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Design Issues for the Michaelis–Menten Model

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We discuss design issues for the Michaelis–Menten model and use geometrical arguments to find optimal designs for estimating a subset of the model parameters, or a linear combination of the parameters. We propose multiple-objective optimal designs when the parameters have different levels of interest to the researcher. In addition, we compare six commonly used sequence designs in the biological sciences for estimating parameters and, propose optimal choices for the parameters for geometric designs using closed-form efficiency formulas.

1. Introduction

The Michaelis–Menten (MM) model is frequently used in the biological sciences to describe the rapid equilibrium kinetics of enzyme systems and in analysing data from drug, neurotransmitter and hormone receptor assays (Cressie & Keightley, 1981; Raaijmakers, 1987; Dunn, 1985). The MM model is also used in compartmental models to modelize the rate of change from one compartment to another. Typically, this model predicts the velocity rate, v, of formation of a product in a chemical reaction given the substrate concentration, s. In its simplest form, the model can be described by

$$E(v) = \frac{Vs}{K+s}, \qquad var(v) = \sigma^2, \quad s \in S = [0, \infty).$$

The parameter V denotes the maximum saturation. The parameter K is the Michaelis–Menten constant and at this value of concentration, half of the maximum saturation is reached. In practice, the design space S is truncated to a closed and bounded interval. Some researchers, Duggleby (1979) and Currie (1982), for example, suggested without reason, that the right end point of the design space S to be a multiple of the nominal value K. We show that an advantage of assuming that $S = [0, bK]$, where b is a user-selected constant, is that behavioral properties of locally optimal designs (Chernoff, 1953) depend on b only.

There is a lot of work for fitting the MM model; the paper by Ruppert *et al.* (1989) and the references therein provide an excellent source of reference. In particular, there are nonlinear algorithms, linearization by transformation and non-parametric procedures for estimating the parameters K and V. Each of these methods has its own advantages and problems and they are discussed abundantly in the above papers. In this paper, we assume least-squares estimates are used and discuss design issues for the MM model. It appears that design issues for the MM model are not well addressed in the literature as many designs used, in practice, are without theoretical justifications. For example, design strategies for

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the MM model suggested in Currie (1982), Cressie & Keightley (1981), Cleland (1967) and Dunn (1985) are all based on simulation studies.

Our discussion focuses on the simplest form of the MM model but the techniques described here can be applied to more complicated forms of the model. Many extensions of the model are used in practice; see, for example, Pang *et al.* (1978) where a sum of MM models was used to study several enzymes simultaneously. In fact, extension of this popular model goes as far back as the paper by Lineweaver & Burk (1934). In Section 6, we discuss a particular extension of the model when there is heteroscedasticity in the model.

Design issues are important because they can greatly affect the accuracy of our inferences. For example, consider the design used in the paper by Watts in Endrenyi (1981). It is a replicated six-point design for enzyme velocity with substrate concentration at 0.2, 0.222, 0.286, 0.4, 0.667, and 2. Table 1 shows the efficiencies of this design and some commonly used designs for estimating V and K: a geometric design, an inverse linear design, a logarithmic design and an arithmetic design. A geometric design has points that form a geometric progression, an arithmetic design has points uniformly spread out, an inverse linear design has points that form a linear sequence along the response axis and a logarithmic design spreads the design points according to the logarithmic law. The sequence designs in Table 1 all have six points and have the same starting and end points as the Watt's design. From the table, it is clear that some designs perform much better than others. This implies that designs can have a substantial impact on the accuracy of the estimates. Watt's design is nearly an inverse linear sequence design and so it is not surprising that the efficiencies of the two designs are quite similar.

TABLE 1

*Efficiencies of Watts' design and four commonly used designs for estimating the parameters K and V in the Michaelis–Menten model**

	eff_K (%)	eff_V (%)
Watts' design	76.7	65.9
Geometric design	66.0	59.1
Inverse linear design	77.5	65.9
Logarithmic design	36.8	40.7
Arithmetic design	52.3	52.1

*The nominal values are $K = 1.70$ and $V = 0.106$.

In this paper, we construct a variety of optimal designs for the MM model using Elfving's geometrical argument. These optimal designs can serve as benchmarks for comparing other designs. In addition, we construct optimal designs for experiments when there are two objectives and there may be unequal interest in each of the objectives. We also evaluate the performance of commonly used designs for the MM model in the biometry literature. In the last section, we show how Elfving's argument can be used to obtain other types of optimal design, including design strategies for the MM model when the variance of the response depends on its mean, as in ligand-binding systems (Raaijmakers, 1987), for example.

