Genetic loads under fitness-dependent mutation rates

A. F. AGRAWAL

Department of Biology, Indiana University, Bloomington, IN, USA

Abstract

Although much theory depends on the genome-wide rate of deleterious mutations, good estimates of the mutation rate are scarce and remain controversial. Furthermore, mutation rate may not be constant, and a recent study suggests that mutation rates are higher in mildly stressful environments. If mutation rate is a function of condition, then individuals carrying more mutations will tend to be in worse condition and therefore produce more mutations. Here I examine the mean fitnesses of sexual and asexual populations evolving under such condition-dependent mutation rates. The equilibrium mean fitness of a sexual population depends on the shape of the curve relating fitness to mutation rate. If mutation rate declines synergistically with increasing condition the mean fitness will be much lower than if mutation rate declines at a diminishing rate. In contrast, asexual populations are less affected by condition-dependent mutation rates. The equilibrium mean fitness of an asexual population only depends on the mutation rate of the individuals in the least loaded class. Because such individuals have high fitness and therefore a low mutation rate, asexual populations experience less genetic load than sexual populations, thus increasing the twofold cost of sex.

Introduction

Because of recurrent mutation, no population can ever be completely free of deleterious alleles. The amount of genetic load borne by populations has been of interest to evolutionary biologists for a variety of reasons, including the maintenance of sex (e.g. Kondrashov, 1988; Charlesworth, 1990; Chasnov, 2000; Agrawal, 2001; Agrawal & Chasnov, 2001; Siller, 2001), the maintenance of outcrossing (e.g. Fisher, 1941; Lloyd, 1979; Charlesworth, 1980), the maintenance of genetic variation (e.g. Barton & Turelli, 1989; Bulmer, 1989), the extinction of small populations (e.g. Lande, 1994; Lynch *et al.*, 1995a,b), speciation (e.g. Kawecki, 1997; Lynch & Force, 2000), and the evolution of specialization (e.g. Kawecki, 1997; Kawecki *et al.*, 1997; Lynch & Force, 2000). The genome-wide rate of spontaneous deleterious mutations plays a key role in determining the amount of

genetic load experienced by a population. Despite its importance, estimates of the mutation rate remain controversial to two to three orders of magnitude (e.g. Keightley & Eyre-Walker, 1999; Lynch *et al.*, 1999; Denver *et al.*, 2000; Keightley & Eyre-Walker, 2000).

Models quantifying genetic load have typically assumed that mutation rate is an invariant property of populations. However, this may not be true. It has been known for some time that environmental stress can increase mutation rates in bacteria (reviewed in Foster, 1993, 1999). Recent evidence suggests that mutation rates in eukaryotes may also be environmentally sensitive. Goho & Bell (2000) exposed replicated lines of the unicellular chlorophyte, *Chlamydomonas reinhardtii*, to a variety of mildly stressful environments for several generations, allowed these lines to recuperate for at least 10 generations in their standard environment, and then assayed their fitnesses. They found that these lines had significantly lower mean fitnesses and higher standardized variances than control lines that had not experienced environmental stress. They interpreted these results as evidence for increased mutation rates in stressful environments. They suggest that mutation rates

Correspondence: A. F. Agrawal, Department of Biology, Indiana University, 1001 E. 3rd Street, Bloomington, IN 47405-3700, USA.
Tel.: 1-812-855-8250; fax: 1-812-855-6705;
e-mail: aagrawal@bio.indiana.edu

are $\sim$10–40-fold higher in mildly stressful environments. There is also evidence for environmentally induced mutations in *Saccharomyces cerevisiae* (Hall, 1992; Steele & Jinks-Robertson, 1992).

