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TWO-STAGE SCREENING WITH COMBINATORIAL DESIGNS FOR EVENT PROBABILITIES $p \geq 0.01$

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Abstract

This paper elaborates on the idea of using partial Steiner systems as two-stage screening schemes. Recently was showed that the expected number E of tests per sample (a natural objective function in this context) is almost minimal in a hypercube scheme with r^k samples (for suitable k and r dependent on the event probabilities p). Our method achieves the same E with much fewer samples in the scheme, which makes it - unlike the hypercube method - useful in practice even for small p. The needed screening schemes are given explicitly which make it easy to implement an optimal two-stage screening for known p.

1. Introduction

In searching for rare binary events (with probability p), for example in blood testing or in polymerase chain reaction (PCR)-based methods, testing each single sample may be ineffective, especially when given a large number N of samples. The simple idea of two-stage screening is to test mixtures in the first stage and only a reduced number of samples in the second stage. For this we assume that the samples are identically

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independently Bernoulli-distributed with event probability p, and that a mixture of samples is positive if and only if at least one of the samples contained in it is positive. Furthermore, we assume perfect specifity and sensitivity of the analytical test used for samples and mixtures. Our objective is to minimize the expected number E of tests per sample needed to identify all positive samples. In the last section we discuss the consequences if the assumption of perfect sensitivity fails. In addition to two-stage-screening, other methods using more than two stages have been developed, however their complex organisation greatly reduces their practical value (Phatarfod and Sudbury [14]).

The general screening procedure is the following: Each sample is divided into $k+1$ parts. One part from every initial sample is reserved for the second stage. The other $k \cdot N$ parts are used to form mixtures. Each mixture contains r parts - of course from different initial samples. At the first stage we test these mixtures. At the second stage only those initial samples are retested which are contained in k positive mixtures.[1] The simplest and oldest example that illustrates two-stage screening is the Dorfman scheme. Dorfman [10] presented the idea of testing the mixture of r samples. If such a mixture is positive, then we waste one test, but if it is negative, then all samples contained must be negative, and accordingly we save $r-1$ tests. The expected number of tests needed for one mixture in the Dorfman scheme $D(r)$ is $1+r(1-q^r)$ with $q = 1-p$. The expected number of tests per sample needed is accordingly:

$$E = \frac{1+r(1-q^r)}{r} = r^{-1} + 1 - q^r,$$

if the number of samples N divided by r is an integer. (Here $k = 1$.)

Phatarfod and Sudbury [14] investigated a two-dimensional generalization of this procedure. Now $k = 2$, and each sample is divided into three parts. Mixtures are formed from parts of the samples contained in one row or one column of a quadratic scheme. At the second stage one has to test only the samples in two mixtures which have tested positive.

[1] We do not consider the possibility of concluding after the first stage that a sample is positive (details for this are in Berger and Levenshtein [5, 6]), because of possible measurement errors.

Here is an example of a 6×6-array scheme: Enumerate the 36 samples $S_{ij} (i,\ j = 1, \ldots, 6)$. The mixture L_i contains one part of all the samples in the i-th line. The mixture C_j consists of parts of all the samples in the j-th column.

S_{11}	S_{12}	S_{13}	S_{14}	S_{15}	S_{16}	L_1
S_{21}	S_{22}	$\boxed{S_{23}}$	S_{24}	$\boxed{S_{25}}$	S_{26}	L_2
S_{31}	S_{32}	S_{33}	S_{34}	S_{35}	S_{36}	L_3
S_{41}	S_{42}	$\boxed{S_{43}}$	S_{44}	$\boxed{S_{45}}$	S_{46}	L_4
S_{51}	S_{52}	S_{53}	S_{54}	S_{55}	S_{56}	L_5
S_{61}	S_{62}	S_{63}	S_{64}	S_{65}	S_{66}	L_6
C_1	C_2	C_3	C_4	C_5	C_6	

If at the first stage the mixtures L_2, L_4, C_3 and C_5 are tested positive, only S_{23}, S_{25}, S_{43} and S_{45} must be tested at the second stage.

Berger et al. [7] extended the investigation from square array schemes to k-dimensional hypercube schemes for arbitrary $k \in N$. Instead of rows and columns, lines parallel to the edges of the hypercube are used. Let each such line contain r elements. At the first stage we test the mixtures corresponding to these lines. At the second stage, only samples at the intersection of k positive lines are tested. Berger et al. [7] were able to prove that hypercube schemes are nearly optimal with respect to the expected number E of needed tests per sample, if size r and dimension k are chosen to fit p. They obtain

$$E = \frac{k}{r} + p + (1 - q^{r-1})^k q.$$

For Table 1 we calculate the optimal E, k and r depending on p with Maple 6 (Monagan et al. [13]). It shows that the number r^k of samples in an optimal hypercube scheme increases very quickly with decreasing event probability p. For applications it is important to reduce the number of samples in the scheme while keeping E optimal.[2] The key for this is our generalization which treats hypercube schemes as special cases of the

[2] Berger et al. [7] already notice this possibility and give two examples. They do not investigate the subject systematically, in particular the reduced number of samples in the given schemes is not minimal.

schemes corresponding to partial Steiner systems of degree r (defined later in Subsection 2.1). In all these schemes E is optimal and the new task is to minimize the number of samples b in the scheme. We imagine a situation in which the user collects new samples continually and can wait until this b is reached.[3]

The second section will explain the link between screening schemes and partial Steiner systems and present all definitions needed from combinatorial design theory. In the third section the needed (partial) Steiner systems are described explicitly which make it easy to implement an optimal two-stage screening for known event probability p. The fourth section deals with a further reduction of the number of samples in the scheme by leaving out single blocks in the case $k = 2$. In the last section we discuss practical questions concerning the situation where the event probability is not precisely known.

2. Modelling as a Combinatorial Design

2.1. The basic combinatorial concept: partial Steiner systems

Here we recall some simple statements about (partial) Steiner systems.

Definition. A design is a pair $(V, \mathcal{B})$, where V is a set consisting of v points and $\mathcal{B}$ is a collection of subsets of V called *blocks*. A design is called a *Steiner system* $S(2, k; v)$ if each block contains k points and each pair of points is contained in exactly one of the blocks.[4]

In the following, if not specified otherwise, we normalize to $V = \{1, ..., v\}$. For convenience of the reader we give the following well-known Lemma with proof.

[3] Berger and Levenshtein [6] also use partial Steiner systems for screening procedures, but they do not try to minimize b. Instead they got asymptotic statements for large b (Berger and Levenshtein[5, 6]; Levenshtein [12]).

[4] The parameter 2 refers to the fact that each pair of points is contained in exactly one block. An $S(c, k; v)$ is a design where each block contains k points and each subset of V with c elements is contained in exactly one block (Beth et al. [8]). We do not need these more general Steiner systems in this paper.

Lemma. *Each point $w \in V$ of a Steiner system is contained in exactly $r = \frac{v-1}{k-1}$ blocks. The number b of blocks in $\mathcal{B}$ is $b = \binom{v}{2} \Big/ \binom{k}{2}$.*

Proof. Consider $w \in V$. Then the number of pairs containing w is $v - 1$, one for each other element of V. Let r be the number of blocks containing w. Each of these blocks has $k - 1$ further elements and since each pair appears only once, these are all different and w is contained in $(k-1)r$ pairs. It follows $(v-1) = r(k-1)$ and so r is independent of w and as stated. The other assertion is obvious.

Definition. The design $(V, \mathcal{B})$ is called a *partial Steiner system* if each block contains k points and each pair of points is contained in at most one of the blocks. We say that the pair $(V, \mathcal{B})$ is a *partial Steiner system* $PS(2, k, r; v)$ of degree r if in addition each point of V is contained in exactly r blocks.[5]

Remark. In the last case the number of blocks b is $b = \frac{vr}{k} \le \frac{\binom{v}{2}}{\binom{k}{2}} = \frac{(v-1)v}{(k-1)k}$ and so $r \le \frac{v-1}{k-1}$ and $v \ge (k-1)r + 1$. Here, equality holds if and only if the partial Steiner system is a Steiner system.

