, it is all right to proceed to the main effects, which can then be presented without any conditions implied by the interactions.

6.4 Higher factorials

Experiments can contain many factors. There is no need to stop at three. If there are more, there will be higher-order interactions. However, it can sometimes be quite difficult to interpret a three-factor interaction; with four or more, the task can be virtually impossible. Nevertheless, in an exploratory situation at the beginning of a research project, it is not unusual to include a lot of factors for the sake of their two-factor interactions. That gives an indication of what factors should go together

in a series of subsequent experiments, i.e. if they interact they should not be separated, at least not until the interaction is better understood. This will be discussed in more detail in Sections 7.7, 7.8, 7.9 and 7.10.

All interactions can be found from summation terms by a simple regular rule. A term like S_{MNP} is said to be of the third order; one like S_{MN} of the second, and so on. Then any interaction sum of squares is found by taking the corresponding summation term, i.e. S_{MNP} for M × N × P or S_{MN} for M × N, subtracting all relevant terms of one order less, adding all relevant terms of two orders less and so on, altering the signs each time until the correction term, S_0 is reached. Thus the sum of squares for the five-factor interaction, A × B × C × D × E, is

$$S_{ABCDE} - (S_{BCDE} + S_{ACDE} + S_{ABDE} + S_{ABCE} + S_{ABCD}) + (S_{ABC} + S_{ABD} + S_{ABE} + S_{ACD} + S_{ACE} + S_{ADE} + S_{BCD} + S_{BCE} + S_{BDE} + S_{CDE}) - (S_{AB} + S_{AC} + S_{AD} + S_{AE} + S_{BC} + S_{BD} + S_{BE} + S_{CD} + S_{CE} + S_{DE}) + (S_A + S_B + S_C + S_D + S_E) - S_0.$$

It will have $(a-1)(b-1)(c-1)(d-1)(e-1)$ degrees of freedom. Thus it will be seen that the rule gives the familiar expressions for interactions with two or three factors.

6.5 Contrasts of factorial designs

Any main effect can be described as a set of contrasts. If it has p levels, $(p-1)$ contrasts are needed. For example, if $p = 4$, the following three will sum up the main effect:

$$\begin{matrix}(+1 & -1 & 0 & 0)\\ (+1 & +1 & -2 & 0)\\ (+1 & +1 & +1 & -3)\end{matrix} \qquad (6.5.1)$$

If the treatment totals were T_A, T_B, T_C, T_D, each based on r plots, the three sums of squares would be

$$\frac{(T_A - T_B)^2}{2r}, \quad \frac{(T_A + T_B - 2T_C)^2}{6r} \quad \text{and} \quad \frac{(T_A + T_B + T_C - 3T_D)^2}{12r}$$

They sum to

$$\frac{T_A^2 + T_B^2 + T_C^2 + T_D^2}{r} - \frac{(T_A + T_B + T_C + T_D)^2}{4r}$$

That is the familiar expression for a main effect, i.e. the difference between two summation terms, one for treatments and the other the correction term.

The method can be extended to any number of levels. Thus, if there were six, five contrasts would be needed, as follows:

$$\begin{array}{rrrrrr} (+1 & -1 & 0 & 0 & 0 & 0) \\ (+1 & +1 & -2 & 0 & 0 & 0) \\ (+1 & +1 & +1 & -3 & 0 & 0) \\ (+1 & +1 & +1 & +1 & -4 & 0) \\ (+1 & +1 & +1 & +1 & +1 & -5) \end{array}$$

There is no need to use these particular contrasts if some other set would be appropriate, e.g. if there was need of linear, quadratic and other effects, etc. It is only that the contrasts just described are always available.

With an interaction the contrasts are found by multiplying out contrasts of two main effects. For example, with an $m \times n$ design, if $m = 3$ and $n = 2$, the contrasts (1) and (2) in Table 6γ represent the first main effect as a linear and a quadratic effect. Contrast (3) represents the main effect of the second factor. Contrasts (4) and (5) which are derived respectively from (1) and (3) and from (2) and (3), represent the components of the interaction.

TABLE 6γ Contrasts in a 3 × 2 design

	(1)	(2)	(3)	(4)	(5)
M_1N_1	−1	+1	−1	+1(= −1 × −1)	−1(= +1 × −1)
N_2	−1	+1	+1	−1(= −1 × +1)	+1(= +1 × +1)
M_2N_1	0	−2	−1	0(= 0 × −1)	+2(= −2 × −1)
N_2	0	−2	+1	0(= 0 × +1)	−2(= −2 × +1)
N_3N_1	+1	+1	−1	−1(= +1 × −1)	−1(= +1 × −1)
N_2	+1	+1	+1	+1(= +1 × +1)	+1(= +1 × +1)

(6.5.2)

That explains why the number of degrees of freedom for an interaction always equals the product of those for the two main effects, i.e. each contrast of one set is multiplied out by each contrast of the other set.

6.6 Yates's algorithm

If all factors are at two levels, the values of the various contrasts can be found by a method that is appealing in its simplicity. The steps are as follows:

In the extreme left-hand column we will make a list of all the treatments. In the case of a quantitative factor we shall represent the higher level by a small letter, corresponding to the capital letter that indicates the factor. The absence of the letter means that the treatment has the factor at its lower level. If all factors are at the lower level we shall designate the treatment by (1). For a qualitative factor it is immaterial which level is

regarded as the higher and which as the lower, so long as consistency is maintained. Further, we shall list the treatments in 'standard order'. That is to say, we shall start with treatment (1) and m and then we shall add factor N to expand the list to (1) m n mn. Then we shall introduce factor P to make it

$$(1) \quad m \quad n \quad mn \quad p \quad mp \quad np \quad mnp.$$

Obviously the process can be continued indefinitely until the last factor has been included. Against each treatment we give the total of its data. We shall call this 'Column 0'. We illustrate the entire algorithm in Table 6δ, where each total represents the sum of four data.

Having formed Column 0 we calculate Column 1 from it. First we write down the total (302) of the first and second entries in Column 0; the next value is the sum of the next two entries in Column 0, i.e., 104 + 257 = 361, and we continue in that way to the bottom of the column, which gives 129 + 274 = 403. Next in Column 1 we write down the corresponding differences, beginning with the second minus the first (181 − 121 = 60). It is important to take them in proper order and to put in the sign, whether positive or negative. Thus the next entry is + 153, namely 257−104. We continue to the bottom of the column, the last entry being +145 (= 274 − 129). Column 2 is calculated from Column 1 in exactly the same way, and we continue until the number of the last column is that of the number of factors, which here equals 3.

TABLE 6δ An example of the application of Yates's algorithm

Column	$\bar{0}$	1	2	3	
(1)	121	302	663	1362	Grand total
m	181	361	699	+ 408	M
n	104	296	+ 213	+ 166	N
mn	257	403	+ 195	+ 188	M × N
p	123	+ 60	+ 59	+ 36	P
mp	173	+ 153	+ 107	− 18	M × P
np	129	+ 50	+ 93	+ 48	N × P
mnp	274	+ 145	+ 95	+ 2	M × N × P
Odds	477	708	1028		
Evens	885	1062	1096		
First half		1362	1770	2124	
Second half		+ 408	+ 354	+ 68	

In the last column we have the contrasts expressed in terms of the treatment totals, so we can use the method at (5.6.8). Further, a simplification is possible because in the contrasts for the various effects each treatment has a coefficient of ± 1. Accordingly U equals the number of treatments and rU equals the number of plots.

In the example of Table 6δ, where there are 32 plots in all, the sum of squares for the main effect of M is $(+408)^2/32$, i.e., 5202.00, while that for the main effect of N is $(+166)^2/32 = 861.125$ and so on. The full partition of the treatment sum of squares is:

Source	d.f.	s.s.
M	1	5202.000
N	1	861.125
P	1	40.500
M × N	1	1104.500
M × P	1	10.125
N × P	1	72.000
M × N × P	1	0.125
Total treatments	7	7290.375

The other components in the analysis of variance ('Error', Stratum total, etc.) are calculated in the usual way.

There are certain checks that should be made, as follows.

We calculate the sub-totals entered below the columns as in the example. Here 'Odds' column 0 = 121 + 104 + 123 + 129 = 477 and 'Evens' column 0 = 181 + 257 + 173 + 274 = 885.

In column 1 'First half' = 302 + 361 + 296 + 403 = 1362
'Second half' = + 60 + (+ 153) + (+ 50) + (+ 145) = + 408.

'First half' column 1 should be the sum of 'Odds' + 'Evens' in column 0, i.e. 302 + 351 + 296 + 403 = 477 + 885. The 'Second half' column 1 should sum to 'Evens' − 'Odds' in column 0, i.e. + 60 + 153 + 50 + 145 = 885 − 477. We repeat these two checks with columns 1 and 2, and again with columns 2 and 3.

A simple rule for writing the contrasts

The contrasts of the various effects can be read downwards in the following table, all coefficients being either + 1 or − 1. They were obtained by the method at (6.5.2).

The Yates table for the example is:

	G	M	N	M×N	P	M×P	N×P	M×N×P
(1)	+	−	−	+	−	+	+	−
m	+	+	−	−	−	−	+	+
n	+	−	+	−	−	+	−	+
mn	+	+	+	+	−	−	−	−
p	+	−	−	+	+	−	−	+
mp	+	+	−	−	+	+	−	−
np	+	−	+	−	+	−	+	−
mnp	+	+	+	+	+	+	+	+

(6.6.1)

The columns of this table give a ready means of calculating the value of any effect. Suppose, for example, that the interaction of M and N is required. Its value can be derived from the appropriate column, i.e. it comes from the contrast

$$(+1 \quad -1 \quad -1 \quad +1 \quad +1 \quad -1 \quad -1 \quad +1) \qquad (6.6.2)$$

The table can be derived very simply. In any main effect a treatment has + 1 for its coefficient if it has the factor at its higher level and − 1 if it has the lower. In an interaction its coefficient will depend upon the number of factors represented at their upper level. If that number is even, the coefficient will be + 1; if it is odd the coefficient will be − 1. Thus, for M × N when there are three factors, M, N and P, the working goes like this:

(1) has no higher level	0 is regarded as even,	hence + 1;
m has one higher level, M,	1 is odd	hence − 1;
n has one higher level, N,	1 is odd	hence − 1;
mn has two higher levels,	2 is even	hence + 1.

We are dealing with the interaction, M × N, P not coming into it. Consequently, it is immaterial whether P has its higher or its lower level. For that reason *p*, *mp*, *np*, and *mnp* will have the same coefficients as (1), *m*, *n* and *mn* respectively. The rule provides an easy way of writing down the contrasts for a 2^k design. (With an interaction of odd order it will reverse all signs, but that is of no importance where the value of the

contrast is to be squared, though it is vital to note what has happened when interpreting the meaning.)

6.7 Quantity–quality interactions

Suppose that an experiment has been conducted to compare two forms of nitrogen (A and B) at three levels (0, 1, 2). In the ordinary way, given a 2 × 3 factorial set of treatments (A0, A1, A2, B0, B1, B2), the contrasts of interest would be:

Main effect of form of fertilizer.

$$(+1 \quad +1 \quad +1 \quad -1 \quad -1 \quad -1)$$

Linear effect of increasing applications

$$(-1 \quad 0 \quad +1 \quad -1 \quad 0 \quad +1) \qquad (6.7.1)$$

Quadratic effect

$$(+1 \quad -2 \quad +1 \quad +1 \quad -2 \quad +1)$$

Interaction

$$(-1 \quad 0 \quad +1 \quad +1 \quad 0 \quad -1) \text{ and} \qquad (6.7.2)$$
$$(+1 \quad -2 \quad +1 \quad -1 \quad +2 \quad -1)$$

They would all be mutually orthogonal, giving a simple covariance matrix like that considered at (5.6.8). However, in this instance A0 is the same as B0, and such contrasts are unsatisfactory. A0 and B0 cannot be used to compare the effect of form of fertilizer, which is better measured by

$$(0 \quad +1 \quad +1 \quad 0 \quad -1 \quad -1) \qquad (6.7.3)$$

Its interaction with increasing level of application is

$$(0 \quad +1 \quad -1 \quad 0 \quad -1 \quad +1).$$

This leaves one contrast still not identified. It must be orthogonal to all those already found and turns out to be

$$(-1 \quad 0 \quad 0 \quad +1 \quad 0 \quad 0)$$

That is to say, it is the contrast between the two identical treatments. Really that belongs to 'error'; it is not a treatment effect at all. When it has been transferred, only four degrees of freedom are left for treatments. On reflection it will be seen that this is correct because there are in fact only five, not six treatments.

More complicated cases arise, e.g. there could be several forms of nitrogen; but the general principle remains, i.e. to isolate the contrasts that really belong to 'error', and to form meaningful contrasts from what remains.

Exercise 6A

An experiment was conducted on sugar cane in five randomized blocks. There were two fertilizer factors, phosphorus at two levels and potassium at four equally spaced levels. Treatment totals in tons per acre were:

	K_0	K_1	K_2	K_3
P_0	180	248	277	285
P_1	251	307	342	346

First of all, work out the treatment sum of squares and partition it thus, using summation terms as in Section 6.2:

P	1
K	3
P × K	3
Treatments	7

Then use orthogonal contrasts to partition further, as in Section 6.5:

P	1
Linear K	1
Quadratic K	1
Cubic K	1
P × Linear K	1
P × Quadratic K	1
P × Cubic K	1
Treatments	7

1 acre = 4047 m² 1 ton = 1016 kg

[Data from W. G. Cochran and G. M. Cox, *Experimental Design*, 2nd edn, 1950, p. 161.]

Exercise 6B

An experiment was conducted to study the control of blight on potatoes. The field plan and the plot yields in pounds are given below. Each plot consisted of four rows, three feet apart, and 16.5 feet long.

	v_2f_1	v_2f_3	v_2f_2	v_1f_2	v_3f_1
	34.1	41.7	36.0	55.9	35.9
Block I	v_3f_4	v_2f_4	v_2f_0	v_3f_0	v_3f_2
	76.3	63.9	21.3	36.6	54.2
	v_3f_3	v_1f_4	v_1f_1	v_1f_3	v_1f_0
	72.2	100.9	55.8	72.7	39.0
	v_1f_1	v_1f_4	v_3f_0	v_3f_4	v_2f_1
	51.9	88.3	35.7	88.2	21.0
Block II	v_3f_2	v_1f_3	v_1f_0	v_2f_2	v_2f_3
	42.9	64.7	42.2	24.5	41.0
	v_2f_4	v_3f_1	v_1f_2	v_3f_3	v_2f_0
	62.1	33.1	65.6	66.5	11.4
	v_3f_4	v_2f_2	v_1f_2	v_3f_3	v_3f_0
	66.3	26.4	60.1	52.7	35.9
Block III	v_3f_2	v_2f_3	v_1f_0	v_2f_0	v_2f_4
	39.1	59.8	45.2	11.8	69.0
	v_1f_3	v_2f_1	v_1f_1	v_3f_1	v_1f_4
	75.1	28.3	47.2	53.1	102.1

[1 foot = 0.305 m 1 pound = 454 grams]

Calculate the analysis of variance, partitioning fertilizer effects into a linear and quadratic component and another for departure from the quadratic form.

[Data from A. A. Rayner, *A First Course in Biometry for Agriculture Students* (1969), p. 466.]

Exercise 6C

An experiment was conducted to find the best level of nitrogen fertilization of rice. Because that might depend upon the variety, a factorial

experiment was decided upon with four randomized blocks. The data, which give grain yields in kilograms per hectare, were as follows:

Variety	Nitrogen in kg/ha	Block I	II	III	IV
A	0	3852	2606	3144	2894
	40	4788	4936	4562	4608
	70	4576	4454	4884	3924
	100	6034	5276	5906	5652
	130	5874	5916	5984	5518
B	0	2846	3794	4108	3444
	40	4956	5128	4150	4990
	70	5928	5698	5810	4308
	100	5664	5362	6458	5474
	130	5458	5546	5786	5932
C	0	4192	3754	3738	3428
	40	5250	4582	4896	4286
	70	5822	4848	5678	4932
	100	5888	5524	6042	4756
	130	5864	5264	6056	5362

Analyse the data, separating the linear and quadratic effects in the treatment sum of squares and the interaction. The form of the linear and quadratic effects can be derived from Section 5.8. (See Exercise 5I.)

[Data from K. A. Gomez and A. A. Gomez, *Statistical Procedures for Agricultural Research with Emphasis on Rice* (1976), p. 58.]

Exercise 6D

An experiment was conducted on maize using two randomized blocks, each of 16 plots. There were three factors:

- A, B Two varieties
- I, U Plots infested or uninfested with witchweed
- F_1–F_4 Four fertilizer treatments, namely
  - F_1 No added fertilizer
  - F_2 Superphosphate alone added
  - F_3 Superphosphate and manure added
  - F_4 Superphosphate, nitrogen and potassium added.

Data were as follows. They represent yield of grain in pounds per plot of 0.01 morgen.

	BIF_3	BUF_1	AIF_3	BIF_4
	13.5	12.8	15.8	11.6
	AIF_1	BUF_4	AIF_2	AUF_1
I	10.4	17.1	12.5	14.8
	BIF_2	BUF_2	BIF_1	AIF_4
	11.8	16.9	9.5	11.3
	BUF_3	AUF_3	AUF_4	AUF_2
	22.3	24.9	19.9	19.7
	BUF_2	AIF_1	BIF_2	AUF_4
	16.0	10.0	9.5	19.2
	AUF_2	BUF_1	BIF_1	AUF_3
II	18.0	13.0	9.6	22.0
	BIF_3	AIF_4	BUF_4	BUF_3
	13.4	11.4	16.6	20.0
	AIF_2	BIF_4	AUF_1	AIF_3
	10.1	9.2	14.0	13.6

1 morgen = 8565 m^2 1 pound = 454 grams

Calculate a complete analysis of variance.

[Data from A. A. Rayner, *A First Course in Biometry for Agricultural Students* (1969), p. 456.]

Exercise 6E

There are four factors, each at two levels, namely, the presence or absence of

m manure
n nitrogen
p phosphorus
k potassium.

The crop is grass and figures represent the total yield of six cuts. The $2^4 = 16$ treatment totals, each representing four plots, are:

(1)	121	*k*	168	
m	181	*mk*	217	
n	104	*nk*	290	
mn	257	*mnk*	321	
p	123	*pk*	173	
mp	173	*mpk*	250	
np	129	*npk*	351	
mnp	274	*mnpk*	362	Grand total 3494

The analysis of variance so far reads:

Source	d.f.	s.s
Treatments	15	26 791.9
'Error'	45	4 074.2
Stratum total	60	30 866.1

Calculate the full analysis of variance, giving a complete partition of the treatment effects into components, each with one degree of freedom. Starting with the highest-order interaction, attempt an interpretation.

[Data from W. G. Cochran and G. M. Cox, *Experimental Designs* (1950), p. 158.]

Chapter 7

Split plots and confounding

7.1 Split-plot experiments

In certain experiments it is necessary or advisable to use two different types of plot, a feature which leads to the split-plot design. In factorial experiments it may be essential for practical reasons to apply the same level of one treatment to several adjacent plots. In that case complete randomization is not possible. Certain treatments such as chemical sprays or fertilizers can be applied over small areas, whereas ploughing methods may require much larger plots. As a result one factor, comprising the 'main treatments', is applied to plots, sometimes called 'main plots' to distinguish them. The other factor, comprising the 'sub-treatments', is applied to 'sub-plots', which are divisions of the main plots. In effect the main plots serve as blocks for the sub-plots. The position is that described in Section 1.9. Another use of split-plot designs comes when it is required to place emphasis, the intention from the beginning being to study one factor more precisely than the other (see Section 7.4).

The procedure for arranging a split-plot design is first to devise a basic experiment which uses the main-plot treatments. The basic experiment is usually arranged in randomized blocks, although it is possible to use a completely randomized design or a Latin square or some non-orthogonal design instead. Each of the main plots is then divided into an equal number of similar units (sub-plots) and the sub-plot treatments are randomly allocated to them in some orthogonal manner. This allocation requires a separate randomization for each main plot.

As a result of the randomization process there are three strata of error variation:

(a) between blocks, if they are present, within the whole;
(b) between plots within blocks if there are any, and otherwise within the whole;
(c) between sub-plots within plots.

The main-plot error is used for comparisons of main-plot treatments, but

both the sub-treatments and the interaction between the two sets of treatments are tested against the sub-plot error.

In a three-factor experiment it is possible to extend the process through the division of the sub-plots into sub^2-plots (or sub-sub-plots), if that seems appropriate. Such an experiment has a further stratum.

7.2 Example of an experiment in split plots

An example from *Statistical Methods* by G. W. Snedecor and W. G. Cochran (1967, p. 370) used three varieties of alfalfa (lucerne) on the main-plots in six randomized blocks. Each of the main plots was divided into four sub-plots, the sub-plot treatments being four cutting schemes. All sub-plots were cut twice: the second cut took place on 27th July. (The data of the first are not given.) Some of the plots received a further cut as follows: B, 1st September; C, 20th September; D, 7th October, but A was not cut further. The yields in tons per acre for the following year are set out in Table 7*a*.

Table 7*a* Yields in tons per acre for an alfalfa (lucerne) experiment with three varieties and four cutting treatments

Variety	Cutting	Block I	II	III	IV	V	VI
Ladak	A	2.17	1.88	1.62	2.34	1.58	1.66
	B	1.58	1.26	1.22	1.59	1.25	0.94
	C	2.29	1.60	1.67	1.91	1.39	1.12
	D	2.23	2.01	1.82	2.10	1.66	1.10
Cossack	A	2.33	2.01	1.70	1.78	1.42	1.35
	B	1.38	1.30	1.85	1.09	1.13	1.06
	C	1.86	1.70	1.81	1.54	1.67	0.88
	D	2.27	1.81	2.01	1.40	1.31	1.06
Ranger	A	1.75	1.95	2.13	1.78	1.31	1.30
	B	1.52	1.47	1.80	1.37	1.01	1.31
	C	1.55	1.61	1.82	1.56	1.23	1.13
	D	1.56	1.72	1.99	1.55	1.51	1.33

Two further tables are required for the analysis. They are formed from the data as two-way tables, the first for blocks and varieties, the second for cutting dates and varieties.

		Block						
		I	II	III	IV	V	VI	Total
Variety	Ladak	8.27	6.75	6.33	7.94	5.88	4.82	39.99
	Cossack	7.84	6.82	7.37	5.81	5.53	4.35	37.72
	Ranger	6.38	6.75	7.74	6.26	5.06	5.07	37.26
	Total	22.49	20.32	21.44	20.01	16.47	14.24	114.97

(7.2.1)

		Cutting date				
		A	B	C	D	Total
Variety	Ladak	11.25	7.84	9.98	10.92	39.99
	Cossack	10.59	7.81	9.46	9.86	37.72
	Ranger	10.22	8.48	8.90	9.66	37.26
	Total	32.06	24.13	28.34	30.44	114.97

The sums of squares can be found from the summation terms as usual, but there are important differences from the general method for factorial experiments. As always with randomized blocks, the main-plot error is the interaction between blocks and main-plot treatments, while the sub-plot error is formed from the interaction of blocks with all other effects.

7.3 The analysis of variance for a split-plot design

If there are b blocks and a factor, M, with m levels is applied to main plots, while another factor, N, with n levels is applied to sub-plots, the analyses of variance are as follows. Note that there are three strata: (a) blocks/total area, to which no treatments have been applied; (b) main plots/blocks which carries M; and (c) sub-plots/plots which carries N and also M × N. However, no treatments have been applied in (a), so it can be ignored.

	Source	d.f.	s.s.
(a)	Blocks	$b-1$	$S_b - S_0$
(b)	M	$m-1$	$S_m - S_0$
	'Error i'	$(b-1)(m-1)$	$S_i - S_b - S_m + S_0$
Stratum total i		$b(m-1)$	$S_i - S_b$
(c)	N	$n-1$	$S_n - S_0$
	M × N	$(m-1)(m-1)$	$S_{mn} - S_m - S_n + S_0$
	'Error ii'	$m(b-1)(n-1)$	$S_{ii} - S_i - S_{mn} + S_m$
Stratum total ii		$bm(n-1)$	$S_{ii} - S_i$

There are two total terms, S_i and S_{ii}, based respectively on the main plots and the sub-plots. This feature will be illustrated below in a worked example. It will be seen from the degrees of freedom for 'Error ii' that effectively a number (m) of experiments in randomized blocks, each with $(b-1)(n-1)$ degrees of freedom for 'error', have been carried out on the plots of each of the main treatments. The form of the sum of squares shows the same. This can be a useful way of looking at a design in split-plots, especially if something goes wrong. Any damage will then extend only to the component for the main treatment involved. The rest should be all right.

If the main treatments had been disposed in some other way, say in a Latin square, the main plot analysis would be altered accordingly, but the sub-plot analysis would have remained the same.

To take the example at (7.2.1), $b = 6$, $m = 3$ and $n = 4$. The analysis of variance requires calculation of the following summation terms:

$$\begin{aligned}
S_i &= (8.27^2 + 6.75^2 + \ldots + 5.07^2)/4 &&= 189.2749\\
S_{ii} &= (2.17^2 + 1.88^2 + \ldots + 1.33^2) &&= 192.7065\\
S_b &= (22.49^2 + 20.32^2 + \ldots + 14.24^2)/12 &&= 187.7346\\
S_m &= (39.99^2 + 33.72^2 + 37.26^2)/24 &&= 183.7628\\
S_n &= (32.06^2 + 24.13^2 + \ldots + 30.44^2)/18 &&= 185.5472\\
S_{mn} &= (11.25^2 + 7.84^2 + \ldots + 9.66^2)/6 &&= 185.9358\\
S_o &= 114.97^2/72 &&= 183.5847
\end{aligned} \qquad (7.3.1)$$

In the sense of Section 1.10, e has been taken as 2.

The analysis of variance is therefore:

	Source	d.f.	s.s.	m.s.	F
(a)	Blocks	5	4.1499	0.8300	
(b)	Varieties (V)	2	0.1781	0.0890	0.65
	'Error i'	10	1.3622	0.1362	
Stratum total i		12	1.5403		

(7.3.2)

(c)	Cuttings (C)	3	1.9625	0.6542	23.36
	V × C	6	0.2105	0.0351	1.26
	'Error ii'	45	1.2586	0.0280	
Stratum total ii		54	3.4316		

Leaving aside for the moment the error variances, the first line to look at is the one for interaction. Clearly there is no reason here to believe that the effect of cuttings depends upon the variety, so it is correct to proceed to the main effect for cuttings and work with means taken over all varieties, namely

A	B	C	S		
1.78	1.34	1.57	1.69	S.E. = 0.039	(7.3.3)

The standard error is obtained from 'Error ii'. Since each mean contains 18 data, S.E. = $\sqrt{0.0280/18} = 0.039$.

From these data, i.e. those at (7.2.1), there is no statistical evidence that the varieties differ, but since they are genetically different it is not to be expected that they will in fact be giving the same yields.

We note that the split-plot design has been very effective in reducing the error of the experiment, at least for the effect of cuttings and the interaction, which appear in the last part of the analysis. The sub-plot 'error' variance of 0.0280 is only 21 percent of that for the main plots. The consequent loss of information in the main plot analysis would be of little importance if, as seems likely, varieties were introduced only to see if they would provoke an interaction.

Some computer packages give the variance ratio between Error i and Error ii. It is useful as showing how far any advantage was gained by splitting the plots.

7.4 Strata in relation to split plots

This is perhaps the moment to relate an analysis of variance like that at (7.3.2) more closely to the consideration of strata in Section 1.9. There are in fact three strata involved, namely:

Blocks/Total area Plots/Blocks and Sub-plots/Plots.

The first is null because no differential treatments have been applied to blocks. Nonetheless the block line was given largely as a matter of convention, though it does have its uses. It must be assumed that blocks have been chosen to pick up differences in the site and that other

differences have become associated with them as part of the administration of the experiment. It is therefore reassuring that quite a lot of variation has been removed by them. Action would be required only if blocks had been ineffective.

The second (Plots/Blocks) carries the varieties. It also is not of great importance if, as appears likely, the varieties were introduced only to provoke an interaction. (If that was the intention things have worked out badly because the varieties are in fact very similar. Of course, they may differ in habit or season and that could be useful.) Finally, the important results are those in the third stratum. It carries cuttings, which had been applied within that stratum, and the interaction, which appears where it does because both factors are involved in its calculation. It is therefore satisfactory that its 'error' variance is so low.

It is sometimes assumed that when a succession of classifications are nested one within another, as here (i.e. Sub-plots → Plots → Blocks → Total Area), the 'error' variances in the strata will decrease as the units become smaller. At (7.3.2) that expectation has been fulfilled, the successive variances being

Blocks/Total Area	0.8300
Plots/Blocks	0.1362
Sub-plots/Plots	0.0280

The first was found as $(S_b - S_0)/5$.

However, this does not necessarily happen and a simple example will show why. Suppose that a block has a pronounced fertility gradient. In particular we will suppose that the north end has better soil than the south. Accordingly, plots are made long in the north–south direction and narrow east–west, so that each shall have its fair share of good and bad soil. Now someone wants to divide plots into sub-plots. If the plots are divided by north–south boundaries so that sub-plots are as long as the plots but narrower, it is to be expected that the 'error' for sub-plots/plots will be less than that for plots/blocks. However, that division may not be possible; it could give sub-plots so narrow that edge-effects were serious. If instead the plots were divided by east–west boundaries, the fertility pattern, which was so carefully eliminated in the plots/blocks stratum, will now appear in that of sub-plots/plots. In each plot one sub-plot will be on the good land in the north, another on the poor land at the south, with others in between. Consequently, the 'error' variance for sub-plots/plots will be appreciably greater than that for plots/blocks, the opposite of what was hoped for.

Much the same problem arises when blocks are chosen wrongly and cross the fertility contours instead of lying along them. (This was discussed in Section 1.8.)

In Section 1.9 a number of other suggestions were made how plots of different sizes might arise, and there is no need to look at them all because they have much the same characteristics. Perhaps the chief problems occur when it is necessary to work out standard errors of treatment differences. They will be considered in the next section.

One special case does deserve notice. It was mentioned in Section 1.6 that a row-and-column design allows for edge-effects. That is true, but difficulties arise if the plots are split. Necessarily some sub-plots will be more subject to those edge-effects than others. This is not a general argument against splitting plots with row-and-column designs. It applies only when there are marked edge-effects that are meant to be removed by the rows and columns. If there are, a discard area round the edges is essential.

Strip-plot or criss-cross designs

Before proceeding to details about split-plot designs we should mention another possibility, namely, the 'strip-plot design' or, as it is often called, the 'criss-cross' design. In a split-plot design plots with the same level of one factor are kept together. A strip-plot design on the other hand contains rows and columns; one factor is applied at random to the rows and the other to the columns. (Naturally there is a fresh randomization in each block.) The result might be like this:

	Ab	Cb	Bb	Db			Cc	Bc	Ac	Dc
Block I	Aa	Ca	Ba	Da		Block II	Cb	Bb	Ab	Db
	Ac	Cc	Bc	Dc			Ca	Ba	Aa	Da

and the other blocks likewise. The strata may be written

Total area → blocks → (rows × columns).

The design is especially useful when there are difficulties about applying either factor on a small area. So long as there is no interaction of rows and columns it gives a particularly good estimate of the interaction of the two factors.

As to analysis, in principle the design is quite straightforward. If there are m row-treatments and n column-treatments, it is easiest to think of the blocks as providing a third factor, B, with b levels. At (7.3.2) the analysis could then have been regarded in these terms:

	Source		d.f.	Composition
(b)	Main effect of M		$(m-1)$	M
	'Error i'		$(b-1)(m-1)$	B × M
(c)	Main effect of N		$(n-1)$	N
	Interaction		$(m-1)(n-1)$	M × N
	'Error ii'		$(b-1)(n-1)$	B × N
		with	$(b-)(m-1)(n-1)$	B × M × N

In the case of a strip-plot design the second analysis is divided to read:

Main effect of N	$(n-1)$	N	(7.4.1)
'Error' for N	$(b-1)(n-1)$	B × N	
Interaction	$(m-1)(n-1)$	M × N	
'Error' for M × N	$(b-1)(m-1)(n-1)$	B × M × N	

Hence each effect, M, N and M × N, has an 'error' variance appropriate to itself. Although the F-test for the interaction is usually sensitive, there are problems when seeking particular effects within the interaction. As the next section shows, there can be difficulties even with a split-plot design. They are much more serious with the designs considered here.

7.5 Standard errors in a split-plot experiment

Before proceeding to work out standard errors of differences there is a point to be cleared up. It relates to the scaling of the main-plot analysis, which at (7.3.2) was actually calculated on a sub-plot basis. This in fact is usual practice but perhaps a little explanation is required. At (7.3.2) everything depends upon S_i, S_b, S_m and S_o, all of which were found at (7.3.1) using divisors derived from the number of sub-plots in each total, not the number of main plots. For example, the divisor for S_m was 24, not six, as might be thought appropriate for a main-plot analysis. In fact it does not matter which basis is adopted so long as everything is consistent. To achieve this, the first part should be worked in sub-plot units. (It is clearly not possible to do the opposite and calculate the second part in main-plot units, so there is not really any alternative.) This was done at (7.3.1). To take an example, the divisor for S_b was 12 not 3. Except possibly for the interaction, the standard errors in a split-plot design do not raise any problems, provided everything be done within a single stratum. The main treatments, for example, lie entirely within the stratum of plots within blocks, being applied to plots and randomized within blocks. If anyone enquires about varieties in (7.3.1) their means are:

Ladak, 1.67 Cossack, 1.57 Ranger, 1.55.

A difference between such means has a variance of

$$(1/24 + 1/24)\ 0.1362 = 0.0135 = (0.106)^2.$$

Note that everything is being worked on a sub-plot basis, as recommended above, even though main-plot comparisons are involved.

For the main effect of sub-treatments everything lies in the stratum of sub-plots within plots, and the only relevant line is that for 'error ii'. The calculations have already been given at (7.3.3).

The interaction is a little more complicated. In most instances only the sub-plot analysis is involved. Thus the F-value at (7.3.2) is 1.26 (= 0.0351/0.0280) with 6 and 45 degrees of freedom. Again if someone should ask about the means of the various cutting treatments for the variety Ladak, they are:

A 1.88 B 1.31 C 1.66 D 1.82.

Differences between them represent comparisons of sub-plots within the same main plots, so 'Error ii' is appropriate and the difference of two such means has a variance of $(1/6 + 1/6)\ 0.0280 = 0.00933 = (0.097)^2$ with 45 degrees of freedom.

The complication comes when someone wants to compare main treatments for a specified sub-treatment, e.g. varieties for cutting method A. The means are:

Ladak 1.88 Cossack 1.76 Ranger 1.70 (7.5.1)

The standard error of such a difference involves both parts of the analysis. (It is no longer true that all comparisons are made between sub-plots within the same main plots). What is needed is a combined variance, found in the following way.

Let the two 'error' variances be V_i and V_{ii} with f_i and f_{ii} degrees of freedom respectively; then the combined variance is

$$[V_i + (s-1)V_{ii}]/s = V_c.$$

where s is the number of sub-plots to a main plot. Thus in the example at (7.3.2)

$$V_c = [0.1362 + (3 \times 0.0280)]/4 = 0.2202/4 = 0.0550.$$

The next step is to find L, where

$$L = \frac{V_i}{V_i + (s-1)V_{ii}} = \frac{0.1362}{0.2202} = 0.6185.$$

Then f_c, the degrees of freedom for V_c, are found thus:

$$\frac{1}{f_c} = \frac{L^2}{f_1} + \frac{(1-L)^2}{f_2}$$

$$= \frac{0.3825}{10} + \frac{0.1455}{45} = 0.0415$$

so $f_c = 24.2$. (Note that degrees of freedom in this context do not have to a whole number.) Hence the standard error of a difference between two of the means at (7.5.1) is $2 \times 0.0550/6 = 0.135$ with 24.1 degrees of freedom. It will be necessary to interpolate in F-tables to find the values required, but that need not be too difficult.

7.6 Split-plot analysis by sweeping

Mostly data from split-plots are analysed using summation terms. Each analysis will contribute its own residual, one component in each stratum, which together will make up the 'total residual' in Section 7.11. In general this will be more useful than their components in detecting where unexpected variation came from, but sometimes the problem is to find which plot within a block is different, or which sub-plot within a plot. It is then that sweeping may be called for. In any case, the operation repays study because it illuminates what goes on in the analysis of split-plot designs and so helps understanding of the way in which many computer programs tackle the problem.

To take the main-plot analysis first, the deviations as given in Section 1.9 equal

Plot mean − Block mean.

As a result each value will occur s times, where s is the number of sub-treatments. Thus, to take the data at (7.2.1), the deviations for Ladak are found in this way. The plot means for the six blocks are respectively:

2.068 1.688 1.582 1.985 1.470 1.205 (7.6.1)

These figures come from the first line of the table at (7.2.1). The block means come from the horizontal margin, i.e.

1.874 1.693 1.787 1.668 1.372 1.187.

Hence, taking the sub-plots of Ladak, the deviations are

+ 0.194	− 0.007	− 0.205	+ 0.317	+ 0.098	+ 0.018
+ 0.194	− 0.007	− 0.205	+ 0.317	+ 0.098	+ 0.018
+ 0.194	− 0.007	− 0.205	+ 0.317	+ 0.098	+ 0.018
+ 0.194	− 0.007	− 0.205	+ 0.317	+ 0.098	+ 0.018

For the two varieties there are likewise six values, each repeated four times, i.e.

Cossack

+ 0.086	+ 0.012	+ 0.056	− 0.216	+ 0.010	− 0.100

Ranger

− 0.279	− 0.006	+ 0.148	− 0.103	− 0.107	+ 0.080.

The sum of squares for 'Error i' and varieties is therefore

$$4[(+0.194)^2 + (-0.007)^2 + \ldots + (+0.080)^2] = 1.5432.$$

This figure does not quite agree with those for the stratum total at (7.3.2) though it is near enough. Where there is a discrepancy the method of summation terms is usually more accurate. The next step is to take variety means of the deviations and to sweep by them. Means are

Ladak + 0.069, Cossack − 0.025, Ranger − 0.045.

That makes the main plot residuals (each arising four times):

Ladak	+ 0.125	− 0.076	− 0.274	+ 0.248	+ 0.029	− 0.051
Cossack	+ 0.111	+ 0.019	+ 0.081	− 0.191	+ 0.035	− 0.075
Ranger	− 0.234	+ 0.039	+ 0.193	− 0.058	− 0.062	+ 0.125

and hence the sum of squares for 'Error i' is

$$4[(+0.125^2) + (-0.076)^2 + \ldots + (+0.125)^2] = 1.3614.$$

Again the figure is near that in (7.3.2). The discrepancies arise from rounding errors, which are made worse because everything has to be multiplied by four. (An additional decimal place in the deviations and residuals would most likely have cleared matters up.)

For the sub-plot analysis the deviations equal

Sub-plot datum − Plot mean.

That is to say, for Ladak, using the plot means found earlier, the sub-plot deviations are:

A	+ 0.102	+ 0.192	+ 0.038	+ 0.355	+ 0.110	+ 0.455
B	− 0.488	− 0.428	− 0.362	+ 0.395	− 0.220	− 0.265
C	+ 0.222	− 0.088	+ 0.085	− 0.075	− 0.080	− 0.085
D	+ 0.162	+ 0.322	+ 0.238	+ 0.115	+ 0.190	− 0.105

(7.6.1)

Similar figures can be found for the other two varieties, making 72 in all. The sum of their squares, 3.4318, is that for the combination of treatments (main effect of cuttings + interaction) and 'Error ii' in the sub-plot analysis at (7.3.2). Note that the main effect of varieties belongs to a different stratum and was removed along with the plot means to which it contributed.

The rest of the detailed calculation is left to the reader. In outline it goes like this. The next step is to sweep for the cutting means, which must of course be based on the deviations, like those at (7.6.1). Taken over all three varieties they are:

A + 0.184 B − 0.256 C − 0.022 D + 0.094.

The values so found have squares that sum to 1.4688. The reduction (3.4318 − 1.4688 = 1.9630) is the cuttings sum of squares at (7.3.2).

The last step is to sweep the figures last found by their means for all combinations, i.e.

Ladak	+ 0.025	− 0.104	+ 0.019	+ 0.060
Cossack	+ 0.009	− 0.014	+ 0.027	− 0.022
Ranger	− 0.033	+ 0.117	− 0.047	− 0.037

(7.6.2)

Sweeping by these means might appear to allow for all the treatment effects, not just the interaction, but this is to miss the point that the data have already been swept by the main effects. (That of varieties was removed in the formation of the deviations for the second stratum. It will be noted that the values at (7.6.2) sum to zero over both margins, so that main effects do not enter further into the calculation.) Sweeping further gives the sub-plot residuals. It will be found that the sum of the squares is 1.2581 which is effectively the value for 'Error ii' at (7.3.2).

A perusal of the above calculations will do much to explain what happens in a series of sweeps—how a sweep once made thereafter gives zero margins for its effect, so that the same sum of squares is not removed

twice, and how each sweep reduces the sum of squares by an amount appropriate to the new effect introduced. (The word 'effect' is used here in the sense of a main effect or a block effect or an interaction.)

7.7 The idea of confounding

In a split-plot design one of the contrasts, the main plot effect of varieties in the example in Section 7.2, is transferred to another stratum. Since comparisons in that stratum are expected to be less good, the effect is to some extent being sacrificed to obtain better comparison of the others. Nevertheless, information about it can commonly be recovered if this is thought to be important. Indeed, that was done in the main-plot analysis at (7.3.2). This was not a very precise analysis, its 'error' variance being 0.1362 with 10 degrees of freedom as compared with 0.0280 with 45 in the sub-plot analysis. In some circumstances one might not want to be bothered with this analysis, but it is there. Such information as it has to impart can always be 'recovered' by working it out.

In many instances it is not a main effect but an interaction that is sacrificed. Given a lot of factors, the two-factor interactions are usually very important. If anyone questions that, he can ask himself why both factors have been introduced into the same experiment if there is no thought that they might interact. It would be simpler to have two experiments. Three-factor interactions are also useful, though not to the same extent, but interactions with four or more factors usually defy interpretation even if they are shown to exist. There is therefore no great objection to sacrificing them if this would help the precision of estimation of other contrasts that are of greater interest. Further, if they are sacrificed for that reason, there is little point in 'recovering' them by a split-plot design; no-one would be interested.

When a contrast is sacrificed in this way, it is said to have been 'confounded'. In a split-plot design, a main effect is confounded and it is usual to recover such information as is available about it. If an interaction is confounded, recovery is not usual.

The relationship between split plots and confounding may be illustrated thus: Let there be two factors A and B giving rise to the treatments

$$(1) \quad A \quad B \quad AB.$$

If now some blocks contain only (1) and A, while the rest contain only B and AB, the result is a design in split-plots. Factor B is on main plots, which are here identical to the blocks, while A is on sub-plot. (It is true that the nomenclature has changed somewhat, but the relationship is clear.) If instead some blocks contain only (1) and AB, while the rest

contain only A and B, it is now the interaction that has been confounded, i.e.,

$$(+1 \quad -1 \quad -1 \quad +1)$$

instead of the main effect

$$(+1 \quad +1 \quad -1 \quad -1)$$

but the idea is the same

The idea can be taken further, as the following will show.

Confounding with a 2^k design

In exploratory experiments it can be helpful to investigate the effect of several factors simultaneously to identify those that show promise and are therefore of special interest. When the most important ones have been found, more detailed experiments may be conducted in which these factors are tested at more levels. Less important factors can be omitted. Factorial experiments can become very large as the number of factors increases, and the size of the experiment can induce unwanted problems. In particular it may not be possible to find enough similar units so that each block shall contain each treatment. In that case only some treatments can occur in any one block. That situation arose in various forms in Chapter 4. Here we will consider another solution. Instead of estimating all contrasts, though allowing some of them to have a reduced efficiency factor, we shall eliminate some altogether in order to obtain better estimates of the rest. In general the least important interaction is the one of highest order, though that is not necessarily so. Mostly a two-factor interaction should be retained. (If it is unimportant, why were the two factors put in the same experiment?) The confounding of a main effect gives a design in split plots.

To take a simple example, we will suppose that an experiment is to have three factors, A, B and C, each at two levels. We shall suppose further that the design is either orthogonal or completely randomized and that all treatments have r replicates, consequently it will be possible to use the methods given at (5.6.7) and (5.6.8). So long as blocks of eight plots are seen as a reasonable solution, there is no great difficulty about design, but what can be done if someone objects that smaller blocks are essential?

If we may sacrifice an interaction, we can design the experiment in blocks of four plots. The obvious one to sacrifice is A × B × C, since the two-factor interactions are probably precious. We know from (6.6.1) that the coefficients of the contrast that we propose to sacrifice are

$$(-1 \quad +1 \quad +1 \quad -1 \quad +1 \quad -1 \quad -1 \quad +1),$$

the treatments being taken in standard order, i.e. (1), *a*, *b*, *ab*, *c*, *ac*, *bc*, *abc*. (As in Section 6.6 we are avoiding confusion between treatments and effects by using small letters for one and capitals for the other.) When we confound, we assign the treatments with a positive coefficient, i.e.,

$$a, \quad b, \quad c, \quad abc$$

to half the blocks, which should be chosen at random, and

$$(1), \quad ab, \quad ac, \quad bc$$

to the other half. The interaction of A × B × C has now become a contrast between blocks instead of within them. If anyone wants to recover it, he or she will have to compare those blocks that contain [*a b c abc*] with those that contain [(1) *ab ac bc*]. This is what was done to recover the main effect confounded in the example of a split-plot design, but here, where an unimportant interaction has been confounded, it is unlikely that anyone will bother. (The situation is a little confused on account of the nomenclature. What are called *blocks* here were *main plots* when we were in a context of split plots, and *plots* here were *sub-plots* then.)

However, all contrasts orthogonal to the one confounded are as they were before. We can now compare the form of the analyses as they would have been without confounding and with it. We shall assume that there are 32 plots.

Degrees of freedom with and without confounding

	Without	With
A	1	1
B	1	1
C	1	1
B × C	1	1
A × C	1	1
A × B	1	1
A × B × C	1	—
'Error'	21	18
Stratum total	28	24

To find the degrees of freedom for the stratum totals, we note that there are four blocks without confounding but eight with. Confounding has given rise to two disadvantages; there are fewer degrees of freedom for 'error' and the three-factor interaction has been lost. The great gain comes from the use of blocks of more acceptable size.

There are other ways in which the two sets of treatments could have been obtained. For example, we could have taken the first set,

$$a \quad b \quad c \quad abc$$

and have 'multiplied' each treatment in it by another treatment that was not contained in it. This operation of multiplying factors and treatments is one to which we shall have to become accustomed. As we saw in Section 6.6, the effect of including a factor in an interaction is to reverse signs if it occurs in the treatment. The effect then of including a factor twice is to restore the original position. Consequently, $A^2 = B^2 = C^2 = 1$. The same rule will apply to treatments. Here, if we multiply each treatment in the first set by *ab*, a treatment that does not occur in it, we get

$$a \times ab = b, \quad b \times ab = a, \quad c \times ab = abc, \quad abc \times ab = c.$$

That is to say, we have generated the second set from the first.

Another way is to expand the polynomial,

$$(1-a)(1-b)(1-c) = (1) - a - b - c + ab + ac + bc - abc$$

which gives the two sets from the positive and negative coefficients in the expansion.

With these methods available, we can ask how we would have confounded, say, the interaction, B × C. (In general we would not want to do so, but we are considering only technique.) To take the first method, from Section 6.6 we see that it is represented by the contrast, (+ 1 + 1 − 1 − 1 − 1 − 1 + 1 + 1), so the two sets of treatments are

$$(1) \quad a \quad bc \quad abc \qquad \text{and} \qquad b \quad ab \quad c \quad ac.$$

When there are only two sets the second method seems superfluous, but we will illustrate it nonetheless. We will take the first set and multiply throughout by *b*. That gives

$$(1) \times b = b, \quad a \times b = ab, \quad bc \times b = c, \quad abc \times b = ac.$$

Finally, we can expand

$$(1 + a)(1 - b)(1 - c) = (1) + a - b - c - ab - ac + bc + abc.$$

The change here, where A does not enter into the interaction, is to use $(1 + a)$ instead of $(1 - a)$. All methods have led to the same result.

The method applies to all cases where all factors are at two levels. If, for example, there were four factors, A, B, C and D, the four factor interaction would be confounded if half the blocks contained

$$(1) \quad ab \quad ac \quad ad \quad bc \quad bd \quad cd \quad abcd \tag{7.7.1}$$

and the rest contained

$$a \quad b \quad c \quad d \quad abc \quad abd \quad acd \quad bcd. \tag{7.7.2}$$

With the unconfounded contrasts there is a simple way of finding the sum of squares for each; it was given in Section 6.6.

