

**HETEROISIS, DIRECT AND MATERNAL GENETIC EFFECTS ON BODY TRAITS IN A COMPLETE DIALLEL CROSS INVOLVING FOUR STRAINS OF RED TILAPIA *OREOCHROMIS SPP***

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**SUMMARY**

Heterosis, direct additive genetic and general reciprocal effects were estimated from a complete diallel cross involving four strains of red tilapia *Oreochromis spp* from Malaysia, Stirling, Taiwan and Thailand. The mating involved 16 parental female and male breeders per strain, producing 64 full sib families in total, with four full-sib families per cross. Statistical analyses were carried out on 1280 performance records collected in both fresh water (0 ppt) and saline water (30 ppt) environments. There was a large additive genetic component for body traits in the four strains of red tilapia. The Malaysian strain was the best (7.4% above the overall mean of the pure strains), whereas the strain from Stirling had the poorest additive performance (13.4% below the overall mean of the pure strains). The average heterosis for body weight across the testing environments was low (4.2%) and the average of all crossbreds was not statistically different from the mean of pure strains. Ranking of strains based on estimates of reciprocal effects was generally similar to that of additive genetic effects. Strategies for the future breeding program in red tilapia are discussed.

**INTRODUCTION**

Information on phenotypic and genetic parameters for red tilapia (*Oreochromis spp.*) is extremely limited. Despite the importance of the species in many Asian countries and the existence of a number of distinct populations (referred to as 'strains' here) there are no published results of strain comparisons or of purebreeding or crossing parameters. In order to gain some understanding of the genetic characteristics of red tilapia, we sampled strains from Malaysia, Stirling, Taiwan and Thailand, and evaluated them in a diallel cross design in both fresh and saline water environments. The main objective of this work was to evaluate relative performance of strains in order to form a synthetic base population for future genetic selection.

**MATERIALS AND METHODS**

**Origin of stocks.** This study included four strains of red tilapia: 1) Malaysian red tilapia obtained from a private hatchery in Malaysia (M), 2) Thai red tilapia *O. niloticus* × *O. mossambicus* from the National Inland Fisheries Institute (N), Department of Fisheries, Thailand, 3) red *O. niloticus* originating in Lake Manzala, Egypt (McAndrew *et al.* 1988), and obtained from Institute of Aquaculture, University of Stirling (S) in United Kingdom, and 4) *O. niloticus* × *O. mossambicus* red tilapia from a private hatchery in Taiwan (T). The exotic strains of red tilapia from Malaysia, Stirling and Taiwan were imported into Thailand in mid-2005. They were kept and reared at the Pathumthani Fisheries Test and Research Center, Aquatic Animal Genetics Research and Development Institute, Department of Fisheries, Pathumthani province, Thailand (latitude 14°N, longitude 100°E, 20 km north of Bangkok). A detailed description of the stocks origin is given in Pongthana *et al.* (2009).

**Family production and rearing.** The pair matings following a complete diallel cross design (Table 1) were conducted in separate 1×1×1 m breeding hapas in February 2006. Sixty four breeding hapas were installed in a pond. In each hapa, one male was mated to one female. A total of 64 full-sib families (four families per cross combination) were successfully produced. After approximately one or two weeks of mating, swim-up fry were collected separately from each hapa and transferred to 1×1×1 m rearing hapas at a stocking density of 500 fry per hapa, one hapa for each full sib group. The date of collection of swim-up fry was recorded. After 3-4 weeks in the rearing hapas, the fry were transferred to B-net hapas (~ 6 mm mesh size) at a stocking density of 300 fry per hapa (1×1×1 m) for further rearing until an average body weight of 5 grams. When the fingerlings reached this weight they were individually tagged using Passive Integrated Transponders (PIT). A total of 20 fingerlings were tagged per family, amounting to 1280 tagged individuals.