2 Background

An experimental design consists of a collection of points, $s_1, s_2, \ldots, s_N$, in some given space S. Some of these N points may be repeated, meaning that several observations are taken at the same value of s. The total number of observations is N and this number is usually pre-determined by cost constraints. A convenient way to understand designs is to treat them as a collection of different points of S, together with the proportion of the N observations to be allocated at the different points. This suggests the idea of the so-called approximate design as a probability measure ξ on S. Thus, $\xi(s)$ is the proportion of observations to be taken at the level s. Kiefer (1959) pioneered this approach and its many advantages are well documented in design monographs, see Silvey (1980) for example. Some recent applications of this approach to solve some dose–response design problems and optimal treatment allocation problems are Huang & Wong (1998a, b) and Zhu & Wong (2000). In what follows, we adopt the approximate design approach and without loss of generality, restrict attention to designs with a finite set of support points. When the design is supported at only two points, we will, for convenience, describe the design using a 2×2 matrix with the support points displayed in the first row and their corresponding mass in the second row.

Let $\theta^{\mathrm{T}} = (K, V)$ and let $f(s) = \partial E(v)/\partial\theta$ evaluated at the nominal value of θ. This nominal value usually represents the best guess for the parameter θ at the start of the experiment. The Fisher information matrix of a design ξ is given by

$$M(\xi) = \sum_{s\in S} f(s) f^{t}(s)\xi(s),$$

apart from an unimportant multiplicative constant, see for example Dette & Wong (1999). For the MM model, this matrix is

$$M(\xi) = \sum_{s\in S} \begin{pmatrix} \dfrac{V^2 s^2}{(K+s)^4} & \dfrac{-Vs^2}{(K+s)^3} \\ \dfrac{-Vs^2}{(K+s)^3} & \dfrac{s^2}{(K+s)^2} \end{pmatrix}.$$

So, when N is large, the covariance matrix of the estimates of θ is approximately σ^2/N times the inverse of this matrix.

A common design criterion for estimating the model parameters is to minimize the volume of the confidence ellipsoid of the parameters. This is D-optimality and formally, the criterion is given by $\Phi_D[M(\xi)] = \det M^{-1/k}(\xi)$, where k is the number of parameters in the model. If the interest is in estimating a linear combination of the parameters, say, $c^{\mathrm{T}}\theta$, we have c-optimality defined by $\Phi_c(M(\xi)) = c^{\mathrm{T}} M^{-1}(\xi) c$. It is known that these criteria are all convex functions of the designs (Silvey, 1980) and so designs with small criterion values are desirable. A design that minimizes one of these functions over all designs on S is called a Φ-optimal design, or more specifically a D- or a c-optimal design, respectively.

An advantage of working with approximate designs is that it is easy to check the optimality of a design. Because the criteria are convex, a standard convex analysis argument using directional derivatives (see Silvey, 1980, Chap. 3) shows that a design ξ^* is Φ-optimal provided

$$\psi(s,\xi^*) = f^{\mathrm{T}}(s)\nabla\Phi(\xi^*) f(s) - \operatorname{tr} M(\xi^*)\nabla\Phi(\xi^*) \geqslant 0, \quad s\in S \qquad (1)$$

with equality at the support points of ξ. Here $\Phi(\xi)$ is $\Phi(M(\xi))$ for short and $\nabla\Phi(\xi)$ denotes the gradient of $\Phi(\xi)$. The above inequality is referred to as an equivalence theorem in optimal design literature and it can be useful to help find the optimal design in simple problems.

The worth of a design is measured by its efficiency defined by $\mathrm{eff}_{\Phi}(\xi) = \Phi(\xi^*)/\Phi(\xi)$. This definition requires that the two designs have the same number of observations. The efficiency is sometimes multiplied by 100 and the efficiency is reported in percentage, see Table (1), for example. If this value is 50%, this means that the design ξ needs double the total number of observations for it to perform as well as the optimal design ξ^*.

3. Elfving's Method

3.1. c-OPTIMAL DESIGNS FOR ESTIMATING V AND K

An elegant method for finding an optimal design to estimate a linear combination of the model parameter, say $c^{\mathrm{T}}\theta$, is given in Elfving (1952). This method is nicely illustrated and explained in Chernoff (1972, pp. 13–16). For a given regression problem with regression function $f(s)$, the method first defines the Elfving set given by $G =$ convex hull of $(f(S)\cup -f(S))$. This means that the set G is the smallest convex set containing the set $(f(S)\cup -f(S))$. The point of intersection of the straight line defined by c with the boundary of the Elfving set determines the c-optimal design, ξ^*, as a convex combination of vertices of G. These vertices form the support points of the optimal design and the weights in the convex combination are the weights of the optimal design. Furthermore, $\Phi_c(\xi^*) = \|c\|/\|c^*\|$, where c^* is the vector defined by the cut point.