7.8 Confounding more than one contrast

The method given in the last section makes it possible to design an experiment with blocks only half the usual size, but that may not be enough. If someone wishes to have block size reduced to a quarter, it is possible to confound further interactions.

We shall take the example at the end of the last section in which the interaction, A × B × C × D, was confounded between two sets of blocks, each block containing eight plots. We shall attempt to divide further into new blocks, each of four plots. To do that we shall confound the three-factor interaction, A × B × C. This involves having

$$(1) \quad d \quad ab \quad ac \quad bc \quad abd \quad acd \quad bcd \tag{7.8.1}$$

in one set and

$$a \quad b \quad c \quad ad \quad bd \quad cd \quad abc \quad abcd \tag{7.8.2}$$

in the other. That enables us to draw up four lists of treatments, α, β, γ and δ, such that A × B × C × D is confounded between lists α and β on the one hand compared with lists γ and δ one the other, and A × B × C is confounded between α and γ on the one hand and β and δ on the other, i.e.

α	(1)	ab	ac	bc
β	ad	bd	cd	$abcd$
γ	d	abd	acd	bcd
δ	a	b	c	abc

We did that by putting into

α all those treatments common to (7.7.1) and 7.8.1),
β all those treatments common to (7.7.1) and (7.8.2),
γ all those treatments common to (7.7.2) and (7.8.1),
δ all those treatments common to (7.7.2) and (7.8.2).

We shall assign the treatments in α to one-quarter of our blocks, which we shall choose at random, β to another quarter, also chosen at random, and so on. (We need scarcely add that the treatments should be assigned at random within the blocks also.)

We have now achieved a design in blocks of four plots such that our two designated contrasts are confounded, but we have done more. Four things can be put into pairs in three ways. We have paired α and β to leave γ and δ and we have paired α and γ to leave β and δ. What happens if we pair α and δ to leave β and γ? That gives us the contrast between

$$(1) \quad a \quad b \quad c \quad ab \quad ac \quad bc \quad abc$$

and

$$d \quad ad \quad bd \quad cd \quad abd \quad acd \quad bcd \quad abcd,$$

It appears that we have also confounded the main effect of D. This is not necessarily a bad thing—we confounded main effects without comment in Section 7.6—but, if we are going to do it, we should act of set purpose and not inadvertently.

In general, if we confound two contrasts, we confound also their 'generalized interaction', i.e., the result of multiplying them following the rule given in the last section that $A^2 = B^2 = \ldots = 1$. That is what happened in the example above, because

$$(A \times B \times C \times D) \times (A \times B \times C) = D.$$

Before leaving this example we may note that the various groups of treatments could have been obtained from one another using the method of multiplication given in the last section. Thus, if α is multiplied throughout by some treatment not contained within itself, say b, the result is [b a abc c], which is δ. If this is now multiplied by some treatment that is in neither α nor δ, say ad, the result is [abd d bcd acd], which is γ. That leaves [ad bd cd $abcd$] for the last list. The method does not help much in forming lists in the first place but it provides a convenient means of checking them.

Usually multiple confounding is not called for with only four factors, because blocks of eight plots can usually be found. Its chief use comes with five factors or more. (If there are five factors, that calls for 32 treatments.

A single confounding will reduce the number of plots needed in a block to 16, but even this may be too many.)

To take an example, we confound A × C × D, B × C × D × E and A × D × E in a 2^5 design. The first, A × C × D, requires division into

a ab ae abe c bc ce bce d bd de bde acd abcd acde abcde

and

(1) *b e be ac abc ace abce ad abd ade abde cd bcd cde bcde.*

What we have done was really very simple. We decided what would be involved in confounding the interaction A × C × D in a 2^3 design, namely, division into

[*a c d acd*] and [(1) *ac ad cd*]. (7.8.3)

Then each of those treatments was multiplied by the treatments, (1), *b*, *e* and *be*, to give the solution, the additional factors being B and E.

The next contrast to be confounded is B × C × D × E, which calls for the division of the treatments into sets,

b ab c ac d ad e ae bcd abcd bce abce bde abde cde acde

and

(1) *a bc abc bd abd be abe cd acd ce ace de ade bcde abcde.*

The additional factor is now A. We have taken the grouping of a 2^4 design with the four-factor interaction confounded, and we have then multiplied each of the treatments so obtained by both (1) and *a*.

We can now form our four lists:

a	*c*	*d*	*ab*	*ae*	*bce*	*bde*	*abcd*	*acde*
β	*a*	*bc*	*bd*	*ce*	*de*	*abe*	*acd*	*abcde*
γ	*b*	*e*	*ac*	*ad*	*bcd*	*cde*	*abce*	*abde*
δ	(1)	*be*	*cd*	*abc*	*abd*	*ace*	*ade*	*bcde*

As before, the first confounded interaction, A × C × D, is given by the difference between lists *a* and *β* as compared with *γ* and *δ*; the second, B × C × D × E, by the difference between *a* and *γ* as compared with *β* and *δ*. This leaves the difference between *a* and *δ* as compared with *β* and *γ*.

Examination shows that it gives the third contrast to be confounded, namely, A × B × E.

If a fourth contrast is added, not only will it be confounded itself but its generalized interaction with the first three will be confounded as well. The following example may be of interest. It represents a 2^5 design in which seven contrasts are confounded, viz. A × C × D, B × C × D × E, A × D × E, A × B × E, C × E, A × B × C, B × D. (The fact that some of the confounded contrasts are two-factor interactions explains why such confounding is not really desirable with fewer than six factors.) The lists are

a	(1)	*abd*	*ace*	*bcde*
β	*a*	*bd*	*ce*	*abcde*
γ	*b*	*ad*	*abce*	*cde*
δ	*c*	*abcd*	*ae*	*bde*
ε	*ab*	*d*	*bce*	*acde*
ς	*ac*	*bcd*	*e*	*abde*
τ	*bc*	*acd*	*abe*	*de*
θ	*abc*	*cd*	*be*	*ade*

We are not now seeking *r* blocks of 32 plots, where *r* is the degree of intended replication, but 8*r* blocks, each of four plots, one-eighth of the blocks to take each of the lists at random. Between 32 treatments there would ordinarily be 31 degrees of freedom, but here seven of the contrasts have been confounded so there are only 24. The outline analysis of variance will have the following form.

Source	d.f.
Unconfounded contrasts	24
'Error'	24 (*r* − 1)
Stratum total	24*r*

A sum of squares can be found for each of the unconfounded contrasts by the use of Yates's algorithm, as set out in Section 6.6, and the method given at (5.6.8).

7.9 Single replication designs

The factors in an exploratory experiment may be quite numerous. Initially there may be no intention of studying the whole subject in depth;

it is just that there are many ideas on how research should proceed, and the proposal is that the factors should be put into one experiment to see which look hopeful. It is true that the investigator could carry out an experiment on each, but that would be wasteful. If all the proposals are put into one 2^k design then, supposing that there are no high-order interactions, each factor will gain from the replication afforded by the others. To take an example, if a 2^5 experiment is conducted and there are no major interactions with more than two factors, then each combination of A and B, i.e., (1), *a*, *b* and *ab*, has its replication increased eight-fold by being studied at eight combinations of C, D and E, i.e., (1), *c*, *d*, *e*, *cd*, *ce*, *de* and *cde*, with which there is no interaction. (This is called 'hidden replication', but we must insist that it exists only if there are no important interactions. Nevertheless, even if three-factor interactions are likely, there is still a four-fold gain in replication if there are no likely four-factor interactions. That surely can safely be assumed.)

Given a large number of factors it is feasible to design a 2^k experiment with a single replication. (For the moment we will not try to introduce any blocks.) To resume the case of five factors, there are five main effects, namely, A, B, C, D and E; there are also ten two-factor interactions, namely, A × B, A × C, A × D, A × E, B × C, B × D, B × E, C × D, C × E and D × E. That leaves 16 degrees of freedom for higher order interactions. Most of them probably do not really exist; any that do are unlikely to be large. It is therefore reasonable to use them collectively as 'error'. The estimate so made may be a little inflated, but that is a fault on the right side. Sometimes there are three-factor interactions that are expected for prior reasons to be important. If that is so, they should be excluded from 'error' and dealt with separately, thereby reducing the degrees of freedom for 'error'; that is quite legitimate. What is wrong is the examination of each interaction with the intention that, if it appears to be having little effect, it can always be put into 'error'. The procedure is wrong because the 'error' is then being formed from interactions selected on account of their low sums of squares. To avoid such bias the components of 'error' should be fixed from the start, and changes made only rarely and for strong reasons.

There is no difficulty about introducing blocks; one of the discarded interactions can be confounded between two halves of the area. If need be, three could be confounded between quarters. Everything is as in Section 7.8 except that the various lists are no longer replicated and, consequently, information about confounded interactions can no longer be recovered by a comparison of blocks. However, it is so unusual for anyone to want to recover information about interactions, especially high-order ones, that the loss is small.

A singly replicated experiment need not have all factors at two levels,

but in an exploratory situation this is usual. There are times, however, when doubts about the correct level of some quantitative factor require that there be three or even more levels. If that is so, there is no objection to using the number required, though it may increase considerably the total number of plots. Some such designs will be considered in Sections 7.11 and 7.12.

7.10 Fractional replication

With numerous factors, especially if all have two levels, it is possible to go even further and to use fractional replication. The problem of total size of the experiment may be serious. For example, if the total number of plots is not to exceed 500, a 2^k experiment with a single replicate must not have more than nine factors. (A 3^k experiment, if one is proposed, must not have more than six.) Some may find a solution by discarding factors, at least for the moment; others may try to minimize the number of levels of each. A third possibility is to use only some of the treatments. That will be done here. Naturally some information will be lost, but since 'error' will have to be estimated from the highest-order interactions this may not matter much.

To illustrate the approach we will suppose that someone wants a 2^6 design. A single replicate would require 64 plots, but this is considered to be too many. We will therefore see what can be done with 32. We shall start by choosing an interaction as the 'defining contrast'. Usually it will be best to take the one of highest order, in this instance

$$A \times B \times C \times D \times E \times F,$$

to separate the 64 treatments into two lists. It is immaterial which of the two we choose. We could make our selection at random or in any other way. Here we shall assume that we have chosen:

(1)	*ae*	*be*	*cf*	*abcd*	*abdf*	*acef*	*bcef*
ab	*af*	*bf*	*de*	*abce*	*abef*	*adef*	*bdef*
ac	*bc*	*cd*	*df*	*abcf*	*acde*	*bcde*	*cdef*
ad	*bd*	*ce*	*ef*	*abde*	*acdf*	*bcdf*	*abcdef*

(7.10.1)

We then allocate those 32 treatments to the plots at random.

We can see the consequences of this procedure better with a simpler example. We shall therefore consider only four factors and consider what interpretation could be made of only eight plots with the treatments

Plot	1	2	3	4	5	6	7	8
Treatment	(1)	*ab*	*ac*	*ad*	*bc*	*bd*	*cd*	*abcd*

(7.10.2)

If we wanted the main effect of A we would have to compare plots 2, 3, 4

and 8 with plots 1, 5, 6 and 7. (We recall what was said in Section 5.2 about all contrasts ultimately being a comparison of data.) Suppose though that we wanted to know about B × C × D, then we would want to compare those plots on which *b*, *c* and *d* were represented an even number of times with those in which the number was odd. (That goes back to Section 6.6.) This being so, we want to compare plots 2, 3, 4 and 8 with plots 1, 5, 6 and 7. In fact, the two contrasts are estimated in exactly the same way. If the difference between the two sets of plots were large, we would not know whether we had a main effect of A or an interaction of B, C and D. Since a main effect is more likely to have a large effect than a three-factor interaction, we would opt for the former. The two effects, A and B × C × D, are said to be 'aliases'. It is like one person having two names, and we cannot be sure which is the right one, though we may make a guess. Any contrast has an alias in the generalized interaction of itself and the defining contrast. Thus,

$$A \times (A \times B \times C \times D) = B \times C \times D.$$

Hence B × C × D is an alias of A and vice versa.

To return to (7.9.1), there are six main effects, those of A, B, C, D, E and F, each with an alias that is a five-factor interaction, so it can be ignored. There are also 15 two-factor interactions, A × B, A × C,..., E × F, each with a four-factor interaction as its alias. Again, if there is a large effect, there will be little doubt which contrast is the cause. Finally there are 20 three-factor interactions that form ten pairs of aliases, namely,

$$\begin{array}{c} A \times B \times C \text{ and its alias } D \times E \times F \\ A \times B \times D \text{ and its alias } C \times E \times F \\ \cdots \quad \cdots \quad \cdots \quad \cdots \\ A \times E \times F \text{ and its alias } B \times C \times D \end{array}$$

Here interpretation is impossible. (Of the two interactions that form a pair there is no reason to prefer one to the other.) Nevertheless, the sums of squares of those ten contrasts can be combined to give an 'error' sum of squares with ten degrees of freedom. In fact, the outline analysis of variance looks like this:

Source	d.f.
Main effects	6
Two-factor interactions	15
'Error'	10
Stratum total	31

Computations required

The calculation of such an analysis is a little lengthy, not on account of any inherent complications but because there are 21 effects to be evaluated. First, the stratum total can be found either by sweeping, as in Section 1.3, or as a difference of two summation terms, one for the data and the other for the general mean (S and S_0 in Section 1.4). Evaluation of the sums of squares for the 21 contrasts of interest is not difficult because a modification of Yates's algorithm is available. (The algorithm is explained in Section 6.6.) We shall illustrate it by a simple example in which there are only three factors, giving rise to eight treatments. If a half-replicate is used, there will in fact be only four of them in the experiment, say *a*, *b*, *c*, *abc*. We shall ascribe data of α, β, and δ respectively to the those plots.

We have four plots, so we think first of four treatments, (1), *a*, *b* and *ab*, factor C being ignored. Of the four treatments thus formed, two, namely (1) and *ab*, do not occur in the experiment, so we shall introduce C by expanding them to c and *abc*. In the table of the algorithm, Column 0 can now be found and the rest of the calculations follow, i.e.,

	0	1	2
$(1) \to c$	γ	$\alpha+\gamma$	$\alpha+\beta+\gamma+\delta$
$a \to a$	α	$\beta+\delta$	$\alpha-\beta-\gamma+\delta$
$b \to b$	β	$\alpha-\gamma$	$-\alpha+\beta-\gamma+\delta$
$ab \to abc$	δ	$-\beta+\delta$	$-\alpha-\beta+\gamma+\delta$
Odds	$\beta+\gamma$	2α	
Evens	$\alpha-\delta$	δ	
First half		$\alpha+\beta+\gamma+\delta$	$2\alpha+2\delta$
Second half		$\alpha-\beta-\gamma+\delta$	$-2\alpha+2\delta$

The three quantities needed to find the sums of squares by use of the method at (5.6.8) are:

Main effect of A (alias B × C)	$\alpha-\beta-\gamma+\delta$;
Main effect of B (alias A × C)	$-\alpha+\beta-\gamma+\delta$;
Main effect of C (alias A × B)	$-\alpha-\beta+\gamma+\delta$.

All can be derived from Column 2.

Of course, the example has concerned an experiment so small that it would never have arisen in practice. It does however illustrate how to deal with the data from any fractionally replicated design. If there are *k* factors, the method is to ignore one of them and to start Column 0 with a complete set of the rest. Then the last factor is introduced where necessary, to make each treatment into one of those in the actual design. After that,

everything is as usual. One advantage is that the sum of squares is calculated for all contrasts. Consequently the 'error' can be found by the addition of sums of squares for all relevant interactions, i.e. those involving three factors or more, but ignoring those of the two highest orders, because they are aliases of the main effects and the two-factor interactions. Because the 'error' sum of squares can be found directly, a check is afforded by everything summing to the stratum total.

More elaborate cases

If there had been seven factors, A, B, ..., G, there would have been seven main effects, each with a six-factor interaction as its alias, and 21 two-factor interactions, each with a five-factor interaction as its alias. Since there would have been 63 degrees of freedom in all, that leaves 35 degrees of freedom for 'error', made up of 35 three-factor interactions, each having as its alias a four-factor interaction, namely,

$$\begin{array}{lll} A \times B \times C & \text{with its alias} & D \times E \times F \times G, \\ A \times B \times D & \text{with its alias} & C \times E \times F \times G, \\ \cdots \quad \cdots \quad \cdots & \cdots \quad \cdots & \cdots \quad \cdots \quad \cdots \\ E \times F \times G & \text{with its alias} & A \times B \times C \times D. \end{array}$$

If the experiment is too large for a single block, one of the contrasts used for 'error' can be confounded to divide the treatments into two lists, one for each block. Multiple confounding can be used, as in Section 7.8, if circumstances call for it. No new principle is introduced by the fractional replication.

If a half-replicate still gives too many plots, it is possible to declare a further defining contrast and to use only a quarter of the treatments. In that case the generalized interaction of the two declared defining contrasts has equal status with them and, in consequence, each contrast under investigation has three aliases, not one. Such a scheme is feasible only when there are many factors.

7.11 Confounding with a 3^k design

So far we have considered only the confounding of a 2^k design, but there are other possibilities. For example, if quadratic effects were of interest, someone might want a 3^k design.

We will first look at the 3^2 case, principally in order to show the approach. The treatments may be written:

00 01 02 10 11 12 20 21 22.

Here the first digit gives the level of factor A, whether 0, 1 or 2, and the second does the same for factor B. Clearly the two degrees of freedom for the main effect of A can be found by adding the totals of three triplets of treatments, i.e.

[00 01 02] [10 11 12] and *[20 21 22].*

They will provide a summation term, S_A, which with S_0 the correction term, will give the desired sum of squares. The same can be done for the main effect of B, the triplets then being

[00 10 20] [01 11 21] and *[02 12 22].*

It is not so obvious that the interaction sum of squares can be calculated similarly. It has four degrees of freedom and it has to be evaluated in two parts. The first comes from the triplets:

[00 11 22], [01 12 20] and *[02 10 21].*

Their three totals will give rise to a summation term, S_I. The second comes from

[00 12 21], [11 20 02] and *[22 01 10].*

That gives a summation term, S_J. Each component will, of course, have two degrees of freedom. The sum of squares for the interaction will be $(S_I - S_J - 2S_0)$ with four.

The conventional way of confounding a 3^2 design is to have six blocks, each of three plots. Each of the triplets associated with the interaction will be assigned to one block, like this:

Block	I	*00*	*11*	*22*
Block	II	*01*	*12*	*20*
Block	III	*02*	*10*	*21*
Block	IV	*00*	*12*	*21*
Block	V	*11*	*20*	*02*
Block	VI	*22*	*01*	*10*

With such a design the main effects are completely unconfounded. Component I of the interaction is confounded between Blocks I, II and III, but may be found from Blocks IV, V and VI. Component J is similarly confounded between Blocks IV, V and VI, but may be found from Blocks I, II and III. (It need hardly be said that really the triplets should be

allocated to blocks at random and the treatments should be assigned at random to the plots of that block. For purposes of explanation, however, it is easier if the underlying pattern can be retained.)

The design just given represents traditional wisdom, but it may be asked if anyone really wants an interaction with an efficiency factor of one-half. (See Section 4.1.) In any factorial design the interactions are rarely well enough determined to carry the weight they are required to bear. If a factor is large, no one should look at the main effects associated with it; only if it is small may one desert the particular effects. It needs to be estimated with considerable precision for such important decisions to depend upon its magnitude. It may be unconventional, but we do suggest that there are times when it would be better to have the main effects at half-efficiency and to estimate the interaction with full efficiency. If the object of the experiment is to find out whether the two factors interacted or operated independently of one another, there would be no doubt about the design to choose. One would assign to blocks the triplets associated with the two main effects. In that case the design would go like this:

Block	I	*00*	*01*	*02*
Block	II	*10*	*11*	*12*
Block	III	*20*	*21*	*22*
Block	IV	*00*	*10*	*20*
Block	V	*01*	*11*	*21*
Block	VI	*02*	*12*	*22*

It would then be necessary to base the calculation of the main effect of A on the data in blocks IV, V and VI and that of B on blocks I, II and III.

The situation is different if there are three factors. We could then be in the familiar situation of wanting the main effects and the two-factor interactions but of being willing to lose the interaction of all three factors. It has eight degrees of freedom, two for each of four components, W, X, Y and Z. In each component there are three sets of nine treatments, and each set will give a data total. The totals of a component will give a summation term from which can be found the sum of squares for the two degrees of freedom. The sets are

W

000	*012*	*021*	*101*	*110*	*122*	*202*	*211*	*220*
002	*011*	*020*	*100*	*112*	*121*	*201*	*210*	*222*
001	*010*	*022*	*102*	*111*	*120*	*200*	*212*	*221*

X

000	*011*	*022*	*102*	*110*	*121*	*201*	*212*	*220*
001	*012*	*020*	*100*	*111*	*122*	*202*	*210*	*221*
002	*010*	*021*	*101*	*112*	*120*	*200*	*211*	*222*

Y

000	*011*	*022*	*101*	*112*	*120*	*202*	*210*	*221*
001	*012*	*020*	*102*	*110*	*121*	*200*	*211*	*222*
002	*010*	*021*	*100*	*111*	*122*	*201*	*212*	*220*

Z

000	*012*	*021*	*102*	*111*	*120*	*201*	*210*	*222*
001	*010*	*022*	*100*	*112*	*121*	*202*	*211*	*220*
002	*011*	*020*	*101*	*110*	*122*	*200*	*212*	*221*

The experiment will need twelve blocks each of nine plots. The sets given above are each allocated one block and the treatments are allocated at random to its plots. Component W will be confounded between the three blocks to which its sets are applied, but its sum of squares can be found from the other nine blocks. It is the same with components X, Y and Z. Hence the whole interaction can be found, but with an efficiency factor of three-quarters.

If the three-factor interaction is indeed of little importance, there will be little occasion to investigate it. The stratum total could be found in the usual way. Then the summation terms could be worked out, as in Section 6.3, to find the sums of squares for the main effects and two-factor interactions. What is left could legitimately be attributed to 'error', the eight degrees of freedom for the three-factor interaction being merged with those that would be used for 'error' anyway. If the sum of squares for the confounded interaction is needed, the somewhat laborious method given above is always available.

Mixed confounding, in which some factors are at two levels and some at three, is decidedly awkward and is better avoided. It is, however, possible to use the method illustrated at (7.8.3). There the interaction of three of t two-level factors was confounded, and each resulting treatment was associated with all combinations from the other factors. There is no requirement that those other factors shall all be at two levels; the method applies however many levels they have.

7.12 Confounding a 4^t design

Given factors each at four levels, confounding is not difficult provided all factors are quantitative with levels equally spaced. The method is to associate each level with a treatment from a 2^2 factorial set. To take an example, we will suppose that factor A has levels 0, 1, 2 and 3. We will associate them thus with factors U and V:

0	1	2	3
(1)	V	UV	U

The contrasts that ordinarily arise from U and V are:

Main effect of U	(−1	−1	+1	+1)
Main effect of V	(−1	+1	+1	−1)
Interaction	(+1	−1	+1	−1)

The main effect of U is effectively a comparison of the two upper levels of A, namely, 2 and 3, compared with the lower levels, namely, 0 and 1. As such it does not pick up the linear effect as effectively as the more usual (−3 −1 +1 +3), but it will serve the same general purpose. The main effect of V is exactly the contrast used at (5.7.4) to show the quadratic effect, which leaves U × V. In so far as it represents an inflexion, it could serve much the same purpose as a cubic effect. In fact, it is more usually confounded. Even if there were only the one factor, confounding U × V would reduce block size from four to two. That is not usual, but if there were two factors, A and B, at four levels, they could be represented respectively by U and V, and by W and X. It could be convenient to confound U × V or W × X or U × V × W × X. Indeed, all three could be confounded at the same time to give blocks of four plots, since any one of them is the generalized interaction of the other two. Another scheme might be to confound only the four-factor interaction, thus using blocks of eight plots and leaving U × V and W × X to give warning of any cubic effects. Also, there is no limit to the number of four-level factors that can be dealt with in this way.

Mixed confounding with factors at three and four levels is particularly awkward, but with factors, some with two levels and some with four, it is quite easy. An experiment with h factors at two levels and k factors with four is readily transformed to one with $(h + 2k)$ factors, each at two levels. Further, as with the 4^k design, there are a number of interactions that can readily be dispensed with.

Given an interest in quadratic effects, which with a 4^k design are estimated without loss of efficiency, as are their interactions, it can be argued that four levels are better than three. An objection comes from the estimation of the linear effects being less than ideal (its efficiency is 0.8, equivalent to the loss of 1/5 of its replication) but that may not matter if, as is usually the case, the linear effects are large compared with the quadratic.

7.13 Total residuals

Whenever we work in several strata—and this happens consistently with split plots and designs in which an interaction is confounded—we need to think carefully about residuals. It follows from what was said in Section 1.9 that each plot will have a residual in each stratum. To anyone who is trying to find out why a certain 'error' sum of squares was larger than expected, this is a convenience. He has to look at the residuals in that stratum to see if any are unduly large. The situation is different if the residuals are being examined to find any features of the land that should be allowed for on another occasion. For that purpose 'total residuals' are more useful, being simply

Datum − treatment mean.

It does not matter what contrasts are confounded, whether main effects or interactions. Neither does it matter how blocks were formed: What is required is a measure of the inherent fertility of the plot. The experiment that happens currently to be on the site and the design adopted to implement it are irrelevant for that purpose. Further, the treatment whose mean is required results perhaps from a combination of many factors, e.g. *abcde*.

Where replication is low, no residuals are going to be of much use because they are calculated under the constraint that they must sum to zero over each treatment. Hence, if there are only two replicates, each residual, p, must have a counterpart, $-p$, at the other plot with the same treatment. Clearly p measures local peculiarities at two locations and, if it is large, there is no way of saying which location brought that about. (This was one reason for not considering residuals in Sections 4.9 and 4.10. Even with three replications residuals are difficult to interpret.)

Where there is only one replication, as with the designs considered in Section 7.9, the calculation of residuals becomes very complicated.* (The 'error' is made up from a number of interactions.) The situation is no less complicated with fractional replication. Some computer packages find the residuals by a method that is completely sound but not of a kind to be attempted by anyone without suitable software. What they do is this: The program evaluates all the contrasts that do not belong to 'error'. Then it takes the deviations, which are found in the usual way, and adjusts each by the values of the relevant contrasts. As we have said, the method gives correct values for the residuals, but the calculations are not of a sort to be undertaken lightly, and they will not be described here.

Exercise 7A

Five varieties of spring wheat were sown in a randomized blocks design

*But for 2^k experiments the Yates table facilitates it.

in four blocks. The soil was treated with three different levels of nitrogen randomly allocated to equal areas within each plot. The design and yields in t/ha were as follows:

Block I	V2			V5			V1			V4			V3		
	N1	N3	N2	N2	N3	N1	N1	N2	N3	N1	N3	N2	N2	N1	N3
	4.6	5.5	5.3	5.0	5.4	4.7	5.5	6.1	6.4	5.0	6.0	5.7	5.5	4.9	5.8
Block II	V1			V3			V2			V5			V4		
	N3	N1	N2	N1	N3	N2	N3	N2	N1	N2	N3	N1	N2	N1	N3
	5.8	5.0	5.5	4.9	5.5	5.4	5.4	5.0	4.7	4.6	5.0	4.2	6.2	5.7	6.5
Block III	V5			V1			V3			V2			V4		
	N2	N3	N1	N2	N3	N1	N3	N1	N2	N1	N3	N2	N1	N3	N2
	4.8	5.0	4.6	5.4	5.9	5.0	5.5	4.8	4.7	5.0	5.8	5.1	5.3	6.7	5.8
Block IV	V2			V3			V4			V1			V5		
	N3	N1	N2	N3	N2	N1	N2	N1	N3	N1	N2	N3	N3	N2	N1
	5.9	5.0	5.6	4.8	4.6	4.0	5.1	4.7	5.4	5.2	5.5	5.8	5.2	4.8	4.4

Analyse and give in outline a report on the results.

Exercise 7B

An experiment was conducted on ways of protecting oats against a pest. On the main plots there were two treatments: seeds infected and not infected. On the sub-plots there were three seed protectants (C = Ceresan M, P = Panogen, A = Agrox) in comparison with an untreated control 0. The data, which represent yields in bushels per acre, were as follows:

Seed	Protectant	Blocks			
		I	II	III	IV
Infected	O	42.9	41.6	28.9	30.8
	C	53.8	58.5	43.9	46.3
	P	49.5	53.8	40.7	39.4
	A	44.4	41.8	28.3	34.7
		190.6	195.7	141.8	151.2
Not infected	O	53.3	69.6	45.4	35.1
	C	57.6	69.6	42.4	51.9
	P	59.8	65.8	41.4	45.4
	A	64.1	57.4	44.1	51.6
		234.8	262.4	173.3	184.0

Analyse and outline a report on the results.
1 bushel = 36.4 litres 1 pound = 454 grams.

[Data from R. G. D. Steel and J. H. Torrie, *Principles and Procedures of Statistics: a Biometrical Approach*, 2nd edn, p. 384.]

Exercise 7C

An experiment on the yield of grass was conducted with five cutting treatments A–E, arranged in a Latin square. With treatments A–C crops of hay were taken on 24th June, 15th July and 31st October. With A, crops had been taken on three previous occasions, with B on two and with C on one. With treatments D and E, crops were taken with relation to the emergence of ears; with D at emergence and 28 and 70 days afterwards; and with E at 28 and 84 days after emergence. Each plot was divided into four sub-plots and four grass mixtures W, X, Y and Z were assigned at random within each plot. Data, which represent weight of dry matter, were as follows:

CW 648	BW 453	EW1032	DW 562	AW 452
X 532	X 463	X 933	X 540	X 427
Y 323	Y 294	Y1215	Y 407	Y 439
Z 434	Z 309	Z 827	Z 511	Z 449
DW 392	EW 781	AW 739	CW 630	BW 624
X 528	X 759	X 826	X 568	X 490
Y 493	Y 588	Y 632	Y 567	Y 618
Z 299	Z 890	Z 550	Z 523	Z 508
AW 489	CW 499	DW 529	BW 456	EW1204
X 620	X 551	X 673	X 366	X 958
Y 400	Y 428	Y 422	Y 554	Y 967
Z 476	Z 294	Z 676	Z 510	Z 950

BW 610	AW 378	CW 759	EW 826	DW 432
X 976	X 436	X 734	X1104	X 677
Y 509	Y 354	Y 457	Y 866	Y 383
Z 457	Z 336	Z 602	Z1380	Z 449

EW1036	DW 289	BW 626	AW 911	CW 596
X 765	X 511	X 445	X 918	X1020
Y 474	Y 339	Y 601	Y 704	Y 438
Z 704	Z 256	Z 466	Z 551	Z 632

Work out an analysis of variance and give an outline of its interpretation.

[Data from S. C. Pearce, *Biological Statistics: an Introduction*, p. 134.]

Exercise 7D

An experiment was conducted on oats in six randomized blocks. Each plot of 1/80 acre was sown with one of three varieties X, Y or Z and then divided into quarters for levels of nitrogen. (It may be assumed that the four levels were equally spaced.) The data, which represent yields in quarter pounds, were as follows:

I	Z	N3	156	N2	118	Z	N2	109	N3	99	II
	Z	N1	140	N0	115	Z	N0	63	N1	70	
	X	N0	111	N1	130	Y	N0	80	N2	94	
	X	N3	174	N2	157	Y	N3	126	N1	82	
	Y	N0	117	N1	114	X	N1	90	N2	100	
	Y	N2	161	N3	141	X	N3	116	N0	62	
III	Z	N2	104	N0	70	Y	N3	96	N0	60	IV
	Z	N1	89	N3	117	Y	N2	89	N1	102	
	X	N3	122	N0	74	X	N2	112	N3	86	
	X	N1	89	N2	81	X	N0	68	N1	64	
	Y	N1	103	N0	64	Z	N2	132	N3	124	
	Y	N2	132	N3	133	Z	N1	129	N0	89	
V	Y	N1	108	N2	126	X	N2	118	N0	53	VI
	Y	N3	149	N0	70	X	N3	113	N1	74	
	Z	N3	144	N1	124	Y	N3	104	N2	86	
	Z	N2	121	N0	96	Y	N0	89	N1	82	
	X	N0	61	N3	100	Z	N0	97	N1	99	
	X	N1	91	N2	97	Z	N2	119	N3	121	

(one acre = 4047 m^2 one pound = 454 g)

Using summation terms, analyse the data, then partition the lines for the main effect of nitrogen and the interaction through the method of contrasts.

[Data from F. Yates, 'Complex experiments', *J. Royal Statist. Soc.*, (1937), Supplement **2**, p. 198.]

Exercise 7E

The following data come from an experiment on peas with eight treatments, formed by factorial combinations of presence or absence of nitrogen (N), phosphorus (P) and potassium (K). There were six blocks, each with four plots, and the data represent yields in pounds per plot. Each plot had an area of 1/70 acre.

I	pk	49.5	(1)	46.8	n	62.0	k	45.5	II
	np	62.8	nk	57.0	npk	48.8	p	44.2	
III	n	59.8	k	55.5	np	52.0	nk	49.8	IV
	npk	58.5	p	56.0	(1)	51.5	pk	48.8	
V	p	62.8	n	69.5	nk	57.2	pk	53.2	VI
	npk	55.8	k	55.0	np	59.0	(1)	56.0	

(1 acre = 4047 m^2 1 pound = 454 grams)

Obtain the appropriate analysis of variance and write notes on the interpretation.

[Data from F. Yates, 'Complex experiments', Supplement to *J. Roy. Statist. Soc.*, **2** (1935), p. 204.]

Exercise 7F

(a) Divide a 2^4 experiment into 4 blocks so as to confound the ABD and BC contrasts.

(b) Divide a 2^5 experiment into 4 blocks so as to confound the ABE and BCDE contrasts.

(c) Divide a 2^5 experiment into 8 blocks so as to confound the ABE, BCDE and ACDE contrasts.

In each case write down the other contrasts which are confounded.

Chapter 8

Correlation, regression and the analysis of covariance

8.1 Correlation

Two measurements have been taken on a number of units. The question may arise whether they are associated or not: it is on such occasions that correlation coefficients can be useful.

To take an example, an experimenter surveys crops at a number of sites and records the yield per square metre at each. It also occurs to him that the differences in yield may be due to differences in soil pH, so he measures that as well. Calling the yields y and pH x, he has ten pairs of figures, like this:

x	6.7	6.6	5.9	7.2	5.7	7.0	7.1	6.9	6.5	6.8
y	13.1	14.6	11.8	16.4	10.9	19.6	17.3	15.5	12.3	18.9

If these are plotted in the usual way, the diagram is as at Fig. 8.1. The dotted lines mark the means of x and y, namely 6.64 and 15.04 respectively. They divide the diagram into four quadrants. It will be seen that nine of the ten pairs of observations lie either in the bottom left-hand quadrant, in which both x and y are below their means, or in the top right-hand one, in which both x and y exceed their means. That is strong evidence that the two are associated. If x and y were distributed independently of one another, the points could be expected to lie about equally in the four quadrants.

The argument is made more precise if actual positions on the diagram are taken into account, not just the quadrants. For example, the last point ($x = 6.8$, $y = 18.9$), where both x and y lie clear of the dotted lines, provides stronger evidence of an association than the first ($x = 6.7$, $y = 13.1$), where neither variable is far from its mean. We shall therefore

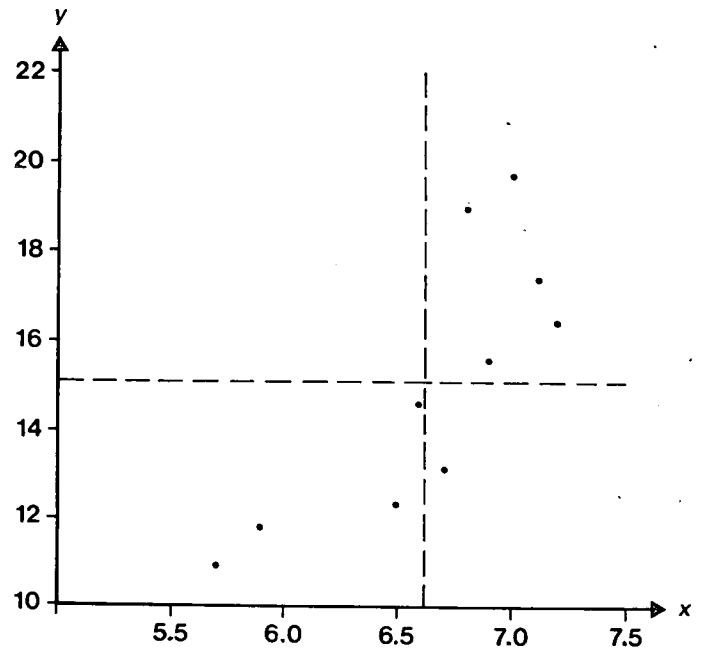


Fig. 8.1 **Yields (y) of survey sites plotted against soil pH (x)**

sweep both x and y by their means, i.e.,

x	+0.06	−0.04	−0.74	+0.56	−0.94	+0.36	+0.46	+0.26	−0.14	+0.16
y	−1.94	−0.44	−3.24	+1.36	−4.14	+4.56	+2.26	+0.46	−2.74	+3.86

The sum of squares for x and y are found in the usual way by squaring residuals—there are no treatments, so the deviations and the residuals are the same—and adding. Calling them E_{xx} and E_{yy},

$$E_{xx} = (+0.06)^2 + (-0.04)^2 + \ldots + (+0.16)^2 = 2.2040$$
$$E_{yy} = (-1.94)^2 + (-0.44)^2 + \ldots + (+3.86)^2 = 81.9640.$$

Both quantities could have been obtained using summation terms, as described in Section 1.4, namely,

$$E_{xx} = (6.7^2 + 6.6^2 + \ldots + 6.8^2) - (66.4)^2/10 = 2.2040$$

$$E_{yy} = (13.1^2 + 14.6^2 + \ldots 18.9^2 - (150.4)^2/10 = 81.9640.$$

So far we have looked at the variability of the two quantities taken separately. To examine how they vary together, we need to multiply them together instead of squaring each. For example, to take the first site, it is the only one where the residuals are of opposite sign. Consequently it will make a negative contribution to a sum of products, namely, $(+0.06) \times (-1.94) = (-0.1164)$. The rest, on the other hand, will all make positive contributions. Furthermore, those where both residuals are large will make a larger contribution than those where one residual (or both) is small. The next step is to work out E_{xy}, which is to be the sum of products of residuals, thus:

$$E_{xy} = (+0.06)(-1.94) + (-0.04)(-0.44) + \ldots + (0.16)(+3.86)$$
$$= +10.7540.$$

The same quantity could have been obtained from summation terms, thus:

$$E_{xy} = (6.7 \times 13.1) + (6.6 \times 14.6) + \ldots + (6.8 \times 18.9) - (66.4 \times 150.4)/10$$
$$= +10.7540.$$

It is important that E_{xy} and quantities derived from it should always be given their signs. Unlike E_{xx} and E_{yy}, which are necessarily positive, E_{xy} could be negative and that would be important.

However, the immediate problem is the measurement of correlation. The basic value is E_{xy}, but it needs to be scaled. As it stands, if y were measured in some other units, e.g. kilograms instead of pounds, it would be multiplied by a suitable constant. In that case E_{yy} would be multiplied by the same constant squared.

To avoid difficulties of scaling, the correlation coefficient, r, is taken to be

$$r = \frac{E_{xy}}{\sqrt{E_{xx}E_{yy}}} \qquad (8.1.1)$$

In the example, $r = +0.800$, which is high. In general, r varies between -1 and $+1$. The extreme values, ± 1, indicate a strict straight-line relationship with no extraneous variation, the slope of the line being upwards or downwards depending on the sign of E_{xy}, whether positive or negative. If $r = 0$, this can imply that there is no association, though other interpretations are possible. Other values indicate some association, either

positive or negative, but not as close as that implied by $r = \pm 1$.

Some care is needed in the interpretation of correlation coefficients. For example, they do not show causation. The data given above do indeed suggest that the species does not like acid soil, but in this particular area it could be that acid soils are mostly shallow. In that event the real interpretation might be that the species does not like shallow soil. (It might even prefer a site of low pH, other things being equal, but other things are not equal.)

It should also be noted that correlation coefficients are based on departures from a steady increase or decrease in y as x changes—in other words, on the idea of a straight-line ('linear') relationship. If however, the relationship is markedly curved, that will lead to a reduction in the value or r. The following is an extreme case:

x	10.9	12.0	13.7	14.8	13.2
y	24.1	20.9	21.1	24.4	19.8

The correlation coefficient, r, is virtually zero, but plotting the points shows that there is in fact a clear relationship between x and y, though it is not given by a straight line.

Many tables have been produced to show whether a given value of r does or does not differ from zero. They are entered with one degree of freedom fewer than those of E_{xx} and E_{yy}. Thus the value of $+0.800$ found above can be tested with eight degrees of freedom. It lies outside the limits of ± 0.765 given in the table for $P = 0.01$ and is therefore unlikely to have arisen by chance from a population in which x and y were in fact uncorrelated. Another method of assessing the relationship will be given in the next section.

8.2 Linear regression

The previous section dealt with correlation, which is the extent to which two quantities depend upon one another for their value. Nevertheless, to know, as in the example of the soil acidity and the crop yields, that there is a significant correlation coefficient is to have evidence only that there is a relationship of some sort with a component of linearity. It does not say exactly what that relationship is.

The simplest case is that in which it is assumed to be a straight line, i.e.

$$y = a + bx + e \qquad (8.2.1)$$

where a and b are constants and e is a residual. This equation is said to give the 'linear regression of y on x', and b is called the 'regression

coefficient'. The value of b indicates the increase (if positive) or the decrease (if negative) in y that goes with one unit increase in x. If (8.2.1) can be taken as a reasonable representation of the relationship, then $\hat{b}$, the best estimate of b, is E_{xy}/E_{xx}. In the example, where $E_{xx} = 2.2040$ and $E_{xy} = +10.7540$, $\hat{b}$ is $+4.88$. That tells us that if there is an increase of 1.0 in pH, we could expect on average an increase of 4.88 units in yield.

Several points need to be noted. As has already been said, causation should not be assumed. To follow up a suggestion already made, there could be a chance relationship, valid only for that locality, between, say, pH and soil depth. Nevertheless for that locality the relationship given at (8.2.1) with b estimated as $+4.88$ is genuine, though it might not hold elsewhere. That is the observed relationship of yield to pH for those sites.

Another point concerns the inverse relationship, which arises when x is estimated from y. The regression coefficient of x on y is $E_{xy}/E_{yy} = +0.1312$. Some would expect the inverse coefficient to be $1/4.88 = 0.2049$, but this would be a mistake. Supposing that there had been no relationship then both regression coefficients would have been zero. In general their product (E_{xy}/E_{xx} multiplied by E_{xy}/E_{yy}) equals r^2, not 1. In this instance $4.88 \times 0.1312 = 0.640 = (+0.800)^2$.

A caution concerns the form of (8.2.1), which implies that plotting y against x will give a straight line. If the line is in fact curved, the approximation will be a bad one. Further, a lot of variation will be ascribed to the random residuals, e, when it is really due to failure of the assumption of linearity.

Given a solution for b, it becomes possible to work out a revised variable, y', in which the disturbing effect of the variable x has been removed.

With a knowledge of the regression coefficient, b, we can estimate what the values of y would have been for any chosen value of x, but which value should we choose? The experimenter may resolve this question by saying that he wants to know y for that range of sites, and the mean value of x is 6.64, so that is what he wants. For the first site, where $x = 6.7$ and $y = 13.1$, y should be adjusted to $13.1 - 4.88(6.7 - 6.64) = 12.8 = y'$ say. For all the sites the y'-values are:

x	6.7	6.6	5.9	7.2	5.7	7.0	7.1	6.9	6.5	6.8
y	13.1	14.6	11.8	16.4	10.9	19.6	17.3	15.5	12.3	18.9
y'	12.8	14.8	15.4	13.7	15.5	17.8	15.1	14.2	13.0	18.1

It will be seen that the mean of the y' is the same as that of the y. (This is because x has been adjusted to its mean.) Also, the sum of squared deviations for y' equals

$$E_{yy}(1 - r^2) = E_{yy} - E_{xy}^2/E_{xx} \qquad (8.2.2)$$

Here it is $81.9640\ (1 - 0.6400) = 29.5070$. That can readily be confirmed from the summation terms, because

$$(12.8^2 + 14.8^2 + \ldots + 18.1^2) - (150.4)^2/10 = 29.0640.$$

(The discrepancy arises from rounding errors. Actually, in this instance 29.5070 is to be preferred because it comes directly from the original data, whereas 29.0640 is derived from the data by a train of calculations, each subject to approximation.) As to degrees of freedom E_{yy} had nine. In passing to $E_{yy}(1 - r^2)$ another quantity had to be estimated and allowed for, namely b, so there remain only eight.

However, the experimenter might have made a different response to our query about the value of x that he wanted. He might have replied that he wanted a figure for neutral soil, i.e. $x = 7.0$. In that case y' gives values of

14.6 16.6 17.2 15.4 17.2 19.6 16.8 16.0 14.7 19.9.

The change in the standard value of x from 6.64 to 7.00 has led to a corresponding change in the mean value of y' amounting to $+1.76 = +4.88\ (7.00 - 6.64)$, but the sum of squares is unchanged, being

$$(14.6^2 + 16.6^2 + \ldots + 19.9^2) - (168.0)^2/10 = 29.6600.$$

However, differences between sites will remain the same whatever value of x is adopted. Thus, to compare the first two sites, $14.6 - 16.6 = -2.00 = 12.8 - 14.8$.

Another important consideration is the precision with which $\hat{b}$ estimates the true value of b. The variance of $\hat{b}$ is in fact

$$\frac{\text{Variance of } y'}{E_{xx}} \qquad (8.2.3)$$

In the present example, the variance of y' is $29.5070/8 = 3.6884$ and $E_{xx} = 2.2040$, so the variance of $\hat{b}$ is 1.6735 and its standard error is $1.294 = \sqrt{1.6735}$ with eight degrees of freedom. The estimated value (4.88) therefore differs from zero by 3.77 times its standard error (1.294).

Another matter is the significance of the departure of $\hat{b}$ from zero, which is the same as the significance of r. If the regression on x is not allowed for, the sum of squares for y is E_{yy}. In the present example, it is 81.9640 with nine degrees of freedom. If the regression is allowed for then the sum of squares is $E_{yy}(1 - r^2) = 29.5070$ with one degree of freedom fewer. That leads to an analysis of variance like this:

Source	d.f.	s.s.	m.s.	F
Regression	1	52.4570	52.4570	14.22***
'Error'	8	29.5070	3.6884	
Total	9	81.9640		

The significance lies rather beyond the level, $P = 0.001$. Actually, the test is readily thought of in algebraic terms. If the degrees of freedom for E_{yy} number f, then

$$F = \frac{(f-1)r^2}{1-r^2} \tag{8.2.4}$$

with 1 and $(f-1)$ degrees of freedom. Here, where $f = 9$ and $r = +0.800$, $F = 14.22$ as before. Further, $3.77^2 = 14.21$, so the test used below (8.2.2) has given the same result (see Section 3.10).

The simple relationship used at (8.2.1) does not necessarily hold, but in many cases a simple tranformation of the variables will justify it. For example, let

$$Y = pX^q$$

be the underlying rule. Transforming X and Y to their logarithms, x and y, gives

$$y = \log p + qx. \tag{8.2.5}$$

That is the same as (8.2.1) with $\log p$ for a and q for b. It could be asked what has happened to the residuals e. In fact, if a random quantity, e, is added to the right-hand side of (8.2.5) it will often be found that it has the properties expected of a residual. That is because a transformation will often change the quantity actually measured, chosen perhaps for practical convenience, into another quantity that is botanically more fundamental. However, this is a large subject and one that will be dealt with more fully in Chapter 9.

The assumptions underlying linear regression can usefully be examined. First of all, it is taken for granted that there is in fact a straight-line relationship between x and y. If the relationship is really curvilinear, the fact will appear from examination of the estimated residuals, e, which will no longer appear to be in random order as x increases. Instead, groups of positive and negative values will be found. It is also assumed that the

residuals are distributed normally about zero. This also may be called into question if the line is not straight. Nevertheless, as a first approximation people do often fit straight lines where a curve would be better. Incidentally, no assumptions are required about the distribution of x. Sometimes it is convenient to use a variable that takes only a limited number of values, e.g. $x = 0$ for male, $x = 1$ for female, and there is no objection to that.

A final point concerns the interpretation of a regression coefficient. It should never be cited as if it represented a law of Nature. The reason is that two quantities, x and y, can often be related by several mechanisms. For example, if x is the height of a plant and y is its number of seed pods, what is the regression of y on x? It all depends on the reason for x being variable. If some plants are in shade and others in full sun, it is possible that the shaded ones will grow higher to find the light and, in consequence, have less resources for reproductive activities, i.e. the regression will be negative. On the other hand, if some plants grow more because they are better fertilized, they could well have more seed pods, i.e. the regression is positive. In many instances both effects will operate and the regression coefficient—and also the correlation coefficient—will be positive or negative according to which dominates. Hence a regression coefficient once found can be taken as summing up the situation for those particular conditions, but it will not necessarily apply in other conditions, where the relative importance of the two mechanisms could quite possibly be different.

