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Agricultural scientists need to apply correct statistical techniques in their experiments. However, many such scientists in developing countries have little training and experience with statistics. This book is for their use. It will help them to grasp statistical concepts and to apply them in practice. The chapters of the book discuss valid experimental designs, single-factor experiments, two-factor experiments, three-or-more-factor experiments, comparisons among treatment means, regression and correlation analyses, covariance analysis, the Chi-square test, problem data, soil heterogeneity, competition effects, sampling in experimental plots, and mechanical errors. The chapter on sampling in experimental plots discusses basic concepts in sampling, the sampling population, sampling unit, sample size, sampling design, simple random sampling, multistage sampling, stratified random sampling, methods of estimation, and development of a sampling plan. The chapter on mechanical errors discusses furrowing, selection of seedlings, thinning, transplanting, fertilizer application, seed mixture and off-types, plot layout, measurement errors, and transcription of data.

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# Statistical procedures for agricultural research with emphasis on rice

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The International Rice Research Institute is grateful to the Literary Executor of the late Sir Donald A. Fisher, F.R.S., to Dr. Frank Yates, F.R.S., and to Longman Group Ltd., London, for permission to reprint Table III "Distribution of t Probability" from their book *Statistical Tables for Biological, Agricultural and Medical Research* (6th edition, 1974).

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The responsibility for all aspects of this publication rests with the International Rice Research Institute.

## Foreword

Several major cooperative research programs of the International Rice Research Institute (IRRI) are planned and implemented in collaboration with rice scientists in national agricultural research organizations of developing countries. In these areas, financial and trained manpower resources for research are limited. Generally, the majority of the research is field based, and thus, permits the assessment of improved technologies under varied ecological conditions where rice and associated crops are grown. Sound statistical techniques are essential if the limited resources available are to be used effectively for such field experiments to provide the desired evaluations of new rices and associated technology.

Most field trials are planned and implemented by agricultural subject matter specialists with limited training in statistics. Furthermore, these researchers may not have ready access to trained statisticians for consultation during the planning and action stages of their experiments or for the analysis and interpretation of the resultant data. Obviously, there is a need for a guide and ready reference to help agricultural researchers who specialize in a scientific discipline to use to full advantage the science of statistics in designing, conducting, and analyzing experiments.

This book is written primarily for agricultural scientists who should apply correct statistical techniques in their experiments, but who have sparse training and experience in statistics. These researchers should also benefit from the sections concerned with the analysis and interpretation of data collected. Likewise, this book will help them grasp the statistical concepts essential for effective agricultural research.

The authors of the book have long experience in conducting field experiments with rice and associated annual crops. Furthermore, they have provided consultative assistance on statistics for other agricultural scientists in a wide range of field research. They have used these experiences effectively in producing this readable book, which field researchers throughout the world should find most helpful.

For illustrative material, the authors have drawn upon their own research, as well as that of their colleagues in the University of the Philippines at Los Baños and in the International Rice Research Institute (IRRI). They used effectively their 1-year sabbatic leaves at Michigan State University, USA, to complete the book.

IRRI is pleased to publish this book and compliments the authors for demonstrating how the science of statistics can be applied readily to the experimental problems not only of scientists working with rice and rice-based cropping systems, but also of agricultural researchers in general.

N. C. Brady, Director-General
The International Rice Research Institute

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CHAPTER 1

# Introduction

Most experiments try to verify certain hypotheses suggested by past experiences and observations. A corn breeder, for example, is apprehensive about the slow adoption rate by farmers in Mindanao of the high-yielding corn varieties developed at Los Baños. He visits the corn-growing areas in Mindanao and observes that the new varieties are much more vigorous and look more productive than the native ones in disease-free areas. However, in fields infested with downy mildew, a very destructive and prevalent disease of corn in that region, the damage to the new varieties is more severe than that to the native varieties. He suspects, and therefore, hypothesizes that the new high-yielding corn varieties developed at Los Baños are more susceptible to downy mildew than are the native varieties.

At other instances, theoretical considerations may play a major role in arriving at a suggested hypothesis. For example, it can be shown theoretically that a rice crop removes more nitrogen from the soil than is naturally replenished during one growing season. Thus, we hypothesize that to maintain the yield potential of a rice farm, supplementary nitrogen fertilizer must be added for every rice crop.

Once a hypothesis is framed, the next step is to design a procedure for its verification. This is the experimental procedure, one that usually consists of four parts: (1) selecting the appropriate materials to test, (2) specifying the characters to measure, (3) selecting the procedure to measure these characters, and (4) specifying the procedure to determine whether the measurements made support the hypothesis. The first two are fairly easy for a subject matter specialist to specify. In our example on corn, the test material would probably be the native and the newly developed varieties. The characters to be measured would probably be disease infection and grain yield. For the example on rice, the test variety would probably be one of the recommended rice varieties and the fertilizer levels to be tested would cover the suspected range of nitrogen needed. As for corn, rice yield and other agronomic characters would have to be measured.

There is not much need for knowledge in statistics to arrive at these decisions. But the procedures on how the measurements are to be made and how these measurements can be used to prove or disprove the postulated hypothesis,

depend heavily on techniques developed by statisticians. The precision in measuring a character, for example, is highly dependent upon the experimental design, number of replications, and plot technique used. Furthermore, the decision whether a certain observed difference is considered real or imaginary is based on probabilistic and statistical reasoning.

Thus, statistical tools and procedures have had and will continue to have universal usefulness for all kinds of research. Because of this and because of the rapid proliferation in areas of research specialization, a wide body of procedures has been developed for the biological, physical, and social sciences. In the biological sciences alone, there are different statistical techniques for crop and animal researches, for laboratory and field experiments, for genetic and physiological researches, etc. Although this diversity indicates the availability of appropriate techniques for most research problems, it also indicates the difficulty of matching the correct technique to a given experiment. Obviously, this difficulty increases as there are more and more procedures to choose from.

Choosing the correct statistical procedure for a given experiment must be based on expertise in both statistics and the experimental subject matter. Thorough knowledge on only one of the two would not be enough. Thus, a good choice could be made by (1) a subject matter specialist with some training in experimental statistics, (2) a statistician with some background and experience in the subject matter under experimentation, or (3) the joint effort and cooperation of a statistician and a subject matter specialist.

For most Asian institutions engaged in agricultural research, the presence of a trained statistician is a luxury. Of the already small number of statisticians in the region, only a small fraction have the interest and experience in agricultural research necessary for effective consultation. Thus, probably the most useful alternative is to give the necessary statistical background to the existing agricultural researchers so that they can correctly choose the statistical technique most appropriate for their planned experiment.

This backgrounding is the major objective of this book. We shall try to cover most of the simple and commonly met problems in crop research. The more complex ones will still have to be tackled in consultation with a competent statistician. But even then, this background in statistics will help the researcher communicate his problems to the statistician.

This book has two main features. First, it is written with a minimum of mathematics and a maximum of actual examples. Our experience as agricultural researchers indicates that using familiar examples makes a text more easily read, more readily understood, and better appreciated than detailing the mathematical proof behind a procedure. Second, we have restricted our discussion and examples to the most important annual food crops in Asia. Thus, the book would be most useful to researchers working with annual grain crops, such as rice, corn, field legumes, and sorghum, but not so much to those working with perennial fruit trees or in the animal sciences. Even though the fairly specialized coverage of the book could reduce its users, we

feel that the prime importance of the annual food crops in both research and actual production justifies our special emphasis.

We have divided the book into three parts: (1) experimental design and analysis of variance, (2) methods of analyses. and (3) field plot techniques. These topics are most pertinent in deciding how measurements are made and how these measurements are used to test the hypothesis. All our examples are from experiments conducted at the International Rice Research Institute (IRRI) and at the University of the Philippines at Los Baños (UPLB), both located at Los Baños in the Philippines, an area typical of the humid tropics of Southeast Asia.

CHAPTER 2

# Valid experimental designs

Consider a plant breeder who wishes to compare the yield potentials of a new rice variety, say variety *B*, to a standard variety of known and tested properties, say variety *A*. He lays out two plots of equal size, side by side, and sows one to variety *A* and the other to variety *B*. Grain yield for each plot is then measured and the variety having higher yields is judged the better of the two. Despite the simplicity and common-sense appeal of the procedure just outlined, it has one important flaw. It presumes that any difference between the yields of the two plots is caused by the varieties and nothing else. This certainly is not true. Even if the same variety were planted on both plots, the yield would still vary from one plot to another. Other factors, such as soil fertility, moisture, and damage by insects, diseases, and birds also affect the yields of plots.

Because these other sources of variability affect the yields of plots, a satisfactory evaluation of the two varieties must involve a procedure that can separate varietal difference from the other extraneous sources of variation. That is, the plant breeder must be able to design an experiment that allows him to decide whether the difference observed is really due to varietal difference or due to other extraneous factors.

The logic behind the decision is simple. Two varieties, *A* and *B*, will be considered different in their yielding ability only if the observed yield difference between plots is larger than that expected if all plots were planted to the same variety. Hence, the researcher needs to know the yield difference between plots planted to the same variety as well as the yield difference between plots planted to different varieties.

This difference among experimental plots treated alike is called "experimental error." This error cannot be totally eliminated, however, since two plots will not perform exactly the same no matter how carefully they are treated alike. But since experimental error is the primary basis for deciding whether an observed difference between various treatments is real or just due to chance, an experiment must be designed so that a measure of experimental error is available. Clearly, then, error plays a major role in modern agricultural research. Equally important are the techniques that help measure and minimize experimental error.

## 2.1 REPLICATION

In the same way that at least two plots of the same variety are needed to determine the difference among plots treated alike, experimental error can be measured only if the same variety (or treatment) is repeated at least once. Thus, to obtain a measure of experimental error, replication is needed.

## 2.2 RANDOMIZATION

But there is more involved than simply planting several plots to the same variety. For example, suppose, in comparing two rice varieties, a plant breeder planted varieties *A* and *B* each in four plots as shown in figure 2.1. If this particular area has a unidirectional fertility gradient so that there is a gradual reduction of productivity from left to right, variety *B* would then be handicapped because it is always on the right side of variety *A* and is always in a relatively less fertile area. Thus, the comparison between the yield performances of variety *A* and variety *B* would be biased in favor of *A*. That is, a part of the yield difference between the two varieties would be due to the difference in the fertility levels and not to the varietal difference.

To avoid such bias, treatments must be assigned to experimental plots so that a particular variety is not consistently favored or handicapped. This can be achieved by assigning treatments to the experimental plots at random. Randomization ensures that each variety will have an equal chance of being assigned to any experimental plot and consequently, of being grown under any particular conditions existing in the experimental site. Thus, both replication and randomization are needed to obtain an unbiased estimate of experimental error.

## 2.3 LOCAL CONTROL

Since the possibility of detecting an existing difference between treatments depends upon the size of experimental error, a good experiment minimizes

| PLOT 1 | PLOT 2 | PLOT 3 | PLOT 4 | PLOT 5 | PLOT 6 | PLOT 7 | PLOT 8 |
|---|---|---|---|---|---|---|---|
| A | B | A | B | A | B | A | B |

**Fig. 2.1** A systematic arrangement of plots planted to two rice varieties, *A* and *B*. This scheme does not provide a valid estimate of experimental error.

experimental error. A good technique to reduce experimental error is to assign treatments to experimental plots in such a way that some of the inherent variation among experimental plots does not enter into the difference among treatments. This technique is commonly known as "local control." Local control is generally accomplished through blocking; i.e., by subdividing the experimental area so that more or less homogeneous plots are grouped together. Treatments can then be assigned to each of the different groups or blocks separately thus ensuring that variations among groups do not contribute to experimental error. (For further discussion on blocking, see section 3.3.1).

## 2.4 CONCLUSIONS

To summarize, the *minimum* requirements for a valid experimental design are:

1. Replication—a treatment appears more than once in an experiment to provide a means for estimating experimental error.
2. Randomization—every treatment has the same chance of being favored or handicapped to provide a valid measure of experimental error.
3. Local control—treatments are arranged so that some known causes of variations are eliminated to reduce experimental error.

CHAPTER 3

# Single-factor experiments

## 3.1 INTRODUCTION

Experiments in which only a single factor varies while all others are kept constant are called single-factor experiments. Some examples of such experiments are: (1) a yield test of several rice varieties, (2) a fertilizer trial involving several rates of only one fertilizer element, and (3) a trial involving several plant densities.

For single-factor experiments, two groups of designs can be used. One group is the family of incomplete block designs, which are appropriate for experiments with large numbers of treatments, e.g., a varietal test involving 100 entries. The other group is the family of complete block designs, which are fitted for experiments with small numbers of treatments, e.g., a fertilizer trial involving one variety and six rates of nitrogen fertilization. This chapter will detail the three most commonly used complete block designs: (a) completely randomized design, (b) randomized complete block design, and (c) Latin square design; and the lattice designs, the most commonly used incomplete block design for agricultural experiments.

## 3.2 COMPLETELY RANDOMIZED DESIGN

The completely randomized design is most appropriate for experiments with homogeneous experimental units. Thus, it is commonly used in laboratory experiments where environmental effects are easily controlled. This design is rarely used in field experiments, but because of its simplicity, a discussion of it will help you understand more complex designs.

### 3.2.1 Randomization and layout

Randomization is accomplished by assigning treatments to experimental units entirely at random. This can be done by using a random number table, by drawing cards, by throwing dice, or by any other operation that serves the same purpose.

For illustration, we shall discuss an experiment involving four treatments, $A$, $B$, $C$, and $D$, each replicated five times. The randomization and layout of plots are accomplished as follows:

1. Determine the total number of experimental plots. For our example, the total number of experimental plots is $5 \times 4 = 20$.

2. Assign a plot number to each experimental plot in any convenient manner, for example, consecutively from 1 to 20.

3. Assign treatments to the experimental plots by using one of the following schemes:

*3.2.1.1 By table of random numbers.*

STEP 1. Locate a starting point in a table of random numbers (appendix 1) by pointing a finger, with your eyes closed, to any position in a page.

STEP 2. Using the starting point obtained in step 1, select $n$ three-digit numbers, where $n$ is the total number of experimental plots ($n = 20$ for our example). Three-digit numbers are preferred since they are less likely to include ties. Indeed, any higher digit numbers can be used.

For example, starting at the intersection of the sixth row and the twelfth column read downward vertically to get 20 three-digit numbers:

| Random number | Sequence | Random number | Sequence | Random number | Sequence | Random number | Sequence |
|---|---|---|---|---|---|---|---|
| 937 | 1 | 749 | 6 | 918 | 11 | 549 | 16 |
| 149 | 2 | 180 | 7 | 772 | 12 | 957 | 17 |
| 908 | 3 | 951 | 8 | 243 | 13 | 157 | 18 |
| 361 | 4 | 018 | 9 | 494 | 14 | 571 | 19 |
| 953 | 5 | 427 | 10 | 704 | 15 | 226 | 20 |

STEP 3. Rank the 20 selected numbers from the smallest to largest.

| Random number | Sequence | Rank | Random number | Sequence | Rank |
|---|---|---|---|---|---|
| 937 | 1 | 17 | 918 | 11 | 16 |
| 149 | 2 | 2 | 772 | 12 | 14 |
| 908 | 3 | 15 | 243 | 13 | 6 |
| 361 | 4 | 7 | 494 | 14 | 9 |
| 953 | 5 | 19 | 704 | 15 | 12 |
| 749 | 6 | 13 | 549 | 16 | 10 |
| 180 | 7 | 4 | 957 | 17 | 20 |
| 951 | 8 | 18 | 157 | 18 | 3 |
| 018 | 9 | 1 | 571 | 19 | 11 |
| 427 | 10 | 8 | 226 | 20 | 5 |

STEP 4. Assign treatments to plots. Use the rank as the plot number and use the sequence in which the random numbers occurred to refer to treatments. Thus in our example, assign treatment $A$ (i.e., the first treatment) to plot numbers 17, 2, 15, 7, and 19; treatment $B$ to plot numbers 13, 4, 18, 1, and 8; treatment $C$ to plot numbers 16, 14, 6, 9, and 12; and treatment $D$ to plot numbers 10, 20, 3, 11, and 5.

| Plot no. → / Treatment → | | | |
|---|---|---|---|
| 1 *B* | 2 *A* | 3 *D* | 4 *B* |
| 5 *D* | 6 *C* | 7 *A* | 8 *B* |
| 9 *C* | 10 *D* | 11 *D* | 12 *C* |
| 13 *B* | 14 *C* | 15 *A* | 16 *C* |
| 17 *A* | 18 *B* | 19 *A* | 20 *D* |

**Fig. 3.1** Field layout of a completely randomized design with four treatments, replicated five times.

The final layout of the experiment is shown in figure 3.1.

*3.2.1.2 By drawing cards.*

STEP 1. Shuffle an ordinary pack of cards and draw 20 cards at random.

STEP 2. Assign ranks to cards according to number and suit. The result may be as follows:

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| J♥ | 3♦ | A♦ | Q♥ | K♣ | 5♥ | 6♣ | 9♠ | 9♥ | 8♠ |
| Rank 14 | 7 | 9 | 15 | 5 | 11 | 2 | 19 | 13 | 18 |
| A♥ | 4♦ | 3♥ | 2♣ | 7♣ | 10♠ | A♣ | 4♠ | 6♥ | 10♣ |
| 16 | 8 | 10 | 1 | 3 | 20 | 6 | 17 | 12 | 4 |

STEP 3. Follow the procedures stated in step 4 of the previous scheme. That is, treatment *A* is assigned to plot numbers 14, 7, 9, 15, and 5; treatment *B* to plot numbers 11, 2, 19, 13, and 18; treatment *C* to plot numbers 16, 8, 10, 1, and 3; and treatment *D* to plot numbers 20, 6, 17, 12, and 4.

Clearly, this procedure is not applicable when the number of experimental units exceeds 52.

*3.2.1.3 By drawing lots.*

STEP 1. Prepare 20 identical pieces of paper and label them with the letters *A*, *B*, *C*, and *D*; each letter appears five times.

STEP 2. Mix the 20 pieces of paper thoroughly and place them in a box (or a hat).

STEP 3. Pick out a piece of paper at random. If it is labeled *B*, the experimental plot number 1 will receive treatment *B*.

STEP 4. Without returning the drawn piece of paper to the box, draw a second piece at random. If it is labeled *A*, then treatment *A* will be assigned to the experimental plot number 2.

**Table 3.1 Grain yield of rice using different types of foliar and granular insecticides for the control of brown planthoppers and stem borers, IRRI.**

| Treatment | Grain yields (kg/ha) | | | | Treatment total | Treatment mean |
|---|---|---|---|---|---|---|
| Dol-Mix (1 kg) | 2,537 | 2,069 | 2,104 | 1,797 | 8,507 | 2,127 |
| Dol-Mix (2 kg) | 3,366 | 2,591 | 2,211 | 2,544 | 10,712 | 2,678 |
| DDT + γ-BHC | 2,536 | 2,459 | 2,827 | 2,385 | 10,207 | 2,552 |
| Azodrin | 2,387 | 2,453 | 1,556 | 2,116 | 8,512 | 2,128 |
| Dimecron-Boom | 1,997 | 1,679 | 1,649 | 1,859 | 7,184 | 1,796 |
| Dimecron-Knap | 1,796 | 1,704 | 1,904 | 1,320 | 7,624 | 1,681 |
| Control | 1,401 | 1,516 | 1,270 | 1,077 | 5,264 | 1,316 |
| Grand total and mean | | | | | 57,110 | 2,040 |

STEP 5. Repeat the process until all 20 pieces of paper have been drawn.

### 3.2.2 Analysis of variance: completely randomized design with an equal number of replications

Analysis of variance is one of the most powerful and commonly used statistical tools in analyzing experimental data. It allows for the subdivision of the total variability into several causal components. For a completely randomized design (CRD), there are two components of variation: (a) that due to treatment and (b) that due to experimental error. The relative size of these two components provides the basis for determining whether the observed differences among treatments are real or not.

We will use as an example an experiment conducted by the Department of Entomology of the International Rice Research Institute to investigate the effects of foliar and granular applications of some selected insecticides for the control of the brown planthopper and stem borers. The experiment involved six treatments and a control, each replicated four times in a completely randomized design.

The steps involved in constructing the analysis of variance are:

STEP 1. Group the data by treatments and calculate the treatment totals, treatment means, and grand total as shown in table 3.1.

STEP 2. Construct an outline of the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Treatment | | | | | | |
| Experimental error | | | | | | |
| Total | | | | | | |

STEP 3. Determine the degrees of freedom (d.f.) for each source of variation as follows:

Total d.f. = total number of observations − 1
= 28 − 1 = 27,
Treatment d.f. = total number of treatments − 1
= 7 − 1 = 6, and
Error d.f. which can be computed in either of two ways:
(1) as total d.f. − treatment d.f. = 27 − 6 = 21, or
(2) as (total number of treatments) (total number of replications − 1)
= (7)(4 − 1) = 21.

STEP 4. Using X's to designate individual measurements from each plot, $t$ as the number of treatments, and $r$ as the number of replications, calculate the sum of squares (SS) as follows:

$$\text{Correction factor (C.F.)} = \frac{(\text{grand total})^2}{\text{total number of observations}} = \frac{(\sum X)^2}{rt} \tag{3.1}$$

$$= \frac{(57{,}110)^2}{(4)\,(7)} = 116{,}484{,}004,$$

$$\text{Total SS} = \sum X^2 - \text{C.F.} \tag{3.2}$$

$$= [(2{,}537)^2 + (2{,}069)^2 + \ldots + (1{,}270)^2 + (1{,}077)^2] - 116{,}484{,}004$$

$$= 7{,}577{,}412,$$

$$\text{Treatment SS} = \sum \frac{(\text{treatment total})^2}{r} - \text{C.F.} \tag{3.3}$$

$$= \frac{(8{,}507)^2 + (10{,}712)^2 + \ldots + (5{,}264)^2}{4} - 116{,}484{,}004$$

$$= 5{,}587{,}175, \quad \text{and}$$

$$\text{Error SS} = \text{total SS} - \text{treatment SS} \tag{3.4}$$

$$= 7{,}577{,}412 - 5{,}587{,}175$$

$$= 1{,}990{,}237.$$

Throughout this book, the symbol $\sum$ will be used to represent "the sum of." For example, the expression $T = X_1 + X_2 + \ldots + X_n$ can be written as $T = \sum_{i=1}^{n} X_i$, or simply $T = \sum X$.

STEP 5. Calculate the mean squares (MS) by dividing each SS by its corresponding degrees of freedom:

$$\text{Treatment MS} = \frac{\text{treatment SS}}{t - 1} \tag{3.5}$$

$$= \frac{(5{,}587{,}175)}{6}$$

$$= 931{,}196, \quad \text{and}$$

$$\text{Error MS} = \frac{\text{error SS}}{t(r - 1)} \tag{3.6}$$

$$= \frac{1{,}990{,}237}{21}$$
$$= 94{,}773.$$

STEP 6. Calculate the observed $F$-value:

$$F = \frac{\text{treatment MS}}{\text{error MS}} \tag{3.7}$$
$$= \frac{931{,}196}{94{,}773}$$
$$= 9.82.$$

STEP 7. Obtain the tabular $F$-values from appendix 5 using the treatment degrees of freedom as $f_1$ and the error degrees of freedom as $f_2$. The tabular $F$-values for 6 and 21 degrees of freedom at 5% and 1% levels of significance then are 2.57 and 3.81, respectively.

STEP 8. Fill up the analysis of variance table with values computed in steps 4 to 7. The final result appears in table 3.2.

STEP 9. Compare the observed $F$-value with the tabular $F$-values based on the following decision rules:

1. If the observed $F$ is as large as or larger than the tabular $F$ at the 1% level, the differences among treatment means are said to be highly significant. This result is indicated by placing two asterisks on the observed $F$-value.
2. If the observed $F$ is as large as or larger than the tabular $F$ at the 5% level but smaller than the tabular $F$ at the 1% level, the differences among treatments are said to be significant. This is indicated by placing one asterisk on the observed $F$-value.
3. If the observed $F$ is smaller than the tabular $F$ at the 5% level, the differences among treatments are said to be nonsignificant. This is indicated by placing *ns* on the observed $F$-value.

In the present example, the observed $F$ is larger than the tabular $F$ at the 1% level. Hence, the differences among treatments are said to be highly significant. In other words, chances are less than 1 in 100 that all the observed differences among the seven treatment means could be due to chance.

Note that we can only conclude that there exist some differences between one or more pairs of the treatments but cannot state which specific pair or pairs are causing the difference. Procedures discussed in chapter 6 can be used to obtain this type of information.

STEP 10. Compute the coefficient of variation (cv):

$$\text{cv} = \frac{\sqrt{\text{error MS}}}{\text{grand mean}} \times 100 \tag{3.8}$$
$$= \frac{\sqrt{94{,}773}}{2{,}040} \times 100 = 15.1\%.$$

**Table 3.2 Analysis of variance for rice yield data of table 3.1.**[1]

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$[2] | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Treatments | 6 | 5,587,175 | 931,196 | 9.82** | 2.57 | 3.81 |
| Experimental error | 21 | 1,990,237 | 94,773 | | | |
| Total | 27 | 7,577,412 | | | | |

[1]cv = 15.1%, ** = significant at 1% level.

The cv value, which indicates the degree of precision in a particular experiment, is generally placed underneath the analysis of variance table as shown in table 3.2.

