



RESEARCH ARTICLE

Design efficiency in genetic association studies

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Abstract

Selecting the best design for genetic association studies requires careful deliberation; different study designs can be used to scan for different genetic effects, and each design has its own set of strengths and limitations. A variety of family and unrelated control configurations are amenable to genetic association analyses, including the case-control design, case-parent triads, and case-parent triads in combination with unrelated controls or control-parent triads. Ultimately, the goal is to choose the design that achieves the highest statistical power using the lowest cost. For given parameter values and genotyped individuals, designs can be compared directly by computing the power. However, a more informative and general design comparison can be achieved by studying the relative efficiency, defined as the ratio of variances of two different parameter estimators, corresponding to two separate designs. Using log-linear modeling, we derive the relative efficiency from the asymptotic variance of the parameter estimators and relate it to the concept of Pitman efficiency. The relative efficiency takes into account the fact that different designs impose different costs relative to the number of genotyped individuals. We show that while optimal efficiency for analyses of regular autosomal effects is achieved using the standard case-control design, the case-parent triad design without unrelated controls is efficient when searching for parent-of-origin effects. Due to the potential loss of efficiency, maternal genes should generally not be adjusted for in an initial genome-wide association study scan of offspring genes but instead checked post hoc. The relative efficiency calculations are implemented in our R package Haplin.

KEYWORDS

case-parent triad, Haplin, parent-of-origin effects, power and sample size, relative (Pitman) efficiency

1 | INTRODUCTION

Optimizing the design of a genetic association study requires careful consideration because (among other things) there are several factors to assess (eg, recruitment costs, genotyping costs, phenotypic costs, statistical power, and design-induced

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biases). The most common design for genetic association analysis is the standard case-control design in which individuals with and without the disease in question are genotyped. By contrast, if case-parent triad data are collected by genotyping cases and their biological parents, parent-of-origin (PoO) effects or direct effects of the maternal genome during fetal development (ie, maternal effects) can also be investigated.¹⁻⁴ Case-parent triads can also be combined with unrelated control-parent triads in a hybrid design.⁵⁻⁹ Although the case-parent triad design is mostly used when the outcome occurs early in life, this design can be used for any condition, provided that parents are available for genotyping.

The statistical power is an important aspect of design comparison. Frequently, study designs are compared directly through a power analysis without considering the total number of individuals that needs to be genotyped. For example, a fixed number of complete case-parent triads could be compared with the same number of case-mother dyads. However, this approach ignores the costs of data collection. In this article, our objective is to present comparisons that enable the highest statistical power to be achieved using the smallest sample collection and assay costs. We assess this through the quantity known as relative efficiency, defined as the ratio of variances of estimators for the same parameter computed from two different designs, or equivalently, the ratio of the sample sizes needed for each of the two designs to achieve the same significance level and power. We demonstrate how the relative efficiency measures relate to the concept of Pitman efficiency.¹⁰

We have previously developed an extensive framework for genetic epidemiological analyses of binary traits based on log-linear modeling, implemented in the R package Haplin.^{4,11-13} Haplin includes a complete setup for power and sample size calculation,^{14,15} which is useful in study planning and in interpreting findings from a genome-wide association study (GWAS). In this article, we present a structured overview of different genetic effects and etiologic scenarios that are applicable to diseases with onset throughout the lifespan, along with appropriate choices of study designs. Our primary focus is on estimating the relative efficiency, which is readily assessed within the power calculation framework of Haplin.

The article is structured as follows. First, we introduce the relevant genetic effects and the family-based designs that are the focus of this article. Second, we describe our sampling and penetrance models, explain the concept of relative efficiency, and illustrate its association with statistical power. Finally, we study the relative efficiency of different designs for different genetic effects, both for single-nucleotide polymorphisms (SNPs) and for haplotypes, that is, the combinations of alleles from several SNPs within a locus. Although we focus on autosomal markers, the methodology presented is readily applicable to SNPs or haplotypes on the X chromosome. A discussion of relative efficiency is provided in Appendix A. In Appendix B, we provide a heuristic derivation of the relative efficiency for regular autosomal effects. To facilitate analysis of other genetic mechanisms, study designs, and input parameters, we provide Haplin commands for various scenarios on the Haplin website at <https://people.uib.no/gjessing/genetics/software/haplin>.

2 | BACKGROUND

The R package Haplin is a comprehensive framework for genetic association analyses of binary traits based on log-linear modeling.⁴ It implements a full maximum-likelihood model for estimation and calculates explicit estimates of relative risks with asymptotic standard errors (SEs) and confidence intervals. Haplin enables the estimation of regular autosomal effects, PoO effects, and maternal effects, as well as interactions between genetic effects and categorical or ordinal exposure variables.^{11,13} It allows for parallel processing of analyses as well as data structure for handling GWAS data. In Haplin, the main unit of analysis is the case-parent triad. However, the log-linear model can readily incorporate unrelated controls or control triads that are population-based (ie, of unknown disease status), or, under the rare disease assumption, unaffected controls or control triads.^{7,16,17} Note that unrelated controls are optional since “pseudocontrols” in principle can be derived from the nontransmitted parental alleles in case-parent triads.¹⁸⁻²¹ To account for unknown parent of origin in ambiguous (uninformative) triads, for example, when the mother, father, and child are all heterozygous for the same two alleles, Haplin uses the expectation maximization (EM) algorithm.²² The EM algorithm also accounts for individuals that are missing “by design,” such as when case-parent triads are reduced to case-mother dyads due to missing data on fathers, assuming that the missingness is random, that is, independent of genotype. The log-linear model in Haplin assumes Mendelian transmission, Hardy-Weinberg equilibrium (HWE), and random mating, although moderate deviations from HWE are unlikely to cause bias.²³ A detailed description of the underlying model is provided in several of our previous publications.^{4,11,13} For applications of Haplin to GWAS data, readers are referred to some of our previous publications.²⁴⁻²⁹

TABLE 1 Overview of genetic effects available in Haplin

Effects	Description
Regular autosomal	A regular autosomal effect is a standard effect of the offspring's own genes. It occurs when a variant allele inherited from one or both parents increases or decreases the risk of a condition.
PoO	A PoO effect occurs if the effect of a variant allele in an individual depends on whether it is inherited from the mother or from the father. Hypothetically, an allele might be protective when inherited from the mother but detrimental when inherited from the father. In statistical terms, we define a PoO effect as an interaction since the effect of an allele is modified by its parent of origin. In contrast, analyses of regular autosomal effects assume that the effect of an allele in an individual is independent of whether it is transmitted from the mother or the father. Note that genomic imprinting may cause PoO effects. ^{42,59} Imprinting is an epigenetic phenomenon where one of the inherited parental alleles is expressed whereas the other is silenced.
Maternal	A maternal genetic effect occurs when a variant allele carried by the mother increases or decreases the risk of a phenotype in her child, regardless of whether the allele has been inherited by the child or not. ³⁴ It is expected to operate mainly via mechanisms in the intrauterine environment. ⁶⁰ This is different from regular autosomal and PoO effects, where we estimate the effects of the child's own alleles. The relevance of maternal effects was recently demonstrated for an individual's educational attainment, ⁶¹ but may be particularly relevant for conditions that depend directly on fetal development.

Note: Adapted from Gjerdevik et al.¹⁴
Abbreviation: PoO, parent-of-origin.

2.1 | Genetic effects

A GWAS scans the entire genome for common variants agnostically, without any prior information about the biological significance of a gene for the trait or disease under investigation. Hence, the selection of an appropriate design for a GWAS requires careful planning and depends heavily on the genetic effect being studied. Haplin enables the estimation of several genetic effects, and we focus here on regular autosomal, PoO, and maternal effects. Table 1 (adapted from Gjerdevik et al¹⁴) provides an explanation of the genetic effects.

2.2 | Study designs

2.2.1 | The case-control design

Similar to classic epidemiological studies of environmental and behavioral risk factors, the case-control design is often used in genetic association analyses (Figure 1A). The allele frequencies of cases and controls are contrasted to identify variants associated with the trait or disease, and familiar methods such as logistic regression and chi-squared tests can be used to discover associations.³⁰ The case-control design is efficient in uncovering regular autosomal effects and their interactions with exposure or stratification variables such as environmental risk factors, study sites, and ethnicity. However, population stratification might lead to spurious associations if not controlled for.

