Asian Fisheries Science 1(1987):75-82.
Asian Fisheries Society, Manila, Philippines
https://doi.org/10.33997/j.afs.1987.1.1.007

Maintenance of Genetic Quality in Cultured Tilapia

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Abstract

Management of a population's effective breeding number (N_e) is crucial, because it is inversely related to genetic drift and to inbreeding. Reductions in N_e can irreversibly damage a population's genetic and biological potential. Recommended N_es for standard reference populations of tilapia are 390-500, and those for farm populations are 100-150. Effective breeding numbers for reference populations must be larger because the management goal is conservation genetics -- preventing detrimental levels of inbreeding, preventing the loss of rare alleles and preserving heterozygosity. Effective breeding numbers for farm populations can be smaller because the management goal is to maximize productivity. Acquisition of a foundation stock is the most important aspect of management of a population's N_e, because the alleles that exist in that sample are those that determine the population's genetic and biological potential. The fecundity of tilapia can facilitate maintenance of desired N_es; females produce only about 150-500 offspring per spawn, so even moderately-sized farms require hundreds of broodfish.

Introduction

Genetic aspects of broodstock management are of the utmost importance, because a population's genome determines its biological potential. Since many hatchery populations are small or had small founder populations, the best way to describe them is not by total number of fish, but by the effective breeding number (N_e), which is a function of the number of males and females that produce viable offspring, the sex ratio of the fish that produce offspring, the system of mating and the variance of family size. Most tilapia culturists use random mating. When this system of mating is used, N_e is:

$$N_e = \frac{4(♀)(♂)}{(♀)+(♂)}$$

where ♀ and ♂ are the number of females and males that produce viable offspring.

Knowledge of a population's N_e is crucial, because it is inversely related to inbreeding and to genetic drift. Consequently, restrictions in N_e can irreversibly damage a population's genetic and biological potential. Studies have suggested that restrictions in N_e resulted in loss of genetic variance in hatchery stocks of rainbow trout, *Salmo gairdneri* (Allendorf and Utter 1979); cutthroat trout, *S. clarki* (Allendorf and Phelps 1980; Leary et al. 1985); brown trout, *S. trutta* (Ryman and Ståhl 1980); Atlantic salmon, *S. salar* (Cross and King 1983; Ståhl 1983); black seabream, *Acanthopagrus schlegeli* (Taniguchi et al. 1983); and *Oreochromis niloticus* (Tave and Smitherman 1980). Because of this, proper broodstock management means managing the population's N_e (Tave 1984, 1986, in press a, in press b).

Managing N_e

Conserving a stock's biological potential involves managing its N_e at or above a pre-determined number. A number of recommendations have been made; the following minimum N_es have been suggested: Ryman and Ståhl (1980) -- 60; Kincaid (1976) -- 200; Kincaid (1979) -- 500; Food and Agriculture Organization of the United Nations (FAO/UNEP 1981) -- 50 for a "short-term" work and 500 for "long-term" work; US Fish and Wildlife Service (1984) -- 1,000; Tave (1986) -- 68 to 685, depending on the program and its goals. However, there is no universal N_e that can be recommended to prevent inbreeding- and genetic drift-related problems.

Minimum desired N_es for a given population can be determined only after answers are provided for the following questions: What is the maximum desirable level of inbreeding? What is the frequency of the rarest allele to be saved? What probability of saving the allele is desired? How many generations will be incorporated into the project?

The answers to these questions are determined by information from previous genetic research, production goals (e.g., management of a reference or a farm population), budget, facilities, generation interval (broodstock replacement interval) of the species being raised and the number of years that will be included in the project. Tave (1986, in press a, in press b) outlined methods to determine minimum N_es that will prevent inbreeding- and genetic drift-related problems.

