The Dominance Theory of HALDANE's Rule

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ABSTRACT

"HALDANE's rule" states that, if species hybrids of one sex only are inviable or sterile, the afflicted sex is much more likely to be heterogametic (XY) than homogametic (XX). We show that most or all of the phenomena associated with HALDANE's rule can be explained by the simple hypothesis that alleles decreasing hybrid fitness are partially recessive. Under this hypothesis, the XY sex suffers more than the XX because X-linked alleles causing postzygotic isolation tend to have greater cumulative effects when hemizygous than when heterozygous, even though the XX sex carries twice as many such alleles. The dominance hypothesis can also account for the "large X effect," the disproportionate effect of the X chromosome on hybrid inviability/sterility. In addition, the dominance theory is consistent with: the long temporal lag between the evolution of heterogametic and homogametic postzygotic isolation, the frequency of exceptions to HALDANE's rule, puzzling Drosophila experiments in which "unbalanced" hybrid females, who carry two X chromosomes from the same species, remain fertile whereas F_1 hybrid males are sterile, and the absence of cases of HALDANE's rule for hybrid inviability in mammals. We discuss several novel predictions that could lead to rejection of the dominance theory.

IN 1922, J. B. S. HALDANE observed that, "When in the F_1 offspring of two different animal races one sex is absent, rare, or sterile, that sex is the heterozygous [heterogametic or XY] sex." Although largely ignored for 50 years, HALDANE's rule has recently become the subject of intensive work. The reason is simple: because HALDANE's rule is obeyed in a wide variety of animals (see COYNE 1992), it suggests some common feature underlying the genetics of postzygotic isolation. The question is: what shared genetic characteristic or evolutionary process gives rise to this pattern in group after group of animals, including birds, flies, mammals and moths?

Many answers have been offered (COYNE and ORR 1989a). Although recent genetic work has shown that many of these are wrong, we still do not know the correct explanation. Recently, ORR (1993a) reconsidered perhaps the simplest possible explanation: the lower fitness of heterogametic hybrids may be a consequence of dominance. As MULLER (1940, 1942) emphasized, many of the alleles lowering hybrid fitness may act as partial recessives. If so, it may not be surprising that the hemizygous sex suffers more in species crosses than the homogametic sex: in homogametic hybrids, many of the alleles causing hybrid problems are partially masked in heterozygous state. ORR (1993a) formalized this dominance argument in a simple population-genetic model.

This paper has two purposes. First, we extend ORR's model by relaxing several of his assumptions. The result is a more general picture of the conditions under which HALDANE's rule is expected. In particular, we model the case in which reproductive isolation involves genes of both large and small effect (the previous model assumed that isolation resulted from many genes each of small effect), incorporate epistasis and allow hybrid fitness to be an arbitrary decreasing function of the number of incompatibilities causing hybrid problems (ORR assumed multiplicative fitness effects), clarify Orr's dominance scheme and consider the simultaneous effects of dominance and different rates of evolution for hybrid male *vs.* female steriles.

Our second, and more important, purpose is to consider several biological problems surrounding the dominance theory of HALDANE's rule. Can it explain, for instance, the large effect of the X chromosome observed in genetic analyses of HALDANE's rule? Can dominance explain HALDANE's rule for both hybrid inviability and hybrid sterility? Is the dominance theory consistent with the observation in Drosophila that exceptions to HALDANE's rule are more common for inviability than sterility? Can we estimate the mean dominance of the alleles causing reproductive isolation from the frequency of exceptions to HALDANE's rule or from the time lag between the evolution of heterogametic inviability and homogametic inviability? Can we estimate the number of genes typically causing HALDANE's rule from the frequency with which hybrid males are sterile or inviable in one direction of a species cross, but fit in the reciprocal cross? And can a dominance theory explain the puzzling results of several critical

This paper is dedicated to our friend and mentor Professor Jerry A. "King" Coyne on the occasion of his 45th birthday.

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genetic experiments; specifically, in cases of HALDANE's rule for hybrid sterility in Drosophila, why do hybrid females carrying both *X*s from the same species often remain fertile?

We argue that the dominance theory of HALDANE's rule offers simple and plausible explanations of these patterns. HALDANE's rule and the large *X* effect may simply be consequences of the partial recessivity of alleles having deleterious effects on hybrid fitness.

MODELS INCORPORATING DOMINANCE

Following DOBZHANSKY (1937, p. 256) and MULLER (1940, 1942), we assume that the alleles causing hybrid inviability and sterility do not have such effects within species. As DOBZHANSKY and MULLER emphasized, postzygotic isolation is likely to result from deleterious *interactions* in hybrids between alleles that are "normal" within each species. Thus, the decay of hybrid fitness follows as a byproduct of normal substitution processes: if two populations begin with genotype *aabb*, one may become *AAbb* and the other *aaBB*. Because *A* and *B* are both viable/fertile on their regular genetic background, these substitutions were presumably favored or at least unopposed by natural selection. If, however, *A* and *B* appear together in hybrids, they may be incompatible and cause some hybrid inviability or sterility. In short, the Dobzhansky-Muller model shows how speciation can occur without maladaptive genetic changes (*i.e.*, without peak shifts).

We will not explicitly model incompatibilities between pairs (or triplets, *etc.*) of loci in hybrids (but see ORR 1995). Instead, we take a simpler approach. When a substitution occurs in one of two diverging populations, we tabulate its marginal effect by asking how much it lowers the fitness of F_1 hybrids. We will be mainly concerned with incompatibilities that involve *X*-linked loci. For these incompatibilities, we will assign the marginal effect of the incompatibility to the *X*-linked allele involved in the interaction. As noted by ORR (1993a, 1995), these interactions may involve either ancestral or derived alleles at the *X*-linked locus. In principle, both alleles at an *X*-linked locus in a hybrid female may reduce fitness; however, we will concentrate on the simpler case where only one allele is involved. We do not attempt to keep track of which other locus or loci this *X*-linked allele interacts with in hybrids, or how these loci interact to lower fitness. We also assume that each species is fixed, rather than polymorphic, for the alleles causing hybrid problems. More detailed models may be worthwhile. Given the primitive state of speciation theory, however, it seems useful to first consider simple models that ignore some biological intricacies.

For most of this paper, we focus on hybrid inviability. This is simpler to analyze than hybrid sterility as there is evidence that the same genes affect viability of both male and female hybrids. Different loci, however, appear to cause hybrid male *vs.* female sterility; we address this complication below.

For convenience, we refer to the heterogametic sex as males, as in Drosophila and mammals. All of our arguments also apply to systems of female heterogamety. By "hybrids" we mean F_1 hybrids, unless otherwise stated. We temporarily ignore any hybrid fitness effects of the *Y* chromosome. For hybrid inviability, this is a safe simplification as, at least in Drosophila, the *Y* has no essential somatic function (ASHBURNER 1989). In the case of hybrid sterility, genetic analyses in Drosophila show that the *Y* has a large effect in some species crosses and none in others (see COYNE and ORR 1989a and below).

We wish to calculate the expected fitness of hybrid males *vs.* females. Hybrid fitness will depend both on the number of substitutions that occur between two diverging populations and on the dominance of alleles in hybrids. Obviously, any particular recessive allele causing hybrid problems will, if *X* linked, affect hybrid males more than females: this allele is fully expressed in hemizygous males but partially masked in heterozygous females. However, as ORR (1993a) emphasized, we must also keep track of the expected *number* of genes producing incompatibilities: although females may partially mask many alleles causing postzygotic isolation, they will on average carry twice as many *X*-linked alleles that reduce hybrid fitness (because they carry twice as many *X* chromosomes). The critical question is: how do these two opposing forces interact? Are males, in the end, worse off because they fully express all *X*-linked alleles or better off because they carry fewer *X*-linked alleles causing hybrid problems? The answer will clearly depend on the dominance of the alleles reducing hybrid fitness.

ORR's (1993a) analysis assumed that postzygotic isolation involves many loci each of small effect. Because we do not know how many genes typically cause postzygotic isolation (COYNE 1992; CABOT *et al.* 1994; WU and PALOPOLI 1994), it is important to consider both this polygenic case and the case in which isolation involves some alleles of large effect. The calculations below make no assumptions about the number of loci causing reproductive isolation.