Both adaptive and nonadaptive explanations exist for environmentally sensitive mutation rates. Adaptive scenarios are generally limited to asexual organisms (Leigh, 1970; Ishii *et al.*, 1989; Taddei *et al.*, 1997). Nonadaptive explanations simply postulate that stress makes it more difficult for an organism to perform any of its normal activities, including the repair and replication of DNA. In this case, an increased mutation rate may be experienced by any organism in poor condition, regardless of whether the underlying reason for poor condition is extrinsic or intrinsic. This results in an interesting positive feedback loop involving deleterious mutations: individuals carrying an above-average number of deleterious mutations will tend to be in below-average condition and consequently experience an above-average mutation rate. Here I examine the consequences of fitness-dependent mutation rates on the genetic loads of sexual and asexual populations.

The model

Analytical approximation for sexual populations

Assuming independent gene action, the fitness of an individual with i mutations is $w_i = (1-s)^i$. Let the genome-wide diploid mutation rate of an individual with i mutations be a function of its fitness, $U_i = U_{max} - (U_{max} - U_{min})w_i^k$. Note that the minimum and maximum mutation rates occur for individuals with zero and infinite numbers of mutations, respectively, $U_0 = U_{min}$ and $U_\infty = U_{max}$. The parameter k determines the curvature of mutation rate as a function of fitness (Fig. 1). For $0 < k < 1$, mutation rate decreases at a diminishing rate as fitness increases. For $k = 1$, mutation rate declines linearly with fitness. For $k > 1$, mutation rate declines at an accelerating rate as fitness increases. The analytical approximation below assumes Hardy–Weinberg proportions, linkage equilibrium and that the frequency of individuals with i mutations follows a Poisson distribution with mean λ. The covariance between the number of mutations carried by an individual and the number of new mutations it produces may cause deviations away from the Poisson distribution. However, the results from the approximation below are in good accordance with computer simulation results that do not rely on these assumptions (see below).

Under these conditions, the mean fitness of the population is

$$W_{sex} = \sum_{i=0}^{\infty} \frac{\lambda^i e^{-\lambda}}{i!}(1-s)^i = e^{-\lambda s}. \tag{1}$$
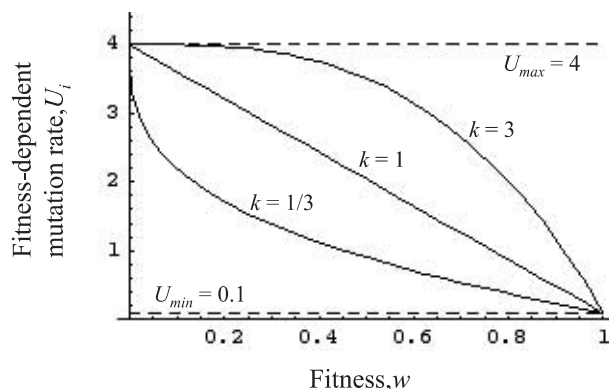
The mean number of mutations after selection is

Fig. 1 Fitness-dependent mutation rate, $U_i = U_{max} + (U_{max} - U_{min})w_i^k$. The shape of the curve relating fitness to mutation rate is shown as a function of the parameter k. For $0 < k < 1$ [or ln $(k) < 0$], mutation rate decreases at diminishing rate as fitness increases. For $k = 1$ [or ln $(k) = 0$], mutation rate decreases linearly with fitness. For $k > 1$ [or $\ln(k) > 0$], mutation rate decreases synergistically as fitness increases. In this example, $U_{min} = 0.1$ (lower dashed line) and $U_{max} = 4$ (upper dashed line).

$$\lambda' = \sum_{i=0}^{\infty} \frac{\lambda^i e^{-\lambda}}{i!}\left(\frac{(1-s)^i}{W_{sex}}\right)i = \lambda(1-s). \tag{2}$$

