Effects of Differential Selection in the Sexes on Cytonuclear Dynamics: Life Stages With Sex Differences

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ABSTRACT

We extend our investigation of cytonuclear selection by determining when differential selection between the sexes will generate allele frequency changes or cytonuclear disequilibria in populations with constant viability selection and an adult census. We demonstrate analytically that there can be a cytonuclear hitchhiking effect upon a selectively neutral marker in either sex provided the other marker is selected in that sex and there is allelic disequilibrium between the loci in females. Cytonuclear disequilibria are generated *de novo* in both sexes when both loci affect fitness in females and there is a nonmultiplicative fitness interaction between them. Similar fitness interactions in males generate male disequilibria only. Through numerical analyses, we investigate the potential magnitude of such disequilibria, their qualitative dynamics, the expected frequency of detectable disequilibria under particular patterns or strengths of selection, and the possible disequilibrium sign patterns resulting from selection. These adult/viability results subsume those for populations with a gamete census and either constant fertility or viability selection. Although previous work suggests that the disequilibria generated by cytonuclear selection may be difficult to detect experimentally, this study shows that cytonuclear disequilibria at life stages with sex differences can be useful markers of the presence and strength of selection.

THE joint analysis of nuclear and cytoplasmic markers and the statistical associations (cytonuclear disequilibria) between them are proving very useful for delineating evolutionary processes that may be undetectable through nuclear data alone. The many applications to date demonstrate how cytonuclear assays can facilitate the detection and estimation of such key factors as nonrandom mating, admixture, gene flow, population subdivision, mutation, and genetic drift in diploid plant and animal populations (Lamb and Avise 1986; Asmussen *et al.* 1987, 1989; Arnold *et al.* 1988; Asmussen and Schnabel 1991; Asmussen and Arnold 1991; Forbes and Allendorf 1991; Fu and Arnold 1991, 1992; Schnabel and Asmussen 1992; Cruzan and Arnold 1993, 1994; Scribner and Avise 1993, 1994a,b; Abernethy 1994; Datta *et al.* 1996a,b; Sites *et al.* 1996; Avise *et al.* 1997; Harrison and Bogdanowicz 1997). In addition, some basic hybrid zone frameworks for using cytonuclear data from haplodiploids or *X*-linked markers in diploids to estimate assortative mating by the pure parental species and differential gene flow through a hybrid zone by direction or sex (Goodisman and Asmussen 1997; Goodisman *et al.* 1998) now exist.

The proper interpretation of cytonuclear data also requires a better understanding of the cytonuclear effects of natural selection because of the growing experimental evidence of detectable selective interactions between the functionally interdependent nuclear and cytoplasmic genomes (Edwardson 1970; Hiraizumi 1985; Clark and Lyckegaard 1988; MacRae and Anderson 1988; Fos *et al.* 1990; Hutter and Rand 1995; Kilpatrick and Rand 1995; Saavedra *et al.* 1996). Initial theoretical studies of the effects of selection on joint cytonuclear genotypes focused on the conditions necessary to maintain joint, nuclear-cytoplasmic polymorphisms (Watson and Caspari 1960; Caspari *et al.* 1966; Clark 1984; Gregorius and Ross 1984; Ross and Gregorius 1985). They concluded that these conditions were very restrictive because simple cytonuclear selection models with random mating and constant viabilities or fertilities cannot maintain cytoplasmic variation (Clark 1984; Gregorius and Ross 1984). Indeed, these early studies identified only one set of circumstances with the demonstrated potential to preserve cytonuclear polymorphism under constant selection: asymmetrical fertility differences between the sexes in mixed-mating populations practicing a mixture of selfing and random outcrossing (Gregorius and Ross 1984).

Motivated by the need to explain recent experimental reports of both dramatic dynamics and polymorphic equilibria for mtDNA frequencies (MacRae and Anderson 1988; Fos *et al.* 1990; Hutter and Rand 1995; Kilpatrick and Rand 1995), we (Babcock and Asmussen 1996) recently readdressed this issue. Our analysis revealed that constant selection can generate both permanent cytonuclear polymorphisms and disequilibria

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in random mating populations provided viability or fertility differences are sufficiently strong both within and between the sexes. In investigating the exact cytonuclear dynamics and disequilibria, which are model dependent, our previous study focused on the three mathematically equivalent models censused at life stages without frequency differences between the sexes. These include constant fertility selection with a zygote or adult census and constant viability selection with a zygote census, which we shall collectively refer to as the "zygote/fertility models."

As a first step toward understanding the expected allele frequency dynamics under joint cytonuclear selection, we derived the precise analytic conditions under which selection will generate cytoplasmic frequency changes or a cytonuclear hitchhiking effect in these systems. We also determined that relatively small, but potentially detectable levels of cytonuclear disequilibria can be generated *de novo* in the zygote/fertility models if the cytoplasmic and nuclear loci each affect female fitness and the two markers have a nonmultiplicative fitness interaction in females (Babcock and Asmussen 1996). However, these disequilibria will usually be only weak markers of cytonuclear selection because they tend to be of fairly short duration and exhibit the same transient sign patterns as those between two neutral markers.

In this article, we extend our analysis of constant cytonuclear selection to the three remaining models in which there *are* sex differences at the time of census. These include viability selection with an adult or gamete census and fertility selection with a gamete census. We primarily focus here on the "adult/viability model," because it conveniently subsumes the two gamete-census models, which are themselves mathematically equivalent. These new models are much more complex than those considered in our original study because of the need to separately monitor the dynamics within each sex, but they also promise to be of greater practical importance, because now the life stage censused provides a more direct reflection of the effects of differential selection. After first delimiting when cytonuclear selection will generate sex-specific allele frequency changes or *de novo* disequilibria in an adult/viability system, we provide detailed numerical investigations of the magnitude, duration, and sign patterns of the resulting disequilibrium trajectories. We also explore whether detectable disequilibria can be correlated with either particular patterns or the strength of selection.

ANALYTIC STUDY

We want to determine the basic effects of constant cytonuclear selection at life stages with frequency differences between the sexes. We assume a diploid, random mating population with discrete, nonoverlapping generations, an autosomal nuclear locus exhibiting Mendelian segregation, a cytoplasmic locus with strict maternal

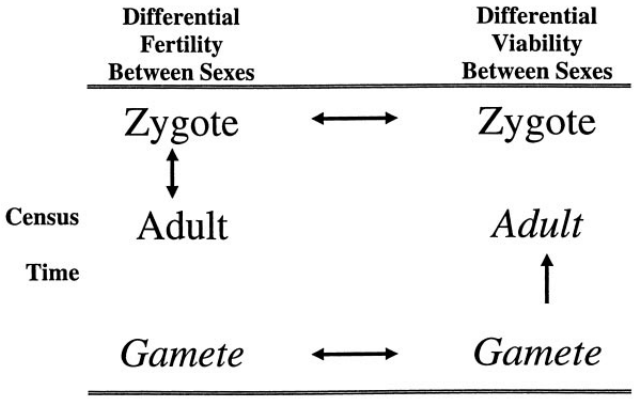

Figure 1.—Six models of differential selection according to life stage censused and selection component analyzed. Double-headed arrows connect mathematically equivalent selection models, while the single-headed arrow indicates that the gamete models are a subset of the adult/viability model. Italics indicate the three models discussed in the text.

inheritance, and no genetic drift, mutation, or gene flow. In the following sections we provide a detailed analysis of differential, constant viability selection between the sexes, with censusing at the adult stage. Later, in the discussion, we indicate how the adult/viability results subsume those for the two gamete-census models, with either differential viabilities or fertilities. Figure 1 provides a schematic summary of the relationships among the six models considered here and in our previous study (Babcock and Asmussen 1996).

Cytonuclear variables: As in our previous models, we assume the population has two alleles, A and a, at the diploid nuclear marker and two cytotypes, C and c, at the haploid cytoplasmic marker. The frequencies of the six possible joint cytonuclear genotypes in adult females are denoted in Table 1, with the column sums providing the marginal frequencies of the three nuclear genotypes (U^f, V^f, W^f) and the row sums providing the marginal frequencies of the two cytotypes (X^f, Y^f). From these, the marginal nuclear allele frequencies in females can be calculated as

$$P^f = U^f + \tfrac{1}{2}V^f = \text{freq}(A)$$

and

$$Q^f = W^f + \tfrac{1}{2}V^f = \text{freq}(a) = 1 - P^f, \qquad (1)$$

where for these and all other female variables freq denotes the frequency in adult females. The joint and marginal frequencies in adult males are analogous to those in Table 1 and (1), with each superscript f replaced by m.