One possible method to represent a partial Steiner system is an incidence matrix. The matrix has v rows corresponding to the points of V and b columns corresponding to the blocks. We have $a_{ij} = 1$ if and only if the i-th point is contained in the j-th block and $a_{ij} = 0$ otherwise. Of course the incidence matrix depends on the labelling, but it is unique up to the permutation of rows or columns. (For an example see Figure 1.) Another way to give a (partial) Steiner system is to describe the blocks by their elements. So to describe the block corresponding to the third column

[5] Partial Steiner systems of degree r are regular graph designs with $\lambda_1 = 0$ and $\lambda_2 = 1$.

of the $S(2, 2; 7)$ in Figure 1 we write $B_3 = \{1, 4\}$.

$$\begin{pmatrix}
1&1&1&1&1&1&0&0&0&0&0&0&0&0&0&0&0&0&0&0&0\\
1&0&0&0&0&0&1&1&1&1&1&0&0&0&0&0&0&0&0&0&0\\
0&1&0&0&0&0&1&0&0&0&0&1&1&1&1&0&0&0&0&0&0\\
0&0&1&0&0&0&0&1&0&0&0&1&0&0&0&1&1&1&0&0&0\\
0&0&0&1&0&0&0&0&1&0&0&0&1&0&0&1&0&0&1&1&0\\
0&0&0&0&1&0&0&0&0&1&0&0&0&1&0&0&1&0&1&0&1\\
0&0&0&0&0&1&0&0&0&0&1&0&0&0&1&0&0&1&0&1&1
\end{pmatrix}$$

Figure 1. The incidence matrix for a Steiner system $S(2, 2; 7)$: The sum in each column is $k = 2$, the sum in each row is $r = \frac{v-1}{k-1} = 6$. In the special case $k = 2$ it is enough to test whether the columns are different to make sure that it is an incidence matrix of a partial Steiner system.

The connection to screening procedures: The motivation to use (partial) Steiner systems for two-stage screening is the observation, that Berger's hypercube schemes of dimension k and length r correspond to partial Steiner systems $PS(2, k, r; v)$ of degree r with $b = r^k$ blocks. Let $\mathcal{S}$ be the set of samples S_i and $\mathcal{M}$ be the set of mixtures M_i. Then

- Each mixture M_i consists of r samples.
- Each sample S_j occurs in exactly k mixtures.
- Two mixtures have at most one sample in common.

We define the corresponding partial Steiner system as following: Let the pointset $V = \{1, \ldots, v\}$ consist of one i for each mixture M_i, and let the blocks B_j correspond to the samples S_j in such a way, that a point belongs to a block if and only if the sample is contained in the mixture:

$$i \in B_j \Leftrightarrow S_j \in M_i.$$

The bijection $\mathcal{M} \to V$, then induces a bijection $\mathcal{S} \to \mathcal{B}$ and the above mentioned properties of $(\mathcal{M}, S)$ correspond exactly to the definition of the partial Steiner system $(V, \mathcal{B})$ of degree r.

Definition. Analogously, we can define a two-stage screening corresponding to a partial Steiner system $(V, \mathcal{B})$. If $\mathcal{B} = \{B_1, \ldots, B_b\}$, then we work with b samples $\mathcal{S} = \{S_1, \ldots, S_b\}$ and define a mixture M_i for each $i \in V$ according to the rule: $S_j \in M_i \Leftrightarrow i \in B_j$. At the first stage we test these mixtures. At the second stage we test the samples which are only in positive tested mixtures. For a first example see Figure 2.

$\boxed{1}$	2	3	4	$\boxed{5}$	$\boxed{6}$	7	8	9	$\boxed{10}$	$\boxed{11}$	12	13	14	15	16	17	18	19	20	$\boxed{21}$	
1	1	1	1	1	1																M_1
1						1	1	1	1	1											M_2
	1					1					1	1	1	1							M_3
		1					1				1				1	1	1				M_4
			1					1				1			1			1	1		M_5
				1					1				1			1		1		1	M_6
					1					1				1			1		1	1	M_7

Figure 2. An example of the screening procedure for the Steiner system $S(2, 2; 7)$ in Figure 1. Let the samples be 1, ..., 21. Each column of the matrix corresponds to a sample, each line to a mixture. The mixture M_i contains the samples j if and only if the entry a_{ij} in the i-th line and the j-th column is 1. For instance, M_6 contains the samples 5, 10, 14, 17, 19 and 21.

If at the first stage the mixtures M_1, M_2, M_6 and M_7 have tested positive, then at the second stage, the samples 1, 5, 6, 10, 11 and 21 must be tested, because they belong only to mixtures which have tested positive.

Example. Choose the Steiner system $S(2, 3; 27)$ for $k = 3$ and $r = 13$ in Subsection 3.3 below. The screening scheme works with 117 samples. Enumerate them by $S_{i,j}$ with $i = 1, \ldots, 9$, $j = 0, \ldots, 12$. At the first stage we form 27 mixtures, say $M_{(a,b)}(a = 0, \ldots, 12, b = 0, 1)$ and M_∞, where the mixture $M_{(a,b)}$ contains the samples $S_{i,j}$ for which $a_b \in B_{i,j}$. For example, the mixture $M_{(7,0)}$ contains $S_{1,8}$ since $B_{1,8} = \{1_1, 10_1, 12_0\} + 8 = \{9_1, 5_1, 7_0\}$ contains 7_0. (The operations are defined in Subsection 3.3, we work modulo 13, so $18_1 = 5_1$.) M_∞ contains the samples $S_{i,j}$ for which $\infty \in B_{i,j}$, i.e., the samples $S_{9,i}$ for $i = 0, ..., 12$.

We give explicitly the mixtures $M_{(7,0)}$ and $M_{(6,1)}$:

$$M_{(7,0)} = \{S_{1,8}, S_{2,9}, S_{3,10}, S_{4,0}, S_{5,3}, S_{6,12}, S_{7,6}, S_{7,4}, S_{7,11}, S_{8,5}, S_{8,1}, S_{8,2}, S_{9,7}\},$$

$$M_{(6,1)} = \{S_{1,5}, S_{1,9}, S_{2,4}, S_{2,12}, S_{3,3}, S_{3,2}, S_{4,0}, S_{4,11}, S_{5,10}, S_{5,7}, S_{6,1}, S_{6,8}, S_{9,6}\}.$$

$M_{(7,0)}$, M_∞ and $M_{(6,1)}$ cannot be the only mixtures tested positive, because there is no sample that is only contained in these three mixtures. Each sample is contained in three mixtures and two mixtures have at most one sample in common. Therefore, it is also impossible that precisely four mixtures are tested positive. Now assume that exactly the mixtures $M_{(7,0)}$, $M_{(6,1)}$ and $M_{(8,1)}$ are tested positive at the first stage. At the second stage all the samples that are only in mixtures which have tested positive must be tested. So, in this case the only sample to be tested at the second stage is $S_{4,0}$.

