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The influence of natural selection in breeding programs: A simulation study

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ABSTRACT

Natural selection, acting directly on fitness or through stabilizing selection for other traits, has always been present in natural populations. In principle, this force will reduce the genetic variability, which is required for artificial selection programs. The genetic improvement of a trait with economic interest by selection programs depends on the amount of genetic diversity for that trait in a particular population. Most studies on the development of breeding programs account only for the genetic variation for the target trait itself. The objective in this work is determining, through computer simulations, the consequences for the evolution of selection programs, considering not only the variability for the trait of interest but also for fitness under different mutational models. Additionally, a scenario where the trait was subjected to stabilizing selection was also studied. Different parameters as effective population size, phenotypic mean, phenotypic variance and heterozygosity were used to monitor the performance in the different scenarios. In conclusion, considering the action of direct natural selection does not lead to lower levels of genetic variability for neutral traits, and thus it does not reduce the ability of populations to respond to artificial selection regardless of the mutational model used. On the other hand, stabilizing selection penalizes individuals with an extreme phenotype (which is the major objective in the artificial selection), reducing their fitness. Therefore, the artificial selection was ineffective in improving traits subjected to stabilizing selection.

1. Introduction

The increment (or modification) of the phenotypic mean for traits of interest is the main objective in genetic selection programs. The magnitude of the response to artificial selection is proportional to the amount of genetic variability for that trait in the population. Therefore, the genetic diversity present at the beginning of the process will be crucial, because it determines the ability to evolve or to be genetically improved. If we only consider the neutral variability, its level depends on the historical effective population size (N_e), being the genetic drift the process determining the rate of loss of genetic variability.

The genetic variability could be described as the existence of different alleles and genotypes and the first action before applying a selection program to a population should be to estimate its initial genetic diversity, and to predict the changes in it for the planned selection intensity. In general, the most frequent measures of genetic diversity based in locus are the observed heterozygosity (OH; proportion of heterozygous individuals), expected heterozygosity (EH; the heterozygosity in a population in Hardy-Weinberg equilibrium with the same allelic frequencies) and the allelic diversity (AD; number of different alleles for a locus). For quantitative traits, diversity is usually measured through the additive genetic variance (V_A) and the heritability (h^2) defined as the proportion of phenotypic variance explained by the additive genetic effects (Falconer and Mackay, 1996).

However, the fate of any population, whatever subjected to a selection program or not, also depends on the fitness of the individuals belonging to it (Reed and Frankham, 2003). Fitness is the capacity of survival, adaptation and reproduction of populations (Roff and Mousseau, 1987). Natural selection directly acts on fitness of population, shaping its genetic diversity in an equilibrium with the input occurring due to mutation. Natural selection may also interacts with the genetic diversity for quantitative (productive) traits through two mechanisms: i) natural selection reduces the census population size (N) leading indirectly to the loss of genetic diversity for any trait; ii) it has a direct effect due to the genetic correlation between fitness and the traits.

A particular way of interaction between fitness and productive traits is the so called stabilizing selection. Stabilizing selection favors

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individuals with phenotypes close to an optimum value (i.e. they have higher fitness) and penalizes individuals far from it (Kingsolver et al., 2001; García-Dorado et al., 2007). Studies as Johnson and Barton (2005) have proven that stabilizing selection leads to the reduction of genetic diversity for the trait.

It is also well known that artificial selection implies the reduction in genetic variability for the target trait but also for fitness due to the reduction in N_e. This may lead to the fixation of deleterious mutations threatening the survival of the population (Lynch et al., 1995).

The objective in this work was to determine, through stochastic simulation, how the combination of natural directional and stabilizing selection affects the response to selection for a trait of interest. The viability of implementing selection programs under different scenarios was evaluated.

2. Materials and methods

2.1. Model assumptions and simulations

Diploid individuals with 20 chromosomes of 1 Morgan each were simulated. The genome comprised two types of biallelic loci: selective marker loci with effect on fitness and neutral marker loci. Each chromosome carried 1000 and 3000 selective and neutral loci, respectively. The selective loci were randomly allocated within the chromosome. One hundred neutral loci were randomly selected to control a quantitative trait, which was the objective of the breeding program. No loci affected fitness and the quantitative trait at the same time and, therefore, in the basic scenario no genetic correlation existed between them, besides the one arising from linkage disequilibrium between loci affecting fitness and the trait, respectively.