The remaining frequency variables are those within the two gamete pools. Female gametes carry the four possible cytonuclear combinations, in the frequencies specified by Table 2. With only viability selection, the female gametic frequencies are simply the frequencies of the four joint allelic combinations in adult females

TABLE 1

Genotypic frequencies in adults

	Nuclear genotype			
Cytotype	*AA*	*Aa*	*aa*	Total
C	$U_1^f = U^f X^f + D_1^f$	$V_1^f = V^f X^f + D_2^f$	$W_1^f = W^f X^f + D_3^f$	X^f
c	$U_2^f = U^f Y^f - D_1^f$	$V_2^f = V^f Y^f - D_2^f$	$W_2^f = W^f Y^f - D_3^f$	Y^f
Total	U^f	V^f	W^f	1.0

Frequencies are given for females. Male frequencies are analogous with each superscript f replaced by m.

and are calculated from the joint genotypic frequencies (Table 1) as

$$P_i^f = U_i^f + \tfrac{1}{2}V_i^f, \quad Q_i^f = W_i^f + \tfrac{1}{2}V_i^f \quad \text{for } i = 1, 2. \tag{2}$$

Because males only transmit the nuclear marker, the male gamete pool is fully described by its nuclear allele frequencies, which in the absence of fertility and gametic selection are the same as those in adult males (P^m, Q^m).

In the adult/viability model, eight disequilibrium statistics are needed to fully describe the departures of cytonuclear frequencies from expectations under random associations, because disequilibria must be measured separately in the adults of each sex. Of these, six are *genotypic disequilibria*, which measure nonrandom associations between the two cytotypes and each of the three nuclear genotypes within each sex. The three female genotypic disequilibria are defined as

$$\begin{aligned} D_1^f &= U_1^f - U^f X^f = \text{freq}(AA/C) - \text{freq}(AA)\,\text{freq}(C) \\ D_2^f &= V_1^f - V^f X^f = \text{freq}(Aa/C) - \text{freq}(Aa)\,\text{freq}(C) \\ D_3^f &= W_1^f - W^f X^f = \text{freq}(aa/C) - \text{freq}(aa)\,\text{freq}(C). \end{aligned} \tag{3}$$

The male genotypic disequilibria are analogous to those in (3) with each f replaced by m and freq now the frequency in adult males. There are also two measures of *allelic disequilibrium*,

$$D^f = U_1^f + \tfrac{1}{2}V_1^f - P^f X^f \tag{4a}$$

$$D^m = U_1^m + \tfrac{1}{2}V_1^m - P^m X^m, \tag{4b}$$

TABLE 2

Female gametic frequencies

	Nuclear allele		
Cytotype	*A*	*a*	Total
C	$P_1^f = P^f X^f + D^f$	$Q_1^f = Q^f X^f - D^f$	X^f
c	$P_2^f = P^f Y^f - D^f$	$Q_2^f = Q^f Y^f + D^f$	Y^f
Total	P^f	Q^f	1.0

defined in each sex as freq (A/C) − freq(A)freq(C), which similarly measure the association between cytoplasmic and nuclear alleles within adult females and males, respectively.

The four disequilibria within each sex are related by the formulas

$$D_1^* + D_2^* + D_3^* = 0, \quad D^* = D_1^* + \tfrac{1}{2}D_2^*, \tag{5}$$

where * is f for females and m for males. Although this reduces the number of independent disequilibrium measures to two in each sex, we analyze all eight because their joint sign patterns can be useful markers of selection. The sex-specific genotypic frequencies and the female gametic frequencies can be parameterized in terms of the marginal frequencies and the disequilibria, as shown in Tables 1 and 2, respectively.

Frequency recursion equations: The basic cytonuclear recursions for the joint genotypic frequencies in adults are easily derived by tracing the changes through the two steps of the generation cycle. Each generation begins with adults in the frequencies shown in Table 1. These individuals first mate at random, after which the interim, zygote frequencies in both sexes are simply products of the gamete frequencies described previously (Table 3). The generation cycle then concludes following differential viability selection, for which the viabilities of female and male genotypes are designated as in Table 4, where the subscript refers to the cytonuclear genotype (with cells numbered across the rows) and the superscript refers to the sex. The recursions for the joint, genotypic frequencies in adult females are thus

$$\begin{aligned} (U_1^f)' &= \frac{\phi_1^f P_1^f P^m}{\bar{\phi}^f}, & (U_2^f)' &= \frac{\phi_4^f P_2^f P^m}{\bar{\phi}^f} \\ (V_1^f)' &= \frac{\phi_2^f (P_1^f Q^m + Q_1^f P^m)}{\bar{\phi}^f}, & (V_2^f)' &= \frac{\phi_5^f (P_2^f Q^m + Q_2^f P^m)}{\bar{\phi}^f} \\ (W_1^f)' &= \frac{\phi_3^f Q_1^f Q^m}{\bar{\phi}^f}, & (W_2^f)' &= \frac{\phi_6^f Q_2^f Q^m}{\bar{\phi}^f}, \end{aligned} \tag{6}$$

where a prime (′) denotes a value in the next generation; the normalization factor

$$\begin{aligned} \bar{\phi}^f = {} & \phi_1^f P_1^f P^m + \phi_2^f (P_1^f Q^m + Q_1^f P^m) + \phi_3^f Q_1^f Q^m \\ & + \phi_4^f P_2^f P^m + \phi_5^f (P_2^f Q^m + Q_2^f P^m) + \phi_6^f Q_2^f Q^m \end{aligned} \tag{7}$$

TABLE 3

Genotypic frequencies in zygotes

	Nuclear genotype			
Cytotype	*AA*	*Aa*	*aa*	Total
C	$u_1 = P_1^f P^m$	$v_1 = P_1^f Q^m + Q_1^f P^m$	$w_1 = Q_1^f Q^m$	x
c	$u_2 = P_2^f P^m$	$v_2 = P_2^f Q^m + Q_2^f P^m$	$w_2 = Q_2^f Q^m$	y
Total	$u = P^f P^m$	$v = P^f Q^m + Q^f P^m$	$w = Q^f Q^m$	1.0

is the mean viability in females; the nuclear frequencies in the male gamete pool (P^m, Q^m) are as defined in (1) with f replaced by m; and the female gamete frequencies (P_i^f, Q_i^f) are as defined in (2). The corresponding recursions for the marginal frequencies of the cytotypes and the nuclear genotypes and alleles in adult females are given in appendix a. The frequency recursions and the mean viability for males have the same form as those in (6–7) and (A1–A5) for females with each ϕ_i^f replaced by ϕ_i^m and $\bar{\phi}^f$ by $\bar{\phi}^m$.

The 12 basic equations in (6–7) and their male counterparts determine the dynamics of the full adult/viability model, which has 10 independent variables, 5 in each sex. However, substitution of the decompositions of the female gametic frequencies shown in Table 2 reveals the important point that the full cytonuclear dynamics are, in fact, determined by the dynamics of just 4 key variables: the nuclear allele frequencies in the two sexes (P^f, P^m), the cytoplasmic frequency in females (X^f), and the allelic disequilibrium in females (D^f).

Cytoplasmic frequency changes: Since the dynamics differ between adult males and females, separate analyses must be made for each sex. In the case of females, the relations in Tables 1 and 2 show in conjunction with the genotypic recursions in (6) that the change in cytoplasmic frequency after one generation of selection,

$$\begin{aligned}\bar{\phi}^f \Delta X^f = {} & [(\phi_1^f - \phi_4^f)P^f P^m + (\phi_3^f - \phi_6^f)Q^f Q^m]X^f Y^f \\ & + (\phi_2^f - \phi_5^f)(P^f Q^m + Q^f P^m)X^f Y^f \\ & + [(\phi_1^f - \phi_2^f)P^m + (\phi_2^f - \phi_3^f)Q^m]Y^f D^f \\ & + [(\phi_4^f - \phi_5^f)P^m + (\phi_5^f - \phi_6^f)Q^m]X^f D^f , \quad (8)\end{aligned}$$

where $\Delta X^f = X^{f'} - X^f$, has many parallels to the composite (non-sex-differentiated) cytoplasmic dynamics under the zygote/fertility models (Babcock and Asmussen 1996). For instance, there must be selective differences among females for there to be a change in adult female cytoplasmic frequencies, and although male selection cannot by itself cause a change in female frequencies it can affect the amount of change generated by female selection through its effect on the nuclear allele frequency in the male gamete pool, P^m. Furthermore, there are exactly two situations that will result in a change in female cytoplasmic frequency: either the cytoplasmic gene must affect the *female* fitnesses directly in at least one nuclear background ($\phi_1^f \neq \phi_4^f$ or $\phi_2^f \neq \phi_5^f$ or $\phi_3^f \neq \phi_6^f$), or the cytoplasmic gene must hitchhike on a nuclear gene whose *alleles* are nonrandomly associated with the cytoplasmic gene *in females* ($D^f \neq 0$) and affect *female* fitness in at least one cytoplasmic background (either $\phi_1^f = \phi_2^f = \phi_3^f$ fails or $\phi_4^f = \phi_5^f = \phi_6^f$ fails). The two key differences from the earlier zygote/fertility models are that the female cytoplasmic changes in adult/viability systems depend on the nuclear allele frequencies rather than the nuclear genotypic frequencies, and cytoplasmic hitchhiking requires allelic disequilibrium in females, rather than overall genotypic disequilibria, with a selected nuclear gene in females.

The change in male cytoplasmic frequency after one generation of selection, directly parallels that for females in (8) only if males and females have the same cytoplasmic frequencies in the previous generation (*i.e.*, $X^f = X^m$). Otherwise, the strict maternal inheritance of the cytoplasmic gene makes the frequency change in males a complicated convolution of the previous male and female cytoplasmic frequencies. The reason for this complexity is that in the absence of male selection ($\phi_1^m = \phi_2^m = \phi_3^m = \phi_4^m = \phi_5^m = \phi_6^m$), the male cytoplasmic frequency does not necessarily remain constant, but instead takes on the cytoplasmic frequency in females of the previous generation (*i.e.*, $X^{m'} = X^f$) paralleling the behavior of selectively neutral X-linked genes.

