



## A comparison of management strategies for conservation with regard to population fitness

Jesús Fernández\* & Armando Caballero

Departamento de Bioquímica, Genética e Inmunología, Facultad de Ciencias, Universidad de Vigo, 36200 Vigo, Spain (\*Author for correspondence: [jesusfm@uvigo.es](mailto:jesusfm@uvigo.es))

Received 24 August 2000; accepted 7 February 2001

*Key words:* ex-situ conservation, fitness, genetic diversity, genetic drift, inbreeding

### Abstract

Computer simulations have been carried out to compare, under realistic genetic models, two methods proposed in the literature to retain genetic diversity in conservation programmes. In a two-step method, contributions of parents are set up to produce minimum coancestry (kinship) among the offspring, and this is independent of the mating system subsequently applied. In a single-step method, contributions and matings are decided simultaneously in order to minimise coancestry. The comparison is made in terms of maintained genetic diversity and in terms of population fitness. We conclude that the two methods maintain approximately the same genetic diversity but the latter induces higher levels of inbreeding, reducing the fitness of the population. Avoidance of close relatives' matings improves this latter method, but the fitness levels do not reach those of the two-step scheme. We also investigate the performances of different mating strategies in combination with minimum coancestry (two-step method), concluding that these mating systems do not substantially affect the effectiveness of the management. Finally, we illustrate how minimum group coancestry can be restricted to a minimum loss of fitness, if a measure of this is available for the individuals.

### Introduction

The final aim of an *ex-situ* conservation programme is the maintenance of captive populations of endangered species or breeds until their census is large enough to assure their survival, or their natural environment is restored (Hedrick and Miller 1992; Ballou and Lacy 1995; Ballou and Foose 1995; Lynch 1996). In any case all efforts must be devoted to guarantee the success of the population when reintroduced in the wild. The critical point from a genetic point of view is to keep the highest levels of genetic diversity, so that the evolvability of the population (the ability to face new environmental challenges) is not at risk, and preventing the adaptation to captive conditions.

The loss of genetic resources (i.e. loss of different alleles) in conserved populations is mainly due to genetic drift, the random variation of allelic frequencies caused by the low census numbers usually involved. Strategies that increase the effective popula-

tion size will lower the effect of genetic drift and, thus, they will maximise the amount of genetic information maintained in the population.

Different methods have been proposed in the literature to minimise or delay the loss of genetic diversity or the appearance of inbreeding. In some methods, the first step to carry out is the election of parents for the next generation or the number of offspring they should contribute. Thus, a strategy has been proposed that searches for the combination that minimises the group coancestry (mean kinship), i.e. the average coancestry between all possible parents, including reciprocals and self coancestries, weighted by the contributions of each individual (Ballou and Lacy 1995). Minimisation of group coancestry has been proven (theoretically and by simulations) to be the most efficient method to preserve genetic information (Meuwissen 1997; Fernández and Toro 1999; Caballero and Toro 2000). In a regular pedigree where relationships between individuals are uniform, the method converges with

the classical method of minimisation of variances of family size (Gowe et al. 1959; Crow and Kimura 1970; Wang 1997).

The second step that should follow, in combination with the previous one, is the way the parents should be mated to create the next generation. In this context there have been proposed strategies such as the ‘maximum avoidance of inbreeding’ method (Wright 1921), the compensatory mating, i.e. the mating between males with high average coancestry with females of low average coancestry (Caballero et al. 1996), or the minimum coancestry mating (Toro et al. 1988). These mating methods have been proven to be effective in controlling inbreeding in artificial selection schemes (Caballero et al. 1996; Sonesson and Meuwissen 2000) but their use in conservation programmes is not evident. For example, Ballou and Lacy (1995) pointed out that the mating between males of high average coancestry with females of low average coancestry (compensatory mating) will result in mixing rare and common alleles. Because, as a result, the number of copies of the rare alleles cannot be increased without also increasing that of the overrepresented alleles, the method should not be used in conservation programmes.

Alternatively, other methods have been proposed to arrange both the individual contributions and the mating scheme in just one step. This procedure has been sometimes called *mate selection*, and it is well known in the animal breeding field (Allaire 1980; Toro and Pérez-Enciso 1990; Klieve et al. 1994). One of these methods, applied to conservation programmes, was presented by Ballou and Lacy (1995). Based on minimisation of mean coancestry, they proposed an iterative method to find, not only which parent will contribute an offspring, but also the specific matings among them. It is not, a priori, evident what method for implementing minimum coancestry is more efficient, either a method that combines contributions and mating in a single step, or another that separates these into two independent steps.

Most studies on the above methods have been focused on the maintenance of neutral genetic variability, but little attention has been paid to the consequences of conservation methods on fitness related traits. In this paper, we use computer simulations with realistic models of fitness variation to compare the performance of two different ways of implementing the optimal management strategy based on minimisation of group coancestry. For one method (Ballou and Lacy 1995) contributions and matings

are decided simultaneously. For the other, the two procedures are decided in independent steps. We study the performance of different mating schemes used in conjunction with minimum group coancestry contributions. The comparison is made both in terms of neutral genetic variability and of the fitness of the population.

### Model assumptions and simulations

We carried out simulations for a dioecious species. An ancestral population was first set up in which gene frequencies are at mutation-selection-drift balance. Fitness was controlled by a large number (up to 5800) of biallelic loci at linkage equilibrium with effects,  $s$ , in homozygosis and dominance coefficients,  $h$ . For each locus, the genotypic fitnesses were 1,  $1-sh$ ,  $1-s$ , with frequencies  $p^2$ ,  $2pq$ ,  $q^2$ , for the  $AA$ ,  $Aa$ ,  $aa$  genotypes, respectively. Fitness action was assumed to be multiplicative among loci and to act through viability differences among individuals.

