

**WINTER SCHOOL**  
**ON**  
**Recent Advances in**  
**Mariculture Genetics**  
**and Biotechnology**

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**Course Manual**



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## **AN OVERVIEW OF THE GENETIC TECHNIQUES FOR IMPROVEMENT OF FISH AND SHELLFISH FOR AQUACULTURE**

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### **Introduction**

There are two ways in which an aquaculturist can increase his yields. The first is by environmental manipulations, such as the optimal use of fertilizers, feeds, improved water quality management etc. The second is by growing genetically improved stock. Any of the different genetic manipulation techniques available can be employed for producing genetically improved brood stock. In addition to the conventional quantitative genetic techniques like selection and breeding, modern tools like chromosomal engineering for production of polyploids, gynogenesis and androgenesis can also be employed. The most modern technique is genetic engineering where in a desirable gene or set of genes from any source can be transferred into a fish for producing a transgenic fish with desired characteristics. However, an in-depth knowledge of the science of fish genetics is a prerequisite for the formulation of the appropriate techniques for their improvement.

### **Quantitative genetic tools**

Breeding is the applied science of genetics. There are a number of selection and breeding programmes that can be used for the genetic improvement of fish. These are the two traditional techniques that have been used for thousands of years to improve all major crops and livestock grown by farmers. Although, farmers have conducted scientific breeding programmes on livestock for thousands of years, fish farmers are only beginning to use selection, hybridisation, or other breeding programmes to improve aquaculture species of food fish. Although some progress has been made, many fish farmers are culturing fish that are essentially wild and unimproved.

The ultimate goal of every selective breeding programme is to improve the breeding value of the population. The fish genes determine the breeding value. A farmer hopes that when he improves the breeding value of his population its monetary value also will be improved, which is determined by the fish's phenotypes. To accomplish this goal, a breeder selects (saves) fish that possess certain desired phenotypes and culls (removes) those that do not. By selecting and mating only the best fish (largest, heaviest, those with the desired colour, etc.) he hopes that the selected brood fish will be able to transmit their superiority to their offspring, thereby creating a genetically improved population. If this occurs, the next generation will be more valuable because the fish will grow faster, which will increase yields; the fish will grow more efficiently, which will lower feed costs; or all fish will have a more

desired body colour, which will increase their market value. There are a number of methods, which can be used for selection of superior genotypes for scientific breeding.

Inbreeding and crossbreeding (hybridisation) are the two traditional breeding approaches that have been successfully used for the improvement of crops and livestock. Inbreeding is often combined with hybridisation to improve the results of the crossbreeding programme. Crossbreeding is a breeding programme that tries to find mating combinations between different populations of fish which produce superior offspring for grow-out, offspring that are said to exhibit hybrid vigour. Although crossbreeding is a tried-and-true method of increasing yields, the results of crossbreeding programmes are impossible to be predicted (unless the mating has been made previously), so the production of superior offspring is a hit-or-miss proposition. Many combinations often have to be evaluated before a combination that produces offspring with hybrid vigour is discovered. Crossbreeding programmes usually involve different strains within a species (intraspecific hybridization), but different species can also be hybridised (interspecific hybridisation). To date, much of the breeding work in aquaculture has been devoted to hybridisation among the different species of tilapia in an attempt to produce all hybrids for grow-out.

In general, crossbreeding is used to produce superior fish for grow-out, while selection is used to create superior brood fish. The hybrids that are created in a crossbreeding programme are usually grown and sold as food. A farmer rarely retains and spawns the hybrids to produce a new generation of production fish. On the other hand, brood fish that are created in a selective breeding programme are created for one purpose – to produce the next generation of genetically superior fish for grow-out and their offspring can, in turn, be retained and selected to continue the process of producing next generation of improved stock.

In recent years, cytogenetic research had led to the development of three additional breeding programmes that can be used to increase yields. One of the most common breeding programmes in aquaculture is the production of sex-reversed brood stock to produce monosex populations for grow-out. This is done either because one sex is superior or more desirable or to prevent reproduction during grow-out. For example female sturgeon are more valuable than males because they are heavier; female salmon are the more valuable sex because sexually precocious males die before they can be harvested; however male tilapia are more desired than females because they grow twice as fast. The major goal in tilapia farming is to prevent reproduction during grow-out; this can be best accomplished by producing a monosex male population.

The production of sex-reversed brood stock is usually accomplished by feeding sex hormones either estrogens or androgens to sexually undifferentiated fry to sex-reverse them. Sex-reversed fish are individuals that are one sex phenotypically but the other genetically. If sex reversal is done properly, sex-reversed fish are capable of producing monosex populations for grow-out/ The type of hormone used- estrogens to produce sex-

reversed females or androgens to produce sex-reversed males depends on the sex-determining system of the species and whether you want to produce an all male or an all female population.