The mean number of *new* mutations generated in the next generation is

$$\bar{U} = \sum_{i=0}^{\infty} \frac{\lambda^i e^{-\lambda}}{i!}\left(\frac{(1-s)^i}{W_{sex}}\right)U_i$$
$$= U_{max} - (U_{max} - U_{min})e^{\lambda\sigma s} \tag{3}$$

where

$$\sigma = \frac{1}{s}(1-s)[(1-s)^k - 1]. \tag{4}$$

Summing the mutations existing after selection and the new mutations, the mean number of mutations in the following generation is

$$\lambda(t+1) = \bar{U} + \lambda'(t). \tag{5}$$

At equilibrium, the mean number of mutations does not change between generations; setting $\lambda(t+1) = \lambda(t)$ in the equation above, the equilibrium mean number of mutations is found to be

$$\lambda_{eq} = \frac{1}{s}\left(U_{max} - \frac{1}{\sigma}ProductLn[\sigma(U_{max} - U_{min}\exp[\sigma U_{max}]]\right) \tag{6}$$

where *ProductLn*[y] is the value of x that satisfies $y = xe^x$. The equilibrium mean fitness of a sexual population is found by substituting this λ_{eq} into eqn (1)

$$W_{sex} = \exp[-[(U_{max} - \frac{1}{\sigma}ProductLn[\sigma(U_{max}$$
$$- U_{min}\exp[\sigma U_{max}]]]. \tag{7}$$

Note that by substituting λ_{eq} into eqn (3), the average mutation rate at equilibrium is

$$\hat{U} = U_{max} - ProductLn[\sigma(U_{max} - U_{min})\exp(\sigma U_{max})]/\sigma. \quad (8)$$

Consequently, we can re-write eqn (6) as

$$W_{sex} = \exp(-\hat{U}), \quad (9)$$

which is similar to the standard equation for the mean fitness of a population at equilibrium under a constant mutation rate, $W = \exp(-U)$ (Kimura & Maruyama, 1966; Crow, 1970; Charlesworth, 1990).

Computer simulations for the mean fitness of sexual vs. asexual reproduction

Assuming multiplicative selection, sexual and asexual populations have equivalent genetic loads when the mutation rate is constant (Kimura & Maruyama, 1966; Kondrashov, 1982; Charlesworth, 1990). Deterministic computer simulations were used to evaluate whether the mode of reproduction influences genetic load under a fitness-dependent mutation rate. These simulations are described below.

The population is defined by the distribution of individuals having different numbers of mutations; let f_i be the frequency of individuals with i mutations. The frequency of individuals with i mutations after selection is $f_i' = f_i e^{-si} / W$ where $W = \sum f_i e^{-si}$ is the mean fitness of the population.

Assuming new mutations follow a Poisson distribution, the probability that an individual with i mutations mutates into one with j new mutations (resulting in a total of $i + j$ mutations) is $p_{i,j} = e^{-U_i} U_i^j / j!$. For an asexual population, the frequency of offspring in the next generation with k mutations is

$$f_k(t + 1) = \sum_{i=0}^{k} f_i'(t) p_{i,(k-i)}.$$

For calculating the distribution of sexually produced offspring, I assumed that mutation occurs after selection but before gamete production and syngamy. Following Kimura & Maruyama (1966) gametes are produced assuming free recombination by using a binomial distribution with h trials and a probability of success equal to 0.5, where h is the number of mutations carried by the parent. The frequency of gametes carrying m mutations is

$$g_m(t + 1) = \sum_{k=0}^{\infty} \left[\left(\sum_{i=0}^{h} f_i'(t) p_{i,(h-i)} \right) \binom{h}{m} \left(\frac{1}{2} \right)^h \right].$$

The gametes combine at random to produce zygotes. For a sexual population, the frequency of offspring with k mutations is

$$f_k = \sum_{m=0}^{k} g_m g_{(k-m)}.$$

Most computer simulations began assuming no mutations (i.e. $f_0 = 1$, $f_{i \neq 0} = 0$). Equilibrium is never actually reached because it is approached asymptotically but it is closely approximated within several hundred generations. Therefore, each simulation was run for at least 300 generations or until the change in mean fitness between generations was less than 10^{-4}, whichever took longer. In several cases, computer simulations were also run from an initial state of many mutations (i.e. more than the expected equilibrium number). For sexual populations, simulations converged towards the same equilibrium regardless of whether the population started with zero or many mutations. This result did not hold for asexual populations because an asexual offspring cannot have fewer mutations than its parent (see below). Analytical calculations and computer simulations were performed using *Mathematica* (Wolfram, 1991).