For the MM model, a direct calculation shows that we have

$$f(S) = \left\{(g_1, g_2) : g_1 = -\frac{V}{K} g_2(1-g_2),\ g_1 \in \left[-\frac{V}{4K}, 0\right],\quad g_2 \in \left[0, \frac{b}{1+b}\right]\right\}.$$

The Elfving set G is shown in Fig. 1. To estimate each of the parameters K and V, we set $c^{\mathrm{T}} = (1,0)$ and $(0,1)$ and call the resulting optimal design K- and V-optimal design respectively. The

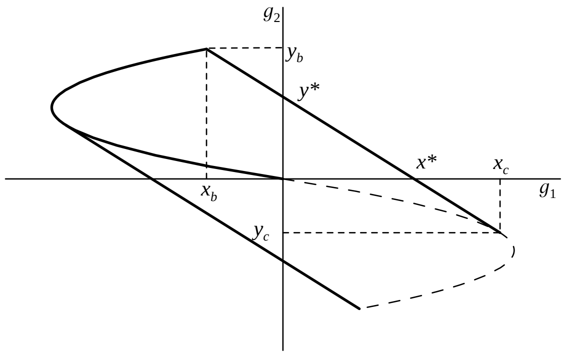

FIG. 1. Elfving set for the Michaelis–Menten Model. The points (x_c, y_c) and (x_b, y_b) define the support points of the K and V optimal designs (i.e. $s_c K$ and bK). The points $(x^*, 0)$ and $(0, y^*)$ are convex combinations of (x_c, y_c) and (x_b, y_b) and the corresponding coefficients of this combination give the weights of the optimal designs.

cut points of the line linking the end point of the curve $f(S)$, $(x_b, y_b) = ((-b/(1+b)^2)V/K, b/(1+b)$, and the tangential point on $-f(S)$, (x_c, y_c), with both axes define the weights for the K- and V-optimal designs. The design points, $s_c K$ and bK are defined by the tangential point (x_c, y_c), and the end point (x_b, y_b) according to the equations defining $-f(S)$ and $f(S)$, respectively (Fig. 1).

From the geometry of Fig. 1 and Elfving's argument, the K-optimal design is

$$\xi_K = \begin{pmatrix} s_c K & bK \\ 1/\sqrt{2} & 1 - 1/\sqrt{2} \end{pmatrix},$$

and the V-optimal design is

$$\xi_V = \begin{pmatrix} s_c K & bK \\ p_V & 1 - p_V \end{pmatrix},$$

where $s_c = (\sqrt{2}-1)b/(1+\sqrt{2}(\sqrt{2}-1)b)$ and $p_V = 1/(\sqrt{2} + (3\sqrt{2}-4)b)$. Both optimal designs are supported at the same two points but only the optimal mass distribution for ξ_V depends on the parameter b. At the point $s_c K$, the velocity of the reaction is $(\sqrt{2}-1) = 0.412$ of the maximum velocity at the end point bK. These designs do not depend on the value of V and the values of the criteria functions (i.e. the variances of the estimates) are

$$\Phi_K(\xi_K) = (3 + 2\sqrt{2})^2 \left(\frac{1+b}{b}\right)^4 \frac{K^2}{V^2}$$

and

$$\Phi_V(\xi_V) = \frac{(3 + 2\sqrt{2} + b)^2 (1+b)^2}{b^4},$$

respectively. The efficiencies of these designs for estimating the other parameter depend only on b and are given by

$$eff_K(\xi_V) = \frac{\Phi_K(\xi_K)}{\Phi_K(\xi_V)} = \frac{1 + (2-\sqrt{2})b}{1 + (2-\sqrt{2})b + (5\sqrt{2}-7)b^2}$$

and

$$eff_V(\xi_K) = \frac{\Phi_V(\xi_V)}{\Phi_V(\xi_K)} = \frac{[1 + (3 - 2\sqrt{2})b]^2}{[1 + (3 - 2\sqrt{2})b]^2 + (5\sqrt{2} - 7)b^2}.$$

Figure 2 shows the behavior of these two efficiencies as the value of the parameter b changes. Both the efficiencies are high for small values of b. For example, if $b = 1$, both efficiencies are greater than 95% and if $b = 3$, both the optimal designs have nearly 80% for estimating the other parameter. For $b = 5$ both efficiencies are over 65% and for $b = 10$, they are still near 50%. Beyond $b = 10$, both efficiencies decrease rapidly; $eff_K(\xi_V)$ tends to 0 and $eff_V(\xi_K)$ tends $1 - 1/\sqrt{2} = 0.293$

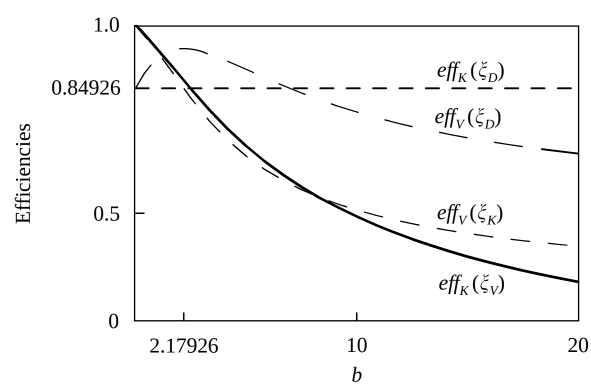

FIG. 2. Efficiencies of D-optimal design for estimating K, $eff_K(\xi_D)$, and V, $eff_V(\xi_D)$, efficiencies of V-optimal design for estimating K, $eff_K(\xi_V)$, and efficiencies of K-optimal design for estimating V, $eff_V(\xi_K)$.

as the right-end point of the design space increases without bound.

