On the contribution of deleterious alleles to fitness variance in natural populations of *Drosophila*

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Summary

I have studied the consequences of habitat patchiness on the persistence times of deleterious alleles in a random mating population. Results based on computer simulations and supported by analytical approximations suggest that deleterious alleles remain approximately $1/(1-2F_{ST})$ more generations in the patchy than in a comparable homogeneous population, where $0 < F_{ST} \leqslant 0\cdot25$ is the fraction of genetic variance due to the sample of families across patches in one generation. In natural populations of *Drosophila*, therefore, the contribution of deleterious mutants to the genetic variance in fitness might be larger than previously thought. A model of density-dependent viability selection, inspired by the suggestion that deleterious effects can substantially increase when the environment becomes harsher, also gives credence to the analytical results and illustrates that mean persistence times are very sensitive to changes in ecological parameters. If the density dependence model can be taken seriously, there is a clear difficulty in comparing observed and expected levels of genetic variance on the basis of the simplest mutation–selection balance model.

1. Introduction

Since the average mutation is deleterious to fitness, most newly arisen mutations will eventually be eliminated from the population. Assuming random mating and relative fitnesses of genotypes *AA*, *Aa*, and *aa* to be 1, $1-hs$, and $1-s$, respectively, where *s* is a positive constant and $0 < h < 0\cdot5$ (as is suggested by the *Drosophila melanogaster* data: Simmons & Crow, 1977; Crow & Simmons, 1983), the standard non-linear recursion equation to obtain the gene frequency of the mutant allele *a* in the next generation is:

$$q' = \frac{q - hspq - sq^2}{1 - 2hspq - sq^2}$$

(Crow & Kimura, 1970). This equation also assumes that members of the population are continuously and uniformly distributed across their spatial range. Evolutionary biologists, however, have accumulated evidence showing that many animals, particularly insects, exploit resources which are discrete, consisting of small, separate units, that sustain a few sibships (Brncic, 1966; Heed, 1968; Jaenike & Selander, 1979; Lacy, 1983; Hoffmann *et al.*, 1984; Santos *et al.*, 1989; Prout & Barker, 1989; Thomas & Barker, 1990; Jaenike & James, 1991; Santos, 1994, 1997). By

means of some algebraic and computer models, I show here that this spatial distribution of eggs or larvae may substantially increase the persistence times of deleterious alleles in the population (Crow, 1979, 1993) and, therefore, their contribution to the genetic variance in fitness might be larger than previously thought. This adds a new dimension to the long-standing, fundamental debate in evolutionary biology concerning the two most plausible mechanisms responsible for the maintenance of genetic variation in natural populations, namely mutation–selection balance and various models of balancing selection (Barton, 1990).

2. Model and analysis

Let us assume a patchy population structure, i.e. a population that exploits ephemeral resources and is not truly subdivided on the demographic time scale (Harrison & Hastings, 1996). Generations are discrete and non-overlapping. Adults mate at random and produce offspring that stay together in sib-groups throughout their early life. The environment is considered as a number of discrete patches into which adults deposit their offspring. For simplicity, Table 1 assumes that each patch is colonized by one mating pair, and each family is infinitely large so that all possible genotypes are present in their expected frequencies. Selection acts on variation in viability

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Table 1. *Recursion equations for a locus with two alleles in a random mating population under selection*

(a) *Non-structured population*

Genotype	AA	Aa	aa
Frequency	p^2	$2pq$	q^2
Relative fitness	1	$1-hs$	$1-s$

$$q' = \frac{q - hspq - sq^2}{1 - 2hspq - sq^2}$$

(b) *Distribution of sib-groups when a single randomly mated (monogamous) female lays eggs in spatially distributed patches*

		Progeny			
Mating pair	Frequency	AA	Aa	aa	Frequency after selection (q')
$AA \times AA$	p^4	1			0
$AA \times Aa$	$4p^3q$	$\frac{1}{2}$	$\frac{1}{2}$		$\dfrac{\frac{1}{4}(1-hs)}{1-\frac{1}{2}hs}$
$AA \times aa$	$2p^2q^2$		1		$\frac{1}{2}$
$Aa \times Aa$	$4p^2q^2$	$\frac{1}{4}$	$\frac{1}{2}$	$\frac{1}{4}$	$\dfrac{\frac{1}{2}-\frac{1}{4}s(h+1)}{1-\frac{1}{2}s(h+\frac{1}{2})}$
$Aa \times aa$	$4pq^3$		$\frac{1}{2}$	$\frac{1}{2}$	$\dfrac{\frac{3}{4}-\frac{1}{4}s(h+2)}{1-\frac{1}{2}s(h+1)}$
$aa \times aa$	q^4			1	1

$$q' = \frac{p^3q(1-hs)}{1-\frac{1}{2}hs} + p^2q^2 + \frac{2p^2q^2[1-\frac{1}{2}s(h+1)]}{1-\frac{1}{2}s(h+\frac{1}{2})} + \frac{pq^3[3-s(h+2)]}{1-\frac{1}{2}s(h+1)} + q^4$$

