A Ruby in the Rubbish: Beneficial Mutations, Deleterious Mutations and the Evolution of Sex

Joel R. Peck

Department of Zoology, The University of Oxford, Oxford OX1 3PS, Great Britain

Manuscript received October 13, 1993
Accepted for publication February 22, 1994

ABSTRACT

This study presents a mathematical model in which a single beneficial mutation arises in a very large population that is subject to frequent deleterious mutations. The results suggest that, if the population is sexual, then the deleterious mutations will have little effect on the ultimate fate of the beneficial mutation. However, if most offspring are produced asexually, then the probability that the beneficial mutation will be lost from the population may be greatly enhanced by the deleterious mutations. Thus, sexual populations may adapt much more quickly than populations where most reproduction is asexual. Some of the results were produced using computer simulation methods, and a technique was developed that allows treatment of arbitrarily large numbers of individuals in a reasonable amount of computer time. This technique may be of prove useful for the analysis of a wide variety of models, though there are some constraints on its applicability. For example, the technique requires that reproduction can be described by Poisson processes.

OVER the long term, adaptation depends on the production and preservation of beneficial mutations. For example, beneficial mutations can play an important role in allowing adaptation after an environmental change. This has been clearly demonstrated in bacteria and yeast, where novel laboratory conditions lead to the regular production of beneficial mutations (PAQUIN and ADAMS 1983; LENSKI *et al.* 1991). Some of the beneficial mutations that arise in these experiments confer rather large advantages, and 10% enhancements of relative fitness from single beneficial mutations are not uncommon.

Beneficial mutations can also be important in stable environments. If deleterious mutations of very small effect occur regularly, then we can expect that some of them will become fixed. This process will lead to a steady decline in the fitness of the population unless back mutations or compensatory mutations also arise and become fixed from time to time. In this context, back mutations and compensatory mutations are beneficial. It seems likely that some such process is going on in natural populations, because there is substantial evidence that deleterious mutations of small effect are common (OHTA 1992). Beneficial mutations may also play a role in stable environments because no species is likely to have become genetically perfect. So long as there is room for improvement of the genome, beneficial mutations are possible. However, in the absence of environmental change, beneficial mutations may become rare.

While it seems likely that beneficial mutations occur in every living species from time to time, it seems even more certain that deleterious mutations are very common occurrences. For example, in Drosophila it appears that the rate of deleterious mutations is on the order of one per genome per generation, and a similar rate is likely to apply to various other "higher" organisms (MUKAI *et al.* 1972; CROW 1979; KONDRASHOV 1988; CHARLESWORTH 1990; CHARLESWORTH *et al.* 1990; KONDRASHOV and TURELLI 1992; HOULE *et al.* 1992).

When a beneficial mutation arises in an organism with a large genome (*e.g.*, in a higher plant or animal), it is likely to occur in a genome that is contaminated with many deleterious mutations. Thus, it is worth asking how sexual reproduction can affect the fate of a beneficial mutation when deleterious mutations are common. This question was addressed verbally by FISHER (1930) and there have also been a few relevant formal theoretical studies. These will be considered in the discussion section.

In addition to the very small amount of research that has been carried out on the interaction between deleterious mutations and beneficial mutations, there is a more substantial literature on the interaction between multiple beneficial mutations at different loci in the absence of deleterious mutations. Interactions of this sort are the basis for one of the best known evolution-of-sex theories, the so-called FISHER-MULLER hypothesis (FISHER 1930; MULLER 1932). The FISHER-MULLER hypothesis says that sexual reproduction and recombination can enhance the probability of fixation of beneficial mutations because they allow two or more beneficial mutations that arise in different individuals to be united in the same genomes.

Theoretical investigations suggest that FISHER and MULLER were right, but it appears that the enhancement that can be gained by the process they described will be

small if beneficial mutations rarely arise (FELSENSTEIN 1974; PAMILO *et al.* 1987). Another problem is that beneficial mutations are generally thought to be very rare (DOBZHANSKY 1970; KIMURA 1983), and so theories that depend on the simultaneous segregation beneficial mutations at multiple loci may have limited applicability.

In the present study, the fate of beneficial mutations that arise in populations where deleterious mutations are common will be studied by means of mathematical analysis and computer simulation. Special attention will be paid to the effects of sexual and asexual reproduction on evolutionary outcomes. The results provide evidence to support the idea that, when deleterious mutations are common, sex and recombination will greatly enhance the probability that a beneficial mutation will increase in frequency from a single initial copy until many copies are present. This idea should be of interest in light of the current debate over the evolution of sex (WILLIAMS 1975; MAYNARD SMITH 1978; BELL 1982; MICHOD and LEVIN 1988; HAMILTON *et al.* 1990). The idea may also have implications for the design of agricultural breeding programs.

THE MODEL

A wide variety of different mathematical models can be used to study the processes that determine the fate of beneficial mutations. However, for the sake of concreteness, let us focus on the case of fertility selection in a diploid population of annual seed-bearing plants. Assume that all reproductively mature population members (*i.e.*, adults) are capable of producing both seeds and pollen. Seeds may be produced either by sexual means, or by asexual means. Asexually produced seeds are genetically identical to the individuals that produced them, except for new mutations. When individuals generate seeds by sexual means they produce an ovule, and this is fertilized by a pollen grain. The production of both ovules and pollen involves standard Mendelian segregation.

Let us number the possible genotypes as 1, 2, 3, . . . , and assume that the number of seeds produced by sexual means for an adult with genotype i follows a Poisson distribution with mean equal to ϕW_i (where $0 \leq \phi \leq 1$ and $0 < W_i$). The number of asexually produced seeds for the same individual follows an independent Poisson distribution with mean equal to $(1 - \phi) W_i$. Thus, on average, the total number of seeds produced by an adult with genotype i is W_i, while ϕ represents the expected proportion of sexually produced seeds for any given adult.

Assume that the number of pollen grains produced by adults with genotype i follows a Poisson distribution with mean equal to $\phi K W_i$, where $K > 1$. Ovules are fertilized in random order, and the pollen grain involved in each case is drawn at random from the collection of all pollen grains that have not yet achieved fertilization. After seed production, N seeds are chosen at random to grow to adulthood, where N is a very large positive integer (in fact, all of the calculations in this study were made for the limiting and biologically unrealistic case where $N = \infty$, but the results will change only slightly in the much more realistic case where N is finite and very large). Thus, N is the number of adults during each successive generation. Because N is infinite and $K > 1$, the pollen grains will not run out before all ovules have been fertilized. In addition, the $N = \infty$ assumption implies that the frequency distribution of genotypes among unused pollen grains will not change during the course of ovule fertilizations.