Proposition.[6] *The expected number of tests per sample E for the screening corresponding to a partial Steiner system PS(2, k, r; v) of degree r is*

$$E = \frac{k}{r} + p + (1 - q^{r-1})^k q \quad (q = 1 - p).$$

Proof. As indicated in the introduction, we use the assumption that the test has perfect specifity and sensitivity for the mixtures, in the sense that all and only the positively tested mixtures contain at least one part, that is positive. Since $v = \frac{bk}{r}$, there are $\frac{k}{r}$ tests per sample at the first stage. The probability $P(T_S)$ that a sample S (corresponding to the block B) must be tested at the second stage (T_S) is

$$P(T_S) = P(T_S \wedge S = +) + P(T_S \wedge S = -)$$
$$= P(T_S \mid S = +)P(S = +) + (T_S \mid S = -)P(S = -)$$
$$= 1 \cdot p + (1 - q^{r-1})^k q.$$

If the sample is positive, then it is clear (because of the perfect sensitivity) that it is contained only in positively tested mixtures and therefore must be tested at the second stage, so $P(T_S \mid S = +) = 1$. If it is negative, then the conditional probability that a mixture M containing S is negative is q^{r-1} since the remaining $r - 1$ samples of the mixture are independent and each of them is negative with probability q. Therefore, $P(M = + \mid S = -) = 1 - q^{r-1}$. The $k(r - 1)$ samples that are mixed with S in all k mixtures $M_1, ..., M_k$ containing S are distinct. Otherwise there would be samples $S = S_i$ and S_j that are in mixtures M_l and M_m. In the corresponding partial Steiner system, $\{l, m\}$ would then be an element of $B_i \cap B_j$ which contradicts the definition of a partial Steiner system. Therefore, the k events $(M_n = +)(n = 1, ..., k)$ are independent and

$$P(M_1 = + \wedge \cdots \wedge M_k = + \mid S = -) = (1 - q^{r-1})^k.$$

Because of perfect specifity we get $P(T_S \mid S = -) = (1 - q^{r-1})^k$ and $E = \frac{k}{r} + p + (1 - q^{r-1})^k q$.

This formula for E is the same as in the hypercube case. Therefore, every partial Steiner system of degree r with k and r chosen with respect to p gives a screening scheme which minimizes the expected number of tests per sample E. The task is then to find such a partial Steiner system of degree r with as few blocks as possible. The Remark in Subsection 2.1 shows that for given k and r a Steiner system $S(2, k; v)$ (if it exists) minimizes the number v of points and therefore minimizes the number of blocks b in the scheme among all partial Steiner systems $PS(2, k, r; w)$ of degree r.

[6] We found this proposition and its proof independently from Berger and Levenshtein [6]. It is a consequence of Theorem 2 there.

2.2. Further concepts from design theory

Here we present concepts and notations that we shall need in the next section.

Definition. A parallel class P in a design $(V, \mathcal{B})$ is a subset of the set of blocks $\mathcal{B}$ such that every element $w \in V$ occurs in exactly one of the blocks of P.

If $\mathcal{B}$ is the disjoint union of parallel classes, then the design is called *resolvable*. (For an example see Figure 3.) We write $RS(2, k; v)$ or $RPS(2, k, r; v)$ for resolvable Steiner systems or resolvable partial Steiner systems of degree r, respectively. (Of course, a partial Steiner system which is not of degree r for any $r \in N$ cannot be resolvable.) A partial resolution class PC is a subset of the blocks of $\mathcal{B}$, so that no point $v \in V$ occurs in more than one of the blocks of PC.

$$\begin{pmatrix}
1 & 0 & 0 & 1 & 0 & 0 & 1 & 0 & 0 & 1 & 0 & 0 \\
0 & 1 & 0 & 0 & 1 & 0 & 0 & 1 & 0 & 1 & 0 & 0 \\
0 & 0 & 1 & 0 & 0 & 1 & 0 & 0 & 1 & 1 & 0 & 0 \\
 & & & & & & & & & & & \\
1 & 0 & 0 & 0 & 0 & 1 & 0 & 1 & 0 & 0 & 1 & 0 \\
0 & 1 & 0 & 1 & 0 & 0 & 0 & 0 & 1 & 0 & 1 & 0 \\
0 & 0 & 1 & 0 & 1 & 0 & 1 & 0 & 0 & 0 & 1 & 0 \\
 & & & & & & & & & & & \\
1 & 0 & 0 & 0 & 1 & 0 & 0 & 0 & 1 & 0 & 0 & 1 \\
0 & 0 & 1 & 1 & 0 & 0 & 0 & 1 & 0 & 0 & 0 & 1 \\
0 & 1 & 0 & 0 & 0 & 1 & 1 & 0 & 0 & 0 & 0 & 1
\end{pmatrix}$$

Figure 3. The incidence matrix of a resolvable Steiner system $RS(2, 3; 9)$. The first parallel class consists of the first three columns, the second of the next three and so on. This Steiner system with $b = 12$ in the case $k = 3$, $r = 4$ shows that the reduction of samples in the scheme mentioned in Berger et al. [7] is not optimal; they give a scheme with $b = 16$ instead of $b = 64$ in the hypercube scheme.

Let $B(k)$ be the set of all v for which a Steiner system $S(2, k; v)$ exists and $RB(k)$ be the set of all v for which a resolvable Steiner system $RS(2, k; v)$ exists. An advantage of resolvable (partial) Steiner systems (of degree r) in our context is that by cutting off a parallel class we obtain a partial Steiner system of degree $r - 1$.

Vector spaces over finite fields give raise to canonical resolvable Steiner systems. Let W be the standard n-dimensional vector space over the field F_q with q elements. The lines in W are the one-dimensional affine subspaces, that mean $a + U$, where U is a linear one-dimensional subspace of W and $a \in W$. Let L be the set of all lines in W. Then (W, L) is a Steiner system, written as $AG(n, q)$, which is resolvable, since the blocks $a + U$ for a fixed U form a parallel class (Beth et al. [8, p. 11]).

Definition. A design is called *divisible* if V can be partitioned into point classes (also called *groups*) such that no block exists which contains points in the same class. If additionally each pair of points from different classes is together in exactly one block, one speaks of a group divisible design. A group divisible design of block size k and group size g with v points is notated $GD[k, g; v]$.

3. Optimal Designs for Event Probability p from 0.12 to 0.01

From Table 1 we get the pairs (k, r), for which we need optimal designs. According to the formula $E = \frac{k}{r} + p + (1 - q^{r-1})^k q$ there are optimal k and r for a given p. Each combination (k, r) is optimal for an interval of p. The p in Table 1 is maximal among those for which (k, r) is optimal,[7] i.e., if the given probability is between two probabilities in the table, we choose the line with the greater p.

[7]There is one exception in the trivial case $k = 1$. Here we give two lines for $r = 3$ because for the maximal $p = 0.3$ is clearly $E \approx 1$.

The second part of the table is a summary of the results of this section. It shows the minimal screening scheme with E optimal, the number b of samples in this and for comparison the number of samples in a hypercube scheme.

For example, let $p = 0.06$. Then $0.063 > p > 0.055$ and we get $k = 3$, $r = 12$. The optimal design is the Steiner system $S(2, 3; 25)$. For this we need 100 samples whereas the hypercube scheme needs 1728.

Table 1. Optimal (k, r) for given p and optimal designs for occurring pairs (k, r)