only within patches. On reaching adulthood individuals leave their patches and form a single homogeneous population. I shall also assume in this section that each patch contributes equally to the adult population; thus, selection within patches is 'soft'. This means that the gene frequencies of the adults after leaving their patches are not weighted by the mean fitness of the whole population (Christiansen, 1975). The recursion equation to obtain the gene frequency of the mutant allele *a* in the next generation is given at the bottom of Table 1, and Fig. 1 shows the results of computer iterations for different strengths of selection against *a* when its initial frequency is 0·05. We can see immediately that the time taken to eliminate the deleterious allele is not only dependent on the selection intensity, but also on the population structure. Roughly speaking, the deleterious allele remains in the population about twice as many generations in the case of a spatially structured population compared with the standard result of considering a homogeneous population.

If q is small (which will indeed be the case in an infinitely large population), the change in frequency of *a* per generation in a homogeneous population equals $\Delta q_{\text{hom}} = -hsq$. In the patchy population (Table 1b), the first term in the recursion equation to obtain the gene frequency in the next generation becomes prevalent. Because experimental evidence from viability mutations in *Drosophila* suggests that the absolute effect in heterozygotes, *hs*, is about the same

(~ 0.02) for mutations that are lethal when homozygous as for the much more frequent mildly deleterious mutations (Simmons & Crow, 1977), the change in allele frequency per generation in this case can be approximated by $\Delta q_{\text{str}} = -\frac{1}{2}hsq$.

Assume that mutation from *A* to *a* occurs at a rate μ per gene per generation, and that reverse mutation can be ignored. At equilibrium, selective elimination is exactly balanced by the input of new mutations. Accordingly (if μ is small),

$$\hat{q}_{\text{hom}} \approx \mu/hs$$
$$\hat{q}_{\text{str}} \approx 2\mu/hs$$

(where the hat denotes the allele frequency at equilibrium).

What happens in essence is that selection is only acting on that fraction of the genetic variance for fitness that occurs within patches, namely $1 - 2F_{ST} = (N-1)/N$, where $0 < F_{ST} \leq 0.25$ is the standardized genetic variance and N is the effective number of locally breeding adults. (Note that the whole population is mating at random and the variance is due to sampling of families across patches in one generation, not the long-term outcome of drift.) A straightforward extrapolation suggests that, in general, the equilibrium frequency of a deleterious allele in a patchy population would be:

$$\hat{q} \approx \left(\frac{N}{N-1}\right)\left(\frac{\mu}{hs}\right) = \left(\frac{1}{1-2F_{ST}}\right)\left(\frac{\mu}{hs}\right).$$
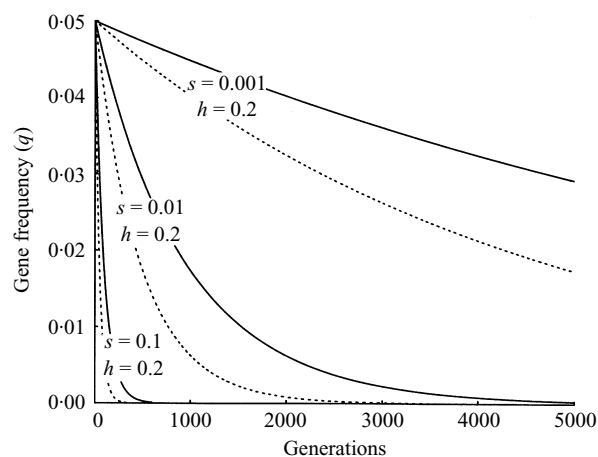
Fig. 1. Selection against a partially recessive allele (*a*) introduced at an initial frequency of $q_0 = 0.05$ in an infinitely large non-structured (homogeneous) population (dotted line), and in an infinitely large patchy population (continuous line) where as single female contributes progeny to each patch.

It is worth mentioning here that in natural populations of *Drosophila* the effect is not trivial (see below) and is likely to be even more important than the effects of permanent subdivision. In this last case, Barton & Whitlock (1997) have shown that the effective selection is reduced by $1/(1-F_{ST})$, and the magnitude of genetic differentiation on a broad geographic scale in *Drosophila* is similar to that observed among breeding sites within a local, presumably panmictic population (cf. Lacy, 1983; McCauley & Eanes, 1987; Santos, 1997).