The change in male cytoplasmic frequency from one generation to the next is consequently best analyzed when decomposed into two distinct components

$$\Delta X^m = X^{m'} - X^f + X^f - X^m \quad (9)$$

corresponding to the change caused by departure from the neutral dynamic ($X^{m'} - X^f$) and the change caused by sex differences in the previous generation ($X^f - X^m$). The first component has the same form as ΔX^f in (8)

TABLE 4

Cytonuclear fitnesses

	Nuclear genotype		
Cytotype	*AA*	*Aa*	*aa*
C	ϕ_1^*	ϕ_2^*	ϕ_3^*
c	ϕ_4^*	ϕ_5^*	ϕ_6^*

* represents f (female) or m (male) values.

with each ϕ_i^f replaced by ϕ_i^m and $\bar{\phi}^f$ by $\bar{\phi}^m$, but with the female cytoplasmic frequencies (X^f, Y^f) and allelic disequilibrium (D^f) unchanged. With this formula in place, the decomposition in (9) reveals that the adult male cytoplasmic frequency will change from one generation to the next provided (1) males and females have different cytoplasmic frequencies in the previous generation; (2) the cytoplasmic gene is under direct selection *in males* (*i.e.*, $\phi_1^m \neq \phi_4^m$ or $\phi_2^m \neq \phi_5^m$ or $\phi_3^m \neq \phi_6^m$); or (3) a form of *crisscross genetic hitchhiking* occurs in which the cytoplasmic gene in males hitchhikes on a selected nuclear locus *in males* whose alleles are nonrandomly associated with the cytoplasmic gene *in females*. In addition, because of the distinctive neutral dynamics prescribed by maternal inheritance, the cytoplasmic frequency changes in males caused by frequency differences in the sexes can be generated by cytoplasmic frequency changes (and selection) *in the previous generation of females*, through either direct selection on the cytoplasmic gene in females or genetic hitchhiking upon a selected nuclear gene in females that is in allelic disequilibrium with the cytoplasmic gene in females.

Nuclear allele frequency changes: Since the nuclear marker is inherited biparentally, the change in the nuclear allele frequencies after one generation of selection has the same basic form in adult females and males,

$$\begin{aligned}\bar{\phi}^*\Delta P^* = {} & (\phi_1^* X^f + \phi_4^* Y^f) P^f P^m Q^* - (\phi_3^* X^f + \phi_6^* Y^f)\, Q^f Q^m P^* \\ & + \tfrac{1}{2}(\phi_2^* X^f + \phi_5^* Y^f)(P^f Q^m + Q^f P^m)(Q^* - P^*) \\ & + [(\phi_1^* - \phi_4^*) P^m Q^* + (\phi_3^* - \phi_6^*) Q^m P^*] D^f \\ & + \tfrac{1}{2}(\phi_2^* - \phi_5^*)(Q^m - P^m)(Q^* - P^*) D^f, \end{aligned} \tag{10}$$

with * representing f for P^f in females and * representing m for P^m in males. The nuclear allele frequency dynamics thus depend in both sexes upon the cytoplasmic frequency and allelic disequilibrium in females (X^f and D^f) as well as upon the nuclear allele frequencies in both females and males (P^f and P^m).

Furthermore, as for the male cytoplasmic frequency, changes can be generated in the nuclear allele frequencies from one generation to the next simply by frequency differences in the sexes within the previous generation. This is because in the absence of selection, $P^{f\prime} = P^{m\prime} = \tfrac{1}{2}(P^f + P^m)$, which shows that $\Delta P^f = -\Delta P^m = \tfrac{1}{2}(P^m - P^f)$ will be nonzero whenever $P^m \neq P^f$. Paralleling (9), the change in the presence of selection in one or both sexes can be decomposed into the change caused by departure from the neutral dynamic [*e.g.*, $P^{f\prime} - \tfrac{1}{2}(P^f + P^m)$ for females] and the change caused by frequency differences between males and females in the previous generation [*e.g.*, $\tfrac{1}{2}(P^m - P^f)$ for females]. However, since the first component is still difficult to interpret in the general case, we will focus as in Babcock and Asmussen (1996) on the important special case giving the conditions for a reverse hitchhiking effect from a selected cytoplasmic locus in either sex.

If the nuclear locus is selectively neutral in females ($\phi_1^f = \phi_2^f = \phi_3^f$ and $\phi_4^f = \phi_5^f = \phi_6^f$), the change per generation in the nuclear allele frequency in adult females is

$$\Delta P^f = \tfrac{1}{2}(P^m - P^f) + \frac{(\phi_1^f - \phi_4^f) D^f}{2(\phi_1^f X^f + \phi_4^f Y^f)}. \tag{11}$$

A change in the female frequency at a nuclear locus with no selective effect in females can therefore be generated either by (1) differences in male and female nuclear allele frequencies in the previous generation or (2) hitchhiking upon a cytoplasmic marker that is both under selection and in allelic disequilibrium with the nuclear marker *in females* ($\phi_1^f = \phi_2^f = \phi_3^f \neq \phi_4^f = \phi_5^f = \phi_6^f$ and $D^f \neq 0$). Selection on the nuclear locus in males can contribute to a change in P^f indirectly through its effect upon P^m. For a selectively neutral nuclear marker in males ($\phi_1^m = \phi_2^m = \phi_3^m$ and $\phi_4^m = \phi_5^m = \phi_6^m$), the per-generation change in the nuclear allele frequency in adult males is equivalent to the female formula in (11) with P^f and P^m interchanged and each ϕ_i^f replaced by ϕ_i^m. Thus, similar to what was found for cytoplasmic markers, a male frequency change at a neutral nuclear locus in males will be generated by either prior frequency differences in the two sexes or *crisscross hitchhiking* upon a cytoplasmic marker that is under direct selection *in males* and in allelic disequilibrium with the nuclear marker *in females*.

Disequilibrium dynamics: The female disequilibrium dynamics are obtained by substituting the recursions for the joint genotypic frequencies from (6–7) and the marginal genotypic and allele frequencies from (A1–A5) into the disequilibrium definitions in (3) and (4). The resulting equations are quite cumbersome, and here we present only the portions that are relevant to the generation of disequilibria *de novo* (*i.e.*, starting with no disequilibria in either sex),

$$\begin{aligned}(\bar{\phi}^f)^2 (D_1^f)' = {} & (\phi_1^f \phi_6^f - \phi_3^f \phi_4^f) P^f P^m Q^f Q^m X^f Y^f \\ & + (\phi_1^f \phi_5^f - \phi_2^f \phi_4^f)(P^f Q^m + Q^f P^m)\, P^f P^m X^f Y^f \\ & + \ldots, \end{aligned} \tag{12a}$$

$$\begin{aligned}(\bar{\phi}^f)^2 (D_2^f)' = {} & (\phi_2^f \phi_4^f - \phi_1^f \phi_5^f)(P^f Q^m + Q^f P^m) P^f P^m X^f Y^f \\ & + (\phi_2^f \phi_6^f - \phi_3^f \phi_5^f)(P^f Q^m + Q^f P^m)\, Q^f Q^m X^f Y^f \\ & + \ldots, \end{aligned} \tag{12b}$$

$$\begin{aligned}(\bar{\phi}^f)^2 (D_3^f)' = {} & (\phi_3^f \phi_4^f - \phi_1^f \phi_6^f) P^f P^m Q^f Q^m X^f Y^f \\ & + (\phi_3^f \phi_5^f - \phi_2^f \phi_6^f)(P^f Q^m + Q^f P^m)\, Q^f Q^m X^f Y^f \\ & + \ldots, \end{aligned} \tag{12c}$$

$$\begin{aligned}(\bar{\phi}^f)^2 (D^f)' = {} & \tfrac{1}{2}(\phi_1^f \phi_5^f - \phi_2^f \phi_4^f)(P^f Q^m + Q^f P^m) P^f P^m X^f Y^f \\ & + \tfrac{1}{2}(\phi_2^f \phi_6^f - \phi_3^f \phi_5^f)(P^f Q^m + Q^f P^m)\, Q^f Q^m X^f Y^f \\ & + (\phi_1^f \phi_6^f - \phi_3^f \phi_4^f) P^f P^m Q^f Q^m X^f Y^f + \ldots, \end{aligned} \tag{12d}$$

where the other terms are of the form $D^f(\ ) + (D^f)^2(\ )$. The full disequilibrium recursions, including the coef-

ficients for D^f and $(D^f)^2$, are given in appendix b. The male disequilibrium recursions have the same form as those in (12) with each ϕ_i^f replaced by ϕ_i^m and $\bar{\phi}^f$ by $\bar{\phi}^m$, but all variables unchanged.

