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Estimating and Interpreting Heritability for Plant Breeding: An Update

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I. THE MEANING OF HERITABILITY

Heritability was originally defined by Lush as the proportion of phenotypic variance among individuals in a population that is due to heritable genetic effects (Nyquist 1991, p. 248). This definition is now termed "heritability in the narrow sense" and is designated h^2 (Nyquist 1991, pp. 248 and 250). Variations on this idea are often also referred to as heritability of one kind or another, such as heritability of family means (h_f^2), the proportion of the phenotypic variance of family means that is due to family genetic effects, and "heritability in the broad sense" (H), the proportion of phenotypic variance that is due to all genetic effects (Nyquist 1991, pp. 239, 312–313; Falconer and Mackay 1996, pp. 123, 232). Whereas Lush's definition was based on his experience as an animal breeder, in which the basic unit of observation and selection is nearly always the individual animal, plant breeders deal with a great diversity of observational units and mating systems. This complicates both the procedures for estimating heritability and the meaning of heritability itself. As Nyquist (1991, p. 238) observed,

> The plant kingdom presents a great diversity of natural modes of reproduction, varying from reproduction without sexuality (asexual) to reproduction by sexual means, cross-fertilization (allogamous), or self-fertilization (autogamous). Mixtures of these main modes of reproduction also exist. With self-fertilization, inbred populations exist and many unique difficulties arise. . . . Considering the diverse array of plant populations which can arise, many different estimators have been labeled heritability, and sometimes it is not clear what the exact nature of the estimator is or what is being estimated.

Hanson (1963) urged plant breeders to unify their concept of heritability as "the fraction of the selection differential expected to be gained when selection is practiced on a defined reference unit." Therefore, throughout this review, various heritability estimators are evaluated in terms of response to selection. Heritability has meaning only in reference to defined selection units and response units, and these can vary among breeding schemes.

Nyquist (1991) critically reviewed the substantial literature on estimating heritability and predicting response to selection in plant populations, and he clarified many of the issues that affect heritability in plants. Little can be added to his review of the topic except to address some newer methods of heritability estimation that have developed and been used in the last ten years. These newer methods include mixed models analysis of unbalanced data, pedigree analysis, and use of DNA markers to estimate genetic components of variation. Mixed models analysis in general terms has been reviewed thoroughly by McLean et al. (1991), Searle et al. (1992), and Littell et al. (1996), but the use of mixed models analysis for plant breeding applications has not been reviewed. Use of pedigree information to estimate genetic variance components in plant breeding was reviewed by Xu (2003). Ritland (2000) reviewed the use of DNA markers for estimating heritability and other population genetic parameters. Marker-based methods will have the greatest impact on studies of natural populations with unknown pedigree relationships and perhaps on domesticated species whose breeding systems are not easily controlled. Recently, these methods have become practical in part because of advances in computing power that have made powerful but previously computationally unmanageable estimating procedures almost routine.

This chapter focuses on placing mixed models analysis procedures in the context of typical plant breeding experiments and provides examples of computing code that can be used to obtain heritability estimates and their standard errors with the commonly used SAS system (see

Appendices 1 to 4). To place the estimation procedures in context, the interpretation of heritability estimators obtained from different mating schemes and generations is discussed.

II. RESPONSE TO SELECTION

A. Applications of Heritability Estimates

The main purpose of estimating heritability and the genetic parameters that compose the heritability estimate is to compare the expected gains from selection based on alternative selection strategies. One can use heritability estimates to predict gain from selection, for example, based on single, unreplicated plot values, and compare this to gain from selection expected if materials are replicated within and across macroenvironments (Hoi et al. 1999). Heritability estimates are useful for comparing the gain from selection under different experimental designs, and this information—combined with information about the relative costs of additional replications within each macroenvironment, additional years of evaluations, and additional locations for evaluations—can be used to design optimal breeding strategies (Milligan et al. 1990). Where genotype-by-environment (GE) interactions cause significant rank changes among families evaluated in different environments, heritability estimates corresponding to response to selection based on means over all environments can be compared with heritability based on means within subsets of local environments to determine the optimal selection strategy (Atlin et al. 2000). Similarly, heritabilities based on different family structures derived from the same base population can be compared to determine which family structure is best for maximizing genetic gain over units of time (Burton and Carver 1993). Heritability may vary among populations, thus, heritability estimates from different populations can be useful for choosing appropriate base populations in which selection will be most effective (Goodman 1965). Because heritabilities vary among traits within a population, heritability estimates of different traits, in addition to genetic correlation estimates among the traits, can be used to identify indirect selection schemes that may be more effective than direct selection schemes (Diz and Schank 1995; Banziger and Lafitte 1997; Rebetzke et al. 2002).

B. Theoretical Basis of Response to Selection

An understanding of the response to selection is needed in order to apply Hanson's (1963) definition of heritability as the fraction of the selection differential expected to be gained when selection is practiced

on a defined selection unit. One way to conceive of the response to selection is as a response or change in the mean of progeny phenotypic values due to a change in the mean value of selection units brought about by selection. The selection differential referred to by Hanson (1963) is the difference between the mean of selected selection units and the overall mean of the initial population. We introduce the notation $S = \mu_s - \mu_0$, where S is the selection differential, μ_s is the mean of the selected selection units, and μ_0 is the overall initial population mean. From elementary statistics, the expected response in any variable, Y, due to a change in a related variable, X, is given as $\Delta Y = b(\Delta X)$, where b is the coefficient of regression of Y on X, ΔY is the change in Y, and ΔX is the change in X (Steel et al. 1997). This general formula can be applied to response to selection by considering X as the variable representing selection unit phenotypic values, and Y as the variable representing phenotypic values of random members of the response units. Thus, ΔX is the selection differential, $\mu_s - \mu_0$, and ΔY is R, the expected response to selection: $\mu_1 - \mu_0$, where μ_1 is the mean (or expected value) of the response unit phenotypes in the first cycle resulting from selection within the initial population. Summarizing, $R = Sb_{YX}$. Therefore, the expected proportion of the selection differential to be achieved as a gain from selection, or heritability, is $R/S = h^2 = b_{YX}$.

The generality of this concept of heritability is very useful for plant breeding, because it is applicable to all plant breeding situations, including selection within randomly-mating cross-pollinated populations, as well as selection among self-fertilized lines (with or without subsequent random-mating), selection among clones, and selection among testcross progenies in hybrid crops. The generality of this concept is also a weakness, because it can have many different genetical meanings, depending on the circumstances and type of selection to which it is applied. We agree with Hanson (1963) and Nyquist (1991, p. 313) that the only remedy for this situation and the possible confusion arising from it is that researchers clearly indicate the basis of their heritability estimates—what is the defined reference unit for selection, and to what method of selection does it refer? Furthermore, we suggest that the reference unit for measuring response also be indicated along with heritability estimates, as this also impacts the interpretation of heritability.

The application of the heritability formula to specific breeding situations is discussed in Section VIII. To specify an appropriate heritability function for any breeding situation, the coefficient of regression of the value of the response unit on the value of the selection unit is required. Mathematically, the regression coefficient is the covariance of the phenotypes of selection and response units divided by the selection unit

phenotypic variance (Nyquist 1991, p. 249). Specifying the response unit phenotypic value as Y, the phenotypic value of the selection unit related to the response unit through its female parent as X_f, and the phenotypic value of the selection unit related to the response unit through its male parent as X_m, we obtain:

$$b_{YXf} = \text{Cov}(X_f, Y)/\text{Var}(X_f),$$
$$b_{YXm} = \text{Cov}(X_m, Y)/\text{Var}(X_m).$$

If selection is practiced on selection units related to both female and male parents, the total expected response to selection is the sum of the two expected responses (Nyquist 1991, p. 272):

$$R = b_{YXf}S_f + b_{YXm}S_m,$$

where S_f and S_m are the selection differentials on female and male sides of the pedigree, respectively.

As shown by Nyquist (1991, p. 272), if selection units related to female and male parents have the same expected value and population variance (i.e., no sexual dimorphism), then $b_{YXf} = b_{YXm}$, $S_f = S_m = S$, and the total response to selection is:

$$R = [2\text{Cov}(X_f, Y)/\text{Var}(X_f)]S = [\text{Cov}(X, Y)/\text{Var}(X)]S$$

where $\text{Cov}(X, Y) = 2\text{Cov}(X_f, Y) = \text{Cov}(X_f, Y) + \text{Cov}(X_m, Y)$. Therefore, the heritability equation that refers to response to selection when selection is practiced on both male and female sides of a pedigree is:

$$h^2 = \text{Cov}(X, Y)/\text{Var}(X). \quad [1]$$

To apply this formula to a specific breeding method, the selection and response units must be specified because their relationship determines the numerator of the equation. For example, response units can be related to the selection units as clonal (asexual) offspring, first-generation progeny of random-matings of the selection units, progeny resulting from self-fertilization of the selection units, or they can be indirectly related to the selected units, such as offspring of relatives of the parents (called "recombination units" by Hallauer and Miranda [1988, p. 170]), rather than direct offspring of the selection units actually evaluated. Each of these situations results in unique covariances between selection and response units. Nyquist (1991, pp. 272–277) presented the selection

pedigree diagrams and covariances between selection units and response units for many commonly used selection schemes.

Specification of the selection unit is also necessary because the denominator of the heritability equation is the variance among selection unit phenotypic values. The variance among the selection units depends on whether individuals or families are evaluated. If families are evaluated, the experimental design used to estimate family means, such as the number of replications and environments in which selection units are evaluated, will impact the variance of selection units, which are family mean phenotypic values in this case.

C. Reference Populations, Assumptions, and Model Definitions

Heritability estimates must refer to a defined population of genotypes (Comstock and Moll 1963; Dudley and Moll 1969). Reference populations are generally assumed to be random-mating populations in Hardy-Weinberg and gametic phase equilibria, although for self-pollinating crops, sometimes the reference population is taken to be completely inbred genotypes derived from a Hardy-Weinberg and gametic phase equilibria reference population by inbreeding without selection. Diploid inheritance is assumed throughout this chapter. To estimate the heritability of the reference population, individuals or families should be sampled at random for measurement. Also, heritability estimates must refer to a specified population of environments (Comstock and Moll 1963; Dudley and Moll 1969; Nyquist 1991, pp. 239–243). Defining the reference population of environments is often more difficult than defining the reference population of genotypes, and reference populations of environments are rarely explicitly defined by researchers. Generally, however, a reference population of environments is defined geographically. For example, public plant breeders often are assigned to develop improved cultivars for a specific state or province of a country, in which case the reference set of populations that is of interest to such a breeder is their state. In contrast, international agricultural research centers are often explicitly concerned with developing germplasm that is broadly adapted to a loosely-defined ecological zone throughout the world. Their reference set of environments may include, for example, all subtropical zones throughout the world. Having defined the target set of environments, the researcher should attempt to sample test environments at random from this population. This is also difficult, because evaluations are often performed on experimental research stations, limiting the plant

breeder's ability to sample target production fields. Similarly, it is rarely feasible for researchers to evaluate material for more than a small number of years, thus limiting the sample of potential climatic variations under which the germplasm of interest can be evaluated. These problems are close to insurmountable, although recent research focused on better defining target production environments may help researchers to better sample the reference population of environments (Gauch and Zobel 1997). We can only emphasize that researchers attempt to sample a range of environments that represent the target production environments for the germplasm, and that at a minimum, this should include a sample of several locations and several years.

Defining and adequately sampling the reference population of genotypes and environments is important for estimating heritability because this provides the context to which the heritability estimate refers. The genotypic values of the individuals in the population may depend on the environment or the conditions under which the experiment was performed (Comstock and Moll, 1963). For example, a drought-tolerant genotype of wheat (*Triticum aestivum*) will most likely be more vigorous under drought conditions compared with a normal genotype, whereas under higher moisture conditions, the normal genotype may be superior. Thus, when the experiment is performed in only a single environment, the estimated genotypic values cannot necessarily be used to make inferences beyond the original environment. The scope of inference of any experiment is an important issue that is often overlooked, but should be as well-defined as possible to avoid any confusion regarding interpretation of the results. The genotypic values refer specifically to the conditions under which the experiment was performed, and it cannot be assumed that the values would be the same in another reference set of environments. Therefore, genetic variance depends on the reference environments as well as the genotypes evaluated. Furthermore, the genetic variance component estimated in the experiment refers only to the population which was sampled for the experiment.

A clear definition of the population being sampled is also important for the estimate of genetic variance to have any meaning. One population of any species will not necessarily have the same amount of genetic variation as another population even from the same species, which can be due to many factors, such as selection, mating behavior, random drift, migration, and mutation. Thus, for example, there is no reason to expect that the genotypic variance estimated for a particular trait for one population of alfalfa (*Medicago sativa*) will have any relevance to another population of alfalfa. Furthermore, the variation observed for any one trait in any population may not hold for another trait in the same

population. For example, a maize (*Zea mays*) population that has been under selection for resistance to a particular disease may eventually become fixed for the resistant phenotype, but it may still have genetic variation for other traits, such as yield or flowering time.

Heritability estimates must be made from data collected in multiple locations and during multiple years representing the target set of environments or else the estimates will be biased unless genotype-by-environment interaction is negligible, which is rarely true for quantitative characters of agronomic importance (Nyquist 1991, pp. 239 and 312). This bias arises because the genotype-by-environment interaction variance is confounded with the genotypic variance component if the genotypic variance component is estimated from a single environment or from a sample of multiple locations or from a sample of multiple years only (Nyquist 1991, pp. 288–289). Another bias can arise if researchers ignore the cross-classified nature of years and locations during the statistical analysis of their experiment. For example, if families are evaluated at three locations across three years, the environments can be classified by year and location, leading to variance components estimates for years, locations, year-by-location interaction, families, and family-by-year, family-by-location, and family-by-year-by-location interactions. Or the analysis can proceed by classifying each year and location combination as one of nine environments, leading to variance component estimates of environments, families, and family-by-environment interaction. The latter choice leads to a simpler statistical model, but also creates bias in the resulting estimate of heritability, because the estimate of family-by-environment interaction variance is smaller than the sum of family-by-year, family-by-location, and family-by-year-by-location variances (Comstock and Moll 1963; Nyquist 1991, pp. 289–290). Throughout this chapter, the model that ignores the cross-classification of families and environments is used only to simplify the presentation of mathematical formulas. This should be avoided if possible in analysis of cross-classified data sets, and formulas for estimating heritability are provided with both approaches to handling environmental classification (Table 2.1, pp. 86-101) at the end of the chapter.

Having defined the reference populations of genotypes and environments, we can define the effects of the statistical model that will be used to estimate heritability. First, assume that the genotypes are sampled at random from the reference population, meaning that the genotypic effects (G_j's) are independent with expected value of zero and a common variance, σ^2_G. Assume also that the environments are sampled at random from the reference population of environments. Further, distinguish between the effects of macroenvironments (which generally refer to a

combination of a geographical location and unique weather pattern, that is, a single year and location combination) and the effects of microenvironments (which refer to environmental variations within macroenvironments). Therefore, we introduce a term for the effect of macroenvironments, E_i, and a term for the effect of microenvironments, ε'_{ijk}. Each is distributed around a mean of zero with variance σ^2_E for macroenvironments and $\sigma^2_{\varepsilon'}$ for microenvironments. We also introduce a term $R_{(i)k}$ for the mean effect of replications (complete blocks) within environments. This leads to a common form of the linear model for data observed on genotypes replicated in multiple blocks within multiple environments on a plot basis:

$$Y_{ijk} = \mu + E_i + R_{(i)k} + G_j + GE_{ij} + \varepsilon'_{ijk}. \qquad [2]$$

This type of model assumes that genotypes can be replicated; in Section V.A we demonstrate how to generalize the model to nonclonal material. The model also assumes that only one phenotypic value is recorded on each plot. If data are taken on individual plants within each plot, the error variance can be partitioned into variance due to random plot effects and within-plot variance. If not, then plot effects and plant-within-plot effects are confounded in the residual effect, which is denoted as ε'_{ijk} in Equation [2], to maintain consistency with Nyquist (1991, p. 258). See Nyquist (1991, pp. 252–259) for details on the definition of residual variances in this model and more complex statistical models. Other than the overall mean effect, μ, all effects in this model are random.

If selection is based on the mean phenotypic value of genotypes evaluated in multiple replications and macroenvironments (r replications within each of e macroenvironments), then the values of interest are mean values of genotypes:

$$\begin{aligned}\bar{X}_{.j.} &= \mu + \frac{\sum_{i=1}^{e} E_i}{e} + \frac{\sum_{i=1}^{e}\sum_{k=1}^{r} R_{(i)k}}{er} + G_j + \frac{\sum_{i=1}^{e} GE_{ij}}{e} + \frac{\sum_{i=1}^{e}\sum_{k=1}^{r} \varepsilon'_{ijk}}{er}. \\ &= \mu + \bar{E}_{.} + \bar{R}_{..} + G_j + \bar{GE}_{..} + \bar{\varepsilon}'_{.j.} \end{aligned} \qquad [3]$$

Similarly, if the genotypes of the next base population are evaluated in replicated, multiple environment trials, their mean phenotypic values ($\bar{Y}_{.j}$) are the response unit values. We assume that the set of environments in which selection units are evaluated and the set of environ-

ments in which response units are evaluated are independent samples of the common reference population of environments. In practical terms, this means that the evaluations of selection units are different from those in which the response units are evaluated.

To specify this in our statistical notation, we write E_i for environments used for selection unit evaluations and $E_{i'}$ for environments used for response unit evaluations, and similarly, $R_{i(k)}$ and $R_{i'(k')}$ for block effects sampled for selection and response unit evaluations, respectively. The number of environments sampled for selection unit evaluations is e_x and the number of environments sampled for response unit evaluations is e_y. The number of complete blocks within each evaluation environment is specified as r_x for selection units, and r_y for response units. Summarizing this notation, we have:

$$\begin{gathered} i = 1, \ldots, e_x, \text{ and } \mathrm{Var}(E_i) = \sigma^2_E, \\ i' = e_x + 1, \ldots, e_x + e_y, \text{ and } \mathrm{Var}(E_{i'}) = \sigma^2_E, \\ k = 1, \ldots, r_x, \text{ and } \mathrm{Var}(R_{(i)k}) = \sigma^2_R, \\ k' = r_x + 1, \ldots, r_x + r_y, \text{ and } \mathrm{Var}(R_{(i')k'}) = \sigma^2_R. \end{gathered}$$

Having defined the statistical model, we can expand the heritability equation to include all of the effects that contribute to selection and response unit phenotypes. Assuming that selection units are chosen based on their mean phenotypic values, and response units are evaluated in replicated multiple environment experiments, we are interested in the following regression coefficient:

$$b_{\overline{XY}} = \frac{\mathrm{Cov}(\overline{X}_{.j.}, \overline{Y}_{.j'.})}{\mathrm{Var}(\overline{X}_{.j.})} =$$

$$\frac{\mathrm{Cov}\left[\left(\mu + \frac{\sum_{i=1}^{e_x} E_i}{e_x} + \frac{\sum_{i=1}^{e_x}\sum_{k=1}^{r_x} R_{(i)k}}{e_x r_x} + G_j + \frac{\sum_{i=1}^{e_x} GE_{ij}}{e_x} + \frac{\sum_{i=1}^{e_x}\sum_{k=1}^{r_x} \varepsilon'_{ijk}}{e_x r_x}\right),\right.}{\left.\left(\mu + \frac{\sum_{i'=e_x+1}^{e_x+e_y} E_{i'}}{e_y} + \frac{\sum_{i'=e_x+1}^{e_x+e_y}\sum_{k'=r_x+1}^{r_x+r_y} R_{(i')k'}}{e_y r_y} + G_{j'} + \frac{\sum_{i'=e_x+1}^{e_x+e_y} GE_{i'j'}}{e_y} + \frac{\sum_{i'=e_x+1}^{e_x+e_y}\sum_{k'=r_x+1}^{r_x+r_y} \varepsilon'_{i'j'k'}}{e_y r_y}\right)\right]}$$

$$\mathrm{Var}\left(\mu + \frac{\sum_{i=1}^{e_x} E_i}{e_x} + \frac{\sum_{i=1}^{e_x}\sum_{k=1}^{r_x} R_{(i)k}}{e_x r_x} + G_j + \frac{\sum_{i=1}^{e_x} GE_{ij}}{e_x} + \frac{\sum_{i=1}^{e_x}\sum_{k=1}^{r_x} \varepsilon'_{ijk}}{e_x r_x}\right).$$

To evaluate the numerator of this expression, we use the formula for the covariance between two variables, X and Y: $\mathrm{Cov}(X, Y) = E[(X-\mu_X)(Y-\mu_Y)]$. Applying this to the numerator of the regression coefficient formula, we obtain:

$$\mathrm{Cov}(\overline{X}_{.j.}, \overline{Y}_{.j'.}) =$$

$$E[(\frac{\sum_{i=1}^{e_x} E_i}{e_x} + \frac{\sum_{i=1}^{e_x}\sum_{k=1}^{r_x} R_{(i)k}}{e_x r_x} + G_j + \frac{\sum_{i=1}^{e_x} GE_{ij}}{e_x} + \frac{\sum_{i=1}^{e_x}\sum_{k=1}^{r_x} \varepsilon'_{ijk}}{e_x r_x})(\frac{\sum_{i'=e_x+1}^{e_x+e_y} E_{i'}}{e_y} +$$

$$\frac{\sum_{i'=e_x+1}^{e_x+e_y}\sum_{k'=r_x+1}^{r_x+r_y} R_{(i')k'}}{e_y r_y} + G_{j'} + \frac{\sum_{i'=e_x+1}^{e_x+e_y} GE_{i'j'}}{e_y} + \frac{\sum_{i'=e_x+1}^{e_x+e_y}\sum_{k'=r_x+1}^{r_x+r_y} \varepsilon'_{i'j'k'}}{e_y r_y})].$$

Working out this expectation in detail is tedious, but we observe that it simplifies greatly because the different model effects are independent. For example, the expectations of cross-products between macroenvironment effects of selection units (E_i) and genetic effects of response units ($G_{j'}$) are all zero because the environment and genotype effects are independent. Applying this rule, we obtain:

$$\mathrm{Cov}(\overline{X}_{.j.}, \overline{Y}_{.j'.}) =$$

$$E[(\frac{\sum_{i=1}^{e_x} E_i}{e_x})(\frac{\sum_{i'=e_x+1}^{e_x+e_y} E_{i'}}{e_y})] + E[(\frac{\sum_{i=1}^{e_x}\sum_{k=1}^{r_x} R_{(i)k}}{e_x r_x})(\frac{\sum_{i'=e_x+1}^{e_x+e_y}\sum_{k'=r_x+1}^{r_x+r_y} R_{(i')k'}}{e_y r_y})] +$$

$$E[(G_j)(G_{j'})] + E[(\frac{\sum_{i=1}^{e_x} GE_{ij}}{e_x})(\frac{\sum_{i'=e_x+1}^{e_x+e_y} GE_{i'j'}}{e_y})] + E[(\frac{\sum_{i=1}^{e_x}\sum_{k=1}^{r_x} \varepsilon'_{ijk}}{e_x r_x})(\frac{\sum_{i'=e_x+1}^{e_x+e_y}\sum_{k'=r_x+1}^{r_x+r_y} \varepsilon'_{i'j'k'}}{e_y r_y})].$$

Next, because we assumed that the environments in which selection and response units were evaluated were independent, their expected cross-product is zero. The same holds true for block effects, genotype-by-environment interaction effects, and error effects. This leaves only the expectation of the cross-product of the selection and response unit genotypic effects, which is not zero, because there is a genetic relationship between them:

$$\mathrm{Cov}(\overline{X}_{.j.}, \overline{Y}_{.j'.}) = E[(G_j)(G_{j'})].$$

The cross-product between selection and response genotypic values depends on their genetic relationship, which requires knowledge of their pedigree relationship. Given the pedigree relationship, the value of this cross-product can be determined using the theory of the covariance of relatives (Section III; see also Nyquist 1991, pp. 268–277 and 296–299). It also remains to determine the phenotypic variance among selection units (the denominator of the response equation) in order to fully evaluate the expected value of the heritability estimator. This phenotypic variance is treated in Section IV.

III. COVARIANCES OF RELATIVES

A. Covariance of Noninbred Relatives

To demonstrate the covariance between genotypic values of relatives, we start by writing the genotypic value (G_{ij}) for an individual as the sum of its additive and dominant effects at the kth locus:

$$G_{ij}^k = \alpha_i^k + \alpha_j^k + \delta_{ij}^k,$$

where α_i^k is the additive statistical effect of allele i at locus k in the defined reference populations of genotypes and environments and δ_{ij}^k is the dominance deviation effect due to the allele pair i and j at locus k in the same reference populations (Falconer and Mackay 1996, pp. 112–113; Holland 2001, p. 37). The genotypic variance caused by locus k in a random-mating population is:

$$\sigma_{G(k)}^2 = \sum_{i=1}^{m} p_i^k(\alpha_i^k)^2 + \sum_{j=1}^{m} p_j^k(\alpha_j^k)^2 + \sum_{i=1}^{m}\sum_{j=1}^{m} p_i^k p_j^k(\delta_{ij}^k)^2,$$

$$= 2E[(\alpha_i^k)^2] + E[(\delta_{ij}^k)^2] = \sigma_{A(k)}^2 + \sigma_{D(k)}^2$$

where p_i^k is the frequency of the ith allele at the locus k, $\sigma_{A(k)}^2$ is defined as the additive genetic variance due to locus k, and $\sigma_{D(k)}^2$ is defined as the dominance genetic variance due to locus k. If the population is in gametic phase equilibrium and epistasis is ignored, these terms can be summed over l loci to obtain the total genetic variance as the sum of total additive and total dominance variances:

$$\sigma_G^2 = \sum_{k=1}^{l} (\sigma_{A(k)}^2 + \sigma_{D(k)}^2) = \sigma_A^2 + \sigma_D^2.$$

Epistasis can be included by expanding the model to include two-locus and higher-order interactions:

$$G_{iji'j'i''j''\ldots} = \sum_{k=1}^{l} (\alpha_i^k + \alpha_j^k + \delta_{ij}^k) + \sum_{k=1}^{l-1} \sum_{k'=k+1}^{l} (\alpha\alpha_{ii'}^{kk'} + \alpha\delta_{ii'j'}^{kk'} + \delta\alpha_{iji'}^{kk'} + \delta\delta_{iji'j'}^{kk'})$$
$$+ \sum_{k=1}^{l-2} \sum_{k'=k+1}^{l-1} \sum_{k''=k'+1}^{l} (\alpha\alpha\alpha_{ii'i''}^{kk'k''} + \alpha\alpha\delta_{ii'i''j''}^{kk'k''} + \ldots) + \ldots,$$

where $\alpha\alpha_{ii'}^{kk'}$ is the additive-by-additive interaction statistical effect of allele i at locus k with allele i' at locus k', $\alpha\delta_{ii'j'}^{kk'}$ is the additive-by-dominance interaction statistical effect of allele i at locus k with allele pair i' and j' at locus k', $\delta\delta_{iji'j'}^{kk'}$ is the dominance-by-dominance interaction statistical effect of allele pair i and j at locus k with allele pair i' and j' at locus k', $\alpha\alpha\alpha_{ii'i''}^{kk'k''}$ is the additive-by-additive-by-additive interaction statistical effect of allele i at locus k with allele i' at locus k' and allele i'' at locus k'', and $\alpha\alpha\delta_{ii'i''j''}^{kk'k''}$ is the additive-by-additive-by-dominance interaction statistical effect of allele i at locus k with allele i' at locus k' and allele pair i'' and j'' at locus k''. The two-locus interaction effects are defined explicitly by Lynch and Walsh (1998, pp. 85–86) and Holland (2001). The higher-order effects are defined analogously (Lynch and Walsh 1998, p. 85). Assuming there is no linkage between loci affecting the trait concerned and assuming the population is in gametic phase equilibrium, the genotypic variance including epistatic terms is:

$$\sigma_G^2 = \sigma_A^2 + \sigma_D^2 + \sigma_{AA}^2 + \sigma_{AD}^2 + \sigma_{DD}^2 + \sigma_{AAA}^2 + \ldots,$$

where σ_{AA}^2 is the variance of additive-by-additive epistatic effects, σ_{AD}^2 is the variance of additive-by-dominance epistatic effects, σ_{DD}^2 is the variance of dominance-by-dominance epistatic effects, σ_{AAA}^2 is the variance of additive-by-additive-by-additive epistatic effects, and variance components of higher-order effects are included as desired.

Using this genetic model, we can determine the genetic components of variance that comprise the covariance of two individuals from the same family, or between a random individual X in the selection unit and random individual Y in the response unit in the context of selection response. The covariance between the genetic effects for two individuals depends on their pedigree relationship, as this influences the prob-

ability that two individuals inherited common genetic effects. The probability that a randomly chosen allele from a given locus in the first individual is identical by descent (IBD) to a randomly chosen allele from the same locus in the second individual is equal to the probability that the two individuals share a common additive statistical effect at all loci. This probability is represented by the symbol θ (or f, as used by Nyquist 1991 and Falconer and Mackay 1996), the coefficient of coancestry. Identical by descent (IBD) means that the two alleles descend from the same allele in a common progenitor (Falconer and Mackay 1996, p. 58; Lynch and Walsh 1998, p. 132). The probability of allele *pairs* being IBD equals the probability that two individuals share a common dominance statistical effect at all loci and is represented by the symbol u. It follows from this and from the assumptions of Hardy-Weinberg and gametic phase equilibria and no linkage, that the genetic covariance between individuals X and Y is:

$$\begin{aligned}\mathrm{Cov}(G_X, G_Y) &= 2\theta\sigma^2_A + u\sigma^2_D + (2\theta)^2\sigma^2_{AA} \\ &+2\theta u\sigma^2_{AD} + u^2\sigma^2_{DD} + (2\theta)^3\sigma^2_{AAA} + \ldots \end{aligned} \quad [4]$$

A formal derivation of this covariance can be found in Lynch and Walsh (1998, pp. 141–145). If individuals are chosen at random from the population, both θ and u for the pair equal zero and their expected genetic covariance is zero. If individuals are related in any way, however, the genetic covariance is greater than zero. Rules for evaluating θ and u and examples of their values for commonly encountered pairs of relatives are given by Falconer and Mackay (1996, pp. 85–88; pp. 152–155) and Lynch and Walsh (1998, pp. 133–145).