### **Chromosomal engineering**

Another programme that is becoming more commonplace is the application of chromosomal manipulation. The most common form of chromosomal manipulation is the production of triploids. This involves the use of temperature or pressure or chemicals to shock newly fertilized eggs. If shock is applied properly, it prevents the second polar body from leaving the egg and therefore, the newly fertilized egg contains a haploid sperm nucleus, a haploid egg nucleus, and a haploid second polar body nucleus. These three haploid nuclei fuse and produce a triploid zygote, which in turn, produces a triploid fish. Triploids are sterile. This type of breeding programme is used to enable farmers to grow exotic species whose culture might otherwise be illegal or to induce sterility in species that become sexually mature before they reach market size. For example, grass carp culture in most of the United States is legal only if a farmer raised triploids. This technique can also be used to improve the results of interspecific hybridization.

Chromosomal manipulation can be used to produce animals with genetic contribution from only the mother (gynogens) other only from the father (androgens). This is done by creating haploid zygotes and by then shocking the zygotes to produce diploid zygotes. Haploid zygotes are produced in one of two ways: a normal egg is fertilized by sperm is used to fertilize an egg whose DNA has been destroyed by UV irradiation (gynogenesis); a normal sperm is used to fertilize an egg whose DNA has been destroyed by UV irradiation (androgenesis). Gynogenesis and androgenesis are techniques that can be used to produce highly inbred lines for breeding purposes. It can also be used to produce super males; such males are capable of producing all-male populations.

### **Genetic engineering**

In recent years, a new, high-tech breeding programme has been developed viz. genetic engineering. This is a breeding programme that transfers a single gene or a set of genes from one individual into another. This transfer can be within a species, between two species, or even across kingdoms. Although genetic engineering has generated lots of publicity, there are only very few cases of successful production of genetically superior fish for farmers. Furthermore, this type of breeding programme is very expensive, highly regulated, and requires highly trained scientists. Scientists working at universities, governmental research stations, can generally conduct this type of breeding programme or at agribusinesses that are capable of supporting expensive research projects with secure containment facilities.

Though selective breeding is the simplest of all, the decision to conduct a selective breeding programme is a decision that must be made for each farmer or each fry/fingerling

production centre on a case-by-case basis. Because selective breeding programmes require dedication, a certain level of sophistication, record keeping, and the investment of extra labour. Additionally, selective breeding programmes are not free; they also require the investment of money. Finally, these programmes usually do not produce immediate improvements. Improvements are usually not seen for at least one growing season, so a farmer must be able to incorporate long-term planning into his farm management programme, and he must be patient. As a result, within a region, only a small percentage of farmers or fingerling production centres should or will ever conduct selective breeding programmes. A final requirement that must be met before a farmer can conduct a selective breeding programme is the existence of proper facilities.

When the uncontrolled reproduction resulting significant suppression of yield where the product is not economically marketable, farmers can benefit from breeding programmes that can produce monosex populations or sterile populations. For example, the biggest problem in tilapia culture is the fact that tilapia become sexually mature before they reach market size and, as a result, reproduces in the grow-out ponds. This uncontrolled reproduction means that a significant percentage of yield is unmarketable. Tilapia farmers may benefit from breeding programmes that can produce monosex male populations far more than from selective breeding programmes that might improve growth rate.

### **Suggested Reading**

- ❖ Introduction to quantitative genetics. 2<sup>nd</sup> Edition. Falcones, D. S. Oliver and Boyd Ltd. Edinburgh and London.
- ❖ Genetics and fish breeding. Colin E. Purdon. Chapman and Hall.
- ❖ Genetics for Fish hatchery managers. Douglas Tave. AVI publishing Co., Connecticut.
- ❖ Genetics of livestock improvement. John.F.Lasley. Prentice Hall of India Ltd.New Delhi.
- ❖ Population genetics and Animal improvement. Lerner, I. M. Cambridge Univ. Press.
- ❖ The genetic basis of selection. Lerner, I. M. John Wiley and Sons Inc. New York.
- ❖ Population genetics in Animal breeding. Pirchner. Panima Publishing Corporation. N.York.
- ❖ Selective Breeding programmes for medium –sized fish farms. FAO-Fisheries Technical Papers
- ❖ Molecular Cloning - Laboratory manual; Sambrook, J., Fritch E. F. and Maniatis, T; Cold spring Harbor Laboratory Press, New York.
- ❖ Recent advances in Marine Biotechnology; Science Publishers, Inc., Enfield, USA.