Let A_i denote a complete haploid set of autosomes from species i, and let X_i denote an *X* chromosome from species i. Let $w(X_1X_2A_1A_2)$ denote the absolute fitness of an F_1 hybrid female, and let $w(X_iYA_1A_2)$ denote the fitness of a hybrid male with mother from species i. HALDANE's rule requires

$$0 = w(X_iYA_1A_2) \ll w(X_1X_2A_1A_2) \quad \text{for at least one } i.$$

ORR (1993a) considered a multiplicative fitness model and sought conditions so that males would be less fit on average than females [*i.e.*, $w(X_iYA_1A_2) < w(X_1X_2A_1A_2)$]. Below we will introduce a simple epistatic model that allows for complete hybrid inviability (or sterility), as specified in (1); the connection of this model to ORR's multiplicative model is discussed in APPENDIX A.

A simple epistatic model: We begin by considering hybrid inviability. For simplicity, we initially ignore maternal effects and alleles having sex-specific effects. Let $B_i(k)$ denote an X-linked allele at locus k from species i that causes some hybrid inviability. The X chromosome from species 1 carries n_1 loci lowering hybrid fitness, while the X_2 carries n_2 such loci. We assume that each incompatibility involving these X-linked alleles contributes to an additive scale of "hybrid breakdown" that maps nonlinearly onto fitness (*cf.*, KONDRASHOV 1988; CHARLESWORTH 1990). Allele $B_i(k)$ contributes an amount $b_i(k)$ to this scale in hemizygous X_iY males and an amount $h_i(k)b_i(k)$ in hybrid females. We assume that the second allele at this locus does not cause hybrid inviability (but see APPENDIX A). Hence, $h_i(k)$ measures the dominance of $B_i(k)$'s deleterious effect on a hybrid genetic background. We map our additive "hybrid breakdown" scale onto viability via a nonincreasing function, $V(x)$. This allows us to compare the viabilities of different genotypes by comparing their positions on the underlying additive scale.

To model complete hybrid inviability, we let $V(x) = 0$ for $x \geq C$. In other words, C is a threshold on our additive hybrid breakdown scale above which hybrids are lethal. Notice that C is the same for males and females, *i.e.*, we do *not* assume that one sex is inherently more sensitive to the effects of hybridization. Although C is a threshold for complete inviability, each hybrid incompatibility probably decreases fitness somewhat; as a result, viabilities for "breakdown scores" below C may be very low. Under this simple model, we have

$$w(X_iYA_1A_2) = V\left(\sum_{k=1}^{n_i} b_i(k) + \nu\right) \quad \text{and} \quad (2a)$$

$$w(X_1X_2A_1A_2) = V\left(\sum_{k=1}^{n_1} h_1(k)\,b_1(k) + \sum_{k=1}^{n_2} h_2(k)\,b_2(k) + \nu\right), \quad (2b)$$

where ν denotes the contribution of autosomal-autosomal interactions to hybrid inviability. For simplicity, we ignore X-X interactions in females; but these should be relatively rare.

For HALDANE's rule, S_f, the position of hybrid females on the "breakdown" scale, must be less than S_{mi}, the position of hybrid X_iY males, for at least one i. For definiteness, suppose that X_1Y males become inviable before hybrid females. In symbols, this implies

$$S_f \equiv \sum_{k=1}^{n_1} h_1(k)\,b_1(k) + \sum_{k=1}^{n_2} h_2(k)\,b_2(k) + \nu < C \leq \sum_{k=1}^{n_1} b_1(k) + \nu \equiv S_{m1}. \quad (3)$$

We will assume that the stochastic processes underlying substitutions in each species and the effects of the resulting incompatibilities (*i.e.*, the hs and bs) follow identical distributions. In particular, $E(n_1) = E(n_2) \equiv E(n)$. Taking expectations in (3), we see that HALDANE's rule will hold "on average" if

$$2E(n)E(hb) < E(n)E(b). \quad (4)$$

Clearly a sufficient condition for (4) is $h < 1/2$, *i.e.*, HALDANE's rule will "usually" hold if the alleles contributing to hybrid inviability are always partially recessive.

More generally, (4) will be satisfied if

$$2[E(b)E(h) + \mathrm{Cov}(b, h)] < E(b). \quad (5)$$

We can define the scaled effect and dominance of each allele by $\tilde{b} = b/E(b)$ and $\tilde{h} = h/E(h)$, and set $\tilde{c} = \mathrm{Cov}(\tilde{b}, \tilde{h})$, the covariance between these scaled values. In this notation, (4) requires

$$d < 1/2 \text{ with } d = E(h)(1 + \tilde{c}). \quad (6)$$

The parameter d summarizes the dominance of alleles contributing to hybrid breakdown.

Condition (4) for HALDANE's rule will clearly be satisfied if $E(h) < 1/2$ and $\mathrm{Cov}(b, h) < 0$. The first condition states that deleterious alleles tend to be recessive. The second condition states that alleles of relatively large effect tend to be more recessive, *i.e.*, if $b_1(k)$ is relatively large, the deleterious effect of allele $B_1(k)$ tends to masked more than usual by its homolog $B_2(k)$ [*i.e.*, $h_1(k) < E(h)$]. This second condition was emphasized by ORR (1993a). As discussed below, we expect on biological grounds that both conditions, $E(h) < 1/2$ and $\mathrm{Cov}(b,h) < 0$, are likely to hold. In APPENDIX A, we discuss an alternative parameterization that allows for deleterious effects in hybrids of both alleles at a locus and elaborate the connection between our analysis and ORR's (1993a).

Sterility *vs.* inviability: Essentially the same calculations as above apply to hybrid fertility, but we now assume that different loci cause male *vs.* female sterility. Let $n_1(m_1)$ denote the number of loci from X_1 that contribute to female (male) hybrid sterility; and let $B_i(k)$, for $k = 1, \ldots, n_1$, denote the female-specific alleles and $S_i(k)$ the male-specific alleles from X_i. As above, $h_i(k)b_i(k)$ gives the contribution of $B_i(k)$ to hybrid female sterility, and $s_i(k)$ gives the contribution of $S_i(k)$ to male sterility. Assuming that the decline in hybrid fitness has the same functional form, V, in both sexes, (2) becomes

$$w(X_iYA_1A_2) = V\left(\sum_{k=1}^{m_i} s_i(k) + \nu_m\right) \quad \text{and} \quad (7a)$$

$$w(X_1X_2A_1A_2) = V\left(\sum_{k=1}^{n_1} h_1(k)\,b_1(k) + \sum_{k=1}^{n_2} h_2(k)\,b_2(k) + \nu_f\right), \quad (7b)$$

where ν_m (ν_f) represents the contribution of autoso-

mal-autosomal interactions to male (female) hybrid sterility. HALDANE's rule for hybrid sterility holds "on average" when

$$2E(n)E(hb) + E(\nu_f) < E(m)E(s) + E(\nu_m). \quad (8)$$

We do not know enough about the genetics of male *vs.* female hybrid sterility to make confident predictions about the random variables describing the numbers or effects of alleles contributing to sex-specific sterility. The simplest assumption is that they follow identical distributions. If so, $E(n) = E(m)$, $E(\nu_f) = E(\nu_f)$ and $E(b) = E(s)$; so that (8) reduces to (4), and recessivity condition (6) applies to both viability and sterility. If, in fact, male sterility evolves more quickly than female sterility (either because incompatibilities evolve more quickly [*i.e.*, $E(m) > E(n)$ and $E(\nu_f) > E(\nu_f)$] or they individually tend to have larger effects [*i.e.*, $E(s) > E(b)$], then condition (6) can be relaxed, *i.e.*, even partially dominant ($d > 1/2$) alleles may be consistent with HALDANE's rule. We elaborate this point below. Our qualitative conclusion is that—irrespective of the relative evolutionary rates or effects of loci contributing to sex-specific sterility—hemizygosity of partially recessive *X*-linked alleles accelerates the evolution of hybrid male relative to female sterility.

IMPLICATIONS OF THE DOMINANCE THEORY

Why would the alleles affecting hybrid fitness tend to be recessive? The calculations above show that, if the alleles reducing hybrid fitness tend to be recessive, HALDANE's rule results. Why should these alleles tend to be recessive?

