

Inbreeding depression and heterosis in a subdivided population: influence of the mating system

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Summary

We investigate the joint effects of gene flow and selfing on the level of inbreeding depression, heterosis and genetic load in a subdivided population at equilibrium. Low gene flow reduces inbreeding depression and substantially increases heterosis. However, in highly self-fertilizing populations, inbreeding depression is independent of the amount of gene flow. When migration occurs via pollen, consanguinity of the reproductive system could have a negative influence on subpopulation persistence, in contrast to the case of isolated populations. However, with only seed migration, genetic load and heterosis depend mildly on the mating system. From an evolutionary point of view, we reach two main conclusions: first, outcrossing is selected for if gene flow is low; second, intermediate levels of gene flow could promote mixed mating systems, especially when migration occurs through pollen.

1. Introduction

Many natural populations are arrayed into complexes of breeding units of subpopulations (Selander, 1970; Chesser, 1983; Barrett & Husband, 1990). Such subdivision will often develop when gene flow is spatially restricted (Ellstrand & Elam, 1993). Together with gene flow, the mating system affects the genetic structure of a population by influencing the genetic differentiation between, and the distribution of genotypes within, subpopulations (Chesser, 1991).

This article investigates how the joint effects of restricted gene flow and mating system affect important genetic properties of a population, such as inbreeding depression and genetic load. The knowledge of these effects is interesting for several reasons.

First, population subdivision can have an important effect on mating system evolution, in particular on self-fertilization (Uyenoyama, 1986; Waller, 1993; Ronfort & Couvet, 1995). The evolution of the selfing rate depends on: (i) the cost of outcrossing (Fisher, 1941) and (ii) the extent of inbreeding depression, which is the relative decrease in fitness of selfed relative to outcrossed progeny (Lande & Schemske,

1985). Restriction of gene flow can promote positive genetic correlations between neighbouring individuals, referred to as biparental inbreeding (Uyenoyama, 1986). Biparental inbreeding has two contrasting effects: it increases the genetic relatedness between parents and offspring, therefore decreasing the cost of outcrossing, while it decreases inbreeding depression by reducing fitness differences between selfed and outcrossed progeny (Waller, 1993).

Hence, the variation in inbreeding depression and genetic correlations with population subdivision will affect mating system evolution. Notice that, although the influence of mating system on inbreeding depression in single populations has been the subject of numerous theoretical and experimental studies (e.g. Charlesworth & Charlesworth, 1987; Barrett & Charlesworth, 1991; Carr & Dudash, 1996; Husband & Schemske, 1996), the effects of the interaction between population subdivision and mating system on the level of inbreeding depression are largely unknown.

Second, restricted gene flow between subpopulations substantially increases genetic load due to the accumulation of deleterious mutations, with severe consequences on population persistence (Lande, 1995; Couvet, 2002). Crosses between genetically differentiated subpopulations are expected to reduce genetic load by producing progeny of enhanced fitness,

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referred to as heterosis or hybrid vigour (Whitlock *et al.*, 2000). However, mating systems influence both the genetic variation between subpopulations and the effectiveness of selection within subpopulations (Barrett & Charlesworth, 1991; Chesson, 1991). Hence, genetic load and heterosis should jointly depend on mating system and level of gene flow.

In conclusion, the examination of inbreeding depression, genetic load and heterosis allows us to give some answers to questions of great biological importance, such as: (i) the influence of population subdivision on the evolution of mating systems, and (ii) the consequences of mating system on the persistence of a subdivided population, through its influence on genetic load. For this purpose, we used an infinite-island model of populations with various selfing rates at mutation–selection–drift–migration balance. Two types of gene flow were distinguished: pollen and seed flow.

2. Methods

(i) The model

In order to study the accumulation of partially recessive deleterious mutants in a subdivided population where there is a certain amount of self-fertilization, we used an island model consisting of an infinite number of identical subpopulations. Each subpopulation receives immigrants from the other subpopulations at a stable rate chosen at random. The assumption of an infinite number of subpopulations allows us to model only one subpopulation because each one is representative of the entire collection (Slatkin, 1985).

We first studied a single-locus model and then extended it to multiple loci by assuming multiplicative fitness effects and independence between loci. A two-allele model was assumed, A being the wild-type allele and a an unconditionally deleterious and partially recessive allele. We denote as D , H and R the frequency of the AA , Aa and aa genotypes in each subpopulation. For diploid species we assume the following scheme:

$$\begin{array}{ccc} AA & Aa & aa \\ \text{Fitness} & 1 & 1 - hs & 1 - s \end{array}$$

where h measures the degree of dominance of the deleterious mutant and s the magnitude of the deleterious effect in homozygotes. Mutation occurs from the allele A to allele a at rate u and at rate v in the other direction.