8.3 Partial and multiple linear regression

By comparison with (8.2.1) y may depend upon two variable quantities, w and x, i.e.

$$y = a + bx + cw + e. \tag{8.3.1}$$

The determination of b and c is now more complicated than that of b in the last section, because each of x and w can affect y indirectly through the other as well as directly. For example, yield (y) could be affected by depth of topsoil (x) and the amount of fertilizer applied (w). If w is disregarded, then $b = E_{xy}/E_{xx}$ as before; if x were disregarded c would equal E_{wy}/E_{ww}. The same equations would hold if x and w were independent of one another, but suppose that someone has been compensating for shallowness of soil by applying more fertilizer, i.e. there is a negative correlation between x and w. Then if x has a high value, this will affect y directly, but it means that w will have to be low, and this also will affect y. (That is the indirect effect of x on y.)

To cope with these complications we will amend the notation. In (8.2.1) we shall in future write b as $b_{y.x}$, i.e. the regression coefficient of y on x. In (8.3.1) we shall write b as $b_{yx.w}$, i.e. the regression coefficient of y on x, w being held constant. Similarly c will be written as $b_{yw.x}$. In that way x and w can be varied independently. In short (8.3.1) will become

$$y = a + b_{yw.x}w + b_{yx.w}x + e. \tag{8.3.2}$$

Further it can be shown that

$$\hat{b}_{yw.x} = \frac{E_{wy}E_{xx} - E_{wx}E_{xy}}{E_{ww}E_{xx} - E_{wx}^2} \tag{8.3.3}$$

$$\hat{b}_{yx.w} = \frac{E_{ww}E_{xy} - E_{wx}E_{wy}}{E_{ww}E_{xx} - E_{wx}^2}$$

It will be recalled that a circumflex accent above b shows that an estimate has been made of the true value, which is not known.

The sum of squares for the regression of y on both w and x takes up two degrees of freedom. It equals

$$\hat{b}_{yw.x}E_{wy} + \hat{b}_{yx.w}E_{xy}$$

leaving as 'error',

$$E_{yy} - \hat{b}_{yw.x}E_{wy} - \hat{b}_{yx.w}E_{xy} \tag{8.3.4}$$

analogous to (8.2.2). This quantity is sometimes written

$$E_{yy}(1 - R^2) \tag{8.3.5}$$

where R is called the 'multiple' correlation coefficient of y on both w and x. Again there is a parallel in (8.2.2).

The following example shows what may happen. Some bushes were planted fairly closely so that they had little opportunity to spread, though they were of course free to grow vertically. After a time their heights and spreads in metres and their crop yields in kilograms were:

		(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)
Height	(h)	2.4	2.6	2.3	2.4	2.6	2.3	2.1	2.0
Spread	(s)	1.6	1.7	1.4	1.5	1.6	1.5	1.4	1.5
Yield	(y)	41	47	39	42	44	40	38	36

As a guide to the future someone wants to know if it would be possible to use h and s as a means of crop forecasting. He points out that h is difficult to measure and is therefore to be avoided if possible, whereas s is readily found. The first task is to work out the sums of squares and products in the usual way.

$E_{hh} = 0.319$ $E_{ss} = 0.075$ $E_{yy} = 84.88$
$E_{sy} = +2.02$ $E_{hy} = +4.94$ $E_{hs} = +0.112.$

Hence from (8.3.2)

$$b_{yh.s} = +12.68 \qquad b_{ys.h} = +8.01.$$

Also, using (8.3.4), the sum of squares with two degrees of freedom, for the double regression, is

$$(+12.68)(+4.94) + (+8.01)(+2.02) = 78.82.$$

That leaves an 'error' sum of squares of 6.06 (= 84.88 − 78.82).

This does not end the matter because the enquiry concerns the possibility of using only one of h and s. If we had regressed y on h alone, this would have given a sum of squares of $E_{yh}^2/E_{hh} = 76.47$. That leaves 2.35 (= 78.82 − 76.47) for any added benefit from knowing s as well. The analysis of variance is therefore

Source	d.f.	s.s.	m.s.	F
Regression on h alone	1	76.47	76.47	63.09***
Gain from using s also	1	2.35	2.35	1.94
'Error'	5	6.06	1.212	
Stratum total	7	84.88		

It is clear that the variation in h has explained most of the variation in y, so h could be used to forecast y. Further, once h is known, there is little advantage in knowing s as well.

As a matter of nomenclature, the first line, corresponding to b_{yh}, is called the 'total regression' of y on h. The second, corresponding to $b_{ys.h}$, is called the 'partial regression' of y on s, h having been allowed for.

The enquirer hoped to be able to use s alone. If he had done so, it would have accounted for a sum of squares of 54.40 ($= E_{sy}^2/E_{ss}$) leaving 24.42

(= 78.82 − 54.40) for the added effect of knowing h as well. The analysis of variance is

Source	d.f.	s.s.	m.s.	F
Regression on s alone	1	54.40	54.40	44.88***
Gain from using h also	1	24.42	24.42	20.15***
'Error'	5	6.06	1.212	
Stratum total	7	84.88		

It appears then that s does provide quite a lot of information about y, but clearly it benefits from supplementation by h, which completes the story. The enquirer must therefore be told that, reluctant though he may be to measure h, this really is required if he is to make a good forecast.

8.4 Curvilinear regression

An ability to use two regressor variables, w and x, instead of only one, x, makes possible the fitting of simple curves. For example, if w is put equal to x^2, (8.3.1) becomes

$$y = a + bx + cx^2 + e \qquad (8.4.1)$$

Here a little caution is necessary. It is conceivable in biology that the relationship between x and y should effectively be a straight line, at least over a limited range, but parabolae like that at (8.4.1), though common enough in physics, are unusual in plant physiology. Going from (8.3.1) to (8.4.1) the most that can justifiably be said is that a significant value for c shows that the relationship is not straight. When that is so, the response curve is better left undecided unless some more thorough study can be undertaken.

If the sole intention is to detect any deviations from linearity, some have thought it better to replace (8.4.1) by

$$y = a + bx + \sqrt{x} + e \qquad (8.4.2)$$

Some people have a reservation about (8.4.1), their reason being the shape of the curve when x^2 is graphed against x. It starts fairly flat, but begins to rise sharply as x increases; it is, in short, rather abrupt and not at all sinuous. For that reason they prefer (8.4.2) as being more likely to fit

the facts. In the main (8.4.2) is quite successful, though (8.4.1) is more usual.

For purposes of illustration we shall use (8.4.1) to try to detect any deviations from the straight-line relationship given at (8.2.1). For that we use the data

$w = x^2$	44.89	43.56	34.81	51.84	32.49	49.00	50.41	47.61	42.25	46.24
x	6.7	6.6	5.9	7.2	5.7	7.0	7.1	6.9	6.5	6.8
y	13.1	14.6	11.8	16.4	10.9	19.6	17.3	15.5	12.3	18.9

Hence

$$E_{ww} = 365.63 \quad E_{xx} = 2.2040 \quad E_{yy} = 81.9640$$
$$E_{xy} = +10.7540 \quad E_{wy} = +138.945 \quad E_{wx} = +28.372$$

and

$$b_{yw.x} = \frac{+1.1223}{0.8781} = +1.278$$

$$b_{yx.w} = \frac{-10.1625}{0.8781} = -11.573$$

From (8.3.4) the sum of squares due to both w and x is equal to 53.1157. This compares with the figure for x alone of 52.4721, leaving only 0.6436 for the added effect of w, which is clearly not significant. The analysis of variance is

Source	d.f.	s.s.	m.s.	F
Regression on x alone	1	52.4721	52.4721	12.73**
Gain from using x^2 also	1	0.6436	0.6436	0.16
'Error'	7	28.8483	4.1212	
Stratum total	9	81.9640		

In fact, the curvilinear component has contributed so little that it has actually led to an increase in the 'error' mean square. Even that does not prove that the relationship of y and x is straight, though one can say without reservation that the present data show no evidence of curvature. However, over the range of x used, x^2 and x are so closely correlated ($r = +28.372/\sqrt{365.63 \times 2.2040} = +0.999$) that it would in any event be

nearly impossible to distinguish their effects apart from one another. The calculations have nevertheless illustrated the method.

8.5 The analysis of covariance—principles

In the analysis of covariance there is a variate, y, that is to be subjected to the analysis of variance, but there are fears that it may be unduly variable on account of its association with x, another variate, which is not for the moment of interest on its own account but could be important as explaining part of the 'error' variance in y. The technique can best be explained by an example.

The following data were presented by Rayner (1969, p. 407). An experiment had been conducted on maize, using five randomized blocks with five treatments, A, B, C, D, and E. The y-variate was pounds per morgen, but it so happened that the same plots had been recorded in the previous year before the treatments had been applied. Those figures will constitute the x-variate. They can be expected to indicate good and bad patches of soil. It is important to note that x could not have been affected by the treatments. If it had been, the whole of the following procedure would be invalid.

The data were as follows, the upper value giving y and the lower x:

	Block I		II		III		IV		V
B	37.1	D	42.7	A	37.1	B	40.4	A	37.3
	51.2		52.9		62.0		54.7		64.0
A	33.7	A	36.5	C	51.0	A	37.1	B	46.2
	53.0		62.8		62.8		63.5		69.5
D	34.3	E	51.3	D	45.8	C	50.2	E	47.0
	48.0		59.4		61.5		66.2		54.1
E	32.6	B	40.7	B	44.4	D	44.0	D	25.8
	44.8		60.9		59.9		62.2		47.9
C	33.7	C	42.7	E	56.5	E	50.4	C	49.0
	47.9		56.7		59.2		56.2		61.2

(8.5.1)

(1 pound = 454 g 1 morgen = 8565 m²)

The following calculations would not ordinarily be used in practice, though they could be and they do show the argument.

First both variates, x and y, are swept by blocks to find their deviations, thus:

B	+ 2.82	D	− 0.08	A	− 9.86	B	− 4.02	A	− 3.76
	+ 2.22		− 5.64		+ 0.92		− 5.86		+ 4.66
A	− 0.58	A	− 6.28	C	+ 4.04	A	− 7.32	B	+ 5.14
	+ 4.02		+ 4.26		+ 1.72		+ 2.94		+ 10.16
D	+ 0.02	E	+ 8.52	D	− 1.16	C	+ 5.78	E	+ 5.94
	− 0.98		+ 0.86		+ 0.42		+ 5.64		− 5.24
E	− 1.68	B	− 2.08	B	− 2.56	D	− 0.42	D	− 15.26
	− 4.18		+ 2.36		− 1.18		+ 1.64		− 11.41
C	− 0.58	C	− 0.08	E	+ 9.54	E	+ 5.98	C	+ 7.94
	− 1.08		− 1.84		− 1.88		− 4.36		+ 1.86

(8.5.2)

These figures lead to the residuals in the usual way, i.e. sweeping by treatments, to give

B	+ 2.96	D	+ 3.30	A	− 4.30	B	− 3.88	A	+ 1.80
	+ 0.68		− 2.44		− 2.44		− 7.40		+ 1.30
A	+ 4.98	D	− 0.72	C	+ 0.62	A	− 1.76	B	+ 5.28
	+ 0.66		+ 0.90		+ 0.46		− 0.42		+ 8.62
D	+ 3.40	E	+ 2.86	D	+ 2.22	C	+ 2.36	E	+ 0.28
	+ 2.22		+ 3.82		+ 3.62		+ 4.38		− 2.28
E	− 7.34	B	− 1.94	B	− 2.42	D	− 2.96	D	− 11.88
	− 1.22		+ 0.82		− 2.72		+ 4.84		− 8.24
C	− 4.00	C	− 3.50	E	+ 3.88	E	+ 0.32	E	+ 4.52
	− 2.34		− 3.10		+ 1.08		− 1.40		+ 0.60

(8.5.3)

Of the 25 pairs of residuals, 19 have the same sign and only six are of opposite sign. That does imply that plots that were good or bad in one year behaved similarly in the year after.

We shall now assess the correlation between the two years' records. Summing squares and products of the residuals gives

$$E_{xx} = 321.22 \quad E_{xy} = +273.72 \quad E_{yy} = 420.66.$$

Hence $r = +0.745$ and $b_{yx} = +0.852$. That is to say, for every one pound of crop above expectation in the preliminary year, a plot had an excess of 0.852 pounds in the experiment itself. This knowledge will now be used to improve precision. In an ordinary analysis of y the 'error' sum of squares is 420.66 with 16 degrees of freedom, which gives an error variance of 26.29. Using the adjustment, from (8.2.2) the 'error' sum of squares is $420.66 - (273.72)^2/321.22 = 187.42$ with 15 degrees of freedom, giving an error variance of 12.49, which is much better.

To complete the analysis using this long method, it is necessary to take sums of squares and products of the deviations at (8.5.2) to produce D_{xx},

D_{xy} and D_{yy}, analogous to E_{xx}, E_{xy} and E_{yy}, which came from (8.5.3). They are:

$$D_{xx} = 492.47 \quad D_{xy} = 171.09 \quad D_{yy} = 851.11$$

all with 20 degrees of freedom. Adjusting y by x gives

$$851.11 - (+171.09)^2/492.47 = 791.67 \text{ with 19 degrees of freedom.}$$

In the adjusted analysis of variance the sum of squares for treatments can be found by subtracting 'error' from the stratum total.

Source	d.f.	s.s.	m.s.	F
Treatments	4	604.25	151.06	12.09***
'Error'	15	187.42	12.49	
Stratum total	19	791.67		

(8.5.4)

If y had not been adjusted, the F-value would have been only 4.09 with 4 and 16 degrees of freedom.

We have given residuals for x and y at (8.5.3), but we might reasonably want to know what the residual is after y has been adjusted by x. There is no problem about that. For the first plot, x and y residuals are respectively $+0.68$ and $+2.96$. Hence the residual of y' is $+2.96 - (+0.852)(+0.68) = +2.381$. All the other plots can be dealt with in the same way. When that is done, it will be found that the sum of the squared residuals is 187.43. Apart from a small effect of rounding, this is the 'error' sum of squares at (8.5.4).

8.6 The analysis of covariance in practice

The method just given is too long for most purposes if everything has to be done by hand, though it would serve very well for a computer program. Also, it is completely general and could be used in conjunction with the Kuiper–Corsten iteration set out in Section 4.2 because that gives both the deviations, as at (8.5.2), and the residuals, as at (8.5.3). After this the calculations can proceed as shown.

Given an orthogonal design, however, the calculations can be done more simply by using summation terms. Those for x^2 and y^2 are the same as for an analysis of variance for those two variates: those for xy are found by multiplying together corresponding values for the two variates instead of squaring each.

To take the total term first, for x^2 it equals

$$51.2^2 + 52.9^2 + \ldots + 61.2^2 = 84\,219.91,$$

and for y^2 it equals

$$37.1^2 + 42.7^2 + \ldots + 49.0^2 = 45\,198.85;$$

so for xy it equals

$$(51.2 \times 37.1) + (52.9 \times 42.7) + \ldots + (61.2)(49.0) = 61\,062.43. \qquad (8.6.1)$$

Similarly for the treatment term, totals are

	A	B	C	D	E
x	305.3	296.2	294.8	272.5	273.7
y	181.7	208.8	226.6	192.6	237.8

(8.6.2)

Hence the treatment terms are

x^2: $[305.3^2 + 296.2^2 + \ldots + 273.7^2]/5 = 83\,403.50$,
y^2: $[181.7^2 + 208.8^2 + \ldots + 237.8^2]/5 = 44\,320.70$
xy: $[(305.3 \times 181.7) + 296.2 \times 208.8) + \ldots + (273.7 \times 237.8)]/5 = 60\,338.12$

and so on for others.

	x^2	xy	y^2
Total	84 219.91	61 062.13	45 198.85
Blocks	83 727.44	60 891.34	44 347.74
Treatments	83 403.50	60 338.12	44 320.70
Corrections	83 232.25	60 440.75	43 890.25

Those terms lead to analyses of variance and covariance as follows:

Source	d.f.	x^2	xy	y^2
Treatments	4	171.25	− 102.63	430.45
'Error'	16	321.22	+ 273.72	420.66
Stratum total	20	492.47	+ 171.09	851.11

(8.6.3)

Total − Blocks

From here it is but a step to (8.5.4). As usual when summation terms have been used, there is an 'error' line which gives an aggregated figure for the action of residuals, but they are nowhere seen as individuals.

From now on we shall be chiefly interested in y after it has been adjusted by x. Treatment totals have already been given at (8.6.2); the means are derived from them, as follows:

	A	B	C	D	E
x	61.06	59.24	58.96	54.50	54.74
y	36.34	41.76	45.32	38.52	47.56

(8.6.4)

The x-means differ only as a result of the sort of variation included in 'error'. Treatments can have had no effect because the x-data were recorded before they were applied. It is therefore correct to adjust y by x using a regression coefficient derived from the 'error' line, namely $\hat{b}_{yx} = +273.72/321.22 = +0.852$.

There is an important point here. As was explained at the end of Section 8.2, a regression coefficient does not express some immutable law. The effect on y of a change in x depends upon the reason for x changing. If it is altering on account of 'error', there could be one regression coefficient; if on account of treatments, there could be another. That indeed is what has happened here. Calculated from the 'error' line the regression coefficient is $+0.852$, but from the treatment line it is -0.599 $(= -102.63/171.25)$. We are justified in using the former figure only if we can be assured that the differences between treatments with respect to their x-means were brought about by the action of 'error'. In this instance there is no room for doubt in the matter. First of all, in the analysis of variance for x^2, given at (8.6.3), the F-value is 2.13, which is not large enough to suggest that there was any effect due to the treatments. There is, however, a more powerful reason. Since the x-values were all measured before the treatments were applied, any differences must be due to 'error', so we were quite right to adopt $+0.852$ as the relevant regression coefficient.

Examination of the x-means at (8.6.4) shows that the treatments had not had equal good fortune from the randomization. Treatment A had in the main been assigned to good plots, while D and E had been rather unlucky. It is now proposed to use the regression coefficient to adjust all y-values to a standard value of x equal to the grand mean, i.e. 57.70. In the case of Treatment A this gives

$$36.34 + (57.70 - 61.06)(+0.852) = 33.48.$$

Hence the adjusted treatment means for y are

A	B	C	D	E
33.48	40.45	44.25	41.25	50.08

(8.6.5)

The mean of these figures is 41.90, the same as that for the unadjusted means at (8.6.4), but this is because the mean of the x-values is unchanged. If all the x-means had been standardized at some other value, the adjusted y-means would all have been raised or lowered by some constant. Consequently, they are not invariant, though differences between them will remain the same.

We shall now look at the standard error of some of those differences, taking as our example that between A and B. The difference is

$$[36.34 + 0.852(57.70 - 61.06)] - [41.76 + 0.852(57.70 - 59.24)]$$
$$= (36.34 - 41.76) + 0.852(-61.06 + 59.24). \qquad (8.6.6)$$

We shall look at the two terms separately.

The first $(36.34 - 41.76)$ is the difference of two unadjusted y-means, each based upon five data from an orthogonal design. Its variance is therefore

$$(1/5 + 1/5)(\text{Error variance}) = 0.4\ (\text{Error variance}).$$

Further, once we have allowed for the effect of x, the error variance in question is that at (8.5.4), namely 12.49. Its multiplier, here 0.4, is the constant, K, defined at (4.1.1) and considered further in Section 5.5.

The second term, $+0.852\ (-61.06 + 59.24)$, may be written $d\,\hat{b}_{yx}$, where d is the difference between the two x-means (in this instance $d = -1.82$). The variance of $\hat{b}_{yx}$ has been given at (8.2.3), so the variance of $d\,\hat{b}_{yx}$ is

$$\left(\frac{d^2}{E_{xx}}\right) \times (\text{Error variance}).$$

Taking the two terms together, the variance of the difference at (8.6.5) is

$$\left(\frac{1}{5} + \frac{1}{5} + \frac{1.82^2}{321.22}\right) \times 12.49$$

$$= 5.125 = 2.26^2.$$

The standard error is therefore 2.26. Since the difference in fact equals -6.97 $(= 33.48 - 40.45)$, it is 3.08 times its own standard error.

Someone may point out that d also is estimated subject to error. That is

true, but it does not affect the argument. We chose to adopt $d = -1.82$, and we could have chosen some other value if we had wished. The fact that the true difference may have been rather different from -1.82 does not affect the other fact that we opted for that figure.

A difficulty arises in practice because each treatment difference has its own d and therefore its own variance, namely

$$\left(K + \frac{d^2}{E_{xx}}\right) \times (\text{Error variance}) \qquad (8.6.7)$$

where K is the multiplier of the error mean square in ordinary analysis of variance for that design. (In the example $K = 0.4$.) It is here useful to note that if all possible values of d are taken, i.e.

A − B	+ 1.82	B − D	+ 4.74
A − C	+ 2.10	B − E	+ 4.50
A − D	+ 6.56	C − D	+ 4.46
A − E	+ 6.32	C − E	+ 4.22
B − C	+ 0.24	D − E	− 0.24

the mean of the squares will always equal

$$\frac{2(D_{xx} - E_{xx})}{v(v-1)}.$$

(It is assumed that the design is orthogonal with v treatments, all of which have the same replication.) Moreover, because F_x, the F-value for treatments in the analysis of variance for x, equals

$$\frac{f(D_{xx} - E_{xx})}{(v-1)E_{xx}}$$

where f is the number of degrees of freedom for 'error' in that analysis, a mean of the values at (8.6.7) is given by

$$\left(K + \frac{2F_x}{fr}\right) \times (\text{Error variance}) \qquad (8.6.8)$$

In this example $F_x = 2.13$, $f = 16$ and $r = 5$, so a mean variance from (8.6.8) is $0.4532 \times 12.49 = 5.661 = (2.38)^2$. The error variance was found from (8.5.4). Hence an average value for the standard error of differences at (8.6.5) is 2.38. That for the difference of A and B was only 2.06 because those treatments had much the same x-mean, as (8.6.4) shows.

Mostly it is enough to report the value given by (8.6.8), though the conscientious may like to add the maximum given by the largest value of d. (In the example this is 2.58, given by comparison of Treatments A and D). If for any particular contrast an exact figure is required, (8.6.7) is always available.

The above results can readily be generalized to other contrasts. Suppose, for example, that someone wanted to know about the extent to which Treatment A differed from the rest, i.e. he was interested in

$$(1 \quad -\tfrac{1}{4} \quad -\tfrac{1}{4} \quad -\tfrac{1}{4} \quad -\tfrac{1}{4}). \qquad (8.6.9)$$

He should first evaluate the contrast for both x and y using the means at (8.6.4):

$$c_x = +4.2 \qquad c_y = -6.95$$

Then, after adjustment this becomes

$$-6.95 + 0.852(0 - 4.2) = -10.53.$$

The same result could have been obtained from the means at (8.6.5). It will be seen that x has been adjusted to zero, that being its value if all the x means are standardized to a constant value.

For the contrast at (8.6.9), if the design is orthogonal and all treatments have five plots, then (from Section 5.5) $K = 0.25$. The adjustment adds c_x^2/E_{xx}, so the variance of the adjusted contrast is

$$\left(K + \frac{c_x^2}{E_{xx}}\right) \times (\text{Error variance}) \qquad (8.6.10)$$

That equals $3.808 = 1.95^2$. Hence the value of the contrast (10.53) is 5.40 times its standard error (1.95).

A similar conclusion could have been obtained from the analysis of variance at (8.6.3). The first task is to extract from the treatment line the component sum of squares or products for the contrast, i.e.

$$x^2,\ 70.56; \quad xy,\ -116.76; \quad y^2,\ 193.21.$$

Here K is taken to be 0.25, as found previously. (This leaves 100.69, + 14.13 and 273.24 respectively for the other three degrees of freedom.)

We can now construct a shorter form of (8.6.3), discarding any components not needed for the moment.

Source	d.f.	x^2	xy	y^2
Contrast	1	70.56	− 116.76	193.21
'Error'	16	321.22	+ 273.72	420.66
Total	17	391.78	+ 156.96	613.87

Adjusting the total line gives

$$613.87 - (156.96)^2/391.78 = 550.99$$

with 16 degrees of freedom. The adjusted line for error is known from (8.5.4), so the new analysis reads

Source	d.f.	s.s.	m.s.	F
Contrast	1	363.57	363.57	29.11***
'Error'	15	187.42	12.49	
Total	16	550.99		

It leads to exactly the same conclusions as the calculations at (8.6.10). With all single-degree effects, F in an analysis equals t^2 when standard errors are used instead (Section 3.10). This is true here ($29.11 = 5.40^2$ as nearly as rounding errors allow).

8.7 Double covariance

Sometimes it would be useful to be able to adjust y by two quantities, w and x. Although more calculations are involved, they are not more difficult. The analyses of variance and covariance have columns for w^2, wx, x^2, wy, xy, and y^2, six in all, compared with the three at (8.6.3). The error line gives E_{ww}, E_{wx}, . . ., E_{yy} and leads to $\hat{b}_{yw.x}$ and $\hat{b}_{yx.w}$ as at (8.3.3). From that point it is easy to find the 'error' sum of squares for y after adjustment by both w and x, the expression being that at (8.3.4).

Where a treatment contrast is to be tested, its line in the analyses of variance and covariance is merged with that for 'error' to give D_{ww}, D_{wx}, . . ., D_{yy}, and a second use of (8.3.4) will give the adjusted sum of squares for the contrast and 'error' combined. Essentially the method is that described above (8.5.4), the adjusted sum of squares for the contrast being

found by difference. Note that the error line has lost two degrees of freedom, not one as before.

The adjusted means for the treatments raise no problems because the partial regression coefficients of y on w and x are known ($\hat{b}_{yw.x}$ and $\hat{b}_{yx.w}$ above). If w needs to be adjusted by d_w and x by d_x, the mean value for y needs to be adjusted by

$$\hat{b}_{yw.x}d_w + \hat{b}_{yx.w}d_x \tag{8.7.1}$$

A mean so adjusted has a variance of

$$K + \frac{E_{xx}d_w^2 - 2E_{wx}d_wd_x + E_{ww}d_x^2}{E_{ww}E_{xx} - E_{wx}^2} \times \text{Error variance} \tag{8.7.2}$$

analogous to (8.6.7). Where differences of means are under study, d_w and d_x are the differences between the w- and x-means of the two treatments. Contrasts can be dealt with as at (8.6.10) but using c_w and c_x, not c_x alone.

Just as it was possible to give a mean value for use at (8.6.8), a mean value can be found for (8.7.2). First, it is necessary to know F_w and F_x. A similar value, G, is required from the column for wx, i.e.

$$G = \frac{f(D_{wx} - E_{wx})}{(v-1)E_{wx}}$$

The correlation coefficient ρ between w and x is also needed, i.e.

$$\rho^2 = \frac{E_{wx}^2}{E_{ww}E_{xx}}$$

A mean of possible values at (8.7.2) is given by

$$\left[K + \frac{2(F_w + F_x - 2\rho^2 G)}{fv(1-\rho^2)}\right] \times (\text{Error variance}) \tag{8.7.3}$$

Covariance efficiency

The use of covariance adjustments leads to a situation very like that at (4.1.1). The variance of a contrast is given by Ks^2. As a result of the adjustment, K is increased to K'. Appropriate expressions have been given at (8.6.7), (8.6.8), (8.6.10), (8.7.2) and (8.7.3). The increase in K is accepted in expectation of a decrease in s^2, and the ratio, K/K', is called the

'covariance efficiency factor' or COVEF. It must be matched by a decrease in s^2 or the covariance analysis will have done harm rather than good.

There is, however, an important difference from the case considered in Section 4.1. The usual efficiency factor (EF) can be calculated before the experiment is initiated, but the COVEF is known only when x has been determined and the randomization is known. It would be wrong to go on randomizing again and again until a high COVEF had been obtained, so anyone who embarks on a covariance analysis has to take a chance. From (8.6.8) it appears that if F equals one,

$$\text{COVEF} = Kfv/(Kfv + 2). \tag{8.7.4}$$

It follows that the covariance efficiency factor is usually high in large experiments because then f will be large. Thus, for a difference of means in a Latin square, where K equals $2/v$,

$$\text{COVEF} = f/(f + 2) \tag{8.7.5}$$

always assuming that F equals one.

This sheds light on the use of an inspired guess for a covariate, which might reduce s^2 but might not. The risks are less in a large experiment than in a small one. If f is large, the covariance efficiency factor will be higher and the loss of one degree of freedom from 'error' of little importance.

8.8 Choice of variables

In the data at (8.5.1) the x-values represent crop in the previous year and a good correlation was found with y, but this cannot be relied upon. As was explained in Section 1.8, the fertility pattern in a wet year can be quite different from that in a dry one or in an early season from a late one, so adjustment of one annual crop by another can often be disappointing, though it has worked very well here. (Perhaps the two seasons were very similar or perhaps there were permanent characteristics of the site.) The correlation between x and y depends quite a lot upon Treatment D in Block V doing badly in both years as is shown at (8.5.3). Was there perhaps something to be seen there? An examination of the site might prove revealing.

The method is usually more effective with perennials, but even then it can fail. One difficulty comes from biennial effects. Crop lost in one year is carried over to the next, so correlations between successive years can be poor, whereas the crops in successive two-year periods may be closely related.

The greatest uses come when something is discovered after an investigation has started. For example, one experiment proved to be very variable in crop and the residuals were examined. They showed a line of poor plots that cut across all the blocks. Local staff were asked to examine the site and they reported a streak of gravel where the low residuals were found. They were told to sample all plots for soil and to measure the gravel content. As a result a covariance adjustment using the gravel content data reduced the error variance to a quarter of its former value. Nevertheless, this was a rescue operation which should not have been necessary. The local staff were at fault for not having examined the site more carefully beforehand.

There are times when the site has been examined beforehand and has been found to vary in so many ways that no single blocking system is going to allow for everything. In such an event it is often best to block as well as possible and to measure characters not otherwise allowed for—soil texture could be one of them—for use as covariates. Deliberate action such as that is good, whereas the carelessness in the previous example was bad.

A common use of covariance is with x-values derived from location or some similar characteristic. For example, an experiment has blocks that form vertical strips on the plan, like those at (8.5.1). After the experiment has started, someone thinks of a reason why they should have gone horizontally. It is perfectly possible to adjust on to a 'pseudo-variate' that will at least remove the linear effect of the trend suspected. At (8.5.1) it would go like this

5	5	5	5	5
4	4	4	4	4
3	3	3	3	3
2	2	2	2	2
1	1	1	1	1

That will be gone into more thoroughly in Section 8.9. Alternatively, covariance might be used to allow for edge-effects. That could be done using a pseudo-variate like this:

1	1	1	1	1
1	0	0	0	1
1	0	0	0	1
1	0	0	0	1
1	1	1	1	1

(It would be better to leave a wider discard area round the experiment.

Although the pseudo-variate might well be effective, it is nonetheless second best.)

There will be more about pseudo-variates later, especially in the next section and in Chapter 12. It should be noted that with pseudo-variates it is not always best to adjust to the mean of x. In the first example above, it would be wise to adjust everything to $x = 3$ so as not to affect the grand mean of y, but in the second example most people would adjust to $x = 0$, that being the value intended.

8.9 Fertility trends and covariance

In some experiments with many treatments it may be difficult to use blocks because they would have to be so large. It is true that a block does not have to contain all the treatments. The designs in Section 4.7 to 4.10 show how this can be arranged, but there are other approaches that can be considered.

To take an example, a large factorial experiment (Lester, *Rothamsted Experimental Station, Report for 1979*, Part 2, pages 17–25) contained 134 plots, all differently treated, in a completely randomized design. Experiments of that sort, like the serially balanced ones to be described in Section 10.6, are especially vulnerable to variation in fertility arising from environmental differences. Many methods of analysis have been used to eliminate, at least in part, the effects of smooth variation, whether in one direction or in two. For example, some have used the total residuals (Section 7.13) to provide a measure of local fertility, and this method will be considered in Section 10.8. In this section we shall consider ways in which the analysis of covariance can be used to allow for trends across the experimental area by adjusting upon pseudo-variates, a method which is an extension of the approach in the last section.

If all the plots of an experiment lie in one line and are equally spaced, it may be convenient to number them 1, 2, 3, . . . from one end and to use the plot number as a pseudo-variate, x. In effect, this is to allow for a steady linear trend, so it may be better to include also $w = x^2$ (or perhaps $w = \sqrt{x}$), which can allow for curvature.

The same approach can be used with long, narrow blocks like those in Exercise 1C. Here the blocks are in effect columns. Clearly it was believed at the planning stage that the main sources of variation would be across the experiment, but there could be differences between the rows as well. Anyone who was bothered by the long blocks could assign an x-value of -9 to the four plots of the top row, of -7 to the plots of the next row and so on, those in the bottom row all being assigned a value of $+9$. If now the yield, y, were analysed with a covariance adjustment on x, allowance

would have been made for any steady trend from top to bottom. A second pseudo-variate, w, could be used to allow for curvature.

There is an alternative. Whether there is a single line of plots or a number of narrow blocks, it may be better to use Fourier functions. The method is as follows. In Exercise 1C the top row could be assigned the angle 0° and the bottom one 180°. Intermediate rows would be assigned other angles going up by equal steps, i.e. 20°, 40°, etc. Then two pseudo-variates could be derived as the sine (x) and the cosine (w) of those angles. To take the example further, for the ten rows the values of x and w would be derived thus:

Row	1	2	3	4	5	6	7	8	9	10
Angle	0°	20°	40°	60°	80°	100°	120°	140°	160°	180°
x	0.00	0.34	0.64	0.87	0.98	0.98	0.87	0.64	0.34	0.00
w	1.00	0.94	0.77	0.50	0.17	−0.17	−0.50	−0.77	−0.94	−1.00

It will be found that such pseudo-variates often provide a covariance adjustment that is both supple and effective. They are able to cope with a wide range of fertility patterns.

That is about as much as can be done using only two variates, x and w, for adjusting y. Those who have to work out all the arithmetic by hand may not wish for more, but those who have computers can be more ambitious. Using powers, x, x^2, x^3, etc., there is usually not much advantage in going beyond x^2 and almost certainly none in going beyond x^4. Fourier functions provide more scope. The use of sin θ and cos θ, where θ is the angle of the plot, can be extended by adding sin (2θ) and cos (2θ) and then sin (3θ) and cos (3θ) and so on. Again, matters should not be taken too far. In particular, when using covariance to allow for row effects, the number of pseudo-variates should not exceed the number of degrees of freedom between rows, supposing that they had been used. This, in effect, turns the original block design into one in rows and columns. If it was necessary, why was it not done in the first place? As it is, the resulting row-and-column design is unlikely to be much good. Although treatments will have been applied to columns in a considered manner, the allocation to rows will have been arbitrary, being the result of randomization. It is likely therefore that standard errors of contrasts will be much inflated to allow for the differential effect of pseudo-variates and a poor experiment will have resulted. (Of course, there are times when the original designer misjudged the fertility pattern and something has to be done to allow for the real one, but it is to be hoped that such occasions are rare.)

When all plots form a single line the situation is different. It is then sound to take as many pairs of Fourier functions as may be needed, but

again with the proviso that there must be enough degrees of freedom left for 'error'.

This deals with the elimination of trends in one direction. Often that is enough. In Exercise 1C, for example, the blocks look after differences across the experiment from left to right. It has been suggested that if there are any differences up and down the experiment, pseudo-variates can be used to remove their effect, though a warning has been given not to be too ambitious. What happens in the genuine two-dimensional case, when there are effects to be removed in both directions?

If only two pseudo-variates can be managed, they obviously have to be x_1 (distance across) and x_2 (distance up or down) but that is unlikely to be effective if trends are not linear, so more are needed. One good plan is to use five:

$$x_1, \quad x_2, \quad x_1^2, \quad x_2^2 \quad \text{and} \quad x_1x_2.$$

This fits a paraboloid and is often very successful. It avoids the difficulty that often besets row-and-column designs. As was explained in Section 1.8, when rows and columns are used, additivity is important and is partly a matter of orientation of the experiment on the land. So long as x_1 and x_2 and x_1^2 and x_2^2 alone are used, the difficulty remains, but it can be met by combining them in x_1x_2. Similar considerations can apply when using Fourier functions. Here two approaches are possible:

$$\cos\theta_1 \cos\theta_2, \qquad \cos\theta_1 \sin\theta_2, \qquad \sin\theta_1 \cos\theta_2, \qquad \sin\theta_1 \sin\theta_2$$

This helps when rows and columns may interact. (θ_1 and θ_2 are angles for rows and columns respectively.) The alternative is to use

$$\cos\theta_1, \qquad \sin\theta_1, \qquad \cos\theta_2, \qquad \sin\theta_2, \text{ etc.}$$

This has the advantage that the number of covariates used for modelling rows need not be the same as the number for columns. Both sets of covariates can be extended by using 2θ, 3θ, etc., as well as θ.

Exercise 8A

In the following body of data, explore the relationship of y to x_1 and x_2:

x_1	x_2	y
9.1	5.4	30.9
10.7	8.0	58.8
11.4	7.3	56.7
13.8	7.9	67.5
14.1	3.9	32.4
14.5	4.1	46.7
8.3	3.7	13.2
12.6	6.4	55.2
7.3	6.3	33.6
7.9	6.4	36.4
9.2	7.2	47.2
15.8	5.9	64.5
12.9	6.4	51.3
5.1	5.3	17.5
10.1	5.5	34.8
10.3	2.6	19.4
10.0	7.8	55.2

How far does the result depend upon the relationship of x_1 and x_2?

[Data devised by D. A. Preece.]

Exercise 8B

Four bodies of data (I, II, III and IV) are given below. For each one calculate the sums of squares and products for x and y and work out the regression equation of y on x.

I		II		III		IV	
x	y	x	y	x	y	x	y
10	8.04	10	9.14	10	7.46	8	6.58
8	6.95	8	8.14	8	6.77	8	5.76
13	7.58	13	8.74	13	12.74	8	7.71
9	8.81	9	8.77	9	7.11	8	8.84
11	8.33	11	9.26	11	7.81	8	8.47
14	9.96	14	8.10	14	8.84	8	7.04
6	7.24	6	6.13	6	6.08	8	5.25
4	4.26	4	3.10	4	5.39	19	12.50
12	10.84	12	9.13	12	8.15	8	5.56
7	4.82	7	7.26	7	6.42	8	7.91
5	5.68	5	4.74	5	5.73	8	6.89

For each body of data, plot y against x.

[Data devised by F. J. Anscombe, Graphs in statistical analysis, *American Statistician*, **27**, pp. 17–21.]

Exercise 8C

An experiment on old apple trees was conducted to try to make them more fruitful again. The treatments, A–E, involved growing various grass mixtures, etc. under the trees to prevent the growth of weeds. In the sixth treatment, O, the local practice was followed by letting the weeds grow but turning them into the soil once a year.

There were two variates:

y, crop in pounds during a four-year period after starting treatments;
x, number of boxes of crop (to the nearest tenth of a box) during a four-year period before starting the treatments.

Data were as follows:

	Block I		II		III		IV	
	X	*Y*	*X*	*Y*	*X*	*Y*	*X*	*Y*
A	8.2	287	9.4	290	7.7	254	8.5	307
B	8.2	271	6.0	209	9.1	243	10.1	348
C	6.8	234	7.0	210	9.7	286	9.9	371
D	5.7	189	5.5	205	10.2	312	10.3	375
E	6.1	210	7.0	276	8.7	279	8.1	344
O	7.6	222	10.1	301	9.0	238	10.5	357

The design was in randomized blocks.

Work out the analysis of variance for y. Then calculate it with a covariance adjustment on x. Note in what ways the assessment of treatment effects is modified.

1 pound = 454 grams One box holds about 40 pounds

[Data from S. C. Pearce, *Field Experimentation with Fruit Trees and Other Perennial Plants*, 1st edn, 1953, p. 113.]

Exercise 8D

An experiment was carried out in New Zealand on grass with four treatments, LP, L, P and (1), where L represents addition of lime and P of phosphate. There were eight randomized blocks. (Only four are given here to ease the load of computation.) Further, each plot was divided into two sub-plots, one of which received a dressing of Ammonium molybdate (M) and the other did not.

The area was first burnt to remove the scrub. It was then sown with grass and clover and given a dressing of urea. Two crops of hay were taken and their combined yield is here called x. After application of treatments, a further crop was taken, here called y. All data represent yields in grams after air drying.

Block		LP		L		P		(1)	
		O	M	O	M	O	M	O	M
I	*x*	1035	1137	887	913	1280	1240	930	891
	y	950	1250	530	491	1140	1135	676	685
II	*x*	433	954	554	839	605	949	571	903
	y	488	800	618	694	925	1406	660	714
III	*x*	523	544	779	321	570	556	648	709
	y	1380	1280	824	566	1212	1043	718	728
IV	*x*	320	646	619	301	418	448	372	629
	y	538	958	492	501	737	863	246	592

Note that with a split-plot design each main effect and interaction is tested using the 'error' mean-square of the stratum to which it is applied. (As explained in Section 7.4, where an effect is partly in one stratum and partly in another, the situation is very complicated and better avoided if possible.)

Carry out an analysis of variance of y with a covariance adjustment on x.

[Data from C. I. Bliss, *Statistics in Biology*, **2** (1970), p. 484.]

Exercise 8E

Someone points out that in Exercise 1C the blocks are rather long and he suggests that it might be advantageous to take out the linear effect of the position in the field. Examine the data with that in mind and see if it helps.

(Hint: In effect this means adjusting on a pseudo-variate, x, that measures distance along the blocks. It will be easier if all values are whole numbers and the mean is zero. That suggests + 9 for all plots in the top row, + 7 for those in the next, and so on to − 9 for those in the bottom row.)

Exercise 8F

After seeing the results from Exercise 8E, our critic is still not satisfied. He asserts that trends are rarely steady and he wants a curvature effect as well. Examine the data, adjusting on the Fourier functions, $\cos\theta$ and $\sin\theta$, as pseudo-variates.

Note: As presented here the critic appears to have asked for one amendment after another in the hope of obtaining results more to his taste. If that was indeed his motivation, he was behaving in a reprehensible way. On the other hand, if he had been apprehensive from the start about those long narrow blocks and had wanted the fullest protection against trends along them, he would have shown a meticulousness that can only be commended. In general, the form of an analysis should be decided without reference to the data. If something objective, like a streak of gravel, is discovered at a late stage, there could justifiably be an attempt to allow for it, because it would certainly have led to modifications in design if it had been found in time. On the other hand, a data analyst who uses first this device and then another to find something that he can declare significant may well succeed, but the resulting analysis will probably be nonsense.

Exercise 8G

The following data come from one of the earlier published examples of the use of randomized blocks. There were four blocks for the comparison of 16 varieties of potato, A to Q with I omitted. The first two blocks raise no obvious queries, but to modern eyes it does look as if the other two blocks had been formed in an unfortunate manner. Data represent yield in pounds per plot (1 pound = 454 grams).

A	L	J	C	P	Q	B	E
351.5	495.5	443.0	383.5	559.0	550.0	359.0	395.5
K	B	G	O	C	H	J	O
472.5	367.5	455.5	502.5	328.5	390.5	483.0	512.0
E	F	Q	D	N	M	A	D
357.5	381.5	531.0	316.0	522.0	444.0	325.0	259.0
N	H	P	M	F	G	K	L
385.5	354.0	496.5	474.5	410.5	351.5	430.0	394.5

The difficulty, as Exercise 1D showed, is that residuals in the lower part are mostly low and those in the upper part are mostly high, which suggests

that the blocks would have been better if they had been two rows deep and eight columns wide.

Apply a covariance adjustment with $x = +3$ in the top row, $+1$ in the second, -1 in the third and -3 in the one at the bottom. Does it reduce the 'error' variance?

[Data from T. Eden and R. A. Fisher, *J Agricultural Science*, **19** (1929), page 207.]

Chapter 9

Transformations

9.1 Justifying assumptions

In Section 1.7 we looked at some of the assumptions made in calculating an analysis of variance and thereafter we have assumed that those assumptions hold. Mostly they do, but sometimes they fail. When they fail, the fault may be with the way in which the various quantities were measured.

To be specific: (1) it is assumed that the various parameters for blocks, treatments, etc. can just be added and do not need to be combined in some other way; (2) it is also assumed that the residuals are all subject to the same sources of variation and therefore all have the same variance of estimation, σ^2, their distribution being normal (see Section 3.1).

If a model is proposed in which parameters are multiplied, $y = ax^p w^q$, then taking logarithms gives $\log y = \log a + p \log x + q \log w$, the parameters $\log a$, p and q now appearing in an additive model that relates $\log y$ to $\log x$ and $\log w$. It may be that $\log y$ is a more basic measurement in a system than is y itself; for example, if y is the size of an organism that is growing exponentially then it is the ratio of final size to initial size that is fundamental biologically.

It may be asked what happens if a transformation is used that enables one assumption to be fulfilled while falsifying the other. The answer is that the difficulty arises only rarely. Transformations well used are more than a statistical expedient to satisfy the mathematicians. As in the example above, they may direct attention to a quantity that is more meaningful than the one actually measured. The point is illustrated by the true story of a botanist who was studying the spread of *Venturia* fungus on apple fruits. At first he was content to measure the diameter of lesions, but later he contrived a means of recording their areas instead. When he gave the data to the statisticians for analysis they found that both assumptions were falsified. The parameters did not combine by addition and the larger data were more variable, defects that could be corrected by using a square-root transformation. In fact, that restored the original basis of measure-

ment. The intention was to measure the spread of *Venturia* from the spore that had been deposited by wind on the apple face, thus initiating a lesion. The measurement really needed was a mean radius from the site of the original spore and that was better measured by a diameter than by an area. (If the intention had been to measure damage to the fruit, a square-root transformation of areas would still have been correct on statistical grounds. The underlying biological phenomenon was the same in each case and that is what needed to be measured.)

9.2 Use of residuals

The assumptions can often be checked by plotting the residuals e against the data y. If the assumption of constant variance and normality is satisfied, the diagram will be as in Case 1 of Fig. 9.1; that is to say, the values will be grouped round the value $e = 0$, and will become increasingly sparse as one moves away from that value. Also the spread of the residuals will be the same for all values of the data, not as in Case 2, where higher values are subject to more variation. Patterns sometimes arise in which all the larger variances are all of one sign and are balanced by a lot of small ones of the opposite sign. That situation also can benefit from a transformation.

A more awkward situation is illustrated by Case 3. Here the residuals come not from one treatment but from two, one represented by crosses and the other by circles. The diagram casts serious doubt on the assumption that data are all equally variable, those from the crosses

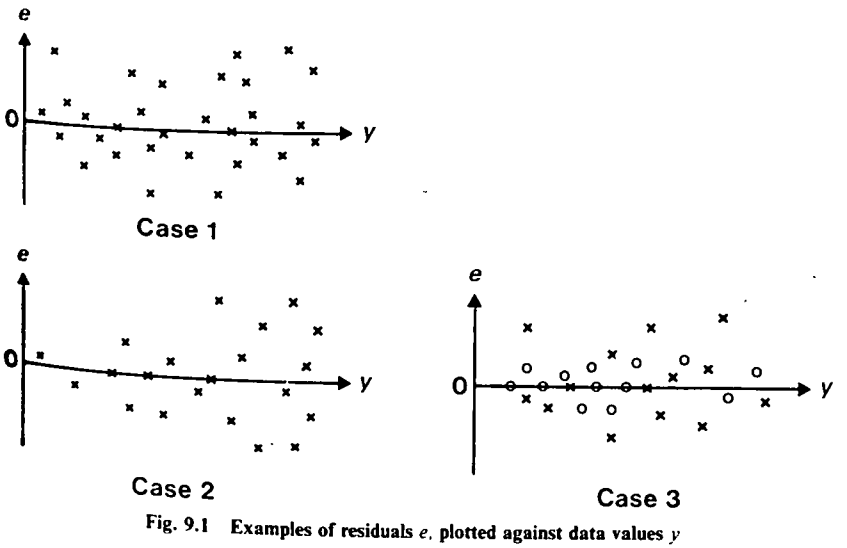


Fig. 9.1 Examples of residuals e, plotted against data values y

showing a wider spread than those from the circles. if there were a difference in means, the difference in variability could be another aspect of Case 2, but that is not so. It appears that the application of one treatment has itself been a source of 'error' when the other has not. (This is quite plausible if the first involved manipulation, like pruning and thinning, absent in the other.) The situation is difficult and will receive attention in Section 9.6. It will not be helped by a transformation because that will affect both treatments and the difficulty will remain.

There are many cases. In skilled hands the device of plotting residuals against data can reveal a lot.

Another useful device is to make a field plan of an experiment and place the value of the residual on each plot; any systematic pattern in those residuals could indicate a fertility trend which the design used has failed to take out. (For example, perhaps a Latin square ought to have been used instead of randomized blocks.) Damage to plots through exposure or accident can also be revealed by this method.

