DIVERGENT SELECTION FOR GROWTH IN THE DEVELOPMENT OF A FEMALE LINE FOR THE PRODUCTION OF IMPROVED GENETICALLY MALE TILAPIA (GMT) IN *Oreochromis niloticus* L.

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Abstract

The culture of genetically male tilapia (GMT) is becoming established as an alternative to hormonal sex reversal in some locations such as in the Philippines. However, concerns exist over the growth performance of the strain of *Oreochromis niloticus* in which GMT have been developed and the consistency of sex ratios. As part of a project to develop improved GMT, a selection programme was initiated in a female line to be crossed with YY males. Three generations of divergent within family selection for 16-week weight were applied to a base population consisting of five strains of pure *O. niloticus* from diverse origins, with each base strain forming a family. Rotational mating was carried out between the families from the base strains. A high intensity of selection was applied with standardized selection differentials ranging from 1.6 to 2.6 per generation. Selection for combining ability for sex ratio was carried out in the selected females (ten heaviest females per family) in generation one and three. After three generations of selection, high line and low line selected males and females were used to produce progeny for growth performance trials in four culture environments to assess the response to selection. Growth differences between high and low lines in the four environments ranged from 37% (extensive ponds) to 102% (intensive tanks) indicating highly significant response to selection ($P < 0.01$). Estimates for realised heritability (h^2) ranged from -0.08 (high line males) to +0.40 (low line females). GMT was produced by crossing YY males from an Egyptian strain with females from the high line. The mean sex ratio of GMT using this selected female line was 97.4% male and preliminary data from growth trials indicate significant increases in growth rate of this improved GMT compared to the original inter-strain GMT.

Introduction

The desirability of culturing monosex male populations of tilapia has been long established as a means to control unwanted reproduction and to exploit the relatively faster growth of males. Monosex male populations can be produced in a number of ways, most

commonly by direct hormonal sex reversal through the oral application of androgens to sexually undifferentiated fry. The YY male technology was developed in response to certain constraints to the commercial scale application of hormonal sex reversal and exploits the genetic basis of sex determination in *Oreochromis niloticus*, which is considered to be predominantly monofactorial (Mair *et al.*, 1991). Mair *et al.*, (1997a) demonstrated the viability of a breeding programme for the mass production of genetically male tilapia (GMT) and growth trials revealed the superior growth rate, survival and yield of GMT compared to mixed sex and hormonally sex reversed tilapia (Mair *et al.*, 1995). The YY male technology has been adopted commercially by farmers in a number of countries, with widespread adoption in the Philippines. Application of the technology is through the provision of males from a true breeding YY male line (utilising sex reversed YY females) and females from a true breeding female line (utilising sex reversed XX males) within the same strain.

Whilst sex determination is considered predominantly monofactorial following the sex chromosome model with heterogametic XY males and homogametic XX females, other factors are known to influence sex ratio. Studies have demonstrated that environmental factors, notably temperature, can influence sex ratio (Abucay *et al.*, 1999, Baroiller *et al.*, 1996) but it is also likely that autosomal sex modifying genes play a role in producing sex ratios deviating from expectation (Mair *et al.*, 1991; Mair *et al.*, 1997a). Mean sex ratios of GMT in the strain in which it has been developed (known in the Philippines as Egypt-Swansea) range from 95-98% but individual family sex ratios can be highly variable. Sex ratios of crossbred GMT using YY males from the Egypt-Swansea strain with females from other strains produces more variable sex ratios with lower mean proportions of males (Mair *et al.*, 1997b and unpublished data). Thus an important consideration in the development of improved GMT is to minimize variability in sex ratio and maximize the proportion of males. Assuming a significant role of autosomal sex modifying genes in the occurrence of sex ratios deviating from expectations, it should be possible to select against such genes by selecting for the highest proportions of males in progeny of females crossed with YY males. Lester *et al.*, (1989) estimated heritability of sex ratio of 0.26 in experiments with a Philippine strain of Nile tilapia.