In populations fragmented into small subpopulations, as assumed in our study, genetic drift has an important effect on the distribution of allele (and genotype) frequencies. Allelic states are correlated in diploids under self-fertilization. Hence, we have to

characterize the sample in terms of genotype rather than allele frequencies (Gale, 1990; Schoen *et al.*, 1998).

To find the genotypic composition of the subpopulation at equilibrium, we used the transition-matrix approach (Ewens, 1979; Hedrick, 1985; Gale, 1990):

$$\mathbf{Q}_{t+1} = \mathbf{P}\mathbf{Q}_t, \quad (1)$$

where \mathbf{Q}_t is the column vector of all the possible genotype states in generation t . For a subpopulation of size N and for a given locus, there are $(N+1)(N+2)/2$ possible states ranging from complete loss ($D = 1, H = 0, R = 0$) to complete fixation ($D = 0, H = 0, R = 1$) of the mutant allele. \mathbf{P} is the square matrix of dimension $(N+1)(N+2)/2$ of transition probabilities. Each element of this matrix is the probability of the subpopulation having a certain genotype composition in generation $t+1$, given the genotype composition in generation t .

Computation of the elements of the transition matrix. Let $R_t = 1/N$, $H_t = k/N$, $D_t = (N-k-1)/N$, ($k, 1 = 0, \dots, N$) denote the fraction of individuals, censused after viability selection, that carry two, one and zero mutant alleles respectively.

The genotypes undergo mutation, which modifies their frequency as follows (neglecting terms in u^2, v^2):

$$D^* = D_t(1 - 2u) + H_tv, \quad (2a)$$

$$H^* = H_t(1 - u - v) + 2uD_t + 2vR_t, \quad (2b)$$

$$R^* = R_t(1 - 2v) + H_tu. \quad (2c)$$

We assume that pollen migration occurs at a rate m_p per generation. The frequency, q_p , of the mutant allele in the *pollen* pool will therefore be

$$q_p = q_o(1 - m_p) + m_pq_m, \quad (3)$$

where $q_o = R^* + H^*/2$, and q_m the frequency of the allele a in the *migrant* pool. Under the infinite island model, q_m equals the overall average frequency of the mutant allele, $q_m = \bar{q}$ (Crow & Kimura, 1970).

Following pollen migration, reproduction occurs. The fraction of ovules self-fertilized is denoted as *Self*. Since ovules are not dispersing, the frequency of the allele a in the ovules will be simply q_o . The genotype frequencies after reproduction are (Holsinger, 1986):

$$D_r = \text{Self} \left[D^* + \frac{H^*}{4} \right] + (1 - \text{Self})(1 - q_p)(1 - q_o), \quad (4a)$$

$$H_r = \text{Self} \frac{H^*}{2} + (1 - \text{Self})[q_p(1 - q_o) + (1 - q_p)q_o], \quad (4b)$$

$$R_r = \text{Self} \left[R^* + \frac{H^*}{4} \right] + (1 - \text{Self})q_pq_o. \quad (4c)$$

The seeds formed after reproduction disperse at a rate

m_s . The genotype frequencies in the progeny after seed migration are:

$$D_s = D_r(1 - m_s) + m_s D_m, \tag{5a}$$

$$H_s = H_r(1 - m_s) + m_s H_m, \tag{5b}$$

$$R_s = R_r(1 - m_s) + m_s R_m, \tag{5c}$$

where D_m , H_m , R_m , are the genotype frequencies among the migrants. The frequency of each genotype among the migrants is equal to the overall average genotype frequency ($D_m - \bar{D}$, $H_m - \bar{H}$, $R_m = \bar{R}$). The total migration rate will be denoted as $m = m_s + m_p/2$, and the number of migrants that enter a subpopulation per generation as Nm . We weight pollen migration rate by one-half because, for the same rate of migration, seeds disperse twice as many genes as pollen.

Finally, offspring undergo viability selection to form the next generation:

$$D = D_s/\bar{w}, \tag{6a}$$

$$H = (1 - hs)H_s/\bar{w}, \tag{6b}$$

$$R = (1 - s)R_s/\bar{w}, \tag{6c}$$

where \bar{w} represents the normalizer which ensures that the genotypes sum to unity.