As an example, for an outbred parent and its outbred offspring resulting from a mating with an unrelated individual, $\theta = (1/4)$ and $u = 0$, leading to:

$$\mathrm{Cov}(G_P, G_O) = (\tfrac{1}{2})\sigma^2_A + (\tfrac{1}{4})\sigma^2_{AA} + (\tfrac{1}{8})\sigma^2_{AAA} + \ldots \ .$$

In Section V, we demonstrate that the covariance of collateral relatives from a systematic mating structure can be estimated with appropriate experimental designs, and this leads to estimates of the genetic components of variance in the regression of response unit phenotypes on selection unit phenotypes needed for the heritability estimator. To interpret the covariance of collateral relatives in terms of genetic components, we use the method described in this section. For example, for outbred half-sibs, $\theta = (1/8)$ and $u = 0$, leading to:

$$\text{Cov}(HS) = (\tfrac{1}{4})\sigma^2_A + (\tfrac{1}{16})\sigma^2_{AA} + (\tfrac{1}{64})\sigma^2_{AAA} + \ldots \ .$$

For outbred full-sibs, $\theta = (1/4)$ and $u = (1/4)$, leading to:

$$\text{Cov}(FS) = (\tfrac{1}{2})\sigma^2_A + (\tfrac{1}{4})\sigma^2_D + (\tfrac{1}{4})\sigma^2_{AA} + (\tfrac{1}{8})\sigma^2_{AD} + (\tfrac{1}{16})\sigma^2_{DD} + (\tfrac{1}{8})\sigma^2_{AAA} + \ldots \ .$$

B. Covariance of Inbred Relatives

When relatives are inbred, Equation [4] and the equations for population genetic components of variance given previously in Section III.A do not hold. With inbreeding, the allelic effects within a locus become correlated, leading to an additional set of three genetic parameters and three additional probability measures required to describe the genetic variance of the population and the covariance of relatives. The total genetic variance of a population inbred to a degree F, where F is the inbreeding coefficient, is (Nyquist 1991, p. 297):

$$\sigma^2_{G(F)} = (1+F)\sigma^2_A + (1-F)\sigma^2_D + 4FD_1 + FD^*_2$$
$$+F(1-F)H^* + (1+F)^2\sigma^2_{AA} + \ldots,$$

where D_1 is the covariance between additive effects and their respective homozygous dominance deviation effects, D^*_2 is the variance of homozygous dominance effects, and H^* is the sum of squared inbreeding depression effects (Cockerham 1983). The covariance of relatives X and Y if either are inbred is:

$$\text{Cov}(G_X, G_Y) = 2\theta_{XY}\sigma^2_A + 2\delta_{\bar{X}+\bar{Y}}\sigma^2_D + 2(\gamma_{\bar{X}Y} + \gamma_{X\bar{Y}})D_1 + \delta_{\bar{X}\bar{Y}}D^*_2$$
$$+(\Delta_{\bar{X}\cdot\bar{Y}} - F_X F_Y)H^* + (2\theta_{XY})^2\sigma^2_{AA} + \ldots, \qquad [5]$$

where θ_{XY} is the coefficient of coancestry between X and Y; $2\delta_{\bar{X}+\bar{Y}}$ is the probability that the allele pair at a locus in X is IBD to the pair at the same locus in Y, and that neither the two alleles at a locus within X nor within Y are IBD (this is equivalent to u if neither X nor Y is inbred); $\gamma_{\bar{X}Y}$ is the probability that the pair of alleles at a locus within X are IBD to each other and to one of the two alleles at the same locus in Y, and $\gamma_{X\bar{Y}}$ is defined similarly; $\delta_{\bar{X}\bar{Y}}$ is the probability that all four genes at a common locus in X and Y are IBD; and $\Delta_{\bar{X}\cdot\bar{Y}}$ is the probability that the two alleles within a locus in X are IBD and the two alleles of Y are IBD (Cockerham 1971, 1983). If the relatives descended by self-fertilization from a com-

mon ancestor, these probability parameters become functions of the inbreeding coefficients of the last common ancestor (F_t), the relative that descended from g generations of self-fertilization (F_g), and the other relative that descended from g' generations of self-fertilization ($F_{g'}$). In this case, the covariance of relatives is denoted $C_{tgg'}$:

$$C_{tgg'} = (1 + F_t)\sigma_A^2 + \frac{(1 - F_g)(1 - F_{g'})}{1 - F_t}\sigma_D^2 + (F_g + F_{g'} + 2F_t)D_1$$

$$+\left[F_t + \frac{(F_g - F_t)(F_{g'} - F_t)}{2(1 - F_t)}\right]D_2^* + \frac{F_t(1 - F_g)(1 - F_{g'})}{1 - F_t}H^* + (1 + F_t)^2\sigma_{AA}^2. \quad [5b]$$

Further simplifications occur when there are only two alleles per locus, in which case $H^* = \sigma_D^2$; if the two alleles are at equal frequency, then $D_1 = D_2^* = 0$ (Cockerham 1983).

Cockerham (1983), Nyquist (1991, p. 299), and Gibson (1996) presented tables of the coefficients of the five genetic parameters that describe the covariance of inbred relatives related by some common ancestor. (A typographical error exists in Nyquist (1991), Table 8, p. 299, for $t = 2$, $g = 2$, and $g' = 3$; change 23/8 to 25/8.) For example, the genetic covariance of an outbred parent (S_0 generation) and its progeny resulting from a single generation of self-fertilization (S_1 generation) is $C_{001} = \sigma_A^2 + (1/2)\sigma_D^2 + (1/2)D_1 + \sigma_{AA}^2$ (Nyquist 1991, p. 299). Note that the F_2 population is considered to be equivalent to the noninbred, random-mating population, even though one generation of selfing has already occurred (Nyquist 1991, p. 297), because it is in Hardy-Weinberg equilibrium.

IV. VARIANCE AMONG SELECTION UNITS

The variance among selection units is obtained from the general formula for the sample variance of a random variable. The population variance of a random variable X is: $\mathrm{Var}(X) = E[X - E(X)]^2$ (Lynch and Walsh 1998, pp. 22–23). The sample variance of X differs slightly because $E(X)$ is not known, but is estimated as $\bar{X}$ from the data; therefore the unbiased estimate of $\mathrm{Var}(X)$ obtained from a sample of size n is:

$$\hat{\sigma}_X^2 = \frac{n}{(n-1)}\sum_{i=1}^{n}\frac{(X_i - \bar{X}_\cdot)^2}{n} = \frac{1}{(n-1)}\sum_{i=1}^{n}(X_i - \bar{X}_\cdot)^2 \quad [6]$$

(Steel et al. 1997, p. 76). Applying this to the selection units, we are interested in estimating the variance of family mean phenotypic values. Recall from Equation [3] in Section II.C that the mean phenotypic value of a selection unit is:

$$\overline{X}_{.j.} = \mu + \frac{\sum_{i=1}^{e} E_i}{e} + \frac{\sum_{j=1}^{e}\sum_{k=1}^{r} R_{(i)k}}{er} + G_j + \frac{\sum_{i=1}^{e} GE_{ij}}{e} + \frac{\sum_{j=1}^{e}\sum_{k=1}^{r} \varepsilon'_{ijk}}{er}.$$

The value for the overall mean is obtained using the same model, introducing g as the total number of families or genotype groups evaluated:

$$\overline{X}_{...} = \mu + \frac{\sum_{i=1}^{e} E_i}{e} + \frac{\sum_{i=1}^{e}\sum_{k=1}^{r} R_{(i)k}}{er} + \frac{\sum_{j=1}^{g} G_j}{g} + \frac{\sum_{i=1}^{e}\sum_{j=1}^{g} GE_{ij}}{eg} + \frac{\sum_{i=1}^{e}\sum_{j=1}^{g}\sum_{k=1}^{r} \varepsilon'_{ijk}}{egr}.$$

Substituting these terms into Equation [6] gives:

$$\hat{\sigma}^2_{(\overline{X}_{.j.})} = \left(\frac{1}{g-1}\right)\sum_{j=1}^{g}[\overline{X}_{.j.} - \overline{X}_{...}]^2 = \left(\frac{1}{g-1}\right)\sum_{j=1}^{g}\Big[\Big(G_j + \frac{\sum_{i=1}^{e} GE_{ij}}{e} + \frac{\sum_{j=1}^{e}\sum_{k=1}^{r} \varepsilon'_{ijk}}{er}\Big) - \Big(\frac{\sum_{j=1}^{g} G_j}{g} + \frac{\sum_{i=1}^{e}\sum_{j=1}^{g} GE_{ij}}{eg} + \frac{\sum_{i=1}^{e}\sum_{j=1}^{g}\sum_{k=1}^{r} \varepsilon'_{ijk}}{egr}\Big)\Big]^2. \quad [7]$$

Again, since the different model effects are independent, we can ignore the cross-products between different model effects (such as between G_i and GE_{ij}). Therefore, the expectation of the formula in Equation [7] simplifies to:

$$E[\hat{\sigma}^2_{\overline{X}_{.j.}}] = E\left[\left(\frac{1}{g-1}\right)\left[\sum_{j=1}^{g}\left(G_j - \frac{\sum_{j=1}^{g} G_j}{g}\right)^2 + \sum_{j=1}^{g}\left(\frac{\sum_{i=1}^{e} GE_{ij}}{e} - \frac{\sum_{i=1}^{e}\sum_{j=1}^{g} GE_{ij}}{eg}\right)^2 + \sum_{j=1}^{g}\left(\frac{\sum_{j=1}^{e}\sum_{k=1}^{r} \varepsilon'_{ijk}}{er} - \frac{\sum_{i=1}^{e}\sum_{j=1}^{g}\sum_{k=1}^{r} \varepsilon'_{ijk}}{egr}\right)^2\right]\right]$$

$$= \left(\frac{1}{g-1}\right)\left[E\left[\sum_{j=1}^{g}\left(G_j - \frac{\sum_{j=1}^{g} G_j}{g}\right)^2\right] + E\left[\sum_{j=1}^{g}\left(\frac{\sum_{i=1}^{e} GE_{ij}}{e} - \frac{\sum_{i=1}^{e}\sum_{j=1}^{g} GE_{ij}}{eg}\right)^2\right] + E\left[\sum_{j=1}^{g}\left(\frac{\sum_{j=1}^{e}\sum_{k=1}^{r} \varepsilon'_{ijk}}{er} - \frac{\sum_{i=1}^{e}\sum_{j=1}^{g}\sum_{k=1}^{r} \varepsilon'_{ijk}}{egr}\right)^2\right]\right]. \quad [8]$$

The expectation of the summation involving G_j in Equation [8] simplifies as follows:

$$E[\sum_{j=1}^{g}(G_j - \frac{\sum_{j=1}^{g} G_j}{g})^2] = E[\sum_{j=1}^{g}(G_j - \frac{1}{g}[G_1 + G_2 + \ldots + G_j + \ldots + G_g])^2]$$

$$= E[\sum_{j=1}^{g}([G_j - \frac{G_j}{g}] - \frac{1}{g}[G_1 + G_2 + \ldots + G_{j-1} + G_{j+1} + \ldots + G_g])^2]$$

$$= E[\sum_{j=1}^{g}(\frac{g-1}{g})G_j)^2] + E[\sum_{j=1}^{g}\sum_{j'=1, j'\neq j}^{g}(\frac{-G_{j'}}{g})^2]$$

$$= (\frac{g-1}{g})^2 \sum_{j=1}^{g} E[G_j^2] + (\frac{1}{g^2})\sum_{j=1}^{g} E[\sum_{j'=1, j'\neq j}^{g} G_{j'}^2]$$

$$= (\frac{g-1}{g})^2 g\sigma_G^2 + (\frac{1}{g^2})g(g-1)\sigma_G^2$$

$$= (\frac{g^2 - 2g + 1 + g - 1}{g})\sigma_G^2$$

$$= (\frac{g^2 - g}{g})\sigma_G^2$$

$$= (g-1)\sigma_G^2.$$

Similarly, the expectation of the sum involving GE_{ij} in Equation [8] simplifies as follows:

$$E\{\sum_{j=1}^{g}[\frac{\sum_{i=1}^{e} GE_{ij}}{e} - \frac{\sum_{i=1}^{e}\sum_{j=1}^{g} GE_{ij}}{eg}]^2\}$$

$$= \frac{1}{e^2} E\{\sum_{j=1}^{g}[(GE_{1j} + GE_{2j} + \ldots + GE_{ej}) - (\frac{\sum_{j=1}^{g} GE_{1j}}{g} + \frac{\sum_{j=1}^{g} GE_{2j}}{g} + \ldots + \frac{\sum_{j=1}^{g} GE_{ej}}{g})]^2\}$$

$$= \frac{1}{e^2} E\{\sum_{j=1}^{g}[(GE_{1j} - \frac{\sum_{j=1}^{g} GE_{1j}}{g}) + (GE_{2j} - \frac{\sum_{j=1}^{g} GE_{2j}}{g}) + \ldots + (GE_{ej} - \frac{\sum_{j=1}^{g} GE_{ej}}{g})]^2\}$$

$$= \frac{1}{e^2} E\{\sum_{j=1}^{g}\sum_{i=1}^{e}[GE_{ij} - (\frac{\sum_{j=1}^{g} GE_{ij}}{g})]^2$$

$$= \frac{e}{e^2} E\{\sum_{j=1}^{g}[GE_{ij} - (\frac{\sum_{j=1}^{g} GE_{ij}}{g})]^2\}.$$

This reduces further in the same way that the expectation of the G_j term simplified; without showing the intermediate steps, this equation becomes:

$$\frac{e}{e^2} E[\sum_{j=1}^{g} GE_{ij} - (\frac{\sum_{j=1}^{g} GE_{ij}}{g})]^2 = \frac{1}{e}(g-1)E[GE_{ij}]^2 = (\frac{g-1}{e})\sigma^2_{GE}.$$

A similar derivation shows the simplification of the term involving error effects in Equation [8] to:

$$E[\sum_{j=1}^{g}(\frac{\sum_{i=1}^{e}\sum_{k=1}^{r}\varepsilon'_{ijk}}{er} - \frac{\sum_{i=1}^{e}\sum_{j=1}^{g}\sum_{k=1}^{r}\varepsilon'_{ijk}}{egr})] = (\frac{g-1}{er})\sigma^2_{\varepsilon'}.$$

Putting these simplified forms back into Equation [8], we obtain:

$$E(\hat{\sigma}^2_{\bar{X}_{j\cdot}}) = (\frac{1}{g-1})\sum_{j=1}^{g}\left[\begin{array}{l} E(G_j - \frac{\sum_{j=1}^{g} G_j}{g})^2 + E(\frac{\sum_{i=1}^{e} GE_{ij}}{e} - \frac{\sum_{i=1}^{e}\sum_{j=1}^{g} GE_{ij}}{eg})^2 \\ +E(\frac{\sum_{i=1}^{e}\sum_{k=1}^{r}\varepsilon'_{ijk}}{er} - \frac{\sum_{i=1}^{e}\sum_{j=1}^{g}\sum_{k=1}^{r}\varepsilon'_{ijk}}{egr})^2 \end{array}\right]$$

$$= (\frac{1}{g-1})\left[(g-1)\sigma^2_G + \frac{(g-1)}{e}\sigma^2_{GE} + \frac{(g-1)}{er}\sigma^2_{\varepsilon'}\right]$$

$$= \sigma^2_G + \frac{\sigma^2_{GE}}{e} + \frac{\sigma^2_{\varepsilon'}}{er}. \qquad [9]$$

This variance can be estimated easily in practice by dividing the mean square for genotypes from the analysis of variance by the total number of observations per genotype, which is *er*, because the expectation of the genotypic mean square from this type of experiment is:

$$E(MS_{Genotype}) = er\sigma^2_G + r\sigma^2_{GE} + \sigma^2_{\varepsilon'}$$

(Nyquist 1991, pp. 256–257; Steel et al. 1997, pp. 379–384).

We can use Equation [9] to obtain the phenotypic variance among individual, unreplicated phenotypic values of selection units, by setting $e = 1$ and $r = 1$. The resulting phenotypic variance is: $\sigma^2_P = \sigma^2_G + \sigma^2_{GE} + \sigma^2_{\varepsilon'}$.

The reason that neither the macroenvironmental nor the block variance contributes to this variance is because we defined it to refer to the expected variation among plots within a single replication of a single environment (Nyquist 1991, pp. 245 and 252). If one were to conduct a selection experiment in which different genotypes were grown in different environments or different blocks and then to select on unadjusted phenotypic values, one would have to add the macroenvironmental and block variance components to the phenotypic variance. Since there is no reason for such an inefficient selection procedure to be conducted, we will not consider that case. Instead, this phenotypic variance can be used to obtain a heritability estimator relevant to the response to selection among plants evaluated with a single replication within the target set of environments. Obviously, if one has data from multiple environments and replications, they should be used for selection purposes. Nevertheless, the heritability on a single-plot basis is often of interest because it suggests the magnitude of selection response that can be expected if future cycles of selection were to be conducted with a single replication. If such a heritability estimate is sufficiently high, the breeder may decide that single-replication evaluations are sufficient, and that evaluation resources can be spent elsewhere.

From Equation [9], it can be seen that increasing the number of replications within each environment will reduce the error portion of the phenotypic variance of genotypic means, but it will not affect the contribution due to GE variance. To reduce the contribution of GE variance to the phenotypic variance of selection units, the parents should be evaluated in more macroenvironments.

V. ESTIMATING HERITABILITY AS A FUNCTION OF VARIANCE COMPONENTS

A. Estimating Genetic Components of Variance from Replicated Family Evaluations

To estimate genetic components of variance in species that are not easily clonally propagated, families or lines containing multiple individuals related in some systematic fashion can be developed, and these families can be evaluated in replicated trials. This permits partitioning of the phenotypic variance into a component due to families (which is due to common genetic effects of the members of the same family) and components due to family-by-environment interaction and residual effects. We use the same model as for clonal genotypes in Equation [2], Section II.C, but instead of genotypic and genotype-by-environment

effects (G_j's and GE_{ij}'s) there are family and family-by-environment effects (F_j's and FE_{ij}'s):

$$Y_{ijk} = \mu + E_i + R_{(i)k} + F_j + FE_{ij} + \varepsilon'_{ijk}.$$

It is important to note that the residual term of this model, ε'_{ijk}, differs from that used in the model for clonally propagated species (Equation [2]). With clonally propagated species, all of the variation among plots of the same genotype grown in the same replication or incomplete block are due to microenvironmental effects. With heterogeneous families, however, differences among plots of the same family in the same replication or incomplete block may be due to three causes: (1) microenvironmental effects, (2) effects of different samples of genotypes of the same family that occur in different plots, and (3) effects due to interaction between genotypes within a family and the macroenvironment (Nyquist 1991, p. 254). The second of these terms is the within-family genetic variance. After accounting for the other model effects, the value of each plot is the mean or total of within-family genotypic and genotypic-by-environmental effects plus the microenvironmental effect of the plot. Therefore, the residual variance component, $\sigma^2_{\varepsilon'}$, equals $\sigma^2_\varepsilon + \frac{\sigma^2_W}{n}$, where σ^2_ε is the variance due to plot effects; σ^2_W is the within-plot component of variance that includes within-family genetic variation, within-plot microenvironmental effect variation, and variation due to interactions between the macroenvironment and genotypes within the family; and n is the number of plants per plot. If there are many plants per plot, the within-family genetic component of variance contributes little to the residual variance of this model. See Nyquist (1991, pp. 254–256) for details on models including individual plant observations.

The family variance component can be estimated and obviously represents an estimate of some portion of the genetic variance. To determine what genetic components of variance comprise the family variance, one must compute the expected genetic covariance of individuals within the family because the covariance of random individuals within the same family grown in independent environments equals the family variance component. The covariance between two random members of the same family grown in independent environments is the covariance between Y_{ijk} and $Y_{i'j'k'}$, where $i \neq i'$, $j = j'$, and $k \neq k'$:

$$\begin{aligned}\mathrm{Cov}(Y_{ijk}, Y_{i'j'k'}) &= \mathrm{Cov}(\mu + E_i + R_{(i)k} + F_j + FE_{ij} + \varepsilon'_{ijk},\ \mu + \\ &\quad E_{i'} + R_{(i')k'} + F_{j'} + FE_{i'j'} + \varepsilon'_{i'j'k'}) \\ &= \mathrm{Cov}(F_j, F_{j'}) = \mathrm{Cov}(F_j, F_j) \text{ (because } j = j') \\ &= \sigma^2_F.\end{aligned}$$

This covariance also equals the genetic covariance between random members of the same family, so we can equate the estimable family component of variance to the genetic covariance of relatives within a family, which is interpretable in terms of genetic components of variance (Section III; Cockerham 1963):

$$E[\mathrm{Cov}(Y_{ijkl}, Y_{i'jk'l'})] = E[\hat{\sigma}_F^2]$$
$$= 2\theta_{XY}\sigma_A^2 + u_{XY}\sigma_D^2 + (2\theta_{XY})^2\sigma_{AA}^2 + \ldots, \quad [10]$$

where Y_{ijkl} and $Y_{i'jk'l'}$ are noninbred members of the same family, and:

$$E[\mathrm{Cov}(Y_{ijkl}, Y_{i'jk'l'}) = E[\hat{\sigma}_F^2]$$
$$= 2\theta_{XY}\sigma_A^2 + 2\delta_{\bar{X}+\bar{Y}}\sigma_D^2 + 2(\gamma_{\bar{X}Y} + \gamma_{X\bar{Y}})D_1 + \delta_{\bar{X}\bar{Y}}D_2^* +$$
$$(\Delta_{\bar{X}\cdot\bar{Y}} - F_X F_Y)H^* + (2\theta_{XY})^2\sigma_{AA}^2 + \ldots, \quad [11]$$

(where Y_{ijkl} and $Y_{i'jk'l'}$ are inbred members of the same family). Cockerham (1963) described a method to translate variance components from more complex mating designs into genetic covariances between relatives.

Since both the genotypic variance component and phenotypic variance among family means can be estimated from replicated family evaluations, it is possible to estimate heritability of family means (h_f^2) as the ratio of the family variance component to the phenotypic variance among family means:

$$\hat{h}_f^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_{\bar{P}}^2}. \quad [12]$$

Interpretation of such estimates may be difficult, however, if the family variance component does not equal the covariance between selection and response units of interest.

As an example, full-sib families can be sampled from a random-mating population and evaluated in r replications within each of e environments. Based on this, the variance component due to families estimates $\sigma_F^2 = \mathrm{Cov}(\text{full sibs}) = (1/2)\sigma_A^2 + (1/4)\sigma_D^2 + (1/4)\sigma_{AA}^2$ (from this point forward, we will follow the convention of including the additive-by-additive epistatic variance component of genotypic variance, but assuming that higher-order epistatic terms are negligible). The family variance component estimate divided by the estimator of the variance among full-sib family means provides a heritability estimate of the form:

$$\hat{h}_f^2 \approx \frac{\hat{\sigma}_F^2}{\sigma_P^2} = \frac{\frac{1}{2}\hat{\sigma}_A^2 + \frac{1}{4}\hat{\sigma}_D^2 + \frac{1}{4}\hat{\sigma}_{AA}^2}{\frac{1}{2}\hat{\sigma}_A^2 + \frac{1}{4}\hat{\sigma}_D^2 + \frac{1}{4}\hat{\sigma}_{AA}^2 + \frac{\frac{1}{2}\hat{\sigma}_{AE}^2 + \frac{1}{4}\hat{\sigma}_{DE}^2 + \frac{1}{4}\hat{\sigma}_{AAE}^2}{e} + \frac{\hat{\sigma}_{\varepsilon'}^2}{er}}.$$

Is this estimator interpretable in terms of response to selection among full-sib families? To determine this, the expected response to selection among full-sib families is calculated as the selection differential times the regression coefficient of random-mated offspring on the full-sib family means representing the maternal and paternal sides of their pedigree. The response units (random-mated offspring) are related to the selection units through untested full-sibs that are used for recombination (X_{f1} and X_{m1} in Fig. 2.1). (Throughout this review, we assume that selection units and response units are related through untested relatives, as in Fig. 2.1 and 2.2. The same relationships hold when the response unit is a direct relative of one of the tested members of the selection unit if many individuals comprise the selection unit. If a small number of individuals comprise the selection unit, the expected covariance requires adjustment; see Nyquist (1991, pp. 291–293) for details.

The expectation of the numerator of the desired regression coefficient is the expected covariance between selection and response units (Fig. 2.1):

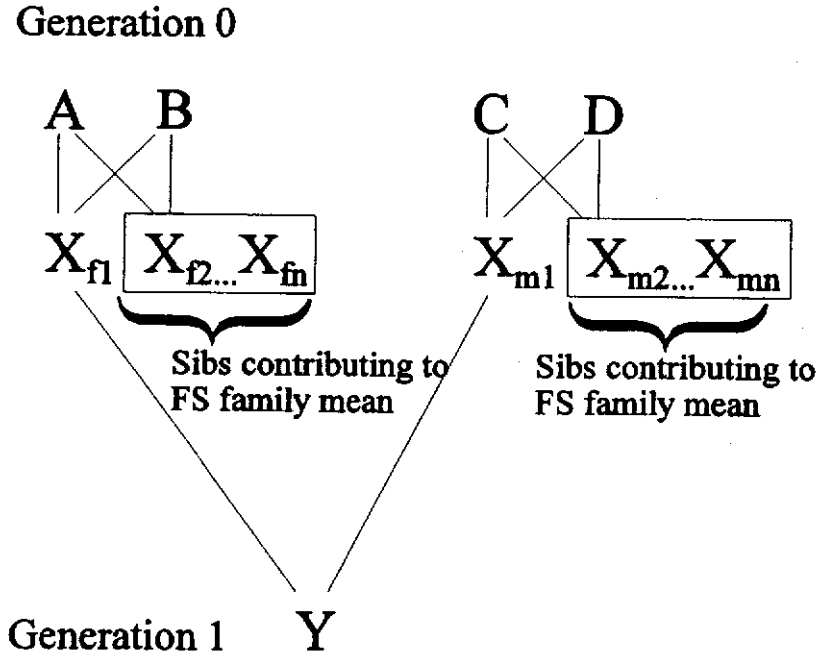

Fig. 2.1. Pedigree relationship between members of full-sib family selection units (*X*) and outbred progeny (response unit, *Y*) created from intermating remnant (untested) full sibs of selected families.

$$E[\text{Cov}(\text{mean of } X_{f2}...X_{fn},\ Y) + \text{Cov}(\text{mean of } X_{m2}...X_{mn},\ Y)]$$
$$= E[\text{Cov}(\text{mean of } X_{f2}...X_{fn},\ Y)] + E[\text{Cov}(\text{mean of } X_{m2}...X_{mn},\ Y)]$$
$$= E[\text{Cov}(X_{f2},\ Y)] + E[\text{Cov}(X_{m2},\ Y)]$$
$$= 2E[\text{Cov}(X_{f2},\ Y)] = 2[2\theta_{Xf2Y}\sigma^2_A + u_{Xf2Y}\sigma^2_D + (2\theta_{Xf2Y})^2\sigma^2_{AA}]$$
$$= 4\theta_{Xf2Y}\sigma^2_A + 2u_{Xf2Y}\sigma^2_D + 8(\theta_{Xf2Y})^2\sigma^2_{AA}.$$

Following Cockerham (1971), the coancestry of X_{f2} and Y is equal to the average coancestry of the parents of X_{f2} and Y:

$$\theta_{Xf2Y} = \theta_{(Xf2)(Xf1\ Xm1)} = (\tfrac{1}{2})(\theta_{Xf2\ Xf1} + \theta_{Xf2\ Xm1}) = (\tfrac{1}{2})[\theta_{(A\ B)(A\ B)} + \theta_{(A\ B)(C\ D)}]$$
$$= (\tfrac{1}{2})[(\tfrac{1}{4})(\theta_{AA} + \theta_{AB} + \theta_{BA} + \theta_{BB}) + (\tfrac{1}{4})(\theta_{AC} + \theta_{AD} + \theta_{BC} + \theta_{BD})]$$
$$= (\tfrac{1}{2})[(\tfrac{1}{4})(\tfrac{1}{2})(1 + F_A) + (\tfrac{1}{4})(\theta_{AB} + \theta_{BA}) + (\tfrac{1}{4})(\tfrac{1}{2})(1 + F_B) + (\tfrac{1}{4})(\theta_{AC}$$
$$+ \theta_{AD} + \theta_{BC} + \theta_{BD})]$$

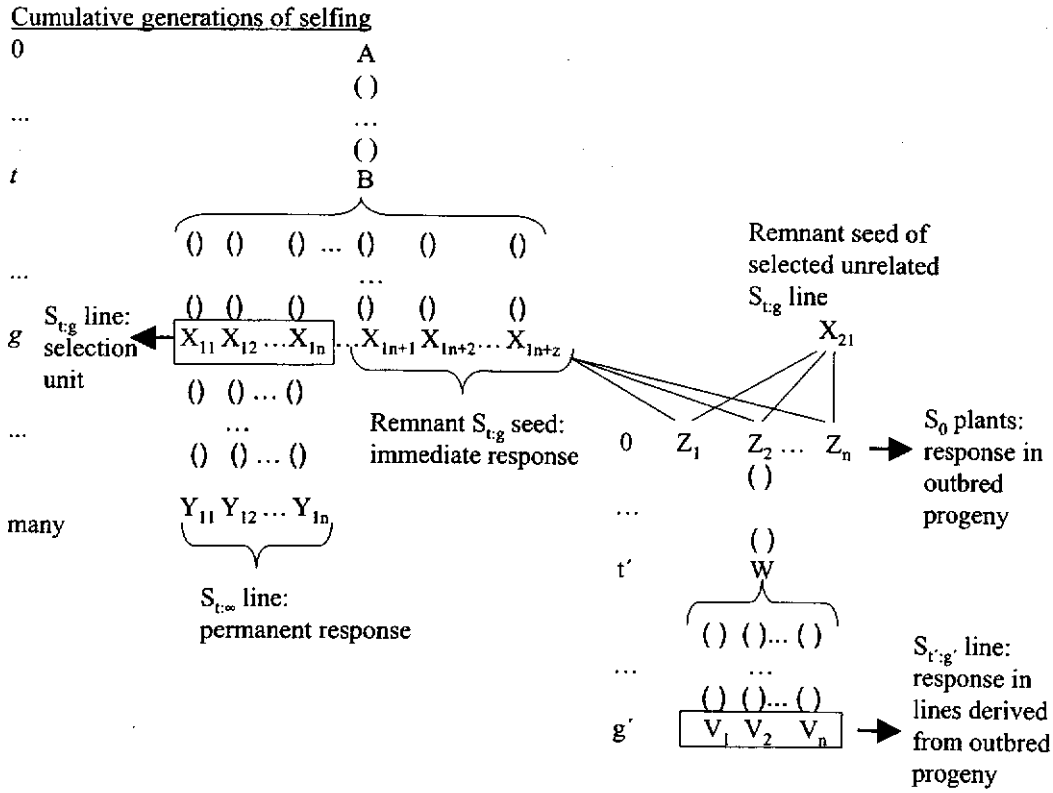

Fig. 2.2. Pedigree relationships among selection units composed of self-fertilized $S_{t:g}$ lines and alternative response units. Response units include "immediate response" units (remnant seed of the same generation line), "permanent response" units (completely inbred progeny of tested line), outbred S_0 generation progeny resulting from intermating unrelated selected lines, and inbred $S_{t':g'}$ lines resulting from self-fertilizing outbred progeny. Each response unit has a corresponding covariance with the selection unit and a corresponding heritability.