Results

Sexual populations

The fitness of a sexual population under a fitness-dependent mutation rate depends on the shape of the mutation rate as a function of fitness; this shape is determined by the parameter k. Sexual populations equilibrate close to the fitness expected under the maximum mutation rate if $k > 1$ (Fig. 2). However, if the mutation rate declines at a diminishing rate as fitness increases ($0 < k < 1$), then sexual populations have an equilibrium mean fitness reflective of a substantially lower mutation rate than U_{max}. For example, with $U_{min} = 0.1$, $U_{max} = 4$, and $k = 0.2$, the equilibrium mutation rate is $\hat{U} \approx 0.38$, resulting in a mean fitness of ~0.68. This mean fitness is ~37-fold greater than that expected under the maximum mutation rate, but ~1.3-fold less than that expected under the minimum mutation rate. When $k > 1$, the average mutation rate at equilibrium is typically close to the maximal rate. In contrast, when $0 < k < 1$, the average mutation rate at equilibrium can be much lower than the potential maximum. This suggests that the mutation rate must decline at a diminishing rate as fitness increases (i.e. $0 < k < 1$) if a population at equilibrium is to have the potential to show a substantially increased mutation rate when stressed. Note that the exact range of k-values over which the transition occurs from high fitness to low fitness depends on the values of U_{min} and U_{max} (compare the three panels in Fig. 2). (Fig. 2 also shows that the analytical approximation provides results that are consistent with computer simulations.)

Assuming multiplicative fitness and a constant mutation rate, mean fitness is independent of the selection coefficient s (Haldane, 1937; Kimura & Maruyama, 1966; Crow, 1970). Under a fitness-dependent mutation rate, W_{sex} is influenced by s through the parameter σ. However, evaluations of σ with reasonable values of s
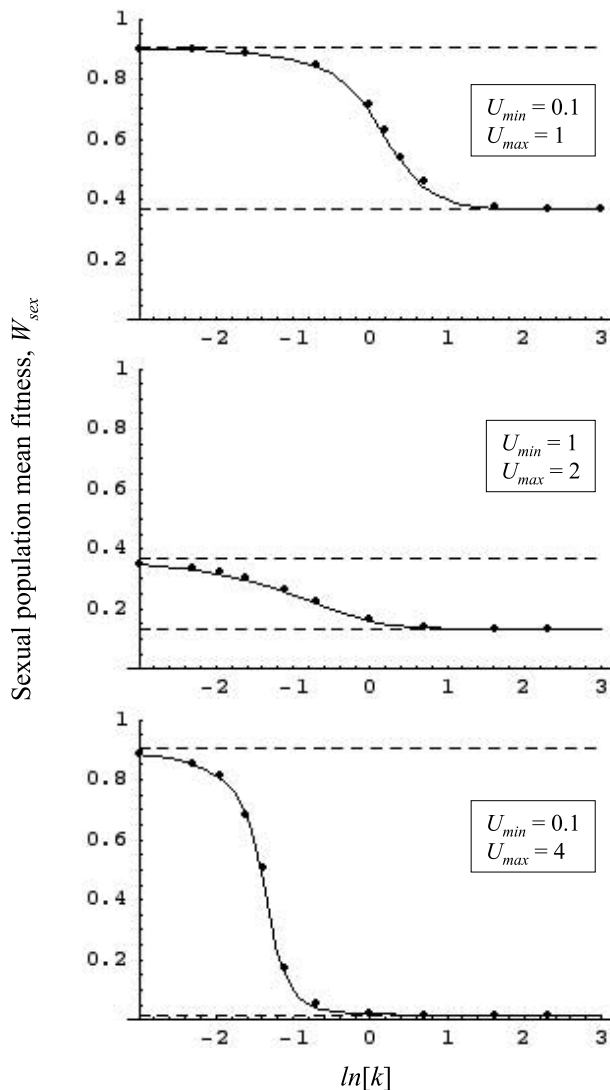
Fig. 2 Mean fitness of sexual populations, W_{sex}, under fitness-dependent mutation rates. W_{sex} is plotted as function of the parameter k, which determines the shape of the curve relating fitness to mutation rate. The analytical approximation to W_{sex} is represented by the solid line. The dots represent computer simulation results. The upper (lower) dashed line represents the fitness expected under a constant mutation rate equal to U_{min} (U_{max}). If an asexual population is initially mutation-free, its mean fitness will be equal to the upper dashed line. In these examples, $s = 0.02$.