These equations are much more complex than the composite, non-sex-differentiated disequilibrium recursions for the zygote / fertility models of differential selection censused at life stages without sex differences (Babcock and Asmussen 1996). However, as in the previous models, it is evident that disequilibria in females can be generated *de novo* only if the cytoplasmic and nuclear loci each affect female fitness with a nonmultiplicative component between the two markers, such that at least two of the following conditions hold in females:

$$\phi_1^f\phi_6^f \neq \phi_3^f\phi_4^f \quad \text{or} \quad \phi_1^f\phi_5^f \neq \phi_2^f\phi_4^f \quad \text{or} \quad \phi_2^f\phi_6^f \neq \phi_3^f\phi_5^f \qquad (13)$$

Two points should be noted regarding this result. First, although each of these three inequalities will generate female disequilibria, at least two must hold because equality in any two of the combinations will ensure equality in all three. Second, while allelic disequilibria can be generated by any of the three fitness interactions in (13), a given genotypic disequilibrium is generated *de novo* by either of the two involving the associated nuclear genotype. For example, nonrandom associations between the cytotypes and the *AA* nuclear genotype (D_1^f) can be generated by the first two interactions, which are the two involving *AA* individuals. Fitness differences in males cannot generate female disequilibria *de novo*, but may affect the amount of disequilibrium generated by selection in females through their influence on the nuclear frequency in the male gamete pool (P^m).

Although disequilibria in males do not contribute in any way to the next generation, male disequilibria can be generated *de novo* by nonmultiplicative cytonuclear fitness interactions in males analogous to those in (13), in the same way that such female fitness interactions generate female disequilibria. Moreover, just as male selection can modify female disequilibria, selective differences in females can affect the amount of disequilibria generated in males through their influence on P^f, X^f, and D^f. Female selection has by far the stronger impact, however, since selective differences in females can even generate male disequilibria *de novo* by generating allelic disequilibria in females (D^f) although it will take two generations to see male disequilibria that are produced solely in this manner.

NUMERICAL STUDY

The primary goal of this study is to determine if measurable cytonuclear disequilibria detected in an adult census of natural populations can be attributed to viability selection. Here we quantify the potential magnitude of disequilibria, qualitatively describe the disequilibrium dynamics, investigate the likelihood of producing detectable associations under particular patterns or strengths of selection, and determine the possible sign patterns of disequilibria in adult males and females caused by viability selection. Following the same basic approach developed in our previous study (Babcock and Asmussen 1996), we have investigated these issues in computer simulations based on random fitnesses and initial conditions, with particular emphasis on the four basic selection regimes:

1. *Differential selection between sexes*: All 12 fitnesses are independently generated by a random number generator with a uniform distribution on [0, 1]. It is important to note that our subsequent use of the phrase "differential selection between sexes" always implies there is selection both within and between sexes.
2. *Equal selection between sexes*: All 6 female fitnesses are randomly generated from [0, 1], and each male fitness is set equal to the corresponding female parameter.
3. *Female selection only*: Female fitnesses are generated randomly from [0, 1], while all male fitnesses are assigned a value of 1.0.
4. *Male selection only*: Male fitnesses are generated randomly from [0, 1], while all female fitnesses are assigned a value of 1.0.

We again consider disequilibria to be measurable if they are greater than 0.01 in magnitude, which is a typical minimum at which cytonuclear associations are detectable at a 0.05 significance level with reasonable marginal frequencies and sample sizes (Asmussen and Basten 1994; Basten and Asmussen 1997). The disequilibrium trajectories are classified as follows: (i) *none*, disequilibria were never measurable in any generation of their trajectory (always <0.01 in magnitude); (ii) *transient*, disequilibria were measurable at some point along their trajectory, but were no longer so at equilibrium; or (iii) *permanent*, disequilibria were measurable at equilibrium. It should also be emphasized that the results below are all based on simulations with no initial cytonuclear associations and are thus caused by selection alone with no confounding effects from preexisting associations in the population. The statistics reported are averages across three replicate runs, each consisting of 100,000 sets of random parameters / initial conditions. In all cases, variance between runs was negligible. A more detailed description of our methods can be found in Babcock and Asmussen (1996).

Initial generation of disequilibria: In censuses without sex differences (Babcock and Asmussen 1996) the average magnitudes of disequilibria produced *de novo* by a single generation of selection are the same for each of the three basic selection regimes with selection in females (differential selection between sexes, equal selection between sexes, and female selection only), which are the only ones capable of generating disequilibria *de novo* in those systems. Here, in censuses with sex

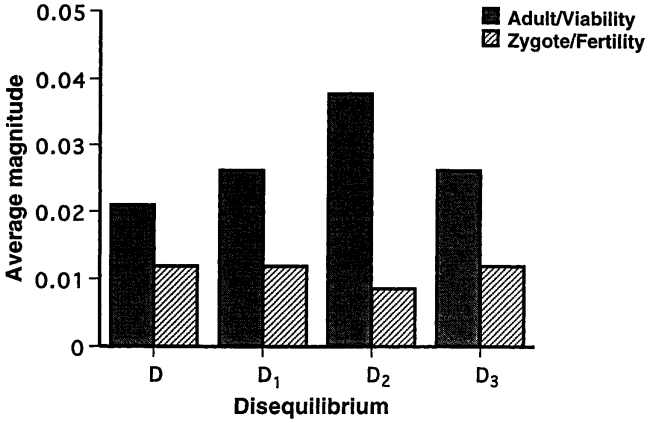

Figure 2.—A comparison of the average magnitudes of cytonuclear disequilibria generated *de novo* by a single generation of random differential selection between the sexes under the adult / viability *vs.* zygote / fertility models. For adults, values are for females; similar results were found for males.

differences, we have already seen analytically that all four basic selection regimes are capable of generating disequilibria *de novo* in at least one sex: differential or equal selection between sexes can immediately generate both male and female disequilibria, while in one generation selection only in females or males produces disequilibria only in the sex with selective differences.

In our simulations, the average magnitudes of adult / viability disequilibria produced by one generation of selection are virtually the same in all cases and are much higher than those generated in the zygote / fertility models. As shown in Figure 2, there is a fourfold difference between the models in the average initial magnitudes of the heterozygote disequilibrium (0.038 *vs.* 0.009) and roughly a twofold difference for the homozygote and allelic disequilibria (0.021–0.026 *vs.* 0.012). Interestingly, the heterozygote association is, on average, the greatest disequilibrium under the adult / viability model, whereas it is the smallest, on average, under the zygote / fertility models of differential selection (Babcock and Asmussen 1996). A comparison of the distributions of normalized disequilibria after one generation of selection (Figure 3) further indicates that differential selection generates disequilibria near their maximum possible magnitude much more frequently in the adult / viability model. Consequently, our initial results suggest that surveys of natural populations are much more likely to detect measurable disequilibria generated by selection if the population is censused at a life stage at which males and females may have different cytonuclear frequencies.

Another noteworthy result regarding the initial disequilibria arises when there are selective differences in only one of the sexes. We already know analytically that male selection can never by itself generate disequilibria in females, while female selection *can* generate disequilibria in males; the latter, however, involves a lag of one

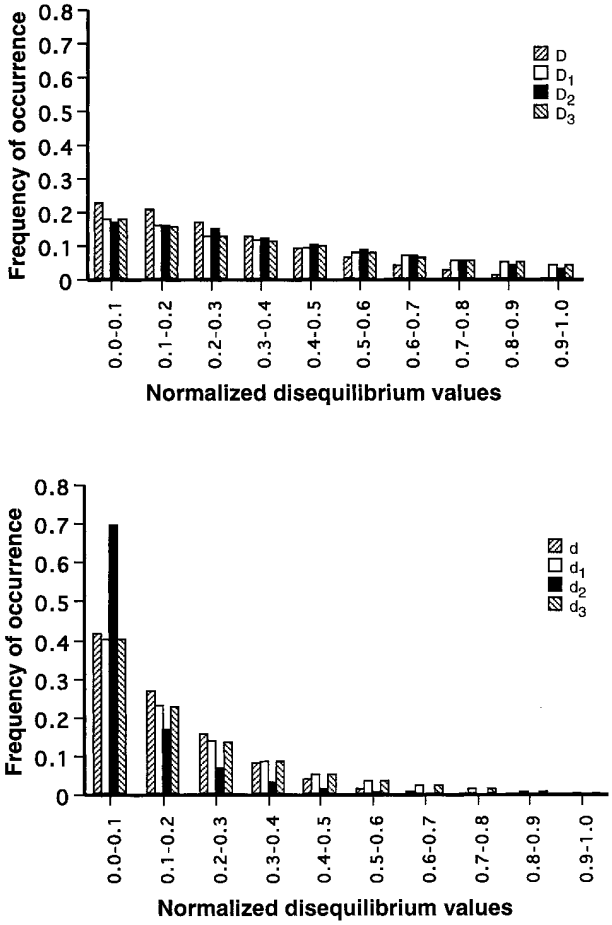

Figure 3.—A comparison of the distributions of normalized cytonuclear disequilibria generated *de novo* by a single generation of random differential selection between the sexes under the adult / viability model (top) versus the zygote / fertility models (bottom). For adults, values are for females; similar results were found for males. Disequilibria are normalized according to their maximum possible magnitudes as described by Asmussen and Basten (1996).