2.2.2 | The case-parent triad and dyad designs

The case-parent triad design involves genotyped cases and their biological parents and is based on the observation that parental genotypes of affected offspring could be used to study associations between a disease and allelic variants.^{31,32} For regular autosomal effects, the frequencies of alleles transmitted to cases are compared to the frequencies of

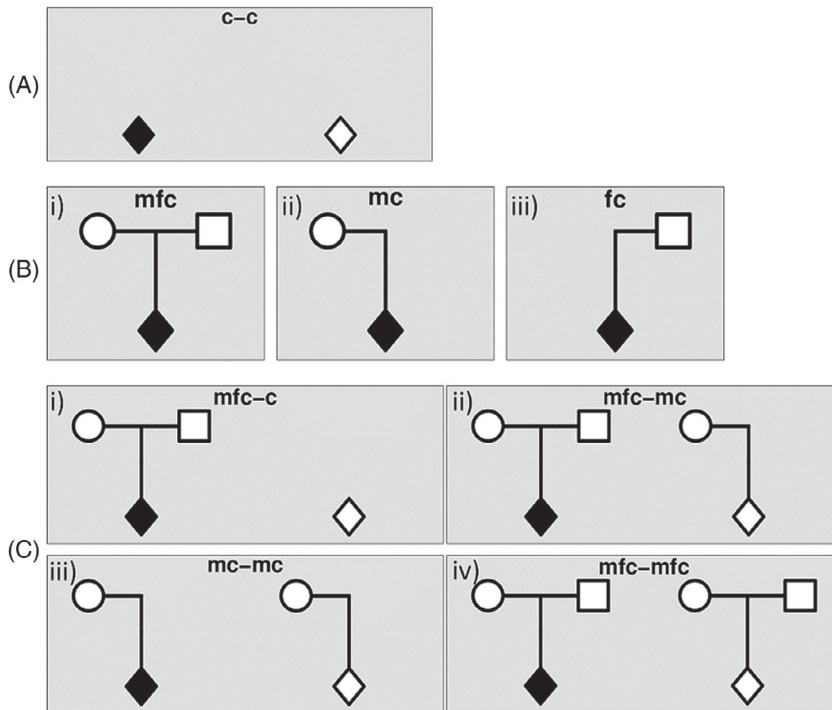


FIGURE 1 A selection of designs for genetic association analyses: (A) case-control design (c-c); (B) various case-parent designs: (i) case-parent triad (mfc); (ii) case-mother dyad (mc); (iii) case-father dyad (fc); (C) a selection of hybrid designs: (i) case-parent triad with independent control (mfc-c); (ii) case-parent triad with independent control-mother dyad (mfc-mc); (iii) case-mother dyad with independent control-mother dyad (mc-mc); (iv) case-parent triad with independent control-parent triad (mfc-mfc)

non-transmitted (pseudocontrol) alleles. Hence, the case-parent triad design does not rely on independent controls and is protected against population stratification since the relevant information is extracted from within-family contrasts. Since Spielman et al³³ proposed the transmission disequilibrium test (TDT) for genetic association testing, exploring family-based designs and their utility for studying different types of genetic effects has been an intense area of research for several decades. Truncated versions of the case-parent triad design have been introduced, with the case-mother and case-father dyad designs comprising genotyped cases and their biological mothers or fathers, respectively. The various constellations are illustrated in Figure 1B. With information on parental genotypes, the case-parent triad and dyad designs allow the estimation and testing of PoO or maternal effects. For PoO and maternal effects, Connolly and Heron³⁴ reviewed different statistical methodologies and compared them according to statistical power and their suitability for studying different etiologic scenarios. Methods for testing PoO effects include extensions of the TDT approach, such as the transmission-asymmetry test (TAT) and the parental-asymmetry test (PAT),³ conditional logistic regression,^{20,21} and log-linear¹⁻⁴ and multinomial modeling.^{17,35,36} With the exception of TAT and PAT, these approaches can also account for maternal effects.³⁴ Despite the inherent strengths of the case-parent triad and dyad designs, there are also some drawbacks. One such drawback is that they rely on Mendelian transmission. Another limitation is that, without independent controls, it is impossible to estimate the main effect of an environmental exposure. There might also be practical concerns, such as obtaining DNA from parents if the disease in question is late onset.

2.2.3 | The hybrid design

To combine the advantages of the case-control and the family-based designs, joint analyses of various combinations of case-parent triads and unrelated controls in a hybrid design have been proposed.^{5-7,17,37} An overview of hybrid designs has been provided by Infante-Rivard et al,³⁸ and different configurations are illustrated in Figure 1C. The full hybrid design comprises complete pairs of case-parent triads and control-parent triads, but truncated versions may include case-parent triads supplemented by control-mother dyads⁹ or case-mother dyads supplemented by control-mother dyads.^{8,39} Analysis methods such as log-linear and multinomial modeling approaches are particularly appealing as they can readily be adapted to accommodate the broad spectrum of various hybrid designs as well as a wide array of causal scenarios and genetic effects.^{11,13,17,24,27,29,35,40} As an example, they can easily be extended to include the maternal-fetal genotype incompatibility test.⁴¹ Nevertheless, although the hybrid design combines the merits of both the case-control and case-parent designs, a straightforward combined analysis may still be influenced by population stratification or non-Mendelian transmission.

2.2.4 | Notation

We use the abbreviations provided in Figure 1 to describe the study designs. The letters c, m, and f denote the child (case or control), mother, and father, respectively. The left side of the hyphen denotes case families, whereas the right side denotes control families. For instance, mfc denotes the case-parent triad, whereas mfc-c denotes a hybrid design consisting of case-parent triads and unrelated controls (ie, the control parents have not been genotyped). We will use the term hybrid design to describe all constellations of study designs consisting of case families and independent control families, except for the straightforward c-c design. Although a case together with a control dyad or control triad can be seen as a hybrid design, these designs are rare in practice and will not be discussed.

3 | METHODS

3.1 | Parameterization of penetrances

We have developed a complete setup for power and sample size calculations in Haplin.¹⁴ The calculations can be performed analytically using the asymptotic variance-covariance structure of the parameter estimator or by a straightforward simulation procedure. Relative efficiency is easily assessed within this framework, and the basic calculations are for regular autosomal, PoO, and maternal effects, with the results depending on the underlying parameterization models. The penetrance models, that is, the probability of a child having the disease conditional on a specific genetic composition, are defined in Table 2 (adapted from Gjerdevik et al¹⁴). For regular autosomal effects, the penetrance model is parameterized as $B \cdot RR_j RR_l RR_{jl}^*$, where B serves as a baseline parameter, and RR_j is the relative risk associated with allele A_j . The double-dose parameter RR_{jl}^* measures the deviation from what would be expected in a multiplicative dose-response relationship, that is, $RR_{jl}^* = RR_j^*$ when $j = l$ and $RR_{jl}^* = 1$ when $j \neq l$. The double-dose estimates provide information about the effect of allele dose on risk. For a diallelic SNP with reference allele A_1 , the penetrance model can be written as $P(D|A_1A_1) = B$, $P(D|A_1A_2) = B \cdot RR$ and $P(D|A_2A_2) = B \cdot RR^2 RR^* = B \cdot \tilde{R}R$. A recessive effect of A_2 would then be seen as $RR = 1$ and $\tilde{R}R \neq 1$, a dominant effect would mean that $RR = \tilde{R}R \neq 1$, and a multiplicative dose-response relationship would be seen as $\tilde{R}R = RR^2$ (see Gjessing and Lie).⁴

Since a mother and her child have one allele in common, maternal effects might be statistically confounded with regular autosomal or PoO effects of the child's own genes.^{42,43} An important feature of the log-linear model is, therefore, the possibility of incorporating and adjusting for maternal effects. Specifically, maternal effects can be addressed simultaneously with regular autosomal or PoO effects by including the maternal risk parameters, as outlined in Table 2. Statistically, we are thus able to separate the effects of maternal alleles from the effect of maternally-derived alleles carried by the offspring.

TABLE 2 Parameterization of penetrances

Effects	Parameterization of Penetrances
Regular autosomal	$B \cdot RR_j RR_l RR_{jl}^*$
PoO	$B \cdot RR_{M,j} RR_{F,l} RR_{jl}^*$
Regular autosomal and maternal	$B \cdot RR_j RR_l RR_{jl}^* \cdot RR_i^{(M)} RR_j^{(M)} RR_{ij}^{(M)*}$
PoO and maternal	$B \cdot RR_{M,j} RR_{F,l} RR_{jl}^* \cdot RR_i^{(M)} RR_j^{(M)} RR_{ij}^{(M)*}$

Note: B is the baseline risk level associated with the (more frequent) reference allele.

RR_j is the risk increase or decrease associated with allele A_j , relative to B .

$RR_{M,j}$ and $RR_{F,j}$ are the relative risks associated with allele A_j , depending on whether the allele is derived from the mother or the father, respectively. Here, we define a PoO effect as the relative risk ratio $RRR_j = RR_{M,j}/RR_{F,j}$, which is a measure of the risk increase (or decrease) associated with A_j when the allele is transmitted from the mother as opposed to from the father.

