Crossover designs: issues in construction, use, and communication

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Abstract

In a trial with a crossover design, participants receive a sequence of treatments over two or more periods, with the outcome measured at the end of each period. In order to estimate contrasts between direct treatment effects and between carryover treatment effects, we model each observation as a linear combination of the effects of period, participant, the direct effect of the current treatment, and, for all except the first period observations, the carryover effect of the treatment in the preceding period.

In this thesis, we will consider some aspects of the design and use of crossover experiments. Our focus will be on methods for construction and comparison of designs which improve performance and are accessible to those researchers who need to use crossover designs but who are not specialists in statistical methodology or the design of experiments. In Chapter 2 we discuss the construction of balanced crossover designs. In Chapter 3 we consider visual methods for determining the connectedness of block, row-column, and a restricted class of crossover designs. In Chapter 4 we discuss participant dropout in crossover designs, and introduce a new criterion for selecting a design that is less likely to result in non-estimable treatment contrasts in the event of some participants not completing the trial. In Chapter 5 we present a review of the use of crossover designs in the scientific literature during a one-year period. In Chapter 6 we discuss the relationship between the theory of crossover designs as described in the earlier chapters, and the reality of the use of crossover designs as described in Chapter 5. We conclude by discussing potential practical approaches for making some experimental design methods more widely known and used by researchers who implement trials with crossover designs.

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Glossary

Notation	First used	Description
B	page 13	block design
b	page 13	number of blocks in a block design
D	page 13	crossover design
d_{ij}	page 13	[in a row-column design] treatment allocated to a plot in row i and column j
d_{ij}	page 13	[in a crossover design] treatment allocated in period i to participant j
E	page 26	set of edges
G	page 26	graph
$H(R)$	page 34	Ghosh graph of row-col design R
i	page 13	block, row or period label ($i = 0, \ldots, r-1$)
j	page 13	column or participant label ($j = 0, \ldots, s-1$)
k	page 13	treatment label ($k = 0, \ldots, t-1$)
$L(B)$	page 27	Levi graph of a block design B
m	page 27	length of a path, walk or cycle in a graph
n_i	page 13	number of plots in block i for $i = 1, \ldots, b$
R	page 13	row-column design
r	page 13	number of rows in a row-col design or number of periods in a crossover design

Continued on next page

Continued from previous page

Notation	**First used**	**Description**
s	page 13	number of columns in a row-col design or number of participants in a crossover design
t	page 13	number of treatments in a design
$T(B)$	page 31	treatment-concurrence graph of a block design B
V	page 26	set of vertices
v	page 26	vertex label
$W(R)$	page 33	Wynn graph of 2-row row-col design R
y_{ik}	page 13	[in a block design] response in a plot in block i to treatment k
y_{ijk}	page 13	[in a row-column design] response in a plot in row i and column j to treatment k
y_{ijk}	page 13	[in a crossover design] response in period i and participant j to treatment k
τ_k	page 13	effect of treatment k

Contents

Chapter 1

Introduction

In this chapter we introduce crossover designs, two notable sources of information on on which are the books by Jones and Kenward [21] and Senn [41]. We also define some concepts and notation for other types of experimental designs: block designs and row-column designs. Finally, we introduce some issues in experimental designs that will be addressed in later chapters. Although crossover designs may be used in a wide range of situations, for the purposes of describing and discussing the design we will assume the application is that of a trial involving human participants.

1.1 Crossover designs

A crossover trial design, which may also be referred to as a changeover trial design, is an experimental design in which each participant receives a sequence of two or more treatments over the course of two or more treatment periods. This is in contrast to a parallel group trial design, where each participant receives only one treatment for the duration of the experiment. Each treatment period lasts for the same amount of time, and there may be a washout period between treatment periods where the participants receive no treatment. As each participant acts as their own control, the obvious advantage of a crossover design is that it allows us to consider the within-participant variability in responses to treatments. Another advantage is that as several observations are made

on each subject, fewer subjects are required than in an equivalently powered parallel group design. From the point of view of the trial participants, crossover designs may be less appealing. This is because the total duration of a crossover design will generally be greater than in an equivalent parallel group design, and the participants may experience greater inconvenience from receiving several types of treatment and undergoing repeated assessments. However, some trial participants may welcome the opportunity to receive different types of treatment.

It is only appropriate to use a crossover design for experiments where the condition to be treated is relatively stable. The treatment must result in a reversible outcome, and participants must return to their pre-treatment condition soon after treatment stops. Examples of studies where a crossover trial might be appropriate are the treatment of chronic conditions such as diabetes mellitus, arthritis, hypertension, asthma, and hypercholesterolemia. An example of an inappropriate use of a crossover design would be in the study of an infectious disease such as bacterial meningitis where the aim of treatment is to cure the patient, and there is a risk of serious deteriation or death. As well as medical applications, other areas where crossover designs are in common use include sensory experiments [10] and animal feeding experiments [42].

The most straightforward situation in which a crossover design may be used is in the comparison of two treatments. In this case, participants initially receive treatment A or B in the first period, and then the alternative treatment in the second period. This two-treatment two-period two-sequence crossover design may be referred to in the literature as the AB/BA crossover design or the 2×2 crossover design. The designs to be considered here are more complex, involving the comparison of at least three treatments. An example of such a design, with three periods, three particpants and three treatments which we will label $\{0, 1, 2\}$, is shown in Figure 1.1. In this design, particpant 1 receives treatment 0 in the first period, treatment 1 in the second period and treatment 2 in the third period, whereas the other two participants receive the treatments in a different order.

		Participants		
		1	2	3
Periods	1	0	1	2
	2	1	2	0
	3	2	0	1

Figure 1.1: A crossover design for 3 treatments, 3 periods and 3 participants.

There are two types of treatment effect to consider in crossover designs: the direct effect due to the treatment in the current period, and the carryover or residual effect due to the treatment or treatments in preceding periods. Even with a washout period between treatment applications, it is conceivable that the treatment in period $i-1$ may impact on the response to the next treatment in period i. This effect may be physical or psychological in nature, for example an amount of a drug remaining in the body or an unpleasant experience of a prior treatment affecting the perception that a participant has of the current treatment. The existence and nature of the carryover effect is somewhat controversial, and how it should be quantified, and even whether experimenters should aim to estimate it at all have been discussed in the literature [38, 39, 40, 36, 37].

Here we will assume the first-order or simple carryover model, which states that the effect on the response of the treatment in the previous period is additive and lasts for one period. This is the standard additive model for a crossover design [21, p.9], as described here in Section 1.2. We will also surmise that estimating the carryover effect of a treatment is indeed of interest to the investigator. The justification for this is that all effects associated with a treatment or intervention should be estimated as they may all be relevant when evaluating the overall comparison between treatments. Additionally, it has been suggested that it is of importance to estimate the *total effect* of a treatment, where the total effect is the sum of the direct effect and the carryover effect [3]. The reason for this is that most treatments evaluated using a crossover design are intended to be used on a long term basis, so the total effect may be in some cases a more pragmatic measure of treatment effect in this context than the direct treatment

effect alone.

1.2 Notation for experimental designs

In this section, we introduce the notation that will be used to describe experimental designs.

In a block design B with b blocks and t treatments, the experimental units are divided up so that each unit appears in exactly one block, with each block containing n_i experimental units ($i = 1, \ldots, b$). Each treatment therefore appears in one or more blocks. The response of each experimental unit is modelled as a linear combination of the treatment effect and the block effect, so the response y_{ik} of a unit in block i to treatment k satisfies $E(y_{ik}) = \mu + \pi_i + \tau_k$, where μ is the overall mean, π_i is the effect of block i, and τ_k is the effect of treatment k.

In a row-column design R with r rows, s columns and t treatments, each experimental unit appears in exactly two blocks: one row and one column. The response of each experimental unit is modelled as a linear combination of the treatment effect, the row effect, and the column effect. If the treatment allocated to the plot in row i and column j is given by d_{ij}, then the response y_{ij} of a unit in row i and column j satisifes $E(y_{ij}) = \mu + \pi_i + \alpha_j + \tau_{d_{ij}}$, where μ is the overall mean, π_i is the effect of row i, α_j is the effect of column j, and $\tau_{d_{ij}}$ is the effect of treatment d_{ij}.

In a crossover design D with r periods, s participants, and t treatments, the treatment allocated in the ith period to participant j is d_{ij}, where $i = 0, 1, \ldots r-1$ and $j = 0, 1 \ldots, s-1$. Assuming the presence of direct and carryover treatment effects, the response y_{ij} in period i of the jth participant satisfies $E(y_{ij}) = \mu + \pi_i + \alpha_j + \tau_{d_{ij}} + \lambda_{d_{i-1,j}}$, where μ is the overall mean, π_i is the effect of period i, α_j is the effect of the jth participant, $\tau_{d_{ij}}$ is the direct effect of treatment d_{ij}, and $\lambda_{d_{i-1,j}}$ is the carryover effect of treatment $d_{i-1,j}$ (equal to zero when $i = 1$).

In all of these experimental designs, we do not aim to estimate the treatment effects $\tau_0, \tau_1, \ldots, \tau_{t-1}$ (and, in the crossover design, $\lambda_0, \lambda_1, \ldots, \lambda_{t-1}$). Instead, we estimate

contrasts between treatment effects, for example $\tau_1 - \tau_2$ (and in the crossover design, $\lambda_1 - \lambda_2$). So in the design in Figure 1.1, the contrast $\tau_0 - \tau_1$ may be estimated. If all simple contrasts of the form $\tau_k - \tau_l$ can be estimated in a design, then it is said to be connected for direct treatment effects. Similarly, in the crossover design, if all simple contrasts of the form $\lambda_k - \lambda_l$ can be estimated, then the design is said to be connected for carryover treatment effects.

1.3 Outline of thesis

In this thesis, we will consider some aspects of the design and use of crossover designs, focussing in particular on methods to improve the performance of designs which are accessible to those who may need to use crossover designs but who are not specialists in statistical methodology or the design of experiments.

In Chapter 2, we define balanced crossover designs and explain how they can be constructed. In Chapter 3, we consider visual methods for determining the connectedness of block designs, row-column designs, and a restricted class of crossover designs. In Chapter 4, we discuss the problem of participant dropout in crossover designs, which leads to missing observations. We review existing approaches to choosing designs so that the impact of dropout is reduced (in particular, on the connectedness of the implemented design), and introduce a new criteria for choosing designs which are less likely to become disconnected due to participant dropout. In Chapter 5, we present a review of the use of crossover designs in research published during a one year period. We survey factors including the dimensions of the crossover designs, the models used, mention of carryover effect, choice of design, and occurrence and impact of participant dropout. In Chapter 6, we discuss the relationship between theoretical issues in crossover designs encountered in Chapters 1 to 4, and the reality of the use of crossover designs in peer-reviewed scientific publications (Chapter 5).

Chapter 2

Balanced designs

Balanced experimental designs are preferable to competing designs of the same dimensions as they have increased efficiency according to various criteria. In this chapter we give the properties required for a crossover design to be balanced, and review some methods for constructing balanced crossover designs.

2.1 Balanced crossover designs

We define a balanced crossover design to be a design which is balanced for a first-order carryover effect, as described by Williams [44].

Definition A crossover design D is balanced under the following conditions: (i) each treatment appears the same number of times in each period; (ii) each participant receives each treatment exactly once; (iii) each ordered pair of distinct treatments appears consecutively in the same number of participants.

It follows that if the design D has t treatments, then the number of periods r and the number of participants s are necessarily whole number multiples of t.

Hedayat and Asfarinejad [17] showed that within the class of crossover designs with r periods, s participants, and t treatments, balanced designs are universally optimal in the sense defined by Kiefer [22]. In particular, the sum of the variances of the

contrasts between direct treatment effects are minimised, and the sum of the variances of the contrasts between carryover treatment effects are minimised. It is therefore important for the experimenter to choose a balanced design in order to obtain the most efficient estimates of direct and residual treatment contrasts. We consider this further in Section 2.2.

2.2 Universal Optimality

In this section, we consider Kiefer's work on Universal Optimality, which gives certain properties of the information matrix (**C**-matrix) of a design that are sufficient to demonstrate that a design is optimum [22]. In order to demonstrate the symmetry that is present in the **C**-matrix of a universally optimal design, we then derive the information matrix of an example balanced crossover design.

2.2.1 Universal Optimality and the C-matrix

Kiefer's important results on universal optimality and the **C**-matrix in [22] form the basis for a large amount of the subsequent literature on design optimality. Other contributions by Kiefer on optimality in experimental design have include work on optimal designs under large-degree polynomial regression [23], and work on block designs and Latin square designs which are optimal under an autocorrelation model [24].

Theorem 2.1 Let D^* be a design in the class of competing designs $\mathcal{D}$, with information matrix $\mathbf{C}_{D^*}$. Then D^* is universally optimal over $\mathcal{D}$ if the following two conditions are satisfied:

1. The matrix $\mathbf{C}_{D^*}$ is symmetric.

2. The trace of $\mathbf{C}_{D^*}$ is maximised compared to all other designs in $\mathcal{D}$.

Proof The proof is by Kiefer [22].

0	1	2	3
1	2	3	0
3	0	1	2
2	3	0	1

Figure 2.1: The balanced crossover design D.

In the specific context of crossover designs under an additive model including period effects, participant effects, direct treatment effects, and carryover treatment effects, this result has been used by Hedayat and Asfarinejad [17] to demonstrate that crossover designs which are balanced according to the definition in Section 2.1 are universally optimal.

2.2.2 The C-matrix for a balanced crossover design

Consider the 4-treatment, 4-period, 4-participant design D shown in Figure 2.1, with periods represented by rows and participants by columns. This is a balanced design as each treatment appears exactly once in every period and in every participant, and each treatment is followed once by every other treatment.

We construct the $\mathbf{X}$-matrix of the design D, which describes how the blocks and treatments are applied to each observation in the design, and use this to derive the $\mathbf{C}$-matrix. As D is balanced, and hence optimal, we will observe that the $\mathbf{C}$-matrix is symmetrical.

The design D consists of 16 observations, and the model for these includes an overall mean, four period effects, four participant effects, four direct treatment effects, and four carryover treatment effects. So the $\mathbf{X}$-matrix for D is a 16×17 matrix which can be partitioned in the following way

$$\mathbf{X} = [\mathbf{1}_{16} \,|\, \mathbf{X}_B \,|\, \mathbf{X}_T \,]$$

where

$\mathbf{1}_{16}$ is a vector of length 16 with all entries equal to 1,

$\mathbf{X}_B$ is a 16×8 indicator matrix defined by the block effects (period and participant) applied to each observation, and

$\mathbf{X}_T$ is a 16×8 indicator matrix defined by the treatment effects (direct and carryover) applied to each observation.