The coefficient of variation is a good index of the reliability of an experiment. It expresses the experimental error as a percentage of the mean, and thus, the higher its value, the lower is the reliability of the experiment.

The coefficients of variation vary greatly with the type of experiment, the crop being tested, and the character being measured. An experienced researcher, however, can make good judgment on the acceptability of a particular cv for a given type of experiment. Our experience with measuring rice yield, for example, indicates that we should aim for a cv of 6 to 8% in varietal tests, 10 to 12% for fertilizer trials, and about 15% for insecticide or herbicide trials. Moreover, the cv for other characters can greatly differ from that of yield. For example, in experiments where the cv for yield is about 10%, that for tiller number can be expected to be about 20% and that for plant height about 3%.

### 3.2.3 Analysis of variance: completely randomized design with an unequal number of replications

Completely randomized design (CRD) is one of the few designs that allows you to use an unequal number of replications and is still easy to compute. This feature is most useful for studies where the experimental material precludes using an equal number of replications for all treatments. Some examples of this are: (a) animal feeding experiments where the number of animals for each breed are not the same, (b) experiments for comparing body length of different species of insect caught in an insect trap, and (c) experiments that may start with an equal number of replications but some experimental units (or plots) will probably be lost or destroyed during the experiment.

When the number of replications per treatment differs, the computational procedures shown in section 3.2.2 are slightly modified. To illustrate the modified procedure, data on grain yield from the performance of post-emergence herbicides in upland rainfed rice trial conducted at IRRI in the 1968 wet season (table 3.3) will be used. The correction factor and the total SS are computed following formulas (3.1) and (3.2).

That is,

**Table 3.3 Grain yield of rice under different types, rates, and times of application of post-emergence herbicides under upland-rainfed condition, IRRI.**

| Treatment | | | Grain yields (kg/ha) | | | | Treatment total | Treatment mean |
|---|---|---|---|---|---|---|---|---|
| Type | Rate[1] (kg a.i./ha) | Time of application[2] (DAS) | | | | | | |
| Propanil/Bromoxynil | 2.0/0.25 | 21 | 3,187 | 4,610 | 3,562 | 3,217 | 14,576 | 3,644 |
| Propanil/2,4-D-Bee | 3.0/1.00 | 28 | 3,390 | 2,875 | 2,775 | | 9,040 | 3,013 |
| Propanil/Bromoxynil | 2.0/0.25 | 14 | 2,797 | 3,001 | 2,505 | 3,490 | 11,793 | 2,948 |
| Propanil/Ioxynil | 2.0/0.50 | 14 | 2,832 | 3,193 | 3,448 | 2,255 | 11,638 | 2,910 |
| Propanil/CHCH | 3.0/1.50 | 21 | 2,233 | 2,743 | 2,727 | | 7,703 | 2,568 |
| Phenyedipham | 1.5 | 14 | 2,952 | 2,272 | 2,470 | | 7,694 | 2,565 |
| Propanil/Bromoxynil | 2.0/0.25 | 28 | 2,858 | 2,895 | 2,458 | 1,723 | 9,934 | 2,484 |
| Propanil/2,4-D-IPE | 3.0/1.00 | 28 | 2,308 | 2,335 | 1,975 | | 6,618 | 2,206 |
| Propanil/Icxynil | 2.0/0.50 | 28 | 2,013 | 1,788 | 2,248 | 2,115 | 8,164 | 2,041 |
| Handweed twice | — | 15 and 35 | 3,202 | 3,060 | 2,240 | 2,690 | 11,192 | 2,798 |
| Control | — | — | 1,192 | 1,652 | 1,075 | 1,030 | 4,949 | 1,237 |

[1]a.i. = active ingredient. [2]DAS = days after seeding.

$$\text{C.F.} = \frac{(\text{grand total})^2}{\text{total number of observations}}$$
$$= \frac{(103{,}301)^2}{40}$$
$$= 266{,}777{,}415, \quad \text{and}$$
$$\text{Total SS} = \sum X^2 - \text{C.F.}$$
$$= [(3187)^2 + (4610)^2 + \ldots + (1030)^2] - 266{,}777{,}415$$
$$= 20{,}209{,}724.$$

Computation of the treatment SS, however, differs slightly from that given by equation (3.3) in that the divisor of the term involving the square of treatment totals varies from one treatment to another. Each divisor is the number of replications corresponding to the treatment concerned. That is,

$$\text{Treatment SS} = \sum_i \frac{T_i^2}{r_i} - \text{C.F.} \tag{3.9}$$
$$= \left[\frac{(14{,}576)^2}{4} + \frac{(9{,}040)^2}{3} + \ldots + \frac{(4{,}949)^2}{4}\right] - 266{,}777{,}415$$
$$= 15{,}090{,}304.$$

The error SS is computed from formula (3.4) as

$$\text{Error SS} = \text{total SS} - \text{treatment SS}$$
$$= 5{,}119{,}420.$$

The mean squares and $F$-values are then calculated using equations (3.5), (3.6), and (3.7). The completed analysis of variance is given in table 3.4. The $F$-test indicates highly significant differences among treatment means.

## 3.3 RANDOMIZED COMPLETE BLOCK DESIGN

The randomized complete block (RCB) design is one of the most widely used experimental designs in agricultural research. It is characterized by blocks of equal size, each of which contains a complete set of all treatments. The chief advantage of this design is that it reduces experimental error (through proper blocking) while retaining much of the flexibility and simplicity of the completely randomized design.

**Table 3.4 Analysis of variance for grain yield data (kg/ha) of Table 3.3.[1]**

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Treatment | 10 | 15,090,304 | 1,509,030 | 8.55** | 2.18 | 3.00 |
| Error | 29 | 5,119,420 | 176,532 | | | |
| Total | 39 | 20,209,724 | | | | |

[1]cv = 16.3%, ** = significant at the 1% level.

### 3.3.1 Blocking

The primary purpose of blocking is to reduce as much as possible heterogeneity among plots within each block. Proper blocking increases the differences among blocks while leaving plots within a block more homogeneous. Knowing the fertility pattern in the experimental area is of great value in achieving this. Some simple rules to follow are:

1. When the fertility pattern of the experimental field is known, orient the blocks so that soil differences between blocks are maximized and those within blocks are minimized. For a field with a unidirectional fertility gradient, long and narrow blocks should be used. Blocks should be oriented such that their length is perpendicular to the direction of the fertility gradient. For example, for a field with a gradient along the length of the field, blocking should be made across the width of the field, cutting across the gradient.
2. When a fertility gradient occurs in two directions, with one direction perpendicular to the other, or nearly so, a Latin square design, described in section 3.4, or a covariance technique, discussed in chapter 8 may be used. If, however, a randomized complete block design must be used, more or less square blocks are recommended.
3. When the fertility pattern is not known or when fertile areas occur in unpredictable spots, blocks should be as square as possible.

Except for the different treatments assigned, plots within each block should be managed as uniformly as possible. Data collection and all cultural and management practices, aside from the treatments being studied, should be made at the same time and as uniformly as possible on all plots in each block. For example, if application of insecticides or harvesting of a field experiment must be done over several days, all plots in a block should be applied or harvested in the same day. Also, if more than one research worker makes observations on the experimental area and if there is any likelihood that observations made on the same plot would differ with the individual, then different persons should be assigned to different blocks; i.e., one person should make the observations for all plots in a block. See chapter 14 for a further discussion on uniform management of experiments.

### 3.3.2 Randomization and layout

Divide the experimental area into as many blocks as the number of replications. Each block is further subdivided into plots that correspond to the number of treatments. Since the randomized complete block design specifies that all treatments must appear once in each replication, randomization is done separately for each block. Randomization procedures similar to those described in section 3.2.1 can be used to assign treatments to plots in each block.

Let us take an experiment where we have six treatments ($A$, $B$, $C$, $D$, $E$, and $F$) to be replicated four times. If the table of random numbers is used, we first select at random six three-digit numbers. For example, starting at the intersection of the sixteenth row and twelfth column of the table in appendix 1, read downward vertically to get six three-digit numbers:

| | *Sequence* |
|---|---|
| 918 | 1 |
| 772 | 2 |
| 243 | 3 |
| 494 | 4 |
| 704 | 5 |
| 549 | 6 |

Second, rank the selected numbers from the smallest to the largest:

| | *Sequence* | *Rank* |
|---|---|---|
| 918 | 1 | 6 |
| 772 | 2 | 5 |
| 243 | 3 | 1 |
| 494 | 4 | 2 |
| 704 | 5 | 4 |
| 549 | 6 | 3 |

Third, use the sequence in which the random numbers occurred as the treatment number, and use the rank as the plot number in the block. Thus, assign treatment *A* to plot 6, treatment *B* to plot 5, treatment *C* to plot 1, treatment *D* to plot 2, treatment *E* to plot 4, and treatment *F* to plot 3.

The layout of the first block is shown in figure 3.2.

Fourth, repeat the first three steps for block II, then for block III, and finally for block IV.

A sample outcome of the layout is given in figure 3.3 (a). A hypothetical layout of completely randomized design (figure 3.3 (b)) is also given to em-

| PLOT no. | BLOCK I |
|---|---|
| 1 | treatment C |
| 2 | treatment D |
| 3 | treatment F |
| 4 | treatment E |
| 5 | treatment B |
| 6 | treatment A |

**Fig. 3.2** Sample layout of the first block of a randomized complete block design with six treatments.

| | (a) RCB layout | (b) CRD hypothetical layout |
|---|---|---|
| Block I | C | B |
| | D | E |
| | F | C |
| | E | F |
| | B | A |
| | A | B |
| Block II | A | C |
| | E | A |
| | F | D |
| | C | C |
| | D | A |
| | B | C |
| Block III | F | E |
| | D | B |
| | C | F |
| | A | E |
| | B | D |
| | E | E |
| Block IV | E | A |
| | C | F |
| | D | D |
| | A | F |
| | F | B |
| | B | D |

**Fig. 3.3** Layouts of randomized complete block (RCB) and completely randomized design (CRD) with six treatments and four replications.

phasize the differences in layouts. Note that in a randomized complete block design, randomization is restricted so that all treatments must appear in the same block, unlike the completely randomized design, which has no restriction whatsoever.

**Table 3.5 Grain yield of rice variety IR8 planted with different rates of seeding, IRRI, 1967 wet season.**

| Treatment (kg seeds/ha) | Grain yield (kg/ha) | | | | Treatment total ($T$) | Treatment mean |
|---|---|---|---|---|---|---|
| | Block I | Block II | Block III | Block IV | | |
| 25 | 5,113 | 5,398 | 5,307 | 4,678 | 20,496 | 5,124 |
| 50 | 5,346 | 5,952 | 4,719 | 4,264 | 20,281 | 5,070 |
| 75 | 5,272 | 5,713 | 5,483 | 4,749 | 21,217 | 5,304 |
| 100 | 5,164 | 4,831 | 4,986 | 4,410 | 19,391 | 4,848 |
| 125 | 4,804 | 4,848 | 4,432 | 4,748 | 18,832 | 4,708 |
| 150 | 5,254 | 4,542 | 4,919 | 4,098 | 18,813 | 4,703 |
| Block total ($B$) | 30,953 | 31,284 | 29,846 | 26,947 | | |
| Grand total | | | | | 119,030 | |
| Grand mean | | | | | | 4,960 |

### 3.3.3 Analysis of variance

To illustrate the analysis of variance for a randomized complete block design, data on grain yield from an experiment comparing six rates of seeding in upland rice will be used (table 3.5). Four replications were used in this experiment.

STEP 1. Group the data by treatments and blocks and calculate treatment totals, block totals, and treatment means as in table 3.5.

STEP 2. Outline the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Block | | | | | | |
| Treatment | | | | | | |
| Error | | | | | | |
| Total | | | | | | |

Note that in addition to the two sources of variation for completely randomized design (section 3.2.2), a third component, block, is also separable for this design.

STEP 3. Determine the degrees of freedom (d.f.) for each source of variation as follows:

$$\begin{aligned}
\text{Total d.f.} &= \text{total number of observations} - 1 \\
&= 24 - 1 = 23, \\
\text{Block d.f.} &= \text{total number of blocks} - 1 \\
&= 4 - 1 = 3, \\
\text{Treatment d.f.} &= \text{total number of treatments} - 1 \\
&= 6 - 1 = 5, \text{ and} \\
\text{Error d.f.} &= \text{total d.f.} - \text{block d.f.} - \text{treatment d.f.} \\
&= 23 - 3 - 5 = 15, \text{ or} \\
&= (\text{block d.f.})(\text{treatment d.f.}) = (3)(5) = 15.
\end{aligned}$$

STEP 4. Calculate the various sums of squares (SS):

$$\begin{aligned}
\text{C.F.} &= \frac{(\text{grand total})^2}{\text{total number of observations}} \\
&= \frac{(119{,}030)^2}{24} \\
&= 590{,}339{,}204, \\
\text{Total SS} &= \sum X^2 - \text{C.F.} \\
&= [(5{,}113)^2 + (5{,}346)^2 + \ldots + (4{,}098)^2] - \text{C.F.} \\
&= 595{,}140{,}272 - 590{,}339{,}204 \\
&= 4{,}801{,}068,
\end{aligned}$$

$$\text{Block SS} = \sum \frac{(\text{block total})^2}{t} - \text{C.F.} \tag{3.10}$$

$$= \frac{(30{,}953)^2 + (31{,}284)^2 + (29{,}846)^2 + (26{,}947)^2}{6} - \text{C.F.}$$

$$= 592{,}283{,}565 - 590{,}339{,}204$$
$$= 1{,}944{,}361,$$

$$\text{Treatment SS} = \sum \frac{(\text{treatment total})^2}{r} - \text{C.F.}$$

$$= \frac{(20{,}496)^2 + \ldots + (18{,}813)^2}{4} - \text{C.F.}$$

$$= 591{,}537{,}535 - 590{,}339{,}204$$
$$= 1{,}198{,}331, \quad \text{and}$$

$$\text{Error SS} = \text{total SS} - \text{block SS} - \text{treatment SS}$$
$$= 4{,}801{,}068 - 1{,}944{,}361 - 1{,}198{,}331$$
$$= 1{,}658{,}376.$$

STEP 5. Divide each sum of square (SS) by its corresponding degrees of freedom to obtain the mean square (MS).

$$\text{Block MS} = \frac{\text{block SS}}{r - 1}$$

$$= \frac{1{,}944{,}361}{3}$$

$$= 648{,}120,$$

$$\text{Treatment MS} = \frac{\text{treatment SS}}{t - 1}$$

$$= \frac{1{,}198{,}331}{5}$$

$$= 239{,}666, \quad \text{and}$$

$$\text{Error MS} = \frac{\text{error SS}}{(r-1)(t-1)} = \frac{1{,}658{,}376}{15} = 110{,}558.$$

STEP 6. Compute the observed $F$-value for testing the treatment effects as in formula (3.7):

$$F = \frac{\text{treatment MS}}{\text{error MS}}$$

$$= \frac{239{,}666}{110{,}558} = 2.17.$$

STEP 7. Obtain the tabular $F$-value for 5 and 15 degrees of freedom from appendix 5. The tabular $F$-value for 5% and 1% levels of significance are 2.90 and 4.56, respectively. Since the observed $F$-value is smaller than the tabular

**Table 3.6 Analysis of variance on grain yield data of table 3.5.**[1]

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Block | 3 | 1,944,361 | 648,120 | 5.86** | 3.29 | 5.42 |
| Treatment | 5 | 1,198,331 | 239,666 | 2.17ns | 2.90 | 4.56 |
| Error | 15 | 1,658,376 | 110,558 | | | |
| Total | 23 | 4,801,068 | | | | |

[1]cv = 6.7%, ** = significant at 1% level, ns = not significant.

$F$-value at 5% level, we conclude that the experiment failed to show any significant difference among the six rates of seeding.

STEP 8. Compute the coefficient of variation:

$$\text{cv} = \frac{\sqrt{\text{error MS}}}{\text{grand mean}} \times 100 = \frac{\sqrt{110{,}558}}{4{,}960} = 6.7\%. \tag{3.11}$$

STEP 9. Enter all values computed in steps 3 to 8 in the analysis of variance table (table 3.6).

### 3.3.4 Block efficiency

The differences among blocks may also be tested, if desired, by dividing the block MS by the error MS to obtain the observed $F$-value. In this instance, we have $F = 648{,}120/110{,}558 = 5.86$, which is highly significant. This indicates that using blocks was effective in reducing the size of the experimental error; i.e., the precision of the experiment was increased by using a randomized complete block design instead of the completely randomized design. To evaluate the gain in precision, we compute the relative efficiency (R.E.) of RCB relative to CRD as

$$\text{R.E.} = \frac{(r-1)E_b + [(t-1) + (r-1)(t-1)]E_e}{(rt-1)E_e}, \tag{3.12}$$

where $E_b$ and $E_e$ are the block and error mean squares. If the error degrees of freedom is under 20, the relative efficiency should be multiplied by the factor

$$k = \frac{[(r-1)(t-1)+1][t(r-1)+3]}{[t(r-1)+1][(r-1)(t-1)+3]}. \tag{3.13}$$

Applying equation (3.12) to the data of table 3.6, we obtain

$$\text{R.E.} = \frac{3(648{,}120) + (5+15)(110{,}558)}{(23)(11{,}056)} = 1.63.$$

Since the error d.f. is only 15, the adjustment factor should be computed using equation (3.13) as

$$k = \frac{(15+1)(18+3)}{(18+1)(15+3)} = 0.982.$$

The adjusted relative efficiency then is

$$k(\text{R.E.}) = (1.63)(0.982)$$
$$= 1.60.$$

Expressing the relative efficiency as a percentage, we get 160%, which indicates that blocking increased the precision of the experiment by 60%.

A word of caution is needed here: Although it has been generally stated that great differences among blocks indicate the success of blocking, the problem of heterogeneity of variance (see chapter 10), as well as the presence of interactions (see chapter 4) between treatment and block, may be encountered when block differences are extremely large. Researchers should not, therefore, choose blocks so different that the response to the various treatments may vary from block to block.

## 3.4 LATIN SQUARE DESIGN

The most important feature of the Latin square design is its capacity to block an area along two directions instead of only one as with the randomized complete block. The two-directional blocking, commonly referred to as rows and columns, is accomplished by arranging the treatments so that every treatment occurs only once in each row and once in each column. This allows us to measure and remove from the experimental error the differences among rows as well as among columns. Thus, the Latin square design is especially adapted for use in an experimental site where there are two fertility gradients running perpendicular to each other.

The stratification into rows and columns, while useful in removing variability along two directions, also becomes a major restriction, requiring that the number of replications must equal the number of treatments. This restriction is very significant because for a large number of treatments, the design becomes impractical. On the other hand, with fewer than four treatments, the number of degrees of freedom associated with the experimental error becomes too small. Thus, the Latin square design is generally used only for experiments with four to eight treatments. Because of such inflexibility and limitation, the Latin square design has not been widely used in agricultural experimentation despite its great potential for controlling experimental error.

### 3.4.1 Randomization and layout

The layout for a Latin square design is set up by selecting a sample Latin square plan from appendix 11 followed by the randomization among rows and among columns.

To illustrate, let us consider an experiment involving five treatments, *A*, *B*, *C*, *D*, and *E*. The randomization procedure is as follows:

STEP 1. Take the 5 × 5 Latin square plan shown in appendix 11:

| | | | | |
|---|---|---|---|---|
| *A* | *B* | *C* | *D* | *E* |
| *B* | *A* | *E* | *C* | *D* |
| *C* | *D* | *A* | *E* | *B* |
| *D* | *E* | *B* | *A* | *C* |
| *E* | *C* | *D* | *B* | *A* |

STEP 2. Randomize the row arrangement as follows:
(a) Select at random five three-digit numbers from appendix 1: e.g., 628, 846, 475, 902, and 452.
(b) Rank these numbers from lowest to highest:

| *Random number* | *Selection sequence* | *Rank* |
|---|---|---|
| 628 | 1 | 3 |
| 846 | 2 | 4 |
| 475 | 3 | 2 |
| 902 | 4 | 5 |
| 452 | 5 | 1 |

(c) Use these ranks as the row number of the selected plan and the sequence as the row number of the rearranged plan. That is, the third row of the selected plan becomes the first row, the fourth row becomes the second, and so on. Thus, the new plan after row rearrangement is:

| | | | | |
|---|---|---|---|---|
| *C* | *D* | *A* | *E* | *B* |
| *D* | *E* | *B* | *A* | *C* |
| *B* | *A* | *E* | *C* | *D* |
| *E* | *C* | *D* | *B* | *A* |
| *A* | *B* | *C* | *D* | *E* |

STEP 3. Randomize the columns using the same procedure as in step 2. For example, five random numbers were again chosen from appendix 1 and ranked as follows:

| *Random number* | *Selection sequence* | *Rank* |
|---|---|---|
| 792 | 1 | 4 |
| 032 | 2 | 1 |
| 947 | 3 | 5 |
| 293 | 4 | 3 |
| 196 | 5 | 2 |

The rank now represents the column number of the plan of step 2, and the sequence that the numbers occurred in represents the column number of the newly rearranged plan. That is, the fourth column of the plan of step 2 be-

comes the first column in the new plan, the first column becomes the second, and so on. Thus, the resulting plan is:

| | | | | |
|---|---|---|---|---|
| *E* | *C* | *B* | *A* | *D* |
| *A* | *D* | *C* | *B* | *E* |
| *C* | *B* | *D* | *E* | *A* |
| *B* | *E* | *A* | *D* | *C* |
| *D* | *A* | *E* | *C* | *B* |

This plan becomes the final layout of the experiment.

### 3.4.2 Analysis of variance

Table 3.7 shows data on grain yield of three promising corn hybrids (*A*, *B*, and *D*) and a check variety (*C*), from an advanced yield test of the Department of Agronomy, UPLB. The experiment was conducted in a Latin square design with four replications.

The steps in constructing an analysis of variance are:

STEP 1. Arrange the data by row and column as in table 3.7. For each observation, the particular treatment involved must be specified.

STEP 2. Compute treatment totals and treatment means as follows:

| | *Yield (cavans/ha)* | |
|---|---|---|
| *Treatment* | *Total* | *Mean* |
| A | 117.1 | 29.3 |
| B | 117.7 | 29.4 |
| C | 85.4 | 21.4 |
| D | 107.1 | 26.8 |

STEP 3. Outline the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed *F* | Tabular *F* 5% | Tabular *F* 1% |
|---|---|---|---|---|---|---|
| Row | | | | | | |
| Column | | | | | | |
| Treatment | | | | | | |
| Error | | | | | | |
| Total | | | | | | |

Note that there are four sources of variation for the Latin square design compared with three for the randomized complete block (section 3.3.3) and with two for the completely randomized design (section 3.2.2).

STEP 4. Determine the degrees of freedom (d.f.) for each source of variation as follows:

**Table 3.7 Grain yield of three promising corn hybrids and of a check variety.**

| Row | Grain yield (cavans/ha) | | | | Row total |
|---|---|---|---|---|---|
| | Col. 1 | Col. 2 | Col. 3 | Col. 4 | |
| 1 | 32.8 (*B*) | 24.2 (*D*) | 28.5 (*C*) | 26.9 (*A*) | 112.4 |
| 2 | 29.5 (*C*) | 23.7 (*A*) | 28.0 (*D*) | 25.8 (*B*) | 107.0 |
| 3 | 33.4 (*A*) | 14.2 (*C*) | 33.3 (*B*) | 23.6 (*D*) | 104.5 |
| 4 | 31.3 (*D*) | 25.8 (*B*) | 33.1 (*A*) | 13.2 (*C*) | 103.4 |
| Column total | 127.0 | 87.9 | 122.9 | 89.5 | |
| Grand total | | | | | 427.3 |

$$\begin{aligned}
\text{Total d.f.} &= \text{total number of observations} - 1 \\
&= 16 - 1 = 15, \\
\text{Row d.f.} &= \text{column d.f.} = \text{treatment d.f.} \\
&= \text{total number of treatments} - 1, \\
\text{Error d.f.} &= \text{total d.f.} - \text{row d.f.} - \text{column d.f.} - \text{treatment d.f.} \\
&= 15 - 3 - 3 - 3 = 6, \text{ or} \\
&= (\text{total number of treatments} - 1)(\text{total number of treatments} \\
&\quad - 2) \\
&= (4 - 1)(4 - 2) = 6.
\end{aligned}$$

STEP 5. Compute the various sums of squares (SS):

$$\begin{aligned}
\text{C.F.} &= \frac{(\text{grand total})^2}{\text{total number of observations}} \\
&= \frac{(427.3)^2}{16} \\
&= 11{,}411.581.
\end{aligned}$$

$$\begin{aligned}
\text{Total SS} &= \sum X^2 - \text{C.F.} \\
&= [(32.8)^2 + (29.5)^2 + \ldots + (13.2)^2] - \text{C.F.} \\
&= 565.569,
\end{aligned}$$

$$\begin{aligned}
\text{Row SS} &= \sum \frac{(\text{row total})^2}{t} - \text{C.F.} \\
&= \frac{(112.4)^2 + (107.0)^2 + (104.5)^2 + (103.4)^2}{4} - \text{C.F.} \\
&= 12.062,
\end{aligned}$$

$$\begin{aligned}
\text{Column SS} &= \sum \frac{(\text{column total})^2}{t} - \text{C.F.} \\
&= \frac{(127.0)^2 + (87.9)^2 + (122.9)^2 + (89.5)^2}{4} - \text{C.F.} \\
&= 330.937,
\end{aligned}$$

$$\text{Treatment SS} = \sum \frac{(\text{treatment total})^2}{t} - \text{C.F.}$$

$$= \frac{(117.1)^2 + (117.7)^2 + (85.4)^2 + (107.1)^2}{4} - \text{C.F.}$$

$$= 170.737, \quad \text{and}$$

$$\begin{aligned}\text{Error SS} &= \text{total SS} - \text{row SS} - \text{column SS} - \text{treatment SS}\\ &= 565.569 - 12.062 - 330.937 - 170.737\\ &= 51.833.\end{aligned}$$

STEP 6. Compute the mean square for each source of variation by dividing the sum of squares by its corresponding degrees of freedom.