Two kinds of mutations were considered: mildly or moderately deleterious and lethals. All mutations were assumed to have the same per locus mutation rate ( $\lambda/5800$  and  $\lambda_L/5800$ , respectively) where  $\lambda(\lambda_L)$  is the average number, Poisson distributed, of new deleterious (lethals) mutations per haploid genome arising per generation and randomly assigned to non-segregating positions of the genome. The ancestral population was generated by running an unmanaged population with 1000 randomly mated breeding individuals. The population was initially devoid of variation, and this was gained by mutation (see mutational models below) until equilibrium with selection and drift was reached after 10000 generations.

Samples of 8, 24 or 48 individuals, the three considered population sizes ( $N$ ), were randomly taken from the ancestral population. Equal numbers of males and females and constant population size over generations were assumed, and management was assayed for a total of 20 discrete generations. The fitness of the conserved population was calculated every generation according to the mutations (deleterious and lethal) carried by every individual, and averaged over 100 replicates.

200 neutral multiallelic loci were also simulated interspersed between the selected loci in order to monitor changes in neutral genetic variation. Individuals in the sample taken out from the ancestral population carried different alleles at all these neutral loci in order to calculate probabilities of identity by

descent per locus. Neutral and selected loci were equally spaced in a genome length of 20 morgans, with cross-overs without interference occurring at randomly chosen places. Genetic diversity preserved in the population was measured by the allelic diversity (percentage of different alleles remaining in the population) and the gene diversity (expected heterozygosity), both averaged over neutral loci and replicates.

### *Mutational models*

Two different models for deleterious mutations were investigated:

– *Model I*: Data on viability in *Drosophila* and other species indicate a large amount of deleterious mutations of small effect (Lynch et al. 1999). For this model we assumed a mean mutational deleterious effect  $\bar{s} = 0.05$  and mean dominance coefficient  $\bar{h} = 0.36$ , with mutations occurring at a rate  $\lambda = 0.5$  per haploid genome and generation. The selection coefficients were sampled from a gamma distribution with shape parameter  $\beta = 1$ . This corresponds to an exponential distribution of gene effects that has been proposed as a likely distribution (see, e.g. Schultz and Lynch 1997). The dominance coefficient of mutations were obtained from an exponential function of the gene effects. The model is that proposed by Caballero and Keightley (1994), for which the dominance coefficient of a mutant is taken from a uniform distribution between 0 and  $\exp(-ks)$ , where  $k$  is a constant allowing the mean dominance coefficient to be the desired one (0.36 in this case).

– *Model II*: Alternative experimental data obtained in *Drosophila*, *C. elegans* and plants suggest that deleterious mutations occur at a low rate but with high average effect (Lynch et al. 1999; Keightley and Eyre-Walker 1999; García-Dorado et al. 1999; Bataillon 2000). For this model we assumed that the rate of mutations is  $\lambda = 0.03$ , the shape parameter of the mutant effects distribution is  $\beta = 2.3$ , and the mean effect is  $\bar{s} = 0.264$ . These parameters have been obtained through minimum distance estimation of *Drosophila* data (García-Dorado et al. 1999). The model of coefficients of dominance followed the same exponential function as above, but the average value was  $\bar{h} = 0.25$ . This figure is suggested by current analysis of *Drosophila* spontaneous mutations (García-Dorado and Caballero 2000).

In addition to the deleterious mutations we also assumed, for both models, that lethal mutations appear at a rate of  $\lambda_L = 0.015$  per haploid genome and generation, having a coefficient of dominance  $h = 0.02$  (Crow and Simmons 1983).

### *Management procedures*

In every replicate the initial individuals were randomly taken from the large ancestral population, so they were assumed to be unrelated. Before any conservation method was implemented the population underwent five unmanaged generations in order to generate a complex pedigree and differential coancestries between individuals. In each replicate, this starting point (generation 0) was identical for all treatments to avoid differences due to initial populations. The management procedures were as follows.

*Random (R)*: This is the reference case, used for comparison. One male and one female were randomly chosen and mated to generate an offspring. The fitness of this individual was calculated according to its genotype (number and effects of deleterious and lethal mutations carried in its genome). A random number between 0 and 1 was drawn and the individual survived (died) if this number was lower or equal (larger) than the fitness value. If the individual died, a new male and a new female were randomly taken from the available parents to generate a new offspring. This process went on until  $N$  offspring were generated. The sex of each offspring was randomly assigned at the end of every generation.

*Method proposed by Ballou and Lacy (1995) (BL)*: For every available parent the mean coancestry (i.e. the average coancestry of this parent with all the individuals in the population, including itself) was calculated. This value was used to sort out males and females separately. The male and female with the lowest mean coancestry were mated to generate an offspring. The descendant was evaluated for fitness and its survival decided as explained above. If the offspring died another individual was generated from the same couple until one survived. To get the second and following offspring, average parental coancestries were re-evaluated but computing only the coancestry with the offspring already created. The pattern was repeated until  $N$  new individuals were generated. Sex was randomly determined at the end of the process.

*BL avoiding full-sib (BL-NFS) or full- and half-sib (BL-NHS) matings:* As suggested by Ballou and Lacy (1995) one way of improving their method could be to avoid the mating between close relatives. We implemented this modification as follows. Once parents were sorted by sex on their mean coancestry, we took the first male and the first female, as before. If they were full-sibs (*BL-NFS*) or full- or half-sibs (*BL-NHS*) we kept the male but took the second female. If the new couple did not fit the restriction either, we took the third female, and so on. When an appropriate couple was found the process continued as in the *BL* method. With low census number, it was sometimes impossible to find a couple not being at least half-sibs. Thus, for the *BL-NHS* method the restriction had to be relaxed in that particular round, and couples were chosen just avoiding full-sib mating.