Assume that deleterious mutations occur at randomly selected loci. Assume further that a very large number of loci are subject to deleterious mutations, and that there is free recombination between loci (note: the calculations were made for the limiting case where there is an infinite number of loci, but the results should change only slightly if the number of loci is finite, but large). The number of new deleterious mutations for asexually produced seeds is assumed to be Poisson distributed with the mean of the distribution equal to U. The number of new deleterious mutations in pollen and ovules is also Poisson distributed, and the mean of the distribution is equal $U/2$ (thus, the mean number of new mutations per seed is U, regardless of the mode of reproduction).

Consider an adult with deleterious mutations in heterozygous state at k loci, and with no beneficial mutations or deleterious mutations in homozygous state. The value of W_i for such an individual is assumed to equal $C(1 - s_d)^k$, where s_d and C are constants that satisfy $0 < s_d < 1$ and $C > e^U$. Populations of this sort were studied by HAIGH (1978) for $\phi = 0$, and he showed that in this case, the equilibrium distribution for the number of deleterious mutations per individual is Poisson with mean equal to U/s_d. In addition, the probability of finding an individual that is homozygous for a deleterious mutation is negligible. Numerical investigations strongly suggest that these same results hold for $\phi > 0$.

Now, consider a population of the sort just described that has come to equilibrium. Say that a beneficial mutation arises in a randomly selected seed, and assume that the seed happens to be among the N seeds that grow to adulthood. Naturally, the seed in question will be heterozygous for the beneficial mutation. Assume that the value of W_i for a heterozygous carrier of the beneficial mutation that also has k deleterious mutations is equal to $C(1 + s_b)(1 - s_d)^k$, where $s_b > 0$ (in other words, assume that the beneficial mutation enhances fitness by a factor of $1 + s_b$).

RESULTS

The analysis will focus on the calculation of E, which is defined as the probability that a beneficial mutation will become established. Establishment of a beneficial

TABLE 1

Probability of establishment of beneficial mutations

Parameter values	ϕ = 0.0	0.05	0.1	0.25	1.0
$U = 0.000$, $s_b = 0.01$	0.0197	0.0197	0.0197	0.0197	0.0197
$U = 0.125$, $s_d = 0.02$, $s_b = 0.01$	3.81×10^{-5}	0.00570 (0.000753)	0.0119 (0.00108)	0.0186 (0.00135)	0.0169 (0.00129)
$U = 0.250$, $s_d = 0.02$, $s_b = 0.01$	7.36×10^{-8}	0.00250 (0.000499)	0.00790 (0.000885)	0.0164 (0.00127)	0.0184 (0.00134)
$U = 0.500$, $s_d = 0.02$, $s_b = 0.01$	2.74×10^{-13}	0.000300 (0.000173)	0.00250 (0.000499)	0.0110 (0.00104)	0.0193 (0.00138)
$U = 0.250$, $s_d = 0.01$, $s_b = 0.01$	2.74×10^{-13}	0.00520 (0.000719)	0.0118 (0.00108)	0.0179 (0.00133)	0.0189 (0.00136)
$U = 0.250$, $s_d = 0.04$, $s_b = 0.01$	3.81×10^{-5}	0.00160 (0.000400)	0.00490 (0.000698)	0.0126 (0.00112)	0.0214 (0.00145)
$U = 0.100$, $s_d = 0.025$, $s_b = 0.10$	0.0510 (0.00220)	0.141 (0.00348)	0.153 (0.00360)	0.168 (0.00374)	0.176 (0.00381)
$U = 0.000$, $s_b = 0.10$	0.176	0.176	0.176	0.176	0.176
$U = 0.250$, $s_d = 0.02$, $s_b = 0.10$	0.000600 (0.000245)	0.0901 (0.00286)	0.109 (0.00312)	0.156 (0.00363)	0.171 (0.00376)

The entries give estimates of probability of establishment (E) of a beneficial mutation for various choices of parameter values. The probability of establishment was calculated by analytic methods in those cases where $U = 0$ or where $\phi = 0$ and $s_b \leq s_d$, and the figures given for these cases represent the results of these calculations (see APPENDIX 2). In all other cases a computer simulation was used to produce the estimate (see APPENDIX 1), and the figures that are not in parentheses represent the proportion of simulation trials for which a beneficial mutation was deemed to be established, while the figures in parentheses give estimates of the standard errors.

mutation means that its loss from the population is virtually impossible. For a beneficial mutation to be established, it must be present in many copies. Also, as we shall see, these copies must have the right distribution with regard to the number of deleterious mutations held by bearers of the beneficial mutation.

To begin the analysis, consider the simple situation where $U = 0$, so that no deleterious mutations ever occur. By extending FISHER and HALDANE's treatment (FISHER 1922; HALDANE 1927) it can be shown that, in this case, the value of E is given by the unique positive solution of the following equation:

$$1 - E = \exp[-(1 + s_b)E]. \quad (1)$$

When s_b is small, this yields the familiar formula, $E \approx 2s_b$. For example, when $s_b = 0.01$ we have $E = 0.0197$, and when $s_b = 0.1$ we have $E = 0.176$. The probability that a new beneficial mutation will be lost from the population is equal to $1 - E$. Loss occurs when all of adults that carry the mutant gene fail to produce any seeds that also carry the mutation, and that grow to adulthood. Note that, in Equation 1, E does not depend on ϕ, and thus the mode of reproduction does not affect the probability of establishment for a beneficial mutation when $U = 0$, so that deleterious mutations are not present in the population. (However, fixation of a beneficial mutation may be forestalled when $\phi = 0$ because asexuality hinders the formation of individuals that are homozygous for the beneficial mutation (KIRKPATRICK and JENKINS 1989; WEINER *et al.* 1992).)

Let us now consider the more complicated situation where $U > 0$, so that non-neutral variation can occur at more than one locus. A multi-locus model provides more scope for an interaction between the mode of reproduction and the probability of establishment of a beneficial mutation. This is because of the HILL-ROBERTSON effect, which describes the way that non-neutral variation at one locus can interfere with the response to selection of allelic frequencies at other loci (HILL and ROBERTSON 1966; FELSENSTEIN 1974, 1988).

