

- Invited Paper -

GENETIC IMPROVEMENT OF AQUACULTURE FINFISH IN JAPAN. PRESENT STATUS AND PROSPECT

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The history of genetic improvement, i.e. breeding in aquaculture organisms is shorter than those in agricultural plants and domestic animals. However, recent progress of aquaculture industries requires active development of better farming races or varieties in important species. The practical breeding of finfishes has been carried out by introduction, crossing (hybridization), selection, ploidy manipulation, cloning and other advanced biotechnologies. In this paper, principle, technique and results of these activities are reviewed and then prospect of genetic improvement in aquaculture is discussed from the viewpoint of sustainable finfish production.

Races of goldfish were introduced from China about 500 years ago and then Japanese varieties were generated by further improvement with cross-breeding and selection techniques. German scaly carp, mirror carp and Donaldson rainbow trout are typical examples of introduced races still utilized in aquaculture. Cross-breeding is a method to integrate desirable traits of one race (species) now is able to reproduce in Japan. Fertile gametogenesis in this hybrid might attribute to the polyploidy of parental two species: probable amphidiploidy might allow synapsis between two homologous chromosomes from each different species and subsequent bivalent formation. Thus, the bester is considered as a new synthetic aquaculture species. Selection programs also generated a new aquaculture race in red sea bream. Growth performance of red sea bream has been improved by selection for more than 25 years and the time required for attaining a commercial size (about 1 Kg) was shortened from initial 3 years to 2 years.

Chromosome manipulation techniques to induce gynogenesis (all-female inheritance), androgenesis (all-male inheritance) and polyploidy (genotype with extra set(s) of homologous chromosomes) have been actively developed and then practically applied for aquaculture of relatively large number of fish species in the last two decades. As triploids are predicted to be sterile due to aberrant meioses caused by additional chromosomes, deleterious influence of maturation on survival, growth and meat quality can be eliminated in triploid population. In most cases, triploids are produced as all-female population by fertilization of sperm of sex-reversed XX males to assure their sterility, because triploid males often exhibited gonad development. Pre-maturation growth of triploids is similar or

inferior to counterpart diploids, but triploids often show better performances in post-maturation season. Triploid hybrids (allotriploids) often exhibited better survival than diploid hybrids. At present, allotriploid salmonids such as rainbow trout x amago and rainbow trout x Japanese charr, are cultured as local products specific to active sight-seeing points, in some prefectures. Although tetraploid line can be useful for large-scale production of triploids by diploid x tetraploid crosses, production of viable and fertile tetraploids is very difficult technically as well as biologically.

Genetically identical, cloned fish has been produced in ayu, amago salmon, hirame flounder, fancy carp and very recently red sea bream, by second cycle of polar-body gynogenesis of the eggs from all-homozygous gynogenetic or androgenetic diploids, produced by inhibition of the first cleavage. Clonal families can be maintained by fertilization using sperm of sex reversed male within a clone. Heterozygous but clonal line produced by hybridization between two different homozygous lines generally outperform than parental homozygous clone in commercially important traits. This can be explained by no or little influence of inbreeding in heterozygosity. Therefore, homozygous clones should be maintained as brood stock, whereas heterozygous ones for aquaculture. At present, some clonal lines of above-mentioned species are reproduced and kept by the Fisheries Experiment Stations belonging to prefecture governments. Cloned fishes are important not only for aquaculture industries but also for basic biology. However, successful cloning of fish is substantially very difficult because the inhibition of first cleavage generally give rise to very poor survival due to technical and genetic reasons. Moreover, a few survivors with all-homozygous genotypes frequently show low fecundity as well as poor egg quality. However, meiotic (polar-body) gynogens are considered more practical to fix desirable traits, because they often show better survival and maturation than mitotic (homozygous) gynogens. "Super growth strain" rainbow trout was established by means of repeating meiotic gynogenesis and almost identical genotypes were reported. Thus, this method may be practically effective to fix genotypes when the survival and reproductive capacity of homozygous gynogens are very poor and cloning is nearly impossible.

Combination of induced gynogenesis or androgenesis with sex steroid treatments provide effective method to control sexes of aquaculture populations. Sex-reversed gynogens are predicted to be XX males. Thus, sperm of such XX males will generate all-female populations by cross-fertilization. All-male population can be achieved by fertilization with Y-sperm of androgenetic diploid super-males (YY). Temperature dependent sex determination (or differentiation) was found in hirame flounder. In such species, sex is easily controlled by environmental factors.

Recent development of hypervariable microsatellite and other polymorphic DNA markers makes it possible to tag individual fish genetically. Such a molecular tagging is highly useful not only for parentage analysis to evaluate the effect of stocking, but also for monitoring of genetic parameters in aquaculture and released populations. DNA markers are now actively mapped on chromosomes of some commercially important fishes, by means of

linkage analysis using reference families constructed by backcross of one homozygous strain to heterozygous strain. When some markers are tightly linked to commercially important quantitative trait loci (QTL), marker-assisted selection (MAS) can be realized in aquatic animals. Thus, integration of classic breeding techniques and DNA techniques will generate new breeding technique in aquaculture. Combination of chromosome manipulation and new biotechnologies such as transgenics, germ-line chimera, nuclear transfer and others will open new possibilities in breeding techniques. Induced gynogenesis or androgenesis is considered a method to fix transgenic gene to homozygous conditions. All-female sterile triploid can be used as a biological containment technique to minimize the risk of escape of genetically modified organisms (GMO). Germ-line chimera and nuclear transplantaion can be utilized for gene-banking. Systematic integrations of these modern biotechnologies with classic methods will produce fruitful results in genetic improvement of aquaculture species.