*Group coancestry (GC):* For this method we determined the individual contributions from parents by finding the combination which minimised the group coancestry of the offspring. We included a restriction to assure that male contributions equal female contributions. The optimisation was performed using the algorithm called ‘*simulated annealing*’ (Press et al. 1989; further details on the implementation can be found in Fernández and Toro 1999). At the end of the process we obtained the number of offspring every parent should contribute to the next generation. Three different mating schemes were associated with this selection method.

- *Random (r):* We randomly chose a male and a female parent from those determined to effectively contribute to the next generation. Offspring were generated until one of them survived. New couples were chosen until the desired number of offspring was reached, in such a way that each parent participated in a number of matings equal to its optimum contribution previously decided.
- *Compensatory (c):* The average coancestry of every individual with all the others was calculated and the parents of each sex were sorted out according to this value. Every parent was repeated in this list as indicated by its optimal contribution. Thus, two vectors with  $N$  positions (the population size) were obtained. Then we mated the male with the lowest mean coancestry (first position in the males’ vector) with the female with the highest mean coancestry (last position in the females’ vector), and so on. For every couple offspring

were generated and evaluated until a descendant survived.

- *Minimum coancestry (mc):* We looked for the combination of matings that yielded the lowest average pairwise coancestry between couples. To do so we constructed two vectors (one for each sex) where every parent appeared as many times as its decided contributions. Then, positions of vectors were matched in such a way that generated the lowest average pairwise coancestry between couples. This minimisation was performed through the ‘*Hungarian algorithm*’ (Dantzig 1963), an exact heuristic procedure to solve assignment problems. Offspring were generated and tested for viability until all couples contributed one offspring.

In the three mating schemes above sex of offspring was assigned at random once all descendants have been obtained.

## Results

Figure 1 shows the evolution of population fitness, scaled relative to that in generation 0, for mutational Models I and II and the three population sizes. Results are shown for random selection with random mating (*R*, line with squares), minimum group coancestry with minimum coancestry mating (*GC/mc*, thick line), the method proposed by Ballou and Lacy (*BL*, solid thin line) and the modified *BL* methods (*BL-NFS*, dotted line; *BL-NHS*, dashed line). Other systems of mating (compensatory or random mating) in combination with group coancestry had a very similar behaviour on the fitness performance to that of *GC/mc*, and are not shown in the figure. This can be seen in Table 1, where the average fitness at generation 20 for the three mating methods is shown.

For mutational Model I, the mean population fitness largely declined with time, as would be expected. The rate at which this occurred was greater as the population size diminished, reflecting two different effects. On the one hand, the higher levels of inbreeding reached with low census number increased inbreeding depression. On the other, the intensity of selection was lowered with low census numbers allowing more deleterious mutations to accumulate in the genome. In general, the method of minimum group coancestry (*GC*) contributions produced the highest fitness. The Random method (*R*) yielded the lowest fitness with  $N = 8$ , but the difference with *GC* dimi-

