MUTATION ACCUMULATION AND THE EXTINCTION OF SMALL POPULATIONS

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Abstract.—Although extensive work has been done on the relationship between population size and the risk of extinction due to demographic and environmental stochasticity, the role of genetic deterioration in the extinction process is poorly understood. We develop a general theoretical approach for evaluating the risk of small populations to extinction via the accumulation of mildly deleterious mutations, and we support this with extensive computer simulations. Unlike previous attempts to model the genetic consequences of small population size, our approach is genetically explicit and fully accounts for the mutations inherited by a founder population as well as those introduced by subsequent mutation. Application of empirical estimates of the properties of spontaneous deleterious mutations leads to the conclusion that populations with effective sizes smaller than 100 (and actual sizes smaller than 1,000) are highly vulnerable to extinction via a mutational meltdown on timescales of approximately 100 generations. We point out a number of reasons why this is likely to be an overly optimistic view. Thus, from a purely genetic perspective, current management policies that provide formal protection to species only after they have dwindled to 100–1,000 individuals are inadequate. A doubling of the deleterious mutation rate, as can result from the release of mutagenic pollutants by human activity, is expected to reduce the longevity of a population by about 50%. As some investigators have previously suggested, the genetic load of a population can be readily purged by intentional inbreeding. However, this effect is at best transient, as intentional inbreeding can only enhance the probability of fixation of deleterious alleles, and those alleles that are purged are rapidly replaced with new mutations.

There has never been any question that the vulnerability of a population to extinction increases with decreasing population size (Ludwig 1976; Leigh 1981; Shaffer 1981; Ginzburg et al. 1982; Goodman 1987; Burgman et al. 1992; Lande 1993; Foley 1994). This is true whether the dominant risk is demographic stochasticity (random variation in birth and mortality rates and gender among individuals), temporal variation in critical environmental factors, or genetic problems (inbreeding depression, mutation accumulation, and loss of adaptive variation). Less clear is how rapidly the risk of extinction declines with increasing population size and the degree to which this relationship is nonlinear. Determination of the scaling between extinction probability and population size is of practical importance since it can help reveal whether a threshold population size exists below

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which there is a rapid increase in vulnerability to extinction. Such information can be of considerable value in the design of breeding and maintenance programs for small captive populations of endangered species, exotic breeds, and inbred lines.

For factors such as demographic and environmental stochasticity, the relationship between expected time to extinction and mean population size can depend rather specifically on the life-history features of the population and on the temporal patterns of environmental variation to which it is exposed. However, for certain genetic problems, general statements may be possible since the basic Mendelian mechanisms of gene transmission are essentially constant across species (Lynch and Lande 1993; Lynch et al. 1993; Lande 1994; Bürger and Lynch 1995; Lynch et al. 1995).

This article is concerned with the consequences of the accumulation of unconditionally deleterious genes for the viability of sexual populations. Our primary focus will be on relatively small populations—those with effective sizes on the order of a few hundred individuals or less, which is a rough approximation of the population size to which species have typically declined by the time they are listed as threatened or endangered under the U.S. Endangered Species Act (Wilcove et al. 1993). In addition to evaluating the degree to which the risk of extinction scales with population size and fecundity, we consider the consequences of an increase in the genomic mutation rate that might result, for example, from an increase in exposure to mutagens promoted by human activities.

BACKGROUND

Deleterious mutations contribute to the mean fitness of an isolated population in three ways. First, segregating mutations inherited from the ancestral population eventually are either eliminated by natural selection or drift to fixation by random chance. Second, mutations arising in an isolated population lead to the establishment of a new quasi-steady-state pool of segregating mutations defined by the balance between the forces of drift, mutation, and selection. Third, a fraction of the mutations entering the population each generation becomes fixed by random genetic drift. Unlike the first two sources of mutation load, the fixation of recurrent mutations leads to a progressive loss in fitness. We will consider how these three sources of fitness decline sequentially in the following sections and then evaluate their joint contribution to the risk to extinction.

Early attempts to model the genetic risk of extinction for small populations focused entirely on deleterious recessive genes carried in the founder individuals, the primary concern being the expected build-up of inbreeding depression (Senner 1980; Lacy 1992, 1993; Halley and Manasse 1993; Hedrick 1994; Mills and Smouse 1994). With the exception of Hedrick’s (1994) study of full-sib mated lines, the models used in these studies have been rather ad hoc. For example, Senner (1980), Halley and Manasse (1993), and Mills and Smouse (1994) simply used fixed mathematical functions to relate the load due to inbreeding depression to the average coefficient of inbreeding at neutral loci. Such treatment ignores the variation in fitness among individuals and, as a consequence, fails to account for selection against deleterious alleles in any explicit way. Lacy
(1992, 1993) modeled inbreeding depression by assuming that all of the recessive load is due to lethals at a single locus, with each founder individual carrying a unique recessive lethal at that locus. Such a scenario is quite inconsistent with extensive observations that the load in populations due to recessive deleterious genes is spread over many loci, with a large fraction of the total load being attributable to mutations with small effects (Charlesworth and Charlesworth 1987).

Recently, it has become clear that the recurrent introduction of new deleterious mutations may also pose a substantial genetic threat to the long-term persistence of small populations (Lande 1994; Lynch et al. 1995). However, in contrast to previous investigations, these more recent studies gave little consideration to mutations inherited from the ancestral population and were restricted primarily to mutations with additive effects. Consequently, they provide little insight into the question as to whether population bottlenecks can result in a purging of inbreeding depression.

The stochastic theory that we develop in this article provides a general approach to predicting the temporal decline in fitness in a small population due to the accumulation of deleterious genes. Both the mutational load inherited from the ancestral population and the load resulting from recurrent mutations are accounted for, and we allow for any degree of dominance. Our methods also provide a basis for predicting the probability distribution of extinction time, which is of special relevance to issues concerning risk. Prior to presenting the theory, we consider the data on the rate and effects of deleterious mutations, since these play a guiding role in quantitative applications of the theory.

Empirical Estimates of the Mutation Parameters

To gain some insight into the genomic mutation rate to deleterious recessives and the number of loci over which these mutations are distributed, we first point out some useful relationships between these parameters and empirical observations on inbreeding depression for fitness. We then use these relationships to infer the average properties of deleterious mutant alleles, primarily by applying them to data on Drosophila.

Provided a founder population is relatively small, it is reasonable to assume that most segregating loci contain only two alleles. Let the fitness of the three genotypes at the ith locus be 1, 1 - 2\(h_is_i\), and 1 - 2\(s_i\). Then, in the absence of epistasis, the expected downward slope of a plot of the logarithm of mean relative fitness on the inbreeding coefficient is

\[ B = 2 \sum_{i=1}^{n_L} q_i(1 - 2h_i)s_i, \tag{1a} \]

where \(q_i\) is the frequency of the deleterious allele at the ith locus, and \(n_L\) is the number of loci with segregating recessive alleles (Morton et al. 1956). Supposing that all of the loci have the same properties, we reduce equation (1a) to

\[ B = 2n_Lq(1 - 2h)s = n_L(1 - 2h)s, \tag{1b} \]
where \( n_T = 2n_L q \) is the expected number of deleterious recessive genes per individual. The variable \( B \) represents an estimate of the number of recessive lethal equivalents per gamete.