STEP 7. Compute the observed $F$-values by dividing each mean square by the error mean square as follows:

$$F \text{ for row} = \frac{4.021}{8.639} < 1,$$

$$F \text{ for column} = \frac{110.312}{8.639} = 12.77,$$

and

$$F \text{ for treatment} = \frac{59.912}{8.639} = 6.59.$$

STEP 8. Compare each of the observed $F$-values with the appropriate tabular values and declare the test for the particular effect significant or not significant following the rules given in section 3.2.2.

STEP 9. Enter all values so far computed in the analysis of variance table. The final results are shown in table 3.8. Significant differences in the mean yields of the four corn varieties tested were obtained. To find out whether every one of the three hybrids gave significantly higher yield than the check, or whether there are any significant differences among the three hybrids, etc., test procedures discussed in chapter 6 may be used.

### 3.4.3 Row and column blocking

The test for column differences in table 3.8 showed a highly significant result while that for row differences gave a nonsignificant result. This indicates that the grouping of experimental plots into rows was not as successful in reducing

**Table 3.8** Analysis of variance on grain yield data of table 3.7.[1]

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Row | 3 | 12.062 | 4.021 | $<1^{ns}$ | 4.76 | 9.78 |
| Column | 3 | 330.937 | 110.312 | $12.77^{**}$ | 4.76 | 9.78 |
| Treatment | 3 | 170.737 | 59.912 | $6.59^{*}$ | 4.76 | 9.78 |
| Error | 6 | 51.833 | 8.639 | | | |
| Total | 15 | 565.569 | | | | |

[1] cv = 11.0%, ** = significant at 1% level, * = significant at 5% level, ns = not significant.

the experimental error as was the grouping by column. This would suggest that a randomized complete block design with columns as blocks would have done similarly well.

## 3.5 LATTICE DESIGNS

Theoretically, the complete block designs discussed in the previous sections can be used for any number of treatments. But in reality they become inefficient when the number of treatments becomes large. The Latin square design becomes impractical because the number of replications must equal the number of treatments. As a result, the size of the experiment increases very fast as the number of treatments increases. For example, with 5 treatments, 25 experimental units are required; with 8 treatments, 64 units are required; and with 10 treatments, 100 units are required. The required number of experimental units quadruples for every doubling of the number of treatments. Thus, as mentioned earlier, Latin square designs are not commonly used for experiments with more than eight treatments.

The other two complete block designs become inefficient as the number of treatments increases because either the number of experimental units or block size must increase substantially. For a completely randomized design, the number of experimental units must increase and this increase results in more heterogeneity among experimental units and, hence, greater experimental error. For a randomized complete block design, block size must increase, and the effectiveness of blocking in controlling experimental error decreases as the block size increases.

Controlling experimental error also becomes more difficult when there are large differences among experimental units or a significant soil heterogeneity in the experimental site. Thus, none of the complete block designs is recommended for use when the number of treatments is large, especially if substantial differences among experimental units are expected. There is, however, an attractive alternative: the family of incomplete block designs. In these designs the number of blocks no longer equals the number of replications. Instead, the replication is further subdivided into smaller blocks in which only a fraction of the total number of treatments is alloted. The name of this family of designs, "incomplete block," describes this important feature.

Another important feature of the incomplete block designs is the flexibility of such in the choice of the number of replications. By choosing an appropriate design, the restrictions on the number of replications becomes so insignificant that practically any number of replications can be used for any number of treatments.

There are too many incomplete block designs to cover in this book, so we have limited our discussion to the lattice designs since this subgroup of designs is more commonly used in agricultural experiments. In opting to include only a few designs we hope to discuss these few as clearly and as completely as possible so that you can then easily understand other incomplete block designs

found in other references. Furthermore, we have deliberately chosen two types of lattice designs that allow us to bring out the important features the researcher needs in choosing an appropriate incomplete block design for his particular experiment.

A requirement for using lattice designs is that the number of treatments must be a perfect square; e.g., $5^2$ or 25. $6^2$ or 36, $7^2$ or 49, $8^2$ or 64, $9^2$ or 81, and $10^2$ or 100. This requirement may seem stringent, but it is not too difficult to satisfy in practice, since, as the number of treatments to be tested becomes large, adding a few treatments or eliminating less important treatments is usually easy to accomplish. For example, if a plant breeder wishes to test the performance of 80 varieties, all he needs to do to use a lattice design is to add one more variety. Or, if he has 82 or 83 varieties to start with, the plant breeder can usu..lly eliminate one or two without much difficulty. However, where adjusting the number of treatments is not easily done, other lattices, such as the rectangular or the cubic, or other incomplete block designs should be considered (see Cochran and Cox, 1957).

We have chosen to discuss the balanced lattice and the partially balanced lattice designs in this section because they illustrate the important trade-offs involved in choosing one incomplete block design over another. An important trade-off between the balanced and partially balanced lattice designs is (a) the balanced lattice design has a more stringent requirement on the number of replications than does the partially balanced lattice, but (b) the precision among treatment comparisons is the same for all pairs of treatments in the balanced lattice but not in the partially balanced lattice design.

For both types of lattice designs, the number of plots in each block is equal to the square root of the total number of treatments. For example, block size would be 8 with 64 treatments, 10 with 100 treatments, etc. For a balanced lattice, the number of replications required is one more than the square root of the total number of treatments; e.g. with 64 treatments, 9 replications are required. For a partially balanced lattice, however, any number of replications can be used. And with 2 replications, the design is called a simple lattice; with 3 replications, a triple lattice; with 4 replications, a quadruple lattice; etc.

Table 3.9 shows a balanced lattice design involving nine treatments arranged in three incomplete blocks each with three treatments. Thus, the number of replications required is four. In this design, any particular pair of treatments appears together in any block once and only once in the experiment. This is why there is an equal degree of precision in the comparisons of all pairs of treatments in the balanced lattice design.

On the other hand, symmetry in the occurrence of the various pairs of treatments together in the same block is not possible in the partially balanced lattice design. Since the number of replications is usually fewer than that required for a balanced lattice, some treatments never appear together in the same block, so the precision of comparison among treatments is not constant for all treatments. There is a higher degree of precision in treatments appearing in the same block relative to those that did not. For example, the plan involving

**Table 3.9 Balanced lattice design for nine treatments in blocks of three units with four replications.**

| Incomplete block no. | Treatment no. | | | |
|---|---|---|---|---|
| | Rep. I | Rep. II | Rep. III | Rep. IV |
| 1 | 1 2 3 | 1 4 7 | 1 5 9 | 1 6 8 |
| 2 | 4 5 6 | 2 5 8 | 2 6 7 | 2 4 9 |
| 3 | 7 8 9 | 3 6 9 | 3 4 8 | 3 5 7 |

only replications I and II of table 3.9 constitutes a partially balanced lattice design for nine treatments with two replications, or a simple lattice design. Note that in this plan, not all pairs of treatments occur together in any incomplete block. For example, treatment number 1 appears with treatment numbers 2, 3, 4, and 7 once in an incomplete block but not with treatment numbers 5, 6, 8, or 9 in any block. Thus, the four comparisons between each of the treatments 2, 3, 4, and 7 with treatment 1 would have higher degree of precision than those between treatment 1 and each of the treatments 5, 6, 8, and 9.

If a triple lattice is used for testing the nine treatments, the plan would be equivalent to taking the first three replications (Replication I, Replication II, and Replication III) from table 3.9. Clearly, in the triple lattice, as in the simple lattice, there are some pairs of treatments that do not occur together in any block. For example, treatment 1 does not occur together in an incomplete block with treatments 6 and 8 if the triple lattice, as just described, is used. Note that there are more pairs occurring together in the same incomplete block in a triple lattice than in a simple lattice; hence, there is a higher degree of precision in the first than in the second design.

Since all comparisons of pairs of treatments have equal precision in the balanced lattice design, it follows that there is only one test of significance that needs to be computed. In contrast, since there is more than one test of significance for comparisons of pairs of treatments in a partially balanced lattice, statistical analysis is also more complicated.

All the major characteristics of the balanced and partially balanced lattice designs so far discussed are summarized in table 3.10.

### 3.5.1 Randomization and layout

The randomization and layout for lattice designs follow these steps:

STEP 1. Divide the experimental area into $r$ replications each containing $t$ experimental plots, where $t$ is the total number of treatments to be tested. Then, divide each replication into $k = \sqrt{t}$ incomplete blocks each containing $k$ experimental plots. There will be a total of $rt = rk^2$ experimental plots for the whole experiment. The blocking technique discussed in section 3.3.1 should be followed in choosing the shape and orientation of the plots, of the incomplete blocks, and of the replications. Note that the criterion established to ensure proper blocking in an incomplete block design allows maximum homogeneity among experimental units within the same incomplete block. Maximum

**Table 3.10 Basic features and requirements of balanced and partially balanced lattice designs.**

| Feature | Balanced lattice | Partially balanced lattice |
|---|---|---|
| Number of treatments | a square number, $k^2$ | a square number, $k^2$ |
| Number of replications | $k + 1$ | any number |
| Block size | $k$ | $k$ |
| Precision of treatment comparison | Same for all pairs of treatments | Higher for treatment pairs appearing in same block relative to pairs that do not. |

homogeneity is achieved by maximizing variations among replications and that among incomplete blocks within a replication. Thus, in applying the blocking technique described in section 3.3.1, the $rk$ incomplete blocks can be treated as if they were $rk$ blocks of a randomized complete block design.

STEP 2. Select a basic plan from appendix 12 based on the number of treatments to be tested. Note that the basic plans given in appendix 12 are for balanced lattice designs. The plans for a partially balanced lattice design can be derived directly from those of the balanced lattice having the same number of treatments by including in the plan only the required number of replications. For example, the first two replications of a balanced lattice plan constitutes a simple lattice; the first three replications, for a triple lattice; and so on. When the number of replications for a partially balanced lattice exceeds three and is an even number, another alternative for deriving the plan is available. Instead of choosing a basic plan corresponding to the desired number of replications from appendix 12, a basic plan corresponding to a number of replications that is only a fraction of the desired number is chosen and repeated as many times as required until it reaches the desired number of replications. For example, for a quadruple lattice (i.e., a partially balanced lattice with four replications), the plan can be obtained by duplicating the simple lattice. In general, the first procedure of using the basic plan without repetition is slightly preferred as compared with the second, which involves some repetitions, because the resulting design comes closer to the symmetry achieved in a balanced lattice.

STEP 3. Randomization is done in two stages: one for the assignment of incomplete blocks within each replication, and another for the assignment of treatments to the plots within each incomplete block. For methods of randomization at either of the two stages, follow the procedures described in section 3.2.1.

For illustration, consider a field experiment using a $3 \times 3$ balanced lattice design to test nine treatments. The steps involved in the randomization and layout are:

First, divide the experimental area into four replications (each containing

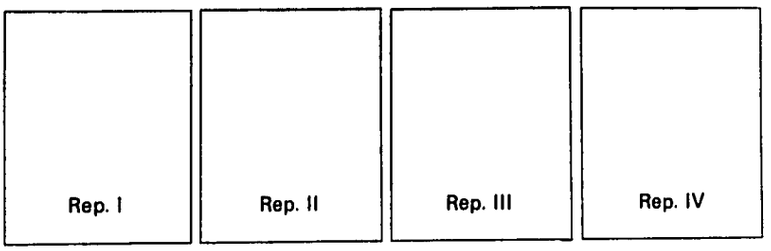


**Fig. 3.4** Division of the experimental area into four replications as the first step in laying out a 3 × 3 balanced lattice design.

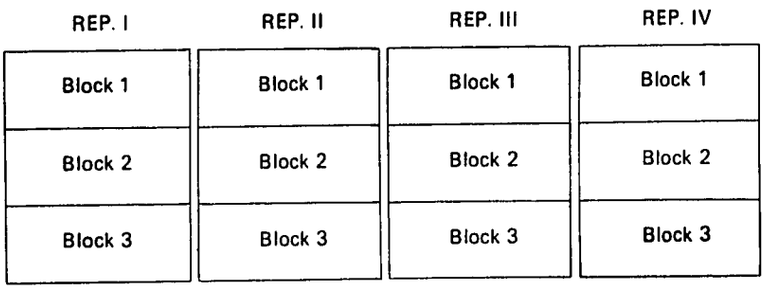


**Fig. 3.5** Division of each of the four replications into three incomplete blocks as a second step in laying out a 3 × 3 balanced lattice design.

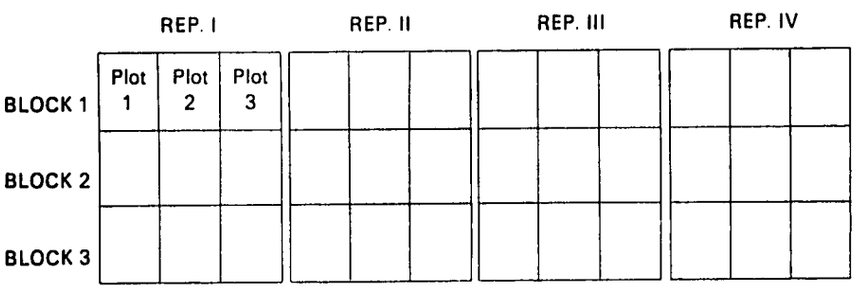


**Fig. 3.6** Division of each of the three incomplete blocks in each replication into three plots as a third step in laying out a 3 × 3 balanced lattice design.

nine experimental plots), as in figure 3.4.

Next, divide each replication into three incomplete blocks (each containing three experimental plots), as shown in figure 3.5.

And finally, divide each of the 12 incomplete blocks into three experimental plots, as in figure 3.6.

The second step is to select a basic plan. We will use the plan given in table 3.9. (For other plans, see appendix 12.)

The third step involves randomization. First, randomize the replications. Using any of the randomization schemes described in section 3.2.1, assign at random the replications appearing in the basic plan to the replications in the field layout. For example, the result of the randomization may be as follows:

| *Replication number in basic plan* | *Replication number in field layout* |
|---|---|
| I | III |
| II | II |
| III | IV |
| IV | I |

Next, randomize the incomplete blocks in each replication. For each of the four replications independently, assign at random the three incomplete blocks of the basic plan to the incomplete blocks in the field layout. The result may be as follows:

| Incomplete block no. in basic plan | Incomplete block no. in field layout | | | |
|---|---|---|---|---|
| | Rep. III | Rep. II | Rep. IV | Rep. I |
| 1 | 3 | 2 | 2 | 1 |
| 2 | 2 | 1 | 3 | 3 |
| 3 | 1 | 3 | 1 | 2 |

That is, incomplete block number 1 of replication I in the basic plan (i.e., replication III in the field layout) would be assigned to incomplete block number 3 of the field layout; block number 2 of the same replication in the basic plan to block number 2 of the field layout; and block number 3 in the basic plan to block number 1 of the field layout. The same implication applied to other replications. The outcome is as follows:

| Rep. III | | | | Rep. II | | | | Rep. IV | | | | Rep. I | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Incomplete block no. | Treatment no. | | | Incomplete block no. | Treatment no. | | | Incomplete block no. | Treatment no. | | | Incomplete block no. | Treatment no. | | |
| 3 | 7 | 8 | 9 | 2 | 2 | 5 | 8 | 2 | 3 | 4 | 8 | 1 | 1 | 6 | 8 |
| 2 | 4 | 5 | 6 | 1 | 1 | 4 | 7 | 3 | 1 | 5 | 9 | 3 | 3 | 5 | 7 |
| 1 | 1 | 2 | 3 | 3 | 3 | 6 | 9 | 1 | 2 | 6 | 7 | 2 | 2 | 4 | 9 |

Finally, randomize the treatments within each incomplete block. For each of the 12 incomplete blocks independently, assign at random the 3 treatments to the 3 plots in each block. The result may be as follows:

| Rep. III | | | | Rep. II | | | | Rep. IV | | | | Rep. I | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Incomplete block no. | Treatment no. | | | Incomplete block no. | Treatment no. | | | Incomplete block no. | Treatment no. | | | Incomplete block no. | Treatment no. | | |
| 3 | 7 | 9 | 8 | 2 | 2 | 8 | 5 | 2 | 8 | 3 | 4 | 1 | 1 | 8 | 6 |
| 2 | 4 | 5 | 6 | 1 | 7 | 4 | 1 | 3 | 5 | 1 | 9 | 3 | 5 | 7 | 3 |
| 1 | 3 | 2 | 1 | 3 | 9 | 3 | 6 | 1 | 6 | 7 | 2 | 2 | 4 | 2 | 9 |

That is, the field layout based on this plan will be as in figure 3.7.

### 3.5.2 Balanced lattices

The analysis of a balanced lattice design will be illustrated using data from a fertilizer trial on rice invoiving 16 treatments. The experiment was conducted in a 4 × 4 balanced lattice with 5 replications. The experimental plan and corresponding data on tiller number are presented in table 3.11. The plan has been rearranged so that it coincides with the basic plan of appendix 12. This is done to help you understand the analysis and is not necessary in the actual analysis. In table 3.11, the values in parentheses placed above the data are the corresponding treatment numbers.

The steps in the analysis are as follows:

STEP 1. Calculate the block totals and replication totals, as shown in table 3.11.

STEP 2. Calculate the treatment totals ($T$) and the grand total ($G$) as given in column 2 of table 3.12.

STEP 3. For each treatment, calculate $B_t$ as the sum of block totals over all blocks in which the particular treatment appears. For example, $B_t$ for treatment number 5 is computed as the sum of block totals (table 3.11) of block number 2, block number 6, block number 9, block number 15, and block number 20. That is,

$$B_t(5) = 616 + 654 + 639 + 675 + 827 = 3{,}441.$$

The results for the 16 values of $B_t$'s are shown in column 3 of table 3.12. To doublecheck the computation make sure that the sum overall $B_t$'s equals

| REP. I | | | REP. II | | | REP. III | | | REP. IV | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 8 | 6 | 7 | 4 | 1 | 3 | 2 | 1 | 6 | 7 | 2 |
| 4 | 2 | 9 | 2 | 8 | 5 | 4 | 5 | 6 | 8 | 3 | 4 |
| 5 | 7 | 3 | 9 | 3 | 6 | 7 | 9 | 8 | 5 | 1 | 9 |

**Fig. 3.7** A sample field layout of a 3 × 3 balanced lattice design showing positions of the nine treatments in each of the four replications.

**Table 3.11 Tiller number per square meter from 16 fertilizer treatments tested in a 4×4 balanced lattice design.[1]**

Rep. I

| Block | | | | | Totals |
|---|---|---|---|---|---|
| | (1) | (2) | (3) | (4) | |
| 1 | 147 | 152 | 167 | 150 | 616 |
| | (5) | (6) | (7) | (8) | |
| 2 | 127 | 155 | 162 | 172 | 616 |
| | (9) | (10) | (11) | (12) | |
| 3 | 147 | 100 | 192 | 177 | 616 |
| | (13) | (14) | (15) | (16) | |
| 4 | 155 | 195 | 192 | 205 | 747 |
| | | | | | 2595 |

Rep. II

| Block | | | | | Totals |
|---|---|---|---|---|---|
| | (1) | (6) | (11) | (16) | |
| 5 | 155 | 162 | 177 | 152 | 646 |
| | (5) | (2) | (15) | (12) | |
| 6 | 182 | 130 | 177 | 165 | 654 |
| | (9) | (14) | (3) | (8) | |
| 7 | 137 | 185 | 152 | 152 | 626 |
| | (13) | (10) | (7) | (4) | |
| 8 | 185 | 122 | 182 | 192 | 681 |
| | | | | | 2607 |

Rep. III

| Block | | | | | Totals |
|---|---|---|---|---|---|
| | (1) | (9) | (5) | (13) | |
| 9 | 140 | 182 | 165 | 152 | 639 |
| | (10) | (2) | (14) | (6) | |
| 10 | 97 | 155 | 192 | 142 | 586 |
| | (7) | (15) | (3) | (11) | |
| 11 | 155 | 182 | 192 | 192 | 721 |
| | (16) | (8) | (12) | (4) | |
| 12 | 182 | 207 | 232 | 162 | 783 |
| | | | | | 2729 |

Rep. IV

| Block | | | | | Totals |
|---|---|---|---|---|---|
| | (1) | (14) | (7) | (12) | |
| 13 | 220 | 202 | 175 | 205 | 802 |
| | (13) | (2) | (11) | (8) | |
| 14 | 205 | 152 | 180 | 187 | 724 |
| | (5) | (10) | (3) | (16) | |
| 15 | 165 | 150 | 200 | 160 | 675 |
| | (9) | (6) | (15) | (4) | |
| 16 | 155 | 177 | 185 | 172 | 689 |
| | | | | | 2890 |

Rep. V

| Block | | | | | Totals |
|---|---|---|---|---|---|
| | (1) | (10) | (15) | (8) | |
| 17 | 147 | 112 | 177 | 147 | 583 |
| | (9) | (2) | (7) | (16) | |
| 18 | 180 | 205 | 190 | 167 | 742 |
| | (13) | (6) | (3) | (12) | |
| 19 | 172 | 212 | 197 | 192 | 773 |
| | (5) | (14) | (11) | (4) | |
| 20 | 177 | 220 | 205 | 225 | 827 |
| | | | | | 2925 |

[1]The values enclosed in parentheses correspond to the treatment numbers.

$k$ times the grand total where $k$ is the block size that is equal to the square root of the number of treatments. In the present example, $k = 4$.

STEP 4. For each treatment, calculate

$$W = kT - (k + 1)B_t + G.$$

For example, the value $W$ for treatment 5 is computed as:

$$W = 4(816) - (5)(3{,}411) + 13{,}746 = -45.$$

**Table 3.12 Treatment totals and adjustment factors for the analysis of data of table 3.11.**

| Treatment no. | T | $B_t$ | W= (4T−5$B_t$+G) | Adjusted total T+$\mu$W | Adjusted treatment mean |
|---|---|---|---|---|---|
| 1 | 809 | 3,286 | 552 | 829 | 166 |
| 2 | 794 | 3,322 | 312 | 805 | 161 |
| 3 | 908 | 3,411 | 323 | 920 | 184 |
| 4 | 901 | 3,596 | −630 | 878 | 176 |
| 5 | 816 | 3,411 | − 45 | 814 | 163 |
| 6 | 848 | 3,310 | 588 | 869 | 174 |
| 7 | 864 | 3,562 | −608 | 842 | 168 |
| 8 | 865 | 3,332 | 546 | 885 | 177 |
| 9 | 801 | 3,312 | 390 | 815 | 163 |
| 10 | 581 | 3,141 | 365 | 594 | 119 |
| 11 | 946 | 3,534 | −140 | 941 | 188 |
| 12 | 971 | 3,628 | −510 | 953 | 191 |
| 13 | 869 | 3,564 | −598 | 848 | 170 |
| 14 | 994 | 3,588 | −218 | 986 | 197 |
| 15 | 913 | 3,394 | 428 | 928 | 186 |
| 16 | 866 | 3,593 | −755 | 839 | 168 |
| G= | 13,746 | 54,984 | 0 | — | — |

The results are presented in column 3 of table 3.12. To doublecheck, the sum of $W$'s should be zero.

STEP 5. Construct an outline of the analysis of variance specifying the sources of variation and their corresponding degrees of freedom as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square |
|---|---|---|---|
| Replication | $k = 4$ | | |
| Treatment (unadjusted) | $k^2 - 1 = 15$ | | |
| Block (adjusted) | $k^2 - 1 = 15$ | | |
| Intrablock error | $(k - 1)(k^2 - 1) = 45$ | | |
| Total | $k^2(k + 1) - 1 = 79$ | | |

STEP 6. Compute the following sums of squares in the usual manner:

$$\text{Total SS} = \sum X^2 - \text{C.F.}$$

$$= [(147)^2 + (152)^2 + \ldots + (225)^2] - \frac{(13{,}746)^2}{80}$$

$$= 58{,}855,$$

$$\text{Replication SS} = \sum \frac{(\text{replication total})^2}{k^2} - \text{C.F.}$$

$$= \frac{(2{,}595)^2 + (2{,}607)^2 + \ldots + (2{,}925)^2}{16} - \frac{(13{,}746)^2}{80}$$

$$= 5{,}946, \quad \text{and}$$

**Table 3.13 Analysis of variance on tiller number data of table 3.11 (a 4×4 balanced lattice design).**

| Source of variation | Degrees of freedom | Sum of squares | Mean square |
|---|---|---|---|
| Replication | 4 | 5,946 | |
| Treatment (unadj.) | 15 | 26,994 | |
| Block (adj.) | 15 | 11,382 | 759 |
| Intrablock | 45 | 14,533 | 323 |
| Total | 79 | 58,855 | |

$$\text{Treatment SS} = \sum \frac{(\text{treatment total})^2}{k+1} - \text{C.F.}$$

$$= \frac{(869)^2 + (794)^2 + \ldots + (866)^2}{5} - \frac{(13{,}746)^2}{80}$$

$$= 26{,}994.$$

STEP 7. Compute the block within replications sum of squares adjusted for treatment effects as

$$\text{Block (adj.) SS} = \frac{\sum W^2}{k^3(k+1)} = \frac{(-598)^2 + (312)^2 + \ldots + (365)^2}{320}$$

$$= 11{,}382.$$

STEP 8. Compute the intrablock error sum of squares as

$$\begin{aligned}\text{Intrablock error SS} &= \text{total SS} - \text{replication SS} - \text{treatment SS} - \\ &\quad \text{block (adj.) SS} \\ &= 58{,}855 - 5{,}946 - 26{,}994 - 11{,}382 \\ &= 14{,}533.\end{aligned}$$

STEP 9. Compute the mean squares for block (adj.) and intrablock error as follows:

$$\text{Block (adj.) MS} = \frac{\text{block (adj.) SS}}{k^2 - 1}$$

$$= \frac{11{,}382}{15} = 759, \quad \text{and}$$

$$\text{Intrablock error MS} = \frac{\text{intrablock error SS}}{(k-1)(k^2-1)}$$

$$= \frac{14{,}533}{45} = 323.$$

STEP 10. Enter all values computed in steps 6 to 9 in the analysis of variance table. The result is shown in table 3.13.