Then the components of the $\mathbf{X}$-matrix are

$$\mathbf{X}_B = \begin{bmatrix} 1 & 0 & 0 & 0 & 1 & 0 & 0 & 0 \\ 0 & 1 & 0 & 0 & 1 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 & 1 & 0 & 0 & 0 \\ 0 & 0 & 0 & 1 & 1 & 0 & 0 & 0 \\ 1 & 0 & 0 & 0 & 0 & 1 & 0 & 0 \\ 0 & 1 & 0 & 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 & 1 & 0 & 0 \\ 1 & 0 & 0 & 0 & 0 & 0 & 1 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 1 & 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 1 & 0 & 0 & 1 & 0 \\ 1 & 0 & 0 & 0 & 0 & 0 & 0 & 1 \\ 0 & 1 & 0 & 0 & 0 & 0 & 0 & 1 \\ 0 & 0 & 1 & 0 & 0 & 0 & 0 & 1 \\ 0 & 0 & 0 & 1 & 0 & 0 & 0 & 1 \end{bmatrix}, \mathbf{X}_T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 1 & 0 & 0 & 1 & 0 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 & 1 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 0 & 0 & 1 \\ 0 & 1 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 1 & 0 & 0 \\ 1 & 0 & 0 & 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 1 & 1 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 & 0 & 1 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 & 0 & 1 \\ 1 & 0 & 0 & 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 & 0 & 0 & 0 \\ 1 & 0 & 0 & 0 & 0 & 0 & 0 & 1 \\ 0 & 0 & 1 & 0 & 1 & 0 & 0 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 & 1 & 0 \end{bmatrix}$$

From [17], the $\mathbf{C}$-matrix is given by

$$\mathbf{C} = \mathbf{X}_T'\mathbf{X_T} - \mathbf{X}_T'\mathbf{X}_B(\mathbf{X}_B'\mathbf{X}_B)^{-}\mathbf{X}_B'\mathbf{X}_T$$

The diagonal of matrix $\mathbf{X}_T'\mathbf{X}_T$ gives the replication of the treatments, with off-diagonal entries taking value zero. So as each treatment appears four times in D as a

direct effect and three times as a carryover effect, then

$$\mathbf{X}_T'\mathbf{X}_T = \begin{bmatrix} 4 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 4 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 4 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 4 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 3 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 3 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 3 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 0 & 3 \end{bmatrix}$$

Similarly, the matrix $\mathbf{X}_B'\mathbf{X}_B$ gives the number of times each of the periods and participants appear in the design. So as each period has four observations, and each participant contributes four observations, then

$$\mathbf{X}_B'\mathbf{X}_B = \begin{bmatrix} 4 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 4 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 4 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 4 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 4 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 4 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 4 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 0 & 4 \end{bmatrix}$$

with

$$(\mathbf{X}_B'\mathbf{X}_B)^- = \begin{bmatrix} \frac{1}{4} & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & \frac{1}{4} & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & \frac{1}{4} & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & \frac{1}{4} & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & \frac{1}{4} & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & \frac{1}{4} & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & \frac{1}{4} & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 0 & \frac{1}{4} \end{bmatrix}$$

The matrix $\mathbf{X}_T'\mathbf{X}_B$ gives the number of times each treatment occurs in each period or participant.

$$\mathbf{X}_T'\mathbf{X}_B = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & 1 & 1 & 1 \\ 1 & 1 & 1 & 1 & 1 & 1 & 1 & 1 \\ 1 & 1 & 1 & 1 & 1 & 1 & 1 & 1 \\ 1 & 1 & 1 & 1 & 1 & 1 & 1 & 1 \\ 0 & 1 & 1 & 1 & 1 & 1 & 0 & 1 \\ 0 & 1 & 1 & 1 & 1 & 1 & 1 & 0 \\ 0 & 1 & 1 & 1 & 0 & 1 & 1 & 1 \\ 0 & 1 & 1 & 1 & 1 & 0 & 1 & 1 \end{bmatrix}$$

Similarly,

$$\mathbf{X}_B'\mathbf{X}_T = \begin{bmatrix} 1 & 1 & 1 & 1 & 0 & 0 & 0 & 0 \\ 1 & 1 & 1 & 1 & 1 & 1 & 1 & 1 \\ 1 & 1 & 1 & 1 & 1 & 1 & 1 & 1 \\ 1 & 1 & 1 & 1 & 1 & 1 & 1 & 1 \\ 1 & 1 & 1 & 1 & 1 & 1 & 0 & 1 \\ 1 & 1 & 1 & 1 & 1 & 1 & 1 & 0 \\ 1 & 1 & 1 & 1 & 0 & 1 & 1 & 1 \\ 1 & 1 & 1 & 1 & 1 & 0 & 1 & 1 \end{bmatrix}$$

Then

$$\mathbf{X}_T'\mathbf{X}_B(\mathbf{X}_B'\mathbf{X}_B)^{-}\mathbf{X}_B'\mathbf{X}_{\mathbf{T}}$$

$$= \begin{bmatrix} 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&1&1\\ 0&1&1&1&1&1&0&1\\ 0&1&1&1&1&1&1&0\\ 0&1&1&1&0&1&1&1\\ 0&1&1&1&1&0&1&1 \end{bmatrix} \begin{bmatrix} \frac{1}{4}&0&0&0&0&0&0&0\\ 0&\frac{1}{4}&0&0&0&0&0&0\\ 0&0&\frac{1}{4}&0&0&0&0&0\\ 0&0&0&\frac{1}{4}&0&0&0&0\\ 0&0&0&0&\frac{1}{4}&0&0&0\\ 0&0&0&0&0&\frac{1}{4}&0&0\\ 0&0&0&0&0&0&\frac{1}{4}&0\\ 0&0&0&0&0&0&0&\frac{1}{4} \end{bmatrix} \begin{bmatrix} 1&1&1&1&0&0&0&0\\ 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&0&1\\ 1&1&1&1&1&1&1&0\\ 1&1&1&1&0&1&1&1\\ 1&1&1&1&1&0&1&1 \end{bmatrix}$$

$$= \begin{bmatrix} \frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}\\ \frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}\\ \frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}\\ \frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}\\ 0&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&0&\frac{1}{4}\\ 0&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&0\\ 0&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&0&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}\\ 0&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&\frac{1}{4}&0&\frac{1}{4}&\frac{1}{4} \end{bmatrix} \begin{bmatrix} 1&1&1&1&0&0&0&0\\ 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&1&1\\ 1&1&1&1&1&1&0&1\\ 1&1&1&1&1&1&1&0\\ 1&1&1&1&0&1&1&1\\ 1&1&1&1&1&0&1&1 \end{bmatrix}$$

$$= \begin{bmatrix} 2&2&2&2&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}\\ 2&2&2&2&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}\\ 2&2&2&2&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}\\ 2&2&2&2&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}\\ \frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{5}{4}&\frac{5}{4}&\frac{5}{4}\\ \frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{5}{4}&\frac{3}{2}&\frac{5}{4}&\frac{5}{4}\\ \frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{5}{4}&\frac{5}{4}&\frac{3}{2}&\frac{5}{4}\\ \frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{3}{2}&\frac{5}{4}&\frac{5}{4}&\frac{5}{4}&\frac{3}{2} \end{bmatrix}$$

So

$$\mathbf{C} = \mathbf{X}_T'\mathbf{X_T} - \mathbf{X}_T'\mathbf{X}_B(\mathbf{X}_B'\mathbf{X}_B)^{-}\mathbf{X}_B'\mathbf{X_T}$$

$$= \begin{bmatrix} 4 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 4 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 4 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 4 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 3 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 3 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 3 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 0 & 3 \end{bmatrix} - \begin{bmatrix} 2 & 2 & 2 & 2 & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} \\ 2 & 2 & 2 & 2 & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} \\ 2 & 2 & 2 & 2 & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} \\ 2 & 2 & 2 & 2 & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} \\ \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{5}{4} & \frac{5}{4} & \frac{5}{4} \\ \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{5}{4} & \frac{3}{2} & \frac{5}{4} & \frac{5}{4} \\ \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{5}{4} & \frac{5}{4} & \frac{3}{2} & \frac{5}{4} \\ \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{3}{2} & \frac{5}{4} & \frac{5}{4} & \frac{5}{4} & \frac{3}{2} \end{bmatrix}$$

$$= \begin{bmatrix} 2 & -2 & -2 & -2 & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} \\ -2 & 2 & -2 & -2 & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} \\ -2 & -2 & 2 & -2 & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} \\ -2 & -2 & -2 & 2 & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} \\ -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & \frac{3}{2} & -\frac{5}{4} & -\frac{5}{4} & -\frac{5}{4} \\ -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{5}{4} & \frac{3}{2} & -\frac{5}{4} & -\frac{5}{4} \\ -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{5}{4} & -\frac{5}{4} & \frac{3}{2} & -\frac{5}{4} \\ -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{3}{2} & -\frac{5}{4} & -\frac{5}{4} & -\frac{5}{4} & \frac{3}{2} \end{bmatrix}$$

So the **C**-matrix for the design in Figure 2.1 is symmetrical, as expected for the information matrix of a design which we know to be universally optimal.

2.3 Williams' designs

Williams [44] described the construction of balanced crossover designs with $r = t$ periods and $s = t$ participants for t even, and $r = t$ periods and $s = 2t$ participants for t odd. Where the number of treatments t is even, the first participant is allocated the sequence of treatments

$$0 \quad 1 \quad t-1 \quad 2 \quad t-2 \quad 3 \quad t-3 \ldots \; t/2$$

For the remaining $t-1$ participants, the treatment sequences are found by adding 1 mod t, 2 mod $t, \ldots,$ $t-1$ mod t to the treatments in this initial sequence. The t treatment sequences form a balanced crossover design with numbers of periods, participants and treatments all equal to t. Alternative treatment sequences for the first participant which are not of this form are also given by Williams, for example

$$0 \quad 2 \quad 1 \quad 4 \quad 5 \quad 3$$

for $t = 6$.

Where the number of treatments is odd, a pair of sequences for an initial two participants are used. These may be of the general form

$$0 \quad 1 \quad t-1 \quad 2 \quad t-2 \ldots \ (t-1)/2 \quad (t+1)/2$$

and

$$(t+1)/2 \quad (t-1)/2 \ldots \ t-1 \quad 2 \quad t-1 \quad 1 \quad 0$$

A further $t-1$ treatment sequences are obtained from each of these by adding 1 mod t, 2 mod $t, \ldots,$ $t-1$ mod t to the treatments as before. This results in a total of $2t$ treatment sequences, which form a balanced crossover design with t periods, $2t$ participants, and t treatments.

The Latin squares that Williams constructed may also, depending on orientation, be known as row- or column-complete Latin squares, and the treatment sequences allocated to the initial participants in the constructions are now recognised as terraces [2]. The formal construction of column-complete Latin squares using terraces is described in the next section.

2.4 Column-complete Latin squares

If the treatments labelled $(0, 1, \ldots, t-1)$ were arranged in a Latin square of side t, then each treatment would appear once in each row and once in each column. In a column-complete Latin square, each ordered vertical pair of treatments appear the same number of times throughout the square. Considering periods as rows and participants as columns, it is clear that a balanced crossover design is a column-complete Latin square. Here we consider a method of constructing column-complete Latin squares, and hence balanced crossover designs. The designs that can be constructed using this method include those given by Williams [44].

Bailey [2] introduces the term *terrace* and discusses the construction of quasi-complete and complete Latin squares using terraces. Terraces can be found for many groups, but we will consider only the cyclic groups $\mathbb{Z}_m$; that is, the integers modulo m under addition.

Definition Let $\mathbf{a} = (a_1, \ldots, a_m)$ be a sequence which is a permutation of the elements $\{0, \ldots, m-1\}$ of $\mathbb{Z}_m$. The differences are given by the sequence $\mathbf{b} = (0, b_2, \ldots, b_m)$, where $b_i = a_i - a_{i-1} \mod m$ for $i = 2, \ldots, m$. Then $\mathbf{a}$ is a **terrace** for $\mathbb{Z}_m$ if for each $x \in \mathbb{Z}_m$ $(x \neq \frac{m}{2})$, either x occurs twice and $-x$ does not occur, or $-x$ occurs twice and x does not occur, or x and $-x$ both occur once in the sequence $\mathbf{b}$. If m is even then $\frac{m}{2}$ must also occur once in $\mathbf{b}$.

Definition If the elements of $\mathbf{b}$ are distinct then $\mathbf{a}$ is a **directed terrace** for $\mathbb{Z}_m$.

For example, if $m = 6$ and $\mathbf{a} = (0, 1, 4, 2, 3, 5)$ then $\mathbf{b} = (0, 1, 3, 4, 1, 2)$ and so $\mathbf{a}$ is an undirected terrace for $\mathbb{Z}_6$. If $\mathbf{a} = (0, 1, 5, 2, 4, 3)$ then $\mathbf{b} = (0, 1, 4, 3, 2, 5)$ and so $\mathbf{a}$ is a directed terrace for $\mathbb{Z}_6$. Although the term is not used, from [44], we know that for any m the sequence $\mathbf{w}_m = (0, 1, m-1, 2, m-2, \ldots)$ is a terrace for $\mathbb{Z}_m$, and for even m it is a directed terrace.

Let t be even, let $\mathbf{a}$ be a directed terrace for $\mathbb{Z}_t$, and let $\mathbf{c}_t$ be the sequence $(0, 1, \ldots, t-1)$. Then using [2] we can construct a Latin square $L(\mathbf{a}, \mathbf{c}_t)$. The ith entry of $\mathbf{a}$ is a_i and

the jth entry of $\mathbf{c}_t$ is $j-1$, so the (i,j)th entry of $L(\mathbf{a},\mathbf{c}_t)$ is given by $l_{ij} = a_i+j-1 \mod t$. We define a crossover design $D(\mathbf{a})$ where the treatment sequences are the columns of $L(\mathbf{a},\mathbf{c}_t)$, so $D(\mathbf{a})$ is a balanced crossover design with t treatments, t periods and t participants. For example, $\mathbf{w}_6 = (0,1,5,2,4,3)$ is a directed terrace for $\mathbb{Z}_6$ so the design $D(\mathbf{w}_6)$ in Figure 2.2 is a balanced six-treatment six-period six-participant crossover design.

If t is odd then we can use an undirected terrace $\mathbf{a} = (a_1,\ldots,a_t)$ and its inverse $-\mathbf{a} = (-a_1,\ldots,-a_t)$, where $-a_i \mod t = t - a_i \mod t$. We form two Latin squares $L(\mathbf{a},\mathbf{c}_t)$ and $L(-\mathbf{a},\mathbf{c}_t)$. The $2t$ columns of the two Latin squares give us a balanced t-treatment t-period $2t$-participant crossover design which we denote by $D(\mathbf{a},-\mathbf{a})$. For example $\mathbf{w}_5 = (0,1,4,2,3)$, $-\mathbf{w}_5 = (0,4,1,3,2)$ and so the balanced five-treatment five-period ten-subject crossover design $D(\mathbf{w}_5,-\mathbf{w}_5)$ is as shown in Figure 2.3.