RR_{jl}^* estimates deviations from the risk that would be expected in a multiplicative dose-response relationship, that is, $RR_{jl}^* = RR_j^*$ when $j = l$ and $RR_{jl}^* = 1$ when $j \neq l$.

$RR_i^{(M)}$ is the relative risk associated with allele A_i carried by the mother, and $RR_{ij}^{(M)*}$ is the maternal double-dose parameter, with an interpretation analogous to RR_{ij}^* .

We set $RR = 1$ for the reference allele to ensure that the model is not overparameterized.

Adapted from Gjerdevik et al.¹⁴

Abbreviation: PoO, parent-of-origin.

We consider a multiplicative dose-response relationship throughout this article, that is, RR_j^* is kept fixed at 1 for all j and only RR_j is estimated (an analogous interpretation applies for the parameterizations of PoO and maternal effects in Table 2). The estimation of RR_j^* is possible and would allow other response models, for example, recessive or dominant, but these situations are not explored herein.

The statistical inference of the log-linear model in Haplin is based on log-transformed relative risks and relative risk ratios using the Wald test. We calculate the relative efficiency based on the asymptotic variance-covariance structure of the parameter estimator, and a derivation of the asymptotic variance-covariance matrix is given by Gjerdevik et al.¹⁴ However, the simulation procedure in Haplin is equally applicable and has been shown to provide similar results within the range of sample sizes and allele frequencies usually studied.¹⁴ For external validation, the power calculation modules in Haplin have previously been compared with the power attained in data simulations by EMIM (Estimation of Maternal, Imprinting, and interaction effects using Multinomial modelling),^{17,35,36} which is another well-established tool for the estimation of various genetic effects based on genotype data from a number of different child-parent configurations. The consistency observed between Haplin and EMIM for regular autosomal, PoO, and maternal effects demonstrates the computational accuracy of the inference methods used in both programs and suggests that power and relative efficiency calculations in Haplin are applicable to genetic association studies based on either log-linear or multinomial modeling.¹⁴

3.2 | Asymptotic relative efficiency

Power analysis allows for a comparison of different designs when all parameter values have been specified. It demonstrates the possible scope of a study, that is, what is feasible logistically, and should, therefore, be an essential part of study planning. However, for “global” comparisons of statistical tests, relative efficiency is a more useful measure. In statistical terms, the relative efficiency of two designs is defined as the ratio of sample sizes required for each of the designs to attain the same significance level and power. This is equivalent to the ratio of variances of two different parameter estimators, corresponding to two separate study designs, taking into account that different designs require a different number of individuals to be genotyped. Figure 2 illustrates the relationship of relative efficiency to sample size and power. For regular autosomal effects, the efficiency of the c-c design is approximately 1.5 relative to the mfc design, which is well known from other studies.⁴⁴ For instance, if 1200 individuals (600 cases and 600 controls) are needed to reach a power of 0.8 with the c-c design, 1800 individuals (600 case-parent triads) are required with the mfc design to achieve the same power.

For the purpose of this article, we aim to compare tests asymptotically. Consider the problem of testing the null hypothesis $H_0 : \beta = 0$ versus the alternative $H_1 : \beta \neq 0$ for a fixed nominal level, α , where β is the log relative risk. With a given sample size N , the power of the test converges to 1 as $|\beta| \rightarrow \infty$. Similarly, when β is fixed, the power converges to 1 as $N \rightarrow \infty$. The limiting power functions are identical for all reasonable tests, and such an approach is, therefore, unhelpful. When N increases, the minimum detectable effect size decreases. To make an informative comparison of different designs,

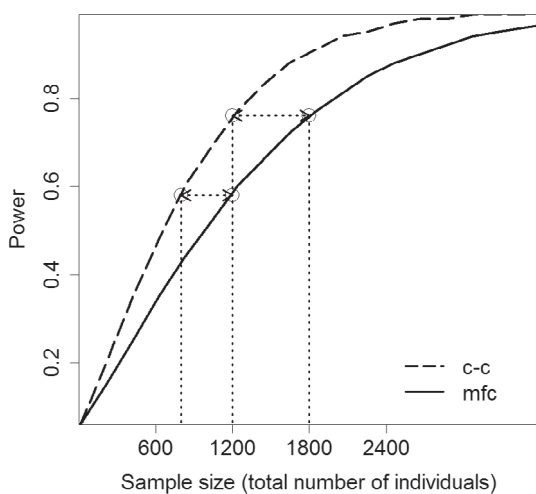


FIGURE 2 Relative efficiency derived from power and sample size. Here, we compare the efficiency of the c-c design relative to the mfc design for regular autosomal effects. The power is calculated for a diallelic SNP at the 5% nominal significance level, using a MAF of 0.2 and an RR of 1.3. The sample size N is defined as the total number of individuals, that is, $N = 1800$ means either 900 cases and 900 controls or 600 case-parent triads. If $N = 1200$, the power is nearly 0.8 for the c-c design. However, approximately $N = 1800$ individuals are required for the mfc design to reach the same power. Similarly, we need $N = 800$ individuals for the c-c design to attain an approximate power of 0.6, whereas $N = 1200$ individuals are required for the mfc design. Hence, the efficiency of the c-c design is 1.5 compared with the mfc design

we, therefore, examine the power at alternatives that approach the null hypothesis, that is, we shrink the alternative as N increases, making it harder to discriminate between the null and alternative hypotheses as the number of observations increases. This is known as the Pitman efficiency,¹⁰ and an explanation of this concept is provided in Appendix A. Most effect sizes reported from genetic association studies of complex traits are small, and empirical studies show that individual relative risks of disease are commonly below two.⁴⁵⁻⁴⁸ Intuitively, the Pitman efficiency is thus a reasonable measure of the asymptotic relative efficiency in our setting.

3.3 | Analyses

We define $k : 1$ as the ratio of control families to case families, regardless of the number of individuals within each family. If $k = 0.5$, we have twice as many case families as control families. For example, for the mfc-mc design, we might have 100 control-mother dyads and 200 case-parent triads. Our main results pertain to the relative efficiency, and we present it here as a function of k on the log-scale. The efficiencies of various study designs are compared with that of the case-parent triad design (mfc), that is, we use the case-parent triad design as a “reference design.” As mentioned previously, the relative efficiency will take into account the total number of genotyped individuals within each design. For example, 150 case-mother dyads are compared with 100 case-parent triads. If $k = 1$, a hybrid design with 50 case-parent triads and 50 control-parent triads is compared with 100 case-parent triads, and if $k = 2$, a hybrid design with 50 case-parent triads and 100 control-parent triads is compared with 150 case-parent triads. Only the ratio of control families to case families, not the actual number of control and case families, affects the relative efficiency estimates.

In genetic association studies, it makes sense to integrate data collection and assay costs with the concept of relative efficiency. For example, if the recruitment of case children occurs at a hospital where parents are likely to be present, parental pseudocontrols would be less expensive than independent controls. However, when studies are nested within a cohort that has already been sampled, the costs of genotyping DNA samples are typically considered equal for all individuals. Hence, for the majority of this article, the data collection costs are simply defined as the number of genotyped individuals. That is, we assume the same costs for all individuals, independent of the individual being a child, mother or father, case or control. However, differential costs of data collection may occur if, for instance, publicly available reference samples (eg, from catalogs such as the Wellcome Trust Case Control Consortium,⁴⁹ the UK Biobank,⁵⁰ and the Norwegian Mother, Father and Child Cohort Study^{51,52}) are included in the study. As a special scenario, we analyze situations in which controls or control families are available without additional costs. For all analyses, we consider well-defined and clinically verified phenotypes, thus ignoring the costs of phenotyping.

The analyses were performed using the Haplin relative efficiency calculator `hapRelEff`. The results were obtained under the null hypothesis, corresponding to the Pitman efficiency.¹⁰ However, we note that relative efficiency estimates in Haplin can also be obtained under alternative (nonnull) hypotheses, and investigators can readily apply our functions to study how alternative effect estimates relevant to their own research question would affect the relative efficiency values.

4 | RESULTS

4.1 | Regular autosomal effects

Figure 3 illustrates the relative efficiency for regular autosomal effects as a function of k , using two different values of the minor allele frequency (MAF). We used the mfc design as the reference, to which the other designs were compared. Unless the ratio of controls to cases is highly skewed, we see that the c-c design provides the best results. The optimal relative efficiency is achieved when $k = 1$. Moreover, we observe that the mfc design is more efficient than the mc or fc design. This result is independent of k , as no control families are sampled. Note that the contribution of a case mother or control mother is equal to the contribution of a case father or control father, respectively. We also see that the relative efficiencies of the hybrid designs decrease when two or three individuals are included in the control family. This is also observed when k becomes sufficiently large. Furthermore, for designs consisting of case dyads or control dyads, that is, mc, fc, mc-mc, fc-fc, mfc-mc, and mfc-fc, the relative efficiency is influenced by the MAF. The MAF does not affect the relative efficiency of the c-c, mfc-c, and mfc-mfc designs.