Assuming the ancestral population to be in selection-mutation balance, with expected allele frequencies \( q = u/(2hs) \) (Crow and Kimura 1970), an estimator for the total number of loci segregating for recessive mutations, \( n_L \), can be obtained by rearranging equation (1b):

\[
\hat{n}_L = \frac{hB}{u(1 - 2h)}. \tag{2}
\]

To obtain an estimator for the genomic mutation rate (\( \mu \)) to deleterious recessives, we make the reasonable assumption that the vast majority of deleterious mutations are in a heterozygous state. Thus, to a good approximation, the average fitness of individuals is expected to be \( \bar{W}_0 = (1 - 2hs)^{n_L} \). Note also that provided the effective size of the ancestral population is greater than \( 5/s \), the mean fitness of founding individuals will be approximately \( \bar{W}_0 = e^{-\mu} \) (Haldane 1937; Kimura et al. 1963; Bürger and Hofbauer 1994). This equilibrium mean fitness, which depends only on the mutation rate (not on the mutational effects), is a result of the balance between the loss of mutations by selection and their recurrent acquisition by mutation. Equating these two quantities, substituting for \( n_L \) in equation (1b), and rearranging, an estimator for the genomic mutation rate is obtained:

\[
\hat{\mu} = \frac{2Bh}{1 - 2h}. \tag{3}
\]

This result implies that, so long as mutations are partially recessive (\( 0 < h < 0.5 \)), an estimate of the genomic mutation rate can be acquired by inbreeding an equilibrium out-breeding population and estimating \( B \). This still requires an estimate of the average degree of dominance, which in principle can be acquired by crossing inbred lines (see, e.g., Comstock and Robinson 1948).

Estimates of the effective number of lethals per gamete are typically on the order of one to two. For human survival to maturity, averaging over three studies, \( B = 1.7 \) (Morton et al. 1956; Slatis et al. 1958; Yamaguchi et al. 1970); for early survival in domesticated chickens, swine, and cattle, \( B \) averages about 1.2 (Pisani and Kerr 1961); and for juvenile survival, the average estimate of \( B \) obtained from captive breeding programs of 38 species of mammals is 2.3 (Ralls et al. 1988). For coniferous trees, \( B \) is substantially higher—averaging over nine species, it is 3.8 for embryonic survival, with one to two additional lethal equivalents influencing germination and the subsequent survival rate (Sorensen 1969, 1970; Koski 1971; Franklin 1972; Coles and Fowler 1976; Sorensen et al. 1976; Park and Fowler 1982; Fowler and Park 1983).

Empirical information on the fitness effects of individual mutations is available for \textit{Drosophila} only. However, as we now show, there is a remarkable consistency between results obtained directly by mutation-accumulation experiments and indirectly by the methods outlined above, within and between species. By using balancer chromosomes with visible markers and exploiting the lack of recombination in males, Mukai (1964, 1979) was able to accumulate mutations on
MUTATION ACCUMULATION AND EXTINCTION 493

intact second chromosomes in isolated sister lines of *Drosophila melanogaster*. Using the Bateman-Mukai technique (reviewed in Lynch 1994), which relies on estimates of the rate of decline in mean fitness and the rate of increase in between-line variance, he obtained a lower-bound estimate of the genomic mutation rate equal to 0.80, as well as an upper-bound estimate of the composite parameter \(2(1 - h)s\) equal to 0.025. Subsequent work done by Ohnishi (1977) and Houle et al. (1992) on the same species estimated \(\mu \geq 0.30\) and 1.05, respectively, and \(2(1 - h)s \leq 0.030\) and 0.061. Averaging over all three studies, \(\mu \geq 0.72\) and \(2(1 - h)s \leq 0.039\). These estimates exclude lethal mutations.

To go any further with these data, we need an estimate of \(h\). Through assays of chromosomal heterozygotes, it has been found that \(h \approx 0.36\) for newly arisen mutations (Mukai et al. 1965; Mukai and Yamazaki 1968; Mukai 1969). We now show that similar estimates are obtained for mutations segregating in natural populations by using information on the relative fitness of chromosomal homozygotes (again produced in laboratory constructs using balancer chromosomes). From the theory developed above, we know that the expected number of homozygous loci for deleterious genes in a completely inbred line is approximately equal to the number of deleterious mutations per gamete \(\mu/(4sh)\). Thus, the expected fitness of a completely homozygous line is \(\bar{W}_1 = (1 - 2s)\mu/(4sh) = e^{-\mu/(2h)}\). Dividing this by the expected fitness of outbred individuals, \(\bar{W}_0 = e^{-\mu}\), and rearranging, we obtain an estimator for \(h\),

\[
\hat{h} = \frac{\mu}{2[\mu - \ln(\bar{W}_1/\bar{W}_0)]}.
\]  

Charlesworth and Charlesworth (1987) summarized data on \((\bar{W}_1/\bar{W}_0)\) for egg-to-adult viability associated with different chromosomes in four species of *Drosophila*. Within species, this ratio is essentially the same for all chromosomes. It is also very similar across species: 0.77 for *D. melanogaster*, 0.76 for *Drosophila pseudoobscura*, 0.85 for *Drosophila subobscura*, and 0.64 for *Drosophila willistoni*. The data are less extensive for total fitness, but they are again strikingly consistent across species: \((\bar{W}_1/\bar{W}_0) = 0.33\) for *D. melanogaster*, 0.36 for *D. pseudoobscura*, and 0.34 for *D. willistoni*. Averaging these values across species and using the lower-bound estimate of \(\mu\) given above, we obtain from equation (4) \(\hat{h} \geq 0.39\) for egg-to-adult viability and \(\hat{h} \geq 0.24\) for total fitness. Thus, all of the available data suggest that deleterious mutations for fitness in *Drosophila* are partially recessive with \(h \approx 0.36\). Applying this value of \(h\) to the results from the mutation-accumulation experiments, we obtain \(s \leq 0.030\).

Our estimate of \(h\) can also be used in conjunction with equation (3) to obtain an estimate of the genomic mutation rate from observations on the decline in fitness in inbred progeny derived from wild-caught parents. Such experiments have been performed for egg-to-adult viability in three species, again yielding fairly consistent results: \(B = 0.88\) for *D. melanogaster* (Mackay 1985), 0.66 for *D. pseudoobscura* (Dobzhansky et al. 1963), and 1.09 for *D. willistoni* (Malogolowkin-Cohen et al. 1964). Averaging over all species, equation (3) yields an estimate of \(\hat{\mu} = 2.26\). This is roughly three times the average lower-bound value obtained from mutation-accumulation experiments (0.72). However, since the
latter estimates excluded lethals, which constitute about 50% of the inbreeding load \( B \) (Charlesworth and Charlesworth 1987), the two estimates are actually in fairly good agreement. This is encouraging and suggests that estimates of the number of recessive lethal equivalents per gamete can in fact be used to successfully estimate the genomic deleterious mutation rate, provided an estimate of \( h \) can be procured. This opens up the possibility of estimating \( \mu \) relatively rapidly for many species, without performing lengthy and laborious mutation-accumulation experiments. Charlesworth et al. (1990) have suggested a similar application to obligately selfing populations.

Some insight into how much the upper-bound estimate of the selection coefficient (0.03) exceeds the true value \( s \) can be acquired by noting that the Bateman-Mukai technique actually estimates \( \bar{s}(1 + \theta_s) \), where \( \bar{s} \) is the mean, and \( \theta_s \) is the squared coefficient of variation of \( s \). Some indirect evidence supports the idea that the distribution of deleterious mutational effects is approximately exponential (Gregory 1965; Edwards et al. 1987; Mackay et al. 1992; Santiago et al. 1992; Keightley 1994)—that is, with a probability density decreasing with increasing \( s \) and a coefficient of variation approximately equal to one. If this is the case, then an unbiased estimate of the mean value of \( s \) is 0.015. Since the Bateman-Mukai technique also estimates \( \mu/(1 + \theta_s) \), similar logic leads to the suggestion that \( \mu \) in *Drosophila* may be close to 1.5.