0	1	2	3	4	5
1	2	3	4	5	0
5	0	1	2	3	4
2	3	4	5	0	1
4	5	0	1	2	3
3	4	5	0	1	2

Figure 2.2: The design $D(\mathbf{w}_6)$

0	1	2	3	4		0	1	2	3	4
1	2	3	4	0		4	0	1	2	3
4	0	1	2	3		1	2	3	4	0
2	3	4	0	1		3	4	0	1	2
3	4	0	1	2		2	3	4	0	1

Figure 2.3: The design $D(\mathbf{w}_5,-\mathbf{w}_5)$

Chapter 3

Visual representations of experimental designs

A visual representation of an experimental design, such as a graph, can be a useful way to express certain properties of the design, and to allow competing designs to be compared and discussed. Here we focus on graphs derived from designs that allow whether the design is connected or not to be determined by inspection of the graph. In this chapter we consider a selection of graphs that represent experimental designs, and introduce a new way of using a graph to represent a crossover design with two periods.

3.1 Some basic graph theory and terminology

The first three chapters of Wilson [45] provide an introduction to the fundamental concepts of graph theory, on which some of the following definitions are based.

Definition A graph G consists of a set of vertices V and a set of edges E. An edge joining vertices v_1 and v_2 ($v_1, v_2 \in V$) is denoted by the unordered pair $\{v_1, v_2\}$. Figure 3.1(a) shows a graph with $V = \{0, 1, 2, 3\}$ and edges $E = \{\{0, 1\}, \{1, 2\}, \{1, 3\}, \{2, 3\}\}$.

Definition A directed graph is a graph in which a direction is applied to each edge. An edge from v_1 to v_2 is expressed as an ordered pair (v_1, v_2). The graph in Figure 3.1(b)

is a directed graph with edges $E = \{(1,0),(1,2),(2,3)\}$.

Definition A walk in G is a finite sequence of edges in which all pairs of consecutive edges share a vertex. So the sequence $(\{v_0,v_1\},\{v_1,v_2\},\{v_2,v_3\},\ldots,\{v_{m-1},v_m\})$ is a walk in G from v_0 to v_m, of length m edges. A path is a walk in which the inner vertices $\{v_1,\ldots,v_{m-1}\}$ are distinct. A cycle is a path which starts and ends at the same vertex and has at least one edge. An elementary cycle is a cycle in which no vertex appears more than once in the path. Figure 3.1(c) shows a path between vertices 0 and 2 within the graph shown in Figure 3.1(a).

Definition A graph is connected if there is a path from v_1 to v_2 for each pair of vertices v_1, v_2. A graph is disconnected if it is not connected. The graph in Figure 3.1(d) is disconnected as there is no path from vertices 0 or 3 to vertices 1 or 2.

Definition A disconnected graph G is the union of two or more connected graphs. Each of these connected graphs is a component of G. The graph shown in Figure 3.1(d) has two components.

Definition If a graph G is *bipartite*, then the vertex set V can be split into two disjoint sets V' and V'' such that each edge of G is of the form $\{v', v''\}$, where $v' \in V'$and $v'' \in V''$. Figure 3.1(e) shows a bipartite graph with disjoint vertex sets labelled with squares and circles.

3.2 The Levi graph for a block design

The bipartite graph known as the Levi graph or incidence graph was initially described by Levi [25, p.5]. As in Section 1.2, block design B has r blocks, t treatments, and n_i plots in each block ($i = 0,\ldots,r-1$), and the response y_{ik} on a plot in block i under treatment k is given by $E(y_{ik}) = \mu + \beta_i + \tau_k$, where $i = 0,\ldots,r-1$ and $k = 0,\ldots,t-1$.

Definition Let $L(B)$ be the Levi graph of block design B. Then $L(B)$ is a bipartite graph with a set of vertices corresponding to the blocks $0,\ldots,r-1$ of B, and a second

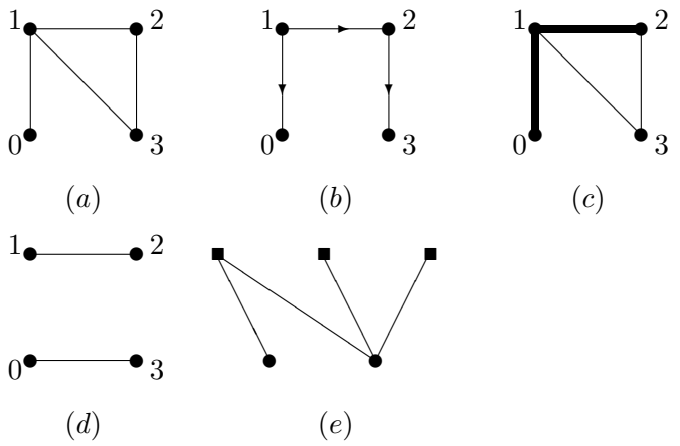


Figure 3.1: Some examples of graphs.

set of vertices corresponding to the treatments $0, \ldots, t-1$ of B. If block i contains treatment k, then $L(B)$ has an edge $\{i, k\}$.

Example 3.1 Figure 3.2 shows the Levi graphs of block designs B_1 and B_2. The square vertices in the upper part of each graph are the block vertices, and the circular vertices in the lower part of each graph are the treatment vertices.

B_1	Block	0	0	1	1	2	2
	Treatment	0	1	0	2	2	3
	Response	y_{00}	y_{01}	y_{10}	y_{12}	y_{22}	y_{23}

B_2	Block	0	0	1	1	2	2
	Treatment	0	1	2	3	0	1
	Response	y_{00}	y_{01}	y_{12}	y_{13}	y_{20}	y_{21}

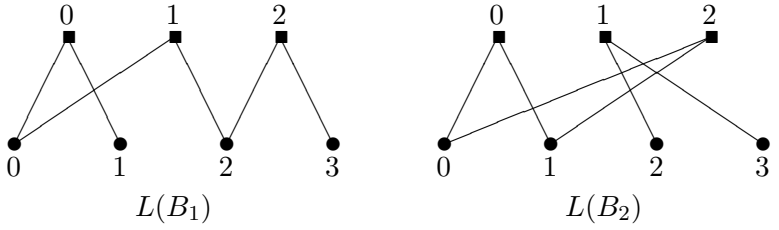


Figure 3.2: Examples of Levi graphs.

In Chapter 1 we defined a design to be connected if all elementary treatment contrasts were estimable. The term *connected* was used in a different way by Bose [5], who first defined a block and a treatment to be associated if the treatment lies within the block, and then:

> "Two treatments, two blocks or a treatment and a block, may be said to be 'connected' if it is possible to pass from one to the other by means of a chain consisting alternately of blocks and treatments, such that any two members of a chain are associated."

It is clear that this definition is equivalent to the existence of a path between the two corresponding vertices in the Levi graph, although Bose does not use the term graph here. Bose goes on to define a design to be connected if every block or treatment is connected to every other block or treatment, a definition which corresponds to the Levi graph of the block design being connected.

Theorem 3.1 (i) A block design B is connected for treatments if and only if the vertices in the Levi graph $L(B)$ corresponding to treatments in B are connected; (ii) a block design B is connected for blocks if and only if the vertices in the Levi graph $L(B)$ corresponding to blocks in B are connected; (iii) the Levi graph $L(B)$ of a block design B is connected if and only if B is connected for all treatment and block contrasts.

Proof The proof (Bose [5] and Chakrabarti [8]) shows that the following are equivalent: a block design B is connected for treatment effects in the usual sense; the information matrix of B has rank $t-1$; and B is connected for treatment effects in the sense of Bose [5]. Consequently B is connected for treatment effects in the usual sense if and only if the treatment vertices in the Levi graph $L(B)$ are connected, so (i) is true. As blocks and treatments may be interchanged in B and $L(B)$, (ii) and therefore (iii) is true.

Furthermore, we may consider this in the following way. Let the Levi graph $L(B)$ be connected for treatments. Then for any pair of treatments k_1, k_m in B there is a

path in $L(B)$ between those vertices which correspond to k_1 and k_m. If k_1 and k_m are in the same block b_1, then the path will consist of the edges $\{k_1, b_1\}$ and $\{k_m, b_1\}$. These edges correspond to the observations in B of y_{b_1,k_1} and y_{b_1,k_m}. Let c_1 be the linear combination of observations from B given by

$$c_1 = y_{b_1,k_1} - y_{b_1,k_m}$$

so then

$$\begin{aligned} E(c_1) &= E(y_{b_1,k_1}) - E(y_{b_1,k_m}) \\ &= (\mu + \beta_{b_1} + \tau_{k_1}) - (\mu + \beta_{b_1} + \tau_{k_m}) \\ &= \tau_{k_1} - \tau_{k_m} \end{aligned}$$

and so the treatment contrast $\tau_{k_1} - \tau_{k_m}$ can be estimated by c_1. However, if k_1 and k_m are not in the same block, then the path between the vertices k_1 and k_m will pass through intermediate treatment vertices $k_2, \ldots, k_{m-1}$ and intermediate block vertices $b_1, \ldots, b_{m-1}$. The path will then consist of pairs of edges $\{k_1, b_1\}$ & $\{k_2, b_1\}$, $\{k_2, b_2\}$ & $\{k_3, b_2\}$, ..., and $\{k_{m-1}, b_{m-1}\}$ & $\{k_m, b_{m-1}\}$. These pairs of edges correspond to observations y_{b_1,k_1} & y_{b_1,k_2}, y_{b_2,k_2} & y_{b_2,k_3}, ..., and $y_{b_{m-1},k_{m-1}}$ & y_{b_{m-1},k_m}. So the linear combinations of observations in B given by

$$c_j = y_{b_j,k_j} - y_{b_j,k_{j+1}}$$

can be estimated, with

$$E(c_j) = \tau_{k_j} - \tau_{k_{j+1}}$$

for $j = 1, \ldots, m-1$. The sum of all such c_j encountered along the path from k_1 to k_m then given by

$$\begin{aligned} \sum_{j=1}^{m} E(c_j) &= \sum_{j=1}^{m-1} (\tau_{k_j} - \tau_{k_{j+1}}) \\ &= \tau_{k_1} - \tau_{k_m} \end{aligned}$$

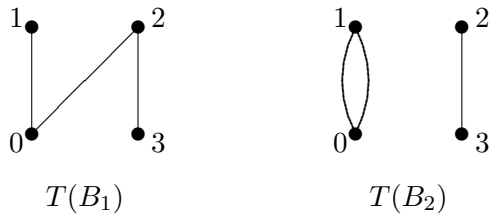
Example 3.1 (revisited) To then illustrate the use of this relationship between a block design and the corresponding Levi graph, we consider the designs B_1 and B_2 and Levi graphs $L(B_1)$ and $L(B_2)$ in Example 3.1. All treatment contrasts are estimable in block design B_1: the elementary contrasts $\tau_0 - \tau_1$, $\tau_0 - \tau_2$ and $\tau_2 - \tau_3$ can be estimated by $\hat{\tau_0} - \hat{\tau_1} = y_{00} - y_{01}$, $\hat{\tau_0} - \hat{\tau_2} = y_{10} - y_{12}$ and $\hat{\tau_2} - \hat{\tau_3} = y_{22} - y_{23}$ respectively, and the remaining contrast estimates $\hat{\tau_0} - \hat{\tau_3}, \hat{\tau_1} - \hat{\tau_2}$ and $\hat{\tau_1} - \hat{\tau_3}$ are linear combinations of these. However, block design B_2 is not connected as the only estimable elementary contrasts are $\tau_0 - \tau_1$ and $\tau_2 - \tau_3$. By inspection of the Levi graphs, it is clear that $L(B_1)$ is connected but $L(B_2)$ is not. The two components of $L(B_2)$ partition the treatment vertices into $\{0, 1\}$ and $\{2, 3\}$, corresponding to the only estimable treatment contrasts in B_2: $\tau_0 - \tau_1$ and $\tau_2 - \tau_3$.

The use of the Levi graph to determine which elementary contrasts are estimable is straightforward, as suggested in Example 3.1. If there is a path between treatment vertices k and l in $L(B)$, then the elementary contrast $\tau_k - \tau_l$ is estimable in B.

3.3 The treatment-concurrence graph

The treatment-concurrence graph $T(B)$ of a block design B is described by John and Williams [20, p.22] and Bailey [1, p.82]. The vertices of $T(B)$ are the treatments of B, and $\{k, l\}$ is an edge whenever treatments k and l are in the same block.

Example 3.2 The graphs $T(B_1)$ and $T(B_2)$ in Figure 3.3 are the treatment-concurrence graphs of the block designs in Figure 3.2.



$T(B_1)$ $T(B_2)$

Figure 3.3: Examples of treatment-concurrence graphs.

As with the Levi graph, the treatment-concurrence graph can be used to determine the connectedness of a block design.

Theorem 3.2 The treatment-concurrence graph of a block design is connected if and only if the block design is connected.

Proof The treatment-concurrence graph $T(B)$ may be constructed by removing the block vertices from $L(B)$. It is therefore clear that there is a path between vertices k and l in $T(B)$ if and only if there is a path between vertices k and l in $L(B)$. Hence $T(B)$ is connected if and only if $L(B)$ is connected, and so from Theorem 3.1, $T(B)$ is connected if and only if the block design B is connected. □

The graph $T(B_1)$ in Figure 3.3 is connected, but $T(B_2)$ is not. The left-hand and right-hand components of $T(B_2)$ correspond to the estimable contrasts $\tau_0 - \tau_1$ and $\tau_2 - \tau_3$ respectively in B_2. It is straightforward to find from $T(B)$ which elementary contrasts are estimable in B: if there is a path between vertices k and l in $T(B)$, then the elementary contrast $\tau_k - \tau_l$ is estimable in B.

3.4 Row-column designs with two rows

Let R be a row-column design with two rows, s columns, and t treatments. The response y_{ijk} of the plot in row i and column j under treatment k is given by $E(y_{ijk}) = \mu + \pi_i +$

$\alpha_j + \tau_k$, where $i = 1, 2,\ j = 1, \ldots, s$ and $k = 1, \ldots, t$. Wynn [46] defines a directed graph for such a design, which we will denote $W(R)$. The graph $W(R)$ has t vertices, which correspond to the treatments of R. The s edges of $W(R)$ are defined by the columns of R, in that (k, l) is an edge if there is a column with treatment k in the first row and treatment l in the second row.

Example 3.3 The examples that we consider here are from Wynn [46]. Figure 3.4 shows the row-column designs R_1 and R_2 which have 2 rows, 5 columns and 5 treatments, and the graphs $W(R_1)$ and $W(R_2)$.

The graph $W(R)$ can be used to determine whether a row-column design with two rows is connected.

Theorem 3.3 A row-column design R with two rows is connected for treatment effects if and only if the graph $W(R)$ satisfies the following two conditions: (i) the underlying undirected graph is connected; (ii) the directed graph $W(R)$ contains at least one elementary unbalanced cycle.

Proof As demonstrated by Wynn [46], using network flow theory.

In Figure 3.4, the design R_1 is connected, and the design R_2 is not. Both of the graphs $W(R_1)$ and $W(R_2)$ are connected, satisfying condition (i), but only graph $W(R_1)$ contains the elementary unbalanced cycle required to satisfy condition (ii). Those elementary contrasts which are not estimable in a row-column design R with two rows cannot be easily determined from the graph $W(R)$. In Figure 3.4, the contrast $\tau_1 - \tau_0$ is not estimable in R_2. This fact could not be derived from simple inspection of the graph $W(R_2)$.