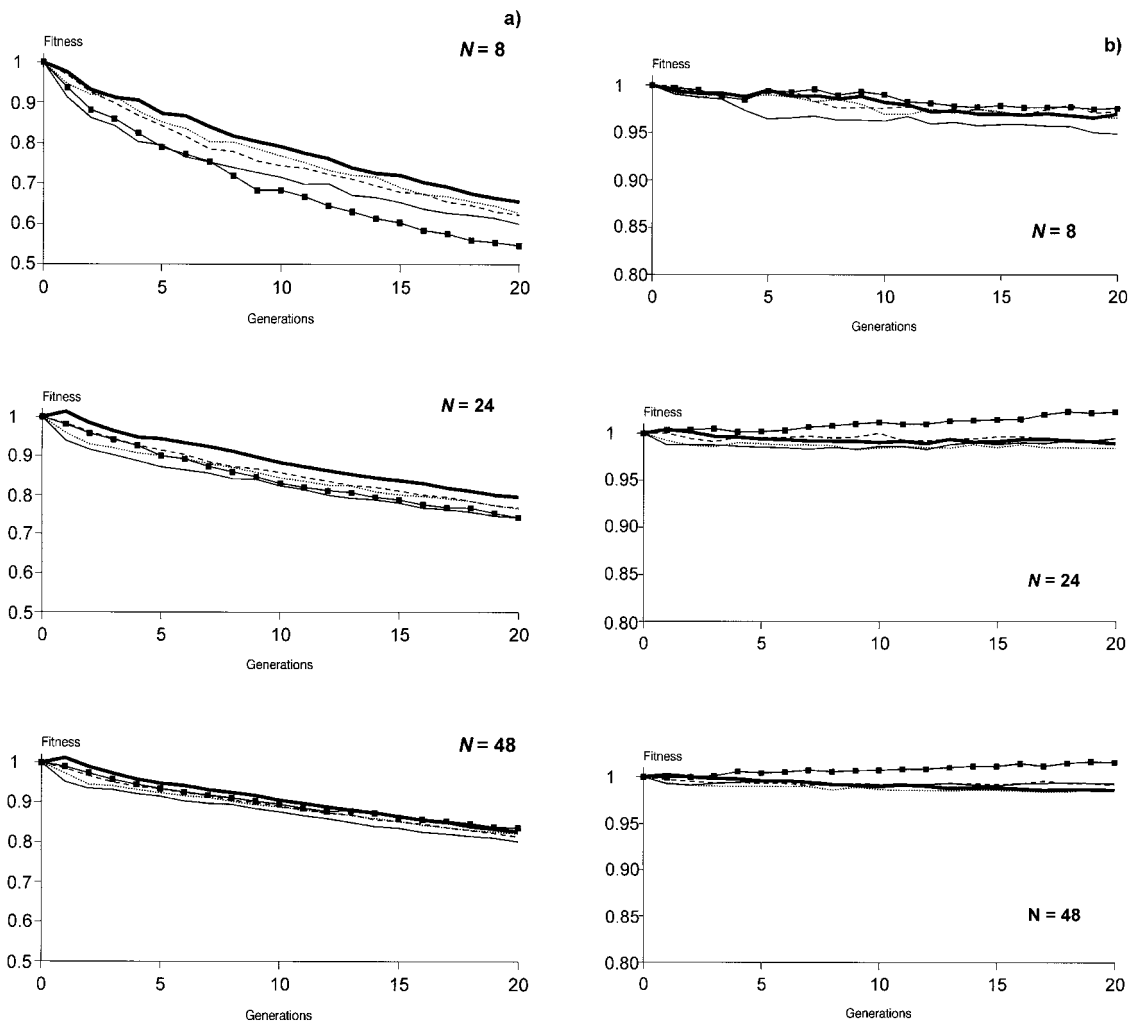


Figure 1. Average population fitness (scaled to that in generation 0), plotted against generations. The thick line corresponds to *GC/mc* method, the thin lines to *BL* methods (solid: *BL*, dotted: *BL-NFS*, dashed: *BL-NHS*) and the line with squares to the *R* method. a) Mutational Model I; b) Mutational Model II. *N*: population size.

nished as the population size increased and the effects of inbreeding depression were lowered.

The *BL* method produced a large drop in fitness in the early generations, even below *R* method levels. This inferiority held along the 20 generations for  $N = 48$  and disappeared with smaller census sizes after a number of generations. The explanation of this result can be clearly seen in Figure 2, where the average inbreeding of the population (calculated using neutral markers) is shown for the five strategies. During the first generations the *BL* method generated fast inbreeding, above the levels of the *R* method. These high levels of inbreeding in the early generations were responsible for a drop in fitness because

of inbreeding depression. For small population sizes, this initial disadvantage was recovered after a few generations. For larger population sizes the recovery took more time. Nevertheless, population fitness under this procedure was always lower than that using the *GC* method (see Figure 1a).

When mating between close relatives (full- or full- and half-sibs) was avoided, the fitness performance of the *BL* method improved (Figure 1a). Declines in the early generations were smaller due to lower levels of inbreeding generated by these procedures (Figure 2). However, the population fitness was still lower than that achieved by the *GC* strategy. A surprising result was that avoidance of full- and half-sib matings (*BL*-

Table 1. Average population fitness, at generation 20, as a percentage of that in generation 0

		<i>GC/r</i>	<i>GC/c</i>	<i>GC/mc</i>
Model I	<i>N</i> = 8	63	64	66
	<i>N</i> = 24	76	77	79
	<i>N</i> = 48	82	82	83
Model II	<i>N</i> = 8	97	98	97
	<i>N</i> = 24	99	99	99
	<i>N</i> = 48	99	99	99

*N* = population size

*GC* = group coancestry, *r* = random mating, *c* = compensatory mating, *mc* = minimum coancestry mating  
Standard errors range from 0.4 to 2.1

*NHS*) was not very different from the avoidance of only full-sib matings (*BL-NFS*). In fact, for *N* = 8, the latter outperformed the first. The reason is that being more strict in the election of couples implies choosing individuals with higher mean coancestry (at lower positions in the ranked list of individuals) and, eventually, higher inbreeding.

Table 2 shows the number and mean effect of deleterious alleles fixed or segregating in the population and the average frequency of the latter. The *R* method implied a higher genetic drift and, therefore, inbreeding depression. Thus, the number of deleterious alleles fixed was the largest for this method, and the number of segregating alleles was the lowest, with alleles at the highest average frequency. As population size became larger the relative effect of drift and inbreeding depression was lowered relative to the effect of selection in purging deleterious genes, and the *R* method performed substantially better in comparison with the management methods. Thus, for small *N* the mean effect of segregating alleles was much larger for the *R* method than for the *BL* and *GC* methods, but as *N* was increased this effect was reversed. Differences between *BL* and *GC* methods were not large with respect to mutations segregating or fixed after 20 generations.

The overall picture was quite different for mutational Model II. In general, the population fitness kept almost at the same level as in the ancestral population for all strategies and, in some cases, even reached higher values (Figure 1b). Thus, differences between methods were small (note the different scale between the graphs for Models I and II in Figure 1). As Model II implied few mutations of large

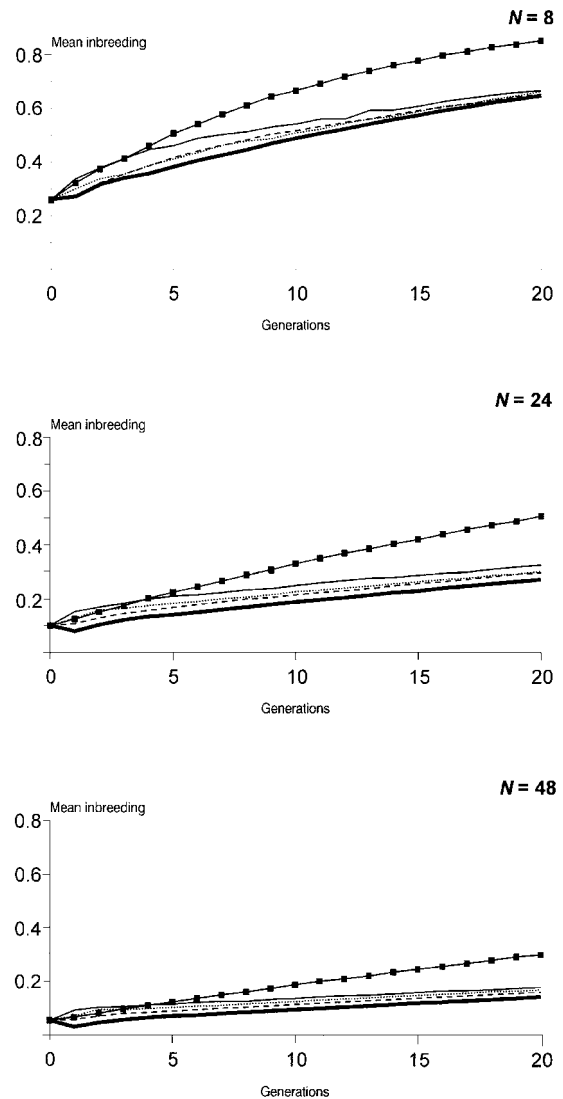


Figure 2. Average inbreeding of the population, plotted against generations, for mutational Model I. Symbols as explained in Figure 1.