generation and requires two generations of selection. A simulation testing the strength of this indirect effect reveals that the average initial magnitudes of male disequilibria produced by two generations of selection in females are substantially less than those in Figure 2 after one generation of direct selection (0.005 *vs.* 0.038 for the heterozygote disequilibrium and 0.011 *vs.* 0.021–0.026 for the other disequilibria). Moreover, the average magnitudes of female disequilibria increase in the meantime (between the first and second generation), because of the additional round of female selection such that there is a sevenfold difference in the heterozygote disequilibrium measures and a threefold difference in the other disequilibrium measures between males and females in the second generation. The upshot is thus that measurable disequilibria can be produced in males by an indirect, time-delayed effect of selection in females, but these appear to be substantially less than those generated by direct selection in the same sex,

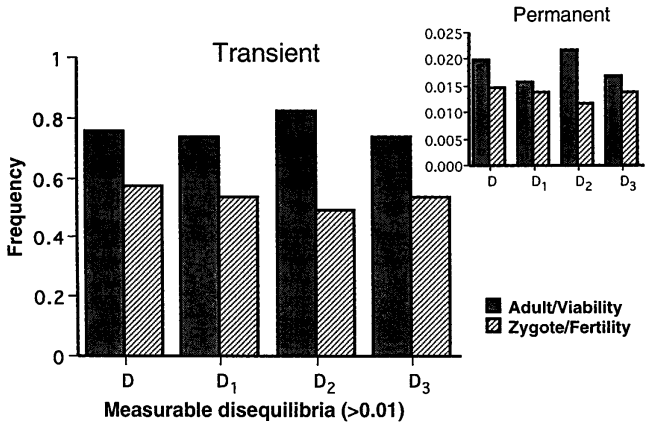

Figure 4.—The frequency of generating measurable transient or permanent (inset) cytonuclear disequilibria by random differential selection between the sexes under the adult/viability model (females only) versus the zygote/fertility models. Male values are similar for permanent associations, but 5–7% higher for transient disequilibria.

presumably because of the diminution by the intervening round of random mating.

Disequilibrium dynamics under differential selection: Moving on to the full disequilibrium trajectories, we find qualitatively similar results to those for the initial disequilibria. We see from Figure 4, for instance, that under random differential selection between the sexes, *measurable* transient and permanent disequilibria are both more likely to be found under the adult/viability model than the zygote/fertility models. On average, measurable transient disequilibria are generated in both sexes by 73–84% of random fitnesses and initial conditions under the adult/viability model *vs.* only 50–58% of the time in the non-sex-differentiated life stages of the zygote/fertility models. There is a similar increase in the overall production of measurable permanent disequilibria under the adult/viability model (1.5–2.1% *vs.* 1.2–1.5%); however, permanent disequilibria are still rare and, as in the zygote/fertility models, account for only ~2.5% of all measurable trajectories under random differential selection. This low incidence of permanent associations is expected, since the opportunity for permanent disequilibria is dependent upon the opportunity for permanent joint cytonuclear polymorphism, which is low (2.6%) and does not differ between models (Babcock and Asmussen 1996). In addition, the two classes of models differ in the frequency of trajectories with measurable heterozygote *vs.* homozygote/allelic disequilibria in the same way that their initial, *de novo* disequilibria vary: under the zygote/fertility models the heterozygote disequilibrium is measurable slightly less often than the others, but is the most likely association to be measurable under the adult/viability model.

Since most of the disequilibria produced by differential selection are transient, it is important to know the duration of these disequilibria (Table 5). Under random differential selection, the average generation past which female disequilibria are no longer measurable is virtually the same as that for the composite, non-sex-differentiated associations in the earlier models (13 generations), although the female, adult/viability disequilibria are on average measurable for a few more generations (11 *vs.* 9 generations). In contrast, the male disequilibria are lost about 1 generation sooner than the others (12 *vs.* 13 generations) and are measurable for the same average number of generations as the zygote/fertility associations (and thus for almost 2 generations less than those in females). For both sexes, however, the ratio between the average number of measurable generations and the average generation lost is high, suggesting that, as in the previous models, most transient disequilibria peak quickly in magnitude and then rapidly decay to zero, rather than have more complex trajectories.

Sex-specific cytonuclear effects: The greatest sex-specific effects of cytonuclear selection are seen by a qualitative comparison of the frequency of generating measurable transient or permanent disequilibria in females and males under the four basic selection regimes of the adult/viability model. In each case, these distinctions are based on the theoretical results of our simulations; the ability to detect these sex differences experimentally will of course depend upon the magnitude of the difference and the sample size employed. One interesting discovery is that, although male disequilibria tend to dissipate sooner (Table 5), random differential selection seems to generate measurable transient associations slightly more often in males (3–5%) than in females (Table 6). However, on average, the maximum magnitude of disequilibria attained along a trajectory is virtually the same in both sexes when averaged across all trajectories that converge to equilibrium, whether or not measurable disequilibria are produced (Table 7). Sex differences are also absent when selection is the same in males and females, which produces measurable transient associations at roughly the same rate (72–83% of the time) and with the same average maximum magnitude (0.03–0.05) in both sexes.

The most striking sex-specific effects are found when there is selection in only one sex. Selection only in females is evidently as effective in generating measurable transient associations in females as the two basic regimes with selection in both sexes (74–82% of the time), but is considerably less successful at generating measurable male disequilibria (38–56% of the time). The maximum magnitudes of the disequilibria generated along a trajectory by female selection show a similar difference; on average, the homozygote and allelic disequilibria in females exceed those in males by a factor of two, and the heterozygote disequilibrium differs by a factor of four (Table 7).

In contrast, male selection alone cannot generate female disequilibria *de novo* and is usually even less effective than the indirect effects of selection in females

TABLE 5

Duration of transient disequilibria under random differential selection

Model	Average generation lost[a]	Average measurable generations[a]	Ratio[b]
Zygote/fertility	13.3	8.7	0.65
Adult/viability, females	13.1	10.9	0.83
Adult/viability, males	11.7	9.4	0.80

[a] Results are averaged over all four disequilibrium measures.
[b] Ratio is computed as the average number of measurable generations/the average generation lost.

at generating measurable male disequilibria (Table 6), despite the higher average maximum magnitudes for male disequilibria under male selection (Table 7). The one anomaly is the male heterozygote disequilibrium, which is generated least often and with the smallest maximum magnitude by selection in females alone. The unusually low incidence of male heterozygote disequilibria under selection only in females is also the one exception to the trend of finding heterozygote disequilibria generated more often than the others in the adult/viability model. This anomaly may be a useful indicator of the form of cytonuclear selection present when analyzing the disequilibria found in natural populations.

Further contrasts are found in the frequency with which the four basic forms of adult/viability selection generate measurable permanent disequilibria (Table 8). On average, permanent disequilibria are produced in nearly equal frequencies in the two sexes, and at rates comparable to those in the zygote/fertility models, if there is differential selection both within and between each sex (the only regime that produces permanent associations in the zygote/fertility models). Permanent disequilibria are never found in any of the models if selection is equal in the two sexes or present only in females, since these forms of selection cannot maintain a cytonuclear polymorphism at any life stage (Babcock and Asmussen 1996).

However, unexpectedly, and in strong contrast to the previous models, permanent disequilibria can be produced in adult/viability systems by male selection only, for which a sizable fraction of the disequilibria produced among males (17–22%) is permanent. In this case cytonuclear polymorphism is maintained in males for 33.3% of random male fitnesses and initial conditions, because with no selection in females the cytotype frequency does not change in zygotes, and, thus, joint cytonuclear polymorphism is preserved at all life stages whenever selection maintains nuclear variation (overdominance in males). Because of the higher incidence of permanent, joint cytonuclear polymorphism, there is an increased opportunity for permanent disequilibria to be generated in males. Interestingly, this opportunity is not realized in the life stages without sex differences, for using the recursions in Babcock and Asmussen (1996) it can be shown analytically that without selection in females the non-sex-differentiated disequilibria always rapidly decay to 0, led by the allelic disequilibrium, which decays at the constant geometric rate of $\frac{1}{2}$ per generation expected for two neutral markers.

Special cases of selection: We have also analyzed 10 special cases of selection to determine if there are particular patterns of selection that have an increased likelihood of generating permanent disequilibria. These include the nine special selective regimes considered and described in Babcock and Asmussen (1996), together

TABLE 6

Frequency of generating measurable (magnitude >0.01) *transient* disequilibria in adults under four modes of viability selection

	Random differential selection		Equal selection between sexes		Female selection only		Male selection only	
Disequilibrium	Females	Males	Females	Males	Females	Males	Females	Males
D	0.74	0.79	0.74	0.74	0.75	0.56	0.0	0.54
D_1	0.73	0.77	0.73	0.72	0.74	0.55	0.0	0.52
D_2	0.81	0.84	0.83	0.82	0.82	0.38	0.0	0.58
D_3	0.73	0.77	0.73	0.73	0.74	0.55	0.0	0.52

TABLE 7

Average maximum magnitude of disequilibria generated along a trajectory by four modes of viability selection

	Random differential selection		Equal selection between sexes		Female selection only		Male selection only	
Disequilibrium	Females	Males	Females	Males	Females	Males	Females	Males
D	0.036	0.033	0.032	0.032	0.034	0.017	0.0	0.025
D_1	0.041	0.039	0.037	0.037	0.039	0.017	0.0	0.031
D_2	0.050	0.051	0.047	0.047	0.047	0.011	0.0	0.043
D_3	0.041	0.039	0.037	0.037	0.039	0.017	0.0	0.031

with *random reverse overdominance*, in which fitnesses are generated randomly from [0, 1] and then reassigned as necessary so that opposite heterozygotes in females and males are overdominant (*e.g.*, $\phi_2^f > \phi_1^f$, ϕ_3^f and $\phi_5^m > \phi_4^m$, ϕ_6^m in Table 4).