Hence, the elements of the transition matrix can be calculated from the polynomial probability distribution with probabilities, D , H and R :

$$p_{ij,kl} = \text{Prob} \left[\left(D_{t+1} = \frac{N-i-j}{N}, H_{t+1} = \frac{i}{N}, R_{t+1} = \frac{j}{N} \right) \middle| (D_t, H_t, R_t) \right] \\ = \frac{N!}{(N-i-j)!i!j!} D^{N-i-j} H^i R^j. \tag{7}$$

To examine the equilibrium frequencies \mathbf{Q}_{eq} , we solved (1), with $\mathbf{Q}_t = \mathbf{Q}_{t+1} = \mathbf{Q}_{\text{eq}}$, using numerical methods (the procedures used are described by Press *et al.*, 1989). According to (3) and (5), the elements of the transition matrix \mathbf{P} are functions of the allele and genotype frequencies in the migrant pool. Under the infinite-island model, these frequencies are equal to the overall average frequencies in the whole population, which depend on the values of \mathbf{Q}_{eq} . Thus, to calculate \mathbf{Q}_{eq} , an iterative procedure was used until the overall average genotype and allele frequencies equal the genotype and allele frequencies among migrants.

(ii) Mean fitness

The mean fitness of a subpopulation at the selected locus was computed as follows:

$$w_t = 1 - hsH - sR, \tag{8}$$

where

$$H = \sum_{i,j=0}^N \frac{i}{N} q_{ij}, \tag{8a}$$

$$R = \sum_{j,i=0}^N \frac{j}{N} q_{ij}, \tag{8b}$$

where q_{ij} is the probability of having i heterozygotes and j homozygotes for the mutant allele at equilibrium, and is calculated by the transition-matrix approach, described above (see also Lynch *et al.*, 1995a). As well as calculating the mean fitness of the subpopulation as a whole, we are also interested in the mean fitness of the outcrossed and inbred progeny, denoted as w_x and w_s respectively.

$$w_x = 1 - 2hs(1 - q)q - sq^2, \tag{9}$$

where

$$q = \frac{H}{2} + R, \tag{9a}$$

$$w_s = 1 - sh\frac{H}{2} - s\left(R + \frac{H}{4}\right), \tag{10}$$

where H and R are calculated as above (8a, b).

Finally, the expected fitness of hybrids between two subpopulations will be

$$w_c = 1 - 2hs\bar{q}(1 - \bar{q}) - s\bar{q}^2, \tag{11}$$

where \bar{q} is the overall average frequency of the mutant allele, as in Whitlock *et al.* (2000).

As already mentioned, the multiple locus case can be treated approximately by assuming no associations between loci. In this case, the product of the values for single locus gives the total effect on mean fitness (Crow & Kimura, 1970). Hence, for either of the fitnesses calculated above, we have

$$W = \prod_{i=1}^K w_i, \tag{12}$$

where K is the number of loci. When slightly deleterious and lethal mutations were considered together, fitnesses for each class of mutants were obtained separately, and their product gave the total fitness (Wang *et al.*, 1999).

Although the hypothesis of unlinked loci is not realistic, the effect of linkage on genetic load is significant only for high rates of inbreeding or very tight linkage (Allard *et al.*, 1968; Simmons & Crow, 1977; Charlesworth *et al.*, 1992). Thus, we did not consider values of selfing rate, *Self*, close to unity. Furthermore, the assumption of independent loci was checked by means of stochastic multi-locus simulations, and was found to yield accurate results, at least for the selfing rates used in this study.

In our results, fitness of a subpopulation is scaled by the fitness expected in an effectively infinite

population. The parameter of interest is the rate of fitness decrease in relation to the degree of population subdivision and mating system, and not the absolute fitness values. The calculation of fitness in a partially self-fertilizing infinite population is based on the analysis of Ohta & Cockerham (1974).

(iii) *Within- and between-subpopulation inbreeding depression and heterosis*

The inbreeding depression due to a single generation of self-fertilization is defined as the proportional decrement in fitness of selfed progeny, w_s , compared with the fitness of outcrossed progeny, w_x , $\delta = 1 - w_s/w_x$ (Lande & Schemske, 1985; Charlesworth & Charlesworth, 1987). Note that outcrossing in this case means reproduction that takes place between randomly chosen individuals within subpopulations.

However, in a subdivided population, a further level of outcrossing exists between subpopulations. It is therefore meaningful to study the relative performance of the selfed progeny versus the progeny of crosses between subpopulations, denoted thereafter as $\delta_{bsc} = 1 - w_s/w_c$, where w_c is the expected fitness of individuals from crosses between different subpopulations.