Parents A, B, C, and D are random members of the initial population, so by definition, their coancestries are zero: $\theta_{AB} = \theta_{BA} = \theta_{AC} = \theta_{AD} = \theta_{BC} = \theta_{BD} = 0$. A, B, C, and D are also noninbred, so $F_A = F_B = 0$. Substituting these values into the equation gives:

$$\theta_{Xf2Y} = (\tfrac{1}{2})[(\tfrac{1}{4})(\tfrac{1}{2}) + (\tfrac{1}{4})(\tfrac{1}{2})] = (\tfrac{1}{2})(\tfrac{1}{4}) = (\tfrac{1}{8})$$

The coefficient of dominance variance in the covariance of X_{f2} and Y is the probability of their having an IBD allele pair. If X_{f2} has alleles a and b at a locus and Y has alleles c and d at the same locus, then X_{f2} and Y have an IBD allele pair if $a \equiv c$ and $b \equiv d$ or if $a \equiv d$ and $b \equiv c$, that is, if they have the same genotype IBD at the locus. By inspection of Fig. 2.1 it is obvious that X_{f2} and Y cannot have an IBD allele pair, because one allele of Y is received from X_{m1}, which is unrelated to X_{f2}. Formally, $u_{Xf2Y} = \theta_{AC}\theta_{BD} + \theta_{AD}\theta_{BC} = 0$.

Substituting these values into the equation for the covariance between selection and response units gives (Nyquist 1991, p. 277):

$$E[\text{Cov}(\text{mean of } X_{f2}...X_{fn},\ Y) + \text{Cov}(\text{mean of } X_{m2}...X_{mn},\ Y)] = (\tfrac{1}{2})\sigma^2_A + (\tfrac{1}{8})\sigma^2_{AA} + \dots .$$

The denominator of the regression coefficient between selection and response units is the variance of family means with the following expectation from Equation [9]:

$$E[\hat{\sigma}^2_{\bar{P}}] = E[\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FE}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}] = \tfrac{1}{2}\sigma^2_A + \tfrac{1}{4}\sigma^2_D + \tfrac{1}{4}\sigma^2_{AA} + \frac{\frac{1}{2}\sigma^2_{AE} + \frac{1}{4}\sigma^2_{DE} + \frac{1}{4}\sigma^2_{AAE}}{e} + \frac{\sigma^2_{\varepsilon'}}{er}.$$

Therefore, the expectation of the regression of response units after the first generation of selection on selection units is:

$$E[b_{Y\bar{X}}] = \frac{\frac{1}{2}\sigma^2_A + \frac{1}{8}\sigma^2_{AA}}{\frac{1}{2}\sigma^2_A + \frac{1}{4}\sigma^2_D + \frac{1}{4}\sigma^2_{AA} + \frac{\frac{1}{2}\sigma^2_{AE} + \frac{1}{4}\sigma^2_{DE} + \frac{1}{4}\sigma^2_{AAE}}{e} + \frac{\sigma^2_{\varepsilon'}}{er}}.$$

This regression coefficient differs from the heritability estimate based on the family variance component given previously by the absence of the dominance variance component in the numerator. In order to properly estimate this regression coefficient, one needs to estimate the additive genetic variance separately from the dominance variance, using a mating design experiment (Hallauer and Miranda 1988, pp. 64–83).

The importance of defining the response unit also can be illustrated with this example. If the response unit is defined as an individual in the gametic phase equilibrium population derived by random mating the first generation population of response units for many generations without selection, the covariance between selection and response units in that gametic phase equilibrium population includes only the additive variance component, and does not include the additive-by-additive epistatic variance component (Nyquist 1991, pp. 250–251; Holland 2001). Therefore, different heritabilities are appropriate for the two different response units, first generation response units and response units after many generations of random-mating without further selection. Nyquist distinguished between these heritabilities with the notation h_1^2 for heritability when individuals in the first generation are the response units and h_∞^2 when individuals in the random-mated equilibrium population are the response units. The full-sib family variance component is biased by $(1/4)\sigma_D^2 + (1/8)\sigma_{AA}^2$ as an estimator of the numerator h_{f1}^2 and biased by $(1/4)\sigma_D^2 + (1/4)\sigma_{AA}^2$ as an estimator of the numerator of $h_{f\infty}^2$.

If selection is conceived of as simply regrowing remnant seed of only the selected full-sib families (without intermating) in an independent sample of environments from the same reference population of environments, the response to this form of "selection" *is* equal to the selection differential times the heritability estimator based on the family variance component. The relevant covariance in this case is the covariance between the mean of the tested family and an untested full-sib from the same family, which is expected to be $(1/2)\sigma_A^2 + (1/4)\sigma_D^2 + (1/4)\sigma_{AA}^2$. Perhaps this is a trivial form of selection, because it is a single-generation "dead end" that does not permit long-term selection response, but at least this provides an interpretation of the heritability estimator based on the family variance component.

With this example in mind, we present heritability estimators based on different experimental and mating designs in Section VIII and Table 2.1 at the end of the chapter, accompanied by an interpretation of each estimator in terms of selection response. We also indicate the bias present in the numerators of these estimators relative to h_1^2. The heritability estimators based on family variance components from outbred clonal families, half-sibs, and families created by testcrosses to an inbred line can all be interpreted directly in terms of response to selection among these family types and measured in corresponding family types developed after intermating the selected parents. In contrast, with full-sib families and self-pollinated lines, the interpretation of heritability estimators based on family variance components is difficult, and such heritability

estimates have less practical utility, being interpretable only in terms of very limited concepts of selection, unless the genetic variance is fully composed of additive genetic variance.

Finally, we caution that aside from the potential biases in estimates of heritability calculated as the ratio of family variance component to phenotypic variance due to nonadditive genetic components of variance, there is also a statistical source of bias in this type of estimator. The difficulty is that the expectation of this ratio does not necessarily equal the ratio of the expectations of family and phenotypic variance components:

$$E\left(\hat{h}_f^2\right) = E\left(\frac{\hat{\sigma}_F^2}{\hat{\sigma}_P^2}\right) \neq \frac{E[\hat{\sigma}_F^2]}{E[\hat{\sigma}_P^2]}.$$

Small portions of the variances and covariances of the estimated variance components (see Section V.C.1) contribute to the expectation of such a heritability estimator (see Lynch and Walsh 1998, pp. 808–809 for the expectation of a complex variable such as heritability). Researchers can compute the bias for specific cases of this type of heritability estimator, but we expect such biases to be small. Furthermore, as sample sizes increase, this source of bias will decrease in magnitude. Thus, this heritability estimator is asymptotically unbiased, in that:

$$E(\hat{h}_{f1}^2) = h_{f1}^2, \text{ as } e, r, f \to \infty.$$

Specifically, when heritability is estimated as a ratio of linear combinations of variance components using the methods described in Section V.B.1 (when data are balanced) and V.B.4 (in the general case), the estimator is consistent, meaning that:

$$\Pr\left[\left(\hat{h}_{f1}^2 - h_{f1}^2\right)^2 > \varepsilon\right] \to 0 \text{ as } e, r, f \to \infty, \text{ for any positive } \varepsilon,$$

or with a slight abuse of notation:

$$\hat{h}_{f1}^2 \to h_{f1}^2, \text{ as } e, r, f \to \infty.$$

B. Variance Component Estimation Procedures

1. Balanced Data. Traditionally, plant breeders and quantitative geneticists have estimated heritabilities based on variance components esti-

Table 2.2. ANOVA layout for balanced data from a replicated evaluation of f half-sib families in r complete blocks within each of e environments.

Source of Variation	Degrees of Freedom	Expected Mean Square
Environment	$e-1$	$\sigma^2_\epsilon + f\sigma^2_R + r\sigma^2_{FE} + rf\sigma^2_E$
Rep (Environment)	$(r-1)e$	$\sigma^2_\epsilon + f\sigma^2_R$
Family	$f-1$	$\sigma^2_\epsilon + r\sigma^2_{FE} + er\sigma^2_F$
Family × Environment	$(f-1)(e-1)$	$\sigma^2_\epsilon + r\sigma^2_{FE}$
Error	$(f-1)(r-1)e$	σ^2_ϵ

mated from ordinary least squares analysis of variance (ANOVA). Observed mean squares were equated to their expectations (linear functions of variance components) and the variance components were estimated algebraically as functions of mean squares. This estimation method is referred to as the *method of moments* (Milliken and Johnson 1992, pp. 233–239). Rules for deriving expected mean squares are given by Steel et al. (1997, pp. 379–384) and by Milliken and Johnson (1992, pp. 216–231), and specific ANOVA layouts including expected mean squares for numerous mating and experimental designs are presented by Hallauer and Miranda (1988) and Nyquist (1991). Equations for computing observed mean squares are given in most standard statistics texts, including Steel et al. (1997). To illustrate the method of moments estimation procedure, an ANOVA layout including expected mean squares for an experiment involving f half-sib families evaluated in r replications within each of e environments is presented in Table 2.2.

The variance component due to half-sib families is estimated with the following linear function of observed mean squares:

$$\hat{\sigma}^2_F = \frac{MS_{Family} - MS_{Family \times Environment}}{er}.$$

By Equation [10], the expectation of this family variance component is the covariance between half-sibs, which is equal to one-fourth of the additive genetic variance plus one-sixteenth of the additive-by-additive genetic variance:

$$E[\hat{\sigma}^2_F] = \tfrac{1}{4}\sigma^2_A + \tfrac{1}{16}\sigma^2_{AA}.$$

The variance among half-sib family means can also be estimated as a function of the family mean-square estimate, based on the following expectation:

$$E[\hat{\sigma}^2_P] = E\left[\frac{MS_{Family}}{er}\right] = \frac{\sigma^2_{\varepsilon'} + r\sigma^2_{FE} + er\sigma^2_F}{er} = \sigma^2_F + \frac{\sigma^2_{FE}}{e} + \frac{\sigma^2_{\varepsilon'}}{er}.$$

The heritability corresponding to selection among half-sib family means is commonly estimated by the ratio of these two estimators using Equation [12] (Nyquist 1991, p. 278):

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{MS_{Family} / er} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FE}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}} \approx \frac{\frac{1}{4}\hat{\sigma}^2_A + \frac{1}{16}\hat{\sigma}^2_{AA}}{\frac{1}{4}\hat{\sigma}^2_A + \frac{1}{16}\hat{\sigma}^2_{AA} + \frac{\frac{1}{4}\hat{\sigma}^2_{AE} + \frac{1}{16}\hat{\sigma}^2_{AAE}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}}.$$

In these equations, we have assumed that the data are balanced, that is, that e, r, and f are constant values and no data are missing. In this case, standard errors of the variance component estimates are estimable (Hallauer and Miranda 1988, p. 91), and methods for estimating confidence intervals for heritability estimates based on nested mating designs were developed by Graybill et al. (1956) (for a biased estimator of heritability) and Broemeling (1969) (for an unbiased estimate of narrow-sense heritability). Knapp et al. (1985), Knapp (1986), and Singh et al. (1993) derived exact confidence intervals for heritability on a family-mean basis for some typical plant breeding experiments. Knapp and Bridges (1987; also see Nyquist 1991, p. 311) developed approximate confidence intervals for family-mean based heritabilities estimated from more complicated plant breeding designs, such as perennial crop traits measured over time and factorial mating designs replicated over environments. All of these methods involved functions of mean squares and assumed that data were balanced.

2. Unbalanced Data. Generally, plant breeding experiments are designed as balanced experiments, but often unbalanced data sets arise for unplanned reasons: seeds of particular families may not be sufficient for complete replication, plots may be lost due to planting or harvesting errors, or plots may be discarded due to exceptional stresses. The effects of unbalanced data include changes in the coefficients of variance components in the expected mean squares, loss of independence between mean squares, and unknown distributional properties of variance component estimates. The changes in the coefficients of expected mean squares can be handled using methods given by Milliken and Johnson (1992, pp. 219–231), and correct coefficients of variance components in the expected mean squares can be computed with software packages

such as SAS (Rawlings 1988, pp. 466–467; SAS Institute 1999). Also, an approximate method to handle missing data in experiments designed to estimate heritability is to predict the missing plots using an analysis of covariance (Steel and Torrie 1980, pp. 426–428; Nyquist 1991, p. 262). Using these methods, variance components and heritability can be estimated using method of moments procedures. The variance-component estimates are unbiased, but no longer are they minimum variance estimators (Shaw 1987; Milliken and Johnson 1992, p. 233). Furthermore, their distributional properties are not known (Shaw 1987; Milliken and Johnson 1992, p. 233), such that estimates of the precision of variance component estimates and heritability estimates are not available. Another approach is to eliminate those families that are missing data, in order to obtain a balanced data set, but this decreases the efficiency of the estimate.

Better estimates (those with smaller variances and with known distributional properties) can be obtained using maximum likelihood methods, specifically, restricted maximum likelihood (REML). When data are completely balanced and there are no negative estimates of variance components under the ANOVA method, the ANOVA and REML variance component estimates are identical (Shaw 1987). When there are missing data, resulting in an unbalanced data structure, however, REML estimates of variance components are more desirable because they are consistent estimators, asymptotically normally distributed, and their asymptotic sampling dispersion matrix is known (Shaw 1987; Searle et al. 1992; Dieters et al. 1995). Modern computers and software have made REML-based estimates of variance components relatively easy to obtain. For example, GENSTAT and PROC MIXED of SAS provide robust and convenient methods for conducting REML analysis of many types of mixed model designs (Littell et al. 1996; Payne and Arnold 1998; SAS Institute Inc. 1999).

3. Maximum Likelihood and Restricted Maximum Likelihood Estimation. In this section, the procedures involved in maximum likelihood estimation are described with a minimum of mathematical detail. This process is also described in Milliken and Johnson (1992, pp. 239–242) and Lynch and Walsh (1998, pp. 853–867). The computational procedures are too complex to be performed by hand reasonably, so they can only be effectively implemented using computers. It may be of use to some readers to understand the basic procedure that occurs in the computing process.

Maximum likelihood estimation is a general method of estimating any sort of parameters from data. It can be used any time that one can

write the likelihood function of the parameters to be estimated. In comparison, the method of least squares in ANOVA involves solving for the parameter estimates that fit a given model with the minimum squared deviations. Using these solutions, the probability of the estimated parameter being greater than the parameter under the null hypothesis (generally that the parameters are zero) is computed, and these probabilities are reported as *p*-values. Maximum likelihood estimation works in a different manner: it begins by postulating the likelihood of different possible parameter values based on the model assumptions, given the observed data. It then chooses the set of parameter estimates with maximum value of the likelihood under those conditions.

The likelihood function of the data depends on the assumptions made about how the data are distributed. The likelihood function indicates how likely it is to observe the data given a model and its distribution. The likelihood function is based on the probability density function (PDF) for the model. For example, the PDF for a single observation of a normally distributed random variable y is:

$$P(y \mid \mu, \sigma^2) = \frac{e^{-(y-\mu)^2/(2\sigma^2)}}{\sqrt{2\pi\sigma^2}}$$

(Casella and Berger 1990, p. 103). The equation involves two parameters: the mean (μ) and the variance (σ^2). The mean indicates where the peak of the distribution is (where a randomly-chosen data point is most likely to be), and the variance indicates how "spread" the distribution is. If the variance is high, it is more likely to observe data points farther from the mean than if the variance were low. So, *given* a mean and a variance for a normal distribution, and given a value of an observation, the probability of observing the data point is obtained with the PDF. Given a different mean and variance, but the same data point, however, one might calculate a different probability for the data point. There are an *infinite* number of possible combinations of means and variances that could be used, but they will differ in terms of how likely it was to observe the data point from any of them. The maximum likelihood estimate of the mean and the variance is that pair of estimates that together gives the highest likelihood of observing the data. From among all of the infinite possible combinations of means and variances, we are interested only in the one with the highest likelihood, that is, the maximum likelihood estimate. Obviously, with only a single data point, one cannot estimate both a mean and a variance. So, we need a function that describes the

likelihood of observing a whole set of data points. This can be derived very simply by remembering that all of the observations from the distribution are drawn randomly and independently (if we do the experiment properly), and, therefore, the joint probability of observing n independent events (i.e., data points) simultaneously is simply the product of the probabilities of observing each event individually:

$$P(y_1, y_2, \ldots, y_n \mid \mu, \sigma^2) = \prod_{i=1}^{n} P(y_i \mid \mu, \sigma^2).$$

The likelihood of the parameters given the data is equivalent to the probability of the data given the parameters:

$$L(\mu, \sigma^2 \mid y_1, y_2, \ldots, y_n) = P(y_1, y_2, \ldots, y_n \mid \mu, \sigma^2).$$

The likelihood function is the same as the joint PDF. However, the joint PDF is regarded as a function of the random variables y_i, conditional on the parameters, whereas the likelihood function is viewed primarily as a function of the parameters, conditional on the observations y_i. Recall that there is an infinite set of means and variances that can be tried by calculating their likelihood. We will describe a combination of mathematical and "searching" methods that can be used to identify the best estimate out of all of the possibilities.

As an example, consider a simple two-factor factorial experiment in which a quantitative trait is observed. The linear additive model for the experiment is:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \alpha\beta_{ij} + \varepsilon_{ijk}.$$

If both A and B are considered random, the following standard model assumptions about the distribution of random model effects are made:

$$\begin{aligned} \alpha &\sim \mathrm{N}(0, \sigma^2_\alpha), \\ \beta &\sim \mathrm{N}(0, \sigma^2_\beta), \\ \alpha\beta &\sim \mathrm{N}(0, \sigma^2_{\alpha\beta}), \\ \varepsilon &\sim \mathrm{NID}(0, \sigma^2_\varepsilon). \end{aligned}$$

The likelihood function of the parameters given the data (with n total observations) is based on the known distributional properties of multivariate normally distributed variables. The PDF for this type of model,

which is an extension of the standard normal distribution, but permitting more than one random effect in the model, is (Lynch and Walsh 1998, pp. 194–195):

$$L(\mu, \sigma^2_\alpha, \sigma^2_\beta, \sigma^2_{\alpha\beta}, \sigma^2_\varepsilon \mid \mathbf{Y}) = \frac{e^{-\frac{1}{2}(\mathbf{Y}-\mu)'\mathbf{V}^{-1}(\mathbf{Y}-\mu)}}{\sqrt{2\pi^n \mid \mathbf{V} \mid}}.$$

The elements of this equation include the mean and the variance component parameters that we wish to estimate; the observed data, **Y**, which here is written as a vector of observations y_1, y_2, ...y_n; and the variance-covariance matrix of the observations, **V**. If we choose a set of values for the parameters of interest (the variance components), we can calculate the likelihood that those parameter values would have produced the data actually observed.

The distributional assumptions inherent in the model are used to determine the structure of variance-covariance matrix, **V**, for the entire data set, **Y**. **V** indicates the covariances between every pair of observations in the entire data set. We can determine the expected covariance between any two observations (Y_{ijk} and $Y_{i'j'k'}$) by expanding their values in terms of the model effects as follows:

$$\begin{gathered}E[\text{Cov}\,(Y_{ijk}, Y_{i'j'k'})] = E[\text{Cov}\,(\mu + \alpha_i + \beta_j + \alpha\beta_{ij} + \varepsilon_{ijk}, \mu + \alpha_{i'} + \\ \beta_{j'} + \alpha\beta_{i'j'} + \varepsilon_{i'j'k'})] = \\ E[\text{Cov}\,(\mu, \mu) + \text{Cov}\,(\mu, \alpha_{i'}) + \text{Cov}\,(\mu, \beta_{j'}) + \text{Cov}\,(\mu, \alpha\beta_{i'j'}) + \\ \text{Cov}\,(\mu, \varepsilon_{i'j'k'}) + \text{Cov}\,(\alpha_i, \mu) \\ + \text{Cov}\,(\alpha_i, \alpha_{i'}) + \text{Cov}\,(\alpha_i, \beta_{j'}) + \ldots + \text{Cov}\,(\beta_j, \beta_{j'}) + \ldots + \\ \text{Cov}\,(\alpha\beta_{ij}, \alpha\beta_{i'j'}) + \ldots + \text{Cov}\,(\varepsilon_{ijk}, \varepsilon_{i'j'k'})].\end{gathered}$$

The covariance between a constant and anything else is zero, so all of the covariances involving the constant μ are zero. All of the expected covariances between different factors, such as $\text{Cov}(\alpha_i, \beta_j)$ are also zero because there is no covariance between the level of one factor and the level of another if we have a properly randomized experiment. So the ellipses in this formula includes many of those covariances, which are all zero. The formula then reduces to:

$$\begin{gathered}E[\text{Cov}(Y_{ijk}, Y_{i'j'k'})] = E[\text{Cov}\,(\alpha_i, \alpha_{i'}) + \text{Cov}\,(\beta_j, \beta_{j'}) + \text{Cov}\,(\alpha\beta_{ij}, \alpha\beta_{i'j'}) + \\ \text{Cov}\,(\varepsilon_{ijk}, \varepsilon_{i'j'k'})].\end{gathered}$$

If $i \neq i'$, then the two observations were made on different levels of A, in which case they have expected covariance of zero, because the levels of

A were drawn at random from the population of A levels. However, if $i = i'$, then $E[\text{Cov}\ (\alpha_i, \alpha_i)] = \sigma^2_\alpha$. This reasoning can be applied to each of the covariances. If the two observations have the same level of B in common, then their covariance includes σ^2_β. If they have both the same levels of A and B in common, then their covariance includes σ^2_α, σ^2_β, and $\sigma^2_{\alpha\beta}$. Finally, the expected covariance of any observation with itself includes σ^2_ε along with σ^2_α, σ^2_β, and $\sigma^2_{\alpha\beta}$.

The **V** matrix includes all of the covariances between each observation and is used in the PDF of the model that includes all of the normally distributed parameters (σ^2_α, σ^2_β, $\sigma^2_{\alpha\beta}$, and σ^2_ε). Using as an example a two-factor factorial experiment in which each factor has two levels, and with two replications of each treatment combination in a completely randomized design, the following data set and models result:

$$
\begin{aligned}
Y_{111} &= \mu + \alpha_1 + \beta_1 + \alpha\beta_{11} + \varepsilon_{111},\\
Y_{112} &= \mu + \alpha_1 + \beta_1 + \alpha\beta_{11} + \varepsilon_{112},\\
Y_{121} &= \mu + \alpha_1 + \beta_2 + \alpha\beta_{12} + \varepsilon_{121},\\
Y_{122} &= \mu + \alpha_1 + \beta_2 + \alpha\beta_{12} + \varepsilon_{122},\\
Y_{211} &= \mu + \alpha_2 + \beta_1 + \alpha\beta_{21} + \varepsilon_{211},\\
Y_{212} &= \mu + \alpha_2 + \beta_1 + \alpha\beta_{21} + \varepsilon_{212},\\
Y_{221} &= \mu + \alpha_2 + \beta_2 + \alpha\beta_{22} + \varepsilon_{221},\\
Y_{222} &= \mu + \alpha_2 + \beta_2 + \alpha\beta_{22} + \varepsilon_{222}.
\end{aligned}
$$

The covariances among these observations are included in the **V** matrix, in which the first row and first column correspond to covariances involving observation Y_{111}, the second row and second column correspond to covariances involving Y_{112}, and so forth. Using the notation $\sigma^2_T = \sigma^2_\alpha + \sigma^2_\beta + \sigma^2_{\alpha\beta} + \sigma^2_\varepsilon$, and $\sigma^2_M = \sigma^2_\alpha + \sigma^2_\beta + \sigma^2_{\alpha\beta}$, the **V** matrix appears as follows:

$$
\mathbf{V} = \begin{bmatrix}
\sigma^2_T & \sigma^2_M & \sigma^2_\alpha & \sigma^2_\alpha & \sigma^2_\beta & \sigma^2_\beta & 0 & 0\\
\sigma^2_M & \sigma^2_T & \sigma^2_\alpha & \sigma^2_\alpha & \sigma^2_\beta & \sigma^2_\beta & 0 & 0\\
\sigma^2_\alpha & \sigma^2_\alpha & \sigma^2_T & \sigma^2_M & 0 & 0 & \sigma^2_\beta & \sigma^2_\beta\\
\sigma^2_\alpha & \sigma^2_\alpha & \sigma^2_M & \sigma^2_T & 0 & 0 & \sigma^2_\beta & \sigma^2_\beta\\
\sigma^2_\beta & \sigma^2_\beta & 0 & 0 & \sigma^2_T & \sigma^2_M & \sigma^2_\alpha & \sigma^2_\alpha\\
\sigma^2_\beta & \sigma^2_\beta & 0 & 0 & \sigma^2_M & \sigma^2_T & \sigma^2_\alpha & \sigma^2_\alpha\\
0 & 0 & \sigma^2_\beta & \sigma^2_\beta & \sigma^2_\alpha & \sigma^2_\alpha & \sigma^2_T & \sigma^2_M\\
0 & 0 & \sigma^2_\beta & \sigma^2_\beta & \sigma^2_\alpha & \sigma^2_\alpha & \sigma^2_M & \sigma^2_T
\end{bmatrix}. \qquad [13]
$$

To simplify the search for the parameter estimates, the derivative of the likelihood function can be taken with respect to a parameter. The

value of that parameter that makes the derivative equal to zero represents the maximum likelihood value of the parameter given the other parameter values. This method is based on the fact that the derivative of any function with respect to a variable equals zero at the point where the variable causes a peak (maximum value) in the function. The natural log of the PDF is usually easier to differentiate than the PDF, and the maximum point of the log of a function is also a maximum point of the function itself. Computation of the likelihoods becomes impractical for very large sample sizes, whereas computation of the log likelihood is much easier. Therefore, in practice, the log of the PDF is differentiated and set to zero to solve for the maximum likelihood value of the variable with respect to which the derivative was taken. The log of the likelihood function equation described above includes some constant, k_1, which will "disappear" upon differentiation:

$$\log[L(\mu, \sigma^2_\alpha, \sigma^2_\beta, \sigma^2_{\alpha\beta}, \sigma^2_\varepsilon \mid \mathbf{Y})] = k_1 - \tfrac{1}{2}\log \mid \mathbf{V} \mid - \tfrac{1}{2}(\mathbf{Y} - \mu)^T \mathbf{V}^{-1}(\mathbf{Y} - \mu).$$

The derivative of this log-likelihood function with respect to one of the parameters (e.g., σ^2_α) is computed, set equal to zero, and the equation is solved for the maximum likelihood value of that parameter. The same is done for the next parameter (e.g., σ^2_β), and so on. A complication is that the likelihood of any parameter depends on the value of the other parameters in the model, because the **V** matrix contains all of the different variance component estimates. Thus, an initial set of estimates for all of the parameters is required and the parameter estimation process must be iterated until a stable solution for all parameters is found. The iterative procedure usually, but not always, converges to the best possible solution.

This roughly describes the process that the MIXED procedure in SAS uses to obtain REML estimates of variance components. REML is a modification of the general maximum likelihood that produces parameter estimates with smaller bias (Lynch and Walsh 1998, pp. 789–791). REML proceeds by first estimating the fixed effects in the model, then by maximizing the likelihood function of n^* residual orthogonal contrasts, where n^* is the number of degrees of freedom remaining after fitting the fixed effects in the model (Lynch and Walsh 1998, pp. 789–791). The method of transforming the original data to the n^* residual orthogonal contrasts is shown in Lynch and Walsh (1998, p. 790). A simple and very instructive example of the difference between maximum likelihood and REML is given by Steel et al. (1997, p. 411).

The PROC MIXED output provides a list of the number of iterations it used to find the estimates. If it cannot converge to a solution, it provides a warning statement that the solutions did not converge. If this happens, the estimates are not unique and should be viewed with suspicion. One can try again by starting with new initial parameter estimates. Having estimated the variance components with REML, heritability estimates can be computed as functions of the variance component estimates.

4. The Likelihood Ratio Test. Mixed models analysis does not involve computation of mean squares of random factors; therefore, F-tests for the effects of random model factors are not available with mixed models analysis. Instead, the null hypothesis that a variance component for factor i is equal to zero (H_0: $\sigma_i^2 = 0$, H_A: $\sigma_i^2 > 0$) can be tested with a likelihood ratio test. To conduct the likelihood ratio test, one must analyze two models separately. One model, referred to as the full model, contains all the parameters of interest, including the variance component for factor i, σ_i^2. A second model, called the reduced model, contains all of the same parameters, except for the one whose significance is to be tested, σ_i^2. The likelihoods or the log likelihoods of the two models are compared using the likelihood ratio (LR) test:

$$\mathrm{LR} = 2\log\frac{L_F}{L_{\mathrm{Red}}} = -2\log\frac{L_{\mathrm{Red}}}{L_F} = -2[\log(L_{\mathrm{Red}}) - \log(L_F)],$$

where L_F is the likelihood of the maximum likelihood full model, L_{Red} is the likelihood of the maximum likelihood reduced model, and "log" refers to the natural log, as in Section V.B.3 (Steel et al. 1997, pp. 412–413; Lynch and Walsh 1998, pp. 857–858). The LR statistic is distributed as a χ^2 with degrees of freedom equal to the number of parameters dropped from the full model to make the reduced model. Typically, each variance component is tested one at a time, leading to LR statistics with one degree of freedom. For example, if there are 100 levels of the random factor i in the experiment and it has 99 degrees of freedom associated with it in the model, the LR test constructed by dropping that factor (and thus the parameter σ_i^2) from the model has only one degree of freedom. For the test of the null hypothesis that a variance component is zero, the p-value of the LR test should be divided by two (Self and Liang 1987). The example given in Steel et al. (1997, pp. 412–414) demonstrates that the p-value of the LR test is approximately

twice as large as the exact F-test of the same hypothesis. Likelihoods (and twice the log likelihoods) of models are part of the default output of SAS PROC MIXED, so the LR test is easily accomplished in practice. Cervantes-Martinez et al. (2001) used likelihood ratio tests to test the significance of family and family-by-environment variance components in a plant breeding experiment.

