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GENETIC EROSION DUE TO FOUNDER EFFECT IN HATCHERY STOCK OF RED TILAPIA (*Oreochromis* sp.) IN SABAH

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ABSTRACT. *Samples of red tilapia (Oreochromis sp.) were collected from three hatcheries namely Kinarut (UMS), Likas and Babagon. These samples were measured for their weight and total length. White muscles were removed and subjected to allozyme electrophoresis. Three enzymes were selected for analysis, which include the phosphoglucomutase (PGM), glucose-6-phosphate isomerase (GPI) and isocitrate dehydrogenase (IDH). Five loci were scored of which three being polymorphic (GPI-2, GPI-3 and IDH) and the remaining two were monomorphic (GPI-1 and PGM). The genetic variability was calculated based on allele frequencies using computer software called BIOSYS-1. The number of allele per locus was low in all population being 1.6 in Babagon, 1.4 in Likas and 1.0 in Kinarut, respectively. The observed heterozygosity was similar in all population (H_0=0.00). The mean F_{IS} value (inbreeding coefficient), was also equal to one, indicating serious degree inbreeding. The length and weight was strongly correlated (R^2 >0.95). The growth exponents recorded in Kinarut was 3.0, and 2.1 in Likas and an interesting value was obtained from Babagon (6.8). This indicates that population in Babagon attains more weight per unit length. Whether or not the situation in Kinarut and Likas populations were due to inbreeding depression remain to be further investigated. This paper however suggests three different approaches in improving genetic variability in hatchery production of red tilapia. First is the use of sufficient number of brood fish, which must consist of equal numbers of female and male. Second is to frequently bring in new brood fish thereby contributing new set of alleles into the gene pool. The third one is to develop broodlines through rotational crossbreeding. These approaches can theoretically control a great number of problems in hatcheries associated with depreciation of genetic variability.*

KEYWORDS. Genetic erosion, red tilapia

INTRODUCTION

Aquaculture of red tilapia (*Oreochromis* sp.) is popular in Sabah. Stocks are reared either in earthen ponds, concrete tanks or high-density polyethylene (HDPE) tanks. The produce is mainly for local markets. Aquaculture of the fish is mainly achieved by controlled breeding using limited number of broodfish.

A survey of red tilapia farming practices in Sabah reveals a limited number of founding individuals are used for hatchery propagation, and many specimens of the same siblings are grown to maturity to serve as broodstock. The analyzed specimens were cultivated stock represents tenth generation of hatchery-propagated fish. Such situation can cause inbreeding depression, which does not compromise with the sustainable high yield. The stocks of tilapia in these hatcheries have been observed and some interesting findings were noticed in UMS stock. The stock has been observed to have size hierarchy among individuals of same age, growth depression and the occurrence of 1-2% of anatomical abnormality (Figure 1). The size hierarchy has also noticed in Likas and Babagon hatcheries, but no anatomical abnormalities were reported in those hatcheries.

Similar investigation by Ransangan *et al.* (1999) on sea bass in Sabah showed that the limited number of broodstock in the propagation of the species in hatchery has contributed to the erosion of genetic diversity as revealed by low number of alleles per locus and low level of heterozygosity. The fish also showed growth depression and physical abnormality such as bending at distal part of the caudal peduncle (Ransangan and Mustafa, 1999; Mustafa *et al.*, 2000).

Considering these factors, a genetic analysis of the red tilapia is necessary for a better understanding of the genetic variability and founder effect, if any, so that measures could be taken to genetically vitalize the stock in the interest of individual fitness, disease resistance and optimum production potential. This investigation of genetic heterozygosity using allozyme electrophoresis of hatchery stock of tilapia was conducted following unconfirmed reports on the low growth rate of the fish in Sabah. The data obtained might lead to the review of the contemporary aquaculture management and policies, and practices.