ORR (1993a) suggested that recessivity is expected if the genes causing hybrid problems tend to act as loss-of-function alleles in a hybrid genetic background. Within species it is well known that deleterious alleles of large effect (*e.g.*, lethals) act as nearly complete recessives, while deleterious mutations of small effect act more additively but still satisfy $h < 1/2$ (SIMMONS and CROW 1977). The negative correlation between effect and dominance has a simple physiological cause (WRIGHT 1969; KACSER and BURNS 1981). Metabolic theory shows that there is a curve of diminishing returns relating fitness (flux through an enzyme pathway) to enzyme activity at a particular locus: while having one functional allele increases flux from 0 to some high level, adding another wild-type allele causes a much smaller increase. Thus, alleles causing drastic reductions in activity act recessively, while those causing small reductions act more additively (KACSER and BURNS 1981).

It seems plausible that a similar phenomenon occurs in species hybrids. In particular, alleles stripped of their normal intraspecific genetic background may sometimes fail to function properly, *e.g.*, an allele $B_1(1)$ may fail to function when interacting with an allele at some second locus from another species. If so, hybrid sterility or inviability may be a consequence of the loss of enzyme activity at locus 1. Because its product is presumably embedded in a chain of enzyme reactions that has been partly or fully broken among hybrids, dominance relations at this locus should mimic those of mutations within species: the greater the loss of flux in homozygotes, the more recessive $B_1(1)$ behaves (KACSER and BURNS 1981). Indeed, the hybrid fitness loss might be equivalent to that for a loss-of-function mutation *within species* that reduces flux by the same amount.

In sum, we need not invoke any special evolutionary explanation for the recessivity of alleles lowering hybrid fitness; it may be a simple consequence of metabolism. Moreover, the recessivity of alleles in species hybrids need not reflect recessivity of allelic effects within species (*cf.* CHARLESWORTH *et al.* 1987).

Can the dominance theory explain the "large *X* effect"? HALDANE's rule is not the only pattern characterizing the genetics of speciation. Mapping experiments reveal that the genes causing hybrid sterility and inviability are very often *X* linked. Indeed the *X* has the largest effect on postzygotic isolation in every one of at least 24 species crosses that has been analyzed (COYNE and ORR 1989a; COYNE 1992). Although many theories have been offered to explain this large *X* effect, none is wholly satisfying and several are demonstrably wrong (COYNE and ORR 1989a; COYNE 1992).

Contrary to COYNE and ORR's (1989a) conclusions, the dominance theory provides a plausible explanation of the *X* effect: if the alleles causing reproductive isolation tend to act recessively, substitution of a hemizygous *X* into a "foreign" genetic background will have more catastrophic effects than substitution of a *heterozygous* autosome into a "foreign" background (see WU and DAVIS 1993). A review of the literature shows that this simple explanation suffices in most reported cases of large *X* effects. Because of the limitations of backcross design (*i.e.*, all hybrids carry a complete haploid set of autosomes from one species), one invariably compares the effect of replacing one hemizygous *X* by another with the effect of replacing *one* autosomal homolog from species 1 with *one* homolog from species 2. Thus, one compares a hemizygous *X* with a heterozygous autosome. The hemizygous substitution will have a larger effect when incompatibilities involve partially recessive alleles.

Early in the evolution of postzygotic isolation, the *X* may have a larger fitness effect than even *homozygous* autosomal substitutions (which appear in F_2, but not backcross, analyses). The reason is that the taxa we genetically analyze are not a random sample of all diverging taxa. To see this, consider the case where hybrid lethals are very recessive. Those taxa that happen to pick up *X*-linked hybrid lethals will produce inviable F_1 hybrids far more often than those that happen to pick up autosomal hybrid lethals. Indeed, we will often not recognize, much less genetically analyze, taxa that pick

up *recessive* autosomal-autosomal incompatibilities. Consequently, even when rates of *X*-linked and autosomal substitution are equal, *X*-linked substitutions will be overrepresented in the sample of taxa we study. Simulations show that this sampling bias can yield a modest excess of *X* to *homozygous*-autosome effects when reproductive isolation involves a fairly small number of genes (results not shown). The effect is greatest just after the first of the two reciprocal hybrid males becomes inviable. It essentially vanishes once both reciprocal males have become inviable.

Neither of these explanations can account for cases of large *X* effects for *homogametic* hybrid sterility or inviability, which led COYNE and ORR (1989a) to reject the dominance theory: because the *X* is no different from an autosome in hybrid females, the dominance theory cannot explain large *X* effects on homogametic fitness. However, two of the four putative cases discussed by COYNE and ORR (1989a)—those involving *Drosophila pseudoobscura* and *D. virilis*—do not in fact demonstrate large homogametic *X* effects (maternally acting alleles are involved in the former case, while the *X* alone was genetically marked in the latter case). Our suspicion is that the remaining two cases, involving *D. montana* and *D. mulleri*, may be statistical flukes. If, however, homogametic sterility/inviability turns out to typically involve large *X* effects, other (or, at least, additional) explanations of the *X* chromosome's role in speciation will be required (*e.g.*, CHARLESWORTH *et al.* 1987).

Can we estimate the dominance of "speciation genes" from the lag between the evolution of hybrid male and hybrid female inviability? The theory above suggests that we can explain HALDANE's rule and the large *X* effect if we assume that the alleles causing hybrid problems tend to act recessively. Now we take the opposite approach and attempt to make inferences about the genetics of hybrid inviability and sterility from data on species hybrids. We begin with a simple estimate of dominance for "hybrid fitness alleles" from published information on the time lag between the evolution of hybrid male *vs.* female inviability/sterility. To simplify the notation, we explicitly consider viability; but the same arguments apply for sterility and the data we use come from studies of both hybrid inviability and sterility.

After a sufficiently long period of divergence, both male and female hybrids become inviable. If the alleles causing hybrid inviability were completely dominant, hybrid females would on average become inviable before males, because the females carry twice as many potentially "bad" alleles on their *X* chromosomes. Conversely, if the alleles causing hybrid inviability were completely recessive, females would never become inviable. Finally, if all relevant substitutions had $h = {}^1/_2$, hybrid males and females would become inviable at the same time on average. Thus, estimates of the relative times at which hybrid males and females become inviable must provide information about the dominance of alleles causing postzygotic isolation.

This relationship is particularly simple under our model with an underlying additive scale. For the homogametic sex, autosomal loci are no different from *X*-linked loci; so both classes of loci must be considered. As before, we apply the convention that the effects of incompatibilities involving *X*-linked loci are assigned to the *X*, and we let n_{x1} (n_{x2}) denote the number of loci on X_1 (X_2) contributing to these incompatibilities. Extending the notation of (3), let

$$S_f \equiv \sum_{k=1}^{n_{x1}} h_1(k)\, b_1(k) + \sum_{k=1}^{n_{x2}} h_2(k)\, b_2(k) + \sum_{k=1}^{I_a} h_a(k)\, b_a(k) \quad \text{and} \quad (9a)$$

$$S_{m1} \equiv \sum_{k=1}^{n_{x1}} b_1(k) + \sum_{k=1}^{I_a} h_a(k)\, b_a(k) \quad (9b)$$

denote the hybrid breakdown scores for hybrid females and X_1Y males, and let $I_x = n_{x1} + n_{x2}$ and I_a denote the number of *X*-autosomal and autosomal-autosomal incompatibilities, respectively, contributing to hybrid inviability. As before, we are ignoring *X*-*X* incompatibilities in this analysis, but they should be relatively rare. Let *C* denote a threshold value for the inviability score at which hybrid lethality occurs. As noted by COYNE and ORR (1989b), molecular data, such as Nei's genetic distance for allozymes or numbers of substitutions estimated from DNA sequences, provide estimates of the age of taxa displaying different degrees of postzygotic isolation.

Once males from both reciprocal crosses become inviable, we know that both $S_{m}1$ and $S_{m}2$ have reached *C*, but we do not know when. However, we can estimate T_m, the average time until hybrid males become inviable, by considering the average age of taxa where hybrid males from *one* of the reciprocal crosses is inviable. In COYNE and ORR's notation, this corresponds to a postzygotic isolation index of 0.25. Similarly, T_f, the average time until $S_f \geq C$, can be estimated using molecular data from taxa displaying an isolation index of 0.75. These cases involve maternal and/or cytoplasmic effects, but they provide a rough guide to the rate at which female inviability evolves.