5. Mixed Models Analysis. Heritability estimation via REML will be beneficial when data are unbalanced (Section V.B.2). REML estimation of heritability has an additional advantage in that it can easily be performed in the context of mixed models analysis methods. We suggest that this will prove useful to plant breeders because it facilitates simultaneous estimation of variance components and heritability in different populations included in the same experiment. Mixed models analysis will also permit estimation of variance components and heritability from incomplete block designs (and perhaps spatial analysis methods), leading to improved heritability estimates through better control of experimental error. Also, with mixed models analysis, heritability estimation can be combined with best linear unbiased prediction (White and Hodge 1989). Combined estimation of genetic components of variance from multiple experiments containing different genetic entries with some known genetic relationships may be possible by augmenting the mixed models approach with pedigree analysis methods. Furthermore, mixed models are more appropriate than ANOVA for handling repeated observations on experimental units, as commonly occurs in perennial crops (Nyquist 1991, pp. 260–264; Littell et al. 1996, pp. 87–134). We confine ourselves in this chapter to briefly demonstrating simultaneous estimation of variance components from different populations grown in the same experiment, including incomplete blocks designs.

The general mixed model has the form:

$$\mathbf{y} = \mathbf{X}\beta + \mathbf{Z}\mathbf{u} + \mathbf{e}, \qquad [14]$$

where $\mathbf{y}$ is a vector of observed values, $\mathbf{X}$ is a design matrix for fixed effects, $\boldsymbol{\beta}$ is a vector of fixed effects, $\mathbf{Z}$ is a design matrix for random effects, $\mathbf{u}$ is a vector of random effects, and $\mathbf{e}$ is a vector of error effects associated with each observation (White and Hodge 1989, pp. 278–280; Lynch and Walsh 1998, p. 746). Using our previous example from Section V.B.3 of a two-factor factorial experiment with two replications of a completely randomized design and two levels of each random

factor, the model can be written in mixed model form using the following matrices:

$$\begin{aligned}
\mathbf{y}^T &= [Y_{111} \quad Y_{112} \quad Y_{121} \quad Y_{122} \quad Y_{211} \quad Y_{212} \quad Y_{221} \quad Y_{222}], \\
\mathbf{u}^T &= [\alpha_1 \quad \alpha_2 \quad \beta_1 \quad \beta_2 \quad \alpha\beta_{11} \quad \alpha\beta_{12} \quad \alpha\beta_{21} \quad \alpha\beta_{22}], \\
\mathbf{e}^T &= [\varepsilon_{111} \quad \varepsilon_{112} \quad \varepsilon_{121} \quad \varepsilon_{122} \quad \varepsilon_{211} \quad \varepsilon_{212} \quad \varepsilon_{221} \quad \varepsilon_{222}].
\end{aligned}$$

The columns of the **Z** matrix correspond to the rows of the **u** matrix (or the columns of the transposed **u** matrix shown). For each observation, the **Z** matrix indicates whether or not it is affected by each random model effect in **u**:

$$Z = \begin{bmatrix}
1 & 0 & 1 & 0 & 1 & 0 & 0 & 0 \\
1 & 0 & 1 & 0 & 1 & 0 & 0 & 0 \\
1 & 0 & 0 & 1 & 0 & 1 & 0 & 0 \\
1 & 0 & 0 & 1 & 0 & 1 & 0 & 0 \\
0 & 1 & 1 & 0 & 0 & 0 & 1 & 0 \\
0 & 1 & 1 & 0 & 0 & 0 & 1 & 0 \\
0 & 1 & 0 & 1 & 0 & 0 & 0 & 1 \\
0 & 1 & 0 & 1 & 0 & 0 & 0 & 1
\end{bmatrix}.$$

X is an 8×1 column vector of 1's, **β** is a 1×1 vector containing the only fixed effect in the experiment, μ. Putting these matrices together generates the data set of eight observations and eight combinations of model effects already given in Section V.B.3:

$$\begin{aligned}
Y_{111} &= \mu + \alpha_1 + \beta_1 + \alpha\beta_{11} + \varepsilon_{111}, \\
Y_{112} &= \mu + \alpha_1 + \beta_1 + \alpha\beta_{11} + \varepsilon_{112}, \\
Y_{121} &= \mu + \alpha_1 + \beta_2 + \alpha\beta_{12} + \varepsilon_{121}, \\
Y_{122} &= \mu + \alpha_1 + \beta_2 + \alpha\beta_{12} + \varepsilon_{122}, \\
Y_{211} &= \mu + \alpha_2 + \beta_1 + \alpha\beta_{21} + \varepsilon_{211}, \\
Y_{212} &= \mu + \alpha_2 + \beta_1 + \alpha\beta_{21} + \varepsilon_{212}, \\
Y_{221} &= \mu + \alpha_2 + \beta_2 + \alpha\beta_{22} + \varepsilon_{221}, \\
Y_{222} &= \mu + \alpha_2 + \beta_2 + \alpha\beta_{22} + \varepsilon_{222}.
\end{aligned}$$

The variance-covariance matrix of the observations, **V**, in this case is the same as in Equation [13], but in the mixed model it is arrived at by equation: $\mathbf{V} = \mathbf{ZGZ}^T + \mathbf{R}$, where **G** is the symmetric matrix that includes the

variances and covariances between random effects in the model (all off-diagonal elements are zero:

$$\mathbf{G} = \begin{bmatrix} \sigma^2_\alpha & & & & & & & \\ & \sigma^2_\alpha & & & & & & \\ & & \sigma^2_\beta & & & & & \\ & & & \sigma^2_\beta & & & & \\ & & & & \sigma^2_{\alpha\beta} & & & \\ & & & & & \sigma^2_{\alpha\beta} & & \\ & & & & & & \sigma^2_{\alpha\beta} & \\ & & & & & & & \sigma^2_{\alpha\beta} \end{bmatrix}.$$

R is a symmetric matrix that generally contains the error variance along the diagonal (Lynch and Walsh 1998, p. 748). In this example, $\mathbf{ZGZ}^T$ is equal to **V** given in Equation [13], except that $\mathbf{ZGZ}^T$ lacks the error variance terms, which are instead partitioned into the **R** matrix. One utility of the mixed model is that it permits estimation of both fixed and random effects (Littel et al. 1996, p. 499):

$$\hat{\boldsymbol{\beta}} = (\mathbf{X}^T\hat{\mathbf{V}}^{-1}\mathbf{X})^{-}\mathbf{X}^T\hat{\mathbf{V}}^{-1}\mathbf{y},$$
$$\hat{\mathbf{u}} = \mathbf{GZ}^T\hat{\mathbf{V}}^{-1}(\mathbf{y} - \mathbf{X}\hat{\mathbf{b}}).$$

The significance of fixed-effect factors is tested with F-tests, whereas the significance of random factors can be tested with likelihood ratio tests (Section V.B.4).

Another major benefit of mixed models analysis is its flexibility in modeling the variances and covariances of random and error effects. Pertinent to the estimation of heritability, the form of the **G** matrix can be modified to fit different variance-covariance structures among the random effects, permitting both the simultaneous fitting of incomplete block effects along with variance component estimation or the estimation of unique variance components for different subsets of genetic entries. We will not explicitly demonstrate the inclusion of incomplete block effects in the model, as it is straightforward to add another random effect to the model and estimate the block effects and variances separately from the genetic components of variance in the **G** matrix.

To separately estimate variance components for different sets of entries in an experiment, one needs to model the **G** matrix so that it permits dif-

ferent subsets of levels of the same factor (e.g., the family factor or the line factor in a heritability estimation study) to have unique variances. For example, we assumed in Section V.B.3 that all levels of factor A were drawn from a common reference population and have the same variance: $\alpha \sim N(0, \sigma^2_\alpha)$. With mixed models procedures, however, we can estimate separate variance components for factor A for different subsets of entries. Formally, we can allow each group g to have a unique variance for factor A: $\alpha_g \sim N(0, \sigma^2_{\alpha(g)})$, and the estimation of these separate variance components for factor A is accomplished by modifying **G**.

To demonstrate by expanding the previous example of the two-factor factorial experiment (Section V.B.3), we assume that four different levels of factor A were sampled and that potentially levels 1 and 2 have a common variance for factor A ($\sigma^2_{\alpha(1)}$), whereas levels 3 and 4 were drawn from a population of factor A with a unique variance, ($\sigma^2_{\alpha(2)}$). If each level of factor A occurs with each of the two levels of factor B and is replicated twice, this experiment would have 16 observations, as follows:

$$
\begin{aligned}
Y_{111} &= \mu + \alpha_1 + \beta_1 + \alpha\beta_{11} + \varepsilon_{111},\\
Y_{112} &= \mu + \alpha_1 + \beta_1 + \alpha\beta_{11} + \varepsilon_{112},\\
Y_{121} &= \mu + \alpha_1 + \beta_2 + \alpha\beta_{12} + \varepsilon_{121},\\
Y_{122} &= \mu + \alpha_1 + \beta_2 + \alpha\beta_{12} + \varepsilon_{122},\\
Y_{211} &= \mu + \alpha_2 + \beta_1 + \alpha\beta_{21} + \varepsilon_{211},\\
Y_{212} &= \mu + \alpha_2 + \beta_1 + \alpha\beta_{21} + \varepsilon_{212},\\
Y_{221} &= \mu + \alpha_2 + \beta_2 + \alpha\beta_{22} + \varepsilon_{221},\\
Y_{222} &= \mu + \alpha_2 + \beta_2 + \alpha\beta_{22} + \varepsilon_{222},\\
Y_{311} &= \mu + \alpha_3 + \beta_1 + \alpha\beta_{31} + \varepsilon_{311},\\
&\cdots\\
Y_{422} &= \mu + \alpha_4 + \beta_2 + \alpha\beta_{42} + \varepsilon_{422}.
\end{aligned}
$$

Compared to the model in Section V.B.3, the $\boldsymbol{\beta}$ vector is unchanged, but the other matrices change. The **X** matrix is augmented from an 8×1 to a 16×1 matrix by adding eight more rows of ones. The **u** vector is augmented from an 8×1 to a 14×1 vector by adding columns corresponding to two additional factor A main effects and four additional AB interaction effects. The **Z** matrix is augmented by adding six additional columns corresponding to the additional rows of the **u** vector, and eight additional rows corresponding to the eight additional observations. The **G** matrix structure is similar to the previous example, except that it would have dimensions of 14 rows and 14 columns, in which the first two diagonal elements are $\sigma^2_{\alpha(1)}$, the next two diagonal elements are $\sigma^2_{\alpha(2)}$,

the next two diagonal elements are σ^2_β, and the final eight diagonal elements are $\sigma^2_{\alpha\beta}$. From this structure, $\mathbf{ZGZ}^T$ models the covariance of two observations with the same level of A to include $\sigma^2_{\alpha(1)}$, if these observations have levels 1 or 2 of factor A, or to include $\sigma^2_{\alpha(2)}$, if the observations have levels 3 or 4 of factor A. The **e** vector increases by eight rows to account for the new observations, and the **R** matrix is augmented with eight more rows and columns, but has the same diagonal structure as before.

Similarly, one could also assume that the subset of entries drawn from the population of levels of A with variance $\sigma^2_{\alpha(1)}$ may also have a unique interaction variance with factor B: $\sigma^2_{\alpha\beta(1)}$, as opposed to $\sigma^2_{\alpha\beta(2)}$ for the second subset. These could also be fitted in the model by modifying the **G** matrix appropriately. Finally, one can also fit unique error variances for different subsets of entries by modifying the **R** matrix such that one group has error effects drawn from the population with variance $\sigma^2_{\varepsilon(1)}$, and the other has error effects drawn from the population with variance $\sigma^2_{\varepsilon(2)}$.

The ability to separately model variance components for different sets of treatments in the same experiment has considerable utility for variance component and heritability estimation experiments in plant breeding. For example, if one wants to compare unselected populations to populations derived from one or more generations of selection from it, it is reasonable to want to study the effect of selection on both population means and variances, as well as on heritability. Previously, plant breeders handled this by estimating population means with an ANOVA based on all of the data, and then by estimating variance components separately for subsets of families belonging to different cycles of selection. The latter procedure is inefficient if incomplete block designs were used: generally, if subsets of entries were analyzed for variance components, incomplete block effects were ignored in the analysis because, otherwise, severe data imbalance resulted. Even if incomplete blocks were maintained in the analysis of separate groups, considerable information on both complete and incomplete block effects was lost. The improved precision of estimates of genetic effects permitted with incomplete block designs was thus lost when estimating variance components. With mixed models analysis, however, a single analysis procedure can be used to estimate complete and incomplete block effects (thus improving precision), and to estimate unique variance components for subsets of genetic entries representing unique reference populations.

Another advantage is that genetic covariances between related families, lines, or individuals in different generations of inbreeding from the same reference population can be estimated, along with estimation of separate

genetic variances for each generation. As discussed in Section III.B, the genetic expectations of variance and covariance components under inbreeding are complex, involving more than simply additive and dominance genetic variances (Equation [5]). By testing related lines in different generations of inbreeding and appropriately modeling the genetic variance components within generations and covariances between generations, it may be possible to obtain better estimates of the nonadditive components of variance than have been reported to date (Cornelius and Dudley 1976). Similarly, it is sometimes of interest to compare the genotype-by-environment interactions of different populations grown in the same sets of environments in order to determine if environmental stability differs among the populations (Holland et al. 2000), and this can be accomplished by modeling unique GE interaction variances as well as unique genetic variances for the different populations. Finally, the method is robust for missing data, as already discussed.

These new modeling approaches are already available in practice to most plant breeders. For example, all of the analyses described can be performed with SAS PROC MIXED (Littell et al. 1996), with careful use of the "group" and "subject" options available with the "random" and "repeated" statements. SAS code for specific examples of models with multiple genetic variances are provided in Appendices 3 and 4, and SAS code and example data sets are available at www4.ncsu.edu/~jholland/heritability.html. Tests are available in PROC MIXED for hypotheses generated with these new analysis approaches. If separate variance components are estimated for different subsets of families, one can test the hypothesis that the different subsets have a common population variance, by also analyzing the data as if all entries were from a single population, and using the likelihoods of the two models to conduct a likelihood ratio test of the hypothesis (Section V.B.4).

6. Difficulties Remaining with Mixed Models Analysis. At least one major practical and one major theoretical difficulty currently hinder wider adoption of mixed models analysis by plant breeders, at least in the short term. The practical difficulty is that the computer memory required to estimate all of the fixed and random effects and the covariance components can easily exceed the computer memory available to most plant breeders if a complex model and a large data set are analyzed. Each additional variance component parameter to be estimated dramatically increases memory required. For example, we attempted to estimate unique variance components for five separate genetic populations simultaneously from data collected by Cervantes-Martínez et al. (2001). The study included 100 genotypes within each population that

were randomly assigned to sets, with each set designed as a lattice within each of four environments. Approximately 15 percent of the data points were missing. Due to memory limitations, it was not possible on either a personal computer with 256MB RAM or on a UNIX server with 2GB of RAM to simultaneously estimate with SAS PROC MIXED the five unique genetic variance components, five GE interaction variances, five unique error variances, and common variances due to environments, sets, complete blocks, and incomplete blocks. Therefore, we resorted to analyzing the populations separately. With less complex experimental designs, however, these types of analyses are feasible today (e.g., Appendix 3), and the more memory-intensive computations may be possible in the near future, given the constant and rapid advances in computing processor speed and memory availability.

Research on alternative algorithms for finding maximum likelihood solutions for large and unbalanced data sets also presents a way to solve the computational difficulties of REML. When the number of parameters in a model becomes large, PROC MIXED requires large amounts of memory to identify the maximum likelihood solution with REML because it uses a ridge-stabilized Newton-Raphson algorithm to maximize the logarithm of the residual likelihood function (REML) (SAS Institute Inc. 1999). This method finds the optimum solution in fewer iterations compared to other methods (Lindstrom and Bates 1989), but to do so, it requires matrix inversion. Inversion of the large, sparse matrices associated with models with many parameters is the most memory-intensive portion of the algorithm. A less memory-intensive alternative to the Newton-Raphson algorithm is the derivative-free (DF) algorithm (Graser et al. 1987) in which the residual likelihood function is evaluated explicitly, and its maximum with respect to the variance-covariance components is located without matrix inversion. Although the DF algorithm requires less central processing unit (CPU) time per round, it often requires many more rounds of iterations to obtain converged estimates (Boldman and Van Vleck 1991), making it slower to converge. However, it has been found that this procedure is computationally feasible for experiments involving very large data sets (Graser et al. 1987; Meyer 1989, 1997). Another approach is the use of the Takahashi algorithm to invert large, sparse matrices, which removes most of the constraints on algorithms to invert large matrices. In particular, average information (AI) REML is a quasi-Newton algorithm which requires first derivatives of the likelihood, but replaces second derivatives with the average of the observed and expected information to approximate the second derivative matrix of the function evaluated at the optima. This algorithm has been found to be computationally highly advantageous over DF procedures

(Meyer 1997). The AI REML procedure also produces approximate standard errors of variance component estimates, which are not available with the DF REML procedure. These standard errors are useful for estimating the precision of heritability estimators (Section V.C). Many strategies to reduce the computations in each round of iteration and the number of rounds required to reach convergence have been included in the derivative-free REML (DFREML) (Meyer 1989) and multiple trait derivative-free REML (MTDFREML) (Boldman et al. 1993) programs. MTDFREML software also can implement the AI REML algorithm (Boldman et al. 1993). However, these programs have been designed specifically for applications in animal breeding, and are not available for application in plant breeding yet.

A theoretical difficulty also needs to be resolved before mixed models can be implemented widely for plant breeding experiments. Mixed models can handle both fixed effect factors and random effect factors simultaneously, but plant breeders often deal with a situation that is not easily handled in current mixed models. Often, plant breeders randomly sample lines or families from an experimental population to estimate variance components and heritability in the reference population, in which case the families are a random effect. Generally, however, check entries (usually widely accepted cultivars) are also included in the same experiment so that breeders can compare the best lines from the experimental population to the check entry. Obviously, the check entries were not drawn from the same reference population as the experimental lines. Nor can it reasonably be argued that the check entries are random samples from some other reference population; the check entries were chosen specifically because they are superior! Thus, one could argue that the check entries represent a group of fixed effects, whereas the experimental entries represent random effects drawn from a separate, but definable, reference population. It is not obvious how to handle this situation with mixed models analysis. Although one can model separate variance components for experimental entries and check entries, it seems that in theory no variance component can be associated with the check entries.

Plant breeders did not face this dilemma previously because ANOVA procedures were used for both estimating family means and variance components, and the same equations were used to estimate the family mean squares from a multiple-environment trial whether families were considered fixed or random. Even the F-test for families was the same whether families were considered fixed or random (but changing environments from fixed to random would cause differences in the F-test for families). Therefore, means for both experimental and check entries and

standard errors for their comparisons could be estimated from the ANOVA including all entries. Then, after dropping check entries, a new ANOVA could be performed to obtain the mean squares, and, consequently, the variance component, due to random experimental families only. A similar approach could be used with mixed models: all entries could be considered fixed effects for the purposes of making comparisons between experimental and check entries. Then a second analysis could be performed on the experimental entries only, considering them random effects, to estimate the genetic variance component in the reference population. This is an entirely reasonable approach, because in this case, for the purposes of making comparisons between experimental families and checks, one is interested only in the families actually included in the experiment, rather than in making inferences to the reference population from which they were sampled. (White and Hodge 1989, pp. 29 and 64, discuss the reasons that families can be treated as fixed for some purposes and random for other purposes.) However, some efficiency may be lost with incomplete block designs, as previously mentioned. If there were some way to treat only the check entries as fixed effects and the experimental entries as random effects in the same analysis, one could use such an analysis to estimate the variance components of the random entries while obtaining information on incomplete and complete blocks from the check entries, maximizing the precision of the estimates. In such a case, it still seems to make more sense to conduct a second analysis, considering all entries as fixed effects, to make comparisons among the lines included in the experiment. Otherwise, using a mixed models analysis, the random family effects are predicted, rather than estimated (Lynch and Walsh 1998, pp. 748–749), and best linear unbiased predictors (BLUPs) of the random families are used for comparisons (Robinson 1991). Thus, even if possible, this type of analysis would raise the issue of how experimental line BLUPs can be compared to check entry means from the same experiment. In any case, we are not satisfied with considering all entries, including both checks and random samples from experimental populations, to be random effects drawn from a common population, for the purpose of making comparisons among them with BLUPs, as we have done in some cases (Cervantes-Martínez et al. 2001) simply to make papers acceptable to journal editors. We know of no theoretical work or guidance on this subject and suggest that such work would be useful to plant breeders.

Finally, we also note that the correct expectations of mean squares in the mixed model remains controversial, and that this can affect the variance component estimates. Statisticians do not agree on whether the

expected mean square of a random factor should include the variance component of the interaction of that random factor with a different fixed factor. The debate centers on the assumptions of the model used, specifically, whether interaction effects between fixed and random factors must sum to zero across all levels of the fixed factor (Rawlings 1988, pp. 468–469; Steel et al. 1997, p. 411). Unfortunately, the model and consequent expected mean squares used in SAS PROC MIXED differ from those traditionally used by plant breeders (Steel et al. 1997, pp. 379–384).

C. Precision of REML-based Heritability Estimators

1. Approximate Standard Errors of REML-based Heritability Estimators. Heritability estimates can be constructed from the REML variance component estimates using Equation [12], but the sampling variance of such a heritability estimate is not immediately obvious. Dickerson (1969) presented an approximate standard error for heritability estimated based on variance component estimates, but it is conservative (Hallauer and Miranda 1988, p. 49) and not generally recommended nowadays (Nyquist 1991, p. 310).

The delta method (Lynch and Walsh 1998, p. 807) provides a general method for obtaining approximate standard errors for any statistic based on estimates with an estimated or known sampling variance-covariance matrix. Gordon et al. (1972), Dieters et al. (1995), Singh and Ceccarelli (1995), and Hohls (1996) proposed approximate standard errors for specific heritability estimators using the delta method. The general form of the approximate standard error estimator for variance component-based heritability estimates proposed by Gordon et al. (1972) is appropriate, but the specific formulas provided by them are based on covariance estimates of the estimated variance components that were derived assuming balanced data. Estimates of the covariances between the estimated variance components are provided directly by REML estimation procedures. The use of these estimates is appropriate whether or not the data are balanced when the sample size is large. Hohls (1996) described an appropriate method to obtain approximate standard errors for heritability estimates from a design II experiment, but his derivation of the standard error was incorrect. Dieters et al. (1995) compared two different approximations and an empirical estimate of the variance of heritability estimates, finding that both approximations performed reasonably well compared with the empirical estimate. Singh and Ceccarelli (1995) derived approximate standard errors for heritability estimates based on REML estimates of variance components for single- and multiple-location trials of random genotypes.

Here, we derive approximate sampling variances and standard errors of heritability estimates based on variance and covariance components estimated by REML. The variance component estimates require the assumption of normally-distributed data, but do not require the assumption of balanced data. The proposed method is robust for missing data, assuming that data are missing at random (Little and Rubin 1987). All components necessary to use the equations are included in the SAS PROC MIXED output, and SAS code to obtain the components and to compute the estimators directly is presented in Appendices 1 to 4.

An estimator for narrow-sense heritability for family means (h_f^2, Section V.A) is:

$$\hat{h}_f^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_{\bar{P}}^2},$$

where σ_F^2 is the family variance component in the reference population, and $\sigma_{\bar{P}}^2$ is the phenotypic variance of family mean deviations in the reference population (Equation [12]). The phenotypic variance of family mean deviations is estimated as the sum of the estimates of the genetic variance component and other variance components multiplied by coefficients. The other variance components generally include those corresponding to family-by-environment interaction (FE) (σ_{FE}^2), experimental error (σ_e^2), and within-plot variance (σ_w^2), if data on individual plants are available. To obtain a general form for the sampling variance of the heritability estimate, the phenotypic variance component will be written as:

$$\hat{\sigma}_{\bar{P}}^2 = \hat{\sigma}_F^2 + c_2\hat{\sigma}_2^2 + c_3\hat{\sigma}_3^2 + \ldots + c_k\hat{\sigma}_k^2,$$

where k is the number of variance components contributing to the phenotypic variance of family means; and σ_2^2, σ_3^2, ..., σ_k^2 refer to the (k-1) other variance components whose estimates are multiplied by coefficients c_2, c_3, ..., c_k, respectively, and summed along with σ_F^2 to estimate phenotypic variance of family mean deviations. To estimate the sampling variance of the heritability estimator, estimates of sampling variances of all of the variance component estimates included in the heritability equation are needed. In addition, estimates of the covariances between all of the variance component estimates included in the heritability formula are required. These elements can be written as a $k \times k$ variance-covariance matrix, **C**:

$$\hat{\mathbf{C}} = \begin{bmatrix} \hat{V}(\hat{\sigma}_F^2) & \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_2^2) & \cdots & \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_k^2) \\ \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_2^2) & \hat{V}(\hat{\sigma}_2^2) & \cdots & \hat{C}(\hat{\sigma}_2^2, \hat{\sigma}_k^2) \\ \vdots & \vdots & \ddots & \vdots \\ \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_k^2) & \hat{C}(\hat{\sigma}_2^2, \hat{\sigma}_k^2) & \cdots & \hat{V}(\hat{\sigma}_k^2) \end{bmatrix},$$

where $\hat{V}(\hat{\sigma}_i^2)$ refers to the estimated variance of the ith variance component estimate, and $\hat{C}(\hat{\sigma}_i^2, \hat{\sigma}_j^2)$ refers to the estimated covariance between the ith and jth variance component estimates.

The variance component estimates computed by REML procedures are asymptotically normally distributed, with variances and covariances given by the **C** matrix. The approximation to normality improves with larger sample sizes (Searle et al. 1992). The delta method can be used to obtain the approximate variance of a function of asymptotically normally distributed estimators (Lindsey 1996; Lynch and Walsh 1998, p. 807). Therefore, given REML estimates of the variance components and of the elements of the **C** matrix, and considering the heritability estimate to be a function of the estimators, $\hat{\sigma}_F^2$, $\hat{\sigma}_2^2$, $\hat{\sigma}_3^2$, ..., $\hat{\sigma}_k^2$, we can apply the delta method to obtain the approximate sampling variance of the heritability estimate. In general, the approximate sampling variance of any estimator, ϕ, that is a function of k moments, m_1, m_2, ..., m_k, is given by:

$$V(\phi) \approx \sum_i \left(\frac{\partial \phi}{\partial m_i} \right)^2 V(m_i) + \sum_{i \neq i'} \frac{\partial \phi}{\partial m_i} \frac{\partial \phi}{\partial m_{i'}} C(m_i, m_{i'}) \qquad [15]$$

[see, for example, Mode and Robinson (1959) or Bulmer (1985, p. 86)]. This formula is obtained as the first two (lowest order) terms in the Taylor series expansion of ϕ around its true value. Equation [15] can also be written in matrix form as:

$$V(\phi) = \begin{bmatrix} \frac{\partial \phi}{\partial m_1} \\ \frac{\partial \phi}{\partial m_2} \\ \vdots \\ \frac{\partial \phi}{\partial m_k} \end{bmatrix}^T \begin{bmatrix} V(m_1) & C(m_1, m_2) & \cdots & C(m_1, m_k) \\ C(m_1, m_2) & V(m_2) & \cdots & C(m_2, m_k) \\ \vdots & \vdots & \ddots & \vdots \\ C(m_1, m_k) & C(m_2, m_k) & \cdots & V(m_k) \end{bmatrix} \begin{bmatrix} \frac{\partial \phi}{\partial m_1} \\ \frac{\partial \phi}{\partial m_2} \\ \vdots \\ \frac{\partial \phi}{\partial m_k} \end{bmatrix}.$$

Applying this quadratic form to the heritability estimator, gives:

$$\hat{V}(\hat{h}_f^2) = \begin{bmatrix} \frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_F^2} \\ \frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_2^2} \\ \vdots \\ \frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_k^2} \end{bmatrix}^T \begin{bmatrix} \hat{V}(\hat{\sigma}_F^2) & \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_2^2) & \cdots & \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_k^2) \\ \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_2^2) & \hat{V}(\hat{\sigma}_2^2) & \cdots & \hat{C}(\hat{\sigma}_2^2, \hat{\sigma}_k^2) \\ \vdots & \vdots & \ddots & \vdots \\ \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_k^2) & \hat{C}(\hat{\sigma}_2^2, \hat{\sigma}_k^2) & \cdots & \hat{V}(\hat{\sigma}_k^2) \end{bmatrix} \begin{bmatrix} \frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_F^2} \\ \frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_2^2} \\ \vdots \\ \frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_k^2} \end{bmatrix}.$$

The $k \times 1$ column vector containing the derivatives of the heritability estimator with respect to the different variance components included in the estimator will be referred to as **d**. In this way, the general formula for the sampling variance for the heritability estimate can be expressed in matrix notation as:

$$\hat{V}(\hat{h}_f^2) = \hat{\mathbf{d}}'\hat{\mathbf{C}}\hat{\mathbf{d}}. \qquad [16]$$

The estimated **C** matrix can be obtained directly from the output of SAS PROC MIXED (SAS Institute Inc. 1999) and from GENSTAT (Singh and Ceccarelli 1995; Hohls 1996; Payne and Arnold 1998). All that remains, therefore, is to determine the derivatives involved in the **d** vector to obtain a specific formula for the sampling variance of a particular heritability estimator.