In summary, the basic conclusions of these results are that deleterious mutations in *Drosophila* are frequent, are partially dominant (\( h = 0.3-0.4 \)), and have individually small effects, which causes an approximately 2% and 4% reduction in fitness in the heterozygous and homozygous states, respectively. Given that other species exhibit similar values of \( B \) to *Drosophila*, these results may be fairly general.

The General Model

We will focus most of our attention on an ideal randomly mating monoecious population growing in discrete generations, with a simple form of density dependence following juvenile production, mutation, and viability selection each generation. Letting \( R \) be the reproductive rate per surviving adult and \( N(t) \) be the number of reproducing adults at time \( t \), we define the number of progeny produced prior to selection as \( RN(t) \). The gametes required to produce these progeny are assumed to be drawn randomly, with free recombination and allowing random selfing, from the \( N(t) \) parents. Newborns are assumed to incur new mutations following a Poisson distribution with an expected genomic mutation rate \( \mu \). All new mutations are assumed to arise at loci that are not currently segregating in the population, a reasonable assumption for small populations. Unless stated otherwise, all mutations are assumed to have the constant properties \( s \) and \( h \).

Following mutation, viability selection operates on the progeny, with the probability of survival to maturity being determined by the fitness function \( W(n_1, n_2) = (1 - 2hs)^{n_1}(1 - 2s)^{n_2} \), where \( 2s \) and \( 2hs \) are the fractional reductions in viability caused by homozygous and heterozygous mutations, and \( n_1 \) and \( n_2 \) are the numbers of loci in the individual that are heterozygous and homozygous for deleterious mutations. This fitness function allows for dominance but assumes that the effects of mutations at different loci are independent. If, following selection, the
number of potential adults exceeds the carrying capacity of the environment, \( K \), the population is assumed to decline to \( K \) by genotype-independent culling prior to the next round of reproduction. So long as the mean viability remains greater than \( 1/R \), the effective population size under this model is very close to \( K \) (Lynch et al. 1995), which we assume below.

We start by presenting some approximations, obtained with transition-matrix (hereafter, abbreviated to TM) and diffusion theory, for the dynamics of fitness decline in small monoecious populations. Our results on expected extinction times are checked by stochastic simulations run on a parallel processing computer (Lynch et al. 1995).

MUTATIONS IN FOUNDER INDIVIDUALS: PURGING VERSUS FIXATION

We first consider the ultimate fate of the deleterious mutations segregating in the founder population, all of which eventually either become eliminated by selection or fixed by random drift. Initially, we will concentrate on mutations with additive effects, primarily to show that they are unlikely to cause a loss of fitness in a bottlenecked population. Throughout, we will assume that the ancestral population is in selection-mutation balance and that the deleterious alleles are not completely recessive. This implies that the mean fitness of founding individuals is approximately \( \bar{W}_0 = e^{-\mu} \).

Mutations with Additive Effects

For a large ancestral population, the expected frequency of a mutant allele with additive effects is \( u/s \) (Crow and Kimura 1970), where \( u \), the genic mutation rate, is expected to be on the order of \( 10^{-4} \) or smaller. With the fitness function that we are using, this implies that the mean number of mutations per individual is approximately \( p/s \). Since \( u/s \) is expected to be quite small, it is reasonable to assume that each individual in a small founder population has a unique set of deleterious alleles (because of their individual rarity). Thus, the founder population can be characterized as initially having \( Kp/s \) polymorphic loci, each with a single deleterious mutation with frequency \( p/(2K) \).

Letting \( u_F \) be the fixation probability of a mutant allele, the expected asymptotic fitness of the population (considering only mutations in the founding individuals and assuming the fixation process acts independently over loci) is

\[
\bar{W}_B = (1 - 2su_F)^{Kp/s}.
\]

For mutations with additive effects,

\[
u_F = \frac{e^{\phi p_0/(1-s)} - 1}{e^{\phi/(1-s)} - 1},
\]

where \( \phi = 4Ks \) and \( p_0 \) and is the initial allele frequency (in this case, \( p_0 = 1/\lfloor 2K \rfloor \)) (Bürger and Ewens 1995; Lynch et al. 1995). This expression for \( u_F \) yields more accurate estimates of the fixation probability than the commonly used expression of Crow and Kimura (1970), which employs \( s \) rather than \( s/(1-s) \) in the exponents.
Fig. 1.—Expected fitness in an isolated population relative to that in an effectively infinite ancestral population, after all of the segregating loci in the founders have become homozygous because of drift and selection. The effective population size is assumed to equal $K$, the deleterious alleles in the founders have additive effects, and new mutations in the isolated population are ignored. The genomic mutation rate in the ancestral population is assumed to be one per individual per generation ($\mu = 1$), so that the mean fitness of founder individuals $W_0 = e^{-\mu} = 0.368$. When all mutations are purged from the population, the relative fitness is $(1/0.368) = 2.718$. Solid lines are for mutations of constant effect $s$; dashed lines assume an exponential distribution of mutational effects with mean $s$. The filled circles denote results obtained from computer simulations, averaging over 256 replicate populations.

The solution of the preceding expressions shows that, for mutations with additive effects, $W_B$ is always greater than $W_0$ (fig. 1). Provided $s > 0.01$, the predicted values are in good agreement with those obtained by computer simulation. For $Ks \ll 0.1$, the deleterious alleles are effectively neutral, each having a probability of fixation approximately equal to $1/2K$. In this case, an expected $(K\mu/s)(1/2K) = \mu/(2s)$ mutations are eventually fixed in the population, which yields $W_B \approx (1 - 2s)^{\mu/2s} = W_0$; that is, the founder effect has essentially no effect on mean fitness. For $Ks > 1$, the fixation probability of a deleterious allele is negligible, and $W_B$ approaches one. Thus, for deleterious genes with additive effects, a population bottleneck leads, on the average, to a purging of the mutation load in the base population, and this purging is essentially complete when $K > 1/s$.

It needs to be emphasized that the preceding results consider only the average long-term consequences of deleterious alleles in a base population. Because the fixation process is stochastic, individual populations may develop values of $W_B$ that are above or below the expectation $W_B$. For issues in conservation biology, it is relevant to consider the probability that the realized value of $W_B$ is less
than the mean fitness of the ancestral population. The statistic of interest is the probability that the number of fixed alleles exceeds $\mu/(2s)$, since beyond this point $W_B < e^{-\mu}$. This probability can be computed by using a binomial distribution involving $K\mu/s$ trials, each with a probability of fixation $u_F$. For $4Ks < 1$, the probability that a given population will decline in fitness relative to the ancestral situation can be quite substantial, even though a purging of the deleterious load is expected on the average (fig. 2). For $2Ks > 1$, the probability of fitness decline is negligible.

Since the equilibrium fitness associated with mutations in the founder population is approached asymptotically, it is of interest to evaluate the timescale over which it evolves. Analytical results given in the appendix indicate that nearly all segregating deleterious mutations in founder individuals will be lost or fixed within a few dozen generations, provided $K$ is on the order of 100 or less.