Taking expectations in (9) and assuming that $E(n_{x1}) = E(n_{x2})$, we have

$$E(S_f) = E(I_x)E(hb) + E(I_a)E(hb) \quad \text{and}$$

$$E(S_{m1}) = E(I_x)E(b)/2 + E(I_a)E(hb). \quad (10)$$

The simplest assumption is that the rate of substitutions causing inviability is the same on the *X* and an equivalent-sized autosome [thus we do not invoke faster evolution of *X*-linked loci due to fixation of partially recessive

advantageous mutations (see CHARLESWORTH *et al.* 1987)]. However, the number of *X*-linked loci is typically smaller than the number of autosomal loci. Thus, even with equal rates per locus, the net rate of *X*-linked substitutions will be smaller than the net rate of autosomal substitutions.

To account for this, we let $I(t) = I_x(t) + I_a(t)$ denote the number of incompatibilities causing hybrid problems at time t. The average fraction of these incompatibilities involving *X*-linked loci is denoted p_x, so that $\mathrm{E}[I_x(t)] = p_x\mathrm{E}[I(t)]$. In Drosophila species such as *D. melanogaster*, which have roughly four times as many autosomal as *X*-linked loci (ASHBURNER 1989), we expect $p_x \approx 1 - (4/5)^2 = 0.36$ for two-locus incompatibilities. Even larger values of p_x result if hybrid incompatibilities involve interactions between more than two genes.

With these assumptions, (10) implies that the average times at which hybrid males and females become inviable satisfy

$$\mathrm{E}[I(T_m)] = \frac{2C}{\mathrm{E}(b)[p_x + 2d(1 - p_x)]} \quad \text{and}$$

$$\mathrm{E}[I(T_f)] = \frac{C}{\mathrm{E}(b)\,d}, \quad (11)$$

where $d = \mathrm{E}(h)(1 + \bar{c})$, as in (6). If the number of incompatibilities separating two taxa increases approximately linearly with time, Equation 11 implies that

$$d \approx \frac{p_x}{2[(T_f/T_m) - 1 + p_x]}. \quad (12a)$$

As expected, $d = 1/2$ when $T_f = T_m$.

A more realistic model must take into account the fact that, although the number of substitutions, K, separating two taxa might increase linearly with time, the number of incompatibilities increases more quickly (ORR 1995). The reason is that if the Kth substitution is equally likely to be incompatible with any of the loci that have previously diverged, incompatibilities become more and more likely with time. Indeed, the number of (pairwise) incompatibilities will rise as $K(K - 1)$. Hence, if $K(t)$ follows a Poisson process, we expect $\mathrm{E}[I(t)] = \beta t^2$ for some constant β. This model conservatively assumes interactions between pairs of genes; interactions between more genes, *e.g.*, triplets, lead to an even faster increase of $\mathrm{E}[I(t)]$. Taking into account this quadratic "snowball" effect, we get

$$d \approx \frac{p_x}{2[(T_f/T_m)^2 - 1 + p_x]}. \quad (12b)$$

We can estimate the ratio T_f/T_m using Nei's genetic distance data from COYNE and ORR (1989b) for taxa showing postzygotic isolation indices of 0.25 and 0.75. The genetic distances reported there imply that $T_f/T_m \approx 3$. Because this ratio is based upon data from a small number of species pairs and it pools data involving both inviability and sterility, it must be viewed as a very rough estimate.

Substituting into Equations 12, we can finally estimate the dominance of the alleles causing hybrid inviability. With $p_x = 0.36$, we obtain $d \approx 0.076$ under the linear model (12a). Under the "snowball" model (12b), we get $d \approx 0.022$. Given the large error in our estimate of T_f/T_m, we cannot take these numbers (much the less the difference between them) too literally. Nonetheless, the qualitative point is clear: to account for the observed large time lag between the evolution of hybrid male and female inviability/sterility in Drosophila, the alleles affecting hybrid fitness must be very recessive.

Can we estimate the dominance of "speciation genes" or their number from the frequency of exceptions to HALDANE's rule? HALDANE's rule does not, of course, hold perfectly. Pooling results from Drosophila, Lepidoptera, birds and mammals, exceptions to HALDANE's rule for hybrid inviability occur about 11% of the time ($n = 82$ species crosses); exceptions to HALDANE's rule for hybrid sterility occur ~2% of the time [$n = 173$ species crosses (data from COYNE 1992), percentages are weighted by the number of crosses from different taxa].

In our simple model (see Equations 9), exceptions to HALDANE's rule occur only when

$$\sum_{k=1}^{n_{x1}} h_1(k)\,b_1(k) + \sum_{k=1}^{n_{x2}} h_2(k)\,b_2(k) > \sum_{k=1}^{n_{xi}} b_i(k)$$

$$\text{for} \quad i = 1, 2. \quad (13)$$

Thus, it might seem that one could estimate the mean and variance of the dominance of *X*-linked alleles causing hybrid problems or their number (I_x) from the observed frequency of exceptions to HALDANE's rule.

Unfortunately, if HALDANE's rule is typically satisfied (*i.e.*, if inequality 6 holds), we can use (13) to predict the chance of violating HALDANE's rule only by making detailed assumptions about the trivariate distributions of homozygous and heterozygous effects and the average number of incompatibilities needed to reach the threshold C. We know too little about these genetic details to have any confidence in such predictions. We can, however, draw some qualitative conclusions.

According to (13), exceptions to HALDANE's rule will be fairly common either if the alleles causing postzygotic isolation have intermediate dominance or if isolation often involves a small number of genes. Given the extreme recessivity implied by our "lag" calculations above, intermediate dominance seems unlikely. Instead, the observed frequency of exceptions may imply that a few genes often cause postzygotic isolation. The reasoning is simple: if the number of substitutions required to exceed C were very large, the law of large numbers implies that exceptions to HALDANE's rule

would be very rare. However, as the number of required substitutions falls, exceptions would become more common if the alleles reducing hybrid fitness are not *always* partially recessive. Tentatively, then, one might explain both the long lag between the evolution of male *vs.* female effects *and* the occurrence of exceptions to HALDANE's rule if postzygotic isolation often involves a modest number of partially recessive alleles.

WU and DAVIS (1993) suggested a third explanation for why exceptions to HALDANE's rule for inviability are more common than for sterility in Drosophila. As they noted, many of the exceptions to HALDANE's rule for inviability involve nonreciprocal lethality: females are inviable in one direction of the cross, but viable in the reciprocal direction. Because these cases must involve maternal effects or interactions between nuclear genes and cytoplasmic factors, (13) is clearly irrelevant. In Drosophila and other developmentally well-known metazoans, maternally acting genes play an important role in the earliest stages of development. Indeed, in Drosophila, products from ~80% of all studied genes are laid down maternally (LAWRENCE 1992, p. 7). As development proceeds, control is gradually handed over to zygotic genes. As a result, there is a great deal of interaction between maternal and zygotic gene products during embryogenesis (*e.g.*, LAWRENCE 1992, Chapters 1–3). WU and DAVIS point out that hybrid *females* suffer a systematic disadvantage with respect to maternal-zygotic interactions. Whereas hybrid males always carry an *X* chromosome and cytoplasm from the same species, hybrid females carry a paternal *X* that derives from a different species than the cytoplasm. Consequently, hybrid females may sometimes suffer nonreciprocal hybrid lethality. However, because maternally acting genes presumably play a smaller role in adult *fertility*, these nonreciprocal exceptions to HALDANE's rule should be rarer for hybrid sterility than inviability, as observed.

Can we estimate the number of incompatibilities causing HALDANE's rule from the frequency of asymmetrical results in species crosses? In ~15% of all cases of HALDANE's rule in Drosophila, hybrid males are sterile or inviable in one direction of the species cross, but not in the other. MULLER (1942, p. 101) argued that the high frequency of these "asymmetric" cases shows that postzygotic isolation often involves a few genes. We have quantified this argument (results not shown). Surprisingly, our analysis showed that, contrary to MULLER's intuition, the frequency of asymmetric crosses tells us very little about the number of genes causing reproductive isolation. Instead, we found that—by varying the detailed genetic assumptions of the model—the observed frequency of asymmetric crosses could be explained by almost any number of genes (including >100) causing isolation. Clearly, direct genetic analyses of "speciation genes" are far more likely to determine the number of genes causing speciation than such theoretical approaches.