More generally, suppose we are interested to estimate $c^{\mathrm{T}}\theta$ and $c^{\mathrm{T}} = (c_1, c_2)$, $c_1 \neq 0$. Clearly, this design problem is the same as that when $c^{\mathrm{T}} = (1, c_2/c_1)$, say $(1, cK/V)$ and c is arbitrary. A similar argument using the Elfving set shows that if $-(1+b) \leqslant c \leqslant -(1+b)/(1+(2-\sqrt{2})b)$, we have a one-point optimal design at $-(1+c)K$ and the criterion value is $(c/(1+c))^2$; otherwise, the optimal design for estimating $c^{\mathrm{T}}\theta$ is a two-point design supported at the same support points as the K- or V-optimal design with the weight at s_cK equal to $(1+b+c)/(\sqrt{2}(1+b+c)+(3\sqrt{2}-4)bc)$. The value of this criterion (i.e. the variance of the estimate) for the c-optimal design is

$$\Phi_c(\xi_c) = \frac{(3+2/\sqrt{2})((1+b+c)+bc)^2(1+b)^2}{b^4}\frac{K^2}{V^2}.$$

3.2. *D*-OPTIMAL DESIGN

A popular design for estimating the two parameters simultaneously is the D-optimal design. This design, ξ, minimizes the generalized variance of the estimated parameters and it is equally supported at s_DK and bK, where $s_D = b/(2+b)$ (Duggleby, 1979). As expected, the D-optimal design does not depend on the parameter V because the MM model is a partially nonlinear regression model (Hill, 1980). To assess the performance of the D-optimal design for estimating each of the parameters, we first verify that

$$\Phi_K(\xi_D) = 40\left(\frac{1+b}{b}\right)^4\frac{K^2}{V^2}$$

and

$$\Phi_V(\xi_D) = \frac{2(1+b)^2(20+4b+b^2)}{b^4}.$$

A direct calculation shows the efficiencies of the D-optimal design for estimating K and V are

$$eff_K(\xi_D) = \frac{\Phi_K(\xi_K)}{\Phi_K(\xi_D)} = 0.84926$$

and

$$eff_V(\xi_D) = \frac{\Phi_V(\xi_V)}{\Phi_V(\xi_D)} = \frac{(3+2\sqrt{2}+b)^2}{2(20+4b+b^2)} \geqslant 0.5.$$

The maximum of the last efficiency is reached at the point $b = 2.1796$ and tends to 0.5 as b becomes arbitrarily large (Fig. 2). For this choice of b, the efficiency for estimating K is about 85% and the efficiency for estimating V is more than 95%. The $K(V)$-optimal design has about 87.2%(84.9%) efficiency for estimating the parameter $V(K)$. This suggests that $b = 2.1796$ is a reasonable choice when we want to estimate both the parameters in the MM model.

4. Compound Optimal Designs

Duggleby (1979) noted that in the MM model, the Michaelis–Menten constant, K, is always more interesting than V. This implies that it is desirable to have a design that can deliver greater efficiency for estimating the more important parameter K. To find such a design, consider the standardized compound criterion given by

$$\Phi_\lambda(\xi) = \lambda\frac{\Phi_K(\xi)}{\Phi_K(\xi_K)} + (1-\lambda)\frac{\Phi_V(\xi)}{\Phi_V(\xi_V)} = \frac{\lambda}{eff_K(\xi)} + \frac{1-\lambda}{eff_V(\xi)},$$

where $0 \leqslant \lambda \leqslant 1$ is a user-selected constant. The criterion is a convex combination of two convex criteria and so the criterion is still convex and the optimization problem is essentially similar to that for a single-objective design problem. A design that minimizes $\Phi_\lambda(\xi)$, for fixed λ, is called a compound optimal design. The value of λ represents the weight to be assigned to each of the criterion; if λ is near unity, this suggests that estimating K is more important. Because it is possible that the variances can have very different magnitudes, we standardize them by dividing by their optimal values. To simplify the search of the optimal design, it is convenient to first fix λ, and find an optimal design to minimize $\Phi_\lambda(\xi)$ among designs of the form

$$\xi_{z,p} = \begin{pmatrix} zK & bK \\ p & 1-p \end{pmatrix}.$$

For fixed b and λ, we seek values for z and p that minimize $\Phi_\lambda(\xi)$. If the resulting design is optimal among all designs, the equivalence theorem (1) requires that these optimal values satisfy

$$\left(\frac{\partial\psi}{\partial s}\right)_{s=zK} = 0 \quad \text{and} \quad \psi(zK, \xi_{z,p}) = 0.$$