Finally, we defined heterosis as the increase in fitness of outcrossed progeny in a subpopulation compared with progeny resulting from crosses between subpopulations (Mitton, 1993; Fry *et al.*, 1998), $H_{et} = 1 - w_x/w_c$. Note that the relation among inbreeding depression, heterosis and between-subpopulation inbreeding depression can be expressed as

$$1 - \delta_{bsc} = (1 - \delta)(1 - Het). \quad (13)$$

(iv) *Numerical values for deleterious mutations*

In most of our analysis, we consider only slightly deleterious mutations, for the following reasons. First, the frequency of lethal mutations at equilibrium is nearly independent of the effective population size (Nei, 1968; Bataillon & Kirkpatrick, 2000); that is, selection acts effectively against lethal mutants within subpopulations and does not allow genetic drift to drive these mutants to high frequencies even when gene flow is restricted. Consequently, inbreeding depression due to lethal mutations remains unchanged for all levels of gene flow, and results are almost identical to those for single populations. Second, lethal mutants do not contribute to the amount of heterosis (Whitlock *et al.*, 2000).

However, a complete estimation of inbreeding depression is important in the study of mating system evolution. Since lethals are expected to contribute significantly to the level of inbreeding depression (Charlesworth & Charlesworth, 1987), we take them

into account in our analysis concerning the evolution of selfing rate.

According to several experimental and theoretical studies concerning mostly viability in *Drosophila melanogaster*, mildly deleterious mutations are estimated to have a total genomic mutation rate of $U = 1$ per generation, fitness effect of $s = 0.01-0.05$, and dominance coefficient of $h = 0.2-0.4$ (Mukai, 1964; Mukai *et al.*, 1972; Simmons & Crow, 1977; Kondrashov & Houle, 1994; Johnston & Schoen, 1995; Lynch *et al.*, 1995*b*). Furthermore, observations on several other species suggest that mutation rates and fitness effects are of the order of magnitude found in *Drosophila* (review in Lynch *et al.*, 1999). For lethal and sublethal mutations, the total genomic mutation rate is estimated to be $U_L = 0.05$ (Simmons & Crow, 1977; Fry *et al.*, 1999), and the average dominance coefficient, $h = 0.02-0.03$ (Crow, 1993).

Results

(i) *Genetic load*

In the absence of gene flow, with the parameter values assumed, deleterious mutations are almost fixed in all subpopulations and fitness is practically zero. Migration reintroduces genetic variability in the subpopulations and therefore fitness increases with migration, whatever the mating system (Fig. 1). However, for high selfing rates, genetic load depends considerably on the agent of gene flow (pollen or seed). When gene flow occurs only via pollen, genetic

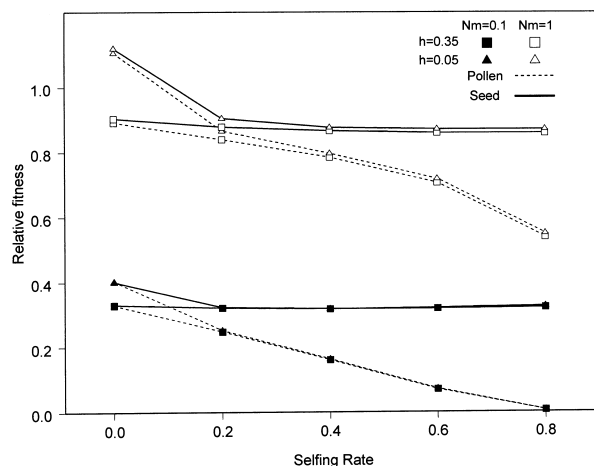


Fig. 1. The effect of selfing rate, number of migrants per generation and type of gene flow on subpopulation fitness. The fitness values are scaled by the fitness expected in an effectively infinite population. The selection coefficient of deleterious mutations is set to $s = 0.02$, the mutation rate per genome to $U = 1$, and the size of each subpopulation to $N = 40$. Two values of dominance were used: $h = 0.35$ and $h = 0.05$. Nm denotes the number of migrants that enter a subpopulation per generation.

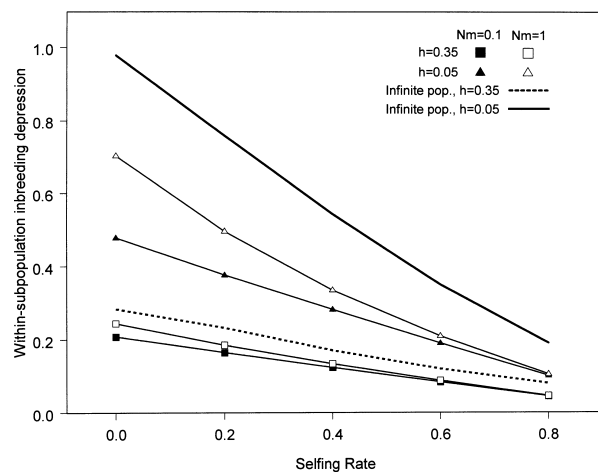


Fig. 2. Relationship between inbreeding depression and selfing rate for different migration rates. Inbreeding depression of the infinite population was calculated as in Kirkpatrick & Jarne (2000). Only pollen migration is considered ($m_p = 0$), results of seed migration being similar. Other parameter values are as in Fig. 1.