Can the dominance theory explain HALDANE's rule for both hybrid sterility and inviability? Because the dominance theory posits that HALDANE's rule reflects recessivity, it predicts that, in species crosses obeying HALDANE's rule for inviability, hybrid *females* who carry both *X* chromosomes from the same species should be inviable (since they fully express all *X*-linked recessives, and we assume that most loci affecting male viability also affect female viability). Experiments in Drosophila have shown that these so-called "unbalanced" hybrid females are indeed inviable (ORR 1993b; WU and DAVIS 1993), and several lines of evidence suggest that the same loci cause both F_1 male and unbalanced female inviability [*e.g.*, "rescue mutations" restore the viability of both genotypes (ORR 1993b)].

Hybrid sterility is different: several experiments have shown that, in crosses obeying HALDANE's rule for sterility, hybrid *females* who carry both *X* chromosomes from the same species remain fertile (reviewed in COYNE and ORR 1989a). In contrast, in mouse *Mus musculus-M. spretus* hybrids, F_1 males are sterile, normal F_1 females are fertile, but *XO* F_1 females are sterile, even though *XO* individuals are typically fertile in mice (BIDDLE *et al.* 1994). Nevertheless, the weight of Drosophila evidence led COYNE and ORR (1989a) to reject MULLER's dominance explanation of HALDANE's rule for hybrid sterility.

However, as COYNE and ORR (1989a) noted and WU and DAVIS (1993) emphasized, there is an important difference between the genes causing inviability and sterility: within species, lethal mutations almost always affect both sexes, while sterile mutations almost always affect one sex only (reviewed in ASHBURNER 1989, Chapter 10). The data from species crosses in Drosophila strongly suggest that the same pattern holds *between* species. Although hybrid male and "unbalanced" female inviability appear to involve the same loci (ORR 1993b), mapping experiments show that hybrid male and female sterility involve different loci (COYNE and ORR 1989a).

This difference might well affect the outcome of unbalanced-female experiments. With hybrid inviability, we can be confident that a recessive *X*-linked allele that kills hybrid males can also—when made homozygous—kill hybrid females. With hybrid sterility, however, we have no similar guarantee that an *X* chromosome that harbors a (recessive) hybrid male sterile also harbors a (recessive) hybrid *female* sterile. Unbalanced females may or may not be sterile. It should be noted, however, that this argument works well only if postzygotic isolation often involves a fairly small number of genes or different numbers of genes for males *vs.* females. If hybrid male sterility reflects the cumulative effects of many loci, then, by the law of large numbers, unbalanced female fitness should closely approximate male

fitness, assuming that each type of sterility requires similar numbers (and kinds) of substitutions.

We can get some feel for the frequency with which unbalanced females should be sterile by assuming that hybrid male and female sterility are independent, but equivalent, evolutionary processes. The cumulative probability of developing *X*-linked hybrid male sterility by time t is $P_m(t)$, while the cumulative probability of developing *X*-linked "unbalanced" hybrid female sterility is $P_f(t)$. The simplest assumption is that substitutions causing male and female sterility accumulate at the same rate, so that $P_m(t) = P_f(t) = P(t)$, *i.e.*, HALDANE's rule merely reflects the *expression* of *X*-linked recessives that affect males. If the cases of HALDANE's rule that are subjected to the "unbalanced female test" are of mean age $\bar{t}$, then the probability that unbalanced females will be sterile is just $P(\bar{t})$ (ORR 1993a).

The mean age of the five Drosophila species pairs that have been tested is $D \approx 0.34$, where D is Nei's genetic distance. We obviously do not know how many taxa of this age have *not* evolved male sterility (we would not see such cases), but we do know that about half of the taxa with $D \leq 0.34$ have already evolved a *second* incompatibility that causes reciprocal hybrid male sterility (COYNE and ORR 1989b, Table 1). Conservatively, then, $P(t) \geq 1/2$ and half of all unbalanced female tests should produce sterile females. Our sample size is too small to reject this possibility with confidence (the probability is $< 1/32$ if we consider only the five Drosophila examples, but the lower bound rises to 0.11 if we include the mouse case). The message, however, is clear: if the dominance theory alone is to explain hybrid sterility as well as hybrid inviability, future unbalanced female tests should reveal a fair share of cases where females are sterile. The recent finding of autosomal *and* *X*-linked regions that, when made homozygous in a "foreign" species background, cause hybrid female sterility (DAVIS *et al.* 1994) is very encouraging: these results prove that hybrid female steriles, although normally masked in heterozygous state, are present in taxa obeying HALDANE's rule.

Finally, it should be understood that the dominance theory does *not* require us to believe that HALDANE's rule is a "composite" phenomenon, having different evolutionary causes for hybrid inviability *vs.* sterility, contrary to WU and DAVIS' (1993) theory and ORR's (1993b) previous interpretation of his data. Instead, HALDANE's rule for both inviability and sterility may have a single simple cause: if the alleles having large effects on hybrid fitness are recessive, heterogametic hybrids will suffer more than homogametic no matter which component of fitness one considers.

If, however, future tests reveal that unbalanced females almost always remain fertile, dominance alone could not account for the sterility data and we would be forced to search for additional genetic causes of HALDANE's rule for sterility. Although we do not believe that present data compel us to embrace these more complex explanations of HALDANE's rule, such "composite" theories need not preclude a role for dominance. Indeed, in APPENDIX B, we show how our model can be generalized to construct a formal composite theory of HALDANE's rule. In particular, APPENDIX B considers the simultaneous effects of three processes: dominance, differential rates of accumulation of male- *vs.* female-specific hybrid steriles, and differential rates of substitution on the *X* *vs.* autosomes. Our results show that the critical level of dominance required for HALDANE's rule does not depend on the rate of substitutions on the *X* *vs.* autosomes; the required dominance is, however, very sensitive to the rates of substitution of male- *vs.* female-specific alleles.

Does the dominance theory work given mammalian dosage compensation? All of the arguments above assume that hybrid females carry two functional *X* chromosomes. In mammals, however, dosage compensation is achieved by inactivating one *X* chromosome early in female development [in the mouse, inactivation begins as early as the 40–50 cell stage (TAKAGI 1974)]. In marsupials, the paternal *X* is almost always inactivated throughout the soma, while in eutherian mammals, inactivation is mosaic, with some patches of female tissue expressing the maternal *X* and others the paternal *X* (MIGEON 1994; GRANT and CHAPMAN 1988).

Thus, female mammals are effectively hemizygous, explaining their well-known expression of recessive *X*-linked coat-color genes (*e.g.*, tortoise-shell and calico cats). The dominance theory predicts that HALDANE's rule for hybrid inviability should be rarer in mammals than in taxa without *X* inactivation. In marsupials, for instance, any hybrid incompatibility involving an *X*-linked recessive that kills hybrid males should also kill females as the two sexes have identical genotypes (at least after the earliest stages of embryogenesis). In eutherians, female hybrids should often die in species crosses that produce inviable males in *both* directions of the hybridization: no matter which *X* chromosome remains active in a patch of hybrid cells, it expresses *X*-linked hybrid lethals. Females might, on the other hand, sometimes survive in those cases where males die in only one direction of the cross: data from humans show that females who are heterozygous for recessive *X*-linked disease alleles sometimes survive. This is often due to proliferation of "healthy" cells, compensating for the loss of mutant cells (reviewed in MIGEON *et al.* 1981).

In sum, mammals might conform to HALDANE's rule before—but rarely after—the evolution of reciprocal male inviability. In other taxa, the vast majority of hybridizations obeying HALDANE's rule show two-way male inviability: because of the long lag before the evolution of female effects, species pairs tend to "pile up" at two-way male inviability. In Drosophila, for instance, of 14 hybridizations obeying HALDANE's rule for hybrid inviabil-

ity, 12 involve reciprocal male inviability (COYNE and ORR 1989b). These cases should not appear in mammals.