The preceding derivations assume that deleterious mutations have a constant effect $s$, which is unlikely to be biologically realistic. To gain some insight into the consequences of variation in mutational effects, we will consider the situation when the selection parameter $s^* = s/(1 - s)$ has an exponential distribution, with mean and standard deviation equal to $\ddot{s}^*$,

$$p(s^*) = \frac{1}{\ddot{s}^*} e^{-s^*/\ddot{s}^*}. \quad (7)$$
We focus on the distribution of $s^*$ rather than $s$ purely for analytical convenience, but since mutations of large effect have a negligible impact on the asymptotic base load, the practical difference between these two distributions is not great. For this distribution, the procedures outlined in Lande (1994) can be used to show that

$$
\hat{W}_B = \int_0^\infty p(s)[1 - 2su/F]^{Ku/s}ds
$$

$$
\approx \exp\left(-\mu\sum_{i=1}^{\infty}(i + \theta)^2\right),
$$

where $\theta = 1/(4Ks^*)$.

With an exponential distribution of $s^*$, $\hat{W}_B$ is lower than that expected with constant $s$ unless $Ks$ is less than 0.1 or so (fig. 1). The reason for this is that, with the exponential distribution, a substantial fraction of mutations may have effects that are effectively neutral (i.e., $s < 1/[4K]$), even though the average mutation has a large enough effect to prevent fixation. On the other hand, for very small populations, variable effects can lead to increased fitness because a fraction of the mutations are selected against, whereas all would be effectively neutral with constant small $s$.

### Recessive Mutations

Estimation of the asymptotic fitness resulting from fixation of recessive mutations in the founders is complicated by two issues. First, since the equilibrium frequency of partially recessive mutations, $q = u/(2hs)$, is greater than that for mutations with additive effects, we are less justified in assuming that mutant alleles in the founder population are always in single copies. Second, a simple expression for the fixation probability of a deleterious recessive mutation does not exist. The development of a strategy for dealing with recessive mutations is desirable, since, as we have seen above, there is compelling evidence that deleterious alleles are partially recessive.

A general approach to the problem uses a transition-matrix approach to describe the complete probability distribution for the frequency of mutant alleles (Lynch et al. 1995). Following Ewens (1979, p. 19), for a population of $K$ individuals, we see that the transition probability from a state of having $i$ mutant alleles at a locus in generation $t-1$ to a state of having $j$ mutant alleles in generation $t$ is

$$
P_{ij} = \binom{2K}{j} \eta^j(1 - \eta)^{2K-j},
$$

where

$$
\eta = \frac{(1 - 2s)^2 + (1 - 2hs)(2K - i)}{(1 - 2s)^2 + 2(1 - 2hs)(2K - i) + (2K - i)^2}.
$$
FIG. 3.—Expected fitness in an isolated population relative to that in an effectively infinite ancestral population, after all of the segregating loci in the founders have become homozygous because of drift and selection. The effective population size is assumed to equal $K$, the deleterious alleles in the founders are recessive with $h = 0.3$, and new mutations in the isolated population are ignored. Results are given for situations in which the effective numbers of lethals per gamete are $B = 1.0$ (solid lines) and 0.5 (dashed lines), and $u =$ in these two cases, when all mutations are purged from the population, the relative fitnesses are 4.48 and 2.12, respectively. The symbols denote results obtained from computer simulations, averaging over 256 replicate populations.

For any locus, there are $2K + 1$ possible states of the population, ranging from complete loss ($i = 0$) to complete fixation ($i = 2K$) of the mutant allele. Letting $x(t)$ be the $(2K + 1) \times 1$ column vector describing the expected number of loci in the $(2K + 1)$ states at time $t$, then the expected distribution for the population in the following generation is

$$x(t + 1) = Px(t),$$

where $P$ is the $(2K + 1) \times (2K + 1)$ matrix of transition probabilities.

To obtain the initial expected state of the founder population, $x(0)$, we require a method for distributing ancestral deleterious mutations over loci. Given $B$, $u$, and $h$, the expected number of polymorphic loci is defined by equation (2). We assumed that the number of mutant genes per locus in the founder population is Poisson distributed with parameter $2Kq$. The expected number of mutant alleles that ultimately become fixed was then obtained by iterating equation (10) until the expected number of segregating loci was negligible.

Results are given in figure 3 for the cases in which the numbers of lethal equivalents per gamete are $B = 0.5$ and 1.0 and the genic mutation rate is $u =$
10^{-4}. The asymptotic fitnesses are given relative to the expectation in the ancestral population, $e^{-\mu}$, where $\mu$ is given by equation (3) once $B$ and $h$ have been defined. As in the case of mutations with additive effects, the results from computer simulations are in excellent agreement with the theory. Unlike the situation with mutations with additive effects, the fixation of deleterious recessive alleles can cause an expected depression in the fitness of a small population. However, unless the selection coefficient is very small, such a reduction in fitness appears to be unlikely for populations larger than a few dozen individuals. Populations with effective sizes larger than this experience a net purging of the mutational load in the founder individuals.

ESTABLISHMENT OF THE NEW LOAD OF SEGREGATING MUTATIONS

When considering the consequences of deleterious mutations on a longer time frame, we realize that the purging or fixation of the ancestral mutation load by no means gives a complete picture of the consequences of segregating mutations. While the deleterious mutations inherited from the ancestral population are going to fixation or being eliminated, new ones are appearing each generation, and the population eventually reestablishes a new load of segregating mutations defined by its own population size.

The development of the new load of segregating mutations can be approximated by considering the cumulative effects of all cohorts of segregating mutations that have arisen since the founding event (Lynch et al. 1995). Each generation, an expected $\mu K$ new mutations are introduced into the population. Making the reasonable assumption that each mutation arises at a unique locus, they each have an initial frequency of $1/(2K)$. Thus, the distribution for the frequency of mutant alleles that arose $n$ generations in the past can be obtained by using the TM approach starting with the vector $x(0)$ with elements $x_i(0) = \mu K$ and $x_{i+1}(0) = 0$, where the subscript on $x$ denotes the number of copies of the mutant allele in the population.

The mean single-locus fitness associated with a mutation whose current frequency is $i/(2K)$ is

$$w(i) = 1 - \frac{si[2h(2K - i) + i]}{2K^2}.$$  \hspace{1cm} (11a)$$

The fitness resulting from the cohort of segregating mutations that arose $n$ generations in the past is then

$$\overline{W}_{s,n} = \prod_{i=1}^{2K-1} w(i)^{x(i)} ,$$  \hspace{1cm} (11b)$$

assuming gametic-phase equilibrium, and the total fitness effect of all mutations that have occurred since the founder event and are still segregating is

$$\overline{W}_s(t) = \prod_{n=1}^t \overline{W}_{s,n} .$$  \hspace{1cm} (11c)$$
As $t \to \infty$, $\bar{W}_i(t)$ converges on an equilibrium defined by the population size ($K$) and the mutational parameters ($\mu$, $s$, and $h$).

An example of the evolution of mean fitness resulting from the loss and fixation of mutations in the founder individuals and the recurrent introduction of new mutations is given in figure 4 for a population of 32 individuals. Using the mutational parameters outlined in the legend, the mean fitness of the population starts out at $\bar{W}_0 = 0.47$ owing to the inheritance of deleterious mutations in the founder individuals. A substantial fraction of this initial load is gradually purged from the population, but enough fixations occur to permanently reduce the mean population fitness by about 20%. From the outset, new mutations accumulate, gradually creating a new fitness associated with segregating mutations, which eventually stabilizes at a value only slightly less than $\bar{W}_0$. Thus, excluding the fixation of new mutations (to be discussed below), the product of the fitness resulting from the fixation of old mutations and the segregation of new ones asymptotes at approximately 0.32 at approximately 30 generations.