MATERIALS AND METHODS

Thirty samples of juveniles (average standard length: 7.6 cm± 1.15) which consist of equal numbers of male and female were collected from three freshwater hatcheries in Sabah, which include UMS hatchery at Kinarut, and two Fisheries Department hatcheries at Likas and Babagon. White muscle from the samples were removed and subjected to allozyme electrophoresis. Weight and length of every specimen was measured using analytical balance and caliper. The white muscle tissue from all specimens were homogenized in 0.1M Tris-HCL, pH 8.0 (Johannessan

et al., 1989), centrifuged at 1,500 rpm for 1 minutes and supernatant was subjected to starch gel electrophoresis according to method described by Ransangan *et al.* (1999). Slight modification has been made on the concentration of starch gel used (12% instead of 11%). Three enzymes namely *glucose-6-phosphate isomerase* (GPI), *isocitrate dehydrogenase* (IDH) and *phosphoglucomutase* (*PGM*) were studied for genetic variability. These enzymes were best resolved in TCE (Aebersold *et al.*, 1987). Genotype frequencies generated based on allozyme bands on starch gel after staining were analyzed using computer software called BIOYSIS-1 (Swofford and Selander, 1981) to obtain genetic parameters such as allele frequencies, number of allele per locus, polymorphic locus and degree of heterozygosity, and Nei's genetic distance. The fixation indices were calculated based on the observed and expected heterozygosities. The F_{IS} (Hardy-Weinberg distribution of genotypes of individuals within subpopulation), F_{IT} (Hardy-Weinberg distribution of individual genotypes within the total population) and F_{ST} (genetic differentiation of subpopulations within the total population). Positive values of these indices demonstrate excess of homozygotes or conversely, a deficiency of heterozygotes, relative to Hardy-Weinberg model. Correlation analysis between weight and standard length of specimens was analyzed using *Statistical Package for Social Science software* (SPSS). Growth exponent was calculated using formula described by Beakman (1948). Nomenclature of genotypes followed the scheme described by Allendorf and Utter (1979).

RESULTS

The study managed to score five loci of which three were polymorphic and the rest were monomorphic. The three polymorphic loci were *GPI-2, GPI-3* and *IDH*, while *GPI-1* and *PGM* were monomorphic. Genetic variability calculated based on allele frequencies are given in table 1.0. The mean number of alleles per locus was low but varied between populations, ranging from 1.0 in Kinarut to 1.4 and 1.6 in Likas and Babagon hatcheries, respectively. The mean observed heterozygosity in all populations was zero. Values of fixation indices calculated based on allele frequencies within and across populations were all positive (Table 1).

Correlation analysis of total body weight and total length in all populations showed a strong positive correlation (R^2>0.95). The length and weight relationship was calculated using formula $W=aL^n$ suggested by Beckman, (1948), where *n* is the growth exponent. The growth exponent was varied among populations with 2.1 in Likas, 3.0 in Kinarut and 6.8 in Babagon, respectively.

Table 1. Allele frequencies, genetic variability and fixation indices of red tilapia (*Oreochromis* sp.) hatchery stocks from Kinarut, Likas and Babagon analyzed using allozyme electrophoresis.

Locus	*Allele frequencies*				*Fixation indices for polymorphic locus*		
	Allele	*UMS (N=30)*	*Likas (N=30)*	*Babagon (N=30)*	F_{IS}	F_{IT}	F_{ST}
GPI-1	*-100*	*1.000*	*1.000*	*1.000*	-	-	-
GPI-2	*117*	*0.000*	*0.133*	*0.000*	*1.000*	*1.000*	*0.850*
	100	*1.000*	*0.867*	*0.600*			
	80	*0.000*	*0.000*	*0.400*			
GPI-3	*100*	*1.000*	*0.867*	*0.767*	*1.000*	*1.000*	*0.514*
	92	*0.000*	*0.133*	*0.233*			
IDH	*118*	*0.000*	*0.000*	*0.467*	*1.000*	*1.000*	*0.432*
	100	*1.000*	*1.000*	*0.533*			
PGM	*100*	*1.000*	*1.000*	*1.000*	-	-	-
Polymorphic locus (P)	-	*0%*	*40%*	*60%*	-	-	-
Number of allele per locus	-	*1.000±0.00*	*1.400±0.24*	*1.6±0.24*	-	-	-
Observed Heterozygosity (H_o)	-	*0.000±0.00*	*0.000±0.00*	*0.000±0.00*	-	-	-
Expected Heterozygosity (H_e)	-	*0.000±0.00*	*0.094±0.054*	*0.276±0.114*	-	-	-
Mean of fixation indices					*1.000*	*1.000*	*0.398*

DISCUSSIONS

The observed heterozygosity ($H_o=0$) of the three hatchery stocks of red tilapia revealed genetic erosion. Positive value of F_{IS} (also called inbreeding coefficient) would indicate that the analyzed stocks are highly inbred. The only possible reason why such observation on heterozygosity was the founder effect due to the fact that the two stocks were founded with limited number of brood fish (pers. com). This is because the species has high fecundity that enables it to produce large quantity of eggs with a limited number of brood fish. Such genetic erosion is commonly experienced in many hatcheries, which due mainly to two possible problems; the elevated cost and the difficulty in maintaining large number of brood fish in a limited space of most hatcheries.