Our argument assumes that lethals are often autonomous: patches of tissue expressing hybrid lethal "X_1" are not rescued by patches of tissue expressing hybrid lethal "X_2" and vice versa. How strong is the evidence for autonomy? A good deal of information is available in genetically well-known organisms, like Drosophila, where the required mosaics are readily constructed. The fact that many lethals can be recovered as small homozygous clones in the fly cuticle (where markers are easily scored) tells us little, as the mutant's lethality presumably has nothing to do with effects in the epidermis (RIPOLL 1977; GARCIA-BELLIDO and ROBBINS 1983). MURPHY (1974), however, tested the autonomy of lethals known to disrupt the imaginal discs; he showed that about half of these lethals were autonomous in the critical disc tissue. Similarly, RIPOLL (1977) produced flies that were hemizygous for half of their tissues throughout development (making it likely that any critical tissue was hemizygous); he estimated that ~75% of *X*-linked lethals are autonomous (also see GARCIA-BELLIDO and ROBBINS 1983). Unfortunately, analogous data are not available in mammals, although some human disease alleles are clearly nonautonomous (MIGEON 1994). To the extent, however, that the Drosophila data can be extrapolated to mammals, hemizygous mammalian females should often suffer the effects of *X*-linked hybrid lethals.

The situation with hybrid sterility is different. Although one *X* appears to be transiently inactivated in oogonia (MIGEON 1994), it is reactivated by meiotic prophase and both *X*s are active in the female germ line early in development (*e.g.*, both *X*s are expressed in mouse oocytes by day 11) (KRATZER and CHAPMAN 1981; GRANT and CHAPMAN 1988). Thus, to the extent that sterility involves meiotic or postmeiotic problems—the rule in Drosophila (WU and DAVIS 1993)—mammals should continue to obey HALDANE's rule for sterility: males are hemizygous and females are heterozygous.

This is, in fact, the pattern observed. COYNE (1992) and WU and DAVIS (1993) recently reviewed the results of species crosses in several animal groups (Drosophila, mammals, lepidoptera, birds), and their conclusions are very similar. Although ~25 cases of one-sex-only hybrid sterility are known in mammals (all of these obey HALDANE's rule), only a *single* case of one-sex-only hybrid inviability is known. Although WU and DAVIS (1993) have argued that the rarity of hybrid inviability reflects the supposed greater sensitivity of spermatogenesis than somatic development to perturbation in hybrids, it seems possible that it instead reflects the fact that mammalian females are—like F_1 males—somatically hemizygous. In sum, mammals may not show an excess of cases of HALDANE's rule for sterility (as claimed by WU and DAVIS), but a shortage of cases for inviability.

Although no other animal group shows this virtually complete absence of inviable hybrids of one sex only, WU and DAVIS point out that HALDANE's rule for hybrid sterility is more common than for inviability in Drosophila. They argue that this pattern reflects the composite nature of HALDANE's rule: hybrid sterility is especially common because spermatogenesis is an inherently sensitive process (but then why are females preferentially sterile in birds and lepidoptera?) and/or because sexual selection causes particularly rapid divergence of the genes affecting male reproduction.

As noted above, we are not convinced that HALDANE's rule for sterility is a composite phenomenon. There is, however, a simple but overlooked reason why hybrid sterility may appear more often than inviability in Drosophila. Although the *Y* chromosome has no essential somatic function in Drosophila (ASHBURNER 1989), it is required for fertility in most species: the *D. melanogaster Y*, for instance, carries six essential male fertility genes (ASHBURNER 1989, p. 692). Because these genes appear to be very large, they may rapidly diverge between species (CHARLESWORTH *et al.* 1987). Although we have focused on *X*-autosomal interactions, there are clearly many more ways of evolving hybrid male sterility than inviability. Hybrid sterility can result from *X*-autosomal, autosomal-autosomal, cytoplasmic-autosomal, *X-Y*, *Y*-autosomal, and *Y*-cytoplasmic incompatibilities. But because the *Y* is very unlikely to affect viability, hybrid male inviability can result only from the first three incompatibilities. Genetic analysis shows that the *Y* does, in fact, often play a major role in hybrid sterility in Drosophila (reviewed in COYNE and ORR 1989a; JOHNSON *et al.* 1993). No cases are known, however, where the *Y* affects hybrid viability in Drosophila.

The near-impossibility of *Y* effects on hybrid viability must contribute to the excess of hybrid sterility over inviability in Drosophila. We do not know if other processes—such as sexual selection (WU and DAVIS 1993)—must be invoked. In any case, it is important to distinguish between two questions: Why do both hybrid sterility and inviability obey HALDANE's rule? and why does hybrid sterility appear more often than inviability? The dominance theory addresses (and, we believe, answers) the first question. One can imagine many possible causes of an excess of hybrid sterility over inviability that have nothing to do with dominance and that have no bearing on the validity of the dominance theory (*e.g.*, selection on the fertility component of fitness might be more intense than on viability, causing a much higher substitution rate for "fertility" than "viability" genes). Nonetheless, the dominance theory would still explain why both sterility and inviability obey HALDANE's rule.

DISCUSSION

The notion that dominance may explain HALDANE's rule is not new. MULLER (1940, 1942) repeatedly em-

phasized that recessive *X*-linked alleles causing hybrid problems will reduce heterogametic fitness more than homogametic. This simple hypothesis was largely abandoned because genetic results from species crosses in Drosophila appeared to rule out any dominance theory: COYNE (1985) showed that, in species crosses obeying HALDANE's rule for hybrid sterility, "unbalanced" females carrying both *X* chromosomes from the same species remain perfectly fertile. Similar results were obtained in several additional Drosophila crosses (reviewed in COYNE and ORR 1989a). These results seemed inconsistent with MULLER's dominance theory: if hybrid females are fertile because they are masking *X*-linked recessives in heterozygous state, why aren't females who carry *both* *X*s from the same species sterile?

Several recent developments render dominance theories more attractive. First, WU and DAVIS (1993) emphasized that hybrid inviability and hybrid sterility may be qualitatively different: *within* species, lethals usually kill individuals of both sexes, while steriles usually afflict one sex only. Thus, cases of HALDANE's rule where unbalanced females remain fertile might not be so surprising: the fact that an *X* chromosome has picked up hybrid male steriles (causing HALDANE's rule) does not guarantee that it has also picked up recessive hybrid female steriles, even when each evolve at the same rate.

This argument implies that, in cases of HALDANE's rule for inviability, unbalanced females *should* be lethal since, at least within species, *X*-linked alleles that kill males can also kill females. ORR (1993b) showed that hybrid females homozygous for an *X* from one species are, in fact, lethal in two Drosophila hybridizations obeying HALDANE's rule for inviability. Several lines of evidence suggest that the same recessive alleles cause both unbalanced female and F_1 hybrid male inviability. These results show that recessive alleles causing hybrid female inviability exist. Had these alleles been dominant, hybrid males and females would have been inviable and we would not have observed instances of HALDANE's rule.

Contrary to the intuition of MULLER (1942) and CHARLESWORTH *et al.* (1987, p. 131), however, the mere existence of some recessive alleles causing hybrid problems does not ensure that hybrid males will be less fit than hybrid females. Instead, as ORR (1993a) pointed out, we must take into account the fact that—although they mask many recessive *X*-linked alleles—hybrid females are twice as likely to carry *X*-linked alleles causing hybrid problems (as they possess two *X* chromosomes). ORR found the conditions under which HALDANE's rule will be obeyed by assuming that: postzygotic isolation results from alleles of individually small effect, hybrid fitness falls off multiplicatively, and alleles at a locus show complementary dominance such that if one allele acts as a recessive in hybrids, the other acts as a dominant.

We have relaxed all of these assumptions. Our results are similar to ORR's, although we find that it may be somewhat easier to obtain HALDANE's rule than previously thought. In particular, if alleles of large effect are allowed in a multiplicative model, hybrid female fitness exceeds hybrid male fitness whenever the geometric mean of heterozygous fitness effects is greater than the geometric mean of homozygous fitnesses (APPENDIX A). This condition will be met if the alleles having large effects on hybrid fitness tend to be recessive *or* if all alleles have additive ($h \equiv 1/2$) effects on hybrid fitness. We have also demonstrated that HALDANE's rule follows from recessivity under a simple epistatic model with an underlying additive scale (see Equation 2, a and b). Here, hybrid females enjoy greater fitness than males whenever alleles affecting hybrid fitness are partially recessive (see inequality 6).