The results in Table 9 suggest that the 10 special forms of selection generate permanent disequilibria at roughly the same frequencies in the two sexes and at roughly the same frequencies as the zygote/fertility models. The one slight exception is *reverse cytoplasmic selection* (for which the fitnesses in cytotype *C* of females are the fitnesses in cytotype *c* of males and vice versa), which generates measurable permanent associations in males at only about 80% the rate for females. None of the tested selection patterns is very likely to produce permanent disequilibria, although *random reverse overdominance/underdominance* (in which opposite cytotypes in females and males are overdominant, and the complementary cytotypes are underdominant) has the highest potential for permanent disequilibria in both types of models (4–8%) along with the highest frequency of permanent cytonuclear polymorphism (8.5%).

Strength of selection: A certain minimal strength of selection is necessary to generate measurable cytonuclear disequilibria in the adult/viability model, just as it is in the zygote/fertility models (Table 10). For both types of models, when genotypic viabilities are chosen at random from intervals of the form [a, 1], the likelihood of measurable transient or permanent disequilibria is low to negligible under very weak selection ($a = 0.9$), and steadily increases as the potential selective differences increase to their maximal levels ($a = 0$). However, as might be expected from the other contrasts between the models, it takes much weaker selection to generate measurable disequilibria in the adult/viability model. For instance, the incidence of measurable transient disequilibria in the adult/viability model is over twice that in the zygote/fertility models when fitnesses are drawn from [0.5, 1] and over four times higher when fitnesses are drawn from [0.7, 1]. The results in Table 10 also suggest that the relative ease of generating male *vs.* female disequilibria depends on the strength of selection. Although measurable disequilibria are usually more common in females, interestingly, males have a higher incidence of measurable transient associations than females when selection is strong.

Sign patterns of disequilibria: The interrelationships among the disequilibria in (5) allow eight possible sign patterns among the four disequilibria within each sex, in terms of which are positive or negative. In our previous study (Babcock and Asmussen 1996), we demonstrated analytically that the sign patterns produced by the zygote/fertility models are not useful markers of cytonuclear selection because they always exhibit one of the four sign patterns found between two selectively neutral markers: in every generation the allelic and *AA*

TABLE 8

Frequency of generating measurable (magnitude >0.01) *permanent* disequilibria in adults under four forms of viability selection

	Random differential selection		Equal selection between sexes		Female selection only		Male selection only	
Disequilibrium	Females	Males	Females	Males	Females	Males	Females	Males
D	0.019	0.019	0.0	0.0	0.0	0.0	0.0	0.169
D_1	0.016	0.015	0.0	0.0	0.0	0.0	0.0	0.168
D_2	0.021	0.021	0.0	0.0	0.0	0.0	0.0	0.224
D_3	0.016	0.015	0.0	0.0	0.0	0.0	0.0	0.168

TABLE 9

Frequency of permanent disequilibria in adults under different patterns of viability selection

Selection regime	Disequilibria in females				Disequilibria in males			
	D^f	D_1^f	D_2^f	D_3^f	D^m	D_1^m	D_2^m	D_3^m
Random differential selection	0.020	0.016	0.021	0.017	0.020	0.016	0.023	0.016
Hybrid inviability selection	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Reverse hybrid inviability	0.056	0.053	0.050	0.053	0.050	0.040	0.051	0.041
Reverse cytonuclear selection	0.011	0.0070	0.013	0.0070	0.0094	0.0099	0.013	0.0095
Reverse cytoplasmic selection	0.057	0.049	0.055	0.049	0.040	0.038	0.049	0.037
Reverse directional selection	0.0071	0.0049	0.0071	0.0056	0.0076	0.0052	0.0093	0.0068
Reverse directional/overdominance	0.046	0.025	0.053	0.037	0.040	0.025	0.053	0.042
Hybrid superiority/overdominance	0.015	0.016	0.0016	0.015	0.013	0.012	0.012	0.013
Reverse overdominance	0.043	0.031	0.046	0.032	0.036	0.028	0.044	0.028
Random reverse overdominance	0.038	0.028	0.040	0.030	0.032	0.026	0.038	0.024
Random reverse over-/underdominance	0.076	0.063	0.083	0.044	0.062	0.064	0.079	0.047

genotypic disequilibria (D and D_1) will have the same sign, and the *aa* genotypic disequilibrium (D_3) will have the opposite, with the sign of the heterozygote disequilibrium (D_2) either the same as or opposite that of D, depending on whether the nuclear gene frequency in the male gamete pool is less or greater than ½.

In contrast, the complexity of the cytonuclear disequilibrium recursions in (12) suggests there may be an opportunity in the adult/viability model for distinctive sign patterns to be indicative of selection. We have investigated this possibility by using computer simulations to compute the frequency with which one generation of random differential selection produces each of the eight permissible disequilibrium sign patterns within adults of each sex. The results are presented in Table 11 along with the frequency distribution under the zygote/fertility model. As proven analytically, the zygote/fertility disequilibria always have one of the four sign patterns (patterns 1–4) expected under selective neutrality (Asmussen *et al.* 1987), and these each occur with equal frequency (25%). However, in the sex-specific disequilibria of the adult/viability model, the four neutral patterns each occur just 16–17% of the time, and over 34% of the time one of the other four possible sign patterns (patterns 5–8) is observed, with frequencies of 8–9% each. In these new, nonneutral patterns either the sign of the heterozygote disequilibrium (D_2) is different than the other three (patterns 5 and 8) or the heterozygote and allelic disequilibria (D_2 and D) have one sign, and the two homozygote disequilibria (D_1 and D_3) have the opposite sign (patterns 6 and 7).

TABLE 10

Frequency of generating measurable (magnitude >0.01) transient and permanent disequilibria in adults under different strengths of viability selection

Selection[a]	Transient disequilibrium[b]	Permanent disequilibrium[b]
[0, 1]	0.75/0.79	0.019/0.018
[0.2, 1]	0.69/0.72	0.013/0.012
[0.5, 1]	0.54/0.58	0.007/0.006
[0.7, 1]	0.33/0.26	0.003/0.002
[0.9, 1]	0.015/0.002	0.000/0.000

[a] Range from which random fitnesses are drawn.
[b] Values for females/males.

We also investigated the frequency of sign patterns produced by one generation of each of the 10 special cases of viability selection considered above (Figure 5). It is evident that some special forms of selection are more likely than others to produce distinctive sign patterns of disequilibria. For example, *random overdominance/underdominance* almost always results in one of the four nonneutral sign patterns, and thus its disequilibria are especially valuable markers of nonneutrality or other deviations from Hardy-Weinberg conditions. The reverse is true of the disequilibria produced by *hybrid superiority/overdominance* (all nuclear heterozygotes have a fitness of 1), which almost always have one of the four neutral sign patterns, and thus their patterns are not informative. *Reverse hybrid inviability selection* (in which different complementary homozygous genotypes are most fit in the two sexes) also only rarely generates a nonneutral pattern within either sex; however, under this selection scheme the female disequilibria almost always have sign pattern 1 or 2 (in which the allelic and *AA* homozygote disequilibria are positive and the *aa* homozygote disequilibrium negative), while the male disequilibria almost always have one of the two complementary sign patterns. Such an asymmetry between the sexes is not consistent with strictly neutral, Hardy-Weinberg conditions. Taken together, our results thus suggest that a joint analysis and comparison of the observed disequilibrium sign patterns within each sex may be a useful new tool for making inferences regarding the

TABLE 11

Frequency of disequilibrium sign patterns following one generation of random differential selection

	Disequilibrium				Model	
Pattern	D	D_1	D_2	D_3	Zygote/fertility	Adult/viability[a]
1	+	+	+	−	0.250	0.163/0.163
2	+	+	−	−	0.250	0.167/0.166
3	−	−	+	+	0.251	0.162/0.160
4	−	−	−	+	0.249	0.161/0.166
5	+	+	−	+	0.0	0.093/0.086
6	+	−	+	−	0.0	0.079/0.084
7	−	+	−	+	0.0	0.089/0.087
8	−	−	+	−	0.0	0.084/0.088

[a]Results are for disequilibria in females followed by disequilibria in males.

presence and form of viability selection on cytonuclear genotypes.

DISCUSSION

This study represents the second step in an ongoing theoretical investigation of the major effects of selection in cytonuclear systems. Two long-term goals are to determine what forms of joint cytonuclear or hitchhiking selection could account for the striking, polymorphic mitochondrial dynamics observed in experimental populations (MacRae and Anderson 1988; Fos *et al.* 1990; Hutter and Rand 1995; Kilpatrick and Rand 1995) and to determine the extent to which distinctive patterns of cytonuclear disequilibria that could be used as markers of selection in natural and experimental populations are produced.