The first of these equations can be simplified using the fact that

$$\nabla\Phi_\lambda(\xi_{z,p}) = \\ -M^{-1}(\xi_{z,p})\begin{pmatrix} \dfrac{\lambda}{\Phi_K(\xi_K)} & 0 \\ 0 & \dfrac{1-\lambda}{\Phi_V(\xi_V)} \end{pmatrix} M^{-1}(\xi_{z,p}).$$

Table 2 shows some of these designs and their efficiencies for $b = 5$ and selected values of λ for estimating K and V. From the table, we deduce that if we wish to have a design that is roughly equally efficient for estimating both V and K, we should set $\lambda = 0.5$. Using the equivalence theorem in eqn (1), one can verify that these designs are also optimal within the class of all designs.

It is instructive to plot the two efficiencies of the compound optimal design for estimating V and K vs. values of λ between 0 and 1. As shown in Cook & Wong (1994), one of the efficiencies is non-increasing as a function of λ, and the other is non-decreasing. The rate of increase or decrease in the plot signifies how much the experimenter has to give up in efficiency under one criterion for a gain in the other. The point of intersection of the two plots gives the value of λ for which the corresponding compound optimal design is equally efficient under both criteria. Imhof & Wong (2000) showed that this same design is also a maximin optimal design in the sense that the design maximizes the minimum efficiency under both criteria. Thus, the efficiency plot can be used to study the tradeoff between the two competing objectives and help the researcher arrives at a compromise design. As an illustration, suppose $b = 5$, and we wish to find a design which will ensure that the more important parameter K is estimated with at least 95% efficiency and, subject to this constraint, estimate V as accurately as possible. The efficiency plot (not shown) provides the answer: it is the compound optimal design corresponding to $\lambda = 0.681$ and supported at $0.55K$ and $5K$ with mass at $0.55K$ equal to 0.54. Justifications of this claim and other applications of the efficiency plots can be found in Cook & Wong (1994), Huang & Wong (1998a, b) and Zhu & Wong (2000).

5. Sequence Designs

5.1. GEOMETRIC DESIGNS

The optimal designs obtained so far for estimating parameters in the MM model have one or two points. Consequently, these designs do not permit one to conduct a lack of fit test for checking model adequacy. To overcome this drawback, designs used, in practice, have more than two points. For instance, Cleland (1967) proposed a range of substrate concentrations from $0.2K$ to $5K$, arranged such that the reciprocal forms an arithmetic sequence. A more popular class of designs is geometric designs. Duggleby (1979) used geometric designs with substrate concentrations set at $0.25K$, $0.5K$, K, $2K$, $4K$ or $0.5K$, K, $2K$. Likewise, Currie (1982) considered only geometric designs in a pharmacokinetic study and found optimal geometric designs by

TABLE 2

*Optimal compound designs and their efficiencies for estimating the parameters K and V, for different values of λ and $b = 5$**

λ	0	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9	1
z	0.527	0.537	0.543	0.547	0.55	0.551	0.551	0.549	0.545	0.538	0.527
p	0.381	0.415	0.447	0.477	0.506	0.536	0.565	0.596	0.628	0.665	0.707
eff_K	68.9	74.3	79.0	83.1	86.7	90.0	92.9	95.5	97.6	99.3	100
eff_V	100	99.5	98.1	96.2	93.7	90.8	87.4	83.5	78.8	73.2	66.0

*The support points of these designs are zK and bK with mass p at zK.

TABLE 3
Efficiencies of optimized 3-, 7- and 10-point geometric designs for estimating the parameter K in the Michaelis–Menten model

a	r			eff_K (ξ_m)			eff_V (ξ_m)		
	$m = 3$	$m = 7$	$m = 10$	$m = 3$	$m = 7$	$m = 10$	$m = 3$	$m = 7$	$m = 10$
0.1	2.01	1.33	1.21	85.3	65.6	60.5	86.3	61.5	55.6
0.2	2.23	1.40	1.27	82.3	63.7	59.6	82.8	55.0	48.8
0.5	2.70	1.59	1.40	70.8	56.3	53.9	66.5	43.3	40.1

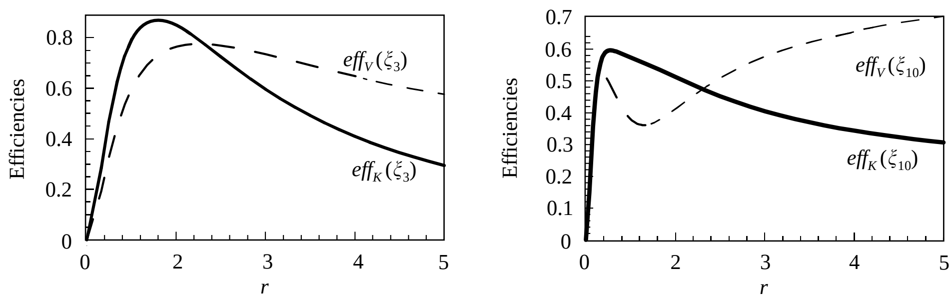

FIG. 3. Efficiencies of ξ_3 for estimating K and V with $a = 0.3$ (left) and efficiencies of ξ_{10} for estimating K and V with $a = 0.2$ (right).

maximizing the determinant of the information matrix when the number of design points is fixed.