Elsewhere (Lynch et al. 1995), we present some analytical results that yield a close approximation to the equilibrium load of segregating loci for the situation...
TABLE 1
EXPECTED SEGREGATIONAL FITNESS UNDER DRIFT-MUTATION-SELECTION BALANCE

\[ s = 0.01 \quad s = 0.05 \]

<table>
<thead>
<tr>
<th>K</th>
<th>Observed</th>
<th>TM</th>
<th>Eqq. (12)</th>
<th>Observed</th>
<th>TM</th>
<th>Eqq. (12)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>0.94</td>
<td>0.94</td>
<td>0.92</td>
<td>0.74</td>
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**Note.**—Results in columns labeled *Observed* were obtained by computer simulation. These results were obtained with both \( R = 10 \) and \( 1,000 \), but to two decimal places, the same results were obtained with both values of \( R \) (except in the cases of \( R = 10, K = 16 \), in which no equilibrium segregational load is attained because of the rapid decline in population size). TM, Results obtained by use of the transition-matrix approach. The mutations have additive effects, and the genomic mutation rate is \( \mu = 1.0 \).

in which mutations have constant additive effects. Noting that \( \mu K \) mutations enter the population each generation, and letting \( t_a \) be the mean time to absorption (loss or fixation) of a mutant allele, the equilibrium number of segregating loci in a population is \( \mu K t_a \). During its sojourn through the population, a mutant allele causes a total (cumulative) load \( L \). Dividing this by the time to absorption yields the average load per generation. Thus, the equilibrium fitness associated with segregating loci is approximately

\[
\bar{w}_S = \left( 1 - \frac{L}{t_a} \right)^{\mu K t_a} \approx (1 - L)^{\mu K}.
\]

(12a)

The cumulative load is closely approximated by

\[
L \approx \frac{(1 - \mu_k)[1 - (1 + \phi)e^{-\phi}] + 2\mu_k K \phi}{K}
\]

(12b)

(Lynch et al. 1995). This approach yields fairly close approximations to results obtained by computer simulation, although not as close as those obtained by iteration of the transition matrix (table 1).

Very small populations with low fecundity do not actually attain an equilibrium number of segregating loci if the mutational parameters are sufficient to cause a rapid reduction of the population size. However, for situations in which there is sufficient time for the development of such an equilibrium, \( L \to 1/K \) with increasing \( K \), and the equilibrium load \( \bar{w}_S \to e^{-\mu} \), in agreement with the predictions of infinite population theory noted above. The relationship between \( K \) and the load of segregating mutations is not monotonic (fig. 5). The equilibrium fitness associated with segregating loci, \( \bar{w}_S \), is close to one with small \( K \) owing to the reduced
MUTATION ACCUMULATION AND EXTINCTION

Fig. 5.—The expected equilibrium segregational fitness in an isolated population with genomic mutation rate $\mu = 1$ and genes with additive effects, given by the dashed lines and obtained with eqq. (12a) and (12b). The minimum segregational fitness occurs at a population size equal to approximately $1/2s$, and the fitness asymptotically approaches $e^{-\mu}$, independent of the mutational effect, with increasing population size. The solid lines are the product of the new segregational fitness and the relative fitness resulting from fixation of genes in the founder population, obtained with eq. (6); at large population sizes, this quantity equals the new segregational fitness due to the fact that all of the mutant alleles in the founders have been purged from the population. The symbols denote the equilibrium segregational fitness obtained from computer simulations.

heterozygosity resulting from small population size, and it declines to a minimum at an intermediate population size (for genes with additive effects, this population size is approximately $1/2s$), thereafter increasing to the constant $e^{-\mu}$ as $Ks > 5$ (see also Kimura et al. 1963; Crow 1992).

**FIXATION OF NEW MUTATIONS**

Although the preceding results imply that very small populations have a reduced load of segregating mutations relative to large ones, this is more than offset by a greater rate of fixation in the former. Noting that $\mu K$ new mutations appear in the population each generation, the cumulative reduction in fitness due to fixation of all mutations that have arisen subsequent to the founder event can be computed by use of the TM approach. Starting with the vector $x(0)$ with elements $x_1(0) = \mu K$ and $x_{i=1}(0) = 0$ and iterating equation (10), the fitness resulting from the cumulative fixation of all new mutations is

$$\bar{W}_F(t) = (1 - 2s)^{\eta(t)},$$

(13)
TABLE 2

FIXATION PROBABILITIES FOR DELETERIOUS MUTATIONS

<table>
<thead>
<tr>
<th>$K$</th>
<th>Observed</th>
<th>TM</th>
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**Note.**—Results in columns labeled *Observed* were obtained by computer simulation. These results are averages from simulations with both $R = 10$ and 1,000, after the segregational fitness had stabilized and during the period in which the population size remained constant at $K$. To two decimal places, the same results were obtained with both values of $R$, except in the cases of $R = 10, K \leq 16$, in which no steady-state fixation rate is attained. *TM,* Results obtained by use of the transition-matrix approach. The mutations have additive effects.

where $n_F(t) = \sum_{n=1}^{s} x_{2K+1}(n)$. Provided the population survives long enough so that the distribution of mutation-frequency classes reaches a steady state, the reduction in fitness per generation due to fixations asymptotically approaches

$$
\bar{w}_F = \lim_{t \to \infty} \left[ \frac{W_F(t + 1)}{W_F(t)} \right] = (1 - 2su_F)^{\mu K}.
$$

(14)

In polygenic systems, the efficiency of selection against deleterious alleles can be reduced by a depression in effective population size (caused by genetic variation in fitness) and by the random generation of linkage disequilibrium (Hill and Robertson 1966; Birky and Walsh 1988). Thus, for the system that we are studying, in which large numbers of deleterious genes enter the population each generation, one may question whether the single-locus diffusion approximation, equation (6), generates an accurate prediction of the fixation probability, $u_F$. In fact, equation (6) yields predictions that are very close to those obtained by the more exact TM approach and, provided $Ks < 1$, these results are very close to observations from computer simulations (table 2). However, when $Ks > 1$, the single-locus theory underestimates the true fixation probabilities, in some cases by a factor of more than two.

**TIME TO EXTINCTION**

As deleterious mutations accumulate by fixation, there is a gradual decline in the mean viability of individuals. Once the mean viability declines below $1/R$, the net reproductive rate is less than one (individuals can no longer replace themselves), and the population size begins to decline. This precipitates a synergistic
interaction between random genetic drift and mutation accumulation, which we refer to as a mutational meltdown—as mutations continue to accumulate, the population size becomes smaller and smaller, which progressively increases the probability of fixation of future mutations by random genetic drift. Relative to the time it takes to decline to this critical viability level, the length of the meltdown phase is generally quite short. Thus, the expected time to extinction is expected to be similar to the time that it takes for the average realized fecundity to decline from $\bar{W}_0 R$ to $\bar{W}(t) R = 1$, where $\bar{W}(t) = \bar{W}_0(t) \bar{W}_8(t) \bar{W}_F(t)$ is the expected viability at time $t$ resulting from the cumulative effects of segregating and fixed mutations arising both prior to and subsequent to the founder event.