STEP 11. To make an $F$-test in the analysis of variance, the treatment SS needs to be adjusted. The treatment mean square, if computed, cannot be

tested against the intrablock error mean square because it contains some block effects. The proper adjustments are made as follows:

Calculate the adjustment factor

$$\mu = \frac{\text{block (adj.) MS} - \text{intrablock MS}}{k^2\,[\text{Block (adj.) MS}]}$$

$$= \frac{759 - 323}{16(759)} = 0.0359.$$

Note: if the intrablock MS happens to be greater than the block (adj.) MS, $\mu$ will be taken to be zero and no further adjustment will be needed. The $F$-test can be computed as the ratio of the treatment MS and the intrablock MS in the usual manner.

Calculate the adjusted treatment total for each treatment as:

$$T(\text{adj.}) = T + \mu W.$$

For example, the adjusted treatment total for treatment 5 is computed as:

$$T(\text{adj.}) = 816 + (0.0359)(-45) = 814.$$

The results for all treatments are shown in column 4 of table 3.12.

Calculate the adjusted treatment mean square as:

$$\begin{aligned}\text{Treatment (adj.) MS} &= \frac{1}{r(k^2-1)}\,[\text{treatment (adj.) total}]^2 \\ &\quad - \frac{(\text{grand total})^2}{k^2}. \\ &= \frac{1}{5(15)}\,[(829)^2 + (805)^2 + \ldots + (839)^2] \\ &\quad - \frac{(13{,}746)^2}{16} \\ &= 1{,}602.\end{aligned}$$

Calculate the effective error mean square as:

$$\begin{aligned}\text{Effective error MS} &= (\text{intrablock error MS})(1 + k\mu) \\ &= 323\,[1 + 4\,(0.0359)] = 369.\end{aligned}$$

Calculate the $F$-ratio as:

$$F = \frac{\text{treatment (adj.) MS}}{\text{effective error MS}}$$

$$= \frac{1{,}602}{369} = 4.34.$$

Compare the observed $F$-value with a tabular $F$-value (appendix 5), with 15 and 45 degrees of freedom, at a specified level of significance in the usual manner.

The effective error MS and not the intrablock error MS will be used as error

MS in all tests for comparisons among treatment means described in chapter 6. For comparisons among treatment means, the adjusted treatment means should be used. They are computed from the adjusted treatment total. For example, the adjusted mean of treatment no. 5 is $\frac{814}{5} = 163$. The adjusted treatment means for all treatments are presented in the last column of table 3.12.

STEP 12. Estimate the gain in precision due to the use of a balanced lattice relative to the use of a randomized complete block design as:

$$\text{R.E.} = 100\left[\frac{\text{block(adj.)SS} + \text{intrablock error SS}}{k(k^2-1)(\text{effective error MS})}\right]$$

$$= 100\left[\frac{11{,}382 - 14{,}533}{60(369)}\right] = 117\%.$$

This result indicates that using a balanced lattice design has increased the precision of the experiment by 17% over that which would have been obtained had the randomized complete block design been used.

### 3.5.3 Triple lattices

The analysis of a triple lattice design will be illustrated by rice yield data from an upland rice varietal test involving 81 entries conducted in a 9 × 9 triple lattice design. The plan and data are given in table 3.14. Values enclosed in parentheses correspond to the treatment numbers.

The steps in the analysis are as follows:

**Table 3.14. Grain yield data from an upland rice varietal trial conducted in a 9×9 triple lattice design.[1]**

| Block no. | Grain yields (t/ha) | | | | | | | | | Block total |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | *Rep. I* | | | | | |
| | (1) | (2) | (3) | (4) | (5) | (6) | (7) | (8) | (9) | |
| I | 2.70 | 1.60 | 4.45 | 2.91 | 2.78 | 3.32 | 1.70 | 4.72 | 4.79 | 28.97 |
| | (10) | (11) | (12) | (13) | (14) | (15) | (16) | (17) | (18) | |
| II | 4.20 | 5.22 | 3.96 | 1.51 | 3.48 | 4.69 | 1.57 | 2.61 | 3.16 | 30.40 |
| | (19) | (20) | (21) | (22) | (23) | (24) | (25) | (26) | (27) | |
| III | 4.63 | 3.33 | 6.31 | 6.08 | 1.86 | 4.10 | 5.72 | 5.87 | 4.20 | 42.10 |
| | (28) | (29) | (30) | (31) | (32) | (33) | (34) | (35) | (36) | |
| IV | 3.74 | 3.05 | 5.16 | 4.76 | 3.75 | 3.66 | 4.52 | 4.64 | 5.36 | 38.64 |
| | (37) | (38) | (39) | (40) | (41) | (42) | (43) | (44) | (45) | |
| V | 4.76 | 4.43 | 5.36 | 4.73 | 5.30 | 3.93 | 3.37 | 3.74 | 4.06 | 39.68 |
| | (46) | (47) | (48) | (49) | (50) | (51) | (52) | (53) | (54) | |
| VI | 3.45 | 2.56 | 2.39 | 2.30 | 3.54 | 3.66 | 1.20 | 3.34 | 4.04 | 26.48 |
| | (55) | (56) | (57) | (58) | (59) | (60) | (61) | (62) | (63) | |
| VII | 3.99 | 4.48 | 2.69 | 3.95 | 2.59 | 3.99 | 4.37 | 4.24 | 3.70 | 34.00 |
| | (64) | (65) | (66) | (67) | (68) | (69) | (70) | (71) | (72) | |
| VIII | 5.29 | 3.58 | 2.14 | 5.54 | 5.14 | 5.73 | 3.38 | 3.63 | 5.08 | 39.51 |
| | (73) | (74) | (75) | (76) | (77) | (78) | (79) | (80) | (81) | |
| IX | 3.76 | 6.45 | 3.96 | 3.64 | 4.42 | 6.57 | 6.39 | 3.39 | 4.89 | 43.47 |
| | | | | | | | | | | 323.25 |

continued on opposite page

Table 3.14 continued

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | *Rep. II* | | | | | |
| | (1) | (10) | (19) | (28) | (37) | (46) | (55) | (64) | (73) | |
| I | 3.06 | 2.08 | 2.95 | 3.75 | 4.08 | 3.88 | 2.14 | 3.68 | 2.85 | 28.47 |
| | (2) | (11) | (20) | (29) | (38) | (47) | (56) | (65) | (74) | |
| II | 1.61 | 5.30 | 2.75 | 4.06 | 3.89 | 2.60 | 4.19 | 3.14 | 4.82 | 32.36 |
| | (3) | (12) | (21) | (30) | (39) | (48) | (57) | (66) | (75) | |
| III | 4.19 | 3.33 | 4.67 | 4.99 | 4.58 | 3.17 | 2.69 | 2.57 | 3.82 | 34.01 |
| | (4) | (13) | (22) | (31) | (40) | (49) | (58) | (67) | (76) | |
| IV | 2.99 | 2.50 | 4.87 | 3.71 | 4.85 | 2.87 | 3.79 | 5.28 | 3.32 | 34.18 |
| | (5) | (14) | (23) | (32) | (41) | (50) | (59) | (68) | (77) | |
| V | 3.81 | 3.48 | 1.87 | 4.34 | 4.36 | 3.24 | 3.62 | 4.49 | 3.62 | 32.83 |
| | (6) | (15) | (24) | (33) | (42) | (51) | (60) | (69) | (78) | |
| VI | 3.34 | 3.30 | 3.68 | 3.84 | 4.25 | 3.90 | 3.64 | 5.09 | 6.10 | 37.14 |
| | (7) | (16) | (25) | (34) | (43) | (52) | (61) | (70) | (79) | |
| VII | 2.98 | 2.69 | 5.55 | 3.52 | 4.03 | 1.20 | 4.36 | 3.18 | 6.77 | 34.28 |
| | (8) | (17) | (26) | (35) | (44) | (53) | (62) | (71) | (80) | |
| VIII | 4.20 | 2.69 | 5.14 | 4.32 | 3.47 | 3.41 | 3.74 | 3.67 | 2.27 | 32.91 |
| | (9) | (18) | (27) | (36) | (45) | (54) | (63) | (72) | (81) | |
| IX | 4.75 | 2.59 | 3.94 | 4.51 | 3.10 | 3.59 | 2.70 | 4.40 | 4.86 | 34.44 |
| | | | | | | | | | | 300.62 |
| | | | | | *Rep. III* | | | | | |
| | (1) | (12) | (20) | (34) | (45) | (53) | (58) | (70) | (77) | |
| I | 3.52 | 2.18 | 3.50 | 3.30 | 3.88 | 2.45 | 3.75 | 4.45 | 4.14 | 31.17 |
| | (2) | (10) | (21) | (35) | (43) | (54) | (59) | (67) | (78) | |
| II | .79 | 3.58 | 4.83 | 3.63 | 3.02 | 4.20 | 3.59 | 5.06 | 6.51 | 35.21 |
| | (3) | (11) | (19) | (36) | (44) | (52) | (60) | (68) | (76) | |
| III | 4.69 | 5.33 | 4.43 | 5.31 | 4.13 | 1.98 | 4.66 | 4.50 | 4.50 | 39.53 |
| | (4) | (15) | (23) | (28) | (39) | (47) | (61) | (72) | (80) | |
| IV | 3.06 | 4.30 | 2.02 | 3.57 | 5.80 | 2.58 | 4.27 | 4.84 | 2.74 | 33.18 |
| | (5) | (13) | (24) | (29) | (37) | (48) | (62) | (70) | (81) | |
| V | 3.79 | .88 | 3.40 | 4.92 | 2.12 | 1.89 | 3.73 | 3.51 | 3.50 | 27.74 |
| | (6) | (14) | (22) | (30) | (38) | (46) | (63) | (71) | (79) | |
| VI | 3.34 | 3.94 | 5.72 | 5.34 | 4.47 | 4.18 | 2.70 | 3.96 | 3.48 | 37.13 |
| | (7) | (18) | (26) | (31) | (42) | (50) | (55) | (66) | (74) | |
| VII | 2.35 | 2.87 | 5.50 | 2.72 | 4.20 | 2.87 | 2.99 | 1.62 | 5.33 | 30.45 |
| | (8) | (16) | (27) | (32) | (40) | (51) | (56) | (64) | (75) | |
| VIII | 4.51 | 1.26 | 4.20 | 3.19 | 4.76 | 3.35 | 3.61 | 4.52 | 3.30 | 32.78 |
| | (9) | (17) | (25) | (33) | (41) | (49) | (57) | (65) | (73) | |
| IX | 4.21 | 3.17 | 5.03 | 3.34 | 5.31 | 3.05 | 3.19 | 2.63 | 4.06 | 33.99 |
| | | | | | | | | | | 301.18 |

[1]The values enclosed in parentheses correspond to the treatment numbers.

STEP 1. Calculate the block totals and the replication totals as shown in table 3.14. Calculate the grand total as 925.05.

STEP 2. Calculate the treatment totals as shown in table 3.15.

STEP 3. Construct an outline of the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square |
|---|---|---|---|
| Replication | $r - 1 = 2$ | | |
| Treatment (unadj.) | $k^2 - 1 = 80$ | | |
| Block (adj.) | $r(k - 1) = 24$ | | |
| Intrablock error | $(k - 1)(rk - k - 1) = 136$ | | |
| Total | $rk^2 - 1 = 242$ | | |

**Table 3.15 Treatment totals computed from data of table 3.14.**

| Treatment | | Treatment | | Treatment | | Treatment | | Treatment | | Treatment | | Treatment | | Treatment | | Treatment | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| no. | total | no. | total | no. | total | no. | total | no. | total | no. | total | no. | total | no. | total | no. | total |
| 1 | 9.28 | 2 | 4.00 | 3 | 13.33 | 4 | 8.96 | 5 | 10.38 | 6 | 10.00 | 7 | 7.03 | 8 | 13.43 | 9 | 13.75 |
| 10 | 9.86 | 11 | 15.85 | 12 | 9.47 | 13 | 4.89 | 14 | 10.90 | 15 | 12.29 | 16 | 5.52 | 17 | 8.47 | 18 | 8.62 |
| 19 | 12.01 | 20 | 9.58 | 21 | 15.81 | 22 | 16.67 | 23 | 5.75 | 24 | 11.18 | 25 | 16.30 | 26 | 16.51 | 27 | 12.34 |
| 28 | 11.06 | 29 | 12.03 | 30 | 15.49 | 31 | 11.19 | 32 | 11.28 | 33 | 10.84 | 34 | 11.34 | 35 | 12.59 | 36 | 15.18 |
| 37 | 10.96 | 38 | 12.79 | 39 | 15.74 | 40 | 14.34 | 41 | 14.97 | 42 | 12.38 | 43 | 10.42 | 44 | 11.34 | 45 | 11.04 |
| 46 | 11.51 | 47 | 7.74 | 48 | 7.45 | 49 | 8.22 | 50 | 9.65 | 51 | 10.91 | 52 | 4.38 | 53 | 9.20 | 54 | 11.83 |
| 55 | 9.12 | 56 | 12.28 | 57 | 8.57 | 58 | 11.49 | 59 | 9.80 | 60 | 12.29 | 61 | 13.00 | 62 | 11.71 | 63 | 9.10 |
| 64 | 13.49 | 65 | 9.35 | 66 | 6.33 | 67 | 15.88 | 68 | 14.13 | 69 | 15.27 | 70 | 10.07 | 71 | 11.26 | 72 | 14.32 |
| 73 | 10.67 | 74 | 16.60 | 75 | 11.16 | 76 | 11.46 | 77 | 12.18 | 78 | 19.18 | 79 | 16.64 | 80 | 8.40 | 81 | 13.25 |

STEP 4. Compute the following sums of squares in the usual manner:

$$\text{Total SS} = \sum X^2 - \text{C.F.}$$
$$= [(2.70)^2 + (1.60)^2 + \ldots + (4.06)^2] - \frac{(925.05)^2}{243}$$
$$= 308.9883,$$

$$\text{Replication SS} = \frac{\sum(\text{replication total})^2}{k^2} - \text{C.F.}$$
$$= \frac{(323.25)^2 + (300.62)^2 + (301.18)^2}{81} - \frac{(925.05)^2}{243}$$
$$= 4.1132, \quad \text{and}$$

$$\text{Treatment SS} = \frac{\sum(\text{treatment total})^2}{r} - \text{C.F.}$$
$$= \frac{(9.28)^2 + (4.00)^2 + \ldots + (13.25)^2}{3} - \frac{(925.05)^2}{243}$$
$$= 256.7386.$$

STEP 5. For each block, calculate $C_b$ as

$$C_b = M - 3B,$$

where $M$ is the sum of treatment totals for all treatments appearing in the particular block, and $B$ is the block total of the particular block concerned. For example, block II of replication II contained the following treatment numbers: 2, 11, 20, 29, 38, 47, 56, 65, and 74. Hence, $M$ for block II of replication II is computed as

$$M = 4.00 + 15.85 + 9.58 + 12.03 + 12.79 + 7.74 + 12.28 + 9.35 + 16.60 = 100.22,$$

and $C_b$ is then computed as

$$C_b = 100.22 - 3(32.36) = 3.14.$$

The $C_b$ values for the 27 blocks are presented in table 3.16.

STEP 6. Calculate the replication totals ($R_c$) of the $C_b$ values for each replication as follows:

*Replication I* : $R_c = 3.25 - 5.33 - 10.15 + \ldots - 10.87 = -44.70$,
*Replication II* : $R_c = 12.55 + 3.14 + 1.32 + \ldots + 6.11 = 23.19$, and
*Replication III* : $R_c = 5.34 + 3.74 - 8.62 + \ldots - 0.83 = 21.51$.

Note: to countercheck, the $R_c$ values should sum up to zero.

STEP 7. Calculate the adjusted sum of squares for blocks as

$$\text{Block (adj.) SS} = \frac{\sum C_b^2}{kr(r-1)} - \frac{\sum R_c^2}{k^2 r(r-1)}$$

**Table 3.16 The values of $C_b$ computed from data of tables 3.14 and 3.15.**

| Rep. I | | Rep. II | | Rep. III | |
|---|---|---|---|---|---|
| Block no. | $C_b$ | Block no. | $C_b$ | Block no. | $C_b$ |
| 1 | 3.25 | 1 | 12.55 | 1 | 5.34 |
| 2 | −5.33 | 2 | 3.14 | 2 | 3.74 |
| 3 | −10.15 | 3 | 1.32 | 3 | −8.62 |
| 4 | −4.92 | 4 | 0.56 | 4 | −2.28 |
| 5 | −5.06 | 5 | 0.55 | 5 | 8.70 |
| 6 | 1.45 | 6 | 2.92 | 6 | 2.97 |
| 7 | −4.64 | 7 | −8.14 | 7 | 6.08 |
| 8 | −8.43 | 8 | 4.18 | 8 | 6.41 |
| 9 | −10.87 | 9 | 6.11 | 9 | −0.83 |
| Total | −44.70 | | 23.19 | | 21.51 |

$$= \frac{(3.25)^2 + (-5.33)^2 + \ldots + (-0.83)^2}{54} - \frac{(-44.70)^2 + (23.19)^2 + (21.51)^2}{486}$$

$$= 12.1492.$$

STEP 8. Calculate the intrablock error sum of squares as

$$\begin{aligned}\text{Intrablock error SS} &= \text{total SS} - \text{replication SS} - \text{treatment SS} \\ &\quad - \text{block (adj.) SS} \\ &= 308.9883 - 4.1132 - 256.7386 - 12.1492 \\ &= 35.9873.\end{aligned}$$

STEP 9. Calculate the mean square for intrablock error and block (adj.) as follows:

$$\text{Intrablock error MS} = \frac{\text{intrablock error SS}}{(k-1)(rk-k-1)} = \frac{35.9873}{136} = 0.2646, \quad \text{and}$$

$$\text{Block (adj.) MS} = \frac{\text{block (adj.) SS}}{r(k-1)} = \frac{12.1492}{24} = 0.5062.$$

STEP 10. Enter all sums of squares and mean squares values computed in steps 4 to 9 in the analysis of variance. The final result is shown in table 3.17.

STEP 11. Calculate the adjustment factor as

$$\mu = \frac{\left(\dfrac{1}{\text{MSE}} - \dfrac{2}{3\text{MSB} - \text{MSE}}\right)}{k\left(\dfrac{2}{\text{MSE}} + \dfrac{2}{3\text{MSB} - \text{MSE}}\right)},$$

**Table 3.17 Analysis of variance for grain yield data of table 3.14 (a 9×9 triple lattice design).**

| Source of variation | Degrees of freedom | Sum of squares | Mean square |
|---|---|---|---|
| Replication | 2 | 4.1132 | |
| Block (adj.) | 24 | 12.1492 | 0.5062 |
| Treatment | 80 | 256.7386 | |
| Intrablock error | 136 | 35.9873 | 0.2646 |
| Total | 242 | 308.9883 | |

where MSE is the intrablock error MS and MSB is the block (adj.) MS. The result is

$$\mu = \frac{\left(\dfrac{1}{0.2646} - \dfrac{2}{3(0.5062) - 0.2646}\right)}{9\left[\dfrac{2}{0.2646} + \dfrac{2}{3(0.5062 - 0.2646}\right]} = 0.0265.$$

If MSB is less than MSE, no further adjustment is needed. The $F$-test required can be made directly from the analysis of variance of table 3.17.

STEP 12. Calculate the adjusted treatment totals as

$$T(\text{adj.}) = T + \mu\sum C_b,$$

where the summation runs over all blocks in which the particular treatment appears. For example, for treatment number 2, the adjusted treatment total is computed as 4.00 + 0.0265 (3.25 + 3.14 + 3.74) = 4.27. The adjusted treatment means can then be computed in the standard form.

STEP 13. The $F$-test of the adjusted treatment totals can be made by computing the adjusted treatment sum of squares as:

$$\text{Treatment (adj.) SS} = \text{treatment (unadj.) SS} - \left(\frac{1}{\text{MSE}} - \frac{2}{3\text{MSB} - \text{MSE}}\right)$$
$$[(\text{MSE})B_u - \text{MSE}(\text{MSB} - \text{MSE})],$$

where $B_u$ is the unadjusted sum of squares for blocks within replications, which is computed as

$$\begin{aligned} B_u &= \frac{\sum(\text{block total})^2}{k} - \frac{\sum(\text{rep. total})^2}{k^2} \\ &= \frac{(28.97)^2 + (30.40)^2 + \ldots + (33.99)^2}{9} \\ &\quad - \frac{(323.25)^2 + (300.62)^2 + (301.18)^2}{81} \\ &= 49.4653. \end{aligned}$$

Therefore, the treatment (adj.) SS

$$= 256.7386 - \left(\frac{1}{0.2646} - \frac{2}{3(0.5062) - 0.2646}\right)[0.2646(49.4653)$$
$$- 0.2646(0.5062 - 0.2646)]$$
$$= 233.9467.$$

Finally, the $F$-test is computed as follows:

$$F = \frac{\text{treatment (adj.)SS}/(k^2 - 1)}{\text{intrablock error MS}}$$
$$= \frac{233.9467/80}{0.2646} = \frac{2.9243}{0.2646} = 11.05,$$

which is compared with the tabular $F$-value with 80 and 136 degrees of freedom. The test indicates a highly significant result.

STEP 14. For comparisons of pairs of treatment means, two different error terms are used: one for comparison between treatments appearing in the same block and another for comparison between treatments that have never appeared in the same block.

*Error MS for comparison between treatments appearing in the same block:*

$$\text{Error MS (1)} = \frac{\text{MSE}}{k}\left[\frac{\dfrac{6}{\text{MSE}}}{\dfrac{2}{\text{MSE}} + \dfrac{2}{3\text{MSB} - \text{MSE}}} - + (k - 2)\right]$$
$$= \frac{(0.2646)}{9}\left[\frac{\dfrac{6}{0.2646}}{\dfrac{2}{0.2646} + \dfrac{2}{3(0.5062) - 0.2646}} + 7\right]$$
$$= 0.2786.$$

*Error MS for comparison between treatments that have not appeared in the same block:*

$$\text{Error MS (2)} = \frac{\text{MSE}}{k}\left[\frac{\dfrac{9}{\text{MSE}}}{\dfrac{2}{\text{MSE}} + \dfrac{2}{3\text{MSB} - \text{MSE}}} + (k - 3)\right]$$
$$= \frac{(0.2646)}{9}\left[\frac{\dfrac{9}{0.2646}}{\dfrac{2}{0.2646} + \dfrac{2}{3(0.5062) - 0.2646}} + 6\right]$$
$$= 0.2856.$$

Note that for most practical purposes, especially when the experiment is large, the average error MS can be used for all comparisons among treatments. The average error MS is computed as:

$$\text{Av. error MS} = \frac{\text{MSE}}{(k+1)}\left[\frac{\dfrac{9}{\text{MSE}}}{\dfrac{2}{\text{MSE}} + \dfrac{2}{3\text{MSB} - \text{MSE}}} + (k-2)\right]$$

$$= \frac{0.2646}{10}\left[\frac{\dfrac{9}{0.2646}}{\dfrac{2}{0.2646} + \dfrac{2}{3(0.5062) - 0.2646}} + 7\right]$$

$$= 0.2835.$$

STEP 15. Compute the precision relative to randomized complete block design as

$$\text{R.E.} = \left[\frac{\text{block (adj.)SS} + \text{intrablock error SS}}{r(k-1) + (k-1)(rk-k-1)}\right]\left[\frac{100}{\text{Av. error MS}}\right]$$

$$= \left[\frac{12.1492 + 35.9873}{24 + 136}\right]\left[\frac{100}{0.2835}\right] = 106.1\%.$$

### 3.5.4 Quadruple lattices

As mentioned in the previous section, the plan for a quadruple lattice can be derived such that each of the four replications comes from a unique basic plan or such that two replications are repetitions of the other two. The second alternative basically consists of two simple lattices, one as a repetition of the other. In this section we shall illustrate the analysis for a quadruple lattice with repetitions, but the procedure for analyzing data from a simple lattice can be directly derived from the procedure discussed here.

Data from a rice varietal trial conducted in a 5 × 5 quadruple lattice design will be used. The plan and data are presented in table 3.18. Replications I and II are from the usual simple lattice plan, and replications III and IV are repetitions of replications I and II, respectively.