load in predominantly self-fertilizing subpopulations is significantly increased relative to the case of seed migration. The reason is that fertilization of ovules by pollen occurs only for the outcrossing proportion of the population. Pollen migration will therefore influence the genotype frequencies only of the progeny of outcrossing (the proportion $(1 - Self)$ in (4)). Pollen and seed migration have nearly equivalent effects when $m_s = (1 - Self)m_p/2$. With outcrossing, decreased dominance of deleterious alleles decreases the genetic load (see also Couvet, 2002). Conversely, in highly selfing populations, mutations are more likely to be homozygous, and thus the level of dominance has no effect on genetic load.

Finally, the influence of subpopulation size on our results was checked. Although larger populations are associated with lower genetic load, the relationship between genetic load and selfing rate remains identical for all population sizes. This is also true for inbreeding depression and heterosis.

(ii) Within-subpopulation inbreeding depression (δ)

In agreement with findings concerning single populations (Lande & Schemske, 1985; Charlesworth & Charlesworth, 1987; Charlesworth *et al.*, 1990), inbreeding depression is a decreasing function of the selfing rate. However, the rate of this decrease depends on two parameters: the gene flow between subpopulations and the dominance of deleterious alleles (Fig. 2).

When gene flow is restricted, inbreeding depression is low whatever the selfing rate. This is because genetic drift drives deleterious mutants to high frequencies for all mating systems. Hence, there is little genetic

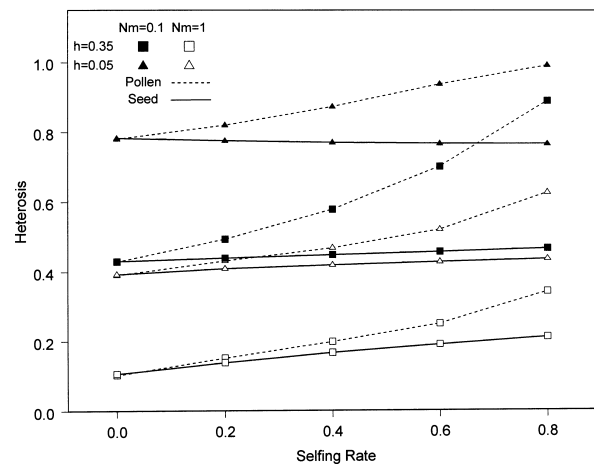


Fig. 3. The effect of mating system on heterosis in relation to migration type (pollen or seed). Parameter values are as in Fig. 1.

variation, and no difference in the fitnesses of selfed and outcrossed progeny. Gene flow, however, increases inbreeding depression in random-mating subpopulations, but has no effect on the level of inbreeding depression in highly self-fertilizing subpopulations. Similarly, low dominance of deleterious alleles leads to a substantial increase in inbreeding depression within outcrossing subpopulations, but has nearly no effect on inbreeding depression in highly selfing subpopulations. This is because within outcrossing populations, highly recessive alleles are masked in heterozygotes. Consequently, a single generation of selfing exposes these mutants in the homozygotes, and therefore results in a high difference in fitness between outcrossed and inbred progeny. Conversely, with high inbreeding most of the mutant alleles are in homozygous form, and therefore the level of dominance has reduced effects on the level of inbreeding depression (see also Charlesworth *et al.*, 1990). The relationship between inbreeding depression and selfing rate is almost independent of the nature of gene flow (pollen- or seed-mediated).

(iii) Heterosis

Heterosis is largest with low levels of dominance, low to intermediate selection coefficients ($Ns \sim 1$), and low migration rates (see also Whitlock *et al.*, 2000). However, pollen and seed migration differ in their influence on heterosis. While for pollen migration heterosis is an increasing function of selfing, for seed migration heterosis depends slightly on the selfing rate (Fig. 3). The reason is that heterosis reveals the degree to which deleterious mutations have accumulated within subpopulations. Consequently, the same relationship between migration mode and mating system is found for heterosis and genetic load.