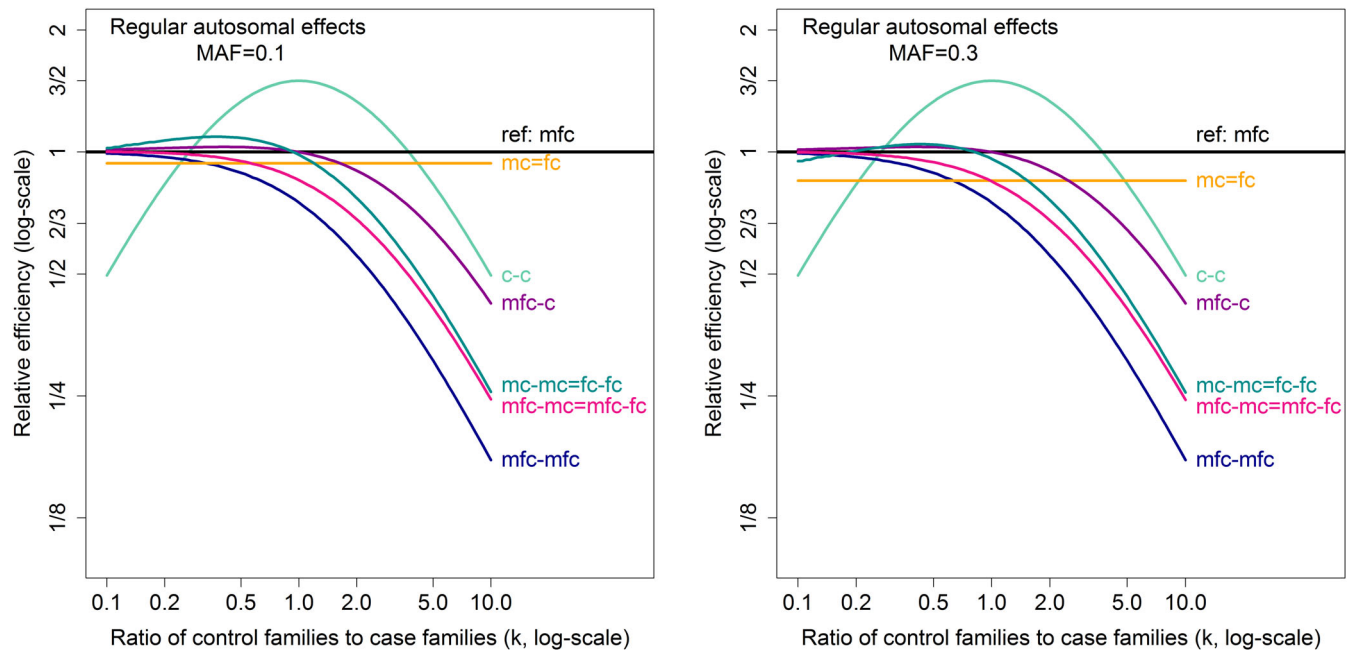


FIGURE 3 Relative efficiency of regular autosomal effects for a given ratio of control families to case families (k). The efficiency of different study designs is compared with that of the case-parent triad design (mfc) under the null hypothesis of $RR=1$. The equality sign (eg, $mc=fc$) denotes that the two designs are interchangeable in terms of relative efficiency [Color figure can be viewed at wileyonlinelibrary.com]

A heuristic formula for the relative efficiency of regular autosomal effects is derived in Appendix B. Equation (B1) verifies the results of Figure 3, and an inspection of the formula provides a better understanding of the observed relationships between the different study designs and each genotyped individual.

4.2 | PoO effects

Figure 4 shows the relative efficiency for PoO effects as a function of k . Again, we compared the relevant study designs with the mfc design under the null hypothesis of $RRR = RR_M = RR_F = 1$. When the MAF is 0.1 (left panel), the mc and fc designs are more efficient than the mfc design. However, this relationship reverses when the MAF is 0.3 (right panel). PoO effects are primarily estimated in case families, by comparing the frequency of alleles transmitted from mother to child with the frequency of alleles transmitted from father to child. Hence, the relative efficiency decreases when k increases or when the number of genotyped individuals within a control family increases. Moreover, the relative efficiencies of the mfc-c, mfc-mc, mfc-fc, and mfc-mfc designs are not influenced by the MAF.

4.3 | Maternal effects

A putative maternal effect detected in a genome-wide scan may, at closer inspection, turn out to be caused by alleles carried by the offspring.^{42,43} In Haplin, maternal effects are therefore assessed while accounting for the effects of the offspring's own alleles (see Table 2). Figure 5 shows the relative efficiency for maternal effects as a function of k while adjusting for possible regular autosomal effects (left panel) and PoO effects (right panel). The results were calculated under the global null, that is, all relative risks are equal to one, using a MAF of 0.1. Overall, the mfc design is a good choice when adjusting for regular autosomal effects. However, when adjusting for PoO effects, a hybrid design generally performs better for small values of k . In both panels, the relative efficiency of the hybrid designs decreases when the number of genotyped individuals within a control family increases, as well as when k becomes sufficiently large. This was also seen in the above analyses of regular autosomal and PoO effects. Note that we excluded the mc, fc, and fc-fc designs when adjusting for PoO effects because the models based on these designs would become overparameterized.

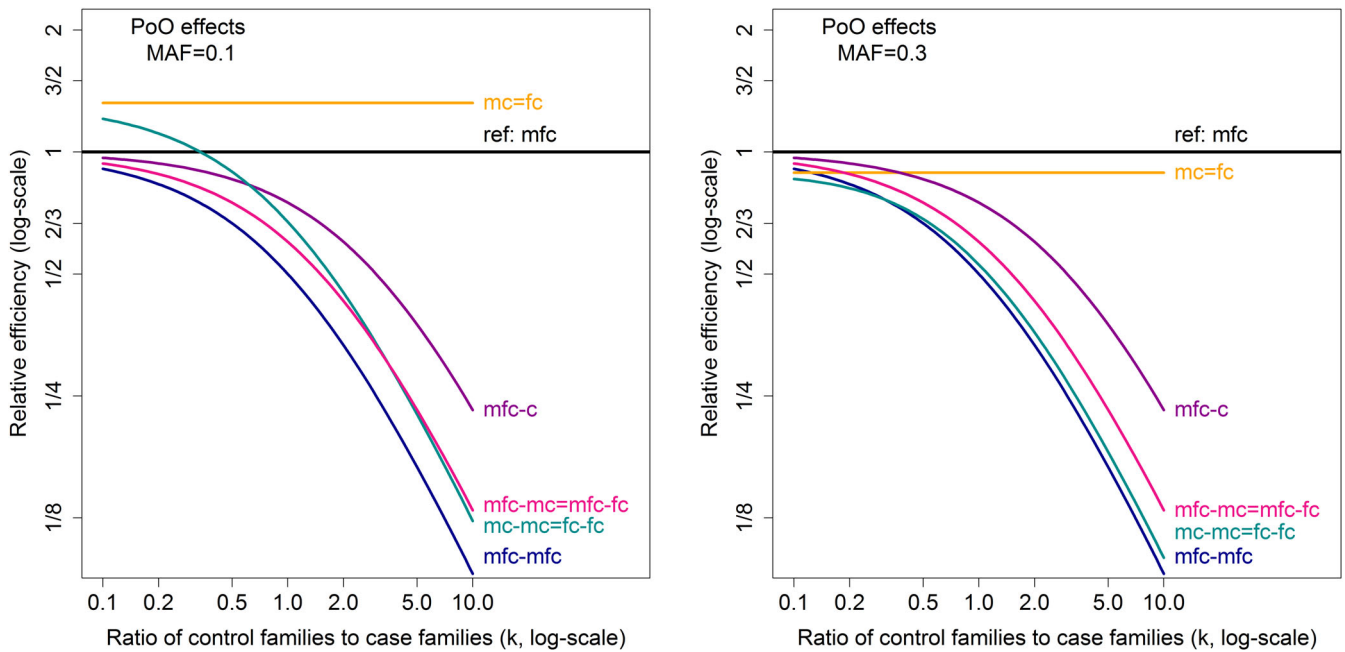


FIGURE 4 Relative efficiency of PoO effects for a given ratio of control families to case families (k). The efficiency of different study designs is compared with that of the case-parent triad design (mfc) under the null hypothesis of $RRR=RR_M=RR_F=1$. The equality sign (eg, $mc=fc$) denotes that the two designs are interchangeable in terms of relative efficiency [Color figure can be viewed at wileyonlinelibrary.com]