mean effect appearing each generation, selection was strong enough to purge such mutations, whatever the management strategy applied.

The observed pattern in this case was that the *R* method had the highest fitness for all population sizes considered (Figure 1b). The reason can be seen in Table 2. For this mutational model a period of 20 generations allowed few or no deleterious mutations to get fixed for any method, even with the lowest census size. In this situation the *R* method became superior by keeping a lower number of segregating mutations.

Table 2. Average number and effects of deleterious alleles fixed or segregating in the population and the mean frequency of these latter at generation 20

		Model I				Model II			
		<i>R</i>	<i>BL</i>	<i>BL-NHS</i>	<i>GC/mc</i>	<i>R</i>	<i>BL</i>	<i>BL-NHS</i>	<i>GC/mc</i>
<i>N</i> = 8	Fixed	118.78	63.48	68.18	67.83	0.49	0.13	0.22	0.21
	Mean effect	0.0095	0.0062	0.0065	0.0064	0.0470	0.0156	0.0196	0.0261
	Segregating	111.84	265.28	251.14	250.49	3.22	6.85	6.77	6.79
	Mean effect	0.0232	0.0186	0.0195	0.0200	0.3151	0.2810	0.3322	0.3371
	Frequency	0.4318	0.4025	0.4065	0.4057	0.2669	0.2376	0.2127	0.2207
<i>N</i> = 24	Fixed	32.43	11.47	11.71	11.61	0.02	0.00	0.00	0.00
	Mean effect	0.0028	0.0011	0.0010	0.0011	0.0005	0.0000	0.0000	0.0000
	Segregating	405.97	700.95	691.56	689.11	8.39	17.94	18.34	18.34
	Mean effect	0.0160	0.0162	0.0166	0.0172	0.3549	0.3252	0.3475	0.3475
	Frequency	0.3319	0.2274	0.2294	0.2308	0.1402	0.0938	0.0910	0.0910
<i>N</i> = 48	Fixed	11.51	4.39	4.73	4.71	0.00	0.00	0.00	0.00
	Mean effect	0.0010	0.0005	0.0006	0.0007	0.0000	0.0000	0.0000	0.0000
	Segregating	690.26	1146.74	1140.94	1136.43	16.08	32.76	32.73	34.04
	Mean effect	0.0148	0.0162	0.0167	0.0171	0.3117	0.3293	0.3517	0.3595
	Frequency	0.2240	0.1448	0.1459	0.1463	0.0802	0.0513	0.0531	0.0550

*N* = population size

*R* = random, *BL* = method proposed by Ballou and Lacy, *GC* = group coancestry, *mc* = minimum coancestry mating, *BL-NHS* = *BL* method avoiding full- and half-sib matings

Standard error's ranges:

Model I – Fixed (0.16 – 1.21), Mean effect (< 0.0001 – 0.0002), Segregating (2.18 – 2.73), Mean effect (0.0001 – 0.0005) and Frequency (0.0003 – 0.0028)

Model II – Fixed (0.01 – 0.06), Mean effect (0.0004 – 0.0071), Segregating (0.18 – 0.46), Mean effect (0.0080 – 0.0180) and Frequency (0.0010 – 0.0140)

Table 3 shows the amount of genetic variation maintained at generation 20 by the different methods measured as the expected heterozygosity and the allelic diversity for neutral loci. Results for both mutational models were identical and, therefore, data are shown for only one of them. Expected heterozygosity was greater as the population size increased, as predicted from population genetics theory, but the behaviour was the contrary for allelic diversity, as this measure is made relative to the total number of alleles present at generation 0. For larger *N* the number of alleles retained would be greater but, as a proportion of the original ones, the allelic diversity would be reduced. As expected, the method that yielded the lowest levels for both measures was the *R* method, for which genetic drift was more important making more alleles to be lost. The other methods performed very similarly. The *BL* method produced a slightly higher benefit, but the *BL-NHS* method was very close to the *GC* method.

## Discussion

The main goal for any conservation plan has been established to keep the largest amounts of genetic information, whether we refer to farm species (Oldenbroek 1999) or wild animals and plants (Ballou and Lacy 1995 and references therein). However, a successful conservation programme should assure also the survival of the population and, therefore, it should also account for the effect of the implemented strategies on the population fitness.