Letting $x(0)$ be the $(2K + 1) \times 1$ vector for which the $i$th element is the number of loci in the founder population with deleterious gene frequency $i/(2K)$ and $x_M$ be the mutational vector whose elements are all zero except $x_{M,i} = \mu K$, then the number of loci with different gene frequencies in generation $t$ is obtained by iterating

$$x(t + 1) = Px(t) + x_M.$$  \hspace{1cm} (15)

The expected fitness in generation $t$ is

$$\bar{W}(t) = \prod_{i=1}^{2K} w(i)^{x(i)},$$  \hspace{1cm} (16)

and the mutational meltdown initiates when $\bar{W}(t = t_M) < 1/R$.

The mean time to extinction can be approximated in the following manner. Prior to the meltdown ($t < t_M$), the population size remains very close to $K$, and the probability of extinction in generation $t$ is simply the product of the probability that the population has not gone extinct previously and the probability that all $RK$ progeny die in the current generation:

$$p_E(t) = [1 - P_E(t - 1)][1 - \bar{W}(t)]^{RK},$$  \hspace{1cm} (17a)

where

$$P_E(t - 1) = \sum_{n=1}^{t-1} p_E(n)$$  \hspace{1cm} (17b)

is the cumulative extinction probability to generation $(t - 1)$. Once the meltdown phase has been entered, we assume that the rate of fitness decline per generation remains approximately constant at $w_M = \bar{W}(t_M)/\bar{W}(t_M - 1)$. Thus, for $t > t_M$,

$$p_E(t) = [1 - P_E(t - 1)] \left(1 - \frac{w_M^{-t_M}}{R} \right)^{Kw_M^{-t_M}}.$$  \hspace{1cm} (17c)

The mean time to extinction is predicted by

$$\hat{t}_E = \sum_{t=1}^{\infty} p_E(t) \cdot t.$$  \hspace{1cm} (18)
For issues in conservation biology, the distribution of extinction times can be quite informative, as, for example, in predicting the probability that a population will go extinct within a specific time frame. The variance of extinction time is a function of two factors. First, demographic stochasticity is induced by the fact that mean viabilities are less than one (i.e., individuals die with a certain probability). We account for this by extending the previous results, focusing on the average fitness trajectory, such that

$$\sigma_D^2(t_E) = \sum_{t=1}^{\infty} p_E(t) \cdot t^2 - (\bar{t}_E)^2$$

(19a)

is the variance in extinction time due to demographic stochasticity.

Second, the stochastic nature of the fixation process results in genetic variation in fitness among replicate populations. From other results (Lynch et al. 1995), it can be shown that the variance among populations in fitness associated with segregating mutations is approximately $\mu s \sigma e^{-2sK}$. Results in Lande (1994) show that if fixations are viewed as occurring by a Poisson process, the variance in fitness among populations due to fixations is approximately $(2s)^2(\mu u K t)$ for large $t$, where $(\mu u K t)$ is the cumulative expected number of fixations by generation $t$. Since populations enter a mutational meltdown when the total number of fixations is approximately $\ln(\bar{W}_S R)/(2s)$, where $\bar{W}_S$ denotes the equilibrium fitness associated with segregating loci, the variance in fitness due to fixations is about $(2s) \ln(\bar{W}_S R)$ when the meltdown is approached. Under most conditions, the latter quantity will be substantially greater than the variance in fitness associated with segregating loci. Thus, we can expect Lande’s (1994) result on the variance due to fixations to be a close approximation to the variance in extinction time due to random genetic differences that develop among replicate populations. Modifying his earlier result, which ignored the depression in mean fitness due to segregating mutations, we obtain

$$\sigma_G^2(t_E) = \frac{\ln(\bar{W}_S R)}{2s(\mu u K)^2}.$$  

(19b)

The sum of equations (19a) and (19b) provides an estimate of the variance in extinction time.

Using mutational parameters similar to those derived for *Drosophila*, we find that simulation results for a range of carrying capacities and reproductive rates are in excellent accord with $\bar{t}_E$ predicted by the TM theory, provided the fecundity is fairly high ($R \geq 100$) (fig. 6). The fit is also quite good for lower fecundities ($R = 10$ in the figure), provided the population size is not too large. However, for $Ks > 1$ and small $R$, the transition-matrix theory can substantially overestimate the mean time to extinction. For most combinations of $R$ and $K$, the theory predicts the coefficients of variation of extinction time to a fairly good degree of accuracy (table 3). To the extent that the *Drosophila* parameters can be extended to other organisms, these results suggest that for genetic reasons alone, sexual populations with effective sizes smaller than 100 individuals are unlikely to persist.
FIG. 6.—Mean times to extinction as a function of the population carrying capacity $K$ and reproductive rate $R$. The populations have a monoecious mating system and are characterized by mutational parameters that are close to those observed in *Drosophila*: $\mu = 1.5$, $s = 0.015$, and $h = 0.35$. The mutations in the founder individuals are assumed to be distributed as described in the text, based on the assumptions that the mean number of lethal equivalents per gamete in the ancestral population is $B = 0.5$ and that the genic mutation rate is $10^{-4}$. The symbols denote results obtained by computer simulations, averaging over at least 256 replicate runs. The dashed lines are the expected times to extinction derived by iteration of the transition matrix and solving eq. (18).

TABLE 3

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<th>Predicted</th>
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Note.—Results in columns labeled *Observed* were obtained by computer simulation. Results in columns labeled *Predicted* were obtained by use of the transition-matrix approach and eqq. (16a) and (16b) in the text. The number of lethal equivalents per gamete in the base population was assumed to be $B = 0.5$ with a per-locus mutation rate of $u = 10^{-4}$ to generate the distribution of mutations in founder individuals. All mutations had effects $s = 0.015$ and $h = 0.35$, and the genomic mutation rate was $\mu = 1.0$. Simulation results are based on 256 or more replications.
for more than a few hundred generations, especially if the fecundity is relatively low.

Since human activities, such as the release of chemical mutagens, have very likely inflated the mutation rates of some species above historical levels, it is of interest to evaluate the consequences of a change in \( \mu \) for the risk of extinction. Recall that the equilibrium fitness from segregating loci is generally \( e^{-\mu} \) for large populations and that small populations suffer from a substantial rate of fixation of deleterious mutations. These points imply that rapid extinction will occur, regardless of population size, when \( \mu > \ln(R) \), the point at which the load due to recurrent segregating mutations exceeds the demographic potential of the population. The results in figure 7, where \( R = 10 \), show that this is indeed the case—the expected extinction times for populations of different sizes converge rapidly above a genomic mutation rate of approximately 2.3.

For populations that are large enough and fecund enough to achieve an equilibrium fitness associated with segregating mutations of approximately \( e^{-\mu} \), the time to enter a mutational meltdown can be shown to be approximately

\[
\hat{t}_M = \frac{\ln(R) - \mu}{2s\mu u_F K}
\]

(from eq. [7] in Lynch et al. 1995). This shows that for low genomic mutation rates, there is an approximately inverse relationship between \( \mu \) and the mean time to extinction; that is, a doubling of the deleterious mutation rate reduces the expected population longevity by approximately 50% (fig. 7).

Finally, we note that all of the preceding results assume that the only risk of extinction is the load due to deleterious mutations. Natural populations experience numerous additional forms of demographic and environmental stochasticity, which through the periodic depression of population size below \( K \) can have substantial synergistic effects on the vulnerability of a population to a mutational meltdown. We consider the joint effects of both types of stochasticity in figure 8, where computer-simulation results are given for monogamous populations subject to intergenerational variation in the carrying capacity, which is assumed to be lognormally distributed without temporal autocorrelation. For these populations, \( R \) is assumed to be constant, and the only source of demographic stochasticity is the random fluctuation in the sex ratio. To draw comparison with monocious populations with the same total productivity, we also assume that monogamous populations have twice the \( R \) per female as monocious individuals. Because density dependence is assumed to occur without respect to gender in our simulations, the expected number of mating pairs is less than \( K/2 \), the effective population size being approximately \( 1 - \sqrt{2/(\pi K)} K \) (Lynch et al. 1995).