The more important assumption mentioned in Section 9.1 is the one that concerns constancy of variance. If we know how the variance of a set of observations is related to their size, it is possible to find a function of y that will have constant variance. If an analysis of variance is carried out using this function then the assumption (2) is satisfied. As we have said, often the other two assumptions, (1) and normality in distribution, will be satisfied better on this new scale of measurement than when using y itself.

9.3 The logarithmic transformation

When the standard deviation of an observation is proportional to its size (Case 2 above), the function $z = \log y$ will have constant variance. The range of a set of observations indicates the size of their standard deviation, and is used as a quick guide whether or not to transform.

Example:

Treatments		A	B	C	D	E
Block	I	14.6	20.2	12.2	25.2	30.1
	II	15.4	20.1	13.5	25.0	31.6
	III	16.8	23.4	14.0	28.7	32.5
	IV	15.8	21.4	13.2	28.0	34.0
Mean		15.6	21.3	13.2	26.7	32.0
Range		2.2	3.3	1.8	3.5	3.9

Plant growth is often analysed in this way, which is natural because the transformation directs attention from actual size to growth rate. That is

what the treatments and the sources of 'error' actually affect.

Nevertheless, the transformation needs to be used with care. First of all, if zeros are possible, even though none may occur in the data set, the transformation needs to be used, if at all, with discretion, because log 0 is minus infinity. As a datum that would be absurd. (Where the transformation is used to convert sizes to growth rates the difficulty does not arise, because a plant cannot be of zero size.)

Another use for the transformation comes with insect colonies, which also can grow exponentially. Here, of course, zeros are possible because there may be no colony. That leads to the question of discontinuity.

As was explained in Section 1.10, data should be recorded to a sufficient degree of precision. If this means measuring to the nearest millimetre instead of the nearest centimetre the effort should be made. Sometimes, however, that will not be possible. Then zeros appear as an approximation, even though a genuine observation of zero is absurd. Also in counting insects, fruits, etc. only whole numbers will make sense. The data may look like this:

4 2 5 1 6;

where such serious discontinuity appears, it is advisable for mathematical reasons to analyse $\log (y + 3/8)$ instead of $\log y$. That also helps with the occasional instance when y appears to equal zero. (This can arise as a result of rounding a small but non-zero datum.)

9.4 The square-root transformation

If the variance, rather than the standard deviation, of an observation is proportional to its size, as when data follow a Poisson distribution (Section 3.8), $z = \sqrt{y}$ will have constant variance. Insect counts often (but not always) need this transformation, as do counts of the number of weeds per unit area of soil or the number of fungal lesions per unit area of leaf. The range of observations is roughly proportional to the square of the mean on these scales; it is only in the transformed units that the residuals have constant variance.

Again there is a limitation. Since z is defined only for zero or positive values of y, the square-root transformation must be wrong—or at least suspect—if there is any possibility of y being negative.

Again, discontinuity can cause difficulties. If any arise, it is better to use $z = \sqrt{y + 3/8}$ than $z = \sqrt{y}$.

9.5 The angular transformation

The third transformation to be considered here is different because it is no more than a statistical expedient without biological meaning. Data are

often presented as a proportion or a percentage. For example, someone examines a sample of n leaves and reports that r of them exhibit a certain deficiency symptom. As a result there is a need for an analysis of $p = r/n$ or perhaps of $100r/n$.

Such a variate can be decidedly awkward. From Section 3.7 it appears that the variance of p is $p(1-p)/n$, which is not constant. The value of n can be standardized. Indeed, it is wise to do so. Sometimes a recorder will examine all the leaves on each shoot, even though the latter vary in length. It would be better to devise a sampling method to obtain a constant number, n, from each plot. That removes one cause of unequal variances, but it does nothing about the other, namely the different values of p itself.

A formal solution of the problem is to analyse not p but an angle, θ, such that

$$\sin^2 \theta = p.$$

The case of highest variance of p comes in the middle of its range, where $p = 0.5$ and $\theta = 45°$. If $p = 0.25$ then $\theta = 30°$; if $p = 0$, $\theta = 0°$ and so on. The transformation cannot be used outside the range of p between 0 and 1.

Although the transformation can be useful, it does make results difficult to interpret because it has no real biological meaning. Fortunately, it can often be avoided. For example, when all data lie within the range $p = 0.15$ to $p = 0.85$, an analysis in terms of θ will give results little different from one in terms of p. Sometimes when there are values of p well below 0.15 or well above 0.85, there is little alternative to the use of θ.

The transformation can be especially awkward with factorial designs. The absence of an interaction usually means that two factors combine by ordinary addition. Thus if Treatment (1) gives a mean of μ, A gives a mean of $a = \mu + (a - \mu)$ and B one of $\beta = \mu + (\beta - \mu)$, then AB is expected to give

$$a + \beta - \mu = \mu + (a - \mu) + (\beta - \mu).$$

That is the interaction as usually understood. To look at the situation more closely, we will suppose that 75 percent of insects survive if nothing is done, but only 50 percent if spray A is applied, and only 25 percent if spray B is used instead. This means that $\frac{2}{3}$ survive when A is used alone and $\frac{1}{3}$ when B is used instead. When used together, it is to be expected that $75 \times \frac{2}{3} \times \frac{1}{3}$ percent will survive, i.e. $16\frac{2}{3}$ percent. Since their effects are expected to combine by multiplication, a logarithmic transformation is required, but that would lead to variances even more diverse than those found before it was used. On the other hand, an angular transformation

will stabilize the variances while making nonsense of the interaction. The θ-values for (1), A and B are respectively 60°, 45° and 30°, so, in the absence of an interaction, AB is expected to give a θ-value of 15°, corresponding to a p-value of 0.07, which is not the same as the 0.17 required by the biological concept of independence of action. It will be found, however, that all transformations give trouble at times. Where difficulties become too great, a possible alternative is the method of the next section.

Given discontinuity, the best form of the angular tranformation is

$$\sin^2\theta = \frac{r + 3/8}{n + 3/4}$$

9.6 Single degree of freedom effects

It sometimes happens that no transformation can be suggested. It could be that the variate for analysis is some 'index' or 'coefficient' that biologists have found useful. It is perhaps calculated in some complex manner, and the statisticians can only guess how it will be distributed. Alternatively it could be that some of the treatments require a lot of manipulation of plants, e.g. tree forming, which leads to differential variability but not of a sort that is associated with high and low means. A third example arises with different modes of application of fungicides or insecticides. Some modes may lead to patches of poor pest control, and this will increase variability for those treatments. Whatever the reason, transformations do not always provide a solution.

In that case it is sometimes possible to take each contrast with its single degree of freedom and to find an estimate of its variance. That can be done if there are enough replications and if the design is in blocks. We shall explain the method using a body of data that raises no special difficulty but is nonetheless very suitable for our purpose. We shall take the data for Variety X in Exercise 7D. They give a six-fold replication of four treatments, i.e. nitrogen fertilizer applied at four equally spaced levels. The data are:

Treatment	Blocks							
	I	II	III	IV	V	VI	Total	
N_0	111	62	74	68	61	53	429	
N_1	130	90	89	64	91	74	538	(9.6.1)
N_2	157	100	81	112	97	118	665	
N_3	174	116	122	86	100	113	711	
	572	368	366	330	349	358	2343	

The corresponding analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Nitrogen levels	3	8136.45	2712.15	16.65
'Error'	15	2443.30	162.89	
Stratum total	18	10579.75		

(9.6.2)

It remains to partition the treatment line into the linear, quadratic and cubic effects, each with a single degree of freedom, as shown at (5.7.4), namely

$$\text{L}, (-3 - 1 + 1 + 3); \quad \text{Q}, (+1 - 1 - 1 + 1); \quad \text{C}, (-1 + 3 - 3 + 1).$$

We begin by evaluating each contrast for each block separately, i.e.,

	I	II	III	IV	V	VI	Total
L	+ 216	+ 172	+ 136	+ 102	+ 123	+ 224	+ 973
Q	− 2	− 12	+ 26	− 22	− 27	− 26	− 63
C	− 18	+ 24	+ 72	− 126	+ 21	− 72	− 99

(9.6.3)

We will take L, the linear effect, first. Its value of U at (5.6.7) is 20 and the replication is six, so its sum of squares is $(+973)^2/(20 \times 6) = 7889.41$. The block values are now squared and added and the sum divided by U to give

$$[(+216)^2 + (+172)^2 + \ldots + (+224)^2]/20 = 8522.25.$$

The difference, $8522.25 - 7889.41 = 632.84$ with five degrees of freedom, is the component of 'error' that specifically relates to the linear effect.

For Q, the quadratic effect, U equals 4. That makes the sum of squares for Q in the treatment line 165.38 and the component of 'error' 512.87. For C, the cubic effect, where U again equals 20, the two figures are respectively 81.68 and 1297.57. We can now take the analysis of variance already found and partition two of its lines, the one for treatments and the one for 'error', as follows:

Source	d.f.	s.s.	m.s.
Linear effect (L)	1	7889.41	7889.41
Quadratic effect (Q)	1	165.38	165.38
Cubic effect (C)	1	81.68	81.68
'Error' (L)	5	632.84	126.57
'Error' (Q)	5	512.87	102.57
'Error' (C)	5	1297.57	259.51
Stratum total	18	10 579.75	

(9.6.4)

That is an extension of what was done at (7.4.1). Apart from rounding errors, everything at (9.6.4) adds correctly to its total at (9.6.2). However, no F-values have been given because there is an ambiguity. Is F for the linear effect 48.43 (= 7889.41/162.89) with 1 and 15 degrees of freedom, or is it 62.33 (= 7889.41/126.57) with 1 and 5 degrees of freedom? In the present instance, where there is no reason to doubt the 'homogeneity' of the 'error', the answer must be 48.43. In the ordinary way no one would have bothered to have calculated the analysis at (9.6.4). However, now that it has been worked out, some may be surprised that the three 'error' components, one for each of L, Q and C, are so different. We will therefore look into the matter formally.

The method is to use 'Bartlett's test', which depends upon the criterion known as χ^2. (That received previous mention in Section 3.5.) The question is: can the three components (126.57, 102.57 and 259.51) really be regarded as random variants of 162.89? We write down the logarithms of the four variances, thus:

$$\log 126.57 = 2.1023$$
$$\log 102.57 = 2.0108$$
$$\log 259.51 = 2.4142$$
$$\log 162.89 = 2.2119$$

Then we multiply each of the logarithms by the corresponding number of degrees of freedom and proceed to take a difference between the figure for the pooled variance and those for the separate ones, i.e., we find

$$A = 15(2.2119) - 5(2.1023) - 5(2.0108) - 5(2.4142) = 0.5420.$$

If the three separate variances had been exactly the same, A would have been zero. It is not so and its value tells us how different the variances are. To apply the test we need a further value, found from the reciprocals of the degrees of freedom, namely,

$$1/5 + 1/5 + 1/5 - 1/15 = 0.5333.$$

We divide that by $3(g-1)$ to obtain W, where g is the number of component variances. Here $g = 3$. That makes $W = 0.08889$. Finally we calculate

$$\chi^2 = 2.3026A/(1 + W)$$
$$= 1.146.$$

The value of χ^2 is referred to tables with $(g-1)$ degrees of freedom. As a result there is no suggestion that the component variances are significantly different, the value of P being rather more than 0.5, so the use of the pooled variance at (9.6.2) appears to be justified.

This approach has led to an expected conclusion. In other circumstances there might have been no reasonable basis for expecting either a homogeneous or a heterogeneous 'error'. In that case we might have been glad to have the evidence of the test. The approach by single degree-of-freedom effects has other uses because the data might have failed to satisfy some other essential assumption. For example, the data at (9.6.1) might have represented a variate that was not necessarily additive. To take another example, the treatments could have been such that the data were correlated within blocks. (That would have happened if the data were diameters taken at different distances along a shoot, each shoot forming one block.) The method is not perfect, but so long as a number of independent estimates can be made of each contrast of interest, it is required only that those estimates shall be distributed in an approximately normal manner. Each contrast can have its own 'error' variance. If it is suggested that the variances should be pooled, we may have to ask whether they are all estimates of some common value. If that question does arise, Bartlett's test is available.

We should note, by the way, that the table at (9.6.3) is not entirely typical. For that particular set of contrasts the column headed 'Total' could have been obtained directly from (9.6.1) or by adding the rows of (9.6.3). That is not always the case. Where the two methods differ, the values required are the row totals.

We may illustrate a different case by considering the well-known law advanced by Mitscherlich. It says that when fertilizer is applied at evenly spaced levels, as at (9.6.1), successive increments in yield bear a constant ratio to one another. Calling the yields for levels 0, 1, 2, 3 respectively a, b, c and d, the law declares that

$$(c-b)/(b-a) = (d-c)/(c-b) \qquad (9.6.5)$$

We shall use the data at (9.6.1) to examine whether that is so.

The first task is to find a variate that will measure the extent to which the two sides at (9.6.5) differ. The obvious solution is to take their difference, but that is not satisfactory because with real data it is quite possible that a will equal b, or c will equal b; in either event our variate will be infinite in value. One possibility is to use

$$H = (c-b)^2 - (b-a)(d-c).$$

If the law holds exactly, H will equal zero. This is not entirely satisfactory either, because H is made up of squares and products of the data; if they are distributed normally it is not to be expected that H will be distributed in the same way. The obvious device is to use a square-root transformation, but that is not feasible here where the variate, H, can take negative values. We shall therefore accept some measure of non-normality, recognizing that the longer tails of the distribution will diminish sensitivity, not increase it.

We proceed thus: for each block we work out the value of H and then we apply the method of Section 3.3 to see whether the mean of H differs significantly from zero, i.e.,

I	II	III	IV	V	VI	Total	
+ 406	− 348	− 551	+ 2200	− 54	+ 2041	+ 3694	(9.6.6)

The sum of squares attributable to the difference between the mean value of H (+ 616) and its expected value (0) is $(+3694)^2/6 = 2\,274\,273$ with one degree of freedom. The sum of squares for 'error' is

$$(+406)^2 + (-348)^2 + \ldots + (+2041)^2 - 2\,274\,273 = 9\,370\,711$$

with five degrees of freedom. That makes F equal to 1.21. Hence the data could well have followed Mitscherlich's law. Nevertheless the outcome has not been entirely satisfactory because of the two large values of H, one for block IV and the other for block VI, both of which arise from a high value of c for those blocks. (There is in fact some evidence of an optimal application somewhere around level 2, in which case the law is not being followed strictly.) On the other hand, the analyses at (9.6.2) and (9.6.4) do not suggest much curvature. The fact is that we do not have enough data to determine the precise form of the response curve, though we have had enough to show the method.

Another example comes from Section 9.5, where Treatment (1) gave a mean kill of μ, A gave α and B gave β. In the absence of an interaction what should we expect from AB? If factor A in the absence of B kills a proportion α/μ, and if factor B in the absence of A kills a proportion β/μ,

we would expect the two working independently to kill a proportion $\alpha\beta/\mu^2$. That would leave an infestation of $\alpha\beta/\mu$. It was pointed out in Section 9.5 that such an interaction can be studied only with a logarithmic transformation, whereas an angular is needed to stabilize the variance. Writing y as the observed result from AB, one solution would be to use the method given above to see whether the mean of $(y - \alpha\beta/\mu)$, when calculated over blocks, did in fact differ from zero.

The same approach could be useful with Case (3) in Section 9.2. As with all these examples, a difficulty could arise from a lack of degrees of freedom to give a satisfactory estimate of the 'error' variance. In Section 1.8 we emphasized that there should at the very least be six or eight, but 20 were desirable. With these partitioned 'errors' there can rarely be any reasonable hope of such standards being attained. The effect is not any loss of validity, but there could be serious loss of sensitivity.

It should be mentioned that the blocks do not have to be complete. For example, we may suppose that treatments A and B concur in b blocks; it would therefore be possible to compare them knowing the variance appropriate to their difference, there being $(b - 1)$ degrees of freedom for that purpose. Treatments A and C may also concur several times, though in a different set of blocks. Nevertheless that does not matter; we require only enough concurrences to provide the degrees of freedom for the calculation of the variance. (We should note however that difficulties might arise if someone wanted to know about a contrast that involved several treatments. We should then be limited to those blocks in which they all occurred.) We should explain that the neat summation of the component 'error' sums of squares at (9.6.4) to the combined value at (9.6.2) can be expected only if the same blocks are used throughout.

9.7 Presenting results after transformation

The results of an analysis of variance are valid only on the transformed scale of measurement, i.e. in terms of the logarithms, square roots or θ-values, as the case may be, and not on the original scale. Means can be compared only on the transformed scales because it is only in those units that they are all subject to the same error. It is often a good plan to transform back again to aid understanding of the results. For example, the mean of 1, 4, 9 and 16 is 7.5, but if their square roots 1, 2, 3 and 4 are used the mean is 2.5, which after back-transformation gives 6.25 $(= 2.5^2)$. People sometimes complain that the result looks arbitrary, but it is not. The original value of 7.5 was obtained giving equal weight to all data. If a square-root transformation is really required, the higher values have a higher standard error and should rightly be accorded less weight. That is why the transformed mean has drifted downwards, i.e. towards the better

established data. Since all means will do the same, the effect on treatment differences may well be small, but there will be protection against attaching too much importance to a mean that was high on the original scale only because of one high but poorly determined value.

Nevertheless, that is not entirely satisfactory. It is true that an experiment does not estimate treatment means as such but only differences between them; but people do quote individual means, especially of crop weights, and they complain if they find a consistent negative bias. There is in fact a conflict and it needs to be recognized. What is clear is that tests need to be based upon the means of transformed variates and that confidence limits should be calculated in the same way, even if it does lead to the limits on the back-transformed variate being asymmetrical. For example, suppose that a treatment gives a mean of 10.0 on the transformed variate using square roots. The standard error is, say, 1.5. Then if the appropriate value of t is 2.20, the confidence limits are $10.0 \pm 3.3 = 6.7$ to 13.3. A back-transformation to restore the original scale of measurement gives limits of $(6.7)^2$ and $(13.3)^2$, i.e. 44.9 to 176.9, the mean itself being back-transformed to 100.0. That may look strange, but higher values have higher variability and the upper limit is correctly further from the mean than the lower. Such apparent discrepancies need to be dealt with sensitively in any report. In many instances the experimenter will have worked out treatment means from the untransformed data, and it may be difficult to convince him that the statistical analysis is correct when each back-transformed mean is lower than the correponding untransformed. Sometimes it is best to explain that the back-transformed values are adjusted for purposes of the analysis; sometimes they are better omitted. That need not be a serious loss, provided all conclusions can be justified from the analysis of transformed values.

9.8 Transformations in the analysis of covariance

The variable y may be transformed if necessary for the same reasons as in analysis of variance, but x is not subject to the same assumptions: it can follow any distribution, its variance need not be constant, and all that is required is a *straight-line* relationship of y on x. Transformation of x may be needed to achieve this, but that is a different problem.

9.9 Some ideal cases

Many years ago, Cochran gave expressions for the resulting error variance if all assumptions are completely justified. That is at once a challenge and a warning. It is a challenge because it sets standards; it is a warning because it reveals that people make assumptions about their data and do so as a matter of course. Whether those assumptions are justified is

another matter. Mostly in calculating an analysis of variance it is taken for granted that the effect of treatments is the same in all blocks and that all residuals are distributed normally (see Section 3.1) with a standard error that is constant over the whole experiment. A transformation is introduced to make both those assumptions hold for that particular body of data. How well does it succeed?

Cochran showed that if the original data follow a Poisson distribution (see Section 3.8) and a square-root transformation is used to correct for the non-constant variance, the error mean-square will equal 0.25. (In fact, there are other distributions for which the square-root transformation is appropriate, so some other figure does not show that anything is seriously wrong, but it does contradict the idea of an underlying Poisson distribution.)

A similar result applies for the case where the data derive from a Binomial distribution. In each plot a sample is taken of n leaves (or fruits or bark samples, etc.) and an analysis is desired of the proportion of leaves, etc. with a certain characteristic. That is the occasion for using the angular transformation. If everything is correct, the error variance will be $821/n$.

Finally there is the case for which a logarithmic transformation is used, i.e. the standard error of any datum is assumed to be k times its value. If that is really so, the error variance will be $0.189/k^2$.

In fact, these ideal values are rarely attained. It is nonetheless useful to know about them. If a test is needed whether the observed variance accords with what was expected, one is given in Section 3.5, namely, if s^2 is the observed variance and σ^2 is its expected value, then fs^2/σ^2 is distributed as χ^2 with f degrees of freedom; f is the number of degrees of freedom used for the estimation of s^2.

Exercise 9A

Three different formulations, A, B, C, of a selective weedkiller were used in an experiment on the growth of soya beans. Eight replicates of each concentration were included in a completely randomized layout, and samples of the soya bean plants were taken from each of the 24 plots after a fixed period. The damage per plot was recorded for analysis as follows:

A:	12	8	15	10	17	19	11	16
B:	32	20	28	37	25	22	29	34
C:	49	60	59	44	40	52	46	48

Examine whether the data require transforming before analysis, and then complete an analysis.

Exercise 9B

An experiment was carried out using five treatments, A–E, in six randomized blocks to promote early germination of seeds, each plot being a pot in the greenhouse. After a fixed period the number of cotyledons to appear was as follows:

Block	A	B	C	D	E
I	8	6	6	5	17
II	18	15	12	10	30
III	10	8	5	2	10
IV	25	27	23	20	48
V	12	8	8	3	25
VI	10	11	6	4	15

Discuss what transformation, if any, to use.

Exercise 9C

The numbers of insects present on each of 8 samples of leaves (of fixed area) of the same crop taken from plants receiving four different spray treatments are as follows:

Variety								
A	3	0	0	6	8	1	12	10
B	21	3	65	8	4	38	11	48
C	25	44	14	18	2	31	0	0
D	15	27	4	0	10	12	0	1

Treatment C is the one currently being used. Do any of the others differ from it in their ability to limit insect damage?

Chapter 10

Some special topics

10.1 Series of experiments

Introduction

If similar experiments are done on several sites in the same season, it is commonly found that the variation of any chosen treatment-contrast between experiments is greater, often much greater, than is indicated by the variation between replicates within each site. In other words there is a treatment × site interaction. If the series of experiments extends over several seasons, there will probably be substantial variations that can be described as treatment × years and treatment × sites × year interactions. The essential problems of designing and reporting a series of similar experiments are the assessment and interpretation of such interactions and the estimation of average treatment-contrasts over all sites and seasons, or of certain sub-sets of sites and seasons, with appropriate measures of confidence, e.g. standard errors.

In a country like England, the weather in one season may be very different from the weather of another, but in any one season most sites will have fairly similar weather. In such conditions we may expect treatment × site interactions to reflect mainly variations between sites in soil or husbandry, whereas treatment × year interactions will be the consequences mainly of variations in weather. There can of course be exceptions; for example, in a generally dry season a few sites may be affected by isolated heavy falls of rain. In other conditions the weather may be less variable from one season to another, or it may be less consistent over the area to be studied. In either case different interactions are to be expected.

Design

The design of a series of experiments has two components, the choice of sites and the design of the experiment at each site. Those two aspects are not independent; they are discussed in the next two sections.

Two main strategies can be distinguished, though there may well be intermediate possibilities.

(a) If the object of the series is to study a particular component (or a few components) of the treatment × environment interaction it is best to choose equal numbers of sites to represent each specified set of environmental conditions. Imaginary examples are:

(1) the four combinations of hillside or valley floor in zones of high or low rainfall;

(2) light or heavy soils after crops of legume or maize.

If the conditions are clearly defined and if each set of conditions can be assumed to represent fairly accurately the conditions in large areas of the country, it may not be necessary to select many sites in each combination of conditions—perhaps two in each will suffice.

(b) Instead, the object may be to formulate recommendations for growers and to estimate the effects on regional or national average yield if the recommendations are widely applied. In this case it is necessary, first, to define precisely the population of fields to be studied, and, second, to choose a truly representative sample of these fields for the sites of experiments of the series. Examples of well-defined populations are:

(1) all fields of maize larger than 0.1 hectare in District A,

(2) all fields where wheat in 1986 follows barley in 1985 on the clay-with-flints soils in England.

Sites may be chosen strictly at random, by stratified sampling (e.g. one field per civil parish) or by other methods; often strictly random sampling is impracticable, but the biometrician must beware of any bias that may be introduced by non-random sampling.

The design (in particular, the number of replications) at each site is related to the number of sites. In general the more sites, the fewer replications are needed at each. The reason is that as the number of sites is increased, the interpretation of the results will depend more on the treatment × sites interaction than on the treatment × replicates interaction (that is, 'error') within sites.

There is a range of possible schemes; most research programmes could be allocated a point somewhere in this range.

(i) The investigation is based on a single experiment at one site, the intention being to generalize over an area typified by that site. The treatments of this single experiment probably have a factorial structure, and it may well be that the 'error' will have to be estimated from high-order interactions. The design may be orthogonal or non-orthogonal, with or without confounding.

(ii) Several rather simpler experiments are dispersed over the area, which

is thereby sampled by more than one site. Such a scheme enables an assessment to be made of the treatments × site interaction in order to find out how far successful treatments depend upon having the correct environmental conditions.

(iii) Many experiments are dispersed over the area. Although they will all be fairly simple, each will have enough degrees of freedom to determine its individual 'error' variance. In that way it would be possible, say, to find the response of rice to different levels of nitrogen fertilizer and to do so over the area as a whole.

(iv) Some very simple trials, each consisting of perhaps two to six plots, are dispersed over many sites to estimate the effect of one or two simple changes in the husbandry of farmers' fields. For example, the investigation might concern the contrast between a new recommended variety of maize as compared with the traditional one. The investigation might well cover the whole country. Since there are no longer enough degrees of freedom to determine the 'error' variance at each site, some pooled figure will have to be used.

(v) In the extreme the investigation becomes a survey and not an experiment at all. There would be very many sites. The intention would be either to estimate mean yield over the whole area or to detect localities in which the farming system under study might prove unsatisfactory.

In general, at all levels apart from the first, the sites should be chosen at random, or as nearly as possible at random, if the intention is to estimate some quantity over the area; but some selection is permissible if the intention is to compare different environments. At level (i) (the single experiment) the choice of site may reasonably be governed, at least partly, by the need to minimize 'error' variation. Moving down the range, especially at level (iii) or more, a rigorous system of selection of sites, with an element of random choice, becomes increasingly important to ensure that the chosen sites shall truly represent the relevant population of fields. It is not permissible at these levels to reject a sub-set of fields because experiments on them are expected to have large 'errors' as such rejection will entail a risk of bias. Ideally, every experiment of a series would have the same design and number of replicates. (There may be exceptions to this rule if complicated confounding is involved but that is beyond the scope of this manual.) If circumstances make it necessary, however, individual experiments may use different designs. (An example would be a series laid down in randomized blocks with a few 'difficult' sites requiring Latin squares.) Similarly, a site with a limited amount of near-uniform land can be included by using fewer replications than the standard, or by use of plots smaller than standard. More extreme cases of the use of different designs will be mentioned below.

The method of data analysis depends upon the level of the investigation. The single experiment at Level (i) should be analysed with care, the purpose always being borne in mind. In a survey at Level (v), however, it may suffice to find a figure for yield per unit area. (To say this is not to decry the considerable skills needed to interpret a survey; they are, however, different from those needed to interpret an experiment. The present text is concerned only with experiments.)

Assuming that there are enough degrees of freedom for 'error' in each experiment to provide a useful estimate of variance, mean squares should be tabulated in case any definable subset of sites (e.g. those on sandy soils) tend to give larger (or smaller) 'errors' than the remainder.

It is possible to test for non-homogeneity (see Sections 3.5 and 9.6), but there is usually no reason why different sites should give equal variances. Any site showing an outstandingly large variance may at this stage be re-examined for hitherto undetected mistakes, e.g. a mistake in recording of one or more yields of misapplied treatments. For any set (or sub-set) of sites believed to have approximately equal 'error' variances we can calculate a pooled estimate. If designs vary, it is probably best to add together the sums of squares (in terms of yield per unit area) and divide by the total degrees of freedom.

Next comes the combined analysis including all (or nearly all) sites. It is reasonable to reject the results of a site only if there is strong evidence of a serious mistake; it should not be omitted solely because its 'error' mean square is unduly large. It is possible to use a weighted analysis, giving each site a weight in inverse proportion to its 'error' mean square. (This rule is subject to modification if different sites have different numbers of replicates.) Nevertheless, this should usually be avoided, for two reasons, first because each 'error' mean square is a sample from a population of possible values, so that the weights usually have a substantial random component; secondly, because undue weight may be given, accidentally, to a particular type of site, perhaps the less weedy ones or those on deeper, less variable soils. Equal weighting avoids these dangers, besides being simpler.

The combined analysis uses as data the estimated treatment-means of all the sites. The simplest analysis takes the form

Sites	S	$(s-1)$ d.f.
Treatments	T	$(v-1)$ d.f.
Sites × Treatments	S × T	$(s-1) \times (v-1)$ d.f.

There are s sites and v treatments; for the moment we assume all the experiments were done in the same season.

Where the same treatments occur at several sites there is a formal

resemblance to an experiment in randomized blocks, the sites taking the place of blocks. Consequently *F*-tests, *t*-tests, standard errors, etc., can all be found using familiar methods. Nevertheless the resemblance is a little superficial. In a randomized block experiment the 'error' sum of squares is usually that for the interaction of blocks and treatments, as was pointed out at (7.4.1). At Level (iii) the interaction of sites and treatments may itself be the subject of study. It is therefore helpful to have an 'error' for each site determined in the usual way. At this stage there are several possibilities. For example, Bartlett's test, described in Section 9.6, may suggest that they could well be pooled. Perhaps some other solution seems better. The point is that it is usually possible to find an 'error' variance with which to compare the interaction of sites and treatments.

Two examples may help to make this clear.

First, a series of variety trials may include some varieties resistant to a particular disease and some varieties susceptible to it. The incidence of the disease may vary widely from site to site. A contrast between a resistant and a susceptible variety will vary more between sites than a contrast between two resistant, or between two susceptible varieties. Second, in a series of factorial experiments testing N, P and K, the crop-response to N may well vary more between sites than the response to P. These considerations, which may rarely apply to variation between replicates on one site, make it necessary to examine the separate components of the sites × treatments sum of squares. In the second example given above we could calculate sums of squares for

N × sites
P × sites
K × sites
N × P × sites
etc.

We can assume that the true (population) values of the corresponding variances are different. (The opposite assumption is usually dangerous.) Then each main effect and interaction needs a separate test of significance (and standard error) based on an appropriate mean square. (In this example we might expect that at least some of the mean squares involving interactions of sites with interactions N × P, etc. would show no clear signs of heterogeneity, and this would justify pooling the corresponding sums of squares.) If in this example N was applied at three or more rates, it would be necessary to examine separately the site-to-site variation of the linear, quadratic, etc. contrasts of the main effect of N. This relates to the methods given in Section 9.6.

Next the analyst should look for any relationship between treatment-

contrasts (e.g. linear N) and the fertility of the site, measured by the general mean of all plots, or perhaps by the mean yield of plots without applied N; there are many possibilities. In a series of variety trials, it may be possible to distinguish between varieties that are more (or less) sensitive to differences in fertility between sites. If other data are available (e.g. soil pH, soil organic matter, incidence of disease), other useful relationships may be detected by appropriate regression analysis. An alternative approach to the assessment of 'sensitivity' is the comparison of the mean square for site differences calculated for each variety separately, but exact tests of significance are not easily derived by this method.

Finally, the tabulated contrasts should be carefully scanned, together with a table of characteristics of the sites, both permanent (e.g. altitude, soil type) and ephemeral (e.g. sowing date). Any relationship that is suggested should be reported, with the reservation that this is of the nature of a correlation and not necessarily a causal relationship.

Experiments over several seasons

If there are several experiments in each of several seasons the preliminary analysis of treatment-estimates of all experiments takes the form

Sites	S	$(s-1)$ d.f.
Years	Y	$(y-1)$
Sites × years	S × Y	$(s-1)(y-1)$
Treatments	T	$(v-1)$
Sites × treatments	S × T	$(s-1)(v-1)$
Years × treatments	Y × T	$(y-1)(v-1)$
Sites × years × treatments	S × Y × T	$(s-1)(y-1)(v-1)$

(s sites, y years, v treatments)

There is a strong resemblance to the analysis at (7.4.1).

Again this analysis can be repeated for any single treatment contrast, or sub-set of treatment contrasts. If the mean squares for T × S, T × Y, T × S × Y show strong evidence of heterogeneity the interpretation of the results is complicated; some relatively simple situations may, however, emerge. If, for example, T × Y is the largest mean square, it is likely that differences in weather are the most important factors determining treatment-effects, and prediction of effects in future seasons is correspondingly uncertain (unless weather can be predicted). If T × S × Y is the dominant source of variation this may be explained by variations in weather at different sites in any one season; again accurate prediction of further effects requires forecasts of weather—though which aspects of weather

(e.g. rainfall early in the season, temperature at anthesis, etc.) are important is difficult to determine. If forecasting future treatment-effects is the object, a rough working rule is to base tests of significance, etc. on T × Y or T × S × Y using the larger of the two mean squares, or, if they differ little, a pooled value.

Making the best of a bad job

If it is necessary to gather together information on, for example, the responses of a crop to N-fertilizer, and there are many experiments already done, each including a test of N but of varied design, some including other factors, an extension of the method described above may be used.

First, if different experiments test N at different rates, a response to a standard rate of N may be estimated for every experiment by use of a suitable fitted response-curve. Secondly, if some experiments include a factor (e.g. organic manure M) that is believed to have an interaction with N-fertilizer while other experiments do not, a response under standard conditions can be calculated and used in a combined analysis.

10.2 Rotation experiments

There are two main types of rotation experiment. They both involve one or more sequences of crops which can be repeated indefinitely; in the experiment the sequences—or perhaps there is only one—may be planned to run once, twice or many times. If the intention is to stop the experiment when the sequence(s) have run once, it is perhaps better to call the experiment a 'crop-sequence' experiment and keep the name 'rotation experiment' for experiments designed to run through the crop-sequence(s) at least twice.

The first type is designed to investigate treatments applied to one or more crops of a rotation, the same rotation being followed on all plots. For example, an organic manure and inorganic fertilizers could be compared in a rotation of wheat, potatoes and barley. The treatments might be applied cumulatively, year after year to each crop on the same plots, or they might be applied to the potatoes only, with residual effects measured in wheat and barley. Such an experiment may be in one phase, e.g.

1985	potatoes
1986	wheat
1987	barley

or it may have one or more replicates in each phase, thus:

Phase:	A	B	C
1985	barley	potatoes	wheat
1986	potatoes	wheat	barley
1987	wheat	barley	potatoes
1988	and so on.		

This more elaborate scheme needs more land and much more work, but it allows us to judge the 'reliability' of the results, i.e. how much the results of past seasons can be expected to hold true for future seasons, more quickly than the simpler scheme above.

The other type of rotation experiment is used to compare different crop rotations, often in conjunction with tests of other factors, for example organic manures and nitrogen fertilizer. An example is a test of the inclusion of a leguminous crop in place of non-legume, perhaps cowpea or groundnut in place of a cruciferous oilseed crop in rotation with cereals.

The range of types of rotation experiment is very great and we shall not attempt to deal with all of them, but almost all such experiments have features in common and here are some of them.

(1) Rotation experiments involve changes in soil properties. (If an experiment does not do that, it does not need to be continued on the same plots year after year.) Movement of soil between plots, therefore, whether by cultivation or by erosion, is a hazard even if the rate is small enough to be unimportant in a one-year experiment. This may indicate large plots with wide discard areas at their edges, or special precautions, e.g. grass paths between plots.

(2) The experimenter, together with the biometrician, must look ahead in time. Will it be possible to introduce a new variety of crop, or a deeper plough, during the experiment, if one becomes available and is favoured by growers? If so, when?

(3) Should spare plots, e.g. duplicates of the simplest cultivation system, or of the growers' commonest crop rotation, be included from the start, to be used if later experience suggests one or more new treatments that were not thought of or did not exist initially?

(4) Alternatively, should plots be made big enough to allow splitting them later for sub-plot tests of a factor or factors that may later be thought necessary, e.g. a test of nitrogen fertilizer in a legume versus graminaceous test, or a date-of-sowing test in a cultivation experiment?

(5) Should all phases be included, or one only, or is a compromise possible, e.g. 3 of the 6 phases of a 6-year rotation? If different phases are included, how do they start: with different crops in the same calendar year, or with the same crop in successive years? (One of the most difficult problems arises when rotations of different lengths have to be compared.)

The analysis of the results of a rotation experiment can be complex. For example, if an experiment runs through several rotations the yield of crop A on Plot *x* in year 1 and the yield of crop A on the same plot in the next rotation will probably be correlated. Normally one would expect the correlation to be positive, but the opposite might happen. Meanwhile the yield of crop B on Plot *x* in year 2 may be correlated, positively or negatively, with the yield of crop A in year 1. Mean crop-yield may show a trend with time, positive or negative, either because the weather is changing with time, or because new crop-varieties are introduced or because of new methods of control of pests, diseases or weeds. (It may just be that the crops are being managed with increasing skill.) Such trends may affect all treatments equally, or they may not. For example, better weather may have more effect on well-fertilized plots than on badly-fertilized ones, and deep cultivation may have cumulative effects.

If an experiment has run through several rotations a preliminary analysis should include, at least:

(a) mean of yields of treatments;
(b) mean values of each useful contrast between treatments (or means of treatments), with standard errors based on the variation of that contrast between years. If all contrasts of a set (e.g. linear and quadratic contrasts for 3 rates of fertilizer) have variation not significantly different, they may perhaps be pooled;
(c) for each contrast the trend, i.e. linear regression on year, with standard error calculated appropriately;
(d) an examination of variation of treatment-contrasts in relation to major differences in weather between years.

The main type of long-term experiment not included under these headings involves long-continued monoculture with tests of fertilizers or varieties or cultivations, etc.

10.3 Experiments with trees and bushes

Sources of variation

Crops grown on trees and bushes are perennial; experiments on such crops last for more than one season, in some cases for very many seasons. During this time the units—the trees or the individual bushes—increase greatly in size. There are also different sources of variability that change in importance (Pearce, 1960b). Three basic sources of variability are:

(A) that inherent in the material at planting;
(B) the different extent of shock experienced by each tree at planting;
(C) the effects of soil variation over the experimental area.

With tree crops, such as apples and pears in the United Kingdom, an experiment needs to be planned at least two seasons ahead, for the individual trees must first be raised in a nursery before they can be planted out into the land used for the permanent experiment. More trees are raised than will be needed; those that grow badly and seem weaker than average are discarded, but a random choice must then be made from the acceptable trees. Unless choice is random, any experimental results can be criticized as not being generally applicable. (In a botanical experiment intended to elucidate some point in physiology, closely standardized tree material may be required, but no-one should apply the result to practical agriculture, for which purpose representative plants are essential.) The initial variability (A) diminishes with the passage of time and can eventually be neglected.

It is also found that the variability described in (B) diminishes fairly quickly; if some trees are removed after a few years, leaving a more widely spaced experimental layout to form a second phase of the experiment, those trees that remain may again suffer something like (B) for a short time. The third source of variation (C) builds up steadily over time. Eventually it will dominate all other sources and can lead to higher variability than is usual with annual crops.

All this affects the selection of material in the nursery. In a short-term experiment, e.g. one to see if trees can be brought into bearing earlier, it is important to have trees closely graded as to initial size, though in that case it may be wise to regard the investigation as 'botanical' rather than 'agricultural'. When the first experiment is finished, the trees can perhaps be used for some other study using the methods to be described in Section 10.8. By that time (C) may have taken over and the initial selection of plants can be forgotten. In a long-term experiment, on the other hand, a wider range of initial size and growth may be completely acceptable, but trees with crooked stems and poor graft unions should be rejected because the cumulative effects of such defects build up over the years and lead to much the same results as (C).

The choice of design is likewise affected by the expected duration of the experiment. In a short-term study there is no need to go to great lengths to control local variation because (C) will not have the time to become important, but when the experiment is intended to go on for a long time a major aim should be to reduce (C) by careful choice of blocks, or, if necessary, the use of rows and columns, based on proper knowledge of the sources of soil variation (and climatic trends, etc.) through the experimental site.

Calibration

When a trial of a particular species is to last a long time, it is useful to be

able to calibrate, i.e. to find a measurement that will predict how crops will vary from one plot to another. That is best done as early as possible in the life of experimental trees, as soon as they are planted into their permanent field positions. With fruit trees one or two seasons' growth is usually allowed before different treatments such as fertilizers are applied; during this time, regular measurements of the circumference of the trees at a standard height above ground (the 'girth') may give a very good way to show how vigorously they are growing. That gives a good indication of future cropping, at least in early years. In the ordinary way calibration is effected by means of the analysis of covariance (see Chapter 8).

With this in mind there is a lot to be said for planting areas to which no treatments are applied initially but the trees are calibrated for future use. Indeed, that can be done very effectively if foresight is exercised. Thus an area could be planted choosing some varieties that grow quickly and others that take longer. As an example, a single unit-plot may contain four varieties laid out A B C D, which after the first phase is thinned to A C ; but that will work only if A and C pollinate one another (or are self-fertile). Alternatively,

A B C D
A B C D

may be thinned to

A C
B D

which keeps all varieties and gives a second phase in which trees are spaced as evenly as possible. (That, however, may not be desirable if B and D are much larger trees than A and C.)

If trees are likely to develop at widely different rates, the experiment can be designed so that the slower-growing varieties are in alternate rows: when the trees have grown enough for the original layout to become crowded, quite a lot of information will already have been discovered about the faster-growing varieties and they can be removed. The remaining layout then contains the slower-growing varieties, which were in alternate rows originally but now form the second phase of the trial at double the previous spacing distance. When planning an experiment in this way, care must be taken that all through the life of the trial any varieties that are not self-fertile will have adequate pollinator varieties remaining in the layout.

At thinning, the trees that remain are likely to suffer different amounts of shock. As a result it may be that no suitable calibrating measurement

can be found for the second phase of the experiment. Of course, if the contrasts sought are those between varieties, or if the treatments have to be applied before planting, calibration is difficult if not impossible, because the calibrating variate must not be subject to the effects of treatments. The reasons were given below (8.6.4).

When mature trees are taken over, perhaps on a commercial planting with no history of differential treatments, calibration by the total crop over the last few years can be very effective, but again a lot depends upon species. An example is given in Exercise 8C.

Guard rows

In a long-term experiment, roots can spread a long way, especially if some plots are given less fertilizer than is needed for healthy growth. Also, when different chemical sprays are used in an experiment, there will be drift through the air, and when trees are older that becomes a serious problem. So rows of 'guard' trees are needed to separate the experimental plots from one another. These are trees that are in neither plot, to left or to right, and they mark the edge of spraying application, the left side receiving a different pesticide from the right. Often they do not receive a spray, because of the danger of drift across into an experimental plot. If an experiment is to go into a second phase after thinning, careful design of the layout is needed to ensure adequate guarding during the second phase. Since guard rows are not in the experiment for measurement purposes, it is sometimes very useful to place in those rows any varieties needed for pollination of the experimental trees. The need for guard rows means that some of the land used is 'wasted' for experimental purposes, and the extent of this can be reduced by using unit plots that consist of several trees, perhaps a tree of each of three or four varieties; to use single trees as plots wastes more material in guarding.

Adding of treatments

Because tree experiments last a long time, and also because they take a while to establish before beginning experimental treatments in the first place, provision should be made wherever possible for using the material again for a second experiment with a new set of treatments, if the first experiment gives all its important information fairly quickly. (That topic will be considered in Section 10.8.) When a new problem is put to the research worker, it is useful to have some ready-made material available for an experiment immediately, because it avoids the delay while a site is prepared and material is set up. (This is also a reason for planting trees with no initial plan beyond their calibration.) Another reason for chang-

ing treatments can be that unexpected problems or effects occur in applying those chosen originally.

Residual effects on the land

When an experiment finally ends and the trees are uprooted, the effects on the soil last a very long time and will have to be taken into account in any future use of an experimental site. Such a site can sometimes be a useful asset to a research institute, but it can also impose annoying restrictions on its future use.

Experiments on bushes

Bush crops share similar problems to tree crops, though experiments on them will be of shorter duration and there will not be the same delay in getting them established. A useful calibration variate in the early seasons of an experiment is often the amount of wood growth made before cropping begins. However, variation in cropping can so disturb wood growth that it is no longer useful for calibration. In many bush crops, size will be regulated by pruning rather than thinning, so that experiments will not have more than one phase; but it may still be useful to change to a new set of treatments before the natural life of the bushes is over.

Recording of harvest

Harvesting usually needs to be done tree-by-tree, and it is essential to have individual trees labelled and numbered very firmly to avoid errors through loss of identity. Since the crop from each tree may be quite large, more than one day is often needed for harvesting the whole area; in case it spreads over several days—through weather delays or sheer volume of work—it is desirable to harvest a block at a time, and so tree experiments are nearly always designed to contain blocking in some form (randomized blocks, or row-and-column designs). When a crop ripens slowly, two harvesting periods may have to be undertaken to gather all the ripe crop.

Analysis of data

Analysis usually involves total crop per season, with perhaps, in addition, a division into top quality and the rest. The effect of seasons may be observed by looking for different patterns in the results of different treatments or varieties. This is often done by working out contrasts between seasons like those between treatments in Chapter 5. Really it is a matter of finding single degree-of-freedom effects that express important

features. To take an example, if there are six seasons the total crop is given by

$$(+1 \quad +1 \quad +1 \quad +1 \quad +1 \quad +1)$$

i.e. in each plot the six crops are added to give a variate, which is then submitted to the analysis of variance in the usual way. However, there might be a biennial effect and the question arises whether or not it is the same for all treatments. In that case

$$(-1 \quad +1 \quad -1 \quad +1 \quad -1 \quad +1)$$

can be dealt with in the same way. Then perhaps someone suggests that some of the treatments are leading to a faster increase in crop than others. Since biennial effects are suspected, it would be better to work with two-year periods and

$$(-1 \quad -1 \quad 0 \quad 0 \quad +1 \quad +1)$$

might be used, as might

$$(-1 \quad -1 \quad +2 \quad +2 \quad -1 \quad -1)$$

to see if the increase in crop was progressing steadily or was slackening off. We should note, however, that though these contrasts would be orthogonal if they were applied to the treatments of an experiment of the kind considered at (5.6.6), there is not the same orthogonality here. The successive crops, unlike the treatment means, are associated, but not in any definable way. Consequently, it is not to be expected that they will all give the same 'error' variance. However, that does not matter if each is to be made the subject of a separate analysis of variance, following the approach suggested in Section 9.6.

10.4 Bivariate analysis and bivariate diagrams

There are occasions when two quantities should really be studied together. For example, the heights and spreads of plants are usually highly correlated, sometimes so much so that there is no need to study both because each tells the same story as the other. Suppose, though, that the experiment concerns irrigation and that plants are beginning to wilt where the supply of water is low. In that case the heights will decrease while the spreads will increase. Taking the two variates separately, the effect of water may not be significant, but taken together the two changes, i.e. in

heights and spreads, may show up because they go against the prevailing positive correlation coefficient. To take another example; in general, plots that grow well will be found to crop well also, but what if the growth is a response to closer spacing and a need to find light? In that case the better growth on some plots could lead to less crop. The two effects may be non-significant when they are considered separately. Nevertheless there could be a marked effect when they are considered together. That is because the separate effects, which go in opposite directions, have to be seen against the positive correlation between the quantities (i.e. growth and crop) when competition is standardized. Such points have a special relevance in the study of intercrops, which will be considered in Chapter 11, but they apply in other situations also.

In dealing with an intercrop the usefulness of bivariate methods is obvious, the two variates being the yield of the two species. Sometimes the 'error' line shows little correlation between the two variates, but in practice it is hard to forecast the sign of the correlation. Some plots will be inherently better than others, and that will cause a positive correlation between the variates. On the other hand, the two species are in competition and that will cause a negative correlation. Either effect might dominate the other, or the two might approximately cancel. So, although there is no certain way of forecasting the degree or sign of the correlation, it would be most unwise to assume independence.

The problem then is this: there are two variates, a and b, which are to be analysed

(i) not separately as in Chapters 1 and 4,
and (ii) not with one serving to adjust the other, as in Chapter 8,
but (iii) together, giving equal weight to each.

To that end, a and b are transformed to two other variates, x and y, such that

(i) x and y have a variance of 1,
(ii) x and y are independent.

This is easily done. First, let

$$x = a/\sigma_A. \tag{10.4.1}$$

Now remove from b the component that could have been predicted from a. This leaves

$$b - \beta a.$$

Here β is the estimated regression coefficient of b on a. In (8.2.1) it is called

b but here the symbol has been changed to avoid confusion. It was mentioned at (8.3.5) that the sum of squares of such an adjusted variate is $S_{yy}(1 - r^2)$, where S_{yy} is the sum of squares before adjustment. Hence, here, the variance is $\sigma_B^2 (1 - r^2)$ and the standard error the square root of that amount. Dividing by that standard error gives

$$y = (b - \beta a)/(\sigma_B\sqrt{1 - r^2}) \tag{10.4.2}$$

The transformation being known, it is an easy matter to find x and y for any plot or treatment, given the values of a and b. The values of x and y so found can be plotted against one another on a sheet of graph paper in the usual way, but that possibility will be examined in the next chapter. The task here is to describe a bivariate analysis of variance.