The steps in the analysis are as follows:

STEP 1. Calculate the block totals as shown in table 3.18. The grand total is computed as 606,426.

STEP 2. Calculate the treatment totals as shown in table 3.19.

STEP 3. Construct an outline of the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square |
|---|---|---|---|
| Replication | $(r-1) = 3$ | | |
| Treatment (unadj.) | $(k^2-1) = 24$ | | |
| Block (adj.): | $r(k-1) = 16$ | | |
| Component ($a$) | $n(p-1)(k-1) = (8)$ | | |
| Component ($b$) | $n(k-1) = (8)$ | | |
| Intrablock error | $(k-1)(rk-k-1) = 56$ | | |
| Total | $k^2np - 1 = 99$ | | |

**Table 3.18 Grain yield data (kg/ha) from a rice varietal trial conducted in a 5×5 quadruple lattice.**

| Rep. I | | | Rep. II | | | Rep. III | | | Rep. IV | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Treatment no. | Yield (kg/ha) | Block total | Treatment no. | Yield (kg/ha) | Block total | Treatment no. | Yield (kg/ha) | Block total | Treatment no. | Yield (kg/ha) | Block total |
| 1 | 4,723 | | 1 | 6,262 | | 1 | 5,975 | | 1 | 5,228 | |
| 2 | 4,977 | | 6 | 5,690 | | 2 | 5,915 | | 6 | 5,302 | |
| 3 | 6,247 | | 11 | 6,498 | | 3 | 6,914 | | 11 | 5,190 | |
| 4 | 5,325 | | 16 | 8,011 | | 4 | 6,389 | | 16 | 7,127 | |
| 5 | 7,139 | | 21 | 5,887 | | 5 | 7,542 | | 21 | 5,323 | |
| | | 28,411 | | | 32,348 | | | 37,735 | | | 28,170 |
| 6 | 5,444 | | 2 | 5,038 | | 6 | 4,750 | | 2 | 5,681 | |
| 7 | 5,567 | | 7 | 4,615 | | 7 | 5,983 | | 7 | 6,146 | |
| 8 | 5,809 | | 12 | 5,520 | | 8 | 5,339 | | 12 | 6,032 | |
| 9 | 5,086 | | 17 | 6,063 | | 9 | 4,615 | | 17 | 7,066 | |
| 10 | 6,849 | | 22 | 6,486 | | 10 | 5,336 | | 22 | 6,680 | |
| | | 28,755 | | | 27,722 | | | 26,023 | | | 31,605 |
| 11 | 5,237 | | 3 | 6,057 | | 11 | 5,073 | | 3 | 6,750 | |
| 12 | 5,174 | | 8 | 6,397 | | 12 | 6,110 | | 8 | 6,567 | |
| 13 | 5,395 | | 13 | 5,214 | | 13 | 6,001 | | 13 | 5,786 | |
| 14 | 5,112 | | 18 | 7,093 | | 14 | 5,486 | | 18 | 7,159 | |
| 15 | 5,637 | | 23 | 7,002 | | 15 | 6,415 | | 23 | 7,268 | |
| | | 26,555 | | | 31,763 | | | 29,085 | | | 33,530 |
| 16 | 5,793 | | 4 | 5,291 | | 16 | 6,064 | | 4 | 6,020 | |
| 17 | 6,008 | | 9 | 4,864 | | 17 | 6,405 | | 9 | 5,760 | |
| 18 | 6,864 | | 14 | 5,453 | | 18 | 6,856 | | 14 | 6,413 | |
| 19 | 5,026 | | 19 | 4,917 | | 19 | 4,654 | | 19 | 5,760 | |
| 20 | 6,348 | | 24 | 6,318 | | 20 | 5,986 | | 24 | 6,856 | |
| | | 30,039 | | | 26,843 | | | 29,965 | | | 30,185 |
| 21 | 5,321 | | 5 | 7,685 | | 21 | 5,750 | | 5 | 7,173 | |
| 22 | 6,870 | | 10 | 5,985 | | 22 | 6,539 | | 10 | 5,626 | |
| 23 | 7,512 | | 15 | 6,107 | | 23 | 7,576 | | 15 | 6,310 | |
| 24 | 6,648 | | 20 | 6,710 | | 24 | 7,372 | | 20 | 6,529 | |
| 25 | 6,948 | | 25 | 6,915 | | 25 | 6,439 | | 25 | 6,677 | |
| | | 33,299 | | | 33,402 | | | 33,676 | | | 32,315 |
| Replication total | | 147,059 | | | 152,078 | | | 151,484 | | | 155,805 |

**Table 3.19 Treatment totals computed from data of table 3.18.**

| Treatment | | Treatment | | Treatment | | Treatment | | Treatment | |
|---|---|---|---|---|---|---|---|---|---|
| no. | total | no. | total | no. | total | no. | total | no. | total |
| 1 | 22,188 | 2 | 21,611 | 3 | 25,968 | 4 | 23,025 | 5 | 29,539 |
| 6 | 21,186 | 7 | 22,311 | 8 | 24,112 | 9 | 19,701 | 10 | 23,796 |
| 11 | 21,998 | 12 | 22,836 | 13 | 22,396 | 14 | 22,464 | 15 | 24,469 |
| 16 | 26,995 | 17 | 25,542 | 18 | 27,972 | 19 | 20,357 | 20 | 25,573 |
| 21 | 22,281 | 22 | 26,575 | 23 | 29,358 | 24 | 27,194 | 25 | 26,979 |

Here $n$ is the number of replications in the basic plan and $p$ is the number of repetitions of the basic plan. In our example, $n = 2$ (i.e., simple lattice is the basic plan) and $p = 2$ (i.e., the basic plan is repeated twice).

STEP 4. Compute the following sums of squares in the usual manner:

$$\begin{aligned}\text{Total SS} &= \sum X^2 - \text{C.F.}\\ &= [(4{,}723)^2 + (4{,}977)^2 + \ldots + (6{,}677)^2] - \frac{(606{,}426)^2}{100}\\ &= 63{,}513{,}101.24,\end{aligned}$$

$$\begin{aligned}\text{Replication SS} &= \frac{\sum(\text{replication total})^2}{k^2} - \text{C.F.}\\ &= \frac{(147{,}059)^2 + (152{,}078)^2 + (151{,}484)^2 + (155{,}805)^2}{25}\\ &\quad - \frac{(606{,}426)^2}{100}\\ &= 1{,}541{,}779.08, \quad \text{and}\end{aligned}$$

$$\begin{aligned}\text{Treatment (unadj.) SS} &= \frac{\sum(\text{treatment total})^2}{r} - \text{C.F.}\\ &= \frac{(22{,}188)^2 + (21{,}611)^2 + \ldots + (26{,}979)^2}{4}\\ &\quad - \frac{(606{,}426)^2}{100}\\ &= 45{,}726{,}281.24.\end{aligned}$$

STEP 5. Rearrange the block totals of table 3.18 by placing any two blocks containing the same set of $k$ treatments side by side as shown in table 3.20. Compute the sum over two block totals for each row as shown in column 4 of table 3.20.

Compute the $C$ values for each group of similar blocks as

$$C = (\text{sum of treatment totals over all treatments appearing in the group}) - n(\text{sum of block totals in the group}).$$

For example, for the sixth row (i.e. block number 1 of replications II and IV), the treatments appearing in this group are treatment numbers 1, 6, 11, 16, and 21 (see table 3.18). Hence, $C$ for this group is computed as

$$C = (22{,}188 + 21{,}186 + 21{,}998 + 26{,}995 + 22{,}281) - 2(60{,}518)$$
$$= 6{,}388.00.$$

The results of all $C$ values are shown in the fifth column of table 3.20.

Compute the sum of $C$ values for each repetition, i.e., $R_c$. Here, there are two repetitions, and, hence, two values of $R_c$ namely, 9,340 and −9,340. The sum of the two $R_c$ values must always be zero.

STEP 6. Compute the two components of the block (adj.) sum of squares:

$$\text{Component (b) SS} = \frac{\sum C^2}{kr(n-1)} - \frac{\sum R_c^2}{k^2 r(n-1)}$$
$$= \frac{(39)^2 + (1{,}550)^2 + \ldots + (-1{,}078)^2}{20}$$
$$- \frac{(9{,}340)^2 + (-9{,}340)^2}{100}$$
$$= 3{,}198{,}864.70.$$

The component (a) SS is needed only with the design having repetition of a certain basic design. Thus, for an ordinary simple lattice, there will be only one component, namely, component (b), in the block (adj.) sum of squares.

Table 3.20 will be used for the base data for computing component (a) sum of squares. Treat the data in columns 2 and 3 of table 3.20 as coming from two $k \times p$ tables, each of which can be analyzed as follows:

| Source of variation | Degrees of freedom |
|---|---|
| Row | $k - 1 = 4$ |
| Column | $p - 1 = 1$ |
| Row × column | $(k - 1)(p - 1) = 4$ |
| Total | $kp - 1 = 9$ |

The component (a) SS is the sum of the row × column interaction sum of squares over all $n$ tables. (In this instance, the sum over two tables.) It can be obtained from the following calculations:

$$\text{Total SS} = \frac{(28{,}411)^2 + (28{,}755)^2 + \ldots + (32{,}315)^2}{k = 5}$$
$$- \frac{(298{,}543)^2 + (307{,}883)^2}{50}$$
$$= 23{,}435{,}940.84,$$
$$\text{Row SS} = \frac{(61{,}146)^2 + (54{,}778)^2 + \ldots + (65{,}717)^2}{pk = 10}$$
$$- \frac{(298{,}543)^2 + (307{,}883)^2}{50}$$

**Table 3.20 Block totals of table 3.18 rearranged in pairs of blocks containing the same set of treatments.**

| Block no. | Block totals | | Sum | C | $\mu C$ |
|---|---|---|---|---|---|
| | *Rep. I* | *Rep. III* | | | |
| 1 | 28,411 | 32,735 | 61,146 | 39 | 6.15 |
| 2 | 28,755 | 26,023 | 54,778 | 1,550 | 244.27 |
| 3 | 26,555 | 29,085 | 55,640 | 2,883 | 454.33 |
| 4 | 30,039 | 29,965 | 60,004 | 6,431 | 1,013.47 |
| 5 | 33,299 | 33,676 | 66,975 | −1,563 | −246.31 |
| (Total) | (147,059) | (151,484) | (298,543) | (9,340) | |
| | *Rep. II* | *Rep. IV* | | | |
| 1 | 32,348 | 28,170 | 60,518 | −6,388 | −1,066.69 |
| 2 | 27,722 | 31,605 | 59,327 | 221 | 34.83 |
| 3 | 31,763 | 33,530 | 65,293 | −780 | −122.92 |
| 4 | 26,843 | 30,185 | 57,028 | −1,315 | −207.23 |
| 5 | 33,402 | 32,315 | 65,717 | −1,078 | −169.88 |
| (Total) | (152,078) | (155,805) | (307,883) | (−9,340) | |

$$= 15{,}364{,}390.84\,, \quad \text{and}$$

$$\text{Column SS} = \frac{(147{,}059)^2 + (152{,}078)^2 + \ldots + (155{,}805)^2}{k^2 = 25} - \frac{(298{,}543)^2 + (307{,}883)^2}{50}$$

$$= 669{,}423.08\,.$$

Component (a) SS is computed as

Total SS − row SS − column SS
= 23,435,940.84 − 15,364,390.84 − 669,423.08
= 7,402,126.92 and
Block (adj.) SS = component (a) SS + component (b) SS
= 7,402,126.92 + 3,198,864.70 = 10,600,991.62.

STEP 7. Compute the intrablock error SS as

Intrablock error SS = total SS − replication SS − treatment (unadj.) SS − block (adj.) SS
= 63,513,101.24 − 1,541,779.08 − 45,726,281.24 − 10,600,991.62 = 5,644,049.30.

STEP 8. Compute the block (adj.) mean square and the intrablock error mean square as

$$\text{MSB} = \frac{10{,}600{,}991.62}{16} = 662{,}561.98\,, \quad \text{and}$$

$$\text{MSE} = \frac{5{,}644{,}049.30}{56} = 100{,}786.59\,.$$

STEP 9. Enter all values of sum of squares and mean squares computed in steps 4 to 8 in the analysis of variance. The final result is shown in table 3.21.

**Table 3.21 Analysis of variance of grain yield data of table 3.18.**

| Source of variation | Degrees of freedom | Sum of squares | Mea squa |
|---|---|---|---|
| Replication | 3 | 1,541,779.08 | |
| Treatment | 24 | 45,726,281.24 | |
| Block (adj.): | 16 | 10,600,991.62 | 662,561 |
| Component (a) | (8) | 7,402,126.92 | |
| Component (b) | (8) | 3,198,864.70 | |
| Intrablock error | 56 | 5,644,049.30 | 100,786 |
| Total | 99 | | |

STEP 10. Calculate the adjustment factor

$$\mu = \frac{p(\text{MSB} - \text{MSE})}{k[(r - p)\text{MSB} - (p - 1)\text{MSE}]}$$
$$= \frac{2(662{,}561.98 - 100{,}786.59)}{5[2(662{,}561.98) - (100{,}786.59)]}$$
$$= 0.15759.$$

Multiply the adjustment factor, $\mu$, to each $C$ value in table 3.20. The resu are presented in the last column of table 3.20.

STEP 11. Calculate the adjusted treatment totals as

$$\text{T(adj.)} = \text{T} + \sum \mu \text{C}$$

where the summation runs over all blocks in which the particular treatme appears. For example, the adjusted treatment total for treatment number 1 computed from data of tables 3.19 and 3.20 as

Adjusted treatment total of treatment number 1
$= 22{,}188 + (6.15 - 1{,}006.69)$
$= 21{,}188.$

The results of all adjusted treatment totals are shown in table 3.22.

STEP 12. For the $F$-test, compute the adjusted treatment sum of square:

$$\text{Treatment (adj.) SS} = \text{treatment (unadj.) SS} - k(n - 1)\mu\left[\frac{n(B_u)}{(n - 1)(1 + k\mu)} - B_a\right].$$

**Table 3.22 Adjusted treatment total for data of tables 3.18 and 3.19.**

| Treatment no. | Treatment total | Treatment no. | Treatment total | Treatment no. | Treatment total | Treatment no. | Treatment total | Treatment no. | Treatment total |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 21,188 | 2 | 21,652 | 3 | 25,851 | 4 | 22,824 | 5 | 29,37 |
| 6 | 20,424 | 7 | 22,590 | 8 | 24,233 | 9 | 19,738 | 10 | 23,87 |
| 11 | 21,446 | 12 | 23,325 | 13 | 22,727 | 14 | 22,711 | 15 | 24,75 |
| 16 | 27,002 | 17 | 26,590 | 18 | 28,863 | 19 | 21,163 | 20 | 26,41 |
| 21 | 21,028 | 22 | 26,364 | 23 | 28,989 | 24 | 26,740 | 25 | 26,56 |

where $B_a$ is the component (b) SS and $B_u$ the row SS used for computing component (a) SS in step 6. Hence, we have

$$\begin{aligned}\text{Treatment (adj.) SS} &= 45{,}726{,}281.24 - 5(1)(0.15759)\\ &\quad \left\{\frac{2(15{,}364{,}390.84)}{(1)[1 + 5(0.15759)]} - 1{,}198{,}864.7\right\}\\ &= 11{,}021{,}656.58.\end{aligned}$$

The $F$-test then is

$$\begin{aligned}F &= \frac{\text{treatment (adj.) SS}/(k^2 - 1)}{\text{intrablock error MS}}\\ &= \frac{11{,}021{,}656.58/24}{100{,}786.59} = 14.35.\end{aligned}$$

Compare the observed $F$-value with a tabular $F$-value with $k^2 - 1 = 24$ and $(k - 1)(rk - k) = 56$ degrees of freedom at a specified level of significance. Here the observed $F$-value is greater than the corresponding $F$-value at 1% level of significance, indicating highly significant differences among treatments.

STEP 13. The effective error mean squares appropriate for making tests on comparisons among treatment means are computed for treatments of the same block as

$$\begin{aligned}\text{Effective error MS} &= \text{MSE}[1 + (n - 1)\mu]\\ &= 100{,}786.59[1 + (0.15759)]\\ &= 116{,}669.62, \quad \text{and}\end{aligned}$$

for treatments not of the same block as

$$\begin{aligned}\text{Effective error MS} &= \text{MSE}[1 + n\mu]\\ &= 100{,}786.59[1 + 2(0.15759)]\\ &= 132{,}552.64.\end{aligned}$$

For most practical purposes, especially when the experiment is large, the average error MS can be used for all comparisons among treatments. The average effective error MS is computed as

$$\begin{aligned}\text{MSE}\left[1 + \frac{nk\mu}{k + 1}\right] &= 100{,}786.59\left[1 + \frac{2(5)(0.15759)}{6}\right]\\ &= 127{,}258.30.\end{aligned}$$

STEP 14. Compute the gain in precision relative to randomized complete block design as

$$\begin{aligned}\text{R.E.} &= \left[\frac{\text{block (adj.) MS} + \text{intrablock error MS}}{r(k - 1) + (k - 1)(rk - k - 1)}\right]\left[\frac{100}{\text{Av. effective error MS}}\right]\\ &= \left[\frac{10{,}600{,}991.62 + 5{,}644{,}079.3}{72}\right]\left[\frac{100}{127{,}258.30}\right]\\ &= 177.3\%.\end{aligned}$$

CHAPTER 4

# Two-factor experiments

## 4.1 INTRODUCTION

Since biological organisms are simultaneously exposed to various factors during their lifetimes, single-factor experiments are sometimes criticized for their narrowness. The usual criticism is that single-factor experiments are not realistic since the response of organisms to a single factor varies with changes in other factors. Thus, many biological experiments involve several variable factors considered simultaneously instead of one at a time. In this chapter we shall examine some experimental designs useful in experiments where two factors are examined simultaneously.

## 4.2 FACTORIAL EXPERIMENTS

Factorial experiments are characterized by treatments that are composed of all possible combinations of levels in each of two or more factors. The simplest example is one with two factors each at two levels, e.g., the response of two varieties of rice to two levels of nitrogen. The treatments for such an experiment might be:

Variety *A* at 0 kg N/ha
Variety *B* at 0 kg N/ha
Variety *A* at 60 kg N/ha
Variety *B* at 60 kg N/ha

Note that the term 'factorial' merely specifies how the treatments are formed. It does not in any way describe how the treatments are to be arranged in the experimental plots as any design should. Thus, a factorial should not be confused with a design. Indeed, a factorial experiment still needs to be fitted to a particular design. For instance, if the rice-nitrogen example were fitted into a randomized complete block design, then the correct description would be a factorial experiment in the randomized complete block design.

A major advantage of factorial experiments over single-factor experiments is their versatility to evaluate response to simultaneous changes in several factors. This ushers in an important concept referred to as *interaction*. When

**Table 4.1 Hypothetical examples of (a) no interaction between variety and nitrogen effects and (b) the presence of an interaction.**

| Variety | Rice yield (kg/ha) | | |
|---|---|---|---|
| | 0 kg N/ha | 60 kg N/ha | Average |
| | | (a) no interaction | |
| *A* | 10 | 30 | 20 |
| *B* | 20 | 40 | 30 |
| Av. | 15 | 35 | |
| | | (b) interaction present | |
| *A* | 10 | 30 | 20 |
| *B* | 20 | 20 | 20 |
| Av. | 15 | 25 | |

two variable factors are being evaluated in a single experiment, one of two things can happen: either the change in one factor does not influence the relative effects of the other factor (the two factors do not interact with each other) or the relative effects of one factor depend upon the level of another factor (one factor interacts with the other). The size of this interaction can be measured only if the factors are tested together in a factorial experiment. To illustrate, let us use the previously described experiment involving two varieties of rice and two levels of nitrogen. Table 4.1a shows a hypothetical case in which the two factors do not interact, while table 4.1b shows a case where there is interaction.

When there is no interaction, the relative yield of one factor (e.g., variety) remains the same regardless of the level of the other factor (e.g., nitrogen). Thus, in table 4.1a the difference between the two varieties remains the same at both 0 N and 60 N. That is, the yield of variety *B* relative to variety *A* is 10 units higher at both fertilizer levels. Similarly, the application of 60 kg N/ha increased yield of both varieties *A* and *B* by 20 units.

Table 4.1b shows the case where varieties interact with fertilizers. Variety *B* is better yielding at 0 N, but variety *A* is better at 60 N. In other words, the relative performance of the two varieties at the two levels of nitrogen are not similar. Likewise, while the application of 60 kg N/ha increased the yield of variety *A* by 10 units, it did not change the yield of variety *B*. When there is an interaction between two factors, two single-factor experiments (one on fertilizer and another on varieties) conducted separately would not have revealed as complete information as that obtained from a factorial experiment involving the two factors. If the two varieties were compared in a single-factor experiment, the researcher would have used only one fertilizer level. But the decision as to which of the two varieties is better depends upon whether 0 or 60 kg N/ha was used. This illustrates how limited, and even at times misleading, the information from a single-factor experiment can be. When interaction is present, a factorial experiment would not only give a broader basis for comparing the varieties but would also give additional information concerning

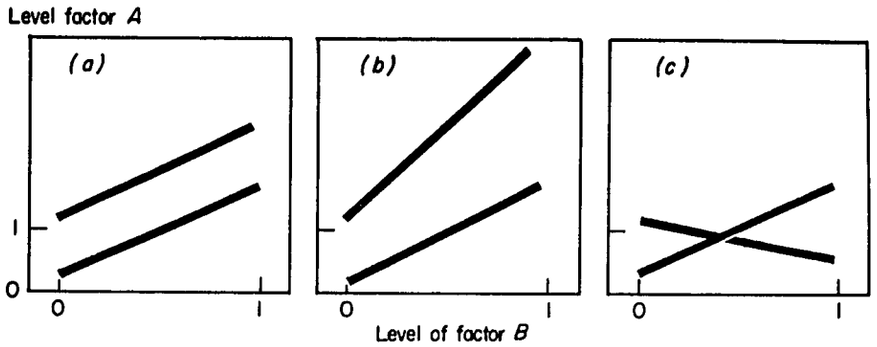


**Fig. 4.1** Graphical illustration of various degrees of interactions between two factors, *A* and *B*, each at two levels: (a) no interaction; (b) mild interaction; and (c) strong interaction.

the desirable fertilizer rate for each of the two varieties. It is thus advisable that a factorial experiment, instead of several single-factor experiments, be conducted whenever possible.

Figure 4.1 illustrates graphically three types of relationship between two factors at two levels each: Figure 4.1a shows no interaction, while figure 4.1b shows a mild interaction where rankings do not change in spite of the difference in response to factor *A* as factor *B* changes, and figure 4.1c shows an extreme interaction where the size of the response in factor *A* is reversed as factor *B* changes.

Because of the wide range of factors that affect biological organisms and because of the need to characterize these effects both separately and in relation to one another, factorial experiments are of major importance and are commonly used in agricultural experiments. This does not imply, however, that one should use factorial experiments uncritically since factorial experiments are usually much larger and therefore more complex and costly to conduct than several single-factor experiments. Furthermore, it is not wise to commit oneself to a large experiment at the investigation's beginning when several small preliminary experiments may indicate promising approaches.

For example, a plant breeder has collected, say, 30 new varieties from a neighboring country. He wants to assess the reaction of these new materials to the local environment. Since the local environment may vary in terms of soil fertility, moisture levels, etc., the ideal case would have been to test the 30 varieties in a factorial experiment involving such other variables as fertilizer, moisture, and population density, The experiment, however, becomes very large as factors other than varieties are added. Even if only three levels of nitrogen were included, the number of treatments would have increased from 30 to 90. This size will mean difficulties in financing, land area, control of soil heterogeneity, etc. Thus, usually the 30 varieties are tested under one environment first, then these results are used to discard the obviously poor varieties and to select a few varieties for a more detailed study. Such a preliminary test, for example, may show that only five varieties are outstanding

**Table 4.2 The 3×5 factorial treatments (treatment combinations of three rice varieties and five nitrogen levels).**

| Nitrogen level (kg/ha) | Variety | | |
|---|---|---|---|
| | 6966 ($V_1$) | PI215936 ($V_2$) | Milfor 6(2) ($V_3$) |
| 0 ($N_0$) | $N_0V_1$ | $N_0V_2$ | $N_0V_3$ |
| 40 ($N_1$) | $N_1V_1$ | $N_1V_2$ | $N_1V_3$ |
| 70 ($N_2$) | $N_2V_1$ | $N_2V_2$ | $N_2V_3$ |
| 100 ($N_3$) | $N_3V_1$ | $N_3V_2$ | $N_3V_3$ |
| 130 ($N_4$) | $N_4V_1$ | $N_4V_2$ | $N_4V_3$ |

enough to warrant further testing. These five varieties could then be put into a factorial experiment with three levels of nitrogen, resulting in a smaller experiment with 15 treatments. Thus, while a factorial experiment will, without question, be more informative than single-factor experiments, practical considerations could limit its use.

In most instances preliminary experiments are very helpful in reducing the levels to be considered in each variable so that the final number of treatments is small enough to be within the limit of the resources of the experimenter.