More important, we have asked if the dominance theory of HALDANE's rule can explain several other patterns characterizing speciation in animals. For instance, can the dominance theory explain the large effect of the *X* chromosome on postzygotic isolation? Can it account for the long temporal lag between the evolution of hybrid male and hybrid female inviability or the observed frequency of exceptions to HALDANE's rule? Can it account for the observed cases of HALDANE's rule in which males are sterile or inviable in one direction of a species cross but fit in the other? Can it explain why unbalanced females usually remain fertile in cases of HALDANE's rule for hybrid sterility? And, most important, can the dominance theory *simultaneously* account for all of these patterns, or are different circumstances required for each?

The qualitative answer is simple. Two conditions must hold to simultaneously explain these patterns: (1) the alleles reducing hybrid fitness must be very recessive and (2) the genes causing hybrid sterility must affect one sex only, while the genes causing hybrid inviability can affect both sexes (when hemizygous or homozygous). There is growing evidence that condition 1 holds: although more data are needed, recessive alleles having large effects on hybrid fitness appear to be common (ORR 1993b; also see BREEUWER and WERREN 1995 who show that recessive alleles cause F_2 female inviability in haplodiploid species). There is also considerable evidence that condition 2 holds (see WU and DAVIS 1993; ORR 1993b).

Two patterns—the frequent fertility of "unbalanced" females and the frequency of exceptions to HALDANE's rule—are most easily explained under the dominance theory if reproductive isolation is often due to a moderate number of genes. There is no consensus on this point. While some studies suggest that a large number of genes cause postzygotic isolation (CABOT *et al.* 1994), others suggest that a fairly small number of genes are involved (CHRISTIE and MACNAIR 1984; WITTBRODT *et al.* 1989; ORR, unpublished data). This disparity may reflect differences in the age of the taxa

studied: because the number of incompatibilities separating species is expected to "snowball" much faster than linearly with time, gross overestimates of the number of genes required to give reproductive isolation are possible if one studies taxa that evolved hybrid male sterility or inviability long ago (ORR 1995). In any case, new data on the number of genes causing reproductive isolation could render our explanations of unbalanced female fertility and of the frequency of exceptions to HALDANE's rule implausible. Most of our conclusions, however—*e.g.*, our explanation of HALDANE's rule, the large *X* effect, and the temporal lag between male and female effects—do *not* depend on any assumptions about the number of genes causing reproductive isolation.

In sum, we believe that the dominance theory offers a simple and compelling explanation of the evolutionary and genetic patterns surrounding HALDANE's rule. The dominance theory also makes sense of several empirical patterns that have not been widely discussed in the literature. For instance, dominance naturally explains why cases of one-sex hybrid inviability almost never occur in mammals: because of their mechanism of dosage compensation (*X* inactivation), female mammals are more-or-less hemizygous for the *X*, just as males. Thus many *X*-linked hybrid lethals that can kill male hybrids can also kill female hybrids, leaving us with few cases of sex-limited hybrid inviability in mammals. Second, the dominance theory makes a specific prediction about the outcome of unbalanced female tests in cases of hybrid inviability *vs.* sterility. If the genes causing hybrid inviability affect both sexes, and the alleles involved are usually recessive, then: in cases of HALDANE's rule for inviability, unbalanced females should be lethal (as noted above), and in cases of HALDANE's rule for sterility, *unbalanced females should not be lethal*. The second prediction follows because observing HALDANE's rule for sterility means that the *X* cannot carry a recessive hybrid female lethal—such a lethal would have also killed F_1 males, precluding HALDANE's rule for *sterility*. Interestingly, this is just the pattern observed: homozygous-*X* females are lethal in cases of HALDANE's rule for inviability (ORR 1993b), but remain perfectly viable in all cases of HALDANE's rule for sterility that have been studied (see COYNE and ORR 1989a). This curious pattern is naturally explained by the dominance theory.

Another simple prediction of the dominance theory is that the frequency of hybridizations obeying HALDANE's rule should be lower in taxa whose *X* chromosome accounts for a smaller fraction of the genome. This is easily demonstrated by considering the average times until male *vs.* female hybrid inviability evolve according to Equation 11, a and b. As the *X* shrinks, the times at which males and female hybrids become inviable should converge (to a relatively large value), and cases of HALDANE's rule should be fairly infrequent. Similarly, in these "small *X*" taxa, we expect that among the cases satisfying HALDANE's rule, male asymmetry may be more common, because relatively few incompatibilities will involve the *X*s.

Because it has been the focus of a great deal of recent work, one pattern associated with HALDANE's rule—the large *X*-effect—merits separate discussion. Following WU and DAVIS (1993), we argue that the well-known large role of the *X* chromosome in postzygotic isolation (COYNE and ORR 1989a) may be a simple consequence of dominance: in backcross analysis, replacement of one hemizygous *X* by another will obviously have a larger effect than replacement of one autosomal homolog by another, if the genes having large effects on hybrid fitness are partially recessive. We also point out that early in speciation, the effect of the *X* may exceed even that of *homozygous* autosomes. This curious effect, which is a consequence of a sampling bias, is explained above.

In retrospect, a dominance explanation of the large *X* effect may seem obvious. Several points should be borne in mind, however. First, as already emphasized, unbalanced female experiments seemed to rule out any dominance explanation of HALDANE's rule and, in turn, of the large *X* effect: if homozygous females are fertile, there seemed little reason to take dominance explanations seriously. Second, in two cases in Drosophila, homogametic hybrid inviability involves a large effect of the *X* chromosome (COYNE and ORR 1989a). Because these cases involve a *heterozygous X*, they remain unexplained by our dominance theory. These cases may be statistical flukes (especially given that Drosophila species often possess only three or four major chromosomes; assuming that incompatibilities are between chromosomes, there is a large chance that the *X* will be involved by chance alone). If, however, the *X* turns out to play a consistently large role in homogametic hybrid sterility/inviability, the dominance theory for the large *X* effect will have to be supplemented or abandoned.

Last, it should be understood that large effects of the *X* chromosome on hybrid fitness were not inevitable. In particular, the factors causing hybrid sterility and inviability did not *have* to behave as partial recessives. As MULLER (1942) pointed out, one can easily imagine that the genes causing postzygotic isolation typically act as gain-of-function "poison" alleles when on a foreign genetic background. In that case, speciation genes would act *dominantly* ($h > 1/2$) and HALDANE's rule and the large *X* effect would not result. Instead, we believe that the last decade of work in the genetics of speciation suggests that the alleles causing postzygotic isolation typically act as partial recessives.

We suggest that this recessivity may have a simple physiological cause that is independent of the dominance of these alleles on their normal species genetic background (contrary to CHARLESWORTH *et al.* 1987). Gene products may often fail to function (or function

less efficiently) when on a hybrid genetic background. If so, hybrid inviability and sterility might be a direct consequence of this loss of function (ORR 1993a). Metabolic theory (WRIGHT 1969; KACSER and BURNS 1981) would predict, therefore, that those alleles showing the greatest loss of function (and so the largest effect on hybrid fitness) should behave more recessively. In short, no particular evolutionary process within species need be invoked to explain the large *X* effect or HALDANE's rule.

This theory also nicely accounts for an important empirical pattern that has, thus far, eluded simple explanation: while postzygotic isolation shows a large *X* effect, the *X* chromosome does *not* play a disproportionate role in either morphological differences or sexual isolation between species (COYNE 1992). This striking difference is expected under the present theory: loss of gene function in species hybrids might cause sterility, inviability or even the appearance of morphological anomalies in hybrids (*e.g.*, missing eyes). Loss of gene function *in hybrids* cannot, however, create morphological differences or sexual isolation between pure-species individuals.

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APPENDIX A: ALTERNATIVE MODELS

ORR's multiplicative model: Here we will allow both alleles at a locus to contribute to hybrid inviability and show that the multiplicative model discussed by ORR (1993a) produces a less stringent condition for HALDANE's rule than the additive-epistatic model presented in the text. As noted by ORR (1993a), a hybrid incompatibility might involve either the derived or ancestral

allele at some locus. Indeed, *both* alleles may cause some inviability when present on a hybrid autosomal background. This assumption does not violate MULLER's (1942) premise that hybrid incompatibilities are asymmetric. The fact that all intermediate genotypes in the evolution of two taxa must be viable/fertile merely precludes the possibility that both alleles at one locus are incompatible with alleles at the *same* other locus.