p	k	r	E	Design	b	r^k
0.3000	1	3	0.990	D(3)	3	3
0.1240	1	3	0.661	D(3)	3	3
0.1230	1	4	0.658	D(4)	4	4
0.1210	2	6	0.653	S(2, 2; 7)	21	36
0.1000	2	7	0.583	S(2, 2; 8)	28	49
0.0770	2	8	0.497	S(2, 2; 9)	36	64
0.0630	3	12	0.438	S(2, 3; 25)	100	1728
0.0550	3	13	0.399	RS(2, 3; 27)	117	2197
0.0480	3	14	0.363	RPS(2, 3, 14; 30)	140	2744
0.0430	3	15	0.336	S(2, 3; 31)	155	3375
0.0390	3	16	0.314	RS(2, 3; 33)	176	4096
0.0350	4	21	0.291	RS(2, 4; 64)	336	$1.94 \cdot 10^5$
0.0320	4	22	0.272	RPS(2, 4, 22; 76)	418	$2.34 \cdot 10^5$
0.0300	4	23	0.259	RPS(2, 4, 23; 76)	437	$2.80 \cdot 10^5$
0.0290	4	24	0.252	RPS(2, 4, 24; 76)	456	$3.32 \cdot 10^5$
0.0270	4	25	0.239	RS(2, 4; 76)	475	$3.91 \cdot 10^5$
0.0260	4	26	0.233	RPS(2, 4, 26; 88)	572	$4.57 \cdot 10^5$
0.0240	4	27	0.219	RPS(2, 4, 27; 88)	594	$5.31 \cdot 10^5$
0.0230	4	28	0.212	RPS(2, 4, 28; 88)	616	$6.15 \cdot 10^5$
0.0220	4	29	0.205	RS(2, 4; 88)	638	$7.07 \cdot 10^5$
0.0210	4	30	0.198	RPS(2, 4, 30; 100)	750	$8.10 \cdot 10^5$
0.0200	4	31	0.191	RPS(2, 4, 31; 100)	775	$9.24 \cdot 10^5$
0.0190	4	32	0.184	RPS(2, 4, 32; 100)	800	$1.05 \cdot 10^6$
0.0182	5	40	0.178	RPS(2, 5, 40; 165)	1320	$1.02 \cdot 10^8$
0.0176	5	41	0.173	RS(2, 5; 165)	1353	$1.16 \cdot 10^8$
0.0170	5	42	0.168	RPS(2, 5, 42; 185)	1554	$1.31 \cdot 10^8$
0.0166	5	43	0.165	RPS(2, 5, 43; 185)	1591	$1.47 \cdot 10^8$
0.0160	5	44	0.160	RPS(2, 5, 44; 185)	1628	$1.65 \cdot 10^8$
0.0156	5	45	0.157	RPS(2, 5, 45; 185)	1665	$1.85 \cdot 10^8$
0.0152	5	46	0.154	RS(2, 5; 185)	1702	$2.06 \cdot 10^8$
0.0148	5	47	0.151	RPS(2, 5, 47; 205)	1927	$2.29 \cdot 10^8$
0.0144	5	48	0.148	RPS(2, 5, 48; 205)	1968	$2.55 \cdot 10^8$
0.0140	5	49	0.144	RPS(2, 5, 49; 205)	2009	$2.82 \cdot 10^8$
0.0136	5	50	0.141	RPS(2, 5, 50; 205)	2050	$3.13 \cdot 10^8$
0.0132	5	51	0.138	RS(2, 5; 205)	2091	$3.45 \cdot 10^8$
0.0130	5	52	0.136	PS(2, 5, 52; 225)	2340	$3.80 \cdot 10^8$
0.0126	5	53	0.133	PS(2, 5, 53; 225)	2385	$4.18 \cdot 10^8$
0.0122	5	54	0.130	PS(2, 5, 54; 225)	2430	$4.59 \cdot 10^8$
0.0120	5	55	0.128	PS(2, 5, 55; 225)	2475	$5.03 \cdot 10^8$
0.0118	5	56	0.126	S(2, 5; 225)	2520	$5.51 \cdot 10^8$
0.0114	5	57	0.123	RPS(2, 5, 57; 245)	2793	$6.02 \cdot 10^8$
0.0112	5	58	0.121	RPS(2, 5, 58; 245)	2842	$6.56 \cdot 10^8$
0.0110	5	59	0.119	RPS(2, 5, 59; 245)	2891	$7.15 \cdot 10^8$
0.0108	5	60	0.118	RPS(2, 5, 60; 245)	2940	$7.78 \cdot 10^8$
0.0104	5	61	0.114	RS(2, 5; 245)	2989	$8.45 \cdot 10^8$
0.0102	5	62	0.112	RPS(2, 5, 62; 265)	3286	$9.16 \cdot 10^8$
0.0100	5	63	0.111	RPS(2, 5, 63; 265)	3339	$9.92 \cdot 10^8$

3.1. The case $k = 1$

For $p > 0.3$ the optimal strategy is to test each sample by itself. For $0.124 \le p \le 0.3$ the Dorfman scheme is optimal with $r = 3$, for $0.121 < p < 0.124$ the Dorfman scheme is optimal with $r = 4$.

3.2. The case $k = 2$

$k = 2$ is optimal for $0.1210 \ge p \ge 0.0630$ and the occurring values for r are 6, 7, 8. For our purpose it is sufficient to find a Steiner system $S(2, 2; v)$. Our formulas give $v = r + 1$ and $b = \frac{vr}{2} = \binom{r+1}{2}$. So we can choose the Steiner system with $r + 1$ points and all different pairs of points as blocks.

3.3. The case $k = 3$

$k = 3$ is optimal for $0.0630 \ge p \ge 0.0350$ with corresponding $12 \le r \le 16$. For a partial Steiner system of degree r we have $v \ge 2r + 1$ and $b = \frac{vr}{3} \ge \frac{(2r+1)r}{3}$. For $r \equiv 0, 1 \pmod 3$ design theory shows that there are Steiner systems with $v = 2r + 1$ and $b = \frac{vr}{3}$. For $r \equiv 2 \pmod 3$ this is not possible since $\frac{(2r+1)r}{3}$ is not an integer. But we can hope for partial Steiner systems of degree r with $v = 2r + 2$ and $b = \frac{(2r+2)r}{3}$. Indeed, design theory shows that such partial Steiner systems exist, which obviously have minimal b.

For our purpose we explicitly need the (partial) Steiner systems for $r = 12, \ldots, 16$. So in the following we give such designs.

r = 12 We give an $S(2, 3; 25)$, referring to Beth et al. [8, p. 483]. The construction goes back to Hwang and Lin [11]. Let $V = Z_{25}$ (the residue classes mod 25). For $i = 0, \ldots, 24$ set:

$$B_{1i} = \{0, 7, 9\} + i, \quad B_{2i} = \{0, 11, 12\} + i,$$

$$B_{3i} = \{0, 6, 10\} + i, \quad B_{4i} = \{0, 5, 8\} + i.$$

r = 13 We give an $S(2, 3; 27)$, referring to Beth et al. [8, p. 528]. The construction goes back to Ray-Chaudhuri and Wilson [15]. Let $V = (Z_{13} \times \{0, 1\}) \cup \infty$ and define for the rest of the paper the notations $(a, b) =: a_b$, $a_b + i := (a+i)_b$, $a_b \cdot i := (a \cdot i)_b$ and $\infty + i := \infty$. For $i = 0, \ldots, 12$ set:

$$B_{1i} = \{1_1, 10_1, 12_0\} + i, \quad B_{2i} = \{2_1, 7_1, 11_0\} + i, \quad B_{3i} = \{3_1, 4_1, 10_0\} + i,$$

$$B_{4i} = \{6_1, 8_1, 7_0\} + i, \quad B_{5i} = \{9_1, 12_1, 4_0\} + i, \quad B_{6i} = \{5_1, 11_1, 8_0\} + i,$$

$$B_{7i} = \{1_0, 3_0, 9_0\} + i, \quad B_{8i} = \{2_0, 6_0, 5_0\} + i, \quad B_{9i} = \{\infty, 0_0, 0_1\} + i.$$

For fixed i these blocks form a parallel class, so the Steiner system is resolvable.

r = 14 We use the 14 parallel classes of a nearly Kirkman triple system (Baker and Wilson [3]). The pointset is $V = (Z_7 \times \{1, 2, 3, 4\}) \cup \{\infty_1, \infty_2\}$. For each $i = 0, \ldots, 6$ one obtains a parallel class consisting of the ten blocks:

$$B_{1i} = \{4_1, 5_2, 0_3\} + i, \quad B_{2i} = \{6_1, 3_2, 4_3\} + i, \quad B_{3i} = \{1_1, 6_2, 5_4\} + i,$$

$$B_{4i} = \{5_1, 1_2, 6_4\} + i, \quad B_{5i} = \{2_1, 6_3, 0_4\} + i, \quad B_{6i} = \{0_1, 1_3, 3_4\} + i,$$

$$B_{7i} = \{3_1, 5_3, 2_4\} + i, \quad B_{8i} = \{0_2, 3_3, 1_4\} + i, \quad B_{9i} = \{2_2, 2_3, \infty_1\} + i,$$

$$B_{10i} = \{4_2, 4_4, \infty_2\} + i.$$

In the same way one obtains for each $i = 0, \ldots, 6$ a parallel class consisting of the ten blocks:

$$C_{1i} = \{0_1, 2_2, 6_3\} + i, \quad C_{2i} = \{6_1, 5_2, 1_4\} + i, \quad C_{3i} = \{0_2, 5_3, 4_4\} + i,$$

$$C_{4i} = \{1_2, 0_3, 3_4\} + i, \quad C_{5i} = \{1_1, 2_1, 4_1\} + i, \quad C_{6i} = \{3_2, 4_2, 6_2\} + i,$$

$$C_{7i} = \{1_3, 2_3, 4_3\} + i, \quad C_{8i} = \{6_4, 0_4, 2_4\} + i, \quad C_{9i} = \{5_1, 5_4, \infty_1\} + i,$$

$$C_{10i} = \{3_1, 3_3, \infty_2\} + i.$$

r = 15 In this case exist many $S(2, 3; 31)$. We give a construction of Colbourn and Mathon [9], referring to Beth et al. [8, p. 478]: Let $V = Z_{31}$, for $i = 0, \ldots, 30$ and $j = 0, \ldots, 4$ set $B_{ij} = \{0, 1, 6\} \cdot 10^j + i$.