3.5 Row-column designs with more than two rows

The Wynn graph for row-column designs with two rows (Section 3.4) cannot be easily extended to row-column designs with more than two rows. The method described by

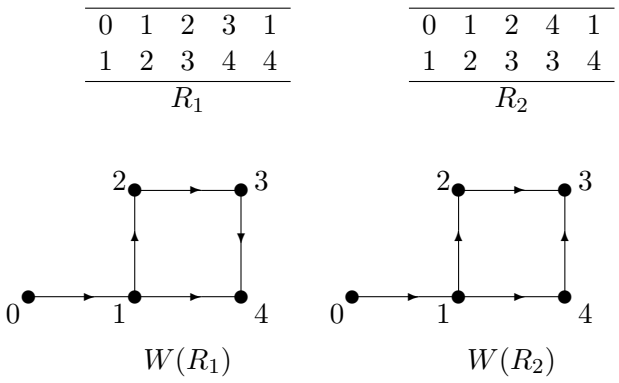


Figure 3.4: Examples of graphs of row-column designs with two rows.

Ghosh [11] for determining whether a general row-column design is connected is quite different to the Wynn graph. Let R be a row-column design with r rows and s columns. Let $H(R)$ denote the graph described by Ghosh, and as in [11], we assume without loss of generality that $r \leq s$. The vertices of $H(R)$ are not individual treatments, but instead pairs of treatments. A *column block* is a pair of columns of R, in which r ordered pairs of treatments appear. The graph $H(R)$ is constructed by first considering the s column blocks consisting of column numbers $(1,2),\ (1,3),\ \ldots,(1,s),\ (2,3)$. The vertices of $H(R)$ are the ordered pairs of treatments appearing in these column blocks. To construct the edges of $H(R)$, an equivalence relation $\sim$ is defined on both the set of treatments $1,2,\ldots,k$ and the set of treatment pairs. When applied to the set of treatments, $k_1 \sim k_2$ if k_1 and k_2 are connected: that is, if the treatment contrast $\tau_{k_1} - \tau_{k_2}$ is estimable in R. When applied to the set of treatment pairs, $(k_1,k_2) \sim (k_3,k_4)$ if the pairs (k_1,k_2) and (k_3,k_4) are connected: that is, if the contrast $(\tau_{k_4} - \tau_{k_3}) - (\tau_{k_2} - \tau_{k_1})$ is estimable in R. A pair of vertices $\{(k_1,k_2),(k_3,k_4)\}$ is an edge if the treatment pairs (k_1,k_2) and (k_3,k_4) are connected by a column block. Treatment pairs can either be connected by a column block if they are in the same column block, or if they can be shown to be connected using the properties of the relation $\sim$

Theorem 3.4 The graph $H(R)$ is connected if and only if the design R is connected in the usual sense.

Proof This is proved by Ghosh [11], using the equivalence classes defined by $\sim$.

This method may not be easy to use in practice as the graph is not as straightforward to construct and inspect as the Levi, treatment-concurrence and Wynn graphs. The number of vertices and edges increases rapidly with the dimensions of the design. For the row-column design R with r rows and s columns, s pairs of columns of R are used to form the column blocks required for constructing $H(R)$. So the maximum number of unique ordered pairs of treatments and therefore vertices in $H(R)$ is equal to $r \times s$. Using this method to determine whether a design R is certainly disconnected may be particularly difficult, as all vertex pairs across presumed separate components of $H(R)$ would have to be assessed to establish that there was no possibility that the corresponding vertex pairs could actually be connected using the properties of the equivalence relation.

3.6 Crossover designs with two periods

Consider a crossover design D with t treatments, 2 periods, and s participants.

Theorem 3.5 A two-period crossover design with $2s$ observations under the model (1.2) is equivalent to a block design with s observations.

Proof This was proven by Hedayat and Asfarinejad [18]. Let x_j be the difference between the period 2 and period 1 observations for a participant j, so $x_j = y_{2j} - y_{1j}$. Then we have

$$\begin{aligned} E(x_j) &= E(y_{2j}) - E(y_{1j}) \\ &= \pi_2 - \pi_1 + \tau_{d_{2j}} - \tau_{d_{1j}} + \lambda_{d_{1j}}. \end{aligned} \tag{3.1}$$

We let $\pi = \pi_2 - \pi_1$ and $\beta_{d_{1j}} = \lambda_{d_{1j}} - \tau_{d_{1j}}$, so we have

$$E(x_j) = \pi + \beta_{d_{1j}} + \tau_{d_{2j}}. \tag{3.2}$$

This is the model for a block design B defined by D. As all t treatments appear in each period, then B has t blocks and t treatments. The block effects are $\beta_{d_{1j}}$ ($d_{1j} = 0, \ldots, t-1$) and the treatment effects $\tau_{d_{2j}}$ ($d_{2j} = 0, \ldots, t-1$). Let C_{dir} be the information matrix for the direct treatment effects in D, and let C_B be the information matrix for the treatments in the block design B. Hedayat and Asfarinejad [18] show that $C_B = 2C_{dir}$. □

We use this relationship between a two-period crossover design and a block design to define the graph G_D of a two-period crossover design D. An example is shown in Figure 3.5. The vertices of G_D are arranged in a $2 \times t$ grid, with each row representing a period and each column representing a treatment. We label the vertex representing treatment k in period i by k_i. Then for each participant in D we draw an edge in G_D joining the first and second period treatments. So G_D has an edge (k_1, l_2) if and only if there is a participant in D with treatment k in the first period and treatment l in the second period. Now consider the block design B which is formed from the two-period crossover design D. It is clear that the bipartite graph G_B as described above is identical to the graph G_D.

Theorem 3.6 The two-period crossover design D is connected for direct and carryover treatment contrasts if and only if the graph G_D of D is connected.

Proof If D is connected, then all elementary contrasts $\tau_k - \tau_l$ and $\lambda_k - \lambda_l$ are estimable in (1.2). So the estimators $\hat{\tau}_k - \hat{\tau}_l$ and $\hat{\lambda}_k - \hat{\lambda}_l$ can be expressed as linear combinations of the observations from D. The expectations of these estimators will clearly not involve the participant effects. Let c be a linear combination of the observations in the crossover design D such that the expectation of c does not involve the participant effects. We will show that c can be expressed as a linear combination of the observations in the block design B. From (1.2),

$$c = \sum_{j=1}^{s} (a_j y_{1j} + b_j y_{2j})$$

where a_j and b_j are the coefficients for the observations on participant j in the first and second periods respectively. Under the model (1.2),

$$\begin{aligned} E(c) &= \sum_{j=1}^{s} [a_j E(y_{1j}) + b_j E(y_{2j})] \\ &= \sum_{j=1}^{s} \left[a_j(\mu + \pi_1 + \alpha_j + \tau_{d_{1j}}) + b_j(\mu + \pi_2 + \alpha_j + \tau_{d_{2j}} + \lambda_{d_{1j}})\right] \\ &= \sum_{j=1}^{s} \left[a_j(\mu + \pi_1 + \tau_{d_{1j}}) + b_j(\mu + \pi_2 + \tau_{d_{2j}} + \lambda_{d_{1j}})\right] + \sum_{j=1}^{s}(a_j + b_j)\alpha_j. \end{aligned}$$

As $E(c)$ does not involve the participant effects, then $a_j + b_j = 0$ for all $j = 1, \ldots, s$. We put $a_j = -b_j$ to get

$$c = \sum_{j=1}^{s} b_j(y_{2j} - y_{1j}) = \sum_{j=1}^{s} b_j x_j,$$

which is a linear combination of observations in B.

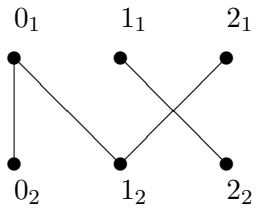
So the estimators of the contrasts may each be expressed as a linear combination of observations x_j from the block design B. Hence we can estimate the elementary treatment contrasts $\tau_k - \tau_l$ in B, and as $\beta_k - \beta_l = (\lambda_k - \tau_k) - (\lambda_l - \tau_l) = (\lambda_k - \lambda_l) - (\tau_k - \tau_l)$ then we can estimate the elementary block contrasts. Now suppose B is connected. Then all elementary contrasts $\tau_k - \tau_l$ and $\beta_k - \beta_l$ are estimable in (3.2). So the estimators $\hat{\tau}_k - \hat{\tau}_l$ and $\hat{\beta}_k - \hat{\beta}_l$ can be expressed as linear combinations of the observations from B. It is obvious from (3.1) that these estimators can then each be expressed as a linear combination of observations from D. So we can estimate the elementary treatment contrasts $\tau_k - \tau_l$ in D, and as $\lambda_k - \lambda_l = (\beta_k + \tau_k) - (\beta_l + \tau_l) = (\beta_k - \beta_l) + (\tau_k - \tau_l)$ then we can estimate the elementary contrasts between carryover treatment effects.

As G_D and G_B are identical, then from Theorem 3.1, G_D is connected if and only if B is connected. As D is connected if and only if B is connected, then we can conclude that the two-period crossover design D is connected for direct and carryover treatment contrasts if and only if the graph G_D of D is connected. □

(a)

Subjects	1	2	3	4
Period 1 treatments	0	0	1	2
Period 2 treatments	0	1	2	1

(b)



(c)

Blocks	0	1	2
Treatments	{0,1}	{2}	{1}

Figure 3.5: (a) The crossover design D, (b) the graph G_D, and (c) the block design B.

We illustrate this in Example 3.4, with the two-period crossover design D shown in Figure 3.5.

Example 3.4 Crossover design D in Figure 3.5 has two periods, four participants, and three treatments $\{0, 1, 2\}$. The graph G_D is also shown, as is the block design B that we can derive from D. It is clear that the Levi graph of B is identical to the graph G_D. Evaluation of D using the method described by Godolphin [15] shows that D is disconnected, with $\tau_0 - \tau_1$ the only estimable elementary contrast. By inspection, G_D is not connected.

The result of Theorem 3.6 gives a simple method of checking if a proposed two-period crossover design is connected, and hence a simple method for checking if a proposed crossover design is connected on the first two periods (Section 4.2).

3.7 General crossover designs

The method described in the previous section cannot be easily extended to crossover designs with more than two periods, as the method relies on the relationship between a two-period crossover design and a block design. Although crossover designs are not explicitly discussed, the methods presented by Butz [6] could potentially be applied to crossover designs. The method presented by Butz can be used to construct a graph for a design with any number of factors (block types and treatment types). However, the method is difficult to apply and not as intuitive as some of the other graphical methods discussed in this chapter. Theoretically, a crossover design can be considered as a 4-factor design with certain restrictions on the factors. These restrictions arise because the levels of the third and fourth factors (the direct and carryover treatment effects) are linked: a direct treatment applied in period i defines the carryover treatment in period $i + 1$. The complexity of Butz's method means that this graph would not be easy to construct or easy to use to determine whether a design is connected.

3.8 Other methods for determining connectedness

The original motivation for visual methods that can be used to determine design connectedness, such as that developed by Ghosh [11], was the computational difficulty of otherwise assessing a design. Generally, the information matrix (C-matrix) would have to be calculated, which was time and resource intensive, particularly if the process was to be repeated for various competing designs. Methods such as Ghosh's [11] and the non-graphical method presented by Park and Shah [33] although less straightforward than the Levi or treatment-concurrence graphs, allowed a design to be assessed without the need to derive the information matrix. As computing facilities have become faster and cheaper, these concerns are less of a priority, but the appeal of straightforward methods to assess experimental designs remains, particularly those which allow the user to easily determine which treatment contrasts are and are not estimable. For

example, a method for detecting disconnected designs which can be applied to block designs, row-column designs and crossover designs with first-order carryover effects has been described by Godolphin [15].

However, such methods do not seem to have been widely used: according to Web of Science (accessed March 2015), Ghosh's paper has only been cited twice, on both occasions in papers also on methods to determine the connectedness of designs. As part of the process of enabling experimenters to understand the concepts of good experimental design, there would be benefits to making such methods more widely known and accessible to those who might use them in practice.

Chapter 4

Dropout in crossover trials

Even the most carefully designed of experiments may not always proceed as planned. Potential problems common to all trial designs include missing data and participant non-compliance. In trials with a crossover design there is the further possibility of participant dropout, where participants complete some but not all of their full allocated sequence of treatments. A possible consequence of this is that some treatment contrasts will be non-estimable, an outcome that means that trial resources have been wasted. In this chapter we consider how crossover trials may be designed so that the impact of potential participant dropout is minimized.

4.1 Participant dropout

If participant j drops out before the end of the ith period of a p-period crossover design, they will only contribute the $i-1$ observations $y_{1j}, \ldots, y_{i-1j}$. Even with careful trial management, the multiple treatment periods that participants are required to complete may mean that some degree of participant dropout is inevitable in crossover designs. Low, Lewis and Prescott [28] state that "experience suggests that a dropout rate of between 5% and 10% is not uncommon and, in some areas, can be as high as 25%". The resulting implemented design may be disconnected, with some direct or carryover treatment contrasts being non-estimable. In order to protect resources invested in the

trial, it is therefore essential to select a design that is more likely to remain connected than competing designs in the event of participant dropout. As well as considering financial and material resources, there are clear ethical implications of using a trial design that is more vulnerable to dropout. This is because participants will have been certainly inconvenienced and possibly exposed to some degree of risk in a trial that might need to be repeated in order to obtain the required treatment contrast estimates.

Block designs have been previously described as *robust* if they are unlikely to become disconnected due to missing observations [12, 13, 14]. Other authors have used the term *robust* slightly differently to describe experiemental designs which perform well according to additional criteria when observations are missing [28, 43]. Here we will focus on reducing the risk of crossover designs becoming disconnected due to dropout, and describe such designs as *protected* against participant dropout.

A method for assessing the robustness of a crossover design to participant dropout is described by Low *et al.* [28]. For a given design, they compare the numerous designs that could be implemented following every possible pattern of dropout. Even for a small design the number of possible implemented designs is large: for example, there are 10^6 possible implemented designs for a three-period design with two participants on each of twelve distinct treatment sequences. The criteria used to compare the robustness of competing designs are that the probability of implementing a disconnected design must be acceptably small, and that implemented efficiency measures for estimating both direct and carryover treatment contrasts must be close to those in the original design. The use of Polya theory (further described in [27]) greatly reduces the amount of computation required, but the resulting method remains computationally intensive.

Low *et al.*'s method is demonstrated by comparing the robustness to dropout of two balanced four-treatment four-period designs, each with 16 participants. One design consists of four copies of a four-participant design, and the other consists of two copies of an eight-participant design. Although dropout may occur at any point, the authors observe that participants are most likely to drop out in the final period. This, along

with computational restrictions, leads to the assumption in their examples that dropout occurs only in the last period of an experiment. Of the 625 possible implemented designs from the four copies of the four-participant design, 113 are disconnected. In contrast to this, no possible implemented designs from the two copies of the eight-participant design are disconnected. With participant dropout, the two copies of the eight-participant design were found to perform better under the A and MV optimality criteria for both direct and carryover treatment effects. A second example with 24 participants compared suitable numbers of copies of the four-participant and eight-participant designs with two copies of a balanced 12-participant design. The two copies of the 12-participant design outperform the other two designs, and so the authors recommend using this design if dropout in the final period is considered to be a likely occurrence.