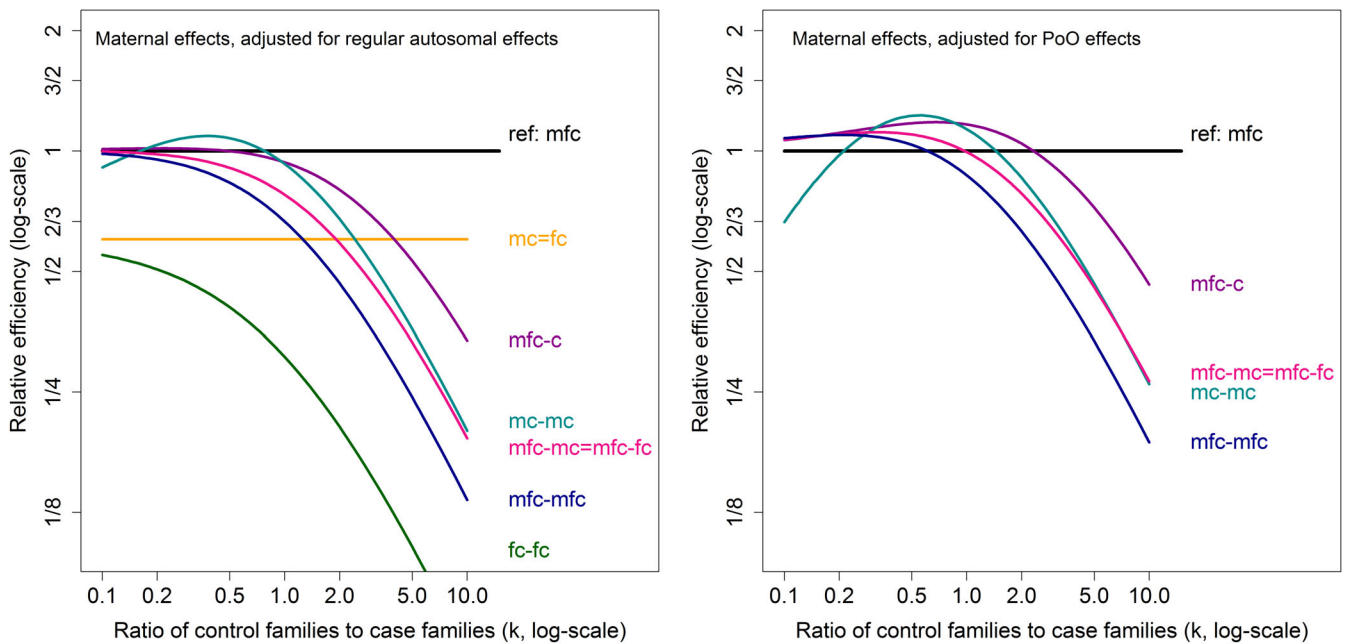


FIGURE 5 Relative efficiency of maternal effects for a given ratio of control families to case families (k). The efficiency of different study designs is compared with that of the case-parent triad design (mfc) under the global null (ie, all RRs are equal to 1). We assumed a MAF of 0.1. The equality sign (eg, $mfc-mc=mfc-fc$) denotes that the two designs are interchangeable in terms of relative efficiency [Color figure can be viewed at wileyonlinelibrary.com]

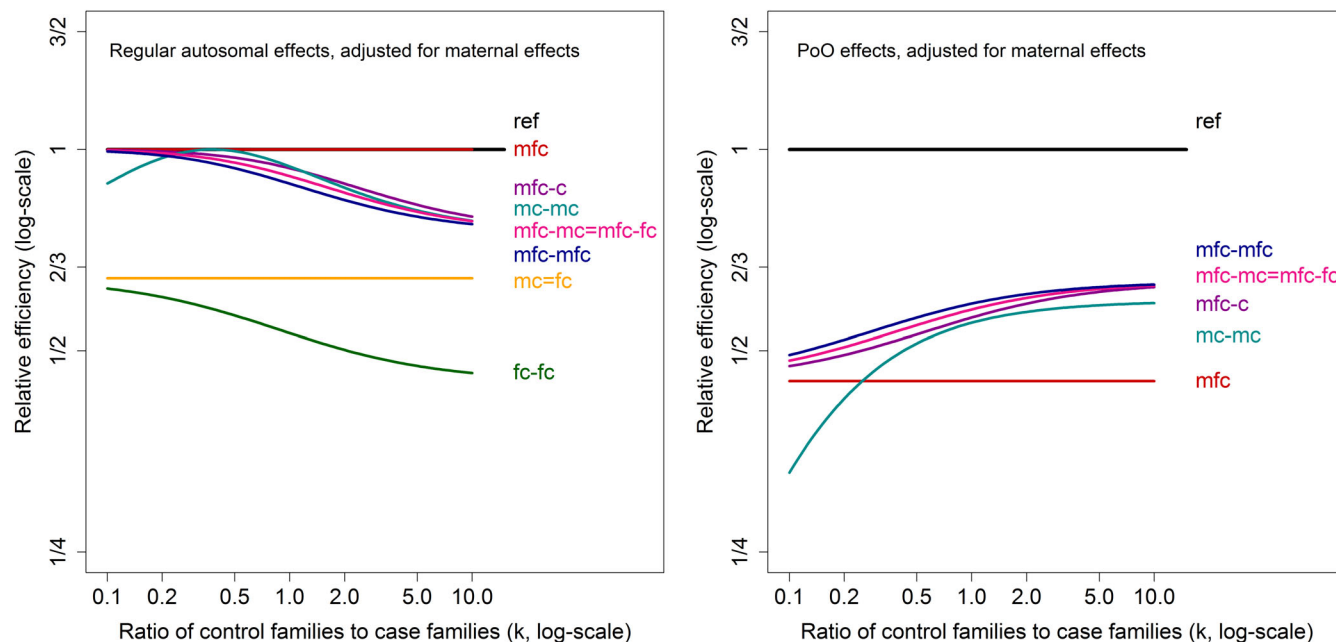


FIGURE 6 Relative efficiency when adjusting for maternal effects for a given ratio of control families to case families (k). For each design, we first adjusted for maternal effects under the global null (ie, all RRs are equal to 1). We then repeated the analysis without adjusting for maternal effects and compared the results. The unadjusted analyses were used as references. We assumed a MAF of 0.1. The equality sign (eg, mfc-mc=mfc-fc) denotes that the two designs are interchangeable in terms of relative efficiency [Color figure can be viewed at wileyonlinelibrary.com]

4.4 | Relative efficiency when adjusting for maternal effects

Including a search for maternal effects in a full GWAS analysis is likely to reduce the power to detect regular autosomal or PoO effects. Figure 6 demonstrates this loss of efficiency as a function of k for regular autosomal effects (left panel) and PoO effects (right panel). We used a MAF of 0.1 in both panels. For each design, we first adjusted for possible maternal effects (even though we did not assume maternal effects in the parameterization model in Table 2, ie, we set $RR^{(M)} = 1$). We then repeated the analysis without adjusting for maternal effects and compared the results. The unadjusted analyses were used as references, and the mfc design is thus no longer a global reference. For regular autosomal effects, adjusting for maternal effects generally decreases the efficiency. However, no loss in efficiency is observed for the mfc design. Although the genotypes of individuals and their mothers are correlated in the population, their contributions to the mfc analysis are close to orthogonal.^{1,2} That is, the estimation of maternal parameters does not affect the estimation of regular autosomal parameters or their SEs, and little bias is introduced for the mfc design (results not shown). When searching for PoO effects, adjusting for maternal effects causes a substantial loss of power for all designs. The efficiency is more than halved for the mfc design.

4.5 | Haplotype reconstruction

The fundamental model in Haplin relates to a single multiallelic locus but extends directly to haplotypes, that is, the sequence of alleles from several closely linked markers within a locus, by statistically reconstructing unknown haplotype phase using the EM algorithm.⁴ A haplotype analysis should enhance the possibility of enclosing a causal variant if the haplotype has a SNP on each side of the variant. However, this analysis might lose power due to haplotype reconstruction and an increased number of degrees of freedom.