Several authors have studied theoretically and experimentally the best strategies to manage conserved populations in order to retain the highest possible genetic information. Most of them conclude that the best way to control the loss of genetic diversity is minimising the group coancestry or average mean coancestry, both in the field of conservation (Lacy 1994; Ballou and Lacy 1995; Montgomery et al. 1997; Caballero and Toro 2000) or in the field of animal

Table 3. Genetic diversity maintained in the population after 20 generations, measured as the expected heterozygosity (%) and the allelic diversity averaged over the 200 neutral loci

		<i>R</i>	<i>BL</i>	<i>BL-NHS</i>	<i>GC/mc</i>	<i>GC/r</i>	<i>GC/c</i>
<i>N</i> = 8	Exp. Heterozygosity	13.0	34.1	31.5	30.8	32.8	32.2
	Allelic Diversity	8.5	12.8	12.3	12.2	12.6	12.4
<i>N</i> = 24	Exp. Heterozygosity	46.9	69.7	69.0	68.7	68.9	69.3
	Allelic Diversity	6.0	11.0	10.8	10.7	10.8	10.9
<i>N</i> = 48	Exp. Heterozygosity	67.8	83.4	83.1	83.0	83.0	82.9
	Allelic Diversity	5.5	10.5	10.4	10.2	10.3	10.3

*N* = population size

Abbreviations for management methods as in Tables 1 and 2. All results for mutational Model I. Standard errors range from 0.05 to 0.5 for expected heterozygosity and from 0.02 to 0.04 for allelic diversity

breeding (Wray and Goddard 1994; Brisbane and Gibson 1995; Meuwissen 1997; Fernández and Toro 1999). Minimising group coancestry outperforms other common strategies such as the equalisation of individual contributions, the equalisation of founder contributions (Alderson 1991) or the breeding priority to animals giving high probability of carrying unique alleles (Ballou and Lacy 1995). This criterion can be easily implemented in a population to decide the contributions of each parent to the next generation, irrespective of the mating system. However, it can also be implemented in such a way that selection and mating scheme are arranged in just one step, as proposed by Ballou and Lacy (1995).

When *GC* is implemented as a separate stage from the mating scheme, the latter seems to be not a critical point, as similar levels of genetic diversity and population fitness are found for different mating schemes in combination with minimum group coancestry contributions (Table 1). Only the mating scheme based on minimising the global pairwise coancestry between couples (*mc*) appears to give a little advantage in the short-term fitness (not shown), associated with lower average inbreeding levels in the first generations. Compensatory mating does not represent a disadvantage to the maintenance of genetic diversity, as was suggested by Ballou and Lacy (1995).

The mean coancestry method proposed by Ballou and Lacy gives a slightly higher level of genetic diversity (Table 3). However, this strategy makes population fitness to be lower than under other management procedures, particularly in the early generations. The cause seems to be the higher increase of inbreeding produced by the method, and the corresponding inbreeding depression (Figures 1 and 2).

Although in later generations the rate of inbreeding for the *BL* method equals those for other strategies, the early effects sometimes made the fitness to be permanently lower. This effect is produced by a tendency of the strategy to mate close related individuals, creating a kind of ‘*inbred lines*’. It is widely known that a simple way to generate the highest heterozygosity level is crossing lines totally inbred for different alleles. This kind of population would have the greatest expected heterozygosity, but it would also suffer the deleterious effects of a large inbreeding depression.

The above explanation for the poor performance of the *BL* method is supported by the following observations. First, if we look at the deviation from Hardy-Weinberg expectations (Wright’s  $F_{IS}$ ), we find higher values for *BL* than for *GC/mc*, indicating a higher proportion of inbred matings in the former. For example, under mutational Model I and  $N = 24$ , the average  $F_{IS}$  for generations 10 to 20 was 0.0293 for *BL* and  $-0.0590$  for *GC/mc*, respectively. This indicates a defect of heterozygotes for the *BL* method and an excess for the *GC* method. The results are similar for other combinations of parameters. Second, as pointed out in the results section, the *BL-NFS* and *BL-NHS* strategies outperform the *BL* method, not suffering from such a large decline in fitness in the early generations. The only difference between the methods is the avoidance of matings between close relatives in the former. Therefore, this should be the reason for the bad performance of the *BL* scheme. In fact, the individuals and contributions chosen by the *BL* and *GC/mc* methods are essentially the same and, usually, they only differ in the way they are mated (data not shown). And finally, to test whether

the explanation was or not correct, we carried out simulations where the contributions to the next generation were arranged to produce the minimum group coancestry (*GC*) but, then, parents were mated as to yield the maximum (instead of minimum) average pairwise coancestry between couples. Although this method provided the highest level of expected heterozygosity along the 20 generations simulated, it also yielded the lowest fitness because of the large average inbreeding generated, specially in the early generations. For example, for Model I and  $N = 24$ , the expected heterozygosity at generation 20 was 2% larger than that obtained with the *BL* method, while population fitness was 8% lower.

The problem of the generation of inbreeding by the *BL* method can be solved by avoiding mating between close relatives (e.g. *BL-NFS* and *BL-NHS*). However, as we mentioned before, being more strict in this respect implies choosing individuals with higher mean coancestry. Thus, *BL-NHS* performed worse, on some occasions, than *BL-NFS* (Figure 1a). In our simulation we avoided mating between sibs for the *BL* method by choosing the first male in the ranked list and exchanging the female. An *a priori* more comprehensive algorithm to avoid mating between relatives in the *BL* method was also tested. The first male and female of the ranked list were chosen. If these were sibs, the first male and the second female were chosen. If these were again sibs, the first female and the second male were chosen. Then, the first male and the third female, the first female and the third male, and so on. If no couples including the first male or female fitted the restriction the process went on with all possible couples including the second male or female, then including the third male or female and so on, until the restriction was fitted. The performance (data not shown) was not better, and even worse than with the previous *BL-NFS* or *BL-NHS*. The reason was, again, the use of individuals with higher mean coancestry because of the increased restriction in the allowed matings. The *GC/mc* method does not have this problem, as the mating scheme is decided after parents' contributions and, therefore, mating does not reduce the average coancestry of individuals.

An additional disadvantage of avoiding mating between close relatives with the *BL* method is that the restrictions imposed could force most of the offspring to be generated by a limited number of parents, specially for small census sizes. This may produce a bottleneck and, therefore, an increase in inbreeding, coancestry and loss of diversity.