The time scale necessary to reach the new equilibrium basically depends on the magnitude of deleterious effects and the mode of selection. Assuming independent effects on fitness (i.e. fitnesses are multiplicative across loci), the proportional decline in deleterious mutations produced in any given generation is $e^{-(hs)t}$ in the homogeneous population, and their number per diploid individual follows a Poisson distribution with mean $n = U/(hs)$, where U is the total genomic deleterious mutation rate (Kimura & Maruyama, 1966; Crow, 1970). For a stable patchy population as in Table 1b, the proportional decline would be $e^{-(\frac{1}{2}hs)t}$. With $hs \approx 0.02$ and $U \approx 1$ (Mukai *et al.*, 1972; Charlesworth *et al.*, 1990; Houle *et al.*, 1992), it would carry the equivalent of about 50 generations of new mutations in a homogeneous population, and the new mean equilibrium number of deleterious mutations per individual would be twice as large (~ 100). For *D. melanogaster*, this might be attained after a few years in those areas where ecological conditions are favourable throughout the seasons.

The contributions of a deleterious allele to the additive and dominance genetic variance components in fitness in a random mating population are given by

$$V_a = 2pqs^2[(p-q)h+q],$$
$$V_d = p^2q^2s^2(1-2h)^2$$

(Mukai *et al.*, 1974). Assuming an effectively infinite homogeneous population at equilibrium ($\hat{q}_{hom} \approx \mu/hs$), the expressions are approximated by

$$\hat{V}_{a(hom)} \approx 2\mu hs,$$
$$\hat{V}_{d(hom)} \approx (1-2h)^2(\mu/h)^2$$

(Mukai *et al.*, 1974; Charlesworth, 1987). For the patchy population in Table 1b, the variance components at equilibrium can be approximated by doubling the mutation rate, namely

$$\hat{V}_{a(str)} \approx 4\mu hs,$$
$$\hat{V}_{d(str)} \approx 4(1-2h)^2(\mu/h)^2,$$

and their ratio, $R = \hat{V}_{d(str)}/\hat{V}_{a(str)}$, would be twice that obtained for the homogeneous population. If, however, $\mu \approx 10^{-5}$ and the average values of h and s for mildly deleterious mutations affecting *Drosophila* viability in natural populations are ~ 0.2 and ~ 0.03, respectively (Crow & Simmons, 1983; Hughes, 1995); R only increases from 0.0075 to 0.015 and the bulk of the variation contributed by mutation would still be additive rather than dominance.

3. Veracity of the assumptions

Two basic assumptions in the model are that adults mate at random with respect to patch of origin, and that selection is soft (i.e. that density regulation occurs within each patch). The empirical evidence available in *Drosophila* clearly indicates that wild females and males engage at random with respect to the traits studied (Gromko *et al.*, 1980; Partridge *et al.*, 1987; Quezada-Díaz *et al.*, 1992; Santos *et al.*, 1992; Barbadilla *et al.*, 1994). On the other hand, there is also evidence that competition mainly occurs in the larval stage in natural populations of *Drosophila*. Thus, Grimaldi & Jaenike (1984), and Quezada-Díaz *et al.* (1997), have shown that up to 5 times as many flies from a particular species emerged from supplemented halves of natural breeding sites compared with the numbers obtained from the control, non-supplemented halves.

Another assumption in the model is that only a few females lay eggs on a patch. The situation depicted in Table 1b is clearly a simplification of the breeding structure of actual populations of *Drosophila*. However, the estimates of the standardized variance (F_{ST}) among breeding sites based on allozyme variation from several *Drosophila* species strongly suggest that few sibships usually grow together in the same habitat patch (Santos, 1997). Additional information comes from the empirical distributions of adult *Drosophila* emerging from natural resources. They show that females usually aggregate their eggs and larvae over patches (Atkinson & Shorrocks, 1984; Rosewell *et al.*, 1990; Kreitman *et al.*, 1992). Aggregation can be measured as the variance to mean ratio or by means of the clumping parameter (*k*) of the negative binomial (Bliss & Fisher, 1953). As $k \to \infty$, the negative binomial

converges to the Poisson distribution and this would mean that females oviposit at random over the available substrates (in practice, values of k above about 10 indicate a random distribution: Atkinson & Shorrocks, 1984). A value of k ranging from 0·5 to 1·5 seems to be representative in many cases (Rosewell *et al.*, 1990). We would expect most patches to be colonized by very few females, and strong competition among larvae in those patches where a relatively high number of females lay their eggs (see Grimaldi & Jaenike, 1984; Quezada-Díaz *et al.*, 1997). There is, in addition, the potential for strong genotype–environment interaction in the magnitude of harmful effects of the mutant alleles (see below).

Because the available experimental information on mutational variance and allelic effects on fitness comes basically from *D. melanogaster*, it might be interesting to know the degree of aggregation in this species. From field experiments in a fruit market, Rosewell *et al.* (1990) found that k ranges between 0·73 and 4·34 depending on the food. Nunney (1990), working under orange trees, found that *D. melanogaster* exhibited very little aggregation ($k \gg 1$), whereas M. Santos, K. T. Eisses & A. Fontdevila (unpublished observations), working under *Opuntia ficus-indica* cacti (prickly pear), found that $k < 1$. It is probably highly unrealistic to assume that the parameter k is constant in any population, and the spatial contagion in the activities and breeding habits of individuals is likely to be dependent on the degree of patchiness and resource abundance.