The first genetic goal in the management of a population is to prevent inbreeding from exceeding detrimental levels. Unfortunately, the effects of inbreeding in tilapia have not been systematically investigated, so the levels of inbreeding which adversely affect different phenotypes are unknown. There has been an inbreeding study in *O. mossambicus* (Ch'ang 1971), but the effects of inbreeding on productivity could not be quantified because the population was already inbred, the inbred group was compared to a crossbred group, and the fish were raised in aquaria. When detrimental levels of inbreeding are unknown, Tave (1986, in press a) suggested using 5% as a conservative estimate and 10% as a liberal estimate.

The second genetic goal is to prevent the loss of alleles (frequency drops to zero) by genetic drift. The probability of losing an allele is proportional to its frequency, so rare alleles are more likely to be lost. Consequently, the goal is to prevent the loss of rare alleles.

In the management of a standard reference population, the goal should be to preserve heterozygosity and preserve polymorphic loci. Alleles whose frequency (f) = 0.01 contribute to polymorphism, so the goal should be to save these alleles. In food fish farming, where the major goal is to maximize yield, rare alleles probably do not contribute significantly to productivity so the standards can be relaxed. It has been suggested that it is necessary to conserve only alleles whose f = 0.05 for farm populations (Tave 1986). It is impractical to maintain an N_e that would give a 100% guarantee that a rare allele was not lost. Consequently, a confidence level for saving the alleles must be established. Tave (1986, in press b) suggested using either P = 0.01 or P = 0.05 (99% and 95% probabilities of keeping the allele).

If N_e is allowed to decline below the desired minimum number, a genetic bottleneck (a severe restriction in N_e) can occur. A bottleneck can permanently damage the genetic quality of a population, because it has a permanent effect on overall N_e. The N_e over a series of generations is the harmonic mean of the N_e in each generation. Thus, the generation with the smallest N_e has the greatest impact on overall N_e.

Bottlenecks accelerate the effect of genetic drift. Nei et al. (1975) estimated that it takes hundreds to thousands of generations to recreate genetic variance that is lost as a result of a bottleneck, other than through the importation of new broodstock.

In addition, bottlenecks can drastically increase inbreeding. Inbreeding and N_e are interlocked in a positive feedback cycle. Inbreeding reduces N_e from what it would have been had there been no inbreeding, and that in turn, increases inbreeding.

When calculating the population's desired N_e, two values are determined: an N_e to prevent inbreeding-related problems and an N_e to prevent genetic drift-related problems. When the two values differ, choose the larger value, because that will allow achievement of both goals.

Reference Populations

Acquisition and maintenance of reference populations of tilapia would be an important development in aquaculture, because they could be used to supply aquaculturists with broodfish of good genetic quality. A number of reference populations should be established, because there are several important species of tilapia and because there are populations or subspecies within each species. Each reference population should be managed so that it will not be contaminated with genetic material from another population.

To properly manage a standard reference population's gene pool, the following genetic goals should be incorporated. First, inbreeding should not exceed 5%. Second, alleles whose $f = 0.01$ should be saved, and the confidence level of saving the alleles should be $P = 0.01$ (a 99% probability of keeping the alleles).

Long-term planning must be incorporated into the management of a standard reference population, and 25-50 generations (25-50 years) is appropriate. To achieve these goals, N_e should be 390-500.

Acquisition

Founder stocks should be gathered from the wild. Most hatchery stocks have some inbreeding and have reduced heterozygosity. In addition, some hatchery populations are contaminated (Taniguchi et al. 1985; Macaranas et al. 1986). Populations of tilapia which contain genes from two species can have more than one system of sex determination, which will cause problems during the production of monosex populations.

Wild populations should be surveyed electrophoretically to determine the geographic ranges of the populations. Depending on goals, the reference population can be a single wild population or can be created from several wild populations.

Care should be taken to ensure that the gene pool is adequately sampled. The N_e for the foundation generation will be determined when the fish reproduce, not when they arrive at the station. Sample sizes should be larger than the desired N_e to account for mortality and lack of spawning success.