In contrast to the final period dropout from several participants considered by Low *et al.* [28], Varghese *et al.* [43] consider t-treatment (and hence t-period) Williams designs (Section 2.3) where the last $t-1$ observations are lost from one participant. They find that all of the implemented designs are connected for direct and carryover treatment contrasts, and that the efficiencies of the implemented designs relative to the original designs are high. Where the number of treatments is equal to 3 or 4, the authors replicate the design twice as they observe that a single replicate Williams design was not sufficiently robust to this pattern of participant dropout.

A procedure is described by Godolphin [15, 16] which allows the identification of *rank reducing observation sets*: sets of observations which would, if missing, result in a disconnected design. To illustrate the importance of evaluating the risk of a design becoming disconnected due to lost observations, an example is given of a crossover design that became disconnected due to participant dropout in the final period. The design consists of two copies of a four-treatment Williams square. Four participants, representing two whole treatment sequence groups, drop out before the end of the final period. In the implemented design, no elementary contrasts between direct or carryover treatment effects are estimable. Godolphin gives an alternative design for the

eight participants consisting of two different Williams squares. The implemented design remains connected even if up to seven participants drop out in the final period. Rank reducing observation sets have also been obtained for designs where the number of trial participants is necessarily small, for example if recruitment is difficult, as it may be if a rare medical condition is under investigation [4].

4.2 Method to select designs protected against dropout

It is clear that the risk of participant dropout in a crossover trial will be higher in the later periods of the design than in the earlier periods. Suppose, in an extreme example, a design D with p periods is terminated at the end of the qth period ($1 \leq q \leq p$). If all direct and carryover treatment contrasts are still estimable in the implemented design, that is if the original design D is connected on the first q periods, then D is protected against any dropout in the later $p-q$ periods. We propose that to reduce the risk of an implemented design being disconnected, a planned crossover design should be connected on the first two periods. This means that any participant dropout during the third or subsequent periods may increase the uncertainty around our estimates but will not compromise the connectedness of the design.

In order to estimate carryover effects, the implemented design must have at least two periods. So to maximize the number of periods during which D is protected against dropout, we choose $q = 2$. It is not suggested that a two-period design should be the aim in such a trial, but rather that choosing a design connected on the first two periods provides the experimenter with a generous safety net to protect against a disconnected design. The graphical method described in Section 3.6 can be used to determine whether the first two periods of a given crossover design are connected. If D is a crossover design with $p \geq 2$ periods, we define the graph G_D to be the graph of the first two periods of D. So D is connected on the first two periods if and only if G_D is connected. If D has t treatments, then by definition G_D has $2t$ vertices. For G_D to be connected, there must be at least $2t-1$ edges, corresponding to at least $2t-1$ participants in D. For D to be

balanced, the number of participants must be a multiple of t. Consequently a balanced t-treatment crossover design which is connected on the first two periods must have at least $2t$ participants.

Insisting that a design be connected on the first two periods still allows for familiar and commonly-used designs which are balanced for carryover effects. In Sections 4.3 and 4.4 we explain how the method described in Section 2.4 can be modified in order to construct designs with these properties.

4.3 Construction of protected designs where t is odd

We first consider the construction of balanced designs protected against participant dropout where there is an odd number t of treatments ($t \geq 3$). From the construction described in Section 2.4, we can use the terrace for $\mathbb{Z}_t$, $\mathbf{w}_t = (0, 1, t-1, 2, t-2, \ldots)$ and its inverse $-\mathbf{w}_t$ to form the balanced design $D(\mathbf{w}_t, -\mathbf{w}_t)$ (Figure 4.1).

In $D(\mathbf{w}_t, -\mathbf{w}_t)$, two participants receive treatment k in period 1, with one such participant then receiving treatment $k-1$ in period 2, and the other receiving treatment $k+1$. Consequently in the bipartite graph $G_{D(\mathbf{w}_t, -\mathbf{w}_t)}$, vertex k_1 is connected to the vertices $(k-1)_2$ and $(k+1)_2$, so there is a path consisting of two edges that connects k_1 to $(k+2)_1$, via $(k+1)_2$, for all $k \in \{0, 1, \ldots, t-1\}$. If we start at 0_1 and proceed along the edges of the graph according to this path, the sequence of period 1 vertices that we visit is $(0_1, 2_1, 4_1, \ldots, (t-1)_1, 1_1, 3_1, \ldots, (t-2)_1, 0_1)$ and the sequence of period 2 vertices that we visit is $(1_2, 3_2, 5_2, \ldots, (t-2)_2, 0_2, 2_2, 4_2, \ldots, (t-1)_2)$. This is a path of length $2t$ starting and finishing at vertex 0_1, passing through all other vertices exactly once. Hence $G_{D(\mathbf{w}_t, -\mathbf{w}_t)}$ is a cycle of length $2t$ and so is connected. So, for t odd, the balanced crossover design $D(\mathbf{w}_t, -\mathbf{w}_t)$ is connected on the first two periods for $t \geq 3$. For example, the design $D(\mathbf{w}_5, -\mathbf{w}_5)$ is shown in Figure 4.1(a) and the graph $G_{D(\mathbf{w}_5, -\mathbf{w}_5)}$ in Figure 4.1(b). The graph $G_{D(\mathbf{w}_5, -\mathbf{w}_5)}$ is connected and so $D(\mathbf{w}_5, -\mathbf{w}_5)$ is connected on the first two periods.

4.4 Construction of protected designs where t is even

We consider an even number of treatments ($t \geq 4$), and aim to find balanced t-treatment t-period designs with $2t$ periods. To construct these designs, we use the lifting method for constructing directed terraces.

4.4.1 The lifting method for constructing directed terrces

Let $\mathbf{a}$ be a terrace for $\mathbb{Z}_t$ (Section 2.4), where $t \geq 2$ and t may be odd or even. It is possible to *lift* $\mathbf{a}$ to construct a directed terrace $\mathbf{a}'$ for $\mathbb{Z}_{2t}$. The lifting method is described by Ollis [32] for a general binary group G with $|G| = 2t$. The unique element z of order 2 in G generates a subgroup $\Lambda(G)$, and a terrace for $G/\Lambda(G)$ is then lifted to construct a directed terrace for G. Here we describe the lifting method in terms of a terrace for $\mathbb{Z}_t$ being lifted to a directed terrace for $\mathbb{Z}_{2t}$. Our terrace for $\mathbb{Z}_t$, $\mathbf{a} = (0, a_2, a_3, \ldots, a_t)$, has differences given by the sequence $\mathbf{b} = (0, b_2, b_3, \ldots, b_t)$, where $b_2 = a_2$ and $b_i = a_i - a_{i-1}$ for $i = 3, \ldots, t$. We want to form a sequence $\mathbf{b}'$ where $\mathbf{b}' = (0, b'_2, \ldots, b'_t, t, -b'_t, \ldots, -b'_2)$ and $b'_i \in \mathbb{Z}_{2t}$, $i = 2, \ldots, t$. We choose values for the b'_i by considering the following cases for each b_i, $i = 2, \ldots, t$.

Case (I). If $b_i \neq \frac{t}{2}$ and $b_i = b_j$ for some $i, j \in \{2, \ldots, t\}, i \neq j$, then either $b'_i = b_i$ and $b'_j = t + b_j \mod 2t$ or $b'_i = t + b_i \mod 2t$ and $b'_j = b_j$.

Case (II). If $b_i \neq \frac{t}{2}$ and $-b_i = b_j \mod t$ for some $i, j \in \{2, \ldots, t\}, i \neq j$, then either $b'_i = b_i$ and $b'_j = b_j$ or $b'_i = t + b_i \mod 2t$ and $b'_j = t + b_j \mod 2t$.

Case (III). If $b_i = \frac{t}{2}$ for some $i \in \{2, \ldots, t\}$, then either $b'_i = b_i$ or $b'_i = t + b_i \mod 2t$.

Forming a sequence $\mathbf{a}'$ with differences given by $\mathbf{b}'$ gives a directed terrace for $\mathbb{Z}_{2t}$.

(a)

$$\begin{array}{ccccc|ccccc}
0 & 1 & 2 & \cdots & t-1 & 0 & 1 & 2 & \cdots & t-1 \\
1 & 2 & 3 & \cdots & 0 & t-1 & 0 & 1 & \cdots & t-2 \\
t-1 & 0 & 1 & \cdots & t-2 & 1 & 2 & 3 & \cdots & 0 \\
2 & 3 & 4 & \cdots & 1 & t-2 & t-1 & 0 & \cdots & t-3 \\
\vdots & \vdots & \vdots & & \vdots & \vdots & \vdots & \vdots & & \vdots \\
\frac{t+1}{2} & \cdots & \cdots & \cdots & \frac{t-1}{2} & \frac{t-1}{2} & \cdots & \cdots & \cdots & \frac{t-3}{2}
\end{array}$$

(b)

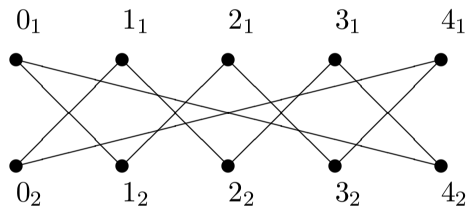


Figure 4.1: (a) The general balanced protected design $D(\mathbf{w}_t, -\mathbf{w}_t)$ for t odd, and (b) the graph $G_{D(\mathbf{w}_5,-\mathbf{w}_5)}$.

Theorem 4.1 Let $\mathbf{a}$ be a terrace for $\mathbb{Z}_t$ of the form $(0, 2, \ldots)$. Then we can use the lifting method to construct a directed terrace $\mathbf{a}'$ for $\mathbb{Z}_{2t}$ of the form $\mathbf{a}' = (0, 2, \ldots)$.

Proof As $\mathbf{a}$ is of the form $(0, 2, \ldots)$, then the sequence of differences $\mathbf{b}$ is also of the form $(0, 2, \ldots)$, so $a_2 = b_2 = 2$. When constructing the sequence $\mathbf{b}'$, regardless of which of Case (I), Case (II) and Case (III) is satisfied by b_2, we are able to choose b'_2 such that $b'_2 = b_2 = 2$. The sequence $\mathbf{a}'$ constructed with differences given by $\mathbf{b}'$ will therefore be a directed terrace for $\mathbb{Z}_{2t}$ of the form $(0, 2, \ldots)$. □

Example 4.1 If $t = 6$, then $\mathbf{a} = (0, 2, 3, 1, 4, 5)$ is an undirected terrace for $\mathbb{Z}_6$. The differences are given by $\mathbf{b} = (0, b_2, b_3, b_4, b_5, b_6) = (0, 2, 1, 4, 3, 1)$. As $b_2 = -b_4 = 2$, case (II) applies. We can choose either $b'_2 = 2$ and $b'_4 = 4$ or $b'_2 = 8$ and $b'_4 = 10$. To construct a terrace of the form $(0, 2, \ldots)$, we choose $b'_2 = 2$ and $b'_4 = 4$. Now $b_3 = b_6 = 1$, so this is case (I) and we can choose $b'_3 = 1$ and $b'_6 = 7$. Finally, $b_5 = 3 = \frac{t}{2}$ so this is case (III) and we can choose $b'_5 = 3$. This gives $\mathbf{b}' = (0, b'_2, \ldots, b'_6, 6, -b'_6, \ldots, -b'_2) = (0, 2, 1, 4, 3, 7, 6, 5, 9, 8, 11, 10)$. So the sequence with differences given by $\mathbf{b}'$ is $\mathbf{a}' = (0, 2, 3, 7, 10, 5, 11, 4, 1, 9, 8, 6)$, which is a directed terrace for $\mathbb{Z}_{12}$.

4.4.2 Even values of t with $t \geq 6$

For even values of t with $t \geq 6$, the lifting method for constructing directed terraces can be used to construct balanced crossover designs which are connected on the first two periods.

Theorem 4.2 For all even t with $t \geq 6$, there exists a directed terrace for $\mathbb{Z}_t$ of the form $\mathbf{e} = (0, 2, \ldots)$.

Proof As t is even we can write $t = 2^k m$ where k is an integer with $k \geq 1$ and m is odd with $m \geq 1$. As m is odd, the map $x \mapsto 2x \mod m$ is an automorphism of $\mathbb{Z}_m$. From [2] we can multiply the elements of the terrace $\mathbf{w}_m$ by 2 to get a terrace for $\mathbb{Z}_m$. As $2\mathbf{w}_m = (0, 2, m-2, 4, \ldots, m-1, 1)$, then there exists an undirected terrace for $\mathbb{Z}_m$ of the form $\mathbf{a} = (0, 2, \ldots)$. We can apply the lifting method to $\mathbf{a}$ and construct a

directed terrace $\mathbf{a}'$ for $\mathbb{Z}_{2m}$ of the form $\mathbf{a}' = (0, 2, \ldots)$. Now suppose that we have a directed terrace $\mathbf{a} = (0, 2, \ldots)$ for $\mathbb{Z}_t$ where $t = 2^k m$ and $k \geq 1$. Then we can apply the lifting method to construct a directed terrace $\mathbf{a}' = (0, 2, \ldots)$ for $\mathbb{Z}_{2t}$. As $2t = 2^{k+1}m$, this completes the induction on k.

Suppose $m = 1$, so $t = 2^k$. First we consider $k = 1$ and $k = 2$, and note that no directed terrace of the form $(0, 2, \ldots)$ exists for $\mathbb{Z}_2$ or $\mathbb{Z}_4$. Now we consider $k = 3$. By exhaustive search, there are exactly 4 directed terraces for $\mathbb{Z}_8$ of the form $(0, 2, \ldots)$: $(0, 2, 1, 5, 3, 6, 7, 4)$, $(0, 2, 3, 6, 5, 1, 7, 4)$, $(0, 2, 5, 1, 7, 6, 3, 4)$ and $(0, 2, 7, 6, 1, 5, 3, 4)$. We can then use the same inductive argument as for the case $m \geq 3$ to show that there exists a directed terrace for $\mathbb{Z}_{2^k}$ of the form $(0, 2, \ldots)$ for all $k \geq 3$. □

Example 4.2 Figure 4.2 shows $D(\mathbf{w}_6, \mathbf{e})$, where $\mathbf{e} = (0, 2, 1, 4, 5, 3)$ is a directed terrace for $\mathbb{Z}_6$, and the connected graph $G_{D(\mathbf{w}_6, \mathbf{e})}$. The terrace $\mathbf{w}_t = (0, 1, t-1, 2, \ldots)$ is a directed terrace for $\mathbb{Z}_t$, so $D(\mathbf{w}_t)$ is a balanced t-treatment, t-period, t-participant design. As $\mathbf{e}$ is also a directed terrace for $\mathbb{Z}_t$, then $D(\mathbf{e})$ is also a balanced t-treatment t-period t-participant design, and hence $D(\mathbf{w}_t, \mathbf{e})$ is a balanced t-treatment t-period $2t$-participant design. In $D(\mathbf{w}_t, \mathbf{e})$, each treatment k in period 1 is followed by treatments $k+1$ and $k+2$ in period 2. So in the bipartite graph $G_{D(\mathbf{w}_t, \mathbf{e})}$, vertex k_1 is connected to the vertices $(k+1)_2$ and $(k+2)_2$, hence there is a path consisting of two edges from k_1 to $(k+1)_1$ passing through $(k+2)_2$. If we start at 0_1 and move along the edges of the graph in this way, the sequence of period 1 vertices that we visit is $(0_1, 1_1, 2_1, \ldots, (t-1)_1, 0_1)$ and the sequence of period 2 vertices that we visit is $(2_2, 3_2, 4_2, \ldots, 0_2, 1_2)$. This is a path of length $2t$ starting and finishing at 0_1, passing through all other vertices exactly once. Hence $G_{D(\mathbf{w}_t, \mathbf{e})}$ is a cycle of length $2t$ and so is connected.