Let $\nu_{12}(k)$ denote the contribution of an incompatibility involving genotype $B_1(k)\,B_2(k)$ at X-linked locus k that reduces hybrid inviability, and let $\nu_{ii}(k)$ denote the analogous quantity for a $B_i(k)$ male. We assume $v_{ij}(k) \leq 1$ for all i, j and k. Because not all hybrid incompatibilities involve X-linked genes, we let $\nu < 1$ denote the cumulative viability effect of autosomal interactions in hybrids. The multiplicative model assumes that

$$w(X_i X_j A_1 A_2) = \nu \prod_{k=1}^{n} \nu_{ij}(k), \qquad \text{(A1)}$$

where $n = n_1 + n_2$, and n_i is the number of incompatibilities associated with X_i. Hybrid males with mothers from species 1 will be less fit than the hybrid females if

$$\prod_{k=1}^{n} \nu_{11}(k) < \prod_{k=1}^{n} \nu_{12}(k). \qquad \text{(A2)}$$

Taking the nth root of each side shows that hybrid females are more fit than hybrid males whenever the geometric mean of the heterozygous effects of substitutions is greater than the geometric mean of the hemizygous effects. This is analogous to the HALDANE and JAYAKAR (1963) "geometric mean overdominance" condition for the maintenance of two-allele polymorphisms with temporally varying viabilities.

To pursue this analogy, we can consider the hybrid fitness effects of incompatibilities as random variables. Following ORR (1993a), we assume that the distribution of homozygous effects across all incompatibilities is the same for both species, so that the value of the left-hand side of (A2) is the same, on average, if $\nu_{11}(k)$ is replaced by $\nu_{22}(k)$. A well-known sufficient condition for geometric mean overdominance is that $\nu_{12}(k) \geq [\nu_{11}(k) + \nu_{22}(k)]/2$. Thus, under this multiplicative model, even alleles with *additive* hybrid effects (*i.e.*, $\nu_{12}(k) = [\nu_{11}(k) + \nu_{22}(k)]/2$ for all k), corresponding to $h \equiv 1/2$ in the context of the epistatic model discussed in the text, would produce HALDANE's rule (*sensu* inequality A2). The present result differs from ORR's because, when calculating a hybrid genetic load, he assumed that the hybrid fitness reductions, $1 - \nu_{ij}(k)$, are small enough that

$$\prod_{k=1}^{n} \nu_{ij}(k) \approx \mathrm{Exp}\left(\sum_{k=1}^{n} - [1 - \nu_{ij}(k)] \right). \qquad \text{(A3)}$$

This approximation effectively replaces the geometric mean implicit in (A2) with an arithmetic mean, somewhat restricting the conditions under which (A2) holds. However, as shown below, ORR's calculations hold exactly for a simple class of models incorporating epistasis.

A more general epistatic model: The model presented in the text can be trivially extended to allow for the possibility that both of the alleles at an X-linked locus contribute to hybrid inviability. Extending the notation of Equation 2, let $b_{ij}(k)$ denote the contribution of $B_i(k)\,B_j(k)$ to the additive scale of hybrid breakdown. If we assume that

$$b_{12} = b_{11}h_1 + b_{22}h_2, \qquad \text{(A4)}$$

where h_i denotes the dominance of allele B_i and $h_i \leq 1$, then $\mathrm{E}(b_{12}) = 2\mathrm{E}(b_{11}h_1)$ and the analogue of (5) is

$$2[\mathrm{E}(b_{11})\mathrm{E}(h_1) + \mathrm{Cov}(b_{11}, h_1)] < \mathrm{E}(b_{11}), \qquad \text{(A5)}$$

which is clearly equivalent. The remaining results in the text are unchanged by this generalization.

ORR (1993a) studied the special case where two alleles at a locus are likely to show complementary dominance relationships: if one allele is relatively recessive, the other is relatively dominant. A simple way to parameterize this constraint is to set $h_2 = 1 - h_1$. Under this parameterization, $\mathrm{E}(h) = 1/2$, where we average over both alleles at a locus whether each causes postzygotic isolation ($b_{11} > 0$, $b_{22} > 0$) or not (*e.g.*, $b_{11} > 0$, $b_{22} = 0$). We ignore those loci where neither allele causes isolation. Orr showed that—if the strength of isolation caused by an allele (b) is independent of its dominance (h)—HALDANE's rule does not result. Instead, HALDANE's rule results only when the more recessive allele at a locus has the larger deleterious effect on hybrid fitness (*i.e.*, when $\mathrm{Cov}(b,h) < 0$). ORR's result is merely a special case of our (A5) with $\mathrm{E}(h) = 1/2$.

APPENDIX B: A COMPOSITE MODEL OF HALDANE'S RULE

Here we consider the interaction between three forces affecting HALDANE's rule for sterility: dominance, different rates of accumulation of incompatibilities contributing to male- *vs.* female-specific sterility, and different rates of substitution on the *X vs.* autosomes. Our epistatic-additive model provides a simple framework in which to partition the effects of these processes. Assume that all alleles affecting hybrid fertility are sex specific and zygotically acting. Following the logic of (8) and (10), the expected values of the hybrid female and male "hybrid breakdown" scores are

$$\mathrm{E}(S_f) = \mathrm{E}(I_f)\mathrm{E}(b_f)\,d_f \quad \text{and} \qquad \text{(B1a)}$$

$$\mathrm{E}(S_m) = \mathrm{E}(I_m)\mathrm{E}(b_m)[p_x/2 + (1 - p_x)\,d_m], \qquad \text{(B1b)}$$

where I_f (I_m) is the number of female-specific (male-specific) incompatibilities, $\mathrm{E}(b_f)$ [$\mathrm{E}(b_m)$] is the expected homozygous (or hemizygous) contribution of each incompatibility to female (male) hybrid sterility,

and d_f and d_m summarize the dominance of the sex-specific incompatibilities (see 6). As before, p_x is the fraction of incompatibilities involving *X*-linked loci (see 11). If we assume that dominance relationships do not differ between the sexes, *i.e.*, $d_m = d_f$, (B1) shows that HALDANE's rule emerges "on average" if

$$d < \frac{\tau p_x}{2[1 - \tau(1 - p_x)]}, \qquad \text{(B2)}$$

where the quantity $\tau = \mathrm{E}(I_m)\,\mathrm{E}(b_m)/\mathrm{E}(I_f)\,\mathrm{E}(b_f)$ measures the relative cumulative effects of incompatibilities affecting hybrid male *vs.* female fertility. If, for instance, the evolution of male steriles is more rapid than female steriles, we expect $\tau > 1$.

CHARLESWORTH *et al.* (1987) and COYNE and ORR (1989a) found conditions under which the *X* contributes disproportionately to hybrid breakdown. Note, however, that if $\tau = 1$, (B2) reduces to $d < {}^1/_2$ *irrespective* of p_x. Hence, the relative rates of *X vs.* autosomal evolution does not affect the degree of dominance required to explain HALDANE's rule. In contrast, the relative evolutionary rates of male-specific *vs.* female-specific incompatibilities, τ, dramatically affects the required dominance. If, for instance, the right-hand side of (B2) exceeds 1, HALDANE's rule results even if all alleles act as complete dominants in hybrids ($h \equiv 1$). This critical value is $\tau_c = 2/[p_x + 2(1 - p_x)]$, which decreases toward 1 as p_x decreases. Even for a genome with a large faction of *X*-linked loci such as Drosophila (which may have $p_x \approx 0.36$), this critical value is only 1.22. Thus, even a fairly small bias toward substitution of male-specific steriles can make the dominance hypothesis unnecessary.

The obvious weakness of this "faster sex" hypothesis is that it requires that the sex that accumulates hybrid steriles fastest switches across taxa so that the faster-evolving sex is always heterogametic: in mammals and Drosophila, male steriles must evolve fastest, while in bird and lepidoptera female steriles must evolve fastest. Such a happy coincidence seems very unlikely.

It is also worth noting that dominance can produce HALDANE's rule even if $\tau < 1$, *i.e.*, even if there is an substitution-rate bias that runs counter to HALDANE's rule. For instance, with $p_x = 0.36$, (B2) is satisfied for $d < 0.13$ (0.21) if $\tau = 0.5$ (0.67). In sum—while dominance is not always required to obtain HALDANE's rule—the recessivity of the alleles causing postzygotic isolation always facilitates the emergence of HALDANE's rule.