



Decreased Genetic Drift under the Overlapping Conservation

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ABSTRACTS

In order to understand the genetic drift under closed population using equaled family size, two overlapping conservation methods proposed (increment and decrement) were introduced to compare with the discrete population. Finally, the article showed body weight controlled by hundred loci having average gene effect. For increment, the second generation had the parents composed of 400 original population and 100 from first generation; the third population consisted of 300 original population, 100 from first generation and 100 from second generation. The fourth population was composed 200 from original population, 100 from first population, 100 from second population, 100 from third population, and 100 from the fourth population. The fifth population was achieved by randomly selecting 100 from each population. For decrement, the method has the opposite direction. For different population, both more than one thousand and less than family sizes 10 have the stable gene frequency despite the different initial gene frequency and sex ratio. Despite several generations were maintained together, less than 5 generations increment conservation can make the gene frequency improved to less than 0.5, while more than 0.5, increment conservation can make them decrease to less than 5 generations and reach the stable higher gene frequency in the following generations. Generally, less than 0.4 gene frequency can use in the increment conservation. More than 0.7, you can use the decrement method. When 100 loci which have average gene effect and random initial gene frequency were assumed, both increment and decrement conservation methods can keep the phenotype stable.

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Authors' Contributions

JZ designed the study. CZ collected the data. SS and FD analyzed the data. CZ and JZ wrote the article.

Key words

Genetic drift, overlapping generation, gene frequency.

INTRODUCTION

In the past years, overlapping conservation method had become the focus of conservation research, which was widely used in wild and local species. In this approach, understanding the age-structured populations will significantly contribute to conservation, which can be assisted by analyzing the genetic background (Ryman, 1997, Jiajia, 2014). This character, combined with birth rates can certainly affect the amount of temporal allele frequency change, one important genetic parameter of conservation in populations with overlapping generations. Also, the temporal estimates of effective population size method with overlapping generations explored must be suitable for age-structured group (Waples and Yokota, 2007; Jorde and Ryman, 1995).

Allele frequency plays a key role in genetic diversity conservation. Nomura (2005) developed two methods to minimize the loss of genetic diversity in

conserved populations with overlapping generations, which can increase the annual effective population sizes 2-3 folds. They also developed the effective size estimation method applied to populations with overlapping generations. Nishio *et al.* (2010) reported that the allele frequency changes under different dominance degrees. In addition, Hoehn *et al.* (2012) reported the genetic and demographic estimators' differences in small overlapping gecko populations. Kuo and Janzen (2003) built the BOTTLESIM software which was used to estimate the genetic consequences of bottlenecks and conservation plans for overlapping generations. The BOTTLESIM includes four simulation models (single locus with constant population size, single locus with variable population sizes, multiloci with variable population sizes) that are useful for evaluating conservation plans of different population sizes based on the empirical genotypic data. One of the population structure analysis methods in the overlapping pattern is to explore the estimation method of effective population size. Engen *et al.* (2005) derived formulas for the variance effective size of populations subject to fluctuations in age structure and total population size produced by a combination of demographic and

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environmental stochasticity. Genetic estimates of effective size of age-structured population using the LD method are strongly affected by both N_e per generation and N_b per season or reproductive cycle (Waples *et al.*, 2014).

In pace with genetic diversity conservation, inbreeding accumulation during the conservation program restricted the efficiency. Improved percentage of younger individuals, coancestry mating style were the usual methods used to change the levels of inbreeding (Bijma *et al.*, 2000; Sonesson and Meuwissen, 2001). Nomura (2005) developed two methods to minimize average coancestry: minimized the average coancestry in the next generation and minimized the long-term accumulation of coancestry in overlapping population. Of them, the minimizing long-term accumulation of coancestry was suggested in the most practical situations. In above researches, how to choose the percentage of different generations in an overlapping population can minimize the inbreeding process is not clearly. So, in this article, we built the increment and decrement overlapping conservation in small population and evaluated their inbreeding processes and phenotypic changes using multiple loci.

MATERIALS AND METHODS

Hypothesis

In the overlapping generation referred in this article, each generation has the same birth time and harvesting time. All of individuals have the same ability of sex maturation and produced offsprings as broodstocks. Epistatic effects and dominant effects were not included in locus analysis.

Initial population setting

Given that one locus with gradient initial gene frequency (0.1, 0.5 or 0.9) was simulated with finite base population size which had 20, 50, 100, 200, 500, 1000 and 2000; for each generation, the sex ratio was arranged into 1:1 and 1:2 (male: female) using equaled family size conservation. Generally, family size was given 5, 10, 20, 50, and 100. Such simulation, we have 50 generations with 20 replicates. Finally, we evaluate the genetic drift using final allele gene frequency.

Overlap generation conservation creation

For increment (such as population size equals to 500), the second generation have the parents composed of 400 from original population (initial generation) and 100 from first generation; the third population consisted of

300 from original population, 100 from first generation and 100 from second generation. The fourth population included 200 from original population, 100 from first population, 100 from second population, 100 from third population and 100 from the fourth population. The fifth population was obtained from each population with 100 randomly. From sixth to sixtieth population, their parent's construction was similar to the fifth population. For decrement, the method took the opposite direction.

Trait change stimulation

It is assume that body weight was controlled by 100 loci with average gene effect. Initial gene frequencies of such 10 loci were randomly created and ranged from 0.1 to 0.9. The next generation genotypes and their body weights can be achieved using equaled family size conservation with a finite population size and family size. When base population with each locus' initial gene frequency (each locus had 0 dominant degree) and randomly body weight were produced; as each generation's body weights can be evaluated. After 50 generations and their body weights were generated by additive effect model, the curve of body weight among such period can be decrypted.