The derivative of the heritability estimator with respect to any of the variance components in the equation can be written as:

$$\frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_i^2} = \frac{\left(\hat{\sigma}_P^2\right)\frac{\partial \hat{\sigma}_F^2}{\partial \hat{\sigma}_i^2} - \left(\hat{\sigma}_F^2\right)\frac{\partial \hat{\sigma}_P^2}{\partial \hat{\sigma}_i^2}}{\left(\hat{\sigma}_P^2\right)^2}$$

Therefore, the **d** vector can be simplified to:

$$\hat{\mathbf{d}} = \frac{1}{\left(\hat{\sigma}^2_{\bar{P}}\right)^2}\begin{bmatrix} \left(\hat{\sigma}^2_{\bar{P}}\right)\frac{\partial\hat{\sigma}^2_F}{\partial\hat{\sigma}^2_F} - \left(\hat{\sigma}^2_F\right)\frac{\partial\hat{\sigma}^2_{\bar{P}}}{\partial\hat{\sigma}^2_F} \\ \left(\hat{\sigma}^2_{\bar{P}}\right)\frac{\partial\hat{\sigma}^2_F}{\partial\hat{\sigma}^2_2} - \left(\hat{\sigma}^2_F\right)\frac{\partial\hat{\sigma}^2_{\bar{P}}}{\partial\hat{\sigma}^2_2} \\ \vdots \\ \left(\hat{\sigma}^2_{\bar{P}}\right)\frac{\partial\hat{\sigma}^2_F}{\partial\hat{\sigma}^2_k} - \left(\hat{\sigma}^2_F\right)\frac{\partial\hat{\sigma}^2_{\bar{P}}}{\partial\hat{\sigma}^2_k} \end{bmatrix} = \frac{1}{\left(\hat{\sigma}^2_{\bar{P}}\right)^2}\begin{bmatrix} \left(\hat{\sigma}^2_{\bar{P}}\right)(1) - \left(\hat{\sigma}^2_F\right)(1) \\ \left(\hat{\sigma}^2_{\bar{P}}\right)(0) - \left(\hat{\sigma}^2_F\right)(c_2) \\ \vdots \\ \left(\hat{\sigma}^2_{\bar{P}}\right)(0) - \left(\hat{\sigma}^2_F\right)(c_k) \end{bmatrix}$$

$$= \frac{1}{\hat{\sigma}^2_{\bar{P}}}\begin{bmatrix} \left(\hat{\sigma}^2_P - \hat{\sigma}^2_F\right) / \hat{\sigma}^2_{\bar{P}} \\ -\left(c_2\hat{\sigma}^2_F\right) / \hat{\sigma}^2_{\bar{P}} \\ \vdots \\ -\left(c_k\hat{\sigma}^2_F\right) / \hat{\sigma}^2_{\bar{P}} \end{bmatrix} = \frac{1}{\hat{\sigma}^2_{\bar{P}}}\begin{bmatrix} 1 - \hat{h}^2_f \\ -c_2\hat{h}^2_f \\ \vdots \\ -c_k\hat{h}^2_f \end{bmatrix}.$$

This can be further generalized by specifying the relevant variance components in a matrix called σ, and defining the family and phenotypic variances as functions of coefficient vectors $\boldsymbol{\lambda}_\mathbf{G}$ and $\boldsymbol{\lambda}_\mathbf{P}$ multiplied by $\boldsymbol{\sigma}$:

$$\boldsymbol{\sigma} = \begin{bmatrix} \sigma^2_F \\ \sigma^2_2 \\ \vdots \\ \sigma^2_k \end{bmatrix},$$

$$\boldsymbol{\lambda}_\mathbf{G} = \begin{bmatrix} 1 \\ 0 \\ \vdots \\ 0 \end{bmatrix}, \qquad [17]$$

$$\boldsymbol{\lambda}_\mathbf{P} = \begin{bmatrix} 1 \\ c_2 \\ \vdots \\ c_k \end{bmatrix}, \qquad [18]$$

$$\sigma_F^2 = \begin{bmatrix} 1 \\ 0 \\ \vdots \\ 0 \end{bmatrix}^T \begin{bmatrix} \sigma_F^2 \\ \sigma_2^2 \\ \vdots \\ \sigma_k^2 \end{bmatrix} = \boldsymbol{\lambda}_G^T \boldsymbol{\sigma},$$

$$\sigma_{\bar{P}}^2 = \begin{bmatrix} 1 \\ c_2 \\ \vdots \\ c_k \end{bmatrix}^T \begin{bmatrix} \sigma_G^2 \\ \sigma_2^2 \\ \vdots \\ \sigma_k^2 \end{bmatrix} = \boldsymbol{\lambda}_P^T \boldsymbol{\sigma}.$$

Using these equations, the **d** vector can be written as:

$$\mathbf{d} = \frac{1}{\sigma_{\bar{P}}^2} \left[\boldsymbol{\lambda}_G - (h_f^2 \boldsymbol{\lambda}_P) \right]. \qquad [19]$$

As an example, we will derive the sampling variance for heritability on an experimental-unit basis (plot basis) estimated from an experiment in which data are taken on a set of random half-sib families grown in randomized complete block trials in multiple environments. In this case, heritability on an experimental-unit basis is estimated by:

$$\hat{h}_f^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \hat{\sigma}_{FE}^2 + \hat{\sigma}_{\varepsilon'}^2} = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_P^2},$$

where σ_{FE}^2 is the family-by-environment interaction variance component, and $\sigma_{\varepsilon'}^2$ is the experimental error variance component.

The variance-covariance matrix of the variance component estimates is:

$$\hat{\mathbf{C}} = \begin{bmatrix} \hat{V}(\hat{\sigma}_F^2) & \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_{FE}^2) & \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_{\varepsilon'}^2) \\ \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_{FE}^2) & \hat{V}(\hat{\sigma}_{FE}^2) & \hat{C}(\hat{\sigma}_{FE}^2, \hat{\sigma}_{\varepsilon'}^2) \\ \hat{C}(\hat{\sigma}_F^2, \hat{\sigma}_{\varepsilon'}^2) & \hat{C}(\hat{\sigma}_{FE}^2, \hat{\sigma}_{\varepsilon'}^2) & \hat{V}(\hat{\sigma}_{\varepsilon'}^2) \end{bmatrix}.$$

The genetic and phenotypic coefficient vectors are:

$$\lambda_G^T = \begin{bmatrix} 1 & 0 & 0 \end{bmatrix},$$
$$\lambda_P^T = \begin{bmatrix} 1 & 1 & 1 \end{bmatrix}.$$

Applying the delta method, we obtain:

$$\hat{\mathbf{d}} = \begin{bmatrix} \frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_F^2} \\ \frac{\partial \hat{h}_f^2}{\partial \hat{\sigma}_{FE}^2} \\ \frac{\partial \hat{h}_f^2}{\partial \sigma_{\varepsilon'}^2} \end{bmatrix} = \begin{bmatrix} \frac{\hat{\sigma}_{FE}^2 + \hat{\sigma}_{\varepsilon'}^2}{(\hat{\sigma}_P^2)^2} \\ \frac{-\hat{\sigma}_F^2}{(\hat{\sigma}_P^2)^2} \\ \frac{-\hat{\sigma}_F^2}{(\hat{\sigma}_P^2)^2} \end{bmatrix} = \frac{1}{\hat{\sigma}_P^2} \begin{bmatrix} \frac{\hat{\sigma}_{FE}^2 + \hat{\sigma}_{\varepsilon'}^2}{\hat{\sigma}_P^2} \\ \frac{-\hat{\sigma}_F^2}{\hat{\sigma}_P^2} \\ \frac{-\hat{\sigma}_F^2}{\hat{\sigma}_P^2} \end{bmatrix}$$

$$= \frac{1}{\hat{\sigma}_P^2} \begin{bmatrix} \frac{\hat{\sigma}_F^2 + \hat{\sigma}_{FE}^2 + \hat{\sigma}_{\varepsilon'}^2 - \hat{\sigma}_F^2}{\hat{\sigma}_P^2} \\ \frac{-\hat{\sigma}_F^2}{\hat{\sigma}_P^2} \\ \frac{-\hat{\sigma}_F^2}{\hat{\sigma}_P^2} \end{bmatrix} = \frac{1}{\hat{\sigma}_P^2} \begin{bmatrix} 1 - \hat{h}_f^2 \\ -\hat{h}_f^2 \\ -\hat{h}_f^2 \end{bmatrix} = \frac{1}{\hat{\sigma}_P^2} [\boldsymbol{\lambda}_{\mathbf{G}} - (\hat{h}_f^2 \boldsymbol{\lambda}_{\mathbf{P}})].$$

$$\hat{V}(\hat{h}_f^2) = \hat{\mathbf{d}}^T \hat{\mathbf{C}} \hat{\mathbf{d}}$$

$$= \frac{1}{(\hat{\sigma}_P^2)^4} \begin{bmatrix} (\hat{\sigma}_{FE}^2 + \hat{\sigma}_{\varepsilon'}^2)^2 V(\hat{\sigma}_F^2) + (\hat{\sigma}_F^2)^2 [V(\hat{\sigma}_{FE}^2) + V(\hat{\sigma}_{\varepsilon'}^2) + 2C(\hat{\sigma}_{FE}^2, \hat{\sigma}_{\varepsilon'}^2)] \\ -2\hat{\sigma}_F^2 (\hat{\sigma}_{FE}^2 + \hat{\sigma}_{\varepsilon'}^2)[C(\hat{\sigma}_F^2, \hat{\sigma}_{FE}^2) + C(\hat{\sigma}_F^2, \hat{\sigma}_{\varepsilon'}^2)] \end{bmatrix}.$$

The estimate of heritability on a family-mean basis has the following form:

$$\hat{h}_f^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \frac{\hat{\sigma}_{FE}^2}{e} + \frac{\hat{\sigma}_{\varepsilon'}^2}{er}},$$

the $\mathbf{C}$ matrix remains unchanged, and the $\boldsymbol{\lambda}_{\mathbf{G}}$ and $\boldsymbol{\lambda}_{\mathbf{P}}$ vectors are:

$$\boldsymbol{\lambda}_{\mathbf{G}}^T = \begin{bmatrix} 1 & 0 & 0 \end{bmatrix} \text{ and } \boldsymbol{\lambda}_{\mathbf{P}}^T = \begin{bmatrix} 1 & \frac{1}{e} & \frac{1}{er} \end{bmatrix},$$

leading to estimates of the phenotypic variance of family mean deviations and heritability as follows:

$$\hat{\sigma}_{\bar{P}}^2 = \boldsymbol{\lambda}_{\mathbf{P}}^T \hat{\boldsymbol{\sigma}}, \quad \hat{h}_f^2 = \frac{\boldsymbol{\lambda}_{\mathbf{G}}^T \hat{\boldsymbol{\sigma}}}{\boldsymbol{\lambda}_{\mathbf{P}}^T \hat{\boldsymbol{\sigma}}} = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_{\bar{P}}^2}.$$

The **d** vector is:

$$\mathbf{d} = \frac{1}{\hat{\sigma}^2_P}[\boldsymbol{\lambda}_G - (\hat{h}^2_f \boldsymbol{\lambda}_P)],$$

and the variance of the estimate of heritability of family means is obtained with Equation [16].

The standard error of the heritability estimate is the square root of the variance (Equation [16]). For example, the standard error of the heritability estimate on a plot basis in the example given is:

$$s.e.(\hat{h}^2_f) = \frac{1}{(\hat{\sigma}^2_P)^2}\sqrt{\left(\hat{\sigma}^2_{FE} + \hat{\sigma}^2_{\epsilon'}\right)^2 \hat{V}(\hat{\sigma}^2_F) + (\hat{\sigma}^2_F)^2[\hat{V}(\hat{\sigma}^2_{FE}) + \hat{V}(\hat{\sigma}^2_{\epsilon'}) + 2\hat{C}(\hat{\sigma}^2_{FE}, \hat{\sigma}^2_{\epsilon'})] - 2\hat{\sigma}^2_F(\hat{\sigma}^2_{FE} + \hat{\sigma}^2_{\epsilon'})[\hat{C}(\hat{\sigma}^2_F, \hat{\sigma}^2_{FE}) + \hat{C}(\hat{\sigma}^2_F, \hat{\sigma}^2_{\epsilon'})]}.$$

This formula is algebraically equivalent to that given by Singh et al. (1995), who also used the delta method. It is also equivalent to the estimator obtainable from a general form given by Gordon et al. (1972) when data are balanced. Gordon et al. (1972) derived their covariance estimators by assuming balanced data and independent mean squares, but this may not be valid when data are unbalanced. On the other hand, whether or not data are unbalanced, the variances and covariances of the elements of the heritability estimator are given by the **C** matrix estimated by REML procedures, assuming large sample sizes. We can be certain, therefore, that the variance estimators described here are valid in the case of unbalanced data.

2. Alternative Methods for Estimating Precision of REML-based Heritability Estimates. The approximate standard errors of heritability estimate may not lead to reliable confidence interval estimators, because of the unknown distribution of the heritability estimates (Lynch and Walsh 1998). The delta method approximate standard errors also assume large sample size, and it is not known exactly how large the number of families, environments, or replications an experiment should have to obtain valid estimates of precision of heritability estimates. Dieters et al. (1995) compared estimates of standard errors of heritability for two traits in pine trees (*Pinus elliottii*) estimated with the delta method to those estimated with a simpler approximation given by Dickerson (1969) (see Nyquist 1991, p. 310) and an empirical estimate. They reported that the delta method approximation of standard error of heritability appeared to be reliable, but seemed slightly less conservative and perhaps more biased than the Dickerson approximation.

An alternative to estimating approximate standard errors based on parametric methods is the use of data resampling techniques to obtain

both standard errors and confidence intervals for heritability estimates (Lynch and Walsh 1998, pp. 569–570). Furthermore, resampling methods to obtain estimates of heritability may be more robust for smaller sample sizes than the delta method estimators. Zhu and Weir (1996) suggested that better estimates of variances of heritability estimates for diallel designs can be obtained with the jackknife method (Miller 1974) than with approximate formulae. This may also be true for other mating designs. Knapp and Bridges (1988) also used jackknife methods for estimating confidence intervals for ratios of variance component estimates, but they noted that the extension of jackknife methods to complex data structures was difficult because the optimal data resampling strategy to use in jackknifing in complex experimental designs is not always clear. For example, if data exist on multiple families evaluated in multiple replications within multiple environments, it is not obvious how to properly resample the factorial data set to simultaneously account for uncertainty in family, FE interaction, and error variances. Further research on the reliability of delta method approximations and resampling methods for estimating the precision of REML-based heritability estimates would be helpful.

D. Accounting for Unbalanced Data in Formulas for Heritability on a Family-Mean Basis

The estimates of the variance among phenotypic means ($\sigma^2_{\bar{P}}$) that serve as the denominators of heritabilities on a family-mean basis in Table 2.1 are correct only if the data are balanced. In the balanced case, the divisor for each variance component comprising $\sigma^2_{\bar{P}}$ represents the number of effects corresponding to that variance component included in each family mean. For example, family j might be evaluated in e environments. In this case, each family mean includes an average over e unique FE effects, and the contribution of σ^2_{FE} to the variance among family means is reduced by a factor of exactly e. Therefore, the divisor of σ^2_{FE} in the formula for the variance among family means is e.

As described in Section IV, the phenotypic variance of family means can be obtained as the mean square for families divided by the total number of observations per family (Nyquist 1991, pp. 256–257). The phenotypic variance of family means is also equal to the variance of a family mean plus the variance component due to families. However, this is no longer true when data are unbalanced because the total number of observations per family is not equal among families. Also, the coefficients on the variance components in the expected mean square for families are not integers corresponding to consistent numbers of levels of the design factors as when data are balanced (Section V.B.2).

Selection among families when data are unbalanced should be based on the least square means of families. Therefore, by analogy to the balanced data situation, when data are unbalanced, one can compute the phenotypic variance of family means as the average variance of the family least square means plus the variance component due to families. Least square means are computed as $\mathbf{L}\hat{\beta}$, where $\hat{\beta}$ is the vector of fixed effects (Section V.B.5) (including family effects in this case), and $\mathbf{L}$ is the vector of coefficients that define a least square mean, according to rules described by Rawlings (1988, pp. 460–462; see also SAS Institute 1999). Differences among least square means are defined as $(\mathbf{L}_i - \mathbf{L}_j)\hat{\beta}$, where $\mathbf{L}_i$ and $\mathbf{L}_j$ are the vectors defining the two least square means. The variance of a difference between least square means is computed as $(\mathbf{L}_i - \mathbf{L}_j)(\mathbf{X}'\hat{\mathbf{V}}^{-1}\mathbf{X})^{-}(\mathbf{L}_i - \mathbf{L}_j)'$ (SAS Institute 1999). The average variance of comparisons of least square means across all pairs of least square means can be computed and summed with the variance component for families to obtain the phenotypic variance of family means. This factor can then be used as the denominator for heritability formulas on a family-mean basis. Another approach is to simply compute all of the family least square means, then calculate the variance among those means. Finally, one could compute the coefficient for the family variance component in the expected mean square for families given the actual unbalanced design of the experiment (using the random statement in PROC GLM of SAS, for example, SAS Institute 1999), and divide the mean square for families by the coefficient to obtain the variance among family least square means. The mean square may have to be constructed as a linear function of variance-component estimates from PROC MIXED, multiplied by the appropriate coefficients in the expected mean squares, obtained from PROC GLM. We are not certain how to simply relate these expressions to the linear combinations of variance component estimates, as was possible for the balanced data situation (Equation [9]). Further research to clarify this issue is needed. From empirical investigation, we have found that Equation [9] is a good approximation to the empirical variance among family least square means if e is replaced by e_h, the harmonic mean of the number of environments per family, and er is replaced by p_h, the harmonic mean of the total number of plots in which each family is observed. The harmonic mean of the number of environments per family is:

$$e_h = \frac{f}{\sum_{j=1}^{f} \frac{1}{e_j}},$$

where e_j is the number of environments in which the jth family is tested. The harmonic mean of the number of plots per family is:

$$p_h = \frac{f}{\sum_{j=1}^{f} \frac{1}{\sum_{i=1}^{e_j} r_{ij}}} = \frac{f}{\sum_{j=1}^{f} \frac{1}{p_j}},$$

where r_{ij} is the number of replications of family j in environment i, and p_j is the total number of replications (plots) of family j across all environments.

VI. ESTIMATING HERITABILITY FROM PARENT-OFFSPRING REGRESSION

A. REML Estimates of the Parent-Offspring Regression Coefficient

REML methods can be used to estimate the parent-offspring covariance and the parent phenotypic variance, leading to a REML-based estimator of heritability. This approach follows that outlined in Section V, and will be the optimal method when data are unbalanced. Furthermore, with mixed models analysis and appropriate experimental design, one can simultaneously estimate heritability from parent-offspring regression and from the ratio of the family variance component to the phenotypic variance of family means. An example of this method is presented in Appendix 3.

B. Heritability Estimated from Parent-Offspring Regression without Inbreeding

We demonstrated in Section III.A that the genetic covariance between outbred parent and outbred offspring is $(1/2)\sigma^2_A + (1/4)\sigma^2_{AA}$ (ignoring higher-order epistatic terms). If parents and progeny are grown in independent environments, then the covariance between their phenotypic values is the genetic covariance (Section II.C). If parents and progeny are grown in the same (or nonindependent) environments, however, the covariance between their phenotypic values will include a portion of the genotype-by-environment variance component, requiring a more complex analysis of covariance to partition the genetic variance component from the genotype-by-environment interaction variance component (Casler 1982; Nyquist 1991, pp. 281–282).

The expectation of the regression of offspring values on parental values is identical whether a single offspring from each parent or the mean of many offspring are measured. If parents are random-mated via open-pollination and the phenotypic value of only one parent of each progeny set is known (corresponding to selection on one parent only), then the regression of offspring on parent phenotypic values is equal to half of the narrow-sense heritability:

$$E\left[\hat{b}_{OP}\right] = E\left[\frac{\mathrm{Cov}(P,O)}{\mathrm{Var}(P)}\right] = \frac{\frac{1}{2}\sigma_A^2 + \frac{1}{4}\sigma_{AA}^2}{\sigma_P^2} = \frac{1}{2}h_1^2,$$

where h_1^2 = narrow-sense heritability corresponding to selection response in response units that are members of the initial population formed by intermating selected parents (Section V.A; Nyquist 1991, pp. 250–251). This is distinguished from the permanent response to selection, as measured in a population derived from the initial response population but resulting from many generations of random mating (Section V.A). The epistatic variance components do not contribute to the numerator of the permanent response to selection (Nyquist 1991, pp. 250–251; Holland 2001), so narrow-sense heritability corresponding to permanent response to selection has the familiar form:

$$h_\infty^2 = \frac{\sigma_A^2}{\sigma_P^2}$$

(Falconer and Mackay 1996, p. 160; Nyquist 1991, p. 251). Formulas for h_∞^2 can be obtained from the formulas for h_1^2 simply by deleting the epistatic components of variance from the numerator.

If the phenotypic values of both parents of each progeny group are known (corresponding to selection on both parents), then the regression of offspring values on mean parental values is directly equal to the narrow-sense heritability:

$$E[b_{O\bar{P}}] = E\left[\frac{\mathrm{Cov}(\bar{P},O)}{\mathrm{Var}(\bar{P})}\right] = \frac{\mathrm{Cov}(\frac{1}{2}P_f + \frac{1}{2}P_m, O)}{\mathrm{Var}[\frac{1}{2}(P_f + P_m)]} = \frac{\frac{1}{2}\mathrm{Cov}(P_f,O) + \frac{1}{2}\mathrm{Cov}(P_m,C}{\frac{1}{4}[\mathrm{Var}(P_f) + \mathrm{Var}(P_m)]}$$

$$= \frac{\frac{1}{2}\sigma_A^2 + \frac{1}{4}\sigma_{AA}^2}{\frac{1}{2}\sigma_P^2} = \frac{\sigma_A^2 + \frac{1}{2}\sigma_{AA}^2}{\sigma_P^2} = h_1^2. \qquad [20]$$

These regression coefficients provide estimators of narrow-sense heritability in Lush's original sense because the phenotypic variance in the denominator is the phenotypic variance of individual plants. Because data are taken from individual parental plants, these estimators are use-

ful only to the extent that individual plant data are useful. Thus, such estimators are probably more useful for traits that are not greatly affected by interplant competition.

A major advantage to estimating heritability from parent-offspring regression is that the sample of parents chosen does not have to be a random sample from the reference population, in contrast to all of the heritability estimators based on variance component estimation (Falconer and Mackay 1996, p. 181; Lynch and Walsh 1998, p. 537; Nyquist 1991, p. 281). In some cases, breeders have data only on selected plants and their offspring, and in such a case, the parent-offspring regression method will provide an unbiased estimator of heritability.

C. Heritability Estimated from Parent-Offspring Regression with Inbreeding

Heritability may be estimated from the regression of self-fertilized offspring phenotypic values on their parental values. For example, parents in a random-mating population may be self-fertilized to form $S_{0:1}$ families and the regression of $S_{0:1}$ line means on S_0 parents has the following expectation:

$$E[\hat{b}_{S_1,S_0}] = \frac{C_{001}}{\text{Var}(S_0\text{plants})} = \frac{\sigma_A^2 + \frac{1}{2}\sigma_D^2 + \frac{1}{2}D_1 + \sigma_{AA}^2}{\sigma_P^2} = h_1^2$$

(Nyquist 1991, p. 303). Such heritability estimators are appropriate only for obtaining the expected response to selection conducted in the same parental generation and evaluated in the same offspring generation as used in the estimation experiment. For example, the heritability estimator based on the regression of $S_{0:1}$ line means on their S_0 parents can be used to obtain the expected response to selection among S_0 plants as evaluated in $S_{0:1}$ lines. However, if one is interested in the response to selection among S_0 plants as evaluated in highly homozygous lines derived from them ($S_{0:\infty}$ lines), one requires the numerator $C_{00\infty}$ (equal to $\sigma_A^2 + D_1 + \sigma_{AA}^2$) in the heritability estimator. Similarly, if one is interested in the response to selection among S_0 plants as measured in outbred progeny developed from intermating the selected S_0's (or the selfed progeny of the S_0's), one requires the narrow-sense heritability estimator given in Equation [20] that does not involve σ_D^2 or D_1 in the numerator.

More frequently, plant breeders have data on earlier and later generations of inbreeding of lines derived from the same common ancestor. For example, F_4 generation lines can be regressed on F_3 generation lines derived from the same common F_2 ancestor. Using the S-generation notation, this is the regression of $S_{0:2}$ lines on their $S_{0:1}$ parents, and the

relevant covariance is $C_{tgg'} = C_{012} = \sigma^2_A + (1/8)\sigma^2_D + (5/4)D_1 + (3/16)D^*_2 + \sigma^2_{AA}$. The relevant phenotypic variance in the denominator of the regression coefficient is the phenotypic variance among $S_{0:1}$ line means, which includes $C_{tgg} = C_{011} = \sigma^2_A + (1/4)\sigma^2_D + D_1 + (1/8)D^*_2 + \sigma^2_{AA}$ as the genotypic variance component. In general, the expectation of the regression coefficient is:

$$E[\hat{b}_{\overline{OP}}] = \frac{C_{tgg'}}{C_{tgg} + \frac{C_{tgg}E}{e} + \frac{\sigma^2_{\varepsilon'}}{er}}. \qquad [21]$$

Such heritability estimators strictly refer to response to selection among $S_{t:g}$ lines as evaluated in $S_{t:g'}$ offspring lines. It also refers to selection among individual inbred plants as measured in later generation inbred lines by setting $t = g$. The permanent response to selection evaluated in highly homozygous offspring lines involves the numerator $C_{tg\infty}$. The response to selection evaluated in outbred progeny involves the additive portion of $C_{tgg'}$ (which also equals the additive portion of C_{tgg}) in the numerator.

Nyquist (1991) suggested that appropriate estimators of heritability corresponding to the response to selection among S_0 plants as measured in $S_{0:1}$ lines can be obtained by adjusting the regression of $S_{t:g}$ lines on S_t parents for any pair of generations t and g (p. 305). Assuming that the genetic variance is completely additive, the heritability estimator can be obtained from the parent-offspring regression of any pair of inbred generations as:

$$h^2 = \frac{b_{OP}}{1 + F_t(1 - b_{OP})},$$

where F_t is the inbreeding coefficient in generation t. Gibson (1996) showed that this estimator can be severely biased for particular allele frequencies if nonadditive effects are important or if genotype-by-environment interactions are important, but in any case, this estimator is closer to the regression of $S_{0:1}$ lines on S_0 parents than is the unadjusted regression coefficient of $S_{t:g}$ lines on S_t parents. Perhaps Nyquist (1991, p. 305) and Gibson (1996) did not emphasize strongly enough that this correction is valid only if the regression estimate to be adjusted involves phenotypic values of individual parents in the S_t generation. If the parental values instead are phenotypic means of $S_{t:g}$ lines, then the phenotypic variance among parental values cannot be adjusted to equal the phenotypic variance among individual S_0 plants.

Holthaus et al. (1996) estimated the heritability of β-glucan content in oat (*Avena sativa*) using the regression of self-fertilized progeny means on individual single-plant values of highly inbred parents. They then adjusted the estimate to refer to selection among noninbred plants using Smith and Kinman's (1965) correction of $h^2 = b_{OP}/(2F_t) = b_{OP}/2$ (for homozygous parents with $F_t = 1$). Nyquist (1991) pointed out that the Smith and Kinman correction factor is incorrect, and the adjusted heritability estimate $[1 + F_t(1 - b_{OP})]$ should be used instead (p. 305). Therefore, Holthaus et al. (1996) should have made the following adjustment under the assumption of predominant additive genetic variance: $h^2 = b_{OP}/[1 + F_t(1 - b_{OP})] = b_{OP}/1.5$, resulting in an estimate of $h^2 = 0.55/1.5 = 0.37$, rather than $h^2 = 0.26$.

VII. ESTIMATING REALIZED HERITABILITY

Distinct from all other estimation procedures discussed in this chapter, realized heritability estimation relies on determining how much of the selection differential applied in previous generations was achieved as a response in progeny. It is a retrospective analysis, although the estimate can be used to make predictions about future responses to selection in similar populations, at least in the short-term. Realized heritability (h_r^2) can be estimated by rearranging the response to selection formula and solving for heritability as a ratio of the observed response to selection (R) to the observed selection differential (S):

$$\hat{h}_r^2 = \frac{\hat{R}}{\hat{S}}.$$

In order for this estimate to be freed from GE interaction bias, the response to selection should be measured in an independent environment from the selection differential. This formula can be generalized to estimating realized heritability from response to multiple generations of selection by performing standard least squares regression of cumulative response on the cumulative selection differential (Hill 1972; Nyquist 1991, p. 283). Walsh and Lynch (1999) proposed a weighted least squares analysis to account for variation due to genetic drift and for correlations between responses observed in different cycles when estimating realized heritability. If selection differentials and responses are measured from n cycles of recurrent selection, the weighted least squares estimate of realized heritability is:

$$\hat{h}_r^2 = (\mathbf{S}^T\mathbf{V}^{-1}\mathbf{S})^{-1}\mathbf{S}^T\mathbf{V}^{-1}\mathbf{R},$$

where **S** is an $n \times 1$ column vector of cumulative selection differentials, **R** is an $n \times 1$ column vector of responses, and **V** is an $n \times n$ variance-covariance matrix of the selection response. The elements of **V** are:

$$w_{ii} = [2F_i h_r^2 + \frac{1}{M_i} + \frac{1}{M_0}]\sigma_{\bar{P}}^2$$

$$w_{ij} = [2F_i h_r^2 \sigma_{\bar{P}}^2 + \frac{1}{M_0}]\sigma_{\bar{P}}^2,$$

where F_i is the inbreeding coefficient in cycle i ($0 < i < j$), M_i is the number of families from cycle i tested in the evaluation trial, and $\sigma_{\bar{P}}^2$ is the phenotypic variance among family means, and h_r^2 is the heritability estimate itself. One can use the standard least squares regression estimate of realized heritability as an initial estimator to use in the **V** matrix of the equation, and then use the equation iteratively until converging on a stable solution. The standard error of the realized heritability estimate is the square root of the sampling variance: $\text{Var}(h_r^2) = (\mathbf{S}^T\mathbf{V}^{-1}\mathbf{S})^{-1}$. Holland et al. (2000) used this method to estimate realized heritability from three cycles of recurrent selection for grain yield in oat.

VIII. EXAMPLES OF HERITABILITY ESTIMATES

A. Broad-Sense Heritability for Clonally Propagated Species

For clonally propagated species, the genotypic content of parents and their offspring are identical, therefore, the expected covariance of parent and offspring phenotypes from independent environments is equal to the total genotypic variance: $\text{E}[\text{Cov}(Y_P, Y_O)] = \text{E}[\text{Cov}(G_P, G_O)] = \sigma_G^2 = \sigma_A^2 + \sigma_D^2 + \sigma_{AA}^2 + \sigma_{AD}^2 + \sigma_{DD}^2 + \ldots$. It follows from this that the response to selection among clonally propagated individuals or families involves the total genotypic variance. Therefore, we seek heritability estimators of the form σ_G^2/σ_P^2, which are referred to as heritability in the broad sense, "*H*" (Nyquist 1991, p. 239), as they refer to the proportion of phenotypic variance due to total genotypic variance. Such estimators are not relevant to selection response in sexually reproducing populations, but in clonally propagated populations they are useful for predicting response to selection.