## 4.3 A TWO-FACTOR EXPERIMENT IN RANDOMIZED COMPLETE BLOCK DESIGN: AN EXAMPLE

Any of the basic designs for single-factor experiments can be applied directly to a factorial experiment. The combination of all the factors involved results

**Table 4.3 Grain yield of three rice varieties under five levels of nitrogen.**

| Nitrogen level (kg/ha) | Grain yield (t/ha) | | | | Treatment | |
|---|---|---|---|---|---|---|
| | Block I | Block II | Block III | Block IV | Total | Mean |
| | | | *6966* | | | |
| 0 | 3.852 | 2.606 | 3.144 | 2.894 | 12.496 | 3.124 |
| 40 | 4.788 | 4.936 | 4.562 | 4.608 | 18.894 | 4.723 |
| 70 | 4.576 | 4.454 | 4.884 | 3.924 | 17.838 | 4.460 |
| 100 | 6.034 | 5.276 | 5.906 | 5.652 | 22.868 | 5.717 |
| 130 | 5.874 | 5.916 | 5.984 | 5.518 | 23.292 | 5.823 |
| | | | *PI215936* | | | |
| 0 | 2.846 | 3.794 | 4.108 | 3.444 | 14.192 | 3.548 |
| 40 | 4.956 | 5.128 | 4.150 | 4.990 | 19.224 | 4.806 |
| 70 | 5.928 | 5.698 | 5.810 | 4.308 | 21.744 | 5.436 |
| 100 | 5.664 | 5.362 | 6.458 | 5.474 | 22.958 | 5.740 |
| 130 | 5.458 | 5.546 | 5.786 | 5.932 | 22.722 | 5.680 |
| | | | *Milfor 6(2)* | | | |
| 0 | 4.192 | 3.754 | 3.738 | 3.428 | 15.112 | 3.778 |
| 40 | 5.250 | 4.582 | 4.896 | 4.286 | 19.014 | 4.754 |
| 70 | 5.822 | 4.848 | 5.678 | 4.932 | 21.280 | 5.320 |
| 100 | 5.888 | 5.524 | 6.042 | 4.756 | 22.210 | 5.552 |
| 130 | 5.864 | 6.264 | 6.056 | 5.362 | 23.546 | 5.886 |
| **Block total** | 76.992 | 73.688 | 77.202 | 69.508 | | |
| **Grand total** | | | | | 297.390 | |
| **Grand mean** | | | | | | 4.956 |

in the total number of treatments, and the randomization schemes are applied as if the treatments are unrelated.

An illustration will be given of a two-factor experiment, involving five levels of nitrogen and three rice varieties (table 4.2), conducted by the Department of Agronomy, IRRI, at Maligaya Experiment Station, Nueva Ecija, in the 1965 wet season. A randomized complete block design with four replications was used. The field layout is shown in figure 4.2, and data on grain yield are given in table 4.3. Note that the layout was constructed using the procedures outlined in section 3.3.2 for a randomized complete block design with 15 treatments.

The steps in the computation are as follows:

STEP 1. Compute treatment totals, treatment means, and the grand total as shown in table 4.3.

STEP 2. Let $r$ be the number of replications, $v$ be the number of varieties, and $n$ be the number of nitrogen levels. Construct an outline of the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Block | $r - 1 = 3$ | | | | | |
| Variety | $v - 1 = 2$ | | | | | |
| Nitrogen | $n - 1 = 4$ | | | | | |
| Variety × nitrogen | $(v - 1)(n - 1) = 8$ | | | | | |
| Error | $(r - 1)(vn - 1) = 42$ | | | | | |
| Total | $rvn - 1 = 59$ | | | | | |

STEP 3. Enter the 15 treatment totals in the two-way table (table 4.4).

STEP 4. Calculate the various sums of squares as follows:

$$\text{C.F.} = \frac{(\text{grand total})^2}{\text{total number of observations}}$$

$$= \frac{(297.390)^2}{60} = 1{,}474.014,$$

$$\text{Total SS} = \sum X^2 - \text{C.F.}$$

$$= [(3.852)^2 + (2.606)^2 + \ldots + (5.362)^2] - \text{C.F.}$$

$$= 1{,}527.544 - 1{,}474.014 = 53.530,$$

$$\text{Block SS} = \frac{\sum(\text{block total})^2}{vn} - \text{C.F.}$$

$$= \frac{(76.992)^2 + \ldots + (69.508)^2}{(3)(5)} - \text{C.F.}$$

$$= 1{,}476.613 - 1{,}474.014 = 2.599,$$

| | | | | | |
|---|---|---|---|---|---|
| | $V_3N_2$ | $V_2N_1$ | $V_1N_4$ | $V_1N_1$ | $V_2N_3$ |
| BLOCK I | $V_3N_0$ | $V_1N_3$ | $V_3N_4$ | $V_1N_2$ | $V_3N_3$ |
| | $V_2N_4$ | $V_3N_1$ | $V_2N_0$ | $V_1N_0$ | $V_2N_2$ |
| | $V_2N_3$ | $V_3N_3$ | $V_1N_1$ | $V_2N_0$ | $V_2N_1$ |
| BLOCK II | $V_1N_3$ | $V_3N_2$ | $V_1N_2$ | $V_1N_4$ | $V_2N_4$ |
| | $V_1N_0$ | $V_3N_4$ | $V_2N_2$ | $V_3N_1$ | $V_3N_0$ |
| | $V_1N_1$ | $V_3N_0$ | $V_1N_0$ | $V_3N_1$ | $V_1N_4$ |
| BLOCK III | $V_2N_2$ | $V_1N_2$ | $V_1N_3$ | $V_2N_4$ | $V_3N_4$ |
| | $V_2N_0$ | $V_3N_2$ | $V_2N_1$ | $V_2N_3$ | $V_3N_3$ |
| | $V_1N_2$ | $V_2N_2$ | $V_2N_4$ | $V_1N_0$ | $V_2N_0$ |
| BLOCK IV | $V_1N_3$ | $V_3N_1$ | $V_1N_4$ | $V_1N_1$ | $V_2N_3$ |
| | $V_3N_0$ | $V_2N_1$ | $V_3N_2$ | $V_3N_3$ | $V_3N_4$ |

**Fig. 4.2** Field layout of the 3 × 5 factorial experiment in a randomized complete block design with four replications.

$$\text{Treatment SS} = \frac{\sum(\text{treatment total})^2}{r} - \text{C.F.}$$

$$= \frac{(12.496)^2 + \ldots + (23.546)^2}{4} - \text{C.F.}$$

$$= 1{,}518.592 - 1{,}474.014 = 44.578,$$

$$\text{Error SS} = \text{total SS} - \text{block SS} - \text{treatment SS}$$

$$= 53.530 - 2.599 - 44.578 = 6.353,$$

$$\text{Variety SS} = \frac{\sum(\text{variety total})^2}{rn} - \text{C.F.}$$

**Table 4.4 The variety × nitrogen table of totals from data in table 4.3.**

| Nitrogen | Yield total (t/ha) | | | |
|---|---|---|---|---|
| | $V_1$ | $V_2$ | $V_3$ | Nitrogen total |
| $N_0$ | 12.496 | 14.192 | 15.112 | 41.800 |
| $N_1$ | 18.894 | 19.224 | 19.014 | 57.132 |
| $N_2$ | 17.838 | 21.744 | 21.280 | 60.862 |
| $N_3$ | 22.868 | 22.958 | 22.210 | 68.036 |
| $N_4$ | 23.292 | 22.722 | 23.546 | 69.560 |
| Variety total | 95.388 | 100.840 | 101.162 | 297.390 |

$$= \frac{(95.388)^2 + (100.840)^2 + (101.162)^2}{(4)(5)} - \text{C.F.}$$

$$= 1{,}475.066 - 1{,}474.014 = 1.052,$$

$$\text{Nitrogen SS} = \frac{\sum(\text{nitrogen total})^2}{rv} - \text{C.F.}$$

$$= \frac{(41.800)^2 + \ldots + (69.560)^2}{(4)(3)} - \text{C.F.}$$

$$= 1{,}515.248 - 1{,}474.014 = 41.234, \quad \text{and}$$

$$\text{Variety} \times \text{nitrogen SS} = \text{treatment SS} - \text{variety SS} - \text{nitrogen SS}$$
$$= 44.578 - 1.052 - 41.234 = 2.292.$$

STEP 5. Calculate mean squares by dividing each SS by its d.f.

STEP 6. Compute observed $F$-values by dividing the mean square for each source of variation by the error mean square.

STEP 7. Look up the corresponding tabulated $F$-values.

STEP 8. Enter all values obtained in steps 4–7 in the analysis of variance. The results are shown in table 4.5.

## 4.4 SPLIT-PLOT DESIGN

A group of designs developed specifically for factorial experiments is the split-plot design. This design's basic feature is the use of two plot sizes, one

**Table 4.5 Analysis of variance on data in table 4.3.**[1]

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Block | 3 | 2.599 | | | | |
| Variety | 2 | 1.052 | 0.526 | 3.48* | 3.22 | 5.15 |
| Nitrogen | 4 | 41.234 | 10.308 | 68.26** | 2.59 | 3.80 |
| Variety × nitrogen | 8 | 2.292 | 0.286 | 1.89ns | 2.17 | 2.96 |
| Error | 42 | 6.353 | 0.151 | | | |
| Total | 59 | 53.530 | | | | |

[1] ** = significant at 1% level; * = significant at 5% level; ns = not significant.

each for the two variables under study. For example, one for variety and one for nitrogen level. The larger plot size is called the mainplot. The number of mainplots in a replication corresponds to the levels of the first factor. Each mainplot is subdivided into smaller size plots, called subplots, such that the number of subplots in each mainplot corresponds to the levels of the second factor. Thus, the relative size of the mainplot and the subplot depends upon the level of the factor assigned to the subplots.

Using two plot sizes results in two levels of precision, one for the mainplot and one for the subplot. The expected precision for the mainplot is lower than that for the subplot. In other words, smaller differences can be detected among subplot treatments than among the mainplot treatments. Consequently, a careful consideration of the relative importance of the factors involved should be made before using the design. For example, do you want more precision in varietal comparisons or nitrogen level comparisons?

The split-plot design is recommended for use when:

1. The practical limit for plot size is much larger in one factor compared with the other. For example, in an experiment to evaluate water managements and population densities, practical considerations necessitate using larger plots for water managements. Water management levels could then be assigned to the mainplot while population densities are allocated to the subplot.
2. Greater precision is desired in one factor relative to the other. For example, in a study to evaluate the performance of varieties at several fertilizer levels, if the factor of primary importance is the variety rather than the fertilizer level, then fertilizer level should be assigned to the mainplot and varieties to the subplot.

### 4.4.1 Randomization and layout

The randomization in a split-plot design is a two-stage one. It involves assigning the levels of the first factor to the mainplot in accordance with any of the basic designs discussed in chapter 3. This is followed by a random assignment of the levels of the second factor to the subplots in each mainplot. In other words, there are two separate randomization processes: one for the mainplots and another for the subplots. For illustration, consider a factorial experiment with six levels of nitrogen as mainplots and four rice varieties as subplots arranged according to a randomized complete block design with three replications.

The steps in the randomization are as follows:

First, divide the experimental area into three blocks of equal size. Each of the three blocks is further subdivided into six mainplots. Starting with the first block, allocate at random the six levels of nitrogen fertilizer ($N_0$, $N_1$, $N_2$, $N_3$, $N_4$, $N_5$) to the mainplots. Repeat the process for the other two blocks independently. Hypothetical results are shown in figure 4.3.

Note that up to this stage, the randomization scheme is merely one of assigning six treatments with three replications in randomized complete block manner.

Next, divide each of the 18 mainplots (six mainplots from each of the three blocks) into four subplots. Assign the four varieties ($V_1$, $V_2$, $V_3$, $V_4$) at random to the four subplots in each mainplot. This process is repeated independently

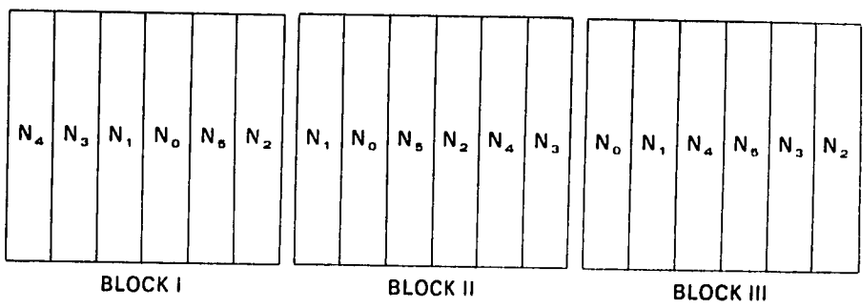


**Fig. 4.3** Random assignment of nitrogen levels to main plots.

18 times since there are 18 mainplots. The layout of this particular experiment is given in figure 4.4.

Note that the randomization procedure assures that all the subplots composing a mainplot are adjoining each other. Thus, in our example, all four varieties in each vertical strip received the same fertilizer level. This strip, then, is considered as only one plot (mainplot), for fertilizer treatment, but is composed of four separate plots (subplots), for varieties.

### 4.4.2 Analysis of variance

For illustration we will use data on grain yield from an experiment involving six levels of nitrogen and four rice varieties conducted in 1969 at Muñoz, Nueva Ecija (table 4.6). A split-plot design was used with the six levels of nitrogen as mainplots and the four rice varieties as subplots. The mainplot treatments were arranged according to the randomized complete block arrangement with three replications. Thus, the layout would be similar to that in figure 4.4.

Let $n$ represent the levels of the mainplot treatment (nitrogen) and $v$ represent the subplot treatment (variety). The steps involved in constructing the analysis of variance of a split-plot design are:

STEP 1. Compute

$$\text{C.F.} = \frac{(\text{grand total})^2}{\text{total number of observations}}$$

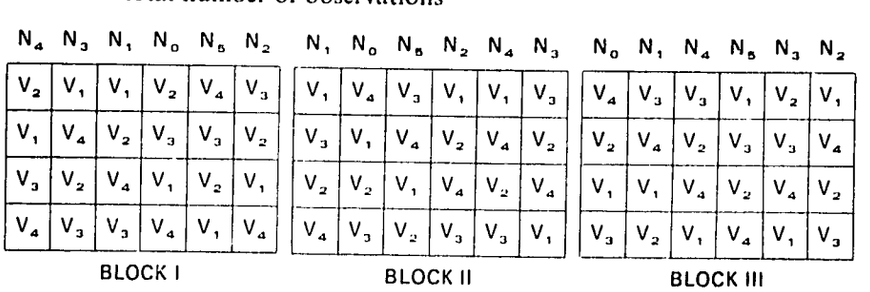


**Fig. 4.4** Hypothetical layout of a split-plot design for testing four rice varieties (subplot) at six nitrogen levels (mainplot) in three replications.

$$= \frac{(394{,}481)^2}{72}$$

$$= 2{,}161{,}323{,}047, \quad \text{and}$$

$$\text{Total SS} = \sum X^2 - \text{C.F.}$$
$$= [(4{,}430)^2 + \ldots + (2{,}014)^2] - 2{,}161{,}323{,}047$$
$$= 204{,}747{,}916.$$

STEP 2. Construct an outline of the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Block | $r - 1 = 2$ | | | | | |
| Nitrogen | $n - 1 = 5$ | | | | | |
| Error ($a$) | $(r - 1)(n - 1) = 10$ | | | | | |
| Variety | $v - 1 = 3$ | | | | | |
| Nitrogen × variety | $(n - 1)(v - 1) = 15$ | | | | | |
| Error ($b$) | $n(r - 1)(v - 1) = 36$ | | | | | |
| Total | $rnv - 1 = 71$ | | | | | |

STEP 3. The computation of sum of squares will be separated into two parts: mainplot and subplot.

(a) *Mainplot analysis.* First, construct the two-way table of mainplot totals (block × nitrogen table of totals) as in table 4.7. Then, compute the following SS's:

$$\text{Block SS} = \frac{\sum B^2}{nv} - \text{C.F.}$$
$$= \frac{(128{,}873)^2 + \ldots + (130{,}004)^2}{(6)(4)} - 2{,}161{,}323{,}047$$
$$= 1{,}082{,}577,$$

$$\text{Nitrogen SS} = \frac{\sum N^2}{rv} - \text{C.F.}$$
$$= \frac{(48{,}670)^2 + \ldots + (69{,}561)^2}{(3)(4)} - 2{,}161{,}323{,}047$$
$$= 30{,}429{,}200, \quad \text{and}$$

$$\text{Error (a) SS} = \frac{\sum (NB)^2}{v} - \text{C.F.} - \text{block SS} - \text{nitrogen SS}$$
$$= \frac{(15{,}964)^2 + \ldots + (22{,}318)^2}{(4)} - 2{,}161{,}323{,}047 -$$

**Table 4.6 Grain yield data for four rice varieties grown under six levels of nitrogen in three replications.**

| Variety | Grain yield (kg/ha) | | |
|---|---|---|---|
| | Rep. I | Rep. II | Rep. III |
| | | $N_1$ (*0 kg N/ha*) | |
| IR8 ($V_1$) | 4,430 | 4,478 | 3,850 |
| IR5 ($V_2$) | 3,944 | 5,314 | 3,660 |
| C4-63 ($V_3$) | 3,464 | 2,944 | 3,142 |
| Peta ($V_4$) | 4,126 | 4,482 | 4,836 |
| | | $N_2$ (*60 kg N/ha*) | |
| IR8 | 5,418 | 5,166 | 6,432 |
| IR5 | 6,502 | 5,858 | 5,586 |
| C4-63 | 4,768 | 6,004 | 5,556 |
| Peta | 5,192 | 4,604 | 4,652 |
| | | $N_3$ (*90 kg N/ha*) | |
| IR8 | 6,076 | 6,420 | 6,704 |
| IR5 | 6,008 | 6,127 | 6,542 |
| C4-63 | 6,244 | 5,724 | 6,014 |
| Peta | 4,546 | 5,744 | 4,146 |
| | | $N_4$ (*120 kg N/ha*) | |
| IR8 | 6,462 | 7,056 | 6,680 |
| IR5 | 7,139 | 6,982 | 6,564 |
| C4-63 | 5,792 | 5,880 | 6,370 |
| Peta | 2,774 | 5,036 | 3,638 |
| | | $N_5$ (*150 kg N/ha*) | |
| IR8 | 7,290 | 7,848 | 7,552 |
| IR5 | 7,682 | 6,594 | 6,576 |
| C4-63 | 7,080 | 6,662 | 6,320 |
| Peta | 1,414 | 1,960 | 2,722 |
| | | $N_6$ (*180 kg N/ha*) | |
| IR8 | 8,452 | 8,832 | 8,818 |
| IR5 | 6,228 | 7,387 | 6,006 |
| C4-63 | 5,594 | 7,122 | 5,480 |
| Peta | 2,248 | 1,380 | 2,014 |

$$1{,}082{,}577 - 30{,}429{,}200$$
$$= 1{,}419{,}678.$$

Note that this analysis is exactly the same as that of a randomized complete block with three replications and six treatments.

(b) *Subplot analysis.* Construct the two-way table of treatment totals (mainplot × subplot), as shown in table 4.8. Then, compute the following SS's:

$$\text{Variety SS} = \frac{\sum V^2}{ru} - \text{C.F.}$$
$$= \frac{(117{,}964)^2 + \ldots + (65{,}558)^2}{(3)(6)} - 2{,}161{,}323{,}047$$
$$= 89{,}888{,}101,$$

$$\text{Variety} \times \text{nitrogen SS} = \frac{\sum (VN)^2}{r} - \text{C.F.} - \text{variety SS} - \text{nitrogen SS}$$

**Table 4.7 The block × nitrogen table of yield totals computed from the data of table 4.6.**

| Nitrogen | Yield total (kg/ha) | | | |
|---|---|---|---|---|
| | Block I | Block II | Block III | N total |
| $N_1$ | 15,964 | 17,218 | 15,488 | 48,670 ($N_1$) |
| $N_2$ | 21,880 | 21,632 | 22,226 | 65,738 ($N_2$) |
| $N_3$ | 22,874 | 24,015 | 23,506 | 70,395 ($N_3$) |
| $N_4$ | 22,167 | 24,954 | 23,252 | 70,373 ($N_4$) |
| $N_5$ | 23,466 | 23,064 | 23,214 | 69,744 ($N_5$) |
| $N_6$ | 22,522 | 24,721 | 22,318 | 69,561 ($N_6$) |
| Block total | 128,873 | 135,604 | 130,004 | |
| Grand total | | | | 394,481 |

**Table 4.8 The nitrogen × variety table of yield totals computed from the data of table 4.6.**

| Nitrogen | Yield total (kg/ha) | | | | |
|---|---|---|---|---|---|
| | $V_1$ | $V_2$ | $V_3$ | $V_4$ | N total |
| $N_1$ | 12,758 | 12,918 | 9,550 | 13,444 | 48,670 ($N_1$) |
| $N_2$ | 17,016 | 17,946 | 16,328 | 14,448 | 65,738 ($N_2$) |
| $N_3$ | 19,200 | 18,777 | 17,982 | 14,436 | 70,395 ($N_3$) |
| $N_4$ | 20,198 | 20,685 | 18,042 | 11,448 | 70,373 ($N_4$) |
| $N_5$ | 22,690 | 20,852 | 20,062 | 6,140 | 69,744 ($N_5$) |
| $N_6$ | 26,102 | 19,621 | 18,196 | 5,642 | 69,561 ($N_6$) |
| Variety total | 17,964 | 110,799 | 100,160 | 65,558 | |
| Grand total | | | | | 394,481 |

$$= \frac{(12{,}758)^2 + \ldots + (5{,}642)^2}{3} - 2{,}161{,}323{,}047$$

$$- 89{,}888{,}101 - 30{,}429{,}200$$

$$= 69{,}343{,}487, \quad \text{and}$$

$$\begin{aligned}\text{Error (b) SS} &= \text{total SS} - \text{the sum of all other SS's}\\ &= 204{,}747{,}916 - (1{,}082{,}577 + 30{,}429{,}200 + 1{,}419{,}678 + \\ &\quad 89{,}888{,}101 + 69{,}343{,}487)\\ &= 12{,}584{,}873.\end{aligned}$$

STEP 4. Compute mean square for each source of variation by dividing the sum of squares by its corresponding degrees of freedom as follows:

$$\text{Nitrogen MS} = \frac{30{,}429{,}200}{5} = 6{,}085{,}840,$$

$$\text{Error (a) MS} = \frac{1{,}419{,}678}{10} = 141{,}968,$$

$$\text{Variety MS} = \frac{89{,}888{,}101}{3} = 29{,}962{,}700,$$

$$\text{Nitrogen} \times \text{variety MS} = \frac{69{,}343{,}487}{15} = 4{,}622{,}899, \quad \text{and}$$

$$\text{Error (b) MS} = \frac{12{,}584{,}873}{36} = 349{,}580.$$

STEP 5. Compute the observed $F$-values. The block and mainplot treatment (in this instance, nitrogen) effects are tested using error (a), while the subplot treatment (variety) and the interaction effects are tested with error (b). Thus,

$$F \text{ for nitrogen} = \frac{6{,}085{,}840}{141{,}968} = 42.87,$$

$$F \text{ for variety} = \frac{29{,}962{,}700}{349{,}580} = 85.71, \quad \text{and}$$

$$F \text{ for interaction} = \frac{4{,}622{,}899}{349{,}580} = 13.22.$$

STEP 6. Compute the coefficients of variation. Since the split-plot analysis has two error terms, there are also two coefficients of variation to be computed, one for the mainplot analysis, cv(a), and another for the subplot analysis, cv(b).

$$\text{cv(a)} = \frac{\sqrt{\text{error (a) MS}}}{\text{grand mean}} \times 100 = \frac{\sqrt{141{,}968}}{5{,}479} \times 100 = 6.9\%, \quad \text{and}$$

$$\text{cv(b)} = \frac{\sqrt{\text{error (b) MS}}}{\text{grand mean}} \times 100 = \frac{\sqrt{349{,}580}}{5{,}479} \times 100 = 10.8\%.$$

The cv(a) indicates the degree of precision attached to the measurement of the mainplot treatment effects and the cv(b) the precision of the subplot treatment and interaction effects. Usually we would expect cv(b) to be smaller than cv(a) since, as indicated earlier, the factor assigned to the mainplot is measured at relatively lower precision than those assigned to the subplot. This trend does not always hold, however, as shown by the present example. The cause for such an unexpected outcome is beyond the scope of this book. You should consult a competent statistician if such results frequently occur in your experiments.

STEP 7. Enter all values obtained from steps 1 to 6 in the analysis of variance table. The completed result is given in table 4.9. Compare the observed $F$-values with their corresponding tabular values and indicate its significance by appropriate asterisk (see section 3.2). In this instance, all the three effects ($N$, $V$, and $N \times V$) are highly significant. When the interaction is significant, the interpretation of the results becomes complicated and procedures given in section 6.5 should be followed.

## 4.5 STRIP-PLOT DESIGN

When the levels of the second factor are laid out in strips, instead of being randomized independently within each mainplot, as in the split-plot design, the resulting design is referred to as strip-plot design. This design is appropriate

**Table 4.9 Analysis of variance for grain yield data of table 4.6.**[1]

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Computed *F* | Tabular *F* 5% | Tabular *F* 1% |
|---|---|---|---|---|---|---|
| Block | 2 | 1,082,577 | 541,228 | | | |
| Nitrogen (N) | 5 | 30,429,200 | 6,085,840 | 42.87** | 3.33 | 5.64 |
| Error (a) | 10 | 1,419,678 | 141,968 | | | |
| Variety (V) | 3 | 89,888,101 | 29,962,700 | 85.71** | 2.86 | 4.38 |
| N × V | 15 | 69,343,487 | 4,622,899 | 13.22** | 1.96 | 2.58 |
| Error (b) | 36 | 12,584,873 | 349,580 | | | |
| Total | 71 | 204,747,916 | | | | |

[1]cv(a) = 6.9%; cv(b) = 10.8%; ** = significant at the 1% level.

for experiments where *both* factors need relatively large plots, and it is accomplished by dividing the block first into horizontal strips for one factor and then dividing the block into vertical strips for the second factor.