Bottleneck analysis

According to above illustration, the 5th conversed population (G5D) using decrement method (approach setting: population size = 500, sex ratio = 1:1, family size = 10, number of overlapping = 5 (exclude the initial generation), number of loci = 100, each of locus initial frequency range from 0.2 to 0.9) was developed (composed of 5 population without initial generation), while G5L: the 5th conversed population using increment method, other steps similar to G5D. Bottleneck analysis of G5D, G5L and initial generation (G0I) was carried out by BottleSim 2.6.1. In the bottleneck analysis process, a series of parameters were setting (1. Generation overlapping setting: 5; 2. Reproductive system of organism: Dioecy with random mating; 3. Longevity of organism: 8; 4. Age of reproductive maturation: 2; 5. Population size before bottleneck: 500; 6. Population size during the bottleneck: 100; 7. Number of females in the population during bottleneck: 50; 8. Number of year to simulate: 10; 9. Number of iterations you want to perform: 10; 10. Simulation module: Diploid multilocus, constant population size).

Statistical method

The simulation program was written by SAS 8.0, while the photograph was built using GraphPad Prism 5.

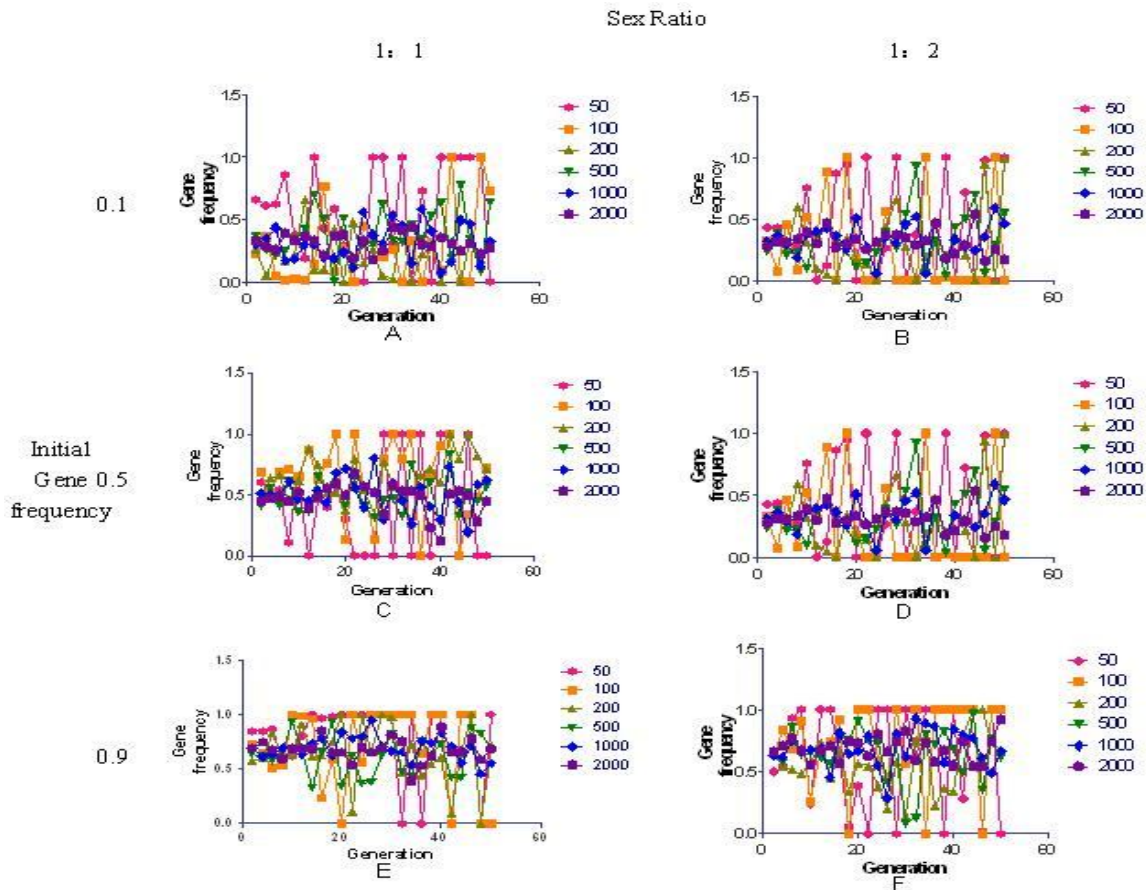


Fig. 1. Influence of population size on genetic drift, under two sex ratios, three initial gene frequencies and six population size with equaled family size conservation (family size=5). First column present the male size equals to female size in different initial gene frequencies, second column depict instead of that female size was two times that of male size in different initial gene frequencies. First row showed 0.1 gene frequency change under different sex ratios, generations and population sizes, second row showed 0.5 gene frequency changes under different sex ratios, generations and population sizes, third row showed 0.5 gene frequency changes under different sex ratios, generations and population sizes.

RESULTS

Different population size

Firstly, the influence of population size on final gene frequency was showed in Figure 1. Under the two sex ratio style and three initial gene frequencies, 50 generations, 6 levels' family size and 5 members in each family were designed to address the effect of population size on genetic drift. Both 1: 1 and 1:2, more than 2000 population size can make a population stable, devoid of genetic drift. These population sizes also can make the different initial gene frequency stable in 50 generations.