It will be supposed that there are eighteen plots in six randomized blocks of three treatments, A, B and C. It will further be supposed that treatment means are:

	A	B	C
a-mean	21	24	30
b-mean	57	63	51

and the analyses of variance and covariance (calculated as in Section 8.3) are as follows:

Source	d.f.	a^2	ab	b^2
Treatments	2	7.00	− 6.00	12.00
'Error'	10	25.00	+ 10.00	32.00
Stratum total	12	32.00	+ 4.00	44.00

(10.4.3)

The standard error σ_A of a is $\sqrt{25.00/10} = 1.581$, so in this instance (10.4.1) gives

$$x = 0.6325a \tag{10.4.4}$$

The regression coefficient of b on a is $+ 10.00/25.00 = + 0.400 = \beta$. Also, the square of the correlation coefficient,

$$r^2 = (+ 10.00)^2/(25.00 \times 32.00) = 0.1250$$

and the standard error of b is $\sqrt{32.000/10} = 1.7889 = \sigma_B$.

Hence, (10.4.2) is now

$$y = (b - 0.400a)/(1.7889 \times \sqrt{0.875}) = 0.5976b - 0.2390a \qquad (10.4.5)$$

In general we may write

$$x = \delta a, \qquad y = \theta b + \gamma a \qquad (10.4.6)$$

where, in this instance, $\delta = 0.6325$, $\theta = 0.5976$ and $\gamma = -0.2390$.

A digression

Although it is not an essential part of the calculations, we shall derive the sums of squares and products for x^2, xy and y^2, because they will show so clearly why the transformations at (10.4.6) are useful. First, a sum of squares for x^2 must equal

$$\delta^2 \times \text{(the corresponding sum of squares for } a^2\text{)}.$$

Accordingly the analysis of variance for x^2 has 2.80 for treatments, 10.00 for 'error' and 12.80 for the stratum total. Similarly a sum of products for xy must be

$$a[\theta^2 \times \text{(sum of products for } ab\text{)} + \gamma^2 \times \text{(sum of squares for } a^2\text{)}].$$

Applying that result to the example, the sums of products for xy are -3.33 for the treatments and the stratum total and 0.00 for the 'error'. Finally, a sum of squares for y will be equal to

$$\theta^2 \times \text{(sum of squares for } b\text{)} + 2\theta\gamma \times \text{(sum of products for } ab\text{)} + \gamma^2 \times \text{(sum of squares for } a\text{)}.$$

In the example this gives 6.40 for treatments, 10.00 for 'error' and 16.40 for the stratum total. Writing the whole as at (10.4.3), that is

Source	d.f.	x^2	xy	y^2
Treatments	2	2.80	−3.33	6.40
'Error'	10	10.00	0.00	10.00
Stratum total	12	12.80	−3.33	16.40

It appears that the transformation has achieved its objectives of giving two variates, x and y, such that each has a standard error of one, with a zero correlation between them. Nevertheless, as has been said, this is a digression, not needed except as a demonstration that all has gone well.

The calculation resumed

From this point it will be helpful to write the analysis of variance of (10.4.3) in a more general form similar to that at (8.5.4):

Source	d.f.	a^2	ab	b^2
Treatments	f	$D_{aa} - E_{aa}$	$D_{ab} - E_{ab}$	$D_{bb} - E_{bb}$
'Error'	e	E_{aa}	E_{ab}	E_{bb}
Stratum total	$e+f$	D_{aa}	D_{ab}	D_{bb}

The next step is to work out the quantity Λ, where

$$\Lambda = \frac{E_{aa}E_{bb} - E_{ab}^2}{D_{aa}D_{bb} - D_{ab}^2} \qquad (10.4.7)$$

In this example, $\Lambda = 700/1392 = 0.5029 = (0.7091)^2$. The bivariate F equals

$$\frac{(1 - \sqrt{\lambda})(e - 1)}{\sqrt{\lambda} f} = \frac{0.2909 \times 9}{0.7091 \times 2} = 1.85 \qquad (10.4.8)$$

with $2f$ and $2(e-1)$ degrees of freedom, i.e. with 4 and 18. It does not appear that the treatment differences were significant at any important level, but perhaps that was not needed. The treatment means for x and y are readily found from (10.4.4) and (10.4.5), namely:

Treatment	a	b	x	y
A	21	57	13.3	29.0
B	24	63	15.2	31.9
C	30	51	19.0	23.3

The values of x and y can be plotted in the usual way with x measured along the horizontal axis and y along the vertical one. This will show that C does appear to stand away from the other two. It should here be explained that the bivariate F defined at (10.4.8) can be used for partitions

of the treatment line just as for the line as a whole. In the present example, If there were good prior reason to regard C as possibly different from A and B, it would be permissible to partition at (10.4.3) thus:

Source	d.f.	a^2	ab	b^2
A *v.* B	1	0.75	+ 1.50	3.00
AB *v.* C	1	6.25	− 7.50	9.00
Treatments	2	7.00	− 6.00	12.00

(10.4.9)

If we are to apply the F-test to the contrast of [AB *v.* C], we should first form the D-line by adding the line for the contrast to the E-line at (10.4.3), i.e. $D_{aa} = 6.25 + 25.00 = 31.25$, $D_{ab} = -7.50 + 10.00 = 2.50$ and $D_{bb} = 9.00 + 32.00 = 41.00$, which makes

$$\Lambda = \frac{700}{1275} = (0.7410)^2$$

so

$$F = 3.15$$

with 2 and 18 degrees of freedom. Significance is now higher, though it still has not attained the level of one in twenty.

If the treatment points are to be plotted on a bivariate diagram in the manner suggested, it would be a convenience to have a quantity corresponding to the least significant difference of univariate analysis. Here a quantity is found equal to

$$t\sqrt{K \text{ Error m} - \text{s}}$$

where t has its usual meaning and K is the constant defined at (4.1.1) and evaluated in various ways in Chapter 5: Then if two treatment means differ by more than that quantity, they are said to be significantly different. In bivariate analysis, a treatment point having been found, a circle can be drawn round it with a radius of

$$\frac{2KF'(e-1)}{e} = L \qquad (10.4.10)$$

where K is as for a univariate analysis and F' is the actual value of F with 2 and $2(e-1)$ degrees of freedom for the significance level required. Then, if two points are so close that the distance between them is less than L, treatments can be said not to differ significantly. However, least significant differences need to be used with discretion in univariate analysis, and the same reservations apply in bivariate cases.

10.5 Fan trials

Spacing trials present special design difficulties. There are two in particular. One concerns the need for plots to be large if they are all to have the same area and each is to contain an exact number of rows.

For example, a comparison of inter-row spacings of 1.0 m, 1.5 m and 2.0 m requires a plot 6 m wide to accommodate respectively six, four and three rows. Another problem concerns guarding. Plants at different spacings give rise to different micro-climates. Further, they throw shadows of wind, rain and sun. A plot adjacent to one closely planted may be more sheltered and less moist and with less light than it would have been if it had a widely spaced neighbour.

For those reasons many people use fan trials. The idea is to plant a series of 'spokes' that radiate from a 'hub', which will ordinarily be outside the experimental area. An example is given in Fig. 10.1. There is a constant angle, θ, between adjacent spokes. On each spoke the distances of plants from the hub are the same, the innermost being at a distance D and the outermost at $D + L$. (Note that L is the length of the spoke in the sense of being the distance between the extreme planting locations. The end plants will each need further space to grow in.)

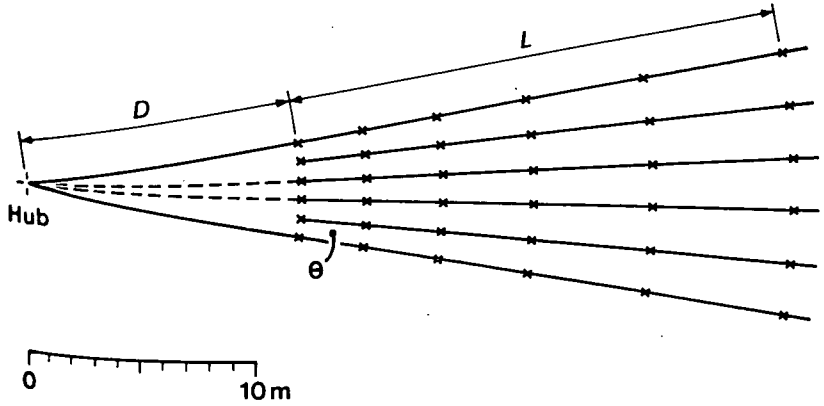


Fig. 10.1 Scheme of a fan trial

Fan trials are of many kinds. The chief distinction is between those intended to study (a) inter-row distances, or (b) the area that a plant needs for good development (A), or (c) rectangularity (R). The last needs a little explanation. By "rectangularity" is meant the ratio of the distance between plants within a row to the distance between rows. It will be seen that R and $1/R$ are the same: the difference is just a matter of the direction in which rows are taken, either along the spokes or across them.

Since plants on the outside spokes suffer less competition, only inside spokes can be used and, even with them, the end plants must be discarded. A plot consists of those plants on inside spokes that are the same distance from the hub. (In Fig. 10.1 there are four plants to each of the four plots.)

To return to kinds of fan trials, if the aim is to compare inter-row spacings, the plants are placed at a uniform distance apart along each spoke, and there are no special difficulties of design. If the intention is to compare areas, rectangularity being kept constant, each plot is an enlargement of its neighbour towards the hub, being C times longer and C times wider, as in Fig. 10.1. Finally, if the intention is to compare rectangularities, areas being kept constant, each plot is C times longer than its inner neighbour but only $1/C$ times as wide, the rectangularity being therefore changed by a factor of C^2. This last sort is rather unusual and will not be discussed further here.

The important case is the second, i.e. the one in which rectangularity is preserved but areas are changed. More specifically, let there be N areas, covering a range from A_1 to A_N. There will in consequence be $(N+2)$ plants on each spoke. The dimensions of the fan can be worked out from four geometrical relationships, of which the first is a slight approximation. They are:

$$L = \frac{2(C^2\sqrt{A_N} - \sqrt{A_1})}{\sqrt{R}(C^2 - 1)} \qquad (10.5.1)$$

$$2(N-1)\log C = \log(A_N/A_1) \qquad (10.5.2)$$

$$D = \frac{2\sqrt{A_1}}{\sqrt{R}(C^2 - 1)} \qquad (10.5.3)$$

$$\theta \text{ in degrees} = 28.65\ R(C^2 - 1)/C. \qquad (10.5.4)$$

The base of the logarithms in (10.5.2) is immaterial. In the example that follows the base will be 10, as is usual in most non-mathematical work.

To take an example, someone may want a fan to explore the range from 3 m^2 ($A_1 = 3$) to 15 m^2 ($A_N = 15$) with a constant rectangularity of 3 ($R = 3$ or 1/3). There is a constraint that L is not to exceed 32 m. (The example has been chosen for its awkwardness.) From (10.5.1) either

$$32 = \frac{2(3.873C^2 - 1.732)}{1.732(C^2 - 1)} \quad (R = 3)$$

or

$$32 = \frac{2(3.873C^2 - 1.732)}{0.577(C^2 - 1)} \quad (R = 1/3)$$

That is to say, C should be about 1.044 or 1.183 according to the chosen value of R. The first is rather small, so the second looks more hopeful. Note, however, that any change from 1.183 must be made cautiously. To use $R = 1/3$ and $C > 1.183$ will lead to $L > 32$.

From (10.5.2) several possibilities appear. In this instance, $(N-1)\log C = 0.3495$ so the choice lies between

$N = 3$	$C = 1.495$
$N = 4$	$C = 1.308$
$N = 5$	$C = 1.223$
$N = 6$	$C = 1.173$

The last looks promising, though deceptively so. To raise C to 1.183 is, from (10.5.3), to increase A_N/A_1 and that will mean a larger value of C to satisfy (10.5.1), which will in turn require a higher value of A_N/A_1. Perhaps a solution can be found along those lines, but it might be better to retreat to $N = 5$, $C = 1.223$ and see what happens there. It appears from (10.5.3) that $D = 12.10$ m and $\theta = 3.87°$. Planting should take place at distances, D, DC, DC^2, ... DC^6, from the hub, i.e. at 12.10 m, 14.80 m, 18.10 m, 22.13 m, 27.07 m, 33.11 m and 40.49 m ($D + L$). That puts L equal to 28.39 m (= 49.49 − 12.10 m), which is less than was intended. If C increases above 1.223, the value of A_N/A_1 will be increased. It could be argued that the ratio of A_N/A_1 of 5 to 1 is quite large enough so an alternative would be to reduce it and seek a solution for $N = 6$. One possibility is $A_1 = 3$ m^2, $A_N = 13.5$ m^2, $C = 1.625$, $D = 17.07$ m, $L = 31.91$ m.

Another useful expression gives the area of the mth plot. It is a generalization of (10.5.3). Because

$$A_1 = \tfrac{1}{2}D^2R(C^2 - 1),$$

$$A_m = A_1C^{2(m-1)} = \left[\frac{D(C^2 - 1)}{2}\right]^2 RC^{2(m-1)} \qquad (10.5.5)$$

Such then are the dimensions of the desired fan, but transferring them from a piece of paper to the field is not easy. The best way is to take one spoke as the base-line and to plot the positions of the end plants on all spokes with precision relative to that line. The task can be accomplished either by trigonometry or alternatively, the whole can be drawn carefully to scale on a large piece of squared paper with the base-line along one of the printed graduations. The important co-ordinates can then be read off. Once the end-points of the spokes have been marked in the field, there is no great difficulty with planting.

The fan will need to be replicated. Some people use complete circles. Certainly that avoids the difficulty of a single fan. If the wind blows into it, this funnelling effect can lead to damage at the narrow end; if the wind blows in the opposite direction, it is divided and the whole fan can be sheltered by the closely planted end. Another idea is to use fans in pairs, side by side, the narrow end of one adjoining the wide end of the other. A second pair at right angles to the first is valuable.

However the fans are disposed, the aim is to find the spacing that gives a maximum of characters like growth and cropping or a minimum of those like disease incidence. The appropriate statistical technique is therefore that of regression rather than the analysis of variance. Fans with different orientations may give different results, which is a reason for studying data from each fan separately, at least initially.

Some workers have found that crop-weights and plant-weights give closer regression relationships when in reciprocal transformation, i.e. when using $1/W$ rather than W. Also, it is better to transform the areas per plant (A_1, A_2, etc.) to plants per unit area (a/A_1, a/A_2, etc.) when seeking a quantity upon which to regress.

10.6 Systematic designs

Spacing trials are not the only sort where extreme treatments should not adjoin. Fertilizer trials are the same. If two treatments, one of high application and the other of low, come together on adjacent plots, there will be robbing as roots from the starved plants find the riches nearby and develop where there is fertilizer. The obvious solution is to leave a guard area between the two plots and to give it an intermediate level of fertilization, but that reduces the problem without eliminating it. Also it wastes space.

If there is only one factor, another possibility is to allot the treatments to plots within a block in ascending (or descending) order of level of application. In that way robbing is kept to a minimum. Further, if the plots are long and narrow and if they adjoin on their long sides, the experimenter can see the response surface before his eyes. There would, of course, have to be several blocks; otherwise the apparent response curve could be the result of a local trend. The only design problem remaining is the allocation of orders (i.e. ascending or descending) to the blocks. Some would randomize; others argue that randomization has been abandoned anyway and they would allocate the two orders alternately to successive blocks. If use is to be made of the method of analysis to be described below, the random approach is better.

The analysis of data is best approached in this way. First, we decide which features of the response curve are important. Here the likely answer is that we should consider linear and quadratic effects as described in Section 5.4. Others may want to carry on to later contrasts, like the cubic, while yet others may have ideas that are quite different. Whatever the answer, the next step is to calculate the desired quantities, q, for each block separately. The rest of the calculations are given in Section 9.6. If the 'error' sum of squares is $(b-1)s^2$, the variance of the mean value is s^2/b where b is the number of blocks. A mean of those values will give an estimate of the quantity under study. The sum of squares of that mean will be given by

$$s^2 = (q_1^2 + q_2^2 + \ldots + q_b^2) - (q_1 + q_2 + \ldots + q_b)^2/b.$$

The mean square will be $s^2/(b-1)$ and the standard error of the mean $\bar{q}$ will be the square root of $s^2/[b(b-1)]$, which is easily found and can be used either to show how well the mean $\bar{q}$ has been estimated or to carry out a t-test. (It is assumed that the null hypothesis implies that the values of $\bar{q}$ have a mean of zero.)

With two factors the situation is more complicated. Plots have to be small and nearly square, if not exactly so. The levels of one factor are applied in either ascending or descending order to the rows, and levels of the other factor are applied similarly to columns. The result is the growing of a response surface. Again the contrasts described in Chapter 5 can be used to estimate single degree of freedom effects, whether of main effects or interactions, and the replication in blocks will enable standard errors, t-values, etc. to be found. When randomizing there are four possibilities, because both rows and columns need to be assigned either an ascending or descending order. Again, the method of analysis is that of Section 9.6.

Serially balanced designs

A type of design intermediate between randomized and systematic has been used in studying powdery mildew, a fungal disease of the leaves of barley (Dyke and Shelley, 1976). It could be useful in other circumstances also. It is believed that the development of the disease in a plot of normal

size may be substantially influenced by the treatment applied to a neighbouring plot; thus a plot downwind of an untreated plot will receive more spores than one downwind of an effectively sprayed plot. Similar considerations probably apply to experiments on many mobile diseases and pests of field crops. Most of the designs used in this work had four treatments, A, B, C, D, and used 38 plots in one line. They are constructed as follows:

Treatment A is tested in 9 situations:

BAB, BAC, BAD, CAB, CAC, CAD, DAB, DAC, DAD.

These, together with the other three treatments in the corresponding sets of situations, can be combined in a design such as

A, BCAD, ABDC, DBAC, ACBD, BDAC, DCAB, ABDC, BCDA, DCBA, B

(Commas indicate blocks, not breaks visible on the land.) This design has the additional property that, apart from the first and last, the plots fall into nine 'blocks' of four plots each, containing a replicate. The extreme plots are needed only as 'neighbours' and their yields need not be recorded. Although, of course, such a design is not fully randomized, it is one of a large sub-set (at least 1566 in number) of all possible randomizations.

This type of design does not include any treatment with the same treatment as its neighbour; to include these combinations would bring problems; for example:

(1) the design could not be divided into 'blocks' each containing a replicate of A, B, C, D;
(2) the larger source of spores from two or (three) adjacent unsprayed plots might appreciably influence plots at a distance.

The intended analysis ignores the division into 'blocks' (though this may be recognized in a 'post mortem' study of 'error' variances). The yields are swept for the effects of treatments applied 'direct' (i.e. to the plot under consideration) and successively for effects of treatments to the left-hand neighbours (e.g. B *v.* C *v.* D as neighbours of A), 8 d.f. in all; and similarly for right-hand neighbours. The sum of squares of the residuals with $35 - 3 - 8 - 8 = 16$ d.f. (which is based on the three-factor interaction) provides an estimate of 'error'.

10.7 Nearest neighbour methods

A long time ago, in 1937, Papadakis made a suggestion that seemed full

of promise but even now few would feel certain about it. What he said was this: each plot has a total residual (Section 7.13) which indicates its fertility. Since good and bad land comes in patches, it should be possible to take each plot in turn and assess its potential performance, x, from the total residuals of its neighbours: Then when an analysis of variance was carried out on its actual performance, y, it should be possible to adjust by covariance upon x.

The difficulty with this suggestion comes from the mathematical complexities of adjusting y by a variate that is itself derived from y. Everything depends upon the correlation between the performance of adjacent plots, and that is a matter about which only the foolish dare to assert anything with confidence. Those who know about fertility patterns in fields will question almost any law supposed to be of universal application.

However, an illustration will show the method. It will represent data from a design without blocks but, if there had been any, they would have been ignored.

A	26	C	31	B	28	
C	29	A	32	B	28	(10.7.1)
B	16	C	21	A	17	

The treatment means are A, 25; B, 24; C, 27.
Hence the total residuals are:

A	+1	C	+4	B	+4
C	+2	A	+7	B	+4
B	−8	C	−6	A	−8

In the absence of any special reason to the contrary it is usually best to estimate the potential performance of each plot from the total residuals of its neighbours, both in rows and columns, in so far as they exist, i.e. x is

A	$(+2+4)/2$	C	$(+1+7+4)/3$	B	$(+4+4)/2$
C	$(+1+7-8)/3$	A	$(+4+2+4-6)/4$	B	$(+4+7-8)/3$
B	$(+2-6)/2$	C	$(-8+7-8)/3$	A	$(-6+4)/2$

which is

A	+3	C	+4	B	+4	
C	0	A	+1	B	+1	(10.7.2)
B	−2	C	−3	A	−1	

The figures at (10.7.2) show a clear fertility pattern from low potential at the bottom to high potential at the top.

It is now a question of adjusting y at (10.7.1) by x at (10.7.2). There is a point to be noted here. Because x is derived from y, the usual rules for finding error degrees of freedom in the analysis of covariance do not apply. If used, they will overestimate the true number by a quantity a which is difficult to estimate but lies between 0 and 1. It is therefore wise to play for safety and take a as 1. That means allowing two degrees of freedom for the regression instead of one.

In this instance, the calculations go like this:

Source	d.f.	x^2	xy	y^2
Treatments	2	0.89	− 2.33	14
Error	6	50.67	+ 88.00	266
Stratum total	8	57.56	+ 84.67	280

leading to

Source	d.f.	s.s.	m.s.	F
Treatment	2	42.28	21.14	0.75
Error	4	113.17	28.29	
Stratum total	6	155.45		

Such then is the method. How valid is it? Most authorities would agree that the analysis of data in the form just given leads to results not far from the truth. How effective is it? Here a lot must depend upon circumstances. With newly planted trees the variability has been brought over from the nursery, and there are no correlations between neighbouring plots to make use of. On the other hand, there are times when the variability is due chiefly to the effect of patchy soil and the method could well be very effective.

A study was carried out to compare the method of Papadakis with randomized blocks (Pearce, 1978, 1980).

The general conclusion was that the two methods were of approximately equal effectiveness, except where a mistake had been made in placing the blocks across fertility contours instead of along them. Then the method of Papadakis, which does not depend upon orientation, was better. Row-and-column designs were in the main better than the use of Papadakis, but, where they fail, they fail disastrously; whereas the nearest

neighbour approach, which sometimes is ineffective, never does much harm. If there are no correlations between the performances of neighbouring plots, no advantage comes from the covariance adjustment and two degrees of freedom have been lost from 'error', but that is all. The method of Papadakis can at least be recommended as safe. That is not to say that it has no worrying features. It is, for example, quite unable to deal with a discontinuity in fertility like that caused by differential former crops, whereas if the position of the break is known, blocks can be formed accordingly. If, on the other hand, the position of the discontinuity is not known, a block design is equally unable to cope. In any case, blocks may be needed for purposes of administration.

There are several points still to be made. One concerns 'iteration'. The difficulty is this. From (10.7.1) the means of A, B and C are respectively 25, 24 and 17. After adjustment by covariance they are 24.6, 23.6 and 27.8. Should we not start again and calculate (10.7.2) afresh and go on round the cycle repeatedly until everything settles down? The question is a reasonable one. The analysis as it stands is inconsistent. It is based on certain values for the treatment means, which it then contradicts. Iteration has raised so many questions in the minds of the mathematicians that it is better avoided, at least for the present.

Others have argued that it would be better if the treatments could be dispersed more, so that two neighbouring plots were never treated alike. Some possible designs are called 'complete Latin squares'. The objectors are possibly right, but it is too early to say definitely.

It may be remarked that if anyone was designing an experiment with the intention of using Papadakis's method of analysis, there is a lot to be said for using long narrow plots, placed side by side so as to encourage a large correlation between adjacent plots. In that way there would be a single strip of plots, which were themselves placed across the strip. The mathematics of the one-dimensional model gives more confidence than that for the two-dimensional in which plots adjoin both at their ends and their sides. Also, it reduces difficulties about missing neighbours at the edges and corners. In the one-dimensional case only the end plots need be considered and it would not be difficult to add two extra plots to provide total residuals with which to assess x for the end plots in the experimental area, as was done in Section 10.6 with the serially balanced designs. The one-dimensional case could also be useful with 'contour blocks' which wind round a hill, giving plots that are admittedly all at the same altitude but very variable with regard to aspect.

Finally, there is a development associated with the name of Wilkinson, which is probably more widely used than that of Papadakis, but it will not be described in detail here because little has been published about it in practice. Essentially it consists in using moving blocks of three plots, the

performance of the treatment on the middle plot alone being assessed, the block then passing on one plot for the assessment of the next 'middle treatment'.

10.8 Changing the treatments of an experiment

Sometimes the treatments are applied and their effect can be assessed soon afterwards. This happens, for example, when sprays are used to check the spread of a fungal infection on leaves. Sometimes too the plants are long-lived and will continue to develop well after the time that a single fertilizer application or a spell of irrigation has spent its force. In such circumstances the experimenter may wish to use the same plants for a second investigation. (With spraying experiments on trees, there could be need of repeated changes of treatments.)

There are three possibilities to be considered.

(a) The treatments have had their respective effects and the plants have become uniform again. That could well be the case, for example, in a trial of insecticides. After a time rain will wash spray residue off the leaves and there will be a migration period during which a new generation of mites will disperse itself. Provided there has been no serious damage to the plants, either from sprays or insects, no special considerations apply if someone wants to use the plants for a further experiment.

(b) There is a residual effect of the first set of treatments, but it is unlikely to interact with the fresh set of treatments to be applied. That is so, for example, when the first set of treatments is no longer operating, the passage of time having removed spray residues. There could still be different levels of insect population or disease infection or plant growth that would need to be allowed for.

(c) The residual effects of the old treatments can be expected to interact with the new ones to be applied. To take an example, any fertilizer regime, if it is continued for long enough, will leave soil differences. A later experiment on the same plants, or even on the same site with different plants, may have to be designed with those former treatments in mind.

As has been said, Case (a) presents no special statistical problems. Case (b) is more difficult though not impossibly so. For example, suppose that the first experiment was in randomized blocks, like this:

I	A	B	C	D
II	A	B	C	D
III	A	B	C	D
IV	A	B	C	D

If there were four new treatments, α, β, γ and δ, they could be disposed in a Latin square. If there were only three, or if there were five, designs like those at (4.12.1) and (4.12.2) are available. The only difference lies in the two prior classifications of plots being by blocks and previous treatments instead of by rows and columns, but that is a change of name rather than a logical difference.

To take another example, if there had been five treatments, i.e.

Block						
	I	A	B	C	D	E
	II	A	B	C	D	E
	III	A	B	C	D	E
	IV	A	B	C	D	E

and someone wanted four new treatments, they could be applied using a design of Type O:TO (Section 4.11) like this:

Block										
	I	A	α	B	δ	C	γ	D	α	E β
	II	A	γ	B	α	C	β	D	δ	E γ
	III	A	β	B	β	C	δ	D	γ	E α
	IV	A	δ	B	γ	C	α	D	β	E δ

If there had been five new treatments, this would have been possible:

Block										
	I	A	δ	B	β	C	α	D	ε	E γ
	II	A	β	B	δ	C	ε	D	γ	E α
	III	A	α	B	ε	C	γ	D	δ	E β
	IV	A	ε	B	γ	C	β	D	α	E δ

It is of type O:OT (Section 4.11). There is no need to go on. An ingenious designer can often find a way. It is true that in all the designs illustrated the number of treatments in the second phase is not very different from the number in the first, but that is not a bad thing because it implies that the degree of replication also will be about the same. If a good choice was made for the first phase, little or no change in the second is to be desired.

The third case (c) can be the hardest. Where interactions between the two sets of treatments are to be expected, one possibility is to see whether the original plots can be split to take the new treatments. Sometimes a different situation arises. Suppose, for example, that an experiment is in progress and someone suggests that the treatment effects, already apparent, would not be there if the soil were irrigated. As a result, water is supplied to some blocks but not to others. The outcome is again a design in split-plots. (The original blocks will have become plots and the original plots will have become sub-plots.) It is true that there will only be a poor

comparison of the irrigation treatments, but that will not matter if they have been introduced only to provoke an interaction.

With a 2^k design there is an easy way of adding another factor. If there is an even number of blocks, each containing a complete set of treatments, the additional factor can always be added by confounding an interaction, e.g.

Block	I	(1)	A	B	AB
	II	(1)	A	B	AB
	III	(1)	A	B	AB
	IV	(1)	A	B	AB

can readily become

Block	I	C	A	B	ABC
	II	(1)	AC	BC	AB
	III	(1)	AC	BC	AB
	IV	C	A	B	ABC

There is, however, the difficulty that the confounding of the highest-order interaction, here A × B × C, inhibits the addition of yet another factor if that should be needed. (The reason is simple. If a further interaction is to be confounded, it cannot be chosen so that both it and its generalized interaction with A × B × C will be of high order.) In the example, which is of a small experiment, a fourth factor is unlikely. (Indeed, three seems rather a lot.) Nevertheless, with more plots it is better not to confound the highest-order interaction, if that can be avoided and if there is much chance of yet more factors being needed.

10.9 The size of an experiment

Experimenters sometimes ask how large their experiment should be and are bewildered when the statistician replies that he has no means of knowing. On the face of it the question is purely quantitative and people suppose that there must be a formula somewhere to give the answer. In fact, a number of considerations have to be taken into account and most of them are non-statistical.

We must start with the expression at (4.1.1). If we are interested in a certain contrast and if we need to estimate its value with a standard error, S, then $S^2 = Ks^2$, where K is a constant derived from both the contrast itself and the design adopted. (All this has been explained in Chapter 5.) Also, s is the estimated standard error of a single observation. It is usually determined by putting s^2 equal to the 'error' mean-square of the analysis of variance.

We must now ask how large S should be. If we are concerned solely with estimation, we probably know the answer to that question. Hence, assuming that we know the value of s, the problem is solved by proposing a design with an appropriate value of K. At least, it is solved if we can rely upon our estimate of s, but mostly we cannot. Some sites are more variable than others. Even if we do know what value of s is usually found, we may still be lucky or unlucky with our particular set of data. Also, we must be careful how we insert blocks (or rows and columns) because we can make things a lot better if we do it well, but we can make things worse if we do it badly. (We have already suggested in Section 10.7 that one advantage of nearest neighbour methods lies in their giving more consistent estimates of s.)

At this point we emphasize how important it is to keep a catalogue of standard errors. Whenever an analysis of variance is concluded, someone should make a record of the square root of the 'error' mean-square along with notes of location, plot size, block size and any attempts to reduce s by a covariance adjustment or similar device. (It is also useful to know the number of degrees of freedom upon which each value of s is based. If that is not recorded, a few high but badly determined estimates may be given undue weight.) As the catalogue lengthens, patterns will begin to appear. Further, if large values are followed up by a 'post-mortem', or at least by an evaluation of the residuals, errors of judgement will be disclosed and mistakes avoided for the future. Without such a catalogue the statistician cannot advise on the size of an experiment unless he has had considerable experience of that crop in those circumstances. Even then he can possibly only guess.

However, we will suppose that somehow we can make a fair estimate of our likely value of s. Turning to testing, we have first to ask how large the value of the contrast must be before it is regarded as important. The question is vital. We do not want to expend resources and energy on an experiment that shows a contrast to be significant if it is so small that no one is going to make changes to secure so trivial a gain. Equally we do not want to find a difference so large that it would revolutionize local agriculture if it could be relied upon, but is so poorly determined that we cannot be sure that it is really genuine. What sort of difference would we be interested in? We will suppose that we receive an answer to the effect that the value of the contrast should exceed some value, D. We next have to consider what significance level is called for. Perhaps the contrast is almost certain to exist and we need little confirmation to convince everyone; perhaps there are grave doubts about it and our evidence will have to be really strong. We shall probably end by following convention and decide on a significance level of $P = 0.05$ because people are very conservative in the matter. Now that we know P we can make a fair guess

at t, which depends also on the number of degrees of freedom for 'error'. That enables us to design an experiment such that

$$D = tS = t(K)^{\frac{1}{2}}s,$$

i.e. such that

$$Ks^2 = D^2/t^2. \qquad (10.9.1)$$

Unfortunately that is not the answer. We do not know s perfectly. We may indeed be able to estimate it without bias; in that case the actual figure may be above our estimate or below it. In fact, we have given ourselves a 50 percent chance of missing the contrast, even if it exists and its value does equal D. At this point we have to ask a further question. Supposing that the contrast does have the minimum value at which it becomes important, how ready are we to miss it? What probability, P', would we be willing to accept? The immediate response may be to insist that in such a case the contrast must be detected with certainty, but that is to set an impossible standard; also, it ignores the fact that we are talking about the critical value, i.e. the point at which people might begin to take note of it. A more reasonable response might be to take P' to be 0.1, but there is no universal answer that will satisfy everybody. Anyhow, the choice of P' finally identifies the problem.

Let us consider the situation carefully. If the true value of the contrast is D the estimates we obtain of it will be distributed about that value with a standard error of S. In deriving (10.9.1) we aimed at the middle of that distribution and arrived at a solution in which half the values lay above the point we hit and half below. What we want is to hit the distribution at a point such that 0.9 lies above and 0.1 below. That produces a situation akin to a one-sided test and leads us to aim not at D but at $(D - t'S)$, where t' has been selected for a probability of $2P'$. In fact, we want S such that

$$D - t'S = tS.$$

Hence (10.9.1) should be replaced by

$$Ks^2 = D^2/(t + t')^2. \qquad (10.9.2)$$

To take an example, we may decide that s is likely to be about 10 and that we can expect to have about 20 degrees of freedom for 'error', the exact figure being known when the design has been chosen. We are given P as 0.05 and P' as 0.1 and we are told that D, the critical value of the

contrast, is 20. How do we proceed? For 20 degrees of freedom t for $(P = 0.05)$ is 2.086 and t' for $(P' = 0.20)$ is 1.325. Hence we want a design such that

$$10^2K = 20^2/(2.086 + 1.325)^2 = 34.4,$$

i.e., K should be about 0.344. Hence, if the contrast is a straightforward difference of two means, an orthogonal design with six replicates could be indicated. That makes K equal to 0.333, which is rather better than is required though not by much.

Several conclusions can be drawn from (10.9.2), of which the most important is the high cost of improving precision by increasing the replication. An experimenter may make a practice of using four replications. One day he decides to carry out a really precise experiment so he increases the number to five. In that way he will multiply the variance of a contrast by 0.8 $(= 4/5)$ or thereabouts, and its standard error by the square root of that amount, i.e. by 0.89. He has therefore obtained an 11 percent reduction in standard error at the expense of doing 25 percent more work. It is true that he will have more degrees of freedom for 'error', but that may not be of great importance.

The formula at (10.9.2) also draws attention to the way in which K and s^2 should be considered together. In all our discussion of non-orthogonality we have had to face the fact that K for a non-orthogonal design is never less than K for one that is orthogonal with the same replication. (Sometimes non-orthogonality will permit unequal replication, which could be to the advantage of certain contrasts. This happens with supplemented balance and is a special case.) The reason for adopting a non-orthogonal design is almost always that it enables the blocking system to fit the land better in the hope that the reduction in s^2 will more than compensate for the increase in K.

If an experiment were intended to examine one contrast for one variate, the expression at (10.9.2) could be used to decide the design, but usually there are several contrasts and several variates and this can raise some difficult decisions. In most factorial experiments it is important to have good estimates of the two-factor interactions. If that is arranged, other contrasts may be determined with greater precision than is really required. To take another example, an experiment may be intended chiefly to determine increase in crop. Then someone wants to take insect counts as well and that variate will be much less uniform. On the other hand, small treatment effects will not matter, so perhaps that is all right, but what if someone wants to study the proportion of blossoms open at a certain time—this can be an awkward variate. Further, he insists that a small difference could be important. It may be necessary to say that the

experiment is not designed for such precision and that the attempt had better not be made. This may cause disappointment at the time but that is better than disappointment when the data have been collected. Most experimenters are tempted to measure a lot of supplementary variates. On general grounds that should be encouraged, but it should not be taken to extravagant lengths. It does however provide an argument for being generous when the size of the experiment is decided. There is another reason too. As Chapter 12 will show, the loss of information from mishaps can be out of proportion to the degree of damage. If accidents are at all likely—and they are always possible—the experiment should not be of minimal size but should have some excess capacity.

Exercise 10A

A maize fertilizer trial was carried out at eight sites on the island of Antigua in the West Indies. The complete set of treatments was rather complex, but by selection of data it is possible to extract a 2^3 factorial design from each site with factors N (Nitrogen), P (Phosphates) and K (Potassium). The three-factor interaction was confounded, there being two blocks with Treatments (1), PK, NK and NP, the other two containing N, P, K and NPK. The data represent weight of good ears in kilograms from an area of 144 square feet.

At Site 3 (Thibou's Estate) the trial suffered damage from animals and is here disregarded. At the other sites treatment means were:

Site	(1)	N	P	NP	K	NK	PK	NPK
1. Friars Hill	3.59	4.94	3.33	5.63	2.09	3.84	2.37	5.08
2. Lower Friars Hill	2.55	4.49	3.26	5.17	1.30	5.15	1.75	5.56
4. Wood's Estate	4.26	4.26	4.85	5.38	3.17	2.98	4.19	4.54
5. Clare Hall	2.12	1.98	2.58	1.73	2.01	2.01	2.77	2.06
6. North Sound	1.53	2.20	2.41	3.20	2.04	2.21	2.03	2.31
7. Orange Valley	4.11	5.05	4.58	4.79	2.80	3.82	4.90	5.20
8. Old Road	5.90	7.83	4.85	6.88	5.94	6.90	4.85	6.97

The respective error mean squares (s^2), each with six degrees of freedom, were:

1. Friars Hill	0.5649	2. Lower Friars Hill	0.2996
4. Wood's Estate	0.5925	5. Clare Hall	0.1405
6. North Sound	0.2154	7. Orange Valley	0.9903
8. Old Road	0.9305		

You are asked to assess the effect of the three elements on maize yields in Antigua, noting especially any interactions of one element with another and of elements with sites.

Note: Anyone who examines the original data and works out the treatment means will find that they do not agree with those given above. What has happened is this. Since it has been assumed that there is no three-factor interaction, the computer has adjusted the treatment means to make the value of that interaction equal to zero, but it has done so in such a way that the values of the main effects and the two-factor interactions are unchanged. Thus, for Friars Hill the value of the three-factor interaction is

$$3.59 - 4.94 - 3.33 + 5.63 - 2.09 + 3.84 + 2.37 - 5.08 = -0.01$$

which is correct within the limits of rounding error, but other contrasts are unaffected. (Computers usually do that, so the warning may be helpful.) If the experimenter really believes that there is no three-factor interaction, the revised figures are an improvement because they make use of the additional information.

A warning should be given about the difficulties that arise if there could be an interaction, but the experimenter takes a chance and confounds it nonetheless. If the three-factor interaction may exist, difficulties arise whether the adjustments are made or not, because interpretation depends upon knowing about it and there is no way of finding out. On account of complexities of this kind, it is much better not to confound interactions if there is a reasonable chance of their existing. (The situation is different in an exploratory experiment, e.g., one with fractional replication, because then the aim is only to discover which factors interact. For that purpose it is enough to know about the two-factor interactions. Questions of interpretation do not arise.)

[Data from Andrews, D.F. and Herzberg, A.M., *Data: A Collection of Problems from many Fields for the Student and Research Worker* (1985), pages 339–46]

Exercise 10B

A set of experiments was conducted in England to study the fertilization of wheat. There were three sites, X, Y and Z. At each site two experiments were conducted. One was on land expected to supply little nitrogen from residues (N-index = 0); the other was on land expected to be rich in such residues (N-index = 1). The six experiments were carried out in three

successive years, 1, 2 and 3, the later ones being always placed on fresh land and not on land occupied by an earlier experiment.

In each of the 18 experiments there were three blocks. Each block contained seven plots, one for each of seven levels of applied nitrogen, namely 0, 2, 3, 4, 5, 6 and 7 times the standard rate.

Among those seven levels there are six effects, (usually linear, quadratic, etc.) but you are here asked to consider only two less conventional ones, namely:

L: The effect of applied nitrogen at low levels, measured for present purposes by the difference in response between levels N_2 and N_0 in each block.

H: The same effect at high levels, here to be measured by the difference in response between levels N_7 and N_6.

Values of these effects are given in Table 10*a*. The data represent crops expressed in tonnes per hectare. Each involves data from two plots, not one, being a difference between N_2 and N_0 or N_7 and N_6 as the case may be. Consequently divisors of summation terms are not the number of figures involved from Table 10*a* but twice that number. You are asked to

Table 10a Values of the effects, L *and* H, *for each of three blocks of the 18 experiments in Exercise 10B*

			L				H		
X	0	1	+ 1.42	+ 3.50	+ 2.82	—	− 0.38	+ 0.98	+ 0.07
X	0	2	+ 3.46	+ 2.65	+ 3.15	—	+ 0.63	+ 0.31	+ 0.02
X	0	3	+ 2.59	+ 3.00	+ 2.93	—	− 0.23	+ 0.09	− 0.06
X	1	1	+ 2.02	+ 2.06	+ 2.16	—	+ 0.03	+ 0.31	+ 0.24
X	1	2	+ 2.28	+ 2.48	+ 2.00	—	+ 0.17	− 0.05	− 0.05
X	1	3	+ 2.98	+ 2.85	+ 3.13	—	+ 0.08	− 0.28	+ 0.18
Y	0	1	+ 2.78	+ 2.89	+ 2.02	—	− 0.04	− 0.11	− 0.55
Y	0	2	+ 2.36	+ 2.43	+ 2.40	—	− 0.14	+ 0.25	− 0.09
Y	0	3	+ 2.15	+ 2.47	+ 2.05	—	+ 0.11	− 0.19	+ 0.47
Y	1	1	+ 2.57	+ 2.32	+ 1.10	—	+ 1.10	− 0.12	− 0.87
Y	1	2	+ 1.24	+ 1.67	+ 1.74	—	+ 0.17	+ 0.77	− 0.53
Y	1	3	+ 2.84	+ 2.80	+ 2.83	—	− 0.15	− 0.59	− 0.15
Z	0	1	+ 1.96	+ 1.86	+ 2.60	—	− 0.16	− 0.04	− 0.02
Z	0	2	+ 2.30	+ 1.81	+ 1.84	—	− 0.10	− 0.25	+ 0.05
Z	0	3	+ 2.38	+ 1.58	+ 2.05	—	− 0.08	—0.15	0.00
Z	1	1	− 0.63	− 0.23	− 0.11	—	− 1.42	+ 0.30	− 0.35
Z	1	2	+ 1.60	+ 0.71	+ 0.84	—	− 0.16	+ 0.02	− 0.37
Z	1	3	+ 1.54	+ 0.86	+ 1.24	—	− 0.16	+ 0.06	+ 0.21

[Data are presented by permission of the Agricultural Development and Advisory Service, Cambridge, who carried out the experiments.]

work out a complete analysis of variance for each variate and to write a careful interpretation.

Hint: The variates in Table 10*a* are in fact the effects under study. It is not necessary therefore to form differences between treatment means to evaluate them; that has already been done. (Indeed, the treatments no longer enter into the table of values to be analysed.) Hence, what in the ordinary way would be the correction term represents the sum of squares for the effect (L or H) itself. It should therefore be included. Further, in randomized block designs the 'error' is the interaction of blocks and treatments. Hence the 'error' sum of squares in these analyses can be found by adding the sums of squares for the block effects, each with two degrees of freedom, for the 18 separate experiments. Hence, the analysis of variance takes the following form:

Source	d.f.	s.s.	m.s.
Effect	1		
Sites (S)	2		
N-Index (I)	1		
Years (Y)	2		
S × I	2		
S × Y	4		
I × Y	2		
S × I × Y	4		
'Error'	36		
Stratum total	54		

Exercise 10C

An investigation is proposed for an area in which traditional farmers use neither fertilizers nor chemical weedkillers (herbicides). They fallow their land every third year, following the rotation:

Fallow—Cotton—Millet.

The fallow is believed to be necessary to restore fertility and to control

weed-seeding, but no one knows how effective it is. Fertilizer and weedkiller have become available at reasonable prices. The Ministry suggest that they should be used to reduce the period under fallow, thus allowing an increase in the production of cotton. Outline the design of an experiment to test whether improvements can be made along those lines.

Exercise 10D

A series of experiments was conducted on a number of islands in the West Indies to investigate the fertilization of maize. The following data come from an experiment on Antigua in which there were many treatments. Here four have been selected. Treatment O, which was duplicated in each block, represented the usual fertilizer practice. Treatments N, P and K were the same except that dressings of nitrogen, phosphates and potassium respectively were applied at three times the usual level, the intention being to discover what would happen if radical changes were made in levels of fertilization. The complete experiment used a non-orthogonal design, but the data selected here make up an orthogonal block design with unequal replication.

The variate *a* represents the number of maize cobs per plot; the variate *b* represents their weight in pounds. Carry out a bivariate analysis of variance on *x* and *y* and assess the conclusions.

Block	I		II		III		IV	
Treatment	*a*	*b*	*a*	*b*	*a*	*b*	*a*	*b*
O	44	6.87	40	7.20	43	6.39	45	6.75
O	41	6.71	48	7.53	40	6.48	45	7.39
N	41	6.75	40	7.55	44	7.52	35	5.27
P	42	6.67	41	7.18	47	7.56	42	6.41
K	43	5.26	38	5.45	46	7.04	37	4.75

[Data: *A collection of problems from many fields for the student and the research worker* (1985), p. 346 (Experiment at Old Road)]

Exercise 10E

A fan trial is needed to cover the range of plot size from $A_1 = 1\ m^2$ to $A_n = 4\ m^2$ with square planting ($R = 1$). The length is not to exceed 20 m ($L = 20$). Determine the other dimensions to secure what is needed.

Exercise 10F

Analyse the data of Exercise 1F, ignoring rows and columns but using the method of Papadakis.

Exercise 10G

An experiment is to be conducted with four treatments, (1), X, Y and XY, interest being centred, as might be expected, on the two main effects and the interaction. There are no special constraints on the land, which lends itself readily to the use of blocks with four plots, so it is proposed to use a design in randomized blocks. In that case s^2 can be expected to be about 50. The question concerns the degree of replication required. It is intended to seek significances at the level $P = 0.01$. Taking D to be 25, the chance, P', of its being missed, supposing that to be its true value, must not exceed 0.05. How many replicates should be used?

Chapter 11

Intercrops and the problems they raise

11.1 The range of intercropping problems

There are many instances in which two or more crops are grown together in order to obtain some advantage. Perhaps the two species will call for water or nutrients at rather different times. On that account they will exploit resources better together than either can alone. Perhaps they benefit from different kinds of weather, so that in, say, a dry season the first will crop well, but in a wet one the yield will come from the second. Either way there will be food.

Intercropping has several variants. For example, the two species are not necessarily sown at the same time, but the second may be introduced before the first is harvested. That is called 'undercropping' or 'relay cropping'. Sometimes a leguminous species is grown as well as the main species, with the idea that it will improve future yields. This is called 'alley cropping', the main crop being in widely-spaced rows. A more intimate mixture, e.g. clover and barley, is often referred to as 'undersowing' or 'mixed cropping'. It is not to be expected that the same statistical approach will suit all problems.

It should be emphasized that diversified cropping is not necessarily intercropping. Anyone with a small area of land and a need for several crops, some to supply food and some to supply cash, will grow them intermingled, filling in gaps as may seem appropriate. Unless some benefit can be claimed for growing species in close association, the result should not be called intercropping.

Statistically there are several distinctions to be made. For example, if the two crops serve similar purposes, it does not matter if one dominates or even kills out the other. Where the two supply different needs, like a cereal and a pulse or a food crop and a cash crop, a balance between the two yields will often be essential.

An important difference is that between experiments in which the treatments, e.g. fertilization, are necessarily applied to both component

crops and those, like variety trials, in which a treatment is applied to one component only.

It should be noted that bivariate analysis (see Section 10.4) is not available unless it can be assumed that the correlation coefficient between the two yields is the same for all treatments. Where spacing is involved and in some other cases also, that could be a risky assumption.

11.2 Land Equivalent Ratios

The Land Equivalent Ratio is a measure of the extent to which more crop can be obtained from a piece of land by intercropping as compared with dividing it into two parts and growing a different sole crop on each. Used for that purpose it can be valuable, but it is not necessarily helpful in other contexts. It is calculated thus:

If a certain area of land is given over to an intercrop it may be expected to give m of the first crop and n of the second. If it were given over to the first as a sole crop, it could be expected to give a crop of a; if to the second, the crop would be b.