**Table 1. Diallel cross mating design involving four strains of red tilapia**

Female/ Male	M	N	S	T
M	MM	MN	MS	MT
N	NM	NN	NS	NT
S	SM	SN	SS	ST
T	TM	TN	TS	TT

**Testing environments.** After tagging, the fish were tested in both freshwater floating cages and saline water concrete tanks. The floating cages were located in a 60,000 m<sup>3</sup> water reservoir inside the Pathumthani Fisheries Test and Research Center. Six cages (4 × 4 × 1.5m) adjacent to each other were assembled, and 2 fish from each family were assigned at random to these cages. The initial stocking density was 5 pieces per m<sup>2</sup> of surface water. The fish were fed twice daily a commercial pellet feed with a dietary protein level of 32%, at the rate of 3-5% of their body weight. Contemporaneously with freshwater cage culture, siblings from the 64 families were tested in a 30 ppt of saltwater environment in four concrete tanks (4 × 10 × 1m). The stocking density was 5 fish per m<sup>2</sup>. The same feeding, culturing and management practices were applied as used for the cages in freshwater. In both environments, water quality parameters (temperature, pH, dissolved oxygen, alkalinity, total ammonia and saline level) were closely monitored once a week.

**Harvest and measurements.** Following a grow-out period of 126 days (range from 118 to 147 days), all fish were harvested and immediately transferred to large hapas for one to two days of conditioning without feeding before the individual identification, body measurements and sex were recorded. Over the growth period in cages and tanks 98% of the tested individuals retained their tags. Survival of the fish in both environments was high, averaging 84.3 per cent, calculated from the difference between the number of fish at stocking and at harvest. This trait was not included in the present analyses. Basic statistics of body weight at harvest is given in Table 2

**Table 2. Descriptive statistics for harvest weight (g)**

Environment	N	Mean	SD	CV
Fresh water	520	579.3	220.8	38.1
Saline water	529	429.2	160.6	37.4
Overall	1049	503.6	206.8	41.1

**Statistical analyses.** Model 1 was used to estimate additive and non-additive genetic components. The fixed effects included in the model were test environments (fresh and saline water), sex (female and male) and their two-way interactions. Age at harvest was fitted as a linear covariate. The genotype effects were partitioned in terms of direct additive, non-additive and reciprocal effects. The model also fitted full-sib group as the random term. The mathematical expression of the model is as follows:

$$y_{ij} = \mu + F + \sum \alpha_i a_i + \sum \alpha_{ij} h_{ij} + \sum \beta_i r_i + s + e_{ijk} \quad [1]$$

where  $F$  are the fixed effects and covariate as described above,  $\alpha_i$  is the proportion of genes contributed by the  $n^{\text{th}}$  individual originating from the  $i^{\text{th}}$  strain ( $\alpha_i = 0.0, 0.5$  or  $1.0$  and  $\sum \alpha_i = 1.0$ );  $a_i$  is the additive genetic effect of genes originating from the  $i^{\text{th}}$  strain;  $\alpha_{ij}$  is the coefficient of the total heterosis effect for the cross between the  $i^{\text{th}}$  and  $j^{\text{th}}$  strains ( $\alpha_{ij} = 0.0$  or  $1.0$ ;  $i \neq j$  and  $ij \neq ji$  and  $\sum \alpha_{ij} = 1.0$ );  $h_{ij}$  is the total heterosis effect for the cross between the  $i^{\text{th}}$  and  $j^{\text{th}}$  strains ( $i \neq j$  and  $ij \neq ji$ );  $\beta_i$  is the coefficient of the general reciprocal effect for the  $i^{\text{th}}$  strain ( $\beta_i = 0$  for purebreds and  $-0.5$  for male strain and  $0.5$  for female strain, for the crossbreds and  $\sum \beta_i = 1.0$ );  $r_i$  is the general reciprocal effect of the  $i^{\text{th}}$  strain;  $s$  is the random effect of full-sib group; and  $e_{ijk}$  is the random residual error for the  $l^{\text{th}}$  individual.