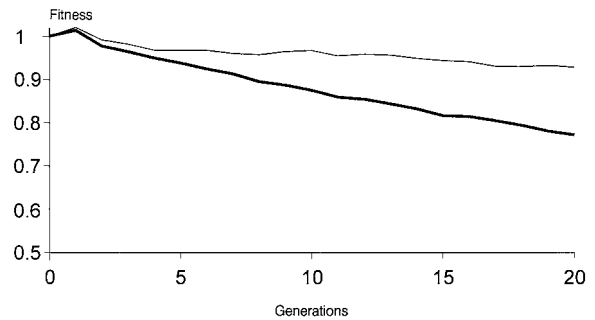


Figure 3. Average population fitness (scaled to that in generation 0), plotted against generations, for *GC/mc* (thick line) and *GC/mc* with restrictions on parents fitness (thin line).  $N = 24$  and mutational Model I are assumed.

If an estimate of the fitness of individuals is available the minimum group coancestry can be carried out taking account of this information. The way of implementing this in a conservation programme would be similar to that used in selection programmes, where contributions are arranged to yield the lower group coancestry assuring a minimum level of response to selection (see, for example, Fernández and Toro 1999). We carried out some simulations to determine the performance of this strategy applied in the context of conservation. For simplicity, we assumed the most favourable situation where we know the genotypic value for fitness of each individual. The objective was to maintain, as far as possible, the initial mean fitness of the population. The procedure was similar to the *GC/mc* method, but the feasible combinations of contributions in every generation were restricted to those having weighted mean fitness of selected individuals equal or greater than the mean population fitness at generation 0. If we assume that fitness is additive, the expected fitness level of the population would not decrease over generations. However, sometimes no combination fulfilled the restriction, so this had to be relaxed and the closer solution was used. An example of these simulations is shown in Figure 3 in comparison with the *GC/mc* method used without restrictions.

The population mean fitness for the restricted method was well above the levels of fitness reached implementing *GC/mc* without restrictions. The effect of restriction on the genetic diversity was negligible, with expected heterozygosities of 67% and 69% at generation 20 for the restricted and the unrestricted method, respectively. Therefore, this approach could be recommended in conservation programmes, when possible, to improve the health of the populations.

However, the method can also lead to adaptation to captive conditions and to loss of adaptive variation useful in its future release to the wild (Lacy 2000). Thus, its implementation should be made carefully, without forgetting the main objective of preserving genetic diversity as high as possible.

Some authors have argued that, as a side-effect of conservation management procedures, selection pressure would be reduced and a greater number of deleterious mutations could get fixed in the population. The long-term fitness impairment, and eventually the extinction, of a population maintained with low census number due to the accumulation of deleterious mutations has been extensively studied for unmanaged populations (Lande 1995; Lynch et al. 1995). Schoen et al. (1998) studied the effect of equalising contributions in the management of a seed collection (germplasm bank). They found a much higher loss of fitness in populations under equalisation of parental contributions than in unmanaged populations. However, this study was restricted to a fecundity model which did not allow for any natural selection when contributions were equalised. As pointed out by Couvet and Ronfort (1994) and Wang et al. (1999), viability selection within the offspring of a family or an individual always takes place. Fernández and Caballero (2001) carried out simulations accounting for this fact, as well as for other factors, such as different reproductive rates for the species, and relaxation of selection in captive conditions. The results indicate that the threat on the survival of populations due to accumulation of deleterious mutations in populations with low census number is not specially greater with equalisation of contributions than in unmanaged populations, at least over the early generations (up to generation 20 or so). The results presented in the present paper completely agree with this conclusion, extending them to other management procedures.

All results presented in the paper were obtained using measures of relatedness calculated from pedigrees. However, if neutral loci information is available, coancestries based on such kind of data could be used as the decision criterion. Recently, Toro et al. (1999) and Wang and Hill (2000) have proposed methods to join information from pedigree and from molecular markers in order to get higher effective population sizes and, thus, keeping a larger amount of genetic diversity. Conservation programmes would benefit from the use of such techniques.

## Acknowledgements

We thank comments on the manuscript from two anonymous referees and R. Frankham. This work was supported by grant BOS2000-0896 (Plan Nacional de Investigación Científica, Desarrollo e Innovación Tecnológica, I+D+I).

## References

- Alderson GHL (1991) A system to maximize the maintenance of genetic variability in small populations. In: *Conservation of Domestic Livestock* (eds. Alderson L, Bodo L), pp. 18–19. CAB International, Wallingford, UK.
- Allaire FR (1990) Mate selection by selection index theory. *Theor. Appl. Genet.*, **57**, 267–272.
- Ballou JD, Lacy RC (1995) Identifying genetically important individuals for management of genetic variation in pedigreed populations. In: *Population management for survival and recovery* (eds. Ballou JD, Gilpin M, Foote TJ), pp. 76–111. Columbia University Press, New York.
- Ballou JD, Foote TJ (1995) Demographic and genetic management of captive populations. In: *Wild Animals in Captivity* (eds. Kleinman DG, Lumpkin S, Allen M, Harris H, Thompson K), pp. 263–283. University of Chicago Press, Chicago, U.S.A.
- Bataillon TM (2000) Estimation of spontaneous genome-wide mutation rate parameters: wither beneficial mutations? *Heredity*, **84**, 497–501.
- Brisbane JR, Gibson JP (1995) Balancing selection response and rate of inbreeding by including relationships in selection decisions. *Theor. Appl. Genet.*, **91**, 421–431.
- Caballero A, Keightley PD (1994) A pleiotropic non-additive model of variation in quantitative traits. *Genetics*, **138**, 883–900.
- Caballero A, Santiago E, Toro MA (1996) Systems of mating to reduce inbreeding in selected populations. *Anim. Sci.*, **62**, 431–442.
- Caballero A, Toro MA (2000) Interrelations between effective population size and other pedigree tools for the management of conserved populations. *Genet. Res.*, **75**, 331–343.
- Couvet D, Ronfort J (1994) Mutation load depending on variance in reproductive success and mating system. In: *Conservation Genetics* (eds. Loescheke V, Tomiuk J, Jain SK), pp. 55–68. Birkhauser, Basel.
- Crow JF, Kimura M (1970) *An Introduction to Population Genetics Theory*. Harper and Row, New York.
- Crow JF, Simmons MJ (1983) The mutation load in *Drosophila*. In: *The Genetics and Biology of Drosophila Vol. 3c* (eds. Ashburner M, Carson HL, Thomson JN), pp. 1–35. Academic Press, London.
- Dantzig GB (1963) *Linear Programming and Extensions*. Princeton University Press, Princeton.
- Fernández J, Caballero A (2001) Accumulation of deleterious mutations and equalisation of parental contributions in the conservation of genetic resources. *Heredity*, 86 (in press).
- Fernández J, Toro MA (1999) The use of mathematical programming to control inbreeding in selection schemes. *J. Anim. Breed. Genet.*, **116**, 447–466.
- García-Dorado A, Caballero A (2000) On the average coefficient of dominance of deleterious spontaneous mutations. *Genetics*, **155**, 1991–2001.