The model also assumes that selective values are constant across patches, i.e. that they are independent of the local density of larvae. Density-independent selection in density-regulated populations would buffer the evolution of allele frequencies from changes in population densities (e.g. Prout, 1980). Kondrashov & Houle (1994; see also Fry *et al.*, 1996), however, have recently suggested that a harsh environment may cause an increase in the magnitude of deleterious effects of all mutations, including those which are roughly neutral under benign conditions. On the other hand, Fernández & López-Fanjul (1997) have failed to find conditional quasi-neutral mutations (quasi-neutral in good environments and deleterious in bad ones), but their experimental procedure fosters the elimination of unconditional deleterious alleles and, therefore, underestimates mutational heritabilities (see also Fernández & López-Fanjul, 1996). In a patchy population the quality of the environment experienced by an individual is determined not by global population averages but by the parameter values in each patch. If global population density is limited by strong intraspecific competition in patches with high local density (Kreitman *et al.*, 1992) then, according to Kondrashov & Houle (1994), some mutations could be essentially neutral in low-density patches ('benign' environments) and largely deleterious in high-density ones ('harsh' environments). In this case, selection against deleterious mutants would be most intense at low resource levels.

4. Computer simulations

The purpose of the Monte Carlo simulations was to check the validity of the approximate results outlined above, and to evaluate the consequences of a patchy population structure on the selective elimination of a deleterious mutation under different situations in a finite population. MATLAB (1992) m-files to carry out the computations are available on request.

(i) *Density-independent selection*

A randomly mating population of diploid individuals of actual size $K = 1000$ was considered in all simulations. It was assumed that each family was infinitely large so that all possible genotypes at the zygotic stage before selection were present in their expected frequencies. The initial frequency q_0 of the deleterious allele was always 1×10^{-3}, and selection acted on viability only. After selection, 1000 randomly sampled individuals (500 females + 500 males) were chosen as the parents for the next generation. In each run, I computed the total number of mutant heterozygotes before the deleterious allele was eliminated from the population, as well as its extinction time (Li & Nei, 1972). In an infinite population the two quantities are equivalent, but in a finite population the mean time to loss is considerably shorter than in an infinite population (Kimura & Ohta, 1969). I will follow Crow (1979, 1993) in calling the first quantity the persistence time. It can be shown to be equal to V_G/V_M, namely the ratio of the equilibrium or standing genetic variance ($V_G = V_M/hs$: Barton, 1990) to the additive genetic variance introduced by mutation each generation ($V_M = U(hs)^2$). The amount of genetic variance at equilibrium under mutation–selection balance models is, therefore, directly related to the average persistence times of deleterious mutations.

For each set of parameter values (hs and s), the program was run for 1000 replicate samples, and the two quantities just described were stored so that their distributions could be calculated. Two situations were simulated: a homogeneous population for comparisons, and a patchy population with constant numbers of breeding females per patch ($n_f = 1$, 2 or 5).

(ii) *Density-dependent selection*

As discussed above, Kondrashov & Houle (1994) observed that the relative fitness against deleterious alleles can change by more than one order of magnitude as the environment becomes harsher. To evaluate their findings in the present context a simple form of density dependence followed juvenile production and selection each generation.

In constructing the density-dependent selection model I considered only the larval stage, and assumed that the environment is more severe as the larval density increases within a patch. Let $N(t)$ be the number of viable eggs laid by one or more females on a patch. Also, assume competition among larvae such that the per capita viability, $s(N(t))$, is a decreasing function of $N(t)$. The form of $s(N(t))$ used here is:

$$s(N(t)) \propto [1 + \Phi N(t)]^{-b},$$

where the parameter Φ characterizes the patch density at which density-dependent effects become important, and b describes the form of density-dependent mortality arising from competition within the patch. This function is taken from the ecological literature as a popular description of competition (see Hassell & Comins, 1976; Goodfray *et al.*, 1992). Larval viability drops more rapidly with increasing $N(t)$ when the parameter b is large, implying severe competition at high densities. For three genotypes, the recursion equations for the total numbers of adults emerging from a patch as a function of the number of larvae can be written as (see Dytham & Shorrocks, 1995):

$$N_{AA}(t+1) = \lambda N_{AA}(t)[1 + \Phi(N_{AA}(t) + \alpha_{ij}N_{Aa}(t) + \alpha_{ik}N_{aa}(t))]^{-b},$$

$$N_{Aa}(t+1) = \lambda N_{Aa}(t)[1 + \Phi(N_{Aa}(t) + \alpha_{ji}N_{AA}(t) + \alpha_{jk}N_{aa}(t))]^{-b},$$

$$N_{aa}(t+1) = \lambda N_{aa}(t)[1 + \Phi(N_{aa}(t) + \alpha_{ki}N_{AA}(t) + \alpha_{kj}N_{Aa}(t))]^{-b},$$