Gene frequencies in the sample should be determined by electrophoresis to ascertain whether the gene frequencies of the sample and the wild population are the same and to produce guideline frequencies for the management of the reference population.

Management

There should be two major goals in the management of reference populations. First, the population's genome should be conserved, so that gene and genotypic frequencies will not change over time. Second, fish should be produced to supply other hatcheries with foundation stocks of good quality.

In order to manage a reference population's N_e, reproduction must be stringently controlled. Traditionally, tilapia are spawned in ponds, but it is difficult to calculate and manage N_e with this type of mating system.

Fish must be paired and spawned in spawning nets or tanks. When equal numbers of males and females reproduce, N_e is maximized in closed populations (Tave 1984). Broodfish sex ratios of 3 females:1 male or 2 females:1 male are often used in tilapia hatcheries to maximize offspring production, while using the fewest number of broodfish (Hughes and Behrends 1983). However, management of N_e requires that the proper number of broodfish are spawned, and offspring production efficiency is of secondary importance.

Pairing must be at random; intentional or unintentional selection of broodfish must be prevented. A fish should not be allowed to spawn more than once, unless all of its offspring die. This type of reproduction will allow calculation of N_e.

Each family should be raised in an individual net or tank for 20-30 days. At this point the fry can be transferred to ponds or tanks. Before stocking, family size should be equalized. Unequal family size lowers N_e.

When the fish become sexually mature, a random sample should be taken to be used as replacement for the previous generation. The sample should be larger than the desired N_e, because some fish will die, some will not spawn and some fish must be sacrificed to determine the effects of genetic drift. If electrophoretic examination reveals that there were drastic changes in gene frequencies and that more rare alleles were lost than expected (based on N_e), the fish should be discarded and the parents should be respawned.

Requests to establish replicate reference populations must be received well in advance so that production of the new generation for the reference population and production of fish for other hatcheries will be coordinated. Each request should be filled by spawning 195-250 pairs in nets or tanks, which is an N_e of 390-500. Total number of requests should be known before the spawning season to determine how many matings will be needed to fill the requests. Each request should be filled by shipping an equal number of fish (minimum of four) from each spawn. By shipping at least four fish per spawn, sample size will be at least 780-1,000 fish, which should compensate for mortality and lack of spawning success.

Farm Populations

Genetic aspects of broodstock management are different for farm populations of tilapia. The major genetic goal in the management of farm populations is to exploit the genetic potential to increase productivity. Because they are not standard reference lines, it is not necessary to practice conservation genetics. Consequently, N_e can be smaller.

Appropriate genetic goals for farm populations are: inbreeding should not exceed 5%; alleles whose $f = 0.05$ should be saved, and the confidence of saving the alleles should be $P = 0.01$ (a 99% probability of keeping the alleles). Effective breeding numbers to achieve these genetic goals for 10-15 generations (10-15 years) are 100 to 150.

Acquisition

Stock can be acquired from either a hatchery or from the wild. If a hatchery population is to be acquired, information about disease history, growth and other production characteristics should be gathered to help determine the most desirable population. Additionally, information about the N_e for each generation should be evaluated. Populations with small N_es may portend problems.

When acquiring a population, make sure that enough fish are obtained to account for mortality and lack of spawning success. As was the case with reference populations, N_e of the foundation population is the most crucial aspect of management because it determines the population's genetic and biological potential.

Management

Management of N_es for farm populations of tilapia is much easier than that for highly fecund species. If the foundation population has the desired N_e and has a broad genetic base, it should be fairly easy to maintain the genetic quality of the population, because of the fecundity of tilapia. Because each female will produce only about 150-500 offspring per spawn, even moderately-sized tilapia farms require hundreds of broodfish. Consequently, with proper management it is relatively easy to maintain the proper N_es for farm populations of tilapia.

Acknowledgement

We thank Katherine B. Tave for critical review of the manuscript.

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