r = 16 We use a construction of Skolem [17] for an $S(2, 3; 33)$, referring to Beth et al. [8, p. 481]. Let $V = Z_{11} \times Z_3$. For $i = 0, ..., 10$ and $j = 0, ..., 2$ set:

$$B_{0i} = \{0_0, 0_1, 0_2\} + i, \qquad B_{1ij} = \{0_{0+j}, 2_{0+j}, 1_{1+j}\} + i,$$

$$B_{2ij} = \{0_{0+j}, 4_{0+j}, 2_{1+j}\} + i, \qquad B_{3ij} = \{0_{0+j}, 6_{0+j}, 3_{1+j}\} + i,$$

$$B_{4ij} = \{0_{0+j}, 8_{0+j}, 4_{1+j}\} + i, \qquad B_{5ij} = \{0_{0+j}, 10_{0+j}, 5_{1+j}\} + i.$$

3.4. The case $k = 4$

Here we have to construct designs for $r = 21, ..., 32$ corresponding to p between 0.035 and 0.019. It is $B(4) = 12N + \{1, 4\}$ and $RB(4) = 12N + 4$, see Beth et al. [8, pp. 637 and 649]. The conditions for partial Steiner systems of degree r take the forms: $v \geq 3r + 1$ and $b = \frac{vr}{4} \geq \frac{(3r+1)r}{4}$.

As above, the first question is, whether a Steiner system with the lower bound of points and blocks exists. If $r \equiv 2 \pmod 4$, then it follows $3r + 1 \equiv 7 \pmod{12}$, if $r \equiv 3 \pmod 4$, then it follows $3r + 1 \equiv 10 \pmod{12}$, i.e., there exists no $S(2, 4; v)$ with $v = 3r + 1$. If $r \equiv 0 \pmod 4$, then it follows $3r + 1 \equiv 1 \pmod{12}$, i.e., there exists an $S(2, 4; v)$ with $v = 3r + 1$. If $r \equiv 1 \pmod 4$, then it follows $3r + 1 \equiv 4 \pmod{12}$, i.e., there exists a resolvable Steiner system $RS(2, 4; v)$ with $v = 3r + 1$.

As mentioned above, from the resolvable Steiner system $RS(2, 4; 3r + 1)$ in the case $r \equiv 1 \pmod 4$ we can easily construct partial Steiner systems of degrees $r - 1, ..., r - 3$ by cutting off parallel classes. For $r - 1$ it would even be possible to construct a Steiner system, but the disadvantage of our choice in using the partial Steiner system ($\frac{3(r-1)}{4}$ samples more in the scheme are required) is not so important compared with the advantage of implementing only one design. For $r - 2$ and $r - 3$ there is no easy prospect for another construction. So from now on we are concerned only with the case $r \equiv 1 \pmod 4$.

r = 21 We give an $RS(2, 4; 64)$, referring to Beth et al. [8, p. 26], namely the affine block design $AG(3, 4)$ with $v = 4^3$ points and $b = 336$ blocks. The underlying field is the finite field $F_4 = F_2[x]/(x^2 - x - 1)$. We define a bijective map $F_4 \to \{0, 1, 2, 3\}$ by $a_0 + a_1 x \mapsto a_0 + 2a_1$. The addition and multiplication tables are shown in Figure 4. Any of the 21 lines through 0 induces a parallel class of 16 blocks. That means for each m a parallel class consists of the 16 blocks we get from B_m $(m = 1, ..., 21)$ by all combinations of $i = 0, ..., 3$ and $j = 0, ..., 3$:

$$B_1 = \{(0, 0, 0), (0, 0, 1), (0, 0, 2), (0, 0, 3)\} + (i, j, 0),$$

$$B_2 = \{(0, 0, 0), (0, 1, 0), (0, 2, 0), (0, 3, 0)\} + (i, 0, j),$$

$$B_3 = \{(0, 0, 0), (1, 0, 0), (2, 0, 0), (3, 0, 0)\} + (0, i, j),$$

$$B_4 = \{(0, 0, 0), (0, 1, 1), (0, 2, 2), (0, 3, 3)\} + (i, j, 0),$$

$$B_5 = \{(0, 0, 0), (0, 2, 1), (0, 3, 2), (0, 1, 3)\} + (i, j, 0),$$

$$B_6 = \{(0, 0, 0), (0, 3, 1), (0, 1, 2), (0, 2, 3)\} + (i, j, 0),$$

$$B_7 = \{(0, 0, 0), (1, 0, 1), (2, 0, 2), (3, 0, 3)\} + (i, j, 0),$$

$$B_8 = \{(0, 0, 0), (2, 0, 1), (3, 0, 2), (1, 0, 3)\} + (i, j, 0),$$

$$B_9 = \{(0, 0, 0), (3, 0, 1), (1, 0, 2), (2, 0, 3)\} + (i, j, 0),$$

$$B_{10} = \{(0, 0, 0), (1, 1, 0), (2, 2, 0), (3, 3, 0)\} + (i, 0, j),$$

$$B_{11} = \{(0, 0, 0), (2, 1, 0), (3, 2, 0), (1, 3, 0)\} + (i, 0, j),$$

$$B_{12} = \{(0, 0, 0), (3, 1, 0), (1, 2, 0), (2, 3, 0)\} + (i, 0, j),$$

$$B_{13} = \{(0, 0, 0), (1, 1, 1), (2, 2, 2), (3, 3, 3)\} + (i, j, 0),$$

$$B_{14} = \{(0, 0, 0), (1, 1, 2), (2, 2, 3), (3, 3, 1)\} + (i, j, 0),$$

$$B_{15} = \{(0, 0, 0), (1, 1, 3), (2, 2, 1)(3, 3, 2)\} + (i, j, 0),$$

$$B_{16} = \{(0, 0, 0), (1, 2, 1), (2, 3, 2), (3, 1, 3)\} + (i, j, 0),$$

$$B_{17} = \{(0, 0, 0), (1, 3, 1), (2, 1, 2), (3, 2, 3)\} + (i, j, 0),$$

$B_{18} = \{(0, 0, 0), (2, 1, 1), (3, 2, 2), (1, 3, 3)\} + (i, j, 0),$

$B_{19} = \{(0, 0, 0), (3, 1, 1), (1, 2, 2), (2, 3, 3)\} + (i, j, 0),$

$B_{20} = \{(0, 0, 0), (1, 2, 3), (2, 3, 1), (3, 1, 2)\} + (i, j, 0),$

$B_{21} = \{(0, 0, 0), (1, 3, 2), (2, 1, 3), (3, 2, 1)\} + (i, j, 0).$

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

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

Figure 4. Operation tables for the case $r = 21$.

r = 25 In this case and the case $r = 29$ we use a construction, referring to Beth et al. [8, p. 504], which gives a resolvable $RS(2, 4; 3r+1)$ if $r \equiv 1 \pmod 4$ is a prime power.