The strip-plot design sacrifices precision on the main effects of both factors. On the other hand, the interaction is measured more accurately than is possible with either a randomized complete block or a split-plot design. Thus, the strip-plot design is appropriate for experiments where (1) practical consideration necessitates relatively large plots for both factors, or (2) the interaction of the two factors is the principal object of study.

### 4.5.1 Randomization and layout

First, one factor is randomly allocated to the horizontal strips and then the second factor is randomly allocated to the vertical strips.

Consider an experiment involving six rice varieties and three nitrogen levels using a strip-plot design with three replications. After dividing the area into three blocks, the randomization scheme starts by assigning the six varieties to horizontal strips. The result is shown in figure 4.5.

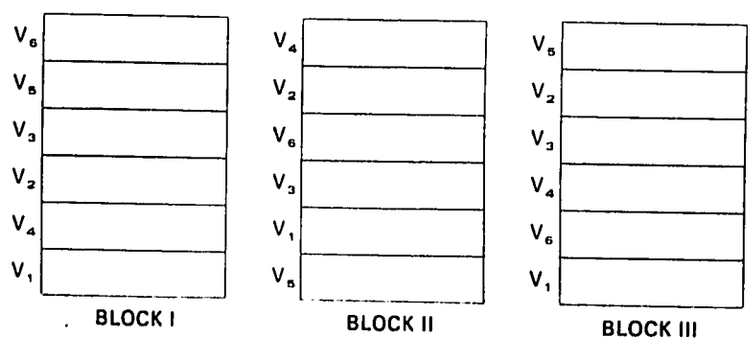


**Fig. 4.5** Random assignment of six varieties to the horizontal strips in strip-plot design.

Then proceed to allocate randomly the three nitrogen levels to vertical strips. The final layout is shown in figure 4.6.
Note that you can assign either the vertical or the horizontal strips first.

### 4.5.2 Analysis of variance

Data from the experiment whose layout is described in the previous section (table 4.10) will be used to illustrate the analysis of variance. Using $v$ as the number of varieties and $n$ as the levels of nitrogen, the steps in computing the analysis of variance are:

STEP 1. Construct an outline of the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Block | $r - 1 = 2$ | | | | | |
| Variety | $v - 1 = 5$ | | | | | |
| Error (a) | $(r - 1)(v - 1) = 10$ | | | | | |
| Nitrogen | $n - 1 = 2$ | | | | | |
| Error (b) | $(r - 1)(n - 1) = 4$ | | | | | |
| Variety × nitrogen | $(v - 1)(n - 1) = 10$ | | | | | |
| Error (c) | $(r - 1)(n - 1)(v - 1) = 20$ | | | | | |
| Total | $rnv - 1 = 53$ | | | | | |

STEP 2. Compute the sums of squares. As usual, we start out with computing

$$\text{C.F.} = \frac{(\text{grand total})^2}{\text{total number of observations}}$$

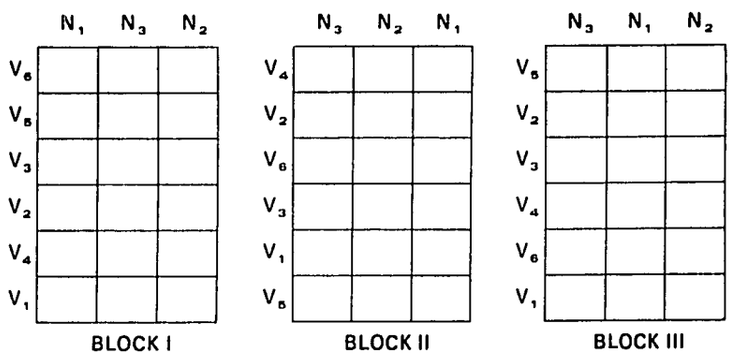


**Fig. 4.6** A hypothetical layout of a strip-plot design with six varieties and three nitrogen levels in three replications.

**Table 4.10 Data on grain yields of six rice varieties grown under three nitrogen levels and under broadcast condition. 1969 dry season.**

| Nitrogen level (kg/ha) | Yield total (kg/ha) | | |
|---|---|---|---|
| | Block I | Block II | Block III |
| | | *IR8* ($V_1$) | |
| 0 ($N_1$) | 2,373 | 3,958 | 4,384 |
| 60 ($N_2$) | 4,076 | 6,431 | 4,889 |
| 120 ($N_3$) | 7,254 | 6,808 | 8,582 |
| | | *IR127-80* ($V_2$) | |
| 0 ($N_1$) | 4,007 | 5,795 | 5,001 |
| 60 ($N_2$) | 5,630 | 7,334 | 7,177 |
| 120 ($N_3$) | 7,053 | 8,284 | 6,297 |
| | | *IR305-4-12* ($V_3$) | |
| 0 ($N_1$) | 2,620 | 4,508 | 5,621 |
| 60 ($N_2$) | 4,676 | 6,672 | 7,019 |
| 120 ($N_3$) | 7,666 | 7,328 | 8,611 |
| | | *IR400-2-5* ($V_4$) | |
| 0 ($N_1$) | 2,726 | 5,630 | 3,821 |
| 60 ($N_2$) | 4,838 | 7,007 | 4,816 |
| 120 ($N_3$) | 6,881 | 7,735 | 6,667 |
| | | *IR665-58* ($V_5$) | |
| 0 ($N_1$) | 4,447 | 3,276 | 4,582 |
| 60 ($N_2$) | 5,549 | 5,340 | 6,011 |
| 120 ($N_3$) | 6,880 | 5,080 | 6,076 |
| | | *Peta* ($V_6$) | |
| 0 ($N_1$) | 2,572 | 3,724 | 3,326 |
| 60 ($N_2$) | 3,896 | 2,822 | 4,425 |
| 120 ($N_3$) | 1,556 | 2,706 | 3,214 |

$$= \frac{(285{,}657)^2}{54}$$

$$= 1{,}511{,}109{,}660\text{, and}$$

$$\text{Total SS} = \sum X^2 - \text{C.F.}$$

$$= [(2{,}373)^2 + \ldots + (3{,}214)^2] - 1{,}511{,}109{,}660$$

$$= 167{,}005{,}649\,.$$

The analysis of variance of a strip-plot design has three parts, the first part for factor $A$, the second for factor $B$, and the third for the interaction between factors $A$ and $B$. The computation of the sums of squares will be made separately for each part as follows:

(a) *Factor A analysis.* Construct the block × variety table of total, as in table 4.11. Then, compute

$$\text{Block SS} = \frac{\sum B^2}{vn} - \text{C.F.}$$

$$= \frac{(84{,}700)^2 + (100{,}438)^2 + (100{,}519)^2}{(6)(3)} - 1{,}511{,}109{,}660$$

$$= 9{,}220{,}962\,.$$

**Table 4.11 The block × variety table of yield totals from data of table 4.10.**

| Variety | Yield total (kg/ha) | | | |
|---|---|---|---|---|
| | Block I | Block II | Block III | Total |
| $V_1$ | 13,703 | 17,197 | 17,855 | 48,755 ($V_1$) |
| $V_2$ | 16,690 | 21,413 | 18,475 | 56,578 ($V_2$) |
| $V_3$ | 14,962 | 18,508 | 21,251 | 54,721 ($V_3$) |
| $V_4$ | 14,445 | 20,372 | 15,304 | 50,121 ($V_4$) |
| $V_5$ | 16,876 | 13,696 | 16,669 | 47,241 ($V_5$) |
| $V_6$ | 8,024 | 9,252 | 10,965 | 28,241 ($V_6$) |
| Block total | 84,700 | 100,438 | 100,519 | |

$$\text{Variety SS} = \frac{\sum V^2}{rn} - \text{C.F.}$$

$$= \frac{(48{,}755)^2 + \ldots + (28{,}241)^2}{(3)(3)} - 1{,}511{,}109{,}660$$

$$= 57{,}100{,}201\,, \quad \text{and}$$

$$\text{Error (a) SS} = \frac{\sum (BV)^2}{n} - \text{C.F.} - \text{block SS} - \text{variety SS}$$

$$= \frac{(13{,}703)^2 + \ldots + (10{,}965)^2}{3} - 1{,}511{,}109{,}660$$

$$- 9{,}220{,}962 - 57{,}100{,}201$$

$$= 14{,}922{,}620\,.$$

(b) *Factor B analysis.* Construct the block × nitrogen table of yield totals as in table 4.12. Then, compute

$$\text{Nitrogen SS} = \frac{\sum N^2}{rv} - \text{C.F.}$$

$$= \frac{(72{,}371)^2 + (98{,}608)^2 + (114{,}678)^2}{(3)(6)} - 1{,}511{,}109{,}660$$

$$= 50{,}676{,}061\,, \quad \text{and}$$

$$\text{Error (b) SS} = \frac{\sum (BN)^2}{v} - \text{C.F.} - \text{block SS} - \text{nitrogen SS}$$

**Table 4.12 The block × nitrogen table of yield totals computed from data of table 4.10.**

| Nitrogen | Yield total (kg/ha) | | | |
|---|---|---|---|---|
| | Block I | Block II | Block III | N total |
| $N_1$ | 18,745 | 26,891 | 26,735 | 72,371 ($N_1$) |
| $N_2$ | 28,665 | 35,606 | 34,337 | 98,608 ($N_2$) |
| $N_3$ | 37,290 | 37,941 | 39,447 | 114,678 ($N_3$) |
| Block total | 84,700 | 100,438 | 100,519 | |
| Grand total | | | | 285,657 |

**Table 4.13 The variety × nitrogen table of yield totals computed from data in table 4.10.**

| Variety | Yield total (kg/ha) | | |
|---|---|---|---|
| | $N_1$ | $N_2$ | $N_3$ |
| $V_1$ | 10,715 | 15,396 | 22,644 |
| $V_2$ | 14,803 | 20,141 | 21,634 |
| $V_3$ | 12,749 | 18,367 | 23,605 |
| $V_4$ | 12,177 | 16,661 | 21,283 |
| $V_5$ | 12,305 | 16,900 | 18,036 |
| $V_6$ | 9,622 | 11,143 | 7,476 |

$$= \frac{(18,745)^2 + \ldots + (39,447)^2}{6} - 1,511,109,660$$

$$- 9,220,962 - 50,676,061$$

$$= 2,974,909.$$

(c) *Factor C analysis.* Construct the variety × nitrogen table of yield totals as in table 4.13. Then, compute

$$\text{Variety} \times \text{nitrogen SS} = \frac{\Sigma(VN)^2}{r} - \text{C.F.} - \text{variety SS} - \text{nitrogen SS}$$

$$= \frac{(10,715)^2 + \ldots + (7,476)^2}{3} - 1,511,109,660$$

$$- 57,100,201 - 50,676,061$$

$$= 23,877,980, \quad \text{and}$$

$$\begin{aligned}\text{Error (c) SS} &= \text{total SS} - \text{the sum of all other SS's}\\ &= 167,005,649 - (9,220,962 + 57,100,201 + 14,922,620\\ &\quad + 50,676,061 + 2,974,909 + 23,877,980)\\ &= 8,232,916.\end{aligned}$$

STEP 3. Compute the mean square for each source of variation by dividing each SS by its degrees of freedom:

$$\text{Variety MS} = \frac{57,100,201}{5} = 11,420,040,$$

$$\text{Error (a) MS} = \frac{14,922,620}{10} = 1,492,262,$$

$$\text{Nitrogen MS} = \frac{50,676,061}{2} = 25,338,031,$$

$$\text{Error (b) MS} = \frac{2,974,909}{4} = 743,727,$$

$$\text{Variety} \times \text{nitrogen MS} = \frac{23,877,980}{10} = 2,387,798, \quad \text{and}$$

$$\text{Error (c) MS} = \frac{8{,}232{,}916}{20} = 411{,}646.$$

STEP 4. Compute the observed $F$-values as follows:

$$F \text{ for variety} = \frac{\text{variety MS}}{\text{error (a) MS}} = \frac{11{,}420{,}040}{1{,}492{,}262} = 7.65,$$

$$F \text{ for nitrogen} = \frac{\text{nitrogen MS}}{\text{error (b) MS}} = \frac{25{,}338{,}031}{743{,}727} = 34.07, \quad \text{and}$$

$$F \text{ for interaction} = \frac{(V \times N)\text{ MS}}{\text{error (c) MS}} = \frac{2{,}387{,}798}{411{,}646} = 5.80.$$

STEP 5. Compute the coefficients of variation as

$$\text{cv(a)} = \frac{\sqrt{\text{error (a) MS}}}{\text{grand mean}} \times 100$$

$$= \frac{\sqrt{1{,}492{,}262}}{5{,}290} \times 100 = 23.1\%,$$

$$\text{cv (b)} = \frac{\sqrt{\text{error (b) MS}}}{\text{grand mean}} \times 100$$

$$= \frac{\sqrt{743{,}727}}{5{,}290} \times 100 = 16.3\%, \quad \text{and}$$

$$\text{cv (c)} = \frac{\sqrt{\text{error (c) MS}}}{\text{grand mean}} \times 100$$

$$= \frac{\sqrt{411{,}646}}{5{,}290} \times 100 = 12.1\%.$$

The cv(a) indicates the degree of precision attached to the measurement of varietal effects, while cv(b) refers to nitrogen effects, and cv(c) to interaction effects. In this instance, cv(c) is the smallest, indicating that the measurement of interaction effects has the highest precision. This trend is normally expected.

**Table 4.14 Analysis of variance for grain yield data of table 4.10.[1]**

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Block | 2 | 9,220,962 | 4,610,481 | | | |
| Variety (V) | 5 | 57,100,201 | 11,420,040 | 7.65** | 3.33 | 5.64 |
| Error (a) | 10 | 14,922,620 | 1,492,262 | | | |
| Nitrogen (N) | 2 | 50,676,061 | 25,338,031 | 34.07** | 6.94 | 18.00 |
| Error (b) | 4 | 2,974,909 | 743,727 | | | |
| V × N | 10 | 23,877,980 | 2,387,798 | 5.80** | 2.35 | 3.37 |
| Error (c) | 20 | 8,232,916 | 411,646 | | | |
| Total | 53 | 167,005,649 | | | | |

[1]cv(a) = 23.1%; cv(b) = 16.3%; cv(c) = 12.1%; ** = significant at the 1% level.

Between cv(a) and cv (b), however, there is no basis to expect one to be greater or smaller than the other.

STEP 6. Enter all values so far computed in the analysis of variance table, as shown in table 4.14. Compare the observed $F$-values with their corresponding tabular values and designate the significant results with the appropriate asterisk (see section 3.2). In this instance, all the three tests show highly significant results. With a significant interaction, caution must be exercised when interpreting the results. See section 6.5. for further details.

CHAPTER 5

# Three-or-more-factor experiments

## 5.1 INTRODUCTION

In the same manner that a single-factor experiment can be expanded to include a second factor, a two-factor experiment can be expanded to include a third, a three-factor experiment to include a fourth, and so on.

The total number of treatments, however, is the product of the levels in each factor so that the size of the experiment expands very quickly with the number of factors. For example, a two-factor experiment with three levels of each factor will have nine treatments. Adding an additional factor with three levels will increase the total number of treatments to 27; adding still another factor, again with three levels, will increase the number of treatments to 81. Thus, the primary limitation to the number of factors that can be included is the size of experiment.

Any of the designs described in chapter 3 for a single-factor experiments and in chapter 4 for two-factor experiments can be used for experiments with three or more factors. For example, a three-factor experiment involving two varieties, four nitrogen levels, and three weed control methods can be put in either a randomized complete block or a split-plot design, as shown in figures 5.1 and 5.2. Obviously, the same experiment can be fitted into any of the other designs previously described. The choice of which alternative to use depends upon the objectives of the experiment and the specific advantages of each design, as described in the previous chapters. In our present example, for instance, the degree of precision for the measurement of the effects of the three factors is equal in the randomized complete block while nitrogen response, which is allocated to the subplots, will be measured with higher precision in the split-plot design.

In this chapter, two other designs that can only be used for three-factor experiments will be described. These are the split-split-plot design and the strip-split-plot design.

## 5.2 SPLIT-SPLIT-PLOT DESIGN

The split-split-plot design is an extension of the split-plot design discussed in section 4.4. To accommodate a third factor, each subplot in the basic split-plot design is further divided into sub-subplots, and the randomization of this

REPLICATION I

| | | | | | |
|---|---|---|---|---|---|
| $V_2$ $N_0$ $W_3$ | $V_2$ $N_3$ $W_1$ | $V_1$ $N_3$ $W_2$ | $V_1$ $N_2$ $W_1$ | $V_1$ $N_3$ $W_3$ | $V_2$ $N_2$ $W_3$ |
| $V_1$ $N_1$ $W_1$ | $V_2$ $N_1$ $W_1$ | $V_1$ $N_0$ $W_3$ | $V_2$ $N_0$ $W_2$ | $V_1$ $N_0$ $W_1$ | $V_2$ $N_2$ $W_1$ |
| $V_1$ $N_1$ $W_2$ | $V_1$ $N_0$ $W_2$ | $V_1$ $N_1$ $W_3$ | $V_1$ $N_2$ $W_1$ | $V_2$ $N_3$ $W_3$ | $V_2$ $N_1$ $W_1$ |
| $V_1$ $N_2$ $W_2$ | $V_1$ $N_3$ $W_1$ | $V_2$ $N_3$ $W_2$ | $V_2$ $N_2$ $W_2$ | $V_2$ $N_0$ $W_1$ | $V_2$ $N_1$ $W_2$ |

REPLICATION II

| | | | | | |
|---|---|---|---|---|---|
| $V_1$ $N_3$ $W_1$ | $V_1$ $N_2$ $W_1$ | $V_2$ $N_0$ $W_2$ | $V_2$ $N_2$ $W_3$ | $V_1$ $N_0$ $W_2$ | $V_1$ $N_3$ $W_2$ |
| $V_2$ $N_1$ $W_1$ | $V_2$ $N_3$ $W_3$ | $V_2$ $N_1$ $W_2$ | $V_1$ $N_0$ $W_3$ | $V_1$ $N_2$ $W_3$ | $V_2$ $N_2$ $W_1$ |
| $V_2$ $N_3$ $W_2$ | $V_2$ $N_1$ $W_3$ | $V_2$ $N_0$ $W_1$ | $V_1$ $N_1$ $W_1$ | $V_1$ $N_1$ $W_2$ | $V_1$ $N_1$ $W_1$ |
| $V_1$ $N_0$ $W_1$ | $V_2$ $N_0$ $W_1$ | $V_1$ $N_3$ $W_1$ | $V_2$ $N_3$ $W_1$ | $V_1$ $N_2$ $W_2$ | $V_2$ $N_2$ $W_2$ |

REPLICATION III

| | | | | | |
|---|---|---|---|---|---|
| $V_1$ $N_2$ $W_1$ | $V_2$ $N_3$ $W_1$ | $V_1$ $N_1$ $W_1$ | $V_2$ $N_3$ $W_3$ | $V_1$ $N_1$ $W_2$ | $V_2$ $N_2$ $W_3$ |
| $V_2$ $N_3$ $W_2$ | $V_1$ $N_0$ $W_2$ | $V_2$ $N_2$ $W_2$ | $V_2$ $N_1$ $W_3$ | $V_2$ $N_0$ $W_2$ | $V_2$ $N_0$ $W_3$ |
| $V_2$ $N_0$ $W_1$ | $V_1$ $N_1$ $W_3$ | $V_2$ $N_2$ $W_1$ | $V_1$ $N_2$ $W_2$ | $V_1$ $N_0$ $W_3$ | $V_1$ $N_3$ $W_3$ |
| $V_2$ $N_1$ $W_1$ | $V_1$ $N_3$ $W_2$ | $V_1$ $N_3$ $W_1$ | $V_1$ $N_2$ $W_3$ | $V_2$ $N_1$ $W_2$ | $V_1$ $N_0$ $W_1$ |

**Fig. 5.1** A layout for an experiment involving two varieties, four nitrogen levels, and three weed control methods, arranged in a randomized complete block design with three replications.

| REPLICATION I | | | | | |
|---|---|---|---|---|---|
| $V_1W_2$ | $V_2W_1$ | $V_1W_3$ | $V_2W_3$ | $V_1W_1$ | $V_2W_2$ |
| $N_2$ | $N_3$ | $N_1$ | $N_2$ | $N_3$ | $N_1$ |
| $N_0$ | $N_2$ | $N_0$ | $N_3$ | $N_0$ | $N_2$ |
| $N_3$ | $N_1$ | $N_2$ | $N_0$ | $N_2$ | $N_0$ |
| $N_1$ | $N_0$ | $N_3$ | $N_1$ | $N_1$ | $N_3$ |

| REPLICATION II | | | | | |
|---|---|---|---|---|---|
| $V_1W_2$ | $V_2W_2$ | $V_2W_3$ | $V_1W_1$ | $V_1W_3$ | $V_2W_1$ |
| $N_2$ | $N_3$ | $N_0$ | $N_0$ | $N_2$ | $N_3$ |
| $N_3$ | $N_1$ | $N_1$ | $N_2$ | $N_1$ | $N_1$ |
| $N_1$ | $N_0$ | $N_3$ | $N_1$ | $N_3$ | $N_0$ |
| $N_0$ | $N_2$ | $N_2$ | $N_3$ | $N_0$ | $N_2$ |

| REPLICATION III | | | | | |
|---|---|---|---|---|---|
| $V_2W_1$ | $V_2W_2$ | $V_1W_2$ | $V_1W_1$ | $V_2W_3$ | $V_1W_3$ |
| $N_2$ | $N_3$ | $N_0$ | $N_1$ | $N_3$ | $N_2$ |
| $N_3$ | $N_1$ | $N_3$ | $N_2$ | $N_1$ | $N_0$ |
| $N_0$ | $N_2$ | $N_1$ | $N_3$ | $N_0$ | $N_1$ |
| $N_1$ | $N_0$ | $N_2$ | $N_0$ | $N_2$ | $N_3$ |

**Fig. 5.2** A layout for an experiment involving two varieties, four nitrogen levels, and three weed control methods, arranged in a split-plot design with nitrogen levels as subplot treatments, in three replications.

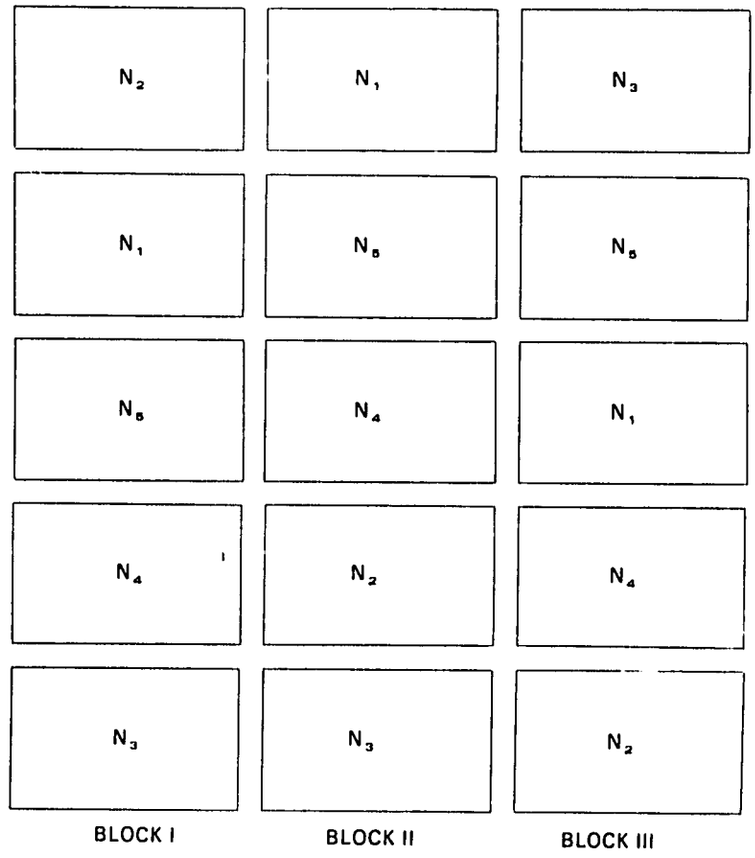


**Fig. 5.3** Random assignment of five nitrogen levels to the mainplots as the first step in a split-split-plot layout.

last factor is made separately for each subplot. Theoretically, subdivision could continue. That is, each sub-subplot in a split-split-plot design can be divided further into sub-sub-subplots and levels of a fourth factor randomly allocated. Practical considerations in terms of total area as well as size of the smallest plot may, however, limit the number of factors that can be included in an experiment.