where $N_{AA}(t)$, $N_{Aa}(t)$ and $N_{aa}(t)$ are the numbers of larvae of each genotype in a patch, λ is the net reproductive rate (assumed to be equal for all three genotypes), and α_{ij}, α_{ik}, etc., are competition coefficients. The parameter Φ is equal to $(\lambda^{1/b} - 1)/N^*$, where N^* is the local equilibrium density. A value of 25 for the parameter b, describing scramble competition among larvae for food (Nicholson, 1954), was chosen in all simulations. I have also assumed that each female lays 10 viable eggs (i.e. $\lambda = 5$) in a single patch. The fitnesses of the three genotypes relative to that of the homozygous AA are:

$$w_{AA} = 1;$$

$$w_{Aa} = \frac{N_{Aa}(t+1)/N_{Aa}(t)}{N_{AA}(t+1)/N_{AA}(t)}$$

$$= \frac{[1 + \Phi(N_{AA}(t) + \alpha_{ij}N_{Aa}(t) + \alpha_{ik}N_{aa}(t))]^b}{[1 + \Phi(N_{Aa}(t) + \alpha_{ji}N_{AA}(t) + \alpha_{jk}N_{aa}(t))]^b};$$

$$w_{aa} = \frac{N_{aa}(t+1)/N_{aa}(t)}{N_{AA}(t+1)/N_{AA}(t)}$$

$$= \frac{[1 + \Phi(N_{AA}(t) + \alpha_{ij}N_{Aa}(t) + \alpha_{ik}N_{aa}(t))]^b}{[1 + \Phi(N_{aa}(t) + \alpha_{ki}N_{AA}(t) + \alpha_{kj}N_{Aa}(t))]^b}.$$

The competition coefficients (Table 2) describe the effect of genotypes on each other. Genotype AA is competitively superior to either Aa or aa, whereas

Table 2. *Competition coefficients describing the effect of one genotype on another. Individuals of a particular genotype have an effect of 1 on individuals of the same genotype. Assuming reciprocal effects, the other interactions are described by three parameters: x, y, z*

Affected genotype	Affecting genotype		
	AA	Aa	aa
AA	1	$1/x$	$1/y$
Aa	x	1	$1/z$
aa	y	z	1

genotype Aa is superior to aa. The values chosen for the three parameters in Table 2 were: $x = 1 \cdot 06$, $y = 1 \cdot 35$ and $z = 1 \cdot 25$. These values provide biologically reasonable figures for the relative viabilities of genotypes Aa and aa in those patches were larvae develop under relatively uncrowded conditions (Simmons & Crow, 1977). For instance, Fig. 2 gives the number of adults that emerge from a patch as a function of the number of eggs. It can be seen that the fitness of genotype Aa, relative to that for the homozygous AA, decreases from $\sim 0 \cdot 98$ (i.e. $hs \approx 0 \cdot 02$) when a single female breeds in a patch, to less than $0 \cdot 74$ when 20 or more females contribute with their eggs. The magnitude of deleterious effects thus increases as the larval environment becomes more stressful, in accordance with what has been suggested by Kondrashov & Houle (1994).

In the basic routine of the model, every generation was set up with 500 available patches. The initial frequency q_0 of the deleterious allele was always 1×10^{-3} as above. In each generation, the number of breeding females per patch was obtained from a negative binomial distribution whose exponent k was the same for each genotype. The breeding individuals in each generation were randomly chosen, and the eggs were independently distributed over the available patches. Mendelian inheritance was assumed. After density-dependent selection within patches (without respect to sex), the number of adults was not rounded down to the nearest integer and so fractional individuals could survive. This avoids small changes in the number of adults produced per site possibly having an important effect on the allele frequencies.