In this section, we consider choosing an optimal geometric sequence design to estimate V or K separately. The geometric design has the form $s_i K = ar^{i-1}K$, $i = 1, 2, \ldots, m$ and our experience suggests that it seems easier to specify the number (m) of design points the researcher is willing to take than pre-selecting the value of a or b. With m and b fixed, it is straightforward to use algebra to determine the optimal value of $a > 0$ or $r > 1$ in the geometric design for the estimation problem. Table 3 displays some of the optimal designs for estimating K when $m = 3$, 7 and 10. In each case, increasing the number of points in these designs always lowers the efficiencies.

Figure 3 shows the efficiencies for estimating K and V for an optimized 3- and 10-point geometric designs. When we use 10 points, the shape for the efficiency for estimating K is similar to that when 3 points are used, but the efficiency pattern for estimating V changes. In both cases, we recommend choosing a small value of a less than unity in line with the common practice of having some observations before K.

Currie (1982) used the sequence design with points at $0.5K$, K, $2K$, i.e. $m = 3$, $r = 2$ and $a = 0.5$. Both the efficiencies of this design for estimating V and K are nearly 70%. For this value of a, the best value for r is 2.70 and the sequence is $0.5K$, $1.35K$, $3.65K$. The efficiencies of this design for estimating the two parameters are shown in Table 3 and so Currie's design is rather efficient. Currie also used other values of m and r in the study and for these values, the optimized geometric designs and their efficiencies are reported in Table 3.

In general, we recommend that the value of a should be small, and subject to this value of a, determine the best value of r for estimating the parameter of interest. For a fixed value of a, the optimal r is found using the efficiency formulas. For instance, suppose we are interested in geometric designs. Let $F(a,r) = (1 + a)(1 + ar)(1 + ar^2)\cdots(1 + ar^{m-1})$, let $F_i(a,r) = F(a,r)/(1 + ar^{i-1})$ and let $F_{i,j}(a,r) = F(a,r)/(1 + ar^{i-1})(1 + ar^{j-1})$. Define $P(a,r) = \sum_{i=1}^{m} r^{2(i-1)}F_i(a,r)^2$, $R(a,r) = \sum_{i=1}^{m} r^{2(i-1)}F_i(a,r)^4$, $Q(a,r) = \sum_{i<j} r^{4i+2j-8}(r^{j-i} - 1)^2 F_{i,j}(a,r)^4$ and denote a generic geometric design on m points with parameters a and r by ξ_m.

A direct calculation shows the efficiencies of ξ_m for estimating V and K are

$$eff_K(\xi_m) = \frac{(1 + ar^{m-1})^2 Q(a,r)}{(3 - 2\sqrt{2})^2 r^{4m-6} F_m(a,r)^2 P(a,r)}$$

and

$$eff_V(\xi_m) = \frac{(1 + ar^{m-1})^2 (1 + (3 - 2\sqrt{2}) ar^{m-1})^2 Q(a,r)}{(3 - 2\sqrt{2})^2 r^{4m-6} m R(a,r)}.$$

When $m = 2$, we have

$$eff_K(\xi_2) = \frac{(r-1)^2 (1 + ar)^2}{2(3 - 2\sqrt{2})^2 (1 + a)^2 r^2 ((1 + a)^2 r^2 + (1 + ar)^2)}$$

and

$$eff_V(\xi_2) = \frac{(r-1)^2 (1 + ar)^2 (1 + 3ar - 2\sqrt{2}ar)^2}{2(3 - 2\sqrt{2})^2 r^2 ((1 + a)^4 r^2 + (1 + ar)^4)}.$$

These formulas are helpful for determining the optimal geometric design. Depending on the problem, the researcher provides a set of nominal values for either one or two of the three parameters and the remaining ones are chosen so that they maximize the efficiency for estimating V or K or both. The formulas are also helpful for understanding the effects of variations in the design parameters. For instance, the degree of the polynomial in a in the numerator of $eff_K(\xi_m)$ is less than that in the denominator. This implies that the efficiency of ξ_m for estimating K tends to 0 when a becomes large.