(e.g. $0.01 < s < 0.1$) show that s has little effect on the value of σ or W_{sex}.

Asexual populations

From inspection of the deterministic simulation results (Table 1), it is evident that the equilibrium fitness of an asexual population is approximately equal to $W_{asex} \sim (1-s)^c \exp(-U_c)$ where c is the number of mutations of the least loaded class. This is true even though most of the

individuals in the population experience higher mutation rates than U_c. When simulations are initiated with individuals with zero mutations (for whom the mutation rate is U_{min}), the equilibrium fitness of the population is $W_{asex} \sim \exp(-U_{min})$ which can be much greater than the mean fitness of a sexual population under the same parameters.

Discussion

Under constant mutation rates, sexual and asexual populations of infinite size have equal fitnesses at equilibrium, assuming that deleterious mutations act independently and that asexuals initially contain no mutations (Kimura & Maruyama, 1966; Crow, 1970; Kondrashov, 1988). This is not true when mutation rates are fitness-dependent. The fitness of a sexual population depends on the shape of the curve relating fitness to mutation rate. In contrast, the fitness of an asexual population depends only on the mutation rate of the least loaded class.

Why are sexual and asexual populations affected so differently by fitness-dependent mutation rates? In both types of populations, there is variance in the number of mutations per individual producing variance in fitness and, consequently, variation in the mutation rate: heavily loaded individuals experience higher mutation rates than lightly loaded individuals. With sexual reproduction, offspring are produced by gametes from different parents; one parent may have a higher than average mutation rate, the other a lower than average mutation rate. Sexual populations truly experience the average mutation rate [as revealed by Eqs (7–9)]. This is not the case for asexual populations. Each asexual individual is affected only by its own mutation rate and not by the average. The offspring of lightly loaded asexuals are not affected by the high mutation rates occurring in the production of offspring by heavily loaded clones. This explanation can be checked by making the mutation rate experienced by each asexual individual a function of mean fitness rather than a function of that individual's own fitness. When this test was carried out, the equilibrium mean fitness of an asexual population was similar to that of a sexual population (unpublished results).

Based on the results of Table 1, it seems that the mean fitness of the entire asexual population is determined by the least loaded class, although this class may only be a small fraction of the population at equilibrium. The simulations investigated here are deterministic and assume an infinite population size. The least loaded class is set by the initial conditions and does not change in the deterministic simulation. In reality, however, the least loaded class can be lost because of stochastic processes. Because mutation is assumed to be unidirectional, this class cannot be regained, which is known as Muller's Ratchet (Muller, 1964; Haigh, 1978; Bell, 1982, 1988).

With each turn of the ratchet, the number of mutations in the least loaded class, c, increases by one,

Table 1 Mean fitness of an asexual population, W_{asex}, under a fitness-dependent mutation rate. Simulation results are shown when initiating the asexual lineage with a least loaded class of individuals carrying 0 or 30 mutations. Simulation results match to at least three decimal places the expected fitness of an asexual population with a constant mutation rate equal to that of the mutation rate of the least loaded class. The mean fitness of a sexual population W_{sex} [calculated from eqn (7)] is shown for comparison. In all cases shown, $s = 0.02$.