In our first study (Babcock and Asmussen 1996), we developed six models of constant, differential selection on cytonuclear genotypes and provided a detailed analysis of the three mathematically identical models for which there are no sex differences at the life stage cen-

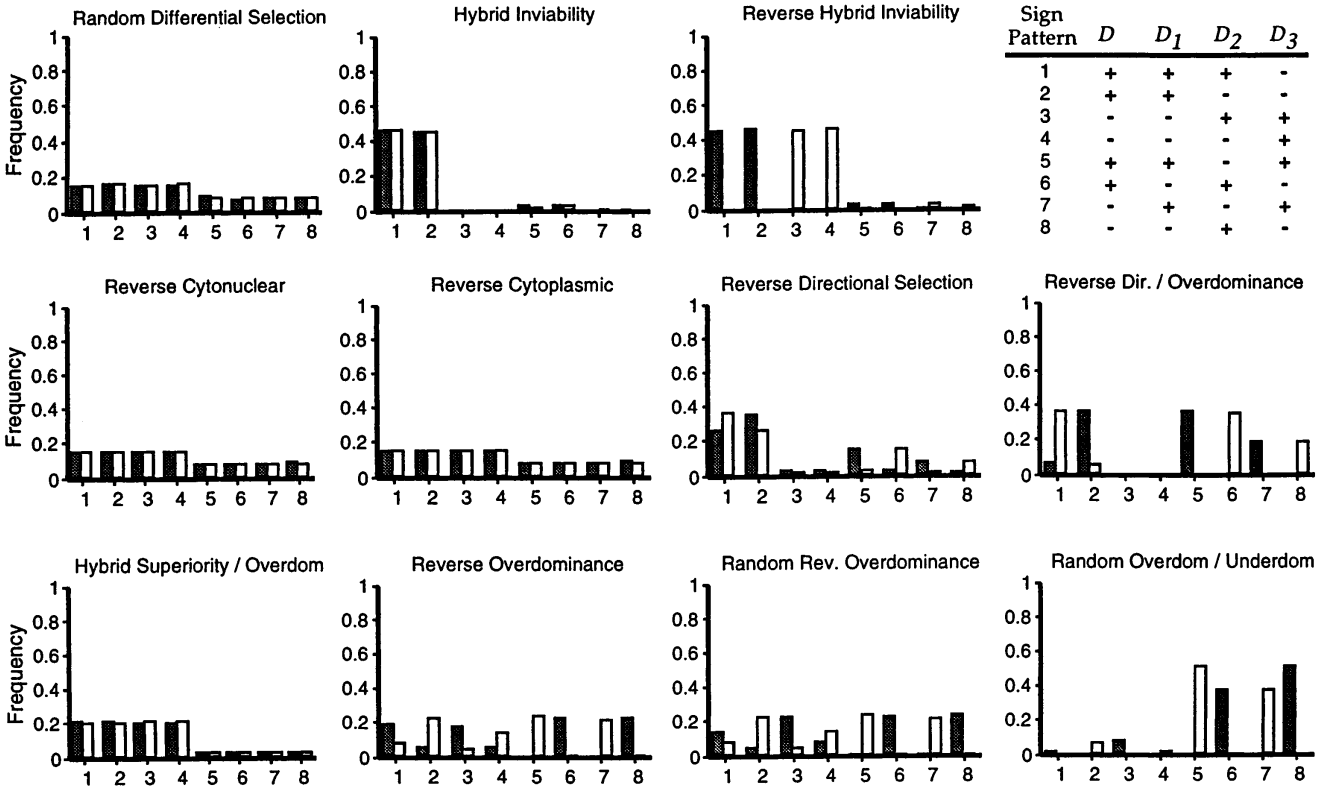

Figure 5.—Frequency of different sign patterns of cytonuclear disequilibria following a single generation of viability selection for random differential selection and 10 special selection regimes. Solid bars indicate female disequilibrium patterns, open bars indicate male disequilibrium patterns, and sign patterns are as indicated in inset.

sused: fertility selection with a zygote or adult census, and viability selection with a zygote census. The fertility selection models assume multiplicative fertilities (*i.e.*, the fertility of a mating pair equals the product of the fertilities of each mate). Here we have extended our analysis to the three remaining models, with sex differences at the life stage censused, which include viability selection with an adult or gamete census and multiplicative fertility selection with a gamete census. Although we have focused on the context of adult/viability selection, this formulation fully subsumes the two gamete systems. The results given here for the allelic frequencies and disequilibrium in adult females, and the nuclear allele frequency in adult males, apply equally to the corresponding variables in the female and male gamete pools, with the viabilities replaced by fertilities in the case of fertility selection.

A key, general discovery, applying to all six models, is that stable, cytoplasmic and joint cytonuclear polymorphisms require strong differential selection both within and between sexes. (The case of male selection only is a degenerate case in this regard, since without selection in females, the cytotype frequency is constant in most life stages, thereby allowing multiple, neutrally stable polymorphic equilibria.) In contrast, the dynamics of cytonuclear frequencies and disequilibria are strongly dependent on the form of selection and life stage censused. At the life stages without sex differences, male selection plays only a subordinate role, serving in most instances only to modify the effects of female selection. For example, the cytoplasmic frequency will change only through direct selection on the cytoplasmic gene in females or through genetic hitchhiking on a selected nuclear gene in females, and cytonuclear disequilibria are only generated *de novo* by nonmultiplicative, cytonuclear fitness interactions in females.

Not surprisingly, the dynamics in populations censused at sex-differentiated life stages provide a much clearer reflection of the presence of cytonuclear selection and its sex-specific effects. Although in females, female selection still plays the predominant role in the generation of cytoplasmic frequency changes, the effects of male selection are now apparent in males. For instance, the male cytoplasmic frequency will change as a result of direct cytoplasmic selection in males or genetic hitchhiking upon a selected nuclear gene in males. On the other hand, the cytoplasmic frequency can change in males simply as a result of prior cytoplasmic frequency differences in males and females, so that male changes can also be caused indirectly by selection in females.

Another striking feature of the allele frequency dynamics in the sex-differentiated life stages is that cytonuclear hitchhiking in males involves a crisscross interaction between the sexes, requiring selection on the other marker in males, but allelic disequilibria between the markers in females. The seemingly paradoxical involvement of female disequilibria is understandable, however, in light of the fact that genetic hitchhiking between two nuclear loci requires linkage disequilibrium between the loci (Thomson 1977; Asmussen and Clegg 1981). Since in our models male cytonuclear disequilibria contribute in no way to the next generation, the cytonuclear associations in females are the only forms of disequilibria through which a hitchhiking effect could occur, and in fact, female allelic disequilibria are able to mediate cytoplasmic and nuclear hitchhiking in both sexes.

Further asymmetrical effects of male and female selection are found in the cytonuclear disequilibria. Whereas nonmultiplicative cytonuclear fitness interactions in females immediately generate female disequilibria *de novo*, they only secondarily generate male disequilibria after a one-generation delay. The corresponding fitness interactions in males can generate disequilibrium *de novo*, but only in males. As might be expected, the male disequilibria produced by direct selection in males are, on average, higher than those produced by indirect, time-delayed selection in females. In other ways, however, female selection still plays the predominant role in that, on average, selection in females generates measurable female disequilibria as often as selection in both sexes and generates measurable male disequilibria even slightly more often than direct selection in males. The potential in males for a double dose of selective effects from the two sexes presumably explains our discovery that under random differential selection, with selection in both sexes, it is generally somewhat easier to generate detectable levels of transient disequilibria in males than in females. In interpreting these results, however, it must be emphasized that they represent the overall potential for cytonuclear disequilibria, based on drawing fitnesses at random from [0, 1], which allows the full range of possible selection strengths. In any particular situation, the sex-specific effects may be different. For instance, the relative ease of generating measurable transient associations in males and females is reversed when selection is weak, for which the reason is unclear.

Together, these results complete our basic understanding of when cytoplasmic frequency changes, genetic hitchhiking, permanent genetic variation, or cytonuclear disequilibria will be generated by cytonuclear selection. The differences between the disequilibria generated at the various possible census times also reveal useful practical guidelines for experimental surveys. In general, the disequilibria at life stages without sex differences will be difficult to detect in experimental and natural populations and be poor markers of cytonuclear selection because they tend to have low magnitude, short duration, and sign patterns consistent with those for two neutral markers. In contrast, measurable disequilibria are produced much more frequently in the adult/viability model, are on average two to four times greater in magnitude, and are likely to remain measur-

able longer. Furthermore, under the adult/viability model, disequilibria are more likely to be generated under weaker selection pressures and to have distinctive sign patterns, either within or between the two sexes, which are not possible between two selectively neutral markers.

Cytonuclear disequilibria generated by joint cytonuclear selection are therefore much more likely to be detected in natural populations and provide useful markers of cytonuclear selection if populations are censused at life stages with frequency differences between the sexes, such as when cytonuclear genotypes differ in either viability or fertility and gametes are sampled, or cytonuclear genotypes differ in viability and adults are sampled. The next, important step will be to delimit the complete cytoplasmic dynamics in these systems, to determine what forms of cytonuclear selection could account for trajectories observed in natural and experimental populations, and to extend this analysis to cytonuclear systems with *X*-linked nuclear markers. Ultimately, for practical applications, it will also be useful to consider the simultaneous effects of random genetic drift and sampling variation.