In terms of the current model, data on deleterious mutations (*e.g.*, MUKAI *et al.* 1972; CROW 1979; KONDRASHOV 1988; CHARLESWORTH 1990; CHARLESWORTH *et al.* 1990; KONDRASHOV and TURELLI 1992; HOULE *et al.* 1992) suggests that parameter values such as $s_d = 0.02$ and $U = 0.25$ are reasonable for both plants and animals (although $U = 0.25$ may substantially underestimate the real rate of deleterious mutations). In a sexual population ($\phi = 1$) with $s_d = 0.02$ and $U = 0.25$, the simulation results lead to an estimate of $E \approx 0.0184$ when $s_b = 0.01$, and $E \approx 0.171$ when $s_b = 0.1$. These figures are not much altered from the figures cited above for the case where no deleterious mutations are present. Furthermore, examination of Table 1 suggests that this sort of result is fairly general when $\phi = 1$. Thus, when offspring are produced sexually, there appears to be a large set of parameter values for which the probability of establishment of a beneficial mutation is only slightly perturbed by frequent deleterious mutations. However, under asexuality ($\phi = 0$) with $s_d = 0.02$ and $U = 0.25$, we have $E = 7.36 \times 10^{-8}$ when $s_b = 0.01$ and $E \approx 0.000600$ when $s_b = 0.1$. Examination of Table 1 and APPENDIX 2 suggests that these results are not unique. It appears that, for a

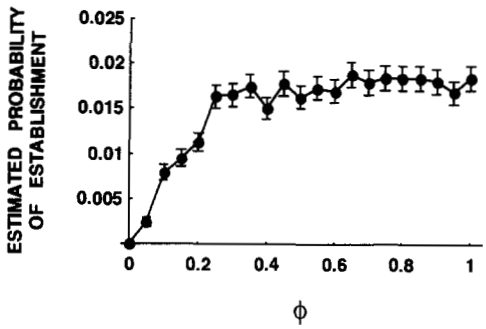

FIGURE 1.—The estimated probability of establishment of a beneficial mutation as a function of ϕ. For this figure, $s_d = 0.02$, $U = 0.25$, and $s_b = 0.01$. The data points were calculated using the methods detailed in APPENDICES 1 and 2. Error bars are twice the size of the standard errors.

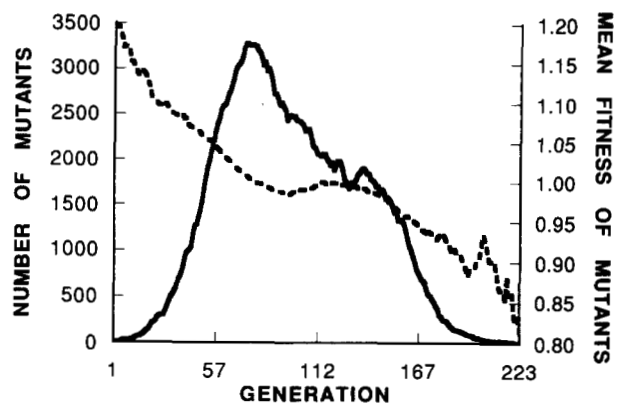

FIGURE 2.—Output from a single trial where the beneficial mutation was initially associated with an adult that had only four deleterious mutations. The solid curve shows the number of adults that carry the beneficial mutation for 223 generations after its introduction, and the scale for this curve is on the left side of the graph. The dashed curve shows the average fitness of adults that carry the beneficial mutation during the same period of time, and the scale is on the right side of the graph. The parameter values used in the simulation were $\phi = 0$, $s_d = 0.02$, $U = 0.25$ and $s_b = 0.01$. Note that, on the axis labels, the word "mutants" refers to individuals that carry the beneficial mutation. The method used to select the trial shown in this graph is described in the text.

wide range of parameter values, a substantial rate of deleterious mutations will cause a large decrease in E when $\phi = 0$. Furthermore, as demonstrated by Figure 1, small but non-zero values of ϕ can also cause a substantial depression in E. However, in all the cases studied where $U > 0$, the value of E for $\phi = 0.05$ is far above the value when $\phi = 0$.

It is interesting to note that, under asexuality, some mutations that are definitely doomed to extinction nevertheless have a tendency to achieve high copy numbers before they are eliminated. This tendency is not apparent in sexual populations. For example, if $s_b < s_d$, then a beneficial mutation can only become established if it arises in an individual that is completely free of deleterious mutations. If it arises in any other sort of individual, then the mutation will definitely be eliminated from the population (see APPENDIX 2 for a proof of this point, and the discussion for an intuitive explanation). However, if the deleterious mutation occurs in an individual that has some deleterious mutations, but a smaller number than average, then it may increase to a large number of copies before disappearing from the population.

As an example of this phenomenon, consider the case of an asexual population ($\phi = 0$) where $s_d = 0.02$, $U = 0.25$ and $s_b = 0.01$. In this situation the average number of deleterious mutations at equilibrium before introduction of a beneficial mutation is 12.5. To demonstrate the phenomenon of transient high copy numbers, the computer simulation methods described above were modified by initiating each of 10,000 trials with a single beneficial mutation associated with an adult with four deleterious mutations (in contrast to the normal method of starting with the beneficial mutation associated with a randomly selected adult). At the equilibrium that is established prior to the introduction of the beneficial mutation, adults with no beneficial mutations and with only four deleterious mutations have a fitness that is 18.4% above the average value of 1.0 (where the fitness of an adult with genotype i is defined as $W_i/\bar{W}$, and $\bar{W}$ is defined as the average number of seeds produced per adult). Shortly after the introduction, an adult who carries the beneficial mutation (in heterozygous state) plus four deleterious mutations will have a fitness that is 19.6% above average. The effects of the elevated fitness levels associated with adults with only four deleterious mutations were readily apparent from the results of the simulation. As predicted by theory, the beneficial mutation was lost from the population in all 10,000 trials. However, in 17.3% of the trials more than 100 copies of the beneficial allele were present at some point in time before the allele went extinct. Furthermore, in 3.3% of the trials, 1,000 copies were present at some point in time before extinction. Extinction results from a gradual accumulation of deleterious mutations among those individuals that carry the beneficial mutation. This accumulation leads to a decline in the average fitness of the beneficial-mutation-bearing adults. Figure 2 shows data from a trial that was randomly selected from among those trials where more than 1,000 copies were present at some point before extinction. Further studies revealed, in line with expectations, that the propensity for a beneficial mutation to rise to high copy numbers before extinction is less pronounced when the beneficial mutation is initially associated with more deleterious mutations, and more pronounced if it is initially associated with fewer deleterious mutations.