In order to assess the relative efficiency when haplotype reconstruction is performed, we considered a situation where one marker with four alleles was compared with two diallelic SNPs. In both scenarios, there were four possible haplotypes (alleles 1, 2, 3, and 4 and SNP-haplotypes 1-1, 2-1, 1-2, and 2-2), with haplotype frequencies 0.1, 0.3, 0.3,

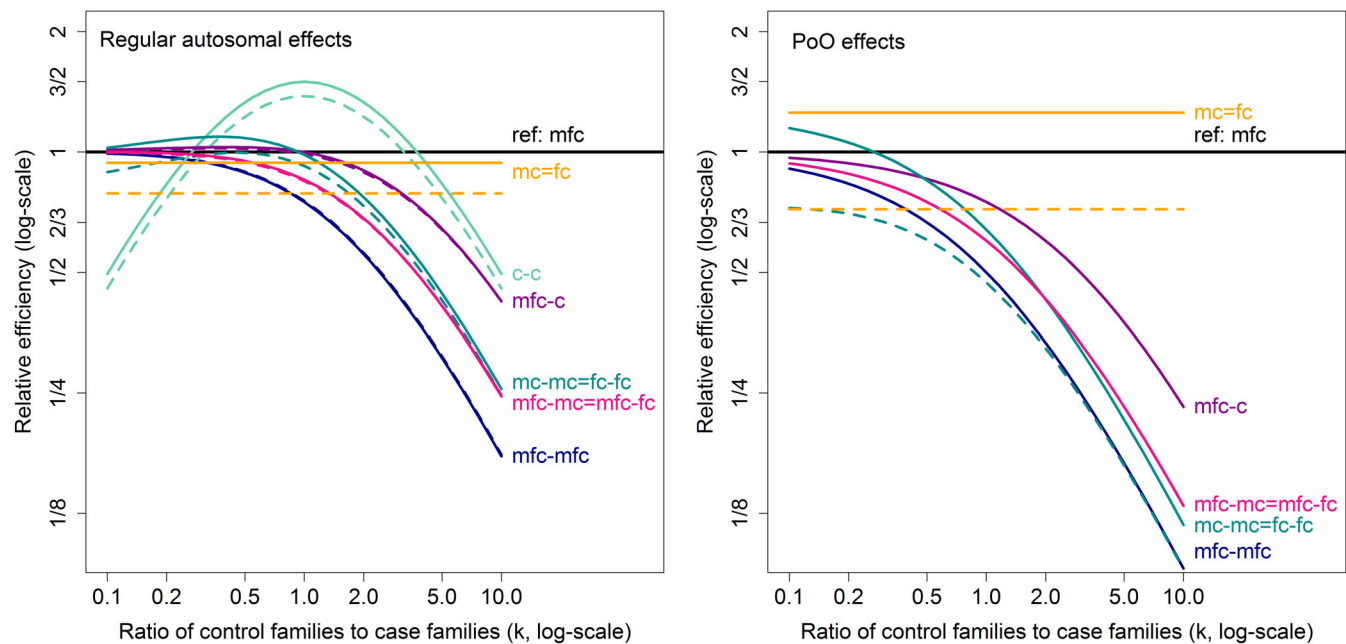


FIGURE 7 Relative efficiency when haplotype reconstruction is performed for a given ratio of control families to case families (k). We constructed four alleles (haplotypes) from a single marker (alleles 1, 2, 3, and 4), and four haplotypes from two diallelic SNPs (haplotypes 1-1, 2-1, 1-2, and 2-2), both with haplotype frequencies 0.1, 0.3, 0.3, and 0.3, respectively, under the global null. A comparison of the solid (single marker, known phase) and dashed (haplotypes from two diallelic SNPs) lines demonstrates the loss of efficiency for the least frequent haplotypes due to haplotype reconstruction, relative to the mfc design. Allele 4 and haplotype 2-2 were used as references. The equality sign (eg, $mc=fc$) denotes that the two designs are interchangeable in terms of relative efficiency [Color figure can be viewed at wileyonlinelibrary.com]

and 0.3, respectively. The alleles are directly observed when derived from a single multiallelic marker, and a haplotype reconstruction is only needed in the analysis of haplotypes from multiple markers. In Figure 7, we considered the efficiency of the least frequent haplotype in all designs, relative to the mfc design, and assessed both regular autosomal and PoO effects. Allele 4 and haplotype 2-2 were chosen as references, respectively. As phase is unknown, haplotype reconstruction for the c-c design is purely a statistical reconstruction. However, if the data from an individual and one or both parents are available at a single locus, the parent of origin can be deduced directly unless all individuals are heterozygous for the same two alleles, such that the EM algorithm is only needed for these ambiguous dyads or triads. Designs that include case-parent triads are, therefore, less vulnerable to unknown phase than the c-c, mc, fc, mc-mc, and fc-fc designs. These findings are in general agreement with those of Douglas et al⁵³ and Schaid.⁵⁴ Note that, in general, the results depend on the haplotype frequencies and also on the reference haplotype (results not shown). The haplotype frequencies used in the example deviate little from their values under linkage equilibrium ($r^2 = 0.0625$). Thus, our analysis demonstrates a larger loss of efficiency than what would be expected when the SNPs are in close linkage disequilibrium. Moreover, haplotype reconstruction in Haplin depends partly on the HWE assumption. Deviations from this assumption can be assessed within the Haplin framework, but such investigations are beyond the scope of this article.

4.6 | The use of external control samples

It has become increasingly common to utilize data from external and publicly available reference or control samples.⁴⁹⁻⁵² Figure 8 illustrates the gains in relative efficiency when external controls or control families are added to the mfc design. The efficiency of the different hybrid designs is compared with that of the mfc design, and the controls are here considered to be free of cost. For regular autosomal effects, we see that the use of freely available control samples increases the efficiency. For PoO effects, however, it has been shown elsewhere that unrelated control samples would not increase the power attained by the mfc design alone.¹⁴ Thus, the relative efficiency of the mfc-c, mfc-mc, mfc-fc, and mfc-mfc designs is equal to 1 for all values of k .

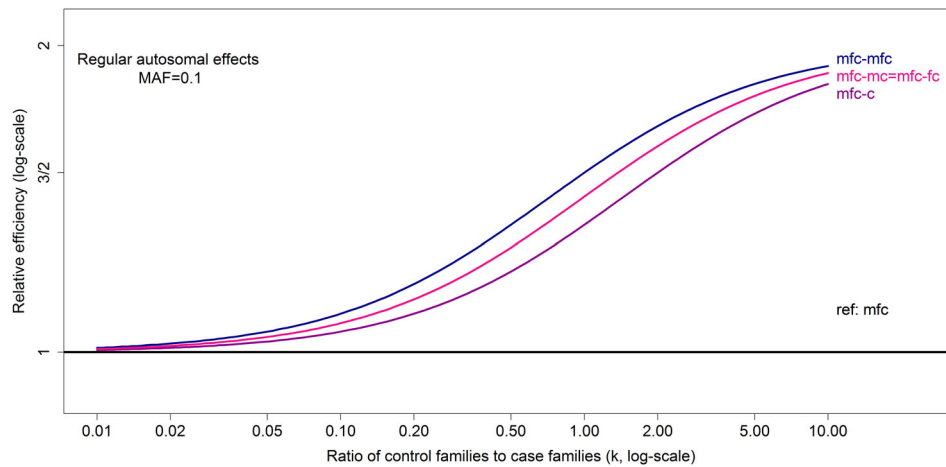


FIGURE 8 Relative efficiency of regular autosomal effects for a given ratio of control families to case families (k). The efficiency of different hybrid designs is compared with that of the case-parent triad design (mfc) under the null hypothesis of $RR=1$. We consider the control samples to be free of charge, that is, without any sampling or genotyping costs. The equality sign (mfc-mc=mfc-fc) denotes that the two designs are interchangeable in terms of relative efficiency [Color figure can be viewed at wileyonlinelibrary.com]

TABLE 3 Application of relative efficiency to cleft palate data

Effects	SNP	MAF ^a	Variance		Empirical Relative Efficiency ^b	Theoretical Relative Efficiency ^b
			Case-mother dyads	Case-parent triads		
Regular autosomal	rs2274616	0.1	0.0387	0.0370	0.96	0.94
	rs12119556	0.3	0.0184	0.0154	0.84	0.85
PoO	rs12137004	0.1	0.0649	0.0868	1.34	1.32
	rs2357649	0.3	0.0399	0.0364	0.91	0.89

^aApproximate estimates.

^bThe case-parent triad design is used as reference.

Abbreviations: MAF, minor allele frequency; PoO, parent-of-origin; SNP, single-nucleotide polymorphism.

4.7 | Application of Haplin to cleft palate only data

Cleft palate only (CPO) is a common craniofacial birth defect in humans, typically classified as to whether the cases occur with (nonisolated) or without (isolated) other congenital anomalies or identifiable malformation syndromes. The overall prevalence of isolated CPO is 5.0 per 10 000 births.⁵⁵ From our previously published GWAS,^{56,57} genotype data from 550 isolated CPO families were available, including 466 complete case-parent triads. These families were primarily of European and Asian ancestry, although other ethnicities were also present in the data. The GWAS data set is available at the dbGaP database (<https://www.ncbi.nlm.nih.gov/gap>) under accession ID phs000094.v1.p1, and information on quality control and detailed characterizations of study participants have been provided elsewhere.²⁵ Background information on the study is given in the original publication,⁵⁶ and ethics approvals were obtained from the respective ethics committees for all the data in the cleft consortium.