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APPENDIX A

For adult females the recursion equations for the marginal nuclear frequencies are

$$\bar{\phi}^{f}(U^{f})' = (\phi_1^{f}P_1^{f} + \phi_4^{f}P_2^{f})P^{m} \tag{A1}$$

$$\bar{\phi}^{f}(V^{f})' = (\phi_2^{f}P_1^{f} + \phi_5^{f}P_2^{f})Q^{m} + (\phi_2^{f}Q_1^{f} + \phi_5^{f}Q_2^{f})P^{m} \tag{A2}$$

$$\bar{\phi}^{f}(W^{f})' = (\phi_3^{f}Q_1^{f} + \phi_6^{f}Q_2^{f})Q^{m} \tag{A3}$$

$$\begin{aligned}\bar{\phi}^{f}(P^{f})' = {} & P_1^{f}(\phi_1^{f}P^{m} + \tfrac{1}{2}\phi_2^{f}Q^{m}) + P_2^{f}(\phi_4^{f}P^{m} + \tfrac{1}{2}\phi_5^{f}Q^{m}) \\ & + \tfrac{1}{2}(\phi_2^{f}Q_1^{f} + \phi_5^{f}Q_2^{f})P^{m}\end{aligned} \tag{A4}$$

and that for the marginal cytoplasmic frequency is

$$\bar{\phi}^{f}(X^{f})' = P_1^{f}(\phi_1^{f}P^{m} + \phi_2^{f}Q^{m}) + Q_1^{f}(\phi_2^{f}P^{m} + \phi_3^{f}Q^{m}), \tag{A5}$$

where $\bar{\phi}^{f}$ is the mean female viability defined in (7). These are more informative, however, when rewritten, using the relations in Table 2, in terms of the marginal frequencies and female allelic disequilibrium:

$$\bar{\phi}^{f}(U^{f})' = (\phi_1^{f}X^{f} + \phi_4^{f}Y^{f})P^{f}P^{m} + (\phi_1^{f} - \phi_4^{f})P^{m}D^{f}$$

$$\begin{aligned}\bar{\phi}^{f}(V^{f})' = {} & (\phi_2^{f}X^{f} + \phi_5^{f}Y^{f})(P^{f}Q^{m} + Q^{f}P^{m}) \\ & + (\phi_2^{f} - \phi_5^{f})(Q^{m} - P^{m})D^{f}\end{aligned}$$

$$\bar{\phi}^{f}(W^{f})' = (\phi_3^{f}X^{f} + \phi_6^{f}Y^{f})Q^{f}Q^{m} + (\phi_6^{f} - \phi_3^{f})Q^{m}D^{f}$$

$$\begin{aligned}\bar{\phi}^{f}(P^{f})' = {} & (\phi_1^{f}X^{f} + \phi_4^{f}Y^{f})P^{f}P^{m} + \tfrac{1}{2}(\phi_2^{f}X^{f} \\ & + \phi_5^{f}Y^{f})(P^{f}Q^{m} + Q^{f}P^{m}) \\ & + [(\phi_1^{f} - \phi_4^{f})P^{m} + \tfrac{1}{2}(\phi_2^{f} - \phi_5^{f})(Q^{m} - P^{m})]D^{f}\end{aligned}$$

$$\begin{aligned}\bar{\phi}^{f}(X^{f})' = {} & [\phi_1^{f}P^{f}P^{m} + \phi_2^{f}(P^{f}Q^{m} + Q^{f}P^{m}) + \phi_3^{f}Q^{f}Q^{m}]X^{f} \\ & + [(\phi_1^{f} - \phi_2^{f})P^{m} + (\phi_2^{f} - \phi_3^{f})Q^{m}]D^{f}.\end{aligned}$$

The marginal frequency recursions for adult males have the same form as those above for females with each ϕ_i^{f} replaced by ϕ_i^{m} and $\bar{\phi}^{f}$ by $\bar{\phi}^{m}$.

APPENDIX B

Here we present the full disequilibrium recursions for the adult/viability model. The genotypic disequilibrium recursions for adult females, in terms of allele frequencies and disequilibrium, are of the form

$$(\bar{\phi}^{f})^2(D_i^{f})' = a_i + b_i(D^{f}) + c_i(D^{f})^2 \quad \text{for } i = 1 \text{ to } 3, \tag{B1}$$

where a_i is the portion of each equation presented in (12) in the text, and letting

$$\begin{aligned}T_1 &= P^{f}X^{f} + Q^{f}Y^{f} \\ T_2 &= P^{f}Y^{f} + Q^{f}X^{f} \\ T_3 &= P^{f}Q^{m} + Q^{f}P^{m} \\ T_4 &= P^{m} - Q^{m},\end{aligned} \tag{B2}$$

the other coefficients are

$$\begin{aligned}b_1 = {} & (\phi_1^{f}\phi_6^{f}T_1 + \phi_3^{f}\phi_4^{f}T_2)P^{m}Q^{m} + \phi_1^{f}\phi_5^{f}(Y^{f}T_3 + P^{f}X^{f}T_4)P^{m} \\ & + \phi_2^{f}\phi_4^{f}(X^{f}T_3 + P^{f}Y^{f}T_4)P^{m}\end{aligned}$$

$$c_1 = [(\phi_1^{f}\phi_5^{f} - \phi_2^{f}\phi_4^{f})T_4 + (\phi_1^{f}\phi_6^{f} - \phi_3^{f}\phi_4^{f})Q^{m}]P^{m}$$

$$\begin{aligned}b_2 = {} & [(\phi_2^{f}\phi_6^{f}X^{f} + \phi_3^{f}\phi_5^{f}Y^{f})T_3 - (\phi_2^{f}\phi_6^{f}Y^{f} + \phi_3^{f}\phi_5^{f}X^{f})T_4Q^{f}]Q^{m} \\ & - [(\phi_2^{f}\phi_4^{f}X^{f} + \phi_1^{f}\phi_5^{f}Y^{f})T_3 + (\phi_2^{f}\phi_4^{f}Y^{f} + \phi_1^{f}\phi_5^{f}X^{f})T_4P^{f}]P^{m}\end{aligned}$$

$$c_2 = [(\phi_2^{f}\phi_4^{f} - \phi_1^{f}\phi_5^{f})P^{m} + (\phi_3^{f}\phi_5^{f} - \phi_2^{f}\phi_6^{f})Q^{m}]T_4$$

$$\begin{aligned}b_3 = {} & -(\phi_1^{f}\phi_6^{f}T_1 + \phi_3^{f}\phi_4^{f}T_2)P^{m}Q^{m} - \phi_3^{f}\phi_5^{f}(Y^{f}T_3 - Q^{f}X^{f}T_4)Q^{m} \\ & - \phi_2^{f}\phi_6^{f}(X^{f}T_3 - Q^{f}Y^{f}T_4)Q^{m}\end{aligned}$$

$$c_3 = [(\phi_2^{f}\phi_6^{f} - \phi_3^{f}\phi_5^{f})T_4 + (\phi_3^{f}\phi_4^{f} - \phi_1^{f}\phi_6^{f})P^{m}]Q^{m}.$$

The allelic disequilibrium recursion in females is

$$(\bar{\phi}^{f})^2(D^{f})' = a + b(D^{f}) + c(D^{f})^2 \tag{B3}$$

with a as presented in Equation 12 in the text,

$$\begin{aligned}b = {} & \tfrac{1}{2}[(\phi_2^{f}\phi_4^{f}X^{f} + \phi_1^{f}\phi_5^{f}Y^{f})T_3 + (\phi_2^{f}\phi_4^{f}Y^{f} + \phi_1^{f}\phi_5^{f}X^{f})T_4P^{f}]P^{m} \\ & + \tfrac{1}{2}[(\phi_2^{f}\phi_6^{f}X^{f} + \phi_3^{f}\phi_5^{f}Y^{f})T_3 - (\phi_2^{f}\phi_6^{f}Y^{f} + \phi_3^{f}\phi_5^{f}X^{f})T_4Q^{f}]Q^{m} \\ & + (\phi_1^{f}\phi_6^{f}T_1 + \phi_3^{f}\phi_4^{f}T_2)P^{m}Q^{m}\end{aligned}$$

and

$$\begin{aligned}c = {} & [\tfrac{1}{2}(\phi_1^{f}\phi_5^{f} - \phi_2^{f}\phi_4^{f})P^{m} - \tfrac{1}{2}(\phi_2^{f}\phi_6^{f} - \phi_3^{f}\phi_5^{f})Q^{m}]T_4 \\ & + (\phi_1^{f}\phi_6^{f} - \phi_3^{f}\phi_4^{f})P^{m}Q^{m},\end{aligned}$$

where T_i are as defined in (B2). The genotypic and allelic disequilibrium recursions for males are the same as those for females in (B1) and (B3) with each ϕ_i^{f} replaced by ϕ_i^{m} and $\bar{\phi}^{f}$ replaced by $\bar{\phi}^{m}$.