The same simulation study just described was re-run, except that obligate sexuality was imposed (*i.e.*, 10,000 trials with $\phi = 1$, $s_d = 0.02$, $U = 0.25$, $s_b = 0.01$, and the beneficial mutation initially associated with an adult that has four deleterious mutations). In this case, the

beneficial mutation was established during 2.4% of the trials. This is an improvement over the 1.8% of trials that led to an establishment in the case where the initial association of the beneficial mutation was random (see Table 1) but the improvement is not dramatic. During only 2.9% of the trials did the copy number of the beneficial mutation rise above 100 at any point, and this is only 16.8% of the rate at which this occurred during the asexual runs described in the preceding paragraph. This observation is made more surprising when one recalls that beneficial mutations always went extinct in the asexual case, while in the sexual case, rising above 100 copies usually led to establishment of the beneficial mutation. In the sexual case, the beneficial mutation rose above 1000 copies during 2.4% of the trials, and on each occasion this was followed by establishment of the beneficial mutation. Recall that the beneficial mutation rose to this level more frequently in the asexual case, even though it never became established.

When $s_b > s_d$ it may be possible for a beneficial mutation to become established in an asexual population even if it does not initially occur in an individual that is entirely free of deleterious mutations. In particular, if the beneficial mutation initially occurs in an individual that has k deleterious mutations, then $E > 0$ so long as s_b is sufficiently large such that $(1 + s_b)(1 - s_d)^k > 1$. However, if the beneficial mutation does initially occur in an individual that carries deleterious mutations and it rises in frequency, the beneficial mutation will drag deleterious mutations with it, and the increase in fitness obtained will be diminished by this factor [see MANNING and THOMPSON (1984) for a discussion related to this point]. In addition, as mentioned previously, under asexuality a single deleterious mutation can never attain a frequency in excess of 0.5 because of the lack of segregation (KIRKPATRICK and JENKINS 1989; WEINER *et al.* 1992). No such obstacle stands in the way of fixation if $\phi > 0$.

DISCUSSION

Why does the mode of reproduction alter the probability of establishment of beneficial mutations when deleterious mutations are common? Roughly speaking, the answer has to do with the fact that, in asexual populations, only the very fittest individuals have a chance of having descendants in the distant future, while this is certainly not the case for sexual populations. Thus, in an asexual population, a beneficial mutation of small effect must arise among the select few if it is to be established, and this requirement decreases the probability of establishment.

To take an example, consider the case of an adult with one deleterious mutation and no beneficial mutations when $\phi = 0$, $s_d = 0.02$, and $U = 0.25$. One deleterious mutation is very few in comparison with the population average of 12.5, and so, the adult in question will have a relatively high fitness (with 1.258 being the expected number of seeds that will grow to reproductive age for this adult). However, adults with one deleterious mutation will produce an average of only 0.980 seeds that will also have just one deleterious mutation, and that will grow to adulthood. This figure is less than one, and so, after a very large number of generations have elapsed, the adult in question has virtually no chance of having descendants who also have only one deleterious mutation. Since descendants with multiple deleterious mutations will be less fit than their ancestor, this argument can be extended to show that the adult in question will leave no descendants at all after a sufficiently large number of generations has elapsed. In fact, in the absence of beneficial mutations, asexuality means that only those adults who are entirely free of deleterious mutations can hope to leave descendants over the long term.

With the foregoing considerations in mind, it should be clear that, if a beneficial mutation has a sufficiently small effect, then it must initially appear in an adult with no deleterious mutations, or it can not become established in an asexual population (this is the case, for example, when $s_d = 0.02$ and $s_b = 0.01$). The probability of such a fortuitous appearance is typically very small. This is because the frequency of members of the mutation-free class is $\exp(-U/s_d)$ (HAIGH 1978). For example, when $s_d = 0.02$ and $U = 0.25$, members of the mutation-free class comprise only 0.000373% of the adult population (at equilibrium). If the effect of the beneficial mutation is larger, then the conditions for establishment are more relaxed, but even when $s_b = 0.1$ it can be shown that the beneficial mutation must initially occur in an adult with no more than four deleterious mutations if it is to have a chance of becoming established. At equilibrium, such individuals comprise only 0.535% of the population (assuming $s_d = 0.02$ and $U = 0.25$).

Figure 2 shows that, under asexuality, even if a beneficial mutation is doomed to extinction by the processes just described, it may rise to a high copy number before it is eliminated from the population. This occurs because asexuality produces a high level of correlation in the average of fitness of beneficial-mutation-bearing individuals from one generation to the next. Thus, if a beneficial mutation is initially associated with a relatively fit genotype, it is likely that adults carrying the beneficial mutation will continue to be relatively fit for a substantial number of generations, even if their fitness eventually erodes. It should be recognized, however, that the pattern shown in Figure 2 might not appear in a relatively small population. As the number of copies of a beneficial mutation increases into the thousands, it can cause an increase in the average fitness of a population that contains, say, only ten or twenty thousand members. This will cause a decrease in the relative fitness of individuals that carry the beneficial mutation. As a result,

in relatively small populations, beneficial mutations that will eventually become extinct might not achieve the sorts of copy numbers that are sometimes observed in the extremely large (effectively infinite) populations studies here.

One problem with the approach taken in this study is that I have assumed an infinite population ($N = \infty$). In an infinite population a beneficial mutation that initially arises in a single copy can never (in finite time) attain a substantial frequency because its initial frequency is infinitesimal. If the mutation becomes established, then its numbers will continually grow, but the frequency of the mutation will always be vanishingly small, except in the limit as the time since the initial appearance of the mutation becomes infinite. Another problem is that, under a biologically reasonable mechanism of mutation, one might expect the constant appearance of all possible beneficial mutations during each successive generation in an infinite population. The probability that a given type of beneficial mutation will arise *de novo* may be tiny for a given newborn individual, but so long as this probability is finite, we can expect an infinite number of new occurrences in an infinite population. This is contrary to my assumption that beneficial mutations are rare.

Fortunately, these difficulties with the $N = \infty$ assumption are more apparent than real. They tell us what we already know, that infinite populations are biologically unrealistic, and that they should only be used to approximate the case of very large populations. In a large (but not infinite) population, established mutations will certainly achieve a substantial frequency in finite time. Furthermore, we have data that strongly suggests that all possible beneficial mutations will not necessarily arise at one time even in very large microbial populations (*e.g.*, PAQUIN and ADAMS 1983; LENSKI *et al.* 1991). The calculations made in this paper will be approximately correct so long as the distribution of deleterious mutations is similar to a Poisson distribution, and this requirement can be fulfilled by a finite population, so long as it is sufficiently large (HAIGH 1978). Thus, there is no real problem with the $N = \infty$ assumption, as long as one keeps in mind that the calculations are meant to be a useful approximation for the case of very large (but finite) populations.