The relative importance of the additive, heterosis and reciprocal effects was assessed using the likelihood ratio test (LRT) by removing each term from the full model at a time (Table 3).

**Table 3. Increase in -2logL for harvest weight when one effect at a time was excluded from the model**

Effect	Fresh water	Saline water	Both
Additive genetic ( $a_i$ )	31.9***	30.9***	30.1***
Total heterosis ( $h_{ij}$ )	62.9***	60.3***	59.9***
General reciprocal ( $r_i$ )	41.4***	42.2***	29.0***

\*\*\*P<0.001

The additive, heterosis and reciprocal effects were estimated as regression coefficients with one degree of freedom. The additive genetic effects were restricted to  $\sum a_i = 0$ . The coefficients of the general reciprocal effects set in the present study assume that the additive genetic effects of a given strain are similar regardless of gender of parental breeders. Total heterosis for a cross between two strains was partitioned as  $h_{ij} = \bar{h} + h_i + h_j + s_{ij}$ , where  $\bar{h}$  is the average heterosis effect for all strains involved in the diallel cross,  $h_i$  and  $h_j$  are the general heterosis effects for the  $i^{\text{th}}$  and  $j^{\text{th}}$  stock, respectively, and  $s_{ij}$  is the specific heterosis effect of strains.

## RESULTS AND DISCUSSION

**Additive genetic effects.** Table 4 presents estimates of strain additive genetic effects on body weight. Across the testing environments, the Malaysian strain ranked highest, whereas the additive performance of the strain from Stirling was poorest (7.4 and 13.4% above and below the overall mean of the pure strains, respectively). Overall, ranking of genotypes changed between the environments.

**Heterosis.** The average heterosis for body weight across the testing environments was low (approximately 4.2%) and not statistically different from the mean of pure strains. The level of

average heterosis was lower in fresh than in saline water, but the difference between the two environments was only about 2% (Table 3). The heterotic effect reported on body traits is generally small in other farmed aquaculture species such as Nile tilapia (Bentsen *et al.* 1998) or Rohu carp (Gjerde *et al.* 2000).

**Reciprocal effects.** In this study reciprocal effects can be considered to be equivalent to maternal components. Across the environments, the Malaysian strain ranked highest, whereas the strain from Stirling was lowest. Furthermore, the general reciprocal effects also varied with testing environments.

**Table 4. Estimates of additive genetic effects and heterosis for body weight at harvest**

	Fresh water		Saline water		Across	
	Estimate	%	Estimate	%	Estimate	%
Mean	532.9± 23.8		395.9 ± 23.7		465.5 ± 20.2	
Additive genetic ( $a_i$ )						
M	83.6	15.7	-0.7	-0.2	34.5	7.4
N	-13.2	-2.5	27.0	6.8	9.3	2.0
S						-
T	-61.7	-11.6	-69.7	-17.7	-62.1	13.4
Maternal effect ( $r_i$ )						
M	-8.7	-1.6	43.5	11.0	18.4	3.9
N						
S	17.3	3.3	34.8	8.8	29.6	6.4
T	43.6	8.3	-7.2	-1.8	16.3	3.5
Average heterosis ( $\bar{h}$ )	-38.2	-7.3	-52.8	-13.3	-43.6	-9.4
	-22.7	-4.3	25.2	6.4	-2.3	-0.5
	15.5	<b>2.9</b>	19.8	<b>5.0</b>	19.5	<b>4.2</b>

## CONCLUSIONS

In contrast to the low level of heterosis for harvest weight, the large additive genetic component estimated in this study suggests that performance improvement of red tilapia could be effectively based on the exploitation of additive genetic variation (i.e. through selective breeding rather crossbreeding). This study also points to the relative performance of strains as a guide for the establishment of a synthetic base population for future genetic selection. The changes in both the additive and non-additive genetic performance of genotypes with testing environments in the present study also merits further examination to evaluate alternative breeding strategies for the future breeding program in red tilapia.

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