Figure 2 shows the effect of family size on final gene frequency. Similarly, two sex ratio style, three initial gene frequency, 50 generations, 5 levels' family

size and 500 population size were considered. Due to more than 1000 population size with stable gene frequency, this approach focused on the 500 population size. Both 1:1 and 1:2, less than 10 family sizes can make the population relatively stable under the little floating, although in the different initial gene frequencies of 50 generations. However, the floating magnitude for family size 10 varied markedly than family size 5.

Overlap generation's conservation under finite population size (500) and family size (10) for 5 years' service life

In order to deal with specie conservation the finite population size (500) and family size (10), the overlap generations conservation method using equal family

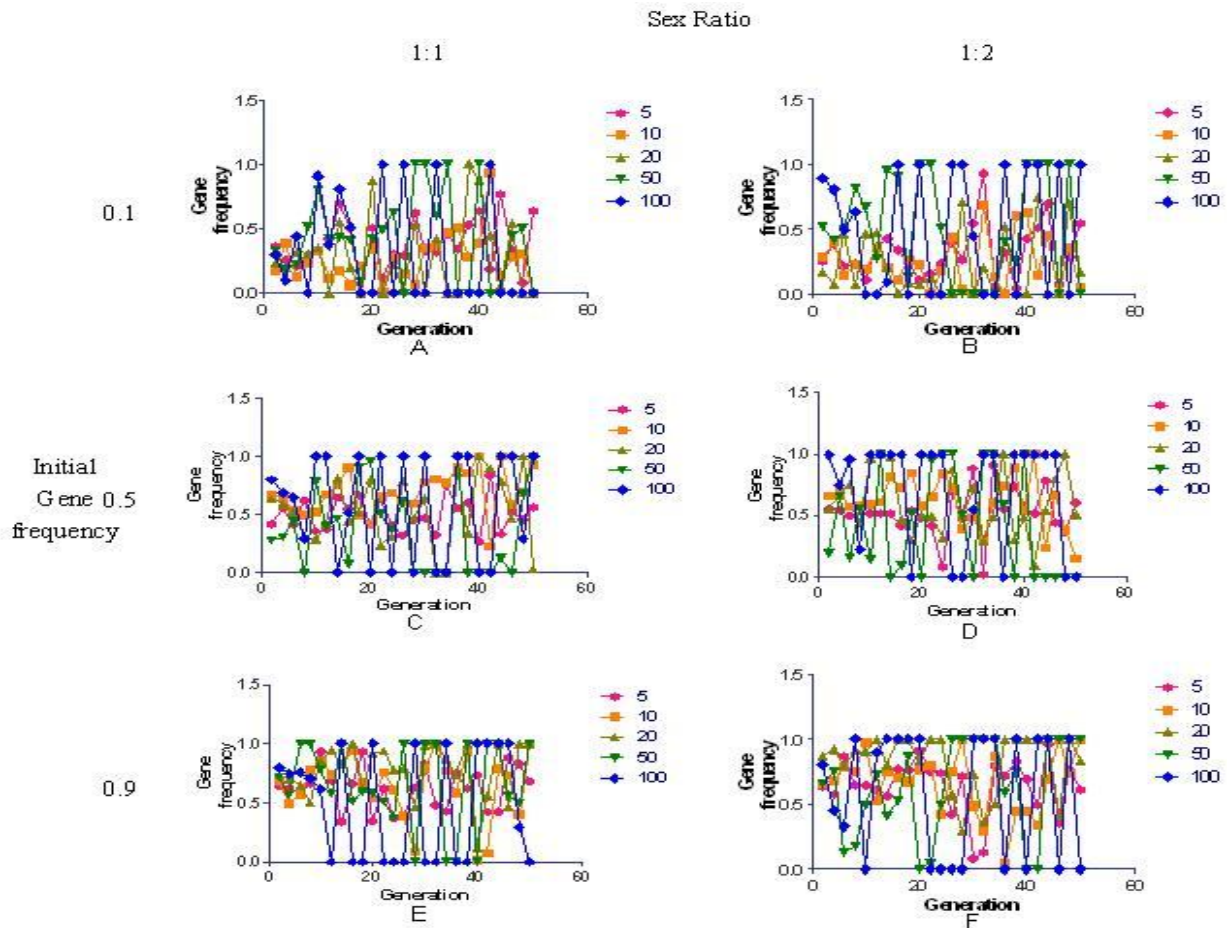


Fig. 2. Influence of family size on genetic drift, under two sex ratio, three initial gene frequency and five family size using equaled family size conservation (population size=500) First column present the male size equals to female size in different initial gene frequencies, second column portray female size was two times that of male size in different initial gene frequencies. First row showed 0.1 gene frequency change under different sex ratios, generations and family sizes, second row showed 0.5 gene frequency changes under different sex ratios, generations and family sizes, third row showed 0.9 gene frequency changes under different sex ratios, generations and family sizes.

conservation was proposed. Firstly, the Overlap original population account for main component in conservation population, named increment conservation, which was thought to be a better conservation because of main original population; its change of gene frequency was evident in the following photograph (Fig. 3). Similarly, from 0.1 to 0.9, two sex ratios gave the same results, and informed, that lower initial gene frequency had bigger gene frequency in the pre-5th population (first, second, third, fourth and fifth population) and stable gene frequency in the following generations. On the contrary, about 0.5, the pre-5th population had better gene frequency and the following generations had higher gene frequency. The 0.9 initial gene frequency of the pre-5th population had lower gene frequency and higher gene

frequency after the sixth population.