It may be worth pointing out that, while the population is assumed to have an infinite number of members, the fate of beneficial mutations (*i.e.*, establishment or loss) is a random process. This may seem surprising to some readers, as infinite populations are usually associated with deterministic processes. The reason for the non-deterministic character of evolution in the case of the models studied here is that the number of copies of a beneficial mutation is finite at any given (finite) time after the initial appearance of the mutation.

It seems that R. A. FISHER was aware of some of the consequences of asexuality that have been described here, though his wording is a bit vague [see FISHER (1930), pp. 121–122]. In addition, KEIGHTLEY and HILL (1983) and BIRKY and WALSH (1988) have produced relevant calculations. The results from both of these studies suggest that the probability of establishment of a beneficial mutation decreases when deleterious mutations are present. However, in contrast to the results reported here, the observed effects in both of these studies were modest in size, even in the absence of recombination. I have carried out preliminary investigations that suggest that the modest size of the effects observed in these studies is a consequence of the fact that both of them used small population sizes (less than 81 adults). Another theoretical study that is closely related to the one presented here was recently published by CHARLESWORTH *et al.* (1993), but they focused on the effects of deleterious mutations on neutral variation, rather than on beneficial mutations.

MANNING and THOMPSON (1984) have produced a model that is nearly identical to the one studied here, except that their formal calculations were restricted to the case of complete asexuality ($\phi = 0$). They do not consider the case of partial sexuality ($0 < \phi < 1$), and they dismiss the case of obligate sexuality ($\phi = 1$) with a single sentence, asserting that the probability of fixation of a beneficial mutation in this case will be equal to $2s_b$. In fact, Table 1 reveals that this assertion may be at least approximately correct for small s_b. However, it appears to be a somewhat wide of the mark for $s_b = 0.1$. Furthermore, it is important to keep in mind that the results I have reported here were generated under the assumption of loose linkage between all loci. I suspect that, with more realistic assumptions about linkage (*i.e.*, tight linkage between adjacent loci) the probability of fixation of beneficial mutations might be considerably less than $2s_b$, even in obligately sexual populations with small values of s_b.

Unfortunately, there are also problems with MANNING and THOMPSON's calculations in the case that they do consider formally (*i.e.*, obligate asexuality, where $\phi = 0$). These problems come from two sources. First, MANNING and THOMPSON develop their analysis by using the approximate solution to Equation 1 (*i.e.*, $E \approx 2s_b$). As pointed out above, this is appropriate only in the case where s_b is small. The second, and probably more important problem with their analysis can be seen after a close inspection of their equations. In making their calculations, MANNING and THOMPSON have ignored the possibility that a beneficial mutation that arises in an individual with k deleterious mutations will become established, even though it is lost from the class of individuals with only k deleterious mutations. However, such an occurrence is possible for many choices of parameter values. Ignoring this possibility can lead to substantial errors in the estimates of E. For example, in the

case where $U = 0.1$, $s_d = 0.025$ and $s_b = 0.1$, MANNING and THOMPSON estimate $E \approx 0.031$. Table 1 gives an estimate of E for the same parameter values that is 65% higher ($E \approx 0.0510$). This suggests that MANNING and THOMPSON's methods can substantially underestimate E when $s_b > s_d$.

The results presented here suggest that, when deleterious mutations are common, sex and recombination can greatly enhance the probability that a beneficial mutation will become established. Thus, sexual species may adapt more quickly than asexual ones, and this may help to explain the apparently short-lived nature of many asexual lines (MAYNARD SMITH 1978; BELL 1982). The results may also have bearing on other phenomena, such as the evolution of outbreeding and the evolution of non-zero recombination rates. In addition, the results suggest that low rates of outcrossing or recombination can lead to a reduction of neutral genetic variation when deleterious mutations are common (as do the results of CHARLESWORTH *et al.* 1993). This may help to explain some of the data on genetic variation (AGUADÉ *et al.* 1989; BEGUN and AQUADRO 1992).

If a sexual population adapts relatively quickly, then it could eventually gain an enormous advantage over competing asexual populations. However, this might take a long time, particularly if beneficial mutations are rare on a population-wide-basis, and if they have very small effects. On the other hand, beneficial mutations that improve fitness by around 10% have been observed to occur regularly in laboratory microbial populations (PAQUIN and ADAMS 1983; LENSKI *et al.* 1991), and if this sort of situation is common in most natural populations, then the processes described here could confer an unchallengeable advantage on sexual populations relatively quickly. Unfortunately, data on beneficial mutations in natural populations is very scarce, and much more information will be required before the power and time scale of the processes described herein can be properly accessed.

The results reported here may be helpful in predicting the likely outcome of competitions between obligately sexual species and asexual "species." As such, they might be usefully integrated with NUNNEY's (1989) theory on group selection and sex, which appears to show that group selection can be an effective force favoring the evolution of sexual reproduction. It is less clear that the results can be useful in explaining how obligately sexual species can be successful when competing against species that produce a substantial fraction of their offspring asexually, and the rest sexually. For the parameter values studied, the difference in the probability of establishment of beneficial mutations seems to become rather small (or absent) as ϕ increases above 0.1 or 0.25. However, it should be recognized that even small differences in the rate of accumulation of beneficial mutations can eventually produce large differences in fitness, and the simulation methods used here are subject to sampling error, so that small differences in E can escape notice. Furthermore, examination of Table 1 suggests that the effect of ϕ upon E becomes more pronounced even for relatively large values of ϕ as U increases. Because of computational constraints, it was not possible to study values of U above 0.5, but such values may well be biologically justified (KONDRASHOV and TURELLI 1992), and incorporation of high values of U might help to illuminate the evolutionary forces that maintain obligate sexuality. In addition, it seems possible that biologically realistic values for the rate of recombination (instead of free recombination, which was assumed here) will help to show the advantage of obligate sexuality.

It would be useful to model a population similar to the one described here, but allow for heritable variation in the propensity to reproduce sexually. Such an approach should help to reveal whether the processes described in this paper can help to explain sex in the context of selection within-populations, rather than in the context of selection between populations. Once again, it seems possible that realistic levels of recombination would help the success of alleles that enhance the rate of sexual reproduction, but this remains to be shown.