To illustrate what the relative efficiencies may amount to with typical MAFs and effect sizes from our example data, we selected a total of 450 complete case-parent triads and chose SNPs with varying MAFs and effect estimates (RR or RRR) close to one for both regular autosomal and PoO effects. For case-mother dyads, the fathers were simply set to missing. To ensure an equal number of genotyped individuals for each design, 300 case-parent triads were randomly drawn from the 450 families using bootstrapping with 101 repetitions. The empirical relative efficiency was then calculated by dividing the median variance of the 101 case-parent triad replicates by the variance of the case-mother dyads. The results are displayed in Table 3, and the findings are in general agreement with the asymptotic calculations shown in Figures 3 and 4.

5 | ADDITIONAL CONSIDERATIONS

5.1 | Gene-environment interactions

A gene-environment interaction (GxE) occurs when a genetic effect is modified by an environmental exposure or a stratification factor such as ethnicity. For example, maternal exposures such as alcohol consumption, smoking, or vitamin intake during the periconceptual period might modify the association between SNPs and a birth defect.^{25,28,29} Interactions between genetic effects and categorical exposure variables are incorporated into the log-linear framework of Haplin by fitting the log-linear model separately for each exposure stratum. A Wald test is then applied to detect whether the relative risk estimates differ significantly across exposure levels.^{11,13} The genetic effect in question might be a regular autosomal, PoO, or maternal effect. Thus, GxE effects can be estimated for all study designs but are restricted to the genetic effects enabled by that design. Note, however, that the main effects of an environmental exposure cannot be estimated from the case-parent triad or dyad design alone without the addition of independent controls.

Because the GxE test stratifies on exposure levels, detecting a GxE effect requires a larger sample size than detecting the genetic effect alone. The SE of a GxE effect is determined by the standard errors of the individual genetic effects in the unexposed and exposed strata.¹³ Provided that the same study design and parameter values are used in each stratum, the relative efficiency estimates are, therefore, directly transferable to GxE effects. Calculated under the global null, that is, $RRR = RR_{\text{exposed}} = RR_{\text{unexposed}} = 1$, Figures 3-5 would also apply to the relative efficiency for GxE effects in these situations.

5.2 | X-chromosome analysis

Haplin allows for analyses of X-linked markers, with corresponding PoO, maternal, and GxE effects. Genetic association analyses of X-linked markers are especially relevant if the prevalence of a complex trait differs systematically between males and females. In Haplin, different X-chromosome models may be fitted depending on the underlying assumptions, including sex-specific baseline risks, shared or different relative risks for males and females, and X-inactivation in females.^{24,40} The methodology presented herein on relative efficiency is readily transferable to genetic effects on X-linked markers. Nevertheless, a discussion regarding sex effects is needed. For instance, when searching for X-linked PoO effects, females are needed to be able to compare maternally- and paternally-derived X-chromosome alleles. However, male individuals and fathers contribute to estimating allele frequencies.^{13,27} They also facilitate haplotype reconstruction because phase can be deduced directly from fathers.

6 | CONCLUDING REMARKS

Statistical power is often a limiting factor for genetic association studies, and no comprehensive software has been available for the full assessment of power and comparison of study designs in such analyses to date. In this article, we provided insights into how relevant designs compare in terms of relative efficiency for a wide range of genetic effects and etiologic scenarios. Furthermore, we illustrated the methodology with extensive analyses and presented results for regular autosomal, PoO, and maternal effects. To facilitate the analysis of power and relative efficiency, the calculations have been implemented in our R package Haplin.¹⁵

The results herein relate to power and efficiency considerations only. Using either a single-SNP or a haplotype approach, the c-c design is recommended when the aim is to search for regular autosomal effects. An equal number of cases and controls maximizes the efficiency. However, additional correction for population stratification may be necessary for the c-c design. For a PoO analysis, the mfc design would be an overall good choice. Note that unrelated control families would not improve the power obtained by the case-parent triad design, as PoO effects are primarily estimated in case families by comparing the frequencies of alleles transmitted from mother to child with the frequencies of alleles transmitted from father to child.¹⁴ Nonetheless, inferences based on the case-parent triad design rely on key assumptions that cannot be fully checked or corrected for without the inclusion of unrelated control families. For maternal effects, the mfc design is appropriate when adjusting for regular autosomal effects, whereas the mfc-c or mc-mc design would be a good choice when adjusting for PoO effects.

Due to the potential loss of power, we do not generally recommend including maternal effects in a full GWAS investigation of regular autosomal or PoO effects. Instead, we suggest additional post-scan analyses to control for possible confounding from maternal effects. As a matter of routine, the most promising SNPs from a GWAS analysis should be further examined for maternal effects.²⁵ However, we note that complex but less likely scenarios where maternal effects cancel out regular autosomal or PoO effects may go undetected by this strategy.

When analyzing real data, one would typically use a combination of several study designs. For example, the data can consist of case-parent triads supplemented by unrelated cases and controls.^{37,38} Such mixture designs are readily handled in Haplin, both in the analysis module and in the power simulation module, but were not illustrated in this article.

The relative efficiency depends on multiple factors, such as the genetic effect in question, the MAF of a given SNP, and the study design. The results are, therefore, hard to summarize. Moreover, the most efficient design to test one hypothesis (ie, casual scenario) is not necessarily the best for testing another hypothesis. If different hypotheses about the modes of inheritance are to be tested, one may prefer a design that is reasonably efficient for a majority of hypotheses rather than the optimal design for a single hypothesis. Hence, since the mfc design is reasonably efficient for the genetic effects studied herein, it may be considered an overall optimal design. The importance of sampling case-parent triads is further strengthened since unrelated, ethnically matched controls have become more easily accessible through publicly available reference samples.⁴⁹⁻⁵²

The concluding recommendations in this article are subject to the log-linear model with the given assumptions, the investigated parameter values, and study designs; they should, therefore, not be interpreted as universal guidelines. Furthermore, practical issues should always be considered, such as the availability of case-parents or suitable controls, as well as recruitment and phenotyping costs. Nevertheless, the methodology presented herein is a useful approach toward optimizing the statistical power using the lowest sample collection and assay cost, and a careful assessment of possible study designs should be routinely performed prior to conducting a GWAS.

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CONFLICT OF INTERESTS

The authors declare that they have no competing interests.

AUTHOR CONTRIBUTIONS


M.G. developed the relative efficiency and power calculation tools in Haplin, conceived, planned, and performed the analyses and drafted the manuscript. J.R., Ø.A.H., A.J., N.O.C., and R.T.L. helped develop the concepts and revised the manuscript. J.R. has also contributed to the recent developments of Haplin. H.K.G. developed the Haplin software, conceived, and planned the analyses and revised the manuscript. All the authors read and approved the final manuscript.

DATA ACCESSIBILITY

Haplin is implemented in the statistical software R and can be installed from the official R package archive, CRAN (<https://cran.r-project.org>).⁵⁸ Standard power calculations in Haplin can be carried out analytically using the asymptotic variance-covariance structure of the parameter estimator (recently implemented in the function `hapPowerAsymp`), or else by a straightforward simulation approach (see functions `hapRun` and `hapPower`). Relative efficiency estimates are readily computed using the function `hapRelEff`.¹⁵ For a thorough description of the Haplin functions and their arguments, please refer to the website at <https://people.uib.no/gjessing/genetics/software/haplin>. The CPO GWAS data are available at the dbGaP database (<https://www.ncbi.nlm.nih.gov/gap>) under accession ID phs000094.v1.p1.

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APPENDIX A

A1 Relative efficiency

For comparisons of statistical tests, the asymptotic relative efficiency is a useful measure.⁶² The asymptotic relative efficiency is defined as the ratio of the asymptotic variances of two different estimators of the same parameter. Under general conditions (see Theorem 14.19 of Reference 63), this ratio corresponds to the ratio of sample sizes needed to achieve the same precision from the two different estimators, or the ratio of sample sizes needed to achieve the same significance level and power for two hypothesis tests about the parameter. In our setting, we compare the variances of the estimators of the same parameter computed from two different study designs, weighted by the number of genotyped individuals within each design. The weights allow us to compare the relative efficiency of the two different designs, subject to the constraint that each design contains the same number of genotyped samples. The relative efficiency thus refers to a ratio of the number of genotyped *individuals*, not a ratio of the number of *families*. Let n denote the number of family structures with a case child. As n varies, we assume the composition of family structures remains the same, relatively speaking. That is, we assume, for instance, that the ratio of case-parent triads to control-mother dyads remains the same, likewise for the ratio of case-mother dyads to complete control-parent triads, and so on.