Broad-sense heritability estimators can vary, depending on the experimental design and on the selection unit, as these will impact the phenotypic variance in the denominator of the heritability function. When data are taken on individual plants within plots in a replicated multiple-environment trial with cross-classified environments, the model for phenotypic data is:

$$Y_{ijklm} = \mu + L_i + Y_j + LY_{ij} + R_{(ij)k} + G_l + GL_{il} + GY_{jl} + GLY_{ijl} + \varepsilon_{ijkl} + w_{(ijkl)m},$$

where L_i is the effect of the ith location; Y_j is the effect of the jth year; LY_{ij} is the effect of the interaction between location i and year j; GL_{il} is the effect of the interaction between genotype l and location i; GY_{jl} is the effect of the interaction between genotype l and year j; GLY_{ijl} is the effect of the interaction between genotype l, location i, and year j; and $w_{(ijkl)m}$ is the effect of the mth plant within the $ijkl$th plot. In this case, the phenotypic variance of individual plant deviations from their block mean is: $\sigma_P^2 = \sigma_G^2 + \sigma_{GL}^2 + \sigma_{GY}^2 + \sigma_{GLY}^2 + \sigma_\varepsilon^2 + \sigma_w^2$, and heritability corresponding to selection among individual plants within a block is:

$$\hat{H} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GL}^2 + \hat{\sigma}_{GY}^2 + \hat{\sigma}_{GYL}^2 + \hat{\sigma}_\varepsilon^2 + \hat{\sigma}_w^2} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_{\bar{P}}^2},$$

where σ_w^2 is the within-plot variance component (Table 2.1.1.A). If selection is based on plot mean values within one replication, the selection units are averages across the n plants per plot, leading to a heritability estimator of the form:

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GL}^2 + \hat{\sigma}_{GY}^2 + \hat{\sigma}_{GYL}^2 + \hat{\sigma}_\varepsilon^2 + \frac{\hat{\sigma}_w^2}{n}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_{\bar{P}}^2} \text{ (Table 2.1.1.B).}$$

If selection is based on family-mean values averaged across years (but from one location), the selection units are averages across n plants per plot, r replications per environment, and y years, leading to a heritability estimator of the form:

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GL}^2 + \frac{\hat{\sigma}_{GY}^2}{y} + \frac{\hat{\sigma}_{GLY}^2}{y} + \frac{\hat{\sigma}_\varepsilon^2}{yr} + \frac{\hat{\sigma}_w^2}{yrn}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_{\bar{P}}^2} \text{ (Table 2.1.1.C).}$$

If selection is based on family-mean values averaged across locations within one year, the selection units are averages across n plants per plot, r replications per environment, and l locations, leading to a heritability estimator of the form:

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \frac{\hat{\sigma}_{GL}^2}{l} + \hat{\sigma}_{GY}^2 + \frac{\hat{\sigma}_{GLY}^2}{l} + \frac{\hat{\sigma}_\varepsilon^2}{lr} + \frac{\hat{\sigma}_w^2}{lrn}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_{\bar{P}}^2} \text{ (Table 2.1.1.D).}$$

Finally, if selection is based on family-mean values averaged across all years and locations sampled, the selection units are averages across n plants per plot, r replications per environment, y years, and l locations, leading to a heritability estimator of the form:

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \frac{\hat{\sigma}_{GL}^2}{l} + \frac{\hat{\sigma}_{GY}^2}{y} + \frac{\hat{\sigma}_{GLY}^2}{ly} + \frac{\hat{\sigma}_{\varepsilon}^2}{lyr} + \frac{\hat{\sigma}_w^2}{lyrn}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_{\bar{P}}^2} \text{ (Table 2.1.1.E).}$$

If a similar evaluation is conducted with cross-classified years and locations, but data are taken on plot totals only, the model for phenotypic data becomes:

$$Y_{ijklm} = \mu + L_i + Y_j + LY_{ij} + R_{(ij)k} + G_l + GL_{il} + GY_{jl} + GLY_{ijl} + \varepsilon'_{ijkl}.$$

If plot means are calculated from the plot totals, then ε'_{ijkl} in the formula replaces

$$\varepsilon_{ijkl} + \frac{\sum_{m=1}^{n} w_{(ijkl)m}}{n}$$

in the previous formula for an individual (Section II.C). Family heritabilities on a plot basis and on a family-mean basis are similar to those just presented, with the substitution of $\sigma_{\varepsilon}^2 + (\sigma_w^2/n)$ for $\sigma_{\varepsilon'}^2$. Also, plot totals themselves can be analyzed (Table 2.1.2.A, B, C, and D; see Nyquist 1991, pp. 259–260).

If data are collected on individual plants within plots and the environments are an *independent* sample of all locations and years (not simply different, random locations within one year or different, random years within one location), or if the cross-classification is ignored (with the introduction of bias as described in Section II.C), the statistical model becomes:

$$Y_{ijkl} = \mu + E_i + R_{(i)k} + G_j + GE_{ij} + \varepsilon_{ijk} + w_{(ijk)l}.$$

The phenotypic variance of individual plant deviations from their block mean is: $\sigma_P^2 = \sigma_G^2 + \sigma_{GE}^2 + \sigma_{\varepsilon}^2 + \sigma_w^2$, and heritability corresponding to selection among individual plants within a block is:

$$\hat{H} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GE}^2 + \hat{\sigma}_{\varepsilon}^2 + \hat{\sigma}_w^2} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_P^2} \text{ (Table 2.1.3.A).}$$

Family heritability corresponding to selection among plot means within one replication is:

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GE}^2 + \hat{\sigma}_\varepsilon^2 + \frac{\hat{\sigma}_w^2}{n}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_P^2} \text{ (Table 2.1.3.B).}$$

Heritability corresponding to selection among family means averaged across all environments is:

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \frac{\hat{\sigma}_{GE}^2}{e} + \frac{\hat{\sigma}_\varepsilon^2}{er} + \frac{\hat{\sigma}_w^2}{ern}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_P^2} \text{ (Table 2.1.3.C).}$$

If data are taken only on plot totals, then the statistical model is modified by substituting ε'_{ijk} for

$$\varepsilon_{ijk} + \frac{\sum_{l=1}^{n} w_{(ijk)l}}{n},$$

and heritabilities on a plot basis and on a family-mean basis follow (Table 2.1.4.A and Table 2.1.4.B). (See the text for Table 2.1.2 for cross-classified environments instead of an independent sample of environments.)

These formulas can also be applied to the situation in which genotypes are replicated in multiple locations in a single year, by substituting σ_{GL}^2 for σ_{GE}^2 and l for e in the preceding formulas, but this results in a positive bias to the estimate of σ_G^2 and a negative bias to the estimate of genotype-by-environment interaction variance (Nyquist 1991, pp. 288–289). Similarly, these formulas can be applied when genotypes are replicated in multiple years at a single location, by substituting σ_{GY}^2 for σ_{GE}^2 and y for e in the preceding formulas, but this also results in a positive bias to the estimate of σ_G^2 and a negative bias to the estimate of genotype-by-environment interaction variance (Nyquist 1991, pp. 288–289).

B. Heritability Estimated from Half-sib Family Evaluations

Half-sib families can serve as selection units, in which case, remnant seed of selected half-sib families is often used for intermating to form a new population. In this case, the response to selection depends on the regression of random-mated offspring derived from remnant half-sib seed of two selected half-sib families on the selection units. The covariance between selection and response units on either the male or female side of the pedigree equals $(1/8)(1 + F_P)\sigma_A^2 + [(1/8)(1 + F_P)]^2\sigma_{AA}^2$. Considering

that selection occurs on both parents of the offspring, the expected response to selection is twice this value, $(1/4)(1 + F_P)\sigma^2_A + (1/32)(1 + F_P)^2\sigma^2_{AA}$, divided by the phenotypic variance of half-sib family means (Nyquist 1991, pp. 274–275).

The variance component due to half-sib families, σ^2_F, is equal to the covariance among half-sibs, $(1/4)(1 + F_P)\sigma^2_A + [(1/4)(1 + F_P)]^2\sigma^2_{AA}$ (Sections III.A and VIII.D). The ratio of the half-sib family variance component to the phenotypic variance of half-sib family means has the expectation:

$$E[\hat{h}^2_{f1}] = E\left[\frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FE}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}}\right]$$

$$\approx \frac{\frac{1}{4}(1 + F_P)\sigma^2_A + \frac{1}{16}(1 + F_P)^2\sigma^2_{AA}}{\frac{1}{4}(1 + F_P)\sigma^2_A + \frac{1}{16}(1 + F_P)^2\sigma^2_{AA} + \frac{\frac{1}{4}(1 + F_P)\sigma^2_{AE} + \frac{1}{16}(1 + F_P)^2\sigma^2_{AAE}}{e} + \frac{\sigma^2_{\varepsilon'}}{er}}.$$

In this case, the expected value of the estimator of heritability is almost equal to the true parameter h^2_{f1}, with a small upward bias of $(1/32)(1 + F_P)^2\sigma^2_{AA}$ in the numerator. Specific formulas for heritability estimates and their standard errors based on evaluation of half-sib families can be obtained by modifying the equations in Section 5 of Table 2.1.

C. Heritability Estimated from Full-sib Family Evaluations

The estimate of the ratio of the full-sib family variance component to the phenotypic variance among full-sib means was given in Section V.A, and was shown to be different from the desired heritability estimator for selection among full-sib families. The desired heritability function for selection among full-sib families can be estimated only if the additive and dominance genetic variance components can be partitioned. Such partitioning is made possible by mating design experiments, as described in Sections VIII.D and VIII.E.

D. Heritability Estimated from NC Design I

The NC Design I involves mating a sample of m plants as male parents each to a separate sample of f females, and evaluating the progenies in r replications within each of e environments. This permits the estimation of the variance components due to male parents and due to female parents nested within male parents, which have the following genetic expectations:

$$E(\hat{\sigma}^2_M) = \text{Cov(HS)} = (\tfrac{1}{4})(1 + F_P)\sigma^2_A + [(\tfrac{1}{4})(1 + F_P)]^2\sigma^2_{AA},$$

$$E(\hat{\sigma}^2_{F(M)}) = \text{Cov(FS)} - \text{Cov(HS)}$$

$$= (1 + F_P)[(\tfrac{1}{2})\sigma^2_A + (1 + F_P)(\tfrac{1}{4})\sigma^2_D + (\tfrac{1}{4})(1 + F_P)\sigma^2_{AA} - (\tfrac{1}{4})\sigma^2_A - (\tfrac{1}{16})(1 + F_P)\sigma^2_{AA}]$$

$$= (1 + F_P)[(\tfrac{1}{4})\sigma^2_A + (1 + F_P)(\tfrac{1}{4})\sigma^2_D + (\tfrac{3}{16})(1 + F_P)\sigma^2_{AA}],$$

where F_P is the inbreeding coefficient of the plants crossed to form the full- and half-sib families.

If one were interested in conducting a recurrent selection program using the data obtained from the nested design, observed means for neither independent half-sib or full-sib families exist. Instead, selection could be conducted on the basis of male group means (the mean of all full-sib families that have a common male parent) or means of nonindependent full-sib families. The heritability of male group means based on plot total data is:

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_M}{\hat{\sigma}^2_M + \dfrac{\hat{\sigma}^2_{F(M)}}{f} + \dfrac{\hat{\sigma}^2_{ME}}{e} + \dfrac{\hat{\sigma}^2_{F(M)E}}{ef} + \dfrac{\hat{\sigma}^2_{\varepsilon'}}{erf}}$$ (Table 2.1.6.D, Table 2.1.7.C).

The phenotypic variance among nonindependent full-sib family means is obtained by adding the sums of squares for males and females within males to obtain the sum of squares for nonindependent full-sib families. That sum of squares is then divided by the corresponding sum of the degrees of freedom to obtain the mean square for nonindependent full-sib families, with the following expectation:

$$E(MS_{\text{nonindep. FS}})$$

$$= \frac{(m-1)(\sigma^2_{\varepsilon'} + r\sigma^2_{F(M)E} + rf\sigma^2_{ME} + er\sigma^2_{F(M)} + erf\sigma^2_M) + m(f-1)(\sigma^2_{\varepsilon'} + r\sigma^2_{F(M)E} + er\sigma^2_{F(M)}}{(m-1) + m(f-1)}$$

$$= \frac{(m-1+mf-m)(\sigma^2_{\varepsilon'} + r\sigma^2_{F(M)E} + er\sigma^2_{F(M)}) + (m-1)(rf\sigma^2_{ME} + erf\sigma^2_M)}{m-1+mf-m}$$

$$= \frac{(mf-1)(\sigma^2_{\varepsilon'} + r\sigma^2_{F(M)E} + er\sigma^2_{F(M)}) + (m-1)(rf\sigma^2_{ME} + erf\sigma^2_M)}{mf-1}$$

$$= \sigma^2_{\varepsilon'} + r\sigma^2_{F(M)E} + er\sigma^2_{F(M)} + \frac{(m-1)}{(mf-1)}(rf\sigma^2_{ME} + erf\sigma^2_M)$$

$$= \sigma^2_{\varepsilon'} + r\sigma^2_{F(M)E} + er\sigma^2_{F(M)} + \frac{rf(m-1)}{(mf-1)}(\sigma^2_{ME} + e\sigma^2_M).$$

This shows that there is less variation among nonindependent full-sib family means because a different male parent is not mated to every female. Thus, the heritability for nonindependent full-sib family means is estimated as:

$$\hat{h}^2_{f1} = \frac{(1 + \frac{f(m-1)}{mf-1})\hat{\sigma}^2_M}{(\frac{f(m-1)}{mf-1})(\hat{\sigma}^2_M + \frac{\hat{\sigma}^2_{ME}}{e}) + \hat{\sigma}^2_{F(M)} + \frac{\hat{\sigma}^2_{F(M)E}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}} \quad \text{(Table 2.1.6.C, Table 2.1.7.B).}$$

Selection acts on the additive genetic variation in both the male and female components, but not on the dominance variation present in the female component.

Hallauer and Miranda (1988, p. 80) suggested the following heritability estimator for the Design I experiment based on noninbred parents ($F_P = 0$) (see Nyquist 1991, pp. 294–295):

$$\hat{h}^2 = \frac{4\hat{\sigma}^2_M}{4\hat{\sigma}^2_{F(M)} + \frac{4\hat{\sigma}^2_{EF(M)}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}}.$$

The expectation of the numerator of this estimator is $\sigma^2_A + (1/4)\sigma^2_{AA}$, which makes it appealing as an estimator of narrow-sense heritability, but it is incorrect as an estimator of narrow-sense heritability because the denominator is not equal to the phenotypic variance among individual plants. Therefore, it is not appropriate to predict the expected response to selection among individual plants. Furthermore, the denominator contains more than one times the genetic variance because error variance in this case contains within-family genetic variation divided by the number of plants per plot. This may contribute only a small fraction of the genetic variance if the number of plants per plot, replications, or environments is sufficiently large. This estimator cannot be interpreted in terms of response to selection among individual plants nor among half-sib or full-sib families, so we do not recommend its use (Nyquist 1991, p. 295).

E. Heritability Estimated from NC Design II

The NC Design II involves a factorial mating each of m plants as males to each of f plants as females and evaluating the mf full-sib families in each of r replications within each of e environments. The variance components due to a common male parent, a common female parent,

and the interaction between male and female parents have the following expectations:

$$E(\hat{\sigma}^2_M) = \text{Cov(HS)} = (\tfrac{1}{4})(1 + F_A)\sigma^2_A + [(\tfrac{1}{4})(1 + F_A)]^2\sigma^2_{AA}$$

$$E(\hat{\sigma}^2_F) = \text{Cov(HS)} = (\tfrac{1}{4})(1 + F_B)\sigma^2_A + [(\tfrac{1}{4})(1 + F_B)]^2\sigma^2_{AA},$$

$$E(\hat{\sigma}^2_{MF}) = \text{Cov(FS)} - 2\text{Cov(HS)} = (\tfrac{1}{4})(1 + F_A)(1 + F_B)\sigma^2_D,$$

where F_A is the inbreeding coefficient of the male plants, and F_B is the inbreeding coefficient of the female plants crossed to form the evaluated families (Cockerham 1963; Nyquist 1991, pp. 269–270). Heritability corresponding to selection among half-sib families can be estimated with either the male or female half-sib variance component, or the average of the two (Table 2.1, Sections 8 and 9). As in the case of the Design I experiment (Section VIII.D), an estimate of narrow-sense heritability is available if individual plant data are collected (Table 2.1.8.A).

Hallauer and Miranda (1988, p. 71) suggested the following heritability estimator for the Design II experiment based on noninbred parents ($F_P = 0$):

$$\hat{h}^2_f = \frac{4\sigma^2_M}{4\sigma^2_M + 4\sigma^2_{FM} + \dfrac{4\sigma^2_{ME} + 4\sigma^2_{FME}}{e} + \dfrac{\sigma^2_{\varepsilon'}}{er}}.$$

However, this estimator is not interpretable in terms of response to selection among individual plants or among families; therefore, we do not recommend its use.

F. Heritability Estimated from Testcross Progenies

Testcross progenies that are evaluated in hybrid crops represent a special case. As an example, we consider a random-mating reference population from which individual plants, families, or inbred lines are sampled, and crossed to an unrelated inbred "tester" line to form testcross progenies. The testcross progenies are evaluated phenotypically and superior progenies selected. The plants, families, or lines that were parents of those superior progenies are intermated to form an improved population. New plants, families, or lines are sampled from the improved population, and these are testcrossed to the same tester line. The gain from selection of interest is the difference between the mean of the testcrosses of the improved population and the mean of testcrosses of the original population.

We define a special genetic model to handle the genetic effects observed in testcross populations. The reference population in this case is *not* a Hardy-Weinberg random-mating population; instead, it is the nonequilibrium first generation population derived from crossing plants or lines sampled from a Hardy-Weinberg equilibrium population to a common inbred tester line. The single-locus genotypic model for the testcross population is:

$$G_{jT} = \mu_T + \alpha_j + \alpha_T + \delta_{jT},$$

where α_j is the effect of the jth allele derived from a plant in the experimental population, α_T is the effect of the allele from the inbred tester, and δ_{jT} is the dominance interaction effect of the pair of the jth allele and tester allele. Melchinger (1987) developed theory for a two-locus, two-allele model with linkage and epistasis, but for simplicity, we will ignore epistasis and assume that the single-locus model generalizes to a multilocus model simply by summing over loci. All of the testcross plants inherit one allele in common from the inbred tester line, so the genotypic model simplifies to: $G_{jT} = \mu^* + \alpha_j^*$, where $\mu^* = \mu_T + \alpha_T$, and the average effect of the jth allele from the experimental population is confounded with its dominance interaction with the tester allele: $\alpha_j^* = \alpha_j + \delta_{jT}$. The total genotypic variance in the testcross population is:

$$\sigma_G^2 = E[G_{jT} - E(G_{jT})]^2 = E[\mu^* + \alpha_j^* - \mu^*]^2 = E[\alpha_j^*]^2 = \sigma_{A(T)}^2.$$

The genetic variance is entirely due to the differences in the average effects of alleles from the experimental population in combination with the tester allele, and we term this variance $\sigma_{A(T)}^2$ to indicate that it is an additive genetic variance only in reference to this specific testcross population.

Consider now the genotypic effects segregating within the equilibrium population from which plants were sampled to cross to the inbred tester: $G_{ij} = \mu + \alpha_i + \alpha_j + \delta_{ij}$. The mean value of testcross progeny derived from a single plant in the experimental population is:

$$G_{ij}^T = (\tfrac{1}{2})(G_{iT} + G_{jT}) = (\tfrac{1}{2})[(\mu^* + \alpha_i^*) + (\mu^* + \alpha_j^*)] = \mu^* + (\tfrac{1}{2})[\alpha_i^* + \alpha_j^*].$$

The variance component due to testcross families derived from individual plants in the experimental populations is then:

$$E[\hat{\sigma}_F^2] = \mathrm{Var}(G_{ij}^T) = E[G_{ij}^T - E(G_{ij}^T)]^2$$

$$= E[\mu^* + (\tfrac{1}{2})(\alpha_i^* + \alpha_j^*) - \mu^*]^2 = (\tfrac{1}{4})E[(\alpha_i^* + \alpha_j^*)]^2.$$

The α_i^* and α_j^* effects are uncorrelated if the experimental population is in equilibrium, so this simplifies to:

$$E[\hat{\sigma}_F^2] = \text{Var}(G_{ij}^T) = (\tfrac{1}{2})[E(\alpha_i^*)^2 + E(\alpha_j^*)^2]$$

$$= (\tfrac{1}{4})(\sigma_{A(T)}^2 + \alpha_{A(T)}^2) = (\tfrac{1}{2})\sigma_{A(T)}^2.$$

The meaning of this is that the genetic component of variance among testcross families is half of the total genetic variance among testcross progeny.

We can generalize this model to include inbreeding in the experimental population, in which case the probability that the two alleles from a single individual are IBD is F. The mean of the testcross population is not affected by inbreeding in the experimental population, however. With probability F, the parent of the testcross family is inbred at an arbitrary locus, and the mean genotypic value of the testcross family at that locus is $\mu^* + \alpha_i^*$. With probability $1 - F$, the parent of the testcross is not inbred and the mean genotypic value of the testcross family at a locus is $\mu^* + (1/2)[\alpha_i^* + \alpha_j^*]$. Therefore, the variance of testcross family mean genotypic values equals:

$$E[\hat{\sigma}_F^2] = E[\text{Var}(G_{ij}^T)] = FE[\alpha_i^*]^2 + (1 - F)E[(\tfrac{1}{2})(\alpha_i^* + \alpha_j^*)]^2$$

$$= F\sigma_{A(T)}^2 + (\tfrac{1}{4})(1 - F)(2)\sigma_{A(T)}^2 = (\tfrac{1}{2})(1 + F)\sigma_{A(T)}^2.$$

If $F = 0$, this reduces to $E[\sigma_F^2] = \text{Var}(G_{ij}^T) = (1/2)\sigma_{A(T)}^2$, as already shown. If $F = 1$, $E[\sigma_F^2] = \text{Var}(G_{ij}^T) = \sigma_{A(T)}^2$.

Now consider the response to selection among testcross progeny. The selection unit is a testcross family derived from a single plant in the experimental population. The response unit of interest is the testcross family of a progeny from the mating of the selected plant (or its selfed progeny) from the experimental population to another plant from the experimental population. The genotypic value of the parent plant based on the mean of its testcross progeny (the selection unit) is:

$$G_{ij}^T = \mu^* + (\tfrac{1}{2})[\alpha_i^* + \alpha_j^*].$$

This parent plant can be inbred to an arbitrary degree, F, which is the probability that $\alpha_i = \alpha_j$, and, therefore, that $\alpha_i^* = \alpha_j^*$. Its random-mated progeny will inherit one allele from it (either i or j) with equal probability and one allele from the other parent (arbitrarily named k). Therefore, the

mean genotypic value of its random-mated progeny in testcross to the same tester (i.e., the mean genotypic value of the response unit to which it is related) is:

$$\text{mean testcross value of progeny of } G_{ij}^{T}$$
$$= \mu^* + (\tfrac{1}{4})[\alpha_i^* + \alpha_k^* + \alpha_j^* + \alpha_k^*] = \mu^* + (\tfrac{1}{4})(\alpha_i^* + \alpha_j^*) + (\tfrac{1}{2})(\alpha_k^*).$$

The covariance between testcross values of parent and offspring is:

$$\text{Cov}[G_{ij}^{T}, \text{progeny of } G_{ij}^{T}] = \text{Cov}[(\mu^* + (\tfrac{1}{2})[\alpha_i^* + \alpha_j^*]),$$

$$(\mu^* + (\tfrac{1}{4})(\alpha_i^* + \alpha_j^*) + (\tfrac{1}{2})(\alpha_k^*))]$$

$$= E[(\tfrac{1}{2})[\alpha_i^* + \alpha_j^*](\tfrac{1}{4})(\alpha_i^* + \alpha_j^*) + (\tfrac{1}{4})(\alpha_k^*)]$$

$$= (\tfrac{1}{8})E[(\alpha_i^*)(\alpha_i^*) + (\alpha_i^*)(\alpha_j^*) + (\alpha_j^*)(\alpha_i^*) + (\alpha_j^*)(\alpha_j^*)]$$
$$(\text{because } E[(\alpha_i^*)(\alpha_k^*)] = 0)$$

$$= (\tfrac{1}{8})(E[\alpha_i^{*2}] + E[\alpha_j^{*2}] + 2E[\alpha_i^*\alpha_j^*])$$

$$= (\tfrac{1}{8})\{\sigma_{A(T)}^2 + \sigma_{A(T)}^2 + 2FE[\alpha_i^{*2}]\} \text{ (because } E[\alpha_i^*\alpha_j^*] =$$
$$E[\alpha_i^{*2}] \text{ with probability } F)$$

$$= (\tfrac{1}{4})(1 + F)\sigma_{A(T)}^2.$$

If $F = 0$, then $\text{Cov}[G_{ij}^{T}, \text{progeny of } G_{ij}^{T}] = (1/4)\sigma_{A(T)}^2$.

If selection is practiced on both parents, then the response to selection involves two times the covariance between selection and response units, and this heritability is estimable as the ratio of the genotypic (testcross family) variance component to the phenotypic variance of testcross families:

$$R = \frac{\frac{1}{2}S[1 + F]\sigma_{A(T)}^2}{\frac{1}{2}[1 + F](\sigma_{A(T)}^2 + \frac{\sigma_{A(T)E}^2}{e}) + \frac{\sigma_{\varepsilon'}^2}{er}} = \frac{S\sigma_F^2}{\sigma_P^2}.$$

If inbred lines from the experimental population are used, F refers to the inbreeding coefficient of the last common parent of the inbred progeny.

Because the ratio of the family variance component to the phenotypic variance is an appropriate estimator of heritability for testcross proge-

nies, then the formulas given in Table 2.1, Sections 10 to 13, are appropriate for testcross progenies.

G. Heritability Estimated from Self-fertilized Family Evaluations

Estimating heritability in self-pollinating species poses an additional complication, because the "response unit" can vary depending on the selection scheme. If the selection units are $S_{t:g}$ families, we can envision at least five different kinds of response units in which response to selection could be measured. First, one might measure the response to selection in the remnant seed of the selected lines, that is, an independent sample of plants within the same selected $S_{t:g}$ lines. This is represented in Fig. 2.2 as individuals X_{1n+1} through X_{1n+z} whose last common ancestor was individual B, and we refer to this as immediate response to selection (Cockerham and Matzinger 1985). Second, one might measure the response in highly inbred lines derived from many generations of self-fertilization from the plants that composed the $S_{t:g}$ generation selection unit. Such lines would be in the $S_{t:\infty}$ generation and we refer to this as permanent response to selection, because the genotypic constitution of such lines is fixed across any further generations of selfing, in the absence of selection. Permanent response units are illustrated in Fig. 2.2 as individuals Y_{11} through Y_{1n} whose last common ancestor was also individual B. Third, $S_{t:g+i}$ lines derived from i additional generations of self-fertilization from the selection units could also be used as response units. These are not illustrated in Fig. 2.2, but would be intermediate between the immediate and permanent response units. Fourth, unrelated remnant seed of selected lines could be intermated to form a new base population, in which individual noninbred S_0 progeny could be used as response units. These are illustrated as individuals Z_1 through Z_n in Fig. 2.2. Finally, the response unit could be $S_{t':g'}$ lines derived from plants produced after t' generations of self-fertilization within the new base population. This could include S_0-derived lines ($t' = 0$) and inbred lines derived from highly homozygous individuals ($t' \rightarrow \infty$). Such lines are illustrated as individuals V_1 through V_n derived from a last common ancestor, W ($W = Z_2$ if $t' = 0$), in Fig. 2.2.

Each of these situations involves a different covariance between selection and response units, and consequently, a unique heritability to predict the response to selection. Cockerham and Matzinger (1985) developed response equations for selection among inbred lines as measured in their self-fertilized progeny or in outbred progeny resulting from intermating selected lines.

A commonly used estimate of heritability in self-fertilized species is based on the ratio of the genetic component of variance to the phenotypic variance of line means estimated from replicated evaluations of inbred lines (Equation [12]). Recall that the variance component due to inbred lines (families) is expected to equal the genetic covariance of any two individuals within such lines (Equation [11]). Using Cockerham's (1983) notation: $\sigma^2_F = C_{tgg'}$, where t equals the number of selfing generations from which the last common ancestor of the progeny were derived, and $g = g'$ equals the number of selfing generations from which the tested plants were derived. For example, the variance component due to $S_{0:1}$ lines is $C_{011} = \sigma^2_A + (1/4)\sigma^2_D + D_1 + (1/8)D^*_2 + \sigma^2_{AA}$, the variance component due to $S_{0:2}$ lines is $C_{022} = \sigma^2_A + (1/16)\sigma^2_D + (3/2)D_1 + (9/32)D^*_2 + \sigma^2_{AA}$, and the variance component due to $S_{1:2}$ lines is $C_{122} = (3/2)\sigma^2_A + (1/8)\sigma^2_D + (5/2)D_1 + (9/16)D^*_2 + (1/16)H^* + (9/4)\sigma^2_{AA}$ (Cockerham 1983; Nyquist 1991, p. 299). In general, and assuming free recombination between genes, $C_{tgg'}$ is given by Equation [5b]. Thus, for example, the common heritability estimator based on the variance among $S_{0:1}$ lines is expected to equal:

$$E[\hat{h}^2_{f1}] = E\left[\frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FE}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}}\right] \approx \frac{\sigma^2_A + \frac{1}{4}\sigma^2_D + D_1 + \frac{1}{8}D^*_2 + \sigma^2_{AA}}{\sigma^2_A + \frac{1}{4}\sigma^2_D + D_1 + \frac{1}{8}D^*_2 + \sigma^2_{AA} + \frac{\sigma^2_{FE}}{e} + \frac{\sigma^2_{\varepsilon'}}{er}}$$

(Nyquist 1991, Equation [86]). This is strictly correct only for predicting immediate response, that is, growing remnant seed of only the selected lines in new environments (Cockerham and Matzinger 1985; Fig. 2.2). The presence of a subscript 1 on h^2_{f1} throughout in Nyquist (1991) implied intercrossing to obtain the response unit. The symbolism of h^2 for self-fertilizing populations in Nyquist (1991) was inadequate. Here, we suggest the use of h^2_{fi} for immediate response to selection. In general, the covariance between selection unit and immediate response unit is $C_{tgg'}$ which is the same as the covariance of relatives within the line, and has the same expectation as the variance component due to lines. In this case, the covariance between selection and response units is not doubled because each response unit is related to only one selection unit.