The advantages and limitations of a split-split-plot design, or any of its extensions, are similar to that of a split-plot design. The precision of the comparisons among the sub-subplot treatments is the best, followed by those among the subplot treatments, with those among the mainplot treatments having the least precision. Therefore, *the experimenter should assign the most important factor to the smallest plot and not to the largest.*

### 5.2.1 Randomization and layout

For illustration, we shall use a 5 × 3 × 3 factorial experiment with three replications in which the five nitrogen levels are the mainplot, the three manage-

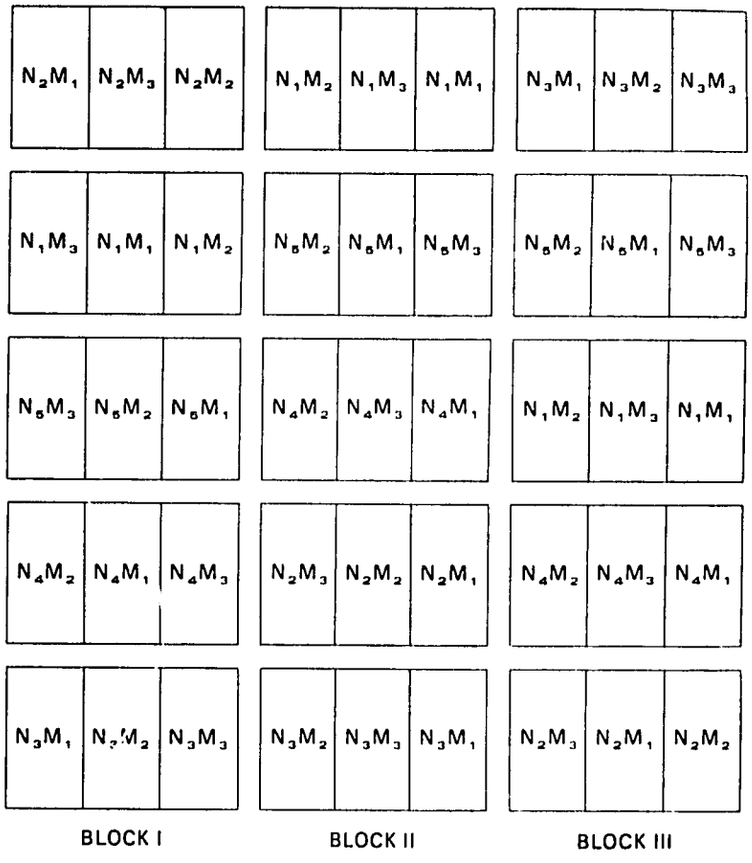


**Fig. 5.4** The layout after the five mainplot treatments ($N_1$, $N_2$, $N_3$, $N_4$, $N_5$) and the three subplot treatments ($M_1$, $M_2$, $M_3$) were assigned.

ment practices the subplots, and the three rice varieties the sub-subplot treatments. The randomization steps are:

STEP 1. Divide the area into three blocks and each block into five equal units to correspond to the five mainplots. Randomly assign the five nitrogen levels ($N_1$, $N_2$, $N_3$, $N_4$, and $N_5$) to the five mainplots separately and independently for each of the three blocks following a randomized complete block design (see section 3.3.2). The results are shown in figure 5.3.

STEP 2. Divide each mainplot into three subplots and randomly assign three management practices ($M_1$, $M_2$, and $M_3$). The randomization process is repeated independently for all the 15 main plots. The result is shown in figure 5.4.

STEP 3. Divide each subplot into three sub-subplots, each of which is randomly assigned to one of the three varieties ($V_1$, $V_2$, and $V_3$). This process is repeated for all the 45 subplots independently. The final layout is shown in figure 5.5.

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| $N_2M_1V_1$ | $N_2M_3V_3$ | $N_2M_2V_3$ | $N_1M_2V_1$ | $N_1M_3V_1$ | $N_1M_1V_2$ | $N_3M_1V_2$ | $N_3M_2V_2$ | $N_3$ |
| $N_2M_1V_3$ | $N_2M_3V_1$ | $N_2M_2V_2$ | $N_1M_3V_3$ | $N_1M_3V_2$ | $N_1M_1V_1$ | $N_3M_1V_3$ | $N_3M_3V_3$ | $N_3$ |
| $N_2M_1V_2$ | $N_2M_3V_2$ | $N_2M_2V_1$ | $N_1M_2V_3$ | $N_1M_3V_3$ | $N_1M_1V_3$ | $N_3M_1V_1$ | $N_3M_2V_1$ | $N_3$ |
| $N_1M_3V_2$ | $N_1M_1V_1$ | $N_1M_2V_1$ | $N_5M_2V_1$ | $N_5M_1V_2$ | $N_5M_3V_1$ | $N_5M_2V_1$ | $N_5M_1V_1$ | $N_5$ |
| $N_1M_3V_1$ | $N_1M_1V_3$ | $N_1M_2V_2$ | $N_5M_2V_3$ | $N_5M_1V_3$ | $N_5M_3V_2$ | $N_5M_2V_3$ | $N_5M_1V_3$ | $N_5$ |
| $N_1M_3V_3$ | $N_1M_1V_2$ | $N_1M_2V_3$ | $N_5M_2V_2$ | $N_5M_1V_1$ | $N_5M_3V_3$ | $N_5M_2V_2$ | $N_5M_1V_2$ | $N_5$ |
| $N_5M_3V_1$ | $N_5M_2V_1$ | $N_5M_1V_3$ | $N_4M_2V_2$ | $N_4M_3V_3$ | $N_4M_1V_3$ | $N_1M_2V_2$ | $N_1M_3V_1$ | $N_1$ |
| $N_5M_3V_3$ | $N_5M_2V_2$ | $N_5M_1V_2$ | $N_4M_2V_1$ | $N_4M_3V_1$ | $N_4M_1V_1$ | $N_1M_2V_3$ | $N_1M_3V_2$ | $N_1$ |
| $N_5M_3V_2$ | $N_5M_2V_3$ | $N_5M_1V_1$ | $N_4M_2V_3$ | $N_4M_3V_2$ | $N_4M_1V_2$ | $N_1M_2V_1$ | $N_1M_3V_3$ | $N_1$ |
| $N_4M_2V_1$ | $N_4M_1V_1$ | $N_4M_3V_3$ | $N_2M_3V_2$ | $N_2M_2V_2$ | $N_2M_1V_1$ | $N_4M_2V_2$ | $N_4M_3V_3$ | $N_4$ |
| $N_4M_2V_2$ | $N_4M_1V_3$ | $N_4M_3V_1$ | $N_2M_3V_3$ | $N_2M_2V_3$ | $N_2M_1V_2$ | $N_4M_2V_1$ | $N_4M_3V_2$ | $N_4$ |
| $N_4M_2V_3$ | $N_4M_1V_2$ | $N_4M_3V_2$ | $N_2M_3V_1$ | $N_2M_2V_1$ | $N_2M_1V_3$ | $N_4M_2V_3$ | $N_4M_3V_1$ | $N_4$ |
| $N_3M_1V_3$ | $N_3M_2V_1$ | $N_3M_3V_3$ | $N_3M_2V_3$ | $N_3M_3V_3$ | $N_3M_1V_1$ | $N_2M_3V_3$ | $N_2M_1V_3$ | $N_2$ |
| $N_3M_1V_1$ | $N_3M_2V_3$ | $N_3M_3V_1$ | $N_3M_2V_2$ | $N_3M_3V_1$ | $N_3M_1V_2$ | $N_2M_3V_2$ | $N_2M_1V_1$ | $N_2$ |
| $N_3M_1V_2$ | $N_3M_2V_2$ | $N_3M_3V_2$ | $N_3M_2V_1$ | $N_3M_3V_2$ | $N_3M_1V_3$ | $N_2M_3V_1$ | $N_2M_1V_2$ | $N_2$ |
| BLOCK I | | | BLOCK II | | | BLOCK III | | |

**Fig. 5.5** The final layout of a 5 × 3 × 3 factorial experiment arranged in a split-split-plot design with $N$ as mainplot, $M$ as subplot, and $V$ as sub-subplot treatments in three replications.

### 5.2.2 Analysis of variance

The steps in computing the analysis of variance will be illustrated using the grain yield data shown in table 5.1. In the computation that follows, $n$, $m$, and $v$ represent the number of treatments in the main plot, the subplot, and the sub-subplot, respectively.

STEP 1. As in any other analysis of variance, the first step is to compute

$$\text{C.F.} = \frac{(\text{grand total})^2}{\text{total number of observations}}$$

$$= \frac{(884.846)^2}{135}$$

$$= 5{,}799.648, \text{ and}$$

$$\text{Total SS} = \sum X^2 - \text{C.F.}$$

**Table 5.1 Data on grain yields of three rice varieties grown under different management practices, IRRI.**

| Management practice | Grain yield (t/ha) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | $V_1$ | | | $V_2$ | | | $V_3$ | | |
| | I | II | III | I | II | III | I | II | III |
| | | | | $N_1$ (*0 kg N/ha*) | | | | | |
| Minimum ($M_1$) | 3.320 | 3.864 | 4.507 | 6.101 | 5.122 | 4.815 | 5.355 | 5.536 | 5.244 |
| Optimum ($M_2$) | 3.766 | 4.311 | 4.875 | 5.096 | 4.873 | 4.166 | 7.442 | 6.462 | 5.584 |
| Intensive ($M_3$) | 4.660 | 5.915 | 5.400 | 6.573 | 5.495 | 4.225 | 7.018 | 8.020 | 7.642 |
| | | | | $N_2$ (*50 kg N/ha*) | | | | | |
| Minimum ($M_1$) | 3.188 | 4.752 | 4.756 | 5.595 | 6.780 | 5.390 | 6.706 | 6.546 | 7.092 |
| Optimum ($M_2$) | 3.625 | 4.809 | 5.295 | 6.357 | 5.925 | 5.163 | 8.592 | 7.646 | 7.212 |
| Intensive ($M_3$) | 5.232 | 5.170 | 6.046 | 7.016 | 7.442 | 4.478 | 8.480 | 9.942 | 8.714 |
| | | | | $N_3$ (*80 kg N/ha*) | | | | | |
| Minimum ($M_1$) | 5.468 | 5.788 | 4.422 | 5.442 | 5.988 | 6.509 | 8.452 | 6.698 | 8.650 |
| Optimum ($M_2$) | 5.759 | 6.130 | 5.308 | 6.398 | 6.533 | 6.569 | 8.662 | 8.526 | 8.514 |
| Intensive ($M_3$) | 6.215 | 7.106 | 6.318 | 6.953 | 6.914 | 7.991 | 9.112 | 9.140 | 9.320 |
| | | | | $N_4$ (*110 kg N/ha*) | | | | | |
| Minimum ($M_1$) | 4.246 | 4.842 | 4.863 | 6.209 | 6.768 | 5.779 | 8.042 | 7.414 | 6.902 |
| Optimum ($M_2$) | 5.255 | 5.742 | 5.345 | 6.992 | 7.856 | 6.164 | 9.080 | 9.016 | 7.778 |
| Intensive ($M_3$) | 6.829 | 5.869 | 6.011 | 7.565 | 7.626 | 7.362 | 9.660 | 8.966 | 9.128 |
| | | | | $N_5$ (*140 kg N/ha*) | | | | | |
| Minimum ($M_1$) | 3.132 | 4.375 | 4.678 | 6.860 | 6.894 | 6.573 | 9.314 | 8.508 | 8.032 |
| Optimum ($M_2$) | 5.389 | 4.315 | 5.896 | 6.857 | 6.974 | 7.422 | 9.224 | 9.680 | 9.294 |
| Intensive ($M_3$) | 5.217 | 5.389 | 7.309 | 7.254 | 7.812 | 8.950 | 10.360 | 9.896 | 9.712 |

$$= (3.320)^2 + \ldots + (9.712)^2 - 5{,}799.648$$
$$= 373.540.$$

STEP 2. Construct an outline of the analysis of variance as follows:

| Source of variation | Degrees of freedom | Sum of squares | Mean square | Observed $F$ | Tabular $F$ 5% | Tabular $F$ 1% |
|---|---|---|---|---|---|---|
| Block | $r - 1 = 2$ | | | | | |
| Nitrogen ($N$) | $n - 1 = 4$ | | | | | |
| Error (a) | $(r - 1)(n - 1) = 8$ | | | | | |
| Management ($M$) | $m - 1 = 2$ | | | | | |
| $NXM$ | $(n - 1)(m - 1) = 8$ | | | | | |
| Error (b) | $n(r - 1)(m - 1) = 20$ | | | | | |
| Variety ($V$) | $v - 1 = 2$ | | | | | |
| $NXV$ | $(n - 1)(v - 1) = 8$ | | | | | |
| $MXV$ | $(m - 1)(v - 1) = 4$ | | | | | |
| $NXMXV$ | $(n - 1)(m - 1)(v - 1) = 16$ | | | | | |
| Error (c) | $nm(r - 1)(v - 1) = 60$ | | | | | |
| Total | $rmnv - 1 = 134$ | | | | | |

STEP 3. Compute the various sums of squares. The computation will be made separately for the mainplot, the subplot, and the sub-subplot parts.

*Mainplot analysis.* First, construct the table of the mainplot totals (block × nitrogen) as in table 5.2. Then compute

$$\text{Block SS} = \frac{\sum B^2}{nmv} - \text{C.F.}$$
$$= \frac{(294.068)^2 + (299.375)^2 + (291.403)^2}{(5)(3)(3)} - 5{,}799.648$$
$$= 0.732,$$

$$\text{Nitrogen SS} = \frac{\sum N^2}{rmv} - \text{C.F.}$$
$$= \frac{(145.387)^2 + \ldots + (195.316)^2}{(3)(3)(3)} - 5{,}799.648$$
$$= 61.641, \quad \text{and}$$

$$\text{Error (a) SS} = \frac{\sum (BN)^2}{mv} - \text{C.F.} - \text{block SS} - \text{nitrogen SS}$$
$$= \frac{(49.331)^2 + \ldots + (67.866)^2}{(3)(3)} - 5{,}799.648 - 0.732$$
$$- 61.641$$
$$= 4.451.$$

*Subplot analysis.* First construct the mainplot × subplot (the nitrogen × management) table of totals, as shown in table 5.3. Then, compute

**Table 5.2 The block × nitrogen table of yield totals computed from data in table 5.1.**

| Nitrogen level (kg/ha) | Yield total (t/ha) | | | |
|---|---|---|---|---|
| | Block I | Block II | Block III | N total |
| 0 | 49.331 | 49.598 | 46.458 | 145.387 ($N_1$) |
| 50 | 54.791 | 59.012 | 54.146 | 167.949 ($N_2$) |
| 80 | 62.461 | 62.823 | 63.601 | 188.885 ($N_3$) |
| 110 | 63.878 | 64.099 | 59.332 | 186.309 ($N_4$) |
| 140 | 63.607 | 63.843 | 67.866 | 195.316 ($N_5$) |
| Block total | 294.068 | 299.375 | 291.403 | |
| Grand total | | | | 884.846 |

$$\text{Management SS} = \frac{\Sigma M^2}{rnv} - \text{C.F.}$$

$$= \frac{(265.517)^2 + (291.877)^2 + (327.452)^2}{(3)(5)(3)} - 5{,}799.648$$

$$= 42.936, \quad \text{and}$$

$$\text{Nitrogen} \times \text{management SS} = \frac{\Sigma(NM)^2}{rv} - \text{C.F.} - \text{nitrogen SS} - \text{management SS}$$

$$= \frac{(43.864)^2 + \ldots + (71.899)^2}{(3)(3)} - 5{,}799{,}648 - 61.641 - 42.936$$

$$= 1.103.$$

Next, construct the block × mainplot × subplot (block × nitrogen × management) table of totals as in table 5.4. Then compute

$$\text{Error (b) SS} = \frac{\Sigma(BNM)^2}{v} - \text{C.F.} - \text{block SS} - \text{nitrogen SS} - \text{error (a) SS} - \text{management SS} - (\text{nitrogen} \times \text{management}) \text{ SS}$$

$$= \frac{(14.776)^2 + \ldots + (25.971)^2}{3} - 5{,}799.648 - 0.732$$

**Table 5.3 The nitrogen × management table of yield totals computed from data in table 5.1.**

| Nitrogen level (kg/ha) | Yield total (t/ha) | | |
|---|---|---|---|
| | Management 1 | Management 2 | Management 3 |
| 0 | 43.864 | 46.575 | 54.948 |
| 50 | 50.805 | 54.624 | 62.520 |
| 80 | 57.417 | 62.399 | 69.069 |
| 110 | 55.065 | 63.228 | 69.016 |
| 140 | 58.366 | 65.051 | 71.899 |
| Management total | 265 51 | 291 877 | 327.452 |

**Table 5.4 The block × nitrogen × management table of yield totals computed from data in table 5.1.**

| Management | Yield total (t/ha) | | |
|---|---|---|---|
| | Block I | Block II | Block III |
| | | *0 kg N/ha* | |
| $M_1$ | 14.776 | 14.522 | 14.566 |
| $M_2$ | 16.304 | 15.646 | 14.625 |
| $M_3$ | 18.251 | 19.430 | 17.267 |
| | | *50 kg N/ha* | |
| $M_1$ | 15.489 | 18.078 | 17.238 |
| $M_2$ | 18.574 | 18.380 | 17.670 |
| $M_3$ | 20.728 | 22.554 | 19.238 |
| | | *80 kg N/ha* | |
| $M_1$ | 19.362 | 18.474 | 19.581 |
| $M_2$ | 20.819 | 21.189 | 20.391 |
| $M_3$ | 22.280 | 23.160 | 23.629 |
| | | *110 kg N/ha* | |
| $M_1$ | 18.497 | 19.024 | 17.544 |
| $M_2$ | 21.327 | 22.614 | 19.287 |
| $M_3$ | 24.054 | 22.461 | 22.501 |
| | | *140 kg N/ha* | |
| $M_1$ | 19.306 | 19.777 | 19.283 |
| $M_2$ | 21.470 | 20.969 | 22.612 |
| $M_3$ | 22.831 | 23.097 | 25.971 |

$$- 61.641 - 4.451 - 42.936 - 1.103$$
$$= 5.236.$$

*Sub-subplot analysis.* First, construct the mainplot × sub-subplot (nitrogen × variety) table of totals as in table 5.5. Then compute

$$\text{Variety SS} = \frac{\sum V^2}{nm} - \text{C.F.}$$
$$= \frac{(230.707)^2 + (287.826)^2 + (366.313)^2}{(3)(5)(3)} - 5{,}799.648$$
$$= 206.013, \quad \text{and}$$

$$\text{Nitrogen} \times \text{variety SS} = \frac{\sum (NV)^2}{rm} - \text{C.F.} - \text{nitrogen SS} - \text{variety SS}$$
$$= \frac{(40.618)^2 + \ldots + (84.020)^2}{(3)(3)} - 5{,}799.648$$
$$-61.641 - 206.013$$
$$= 14.144.$$

Next, construct the subplot × sub-subplot (management × variety) table of totals as in table 5.6. Then, compute

$$\text{Management} \times \text{variety SS} = \frac{\sum (MV)^2}{rn} - \text{C.F.} - \text{management SS}$$

**Table 5.5 The nitrogen × variety table of totals computed from data in table 5.1.**

| Nitrogen level (kg/ha) | Yield total (t/ha) | | |
|---|---|---|---|
| | $V_1$ | $V_2$ | $V_3$ |
| 0 | 40.618 | 46.466 | 58.303 |
| 50 | 42.873 | 54.146 | 70.930 |
| 80 | 52.514 | 59.297 | 77.074 |
| 110 | 49.002 | 62.321 | 75.986 |
| 140 | 45.700 | 65.596 | 84.020 |
| Variety total | 230.707 | 287.826 | 366.313 |

**Table 5.6 The management × variety table of totals computed from data in table 5.1.**

| Management | Yield total (t/ha) | | | |
|---|---|---|---|---|
| | $V_1$ | $V_2$ | $V_3$ | *N* total |
| $M_1$ | 66.201 | 90.825 | 108.491 | 265.517 |
| $M_2$ | 75.820 | 93.345 | 122.712 | 291.877 |
| $M_3$ | 88.686 | 103.656 | 135.110 | 327.452 |
| Variety total | 230.707 | 287.826 | 366.313 | |

$$- \text{ variety SS}$$
$$= \frac{(66.201)^2 + \ldots + (135.110)^2}{15} - 5{,}799.648$$
$$- 42.936 - 206.013$$
$$= 3.852.$$

Finally, construct the mainplot × subplot × sub-subplot (nitrogen × management × variety) table of totals as in table 5.7. Then compute

Nitrogen × management × variety SS

$$= \frac{\sum(NMV)^2}{r} - \text{C.F.} - \text{nitrogen SS} - \text{management SS} - \text{variety SS}$$
$$- (\text{nitrogen} \times \text{management})\text{SS} - (\text{nitrogen} \times \text{variety})\text{SS}$$
$$- (\text{management} \times \text{variety})\text{SS}$$
$$= \frac{(11.691)^2 + \ldots + (29.968)^2}{3} - 5{,}799.648 - 61.641 - 42.936$$
$$-206.013 - 1.103 - 14.144 - 3.852$$
$$= 3.699, \text{ and}$$

Error (c) SS = Total SS − all other SS's.
$$= 373.540 - 0.732 - 61.641 - 4.451 - 42.936 - 1.103$$

**Table 5.7 The nitrogen × management × variety table of yield totals computed from data in table 5.1.**

| Management | Yield total (t/ha) | | |
|---|---|---|---|
| | $V_1$ | $V_2$ | $V_3$ |
| | | *0 kg N/ha* | |
| $M_1$ | 11.691 | 16.038 | 16.135 |
| $M_2$ | 12.952 | 14.135 | 19.488 |
| $M_3$ | 15.975 | 16.293 | 22.680 |
| | | *50 kg N/ha* | |
| $M_1$ | 12.696 | 17.765 | 20.344 |
| $M_2$ | 13.729 | 17.445 | 23.450 |
| $M_3$ | 16.448 | 18.936 | 27.136 |
| | | *80 kg N/ha* | |
| $M_1$ | 15.678 | 17.939 | 23.800 |
| $M_2$ | 17.197 | 19.500 | 25.702 |
| $M_3$ | 19.639 | 21.858 | 27.572 |
| | | *110 kg N/ha* | |
| $M_1$ | 13.951 | 18.756 | 22.358 |
| $M_2$ | 16.342 | 21.012 | 25.874 |
| $M_3$ | 18.709 | 22.553 | 27.754 |
| | | *140 kg N/ha* | |
| $M_1$ | 12.185 | 20.37 | 25.854 |
| $M_2$ | 15.600 | 21.253 | 28.198 |
| $M_3$ | 17.915 | 24.016 | 29.968 |

$$- 5.236 - 206.013 - 14.144 - 3.852 - 3.699$$
$$= 29.733.$$

STEP 4. Compute all the mean squares by dividing each sum of squares by its degrees of freedom. The $F$-values are then obtained for each effect by dividing its mean square by the appropriate error mean square, as follows:

$$F \text{ for nitrogen} = \frac{\text{nitrogen MS}}{\text{error (a) MS}}$$
$$= \frac{15.4102}{0.5564}$$
$$= 27.70,$$

$$F \text{ for management} = \frac{\text{management MS}}{\text{error (b) MS}}$$
$$= \frac{21.4680}{0.2618}$$
$$= 82.00,$$

$$F \text{ for nitrogen} \times \text{management} = \frac{(N \times M)\text{ MS}}{\text{error (b) MS}}$$
$$= \frac{0.1379}{0.2618}$$
$$= 0.53,$$

$$F \text{ for variety} = \frac{\text{variety MS}}{\text{error (c) MS}} = \frac{103.0065}{0.4956} = 207.84,$$

$$F \text{ for nitrogen} \times \text{variety} = \frac{(N \times V)\text{ MS}}{\text{error (c) MS}} = \frac{1.7680}{0.4956} = 3.57,$$

$$F \text{ for management} \times \text{variety} = \frac{(M \times V)\text{ MS}}{\text{error (c) MS}} = \frac{0.9630}{0.4956} = 1.94, \text{ and}$$

$$F \text{ for nitrogen} \times \text{management} \times \text{variety} = \frac{(N \times M \times V)\text{ MS}}{\text{error (c) MS}} = \frac{0.2312}{0.4956} = 0.4665.$$

STEP 5. Obtain the tabular $F$-values for each effect.

STEP 6. Compute the coefficients of variation as follows.

$$\text{cv (a)} = \frac{\sqrt{\text{error (a) MS}}}{\text{grand mean}} \times 100 = \frac{\sqrt{0.5564}}{6.55} \times 100 = 11.4\%,$$

$$\text{cv (b)} = \frac{\sqrt{\text{error (b) MS}}}{\text{grand mean}} \times 100 = \frac{\sqrt{0.2618}}{6.55} \times 100 = 7.8\%, \text{ and}$$

$$\text{cv (c)} = \frac{\sqrt{\text{error (c) MS}}}{\text{grand mean}} \times 100 = \frac{\sqrt{0.4956}}{6.55} \times 100 = 10.7\%.$$

The cv(a) indicates the degree of precision attached to the measurements of the effects of the mainplot treatments, cv(b) indicates the precision of the subplot treatments and their interaction with the mainplot, and cv(c) indicates the precision of the sub-subplot treatments and all their interactions with the other factors. Normally, we would expect the sizes of these coefficients of variation to decrease from cv(a) to cv(c). In the present example cv(a) is the largest, as expected, but cv(b) and cv(c) do not follow the expected trend. As mentioned earlier (section 4.4.2), such outcomes are encountered occasionally but if they occur frequently, you should consult a competent statistician.

STEP 7. Enter all values computed in steps 1 to 6 in the analysis of variance table as shown in table 5.8.