As a final point, it is important to recognize that the rate at which beneficial mutations appear in a population is likely to increase with the size of that population (WEBER and DIGGINS 1990; CABALLERO *et al.* 1991), and this may limit the relative adaptability of low density sexual populations. Nevertheless, in light of the very large enhancement in the probability of establishing beneficial mutations that can be conferred by sexuality, it seems possible that a sexual species could adapt more quickly than an asexual one, even if its population density was tiny in comparison to the density of its asexual competitor.

Thanks are due to M. ANDERSON, N. H. BARTON, M. BULMER, W. D. HAMILTON, W. G. HILL, A. S. KONDRASHOV, R. E. LENSKI, S. C. HEATH and M. TURELLI for useful discussion and assistance. This research was supported by Natural Environment Research Council grant GR3/8367 and Science and Engineering Research Council grant GR/H/09928.

LITERATURE CITED

AGUADÉ, M., N. MIYASHITA and C. H. LANGLEY, 1989 Reduced variation in the yellow-achaete-scute region in natural-populations of *Drosophila melanogaster*. Genetics **122:** 607–615.

BEGUN, D. J., and C. F. AQUADRO 1992 Levels of naturally-occurring DNA polymorphism correlate with recombination rates in *Drosophila melanogaster*. Nature **356:** 519–520.

BELL, G., 1982 *The Masterpiece of Nature*. University of California Press, San Francisco.

BIRKY, C. W., and J. B. WALSH, 1988 Effects of linkage on rate of molecular evolution. Proc. Natl. Acad. Sci. USA **85:** 6414–6418.

CABALLERO, A., M. A. TORO and C. LÓPEZ-FANJUL, 1991 The response to artificial selection from new mutations in *Drosophila melanogaster*. Genetics **128:** 89–102.

CHARLESWORTH, B., 1990 Mutation-selection balance and the evolutionary advantage of sex and recombination. Genet. Res. **55:** 199–221.

CHARLESWORTH, B., D. CHARLESWORTH and M. T. MORGAN, 1990 Genetic loads and estimates of mutation rates in highly inbred plant populations. Nature **347:** 380–382.

CHARLESWORTH, B., M. T. MORGAN and D. CHARLESWORTH, 1993 The effect of deleterious mutations on neutral molecular variation. Genetics **134:** 1289–1303.

CROW, J. F., 1979 Minor viability mutants in Drosophila. Genetics **92** (Suppl.) s165–s172.

DOBZHANSKY, T., 1970 *Genetics of the Evolutionary Process.* Columbia University Press, New York.

FELSENSTEIN, J., 1974 The evolutionary advantage of recombination. Genetics **78:** 737–756.

FELSENSTEIN, J., 1988 Sex and the evolution of recombination, pp. 74–86 in *The Evolution of Sex: An Examination of Current Ideas,* edited by R. E. MICHOD and B. R. LEVIN. Sinauer Associates, Sunderland, Mass.

FISHER, R. A., 1922 On the dominance ratio. Proc. R. Soc. Edinb. **42:** 321–341.

FISHER, R. A., 1930 *The Genetical Theory of Natural Selection.* Clarendon Press, Oxford.

HAIGH, J., 1978 The accumulation of deleterious genes in a population– Muller's ratchet. Theor. Popul. Biol. **14:** 251–267.

HALDANE, J. B. S., 1927 A mathematical theory of natural and artificial selection. V. Selection and mutation. Proc. Camb. Phil. Soc. **23:** 838–844.

HAMILTON, W. D., R. AXELROD and R. TANESE, 1990 Sexual reproduction as an adaptation to resist parasites (a review). Proc. Natl. Acad. Sci. USA **87:** 3566–3573.

HILL, W. G., AND A. ROBERTSON, 1966 The effect of linkage on limits to artificial selection. Genet. Res. **8:** 269–294.

HOULE, D., D. K. HOFFMASTER, S. ASSIMACOPOULOS and B. CHARLESWORTH 1992 The genomic mutation rate for fitness in Drosophila. Nature **359:** 58–60.

KEIGHTLEY, P. D., and W. G. HILL, 1983 Effects of linkage on response to directional selection from new mutations. Genet. Res. **42:** 193–206.

KIMURA, M., 1983 *The Neutral Theory of Molecular Evolution.* Cambridge University Press, Cambridge.

KIRKPATRICK, M., and C. D. JENKINS, 1989 Genetic segregation and the maintenance of sexual reproduction. *Nature* **339:** 300–301.

KONDRASHOV, A. S., 1988 Deleterious mutations and the evolution of sexual reproduction. Nature **336:** 435–440.

KONDRASHOV, A. S., and M. TURELLI, 1992 Deleterious mutations, apparent stabilizing selection and the maintenance of quantitative variation. Genetics **132:** 603–618.

LENSKI, R. E., M. R. ROSE, S. C. SIMPSON and S. C. TADLER, 1991 Long-term experimental evolution in *Escherichia coli.* I. Adaptation and divergence during 2,000 generations. Am. Nat. **138:** 1315–1341.

MANNING, J. T., and D. J.THOMPSON, 1984 Muller's ratchet accumulation of favourable mutations. Acta Biotheor. **33:** 219–225.

MAYNARD SMITH, J., 1978 *The Evolution of Sex.* Cambridge University Press, Cambridge.

MICHOD, R. E., and B. R. LEVIN (Editors), 1988 *The Evolution of Sex: An Examination of Current Ideas.* Sinauer Associates, Sunderland, Mass.

MUKAI, T., S. T. CHIGUSA, L. E. METTLER and J. F. CROW, 1972 Mutation rate and dominance of genes affecting viability in *Drosophila melanogaster.* Genetics **72:** 335–355.

MULLER, H. J., 1932 Some genetic aspects of sex. Am. Nat. **66:** 118–138.

NUNNEY, L., 1989 The maintenance of sex by group selection. Evolution **43:** 245–257.

OHTA, T., 1992 The nearly neutral theory of molecular evolution. Annu. Rev. Ecol. Syst. **23:** 263–286.

PAMILO, P., M. NEI and W. H. Li, 1987 Accumulation of mutations in sexual and asexual populations. Genet. Res. **49:** 135–146.

PAQUIN, C. E., AND J. ADAMS, 1983 Relative fitness can decrease in evolving asexual populations of *S. cerevisiae.* Nature **306:** 368–371.

