Development of Genetics and Breeding in Abalone Culture

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SUMMARY: Abalone is one of the most important mollusca species in global for aquaculture. Technology for artificial seed production and a rearing system has been developed and established for the species. Specialized culture strains are expected. Genetic differences among species and local races have been analyzed by genetic markers for the purpose of introducing a new culture species. Heritability for growth has been calculated from the data of mating experiments for selective breeding. Chromosome manipulation techniques have been developed for the fixation of traits. We discuss the present and future of abalone culturing.

KEY WORDS: abalone, genetics and breeding, marker genes, chromosome manipulation

There are more than 70 species of abalone inhabiting tropical to subarctic zones. Among these, eleven species inhabit Japan, where *Haliotis discus hannai* is one of the most important species to aquaculture. Technology for artificial seed production and a rearing system has been developed and established for this species,^{1,2)} and cultured strains are needed and expected. Before producing a culture strain, it is necessary to accumulate a lot of genetic information about the abalone.³⁾ Genetic differences among and within species have been analyzed by several genetic markers for the purpose of developing a new culture species. Heritability for growth has been calculated from the data of mating experiments designed for selective breeding. Chromosome manipulation techniques have been developed for fixation of the traits. Until now, however, we had not established any culture strain. Here, findings from previous studies on genetics and breeding for abalone are arranged into the following three sections: (1) Studies on the development of genetic markers and analysis of genetic population structure, (2) Genetic study of target quantitative traits for breeding, and (3) Study of chromosome manipulation for establishing a new strain. In addition, the future of abalone culture will be discussed.

Studies on the development of genetic markers and analysis of genetic population structure: Study of the genetic population structure began with a comparison by Fujino⁴⁾ of isozyme patterns between *H. discus discus* and *H. d. hannai*. Subsequently, Fujio et al.⁵⁾ evaluated the genetic variability of the Pacific abalone using isozyme genes.

For purpose of species differences in abalone,

isozyme analyses^{6,7)} have been used. Recently, DNA polymorphisms have been applied to genetic analyses. In 1995, species differences and relationships among 27 abalone species were analyzed by using sequences of lysin.⁸⁾ Microsatellite DNA polymorphism,⁹⁾ RAPD analysis,¹⁰⁾ and sequences of 18SrDNA¹¹⁾ have been used for making comparisons among abalone species. Based on the analysis of 18SrDNA sequences,¹¹⁾ among *Haliotis* species, *H. discus discus* and *H. d. hannai* are closely related.

Studies of genetic population structure within a species have been carried out in *Haliotis discus*, including *H. d. hannai*,^{5,12,13,14)} *H. rubra*,¹⁵⁾ *H. cracherodii*,¹⁶⁾ and *H. fulgens*,¹⁷⁾ by using isozyme marker genes. Recently, DNA polymorphisms have been developed as genetic markers,^{18,19,20,21,22,23,24,25,26)} though they have not yet been applied for genetic analyses. Park et al.²⁷⁾ investigated the relationships among four abalone species (*H. discus hannai* collected from Japan and China, *H. rufescens* from California, *H. rubra* from Australia, and *H. midae* from South Africa) by using several genetic markers (isozyme genes, RFLP at 18SrDNA, and RAPD, AFLP, and RFLP at the mtDNA COI region). These species are clearly distinguished geographically and morphologically. As a result, each genetic marker has specific characteristics. Isozyme genes and mtDNA are available for the detection of species and/or local populations. RFLP and AFLP are suitable markers for detection of individuals. Because RFLP of 18SrDNA did not detect genetic variation, this marker can be expected to be useful for analyzing higher taxa. Isozyme genes are good markers of chromosomes but another genetic markers having higher variability, such as

microsatellite DNA polymorphisms are needed in the future. For purposes of abalone culture, the characteristics of variation of several marker genes should be checked, and these markers subsequently used for suitable subjects. Development of genetic markers is necessary, and quite variable marker genes will be effective not only for population analysis but also for linkage analysis with economic traits of abalone.

Genetic study of target quantitative traits for breeding: Genetic study of the target traits for breeding of abalone was initiated by isozyme analysis, which suggested homozygote excess and inbreeding depression.^{28,29)} Growth-related traits have been analyzed in relation to isozymes in *H. discus hannai*^{5,30,31,32,33,34,35)} and *H. rufescens*,³⁶⁾ and findings suggested a change in genetic composition by artificial seed production and the possible existence of recessive deleterious genes. The trait of temperature tolerance has been investigated in the Pacific abalone collected from the natural population.^{13,37,38,39)} Other studies have clarified the genetic factors of target traits for breeding by conducting mating experiments and rearing experiments.^{40,41,42,43,44)} Kawahara et al.⁴⁴⁾ estimated the heritability of growth by several full-sib matings and quantitatively indicated the ability of effective selection. Regarding the traits of survival (viability), Hara and Sekino²⁵⁾ showed a clear relationship between mortality by withering syndrome and pedigree detected by microsatellite DNA polymorphisms. Kijima et al.⁴⁵⁾ discovered null alleles at *Pgm-1* and *Pgm-2* loci in the population of the Pacific abalone by pedigree analysis produced by 10 pair-mating experiments. Reviews of abalone culture from the point of view of genetics and breeding have been published.^{42,46,46,48)} However, few reports have clarified the genetic control of target traits. To that end, establishment of inbred lines (strains) is required. Using these lines and several genetic markers, genetic control of the traits will be detected. Among various techniques used for this purpose, chromosome manipulation is one of most promising.

Study of chromosome manipulation for establishing a new strain: Many reports on the production of gynogenetic diploid^{49,50,51,52,53,54)} and polyploid (triploid and tetraploid) have appeared.^{55,56,57,58,59,60,61,62)} In chromosome manipulation techniques, production of an artificial gynogenetic diploid (G2n) is considered vital to the production of new culture species and strains in abalone. However,

an effective procedure for G2n production has not yet been established. One approach is to decrease the fertilization rate by UV-irradiation of sperm, which destroys the acrosome and flagellum.^{51,52,53,54)}

It has been indicated that producing a sufficient number of G2n successively will be difficult. Androgenetic diploids that can be produced by UV-irradiated eggs and diploidization by protecting of 1st cell division are expected in the future. Chromosome manipulation techniques are also expected to be utilized for clarifying genetic control of the target traits by means of chromosome combinations such as haplotypic gene expression.

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