Determination of the initial gene frequency during the increment conservation under equaled sex ratio

It is known that the increment conservation method is not suitable with more than 0.5 initial gene frequency conservation. But which thread value between 0.1 and 0.5 for initial gene frequency can be determined for such conservation approach? The present study starts the further research (Fig.4). It was found that all initial gene frequency, less than 0.4, can used in increment overlap generations. For 0.4, its average gene frequency in each generation can reach 0.7 levels, which is better than 0.5.

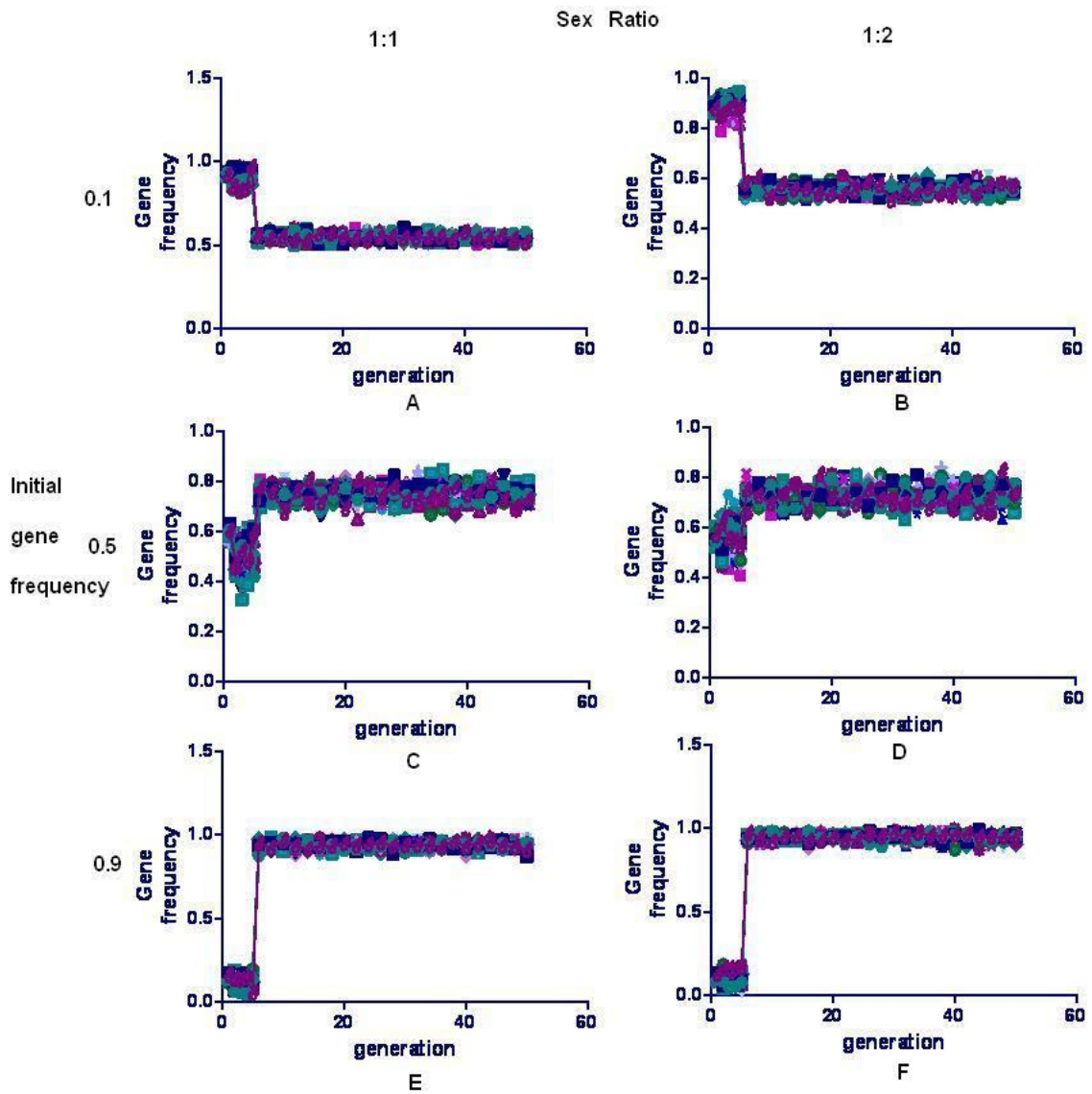


Fig. 3. Influence of overlap generation on genetic drift, under two sex ratio, three initial gene frequency and ten family size using equaled family size conservation approach (population size=500) First column present the male size equal to female size in different initial gene frequencies, second column instead of female, size was two times that of male size in different initial gene frequencies. First row showed 0.1 gene frequency change under different sex ratios and generations, second row showed 0.5 gene frequency changes under different sex ratios and generations, third row showed 0.9 gene frequency changes under different sex ratios and generations.

Conservative effect and determination of the initial gene frequency during decrement conservation under equaled sex ratio

In the above descriptions, more than 0.5, ten family size and 500 population sizes was not appropriate for increment conservation; to deal with such problem, the paper paid more attention to adjusting increment

parameters. Finally, the decrement method can be used to launch conservation with a 0.9 initial gene frequency. A threshold during the bigger initial gene frequency was then determined (Fig. 5). Both 0.9 and 0.8 had stable gene frequency and smooth platform after the 6th generation. However, 0.7 and 0.6 levels still had higher gene frequencies.