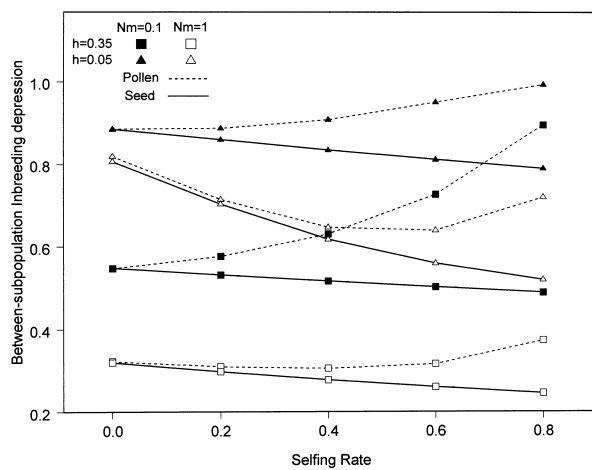


Fig. 4. The joint effect of gene flow and selfing rate on between-subpopulation inbreeding depression (δ_{bsc}). Parameter values are as in Fig. 1.

(iv) *Between-subpopulation inbreeding depression* (δ_{bsc})

Between-subpopulation inbreeding depression (δ_{bsc}) is greatest when gene flow is low. Moreover, although gene flow restores some genetic variability, and therefore fitness within subpopulations, high δ_{bsc} values are observed even for intermediate rates of gene flow (Fig. 4).

The relationship between δ_{bsc} and the selfing rate depends on the type of gene flow (seed or pollen flow). When gene flow is seed-mediated, δ_{bsc} always decreases with the selfing rate. In contrast, when only pollen disperses, the relationship between δ_{bsc} and selfing rate changes according to the degree of population subdivision. With restricted pollen migration, δ_{bsc} increases as the selfing rate increases, because higher genetic load is present in highly self-fertilizing populations. For intermediate rates of pollen migration, δ_{bsc} is minimized for intermediate rates of self-fertilization (Fig. 4). Purging within subpopulations occurs for low and intermediate selfing rates, therefore decreasing δ_{bsc} . However, for high selfing rates, purging is less efficient because of restricted genetic variability, and δ_{bsc} remains high (see the analysis in the paragraph concerning genetic load).

Moreover, with low dominance of deleterious alleles, δ_{bsc} increases substantially for all mating systems, the selection coefficient having a moderate effect on δ_{bsc} . For a given number of migrants and selfing rate, δ_{bsc} is generally greatest when both h and s are low. As seen, from equation (13), δ_{bsc} is maximized when both heterosis and inbreeding depression are maximized. It is already well established that inbreeding depression is greatest for highly recessive alleles (Charlesworth *et al.*, 1992; Byers & Waller, 1999) and heterosis for low dominance and selection coefficients (Whitlock *et al.*, 2000).

4. Discussion

The main goal of this study was to examine the joint effects of population subdivision and mating system on genetic load and inbreeding depression. Subdivision of a population tends to decrease within-subpopulation variation while increasing between-subpopulation variation. Decreasing gene flow will therefore result in a decrease of both inbreeding depression and fitness within subpopulations.

Mating systems can modify the effect of subdivision on genetic load and inbreeding depression. In the case of outcrossing, within-subpopulation inbreeding depression increases with gene flow. For selfing subpopulations, inbreeding depression is independent of the population subdivision; the level of heterozygosity, and therefore inbreeding depression, in such inbred populations is always low, regardless of the amount of gene flow. Moreover, enhanced selfing can substantially increase genetic load and heterosis when gene flow is pollen mediated.

(i) *The evolution of the selfing rate in a subdivided population*

One could argue that decreased inbreeding depression in subdivided populations, relative to a large undivided one, should lead to selection for higher selfing rates. However, this ignores the presence of biparental inbreeding resulting from restricted gene flow. With biparental inbreeding, the relatedness between parents and offspring is greater than when mating is between unrelated individuals. As a result, the ‘cost of outcrossing’ – the increase in parent-offspring relatedness under uniparental reproduction compared with random mating – is lowered; therefore the transmission advantage of genes increasing selfing is lowered with biparental inbreeding (Uyenoyama, 1986; Jarne & Charlesworth, 1993; Waller, 1993). Hence, selfing is promoted only when

$$c > \delta, \quad (14)$$

where c is the cost of outcrossing (Waller, 1993, Ronfort & Couvet, 1995). In order to estimate the impact of population subdivision on the selection of the selfing rate, calculation of the cost of outcrossing is therefore necessary.

The cost of outcrossing can be estimated as, $c = (1-r)/2$ (Ronfort & Couvet, 1995), where $r = 2F_{ST}/(1+F_{IT})$ (Michod & Hamilton, 1980) is the coefficient of relatedness between outcrossed individuals; $F_{ST} = 1/[1+2N(\frac{1}{2}m_p+m_s)(2-Self)]$ (Wang, 1997) and $F_{IT} = 1-(1-F_{ST})(1-F_{IS})$, with $F_{IS} = Self/(2-Self)$ (Crow & Kimura, 1970).