Our simulations for the different scenarios can be divided in two steps: first, we generated base populations in mutation-drift or mutation-selection-drift equilibrium; second, we implemented classic artificial selection on the trait of interest.

We simulated 5000 discrete generations for a randomly mating population of constant size ($N = 100$), where half the population were females and half males. Genotypes for the neutral loci were generated in the initial population by randomly choosing alleles between two possibilities (i.e. initial frequencies were 0.5). Selective loci were all fixed for the 'wild' type allele. We verified that the mutation-drift or mutation-drift-selection equilibrium has been achieved, by detecting the stabilization of the allelic frequencies and the expected heterozygosity. The rationale behind this first step was getting a sensible distribution of allelic frequencies (not a high proportion of fixed loci) and a reasonable linkage disequilibrium between all types of loci.

Every generation, gametes were created allowing for recombination between the pairs of homologous. Accordingly with the simulated chromosome length, the actual number of recombinations per chromosome was obtained from a Poisson distribution with $\lambda = 1$ and randomly allocated along the chromosome.

The neutral loci mutation rate was 2.5×10^{-3} by locus and it could occur in both alleles, switching allele 0 to allele 1 and vice versa. Lower mutation rates would imply lower levels of heterozygosity at the equilibrium and few segregating loci. This could be compensated by enlarging the population size and the number of generations in the first step. But general results would not change substantially (Gómez-Romano et al., 2013).

2.2. Fitness mutational models

For selective loci mutations, we consider two contrasting models regarding the mutation rate and the distribution of the effects of mutations:

- Mukai et al. (1972), numerous mutations with little effect occur in populations. They are difficult to be eliminated by natural selection, as only individuals with many mutations do not survive. Mutation rate used was $\mu = 0.5$ per haploid genome and generation.
- CGD (García-Dorado and Caballero, 2000) the mutations are few but with great effect on the individual fitness. They are easier to remove, because carriers die and do not transmit the deleterious mutations (natural selection is more effective in the removal). Mutation rate was $\mu = 0.03$ per haploid genome and generation (see selection coefficients below).

The number of selective loci mutations per individual were also obtained from a Poisson distribution with mean equal to twice the haploid mutation rate (i.e. 1 and 0.06 for Mukai and CGD models, respectively), and randomly allocated along the whole genome. The fitness of an individual was calculated as the product of the values for each selective *locus* (Lynch and O'Hely, 2001), being 1 for 'wild' type homozygotes, (1 - s) for 'mutant' type homozygotes and (1 - hs) for heterozygotes. The initial fitness was 1 (without deleterious 'mutant' type alleles) and consequently, the more mutations accumulated in selective loci the lower the individual fitness. Backward mutation was not allowed, thus, only changes from 'wild' type to deleterious 'mutant' type alleles were allowed.

The mean effect s and the mean dominance coefficient h of mutations were $s = 0.05$ and $h = 0.36$ under Mukai's model and $s = 0.26$ and $h = 0.20$ for CGD. The effect of each selective locus was sampled from a gamma distribution (with parameters α and β, being β the shape parameter). The values for each of the models were $\beta = 1$ and $\alpha = 0.05$ for Mukai and $\beta = 2.3$ and $\alpha = 0.61$ for CGD model. The dominance coefficient of mutations was obtained from a uniform distribution between 0 and exp(-ks), where k is a constant allowing the mean dominance coefficient to be the desired one. Actual values used were $k = 991$ and $k = 26$ for Mukai and CGD, respectively. The Mukai model parameters corresponds to an exponential distribution of gene effects that has been proposed as a likely distribution (see, e.g. Schultz and Lynch, 1997). The CGD model parameters have been obtained through minimum distance estimation of Drosophila data (García-Dorado et al., 1999). The particular combinations of alpha and beta used to define both models are the same as found in de Cara et al. (2013).

Offspring for the next generation were created from random chosen males and females. To decide if a newborn individual survived, we drew a random number to simulate environmental stochasticity from a uniform distribution between 0 and 1 and compared with the fitness for that individual (see the calculations of fitness above). If the fitness was less than this random value, the individual died and a new offspring was generated from new randomly chosen parents. This process was repeated until 100 offspring survived. In this study fitness was expressed as differences in viability, with no effect on fecundity or fertility. In general, larger number of matings (attempts) were needed to maintain the population size as the generations passed.