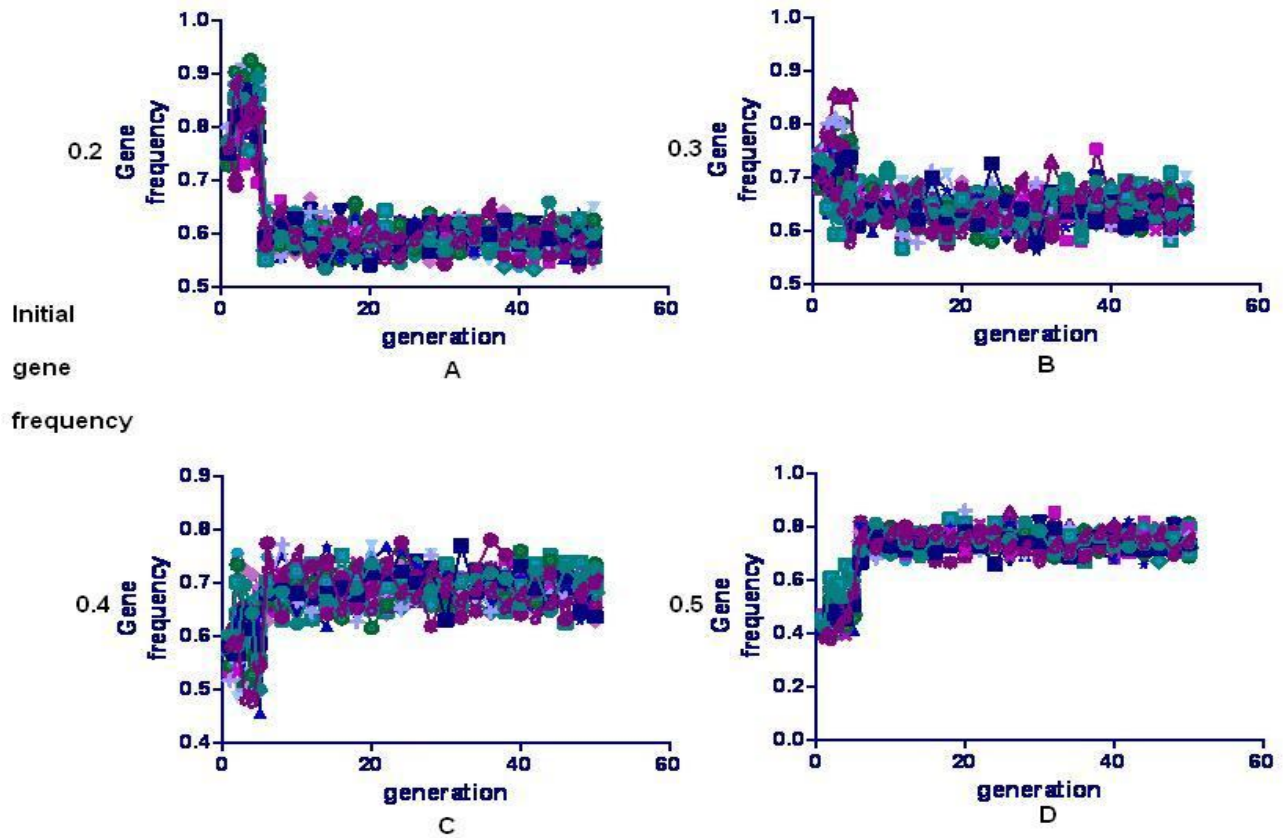


Fig. 4. Determined the initial gene frequency during the increment conservation (50 generations) under equaled sex ratio, ten family size using equaled family size conservation approach (population size=500). An initial gene frequencies equals 0.2, the platform period showed smooth; B initial gene frequencies equals 0.3, the platform period showed less smooth than 0.2; C initial gene frequencies equals 0.4, the platform period showed as the same smooth as 0.3; D initial gene frequencies equals 0.5, the platform period showed more smooth than 0.4.

Multiple loci stimulation in increment and decrement conservation method

To explore further effect on species' conservation, the present research had multiple loci (100 loci) with averaged gene effect on body weight (Fig. 6). Assuming that 100 loci had each random initial gene frequency and sole additive effect, the body weight can be achieved. Then mating strategy was based on the increment and decrement principles. The result showed that both increment and decrement had stable economic trait-body weight and smooth platform.

Un-changed inbreeding coefficient using overlapping conservation

In order to understand the variation of inbreeding coefficients in the above 3 methods, the present study take the previous 6 generations for example because of

defined close relative within such generations. When the conservation last such periods, decrement had the lower value, followed the increment approach (Fig. 7). However, decrement method had the stable condition from 3rd. Non-overlapping had the worst conservation.

Focusing on the 5th overlapping generations using increment and decrement methods (excluding initial generation), we explored the tendency of number of alleles and heterozygosity using BottleSim2.6.1 software. Both increment and decrement methods have higher effective number of alleles and expected heterozygosity compared to initial population during the bottleneck process. On the 7th year, observed heterozygosity of decrement method enters into the increasing phase and has the higher number compared to others, while that of increment method is lower than that of initial population (Fig.8).