If we set out to produce the same yield as the intercrop, and to do so using sole crops, we should need an area of m/a of the first crop and n/b of the second. That makes a total of

$$m/a + n/b = \text{LER}, \tag{11.2.1}$$

the 'Land Equivalent Ratio'.

Suppose, however, that the intercrop does not yield as much of the second crop as we would have wished. If we want a ratio of r instead of n/m ($r > n/m$), we would need to supplement the intercrop with an area of the second crop grown sole. If we assigned a proportion θ of the land to the intercrop and $(1 - \theta)$ to the sole crop, we should get

(i) θm of the first
and (ii) $\theta n + (1 - \theta)\, b$ of the second.

We wish to choose θ so that

$$\frac{\theta n + (1 - \theta)b}{\theta m} = r$$

i.e.

$$\theta = b/(b - n + rm).$$

In that case the yields are:

(i) $bm/(b - n + rm)$ of the first
(ii) $bmr/(b - n + rm)$ of the second.

This gives an Effective LER (ELER) of

$$\frac{bm}{a(b-n+rm)} + \frac{mr}{(b-n+rm)} = \frac{m(b+ar)}{a(b-n+rm)} \qquad (11.2.2)$$

Note that if $r = n/m$, ELER = LER.

It could be that we are not concerned to obtain a given ratio between the crops so much as to obtain a given target, t, of the second crop, here called the 'staple crop'.

In that case, if we give θ to the intercrop and $(1 - \theta)$ to the staple, we shall get

$$\theta n + (l - \theta)b = t$$

of the staple. We must therefore choose θ so that

$$\theta = \frac{b-t}{b-n} \quad \text{and} \quad 1 - \theta = \frac{t-n}{b-n},$$

since $n < t < b$, $0 < \theta < 1$. With that θ we get

(i) $m(b - t)/(b - n)$ of the first crop
(ii) $n(b - t)/(b - n) + (b(t-n)/(b - n) = t$ of the second.

That gives a Staple Land Equivalent Ratio (SLER) of

$$1 + \frac{b-t}{b-n}(\text{LER} - 1). \qquad (11.2.3)$$

It will be seen that SLER will exceed 1 only if LER does. Also any gain in SLER is only $(b - t)/(b - n)$ times the gain in LER.

If Land Equivalent Ratios need to be compared, provided both sole crops and all the relevant intercrops are represented in each block, the second method in Section 9.6 is available.

11.3 Bivariate diagrams as used with intercropping

We have two species, A and B, which we have grown in a range of conditions, e.g. in several seasons or at several sites or under different treatments. For each of these conditions we have values, a and b respectively, for the two crops. Naturally they will be different for the various conditions, but we will assume that the standard deviations, σ_A and σ_B respectively, are constant over the whole. We shall also allow for the possibility that a and b are not estimated independently, i.e. that they have a correlation coefficient, ρ, which is not necessarily zero.

To form a bivariate diagram, we transform a and b to x and y, as described in (10.4.1) and (10.4.2). To examine our new variables more closely, x represents the crop of the first species expressed in terms of its own standard error. The second, y, represents that part of the crop of the second species that cannot be explained by the crop of the first, bearing in mind that the two are correlated. That also is expressed in terms of its own standard error. (The crop of the second species is in fact given by the ordinate of its point from the $0z$ axis described at the start of the next section.)

Bivariate diagrams are especially useful in the study of intercrops. First, we will take the two yields from the intercrop, either in a range of seasons, or at a range of sites, or more probably under a range of treatments, and then we will represent each by a point, surrounded by a circle with a radius of $1\sqrt{r}$, where r is the replication of the treatment concerned. A little care is needed here. Strictly speaking, field experiments estimate differences between treatment means, not the means themselves. As Chapter 5 showed, the variance of the difference between two treatment means is not necessarily

$$\left(\frac{1}{r_1} + \frac{1}{r_2}\right) s^2.$$

If it is not, the circles could cause some confusion and it might be better to omit them. Sometimes, in any case, it is better to put a circle of radius L, calculated from (10.4.10), round the point for the standard treatment. Points that lie outside the circle then indicate a treatment that differs significantly from the standard at the level used.

If there is reason to think that the correlation coefficient between a and b is not constant but depends upon the treatment, the bivariate diagram cannot be used, though an algebraic solution can sometimes be given. The situation is explored in the paper by Singh and Gilliver (1983).

11.4 Land equivalent ratios on a bivariate diagram

The diagram can be used to show LERs as in (11.2.1). First, though, we should see where sole crops go on it. If $a = 0$, $x = 0$, and the point lies up the y-axis. If, however, $b = 0$,

$$x = a/\sigma_A, \quad \text{as usual}$$

$$y = -a\rho/(\sigma_A\sqrt{1-\rho^2}).$$

It will be seen that $y/x = -\rho\sqrt{1-\rho^2}$, i.e. all sole crops of A will give points on a line, Oz, through the origin. That line also is shown in Fig. 11.1. The angle, Φ, is such that $\cos\Phi = \rho$. Consequently, if ρ is negative, the line will lie above the x-axis; if ρ is positive, the line will lie below it.

To return to LER's, any sole crop necessarily has an LER of 1. If the yield of each is plotted, one on Oy and the other on Oz, and a straight line is drawn between them, all points on that line will have the same LER. If now we think of the two sole crops as multiplied by α, we can plot these end-points and draw a line between them. All points on it will indicate an LER of a. Further, any point above the line or to the right of it will show

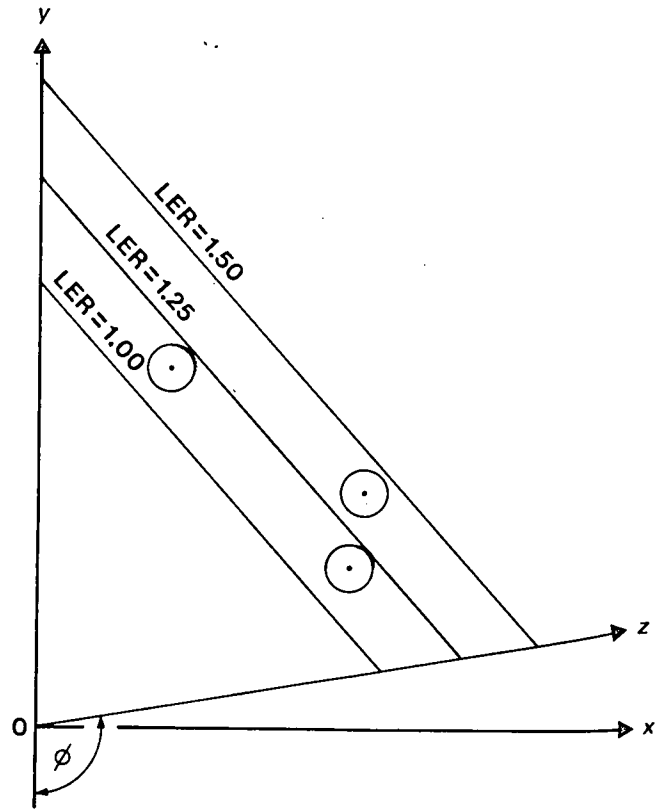


Fig. 11.1 Contours of LER

an LER greater than a, while all other points will indicate an LER less than a. This also is shown in Fig. 11.1.

It could be that we need a certain ratio between the yields of the two species, e.g. we might want the crop of the second to be r times that of the first. In that case $b = ra$. Since the regression coefficient can be written $\rho\sigma_B/\sigma_A$,

$$x = a/\sigma_A$$

$$y = a(r - \rho\sigma_B/\sigma_A)/(\sigma_B\sqrt{1-\rho^2})$$

and y/x is a constant, i.e.

$$y/x = \frac{r\sigma_A - \rho\sigma_B}{\sigma_B\sqrt{1-\rho^2}}$$

Consequently, for any r we can draw a line through the origin with that slope, and all points on it will give the desired value of $b/a = r$. Further, a line between the point for an intercrop and a sole crop will intersect that line in the ratio of θ to $(1-\theta)$, as shown in Fig. 11.2. (The same is true of a line joining the points of two intercrops. That also is shown in Fig. 11.2.) We can still use the lines of constant LER to find the ELER of the combination of intercrop and sole crop or of two intercrops. Note that ELERs have been defined in (11.2.2).

Alternatively we might want a certain amount of one crop. Suppose we require that b shall equal t then, a now being undetermined,

$$x = a/\sigma_A$$

$$y = (t - a\rho\sigma_B/\sigma_A)/\sigma_B\sqrt{1-\rho^2}).$$

As we vary a the point (x, y) will always lie on a line parallel to Oz and at a distance $t/(\sigma_B\sqrt{1-\rho^2})$ above it. Again, we can find combinations of intercrops and sole crops with suitable values of θ by joining points and seeing where they intercept the lines just given. Contours for LER can be used in that case to show the SLER, as defined at (11.2.3).

11.5 Interactions in a bivariate diagram

Bivariate diagrams can also be used to show the form of interactions. Let the point for a standard treatment, O, be at (x_0, y_0) and let two modifications, A and B, give points at (x_A, y_A) and (x_B, y_B) respectively. What would we expect to happen if A and B were tried together? Supposing that the two modifications act independently, we can find the point by completing the parallelogram, i.e. we should expect

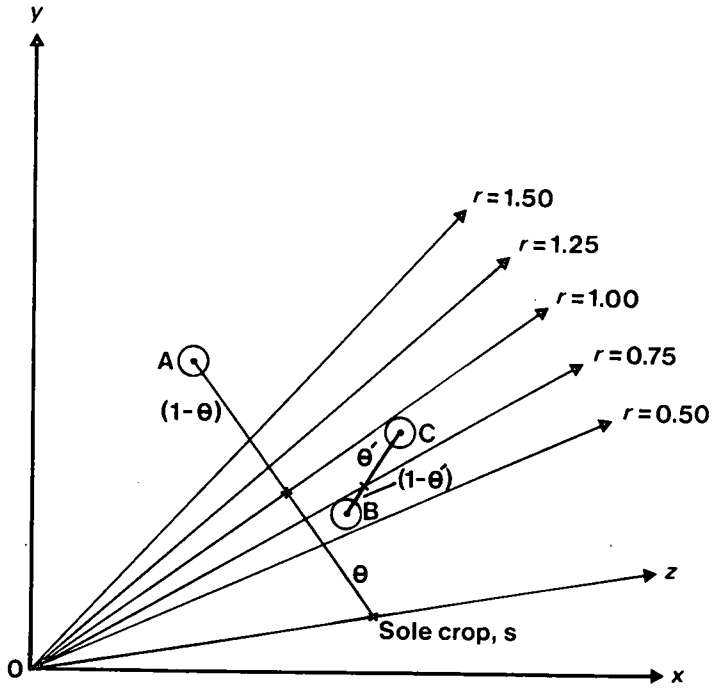


Fig. 11.2 Contours of equal ratio (r = second crop/first crop)

If it were required that Intercrop A be supplemented by a sole crop S to make $r = 1.00$, the proportion of AS that lies above the $r = 1.00$ contour gives (1–0), the proportion that needs to be given to the sole crop, the proportion of A that lies below $r = 100$ giving θ, the proportion that should be given to A. (Note that in this case it is the first crop that is being supplemented, not the second.) Similarly, if it were proposed to apportion land between Intercrops B and C to obtain $r = 0.75$, θ′ should be given to B and (1–θ′) to C.

$(x_A + x_B - x_0,\ y_A + y_B - y_0)$. If then we try them together and obtain instead the point $(x_{AB},\ y_{AB})$, the displacement shows the additional effect brought about by their being in combination, i.e. their interaction. The approach is shown in Fig. 11.3.

Where the factors have more than two levels the diagram can become confusing, though usually its interpretation is clear enough. The method is this: if there are several factors, p in number, in the absence of an interaction between them the diagrams for $(p - 1)$ of them should be the same for all levels of the remaining factor. However, the standard errors of contrasts can vary so much that it is difficult to judge much by eye. It is better to explore the situation carefully by partitioning the treatment effects in the analysis of variance and testing each component separately.

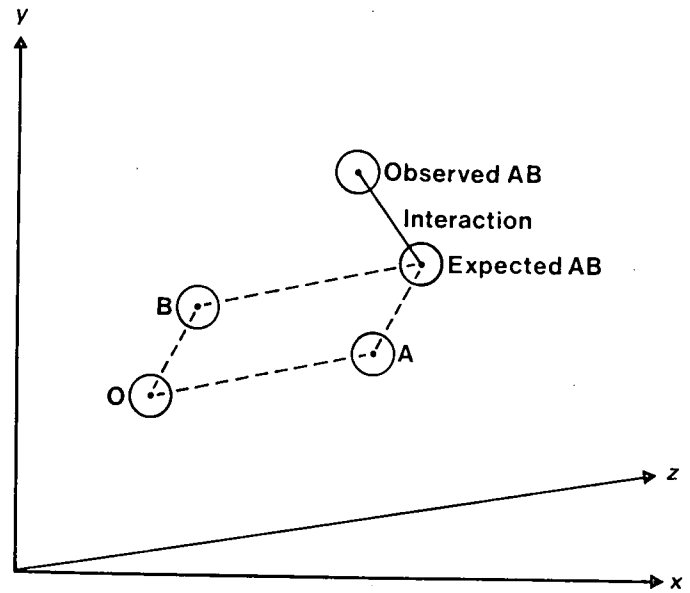


Fig. 11.3 An interaction

beginning with the highest-order interaction. The method of testing is quite simple. It has been given at (10.4.7). The only difference is that D_{aa}, D_{ab} and D_{bb} are no longer the sum of the 'error' and the treatment lines but of the 'error' and the component of treatments (main effect, interaction, etc.) that is under test. Once it is known what interaction is being sought, the diagram will help to show its form.

To take an example, with a 2^3 set of treatments, if there is no three-factor interaction the quadrilateral given by treatments (1), X, Y and XY should have the same shape as that given by Z, XZ, YZ, and XYZ. The difference in shape shows the form of the interaction.

11.6 Contours on the bivariate diagram

In assessing intercrops there can be different standards. Someone might want to consider monetary values in the current year, another biomass, and yet a third might be interested in protein. The best thing is to take contours of equal value. Thus, suppose someone proposed energy as a good basis of assessment. He has first to find conversion factors to change yields into energy and he suggests multipliers of C_A for the first species and C_B for the second. He now says that he wants the contour for E_1 units of energy. It can be found by asking what yield of each species would supply

that need. If he used only the first he would need a crop of E_1/C_A. This would give a point on Oz. If, on the other hand, he were to get the required energy from the second species alone, he would need a crop of E_1/C_B), which is on Oy. A straight line joining them gives all combinations of the two species that give the required amount of energy. He could then draw further contours for E_2, E_3, etc. They will be straight lines parallel to that for E_1. When they are all drawn, it is a simple matter to assess the energy given by any treatment, the position of its point relative to the contours showing all that is needed.

If someone else comes along and says that he is interested in monetary value, M, then he will need other conversion factors, D_1 and D_2 instead of C_1 and C_2. He will therefore obtain different contours and he might well make a different choice of best intercrop.

11.7 The design of intercropping experiments

The preceding sections of this chapter show how diverse are the questions raised by intercrops. (Some of them indeed have no analogue at all with sole crops.) There is therefore greater danger of an imperfect appreciation of the problem leading to a poor design. That risk always exists, but with intercrops it is more likely to cause difficulties because the possibilities are more numerous.

The first question often is whether or not to include sole crops. Mostly experimenters include them, but if the object is to find the best cultural method for an established intercrop, they serve no useful purpose. The usual justification for including them is the need to show a good LER, but why? If the intercrop is really established, its LER must be good enough. If someone can improve it, the LER will supposedly be even better.

The opposite fault can arise if for any reason the sole crops are included. Given a range of fertilization levels for the intercrop and a need to find the LER for each, not only are the sole crops needed but they should also be represented at all levels. More usually the experimenter argues that he knows how to grow the sole crops and he will only introduce each at the level of fertilization that he would recommend. The argument may be sound, but it requires a sceptical examination on each occasion it is advanced.

Finally on the matter of sole crops, if they are needed only to establish the LERs, is it necessary to assign them to small plots and to randomize them? Would it not be easier to grow them in strips with blocks between the strips? After all, if they are only needed as a base-line to estimate a and b for (11.2.1), (11.2.2) or (11.2.3), they are not themselves the objects of study. Only if they are being studied for their own sake are all the sole crops needed in each block.

Other questions arise when one species dominates the other, usually on account of height but sometimes for other reasons. In such a case there could well be questioning whether it could not be grown in some other way, e.g. with wider spacing between rows, to allow more room for the other species. That can be a most useful enquiry, but if the aim is to find out what happens to the taller species using both configurations, i.e. if it is grown, say, at wider spacing than usual between rows but at closer spacing within them, what useful purpose is served at that stage by introducing the other species? When it has been found how to grow the first species to give as much space as possible to the second, then is the time to add it to the system.

It should also be borne in mind that a trial of configurations is not the same as an investigation of competition. If someone wants to know in a botanical sense whether A or B is the stronger competitor with X, then clearly A and B should be compared using the same configuration, but that is not the agronomic problem. If B is more spreading than A, in an intercrop it may well need more space, i.e. the rows should be further apart.

Finally, before starting to design an experiment some thought should be given to the method of dealing with the data. If the intention is to use a bivariate analysis (Section 10.4) it should be remembered that there must be a reasonable expectation of all treatments giving the same correlation coefficient between the two crops. If that is unlikely, as it would be in a spacing experiment, the task of analysis is going to be much more difficult. It will be impossible if there is not enough replication to estimate the correlation coefficient for each treatment separately.

Exercise 11A

An intercrop of maize and pigeonpea was tried out at three spacings (3, 6 or 9 plants per m^2). Yields, expressed in kilograms per hectare, were as follows:

Density	Maize (S)	Pigeonpea (S)	Maize (I)	Pigeonpea (I)
3	2679	988	1637	780
6	3398	1035	2205	672
9	3331	954	2400	768

(S indicates sole crop. I is a component of the intercrop.)

(a) Work out the LER for each density.
(b) Assuming that maize and pigeonpea were desired in a ratio of 2 to 1, find the ELER for each density.

(c) Assuming that at least 900 kilograms of pigeonpea was required per hectare, work out the SLER for each density.

[Data from the Annual Report of the International Crop Research Institute for the Semi-Arid Tropics (ICRISAT) for 1976–7, p. 159.]

Exercise 11B

An intercropping experiment was carried out in four randomized blocks, I–IV. There were two strains of sorghum:

A An early variety
B A mid-season variety

and two strains of pearl millet

P A short-stemmed variety
Q A long-stemmed variety

Data were as follows:

Block		I	II	III	IV
AP	*a*	0.18	0.62	0.28	0.85
	b	1.95	1.70	2.12	1.49
BP	*a*	0.24	0.09	0.52	0.41
	b	2.99	4.24	3.15	3.13
AQ	*a*	1.59	1.81	1.12	2.12
	b	1.03	1.10	1.29	1.51
BQ	*a*	2.10	1.52	1.20	1.25
	b	2.10	2.63	2.42	1.69

The quantities *a* and *b* are respectively yields per plot of sorghum and millet.

Work out the bivariate analysis of variance with the treatments effects partitioned into two main effects and the interaction. Draw the bivariate diagram and offer an interpretation.

[Data published by permission of the International Crop Research Institute for the Semi-Arid Tropics (ICRISAT), where the experiment was conducted.]

Exercise 11C

An experiment concerned the time of planting of the cassava in a maize/cassava intercrop. The cassava was planted either at the time the maize was sown (0) or at 4, 8, 12 or 16 weeks afterwards. In half the plots melons were grown as well but in the other half they were omitted. Taking *a* as the yield of the cassava and *b* as the yield of the maize, the analyses of variance and covariance were as follows:

Source	d.f.	a^2	*ab*	b^2
Times of planting cassava (T)	4	4263	2790	6627
Melons (M)	1	17	35	71
Interaction (T × M)	4	882	262	212
'Error'	27	11 762	− 1362	1662
Stratum total	32	16 924	1725	8572

Treatment means were:

		T0	T4	T8	T12	T16
O	*a*	66.2	88.0	64.2	87.7	69.0
	b	61.0	57.2	41.9	52.1	30.2
M	*a*	84.0	79.2	60.2	93.0	65.2
	b	71.6	60.5	45.0	50.6	27.9

All figures represent crop in kilograms from a plot of 32 m^2. There were four randomized blocks, each of ten plots.

Calculate the bivariate analysis of variance and draw the bivariate diagram. Assess the results.

Note: There is an apparent inversion of Treatments T8 and T12 that no-one could explain. For present purposes there is no need to be worried by it.

[Data from S. C. Pearce and B. Gilliver. *J. Agricultural Science* (Cambridge), **91** (1978), p. 629.]

Exercise 11D

A 2^3 factorial experiment was carried out on an intercrop of sorghum (a) and pigeonpea (b). After transformation the treatment means were:

Treatment	x	y
$V_1P_1F_1$	0.86	4.26
$V_1P_1F_2$	2.21	1.80
$V_1P_2F_1$	1.24	5.59
$V_1P_2F_2$	2.83	4.12
$V_2P_1F_1$	3.26	2.76
$V_2P_1F_2$	8.39	−0.98
$V_2P_2F_1$	4.40	4.59
$V_2P_2F_2$	9.67	2.34

The two levels of Factor V were:

1 Local variety of sorghum
2 Introduced variety

Those of Factor P were:

1 Planting on the flat
2 Planting on raised beds

Those of Factor F were

1 Farmyard manure at 10 tonnes per hectare
2 Ammonium sulphate at 0.4 tonnes per hectare

There was no evidence of a three-factor interaction, but there was a significant interaction of V and F. Draw it on the bivariate diagram and derive the form of the interaction.

[Data from B. Gilliver and S. C. Pearce, *Experimental Agriculture*, **19**, p. 28.]

Exercise 11E

In Exercise 11C draw contours on the bivariate diagram to show calorific values of 30 000 MJ and 40 000 MJ per hectare. You may take a kilogram of maize to give 0.979 MJ and a kilogram of cassava to give 0.898 MJ. In order to obtain the greatest calorific value, would you plant cassava early or late?

Chapter 12

Defective data

12.1 The consequences of losing data

The analysis of data when some plots are missing is one of the classical problems in statistics and the subject has acquired a large literature. The development of completely general methods of analysis, e.g. the Kuiper–Corsten iteration, has made much of that work unnecessary, at least as far as block designs are concerned. Row-and-column designs raise further questions, chiefly on account of lost data causing the rows and columns to become non-orthogonal to one another. (They will therefore be considered separately in Section 12.5.)

It should be emphasized right from the start that there is always a price to be paid for lost data, just as there is for any other defect in the design of the experiment or for any other deficiency in the data. (This will have become apparent from a study of Chapter 4. In Exercises 4A and 4B it is indeed possible to derive analyses that are formally correct, but some standard errors of treatment differences are larger than they would have been if nothing had gone wrong.)

In the case of a block design the position is fairly well understood. To take an example, suppose that an experiment has been designed with four treatments in five randomized blocks, like this:

Block					
	I	A	B	C	D
	II	A	B	C	D
	III	A	B	C	D
	IV	A	B	C	D
	V	A	B	C	D

Now suppose that the plot in Block I with Treatment A has been lost for some reason that has nothing to do with the treatment, e.g., the plants have not died from low fertilization nor from a disease that the treatment was intended to control. The plot is missing on account of some other cause unrelated to the treatment. Accepting that the data from the

remaining 19 plots can be satisfactorily analysed, how much has been lost?

The answer is that there has been a loss of information concerning the contrast between the treatment of the missing plot and other treatments in the damaged block. To be more specific, the contrast

A	B	C	D
(+3	−1	−1	−1)

has suffered a loss of one of its effective replications, since Block I no longer has anything to say about it. That is to say, its standard error is not

$$\sqrt{\frac{12 \times \text{Error mean square}}{5}}$$

but

$$\sqrt{\frac{12 \times \text{Error mean square}}{4}} \qquad (12.1.1)$$

(Note that $(+3)^2 + (-1)^2 + (-1)^2 + (-1)^2 = 12$.) There is now one contrast with an effective replication of four, namely (3 − 1 − 1 − 1), and two others orthogonal to it, say (0 2 − 1 − 1) and (0 0 + 1 − 1) with five, a total of 14. Before, there were three orthogonal contrasts, each with an effective replication of five, a total of 15. That is to say, the loss of 1/20 of the data has led to a loss of 1/15 of the information; as usual the information loss is proportionately greater than the data loss.

The above would be helpful if (+3 −1 −1 −1) were a contrast of interest. Quite possibly it is, but not necessarily so. It could be that the four treatments form a factorial set, the contrasts of interest being (−1 −1 +1 +1), (−1 +1 −1 +1) and (+1 −1 −1 +1) as in Section 5.2. The general rule is that any contrast loses in effective replication by an amount ρ^2, where ρ is the correlation coefficient between the contrast concerned and the one that has suffered the direct damage, which in this instance was (+3 −1 −1 −1). Thus, for (−1 −1 +1 +1) $\rho^2 =$

$$\frac{[(+3 \times -1) + (-1 \times -1) + (-1 \times +1) + (-1 \times +1)]^2}{[(+3)^2 + (-1)^2 + (-1)^2 + (-1)^2][(-1)^2 + (-1)^2 + (+1)^2 + (+1)^2]}$$

$$= \frac{(-4)^2}{12 \times 4} = 1/3.$$

It is the same for the other two contrasts of interest. It follows that each has a standard error of

$$\sqrt{\frac{12 \times \text{Error mean square}}{4\frac{2}{3}}} \qquad (12.1.2)$$

instead of

$$\sqrt{\frac{12 \times \text{Error mean square}}{5}}$$

The position now is that there are three contrasts, each with an effective replication of $4\frac{2}{3}$, making a total of 14 as before, the information loss still being 1/15. The same approach applies to data missing from block designs that are non-orthogonal.

It should be explained that the values calculated at (12.1.2) are sometimes approximate. In technical parlance they are exact only if the contrast of interest is an eigenvector of the coefficient matrix of the design achieved, but the ordinary practitioner does not need to bother about that. Always to accept the loss of ρ^2 in the effective replication is much more realistic than to pretend that nothing has happened. In the example given, exact methods show that the true effective replication is 4.62, which is much nearer to $4\frac{2}{3}$ than to 5.

12.2 Missing-plot values in a block design

The method to be described is the oldest and one that is still commonly used. In essence the idea is to fill each gap in the data with a figure that will give a zero residual for that plot. The fitted value then represents what the injured plot would have achieved according to the testimony of those that remain. There is the loss of a degree of freedom from the stratum total on account of each missing plot, with a similar loss from the 'error' line since the missing plots have been constrained to make no contribution to it.

The method will be illustrated using the following contrived data:

Block		I	II	III	IV	
Treatment	A	m	21	21	18	$60 + m$
	B	13	15	19	13	60
	C	12	14	17	13	56
	D	11	18	15	20	64
		$36 + m$	68	72	64	$240 + m$

(12.2.1)

Where it is possible to express the 'error' sum of squares using summation terms, there is no difficulty about writing down a formula for a residual. Here the design is in randomized blocks, so the 'error' sum of squares equals

Total term − block term − treatment term + correction term. (12.2.2)

The four quantities derive respectively from the data, the block means, the treatment means and the grand mean. Following the form of (12.2.2), a residual equals

Datum − block mean − treatment mean + grand mean. (12.2.3)

In the case of the missing plot, its residual equals

$$m - (36 + m)/4 - (60 + m)/4 + (240 + m)/16$$

so $9m - 4(36) - 4(60) + 240 = 0$
and $m = 16$. (12.2.4)

It is instructive to analyse the data at (12.2.1) by sweeping, having first set m to 16. The block means are

I, 13; II, 17; III, 18 and IV, 16.

Sweeping by them gives

A	+3	+4	+3	+2
B	0	−2	+1	−3
C	−1	−3	−1	−3
D	−2	+1	−3	+4

(12.2.5)

The sum of those values squared is 102 and the treatment means are: A, +3; B, −1; C, −2 and D, 0. That gives residuals of

0	+1	0	−1
+1	−1	+2	−2
+1	−1	+1	−1
−2	+1	−3	+4

(12.2.6)

The 'error' sum of squares is 46 with $(9 - 1) = 8$ degrees of freedom, one having been lost on account of the missing value. It will be seen that the missing plot is indeed giving a zero residual. So is another plot, but that is a coincidence.

It would however be a mistake to adopt 102, derived from (12.2.5), as the stratum total. Just as the missing plot must not contribute anything to the residuals, so it must have a zero deviation in order to contribute nothing to the stratum total. In (12.2.5) the value is +3, so there is an

overestimation of the stratum total and hence of the treatment sum of squares. It is necessary to find a new missing plot value, m', that will make the deviation zero. Since the total sum of squares for the stratum equals

Total term − block term,

m' should be such that

datum − block mean = 0

i.e.

$$m' - (36 + m')/4 = 0 \qquad (12.2.7)$$

i.e. $m' = 12$.

The deviations are found to be:

A	0	+4	+3	+2
B	+1	−2	+1	−3
C	0	−3	−1	−3
D	−1	+1	−3	+2

(12.2.8)

(Block means are as before except for Block I, which now has a mean of 12 instead of 13.) The sum of squared deviations is 90 with 11 degrees of freedom, making the analysis of variance to be

Source	d.f.	s.s.	m.s.	F
Treatments	3	44	14.67	2.55
'Error'	8	46	5.75	
Stratum total	11	90		

(12.2.9)

Some will wonder how the missing value can be both m and m', which are usually not the same. The answer is that the two values serve different purposes. The deviations represent the unexplained variation when treatments are ignored; the residuals represent the same variation when they are allowed for. On the testimony of the other plots, the missing value should be 12, supposing there to be no treatment effects, but 16 if there are.

In practice many people do not calculate m' at all, but use the sum of squared values in (12.2.5) instead of those in (12.2.8). Since that might increase the apparent significance of treatments with no possibility of decreasing it, the approximation will cause little harm if the value of F is

plainly not significant or if it is so large that a small reduction will not change significance to non-significance. In those circumstances it may be permissible to avoid calculating m'. Sometimes, indeed often, no significance test is required, the aim being only to estimate means. In that case there is no need of m' because treatment means must obviously allow for treatment effects.

In general it is wise to calculate m and m' to one decimal place more than is needed for the data (see Section 1.9).

When several plots are missing, the method can still be used, the outcome being a set of simultaneous equations. For example, suppose that the datum for Treatment B in Block I was missing as well. Calling the two missing values m_1 and m_2, the data read:

Block		I	II	III	IV
Treatment	A	m_1	21	21	18
	B	m_2	15	19	13
	C	12	14	17	13
	D	11	18	15	20

(12.2.10)

Hence, by the same method as before,

$$m_1 - (23 + m_1 + m_2)/4 - (60 + m_1)/4 + (227 + m_1 + m_2)/16 = 0$$
$$m_2 - (23 + m_1 + m_2)/4 - (47 + m_2)/4 + (227 + m_1 + m_2)/16 = 0.$$

Simplifying these equations gives

$$9m_1 - 3m_2 = 105$$
$$-3m_1 + 9m_2 = 53$$

and so $m_1 = 15.3 \quad m_2 = 11.0.$ (12.2.11)

Turning now to the deviations, by the same method as for residuals it emerges that

$$m_1' - (m_1' + m_2' + 23)/4 = 0$$
$$m_2' - (m_1' + m_2' + 23)/4 = 0$$

Hence $3m_1' - m_2' = 23$

$$-m_1' + 3m_2' = 23$$

so $m_1' = m_2' = 11.5.$

In this instance $m_1 = m_2$ because they refer to the same block, treatments being ignored, but that will not always be so.

The weakness of this method lies in the difficulty of finding the formulae

for m and m' when the 'error' sum of squares cannot readily be found from summation terms. For that reason, it needs to be supplemented by some method of wider application.

12.3 Rubin's method for fitting missing-plot values

By contrast with the last method, the procedure suggested by Rubin (1972) is of completely general application. Conceptually it is very simple. Two parallel bodies of data are set up. The first of these, x, is called a 'pseudo-variate'; it has a datum of one for the missing plot and zero for all the others. The second, y, consists of zero for the missing plot and of the known values elsewhere, like this:

	x				y			
	I	II	III	IV	I	II	III	IV
A	1	0	0	0	0	21	21	18
B	0	0	0	0	13	15	19	13
C	0	0	0	0	12	14	17	13
D	0	0	0	0	11	18	15	20

(12.3.1)

Sweeping by blocks gives deviations of

A	$\frac{3}{4}$	0	0	0	-9	$+4$	$+3$	$+2$
B								
C	Not needed				Not needed			
D								

Sweeping now by treatments, the only residuals needed are those for the missing plot, namely, 9/16 and -9. (The first of these will be needed later, so it will be given a symbol, namely ψ. The corresponding deviation namely $\frac{3}{4}$, will be called φ.) The two missing plot values can now be found thus:

m' = (deviation of y for the missing plot)/(deviation of x)
$= -(-9)/(\frac{3}{4}) = 12$, and.

m = $-$ (residual of y for the missing plot)/(residual of x)
$= -(-9)/(9/16) = 16.$

It follows that any method that gives deviations and residuals, e.g. the Kuiper–Corsten iteration, provides a means of finding m and m'.

It may here be noted that the calculation of ψ is usually quite simple, because the ψ values for all plots must sum to the number of degrees of freedom for error. Since in the example all 16 plots have the same status

(i.e. all blocks contain four plots, and all treatments are replicated four times, so there is no reason to pick on some plots as being different from the rest), there are 16 similar ψ values which must add up to 9. Hence each must equal 9/16. The argument holds not only for randomized blocks but for Latin squares, balanced incomplete blocks (Section 4.7), and lattices (Section 4.9). On the other hand, it breaks down when blocks were intended to be of unequal size, a case not considered in this manual, or when, as in designs with supplemented balance (Section 4.8), the replication was intended to be unequal. Even so, in those cases the evaluation of ψ is rarely difficult.

Similar to ψ is the deviation for the missing plot in the pseudo-variate, which has been written φ. (In the example, $\varphi = 0.75$.) The sum of φ-values over all the plots must give the degrees of freedom for the stratum total. If all blocks are the same size, all plots will have the same φ)-value, which must therefore be

(Stratum total degrees of freedom)/(number of plots).

In the example that gives 12/16, which is correct. The same argument applies for a row-and-column design.

The method can be extended to cover any number of missing plots, but the general case requires a matrix inversion. It will therefore be avoided here. The calculations are, however, quite simple when only two plots are missing.

The method is this. Set up a pseudo-variate for the first plot and find residuals. ψ_{11} and ψ_{12}, for the two plots that are missing. Then do the same for the second plot and find corresponding residuals, ψ_{21} and ψ_{22}. (It is no coincidence that ψ_{21} always equals ψ_{12}.) Next find $\kappa = \psi_{11}\ \psi_{22} - \psi_{12}^2$. Finally, it is necessary to fill the gaps in the data with two zeros and to find ρ_1 and ρ_2, the residuals for the two missing plots. Then the values sought are respectively:

$$m_1 = (\psi_{21}\rho_2 - \psi_{22}\rho_1)/\kappa$$
$$m_2 = (\psi_{12}\rho_1 - \psi_{11}\rho_2)/\kappa. \qquad (12.3.2)$$

The method will be illustrated by the example at (12.2.10). In the pseudo-variate for the first missing plot, $\psi_{11} = 9/16$ and $\psi_{12} = -3/16$; in that for the second, $\psi_{21} = -3/16$ and $\psi_{22} = 9/16$. Hence $\kappa = 9/32$. Filling in both gaps with zeros gives corresponding residuals of $-105/16$ ($=\rho_1$) and $-53/16$ ($=\rho_2$); hence from (12.3.2),

$$m_1 = \left[\frac{(-3)}{16} \times \frac{(-53)}{16} - \frac{(+9)}{16} \times \frac{(-105)}{16}\right] \frac{32}{9} = \frac{1104}{72} = 15.3$$

$$m_2 = \left[\frac{(-3)}{16} \times \frac{(-105)}{16} - \frac{(+9)}{16} \times \frac{-53)}{16}\right] \frac{32}{9} = \frac{792}{72} = 11.0$$

Obviously the calculations are very similar to what they were before. The choice between the two methods must depend upon convenience. The first way of calculating m_1 and m_2 needed the summation terms. It is therefore available only for orthogonal designs, whereas the second is completely general, provided a method is available for calculating residuals. Use of φ_{11}, φ_{12} ($=\varphi_{21}$) and φ_{22} will give m_1' and m_2', just as ψ_{11}, ψ_{12} and ψ_{22} give m_1 and m_2.

12.4 Use of the analysis of covariance with incomplete data

Given a computer program able to make adjustments by covariance, i.e. by the method set out in Sections 8.5 to 8.7, there is a simple method of analysing incomplete data. To take the example given in (12.2.1), it is sufficient to use the same data as at (12.3.1), the right-hand table, i.e. the known data with a zero for the missing plot, being taken to be y, the incomplete data to be analysed. The left-hand data provide x, the pseudo-variate by which y is to be adjusted. It is intended to adjust all values of y to a zero value of x. First, the summation terms are:

	x^2	xy	y^2
Total	1.0000	0	3998
Blocks	0.2500	9	3800
Treatments	0.2500	15	3608
Correction	0.0625	15	3600

This leads to the analyses of variance and covariance:

Source	d.f.	x^2	xy	y^2
Treatments	3	0.1875	0	8
'Error'	9	$0.5625 = \psi$	−9	190
Stratum total	12	$0.7500 = \varphi$	−9	198

Adjusting the analysis of variance of y^2 by the regression of y on x as in (8.5.4) gives

Source	d.f.	s.s.	m.s.	F
Treatments	3	44	14.67	2.55
'Error'	8	46	5.75	
Stratum total	11	90		

as at (12.2.9).

The one value of y that needs to be adjusted is that for the missing plot. Since x is being changed by -1, i.e. from $+1$ to 0, y has to be changed by $-b$, where b is the regression coefficient. That is, the zero value of y for the missing plot should become

$$-(\text{'Error' for } xy)/(\text{'Error' for } x^2)$$
$$= -(-9)/(0.5625) = 16 \text{ as at (12.3.3).}$$

If there are several missing plots, a separate pseudo-variate is needed for each. For example, for the problem given at (12.2.10), it would be necessary to use the variate, y, with two zeros, i.e.

Block		I	II	III	IV
Treatment	A	0	21	21	18
	B	0	15	19	13
	C	12	14	17	13
	D	11	18	15	20

and to adjust it by two pseudo-variates, w and x, like this:

Block		I		II		III		IV	
		w	x	w	x	w	x	w	x
Treatment	A	0	1	0	0	0	0	0	0
	B	1	0	0	0	0	0	0	0
	C	0	0	0	0	0	0	0	0
	D	0	0	0	0	0	0	0	0

The advantage of the method lies in its giving correct standard errors of treatment differences as well as a correct analysis of variance. Its disadvantage lies in requiring an additional pseudo-variate for each additional missing value. It is therefore limited by the capacity of the available computer program to adjust y by many variates.

12.5 Missing data in more difficult circumstances

With row-and-column designs the loss of data is more important than with block designs because it also gives rise to non-orthogonality between rows and columns. As a result, the loss of information can be more serious and more difficult to calculate. Clearly the Kuiper–Corsten iteration is not going to provide the ultimate protection that it does with block designs. The other methods are all available, but the preferred one must be use of the analysis of covariance because it gives correctly the standard errors of contrasts, which are otherwise rather elusive.

This still does not meet the need of someone who loses three or more plots from a row-and-column design. Also, to avoid a matrix inversion, we have not shown you how Rubin's method works with several missing values; for the same reason we have not gone beyond double covariance. There is, however, a method that can be used with any orthogonal design, whether in blocks or in rows and columns. It is rather laborious, but its advantage lies in its wide availability. We shall illustrate it using the data at (12.2.10).

The equations at (12.2.11) may be written like this:

$$m_1 = 105/9 + 3m_2/9 = 11.67 + m_2/3 \qquad (12.5.1)$$
$$m_2 = \ 53/9 + 3m_1/9 = \ 5.89 + m_1/3 \qquad (12.5.2)$$

In that form they are called 'leading equations' for m_1 and m_2 respectively.

We will start by choosing a likely value for one of the missing values. We might, for example, put m_2 equal to $(15 + 19 + 13)/3 = 15.67$, that being the mean value of other data from the same treatment. Then from (12.5.1), $m_1 = 16.84$; then from (12.5.2), $m_2 = 11.45$. Clearly we can continue the process, the two equations being taken in turn, like this:

Cycle	m_1	m_2
0		$15.67 = m_{20}$
1	$16.89 = m_{11}$	$11.52 = m_{21}$
2	$15.51 = m_{12}$	$11.06 = m_{22}$
3	$15.36 = m_{13}$	$11.01 = m_{23}$
4	$15.34 = m_{14}$	$11.00 = m_{24}$
5	$15.34 = m_{15}$	

The two equations have in fact 'led' us to the correct conclusion. (There will be no further advance because $m_{15} = m_{14}$.)

The method can be used for any orthogonal design whatever the number of missing plots. There are a few points to notice. One is that an equation will lead only if it is derived from an earlier one in which the

highest coefficient is that of the appropriate unknown value. At (12.2.11) this was so for m_1 in the first equation and for m_2 in the second. The other point concerns procedure when there are three or more values, m_1, m_2, m_3, etc. to be found. (We shall assume for the sake of simplicity that there are only three.) First, approximations m_{20} and m_{30} are found for m_2 and m_3 in any way that seems reasonable. Then m_{11} is found from them using the first leading equation. Then m_{21} is found from m_{11} and m_{30}, then m_{31} from m_{11} and m_{21}, then m_{12} from m_{21} and m_{31}, and so on, i.e., each missing value in turn is found from the rest using the latest value for the other unknowns.

The last difficulty to be considered is that of missing sub-plots in a split-plot design. In fact the solution is very simple when, as is usually the case, each sub-plot treatment is represented once on each main plot. It arises because the sub-plot analysis can be regarded as the aggregation of the analyses given by a randomized block design on each of the main treatments. Thus, at (7.2.1) the analyses for the three variates separately are

	Source	d.f.	s.s.
Ladak	Cuttings	3	1.1792
	'Error'	15	0.3726
	Stratum total	18	1.5518
Cossack	Cuttings	3	0.6926
	'Error'	15	0.7200
	Stratum total	18	1.4126
Ranger	Cuttings	3	0.3012
	'Error'	15	0.1660
	Stratum total	18	0.4672

It will be seen that the 'Error' sum of squares at (7.3.2) was 1.2586, which equals the sum of the 'error' sums of squares in the component analyses, i.e. 0.3726 + 0.7200 + 0.1660. Also, the sum of squares for treatments in the combined analysis at (7.3.2), i.e. 2.1730 = 1.9625 + 0.2105, equals the sum of the treatment sums of squares in the component analyses, i.e. 1.1792 + 0.6926 + 0.3012. It follows that if a sub-plot is defective, either by being missing or in one of the other ways to be considered, it is enough

to isolate the component analysis, to deal with the difficulty as if the design were in randomized blocks, and finally to constitute the combined analysis from its components. (That is the explanation of Exercise 12A. The complete experiment was in split-plots. The data given are from one component.)

12.6 Approximate missing-plot values

There are several contexts, which will be described later, in which it is helpful to know the effect of choosing a *wrong* missing-plot value. There is in fact a simple rule. If $m + d$ is used when m would have been correct, the 'error' sum of squares will be increased by

$$\psi d^2. \tag{12.6.1}$$

Similarly, if $m' + d'$ is used instead of m' the stratum sum of squares will be increased by

$$\varphi d' \tag{12.6.2}$$

An example of the use of these results is afforded by the figures in Section 12.2. Fitting the missing plot value as $m = 16$ gave an 'error' sum of squares of 46, which is correct, but a stratum sum of squares of 102, which is wrong. It is overestimated because it should have been calculated using $m' = 12$ instead of $m = 16$. Hence $d' = 4$. Hence the overestimation equals $\varphi d'^2 = \frac{3}{4}(4)^2 = 12$, the true value being $102 - 12 = 90$, as given at (12.2.9). That calculation provides a ready means of judging the importance of the difference between m and m'.

12.7 Questionable data

Sometimes a plot is known to have suffered damage, and the question arises whether its datum should or should not be accepted for purposes of analysis. The decision can be a difficult one. The experimenter who discards every plot that is open to some criticism could well find himself with nothing left. On the other hand it would be absurd to retain everything even when it was clear that the plot had been damaged severely by some mishap. Some procedure is necessary for guidance with problems of that kind. It can be found by use of (12.6.1) and (12.6.2).

The following data will illustrate the method. They come from an experiment on Thibou's estate in Antigua, one of the series considered in Exercise 10A and similar to the one in Exercise 10D. The same general description applies. There were four treatments, O (duplicated), N, P and K, disposed orthogonally in four blocks. Goats had been observed eating

the maize on the plot indicated by the asterisk and had been driven off. Had they done so much damage that the plot should be discarded?

The data, which represented crop weight per plot, were:

	O	O	N	P	K
I	2.10	3.20	3.69	3.16	1.27
II	2.18	4.20	2.02	2.01	3.91
III	2.57	3.01	3.25	3.92	2.87
IV	3.61	3.42	1.14	1.06*	3.05

The data should first be analysed as if there were no doubts about them. That gives:

Source	d.f.	s.s.	m.s.	F
Treatments	3	1.0206	0.3402	0.30
'Error'	13	14.9417	1.1494	
Stratum total	16	15.9623		

The missing plot value for the dubious datum is $m = 2.979$, which makes $d = 1.919$ (i.e. 2.979 − 1.06). Since $\psi = 1 - 1/5 - 1/4 + 1/20 = 0.60$, from (12.6.1) the use of m instead of the observed value of 1.06 will reduce the error sum of squares by $\psi d^2 = 2.2095$. That will leave 12.5480 with 12 degrees of freedom and gives rise to another analysis of variance, namely,

Source	d.f.	s.s.	m.s.	F
Due to the dubious plot	1	2.2095	2.3937	2.29
Current 'error'	12	12.5480	1.0457	
Former 'error'	13	14.7575		

(12.7.1)

It cannot be said that F is significant at any important level. However, since the goats can only have diminished the recorded crop and not increased it, this is really a one-sided test. Hence P can be halved. Even so, it still exceeds 0.05, but there is now quite strong evidence that the goats did in fact do appreciable damage. Since they were in fact observed eating, most people would omit the plot, but the decision really depends upon the assessment of damage made at the time. It could have appeared to be so

serious that no statistical analysis was needed to justify the exclusion of the plot; it could have been so slight that no one bothered further about it. The significance level required to convince the experimenter that the goats had done important damage must depend upon the prior evidence. (The whole episode underlines the need for a diary in which events are recorded as they occur.) Here we shall assume general agreement that the plot should be taken out of the experiment.

The decision having been made, the next step is to find m'. It equals 2.805, so $d' = 2.805 - 1.06 = 1.745$.

Since φ for the damaged plots equals 0.8, the sum of squares for the stratum total, 15.9623, should be reduced according to (12.6.2) by $\varphi d'^2 = 2.4360$, which leaves 13.5263 with 15 degrees of freedom. Hence the final analysis is:

Source	d.f.	s.s.	m.s.	F
Treatments	3	0.9783	0.3261	0.31
'error'	12	12.5480	1.0457	
Stratum total	15	13.5263		

Regarding the doubtful plot as missing has increased the mean for Treatment P from 2.54 to 3.02 and that is important. In other respects everything is much as before, except that the results are more soundly based now that the ambiguity has been resolved.

In the example considered above there was a clear, objective reason for thinking that the datum might be wrong. The analysis at (12.7.1) measured the weight of evidence that the value observed was out of line with the others. Taken in conjunction with the prior evidence, that clinched the argument for excluding the plot. Much more difficult is the situation when a datum looks absurd but no one can suggest why it should be so.

Much care is needed. It should be recalled that if there are 60 plots and each residual is tested at a level of $P = 0.05$, it is to be expected that three will appear significantly large purely by chance. (The argument is not completely sound because residuals are not estimated independently, but it will serve.) The matter can be looked at more generally. Suppose there are n plots and each has a probability, p, of being rejected by some test. Then it has a probability of $(1 - p)$ of being acceptable; collectively the n plots have a probability of $(1 - p)^n$ of all being acceptable. That leaves a probability of $1 - (1 - p)^n$ that one or more will be wrongly rejected. To take an example, let $n = 60$; then

$$1-(1-p)^n = 0.05, \text{ say}$$

so $(1-p)^n = 0.95$

and $n \log (1-p) = \log 0.95$

$= 1.977\,72 = -0.022\,28.$

Hence $\log (1-p) = 0.000\,371\,3$ and p is 0.000 832 or about 1 in 1200. Hence, if each residual were tested at that level, there is one chance in 20 ($P = 0.05$) that one plot, or perhaps more, would be rejected even though nothing was wrong. The calculations will serve as a warning that data should not be rejected casually just because they look different. (Occasions do arise when a datum is so different from the rest that no one can believe in it, but they do not occur often.) Also there is no reason to adopt $P = 0.05$ as standard. If all data were checked carefully as they were recorded, the experimenter may be unwilling to believe that there was a mistake. He might be more easily convinced if there had been a rush that invited faults and left no time for checking.