2.3. Quantitative trait

Genetic value (G_i) for the target trait was the summation of the individual values of each locus along the 100 neutral loci with effect on the trait (which were randomly chosen among the neutral loci), following the expression:

$$G_i = \sum_{j=1}^{j=n} a_{ij} \quad (1)$$

where a_{ij} is the genetic value of individual i for locus j. This implies the trait was additive across loci (i.e., no epistasis). The value of each locus was assigned through an allele coding system (Strandén and Christensen, 2011), being 1 for the homozygotes (11), 0 for the heterozygote, and −1 for the opposite homozygotes (00). The trait was also additive within locus. Therefore, the maximum genetic value for the trait was 100, when all loci were fixed for the allele 1, and the minimum −100, in the case of every loci homozygous for the allele 0.

The phenotype of individuals (P_i) was calculated with the following equation:

$$P_i = \mu + G_i + e_i \quad (2)$$

where μ is the mean population arbitrarily fixed in 100, and e_i the environmental deviation, obtained from a Gaussian distribution (0, 100).

2.4. Selection program

From the equilibrium population, 2000 discrete generations under a truncation artificial selection program based on phenotypic values were simulated (second step of this study). The 10 males and 20 females with the highest phenotypic value for the quantitative trait were chosen and mated randomly to create the offspring (100) for the next generation.

Natural directional selection was also accounted for in the artificial selection, step. Survival of the offspring was determined in the same way as in the equilibrium population step. Thus the number of the offspring per couple depends on the fitness of the parents, more individuals surviving from those with higher fitness among the selected based on their phenotype.

2.5. Stabilizing selection

In some scenarios, stabilizing selection on the trait of interest was also simulated during this second step of artificial selection, setting a relationship between the trait and fitness. When stabilizing selection was present, individuals with intermediate phenotype for the trait near an optimum value were favored as they showed a higher fitness (i.e. higher probability of survival, Haldane, 1935). Consequently, this kind of selection tends to decrease phenotypic and genetic variance. The stabilizing selection effect on the population will be higher the larger the phenotypic variance is. Optimum value in our simulations was set to the phenotypic mean of generation 5000 (mutation-selection-drift equilibrium), which was 100.

The fitness due to stabilizing selection was calculated from:

$$W_i = \exp\left(-\frac{(P_i - P_{opt})^2}{2\cdot\omega^2}\right) \quad (3)$$

(Turelli, 1984; Bürger et al., 1989) being W_i the fitness for individual i, P_i the phenotype of individual i for the trait, P_{opt} the optimum phenotype, and ω^2 is a parameter that depends on the selection pressure (reflected in the amplitude of the fitness distribution). The strength of stabilizing selection is usually expressed adding the environmental variance ${\sigma_E}^2$ (Kondrashov and Turelli, 1992) as $Vs = \omega^2 + {\sigma_E}^2$. If selection pressure is very strong (small values of ω^2), only individuals with phenotype close to the optimum value survive. As it relaxes (higher ω^2 values), there are more individuals with extreme phenotype that survive. In the present study, ω^2 took values of 25, 50, 75, 100 and 200 in order to cover the wide range of selection intensities that are described in the literature (García-Dorado and González, 1996; Wayne and Mackay, 1998; Johnson and Barton, 2005).

Total fitness in these scenarios was the product of both fitness: that determined by the genotypes for the selective loci and fitness due to stabilizing selection depending on the trait performance. As before, survival of an individual was decided by comparing its fitness with a random value sampled from a uniform distribution.

2.6. Scenarios

In the first simulated scenario -NF or no fitness- neutral variability and, consequently, variability for the quantitative trait initially depended only on drift and mutation. In the second step of the simulations diversity was also affected by artificial selection. All individuals were viable – equal fitness – and they had the same probability to contribute offspring to the next generation, except during the artificial selection period.