U_{min}	U_{max}	k	Mutation no. in least loaded class, c	W_{asex} (simulation results)	W_{asex} [constant U expectation, $W_{asex} = (1-s)^c \exp(-U_c)$]	W_{sex}
1	2	0.1	0	0.368	0.368	0.332
		1		0.368	0.368	0.160
		10		0.368	0.368	0.135
1	2	0.1	30	0.189	0.189	0.332
		1		0.127	0.127	0.160
		10		0.074	0.074	0.135
0.1	4	0.1	0	0.905	0.905	0.850
		1		0.905	0.905	0.02
		10		0.905	0.905	0.018
0.1	4	0.1	30	0.392	0.392	0.850
		1		0.084	0.084	0.02
		10		0.010	0.010	0.018

reducing the mean fitness of an asexual population. Eventually, the mean fitness of an asexual population completely erodes the twofold advantage of asexual reproduction. With respect to the maintenance of sex, there are two questions facing Muller's Ratchet. First, how many times must the ratchet turn to offset the twofold cost of sex? Secondly, how much time is required for each of these turns?

Under a constant mutation rate the fitness of an asexual population is $W_{asex} = (1-s)^c \exp(-U)$ whereas that of a sexual population is $W_{sex} = \exp(-U)$. To offset the twofold cost of sex, the minimum necessary number of mutations in the least loaded class is found by setting $W_{sex} = 2 W_{asex}$ and solving for c. The required number of mutations is $c^* = -\ln(2)/\ln(1-s)$. If the initial clone carries only c_o mutations, the ratchet must click $(c^* - c_o)$ times. For example, assuming $s = 0.02$, if the initial clone is mutation free the ratchet must click approximately 34 times (i.e. $c^* \sim 34$). New asexual clones will most frequently arise carrying as many mutations as the average sexual individual. However, such clones will generate an asexual population with a heavy genetic load and therefore do not represent the greatest threat to sexual populations. The primary challenge for Muller's Ratchet, with respect to the maintenance of sex, occurs when the initial clone contains zero or few mutations (Bulmer, 1994; Howard & Lively, 1998).

Under a fitness-dependent mutation rate, $W_{sex} = \exp[-\hat{U}]$ and, from inspection of simulation results, $W_{asex} \sim (1-s)^c \exp(-U_c)$. The minimum number of mutations in the least loaded class required to offset the twofold cost of sex is

$$c^* = \Delta - \frac{ProductLn[(U_{max} - U_{min})k(1-s)^{k\Delta}]}{\ln[1-s]k}$$

where $\Delta = (U_{max} - \bar{U}_{sex}) - \ln[2])/\ln[1-s]$. Unlike the formula for c^* under a constant mutation rate, the equation above is not just a simple function of s. This is because asexuals have a different mutation rate than sexuals, and this mutation rate changes as the ratchet turns. Under a fitness-dependent mutation rate c^* can be greater or less than c^* under a constant mutation rate, although it is surprisingly similar in many cases (Fig. 3).

Under a constant mutation rate, the time for each turn of the ratchet is expected to be constant (Haigh, 1978; Bell, 1988; Bulmer, 1994), assuming population size is constant. Under a fitness-dependent mutation rate, the
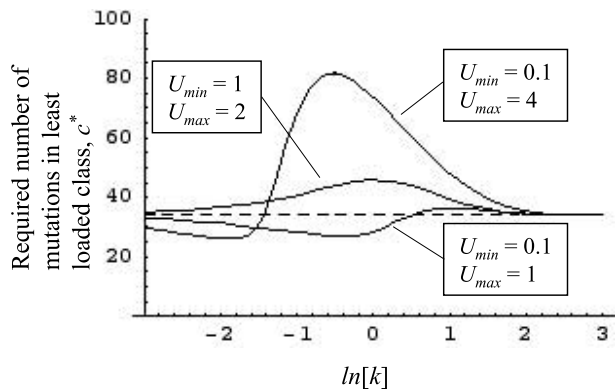

Fig. 3 Minimum number of mutations in the least loaded class of an asexual population required to offset the two-fold cost of sex, c^*. For a constant mutation rate, $c^* = -\ln(2)/\ln(1-s)$. In this example, $s = 0.02$ so $c^* \sim 34$. The dashed line represents this value of c^*. The solid lines represent the c^* under different fitness-dependent mutation rates. Note that the value of c^* under a fitness-dependent mutation rate can be greater or less than that under the constant mutation rate.