A1.1 The asymptotic SE of the log-scale parameter estimator

In Haplin, we use the Wald test to conduct post-hoc inference on the log-transformed relative risk parameters, based on asymptotic normality (see Chapter 1.3 of Reference 64). The main univariate outcome measure is the log relative risk of the relevant genetic effect, that is, $\beta = \log(\text{RR})$. For PoO effects, the parameter of interest is the ratio of two relative risks, which means looking at the difference between the corresponding β values, so the theory is the same. Based on the standard maximum likelihood theory, Haplin computes the SE $\sigma_n(\hat{\beta})$ of $\hat{\beta}$ from the observed Fisher information, using all available data, that is, with n cases. If the composition of family structures is kept fixed as n increases, we have that $\sqrt{n}\sigma_n(\hat{\beta}) \approx \omega(\beta)$, where $\omega(\beta)$ is the asymptotic SE of $\hat{\beta}$ computed from the Fisher information in the maximum likelihood model.¹⁴ The value of $\omega(\beta)$ can thus be seen to represent a sample with only one case ($n = 1$). For instance, in a setting with 200 case-parent triads and 100 control-parent triads, $\omega(\beta)$ would, theoretically, correspond to a family structure with one case triad and half a control triad. The derivation of the asymptotic multivariate variance-covariance matrix is provided in a previous article.¹⁴

A1.2 Asymptotic relative efficiency

The asymptotic SE is characteristic of the design used in the estimation. When comparing two designs 0 and 1, with design 0 as reference, the asymptotic relative efficiency of design 1 over design 0, that is, using design 0 as reference, is

$$\left\{ \omega^{(0)}(\beta) / \omega^{(1)}(\beta) \right\}^2 \cdot \frac{m_0}{m_1}, \quad (\text{A1})$$

where m_0 and m_1 are the number of individuals to be genotyped in designs 0 and 1, respectively. For instance, the asymptotic relative efficiency of the case-control design over the case-parent triad design uses $m_0 = 3$ and $m_1 = 2$ (the case-parent triad design is used as reference). Note that a ratio larger than one favors design 1.

Comparing the asymptotic variances of estimators of the same parameter from different designs provides an intuitive understanding of relative efficiency. Alternatively, one can consider relative efficiency in terms of hypothesis testing. Consider the problem of testing the null hypothesis $H_0 : \beta = 0$ versus the alternative $H_1 : \beta \neq 0$ for a fixed nominal level. Let $\gamma_n(\beta)$ be the power of the Wald test based on n cases (ie, n family structures with one case child in each). Clearly, for a fixed alternative $\beta \neq 0$, $\lim_{n \rightarrow \infty} \gamma_n(\beta) = 1$. That is, with enough data, a relative risk RR different from one will eventually

be detected by increasing the sample size sufficiently. To make an informative asymptotic comparison of two tests, that is, of tests for the same null hypothesis but based on two different designs, it is better to compare the efficiency of the tests when testing steadily decreasing effect sizes as the sample size increases. Here, we let the alternative to be tested for be $\beta_n = h/\sqrt{n}$, where h is a fixed constant. Under general conditions,

$$\lim_{n \rightarrow \infty} \gamma_n(\beta_n) = \gamma(h),$$

where $\gamma(h)$ is the so-called local limiting power function (see Theorem 14.7 of Reference 63). In our setting, the limiting power function γ of the Wald test with level α can be written

$$\gamma(h) = 1 - F_{\lambda(h)}(\chi_\alpha^2),$$

where χ_α^2 is the upper- α quantile of the chi-squared distribution with one degree of freedom and F_λ is the cumulative distribution function of a one degree of freedom non-central chi-squared distribution, with $\lambda = \lambda(h)$ as the noncentrality parameter. The noncentrality parameter can be expressed as $\lambda(h) = (h/\omega(0))^2$, where $\omega(0)$ is the asymptotic SE of $\hat{\beta}$ under the null hypothesis. Hence, comparing two parameter estimators corresponding to different study designs is equivalent to comparing the locally attained power of the Wald test. That is, the asymptotic relative efficiency of two designs when testing the null hypothesis can be found from Equation (A1) by setting $\beta = 0$. Note that Equation (A1) is independent of α and h when $\beta = 0$ (see Theorem 14.19 of Reference 63). This type of asymptotic relative efficiency for hypothesis tests is referred to as the Pitman efficiency.¹⁰

APPENDIX B

B1 An explicit formula for the asymptotic relative efficiency of regular autosomal effects for a diallelic SNP under H_0

For regular autosomal analyses of a diallelic SNP under H_0 , a formula for the relative efficiency is easily derived by heuristic arguments. We quantify the statistical contribution of a genotyped individual by its “design factors” and count the effective number of cases and controls while assuming a multiplicative dose-response relationship. The case (affected individual) forms the basis of the family-based designs and is always assumed to be genotyped. We, therefore, define the effective number of cases as $n_1 = 1$. The total effective number of controls can be written as $n_0 = d_1 + kd_0$, where d_1 is the effective number of controls from a case family, d_0 is the effective number of controls from a control family, and $k : 1$ is the ratio of control families to case families.

A single case or control (without their genotyped parents) identifies only two case or control alleles, respectively. Hence, the design factors are $d_1 = 0$ and $d_0 = 1$. However, a single case-parent triad encompasses four alleles, two of which are inherited by the case child, two of which are not. The nontransmitted parental alleles form the so-called pseudocontrols.^{20,21} Effects are seen as a contrast between the alleles of the pseudocontrols and the cases, similar to the approach used with a regular case-control design. A case-parent triad thus represents one case and one control ($d_1 = 1$). Conversely, a complete control-parent triad adds a single control offspring. Moreover, a pseudocontrol can also be formed, effectively resulting in two controls ($d_0 = 2$). Because these two controls together carry the same alleles as their parents, there is no need to genotype the original control child when both control parents have been genotyped.⁷

The issue of determining the design factor gets more complex when case dyads or control dyads are genotyped. If the case and only one of his/her parents are available, there are two case alleles and one control allele. However, deciding which of the parent's two alleles should be the control allele is not always possible when the other parent is missing. This results in a loss of efficiency, which leads to a design factor $d_1 < 1/2$, depending on the minor allele frequency (MAF).⁶⁵ If only one of the control parents is available for genotyping, genotyping the control offspring and his/her parent produces three control alleles. However, similar to the case-dyad scenario, if both the control offspring and his/her parent are heterozygous, one cannot distinguish which allele has been transmitted from the genotyped parent. Again, this leads to a loss of efficiency and a design factor $d_0 < 3/2$. The results are summarized in Table B1.

B1.1 An explicit formula

The total (actual) number of genotyped individuals is equal to $G = l_1 + kl_0$, where l_1 and l_0 are the number of genotyped individuals within a case and control family, respectively, with the possible values 0, 1, 2, or 3. Under H_0 , the SE of the

TABLE B1 Design factors for regular autosomal effects under H_0 in the single-SNP situation

	Control Family		Case Family	
	d_0	l_0	d_1	l_1
MFC	2	3	1	3
MF	2	2	—	—
MC or FC	$(3 - \text{MAF} \cdot (1 - \text{MAF}))/2^a$	2	$(1 - \text{MAF} \cdot (1 - \text{MAF}))/2^a$	2
M or F	1	1	—	—
C	1	1	0	1

Note: d_0 is the effective number of controls from a control family; l_0 is the number of genotyped individuals within a control family; d_1 is the effective number of controls from a case family; l_1 is the number of genotyped individuals within a case family.

Abbreviations: MAF, minor allele frequency.

^aThe effective number of controls is derived by subtracting the subset of ambiguous dyads.

difference between cases and controls is expected to be proportional to $\sqrt{1/n_0 + 1/n_1}$. Because $n_1 = 1$, the effective sample size for design i can be written as

$$N_i \propto \frac{1}{\text{SE}_i^2} \propto \frac{n_0}{n_0 + 1}.$$

Relative to the number of genotyped individuals, the effective sample size for design i is

$$\frac{N_i}{G_i}(k) = \frac{d_1 + kd_0}{(d_1 + kd_0 + 1)(l_1 + kl_0)}.$$

In this article, the case-parent triad design (mfc) is used as the reference, and we have that $\frac{N_{\text{mfc}}}{G_{\text{mfc}}}(k) = \frac{1}{6}$. Under H_0 , the efficiency of design i relative to the mfc design is thus

$$\frac{N_i/G_i}{N_{\text{mfc}}/G_{\text{mfc}}}(k) = \frac{6(d_1 + kd_0)}{(d_1 + kd_0 + 1)(l_1 + kl_0)}. \quad (\text{B1})$$

When $k = 1$, we see that the relative efficiency is 3/2 for the case-control (c-c) design and 3/4 for the full hybrid (mfc-mfc) design, independent of the MAF. This corresponds to the results of Figure 3 in this article.