In the rest of scenarios, fitness was included with a genetic control. Two different situations were simulated:

- GF or general fitness. Fitness (determined by the selective loci) was not correlated with the quantitative trait except by the linkage disequilibrium (LD) between selective loci and the ones controlling the trait.
- SS or stabilizing selection scenario. Besides the effect of selective loci on the fitness of the individuals, the trait of interest was correlated with fitness with intermediate phenotypes being more fit than extremes.

2.7. Parameters for monitoring

The levels of total fitness were calculated in all generations for those scenarios including a genetic control of the fitness.

As a measure of genetic variability, the expected heterozygosity (EH) in generations 5000, 6000 and 7000 was calculated for a single locus following to formula:

$$EH = 1 - \sum_{i=1}^{2} p_i^2 \quad (4)$$

being p_i the frequency of allele i. The global value was the average of EH across all the neutral loci.

The heritability (h^2) of the productive trait was estimated in generation 5001, through the additive genetic variance (V_A) and phenotypic variance (V_P). The additive genetic variance was calculated from the allele frequencies on the 100 individuals for each 100 neutral loci with effect on the trait. In the second step of the simulations (in the artificial selection program) phenotypic mean and variance were calculated every generation using the values of all individuals.

Pedigree was recorded during the simulation and, thus, inbreeding coefficient (F) and its corresponding rate (ΔF) could be calculated for different periods. The effective population size (N_e) was calculated through two methodologies: i) a demographical estimate which calculate the expected N_e corresponding to the particular sex ratio in the selection program

$$\frac{1}{N_e} = \frac{1}{4N_m} + \frac{1}{4N_f} \quad (5)$$

being N_m the number of males and N_f the number of females which are selected; ii) from the individual rate of inbreeding (ΔF_i) following

$$N_e = \frac{1}{2\Delta F}. \quad (6)$$

(Falconer and Mackay, 1996).

The individual inbreeding (F_i) and the individual rate of inbreeding (Gutiérrez et al., 2008) were calculated using the software ENDOG v4.8 (Gutiérrez and Goyache, 2005) from pedigree data, following the expression:

$$\Delta F_i = 1 - \sqrt[g_i - 1]{1 - F_i} \quad (7)$$

where ΔF_i is the increment in inbreeding of animal i, F_i is its inbreeding coefficient and g_i is the mean equivalent number of generations above individual i. Only pedigree from generation 5000 and henceforth was accounted in the calculations of F, ΔF and N_e.

Every scenario was replicated 10 times and results are averages across replicates.

3. Results and discussion

Evolution of fitness during the second step (i.e. breeding program) for the different scenarios and generations can be found in Table 1. As

Table 1
Fitness mean along generations (G) under different mutational models (Mukai, CGD), in general fitness (GF) scenario and in fitness and stabilizing selection (SS) scenarios with selection pressures 25, 50, 75, 100 and 200.

G	*GF*		*SS*									
	Mukai	**CGD**	**Mukai**					**CGD**				
			25	**50**	**75**	**100**	**200**	**25**	**50**	**75**	**100**	**200**
5000	0.34 ± 0.03	0.92 ± 0.03	0.34 ± 0.03	0.34 ± 0.03	0.34 ± 0.03	0.34 ± 0.03	0.34 ± 0.03	0.92 ± 0.02	0.92 ± 0.02	0.92 ± 0.02	0.92 ± 0.02	0.92 ± 0.02
5500	0.20 ± 0.03	0.87 ± 0.08	0.15 ± 0.02	0.17 ± 0.02	0.15 ± 0.02	0.15 ± 0.02	0.12 ± 0.02	0.65 ± 0.04	0.63 ± 0.04	0.58 ± 0.06	0.60 ± 0.04	0.49 ± 0.07
6000	0.13 ± 0.02	0.83 ± 0.06	0.10 ± 0.02	0.12 ± 0.01	0.10 ± 0.02	0.10 ± 0.02	0.07 ± 0.02	0.60 ± 0.05	0.60 ± 0.04	0.57 ± 0.06	0.57 ± 0.04	0.48 ± 0.05
6500	0.09 ± 0.02	0.79 ± 0.08	0.07 ± 0.02	0.08 ± 0.01	0.07 ± 0.01	0.06 ± 0.02	0.05 ± 0.02	0.57 ± 0.03	0.56 ± 0.06	0.55 ± 0.07	0.53 ± 0.07	0.43 ± 0.07
7000	0.06 ± 0.01	0.73 ± 0.08	0.04 ± 0.01	0.05 ± 0.01	0.05 ± 0.01	0.04 ± 0.01	0.03 ± 0.02	0.52 ± 0.06	0.54 ± 0.05	0.52 ± 0.05	0.52 ± 0.05	0.40 ± 0.07