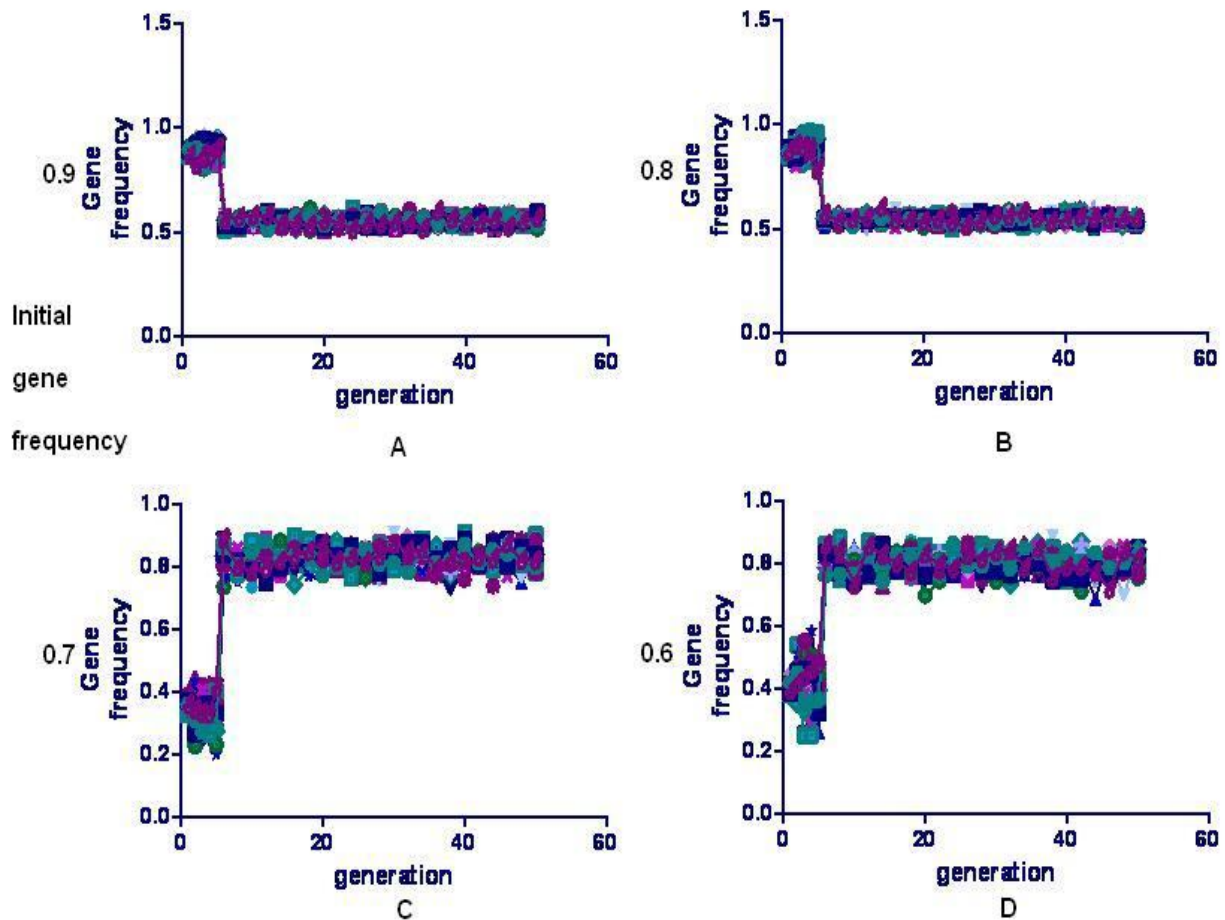


Fig. 5. Determined initial gene frequency during the decrement conservation (50 generations) under equal sex ratio, ten family size with equaled family size conservation approach (population size=500). An initial gene frequency of 0.9, the platform period showed smooth, the pre-5th population had the higher gene frequency, from the 6th population, the gene frequency keep stable; B initial gene frequency equals 0.8, the platform period showed smooth like 0.9, the pre-5th population had the higher gene frequency, from the 6th population, the gene frequency keep stable; C initial gene frequencies equals 0.7, the platform period showed the smooth is less than 0.8, the pre-5th population keep the middle gene frequency, the following generations keep the higher gene frequency; D initial gene frequencies equals 0.6, the platform period showed less smooth than 0.7, the pre-5th population keep the middle gene frequency, the following generations keep the higher gene frequency.

DISCUSSION

Overlapping can change genetic gain

In practice, overlapping population conservation was usually applied in multiple species and focused on by its mating schemes. Sonesson and Meuwissen (2002) found that minimum coancestry mating with a maximum of one offspring per mating pair can increase the genetic gain from 11-18% for overlapping and 1-4% for discrete generations, when comparing schemes with similar genetic gain and size. In the present study, we found

overlapping generations can improve the gene frequency in the 5th generations and keep stable in the subsequent years. This result illustrated that overlapping selection can change the genetic gain in the few years by gene frequency. Actually, Villanueva *et al.* (2000) propose optimum breeding schemes for maximizing the rate of genetic progress with a restriction on the rate of inbreeding for populations with overlapping generations, which simulated by annealing algorithm undergoing mass selection.