Fluctuations in the sex ratio alone cause a large depression in the mean time to extinction, approaching a factor of two in large populations (fig. 8). For monogamous populations, the addition of only a moderate amount of stochasticity in \( K \) causes little change in \( \hat{t}_F \) when the average \( K \) is small because demographic stochasticity is the predominant source of extinction (fig. 8). However, for large average \( K \), moderate amounts of environmental variation in \( K \) can cause a several-fold reduction in \( \hat{t}_F \).
Our results extend earlier work (Lande 1994; Lynch et al. 1995) in demonstrating that the accumulation of spontaneously arising mutations can pose a substantial threat to the survival of small populations. For populations with long-term effective sizes below 100 individuals, a substantial load of deleterious mutations can be expected to develop within a few dozen generations. Because much of this load is due to the fixation of mutations, it may be essentially irreversible on timescales of several hundreds to thousands of generations. For small populations of low-fecundity species, such as birds and mammals, the expected cumulative build-up of deleterious mutations appears sufficient to lead to extinction within a few tens to hundreds of generations. Thus, because the effective sizes of natural populations are often on the order of a tenth to a third of the actual number of breeding adults (Heywood 1986; Lande and Barrowclough 1987; Briscoe et al. 1992), avoidance of extinction by a mutational meltdown over time periods of 100 or more generations appears to require the stable persistence of well over 1,000 breeding adults per generation.

Although this conclusion is based largely on computer simulations, it is likely to be on the conservative side, since we have not allowed for variable mutational effects and have not, for the most part, incorporated demographic or environment-
Population Size, $K$

**Fig. 8.**—Mean times to extinction as a function of the population carrying capacity $K$ for monogamous populations. In these simulations, individuals surviving to maturity pair randomly and permanently; because of random variation in the sex ratio, this causes the number of mating pairs in each generation to be less than or equal to half the current carrying capacity. The carrying capacity is assumed to be lognormally distributed over generations, without temporal autocorrelation; the coefficients of variation (CV) of these distributions are denoted in the plot. In all generations, $R = 10$, and the mutational parameters are the same as those given in fig. 6. The results were obtained by computer simulation, averaging over at least 256 replicate runs. For comparative purposes, the results for monoecious populations with constant carrying capacity are given by the open circles and dashed line.

Stochasticity, both of which tend to exacerbate the mutation-accumulation process (Lande 1994; Lynch et al. 1995). A variable spectrum of mutational effects increases the vulnerability of a population to mutation accumulation by increasing the range of population sizes within which some deleterious mutations are immune to elimination by natural selection, that is, by increasing the fraction of mutations for which $s < 1/(2K)$. Lande (1994) found that reasonable distributions of mutational effects can reduce the mean time to extinction by orders of magnitude relative to the situation in which mutations have fixed effects. Sources of demographic and environmental stochasticity increase the rate of fixation of deleterious mutations through a reduction in the effective population size (fig. 8).

Our results provide no evidence for the existence of a threshold population size beyond which a population is completely invulnerable to a mutational meltdown. However, the mean time to extinction via a mutational meltdown ($\bar{t}_E$) does increase rapidly with increasing population carrying capacity ($K$). For very small $K$ (<30 or so), $\bar{t}_E$ increases very rapidly, but at a decreasing rate, with $K$ (fig. 6). Beyond this point, $\bar{t}_E$ increases at a slowly increasing rate with $K$. This behavior
is largely due to the exponential decline in the fixation rate of a deleterious mutation with increasing $K$.

On the other hand, the mean time to extinction is relatively insensitive to changes in a population’s reproductive capacity. For $R > 50$ or so, $\bar{t}_E$ increases with the logarithm of $R$ (fig. 6). This slow scaling can be understood by considering the situation for populations that survive long enough to reach an equilibrium fitness associated with segregating loci ($\bar{W}_S$). For such populations, the mutational meltdown is entered when the mean viability due to fixed mutations declines to $1/(\bar{W}_S R)$. Letting $\bar{W}_F$ be the fitness due to $T$ generations of fixed mutations, the approximate time to the mutational meltdown is $-\ln (\bar{W}_S R)/\ln(\bar{W}_F)$ generations (Lande 1994; Lynch et al. 1995). Thus, a doubling of the carrying capacity of a population will generally have a greater impact on the risk of extinction than a doubling of the reproductive rate.

Over the past several years, the United States government has avoided bestowing protected status on animal species until their population sizes have dwindled to an average of 1,000 individuals (Wilcove et al. 1993). Many animal species have not been formally recognized as endangered and/or threatened until their numbers declined to a few tens or hundreds, and the average population size of plants at the time of listing is only 100 individuals. Our results imply that even if management plans, such as captive breeding programs, can secure such small populations from short-term risks of demographic and/or environmental stochasticity, without an expansion to larger population sizes, they will have a high likelihood of experiencing substantial mutational degradation within one or two centuries, the usual time frame within which most conservation policies focus (Franklin 1980; Soulé 1980; Soulé et al. 1986; Mace and Lande 1991). Thus, the short-term management of populations at densities below several hundred individuals virtually guarantees a need for even more intensive management for survival in the future.

Some types of management programs may actually exacerbate the mutation-accumulation process beyond that described in this article. For example, for captive populations living in benign environments (with services from dieticians and veterinarians, artificial insemination, etc.), mutations that would be deleterious in natural settings may be rendered nearly neutral. This speculation is supported by recent work that shows that the fitness of mutation-accumulation lines of Drosophila melanogaster relative to controls is greatly depressed in harsh environments (A. Kondrashov and D. Houle, unpublished data). It is also conceivable that adaptation to a captive environment may selectively advance alleles that are ordinarily selected against in nature (Frankham and Loebel 1992). In either case, the accumulation of mutations (deleterious only in the wild) would be expected to occur at the neutral rate, $\mu/2$ per generation, or faster, and their deleterious effects would go largely undetected until the population was reintroduced into the wild.

Considerable attention has recently been given to the idea that the genetic integrity of small captive populations might be improved by purging the genetic load through intentional inbreeding (Templeton and Read 1983, 1984; Foose et al. 1986; Ralls and Ballou 1986; Hedrick and Miller 1992; Frankham et al. 1993;
Hedrick 1994). However, the debate on this issue has focused entirely on the deleterious mutations inherited from an ancestral population. Our results show that, even without intentional inbreeding, much of the genetic load of a founder population can, in fact, be purged by natural selection, provided the effective population size is at least a few dozen individuals. However, a solitary focus on ancestral mutations can be misleading. Because of the high genomic rate of deleterious mutation, the elimination of deleterious ancestral genes by selection generally will not proceed much more rapidly than their reintroduction by mutation. Moreover, the intentional bottlenecking of a population can only enhance the probability of fixation of mildly deleterious mutations. Thus, at best, management programs intended to purge the ancestral mutation load can only have transient effects. In the long run, maximization of the effective population size appears to be the only reliable means of minimizing the rate of accumulation of deleterious genes in a population.