Cockerham and Matzinger (1985) refer to response to selection measured in completely homozygous lines developed by selfing without selection from the selected lines as "permanent response" (Fig. 2.2). We suggest the use of h^2_{fp} for heritability related to permanent response to selection among self-fertilized families. The covariance between selected family means and the response unit in this case is $C_{tg\infty} = (1+F_t)\sigma^2_A + (1 + 2F_t + F_g)D_1 + (1/2)(F_t + F_g)D^*_2 + (1 + F_t)^2\sigma^2_{AA}$ (Cockerham and

Matzinger 1985). The difference between the family variance component (C_{tgg}) and the covariance between selected line means and their homozygous selfed progeny ($C_{tg\infty}$) can be easily observed in the tables given by Cockerham (1983) or Nyquist (1991, p. 299). For example, the variance among $S_{0:1}$ lines, as already given, is expected to be:

$$E(\sigma^2_{S0:1}) = C_{011} = \sigma^2_A + (\tfrac{1}{4})\sigma^2_D + D_1 + (\tfrac{1}{8})D^*_2 + \sigma^2_{AA},$$

whereas the covariance between $S_{0:1}$ family means and their $S_{0:\infty}$ progeny is expected to be:

$$C_{01\infty} = \sigma^2_A + (\tfrac{3}{2})D_1 + (\tfrac{1}{4})D^*_2 + \sigma^2_{AA}.$$

In all such cases, the additive genetic variance component is identical in the family variance component and the covariance between selection lines and progeny homozygous lines. Therefore, if the additive genetic variance is the predominant component of genetic variance in the selection populations, then the heritability estimator based on the variance component due to lines should be adequate to predict the permanent response to selection (without intermating). If dominance variance or any of the other variance or covariance components that involve dominance are important, however, then the heritability estimator based on the family variance component may inaccurately predict the permanent response to selection. The regression coefficient of response units on selection units involves the genetic component D_1, which is a covariance component, and may be negative or positive, resulting in an upward or downward bias in the heritability estimate based on the family variance component. In this case, estimating heritability appropriately may require direct estimation of the covariance between a random individual in the selection unit and a random individual in a homozygous progeny line.

As the selected lines become more inbred (being derived from more generations of selfing from the last common ancestor), the differences between the family variance component and the covariance of selection units and homozygous progeny decrease. At the extreme, if highly inbred lines are the selection units, then the variance component due to lines ($C_{t\infty\infty}$) is equal to the covariance between the selection lines and their selfed offspring ($C_{t\infty\infty}$), and the expected immediate and permanent responses to selection are equal.

Cockerham and Matzinger (1985) also developed equations for the expectations of responses to selection among inbred lines as measured in outbred progenies (Fig. 2.2). They introduced the notation of $C_{tgg'1}$ for the covariance between the selection units, which are $S_{t:g}$ lines, and

outbred progeny in the first generation developed from intermating plants from selected lines. They also introduced the notation of $C_{tgg'\infty}$ for the covariance between the selection units, which are $S_{t:g}$ lines, and outbred progeny in the equilibrium random-mated population developed from intermating plants from selected lines: $C_{tgg'\infty} = (1/2)[(1 + F_t)\sigma^2_A + (F_t + F_g)D_1]$. The two terms differ only by the inclusion of an epistatic variance component in the response measured in the first randomly mated generation: $C_{tgg'1} = C_{tgg'\infty} + (1/4)(1 + F_t)^2\sigma^2_{AA}$ (see Section V.A). This covariance is doubled in the heritability function that will give the correct expected response to selection among $S_{t:g}$ lines as measured in outbred progeny because selection is practiced on both lines giving rise to the outbred progeny:

$$\hat{h}^2_{f1} = \frac{2\hat{C}_{tgg'\infty}}{\hat{\sigma}^2_{\bar{P}}} \approx \frac{(1 + F_t)\hat{\sigma}^2_A + (F_t + F_g)\hat{D}_1}{C_{tgg} + \dfrac{C_{tgg}E}{e} + \dfrac{\hat{\sigma}^2_{\varepsilon'}}{er}}.$$

Note that, again, the coefficient of the additive variance in the numerator of this heritability estimator is equal to the coefficient of the additive variance in the covariance of relatives within the line. Thus, if the genetic variance for the trait is predominantly additive, the heritability estimator based on the ratio of the variance component due to lines to the phenotypic variance of line means should be appropriate.

Finally, what heritability function is appropriate for selection among $S_{t:g}$ lines developed from the initial population, followed by intermating the lines to form a new random-mated population, then deriving $S_{t':g'}$ lines from the new population and measuring the response in these inbred lines (Fig. 2.2)? The relevant covariance between selection and response units for this situation has not been published to our knowledge, but we can at least state that the additive portion of the covariance will be equal to the additive portion of $C_{tgg'\infty}$ as given previously.

In summary, in self-pollinated species, a commonly used estimator of heritability is the ratio of the variance component due to inbred lines to the phenotypic variance of inbred line means (Table 2.1, Sections 10 to 13). This estimator is exact only for immediate response to selection, that is, the growing of remnant seed of selected lines in new (independent) environments. This estimator does not exactly provide the expected response to permanent selection (response as measured in highly homozygous progeny developed by self-pollination without selection from the selected lines) unless the selection units are already highly homozygous lines, or unless the genetic variance is completely additive in nature. Nor does this estimator provide the expected response to selection measured

in outbred progenies developed from intermating selected lines, unless the genetic variance is completely additive. The difficulty that this situation presents is that the heritability function that is easily estimated may not be useful for predicting response to selection in progeny types of interest, whereas the appropriate heritability estimators for outbred progeny cannot be easily obtained except by specialized and complex mating designs or by direct estimation of the relevant covariance. We suggest that the scope of inference for such heritability estimators be limited to immediate response unless sufficient evidence exists to broaden its scope of inference to permanent response or response in outbred progenies.

H. Heritability Corresponding to Selection among Self-fertilized Half-sib and Full-sib Families

Burton and Carver (1993) derived expectations for response to selection on the basis of self-fertilized progeny of half-sib and full-sib families (HS-S_1 and FS-S_1 families). Including an additive-by-additive variance component, the response to selection among HS-S_1 families is expected to be:

$$R = \frac{S\left(\frac{7}{16}\sigma_A^2 + \frac{7}{32}D_1 + \frac{49}{512}\sigma_{AA}^2\right)}{\sigma_P^2}.$$

The response to selection among FS-S_1 families is expected to be:

$$R = \frac{S(\frac{7}{8}\sigma_A^2 + \frac{7}{16}D_1 + \frac{49}{128}\sigma_{AA}^2)}{\sigma_P^2}.$$

Based on estimates of family variance components and phenotypic variances of HS-S_1 and $S_{0:1}$ families in soybean (*Glycine max*) and FS-S_1 and $S_{0:1}$ families in wheat, Burton and Carver (1993) suggested that selection among FS-S_1 families would provide optimum response to selection, despite the larger coefficient of additive genetic variance in the numerator of the expected response to selection among $S_{0:1}$ families. The reason for this was that more seed could be produced of FS-S_1 families, permitting larger plot sizes and more replication of FS-S_1 than $S_{0:1}$ families, resulting in a reduction of the phenotypic variance among family means that more than compensated for the reduction in genetic variance in the numerator of the heritability equation. Holland et al. (2000) used FS-S_1 families for recurrent selection in oat in order to obtain sufficient seed to conduct replicated trials with three replications at each of five locations and still conduct one cycle of selection per year.

Table 2.1. Heritability estimators and their biases,[†] and elements of λ_G and λ_P vectors used in estimating variances of heritability estimates for various experimental and mating designs.[‡] In each case, the variance components (without coefficients) in the denominator of the heritability estimate are the components of the vector σ (described in Section V.C.1), with the first variance component in the denominator being the first row of σ, the second variance component in the denominator being the second row of σ, and so forth.

1. **Broad-sense heritability estimated from data obtained on *individual plants* of a clonally propagated population from multiple environment evaluations in cross-classified locations and years (Section VIII.A). The estimators are unbiased.**

   A. Heritability on an individual-plant basis, giving the response to selection among individual plants within one replication of one environment as measured in independent environments (see Nyquist 1991, p. 267).

$$\hat{H} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GL}^2 + \hat{\sigma}_{GY}^2 + \hat{\sigma}_{GLY}^2 + \hat{\sigma}_\varepsilon^2 + \hat{\sigma}_w^2} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_P^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & 1 \end{bmatrix}$$

   B. Family heritability on a plot basis, giving the response to selection among plot means within one replication of one environment as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GL}^2 + \hat{\sigma}_{GY}^2 + \hat{\sigma}_{GLY}^2 + \hat{\sigma}_\varepsilon^2 + \frac{\hat{\sigma}_w^2}{n}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_P^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & \frac{1}{n} \end{bmatrix}$$

   C. Heritability of family means, giving the response to selection among family means averaged across years within one location as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GL}^2 + \frac{\hat{\sigma}_{GY}^2}{y} + \frac{\hat{\sigma}_{GLY}^2}{y} + \frac{\hat{\sigma}_\varepsilon^2}{yr} + \frac{\hat{\sigma}_w^2}{yrn}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_P^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & \frac{1}{y} & \frac{1}{y} & \frac{1}{yr} & \frac{1}{yrn} \end{bmatrix}$$

   D. Heritability of family means, giving the response to selection among family means averaged across locations within one year as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \frac{\hat{\sigma}_{GL}^2}{l} + \hat{\sigma}_{GY}^2 + \frac{\hat{\sigma}_{GLY}^2}{l} + \frac{\hat{\sigma}_\varepsilon^2}{lr} + \frac{\hat{\sigma}_w^2}{lrn}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_P^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & \frac{1}{l} & 1 & \frac{1}{l} & \frac{1}{lr} & \frac{1}{lrn} \end{bmatrix}$$

E. Heritability of family means, giving the response to selection among family means averaged across locations and years as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_G + \frac{\hat{\sigma}^2_{GL}}{l} + \frac{\hat{\sigma}^2_{GY}}{y} + \frac{\hat{\sigma}^2_{GLY}}{ly} + \frac{\hat{\sigma}^2_{\varepsilon}}{lyr} + \frac{\hat{\sigma}^2_{w}}{lyrn}} = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_{\bar{P}} = \begin{bmatrix} 1 & \frac{1}{l} & \frac{1}{y} & \frac{1}{ly} & \frac{1}{lyr} & \frac{1}{lyrn} \end{bmatrix}$$

2. Broad-sense heritability estimated from data obtained on *total plot values* of multiple-plant plots of a clonally propagated population from multiple environment evaluations in cross-classified locations and years (Section VIII.A). The estimators are unbiased.

A. Family heritability on a plot basis, giving the response to selection among plot values within one replication of one environment as measured in independent environments (see Nyquist 1991, pp. 267–268).

$$\hat{H}_f = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_G + \hat{\sigma}^2_{GL} + \hat{\sigma}^2_{GY} + \hat{\sigma}^2_{GLY} + \hat{\sigma}^2_{\varepsilon'}} = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_P} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 \end{bmatrix}$$

B. Heritability of family means, giving the response to selection among family means averaged across years within one location as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_G + \hat{\sigma}^2_{GL} + \frac{\hat{\sigma}^2_{GY}}{y} + \frac{\hat{\sigma}^2_{GLY}}{y} + \frac{\hat{\sigma}^2_{\varepsilon'}}{yr}} = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & \frac{1}{y} & \frac{1}{y} & \frac{1}{yr} \end{bmatrix}$$

C. Heritability of family means, giving the response to selection among family means averaged across locations within one year as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_G + \frac{\hat{\sigma}^2_{GL}}{l} + \hat{\sigma}^2_{GY} + \frac{\hat{\sigma}^2_{GLY}}{l} + \frac{\hat{\sigma}^2_{\varepsilon'}}{lr}} = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{l} & 1 & \frac{1}{l} & \frac{1}{lr} \end{bmatrix}$$

D. Heritability of family means, giving the response to selection among family means averaged across locations and years as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \frac{\hat{\sigma}_{GL}^2}{l} + \frac{\hat{\sigma}_{GY}^2}{y} + \frac{\hat{\sigma}_{GLY}^2}{ly} + \frac{\hat{\sigma}_{\varepsilon'}^2}{lyr}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_{\bar{P}}^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & \frac{1}{l} & \frac{1}{y} & \frac{1}{ly} & \frac{1}{lyr} \end{bmatrix}$$

3. Broad-sense heritability estimated from data obtained on *individual plants* of a clonally propagated population from multiple independent environment evaluations (Section VIII.A). The estimators are unbiased.

A. Heritability on an individual-plant basis, giving the response to selection among individual plants within one replication of one environment as measured in independent environments.

$$\hat{H} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GE}^2 + \hat{\sigma}_\varepsilon^2 + \hat{\sigma}_w^2} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_P^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 \end{bmatrix}$$

B. Family heritability on a plot basis, giving the response to selection among plot means within one replication of one environment as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \hat{\sigma}_{GE}^2 + \hat{\sigma}_\varepsilon^2 + \frac{\hat{\sigma}_w^2}{n}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_{\bar{P}}^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & \frac{1}{n} \end{bmatrix}$$

C. Heritability of family means, giving the response to selection among family means averaged across environments as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_G^2 + \frac{\hat{\sigma}_{GE}^2}{e} + \frac{\hat{\sigma}_\varepsilon^2}{er} + \frac{\hat{\sigma}_w^2}{ern}} = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_{\bar{P}}^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & \frac{1}{e} & \frac{1}{er} & \frac{1}{ern} \end{bmatrix}$$

4. Broad-sense heritability estimated from data obtained on *total plot values* of multiple-plant plots of a clonally propagated population from multiple independent environment evaluations (Section VIII.A). The estimators are unbiased.

A. Family heritability on a plot basis, giving the response to selection among plot values within one replication of one environment as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_G + \hat{\sigma}^2_{GE} + \hat{\sigma}^2_{\varepsilon'}} = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_P} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & 1 \end{bmatrix}$$

B. Heritability of family means, giving the response to selection among family means averaged across environments as measured in independent environments.

$$\hat{H}_f = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_G + \frac{\hat{\sigma}^2_{GE}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}} = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{e} & \frac{1}{er} \end{bmatrix}$$

5. Narrow-sense heritability estimated from a half-sib family experiment with data obtained on *individual plants* in multiple independent environments (Section VIII.B).

A. Heritability on an individual-plant basis, giving the response to selection among individual plants within one replication of one environment as measured in outbred progeny grown in independent environments.

$$\hat{h}^2_1 = \frac{\left(\frac{4}{(1+F_P)}\right)\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \hat{\sigma}^2_{FE} + \hat{\sigma}^2_\varepsilon + \hat{\sigma}^2_w} = \frac{\hat{\sigma}^2_A + \left(\frac{(1+F_P)}{4}\right)\hat{\sigma}^2_{AA}}{\hat{\sigma}^2_P}$$

$$\text{Bias} = \frac{\frac{1}{4}(F_P - 1)\sigma^2_{AA}}{\sigma^2_P} \qquad \lambda^T_G = \begin{bmatrix} \frac{4}{1+F_P} & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & 1 & 1 \end{bmatrix}$$

B. Family heritability on a plot basis (half-sib family, single-plot mean value), giving the response to selection among plot means within one replication of one environment as measured in outbred progeny grown in independent environments.

$$\hat{h}^2_{f1} = \frac{\sigma^2_F}{\sigma^2_F + \sigma^2_{FE} + \sigma^2_\varepsilon + \frac{\sigma^2_w}{n}} = \frac{\frac{1}{4}(1+F_P)\hat{\sigma}^2_A + \frac{1}{16}(1+F_P)^2\hat{\sigma}^2_{AA}}{\hat{\sigma}^2_{\bar{P}}}$$

$$\text{Bias} = \frac{\frac{1}{32}(1+F_P)^2\sigma^2_{AA}}{\sigma^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_G = \begin{bmatrix} 1 & 1 & 1 & \frac{1}{n} \end{bmatrix}$$

C. Family heritability, giving the response to selection among half-sib family means averaged across environments as measured in outbred progeny grown in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FE}}{e} + \frac{\hat{\sigma}^2_\varepsilon}{er} + \frac{\hat{\sigma}^2_w}{ern}} = \frac{\frac{1}{4}(1+F_P)\hat{\sigma}^2_A + \frac{1}{16}(1+F_P)^2\hat{\sigma}^2_{AA}}{\sigma^2_{\bar{P}}}$$

$$\text{Bias} = \frac{\frac{1}{32}(1+F_P)^2\sigma^2_{AA}}{\sigma^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{e} & \frac{1}{er} & \frac{1}{ern} \end{bmatrix}$$

6. Narrow-sense heritability estimated from a Design I experiment with data obtained on *individual plants* in multiple independent environment evaluations (Section VIII.D).

A. Heritability on an individual-plant basis, giving the response to selection among individual plants within one replication of one environment as measured in outbred progeny grown in independent environments.

$$\hat{h}_1^2 = \frac{\left(\frac{4}{(1+F_P)}\right)\hat{\sigma}_M^2}{\hat{\sigma}_M^2 + \hat{\sigma}_{F(M)}^2 + \hat{\sigma}_{ME}^2 + \hat{\sigma}_{F(M)E}^2 + \hat{\sigma}_\varepsilon^2 + \hat{\sigma}_w^2} = \frac{\hat{\sigma}_A^2 + \left(\frac{(1+F_P)}{4}\right)\hat{\sigma}_{AA}^2}{\hat{\sigma}_P^2}$$

$$\text{Bias} = \frac{\frac{1}{4}(F_P - 1)\sigma_{AA}^2}{\sigma_P^2} \qquad \lambda_G^T = \left[\frac{4}{1+F_P} \quad 0 \quad 0 \quad 0 \quad 0 \quad 0\right] \qquad \lambda_P^T = \left[1 \quad 1 \quad 1 \quad 1 \quad 1 \quad 1\right]$$

B. Family heritability on a plot basis (nonindependent full-sib family, single-plot mean value), giving the response to selection among plot means within one replication of one environment as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{(1 + \frac{f(m-1)}{mf-1})\hat{\sigma}_M^2}{\frac{f(m-1)}{mf-1}(\hat{\sigma}_M^2 + \hat{\sigma}_{ME}^2) + \hat{\sigma}_{F(M)}^2 + \hat{\sigma}_{F(M)E}^2 + \hat{\sigma}_\varepsilon^2 + \frac{\hat{\sigma}_w^2}{n}} = \frac{[1 + \frac{f(m-1)}{mf-1}][\frac{1}{4}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{16}(1+F_P)^2\hat{\sigma}_{AA}^2]}{\hat{\sigma}_{\bar{P}}^2}$$

$$\lambda_G^T = \left[1 + \frac{f(m-1)}{mf-1} \quad 0 \quad 0 \quad 0 \quad 0 \quad 0\right] \qquad \lambda_P^T = \left[\frac{f(m-1)}{mf-1} \quad \frac{f(m-1)}{mf-1} \quad 1 \quad 1 \quad 1 \quad \frac{1}{n}\right]$$

C. Family heritability, giving the response to selection among nonindependent full-sib family means averaged across environments as measured in outbred progeny in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{(1 + \frac{f(m-1)}{mf-1})\hat{\sigma}_M^2}{\frac{f(m-1)}{mf-1}(\hat{\sigma}_M^2 + \frac{\hat{\sigma}_{ME}^2}{e}) + \hat{\sigma}_{F(M)}^2 + \frac{\hat{\sigma}_{F(M)E}^2}{e} + \frac{\hat{\sigma}_\varepsilon^2}{er} + \frac{\hat{\sigma}_\varepsilon^2}{ern}} = \frac{[1 + \frac{f(m-1)}{mf-1}][\frac{1}{4}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{16}(1+F_P)^2\hat{\sigma}_{AA}^2]}{\hat{\sigma}_{\bar{P}}^2}$$

$$\lambda_G^T = \left[1 + \frac{f(m-1)}{mf-1} \quad 0 \quad 0 \quad 0 \quad 0 \quad 0\right] \qquad \lambda_P^T = \left[\frac{f(m-1)}{mf-1} \quad \frac{f(m-1)}{e(mf-1)} \quad 1 \quad \frac{1}{e} \quad \frac{1}{er} \quad \frac{1}{ern}\right]$$

D. Family heritability, giving the response to selection among male group means averaged across environments as measured in outbred progeny grown in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_M}{\hat{\sigma}^2_M + \frac{\hat{\sigma}^2_{F(M)}}{f} + \frac{\hat{\sigma}^2_{ME}}{e} + \frac{\hat{\sigma}^2_{F(M)E}}{ef} + \frac{\hat{\sigma}^2_{\varepsilon}}{erf} + \frac{\hat{\sigma}^2_{w}}{erfn}} = \frac{\frac{1}{4}(1+F_P)\hat{\sigma}^2_A + \frac{1}{16}(1+F_P)^2\hat{\sigma}^2_{AA}}{\hat{\sigma}^2_P}$$

$$\text{Bias} = \frac{\frac{1}{32}(1+F_P)^2\sigma^2_{AA}}{\sigma^2_P} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{f} & \frac{1}{e} & \frac{1}{ef} & \frac{1}{erf} & \frac{1}{erfn} \end{bmatrix}$$

7. Narrow-sense heritability estimated from a Design I experiment with data obtained on *total plot values* of multiple-plant plots in multiple independent environment evaluations (Section VIII.D).

A. Family heritability on a plot basis (nonindependent full-sib family, single-plot value), giving the response to selection among plot values within one replication of one environment as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h^2_{f1}).

$$\hat{h}^2_{f1} = \frac{(1+\frac{f(m-1)}{mf-1})\hat{\sigma}^2_M}{\frac{f(m-1)}{mf-1}(\hat{\sigma}^2_M + \hat{\sigma}^2_{ME}) + \hat{\sigma}^2_{F(M)} + \hat{\sigma}^2_{F(M)E} + \hat{\sigma}^2_{\varepsilon'}} = \frac{[1+\frac{f(m-1)}{mf-1}][\frac{1}{4}(1+F_P)\hat{\sigma}^2_A + \frac{1}{16}(1+F_P)^2\hat{\sigma}^2_{AA}]}{\hat{\sigma}^2_P}$$

$$\lambda^T_G = \begin{bmatrix} 1+\frac{f(m-1)}{mf-1} & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} \frac{f(m-1)}{mf-1} & \frac{f(m-1)}{mf-1} & 1 & 1 & 1 \end{bmatrix}$$

B. Family heritability, giving the response to selection among nonindependent full-sib family means averaged across environments as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h^2_{f1}).

$$\hat{h}^2_{f1} = \frac{(1+\frac{f(m-1)}{mf-1})\hat{\sigma}^2_M}{\frac{f(m-1)}{mf-1}(\hat{\sigma}^2_M + \frac{\hat{\sigma}^2_{ME}}{e}) + \hat{\sigma}^2_{F(M)} + \frac{\hat{\sigma}^2_{F(M)E}}{e} + \frac{\hat{\sigma}^2_{\varepsilon'}}{er}} = \frac{[1+\frac{f(m-1)}{mf-1}][\frac{1}{4}(1+F_P)\hat{\sigma}^2_A + \frac{1}{16}(1+F_P)^2\hat{\sigma}^2_{AA}]}{\hat{\sigma}^2_{\bar{P}}}$$

$$\lambda^T_G = \left[1+\frac{f(m-1)}{mf-1} \quad 0 \quad 0 \quad 0 \quad 0\right] \qquad \lambda^T_P = \left[\frac{f(m-1)}{mf-1} \quad \frac{f(m-1)}{e(mf-1)} \quad 1 \quad \frac{1}{e} \quad \frac{1}{er}\right]$$

C. Family heritability, giving the response to selection among male group means averaged across environments as measured in outbred progeny grown in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_M}{\hat{\sigma}^2_M + \frac{\hat{\sigma}^2_{F(M)}}{f} + \frac{\hat{\sigma}^2_{ME}}{e} + \frac{\hat{\sigma}^2_{F(M)E}}{ef} + \frac{\hat{\sigma}^2_{\varepsilon'}}{erf}} = \frac{\frac{1}{4}(1+F_P)\hat{\sigma}^2_A + \frac{1}{16}(1+F_P)^2\hat{\sigma}^2_{AA}}{\hat{\sigma}^2_{\bar{P}}}$$

$$\text{Bias} = \frac{\frac{1}{32}(1+F_P)^2\sigma^2_{AA}}{\sigma^2_{\bar{P}}} \qquad \lambda^T_G = \left[1 \quad 0 \quad 0 \quad 0 \quad 0\right] \qquad \lambda^T_P = \left[1 \quad \frac{1}{f} \quad \frac{1}{e} \quad \frac{1}{ef} \quad \frac{1}{erf}\right]$$

8. Narrow-sense heritability estimated from a Design II experiment with data obtained on *individual plants* in multiple independent environment evaluations (Section VIII.E).

A. Heritability on an individual-plant basis, giving the response to selection among individual plants within one replication of one environment as measured in outbred progeny grown in independent environments.

$$\hat{h}^2_1 = \frac{\frac{4}{(1+F_P)}\hat{\sigma}^2_M}{\hat{\sigma}^2_M + \hat{\sigma}^2_F + \hat{\sigma}^2_{MF} + \hat{\sigma}^2_{ME} + \hat{\sigma}^2_{FE} + \hat{\sigma}^2_{MFE} + \hat{\sigma}^2_\varepsilon + \hat{\sigma}^2_w} = \frac{\hat{\sigma}^2_A + \left(\frac{1+F_P}{4}\right)\hat{\sigma}^2_{AA}}{\hat{\sigma}^2_P}$$

$$\text{Bias} = \frac{\frac{1}{4}(F_P-1)\sigma^2_{AA}}{\sigma^2_P} \qquad \lambda^T_G = \left[\frac{4}{1+F_P} \quad 0 \quad 0 \quad 0 \quad 0 \quad 0 \quad 0 \quad 0\right] \qquad \lambda^T_P = \left[1 \quad 1 \quad 1 \quad 1 \quad 1 \quad 1 \quad 1 \quad 1\right]$$

B. Family heritability on a plot basis (full-sib family, single-plot mean value), giving the response to selection among plot means within one replication of one environment as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{2\hat{\sigma}_M^2}{\hat{\sigma}_M^2 + \hat{\sigma}_F^2 + \hat{\sigma}_{MF}^2 + \hat{\sigma}_{ME}^2 + \hat{\sigma}_{FE}^2 + \hat{\sigma}_{MFE}^2 + \hat{\sigma}_\varepsilon^2 + \frac{\hat{\sigma}_w^2}{n}} = \frac{\frac{1}{2}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{8}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_P^2}$$

$$\lambda_G^T = \begin{bmatrix} 2 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & 1 & 1 & \frac{1}{n} \end{bmatrix}$$

C. Alternative estimate of family heritability on a plot basis (full-sib family, single-plot mean value), giving the response to selection among plot means within one replication of one environment as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_M^2 + \hat{\sigma}_F^2}{\hat{\sigma}_M^2 + \hat{\sigma}_F^2 + \hat{\sigma}_{MF}^2 + \hat{\sigma}_{ME}^2 + \hat{\sigma}_{FE}^2 + \hat{\sigma}_{MFE}^2 + \hat{\sigma}_\varepsilon^2 + \frac{\hat{\sigma}_w^2}{n}} = \frac{\frac{1}{2}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{8}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_P^2}$$

$$\lambda_G^T = \begin{bmatrix} 1 & 1 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & 1 & 1 & \frac{1}{n} \end{bmatrix}$$

D. Family heritability, giving the response to selection among full-sib family means averaged across environments as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{2\hat{\sigma}_M^2}{\hat{\sigma}_M^2 + \hat{\sigma}_F^2 + \hat{\sigma}_{MF}^2 + \frac{\hat{\sigma}_{ME}^2}{e} + \frac{\hat{\sigma}_{FE}^2}{e} + \frac{\hat{\sigma}_{MFE}^2}{e} + \frac{\hat{\sigma}_\varepsilon^2}{er} + \frac{\hat{\sigma}_w^2}{ern}} = \frac{\frac{1}{2}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{8}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_P^2}$$

$$\lambda_G^T = \begin{bmatrix} 2 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & \frac{1}{e} & \frac{1}{e} & \frac{1}{e} & \frac{1}{er} & \frac{1}{ern} \end{bmatrix}$$

E. Alternative estimate of family heritability, giving the response to selection among full-sib family means averaged across environments as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h^2_{f1}).

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_M + \hat{\sigma}^2_F}{\hat{\sigma}^2_M + \hat{\sigma}^2_F + \hat{\sigma}^2_{MF} + \frac{\hat{\sigma}^2_{ME}}{e} + \frac{\hat{\sigma}^2_{FE}}{e} + \frac{\hat{\sigma}^2_{MFE}}{e} + \frac{\hat{\sigma}^2_{\varepsilon}}{er} + \frac{\hat{\sigma}^2_{w}}{ern}} = \frac{\frac{1}{2}(1+F_P)\hat{\sigma}^2_A + \frac{1}{8}(1+F_P)^2\hat{\sigma}^2_{AA}}{\hat{\sigma}^2_{\bar{P}}}$$

$$\lambda^T_G = \begin{bmatrix} 1 & 1 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & 1 & \frac{1}{e} & \frac{1}{e} & \frac{1}{e} & \frac{1}{er} & \frac{1}{ern} \end{bmatrix}$$

F. Family heritability, giving the response to selection among paternal half-sib family means averaged across environments as measured in outbred progeny grown in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_M}{\hat{\sigma}^2_M + \frac{\hat{\sigma}^2_F}{f} + \frac{\hat{\sigma}^2_{MF}}{f} + \frac{\hat{\sigma}^2_{ME}}{e} + \frac{\hat{\sigma}^2_{FE}}{ef} + \frac{\hat{\sigma}^2_{MFE}}{ef} + \frac{\hat{\sigma}^2_{\varepsilon}}{erf} + \frac{\hat{\sigma}^2_{w}}{erfn}} = \frac{\frac{1}{4}(1+F_P)\hat{\sigma}^2_A + \frac{1}{16}(1+F_P)^2\hat{\sigma}^2_{AA}}{\hat{\sigma}^2_{\bar{P}}}$$

$$\text{Bias} = \frac{\frac{1}{32}(1+F_P)^2\sigma^2_{AA}}{\sigma^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{f} & \frac{1}{f} & \frac{1}{e} & \frac{1}{ef} & \frac{1}{ef} & \frac{1}{erf} & \frac{1}{erfn} \end{bmatrix}$$

G. Family heritability, giving the response to selection among maternal half-sib family means averaged across environments as measured in outbred progeny grown in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_M}{m} + \frac{\hat{\sigma}^2_{MF}}{m} + \frac{\hat{\sigma}^2_{FE}}{e} + \frac{\hat{\sigma}^2_{ME}}{me} + \frac{\hat{\sigma}^2_{MFE}}{me} + \frac{\hat{\sigma}^2_{\varepsilon}}{mer} + \frac{\hat{\sigma}^2_{w}}{mern}} = \frac{\frac{1}{4}(1+F_P)\hat{\sigma}^2_A + \frac{1}{16}(1+F_P)^2\hat{\sigma}^2_{AA}}{\hat{\sigma}^2_{\bar{P}}}$$

$$\text{Bias} = \frac{\frac{1}{32}(1+F_P)^2\sigma^2_{AA}}{\sigma^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{m} & \frac{1}{m} & \frac{1}{e} & \frac{1}{me} & \frac{1}{me} & \frac{1}{mer} & \frac{1}{mern} \end{bmatrix}$$

9. Narrow-sense heritability estimated from a Design II experiment with data obtained on *total plot values* of multiple-plant plots in multiple independent environment evaluations (Section VIII.E).