4.4.3 The case $t = 4$

We now consider the special case $t = 4$. We cannot use the same construction as for all even $t \geq 6$ as no directed terrace of the form $(0, 2, \ldots)$ exists for $\mathbb{Z}_4$. The only possible candidates are $(0, 2, 1, 3)$ and $(0, 2, 3, 1)$, which have sequences of differences $(0, 2, 3, 2)$

(a)

0	1	2	3	4	5		0	1	2	3	4	5
1	2	3	4	5	0		2	3	4	5	0	1
5	0	1	2	3	4		1	2	3	4	5	0
2	3	4	5	0	1		4	5	0	1	2	3
4	5	0	1	2	3		5	0	1	2	3	4
3	4	5	0	1	2		3	4	5	0	1	2

(b)

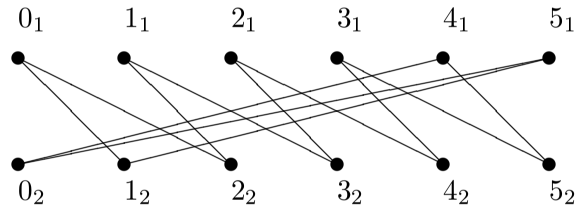


Figure 4.2: Using the directed terrace $\mathbf{e} = (0, 2, 1, 4, 5, 3)$ we can construct (a) the design $D(\mathbf{w}_6, \mathbf{e})$ with (b) graph $G_{D(\mathbf{w}_6,\mathbf{e})}$.

and $(0, 2, 1, 2)$ respectively, and so are not directed terraces for $\mathbb{Z}_4$. In fact we can show that no design with the desired properties exists, and so for a balanced design with $t = p = 4$ to be connected on the first two periods, at least $n = 12$ participants are required.

Theorem 4.3 There does not exist a balanced design D with $t = p = 4$ and $s = 8$ which is connected on the first two periods.

Proof Suppose that such a design D exists. Then the graph G_D of the first two periods of D is a bipartite graph with 8 vertices and 8 edges. As D is balanced, each vertex of G_D has degree 2. As D is connected on the first two periods, it is clear that G_D is a cycle as shown in Figure 4.3(a). The unknown second period treatments are labelled a, b, c, d. The first two periods of design D (Figure 4.3(b)) are unique up to permutation of the treatment labels. As each treatment appears only once in each sequence of the full design D, $a \neq 0$ and $a \neq 1$. So $a \in \{2, 3\}$, and similarly $b \in \{0, 3\}$, $c \in \{0, 1\}$ and $d \in \{1, 2\}$. Choosing $a = 2$ forces $d = 1$, $c = 0$, $b = 3$, which gives the possible first two periods A of D in Figure 4.4. Choosing $a = 3$ forces $b = 0$, $c = 1$, $d = 2$, giving the possible first two periods B of D in Figure 4.4. However, A is a permutation of B formed by interchanging the treatments 0 and 2. So we need only consider A as the first two periods of D.

We now consider the treatments in the remaining periods of D. Suppose the treatment allocated to participant 1 in period 3 is $d_{3,1} = 2$. The ordered pair $(1, 2)$ appears in both participant 1 and in participant 3, so $d_{3,8} = 0$. This forces $d_{4,8} = 2$, so the ordered pair $(0, 2)$ appears in participants 2 and 8. So $d_{3,7} = 1$, which forces $d_{4,7} = 2$, resulting in the ordered pair $(1, 2)$ appearing more than twice in D.

Now suppose $d_{3,1} = 3$. If $d_{3,2} = 1$ then $d_{4,2} = 3$ and so the ordered pair $(1, 3)$ would appear in participants 1, 2 and 4. So we must instead have $d_{3,2} = 3$. Treatment 3 now appears twice in period 3, forcing $d_{3,6} = 1$ and so $d_{4,6} = 3$. This results in the ordered pair $(1, 3)$ appearing in participants 1, 2 and 6. So the construction of D has failed, and we conclude that such a design does not exist. □

4.5 Performance and application of protected designs

We have shown that balanced t-treatment t-period $2t$-participant crossover designs that are connected on the first two periods exist for all odd t with $t \geq 3$ and for all even t with $t \geq 6$. For odd t we have shown how to construct such designs using the terrace $\mathbf{w}_t = (0, 1, t-1, 2, t-2, \ldots)$ and its inverse $-\mathbf{w}_t = (0, t-1, 1, t-2, 2, \ldots)$. For even t we use the directed terrace $\mathbf{w}_t$ to form a balanced t-period t-participant design, and we use a directed terrace of the form $(0, 2, \ldots)$ to form another balanced t-period t-participant design. Together these give a balanced t-period $2t$-participant design which is connected on the first two periods.

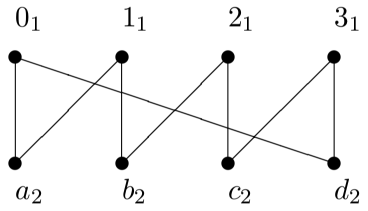
4.5.1 Simulation

In order to compare the performance of a protected design to that of a competing design which is balanced but which is not connected on the first two periods, we consider a simulation of participant dropout. Suppose a balanced design with $t = p = 6$ is required for $s = 12$ participants. Figure 4.5 shows two possible choices: two copies of the six-participant design $D(\mathbf{w}_6)$, or a single copy of the 12-participant design $D(\mathbf{w}_6, \mathbf{e})$ (using $\mathbf{e} = (0, 2, 1, 4, 5, 3)$).

The design $D(\mathbf{w}_6, \mathbf{e})$ has been constructed to be connected after the first two periods, and it can be shown that $D(\mathbf{w}_6, \mathbf{w}_6)$ is connected only after the first three periods. This suggests that choosing $D(\mathbf{w}_6, \mathbf{e})$ would reduce the risk of a disconnected design. We compared these designs using a simulation of dropout. For each participant, we assumed the following probabilities of dropping out before the end of the given period, conditional on being in the trial at the start of the period:

Period	1	2	3	4	5	6
P(dropout)	0.05	0.1	0.15	0.2	0.25	0.3

The simulation included 10000 runs for each of the two designs, and for each run the process involved the following steps:



0	0	1	1	2	2	3	3
d	a	a	b	b	c	c	d

(b)

Figure 4.3: (a) The first two periods of the design D and (b) the graph G_D.

0	0	1	1	2	2	3	3
1	2	2	3	3	0	0	1

A

0	0	1	1	2	2	3	3
2	3	3	0	0	1	1	2

B

Figure 4.4: Possible first two periods of design D.

$$D(\mathbf{w}_6, \mathbf{w}_6) \quad \begin{matrix} 0 & 1 & 2 & 3 & 4 & 5 \\ 1 & 2 & 3 & 4 & 5 & 0 \\ 5 & 0 & 1 & 2 & 3 & 4 \\ 2 & 3 & 4 & 5 & 0 & 1 \\ 4 & 5 & 0 & 1 & 2 & 3 \\ 3 & 4 & 5 & 0 & 1 & 2 \end{matrix} \qquad \begin{matrix} 0 & 1 & 2 & 3 & 4 & 5 \\ 1 & 2 & 3 & 4 & 5 & 0 \\ 5 & 0 & 1 & 2 & 3 & 4 \\ 2 & 3 & 4 & 5 & 0 & 1 \\ 4 & 5 & 0 & 1 & 2 & 3 \\ 3 & 4 & 5 & 0 & 1 & 2 \end{matrix}$$

$$D(\mathbf{w}_6, \mathbf{e}) \quad \begin{matrix} 0 & 1 & 2 & 3 & 4 & 5 \\ 1 & 2 & 3 & 4 & 5 & 0 \\ 5 & 0 & 1 & 2 & 3 & 4 \\ 2 & 3 & 4 & 5 & 0 & 1 \\ 4 & 5 & 0 & 1 & 2 & 3 \\ 3 & 4 & 5 & 0 & 1 & 2 \end{matrix} \qquad \begin{matrix} 0 & 1 & 2 & 3 & 4 & 5 \\ 2 & 3 & 4 & 5 & 0 & 1 \\ 1 & 2 & 3 & 4 & 5 & 0 \\ 4 & 5 & 0 & 1 & 2 & 3 \\ 5 & 0 & 1 & 2 & 3 & 4 \\ 3 & 4 & 5 & 0 & 1 & 2 \end{matrix}$$

Figure 4.5: Competing designs with $t = p = 6$ and $s = 12$.

1. Begin with $N = 12$ participants. Independently, each have a 5% probability of dropping out before the end of the 1st period. Observations from the 1st period are available for the remaining $N_1 \leq 12$ participants.

2. The remaining N_1 participants start the 2nd period. Independently, each have a 10% probability of dropping out before the end of the 2nd period. Observations from the 2nd period are available for the remaining $N_2 \leq N_1$ participants.

3. The remaining N_2 participants start the 3rd period. Independently, each have a 15% probability of dropping out before the end of the 3rd period. Observations from the 2nd period are available for the remaining $N_3 \leq N_2$ participants.

4. The remaining N_3 participants start the 4th period. Independently, each have a 20% probability of dropping out before the end of the 4th period. Observations from the 4th period are available for the remaining $N_4 \leq N_3$ participants.

5. The remaining N_4 participants start the 5th period. Independently, each have a 25% probability of dropping out before the end of the 5th period. Observations from the 5th period are available for the remaining $N_5 \leq N_4$ participants.

6. The remaining N_5 participants start the final period. Independently, each have a 30% probability of dropping out before the end of the final period. Observations from the final period are available for the remaining $N_6 \leq N_5$ participants.

After 10000 runs, 94 of the designs implemented from $D(\mathbf{w}_6, \mathbf{w}_6)$ were disconnected, and 15 of the designs implemented from $D(\mathbf{w}_6, \mathbf{e})$ were disconnected. Although the absolute probability of a design becoming disconnected is small, it is desirable to avoid such an event, and the protected design offers a substantial reduction in risk.

4.5.2 Application

If the number of treatments t is odd ($t \geq 3$), then we can construct a protected and balanced design with $2t$ participants. Note that standard balanced designs for odd

t require $2t$ participants, although for some larger values of t there exist balanced designs with only t participants. The protected and balanced design $D(\mathbf{w}_t, -\mathbf{w}_t)$ given in Figure 4.1 can be used for all odd values of t ($t \geq 3$).

If t is even ($t \geq 4$), then some modification is needed to the designs most commonly used. In Section 4.4 we saw that for $t = 4$ we need 12 participants for a balanced design connected on the first two periods, and for even $t \geq 6$ we need $2t$ participants. It is currently common practice for experimenters to use several copies of a balanced t-participant design if t is even. For example, [21, p.198] explicitly recommend using a number of copies of a Williams square. As an alternative approach, we suggest that the protected design is used but with fewer copies. So if 24 participants are available and $s = 4$, then two copies of the protected and balanced four-treatment four-period 12-participant design should be used instead of six copies of the four-treatment four-period four-participant Williams square. Note that this is the recommendation made by Low *et al.* [28], using their robustness criteria and assuming dropout only in the final period.

If $t \geq 6$ then the protected and balanced $2t$-participant design consists of two balanced t-participant designs. Hence we do not necessarily need to have a number of participants which is a multiple of $2t$, as for balance we only need whole copies of both of the individual t-participant designs. For example, if we have 18 participants and $t = 6$ treatments, we could use the design $D(\mathbf{w}_6, \mathbf{w}_6, \mathbf{e})$, which would be formed of two copies of the left-hand square and one copy of the right-hand square of the design shown in Figure 4.2.

From Section 4.4, if t is even and $t \geq 6$ then we need the directed terrace $\mathbf{w}_6$ and a directed terrace $\mathbf{e}$ for $\mathbb{Z}_t$ of the form $\mathbf{e} = (0, 2, \ldots)$. Some examples of terraces of this form are given in Figure 4.6.

This is not an exhaustive list, and other examples for these values of t and for other suitable t may be found using the described 'lifting' method.

t	$\mathbf{e}$
6	$(0, 2, 1, 4, 5, 3)$
8	$(0, 2, 1, 5, 3, 6, 7, 4)$
10	$(0, 2, 3, 9, 6, 1, 4, 8, 7, 5)$
12	$(0, 2, 3, 7, 10, 5, 11, 4, 1, 9, 8, 6)$

Figure 4.6: Directed terraces $\mathbf{e} = (0, 2, \ldots)$ for $\mathbb{Z}_t$ when t is even

4.6 Alternative model and connectedness conditions

In this section, we discuss two modifications to the problem of selecting designs which are connected on the first two periods, and which are balanced. In Section 4.6.1, we consider removing the carryover effects from the model. In Section 4.6.2, we consider keeping the carryover effects in the model, but requiring only that the direct treatment effects are connected. Finally, in Section 4.6.3 we apply these results to the special case $t = 4$, which under the original conditions of the problem requires 12 (i.e. $3t$) participants for a balanced design which is connected on the first two periods (Section 4.4.3), whereas all other $t \geq 3$ require only $2t$ participants.

4.6.1 Removing the carryover effect from the model

If the carryover effect is removed from the model in Section 1.2 for a crossover design D, observation y_{ij} in period i and participant j will satisfy

$$E(y_{ij}) \quad = \quad \mu + \pi_i + \alpha_j + \tau_{d_{ij}}$$

and D may be considered as a row-column design with periods as rows and participants as columns.

When determining the connectedness of the first two periods of D, we can use the Wynn graph for determining the connectedness of row-column designs with two rows (Section 3.4, [46]). For the crossover design D, this defines a directed graph $W(D)$ where each vertex represents a treatment and each edge (k, l) represents a participant receiving treatment k in period 1 and treatment l in period 2. The connectedness of the

first two periods of D can then be determined by inspecting $W(D)$, as D is connected on the first two periods if the underlying undirected graph of $W(D)$ is connected, and if $W(D)$ contains an elementary unbalanced cycle (Theorem 3.3).

Theorem 4.4 If carryover effects are not included in the model, then for all $t \geq 3$ a t-treatment, t-period, t-participant crossover design exists which is connected on the first two periods, and which is universally optimal for the estimation of direct treatment effects.