ratchet is likely to turn at an accelerating rate as the mutation rate of the least loaded class will increase with each turn. However, the average time for each of the necessary turns may be greater under a fitness-dependent mutation rate than under a constant mutation rate equal to $\hat{U}_{sex}$. This is because U_c will be less than $\hat{U}_{sex}$ for most, if not all, of the necessary turns for the critical case when the initial asexual clone contains fewer mutations than the average sexual individual. Further, the time to extinction of the least loaded class depends on the frequency of this class, f_c. Under a constant mutation rate, this frequency is exp $(-U/s)$. Simulation results indicate that, under a fitness-dependent mutation rate, the equilibrium frequency is greater than exp $(-U_c/s)$, often by more than one order of magnitude. This higher frequency makes it more difficult to stochastically lose the least loaded class. A formal analysis of the time to extinction of the least loaded class under fitness-dependent mutation rates is needed.

In summary, fitness-dependent mutation rates make it even more difficult to account for the maintenance of sex. Assuming that both sexual and asexual populations are infinite and initially contain none or only a few mutations, sexual populations will have a higher genetic load at equilibrium than asexual populations. Asexual populations have a greater than twofold advantage because they have less load than sexual populations. For example, in Fig. 2, an infinite population of asexuals initially containing no mutations will have an equilibrium fitness equal to the upper dashed line, whereas the equilibrium fitness of a sexual population is represented by the solid line. In finite populations, the ratchet will work to reduce the fitness of asexuals, though perhaps more slowly than it would under a constant mutation rate.

If mutation rates are fitness-dependent, mutation seems to make the situation worse for sex rather than better. In this light, ecological explanations for the maintenance of sex such as the Red Queen hypothesis (Jaenike, 1978; Bremerman, 1980; Hamilton, 1980; Bell, 1982) may be more important. It is possible that ecological factors interact with fitness-dependent mutation rates to reverse this bleak outlook for sexual populations. Previous theoretical investigations suggest that important interactions may occur between the Red Queen and Muller's Ratchet (Howard & Lively, 1994, 1998; West et al., 1999).

Fitness-dependent mutation rates could play important roles in other areas of evolutionary biology. In mutational meltdown models for the extinction of small populations, two phases occur (Lynch et al., 1993; 1995a,b). In the first phase, mutations drift to fixation in a population of small but steady size. The second phase begins after enough mutations have accumulated that the population growth rate is less than one and population size begins to decline. In this second phase, mutations drift more easily to fixation with each

passing generation as the population size declines. Fitness-dependent mutation rates could accelerate both phases of the mutational meltdown.

One of the major theories for the maintenance of outcrossing in hermaphrodites is based on inbreeding depression (e.g. Fisher, 1941; Lloyd, 1979; Charlesworth, 1980). A fitness-dependent mutation rate could make the inbreeding depression hypothesis more powerful than under a constant mutation rate. Inbred individuals have lower fitness because of inbreeding depression and would therefore have higher mutation rates. These new mutations would soon be in a homozygous state because of inbreeding, making them more deleterious, and further increasing the mutation rate. Purging would be more difficult with fitness-dependent mutation rates because the mutation rate of inbred individuals would be higher than that of outbreeders.

Mutation-based theories depend critically on mutation rate; these ideas have little explanatory power if mutation rate is low. However, the possibility of fitness-dependent mutation rates suggests that there may be no true mutation rate. Goho & Bell (2000) estimate that mutation rate was at least 10–40-fold higher in a mildly stressful environment. This means that mutation rate can be low (e.g. $U_{min} \sim 0.1$) under some conditions or for some individuals but much higher under other conditions or for other individuals (e.g. $U_{max} \sim 4$). However, it is not obvious how such a plastic mutation rate will affect the various mutation-based theories. More importantly, the ubiquity and magnitude of fitness effects on mutation rate must be established empirically.

Acknowledgments

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