When several data are suspect, there are two cases. In one, they are all suspected of the same defect, as when flood water has covered some plots and not others. It is then permissible to assign $x = 1$ to those subject to possible damage, and $x = 0$ to the rest. An analysis of covariance is then worked out on y, the yield being adjusted by x. Whether there is a significant reduction of the 'error' sum of squares or not, the adjustment can still be made, very little being lost if it is not needed. In the other case, each plot is suspected of a different defect, so the case for exclusion is separate for each. It is better to fit all by missing plot values and then to take the doubtful plots one by one, assigning each its observed value to see if that leads to a significant increase in the 'error' sum of squares. After each plot is dealt with, its missing-plot value is restored before another plot is considered. In that way a decision can be reached as to which values to reject and which to accept. (The procedure is not quite correct because, strictly speaking, the missing-plot values should be worked out afresh each time, but it will serve.)

12.8 Mixed-up plots

Another defect of data arises when there is doubt whether the yield of two adjacent plots has been correctly apportioned between them. That is to say, the total for the two plots, T, is known, but no one can be certain how much belongs to each. A similar difficulty arises when two samples become mixed.

In that case the recognized method is to ascribe T to one plot and 0 to the other. A pseudo-variate, x, is then formed in which the plot with T is assigned the value + 1, the plot with 0 is assigned − 1 and all the others

have $x = 0$. An analysis of covariance of y and x will give a valid analysis, but some standard errors will be increased, especially that between the treatments of the two plots involved in the muddle. One advantage of using covariance is the ability to give exact standard errors of contrasts.

Another method and one that is often simpler is this. Suppose that the two missing plot values are m_1 and m_2; then there is an amount, $T - m_1 - m_2$, which represents the discrepancy between actuality (T) and expectation ($m_1 + m_2$). It should be allocated to the first and second missing plots in the ratio of $(\psi_{22} - \psi_{12})$ and $(\psi_{11} - \psi_{12})$ respectively (see Section 12.3). Thus, to take the example at (12.2.10), $m_1 = 15.3$ and $m_2 = 11.0$. Suppose that the sum of yields from the two plots was in fact known to be 24.1, that is a discrepancy of − 2.2. Since in this instance $\psi_{22} - \psi_{12} = \psi_{11} - \psi_{12}$, the discrepancy should be apportioned equally between the two plots, i.e. the quantity 1.1 should be subtracted from each missing plot value to give 14.2 and 9.9 respectively. The true sum of squares for the stratum total can be found in the same way by taking the discrepancy between T and $(m_1' + m_2')$ and apportioning it between the plots in the ratio of $(\varphi_{22} - \varphi_{12})$ and $(\varphi_{11} - \varphi_{12})$, where φ_{11}, φ_{12} and φ_{22} are deviations corresponding respectively to φ_{11}, φ_{12} and φ_{22}. If all blocks are of the same size, $\varphi_{11} = \varphi_{22}$, so this allocates half the discrepancy to each plot. Unfortunately there is no simple way from this point of finding the variance of a difference of treatment means.

12.9 Special problem with confounded designs

So far in this chapter it has been implicitly assumed that the design is not confounded. If it is, there will be a 'disconnection', i.e. some of the treatments will occur only in certain blocks while the other treatments occur only elsewhere. (There may be several disconnections, but that does not alter what follows.) In itself a disconnection is not serious, provided no one tries to estimate the contrast between the two parts, which has been lost. (To be more exact, it has been transferred to another stratum and could be recovered from there if needed, but in most instances for practical purposes it has indeed been 'lost'.) The special difficulty is that mishaps can create a disconnection where there was none before; they can also remove one. In a design as well connected as randomized blocks, only a disaster could bring about a disconnection but quite a minor accident could disconnect a simple lattice. Given an accident, the possibility has to be borne in mind. As to removal, a mishap of the kind considered in Exercise 4A could lead to the two sets of treatments becoming connected.

The Kuiper–Corsten iteration is always available for the analysis of the data themselves (Section 4.2), but when it comes to finding the variances (Section 4.3), if there is a disconnection the iteration should be applied in

each part separately. There is no great difficulty in putting the distinct matrices together to generate a single covariance matrix of the kind used in Section 5.2.

The method, which is really very simple, will be illustrated by considering a 2^3 factorial set in four blocks with the three-factor interaction confounded, with treatments of two plots interchanged:

Block					
	I	(1)	BC	AC	AB
	II	A	A	C	ABC
	III	B	B	C	ABC
	IV	(1)	BC	AC	AB

It will be supposed further that Treatment (1) has been lost in Block I. In the first disconnected part, (1), BC, AC and AB, the covariance matrix is shown by the Kuiper–Corsten iteration to be:

0.980	− 0.163	− 0.163	− 0.163
− 0.163	0.361	− 0.139	− 0.139
− 0.163	− 0.139	0.361	− 0.139
− 0.163	− 0.139	− 0.139	0.361

For the second, A, B, C and ABC, it is

0.625	− 0.375	− 0.125	− 0.125
− 0.375	0.625	− 0.127	− 0.125
− 0.125	− 0.125	0.375	− 0.125
− 0.125	− 0.125	− 0.125	0.375

Taking the treatments in standard order, (1), A, B, AB, C, AC, BC, ABC, the two matrices just found can be combined. To show the method, elements marked * come from the first disconnected part and those marked § from the second. Elements that relate to treatments in different parts are set equal to zero.

$$\begin{pmatrix}
0.980^* & 0.000 & 0.000 & -0.163^* & 0.000 & -0.163^* & -0.163^* & 0.000 \\
0.000 & 0.625\S & -0.375\S & 0.000 & -0.125\S & 0.000 & 0.000 & -0.125\S \\
0.000 & -0.375\S & 0.625\S & 0.000 & -0.125\S & 0.000 & 0.000 & -0.125\S \\
-0.163^* & 0.000 & 0.000 & 0.361^* & 0.000 & -0.139^* & -0.139^* & 0.000 \\
0.000 & -0.125\S & -0.125\S & 0.000 & 0.375\S & 0.000 & 0.000 & -0.125\S \\
-0.163^* & 0.000 & 0.000 & -0.139^* & 0.000 & 0.361^* & -0.139^* & 0.000 \\
-0.163^* & 0.000 & 0.000 & -0.139^* & 0.000 & -0.139^* & 0.361^* & 0.000 \\
0.000 & -0.125\S & -0.125\S & 0.000 & -0.125\S & 0.000 & 0.000 & 0.375\S
\end{pmatrix}$$

In using such a matrix it is important to be clear what is confounded and what is not. If a contrast is to be unconfounded, its coefficients must sum

to zero in each disconnected part separately. Thus, the particular effect of the interaction of A and B in the absence of C is confounded. Its coefficients are

$$(+1 \quad -1 \quad -1 \quad +1 \quad 0 \quad 0 \quad 0 \quad 0)$$

The two treatments, (1) and AB, in the first part both have coefficients of + 1 and those in the second part (A and B) both have − 1, so that contrast fails the test. If, however, someone wanted to know about the main effect of B, namely,

$$(-1 \quad -1 \quad +1 \quad +1 \quad -1 \quad -1 \quad +1 \quad +1)$$

there is no difficulty. The treatments in the first disconnected part, i.e. (1), BC, AC and AB, have coefficients respectively of − 1, + 1, − 1 and + 1 which sum to zero, as do the coefficients for the treatments in the second part.

Exercise 12A

An investigation was conducted on the growth of soya beans in nutrient solution. The following data have been extracted from those of a larger experiment. They give the dry weight of individual whole plants after 60 days for three concentrations of iron, using five randomized blocks.

	Fe 1	Fe 2	Fe 3
I	0.607	0.762	1.083
II	0.588	0.660	*m*
III	0.534	0.662	1.181
IV	0.555	0.674	1.123
V	0.449	0.601	1.061

Find m and m'. Complete the analysis of variance. Give the approximate effective replication of the linear and quadratic effects and assess their significance.

[Data from C. I. Bliss, *Statistics in Biology*, **2** (1970), p. 400.]

Exercise 12B

The following data are of interest because they come from the first paper ever published on missing data.

An experiment on sugar beet had five treatments arranged in a Latin square. One of the corner plots was trampled upon during the growing season and in consequence was discarded from the experiment. Data in pounds per plot were as follows, each plot having an area of 0.025 acres:

A	306	B	556	C	369	D	332	E	396
B	357	E	485	D	358	C	317	A	325
C	309	D	467	E	367	A	275	B	413
D	418	A	453	B	389	E	324	C	335
E	503	C	572	A	346	B	397	D	m

Analyse the data by at least two methods and check that you get the same results whichever method is used.

1 pound = 454 grams 1 acre = 0.405 ha.

[Data from F. E. Allen and J. Wishart, *J. Agricultural Science*, **20** (1930), pp. 399–406.]

Exercise 12C

We may ask ourselves what would have happened in Exercise 4A if the person responsible for the analysis of the data had been forced by lack of knowledge to regard all plots that had received a wrong treatment as missing. The data would then have read:

	A	B	C	D
I	14.7	m_1	12.5	14.2
II	m_2	17.1	13.3	15.0
III	15.4	17.6	15.0	16.7
IV	16.3	18.2	16.6	17.4
V	16.8	19.1	17.8	18.9

Find the missing plot values, m_1, m_2, m_1', m_2' and calculate the analysis of variance. Then study the data using the analysis of covariance and adjusting upon the two pseudo-variates. Check that you get the same analysis of variance. Then find the variances of estimation for the contrasts (i) between A and B, (ii) between A and C, (iii) between B and C. Compare them with the corresponding values in Exercise 4A and assess any gain from using all the data available.

Exercise 12D

An experiment was being conducted on six turnip variates using a Latin square, when thieves stole the crop of three plots in a corner. The remaining data were:

E	9.0	F	14.5	D	20.5	A	22.5	B	16.0	C	6.5
B	17.5	A	29.5	E	12.0	C	9.0	D	33.0	F	12.5
F	17.0	B	30.0	C	13.0	D	29.0	A	27.0	E	12.0
A	31.5	D	31.5	F	24.0	E	19.5	C	10.5	B	21.0
D	25.0	C	13.0	B	31.0	F	26.0	E	19.5	A	m_1
C	12.2	E	13.0	A	34.0	B	20.0	F	m_2	D	m_3

Data represent fresh weight of crops (including the tops) in pounds per plot, each plot being 15 ft square (1 pound = 454 grams; 1 foot = 0.305 m).

Use the method of leading equations to find values for m_1, m_2 and m_3.

[Data from A. A. Rayner, *A First Course in Biometry for Agriculture Students* (1969).]

Exercise 12E

In Exercise 4F a block was omitted because it contained two missing plots. We are now at stage when we can see how to deal with it. The additional data (i.e. those in Block III) are:

A	235	B	154	C	110
C	151	D	145	D	159
B	249	A	248	A	m_1
C	152	B	221	D	m_2

Analyse the complete data, missing plots included.

Exercise 12F

In Exercise 12B the plot that had earlier been trampled upon gave a yield of 279 pounds. Do you think that its rejection was justified?

Exercise 12G

The plot regarded as missing in Exercise 12A had given a datum of 0.710. That was regarded as so improbable that it must be wrong. Examine the situation.

Chapter 13

Writing the report

13.1 Introduction

Two questions must be considered before the report of a field experiment, or of a series of experiments, is written.

(1) Who is going to read the report immediately, and who may perhaps read it years later? Farmers? Extension workers? Fellow scientists? Administrators? The report should, as far as possible, be intelligible to all likely readers.

(2) What questions was the experiment designed to answer? Or, in other words, which contrasts were the main subject of interest? (Sometimes an experiment may give an unexpected, unplanned result. No hypothesis can be proved by the data that suggested it, but the wise can often take a hint, in this instance where to look in a future experiment.)

13.2 General

Estimates of treatment effects are almost always the most important results of an experiment; tests of significance, standard errors, etc. are less important. An estimate tells how much the yield is expected to be changed by using one treatment in place of another; an F-test or a t-test indicates the degree of confidence with which the estimate may be stated.

The magnitude of a treatment effect has to be seen in the light of the cost of making the change of treatment indicated. Extension workers seldom recommend to farmers a change of practice, e.g. a new variety, unless it is expected to increase crop by at least 5 percent. If the new practice is going to be costly, e.g. additional fertilization or increased seed rate, 10 percent may be a more likely critical figure. So for practical purposes, the question is not so much 'Can we detect any change in the value of the variate?' as 'Is the gain in excess of what would justify the change?' All this was touched on in Section 10.9.

In reporting on any experiment we should avoid making too much of differences that are too small to matter. Some minimum should be borne in mind. Sometimes one encounters an effect that appears to be large but is

so poorly determined that it cannot be regarded as proven. It should be reported only as a possible candidate for further investigation. The opposite also occasionally occurs, i.e., a small effect has a small standard error. In that case one should report it with the comment that the effect, though clearly established, is too small to be of much importance. A report like that, written with practical considerations in mind, will carry more weight than one that looks as if the writer had never stepped far from the statistics laboratory.

13.3 Presentation of tables of means, etc.

Tables of mean yields should be included in the report, and perhaps also tables of mean differences or of other contrasts. The report of any factorial experiment should include at least the 2-factor tables (or their equivalent, e.g. a table of differences), even if the relevant interactions are not significant. Each table should have attached standard errors ('SEs') or standard errors of differences ('SEDs'). There are times when the least significant differences ('LSDs') are needed, but this figure should be used with discretion. The general mean has no standard error.

13.4 Commenting on tables

When an interaction between two factors is found to be significant at the chosen level of probability (commonly 5%, or $P=0.05$), some comment is needed on the effects of one factor at each level of the other. If an interaction with three or more factors is significant, we should try to detect an intelligible pattern in the results. For example, suppose an unconfounded 2^3 experiment gave the following results:

Factor B		0		1	
Factor C		0	1	0	1
Factor A	0	6	10	12	16
	1	7	10	14	26

SE (table entries) ± 1.0 (21 d.f.)

A suitable comment on the 3-factor interaction would be: 'Factor A had a positive effect on yield (+10 ± 1.4 units) where both B and C were applied, but little or no effect (0, +2, +1 ± 1.4 units) where B or C or both were omitted'. (Here ± indicates the standard error of a mean).

If the experiment is of a complex factorial design, there will be many separate treatment-contrasts; in an unconfounded 2^6 experiment, for example:

Main effects	6
2-factor interactions	15
3-factor interactions	20
4-factor interactions	15
5-factor interactions	6
6-factor interaction	1
Total	63

If there are in reality no treatment effects (imagine for example that the field workers had forgotten to apply the treatments!), about three of the 63 contrasts may be expected to exceed the 0.05 significance level. Further, one may well exceed the 0.01 level. So, if complex interactions (e.g. those involving three or more factors) were considered to be unlikely, it may be reasonable to dismiss one or two of the 42 such contrasts as the results of chance variation, even if they exceed the 0.05 level.

In such complex experiments the 'error' sum of squares may be far from homogeneous; some contrasts used to estimate 'error' may be high-order interactions, judged to have very small values. (In an experiment with fractional replication, such contrasts are the only ones available for estimation of 'error'.) Some may be of the nature of treatment × block interactions, and if each block contains replicate plots of one or more treatments, some are derived from plot differences within blocks. If some of the assumptions on which this pooling of 'error' contrasts is based turn out to be false, it may be necessary to examine the components separately. Occasionally an 'error' mean square can be greatly inflated by an unexpected interaction, either between several treatment contrasts, or between one treatment contrast and a block contrast, e.g., if the soils of different blocks are very different. Sometimes this phenomenon is indicated by several variance ratios being significantly *small*; this can be tested by entering the table of F with the reciprocal of the ratio, and with numbers of degrees of freedom interchanged. (With a row-and-column design it may indicate an interaction of rows and columns, leading to inflation of the 'error' sum of squares, a matter discussed in Section 10.8.)

13.5 Treatment contrasts

We should comment on the treatment contrasts of prime interest even if they are non-significant; if one is nearly significant, e.g. if it exceeds the $P=0.10$ level, the fact should be mentioned. Also, we should be on the alert for simplifications, e.g. perhaps a complex interaction between two multilevel factors is dominated by the linear × linear component, with other components relatively small, even though some of them may be just significant.

We should remember too that an important and highly significant contrast may be overlooked if it is pooled with others that are not significant. For example, if the true response is exactly linear, and if there are six levels, a value of the linear contrast that is just significant ($P = 0.05$) when treated individually may need to be twice as large if it is to give a significant mean square for the five degrees of freedom for all the levels. Similarly, if the experiment compares many insecticides, all equally effective, with a single untreated control, the mean square for all treatment contrasts may be non-significant, even though the contrast 'nil versus treated' is highly significant.

13.6 Multiple variates

If two or more variates were recorded for each plot, e.g. weights of grain and of straw, or weights of harvests at different dates, several of them may show significant effects of one particular factor. We must be conscious that there are probably correlations between the different variates. Such correlations are often positive, for example, because plots on fertile soil give more grain and more straw than plots on poorer land; but negative ones may occur, even between yields of different fractions of one crop. For two variates, the method of bivariate analysis (see Section 10.4) allows correctly for such correlations.

If an analysis of covariance has been calculated, with a covariate ('independent variate') not affected by the treatments, the results may be used in the report. The nature of the analysis and the value of the regression coefficient, with its standard error, should be given. If it is believed that the covariate was affected by treatments, the analysis of covariance may be used, but only with the greatest caution. An example of misuse, often quoted, is the adjustment of yields by covariance with plant density in an experiment in which the treatments have produced different populations of plants. Covariance in such circumstances can, nevertheless, help to interpret the mode of action of the treatments, even though discretion is needed.

13.7 Graphs

Graphs are often helpful to the reader. They may be used to illustrate results given in tables. If one or more factors had many levels (e.g. 4 or 6), a graph will show the form of the response curve, and give the reader a chance to assess the position of any maximum (or minimum) values within the range of levels tested. The report may mention such estimates. To obtain them by strict mathematical methods is beyond the scope of this manual, but with reasonably well-determined response curves, curves drawn freehand are usually satisfactory for practical purposes. A graph may also be useful in assessing the value of a new material in relation to a standard one.

13.8 Miscellaneous

Most reports should include at least most of the following information: Design, plot dimensions, area harvested for yield, soil type, previous crop(s), location, standard error (s) per plot, degrees of freedom for 'error', coefficient of variation (CV) being s as a percentage of the general mean.

Exercise 13

The exercises in earlier chapters provide plenty of material for writing full reports, whether for research supervisors, other scientists, or farmers. Those at the ends of Chapters 6 and 7 are especially suitable, though Exercises 4A, 4F, 4G, 4H, 8C, 8D, 9A, 11B, 12D, 12E and 12F all present individual challenges.

APPENDIX

Confusing and Ambiguous Words

Several words and phrases are used by different writers in different senses. Often, but not always, it is possible to decide by considering the context which sense is intended. We discuss in this appendix some of the ones that are most likely to cause confusion.

(1) *Response*

Some writers use 'response' to mean the measurement made on any single unit, i.e. plot or sub-plot in a field experiment. We prefer 'observation', 'record', 'yield', or 'count' as appropriate. This leaves 'response to nitrogen fertilizer' to indicate a difference between means of treatments with and without nitrogen. The phrase 'response curve' follows naturally from this usage.

(2) *Effect*

In a 2^k experiment the word 'effect' is often used in the sense of 'main effect'. More generally, if (in any experiment) a factor is tested at two levels, the difference between the two relevant mean yields is called the 'effect' of that factor. Sometimes, however, the word 'effect' is used to indicate the deviation of the mean of one treatment from the general mean of all treatments.

(3) *Interaction*

In a 2^k experiment with r replicates the main effect of factor A is defined as follows:

$$(T_{A+} - T_{A-})/(r.2^{k-1}),$$

where T_{A+} and T_{A-} are the totals of the plots with the higher and lower levels of factor A. Each total has $r.2^{k-1}$ plots, and so the main effect is the difference between the two means.

Yates (1937) also used the same divisor ($r.2^{k-1}$) for all interactions, so that, for example,

$$\text{interaction A/B} = (T_{++} - T_{+-} - T_{-+} + T_{--})/(r.2^{k-1})$$

where T_{++} etc. are totals of $r.2^{k-2}$ plots each. Unfortunately, an alternative system has been proposed; in this, although the same divisor is used for main effects, a different one is used for two-factor interactions, another for three-factor interactions, and so on. Either option may be specified in Genstat output.

(4) *Control*

Since entomologists speak of the 'control' of a pest (e.g. by an insecticide), the use of the phrase 'control plot', meaning a plot *not* treated with insecticide or other experimental treatment, may be confusing. 'Nil plot' or 'untreated plot' is safer.

(5) *Residual*

This word has three senses:

(a) = plot residual, as calculated, for example, by sweeping;
(b) 'residual effect' = effect of a treatment applied to an earlier crop on the plots of an experiment;
(c) 'residual herbicide' = weedkiller that remains active in soil for a prolonged period.

(6) *Error*

(a) Mistake, as in 'gross error' of weighing.
(b) In the statistical sense, referring to the unexplained component of a plot-yield, or of a sum of squares for unassigned variation (= 'residual' or 'error' sum of squares).

(7) *Sum of squares*

Sometimes used of 'raw' sums of squares, e.g.

$$x_1^2 + x_2^2 + \ldots + x_n^2$$

but also of 'corrected' sums of squares

$$(x_1 - m)^2 + (x_2 - m)^2 + \ldots (x_n - m)^2 \quad \text{(where } m = \text{mean)}.$$

The difference between these two quantities, nm^2, should be called 'correction for the mean' (CFM), or 'correction term' rather than 'correction factor' because the word 'factor' suggests multiplication rather than subtraction.

(8) *Normal*

To avoid confusion with other uses of the word 'normal', the 'normal distribution' may be called the 'Gaussian' or 'Laplacian' distribution.

(9) *Variance ratio*

Some tables of analysis of variance (ANOVA) include a column labelled 'F', others label the corresponding column 'variance ratio' or 'VR'. Strictly 'F' refers to a theoretical distribution; each tabulated value may (or may not) be a sample from an *F*-distribution.

(10) *Parabolic*

Used as a synonym of 'quadratic'.

(11) *Split-plot*

A synonym of 'sub-plot'.

Solutions to Exercises

1A For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	*F*
Treatments	3	340.000	113.333	19.17
'Error'	23	136.000	5.913	
Stratum total	26	476.000		

The residuals are:

0	+1	−1	+2	+1	−1	+3	−1
0	+1	+3	+3	+1	+3	+2	−2
+2	0	−1	+1	−1	−6		
0	−2	0	−3	−5			

The two low residuals (−5 and −6) suggest that the spray may have caused damage over a wider area than anyone had supposed.

1B For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	*F*
Treatments	5	444.434	88.887	23.48
'Error'	15	56.795	3.786	
Stratum total	20	501.229		

The residuals are as overleaf:

− 1.492	− 1.867	+ 2.183	− 1.167	+ 1.258	− 1.067
+ 1.083	− 0.517	+ 0.608	+ 0.833	− 0.242	+ 0.383
− 1.900	+ 0.850	0.000	+ 0.083	+ 0.633	+ 0.783
+ 0.825	+ 2.800	− 2.575	− 1.192	+ 2.808	− 3.117

The data were presented as kilograms per plot of 36 sq. metres, which calls for multiplication by 0.278 to convert to tonnes per hectare. That would be more generally understood, though other units might be preferred. It is better not to multiply the data themselves because that would lead to rounding errors, the likelihood of copying errors, and difficulty in applying the rule for determining e (Section 1.9). It is better to work with the data as they are on the field sheets and to use the multiplier to convert treatment means, standard errors, etc., at the end when the analysis is complete.

1C For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	9	287.576	31.953	8.27
'Error'	27	104.362	3.865	
Stratum total	36	391.938		

The residuals are:

− 3.250	− 0.125	− 1.045	+ 3.345
+ 1.025	+ 2.175	+ 1.555	+ 2.645
+ 0.675	+ 1.125	+ 0.830	− 0.155
− 0.225	− 2.075	− 0.095	− 1.605
+ 1.650	+ 3.400	+ 0.705	− 0.280
− 1.925	− 0.775	+ 0.955	+ 1.345
− 0.675	− 2.000	− 1.145	− 2.855
− 0.125	− 1.275	− 1.545	− 1.405
+ 0.285	− 1.075	+ 1.400	+ 0.555
+ 2.575	− 0.075	− 1.645	− 1.580

There is no obvious pattern in the residuals to suggest that the blocks were formed badly. The matter will be studied further in Exercises 8E and 8F.

1D The residuals are:

+ 11.6	+ 48.8	− 21.7	+ 25.8	+ 32.9	+ 11.2	− 2.6	+ 20.7
+ 19.6	+ 2.6	+ 50.3	− 6.4	− 25.8	+ 19.9	+ 21.7	+ 6.4
− 20.7	− 16.2	− 11.2	+ 26.8	+ 69.9	− 13.6	− 11.6	− 26.8
− 69.9	− 19.9	− 32.9	+ 13.6	+ 16.2	− 50.3	− 19.6	− 48.8

Broadly speaking, the first row contains high values and the last contains low ones. It could be that the blocks would have been more effective if each had consisted of two rows and had extended across the whole area. The situation will be examined in more detail in Exercise 8G. It may be noted that the blocks actually used led to an 'error' mean square of 19.609. Using methods to be described in Chapter 4, it emerges that the alternative blocking system proposed above would have reduced that figure to 11.966. This gives a strong indication that the blocks could have been formed better, but the alternative analysis of variance should not be used in the interpretation of the present data. ('As the randomization is, so must the analysis be.') It could, however, be used as guidance if a future experiment were designed on the same site or in similar circumstances.

1E For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	3	170.736	56.912	6.59
'Error'	6	51.834	8.639	
Stratum total	9	222.570		

The unit of cavans per hectare is no doubt well understood in some places, but for a scientific publication it is better to convert figures to units more widely understood. In any case, it is better not to convert the data themselves.

1F For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	5	52.498	10.500	3.44
'Error'	20	60.949	3.047	
Stratum total	25	113.447		

It would be better to express results in litres per hectare and that requires a conversion factor of 89.9.

1G For $e = -\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	4	53 068.4	13 267.1	13.88
'Error'	21	20 068.0	955.6	
Stratum total	25	73 136.4		

It is a pity that the plot size is nowhere stated. (The same comment can be made of a number of other exercises.)

2B

Block		
	I	EBDAFC
	II	DFCBAE
	III	FABECD
	IV	AFECDB

2E We suggest:

3	4	9	10	15	16	21	22	27	28	33	34	39	40	45	46
2	5	8	11	14	17	20	23	26	29	32	35	38	41	44	47
1	6	7	12	13	18	19	24	25	30	31	36	37	42	43	48
96	91	90	85	84	79	78	73	72	67	66	61	60	55	54	49
95	92	89	86	83	80	77	74	71	68	65	62	59	56	53	50
94	93	88	87	82	81	76	75	70	69	64	63	58	57	52	51

But do the field staff agree? Also, would it help to renumber the blocks?

3A The variance of the mean is $25/64 = (0.625)^2$. The value of Z in Section 3.2 is therefore $(11.1 - 10.0)/0.625 = 1.76$, which raises no serious doubts about the null hypothesis that the mean is really 10.

3B Note that nothing has been said about the form of the distribution. With 200 observations we can gauge if it is normal or nearly so; also their mean will come near to being normally distributed even if the observations themselves are not. We therefore note that the variance of the mean is $8/200 = (0.2)^2$, so Z equals $(4.77 - 5.00)/0.2 = -1.15$, which does not lead us to reject the null hypothesis.

If there had been only 20 observations there would have been even less evidence for doing so, and we would not know as much about the normality of distribution.

3C The variance of the mean is 8.4, so $Z = 1.38$. We have here a one-tail test because no one expects the soil conditioner to do harm, but there is little evidence that it has done good.

3D The variance of the mean is $49/100 = (0.7)^2$, so $Z = -4.29$. This is a two-tail test and the null hypothesis must be rejected, i.e. the two regions show a difference.

As in Exercise 3B the test can be relied upon only if the observations are normally distributed or if there are enough of them to give a normally distributed mean. Experience shows that the distribution of plant weights is usually skew (i.e. asymmetrical), so with only five observations neither condition holds.

3E From (3.3.1) the variance can be estimated by

$$s^2 = [(+1.2)^2 + (-1.2)^2 + (-0.3)^2 + (-0.8)^2 + (0.8)^2 + (+0.3)^2 + (-0.1)^2 + (+0.1)^2]/7 = 0.6229$$

with 7 degrees of freedom. (Note that $\bar{x}$, the mean, $= 0.4$.) The variance of the mean is therefore estimated as $0.6229/8 = (0.279)^2$ and $t = (0.4 - 0.1)/0.279 = 1.08$. Hence there are no grounds for rejecting the null hypothesis that the true mean is 0.1.

The actual values of t with 7 degrees of freedom and for $P = 0.05$ and 0.01 are respectively 2.365 and 3.499. Hence confidence limits for the observed mean are:

$(P = 0.05)$ $0.4 \pm (2.365)(0.279)$, i.e. -0.26 and 1.06
$(P = 0.01)$ $0.4 \pm (3.499)(0.279)$, i.e. -0.58 and 1.38.

3F The variance of the mean is $4.84/25 = (0.44)^2$ estimated with 24 degrees of freedom. Hence $t = (5.85 - 4.00)/0.44 = 4.20$, so the null hypothesis must be rejected. For $P = 0.01$, $t = 2.797$, so confidence limits are $5.85 \pm (2.797)(0.44)$, i.e. 4.62 and 7.08.

3G The ratio of the two variances (F) is $15.2824/8.0275 = 1.90$ with 10 and 15 degrees of freedom, so there is no objection to pooling. The result is a variance of

$$(10 \times 15.2824 + 15 \times 8.0275)/25 = 10.9295$$

with 25 degrees of freedom. The variance of the difference of means is therefore

$$10.9295(1/11 + 1/16) = (1.294)^2$$

so $t = (6.65 - 4.28)/1.294 = 1.83$ and the null hypothesis can be accepted.

For $P = 0.05$ critical value of t with 10 degrees of freedom is 2.228 and for 15 it is 2.131, so confidence limits for

A are $6.65 \pm (2.228)(1.179)$, i.e. 4.02 and 9.28 and for
B they are $4.28 \pm (2.131)(0.708)$, i.e. 2.77 and 5.79.

It will be seen that the two sets of limits overlap, though not by much.

3H Mean difference is 2.65 with an estimated variance of $15.8307/11 = (1.200)^2$. The null hypothesis is that the difference is really zero, so $t = (2.65 - 0.00)/1.200 = 2.21$ with 10 degrees of freedom. That makes P approximately equal to 0.05, so more data are called for if a decision is to be made. The confidence limits ($P = 0.05$) are

$$2.65 \pm (2.228)(1.200) = -0.02 \text{ and } 5.23.$$

Zero is only just within the limits.

3I The sum of squared deviations is

$$(18^2 + 21^2 + 12^2 + 16^2 + 25^2 + 20^2) - 112^2/6 = 99.33 = (n-1)s^2$$

so χ^2 with 5 degrees of freedom is $99.33/15 = 6.62$, which is well within the upper and lower limits for $P = 0.025$.

3J The value of χ^2 is also below the upper point for $P = 0.05$.

3K For A, variance is 689.43 and for B it is 255.06. Hence $F = 689.43/255.06 = 2.70$ with 9 and 11 degrees of freedom. There is no reason to think that the two variances differ.

For A with 9 degrees of freedom critical values of χ^2 with $P = 0.025$ and 0.975 are 2.700 and 19.023. Also $9s^2 = 6204.9$, so σ^2 lies between 326.2 and 2298.1 with 0.95 confidence. Corresponding limits for B are 128.0 and 735.2.

3L

(a) $100 + 1.96\sqrt{10}$, i.e. 93.8 and 106.2

(b) $100 + 1.96\sqrt{10/10}$, i.e. 98.0 and 102.0
(c) $100 + 1.96\sqrt{10/100}$, i.e. 99.4 and 100.6

If σ^2 were unknown, with (a) there would be no way of estimating s^2, but with (b) s^2 could be found from (3.3.1). With (c) the same method could be used, but s^2 would be so well known that little harm would result from using 1.96 instead of t for $P = 0.05$.

3M For A, $\bar{x} = 25.78$ and $s^2 = 13.4119$ with 8 degrees of freedom; for B, $\bar{x} = 28.24$ and $s^2 = 5.2525$ with 10. Hence $13.4119/5.2525 = 2.55$, which does not approach significance, so the two variances can be pooled to give $(8 \times 13.4119 + 10 \times 5.2525)/18 = 8.8789$ with 18 d.f.

The variance of the difference between the means is therefore

$$8.8789(1/9 + 1/11) = (1.339)^2,$$

so $t = (28.24 - 25.78)/1.339 = 1.84$ and there is no reason to think that the means differ.

3N For preparation A, the estimated probability of its proving effective is $172/250 = 0.688 = p_A$. For preparation B, it is $158/200 = 0.790 = p_B$. On the null hypothesis that there is really no difference between the preparations, $p = (172 + 158)/(250 + 200) = 0.733$. In that case the variance of the difference between p_A and p_B is $p(1-p)(1/250 + 1/200) = (0.042)^2$.

Since $Z = (p_A - p_B)/0.042 = 2.43$, the two preparations differ at the level, $P = 0.05$.

Confidence limits ($P = 0.05$) are:

$$\text{A, } 0.688 \pm 1.96\sqrt{\frac{0.688 \times 0.312}{250}} = 0.631 \text{ and } 0.745$$

$$\text{B, } 0.790 \pm 1.96\sqrt{\frac{0.790 \times 0.210}{200}} = 0.734 \text{ and } 0.846.$$

For the confidence limits to be reduced from ± 0.06 (approx) to ± 0.03, i.e. halved, it would be necessary to take samples four times as large.

3O We here have a sample that probably follows the Poisson distribution with a mean of 4. The variance of a single observation will also equal 4 and that of a mean of 50 will be $4/50 = (0.283)^2$. Further, from a sample of that size its distribution will approximate to the normal. The confidence limits ($P = 0.05$) can therefore be set at

$$4 \pm 1.96(0.283) = 3.45 \text{ and } 4.55.$$

4A For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	3	16.281	5.427	13.79
'Error'	12	4.722	0.393	
Stratum total	15	21.003		

For the intended design, which was orthogonal, K at (4.1.1) would have been $(1/5 + 1/5) = 0.4$ for all the contrasts mentioned. For the achieved design, K is 0.5 for A *versus* B, 0.425 for A *v.* C and 0.4 for C *v.* D. The 'error' mean square provides an estimate of s^2.

4B For $e = +1$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	3	0.0333	0.011 1	5.63
'Error'	15	0.0296	0.001 97	
Stratum total	18	0.0629		

For a design in randomized blocks K would have been $(1/6 + 1/6) = 0.333$. With the design used K is 0.429 for A *v.* B, 0.440 for A *v.* C, 0.298 for B *v.* C and 0.333 for C *v.* D. The 'error' mean square of 0.001 97 provides an estimate of s^2.

4C For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	4	186.800	46.700	11.82
'Error'	16	63.200	3.950	
Stratum total	20	250.000		

The residuals are:

+ 1.133	+ 0.333	− 1.467
+ 1.800	+ 1.200	− 3.000

− 0.533	+ 1.467	− 0.933
+ 1.000	+ 0.800	− 1.800
− 0.400	+ 0.600	− 0.200
+ 3.133	− 1.267	− 1.867
+ 2.267	− 0.133	− 2.133
+ 0.467	+ 0.667	− 1.133
− 0.467	− 0.067	+ 0.533
+ 1.200	+ 1.400	− 2.600

There is a strong tendency for the residuals in the last column to be negative. Probably two blocks of five plots in each column would have given a reduced value of s^2 in (4.1.1). That would also have avoided the non-orthogonality, which reduced an actual replication (r) of 6 to an effective replication (R) of 5. However, this is only a hint for the future.

4D For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Varieties	12	327.891	27.324	1.22
'Error'	27	603.015	22.334	
Stratum total	39	930.906		

Here, where $R = 3.25$, the contrast between any two adjusted treatment means has a K equal to 0.615. Since $s^2 = 22.334$, the variance of any such contrast is 13.735, giving a standard error of 3.71. Since t for $P = 0.05$ and 27 degrees of freedom is 2.052, any difference between the adjusted means of a standard treatment and a new one should be regarded as significant ($P = 0.05$) if it exceeds 7.6.

The adjusted means of the standard varieties are X, 23.8; 6, 29.3 and Z, 34.6. For the new varieties they are:

A 32.6	B 27.9	C 29.8
D 28.1	E 30.0	F 27.1
G 30.1	H 34.1	I 29.4
	J 30.4	

so none has been shown to yield more heavily than Y and Z. On the other hand, both A and H crop more heavily than X, which did not do very well. Was it perhaps introduced as a standard on account of some special property like disease resistance?

4E For $e = +\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Spray treatments	4	51.052	12.763	33.89
'Error'	8	3.013	0.377	
Stratum total	12	54.065		

Adjusted treatment means are:

A	B	C	D	O
4.68	3.35	2.65	8.23	4.48

For this design $R = 2.75$ and $R_o = 4.125$, so K for the contrast between O and one of the spray treatments is $(1/2.75 + 1/4.125) = 0.606$, making the variance $0.2285 = 0.478^2$. It appears then that the adjusted means for sprays B and C lie respectively 2.36 and 3.83 times the standard error below that for O. Also, the adjusted mean for D lies 7.85 times the standard error above. That provides a reservation about the use of one-tail tests. The spray substance itself may have done no harm, but how about the water with which it was formulated? (*Venturia* is a water-borne fungus.)

4F The design is in supplemented balance with $R = 13$ and $R_o = 351/43$, the supplementing treatment being A, so K for the contrast between A and any of B, C and D is 0.1994. Between any pair of B, C and D it is 0.1538.

For $e = -\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	3	13 305.7	4435.2	3.59
'Error'	41	50 643.1	1235.2	
Stratum total	44	63 948.8		

Adjusted treatment means are:

A	B	C	D
172.7	181.8	139.0	156.2

Here it should be recalled that we are dealing with yields, and the real object of the study was to try to alter fruit size. It is of interest, nevertheless, to note that B, in which only king fruits were removed, had no discernible effect on yield, whereas C in which fruit was removed from lateral branches, led to a loss of crop, $(172.7 - 139.0) = 33.7$ with a standard error of 15.7. Treatment D, in which both kinds of blossom would be damaged, was intermediate between B and C, though, of course, the general level of damage was not controlled and may well have been different from that for B and C.

With tree crops the presentation of yields as crop per tree (as has been done here) is usually more intelligible than crop per unit area. It is desirable though to give the density of planting, so that the figures can be converted to an areal basis if anyone wants them in that form.

4G For $e = -\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Varieties	15	15 070.6	1004.7	4.68
'Error'	9	1 931.4	214.6	
Stratum total	24	17 002.0		

For a contrast between two concurring varieties, $K = 5/4$; for other contrasts between two varieties, $K = 3/2$.

Adjusted means are:

A 140	B 160	C 183	D 150
E 145	F 157	G 164	H 186
I 168	J 111	K 202	L 206
M 141	N 191	O 181	P 179

Tillers per square metre of rice is a readily understood measure.

4H For $e = -\frac{1}{2}$, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Varieties	15	17 972.3	1198.2	4.62
'Error'	21	5 441.4	259.1	
Stratum total	36	23 413.8		

For a contrast between two concurring varieties, $K = 5/6$; for other contrasts between two varieties, $K = 11/12$.

Adjusted means are:

A	150	B	152	C	179	D	152
E	164	F	162	G	160	H	182
I	165	J	101	K	191	L	189
M	149	N	197	O	181	P	170

4I At (4.12.1), $R = 5/2$ and $R_o = 30/7$.
At (4.12.2), $R = 9/2$ and $R_o = 117/23$.

5A The treatment sum of squares is 982.15. The contrasts have been taken as:

(1)	(+3	−1	−1	−1)
(2)	(0	+1	−2	+1)
(3)	(0	+1	0	−1)

From the rule at (5.6.8) they are orthogonal, as the following partition confirms.

Contrast	Value	K	Contribution
(1)	−24.6	2.40	252.15
(2)	−27.0	1.20	607.50
(3)	+ 7.0	0.40	122.50
			982.15

5B The contrast between the two varieties is

(+1 +1 +1 +1 +1 −1 −1 −1 −1 −1)

What is left represents the variation of strains within a variety. That leads to the partition:

Source	d.f.	s.s.	m.s.	F
Between varieties	1	57.600	57.600	14.90
Between strains within a variety	8	229.976	28.747	7.44
Between strains	9	287.576		

Even after taking out the effect of varieties there are still considerable differences between strains.

5C It is always difficult to decide after the event what the experimenter had in mind. That is a good reason for always writing down the contrasts of interest at the time of inception.

Taking treatments in the order, S, M, C, D, O, there could well be interest in

(+1 −1 0 0 0)

and in

(0 0 +1 −1 0)

There is perhaps interest in

(+1 +1 −1 −1 0).

Supposing that none of the above contrasts shows anything, the question will arise whether there was any effect at all of nitrogen. This would be shown by

(+1 +1 +1 +1 −4).

The conversion factor to tonnes per hectare is 0.0448.

5D This is an example of the way in which experiments sometimes form a cascade of treatments, each being a development of the one before. Here the idea was perhaps to study:

Nitrogen	(−1	+1	0	0	0	0)
Phosphorus	(0	−1	+1	0	0	0)
Lime	(0	−1	0	+1	0	0)
Potassium	(0	0	−1	0	+1	0)
Special mixture	(0	0	0	0	−1	+1)

If that was the idea, we suggest that a factorial design would have been better with, perhaps, D and F as supernumerary treatments. (By that we mean that they were added to the factorial set to provide special contrasts additional to those that would ordinarily be studied.)

The conversion factor to tonnes per hectare is 0.0953.

5E Again, without guidance from the experimenter, it is not clear which contrasts were under study. Little and Hills (1978) point out that Treatment C involves an organic compound, all the rest being inorganic. Further, two of the others (A and B) are salts of ammonium; two (D and E) are metallic salts. That suggests the contrasts:

(1)	(−1	−1	−1	−1	−1	+5)
(2)	(−1	−1	+4	−1	−1	0)
(3)	(+1	+1	0	−1	−1	0)
(4)	(+1	−1	0	0	0	0)
(5)	(0	0	0	+1	−1	0)

Since the propsed contrasts are in fact orthogonal, they could well be those intended. If so, the analysis of variance reads:

Source	d.f.	s.s.	m.s.	*F*
(1)	1	180.200	180.200	119.10
(2)	1	3.816	3.816	2.52
(3)	1	0.202	0.202	0.13
(4)	1	1.333	1.333	0.88
(5)	1	0.213	0.213	0.14
'Error'	20	30.256	1.513	
Stratum total	25	216.020		

There appears to be little difference in the effect of the various sources of nitrogen, but it is clearly important that the element be supplied.

The conversion factor to tonnes per hectare is 2.511.

5F In Exercise 1E the contrasts of interest are:

(−1	+1	0	0)
(−1	0	+1	0)
(−1	0	0	+1)

(The varieties are taken in the order R, X, Y, Z.) For each contrast, $K = 0.5$. The three contrasts give respectively the values:

+7.925, +8.075, +5.425.

The error mean square is 8.6390, so each has a variance of 4.3195. Dividing each value by its standard error, 2.0783, gives *t*-values of respectively

3.813, 3.885, 2.610.

Using the rule given at the beginning of Section 5.6, it will be found that the three contrasts contribute sums of squares of

125.611, 130.411, 58.861

respectively to the analysis of variance, each having one degree of freedom. This gives respective *F*-values of 14.54 ($= 3.813^2$), 15.10 ($= 3.385^2$) and 6.81 ($= 2.610^2$).

5G The initial partition reads:

Source	d.f.	s.s.	m.s.	*F*
L	1	76 0035	76 0035	6.87
Q	1	7 4405	7 4405	0.67
Departures	3	36 3890	12 1297	1.10
'Error'	15	165 8375	11 0558	
Stratum total	20	285 7705		

The line for departures, which was found by difference, does not look very interesting and the matter need not be pursued. Even if all the sum of squares for departures were due to one degree of freedom this would give an *F*-value of 3.29, which would be significant only at about $P = 0.1$, so the subject scarcely needs to be pursued. If anyone is interested in the further partition, the three remaining effects give:

Source	df.	s.s.
Cubic	1	11 3301
Quartic	1	9 0630
Quintic	1	15 9960
Departures	3	36 3891

5H The contrasts are:

Linear	(−7	−3	+1	+9)
Quadratic	(+7	−4	−8	+5)
Cubic	(+3	−8	+6	−6)

5I The contrasts are:

Linear	(−34	−14	+1	+16	+31)
Quadratic	(+1830	−1242	−1747	−710	+1869)

6A The first partition, which can be carried out using summation terms, reads:

Source	d.f.	s.s.
P	1	1638.40
K	3	2518.40
P × K	3	8.40
Treatments	7	4165.20

The further partition needs the following contrasts:

Main effect of K

L	(−3	−1	+1	+3	−3	−1	+1	+3)
Q	(+1	−1	−1	+1	+1	−1	−1	+1)
C	(−1	+3	−3	+1	−1	+3	−3	+1)

Interaction

L	(−3	−1	+1	+3	+3	+1	−1	−3)
Q	(+1	−1	−1	+1	−1	+1	+1	−1)
C	(−1	+3	−3	+1	+1	−3	+3	−1)

It is:

Source	d.f.	s.s.
P	1	1638.40
Linear K	1	2204.48
Quadratic K	1	313.60
Cubic K	1	0.32
P × Linear K	1	2.88
P × Quadratic K	1	1.60
P × Cubic K	1	3.92
Treatments	7	4165.20

Here again the data are presented after conversion from the field records. For reasons given above (Exercise 1B) this is not altogether wise. Conversion to tonnes per hectare requires further multiplication by 2.509.

6B The provisional analysis of variance obtained from summation terms reads:

Source	d.f.	s.s.	m.s.	*F*
Fungicide (F)	4	13 250.7	3312.7	64.11
Varieties (V)	2	5 762.1	2881.0	55.75
F × V	8	259.5	32.44	0.63
'Error'	28	1 446.9	51.675	
Stratum total	42	20 719.2		

There is, however, a clear structure among the treatments, namely, a factorial set of treatments with a supernumerary untreated control, which suggests the use of the following orthogonal set of contrasts of interest:

Substance (S)	(0	+1	+1	−1	−1)
Time of application (T)	(0	−1	+1	−1	+1)
S × T	(0	−1	+1	+1	−1)
Use of fungicides (V)	(−4	+1	+1	+1	+1)

Taken in conjunction with two contrasts for the varieties, say, (−1 +1 0) and (−1 −1 +2), this leads to a more detailed analysis of variance:

Source	d.f.	s.s.	m.s.	*F*
S	1	6900.7	6900.7	133.54***
T	1	1284.1	1284.1	24.83***
S × T	1	443.8	443.8	8.59**
U	1	4622.8	4622.8	89.46***
V	2	5762.1	2881.0	55.75***.

V × S	2	0.6	0.3	0.01
V × T	2	143.3	21.6	0.42
V × S × T	2	35.8	17.9	0.35
V × U	2	79.6	39.8	0.77
'Error'	28	1446.9	51.675	
Stratum total	42	20 719.1		

We meet here a phenomenon that will become increasingly common, namely, occasions in which the sums of squares in a partition sum to a figure that is slightly wrong. Thus, here, (0.6 + 143.3 + 35.8 + 79.6) = 259.3, not 259.5. That, of course, is due to rounding error and need cause no concern so long as the discrepancy is small.

To turn to interpretation, there is a marked effect of varieties, but no interaction of varieties with fungicide treatments, which may therefore be considered without reference to varieties. The four treatments in the factorial set averaged over varieties give means of

	4 times	10 times	
Copper oxychloride	40.0	45.0	(A)
Dithane	60.7	79.7	

Dithane has been more effective than copper oxychloride, especially when applied ten times. (The standard error of a difference of two means in Table A is 3.39.) The mean for the unsprayed control is 31.0, so the difference between this and the least effective treatment is 9.0, which is 2.65 times its standard error. It may be safely asserted that applying any of the four fungicide treatments gave better crops than doing nothing.

The conversion factor to tonnes per hectare is 0.247.

6C The preliminary analysis of variance reads:

Source	d.f.	s.s.	m.s.	*F*
Varieties (V)	2	882 917	441 458	2.99
Nitrogen (N)	4	39 621 078	9 905 270	243.52***
V × N	8	2 447 594	305 949	2.07
'Error'	42	6 209 863	147 854	
Stratum total	56	49 161 452		

The partition of the nitrogen effect into a linear and a quadratic effect has already been considered in Exercise 5I. The departures from quadratic form (i.e. the cubic and quartic effects) will have to be found by difference.