5.2. COMPARISON OF SEQUENCE DESIGNS

We now compare the performance of different types of sequence designs for estimating the parameters in the MM model. The sequence designs of interest are geometric, harmonic, arithmetic, logarithmic, c^2-arithmetic and inverse linear designs. Harmonic designs have the property that the reciprocal of the design points form a linear sequence. The c^2-arithmetic design have points that are spaced according to the rule $(b - c(i-1)^2)K$, $c > 0$, $i = 1, 2, \ldots$ for some user-selected constant c. For the purpose of comparison, all these designs have the same number of points and are defined by any two of the following three parameters: the first design point (aK), the last design point (typically bK) and a spacing parameter which determines the spread between points. For example, the spacing parameter for the geometric design is the constant ratio (r) between two consecutive points and for the arithmetic design, the spacing parameter is the uniform distance (d) between two consecutive points. One may wish, for instance, to fix the last point and the number of points to find the optimal value of a for estimating V and K using sequence designs with $m = 3$, 5 and 10 points and $b = 3, 5, 10$.

Figure 4 is an illustrative plot showing the efficiencies of the various sequence designs for estimating K and V when the position of the first point varies from 0 to 1, and $b = m = 5$. We observe that when a is very small, all the designs are inefficient for parameter estimation. When $a \geqslant 0.1$, the harmonic sequence design has the highest efficiency for estimating K among the six sequence designs but its efficiency for estimating V drops as a increases. When a approaches unity, the harmonic sequence design has the lowest efficiency for estimating V among the six designs. The geometric sequence is quite stable and has relatively high efficiencies for estimating both parameters for a large range of values of a. The arithmetic design and the c^2-arithmetic designs are generally inefficient. The properties of the inverse linear designs for estimating both the parameters are similar to the geometric designs.

For each of the design sequences, we computed the optimal value of a for estimating V and the optimal value of a for estimating K when $b = 3$, 5 and 10. We studied the pattern of the efficiencies of various sequence designs for estimating each of these parameters and noted that the efficiencies of the optimized sequence for estimating V are quite similar. This suggests that the researcher may want to focus on estimating the more interesting parameter K. As the number of

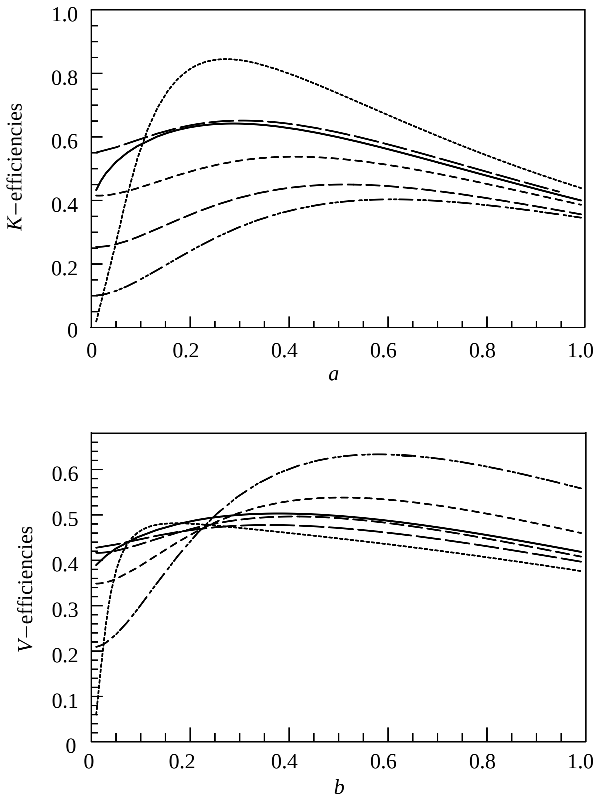

FIG. 4. Efficiencies of estimating K (top) and V (bottom) using different 5-point optimized sequence designs with $b = 5$: harmonic (┈┈┈), inverse linear (——), geometric (——), logarithmic (- - - -), arithmetic (— — —) and c^2-arithmetic (-·-·-·-) sequences designs.

design point increases, the efficiencies of these designs for estimating V decrease rapidly but their efficiencies for estimating K tend to be more stable.

Cressie & Keightley (1981, Table 1) used a 10-point design which can be regarded as nearly a 5-point duplicated design for a hormone-receptor assay with five concentrations of estradiol-17β. The design is not a geometric design but is close. The independent variable corresponding to the amount of hormone not bound is calculated directly from these concentrations. The response variable is the amount of hormone bound to the receptor. We can find improved designs by fixing the end point (as is used in the study) and optimize the first point of the design sequence. The criterion could be for estimating the parameter K or V and the optimal value of a for each of the sequence designs can be computed. In our computation, we used the same set of nominal values for K and V, i.e. $K = 227.27$ and $V = 43.73$ provided in Cressie & Keightley (1981).