For each set of conditions, the program was run for 1400–2000 replicate samples. In each run, the total number of mutant heterozygotes (i.e. the persistence times) before the deleterious allele was selectively eliminated from the population was stored. To proceed any further with the numerical results, we also need an estimate of the mean persistence times for the appropriate control (homogeneous) population for each set of parameter values. These numbers were obtained by averaging genotype frequencies over patches after the breeding individuals in each gen-
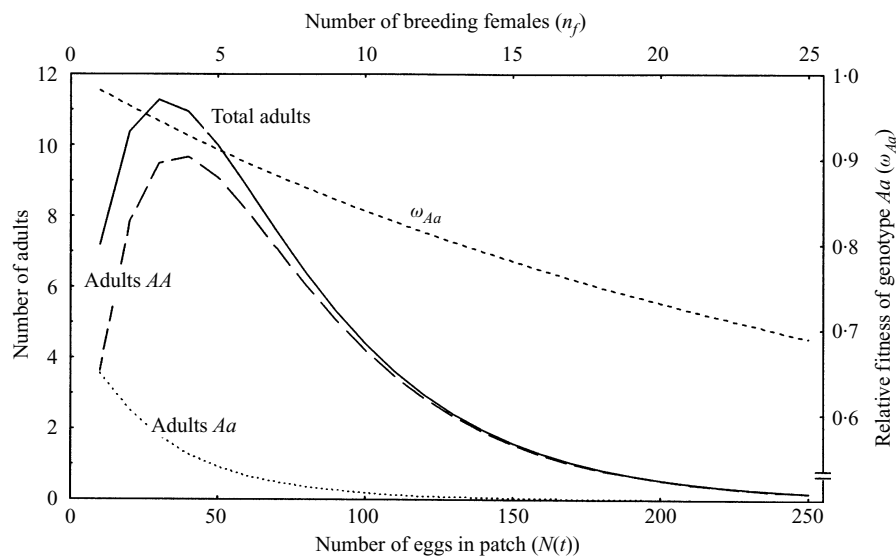
Fig. 2. Number of adults that emerge from a patch, and fitness (w_{Aa}) of larvae with genotype *Aa* relative to those with genotype *AA*, as a function of the number of eggs in the patch. The reproductive output for each breeding female is assumed to be 10 viable eggs. Because the frequency of the deleterious allele *a* will be small in a relatively large population, a single parental heterozygous fly is assumed to contribute offspring to a patch in all cases. The values used for the parameters were $\lambda = 5$, $b = 25$, and $N^* = 50$ (see text for details).

eration were randomly chosen and assuming that all patches had the same genotype input.

4. Numerical results

(i) *Density-independent selection*

From the previous analytical arguments, the ratio of mean persistence times between a patchy and an equivalent homogeneous population is expected to be equal to $N/(N-1)$ (or to $1/(1-2F_{ST})$), where N is the effective number of locally breeding adults. The simulation results in Table 3 show that this expectation

holds up reasonably well for different values of h and s. The averages of the ratios between the patchy and the homogeneous populations are 2·02 when $n_f = 1$ (i.e. $N = 2$), 1·36 when $n_f = 2$, and 1·07 when $n_f = 5$. Although I have not explored a broad range of values, these results clearly suggest an approach to the actual persistence times of deleterious alleles in patchy populations when empirical estimates of the standardized genetic variance (F_{ST}) among breeding sites can be obtained.

I have previously summarized a number of F_{ST} values based on allozyme variation across breeding

Table 3. *Total number of mutant heterozygotes affected by a deleterious allele introduced in the population at an initial frequency $q_0 = 1 \times 10^{-3}$, and number of generations to loss (in parentheses)*

			Patchy population with a constant number of founder females per patch (n_f)		
hs	s	Non-structured population	$n_f = 1$	$n_f = 2$	$n_f = 5$
0·04	0·4	47·1 (10·3)	78·3 (12·1)	60·0 (11·1)	49·5 (10·4)
0·04	0·2	49·5 (10·3)	81·3 (11·9)	57·8 (11·2)	52·5 (10·6)
0·04	0·1	56·8 (10·9)	92·7 (12·5)	52·8 (10·9)	69·7 (11·3)
0·04	0·04	49·6 (10·4)	106·1 (12·5)	67·2 (11·2)	49·8 (10·7)
0·02	0·4	80·6 (11·8)	132·6 (13·7)	118·2 (13·0)	112·2 (13·9)
0·02	0·2	80·3 (11·8)	132·9 (14·4)	170·4 (14·8)	108·3 (13·6)
0·02	0·1	95·7 (12·9)	148·1 (14·7)	115·2 (12·9)	88·4 (12·5)
0·02	0·04	81·5 (11·8)	268·1 (16·6)	127·2 (14·1)	105·1 (12·9)
0·01	0·4	133·6 (14·4)	187·8 (15·1)	139·9 (14·3)	143·6 (14·5)
0·01	0·2	144·2 (14·8)	297·0 (18·4)	183·8 (15·3)	115·7 (13·3)
0·01	0·1	154·7 (14·9)	554·8 (20·1)	204·4 (16·2)	127·4 (14·0)
0·01	0·04	195·4 (15·5)	384·3 (17·4)	312·4 (16·4)	165·7 (14·3)

The actual size of the population is $K = 1000$, and it is assumed that each family is infinitely large so that all possible genotypes at the zygotic stage before selection are present in their expected frequencies. Each value is the average of 1000 independent runs.