WEBER, K. E., and L. T. DIGGINS, 1990 Increased selection response in larger populations. II. Selection for ethanol resistance in Drosophila melanogaster at two population sizes. Genetics **125:** 585–597.

WILLIAMS, G. C., 1975 *Sex and Evolution.* Princeton University Press, Princeton, N.J.

WIENER, P., M. W. FELDMAN and S. P. OTTO, 1992 On genetic segregation and the evolution of sex. Evolution **46:** 775–782.

Communicating editor: M. BULMER

APPENDIX 1

In the simulations, a total of 10,000 trials were run for each choice of parameter values. Before running any of the trials for a given set of parameter values the equilibrium frequency distribution in the absence of beneficial mutations was produced. (The generation of this distribution is a deterministic process, so it was only necessary to do it once for each set of parameter values.) In the general case (where $0 < \phi < 1$) the frequency distribution was generated by iterating for a number of generations until an equilibrium was approached. These iterations were initiated with a mutation-free infinite population. Each generation, the frequency distribution of asexually produced offspring was created by first calculating the relative contribution of each class of adult (*i.e.*, contributions from adults with zero deleterious mutations, one deleterious mutation and two deleterious mutations). The relative contribution of each class was assumed to be proportional to $(1 - s_d)^k$, where k is the number of deleterious mutations for that class. The probability that an asexually produced offspring from a parent with k deleterious mutations would have j deleterious mutations was assumed to equal zero if $j > k$, and was assumed to equal $P(j - k, U)$ if $j \geq k$, where $P(i, x)$ gives the height of the Poisson distribution for i successes when the parameter of the distribution is x. A similar process was used to generate the frequency distribution of the sexually produced offspring. In particular, the contribution of adults with k deleterious mutations to the "gamete pool" was assumed to be proportional to $(1 - s_d)^k$. Under the assumptions of the model, the probability that a gamete produced by an adult with k deleterious mutations will have j deleterious mutations is equal to $(\sum_{i=0}^{j} B(i, k, 0.5))P(j - i, (U/2))$, where $B(b, d, x)$ is the height of the binomial distribution for b successes in d trials, given that the probability of a success on each trial is equal to x. This expression embodies the assumptions of the model, since the infinite population and infinite number of loci assumptions imply that deleterious mutations will always be in heterozygous state, and thus, with free recombination, the probability that a given deleterious mutation will be incorporated into a particular gamete is 0.5, regardless of whether or not other mutations are also incorporated. Furthermore, to produce a gamete with j deleterious mutations, i mutations must be transferred from the parent, and $j - i$ new mutations must also be incorporated, for some value of i that satisfies $0 \leq i \leq k$. After gamete formation there is random pairing of gametes to form zygotes. Note that there is no need to distinguish between the two different gamete pools (for ovules and pollen) because the infinite-population assumption assures that they will both have the same distribution of genotypes at all times.

The only exception to the procedures just described arose when the number of deleterious mutations in the class of gametes or zygotes being produced was in excess of a pre-set maximum, which will be called Z. Selection of some maximum value was necessary for computer analysis, as without it there would have been an infinite number of different types of individual (although those with very large numbers of deleterious mutations would be exceedingly rare). Experimentation revealed that, if Z was in excess of a certain level, then its exact value had very little effect on the results. This observation is in line with expectations. After further experimentation, the value of Z selected was 34 for those simulations in which $U/s_d = 12.5$. Furthermore, the Z was set at 54, 20 and 14 for U/s_d values of 25, 6.25 and 4, respectively. Zygotes or gametes with more than Z deleterious mutations were treated as identical to zygotes or gametes with exactly Z deleterious mutations. Thus, for example, if $U/s_d = 12.5$, then the union of two gametes each with 20 deleterious mutations was presumed to produce a zygote with only 34 deleterious mutations, rather than 40. Of course, this is at variance with the biological facts, but it has virtually no effect on the results because, when $U/s_d = 12.5$, zygotes with 40 deleterious mutations are exceedingly rare.

After generating the distribution of sexually produced and asexually produced offspring, the two distributions were combined in proportions ϕ and $1 - \phi$, respectively, to create the distribution of adult genotypes. In other words, the proportion of adults with k deleterious mutations in the next generation was produced by summing the product of ϕ and the proportion of offspring with k deleterious mutations among the sexually produced offspring with the product of $1 - \phi$ and the proportion of offspring with k deleterious mutations among the asexually produced offspring. Because of the $N = \infty$ assumption, this procedure is equivalent to choosing N offspring at random from the total pool of offspring to produce the population of adults for the next generation. Of course, when $\phi = 1$ it was not necessary to generate asexually produced offspring, and when $\phi = 0$ it was not necessary to generate sexually produced offspring. Thus, in these two cases, the appropriate omissions were made to save computer time.

The iterations were stopped and the equilibrium frequency-distribution was recorded once the absolute value of the per-generation change in average number of deleterious mutations per individual was less than 10^{-8} for 100 consecutive generations. In all cases, the resulting distribution was virtually indistinguishable from a Poisson distribution with mean equal to U/s_d for the first Z classes, and these classes include all but a tiny fraction of the population. The distribution of gametes that is produced by the equilibrium distribution of adults was also recorded for future use.

It is straightforward to show that, under the assumptions of the model, the number of offspring that are destined to grow to adulthood that an adult with genotype i produces by asexual means follows a Poisson distribution with the mean number given by $(1 - \phi) W_i/\bar{W}$, where $\bar{W}$ is the average number of seeds produced per adult. If the adult carries a beneficial mutation, then all of the asexually produced offspring will also carry the beneficial mutation. The number of offspring destined to mature to adulthood to which an adult with genotype i contributes genetic material by sexual means also follows a Poisson distribution, and the mean in this case is given by $2\phi W_i/\bar{W}$. The adult can contribute to these offspring by means of pollen or ovules. Because $N = \infty$, we are guaranteed that the proportion of individuals that carry a beneficial mutation will always be negligible for any finite number of generations after the first appearance of the mutation. This observation implies that the beneficial mutation will always be in heterozygous state for any finite number of generations after the introduction of the mutation. Thus, an adult who carries the beneficial mutation has a 50% chance of passing it on to a given sexually derived offspring. This means that the number of offspring that are destined to mature to adulthood to which a beneficial-mutation-bearing adult contributes genetic material by sexual means, and that also carry the beneficial mutation, follows a Poisson distribution with mean equal to $\phi W_i/\bar{W}$.

