

Population Genetics for Management and Conservation of Aquatic Resources and DNA Fingerprinting in Fishes*

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Introduction

The river catfish *Mystus nemurus* is widely distributed in both mainland as well as archipelago Southeast Asia. Due to its good flesh quality it is a popular fresh water food fish throughout the region and is therefore of interest to aqua culturists. Not much genetic studies had been done on this fish so far. We typed five populations of river catfish from Peninsular Malaysia for 16 *loci* coding for nine enzymes and sarcoplasmic protein using horizontal polyacrylamide gel electrophoresis. These populations were from Kedah, Kedah, Banded, Perak, Serdang, Selangor, Rengam, Johor and Kuala Terengganu, Terengganu. Four polymorphic *loci* were found and the level of heterozygosities ranged from 0.132 in the Selangor population to 0.031 in the Terengganu population (Siraj *et al.*, 1998). Tilapia is an important aquaculture fish in Malaysia although it is an introduced African fish. Our breeding results showed that different strains of *Oreochromis niloticus* and crosses between them gave fishes with the best commercial demand. Therefore, it is necessary to develop molecular markers that can distinguish between strains of the same tilapia species.

Materials and Methods

River catfish samples from Perak, Kedah, Johor, Selangor and Sarawak were collected and brought back to our laboratory. Sample sizes ranged from 25 to 50 fish from each location. They were typed for DNA markers by using the Random Amplified Polymorphic DNA (RAPD) and the Amplified Fragment Length Polymorphism (AFLP) techniques. A RAPD based technique was used to identify microsatellite repeats in *Mystus nemurus*. RAPD bands generated from OP kit A were cloned and screened for micro satellites. Other

approaches such as Random Amplified Hybridisation Microsatellites (RAHM) and Direct Amplification of Length Polymorphisms (DALP) were also used to obtain codominant DNA markers for this species in the absence of DNA sequence data (Usmani *et al.*, 1999). In the case of tilapia, 8 strains of local tilapia being maintained by our related breeding programme were typed by using 25 microsatellite *loci* developed by Lee *et al.* and Kocher (1996).

Results and Discussion

Initially 40 RAPD primers and 64 AFLP primer pair combinations were tested. Of these, 9 RAPD primers and 4 AFLP primer pair combinations detected a total of 42 and 158 polymorphic markers respectively. These AFLP and RAPD polymorphic markers were used to type fish from Perak, Kedah, Johor, Sarawak and Selangor. As both RAPD and AFLP bands were inherited as dominance-recessive markers, the presence and absence of bands were used to estimate similarity indices. A correlation value of 0.69 between the RAPD and AFLP similarity matrices indicated that the data generated by both methods were not identical but showed agreement (Chong *et al.*, 2000). In order to get an idea of the genetic relationships among the populations based on RAPD and AFLP data respectively. An Unweighted Pair Group Method with Arithmetic Averaging (UPGMA) dendrogram was generated based on the similarity matrices for each of the DNA typing techniques used. In both dendrograms, the four populations from Peninsular Malaysia clustered together whereas the Sarawak population from Borneo Island clustered by itself. When the fishes were clustered on an individual basis, three subgroups each from Kedah, Perak and Sarawak populations were detected by

AFLP but not by RAPD. Unique AFLP fingerprints were also observed in some unusual genotypes sampled in Sarawak. This indicated that AFLP might be a more efficient marker system than RAPD for identifying genotypes within populations. In addition the Mendelian dominance-recessive mode of inheritance for 9 RAPD and 24 AFLP markers were confirmed through the use of family studies. These dominant markers are excellent for genetic characterisation of the populations and we can expect hybrid vigour in crosses between Sarawak and Peninsula fishes but they are not efficient for testing for possible associations between quantitative traits of economic importance such as growth rates, disease resistance and molecular markers. A RAPD based technique was used to identify microsatellite repeats in *Mystus nemurus*. A total of 40 plasmids have been sequenced out of which 13 were microsatellite repeats. Primers were designed for four of the microsatellites. Two primers were tested one was polymorphic while the other was monomorphic. For the studies of tilapia, the mean heterozygosities within populations ranged from 0.22 to 0.36 and UPGMA clustering based on Nei's genetic distances calculated from microsatellite *loci* allele frequencies showed that Taiwan A and B strains cluster together distinctly apart from the local black, local hybrid, Israel, Thai and Phillipines strains and from *O. mossambicus* (Bhassu *et al.*, 1998). Work is currently in progress to find correlations between the microsatellite markers and quantitative traits.

Conclusions

Genetic markers are useful for characterisation of the two species, which will help in choosing the right stocks for breeding programmes.

Benefits from the study

Knowledge as reported in scientific publications, which includes journal, conference and seminar papers as well as theses of undergraduate and post-graduate students.

Literature cited in the text

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Project Publications in Refereed Journals

- Chong, L.K., Tan, S.G., Yusoff, K., Siraj, S.S. 2000. Identification and characterization of Malaysian river catfish, *Mystus nemurus* (C & V): RAPD and AFLP analysis. *Biochem. Genetics*. 38: 63-75
- Chong, L.K., Tan, S.G., Siraj, S.S., Christianus, A. and Yusoff, K. 1999. Mendelian inheritance of random amplified polymorphic DNA (RAPD) markers in the river catfish. *Malaysian Applied Biology*. 28: 79-84.

Project Publications in Conference Proceedings

None.

Graduate Research

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