The detailed analysis reads:

Source	d.f.	s.s.	m.s.	*F*
Varieties (V)	2	882 917	441 458	2.99
N, linear (L)	1	36 004 833	36 004 833	243.52***
N, quadratic (Q)	1	3 065 044	3 065 044	20.73***
N, departures (D)	2	551 201	275 600	1.86
V × L	2	723 240	361 620	2.45
V × Q	2	291 044	145 522	0.98
V × D	4	1 433 310	358 328	2.42
'Error'	42	6 209 863	147 854	
Stratum total	56	49 161 452		

It emerges then that there is a marked effect of added nitrogen; the response curve is not a straight line, but it is fairly well represented by a parabola. The varieties do not differ much and do not have any noticeable effect on the response curve, which is much the same for all of them. (The interaction V × D comes so near to significance at $P = 0.05$ that it perhaps deserves a mention, none the less. It is not clear what it means.)

6D The analysis of variance reads:

Source	d.f.	s.s.	m.s.	*F*
Fertilizer (F)	3	167.745	55.915	95.20**
Varieties (V)	1	19.220	19.220	32.72**
Infestation (I)	1	338.000	338.000	575.48**
V × I	1	3.645	3.645	6.21*
F × I	3	22.225	7.408	12.61***
F × V	3	0.705	0.235	0.40
F × V × I	3	0.330	0.110	0.19
'Error'	15	8.810	0.5873	
Stratum total	30	560.680		

All three factors enter into interactions of some sort, and some statisti-

cians would insist that only a three-way table would suffice to show the results. We do, however, suggest an alternative. The effect of infestations is very great and it interacts with each of the other two factors. In those circumstances it would simplify interpretation to consider the results in two parts. For infested plots, the treatment means are:

	F_1	F_2	F_3	F_4	Means
A	20.4	22.6	29.4	22.7	23.88
B	19.1	21.3	26.9	20.8	22.02
Mean	19.75	21.95	28.35	21.75	22.95
Diff.	1.3	1.3	2.9	1.9	1.85

If this approach is to be followed, we should re-partition, thus:

Source	d.f.	s.s.	m.s.	*F*
Infestation (I)	1	338.000	338.000	
Fertilizer within I(F_I)	3	39.710	13.237	22.54***
Varieties within I(V_I)	1	3.062	3.062	5.21*
F × V within I($F_I \times V_I$)	3	0.248	0.083	0.14
Fertilizer within U(F_u)	3	150.260	50.087	85.28***
Varieties within U(V_u)	1	19.802	19.802	33.72***
F × V within U($F_u \times V_u$)	3	0.788	0.263	0.45
'Error'	15	8.810	0.5873	
Stratum total	30	560.680		

The sums of squares were obtained from two sets of summation terms, namely, S_{vF}, S_v, S_F and S_o, one worked out using only data from infested plots and the other only data from uninfested.

The revised analysis is quite easy to interpret. The only remaining question concerns the partition of the effect of Factor F, but here there is the cascade pattern noted in Exercise 5D and standard errors of treatment differences could well be most effective.

The conversion factor to tonnes per hectare is 0.0530.

6E The partition gives:

Source	d.f.	s.s.	*F*
M	1	5184.0	57.26***
N	1	7267.6	80.27***

P	1	484.0	5.35*
K	1	9264.1	102.32***
M × N	1	169.0	1.87
M × P	1	1.6	0.02
M × K	1	900.0	9.94
N × P	1	196.0	2.16
N × K	1	1914.1	21.14*
P × K	1	169.0	1.87
N × P × K	1	4.0	0.04
M × P × K	1	10.6	0.12
M × N × K	1	1156.0	12.77**
M × N × P	1	33.1	0.37
M × N × P × K	1	39.1	0.43
'Error'	45	4074.2	MS = 90.537
Stratum total	60	30 866.1	

As to interpretation, the difficulty comes from the three-factor interaction, M × N × K. Of its two-factor components, only N × K appears to be of much importance, so it should be helpful to write out the table of means, thus:

	(1)	N	K	NK	
(1)	122.0	116.5	170.5	320.5	(A)
M	177.0	265.5	233.5	341.5	
	149.5	191.0	202.0	331.0	

From the horizontal margin it is clear that N and K operating together are on average more effective than would be expected from their separate effects (331.0 against 191.0 + 202.0 − 149.5 = 243.5, a difference of 87.5). However, when this interaction is examined for the two levels of M separately, it emerges that it is marked in the absence of M (155.5) but scarcely exists (19.5) in its presence. Essentially the table at (A) sets out the conclusions relating to those three factors; its inclusion is essential to any report. The last factor, P, shows a difference of 22.0 which does not enter into any interactions, i.e. it does not depend in any way on the levels of the other factors. That also must be presented. The results are not complete without standard errors. Whether they are those of stated contrasts or of simple differences is a matter for local decision, though they should be pertinent to any discussion of results. Convention requires the presen-

tation of the standard error of the means themselves, though nowadays some will object that field experiments do not estimate means but only contrasts between them.

7A Since the nitrogen factor is quantitative it will be divided into a linear and a quadratic effect. The analysis of variance reads:

Source	d.f.	s.s.	m.s.	*F*
Varieties (V)	4	6.4493	1.6123	5.04
'Error' i	12	3.8387	0.3199	
Stratum total i	16	10.2880		
N. linear (L)	1	6.4802	6.4802	293.89***
N. quadratic (Q)	1	0.0068	0.0068	0.03
V × L	4	0.0388	0.0097	0.44
V × Q	4	0.0659	0.0165	0.75
'Error' ii	30	0.6616	0.02205	
Stratum total ii	40	6.5917		

The following points could well be included in any report. (Note: the fact that varieties are different merely confirms what must have been assumed when they were introduced.)

(1) There is a marked effect of nitrogen, the response curve being virtually a straight line.
(2) The response to nitrogen is much the same for all varieties.

7B The analyses read:

Source	d.f.	s.s.	m.s.	*F*
Infection (I)	1	959.22	959.22	28.89*
'Error' i	3	99.61	33.203	
Stratum total i	4	1058.83		
Protectant (P)	3	421.60	140.53	7.29**
P × I	3	207.11	69.04	3.58*
'Error' ii	18	346.76	19.264	
Stratum total ii	24	975.47		

It is scarcely to be supposed that anyone wants to know that Factor I has had an effect, but the amount should be stated. Obviously it was introduced to see if it would provoke an interaction. Clearly it has done so. Consequently the essential table is the following:

Effect of Caresan compared with the control
Infected 14.6
Not infected 4.5

Effect of Panogen compared with the control
Infected 9.8
Not infected 2.2

Effect of Agrox compared with the control
Infected 1.2
Not infected 1.0

All these differences have a standard error of 3.10 because the variance is

$$19.264(\tfrac{1}{4} + \tfrac{1}{4}) = (3.10)^2.$$

(Note that all contrasts under study are between protectants within infection status. Consequently only the second analysis is involved.) It appears then that no protectant can be said to have led to an improved crop with oats that were not infected, but Caresan and Panogen did so when there was infection.

The conversion factor to litres per hectare is 89.9.

7C It is not stated what contrasts were of interest, but it seems fair to regard A, B and C as one group with D and E in another. If that was the intention, reasonable contrasts are:

α	(+2	+2	+2	−3	−3)
β	(+1	0	−1	0	0)
γ	(+1	−2	+1	0	0)
δ	(0	0	0	+1	−1)

The analyses of variance read:

Source	d.f.	s.s.	m.s.	*F*
α	1	514 332	514 332	13.30**
β	1	548	548	0.01

γ	1	22 743	22 743	0.59
δ	1	1976 247	1976 247	51.22***
'Error' i	12	462 998	38 583.2	
Stratum total i	16	2976 866		
Grasses (G)	3	298 350	99 450	5.99*
G × α	3	44 967	14 989	0.90
G × β	3	16 776	5 592	0.34
G × γ	3	41 676	13 892	0.84
G × δ	3	74 321	24 774	2.49
'Error' ii	60	995 390	16 589.8	
Stratum total ii	75	1471 480		

There is clearly a difference (α) between ABC as a group and DE as another. Since there are no discernible differences within the first group (β and γ) they can be regarded as a whole with a mean of 544. The principal difference lies in the second group, the mean of Treatment E being 913, in comparison with 468 for Treatment D. (In fact, the difference between making the final cut 84 after emergence as compared with 70 is extremely high and does perhaps provide the main conclusion.) There are no interactions with grass mixtures, so results can be presented without qualification in that respect.

Interpretation must depend upon the time of emergence, which is not given. If 30 October lies between 70 and 84 days after emergence, it would be clear that the date of final cut was of paramount importance. The results could then perhaps be best expressed by taking means for the final date of cutting, like this:

D	ABC	E
468	544	913

The standard error of the difference between D and E is 80.2. Between ABC as a group and either D or E it is the square root of

$$38\,583.2(1/36 + 1/12) = (65.5)^2.$$

Note that all contrasts under consideration relate to the first analysis.

The conversion factor to tonnes per hectare is 0.0224.

7D The analyses read:

Source	d.f.	s.s.	m.s.	*F*
Varieties (V)	2	1 905.5	952.8	1.64
'Error' i	10	5 799.1	579.91	
Stratum total i	12	7 7704.6		
N, linear (L)	1	19 096.1	19 096.1	107.43***
N, quadratic (Q)	1	430.2	430.2	2.42
N, cubic (C)	1	5.9	5.9	0.03
V × L	2	227.8	113.9	0.64
V × Q	2	4.7	2.4	0.01
V × C	2	126.7	63.4	0.36
'Error' ii	45	7 998.7	177.75	
Stratum total ii	54	27 890.1		

There is a marked effect of nitrogen, which appears to follow a straight-line relationship and to be the same for all three varieties.

7E The analyses read:

Source	d.f.	s.s.	m.s.	*F*
N	1	189.28		12.26**
P	1	8.40		0.54
K	1	95.20		6.17*
P × K	1	0.48		0.03
N × K	1	33.14		2.15
N × P	1	21.28		1.38
'Error'	12	185.29	15.441	
Stratum total	18	533.07		

There is a marked effect of nitrogen and a less well marked effect of potassium. There is some evidence that they interact, but it is not significant.

7F

(a)

I	(1)	d	bc	bcd
II	a	ad	abc	abcd
III	b	c	bd	cd
IV	ab	ac	abd	acd

A, B × C and A × B × C are all confounded.

(b)

I	(1)	be	cd	abc	abd	ace	ade	bcde
II	a	bc	bd	ce	de	abe	acd	abcde
III	b	e	ac	ad	bcd	cde	abce	abde
IV	c	d	ab	ae	bce	bde	abcd	acde

A × B × E, A × C × D and B × C × D × E are all confounded.

(c)

I	(1)	cd	abc	abd
II	be	ace	ade	bcde
III	a	bc	bd	acd
IV	ce	de	abe	abcde
V	b	ac	ad	bcd
VI	e	cde	abce	abde
VII	c	d	ab	abcd
VIII	ae	bce	bde	acde

The following contrasts are confounded in addition to those in (b): E, A × B, B × C × D and A × C × D × E.

8A The analysis of variance reads:

Source	d.f.	s.s.
Total regression of y on x	1	1806.5
Gain from using x_2 also	1	2440.2
Total regression of y on x_2	1	2471.7
Gain from using x_1 also	1	1775.0
Multiple regression of y on x and x_2	2	4246.7
'Error'	14	198.7
Total	16	4445.4

The partial and total regression coefficients are virtually the same because x_1 and x_2 are uncorrelated.

8B All four bodies of data lead to the relationship.

$$y = 3 + x/2.$$

Nevertheless, they correspond to very different cases:

(I) Here the regression line appears to be straight, but the values of y contain a large random component.
(II) Here the regression relationship is given by a curve and all the points lie close to it.
(III) Again the regression relationship appears to be straight, but there is little random variation apart from the one aberrant pair of values, (13, 12.74), which really requires further investigation.
(IV) Here everything depends upon one value of x that is different from the rest. A regression line, if straight, is fairly well determined, though it would be known better if there were more points with $x = 19$. There is, however, no way of judging from these data whether the regression line is straight or not.

8C Without a covariance adjustment, the analysis of variance of y reads:

Source	d.f.	s.s.	m.s.	F
Treatments	5	749	149.8	0.10
'Error'	15	23 433	1562.2	
Stratum total	20	24 182		

There is no evidence that the treatments are having any effect. The treatment means are:

A	B	C	D	E	O
285	268	275	270	277	280

After a covariance adjustment has been applied, the analysis of covariance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	5	4359	871.8	3.15
'Error'	14	3881	277.2	
Stratum total	19	8240		

The treatment means are:

A	B	C	D	E	O
281	267	274	281	301	252

The adjustment has had two effects:

(a) It has explained much of the 'error' variation and has so reduced the estimated variance from 1562.2 to 277.2.
(b) It has shown that the treatment O (the control) had been assigned most of the best plots. Once this is allowed for, it becomes clear that it is rather an unproductive treatment.

8D To provide a check at an intermediate point in the lengthy calculations, we give the full analyses of variance and covariance.

Source	d.f.	x^2	xy	y^2
L	1	26 107	31 990	39 200
P	1	19 602	157 658	1268 028
L × P	1	36	546	8 256
'Error' i	9	168 543	60 952	434 947
Stratum total	12	214 288	251 146	1750 431
M	1	64 441	70 544	77 224
L × M	1	5 670	1 757	545
P × M	1	40 898	41 398	41 906
L × P × M	1	93 096	60 949	39 903
'Error' ii	12	337 844	196 772	263 583
Stratum total	16	541 949	371 420	423 161

Regression coefficients are: $b_i = +0.3616$: $b_{ii} = +0.5824$.

The adjusted analyses of variance are:

Source	d.f.	s.s.	m.s.	F
L	1	16 865	16 865	0.33
P	1	1 036 063	1 036 063	20.07
L × P	1	7 864	7 864	0.15
'Error' i	8	412 904	51 613	
Stratum total i	11	1 473 696		
M	1	14 201	14 201	1.05
L × M	1	415	415	0.03
P × M	1	6 741	6 741	0.50
L × P × M	1	382	382	0.03
'Error' ii	11	148 976	13 543	
Stratum total ii	15	168 612		

A covariance adjustment leads to non-orthogonality of contrasts that would otherwise be orthogonal. That is why the adjusted sums of squares do not add to the stratum totals.

As to conclusions, there is clearly a highly significant main effect of phosphate, but nothing else requires consideration. (It would, nonethe less, be wise in any report to give adjusted means for all main effects, whether significant or not.) The factor P, lies in the first analysis, so b_i applies, not b_{ii}. Hence, after adjustment of x to its mean, i.e. 703.9, the y-means are

(1). 617; P, 998.

The difference (381) has a standard error equal to the square root of

$$51\,613(1/16 + 1/16 + (49.5)^2/168\,543)$$

i.e. 84.9, so it equals 4.48 times its own standard error.

If there had been any significant effects involving Factor M, b_{ii} and E_{xx} would have been taken from the second analysis. Also, interactions would have been most conveniently expressed in terms of the effect of M depending upon levels of L and P.

8E After adjustment by the covariate, the analysis of variance read:

Source	d.f.	s.s.	m.s.	*F*
Treatments	9	276.33	30.70	8.16
'Error'	26	97.78	3.760	
Stratum total	35	374.11		

The 'error' mean square has been reduced from 3.865 to 3.760, which is not of much importance. On the other hand, K, according to (8.6.8), has been increased from 0.500 to 0.515, so the situation is little changed.

8F After adjustment by $w = \sin\theta$ and $x = \cos\theta$, the 'error' mean square becomes 3.811 with 25 degrees of freedom. Again, nothing has been gained. There are in fact no grounds for supposing that the long, narrow blocks were badly chosen.

8G The effect of the covariance adjustment is to reduce the 'error' mean square to 11.892.

9A The means and ranges of the three treatments are as follows:

Treatment	Mean	Range
A	13.5	11
B	28.4	17
C	49.8	20

The ranges increase roughly with the square roots of the means, which indicates a square-root transformation, and that is not unreasonable. Taking square roots to two decimal places leads to this analysis:

Source	d.f.	s.s.	m.s.	*F*
Treatments	2	46.179	23.090	83.03***
'Error'	21	5.841	0.2781	
Stratum total	23	52.020		

There are clearly large differences between the treatments. Results are best expressed by squaring the means of the transformed variates, i.e.

A, 13.2; B, 28.1; C, 49.5

rather than the means of the untransformed variate as given above.

9B Here there is the complication of blocks, but again the simplest approach is to relate means and ranges, i.e.

Treatment	Mean	Range
A	13.8	17
B	13.5	21
C	10.0	18
D	7.3	18
E	24.2	38

The mean value for E is about twice that for A–D taken together, and the same can be said of the ranges. That implies a need for a logarithmic transformation. On the other hand, it is more reasonable in the present instance to expect a Poisson distribution (see Section 3.8), in which case the standard error (and hence the range) would vary, so the square root is the mean. Whichever transformation is used, there will be reservations, but either would be better than no transformation at all.

If these data stand in isolation, the cautious may take refuge in the method of Section 9.6. That is to say, the differences between Treatments, A, B, C and D are unimportant, whereas E does look successful. The six differences between F and the mean of the rest are:

I	II	III	IV	V	VI
+ 10.8	+ 16.2	+ 3.8	+ 24.2	+ 17.2	+ 7.2

That leads to an analysis of variance, thus:

Source	d.f.	s.s.	m.s.	*F*
Contrast	1	1050.73	1050.73	19.03**
'Error'	5	276.11	55.222	
Total	6	1326.84		

Critics may complain that information is being wasted, but at least the method is fairly safe.

If the data are part of a series, the other experiments will give a clue which transformation to choose.

9C Again the first task is to relate means and ranges:

Treatments	Means	Range
A	5.0	12
B	24.8	61
C	16.8	44
D	8.6	27

In general the ranges are in much the same proportion as the square roots of the means. That suggests the need for a square-root transformation, However, discontinuity is serious, so the square root of $(y + 0.375)$ would be better.

The analysis accordingly reads:

Source	d.f.	s.s.	m.s.	F
Treatments	3	28.435	9.478	1.88
'Error'	21	106.096	5.0522	
Stratum total	24	134.531		

On the transformed scale the variety means are:

A, 2.05; B, 4.51; C, 3.52; D, 2.55.

The standard error of C difference between two treatments is 1.124. For 21 degrees of freedom and $P = 0.05$, $t = 2.080$, so a difference larger than 2.34 must be accounted significant at that level. There is no variety with a mean less than $(3.52 - 2.34)$, so none show any improvement. Even on a one-tailed test that remains better, but only just.

10A This is a particularly awkward body of data because it exhibits both the features that raise difficulties of analysis. First, there are signs of strong interaction of sites with one of the factors, namely, nitrogen. At Friars Hill, Lower Friars Hill and Old Road it gives a marked positive effect; elsewhere the effect, if it exists at all, is negligible. (To anyone who knows Antigua the names will be meaningful, but clearly soil response to nitrogen differs from one part of the island to another.) Secondly, the various experiments gave widely different error mean squares, which implies the need to give a higher weighting to some (the more precise) than to others. Each of these features could be established by a formal test, but the exercise hardly seems necessary. (An analysis of variance could be used for the interaction of treatments and sites: Bartlett's test, explained in Section 9.6, for the homogeneity of variances.)

With this double difficulty the best course is probably to consider each site in isolation. There are enough data to estimate the three main effects and the three two-factor interactions for each of the eight localities, and this would give an indication of how to fertilize at each. If the values of those six quantities were marked on a map they might well show the responses to be expected in different areas.

If, on the other hand, there had been only a few observations at each site this procedure would not prove very informative. In that case the best procedure might well be to test each of the six effects against its own interaction with sites. Strictly speaking, that does no more than indicate whether or not each effect exists on average over the island as a whole, but that would be worth knowing and, in the absence of enough data at each site, is as much as can be expected.

10B The two analyses of variance read:

		L		H	
Source	d.f.	s.s.	m.s.	s.s.	m.s.
Effect	1	116.8544	116.8544	0.0138	0.0138
Sites (S)	2	7.9574	3.9787	0.3069	0.1534
Year (Y)	2	1.1751	0.5876	0.0470	0.0235
N-Index (I)	1	3.1896	3.1896	0.0370	0.0370
Y × I	2	1.6336	0.8168	0.0008	0.0004
S × I	2	1.7099	0.8550	0.0158	0.0079
S × Y	4	0.6641	0.1660	0.2141	0.0535
S × I × Y	4	0.9123	0.2281	0.3632	0.0908
'Error'	36	3.2087	0.08913	3.2627	0.09063
Stratum total	54	137.3051		4.2613	

To take L first, there is clearly a large effect. Its mean square is much the largest of any in the analysis and enables one to say, not just that there is

an effect overall, but that there is one for all sites and years regardless of N-Index. The existence of other significant effects in the analysis shows, however, that its magnitude depends upon circumstances. For example, those for sites ($F = 44.64$) and N-Index ($F = 35.79$) are especially marked and show that the value of the effect varies markedly with those factors. For years, $F = 6.59$ and that also is significant, though one might have expected it to be larger. Further it appears to interact with both sites and years. Some might argue that all these effects would have been foreseen; consequently they require estimation rather than testing. That may well be so, but the analysis of variance needs to be calculated, and a preliminary survey of what it indicates does no harm, provided previous knowledge is not forgotten. (An experiment is rarely the sole source of information about the subject; it needs to be interpreted in the light of its predecessors.)

Estimation is, nonetheless, essential, whether there have been preliminary tests or not. Here the most important differences are associated with sites, N-Index and their interaction, so a table to show those effects should be helpful. It reads:

	X	Y	Z	Index means
Index = 0	+ 2.84	+ 2.39	+ 2.04	+ 2.42
= 1	+ 2.44	+ 2.12	+ 0.65	+ 1.74
Site means	+ 2.64	+ 2.26	+ 1.34	
Differences	+ 0.40	+ 0.27	+ 1.39	

There is an interaction. Though it is small relative to the main effects, it should, nonetheless, be looked at first. In general it may be summed up by saying that the effect of residual nitrogen (Index 1 compared with Index 0) is more marked when, as at Site Z, the crop is less good, supposedly on account of poor soil. Differences within the table have a standard error of 0.141. (It is found by taking 0.08913, multiplying by 4, dividing by 18, the number of data used to calculate each mean, and taking the square root. We recall that the figures in the table are themselves each the mean of two data, and a difference of two such quantities involves four data. That explains the use of 4 as a multiplier and not 2.) Here it should be remembered that the figures represent the effect of applied nitrogen ($N_2 - N_0$). On poor soil with good supplies of residual nitrogen, applications of that element have had little effect (0.65) compared with figures elsewhere in the table. (Perhaps there was some inhibition of cropping that had little to do with nitrogen, so, where there was already enough of that element, further supplies did no good. Questions of that sort can be

decided only by those with local knowledge.) The interpretation of the main effects is obvious, but must be made with the interaction in mind.

The figures given say nothing about the effect of years, which was not very large anyway. Since no one can control the weather there is often little point in saying much about the year differences, unless one of the seasons was remarkable on account of drought or some similar feature. In this instance we have no information about that, but we do note that there was an interaction of years and N-Index, as follows:

	1	2	3
N-Index = 0	+ 2.43	+ 2.49	+ 2.36
N-Index = 1	+ 1.25	+ 1.62	+ 2.34
Year means	+ 1.84	+ 2.05	+ 2.35

The standard error is as before, since each figure is again the mean of 18 data. The result here is surprising and not easy to interpret. The figures suggest strongly that residual nitrogen had little effect in the third year, the effect of applied nitrogen being much as usual. Could it have rained hard at the beginning of the season so that residual nitrogen was leeched out? Again, more intimate knowledge of all the circumstances would be a help.

Turning to the other variate, the 'error' sum of squares was much the same as for L, but there is no suggestion of anything being significant, not even the effect itself ($F = 0.0138/0.090\,63 = 0.15$). There is therefore in a sense nothing to interpret, but it would still be wise to set out means and standard error as for L.

10C Any suggestions for the design of a rotation experiment with different lengths of rotation will be criticized by someone, but here is one approach.

First, we can compare the three-year rotations:

I	F	C	M	...
II	C	C	M	...
III	C	M	M	...
IV	X	C	M	...

(We are assuming that it is possible to grow cotton after cotton.) Next we can add the six-year rotations, including one year's fallow, like this:

I/II	F	C	M	C	C	M	...
I/III	F	C	M	C	M	M	...
I/IV	F	C	M	X	C	M	...

These latter rotations require six phases, but I–IV require only three. We can economize a little by associating plots that differ in phase by three years, thus:

Year	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	
I/II	F	C	M	C	C	M	F	C	M	C	C	M	...			
I/III	F	C	M	C	M	M	F	C	M	C	M	M	...			
I/IV	F	C	M	X	C	M	F	C	M	X	C	M	...			
I	F	C	M	F	C	M	F	C	M	F	C	M	...			
II	C	C	M	C	C	M	C	C	M	C	C	M	...			
III	C	M	M	C	M	M	C	M	C	M	C	M	...			
IV	X	C	M	X	C	M	X	C	M	X	C	M	...			
I/II	[F	C	M]	F	C	M	C	C	M	F	C	M	C	C	M	...
I/III	[F	C	M]	F	C	M	C	M	M	F	C	M	C	M	M	...
I/IV	[F	C	M]	F	C	M	X	C	M	F	C	M	X	C	M	...

The ten treatments may be arranged in one randomized block, or preferably more. Useful information on yields of cotton as affected by previous cropping will be obtained in years 2, 4, 5, 8, 10, 11, etc. and on millet in years 3, 5, 6, 8, 9, 12, etc. A little information about the new crop, X, will be found from years 4, 7, 10, etc. For completeness of information and to guard against the mishaps of season (e.g. a failure of millet in year 6), it would be wise to start replicate blocks one year later and two years later, so as to give all six phases in each year. It would also be desirable to split the plots, perhaps into four sub-plots each, so as to add the factors of weedkiller and fertilizer, each at two levels.

10D The analyses of variance and covariance read:

Source	d.f.	a^2	ab	b^2
Treatments	3	36.30	+ 7.017	5.193
'Error'	13	144.50	+ 22.631	5.583
Stratum total	16	180.80	+ 29.648	10.776

Hence $\cos\theta = 0.635$, $\theta = 0.50°$.

Transformations are:

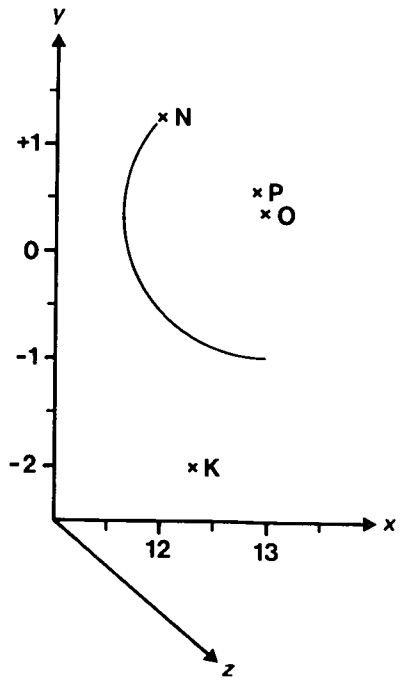
$$x = +0.300a$$
$$y = -0.395a + 2.525b.$$

Bivariate $F = 3.62$ with 6 and 24 degrees of freedom.

Treatment means are:

	a	b	x	y	Distance from O
O	43.2	6.92	12.97	0.36	—
N	40.0	6.77	12.00	1.28	1.34
P	43.0	6.96	12.90	0.56	0.21
K	41.0	5.62	12.30	− 2.01	2.46

For the contrast between O and any other treatment, $K = (1/8 + 1/4) = 0.375$, so L in (10.5.1) = 1.32 for $P = 0.05$.



Compared with 0, a heavy dressing of nitrogen had led to small decreases both in number of cobs and their weight, which together are just significant ($P = 0.05$). (Note that y indicates weight of cobs after an adjustment for their number. If that adjustment is omitted, the value for n is slightly less than that for 0.) A large application of potassium has led to

the same two effects, but more marked in both cases. Using two-univariate analysis of variance, the effect of potassium on crop weight comes through clearly, but nothing else does.

10E We suggest $C = 1.061$, $N = D = 15.9$, $\ominus = 3.39°$.
$L = 18.4$.
It is a little on the short side since L could go to 20.

10F The Papadakis covariate is:

+ 0.72	+ 1.80	− 1.48	− 0.66	− 1.35	− 2.19
+ 1.30	− 0.85	− 1.12	− 3.40	− 2.39	− 3.11
+ 1.41	− 0.85	− 2.12	− 2.48	− 3.51	− 1.92
+ 0.21	+ 1.67	− 0.43	− 0.42	− 0.28	− 1.82
+ 3.92	+ 1.68	+ 2.14	+ 0.77	+ 0.88	+ 0.41
+ 1.86	+ 3.90	+ 1.67	+ 2.28	− 0.06	+ 0.77

As a result of using it, the 'error' mean square is reduced to 5.516 with 28 degrees of freedom. Without local control it was 8.582 with 30 degrees of freedom. Using the Latin square (Exercise 1F) it was 3.047 with 20 degrees of freedom.

10G We first guess $r = 6$, which would make $f = 15$, $t = 2.947$ and $t' = 1.753$. The expression at (10.9.2) leads to $r = +7.07$. Taking $r = 7$, f becomes 18, t becomes 2.878 and t' becomes 1.734, so r, the required replication, is now estimated as 6.81, which is less than the figure of 7 proposed. Nevertheless, we advise that replication should not be minimal. Eight replicates would be better but nine excessive.

11A

Density	*3*	*6*	*9*
LER	1.38	1.30	1.53
ELER ($r = \frac{1}{2}$)	1.37	1.21	1.36
SLER ($t = 900$)	1.16	1.11	1.15

11B The transformations are:

$$x = 2.81a$$
$$y = 2.62b - 0.37a.$$

Treatment means are:

	a	*b*	*x*	*y*
A P	0.48	1.82	1.36	4.93
Q	0.32	3.38	0.88	8.93
B P	1.66	1.23	4.66	3.85
Q	1.52	2.21	4.26	6.36

The bivariate F for all treatments is 12.89 with 6 and 16 degrees of freedom. For the main effect of sorghum varieties, the main effect of millet varieties and the interaction, the bivariate F had values of 13.88, 11.32 and 0.97, respectively.

Looking at the means of x and y, those for sorghum are:

	x	*y*
Early	1.12	6.93
Mid-season	4.46	5.10

The mid-season variety has given more sorghum with some loss of millet, having proved a strong competitor.

For millet the means are:

	x	*y*
Short-stemmed	3.01	4.39
Long-stemmed	2.57	7.64

The long-stemmed variety has given more millet, but there may be a slight loss in the yield of sorghum. However, judging from the invariate analysis of x, that is not significant.

11C The transformations are:

$$x = 0.0479a$$
$$y = 0.0155a + 0.1340b.$$

Consequently the treatment means for the eight treatments are:

	T0		T4		T8		T12		T16	
	x	*y*	*x*	*y*	*x*	*y*	*x*	*y*	*x*	*y*
O	3.17	9.20	4.22	9.03	3.08	6.61	4.20	8.34	3.31	5.12
M	4.02	10.90	3.79	9.34	2.88	6.96	4.45	8.22	3.12	4.75

It is a simple matter to plot those points on a bivariate diagram.

After transformation the analyses of variance and covariance become:

Source	d.f.	x^2	xy	y^2
Time of planting cassava	4	9.7793	21.0722	131.6072
Melons	1	0.0390	0.2373	1.4243
Interaction	4	2.0233	2.3362	5.1067
'Error'	27	26.9820	− 0.0153	27.0080
Stratum total	36	38.8237	23.6304	165.1462

It will be seen that the two 'error' sums of squares do not exactly equal the 'error' degrees of freedom, nor is the 'error' sum of products exactly equal to zero, but that is the result of rounding. If the bivariate analysis is to be obtained precisely, more decimal places are needed in the coefficients of the transformation. However, although the bivariate analysis lies at the heart of the method, it is not always necessary to calculate it. The mean values of x and y can be found without it, while the values of the bivariate F are better found from the analyses of variance and covariance of a and b, i.e. as given in the Exercise, because to use x and y would be to incorporate the rounding errors already noted.

To consider the interaction first, A equals

$$(11\,762 \times 1662 - 1362^2)/(12\,644 \times 1874 - 1100^2)$$

which equals 0.7869, showing the bivariate F to be equal to 0.83 with eight and 26 degrees of freedom. That is clearly not significant, so the correct procedure is to examine the two main effects. The bivariate F for the melons is 0.69, so again there is no sign of any effect. Hence it may be concluded that the interplanting of melons has had no discernible effect, so anything gained from their crop has been obtained without loss of cassava. The last bivariate F is that for the main effect of time of planting cassava. It equals 11.60 with 8 and 26 degrees of freedom. Clearly that gives strong evidence of an effect, and the tables show that there is advantage to the cassava in planting it early.

11D

The means for the combinations of V and F are:

	x	y
V_1F_1	1.05	4.92
V_1F_2	2.52	2.96
V_2F_1	3.83	3.60
V_2F_2	9.03	0.68

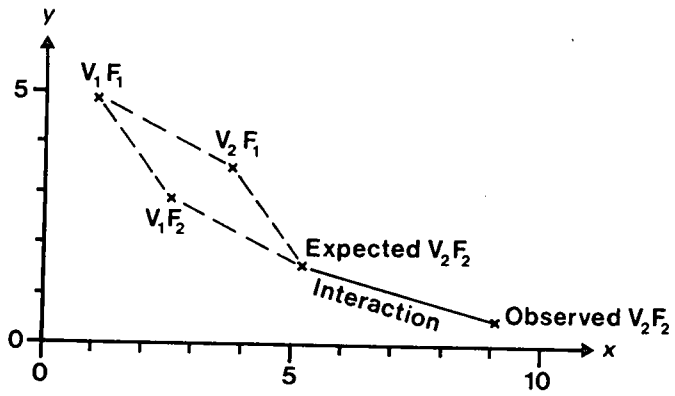
In the absence of an interaction, V_2F_2 would be expected at the point

$$(3.83 + 2.52 - 1.05,\ 3.60 + 2.96 - 4.92)$$

i.e. at (5.30, 1.64). It is in fact, at (9.03, 0.68), displaced by a distance equal to the square root of

$$(9.03 - 5.30)^2 + (0.68 + 1.64)^2 = 3.85^2$$

The following diagram shows the form of the interaction:



It appears V_2F_2 has given much more sorghum than would be expected from the performance of the other treatments and has done so without having had much effect on y.

11E To take the contour for 30 000 MJ first, this calls for 96 MJ per plot of 32 m². We will start with $a = 60$. In that case the maize will be contributing 58.74 MJ, leaving the cassava to provide 37.26 MJ, so b must equal 41.49 MJ. Hence $x = 2.874$ and $y = 6.490$. Another point is that for $a = 75$, $b = 25.24$, $x = 3.592$, $y = 4.531$. The simplest way of drawing the contour is to put a straight line between the two points just found. If the equation of the line is needed, it is

$$(x - 2.874)/(3.592 - 2.874) = (y - 6.490)/(4.531 - 6.490)$$

or

$$x + 0.367y = 5.25.$$

Turning to the contour for 40 000 MJ, corresponding points are given by:

$$a = 60, \quad b = 77.13, \quad x = 2.874, \quad y = 11.265;$$
$$a = 75, \quad b = 60.77, \quad x = 3.592, \quad y = 9.306.$$

This leads to the equation

$$x + 0.367y = 7.00.$$

It will be noted that 5.25/7.00 = 30 000/40 000.

Once the contours have been marked on the bivariate diagram, it will be apparent that higher calorific values for the combined crops are obtained when the cassava is planted early.

12A

$$m = 1.130 \qquad m' = 0.624.$$

The analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	2	0.723 31	0.361 66	174.63***
'Error'	7	0.014 50	0.002 071	
Stratum total	9	0.737 81		

The damaged contrast is $(-1 \; -1 \; +2)$, so the approximate effective replications of treatments for study of the linear, $(-1 \quad 0 \; +1)$, and quadratic, $(-\frac{1}{2} \; +1 \; -\frac{1}{2})$, effects are respectively 4.25 and 4.75.

The linear effect is estimated as 0.569 and its variance is 0.002 071 (2/4.75) = 0.0312^2. It is therefore very highly significant. The quadratic effect is estimated as −0.159 with a variance of 0.002 071 (1.5/4.97) = $(0.0250)^2$, so the figure for Fe2 is significantly below the mean of the other two.

12B Essentially there are three main methods:

(a) The fitting of missing-plot values, whether by the method of Section 12.2, by Rubin's method or in any other way.
(b) The use of covariance.
(c) By the Kuiper–Corsten iteration. However, in this text we have not shown the extension to row-and-column designs, so that is not available in this instance.

(a) *Missing-plot values*

$$m = 464.3 \qquad m' = 441.6.$$

The analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	4	21 828	5457	5.05
'Error'	11	11 898	1081.6	
Stratum total	15	33 726		

(b) *Covariance*

The analyses of variance and covariance read, m having been put equal to zero, like this:

Source	d.f.	x^2	xy^2	y^2
Treatments	4	0.16	− 59.76	43 159
'Error'	12	0.48	− 222.88	115 387
Stratum total	16	0.64	− 282.64	158 546

Adjusting y by x gives the analysis of variance at (a). Further, 222.88/0.48 = 464.3 = m, so means, etc., will be the same by either method.

The conversion factor to tonnes per hectare is 0.448.

12C

$$12m_1 + m_2 = 202.4$$
$$m_1 + 12m_2 = 187.2$$

Hence $m_1 = 15.68$ and $m_2 = 14.29$.

Further, $m_1' = 13.80$ and $m_2 = 15.13$, so the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Treatments	3	15.1211	5.040 4	10.78
'Error'	10	4.6738	0.467 38	
Stratum total	13	19.7949		

Using covariance, setting $m_1 = m_2 = 0$, the analyses of variance and covariance read:

Source	d.f.	w^2	x^2	y^2
Treatments	3	0.15	0.15	37.286
'Error'	12	0.60	0.60	297.099
Stratum total	15	0.75	0.75	334.385

Source	d.f.	xy	wy	wx
Treatments	3	− 0.23	− 1.99	− 0.05
'Error'	12	− 10.12	− 9.36	+ 0.05
Stratum total	15	− 10.35	− 10.35	0.00

From the 'error' line the partial regression coefficients are − 14.29 (= − m_1) and − 15.68 (= − m_2). From the 'stratum total' line they are similarly − 15.13 and − 13.80. It will be found that if y is adjusted by both w and x, the resulting analysis of variance will be that given above.

For the three contrasts the value of K from (8.7.2) become respectively 0.545, 0.467 and 0.400. Since s^2 is 0.467 38, the respective standard errors are 0.505, 0.467 and 0.432. Using the methods of Exercise 4A, the values of K are respectively 0.500, 0.425 and 0.400, so, as far as the damaged treatments were concerned, it was helpful to have data from all the plots, even if some of them involved a mistake.

12D The equations for the missing-plot values are:

$$\begin{aligned} 10m_1 + m_2 - 2m_3 &= 270.8 \\ m_1 + 10m_2 - 2m_3 &= 175.4 \\ -2m_1 - 2m_2 + 10m_3 &= 148.4. \end{aligned}$$

They convert into three leading equations:

$$\begin{aligned} m_1 &= 27.08 - 0.1m_2 + 0.2m_3 \\ m_2 &= 17.54 - 0.1m_1 + 0.2m_3 \\ m_3 &= 14.84 + 0.2m_1 + 0.2m_2. \end{aligned}$$

We will start by putting m_2 and m_3 equal to their respective treatment means. The iteration proceeds thus:

Cycle	m_1	m_2	m_3
0	—	18.80	27.80
1	19.64	24.36	23.64
2	29.37	19.33	24.58
3	30.06	19.45	27.74
4	30.08	19.48	24.75
5	30.08	19.48	24.75

12E Although the complete data could be analysed using the Kuiper–Corsten iteration, it is easier to use the method of covariance. (There is no simple way of expressing a residual, so missing-plot values will not be found easily.)

It is therefore proposed to put $m_1 = m_2 = 0$. The effect of this is to add 78 350 to the stratum total sum of squares in Exercise 4F and + 27.0, + 168.0, − 43.0 and − 152.0, respectively, to Q_A, Q_B, Q_C and Q_D. With the enlarged design $\lambda = 52$, $\lambda_o = 36$, $R = 16$, $R_o = 144/13$.

Two pseudo-variates are needed, one for the plot missing from Treatment A(w) and the other for the plot missing from D(x). Both give a stratum total sum of squares of 11/12, being zero in all blocks except 111. The values of Q are:

	Q_A	Q_B	Q_C	Q_D
w	+ 0.75	− 0.25	− 0.25	− 0.25
x	− 0.25	− 0.25	− 0.25	+ 0.75
y	+ 119.3	+ 433.7	− 333.3	− 219.3

Accordingly the analyses of variance and covariance are:

Source	d.f.	w^2	x^2	y^2
Treatments	3	9/144	− 3/144	22 991
'Error'	52	123/144	− 9/144	119 308
Stratum total	55	132/144	− 12/144	142 299

Source	d.f.	xy	wy	wx
Treatments	3	− 1 454	+ 944	− 3/144
'Error'	52	− 13 746	− 16 194	− 9/144
Stratum total	55	− 15 200	− 15 200	− 12/144

It will be noticed that the sums of squares and products for the covariates w and x have been expressed as fractions and not in decimals, as were the effective replications, R and R_o. Since the fractions are exact, whereas the decimals would involve rounding, there are advantages in doing so.

The resulting analysis of variance for y adjusted by w and x reads:

Source	d.f.	s.s.	m.s.	F
Treatments	3	24 059	8020	6.51
'Error'	50	62 790	1231.2	
Stratum total	53	86 849		

The treatment means, after adjusting all values of w and x to zero, are:

A	B	C	D
182	187	139	157

Standard errors of contrasts can be found in the usual way from the expression at (8.7.2). It can be generalized, if desired, in the same way that (8.6.7) was generalized to (8.6.10).

12F The value of 279 shows a discrepancy from m of 185.3. All plots will have the same value of ψ, which must therefore equal $12/25 = 0.48$. Hence, if 279 were used instead of m, the 'error' sum of squares would be increased by $0.48(185.3)^2 = 16\,481$. Hence the analysis of variance reads:

Source	d.f.	s.s.	m.s.	F
Discrepancy	1	16 481	16 481	15.24**
'Error'	11	11 898	1 081.6	
Total	12	28 379		

The discrepancy is too large to be explained as chance ($P < 0.01$).

12G The discrepancy is $(1.130 - 0.710) = 0.420$. All plots must have the same value of ψ, namely 8/15, so the sum of squares for the discrepancy is $(8/15)(0.429)^2 = 0.094\,08$. The analysis of variance therefore reads:

Source	d.f.	s.s.	m.s.	F
Discrepancy	1	0.094 08	0.094 08	45.43***
'Error'	7	0.014 50	0.002 071	
Total	8	0.0108 58		

The discrepancy is clearly significant ($P < 0.001$).

Suggestions for Further Reading

General There are many books on crop experimentation, though some of the best are now rather out-of-date. Among recent publications are those of Preece (1982) and Pearce (1986a).

Chapter 1 The method of sweeping derives from the paper of Wilkinson (1970). The concept of strata was advanced by Nelder (1965a,b). On precision of measurement we mention the papers of Yule (1927), Preece (1981) and Riley *et al.* (1983).

There is an interesting historical survey in the paper by Kempton (1984), which also considers some alternatives to traditional methods.

Chapter 2 The law of environmental variation given by Fairfield Smith (1938) has been widely used to decide questions of plot size and shape, but not always with happy results. A recent paper by Brewer and Mead (1987) goes into the matter in some detail, besides giving a useful survey of the literature.

Chapter 3 There are many texts that advocate using multiple comparisons as if that represented accepted wisdom. In fact, ever since the methods were introduced, they have occasioned dissent. Papers that put the case for the other side are those of Chew (1976), Little (1981), Bryan-Jones and Finney (1983), Pearce (1983b) and Perry (1986).

Chapter 4 In essence this chapter is an attempt to take some established mathematical results and to make them accessible to those who may have occasion to use them. The underlying results are mostly to be found in the book by Pearce (1983a) in Sections 3.5, 5.2, 5.3, 5.8, 6.1, 6.2, 6.6 and in Example 6C. There is also the paper of Pearce (1987).

Supplemented balance has been studied by Pearce (1960a), Beckhofer (1969) and by Beckhofer and Tamhane (1981). Lattice designs were developed by Yates (1936a).

Chapters 5 and 6 The idea of contrasts is a very old one, though only recently has it begun to play much part in the general theory of statistics. Special cases, however, have been studied extensively. Thus, polynomials to a high order were presented a long time ago by Fisher and Yates (1938).

As to other partitions, the first paper on the analysis of variance (Fisher and Mackenzie, 1923) dealt with a factorial design. The two classic publications of Yates (1935, 1937) remain useful to this day, not least for their treatment of confounding.

Chapter 7 Designs with split plots have a reputation for being rather mysterious; it is hoped that this chapter will have dispelled some of the darkness. Much of the difficulty comes from teachers making an early distinction between blocks and plots and then having to minimize it. That is where an understanding of strata helps (Nelder 1965a,b). Single replication experiments have been considered by Kempton (1984). Fractional replication was developed by Finney (1945, 1946, 1950).

Chapter 8 Considering that the analysis of covariance was developed in an agricultural context by R. A. Fisher over fifty years ago, there is surprisingly little about it in the literature, though it is used by many people as a standard technique and many computer packages facilitate its use. A complete number of *Biometrics* (Vol. 13, Part 2, 1957) was dedicated to it; then, to commemorate the 25th anniversary, a further group of papers was published in Volume 38, Part 2 (1982). Volume 8, Part 8 (1979) of *Communications in Statistics* was similarly dedicated to the subject. Useful accounts of the method have been given by Bliss (1970, chapter 20) and Finney (1962). The paper of Preece (1980) expresses some necessary warnings. That of Wishart (1934) gives the modern form of the technique. The data of Exercise 8C have been studied by a number of authors, the remarks of Cox (1958) being especially illuminating.

Local control by covariance on pseudo-variates was suggested by Federer and Schlottfeldt (1954) and taken further by Outhwaite and Rutherford (1955). The use of Fourier functions was suggested by Dyke *et al.* (1982).

Chapter 9 It would be a pity if some of the earlier papers were overlooked, especially that of Cochran (1938a). Hoyle (1973) has given a useful bibliography. Corrections for discontinuity were investigated by Anscombe (1948). The paper by Box and Cox (1964) shows how powerful transformations can be at stabilizing variance. The method of Section 9.6 is very old (Tharp *et al.*, 1941).

Chapter 10 On the subject of series of experiments there are some established classics, notably the papers of Yates and Cochran (1938) and the relevant passages in the book by Cochran and Cox (1950). For rotation experiments useful papers are those of Yates (1954), Patterson (1964) and Preece (1986). The last contains a short bibliography.

On experiments with long-term crops there are the two editions of the book by Pearce (1953, 1976); each contains material lacking in the other. Sources of variation were investigated by Pearce (1960b). Those interested in long-term experiments with annual species will find much of value in the paper of Patterson and Lowe (1970).

Multivariate methods go back to Wilks (1932) and were taken further, among others, by Rao (1952). Pearce and Gilliver (1978, 1979) pointed out the special interest of bivariate methods.

Fan trials owe much to the paper by Nelder (1962). A bibliography was given by Pearce (1976). The papers by Freeman (1964, 1969) and Cleaver *et al.* (1970) show how useful systematic designs can be. Serial balance was investigated by Dyke and Shelley (1976) and by Jenkin *et al.* (1979).

Nearest neighbour methods again have a large literature, starting with two papers by Papadakis (1937, 1940) and one by Bartlett (1938). More recent work has been set out by Bartlett (1978) and Wilkinson *et al.* (1983); with both, the discussion contains much of value. Mention should also be made of the papers by Besag and Kempton (1986).

An introduction to the changing of treatments has been given by Freeman (1959).

Chapter 11 Among general papers on the statistical aspects of intercropping experiments, the following should be mentioned: Federer (1979), Mead and Stern (1980), Mead and Riley (1981), Pearce and Edmondson (1983, 1984).

The use of bivariate analysis of variance has been considered by Pearce and Gilliver (1978, 1979) and by Gilliver and Pearce (1983).

Land Equivalent Ratios were considered by Mead and Willey (1980) and have been taken further by Riley (1984) and by Reddy and Chetty (1984).

The paper by Oyejola and Mead (1982) considers the behaviour of Land Equivalent Ratios in the analysis of variance.

Chapter 12 Missing data also have a large literature; it goes back to the paper by Allen and Wishart (1930). More recent papers are those of Rubin (1972) and of Pearce and Jeffers (1971). Recently Pearce (1986) has examined the consequences of a range of data defects.

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