expected, values in the mutation-selection-drift equilibrium (i.e. generation 5000) were higher under CGD mutational model than under Mukai model (0.92 and 0.34, respectively). This observation is in agreement with previous studies (Schoen et al., 1998) which established that when mutations are numerous and with little effect they tend to accumulate lowering the fitness of the population. In contrast, under CGD model there are few mutations with larger effects on fitness, being easier to be removed because carriers do not survive.

When artificial and/or stabilizing selection were simulated, the total fitness further decreased due to the smaller N_e that selection implies, being the purge of deleterious alleles less effective (see Table 1). This led to the fixation of more deleterious mutations and the increase of inbreeding, which allows the expression of recessive mutations and, thus, the rise of inbreeding depression for fitness. From generation 5000 (i.e. when artificial and/or stabilizing selection began) the initial descent of fitness was higher in CGD mutational model, but the slope was lower than Mukai model afterwards (data not shown). For scenarios under CGD model, it could be observed that the decrease in fitness was greater when both types of selection (i.e. artificial and stabilizing) were implemented (0.73 for general fitness scenarios with only artificial selection vs. 0.40–0.54 for stabilizing and artificial selection scenarios in generation 7000).

However, smaller differences were observed for Mukai scenarios, as accumulation must be high enough with artificial selection and no extra effect was detected when adding stabilizing selection.

A somehow counterintuitive performance was observed in SS scenarios. Less restrictive cases (with lower stabilizing selection pressures; $\omega^2 = 200$) produced a larger decrease in fitness. Notice that we were forcing a relation between trait and fitness: fitness of individuals was higher when the phenotypic value of the trait approaches the average – the optimum phenotype. When stabilizing selection is strong, individuals departing from the optimum are eliminated. However, under low selection pressures some individuals with extreme phenotypic values may survive, reducing total fitness of populations.

It must be highlighted that the simulations performed assumed a high reproductive rate, being a breeder able to generate as many individuals as required to complete the desired number of offspring. Therefore, even breeders with a low fitness may end up contributing descendants just by participating in a huge number of matings. If we restrict the number of attempts for a particular individual, mimicking species with a low reproductive rate, no offspring from individuals with many deleterious mutations will be obtained. This will allow for a higher purging effect of natural selection but may also lead to the extinction of populations if few offspring survive (Lynch et al., 1995).

Table 2 shows that the expected heterozygosity (EH) in generation 5000, for the 3000 neutral loci was around 0.33 for all scenarios and with both mutational models. This could be the reflection of the low linkage disequilibrium (LD) generated between the selective and the neutral loci. In generation 5000, the mean heterozygosity in the loci controlling the trait was 0.35 for NF, 0.33 for CGD and 0.33 for Mukai scenarios.

It can be seen that results for EH were almost identical for both mutational models in the GF scenario as well as for all SS scenarios. Thus, it seems that mutational models only have effect on the fitness of populations. When the selection period started expected heterozygosity fell rapidly until it reached a new equilibrium (data not shown). But the dynamic mostly depended on artificial selection and drift, and the presence of direct natural selection had not effect due to the low level of LD between the selective and neutral loci already pointed.

The similarity of results for scenarios NF and GF (not including stabilizing selection) was also observed for the phenotypic mean and variance. The mean phenotype (P) along the generations under artificial selection is also shown in Table 2 and Fig. 1. In generation 5000, P was around 100 in all scenarios, which was the fixed value added to all individuals when calculating the phenotype. This occurs because loci controlling the quantitative trait were randomly chosen and, thus, the probability of one of the alleles having a high frequency (or eventually get fixed) is the same than for the opposite allele. Therefore, the mean genetic value was zero. This performance happened even when fitness was taken into account, as alleles with effect on the trait could be linked to 'wild' type alleles or to mutant randomly.