However, in a subdivided population there are two levels of outcrossing: within and between sub-

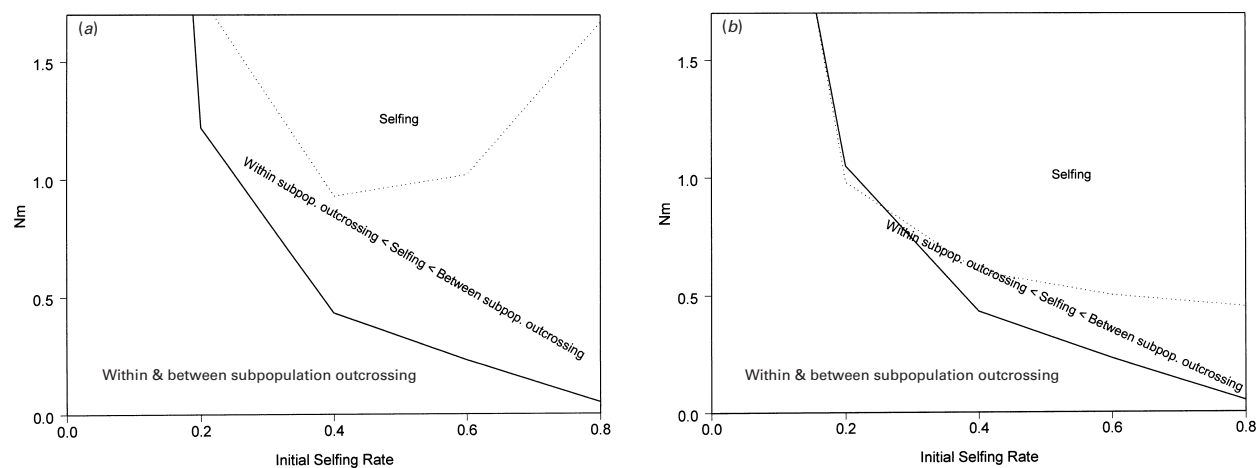


Fig. 5. Selection in selfing, depending on gene flow and initial selfing rate. The continuous curve corresponds to parameter combinations for which inbreeding depression equals the cost of outcrossing. The region below the curve favours outcrossing within subpopulations, while the region above the curve favours selfing. The dashed curve corresponds to parameter combinations for which between-subpopulation inbreeding depression (δ_{bsc}) equals one-half. The region below the dashed curve favours outcrossing between subpopulations, while the region above the dashed curve favours selfing. (a) Only pollen migration occurs ($m_p = 0$). (b) Only seed migration occurs ($m_s = 0$). The size of each subpopulation is set to $N = 20$. The parameter values of detrimental mutations are set to $h = 0.2$, $s = 0.02$, $U = 1$, and those of lethals to $h = 0.02$, $s = 1$, $U_L = 0.05$.

populations. Even if selfing is favoured in relation to within-subpopulation crosses, the substantial levels of δ_{bsc} and heterosis observed in our results suggest that crosses between subpopulations could prevent complete selfing from occurring. It is necessary, therefore, to examine under what conditions crosses between subpopulations would be promoted. Under the infinite-island model, individuals from different subpopulations are genetically unrelated. Selfing alleles therefore have a 50% transmission advantage relative to alleles promoting outcrossing between subpopulations. For crosses between subpopulations to be favoured, between-subpopulation inbreeding depression must counteract the transmission advantage of selfing, or $\delta_{bsc} > 0.5$.

In order to predict the evolutionary stability of a given mating system, we proceeded as follows. We allowed the population to reach equilibrium, with regard to the frequency of deleterious alleles, with a fixed selfing rate. A new mutant that promotes selfing can invade a subpopulation if two conditions are met: (i) the cost of within-subpopulation crosses, c , is higher than inbreeding depression, δ , and (ii) the cost of between-subpopulation crosses is higher than δ_{bsc} . To calculate inbreeding depression we considered both lethals and detrimental. Fig. 5 shows the outcome of this analysis.

When gene flow is large, and thus the population behaves almost as a single undivided one, our results are in agreement with previous studies, i.e. there are two evolutionarily stable strategies: complete outcrossing or complete selfing; which will be selected depends on the initial selfing rate (Charlesworth *et al.*, 1990, 1992).