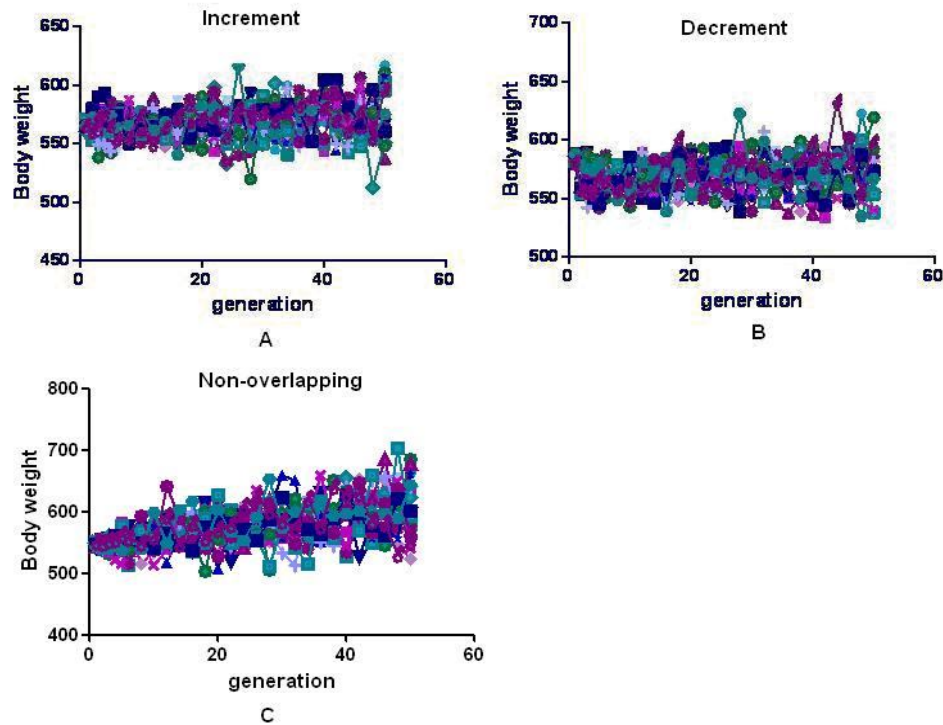


Fig. 6. Multiple loci simulation in increment and decrement conservation method during the 50 generations conservation under equaled sex ratio, ten family size with equaled family size conservation approach (population size=500). A, increment method conservation; B decrement method conservation. Both increment and decrement have stable conservation for single economic trait and multiple loci with random initial gene frequency. C Non-overlapping generation conservation. With increased generations, the variation of body weight changed markedly.

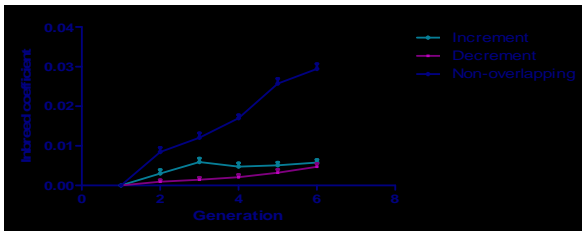


Fig. 7. Inbreed coefficient estimation in increment and decrement conservation method compared to the non-overlapping approach during 6 generations conservation under equaled sex ratio, ten family size using equaled family size conservation approach (population size=500). For 6 generations, increment method conservation had the platform located at the middle position from increment, decrement to non-overlapping method. Compared to this, decrement method conservation had lowest inbreed coefficients and the growth curve can not stop. Non-overlapping methods had the highest coefficients and keep the value rise step by step.

Besides this, the same population size was set in order to contrast with discrete population having same family size and number of individuals in the study. Moreover, after 6 years, the gene frequency enters into platform because of the same individuals per generation in overlapping population. This may be approved by the research done by Hill (1979), in which the effective population size was the same as that of a population with discrete generations having the same variance of lifetime family size and the same number of individuals entering the population per generation. All of these results were derived under the family size, sex ratio, initial gene frequency, population size. Similarly, Engen *et al.* (2007) generated formulas that estimated the effective population size considering sex ratio, age, temporal fluctuations in population size.

Overlapping mating and inbreeding coefficients

Another problem for overlapping conservation is how to minimize the inbreeding coefficient. Firstly, Johnson (1977) constructed age, sex and number of individual based inbreeding matrix. In the following stage,