At first we deal with the case $r = 25$. Let $V = (F_{25} \times Z_3) \cup \infty$. Identify $F_{25} = F_5[x]/(x^2 - 3)$ with $\{0, ..., 24\}$ via the bijective map $a_0 + a_1 x \mapsto a_0 + 5a_1$ and write the normal symbols for the induced operations $+, \cdot$ on $\{0, ..., 24\}$. For each $m = 0, ..., 24$ the following 19 blocks yield a parallel class: For $i = 0, ..., 5$, $j = 0, 1, 2$ set:

$$B_0 = \{\infty, 0_0, 0_1, 0_2\} + m,$$

$$B_{ij} = \{(6^i)_j, (-6^i)_j, (3 \cdot 6^i)_{j+1}, (-3 \cdot 6^i)_{j+1}\} + m.$$

r = 29 We set $V = (Z_{29} \times Z_3) \cup \infty$. For each $m = 0, ..., 28$ a parallel class of the constructed $RS(2, 4; 88)$ is formed by the following 22 blocks: For $i = 0, ..., 6$ and $j = 0, 1, 2$ set:

$$B_0 = \{\infty, 0_0, 0_1, 0_2\} + m,$$

$$B_{ij} = \{(2^i)_j, (-2^i)_j, (12 \cdot 2^i)_{j+1}, (-12 \cdot 2^i)_{j+1}\} + m.$$

r = 33 We use a construction of Beth et al. [8, p. 529] to get a resolvable $RS(2, 4; 100)$. Let $V = F_{25} \times \{1, 2, 3, 4\}$. Identify the set $\{0, ..., 24\}$ with F_{25} as above. For each $m = 0, ..., 24$ a parallel class is formed by the following 25 blocks:

$$B_0 = \{0_1, 0_2, 0_3, 0_4\} + m, \quad B_x = \{x_1, 2 \cdot x_2, 3 \cdot x_3, 4 \cdot x_4\} + m,$$

$$B_{i1} = \{i \cdot 3_i, i \cdot 4_i, i \cdot 8_i, i \cdot 15_i\} + m, \quad B_{i2} = \{i \cdot 2_i, i \cdot 11_i, i \cdot 17_i, i \cdot 19_i\} + m$$

$$(i = 1, 2, 3, 4, x \in \{1, ..., 24\} - \{2, 3, 4, 8, 11, 15, 17, 19\}).$$

For each $x \in \{2, 3, 4, 8, 11, 15, 17, 19\}$ the 25 blocks B_x (for $m = 0, ..., 24$) form one of the remaining eight parallel classes.

3.5. The case $k = 5$

In this case we are searching for designs with $r = 40, ..., 63$. The corresponding probabilities are $0.010 \le p \le 0.018$. It is known that $B(5) = 20N + \{1, 5\}$ and that for $v \in RB(5)$ it is necessary that $v \equiv 5 \pmod{20}$. Our formulas give: $v \ge 4r + 1$ and $b = \frac{vr}{5} \ge \frac{(4r+1)r}{5}$ with equality only in the Steiner system case. Only in the case $r \equiv 1 \pmod 5$ is $v \equiv 5 \pmod{20}$ and we can hope to find a resolvable Steiner system.

Because we have to cover 24 values of r we proceed as in the previous paragraph: We use Steiner systems with $r \equiv 1 \pmod 5$ and at least 4 parallel classes to give partial Steiner systems of degree $r, ..., r-4$ by cutting parallel classes.

r = 41 Abel et al. [1] gave a resolvable Steiner system $RS(2, 5; 165)$: Let $V = (Z_2 \times Z_2 \times Z_{41}) \cup \infty$. For each $m = 0, ..., 40$ the following 33 blocks form a parallel class $(i, j \in \{0, 1\})$:

$$B_0 = \{\infty, (0, 0, 0), (0, 1, 0), (1, 0, 0), (1, 1, 0)\} + (0, 0, m),$$

$$B_{1ij} = \{(0, 0, 1), (0, 0, 40), (0, 0, 7), (0, 1, 15), (1, 0, 6)\} + (i, j, m),$$

$$B_{2ij} = \{(0, 0, 2), (0, 1, 17), (0, 1, 36), (1, 0, 5), (1, 0, 20)\} + (i, j, m),$$

$$B_{3ij} = \{(0, 0, 10), (0, 0, 31), (0, 0, 34), (1, 0, 21), (1, 1, 38)\} + (i, j, m),$$

$B_{4ij} = \{(0, 0, 24), (1, 0, 3), (1, 0, 39), (1, 1, 26), (1, 1, 35)\} + (i, j, m),$

$B_{5ij} = \{(0, 0, 9), (0, 0, 32), (0, 0, 22), (0, 1, 12), (1, 0, 13)\} + (i, j, m),$

$B_{6ij} = \{(0, 0, 18), (0, 1, 30), (0, 1, 37), (1, 0, 4), (1, 0, 16)\} + (i, j, m),$

$B_{7ij} = \{(0, 0, 8), (0, 0, 33), (0, 0, 19), (1, 0, 25), (1, 1, 14)\} + (i, j, m),$

$B_{8ij} = \{(0, 0, 11), (1, 0, 27), (1, 0, 23), (1, 1, 29), (1, 1, 28)\} + (i, j, m).$

r = **46** We give an $RS(2, 5; 185)$ referring to Abel et al. [2]. Let $V = (Z_3 \cup \{\infty_1, \infty_2\}) \times Z_{37}$ and $\infty_j + i = \infty_j$. For each m the following 37 blocks form a parallel class $(i = 0, ..., 2)$:

$B_0 = \{(0, 0), (1, 0), (2, 0), (\infty_1, 0), (\infty_2, 0)\} + (0, m),$

$B_{1i} = \{(0, 1), (0, 2), (0, 17), (1, 34), (\infty_1, 13)\} \cdot (1, 10^i) + (i, m),$

$B_{2i} = \{(0, 4), (0, 27), (0, 29), (2, 28), (\infty_1, 12)\} \cdot (1, 10^i) + (i, m),$

$B_{3i} = \{(0, 7), (0, 13), (1, 4), (1, 28), (\infty_1, 11)\} \cdot (1, 10^i) + (i, m),$

$B_{4i} = \{(0, 23), (0, 34), (1, 6), (1, 21), (\infty_1, 10)\} \cdot (1, 10^i) + (i, m),$

$B_{5i} = \{(0, 19), (1, 24), (1, 30), (2, 22), (\infty_2, 11)\} \cdot (1, 10^i) + (i, m),$

$B_{6i} = \{(0, 6), (0, 9), (0, 16), (1, 32), (\infty_2, 18)\} \cdot (1, 10^i) + (i, m),$

$B_{7i} = \{(0, 10), (0, 14), (1, 13), (1, 18), (\infty_2, 25)\} \cdot (1, 10^i) + (i, m),$

$B_{8i} = \{(0, 11), (0, 20), (\infty_1, 6), (\infty_2, 2), (\infty_2, 19)\} \cdot (1, 10^i) + (i, m),$

$B_{9i} = \{(0, 12), (0, 36), (\infty_1, 18), (\infty_2, 16), (\infty_2, 34)\} \cdot (1, 10^i) + (i, m),$

$B_{10i} = \{(0, 22), (1, 14), (\infty_1, 2), (\infty_2, 3), (\infty_2, 10)\} \cdot (1, 10^i) + (i, m),$

$B_{11i} = \{(0, 15), (\infty_1, 4), (\infty_2, 8), (\infty_2, 29), (\infty_2, 35)\} \cdot (1, 10^i) + (i, m),$

$B_{12i} = \{(0, 26), (\infty_1, 7), (\infty_1, 17), (\infty_1, 25), (\infty_1, 29)\} \cdot (1, 10^i) + (i, m).$

The remaining 9 parallel classes arise from the following blocks, one for each C_{ji} $(i = 0, 1, 2;\ j = 1, 2, 3)$. They consist of the 37 blocks one obtains for the different values of m $(m = 0, ..., 36)$:

$C_{1i} = \{(0, 1), (1, 3), (2, 12), (\infty_1, 32), (\infty_2, 22)\} \cdot (1, 10^i) + (i, m),$