Proof For $t \geq 3$, let the t-treatment, t-period, t-participant crossover design D be described by the $t \times t$ Latin square of the form shown in Figure 4.7, where the entry in the ith row and jth column is given by $(i-1)+(j-1) \mod t$. As the model does not include carryover effects, D may be considered as a row-column design, and so, as a Latin square, is balanced and so universally optimal for the estimation of direct treatment effects [22]. In order to determine whether D is connected on the first two periods, we consider the Wynn graph $W(D)$ of the first two periods of the design shown in Figure 4.7. Each treatment k $(k = 0, 1, 2, \ldots, t-1)$ appears exactly once in the first period, and is followed by treatment $k+1 \mod t$ in the second period. The directed graph $W(D)$ is then a cycle with t edges $(0,1),(1,2),(2,3),\ldots,(t-1,0)$. As this is connected and also an elementary unbalanced cycle, then $W(D)$ satisfies the conditions of Theorem 3.3. Consequently, D is connected on the first two periods under the model which excludes carryover effects. □

$$\begin{array}{ccccc} 0 & 1 & 2 & \ldots & t-1 \\ 1 & 2 & 3 & \ldots & 0 \\ 2 & 3 & 4 & \ldots & 1 \\ \vdots & \vdots & \vdots & & \vdots \\ t-1 & 0 & 1 & \ldots & t-2 \end{array}$$

Figure 4.7: Design D of Theorem 4.4, a $t \times t$ Latin square with entry $(i-1)+(j-1)$ mod t in row i and column j.

For all $t \geq 3$, removing carryover effects from the model therefore allows us to choose

designs with only t participants, which satisfy our requirements (optimal, and connected on the first two periods).

4.6.2 Removing the requirement for a design to be connected for carryover effects

Now we consider the problem where carryover effects remain in the model, but connectedness is only required for the direct treatment effects.

Theorem 4.5 Let D be a balanced t-treatment, t-period, s-participant crossover design which is connected for direct treatment effects on the first two periods. Then D is also connected for carryover treatment effects on the first two periods.

Proof Let k and k' be treatments in the crossover design D, with $k \neq k'$. As D is a balanced design, then from the definition in Section 2.1, k and k' both appear in the first period, allocated to participants j and j' respectively, and are followed in the second period by treatments l and l' respectively, such that $k \neq l$ and $k' \neq l'$. The difference between the two observations from participant j receiving treatments k in the first period and l in the second period satisfies

$$\begin{aligned} E(y_{2j} - y_{1j}) &= (\mu + \pi_2 + \alpha_j + \tau_l + \lambda_k) - (\mu + \pi_1 + \alpha_j + \tau_k) \\ &= (\pi_2 - \pi_1) + (\tau_l - \tau_k) + \lambda_k \end{aligned}$$

Similarly, the difference between the two observations from participant j' satisfies

$$E(y_{2j'} - y_{1j'}) = (\pi_2 - \pi_1) + (\tau_{l'} - \tau_{k'}) + \lambda_{k'}$$

So, subtracting this value for particpant j' from the value for participant j gives

$$\begin{aligned} E[(y_{2j} - y_{1j}) - (y_{2j'} - y_{1j'})] &= [(\pi_2 - \pi_1) + (\tau_l - \tau_k) + \lambda_k] - [(\pi_2 - \pi_1) + (\tau_{l'} - \tau_{k'}) + \lambda_{k'}] \\ &= (\tau_l - \tau_k) - (\tau_{l'} - \tau_{k'}) + (\lambda_k - \lambda_{k'}) \end{aligned}$$

As the first two periods of D are connected for direct treatment effects, then $\tau_k - \tau_l$ and $\tau_{k'} - \tau_{l'}$ are estimable. Therefore the elementary contrast $\lambda_k - \lambda_{k'}$ is also estimable in the first two periods of D, and so the first two periods of D are connected for carryover effects. □

For all $t \geq 3$, keeping carryover effects in the model but removing the requirement for carryover treatment contrasts to be estimable reduces to the original problem, as in balanced designs connectedness for direct treatment effects on the first two periods implies connectedness for carryover treatment effects on the first two periods.

4.6.3 Implications for the case $t = 4$

As the main results for this chapter do not apply for the case $t = 4$, it is of interest to consider what results can be obtained with alternative model specifications or connectedness conditions. In Section 4.6.1 we showed that if carryover effects were removed from the model, a balanced t-treatment t-period design connected on the first two periods can be found with only t participants for $t \geq 3$. Such a design for $t = 4$ is shown in Figure 4.8. Without carryover effects in the model, this design is balanced. Additionally, the design is connected on the first two periods, as can be determined from the Wynn graph in Figure 4.9.

0	1	2	3
1	2	3	0
2	3	4	1
3	0	1	2

Figure 4.8: A crossover design for $t = 4$.

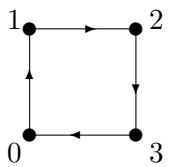


Figure 4.9: The Wynn graph for the first two periods of the design in Figure 4.8.

In Section 4.6.2 we considered the effect of keeping the carryover effects in the model, but removing the requirement that the carryover treatment contrasts be estimable.

However, in Theorem 4.5 we showed that if the design is balanced, and if the direct treatment contrasts are estimable on the first two periods, then the carryover treatment contrasts are necessarily also estimable on the first two periods. Consequently, attempting to apply this adjusted criteria to the case $t = 4$ will not represent any real change, and so $3t = 12$ participants will still be required for a balanced design which is connected on the first two periods.

Chapter 5

Survey of crossover trials reported in the general scientific literature

5.1 Introduction

It is important to investigate how trials with crossover designs are implemented in applied research, so that the nature of any translational gap between the theory of design of experiments and the reality of the use of such designs can be better understood. A previous review of the methodological aspects of crossover trials has been performed, which focussed on reports of crossover trials published during December 2000 and indexed on PubMed [31]. That review included 116 papers reporting a total of 127 crossover trials, of which 72% were trials involving only two treatments, and of the AB/BA design. It was reported that although most (70%) of the trials either included or discussed a washout period, carryover effects were only discussed in 29% of cases. Overall, the review authors found that key methodological issues were often absent in reports of crossover trials.

The review of the use of crossover designs that we have undertaken and describe in

this chapter differs in scope to the review by Mills *et al* [31]. Firstly, as the crossover designs of interest elsewhere in this thesis are those with three or more treatments, we did not consider crossover trials involving two treatments. Secondly, as many trials involving a crossover design are found outside of the field of biomedical research, we searched for publications indexed in the general scientific database Web Of Science. We considered papers published during the whole of 2009.

5.2 Methods

The Web Of Science abstract database was searched for the following terms in the title, abstract or keywords: ((trial OR experiment) AND (crossover OR cross-). Restricting the search to items published during 2009, 3165 abstracts were identified. When these were restricted to articles only, and then to those published in English, 2554 abstracts remained under consideration. Each of these abstracts was inspected in the Web Of Science interface in order to determine whether the terms 'crossover' or 'cross-' were used in the appropriate context. When abstracts are viewed in Web Of Science following a search, the search terms in the abstract, title and keywords are highlighted: this allows the context in which the search terms are used to be determined accurately (Figure 5.1). Following this process, 1851 abstracts were judged to be unrelated to crossover trials and so were disregarded, with 703 abstracts remaining under consideration. Each of these abstracts was then considered in more detail, to establish whether the corresponding article was eligible for inclusion in this review.

Of the 703 abstracts under close consideration, 82 were excluded as they were found to not be crossover designs, and 463 were excluded as they described crossover trials with only two treatments. The number of studies remaining at this point and for which the full-length articles were to be considered was 158: details of these are given in the review bibliography.

Data from the articles selected for inclusion was entered into a form in an Access database (Figure 5.2). Information collected included basic information about the size

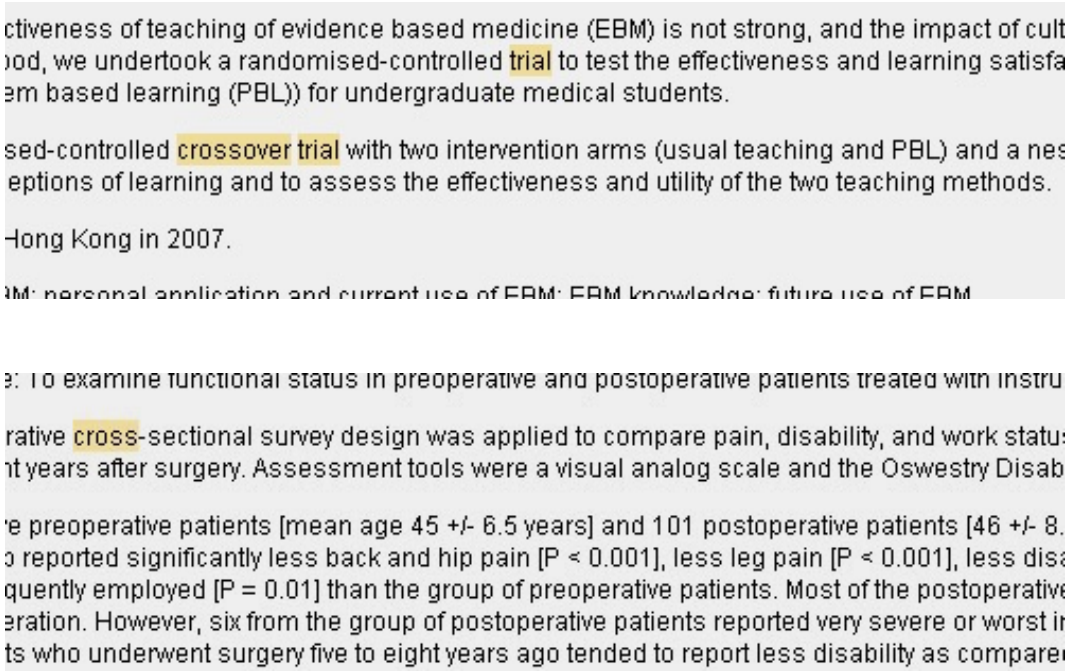


Figure 5.1: Partial abstracts displayed in the Web Of Science interface with search terms highlighted.

of the trial (number of treatments, number of periods, number of sequences, number of participants), the general research area, and information about the design of the trial, the analysis, and whether concepts such as carryover or a washout period were mentioned.

5.3 Results

5.3.1 Characteristics of included studies

A total of 158 study articles were selected for inclusion on the basis of the two-stage abstract screening process. Of these, 14 were found to be crossover trials with two treatments, 14 were found to not be crossover trials, two reported on studies that were not fully randomised, three were duplicate studies represented elsewhere in the sample, and one article could not be obtained. These 34 studies were excluded, leaving a sample of 124 study articles to be included (Figure 5.3). Some study articles reported on more than one crossover design: in these cases, the first design mentioned in the article was included. The characteristics of the included crossover trials are summarised in Figures 5.4 and 5.5.

The included studies covered a wide range of research areas, including pain medication, pharmacology, nutrition, and livestock management. Most involved human research participants, with 65 (52%) of trials involving healthy volunteers and 45 (36%) involving patients.

Most of the included trials involved three treatments (66, 53%) or four treatments (42, 34%) (Figure 5.4). The maximum number of treatments in a trial was nine. Most trials took place over three periods (63, 51%) or four periods (41, 33%), and only four trials had a number of periods that was not equal to the number of treatments (Figure 5.5). The smallest trial involved three participants, and the largest involved 207 participants, with the median (interquartile range) of 20 (12 to 33) participants.

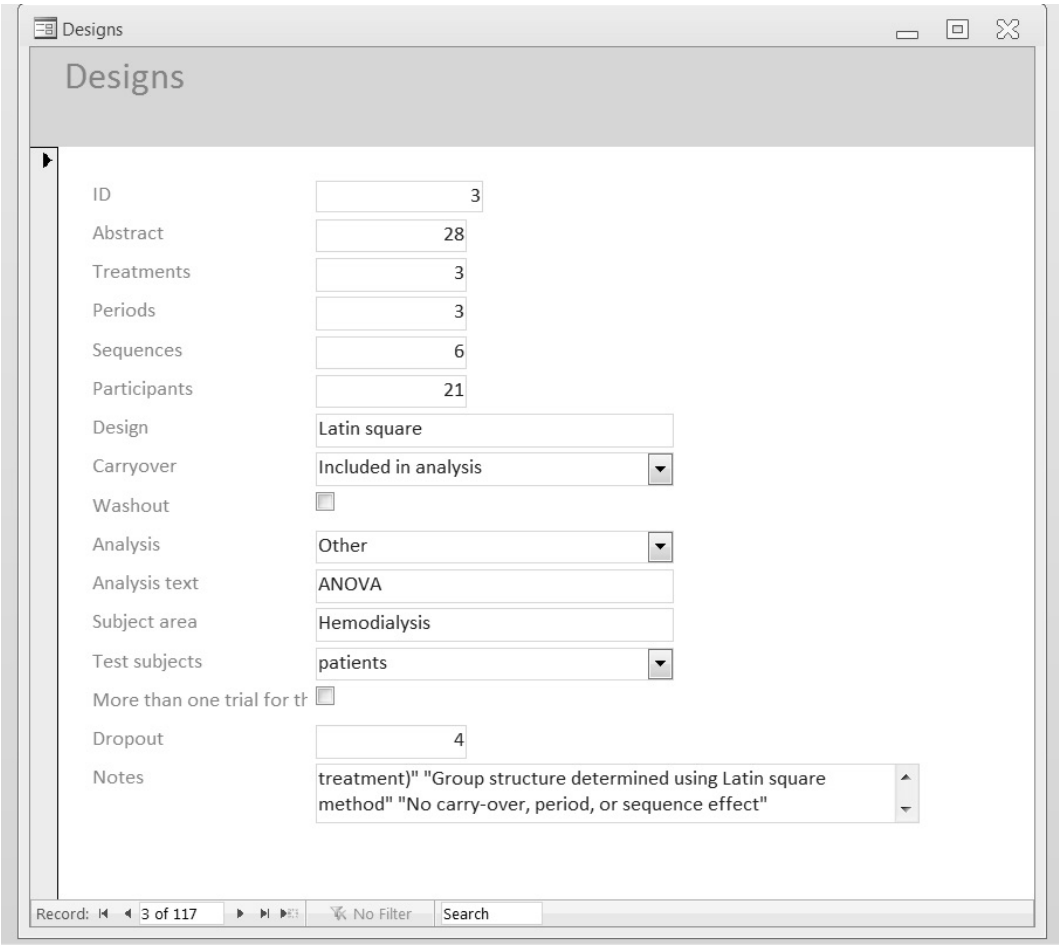


Figure 5.2: Access database form used for capturing data on included studies.