- García-Dorado A, López-Fanjul C, Caballero A (1999) Properties of spontaneous mutations affecting quantitative traits. *Genet. Res.*, **74**, 341–350.
- Gowe RS, Robertson A, Latter BDH (1959) Environment and poultry breeding problems. 5. The design of poultry control strains. *Poult. Sci.*, **38**, 462–471.
- Hedrick PW, Miller PS (1992) Conservation genetics: Techniques and fundamentals. *Ecol. Appl.*, **2**, 30–46.
- Keightley PD, Eyre-Walker A (1999) Terumi Mukai and the riddle of deleterious mutation rates. *Genetics*, **153**, 515–523.
- Klieve HM, Kinghorn BP, Barwick SA (1994) The joint regulation of genetic gain and inbreeding under mate selection. *J. Anim. Breed. Genet.*, **111**, 81–88.
- Lacy RC (1994) Managing genetic diversity in captive populations of animals. In: *Restoration and Recovery of Endangered Plants and Animals* (eds. Bowles ML, Whelan CJ), pp. 63–89. Cambridge University Press, Cambridge, UK.
- Lacy RC (2000) Should we select genetic alleles in our conservation breeding programs? *Zoo Biol.*, **19**, 279–282.
- Lande R (1995) Mutation and conservation. *Conserv. Biol.*, **9**, 782–791.
- Lynch M (1996) A quantitative-genetic perspective on conservation issues. In: *Conservation Genetics: Case Histories from Nature* (eds. Avise J, Hamrick J), pp. 471–501. Chapman and Hall, New York, U.S.A.
- Lynch M, Blanchard J, Houle D, Kibota T, Schultz S, Vassilieva L, Willis J (1999) Perspective: Spontaneous deleterious mutation. *Evolution*, **53**, 645–663.
- Lynch M, Conery J, Bürger R (1995) Mutation accumulation and the extinction of small populations. *Am. Nat.*, **146**, 489–518.
- Meuwissen THE (1997) Maximizing the response of selection with a predefined rate of inbreeding. *J. Anim. Sci.*, **75**, 934–940.
- Montgomery M, Ballou JD, Nurthen RK, England P, Briscoe D, Frankham R (1997) Minimizing kinship in captive breeding programs. *Zoo Biol.*, **16**, 377–389.
- Oldenbroek JK (1999) *Genebanks and the Conservation of Farm Animal Genetic Resources*. DLO Institute for Animal Science and Health, Lelystad, The Netherlands.
- Press WH, Flannery BP, Teukolsky SA, Vetterling WT (1989) *Numerical Recipes*. Cambridge University Press, Cambridge, UK.
- Schoen DJ, David JL, Bataillon TM (1998) Deleterious mutation accumulation and the regeneration of genetic resources. *Proc. Natl. Acad. Sci. U.S.A.*, **95**, 394–399.
- Schultz ST, Lynch M (1997) Mutation and extinction: The role of variable mutational effects, synergistic epistasis, beneficial mutations, and degree of outcrossing. *Evolution*, **51**, 1363–1371.
- Sonesson A, Meuwissen THE (2000) Mating schemes for optimum contribution selection with constrained rates of inbreeding. *Genet. Sel. Evol.*, **32**, 231–248.
- Toro MA, Nieto B, Salgado C (1988) A note on minimization of inbreeding in small-scale selection programmes. *Liv. Prod. Sci.*, **20**, 3171–323.
- Toro MA, Pérez-Enciso M (1990) Optimization of selection response under restricted inbreeding. *Genet. Sel. Evol.*, **22**, 93–107.
- Toro MA, Silió L, Rodríguez MC, Rodríguez J, Fernández J (1999) Optimal use of genetic markers in conservation programmes. *Genet. Sel. Evol.*, **31**, 255–261.
- Wang J (1997) More efficient breeding systems for controlling inbreeding and effective size in animal populations. *Heredity*, **79**, 591–599.
- Wang J, Hill WG (2000) Marker-assisted selection to increase effective population size by reducing Mendelian segregation variance. *Genetics*, **154**, 475–489.
- Wang J, Hill WG, Charlesworth D, Charlesworth B (1999) Dynamics of inbreeding depression due to deleterious mutations in small populations: Mutation parameters and inbreeding rate. *Genet. Res.*, **74**, 165–178.
- Wray NR, Goddard ME (1994) Increasing long-term response to selection. *Genet. Sel. Evol.*, **26**, 431–451.
- Wright S (1921) Systems of mating. *Genetics*, **6**, 111–178.