Table 4. *Ratio of mean persistence times (i.e. ratio of the averages of the total number of mutant heterozygotes affected by a single deleterious mutant before its selective elimination) between a patchy and a comparable homogeneous population when the deleterious allele is introduced in the population at an initial frequency $q_0 = 1 \times 10^{-3}$, and selection is density-dependent*

n_f	$k = 1$			$k = 5$		
	$N^* = 25$	$N^* = 50$	$N^* = 100$	$N^* = 25$	$N^* = 50$	$N^* = 100$
0·5	1·82	1·22	1·43	1·61	1·78	2·07
1	1·44	1·33	1·20	1·45	1·37	1·29
2	1·24	1·15	1·22	1·43	1·23	1·18
5	1·27	1·09	1·14	1·16	1·11	1·12

For each set of parameter values (average number of breeding females per patch (n_f), degree of aggregation measured by the clumping parameter of the negative binomial (k), and local equilibrium density (N^*)), the ratio is based on the averages of 1400–2000 simulations.

sites in several species of *Drosophila* (Santos, 1997). For instance, a value of 0·038 has been reported in an Australian population of *D. melanogaster* (Hoffmann *et al.*, 1984). This would suggest that deleterious alleles segregate in natural populations for approximately 10 % more generations than is usually assumed. In absolute numbers (under a mutation–selection balance model), that figure translates to approximately 5 generations for mutations affecting life-history traits, and approximately 12 generations for those affecting morphological traits (see Houle *et al.*, 1996). It may seem a small increase, but its biological importance should probably not be dismissed.

(ii) Density-dependent selection

Table 4 gives the ratios of mean persistence times between a patchy and a homogeneous population for the presumably more realistic density-dependent selection model. The values of N^* chosen in the simulations are within the range of values expected in wild drosophilids (from 10 to 500; see Shorrocks & Rosewell, 1986). As indicated above, the reproductive capacity of individuals was fixed and the population size was kept constant. Under most conditions, the ratio of the number of breeding adults to the carrying capacity of the environment was lower than 1 (from 0·128 when $k = 5$, $N^* = 100$ and $n_f = 0·5$; to 0·758 when $k = 1$, $N^* = 50$ and $n_f = 5$), indicating that the population could be sustained. However, when $N^* = 25$ and $n_f = 5$, the carrying capacity was lower than the number of breeding adults. Although in these cases the simulations were based on ecologically unrealistic assumptions, the corresponding figures in Table 4 are not likely to be very biased.

In general, the results suggest that the oviposition behaviour of females, i.e. whether they heavily aggregate their eggs or not, does not make a large
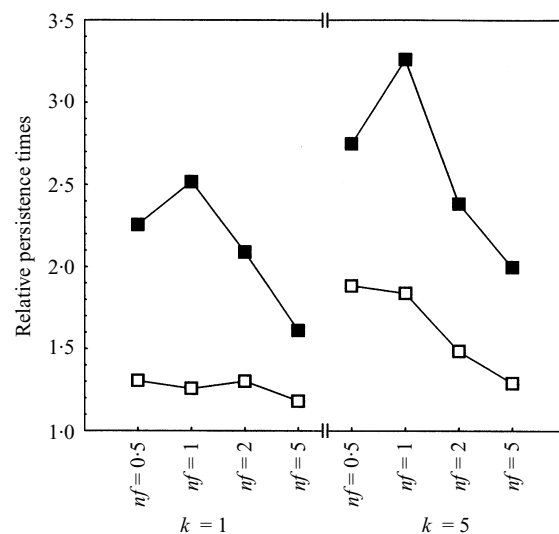

Fig. 3. Relationship between the mean persistence time of a deleterious allele in a patchy population as a function of the local equilibrium density parameter N^*. For each set of conditions (average number of breeding females per patch (n_f), and degree of aggregation measured by clumping parameter of the negative binomial (k)), the plotted symbols denote the ratios of averages obtained from the 1400–2000 replicate runs. □, Ratio: $N^* = 50/N^* = 25$; ■, Ratio: $N^* = 100/N^* = 25$.

difference to the relative persistence times. What seems to be more important, as expected, is the average number of females breeding on a single patch. However, a patchy environment might also have important additional consequences if selection is density-dependent. Thus, the number of generations a deleterious allele can segregate in the population would be very sensitive to changes in ecological parameters, and this is clearly illustrated in Fig. 3 where the ratios of mean persistence times obtained in the simulations for a patchy population are plotted as a function of N^*, the local equilibrium density.

Depending on the quantity and quality of the local resources, the coexistence of other species which use the same habitat patches, and the local density of larvae, the mean persistence times of conditional quasi-neutral mutations could be highly variable. If this model can be taken seriously, there is a clear difficulty in comparing observed and expected levels of genetic variance across populations on the basis of the simplest mutation–selection balance model.

5. Discussion

In the simulations I have assumed that all selection operates through egg-to-adult differential viability. There is evidence for strong positive correlations between the effects of detrimental mutations on different fitness components, and the available data suggest that the net effect of a detrimental mutation on viability can be approximately one-half its deleterious effect on fitness as a whole (Charlesworth & Charlesworth, 1987; Houle et al., 1992, 1994; Hughes, 1995). In a random mating population, however, ignoring fertility differences is generally equivalent to assuming that viability effects in both sexes are the same (Bodmer, 1965). It is accordingly reasonable to have modelled deleterious alleles ignoring differences in fertility, because I was primarily interested in the relative effect of habitat patchiness on the mean persistence times. Therefore, to the extent that individuals exploiting resources patchily distributed in space are also patchily distributed during the juvenile stages, the conclusion that standard single-locus theory underestimates the true persistence times of deleterious alleles in natural populations seems to be inescapable. An area of uncertainty, however, concerns the comparison of persistence times across populations. Kondrashov & Houle (1994) posed the interesting question regarding the strength of selection at different resource levels, and the density-dependent selection model above clearly illustrates that mean persistence times are very sensitive to changes in the ecological parameters.