In the scenarios NF and GF there was a clear and similar selection response with the mean phenotype steadily increasing. In the GF scenarios the response was the same, whatever the mutational model assumed. Phenotypic mean reached a value of 196 in 100 generations, with no extra improvement thereafter. Selection during this period exhausted the genetic variability for the trait and mutation was not able to counteract this effect.

In SS scenarios (i.e. with stabilizing selection), individuals with extreme phenotypes were eliminated. Therefore, artificial selection was unable to improve the phenotype of individuals, especially for high selection pressures where mean phenotype remained close to the optimum value of 100. If selection pressure decreases ($\omega^2 = 200$) there is a little margin for the improvement of phenotype and values reached around 118. The subject of the interaction between directional and stabilizing selection has been studied for a long time from a theoretical point of view as well as in the consequences on real populations. With both types of approaches the conclusion is that, in such scenarios, the fate of the population is reaching an equilibrium with the phenotype for the trait of interest fixed at a value instead of increasing generation after generation, as it is the objective of directional selection. But no study has been published, to our knowledge, on the extra costs incurred for the stabilizing selection acting against directional selection. The profitability of the program from the beginning of the selection until the equilibrium is reached will depend on the economic value of the increase in units of the trait. But once at the equilibrium no extra gain will be obtained but the costs associated to the selection process (i.e. recording the phenotype, keeping the genealogies, …) will still exist. If the selection pressure cease at this moment, the stabilizing selection will make the mean phenotype go back to the original optimum and, thus, gain will disappear.

Phenotypic variance mean (V_P) – Table 2 and Fig. 2- dropped to

Table 2
Expected heterozygosity (EH), phenotypic mean (P) and phenotypic variance (V_P) in generations 5000 and 7000, and EH of the trait in generation 5000 for scenarios NF[1], GF[2] and SS[3] (selection pressures 25, 50, 75, 100 and 200) under different mutational models (Mukai, CGD).

	NF	*GF*		*SS*									
		Mukai	CGD	Mukai					CGD				
				25	50	75	100	200	25	50	75	100	200
EH_{5000}	0.34 ± 0.000	0.33 ± 0.002	0.33 ± 0.003	0.33 ± 0.002	0.33 ± 0.002	0.33 ± 0.002	0.33 ± 0.002	0.33 ± 0.002	0.33 ± 0.002	0.33 ± 0.002	0.33 ± 0.002	0.33 ± 0.002	0.33 ± 0.002
EH_{7000}	0.17 ± 0.003	0.17 ± 0.004	0.17 ± 0.002	0.18 ± 0.004	0.18 ± 0.004	0.18 ± 0.004	0.18 ± 0.004	0.18 ± 0.004	0.18 ± 0.007	0.18 ± 0.007	0.18 ± 0.007	0.18 ± 0.007	0.18 ± 0.007
EH_{trait}	0.35 ± 0.02	0.33 ± 0.01	0.33 ± 0.01	0.33 ± 0.02	0.33 ± 0.02	0.33 ± 0.02	0.33 ± 0.02	0.33 ± 0.02	0.34 ± 0.01	0.34 ± 0.01	0.34 ± 0.01	0.34 ± 0.01	0.34 ± 0.01
P_{5000}	103 ± 6.77	100 ± 5.15	98 ± 4.85	99 ± 7.78	99 ± 7.78	99 ± 7.78	99 ± 7.78	99 ± 7.78	102 ± 3.77	102 ± 3.77	102 ± 3.77	102 ± 3.77	102 ± 3.77
P_{7000}	194 ± 1.62	196 ± 1.12	195 ± 1.75	100 ± 7.61	102 ± 7.80	104 ± 8.02	106 ± 8.40	115 ± 7.76	104 ± 3.75	105 ± 3.72	107 ± 3.41	109 ± 3.89	118 ± 3.18
V_{P5000}	123 ± 16.24	124 ± 13.13	141 ± 29.24	133 ± 15.40	133 ± 15.40	133 ± 15.40	133 ± 15.40	133 ± 15.40	127 ± 16.50	127 ± 16.50	127 ± 16.50	127 ± 16.50	127 ± 16.50
V_{P7000}	97 ± 17.40	103 ± 19.02	99 ± 12.87	21 ± 2.78	33 ± 4.59	45 ± 6.02	53 ± 8.30	73 ± 8.84	20 ± 2.19	33 ± 3.54	44 ± 9.04	54 ± 7.83	72 ± 10.57

[1]NF: without fitness scenario. [2]GF: general fitness scenario. [3]SS: with fitness and stabilizing selection scenarios.