For intermediate migration and selfing rates, selfing is promoted in relation to within-subpopulation outcrossing and, at the same time, between-subpopulation outcrossing is promoted in relation to selfing. Crosses between subpopulations could therefore prevent a modifier causing selfing from invading a population, and intermediate selfing rates would be preserved. Thus, mixed-mating systems could occur as a result of restricted migration. This result depends mildly on the type of migration: seed-mediated gene flow reduces the range of stability of mixed mating systems (Fig. 5b). Ronfort & Couvet (1995), with a model simulating a continuous population, where selection on the mating system was determined by modifier loci affecting the selfing rate, reached similar conclusions: population structure, resulting in their model from restricted seed and pollen dispersion and density-dependent recruitment, can maintain intermediate selfing rates.

Moreover, Fig. 5 suggests that a change in the population structure can lead to a shift in the mating system. Surprisingly, an increase in isolation between subpopulations is expected to lead to selection for lower selfing rates. The reason is that although inbreeding decreases with restricted gene flow, this is compensated by an even greater reduction in the cost of outcrossing (see also Ronfort & Couvet, 1995). A change in the population structure could occur when, for instance, insular habitats are occupied as a result of enhanced intra-island isolation. Indeed, loss of dispersibility occurs prominently on oceanic islands, due, for example, to modifications in the guild of pollinators (Carlquist, 1966a; Delph, 1990). Our results could therefore be consistent with an extensive

survey on the reproductive system of oceanic island populations which showed that these populations have developed mechanisms that promote outcrossing (dioecy, monoecy, dichogamy and various other floral conditions) in a much higher frequency than have continental populations (Carlquist, 1966*b*).

Note that the level of dominance has a marked influence on the pattern observed. We have deliberately chosen $h = 0.2$ for slightly deleterious alleles. For this value of dominance and mutation rate per genome, $U = 1$, inbreeding depression is greater than 0.5, and thus stability of outcrossing in infinitely large populations is ensured (Lande & Schemske, 1985). For higher levels of dominance, selection in favour of selfing appears for lower values of gene flow and lower selfing rates. With, for instance, $h = 0.35$, selfing is promoted even in an initially outcrossing population for almost all values of gene flow (as in Charlesworth *et al.*, 1990, for an infinite population). Furthermore, the range of gene flow values where potential stability of intermediate selfing rates occurs, diminishes.

Decreasing the total genomic mutation rate has similar effects to increasing dominance (results not shown). However, as long as the mutation rate is not too low ($U \geq 0.5$), the two main conclusions of our analysis hold: (i) outcrossing is preferentially selected for when gene flow is low; (ii) mixed-mating systems represent evolutionarily stable states for intermediate levels of gene flow, especially when migration occurs through pollen.

Finally, although our model should correctly predict whether alleles causing small changes in selfing rates will invade populations, further study of the evolution of selfing rate requires a more realistic model, which takes into account the coevolution of the cost of outcrossing and inbreeding depression with population structure as well as linkage for high selfing rates.

(ii) *Gene flow and population persistence*

Our findings suggest that subpopulation persistence could be largely determined by the interaction between population subdivision and mating system. In a subdivided population consanguinity of the mating system can have a negative influence on subpopulation persistence. In particular, when only pollen migration occurs, genetic load increases substantially with self-fertilization (Fig. 1). This result differs from that predicted for a single population, in which genetic load is a decreasing function of selfing rate (Charlesworth *et al.*, 1990, 1992). As a result, the decrease in genetic load from crosses between subpopulations is higher for higher selfing rates (Fig. 3). Evidence for differential pollen success of outcrossed

pollen has been documented in several self-compatible species (Richards, 2000 and references therein). In agreement with our results, Richards demonstrated that pollen-mediated gene flow into patches of consanguineous mating system (in this case, full-sib mating) generates higher heterosis than into patches comprised of unrelated individuals. Enhanced siring ability of outcrossed pollen in inbred subpopulations may act to reduce genetic differentiation among subpopulations and compensate the negative effects of inbreeding.

Note, however, that in highly selfing populations the relative benefits from gene exchange between subpopulations would depend on the migration mode. For the same number of migrating genes, seed flow is more effective in decreasing genetic load than pollen flow (Figs 1, 3). Hence, when we allow for seed migration, higher selfing rates result in only a mild increase of genetic load and heterosis.

(iii) *Limitations*

A limitation of the predictive value of our results arises from the fact that we did not consider alleles that represent local adaptations. In natural populations, environmental variability and subdivision could lead to subpopulations adapting to differential local conditions. In this case, crosses between subpopulations may result in outbreeding depression and therefore limit the benefits from enhanced gene flow (Waser & Price, 1994; Richards, 2000). One would expect that intermediate values of migration rate would be favoured in order to balance these two conflicting forces. It seems, however, difficult to derive general conclusions for the optimum level of genetic differentiation among subpopulations. The number of studies that would permit an evaluation of the intensity and the frequency of the phenomenon of local adaptation is still limited.

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