The two most plausible mechanisms for the maintenance of genetic variation in natural populations are mutation–selection balance and various models of balancing selection (Barton, 1990). An important reason to reject the simplest mutation–selection balance model comes from the empirical evidence of a latitudinal cline in the additive variance component of egg-to-adult viability in D. melanogaster (Mukai, 1985, 1988; Charlesworth, 1987; Barton, 1990; Houle et al., 1996). The variance of breeding values in the populations at high latitudes is approximately 0·003, which seems to agree with the expected value under mutation–selection balance in a homogeneous random mating population. On the other hand, the populations at low latitudes have between 5 and 8 times the genetic variance of those at high latitude (Mukai, 1985, 1988; Charlesworth, 1987). Mukai & Nagano (1983; see

also Mukai, 1988) suggested that diversifying selection promoted by a larger variation of ecological niches in southern populations is the most likely explanation. Genotype–environment interaction, and other forms of balancing selection different from classic over-dominance (i.e. frequency dependence and antagonistic pleiotropy), could also explain the absence of dominance variance observed by Mukai and his colleagues (see Charlesworth & Hughes, 1996). It is known, however, that the conditions for stable genetic polymorphisms are quite restricted in this kind of models (Maynard Smith & Hoekstra, 1980; Rose, 1982; Curtsinger et al., 1994). The number of loci on which these forms of balancing selection could be operating is probably very small, if any. On the other hand, if variation in fitness is mainly contributed by mildly deleterious alleles with an average dominance at equilibrium of approximately 0·2 (Simmons & Crow, 1977; Crow & Simmons, 1983; Hughes, 1995), the ratio V_d/V_a is also expected to be very low (see above).

Despite Mukai's (1988, p. 23) remarks that the amounts of inbreeding depression due to nonlethal-bearing chromosomes do not show a parallel north-to-south cline, there is indeed some indication of a positive correlation between the additive genetic variance and the detrimental load in the six populations analysed (Kendall $\tau = 0·69$; $P = 0·052$). This result can be interpreted in at least two ways. More deleterious alleles can be concealed in those populations with larger additive genetic variance in fitness. Conversely, a higher inbreeding decline of nonlethal homozygotes occurs in those populations where a larger fraction of loci are overdominant. Because diversifying selection effectively results in a higher fitness of heterozygotes, the second alternative would support the claim that this model accounts for the excess of additive genetic variance in some populations. On the other hand, the hypothesis advanced here that persistence times of unconditionally deleterious alleles may be determined primarily by ecological conditions (i.e. degree of habitat patchiness, resource abundance, etc.), could also explain the pattern and would fit better with the available evidence that indicates that overdominant loci do not contribute much to the genetic load in natural populations (Charlesworth & Charlesworth, 1987; Houle, 1989; Barret & Charlesworth, 1991).

If the genomic deleterious mutation rate in D. melanogaster has been grossly underestimated, as suggested by Kondrashov & Houle (1994) and supported by recent empirical studies that provide an estimated genotypic variance for net fitness of 0·45 when extrapolating from the third chromosome to the whole genome (Fowler et al., 1997), the level of genetic variation found in those populations at low latitudes might not be substantially higher than that expected under mutation–selection balance. The low levels of variation observed in high-latitude environ-

ments could thus be explained by assuming that these populations face harsher environmental conditions (see Houle *et al.*, 1996; Fowler *et al.*, 1997). There is a touch of irony here: the density-dependent model I used in the simulations builds upon habitat patchiness a defence of the mutation–selection balance model, but the same population structure is required in Levene's (1953) and related models of selection in heterogeneous environments. The important point here is that theoretical predictions of genotypic variance for fitness-related traits under the mutation–selection balance model are not as straightforward as usually assumed when applied to natural populations of *Drosophila*.

A number of additional observations may also be relevant to the present discussion. Sperlich *et al.* (1980) analysed the frequency of lethal-bearing O chromosomes in various European populations of *D. subobscura* that were chosen following criteria of ecological centrality or marginality. They found a higher frequency of lethals in central than in marginal populations, a finding further corroborated by Saura *et al.* (1990). On the assumption of negligible local inbreeding, there is no clear explanation for this pattern because it is difficult to understand why lethals should accumulate in any particular population (Lewontin, 1974). If, however, their criteria of ecological 'centrality' or 'marginality' are somewhat related to levels of resource abundance and habitat patchiness, the results of the present study could help to explain why lethal-bearing chromosomes are about twice as frequent in Italy ('central') than in Sweden ('marginal').

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