Specific quantitative details of the conclusions in this article rest on the assumption that the average effects of spontaneous mutations observed in Drosophila are representative of those in other species, validation of which will require additional empirical study. As noted above, if the mean mutational effect is less than that observed in Drosophila, and/or the distribution of mutational effects is highly skewed toward the effectively neutral range, then the range of population sizes that is vulnerable to a mutational meltdown is expected to expand and the mean time to extinction may decline dramatically (Lande 1994; Lynch et al. 1995). An elevation of the genomic mutation rate, resulting, for example, from environmental mutagens, will also increase the vulnerability of a population to a mutational meltdown, although the magnitude of this effect will depend on the spectrum of fitness effects of induced relative to spontaneous mutations.

Unfortunately, there is very little in the empirical literature that can be drawn upon as evidence in support of the mutational meltdown model. To our knowledge, no studies have been done on the population-level consequences of spontaneous deleterious mutations. However, in the 1950s and 1960s, several long-term irradiation experiments were performed with populations of fruit flies and mammals (Sankaranarayanan 1964, 1965; Green 1968; Nöthel 1987; Wallace 1991). A consistent feature of these experiments was that although irradiation caused an immediate and very substantial decline in mean fitness, upon relaxation of the treatment, mean fitness rapidly returned to nearly pretreatment levels. These observations are not too surprising, since the population sizes and time spans involved in virtually all of the experiments were such that few fixations of deleterious mutations were likely to occur. For example, in the most detailed Drosophila experiments, those of Sankaranarayanan (1964, 1965), the cage populations typically contained 600–800 adult flies, and the relaxation experiments were all run within 20 generations of the initiation of irradiation.

Sankaranarayanan’s (1964) results provide support for the idea that the low fitness of the irradiated populations is due largely to the segregation of recently derived mutations and that the rapid recovery upon cessation of mutation pressure is due to the efficient purging of the deleterious alleles by natural selection. For populations regularly irradiated with 2,000, 4,000, and 6,000 r of X-rays/
generation, he found that egg hatchabilities quickly stabilized at 0.682, 0.447, and 0.290 relative to controls. Assuming that these fitness measures reflect the equilibrium fitness effects of segregating loci, caused by the balance between recurrent mutation and natural selection, and setting them equal to $e^{-\mu}$, we find estimates of the induced genomic mutation rate for the three treatments are $\mu = 0.38$, $0.80$, and $1.24$. Noting that only males were irradiated in these experiments, we believe these results are quite consistent with the hypothesis that the genomic deleterious mutation rate for egg hatchability increases by approximately 0.8 for every 2,000 r of X-rays/d. With the same method applied to larval-to-adult viability data, estimates of $\mu$ are 0.065, 0.129, and 0.196, which implies that the genomic deleterious mutation rate for this fitness component increases by approximately 0.13 for every 2,000 r of X-rays/d. For both sets of results, the exponential decline of fitness with X-ray dosage is strikingly consistent with the theory for the equilibrium fitness effects of segregating loci that we used above.

Longer-term experiments than these, with or without mutagens, over a range of population sizes will be useful as a means of validating the basic qualitative properties of the mutational meltdown model. Many model systems with short generation times might be profitably exploited for these purposes.

Finally, we note that because the computer simulations on which much of our work is based can be quite time-consuming, it is useful to have some analytical theory that yields good approximations to the mean time to extinction. Using the diffusion approximation for the fixation probability of a deleterious allele with additive effects, Lande (1994) obtained a simple analytical expression for the time to entry into a mutational meltdown as a function of $R$ and $K$. Because no simple expression for the fixation probability of a recessive mutation exists, this approach is not so easy to extend to recessive mutations. Moreover, it can describe the meltdown process only in populations that are large enough to settle into a steady-state fixation process well in advance of the mutational meltdown. We have shown that a transition-matrix approach provides a more general solution to the time of extinction since it incorporates segregating as well as fixed mutations and does not require the attainment of an equilibrium fixation phase (Lynch et al. 1995). In this article, we have extended the TM approach to allow for recessive mutations and to account for deleterious mutations segregating in founder individuals. For a range of population sizes, reproductive rates, and mutation rates, this approach predicts the mean and variance of the time to extinction remarkably well, which, if nothing else, provides a check on the validity of our simulation results.

However, the TM approach also has some significant limitations. First, when the product $Ks \geq 1$, the single-locus theory begins to underestimate the fixation probabilities of deleterious mutations and hence to overestimate the mean time to extinction (fig. 6). Elsewhere (Lynch et al. 1995), we have argued that this phenomenon is due to the fact that simultaneously segregating mutations interfere with each others' elimination by natural selection (Hill and Robertson 1978; Birky and Walsh 1988). Single-locus theory assumes that the dynamics of individual mutations are independent of the genetic background. In a somewhat different context, directional selection for favorable mutations, Keightley and Hill (1983)
also found a discrepancy between the observed fixation rate and that predicted by single-locus theory, which increased with increasing population size. This effect appears to be most pronounced when mutually segregating mutations have identical effects (Hill and Robertson 1966), the situation in our simulations. Thus, it is plausible that the single-locus theory will yield more accurate predictions when mutations are allowed to have variable effects. Second, extension of the TM approach to situations in which population size and/or sex ratio are fluctuating due to demographic and/or environmental stochasticity raises a number of technical difficulties (e.g., the need to generate separate probability distributions of alleles in each of the sexes and the need to stochastically vary the dimensions of the transition matrix), which, although not insurmountable, would require considerable computational time.

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APPENDIX

TIMESCALE TO APPROACH FIXATION OF FOUNDER ALLELES

To achieve an approximate result for the timescale over which the variation in a founder population is eliminated, we use the formula for the expected dynamics of an allele's frequency under weak selection \((4Ks < 1)\),

\[
E(p_t) = p_0 - 2Ksp_0(1 - p_0) \left[ 1 - e^{-t/K} \right] + \frac{(1 - 2h)(1 - 2p_0)}{3} \left[ 1 - e^{-3t/K} \right],
\]

where \(h = 0.5\) for mutations with additive effects (Robertson 1960; Silvela 1980). Assuming that deleterious alleles in founder individuals are rare, so that \(p_0 = 1/(2K)\), and noting that the expected fitness at time \(t\) associated with founder mutations is \(W_B(t) = [1 - 2sE(p_t)]^{1/2}\), the time to reach a fraction \(x = W_B(t)/W_B\) of the asymptotic value (given by eq. [8]) is found to be

\[
t_x \approx -2\ln \left[ 1 + \frac{1 - (s/K)(1 - 2s\mu)x^{1/2}}{2s^2} \right].
\]

Solution of this equation shows that \(W_B\) is approached fairly rapidly. For example, with \(s = 0.01\) and \(\mu = 1\), equation (A2) yields \(t_{0.95} = 21\) and 65 generations with \(K = 10\) and 25.

With strong selection \((4Ks > 1)\), the change in gene frequency can be assumed to be essentially unmodified by random genetic drift:
\[ E(p_t) = 1 - \frac{1}{1 + \left[p_0/(1 - p_0)\right]e^{-st}} \]  
(A3)

(Crow and Kimura 1970, p. 192), which leads to

\[ t_x = -\frac{1}{s}\left(\frac{2K[1 - x^{s/(s+K)}]}{2s - 1 + x^{s/(s+K)}}\right) = -\frac{1}{s}\ln\left(\frac{1 - x}{\mu}\right). \]  
(A4)

With \( \mu = 1 \), equation (A4) implies \( t_{0.5} = 0.7/s \) and \( t_{0.95} = 3/s \).

LITERATURE CITED


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