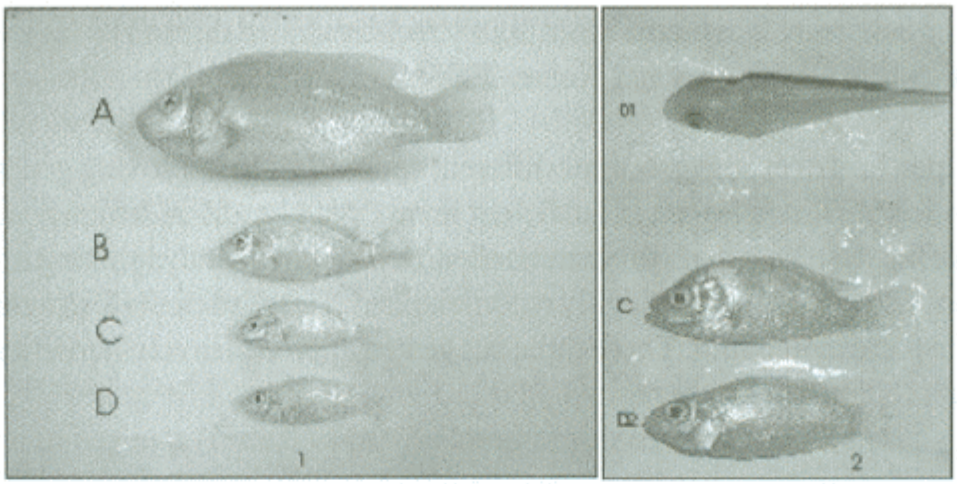

Figure 1. Picture 1 shows size hierarchy among fingerlings of the same age where A being the larger and C beings smallest in the stock while D is experienced a physical abnormality at the anterior part (the head is slightly twisted). The picture 2 shows the difference between abnormal fish (D1& D2) and the normal fish (C).

Genetic studies on hatchery stocks showed that loss of genetic diversity is common in salmoid fish (Allendorf and Phelps, 1980; Cross and King, 1983; Leary *et al.*, 1985; Verspoor, 1988), seabream in Japan (Taniguchi *et al.*, 1983; Suguma *et al.*, 1988), muscle (Gosling, 1982; Gaffney *et al.*, 1992; Hedgecock and Sly, 1990), shrimp (Sbordoni *et al.*, 1986) and Asian sea bass (Ransangan *et al.*, 1999).

Although there was strong relationship between weight and length of the red tilapia, the growth exponents are varied among populations. The higher growth exponent was recorded in Babagon population (6.8) followed by Kinarut (3.0) and Likas (2.1). These mean that the stock in Babagon attaining more weight per unit length as compared to the Likas stock. The situation in Babagon stock is that it was founded with large number of brood fish (>500). This is evident since the brood fish are cultured in earthen ponds of 10 by 5 meter. The hatchery has an equal number of males and females broodfish and is frequently adding new fish collected from different hatcheries (Gidius; pers.com). Different scenario is seen in Likas stock where the stock was cultured in HDPE tanks. There is no however history of broodfish was obtained from the UMS hatchery. As far as growth is concerned, many factors are involved in determining growth rate. The study was able to correlate the occurrence of growth depression and the anatomical abnormality in UMS stock with the low level of genetic diversity as given by the zero mean observed heterozygosity of the stock.

Selection program for brood fish is crucial in aquaculture to ensure a continuous production in hatchery. Homozygous individuals normally have decreased in growth rate and reduced resistance to diseases as compared to heterozygous individuals (Sbordoni *et al.*, 1987). As far as tilapia in Sabah is concerned, there is report on reduced growth rate of tilapia in Sabah. However, the

selection program for growth in tilapia is not highly recommended due to low level of heritability ($h^2=0.12$) of such trait (Beaumont and Hoare, 2003).

This paper, however suggests three different approaches in improving genetic variability of red tilapia in Sabah. First, the use of sufficient number of broodfish, which consists of equal numbers of male and female is highly recommended. Second, frequently bringing in new genes in the form of broodfish into the pool is highly recommended. The third one is to develop broodlines through rotational crossbreeding. These three suggested approaches can theoretically control a great number of problems in hatcheries.

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