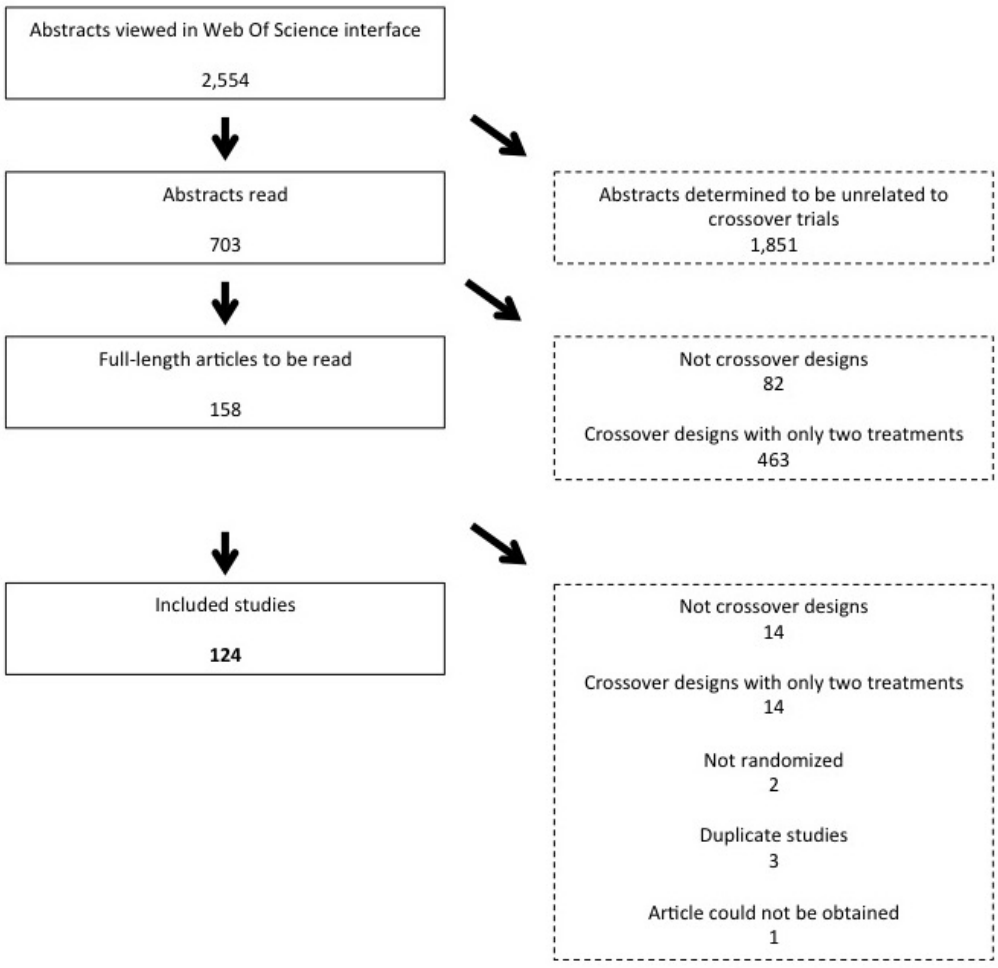


Figure 5.3: Flowchart showing studies selected for inclusion in review.

Characteristic		Number and % of trials	
Washout	included	83	67%
Carryover	included in analysis	14	11%
	mentioned only	14	11%
Sample size	included details of calculation	31	25%
Participants			
type	healthy volunteers	65	52%
	patients	45	36%
	animals / livestock	13	10%
	other	1	1%
number	*median*	*20*	
	interquartile range	*12 to 33*	
	range	*3 to 207*	

Figure 5.4: Characteristcs of the included crossover trials.

		Periods								
		2	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**Total**
Treatments	**3**	2	63	0	0	0	0	0	1	66
	4	1	0	41	0	0	0	0	0	42
	5	0	0	0	7	0	0	0	0	7
	6	0	0	0	0	6	0	0	0	6
	7	0	0	0	0	0	1	0	0	1
	8	0	0	0	0	0	0	1	0	1
	9	0	0	0	0	0	0	0	1	1
	Total	3	63	41	7	6	1	1	2	**124**

Figure 5.5: Dimensions of the included crossover trials. Shaded cells indicate equal numbers of treatments and periods.

5.3.2 Design

There was wide variation in how the study design was addressed, with the concept of a design with different treatment sequences mentioned in 37 (30%) of studies. Few reported details of specific crossover designs used, with most giving no more information than that participants had been randomly assigned to treatments. However, Latin squares were explicitly mentioned in 14 (11%) of studies, and several studies reported the exact treatment sequences used.

5.3.3 Carryover effects

Most (83, 67%) of studies reported the use of a washout period between treatment periods. Most did not address the issue of carryover or residual effects, although 14 (11%) accounted for carryover effects in the analysis, and a further 14 (11%) mentioned the concept of carryover effects.

5.3.4 Sample size estimation

Details of the calculation performed to obtain the sample size estimation were reported in 31 (25%) of studies, including power, significance level, effect size, and (in eight studies) anticipated participant droput. Additionally, eight studies reported having undertaken some form of sample size estimation, but did not report full details. Four further studies had no mention of sample size estimation, but included remarks that the sample size may have been too small.

5.3.5 Dropout

There was no report of participant dropout in 73 (59%) of trials. In the 51 studies where dropout was reported, the median (interquartile range) proportion of participants dropping out was 13% (8% to 20%). The largest reported dropout rate in a trial was 40%.

5.4 Discussion

This exhaustive review of the use and reporting of crossover trials across a range of scientific disciplines during a single year has provided a useful snapshot of the use of this type of trial. We have found evidence that crossover trials with three or four treatments are are actually used in practice, with occasional trials involving much larger numbers of treatments. What is particularly clear from the studies considered here is that the implementation of crossover designs in applied research goes far beyond the AB/BA design which many may consider to be synonymous with the term 'crossover design'. There is evidence that some quite complex crossover designs are being undertaken, with large numbers of treatments.

As observed in the review by Mills *et al.* [31], reporting of information such as design and the consideration of carryover effects is limited. However, in concordance with their observations, we also found that the majority of researchers have considered washout periods when implementing a trial with a crossover design.

Participant dropout is shown to be a concern, with one trial experiencing a 40% dropout rate. Although most trials had no report of dropout, this does not necessarily indicate an absence of dropout. While some studies provided full information about the flow of their participants through the design, others reported only the number who completed the trial, rather than also the number who were initially randomized. Although no studies reported problems with estimating treatment contrasts due to dropout, it is plausible that studies where there have been serious problems of this nature may not be published.

This review only provides a limited snapshot of the use of crossover designs by researchers, as many trials, particularly ones involving small numbers of particpants, may not result in publication. It is also important to consider that what is published on a study may not reflect exactly what the researchers wish to say about a study [9]. Possible reasons for this might include authors removing text to keep within the word limit for a manuscript submission, or on acceptance a journal may request that some

of the more technical detail in a paper is removed. Consequently researchers may have considered issues such as choosing a balanced design, and carryover effect, but the information has not been published. Another limitation of this review concerns the search terms used. Restricting to abstracts where terms such as "crossover" have been used was a practical approach to the otherwise wide-ranging search, but it does mean that all of the included studies are those which the researchers have explicitly identified as being crossover designs. Other studies may have used designs that would be recognisable as crossover designs, but may be referred to as something else if the researchers were unfamiliar with this type of design. Consequently, in the sample of studies captured from our search, researchers more familiar with crossover designs may be overrepresented. It is of interest that in the review by Mills *et al.* [31], one third of included studies did not use the term "crossover" in either title or abstract.

In summary, although our sample may not be entirely representative of all crossover trials actually being performed, it is clearly representative of crossover designs as reported in the scientific literature. Crossover trials are used in many different disciplines, with varying degrees of standards of reporting. Although a small proportion of researchers appear to undertake crossover trials while considering carefully the relevant methodological issues, another group of researchers seem to undertake potentially quite complex trial designs with little or no reference to the supporting methodology. So that the standard of such trials and their reporting might improve in the future, it is this group that we will consider in Chapter 6.

Chapter 6

Crossover design methodology and crossover trials in practice

6.1 The translational gap

There is a large amount of research which is concerned with the design of crossover trials, and, as we have seen in Chapter 5, there is a wide range of research situations in which trials with a crossover design with three or more treatments are used. However, what we have seen in Chapter 5 suggests that the relevant methodological issues may not be considered for all such studies. This translational gap needs to be adressed, for a number of reasons, and not only for the specific case of crossover trials. Firstly, the methodology of a trial must be correct for the results to be considered valid. Secondly, performing trials which are methodologically substandard may lead to problems such as nonestimable treatment contrasts, or contrasts estimated with unnecessarily high degrees of uncertainty. This can lead to wasted time and resources. Additionally, it is important to consider the ethical implications of recruiting participants into a trial which has been poorly designed and so has a higher risk of failure than if it had been designed appropriately.

6.2 Existing barriers to closing the gap

There are a number of issues that may be preventing the effective use of crossover design methodology in applied research. One possibility is that often a statistician may not be consulted about a trial until it has been completed and the results are to be analysed. This enduring problem was acknowledged by R.A. Fisher in an address to the Indian Statistical Congress in 1938:

> "To call in the statistician after the experiment is done may be no more than asking him to perform a postmortem examination: he may be able to say what the experiment died of."

The audience that methodology researchers have for their work should also be considered: they may write up their research primarily for other methodological researchers, so that their work is not accessible to those who might apply it. It may be that methodology researchers will make developments in their work in response to an applied problem that they have perhaps been consulted on, but the communication between the methodology researcher and a specific applied researcher at that time may not be replicated more generally. So a methodological development is communicated only to a number of other methodology researchers by publication, and to one applied research by the consultancy on the original problem.

It is likely that applied researchers will work within a particular research area, but use a number of different techniques, tools and experimental designs. Consequently, when faced with an unfamiliar study design, a lack of familiarity with the terms used in describing aspects of the design may be a barrier to accessing the correct information to be able to design the study appropriately. As an example of this, in the review of crossover trials in Chapter 5, some studies that were not included had used the term 'crossover design' to describe trials which were not crossover designs as generally defined by statisticians.

In the context of clinical trials for pharmaceutical interventions where the aim is to gain approval from some agency such as the Food and Drug Administration in the US, the trial design must adhere to very specific prescribed guidelines. As an additional barrier, it could be argued that this encourages a conservative culture with respect to trial design and statistical methodology, even in situations where the strict requirements for FDA approval do not apply. For example, pharmaceutical companies may be reluctant to engage with unfamiliar methodological issues in their wider research, as their concerns are primarily with meeting FDA approval in those situations where it is relevant.

6.3 Possible solutions

We have identified potential three barriers to closing the gap between trial design methodology and trial implementation by applied researchers: lack of communication from methodology researchers to the applied researcher community; lack of familiarity with specific design terms and issues making finding the right information more difficult; and a conservative culture resistant to change. The third of these is the most difficult to change, so we will consider potential solutions to barriers related to communication.

A possible approach is that methodology researchers could be encouraged, or required, to make available plain language summaries for methodological papers so that applied researchers can easily establish whether the methodology might be useful for them, providing a route for future collaborations. Additionally, methodology researchers could write up a more practical example-led version of any methodological paper for the clinical or applied literatue.

Furthermore, information about appropriate experimental design could be made available through a website which could act as a dynamic repository of designs. There are currently a number of websites that aim to assist researchers with experimental design choices, but many require the researcher to be familiar with the terminology around the theory of experimental design. Examples are a website that includes a

question-based selection key for choosing a design (http://dawg.utk.edu/choose.htm, accessed March 2015), and a website for generating nested or crossed block designs (http://www.expdesigns.co.uk, accessed March 2015). As one of the potential barriers to accessing information may be unfamiliarity with the terminology, an appropriate interactive graphical interface would allow the applied researcher to search the site by characteristics of their design problem without requiring knowledge of the precise terminology used. Additionally, as a potential application of some of the visual representations of designs presented here in Chapter 3 could be to further demystify experimental design methodology, similar visual representations could be included on such a website.

Chapter 7

Discussion

7.1 Conclusions

From Chapters 2 and 3 we have seen that there are methods by which designs can be constructed, compared and discussed by non-specialists, for example by considering a visual representation of the design. In Chapter 4 we introduced a straightforward characteristic that a crossover design should have to reduce the risk of it becoming disconnected due to participant dropout. Using designs which are connected on the first two periods still allows the use of familiar balanced designs; just making a slightly different, and still straightforward, choice of design will have no other impact than to reduce the risk of a disconnected design. In Chapter 5 we saw that, despite there being easily attainable benefits to good design choices, there was great variability in the implementation of crossover designs in applied research, with many studies apparently not engaging with the concept of an experimental design at all beyond random assignment to treatments. In Chapter 6 we discussed why that might be, and what might be done to improve communication so that more applied researchers can benefit from the work of methodological researchers.

7.2 Further issues resulting from participant dropout

In this thesis, we have focussed on the problem of avoiding disconnected crossover designs following participant dropout. However, there are other consequences which may arise when planned observations are not available in a crossover trial, which we will briefly discuss here. One possible consequence, which we have considered in Section 4.1, is that the efficiencies of the estimators of the treatment contrasts may be markedly reduced compared to the efficiencies in the planned design. Another important possible consequence of data being missing is that, due to the reasons for the data being missing, the resulting estimates may be biased.

Various analysis approaches may be used in the presence of missing data, the appropriateness of which vary according to the assumed underlying missingness mechanism of the data. Missing data mechanisms are generally classified as missing completely at random (MCAR), missing at random (MAR), or missing not at random (MNAR) [35, 26]. Under the MCAR assumption, the probability of an observation being missing is independent of any observed or unobserved values. Under the MAR assumption, the probability of an observation being missing depends only on the observed values. Under the MNAR assumption, the probability of an observation being missing depends on both observed and on unobserved values.

Where data are assumed to be MCAR, a complete case analysis would be appropriate: only those participants without any unobserved data would be included. If the underlying missingness mechanism is MAR, a complete case analysis would induce bias. However, other approaches such as multiple imputation could be used, where missing values are predicted using existing values elsewhere in the dataset [7]. In the case of a MNAR missingness mechanism, other approaches are needed in order to avoid bias.

A justification for an assumption of a MNAR missingness mechanism in a crossover trial is that participants who have a poorer experience in one period may be more likely to drop out in the next period [29]. Where the missingness mechanism is MNAR, an inverse probability weighting approach can be used to avoid bias [34]. This method has

been applied to a two-treatment two-period crossover trial with an AB/BA design [19], where it was observed that there were practical difficulties involved in the use of the method in this setting.

It has been argued that for a two-treatment crossover design, even if the missingness mechanism is MNAR, a complete case analysis may in fact be the most appropriate method [30]. The justification for this is that a complete case analysis compares the two treatments within participants who are able to tolerate both treatments: in generalising study results, this comparison would only be relevant to those who could tolerate both treatments.

7.3 Future research

The survey of designs in Chapter 5 has provided a overview of what types of crossover designs are being used and how the associated methodological issues are being addressed. In order to better understand the gap between crossover design methodology and implementation in research, and to investigate how it can be addressed, it would be beneficial to find out from applied researchers about their knowledge of experimental design, and where they get their information from. One possible way to do this would be to identify a number of published studies with crossover designs in a recent given time period, perhaps using a similar approach to that in Chapter 5, and contact the study authors with some survey questions on their experience of using a crossover design. This would allow a wide range of views to be canvassed. Additionally, it could be determined whether design details were in fact being considered by researchers undertaking experiments, but not being included in resulting publications.

An alternative approach would be to undertake some qualitative interviews with a smaller selection of applied researchers, which would be more resource-intensive, but may allow for a deeper understanding of the issues involved, and better inform the potential development of resources to improve researcher engagement with the methodology of crossover and other designs.

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