$C_{2i} = \{(0, 4), (1, 22), (2, 36), (\infty_1, 33), (\infty_2, 35)\} \cdot (1, 10^i) + (i, m),$

$C_{3i} = \{(0, 5), (1, 11), (2, 2), (\infty_1, 21), (\infty_2, 8)\} \cdot (1, 10^i) + (i, m).$

r = **51** Here $v = 205$ and the construction is parallel to the one used in the case $r = 33$. Let $V = Z_{41} \times \{1, 2, 3, 4, 5\}$. For each $m = 0, ..., 40$ a parallel class is formed by the following 41 blocks:

$B_0 = \{0_1, 0_2, 0_3, 0_4, 0_5\} + m,$

$B_x = \{x_1, 2 \cdot x_2, 3 \cdot x_3, 4 \cdot x_4, 5 \cdot x_5\} + m,$

$B_{i1} = \{i \cdot 5_i, i \cdot 12_i, i \cdot 15_i, i \cdot 16_i, i \cdot 28_i\} + m,$

$B_{i2} = \{i \cdot 4_i, i \cdot 9_i, i \cdot 18_i, i \cdot 24_i, i \cdot 26_i\} + m$

$(i = 1, 2, 3, 4, 5,\ x \in \{1, ..., 40\} \setminus \{4, 5, 9, 12, 15, 16, 18, 24, 26, 28\}).$

For each $x \in \{4, 5, 9, 12, 15, 16, 18, 24, 26, 28\}$ the 41 blocks B_x (for $m = 0, ..., 40$) form one of the remaining ten parallel classes.

r = **56** It is not known whether a resolvable Steiner system $RS(2, 5; 225)$ exists. We describe the construction of a Steiner system $S(2, 5; 225)$ (Beth et al. [8, pp. 503 and 623]) with at least six parallel classes which fits perfectly with our purpose. We proceed in several steps:

1. We start with a $GD[5, 5; 45]$ and get an $S(2, 5; 45)$ by attaching a parallel class which contains nine blocks, each containing the five elements of one of the groups. Let M be the 45×99 -incidence matrix of this $S(2, 5; 45)$ and P be the 45×9 -submatrix corresponding to the attached parallel class.

2. We start with $RS(2, 5; 25) = AG(2, 5)$ and get a $GD[5, 5; 25]$ by cutting of a parallel class and define the groups according to the blocks

we have cut. We choose to label V such that the first five points of V form a group, and then the next five and so on. Then we choose to label $\mathcal{B}$ so that the first five blocks form a parallel class, the next five as well and so on. Let M_i be the 5×25-submatrix consisting of the rows $5(i-1)+1, \ldots, 5i$ $(i = 1, \ldots, 5)$, i.e., the submatrix corresponding to the i-th group.

3. Now we combine the incidence matrices of these designs. In each column of the incidence matrix M of the $S(2, 5; 45)$ we replace the first by M_1, the second by $M_2, \ldots,$ and the last by M_5. The zeroes we replace by 5×25-zero matrices. This means from each column we obtain a 225×25-matrix. The resulting 225×2475-matrix is the incidence matrix of a $GD[5, 5, 225]$.

4. We construct an $S(2, 5; 225)$ out of this $GD[5, 5; 225]$ by attaching a parallel class, which consists of 45 blocks, each containing the five elements of one of the groups.

5. Finally we have to identify five further parallel classes. For the first class we take for each column of P (see step 1) the first five columns of the 225×25-matrix replacing it as in step 3, obtaining 45 blocks forming a parallel class. In general, for the i-th parallel class for each column of P we take the $5(i-1)+1$-th to $5i$-th columns of the 225×25-matrix replacing it as in step 3 $(i = 1, \ldots, 5)$. For the realization of this procedure one surely needs a computer.

r = 61 There is an $RS(2, 5; 245)$ as shown in Abel et al. [1]. Let $V = (Z_2 \times Z_2 \times Z_{61}) \cup \infty$. The Steiner system consists of 61 parallel classes, one for each value of $m = 0, \ldots, 60$. The 25 blocks of such a parallel class are $(i = 0, 1,\ j = 0, 1,\ t = 0, \ldots, 5)$:

$$A_{ijt} = \{(0, 0, 1), (0, 0, 60), (0, 0, 6), (0, 1, 55), (1, 0, 18)\} \cdot (1, 1, 2^{5t}) + (i, j, m),$$

$$B_{ijt} = \{(0, 0, 7), (0, 1, 33), (0, 1, 54), (1, 0, 43), (1, 0, 28)\} \cdot (1, 1, 2^{5t}) + (i, j, m),$$

$$C = \{\infty, (0, 0, 0), (0, 1, 0), (1, 0, 0), (1, 1, 0)\} + (0, 0, m).$$

r = 66 It is possible to construct an $RS(2, 5; 265)$ using an existing $S(2, 6; 66)$ (see Beth et al. [8, pp. 49 and 524]). The construction is too complicated to be given here and requires a computer for implementation. It is much simpler to use the following $RS(2, 5; 285)$ for the values $r = 71, \ldots, 62$ by cutting off one to nine parallel classes.

r = 71 we give this $RS(2, 5; 285)$ referring to Abel et al. [1], constructed as in the case $r = 61$ above. Let $V = (Z_2 \times Z_2 \times Z_{71}) \cup \infty$. The Steiner system consists of 71 parallel classes, one for each value of $m = 0, \ldots, 70$. The 57 blocks of such a parallel class are $(i = 0, 1,\ j = 0, 1,\ t = 0, \ldots, 6)$:

$$A_{ijt} = \{(0, 0, 1), (0, 0, 70), (0, 0, 11), (0, 1, 60), (1, 0, 9)\} \cdot (1, 1, 7^{5t}) + (i, j, m),$$

$$B_{ijt} = \{(0, 0, 36), (0, 1, 29), (0, 1, 35), (1, 0, 62), (1, 0, 42)\} \cdot (1, 1, 7^{5t}) + (i, j, m),$$

$$C = \{\infty, (0, 0, 0), (0, 1, 0), (1, 0, 0), (1, 1, 0)\} + (0, 0, m).$$

4. Finer Considerations in the Case $k = 2$

For a given probability p let D_1 be an optimal design in a specified class. By reducing p we come to a point at which D_1 is no longer optimal but the design D_2 is optimal. We say that these designs are *neighboured*. In the hypercube case (see Table 1) there are two effects: if k remains constant, then r grows by one, but if k grows by one, then r may jump.

Schuster [16] shows that there are optimal hypercuboids between optimal hypercubes. More precisely, if the hypercube r^k is optimal for the probability p_0 and the hypercube $(r+1)^k$ is optimal for $p_k < p_0$, then there exists a chain of probabilities $p_k < p_{k-1} < \cdots < p_1 < p_0$ such that p_i is optimal for the hypercuboid $(r+1)^i r^{k-i}$. So we can improve the transitions by considering the greater class of hypercuboids.

In this section we are concerned with transitions from partial Steiner systems $PS(2, k, r; v)$ of degree r to the more general partial Steiner systems with the same k. We proceed by cutting off single blocks of a parallel class or a partial resolution class. In this way, the number of elements of V contained in r blocks is reduced step-by-step while the number of elements contained in $r-1$ blocks grows.

We treat the case $k = 2$, i.e., the starting point is an $S(2, 2; v)$, with $b = \frac{v(v-1)}{2}$ blocks. Clearly in this case a (partial) resolution class exists and we can cut of $0 \le c < \frac{v}{2}$ blocks. Let $b^* = b - c$ be the modified number of blocks.