6. Extensions

The previous sections discussed designs for estimating parameters in the MM model but Elfving's method has more general applications. We describe two here; one, where there is interest to estimate the ratio $\mu = V/K$ in the MM model and second, there is interest in parameter estimation in the presence of heteroscedasticity. The first ratio is the steepest slope of the MM-curve (at $s = 0$) and is frequently of interest in enzyme-catalysed reactions (Duggleby, 1979). Given a design, ξ, the asymptotic variance of the estimate of the ratio V/K is proportional to $\mathrm{var}_\xi(\hat{\mu}) = \nabla\mu^{\mathrm{T}} M^{-1}(\xi) \nabla\mu$. It is convenient to reparametrize the model as $E(v) = (V/K)s/((s/K) + 1)$ before applying Elfving's method to determine the optimal design for minimizing the above variance. The Elfving set is now generated by $f(S) = \{g_2 = -\mu g_1^2\colon\ g_1 \in [0, [K/(1 + b)]b]\}$ and using Elfving's argument, the optimal design is

$$\xi_\mu = \begin{pmatrix} s_\mu b & bK \\ \dfrac{2 + \sqrt{2}}{4} & \dfrac{2 - \sqrt{2}}{4} \end{pmatrix},$$

where $s_\mu = b/(1 + \sqrt{2} + \sqrt{2}b)$.

The variance of $\hat{\mu}$ using the optimal design is $2(1 + b)/(\sqrt{2} - 1)bK)^2$. The variances of the same estimate using the D- and K-optimal designs are

$$\mathrm{var}_{\xi_D}(\hat{\mu}) = \frac{34(1 + b)^2}{b^2K^2}$$

and

$$\mathrm{var}_{\xi_K}(\hat{\mu}) = \frac{(9\sqrt{2} + 13)(1 + b)^2}{b^2K^2}.$$

A direct calculation shows the efficiencies of these two designs for estimating V/K are 68.6 and 90.6%, respectively, independent of b. The efficiency of the V-optimal design for estimating V/K is

$$\frac{4(1+\sqrt{2}(\sqrt{2}-1)b)}{3+\sqrt{2}+2(3-\sqrt{2})b+(\sqrt{2}-1)b^2}$$

and this ratio approaches 0 when b increases without bound. Duggleby (1979) claimed without justification that the D-optimal design is also optimal for estimating the steepest slope V/K. Our results show that the D-optimal design is not optimal for estimating V/K.

Endrenyi (1981, p. 137) and Currie (1982) incorporated heteroscedasticity in the MM model. Endrenyi (1981) assumed the error variance is proportional to the mean and the Currie (1982) extended it to the case when the variance is a simple function of the mean. Elfving's method can also be applied to obtain optimal designs for this more complicated situation. As an illustrative example, we consider one of the error variance functions discussed in Currie (1982) and Dette & Wong (1999) where $\mathrm{var}(v) = d\exp(eVs/(K+s)), s \in [0.125, 64]$, e is arbitrary and d is any positive number. When $s \in [0, b]$, the regressor vector is

$$\left(\frac{-Vs}{(K+s)^2}, \frac{s}{K+s}\right)\sqrt{\frac{1}{d}\exp\left(\frac{-eVs}{K+s}\right)+\frac{e^2}{2}}$$

and

$$f(S) = \left\{(g_1, g_2) = \left(\frac{-Vs}{(K+s)^2}, \frac{s}{K+s}\right) \times \sqrt{\frac{1}{d}\exp\left(\frac{-eVs}{K+s}\right)+\frac{e^2}{2}} : s \in [0, b]\right\}.$$

The Elfving set (not shown) is similar to that of the homoscedastic case (Fig. 1) except that it is not as symmetric as before. The c-optimal designs for estimating each of the parameters V and K are determined as before from the Elfving set. Both designs are supported at s_cK and bK with mass at s_cK equal to p_c. If we denote derivatives by primes, it can be shown that the value of s_c is the root of the equation

$$\frac{g_1(b)+g_1(s_c)}{g_1'(s_c)} = \frac{g_2(b)+g_2(s_c)}{g_2'(s_c)}$$

and p_c is the weight found geometrically from the Elfving set.

7. Summary

In this paper, we addressed design issues for the MM model. Our objective was to find a design that would provide the best-possible estimate for one or two of the parameters, or some function of the model parameters. We also constructed compound optimal designs when there are two objectives in the experiment and there may be unequal interest in each of the objectives. The compound optimal designs can balance the competing needs of the study according to the user requirements. It is clear that the same methodology can be extended to situations when there are more than two objectives in the experiment.

In addition, we studied properties of these designs and used them as benchmarks to compare the merits of several widely used design sequences for estimating the model parameters. Our results suggest that the geometric and the inverse linear designs tend to outperform other design sequences for estimating the parameters in the MM model. It is interesting to note that our results here show that some of the designs used in practice are actually quite efficient. For example, Watts' and Cressie and Keightley provided no theoretical justifications for their designs and yet their designs compare quite favorably with our optimized sequence designs. The work here attempts to bridge the gap between theory and practice, and demonstrate the usefulness of Elfving's method for finding optimal designs in biometry.

The computer algorithm developed here for optimizing the parameters of a geometric design under various scenarios is based on the software *Mathematica*. A computer code is available from the first author. The computer program generates the best geometric design after the user fixes some of the geometric design parameters. Currently, we are constructing additional user-friendly algorithms that would automatically generate other types of optimized design sequences for the MM model and related models.

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