Fig. 1. Phenotypic mean at generations 5000–5200 in without fitness (NF), general fitness (GF) and with fitness and stabilizing selection (SS) scenarios with different selection pressures (25, 200) and different mutational models (Mukai and CGD).

Fig. 2. Phenotypic variance at generations 5000–5200 in scenarios without fitness (NF), with general fitness (GF) and with fitness and stabilizing selection (SS) with different selection pressures (25, 200) and different mutational models (Mukai and CGD).

around 100 in NF and GF scenarios in generation 7000. Although starting from different levels (123 for NF, 124 for GF Mukai and 141 for GF CGD scenarios), in few generations V_P levels reach the same equilibrium value (corresponding to the simulated environmental variance). This performance reflects the faster response observed which exhausted the genetic variance for the trait quickly. The lowest levels of V_P were obtained in SS scenarios. As we relaxed the pressure of stabilizing selection, higher values for V_P were observed, as individuals with diverse and extreme phenotypes survived.

The heritability (h^2) calculated by dividing V_A by V_P was in the range 0.20–0.30 for all scenarios in generation 5000. The realized heritabilities from generation 5000–5100 were 0.28, 0.24 and 0.27 for NF, GF CGD and GF Mukai scenarios, dropping to 0.04–0.17 for SS CGD scenarios and 0.04–0.09 for SS Mukai scenarios due to the effect of stabilizing selection.

Deterioration of fitness and the artificial selection processes itself will decrease genetic diversity and increase inbreeding with their detrimental consequences. Therefore, checking their evolution is also important. This monitoring of the increases in inbreeding and the corresponding effective population size was carried out in the second period. The expected N_e if we only account for the actual number of males and females contributing each generation was $N_e = 26.7$. The observed N_e, calculated from the rate of inbreeding in the first 50 generations of artificial selection, was quite close to that expectation ranging from 25.6 in Mukai GF to 29.1 in scenario CGD SS. The inbreeding was higher in Mukai mutational models than in CGD mutational models because more individuals die and those surviving tend to be more related to each other. Contrarily, inbreeding is lower (*Ne* higher) in stabilizing selection scenarios (SS) than in scenarios only with general

fitness (GF) because in the latter only individuals with extreme (high) phenotype are kept (which are likely close relatives) while under stabilizing selection individuals with intermediate phenotypes are selected reducing the increase in inbreeding.

Although, actually, there is no trait which is totally uncorrelated to fitness, in the basic scenario (GF) we decided to simulate the trait subject to artificial selection without direct effect on fitness (besides the correlation arising from linkage disequilibrium between loci affecting fitness and the trait, as already stated in the Section 2). Obviously, a percentage of the loci could be simulated with effect on both traits, but this would lead to a huge number of possible situations regarding the number of common loci and the correlation of effects within loci. Nevertheless, if there was a positive correlation between the quantitative trait and fitness, the artificial selection would improve faster the phenotype and an equilibrium phenotype would be reached with a negative correlation. The second scenario, (SS) with stabilizing selection, does imply correlation between fitness and the trait of interest and was considered more meaningful in the context of breeding programs.

4. Conclusions

The amount of preserved genetic diversity for a quantitative trait is not affected by natural selection during the evolution of the population prior to the start of the process of artificial selection on the trait, provided there is not genetic correlation between fitness and the trait of interest. Thus, if the reproductive rate is high enough, the influence of direct natural selection on fitness does not compromise the ability of populations to respond to selection within a breeding program.

Nevertheless, if the target trait is affected by stabilizing selection, even with a weak selection pressure, the response to selection is greatly compromised. Thus, it is probably not worthy to include the trait within the objectives of selection of the population. In addition, artificial selection may also fail to succeed when the affected trait is negatively correlated with another trait that it is also worthy to be improved. This could be the case of disease resistance and growth related traits in some species (for example turbot; Rodríguez-Ramilo et al., 2014).

Conflict of interest

All author's declare that there are no known conflicts of interest associated with this publication, and there has been no financial support for this work that could have influenced its outcome.

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