A. Family heritability on a plot basis (full-sib family, single-plot value), giving the response to selection among plot values within one replication of one environment as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{2\hat{\sigma}_M^2}{\hat{\sigma}_M^2 + \hat{\sigma}_F^2 + \hat{\sigma}_{MF}^2 + \hat{\sigma}_{ME}^2 + \hat{\sigma}_{FE}^2 + \hat{\sigma}_{MFE}^2 + \hat{\sigma}_{\varepsilon'}^2} = \frac{\frac{1}{2}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{8}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_P^2}$$

$$\lambda_G^T = \begin{bmatrix} 2 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & 1 & 1 \end{bmatrix}$$

B. Alternative estimate of family heritability on a plot basis (full-sib family, single-plot value), giving the response to selection among plot values within one replication of one environment as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_M^2 + \hat{\sigma}_F^2}{\hat{\sigma}_M^2 + \hat{\sigma}_F^2 + \hat{\sigma}_{MF}^2 + \hat{\sigma}_{ME}^2 + \hat{\sigma}_{FE}^2 + \hat{\sigma}_{MFE}^2 + \hat{\sigma}_{\varepsilon'}^2} = \frac{\frac{1}{2}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{8}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_P^2}$$

$$\lambda_G^T = \begin{bmatrix} 1 & 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & 1 & 1 \end{bmatrix}$$

C. Family heritability, giving the response to selection among full-sib family means averaged across environments as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{2\hat{\sigma}_M^2}{\hat{\sigma}_M^2 + \hat{\sigma}_F^2 + \hat{\sigma}_{MF}^2 + \frac{\hat{\sigma}_{ME}^2}{e} + \frac{\hat{\sigma}_{FE}^2}{e} + \frac{\hat{\sigma}_{MFE}^2}{e} + \frac{\hat{\sigma}_{\varepsilon'}^2}{er}} = \frac{\frac{1}{2}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{8}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_{\bar{P}}^2}$$

$$\lambda_G^T = \begin{bmatrix} 2 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & \frac{1}{e} & \frac{1}{e} & \frac{1}{e} & \frac{1}{er} \end{bmatrix}$$

D. Alternative estimate of family heritability, giving the response to selection among full-sib family means averaged across environments as measured in outbred progeny grown in independent environments (the estimator is unbiased relative to h_{f1}^2).

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_M^2 + \hat{\sigma}_F^2}{\hat{\sigma}_M^2 + \hat{\sigma}_F^2 + \hat{\sigma}_{MF}^2 + \frac{\hat{\sigma}_{ME}^2}{e} + \frac{\hat{\sigma}_{FE}^2}{e} + \frac{\hat{\sigma}_{MFE}^2}{e} + \frac{\hat{\sigma}_{\varepsilon'}^2}{er}} = \frac{\frac{1}{2}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{8}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_{\bar{P}}^2}$$

$$\lambda_G^T = \begin{bmatrix} 1 & 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & \frac{1}{e} & \frac{1}{e} & \frac{1}{e} & \frac{1}{er} \end{bmatrix}$$

E. Family heritability, giving the response to selection among paternal half-sib family means averaged across environments as measured in outbred progeny grown in independent environments.

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_M^2}{\hat{\sigma}_M^2 + \frac{\hat{\sigma}_F^2}{f} + \frac{\hat{\sigma}_{MF}^2}{f} + \frac{\hat{\sigma}_{ME}^2}{e} + \frac{\hat{\sigma}_{FE}^2}{ef} + \frac{\hat{\sigma}_{MFE}^2}{ef} + \frac{\hat{\sigma}_{\varepsilon'}^2}{erf}} = \frac{\frac{1}{4}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{16}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_{\bar{P}}^2}$$

$$\text{Bias} = \frac{\frac{1}{32}(1+F_P)^2\sigma_{AA}^2}{\sigma_{\bar{P}}^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & \frac{1}{f} & \frac{1}{f} & \frac{1}{e} & \frac{1}{ef} & \frac{1}{ef} & \frac{1}{erf} \end{bmatrix}$$

F. Family heritability, giving the response to selection among maternal half-sib family means averaged across environments as measured in outbred progeny grown in independent environments.

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \frac{\hat{\sigma}_M^2}{m} + \frac{\hat{\sigma}_{MF}^2}{m} + \frac{\hat{\sigma}_{FE}^2}{e} + \frac{\hat{\sigma}_{ME}^2}{em} + \frac{\hat{\sigma}_{MFE}^2}{em} + \frac{\hat{\sigma}_{\varepsilon'}^2}{erm}} = \frac{\frac{1}{4}(1+F_P)\hat{\sigma}_A^2 + \frac{1}{16}(1+F_P)^2\hat{\sigma}_{AA}^2}{\hat{\sigma}_{\bar{P}}^2}$$

$$\text{Bias} = \frac{\frac{1}{32}(1+F_P)^2\sigma_{AA}^2}{\sigma_{\bar{P}}^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & \frac{1}{m} & \frac{1}{m} & \frac{1}{e} & \frac{1}{em} & \frac{1}{em} & \frac{1}{erm} \end{bmatrix}$$

10. Narrow-sense heritability estimated from data obtained on *individual plants* from multiple environment evaluations of families in cross-classified locations and years, giving the response to selection among testcross progenies (Section VIII.F) or "immediate response" to selection among inbred families (Section VIII.G) as measured in independent environments. The estimators are unbiased relative to h_1^2 for these situations, but are biased when applied to selection among inbred families when response units are outbred or evaluated at a different level of inbreeding than the selection units (Section VIII.G).

A. Heritability on an individual-plant basis, giving the response to selection among individual plants within one replication as measured in independent environments.

$$\hat{h}_1^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \hat{\sigma}_{FL}^2 + \hat{\sigma}_{FY}^2 + \hat{\sigma}_{FLY}^2 + \hat{\sigma}_\varepsilon^2 + \hat{\sigma}_w^2} = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_P^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & 1 \end{bmatrix}$$

B. Family heritability on a plot basis, giving the response to selection among plot means within one replication of one environment as measured in independent environments.

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \hat{\sigma}_{FL}^2 + \hat{\sigma}_{FY}^2 + \hat{\sigma}_{FLY}^2 + \hat{\sigma}_\varepsilon^2 + \dfrac{\hat{\sigma}_w^2}{n}} = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_{\bar{P}}^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 & \dfrac{1}{n} \end{bmatrix}$$

C. Heritability of family means, giving the response to selection among family means averaged across years within one location as measured in independent environments.

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \hat{\sigma}_{FL}^2 + \dfrac{\hat{\sigma}_{FY}^2}{y} + \dfrac{\hat{\sigma}_{FLY}^2}{y} + \dfrac{\hat{\sigma}_\varepsilon^2}{yr} + \dfrac{\hat{\sigma}_w^2}{yrn}} = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_{\bar{P}}^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & \dfrac{1}{y} & \dfrac{1}{y} & \dfrac{1}{yr} & \dfrac{1}{yrn} \end{bmatrix}$$

D. Heritability of family means, giving the response to selection among family means averaged across locations within one year as measured in independent environments.

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \dfrac{\hat{\sigma}_{FL}^2}{l} + \hat{\sigma}_{FY}^2 + \dfrac{\hat{\sigma}_{FLY}^2}{l} + \dfrac{\hat{\sigma}_\varepsilon^2}{lr} + \dfrac{\hat{\sigma}_w^2}{lrn}} = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_{\bar{P}}^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & \dfrac{1}{l} & 1 & \dfrac{1}{l} & \dfrac{1}{lr} & \dfrac{1}{lrn} \end{bmatrix}$$

E. Heritability of family means, giving the response to selection among family means averaged across locations and years as measured in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FL}}{l} + \frac{\hat{\sigma}^2_{FY}}{y} + \frac{\hat{\sigma}^2_{FLY}}{ly} + \frac{\hat{\sigma}^2_{\varepsilon}}{lyr} + \frac{\hat{\sigma}^2_{w}}{lyrn}} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_P} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{l} & \frac{1}{y} & \frac{1}{ly} & \frac{1}{lyr} & \frac{1}{lyrn} \end{bmatrix}$$

11. Narrow-sense heritability estimated from data obtained on *total plot values* from multiple environment evaluations of families in cross-classified locations and years, giving the response to selection among testcross progenies (Section VIII.F) or "immediate response" to selection among inbred families (Section VIII.G) as measured in independent environments. The estimators are unbiased relative to h^2_1 for these situations, but are biased when applied to selection among inbred families when response units are outbred or evaluated at a different level of inbreeding than the selection units (Section VIII.G).

A. Family heritability on a plot basis, giving the response to selection among plot values within one replication of one environment as measured in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \hat{\sigma}^2_{FL} + \hat{\sigma}^2_{FY} + \hat{\sigma}^2_{FLY} + \hat{\sigma}^2_{\varepsilon'}} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_P} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & 1 & 1 & 1 \end{bmatrix}$$

B. Heritability of family means, giving the response to selection among family means averaged across years within one location as measured in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \hat{\sigma}^2_{FL} + \frac{\hat{\sigma}^2_{FY}}{y} + \frac{\hat{\sigma}^2_{FLY}}{y} + \frac{\hat{\sigma}^2_{\varepsilon'}}{yr}} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_P} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & \frac{1}{y} & \frac{1}{y} & \frac{1}{yr} \end{bmatrix}$$

C. Heritability of family means, giving the response to selection among family means averaged across locations within one year as measured in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FL}}{l} + \hat{\sigma}^2_{FY} + \frac{\hat{\sigma}^2_{FLY}}{l} + \frac{\hat{\sigma}^2_{\varepsilon'}}{lr}} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_P} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{l} & 1 & \frac{1}{l} & \frac{1}{lr} \end{bmatrix}$$

D. Heritability of family means, giving the response to selection among family means averaged across locations and years as measured in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FL}}{l} + \frac{\hat{\sigma}^2_{FY}}{y} + \frac{\hat{\sigma}^2_{FLY}}{ly} + \frac{\hat{\sigma}^2_{\epsilon'}}{lyr}} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_{\bar{P}}} \quad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \end{bmatrix} \quad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{l} & \frac{1}{y} & \frac{1}{ly} & \frac{1}{lyr} \end{bmatrix}$$

12. Narrow-sense heritability estimated from data obtained on *individual plants* from multiple independent environment evaluations of families, giving the response to selection among testcross progenies (Section VIII.F) or "immediate response" to selection among inbred families (Section VIII.G) as measured in independent environments. The estimators are unbiased relative to h^2_1 for these situations, but are biased when applied to selection among inbred families when response units are outbred or evaluated at a different level of inbreeding than the selection units (Section VIII.G).

A. Heritability on an individual-plant basis, giving the response to selection among individual plants within one replication of one environment as measured in independent environments.

$$\hat{h}^2_1 = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \hat{\sigma}^2_{FE} + \hat{\sigma}^2_\epsilon + \hat{\sigma}^2_w} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_P} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & 1 & 1 \end{bmatrix}$$

B. Family heritability on a plot basis, giving the response to selection among plot means within one replication of one environment as measured in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \hat{\sigma}^2_{FE} + \hat{\sigma}^2_\epsilon + \frac{\hat{\sigma}^2_w}{n}} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & 1 & 1 & \frac{1}{n} \end{bmatrix}$$

C. Heritability of family means, giving the response to selection among family means averaged across environments as measured in independent environments.

$$\hat{h}^2_{f1} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_F + \frac{\hat{\sigma}^2_{FE}}{e} + \frac{\hat{\sigma}^2_\epsilon}{er} + \frac{\hat{\sigma}^2_w}{ern}} = \frac{\hat{\sigma}^2_F}{\hat{\sigma}^2_{\bar{P}}} \qquad \lambda^T_G = \begin{bmatrix} 1 & 0 & 0 & 0 \end{bmatrix} \qquad \lambda^T_P = \begin{bmatrix} 1 & \frac{1}{e} & \frac{1}{er} & \frac{1}{ern} \end{bmatrix}$$

13. Narrow-sense heritability estimated from data obtained on *total plot values* from multiple independent environment evaluations of families, giving the response to selection among testcross progenies (Section VIII.F) or "immediate response" to selection

among inbred families (Section VIII.G) as measured in independent environments. The estimators are unbiased relative to h_1^2 for these situations, but are biased when applied to selection among inbred families when response units are outbred or evaluated at a different level of inbreeding than the selection units (Section VIII.G).

A. Family heritability on a plot basis, giving the response to selection among plot means within one replication of one environment as measured in independent environments.

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \hat{\sigma}_{FE}^2 + \hat{\sigma}_{\epsilon'}^2} = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_P^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & 1 & 1 \end{bmatrix}$$

B. Heritability of family means, giving the response to selection among family means averaged across environments as measured in independent environments.

$$\hat{h}_{f1}^2 = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_F^2 + \frac{\hat{\sigma}_{FE}^2}{e} + \frac{\hat{\sigma}_{\epsilon'}^2}{er}} = \frac{\hat{\sigma}_F^2}{\hat{\sigma}_P^2} \qquad \lambda_G^T = \begin{bmatrix} 1 & 0 & 0 \end{bmatrix} \qquad \lambda_P^T = \begin{bmatrix} 1 & \frac{1}{e} & \frac{1}{er} \end{bmatrix}$$

---

†Bias measured as $E(\hat{h}_1^2) - h_1^2$ or $E(\hat{h}_{f1}^2) - h_{f1}^2$, where h_1^2 or h_{f1}^2 is the regression of response units (members of the first generation of the next base population) on selection units. The bias will differ if the response unit is a gametic phase equilibrium population resulting from many generations of random mating (Nyquist 1991, pp. 250–251).

‡σ_G^2 is the genotypic variance component, σ_ϵ^2 is the experimental error variance component, $\sigma_{\epsilon'}^2$ is the experimental error variance when data are not available on individual plants within plots, component σ_w^2 is the plant-to-plant-within-plot variance component, σ_{GL}^2 is the genotype-by-location interaction variance component, σ_{GY}^2 is the genotype-by-year interaction variance component, σ_{GLY}^2 is the genotype-by-location-by-year interaction variance component, σ_{GE}^2 is the genotype-by-environment interaction variance component, σ_M^2 is the male (or half-sib family) variance component, $\sigma_{F(M)}^2$ is the female nested within male variance component, σ_A^2 is the additive genetic variance component, σ_{ME}^2 is the male-by-environment interaction variance component, $\sigma_{F(M)E}^2$ is the female-within-male-by-environment interaction variance component, σ_F^2 is the family variance component (or the female variance component in Design II experiments), σ_{MF}^2 is the male-by-female interaction variance component, σ_{FE}^2 is the family-by-environment interaction variance component (or female-by-environment interaction variance component in Design II experiments), σ_{MFE}^2 is the male-by-female-by-environment interaction variance component, σ_{FL}^2 is the family-by-location interaction variance component, σ_{FY}^2 is the family-by-year interaction variance component, and σ_{FLY}^2 is the family-by-location-by-year interaction variance component, r is the number of replications per environment, e is the number of environments, l is the number of locations, y is the number of years, f is the number of female parents mated to each male parent in Designs I and II, m is the number of male parents mated to each female parent in Design II and the total number of male parents in Design I, and F_P is the inbreeding coefficient of parents used in the mating design.

APPENDICES

SAS CODE FOR ESTIMATING HERITABILITY WITH REML

The SAS codes presented in the appendices and sample data sets for which these examples were written are freely available on the Internet at www4.ncsu.edu/~jholland/heritability.html. The symbol H is used for h_f^2 in the codes due to limitations on typeset.

Appendix 1. Estimating Heritability from Multiple Environments, One Replication per Environment

SAS version 8.0 code for estimating heritability and its standard for a trait measured on one replication at multiple environments is given here. As an example, we assumed that family means were based on data from six independent environments. The equation for family heritability on a plot basis from Table 2.1.13.A and the equation for family heritability on a mean basis from Table 2.1.13.B were used. Heritability on a family-mean basis was approximated by setting $e = 6$ and $r = 1$ in this example.

```
proc mixed asycov; class env geno; model trait = ; random
  env geno;
ods listing exclude asycov covparm; ods output asycov =
  covmat covparms = estmat;
proc iml;
start seh(V, C, LG, LP, H, SE);
Vp = LP`*V;
Vg = LG`*V;
H = Vg/Vp;
d = (1/Vp)*(LG - (LP*H));
VH = d`*C*d;
SE = sqrt(VH);
finish seh;

use estmat; read all into v; use covmat; read all into c;
* Note that SAS introduces an extra first column into the
  matrix which must be removed;
C = C(|1:nrow(C), 2:ncol(C)|);
*order of variance components in v and c matrices is σ²E,
  σ²G, residual (=σ²GE);
LG = {0, 1, 0};
LP = {0, 1, 1};
```

```
call seh(V, C, LG, LP, H, SE);
print "Heritability on a Plot Basis", H, SE;
e = 6;
LP = 0//1//(1/e);
call seh(V, C, LG, LP, H, SE);
print "Heritability on a Family-Mean Basis", H, SE;
quit; run;
```

Appendix 2. Estimating Heritability from Multiple Environments, Several Replications per Environment

SAS version 8.0 code for estimating heritability and its standard error for a trait measured on several replications at each of multiple environments is given here. As an example, we assumed that family means were based on data from three replications within each of six independent environments. The equation for family heritability on a plot basis from Table 2.1.13.A and the equation for family heritability on a mean basis from Table 2.1.13.B were used. Heritability on a family-mean basis was approximated by setting $e = 6$ and $r = 3$ in this example.

```
proc mixed asycov; class env rep geno; model trait = ;
  random env rep(env) geno env*geno;
ods listing exclude asycov covparm; ods output asycov =
  covmat covparms = estmat;
proc iml;
start seh(V, C, LG, LP, H, SE);
Vp = LP`*V;
Vg = LG`*V;
H = Vg/Vp;
d = (1/Vp)*(LG - (LP*H));
VH = d`*C*d;
SE = sqrt(VH);
finish seh;

use estmat; read all into v; use covmat; read all into c;
* Note that SAS introduces an extra first column into the
  C matrix which must be removed;
C = C(|1:nrow(C), 2:ncol(C)|);
*order of variance components in v and c matrices is σ²E,
  σ²R, σ²G, σ²GE, residual;
LG = {0, 0, 1, 0, 0};
LP = {0, 0, 1, 1, 1};
call seh(V, C, LG, LP, H, SE);
print "Heritability on a Plot Basis", H, SE;
```

```
r = 3;
e = 6;
LP = 0//0//(1/e)//(1/(e*r));
call seh(V, C, LG, LP, H, SE);
print "Heritability on a Family-Mean Basis", H, SE;
quit; run;
```

Appendix 3. Estimating Heritability in Multiple Populations Grown in a Common Experiment

SAS version 8.0 code for estimating heritability and its standard error within two different populations for a trait measured on several replications at each of multiple environments is given here. As an example, the treatment design consists of equal number of genotypes from each population randomly assigned to different sets, and the experimental design is a replications-within-sets layout replicated three times within each of four independent environments. The equation for family heritability on a plot basis from Table 2.1.13.A and the equation for family heritability on a mean basis from Table 2.1.13.B were used. Heritability on a family-mean basis was approximated by setting $e = 4$ and $r = 3$ in this example. Unique family and family-by-environment interaction variances are estimated for the two populations, but a common error variance is assumed.

```
proc mixed asycov; classes env rep set geno pop;
model trait = pop;
random env set rep(env*set) pop*env set*env;
random geno(set) env*geno(set)/group = pop;
lsmeans pop/pdiff;
ods output asycov = covmat covparms = estmat;
run;


proc iml;
start seh(V,C,LG,LP,H,SE);
Vp = LP`*V;
Vg = Lg`*V;
H = Vg/Vp;
d = (1/Vp)*(LG - (Lp*H));
VH = d`*C*d;
SE = sqrt(VH);
finish seh;
```

```
use estmat; read all into v;use covmat; read all into c;
*Note that SAS introduces an extra first column into the C
  matrix which must be removed;
C = C(|1:nrow(C), 2:ncol(C)|);

*order of variance components in V and C matrices is σ²Env,
  σ²Set, σ²Rep, σ²Env*Pop, σ²Env*Set, σ²Geno(Set)Pop1, σ²Geno(Set)Pop2,
  σ²Env*Geno(Set)Pop1, σ²Env*Geno(Set)Pop2, σ²Error.;

*get heritability for first population;

*LG and LP vectors for Population 1;
LG = {0,0,0,0,0,1,0,0,0,0};
LP = {0,0,0,0,0,1,0,1,0,1};

call seh(V,C,LG,LP,H,SE);
print "Heritability on a Plot Basis - Population 1", H,
  SE;
e = 4;
r = 3;
LP = 0//0//0//0//0//1//0//(1/e)//0//(1/(e*r));print LP;
call seh(V,C,LG,LP,H,SE);
print "Heritability on a Family-Mean Basis - Population
  1", H, SE;

*LG and LP vectors for Population 2;
LG = {0,0,0,0,0,0,1,0,0,0};
LP = {0,0,0,0,0,0,1,0,1,1};

call seh(V,C,LG,LP,H,SE);
print "Heritability on a Plot Basis - Population 2", H,
  SE;
e = 4;
r = 3;
LP = 0//0//0//0//0//0//1//0//(1/e)//(1/(e*r));
call seh(V,C,LG,LP,H,SE);
print "Heritability on a Family-Mean Basis - Population
  2", H, SE;
quit;
run;
```

Appendix 4. Estimating Heritability via Parent-Offspring Regression and from Replicated Family Evaluations

SAS version 8.0 code for estimating heritability and its standard error for a trait measured on parental and offspring genotypes in different environments is given here. This code was used to analyze the following data set: disease ratings were made on 140 F_2 (S_0) parents (individual plants) in Florida, 1991; disease ratings were made on two replications of 24-plant row plots of all 140 self-fertilized progeny ($F_{2:3}$ or $S_{0:1}$) in North Carolina, 1991; disease ratings were made on two replications of 24-plant row plots of a selected group of 52 $S_{0:1}$ progeny in North Carolina 1994. Holland et al. (1998) used these data to estimate heritability from regression of 140 $F_{2:3}$ means from the North Carolina, 1991, environment; from the ANOVA of $S_{0:1}$ families grown in North Carolina, 1991, environment (biased by family-by-environment interactions in numerator); and they estimated repeatability unbiased by FE in the numerator using the selected set of 52 $S_{0:1}$ lines grown in both North Carolina, 1991 and 1994, environments. Using mixed models approaches, a single estimator of heritability based on the variance of all 140 $S_{0:1}$ families can be obtained, including the data from some families in 1994 to remove FE bias from the numerator. The equation for family heritability on a plot basis from Table 2.1.13.A and the equation for family heritability on a mean basis from Table 2.1.13.B were used. Heritability on a family-mean basis was approximated by setting *e* equal to the harmonic mean of the number of environments in which each family was evaluated (1.23) and setting *er* equal to the harmonic mean of the number of plots in which each family was evaluated (2.42). In the same analysis, the parent-offspring regression analysis can be performed to obtain the heritability estimator with individual S_0 plants as the phenotypic variance using Equation [21]. Before implementing the mixed models analysis, ANOVAs were conducted to provide initial estimates of the variance and covariance parameters to promote faster computation and convergence on the maximum likelihood estimates. Initial parameter estimates are specified in the "parms" statement in PROC MIXED. Multiple random statements in PROC MIXED are used to permit modeling the **G** matrix effects separately for different variance components. Variables coding for the environment ("env"), the block within environment ("rep"), the generation ("gen" = "F2" or "F3"), and the family number code are associated with each observation.

```
proc mixed asycov; class env rep family gen;
model rust = ;
*the macroenvironment and block within environment effects
  are treated as random variables each with a single
  variance component;
```

```
random env rep(env) ;
*unique family - by - environment interaction variances
  are modeled for the two different generations by using
  the group option;
random family*env/group = gen;
*unique family variances are modeled for the two different
  generations, and the covariance between families with
  the same code is modeled by using the subject option and
  specifying an unstructured covariance matrix and
  specifying that each family has the same variance-
  covariance structure;
random gen/subject=family type = un;
*unique error variances are modeled for the two different
  generations with the repeated command and group option;
repeated /group = gen;
* initial values of variance-covariance parameters based
  on the preliminary ANOVAs are introduced with the parms
  command - in the order that effects are specified in the
  random statements (σ²E, σ²R(E), σ²FE(S0), σ²FE(S1), σ²F(S0), σG(S0,S1),
  σ²F(S1), σ²e(S0), σ²e(S1)). The variance components for GE and
  Error within the F2 generation (σ²FE(S0) σ²e(S1)) are forced
  to be zero with the hold option - this is necessary
  because only one variance component is estimable in the
  F2 generation, as data were taken on individual plants,
  so the component σ²F(S0) is actually the phenotypic
  variance in the S0 generation;
parms (0.1451) (0.0057) (0) (0.146) (4.274) (2.5741)
  (3.0086) (0) (0.7515)/ hold=3,8;
ods listing exclude asycov covparm; ods output asycov =
  covmat covparms = estmat;

proc iml;
start seh(V, C, LG, LP, H, SE);
Vp = LP`*V;
Vg = LG`*V;
H = Vg/Vp;
d = (1/Vp)*(LG - (LP*H));
VH = d`*C*d;
SE = sqrt(VH);
finish seh;

use estmat; read all into v; use covmat; read all into c;
* Note that SAS introduces an extra first column into the
  matrix which must be removed;
C = C(|1:nrow(C), 2:ncol(C)|);
*Note carefully the order of variance components in v and
  c matrices:σ²E, σ²R(E), σ²FE(S0), σ²FE(S1), σ²F(S0), σG(S0,S1), σ²F(S1),
```

```
  σ²e(S0), σ²e(S1). The vector of variance components estimates
  that is of interest for estimating heritability based on
  S0:1 line variances includes only σ²F(S1), σ²FE(S1), and
  σ²e(S1); these are the 7th, 4th, and 9th components of the V
  and C matrices, respectively;
v = v(|7|)//v(|4|)//v(|9|)
c = c(|{7 4 9}, {7 4 9}|);
LG = {1, 0, 0};
LP = {1, 1, 1};
call seh(V, C, LG, LP, H, SE);
print "Heritability on a Plot Basis", H, SE;

*the harmonic mean of the number of plots per S0:1 family
  is 2.42 and the number of environments in which each
  family was tested is 1.23;
eh = 1.23;
ph = 2.42;
lp = 1//(1/eh)//(1/ph);
call seh(V, C, LG, LP, H, SE);
print "Heritability on a Family-Mean Basis", H, SE;

*now create a new pair of v and c matrices to estimate
  heritability from parent offspring regression. In this
  case the variance components of interest are the 6th and
  5th, respectively: σG(S0,S1) and σ²F(S0);
use estmat; read all into v; use covmat; read all into c;
v = v(|6|)//v(|5|);
C = C(|1:nrow(C), 2:ncol(C)|);
c = c(|{6 5}, {6 5}|);
LG = {1, 0};
LP = {1, 1};
call seh(V, C, LG, LP, H, SE);
print "Heritability from regression of S1 offspring on
  individual parents", H, SE;
quit;
run;
```

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Errata for Table 2.6 from Holland, J. B. 2001. Epistatsis and Plant Breeding, in **Plant Breeding Reviews,** *Volume 21, edited by Jules Janick. John Wiley & Sons, New York.*

Table 2.6. Statistical genetic parameters of the two-locus, two-allele model, assuming Hardy-Weinberg and gametic phase equilibria.

Model for genotypic value: $G_{ijkl} = \mu_{....} + \alpha_i^A + \alpha_j^A + \alpha_k^B + \alpha_l^B + \delta_{ij}^A + \delta_{kl}^B + \alpha\alpha_{ik} + \alpha\alpha_{il} + \alpha\alpha_{jk} + \alpha\alpha_{jl} + \alpha\delta_{ikl} + \alpha\delta_{jkl} + \delta\alpha_{ijk} + \delta\alpha_{ijl} + \delta\delta_{ijkl}$

Restrictions of genotypic model:

$$\sum_{i=1}^{2} p_i\alpha_i^A = 0, \sum_{k=1}^{2} q_k\alpha_k^B = 0$$

$$\sum_{i=1}^{2}\sum_{j=1}^{2} p_ip_j\delta_{ij}^A = 0, \sum_{k=1}^{2}\sum_{l=1}^{2} q_kq_l\delta_{kl}^B = 0, \sum_{i=1}^{2} p_i\delta_{ij}^A = 0, \sum_{k=1}^{2} q_k\delta_{kl}^B = 0$$

$$\sum_{i=1}^{2}\sum_{k=1}^{2} p_iq_k\alpha\alpha_{ik} = 0, \sum_{k=1}^{2} q_k\alpha\alpha_{ik} = 0, \sum_{i=1}^{2} p_i\alpha\alpha_{ik} = 0$$

$$\sum_{i=1}^{2}\sum_{k=1}^{2}\sum_{l=1}^{2} p_iq_kq_l\alpha\delta_{ikl} = 0, \sum_{k=1}^{2}\sum_{l=1}^{2} q_kq_l\alpha\delta_{ikl} = 0, \sum_{i=1}^{2} p_i\alpha\delta_{ikl} = 0$$

$$\sum_{i=1}^{2}\sum_{j=1}^{2}\sum_{k=1}^{2} p_ip_jq_k\delta\alpha_{ijk} = 0, \sum_{i=1}^{2}\sum_{j=1}^{2} p_ip_j\delta\alpha_{ijk} = 0, \sum_{k=1}^{2} q_k\delta\alpha_{ijk} = 0$$

$$\sum_{i=1}^{2}\sum_{j=1}^{2}\sum_{k=1}^{2}\sum_{l=1}^{2} p_ip_jq_kq_l\delta\delta_{ijkl} = 0, \sum_{j=1}^{2}\sum_{k=1}^{2}\sum_{l=1}^{2} p_jq_kq_l\delta\delta_{ijkl} = 0, \sum_{i=1}^{2}\sum_{j=1}^{2}\sum_{k=1}^{2} p_ip_jq_k\delta\delta_{ijkl} = 0,$$

$$\sum_{i=1}^{2}\sum_{j=1}^{2} p_ip_j\delta\delta_{ijkl} = 0, \sum_{k=1}^{2}\sum_{l=1}^{2} q_kq_l\delta\delta_{ijkl} = 0, \sum_{i=1}^{2}\sum_{k=1}^{2} p_iq_k\delta\delta_{ijkl} = 0, \sum_{i=1}^{2} p_i\delta\delta_{ijkl} = 0, \sum_{k=1}^{2} q_k\delta\delta_{ijkl} = 0$$