The fact that, under the $N = \infty$ assumption, the frequency of the beneficial mutation remains negligible for any finite number of generations after the initial introduction of the beneficial mutation is convenient for several additional reasons. First, it means that the value of $\bar{W}$ calculated at the pre-introduction equilibrium can be used to calculate the parameters of the Poisson distributions mentioned in the preceding paragraph. This is because there will be no appreciable change in the value of $\bar{W}$ at any time that is a finite number of generations after the introduction of the beneficial mutation. Thus, the parameters of these distributions can be calculated once only for each set of parameter values. Similarly, because for any finite number of generations after the introduction of the beneficial mutation, the distribution of genotypes that do not incorporate the beneficial mutation will undergo only a negligible disruption, there is no appreciable change during a finite period of time in the probability of a union between any given type of beneficial-mutation-bearing gamete and any given gamete that does not bear the beneficial mutation. Furthermore, the probability of a union between two beneficial-mutation-bearing gametes is so small that it can be ignored without any appreciable alteration of the results.

Consider an adult that carries the beneficial mutation (in heterozygous state) and i deleterious mutations. Using the points outlined above in this appendix, it is straightforward to calculate a probability distribution for the number of offspring to which this adult contributes genetic material (including the beneficial mutation) by means of both sexual and asexual reproduction given that the offspring in question mature to adulthood and

carry exactly j deleterious mutations. When $\phi > 0$, these calculations must take into account the equilibrium distribution of genotypes that is established before introduction of the beneficial mutation. The resulting distribution is Poisson, the mean of the distribution will not change during any finite number of generations. These calculations were made for $0 < i, j > Z$. The resulting $(Z + 1) \times (Z + 1)$ matrix was then employed to carry out the 10,000 trials that were completed for each set of parameters.

Each of the 10,000 trials run for a given set of parameters was initiated by randomly selecting a genotype from the equilibrium distribution that is established in the absence of beneficial mutations, and then incorporating a single beneficial mutation into that genotype. The resulting individual is assumed to be among the N adults that can contribute offspring to the next generation. The $(Z + 1) \times (Z + 1)$ matrix described above is used to generate the collection of beneficial-mutation-bearing adults during each successive generation. There are always a finite number of beneficial-mutation-bearing adults, and so the progress of evolution after introduction of the beneficial mutation is a stochastic process.

Trials were terminated when the beneficial mutation was lost, or when it was deemed to be established. A beneficial mutation was deemed to be established when the all of the following conditions were satisfied. (1) At least 1,000 copies of the beneficial mutation were present in the population. (2) The average fitness of adults carrying the beneficial mutation was in excess of one. (3) The absolute value of the per-generation change in the average fitness of adults carrying the beneficial mutation was less than 10^{-8} for at least 100 consecutive generations.

Poisson processes are additive in the sense that, if we combine L Poisson processes with parameters λ_1, λ_2, . . . , λ_L, we obtain a new Poisson process with parameter λ^*, where $\lambda^* = \sum_{k=1}^{L} \lambda_k$. The computer program used in the simulation studies exploited this fact to decrease the time required to make the calculations necessary to proceed from one generation to the next. This was possible because, as explained above, the production of offspring of type j that are destined to mature to adulthood by an individual of type i is a Poisson process. Thus, it was possible to select the number of adults of type j that carry the beneficial mutation during each successive generation by calculating the total expected number of such individuals, and then using the result of this calculation as the parameter of a Poisson process. Consequently, the time it took for the computer program to proceed from one generation to the next did not depend directly on the number of adult bearers of the beneficial mutation. Rather, it depended on the number of different types of adult bearers of the beneficial mutation. As there is a definite upper bound to this quantity (namely, $Z + 1$) there is also an upper bound to the length of time that it can possibly take to make the calculations necessary to proceed from one generation to the next. A similar technique can almost certainly be used in the context of other models to increase our understanding of evolution in large but finite populations.

The Poisson variates used in the course of the simulations were mostly generated using standard numerical methods. However, when the mean of a required Poisson distribution was in excess of 200, a normal distribution with equal mean and variance was used in place of a Poisson distribution. However, this should not cause any substantial alteration of the results because, when the mean of the distribution is in excess of 200, the normal approximation to the Poisson is very accurate.

APPENDIX 2

In those cases where $U = 0$, FISHER (1922) and HALDANE's (1927) analysis can be used to show that the value of E is given by the positive solution of Equation 1. When $\phi = 0$ and $s_b < s_d$ the value of E is calculated by multiplying $\exp(-U/s_d)$ by the positive solution of Equation 1. To prove that this calculation yields the correct result, assume $\phi = 0$ and consider a beneficial mutation that is destined to be established. Using HAIGH's (1978) analysis it is straightforward to show that the number of deleterious mutations among carriers of the beneficial mutation will eventually approach a distribution such that the proportion of carriers with i deleterious mutations is 0 for $i < M$ and $P(i - M, U/s_d)$ for $i \geq M$, where $P(i, x)$ is defined as in APPENDIX 1, and M is the smallest number of deleterious mutations associated with any carrier of the beneficial mutation (thus, when $\phi = 0$, M may increase during the early stages of the establishment, but it can never decrease, and it can not be less than the number of deleterious mutations in the genome of the individual within which the beneficial mutation initially arose). From this distribution, one can show that the average number of seeds for carriers of the established beneficial mutation will eventually approach $C(e^{-U})(1 + s_b)(1 - s_d)^M$, and their average fitness (as defined in APPENDIX 1) will approach $(1 + s_b)(1 - s_d)^M$ (although average fitness may initially exceed this value). Clearly, when $s_b < s_d$ the average fitness of carriers of the established beneficial mutation will exceed one if and only if $M = 0$. This implies that, when $s_b < s_d$, establishment is possible only if the first adult to carry the beneficial mutation is entirely free of deleterious mutations, and if deleterious-mutation-free individuals who carry the beneficial mutation are never lost from the population. HAIGH's (1978) analysis shows that the frequency of mutation-free adults before the beneficial mutation arises is $\exp(-U/s_d)$, and under the $\phi = 0$ assumption we can demonstrate that, for a mutation-free adult, the number of mutation-free seeds that will mature to adulthood follows a Poisson distribution with mean equal to $1 + s_b$. Thus, we can apply FISHER (1922) and HALDANE's (1927) analysis to prove that, when $\phi = 0$ and $s_b < s_d$, the probability of establishment is given by multiplying $\exp(-U/s_d)$ times the positive solution of Equation 1.