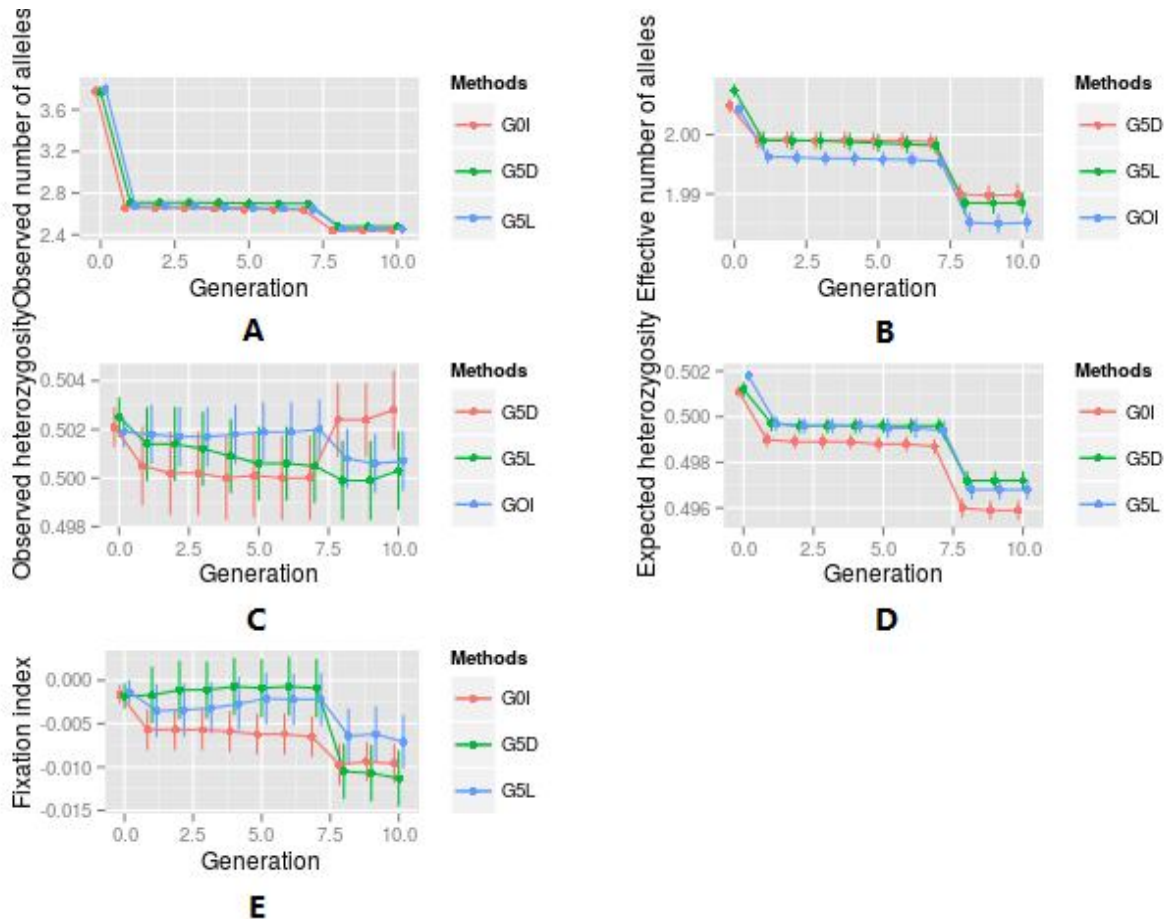


Fig. 8. Population bottleneck analysis of both increment and decrement conservation methods compared to the non-overlapping approach using BottleSim2.6.1. In the bottleneck analysis process, a series of parameters were setting (1. Generation overlapping setting:5; 2. Reproductive system of organism: Dioecy with random mating; 3. Longevity of organism: 8; 4. Age of reproductive maturation: 2; 5. Population size before bottleneck: 500; 6. Population size during the bottleneck: 100; 7. Number of females in the population during bottleneck: 50; 8. Number of year to simulate: 10; 9. Number of iterations you want to perform: 10; 10. Simulation module: Diploid multilocus, constant population size). G0I: initial generation; G5D: according to this article, the 5th conversed population using decrement method (approach setting: population size = 500, sex ratio = 1:1, family size =10, number of overlapping = 5 (exclude the initial generation), number of loci = 100, each of locus initial frequency range from 0.2 to 0.9) was developed(composed of 5 population without initial generation); G5L: the 5th conversed population using increment method, other steps similar to G5D.

A: All of 3 methods have the similar observed number of alleles ; B: Both increment and decrement methods have higher effective number of alleles compared to initial population during the bottleneck process; C: On the 7th year, observed heterozygosity of decrement method enters into the increasing phase and has the higher number compared to others, while that of increment method is lower than that of initial population; D:Both increment and decrement methods have higher expected heterozygosity compared to initial population during the bottleneck process; E: After the 8th year, fixation index of decrement method is lower than that of initial population contrast to that of increment.

Choy (1978) provided the methods which can calculate the inbreeding coefficients in populations with overlapping generations exactly. Secondly, how to use the overlapping mating to reduce the inbreeding gain? Bijma *et al.* (2000) reported that F/year decreases much

more slowly compared to $F/\text{generation}$ as the number of younger individuals increases, whereas the decrease is more similar as the number of older individuals increases. The result is similar to that both the decrement and increment approach had lower inbreeding coefficient

value compared to discrete population. And decrement method (higher percentage of younger individuals) had the stable condition from 3rd generation. Further more, Sonesson and Meuwissen (2001) proposed the minimum coancestry mating method which resulted in lower levels of inbreeding than random mating, but F was approximately the same. Nomura (2005) compared two mating design and found that minimizing the average coancestry in the next generation had the better result in a overlapping population compared to minimizing the long-term accumulation of coancestry because of 5~10% larger annual effective size in subsequent years. In addition, the effective number of alleles and effective heterozygosity were still higher after the following 5 overlapping generations using bottleneck analysis methods compared to the initial population. This may be a suitable approach to have less inbreeding and more heterozygosity.

Economic traits conservation in overlapping population

The difference between overlapping population and discrete population is age-structured group. Mueller and James (1984) built the procedure which was used to analysis the phenotypic response to selection of traits with direct and maternal components when generations overlap by adding the age-sex distributions. In the subsequent research, multiple loci were added into simulating the phenotype response. For example, Ellner and Sasaki (1995) reported that under the randomly fluctuating selection in overlapping generations, the quantitative traits' fluctuations variance were related with number of polymorphic loci and distribution of the phenotypic optimum θ . In this paper, we assumed that the 10 loci (additive effect) had each random initial gene frequency and sole additive effect on body weight. Both increment and decrement had stable economic trait-body weight and smooth platform. Compared to discrete population, overlapping conservation in the subsequent year had stable and lower variance. Wang *et al.* (2009) proposed an estimator (EPA, estimator by parentage assignments) of current N_e of populations with overlapping generations, using sex, age, and multilocus genotype information of a single sample of individuals taken at random from the population in order to get the effective number of breeder of each age class. However, the number of each age class was set as same size as in the present study. Thus, however, the use of age class information to achieve better economic traits needs further research.

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