Proposition 4.1. *If we cut off c blocks of an* $S(2, 2; v)$, *then E of the corresponding screening scheme is*

$$E(S(2, 2; v), c) = \frac{kb}{rb^*} + p + \frac{q}{b^*}\left[\frac{(v-2c)(v-2c-1)}{2}(1-q^{r-1})^2 + 2c(v-2c)(1-q^{r-1})(1-q^{r-2}) + 2c(c-1)(1-q^{r-2})^2\right].$$

Proof. At the first stage we have $\frac{kb}{rb^*}$ tests per sample. We use the formula from the proof of the Proposition in Subsection 2.1, but $P(T_S \mid S = -)$ now depends on c and we have different types of blocks: There remain $v - 2c$ mixtures of r samples. They correspond to $\frac{(v-2c)(v-2c-1)}{2}$ blocks and for each of these $P(T_S \mid S = -) = (1-q^{r-1})^2$. Furthermore, there are $2c(v-2c)$ blocks with one mixture consisting of r samples and one consisting of $r-1$ samples, i.e., for these blocks $P(T_S \mid S = -) = (1-q^{r-1})(1-q^{r-2})$. Finally we have $\frac{c(c-1)}{2}$ combinations of cut off blocks with altogether $2c(c-1)$ blocks for which $P(T_S \mid S = -) = (1-q^{r-2})^2$. If we add $P(T_S \mid S = -)$ for all blocks and divide by the number b^* of blocks, then we get the expected probability for an arbitrary block and the desired formula. Table 2 gives optimal partial Steiner systems for the probabilities $0.121 \ge p \ge 0.07$ with $k = 2$, i.e., the number of blocks to cut. For $p < 0.07$ is $k \ge 3$. In this cases analogous calculations are only possible for $c = 1$ and $c = 2$ whereas for $c \ge 3$ the required probabilities could depend on the special design and the chosen blocks to cut.

Table 2. Optimal number c of blocks to cut for given p in the case $k = 2$, i.e., $0.121 \ge p \ge 0.070$

p	r	b	c	b^*	E^*	E
0.121	6	21	0	21	0.653	0.653
0.107	7	28	3	25	0.607	0.610
0.103	7	28	2	26	0.593	0.595
0.099	7	28	1	27	0.579	0.5795
0.096	7	28	0	28	0.568	0.568
0.081	8	36	4	32	0.512	0.514
0.079	8	36	3	33	0.504	0.506
0.077	8	36	2	34	0.497	0.497
0.075	8	36	1	35	0.488	0.489
0.073	8	36	0	36	0.480	0.480

Note. b is the number of samples in the $S(2, 2; r+1)$-scheme, b^* is the number of samples in the finer scheme, E is the expected number of tests per sample in the $S(2, 2; r+1)$ and E^* is the expected number of tests per sample in the final screening scheme. (For p between two entries in the table chooses the upper line.)

5. Discussion of Practical Issues

In Table 1 we suggest designs for pairs (k, r). The only consequence of choosing a pair (k, r) which is not optimal, is a higher value for E. So it is even possible to choose a more simple design if this higher E is acceptable.

5.1. We only know $c_1 \le p \le c_2$

For fixed k and r and $0 \le p \le 1$ the function $E(p)$ is increasing. To show this, we differentiate $E = \frac{k}{r} + p + (1 - q^{r-1})^k q$:

$$\frac{dE}{dp} = 1 - (1-(1-p)^{r-1})^k + (1-p)k(1-(1-p)^{r-1})^{k-1}(-(r-1)(1-p)^{r-2}(-1))$$

$$= 1 - (1-(1-p)^{r-1})^{k-1}[1-(1-p)^{r-1} - k(r-1)(1-p)^{r-1}]$$

$$= 1 - (1-(1-p)^{r-1})^{k-1}[1-(1-p)^{r-1}(1+k(r-1))] \ge 0,$$

since $0 \le (1-(1-p)^{r-1})^{k-1} \le 1$ and $[1-(1-p)^{r-1}(1+k(r-1))] \le 1$.

We suggest a minimax-strategy: Choose an optimal strategy for $p = c_2$. The true E^* is at most $E(c_2)$.

Another possibility works with the assumption that p is uniformly distributed in the interval $i = \{p \mid c_1 \le p \le c_2\}$. Let $D_1, \ldots, D_j$ be all the designs that are optimal at one point in the interval. Now we look at n equidistant points in the interval and calculate the average E for every D_i. We choose the design with the minimal average E. This procedure is a simplified numerical integration and requires elaborate calculation whereas the minimax-suggestion can be read off from Table 1.

5.2. Mixing influences specifity or sensitivity of the test

We concentrate on the influence of the screening procedure so we still assume perfect specifity and sensitivity for the test of a single sample. In our model the problem of mixing is a possible low concentration, not a possible interaction between the samples. Therefore, false positive testing of a mixture is unlikely. We do not discuss this here, but notice that it would lead to unnecessary tests at the second stage, i.e., a greater E.

We now discuss the case of imperfect sensitivity. Let M_i be a mixture and s_{ij} be the part of the sample S_j contained in M_i. We describe the problem of mixing in terms of s_{ij}, i.e., we assume that a case of false negative testing can be identified as a problem of the type $(S_j = +) \wedge (s_{ij} = -)$. Then the mixture is positive if at least one of its parts s_{ij} is positive: $P(M_i = +) = P(\bigcup_j (s_{ij} = +))$. We assume the following conditional probabilities:

$$P(s_{ij} = - \mid S_j = -) = 1, \; P(s_{ij} = + \mid S_j = -) = 0,$$

$$P(s_{ij} = + \mid S_j = +) = s, \; P(s_{ij} = - \mid S_j = +) = 1 - s = t.$$

We assume that the errors $(S_j = +) \wedge (s_{ij} = -)$ are independent, so we can calculate the probability that the mixture M_i is tested positive:

$$P(M_i = +) = P(\bigcup_j (s_{ij} = +)) = 1 - P(\bigcap_j (s_{ij} = -)) = 1 - \prod_j P(s_{ij} = -)$$

$$= 1 - \prod_j [P(s_{ij} = - \mid S_j = -)P(S_j = -) + P(s_{ij} = - \mid S_j = +)P(S_j = +)]$$

$$= 1 - \prod_j (1 \cdot q + t \cdot p) = 1 - (q + tp)^r.$$

For fixed j we get $P(M_i = + \mid S_j = +) = 1 - (1 \cdot 0 + t \cdot 1)(q + t \cdot p)^{r-1} = 1 - t(q + tp)^{r-1}$ and $P(M_i = + \mid S_j = -) = 1 - (1 \cdot 1 + t \cdot 0)(q + t \cdot p)^{r-1} = 1 - (q + tp)^{r-1}$. In the special case $t = 0$ we obtain the formulas of Section 2.

Now fix S_j and look at all M_i that contain parts of S_j. Since the M_i are independent, we can calculate the probabilities that all the mixtures test positive:

$$P(\bigcap_i (M_i = + \mid S_j = +)) = (1 - t(q + tp)^{r-1})^k,$$

$$P(\bigcap_i (M_i = + \mid S_j = -)) = (1 - (q + tp)^{r-1})^k.$$

Now it is easy to calculate the probability that at least one of the mixtures containing parts of S_j has tested false negative:

$$P(\bigcup_i (M_i = - \mid S_j = +)) = 1 - P(\bigcap_i (M_i = + \mid S_j = +)) = 1 - (1 - t(q + tp)^{r-1})^k.$$

For small t the first approximation $(t^2 = 0)$ of this is kt, i.e., the probability that a positive sample is not identified, increases linearly with k.

So if the probability t for a false negative testing of a mixture with r components is known, the probability of a false negative tested sample can be calculated. If a maximal acceptable probability for a false negative tested sample and $t(r)$ for the respective r has been given, a screening scheme with k and r small enough according to this condition and the above formula can be chosen.

5.3. Decisions for bounded r and b

If it is possible to mix only r_0 samples and the optimal scheme for the given probability p works with $r > r_0$, it is optimal to choose a screening scheme with the maximal $r \leq r_0$ occurring in Table 1. If it is not possible to collect the samples for the optimal screening design according to the known probability p, but only b_0 samples are to be tested, choose in Table 1 the design with the maximal $b \leq b_0$. If the number of samples that can be mixed without false negative testing is not known use other methods, e.g., Balding and Tourney [4].

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