The YY male technology was originally researched and developed in a domesticated strain of *O. niloticus* originating from Lake Manzala in Egypt. Commercial scale production originated from a stock introduced into the Philippines from the U.K. This stock is likely to have passed through a number of genetic bottlenecks associated with the transfer and establishment of U.K. laboratory populations and with the development of the YY male and all-female broodstock lines. It is thus likely that inbreeding may be affecting the fitness of commercially produced intra-strain GMT and depressing growth rates. This hypothesis is supported by the superior growth rate exhibited by crossbred GMT using females from several different strains (Mair *et al.*, 1997b and unpublished data) indicating the presence of heterosis that might be expected if the parental lines were inbred. To counteract the potential problem of inbreeding and to develop GMT with superior growth performance, a selection programme was initiated in a synthetic line of tilapia with the ultimate objective of using this line in crosses with YY male to produce improved GMT.

The results from selection programmes in tilapia have been quite varied. An early attempt to apply mass selection for growth rate in an Israeli cultured strain (Hulata *et al.,* 1986) was not successful with no response detected after two generations compared to unselected controls. However, a significant but asymmetric response was detected in divergent selection for growth rate in *O. aureus* (Bondari *et al.*, 1983). Since these early experiments, there have been a number of successful selection programmes for tilapia, the most notable being ICLARM's GIFT project. Under this large-scale project, six generations of selection for growth performance were carried out on a synthetic base population created from eight distinct strains of Nile tilapia. Average response to selection was 13.2% per generation with estimated realised heritabilities ranging from 0.07 to 0.25 (Eknath and Acosta, 1998). This level of response and moderate heritabilities indicate that selection is well worthwhile. A number of other studies have selected tilapia for growth with varying levels of success. A within family selection programme has produced significant production gains over 13 generations with a cumulative response to selection of up to 68% by generation 10 (Bolivar and Newkirk (2000). Uraiwan and Doyle (1986) reviewed data related to different methods of selection for tilapia in Thailand and concluded that within family selection was likely to be more appropriate for small to medium scale selection programmes in the region.

Based on past results from previously conducted selection programmes and on the assumption that adequate genetic variation exists in the five base strains used to form the selected lines in our study, it was decided to practice within family selection for growth rate with a high selection intensity. Long term concerns over accumulation of inbreeding were not of primary consideration due to the fact that the strain was ultimately being selected for a crossbreeding programme (i.e. for combining ability in crosses with YY males). To maximise the prospect of detecting response to selection in the shortest time and to best utilise limited resources (in terms of facilities and manpower) it was further decided to practice only divergent selection, without an unselected control, which would have been difficult to maintain over the medium to long term without bias. This paper reports the preliminary results from the evaluation of growth performance following three generations of divergent selection for growth rate.

Materials and methods

The strains were selected as the base for this selection programme on the basis of comparative growth performance trials under mixed sex and monosex male environments (Capili, 1995). The strains selected predominantly based on their relative ranking for relative growth performance in communally stocked trials of mixed sex tilapia.

The base strains

The strains, described using nomenclature adopted in the Philippines to label strains of tilapia being held in live gene banks at the Freshwater Aquaculture Center (FAC), of the Central Luzon State University on which this selection programme was based were chosen based on results from comprehensive growth trials conducted by Capili (1995). The following five strains were used in the first generation of rotational mating:

Egypt-ICLARM (1): This strain was collected by ICLARM as one of the base strains for the development of the GIFT fish with 30 individuals being collected from three different locations in Egypt (Eknath *et al.*, 1993). The fish obtained by FAC were the result of a single pair mating in this strain. Growth trials of multiple strains of tilapia showed this strain to have superior growth rates to the majority of wild caught and domesticated strains of diverse origin.

Egypt-AIT (2): This strain originated in Egypt and was transferred to Thailand via Japan. Stock (50 individuals) was transferred to the Philippines from the Asian Institute of Technology in 1991. Common with most strains of Egyptian origin, this strain showed superior growth rates in communally stocked comparisons of 11 strains of tilapia (Capili, 1995).

Kenya-Turkana (3): This strain originated from Crater Lake A in Lake Turkana, Kenya and with high levels of genetic variation, may represent a sub-species *O. n vulcani* (Macaranas *et al.*, 1993). Twenty fish were transferred to the University of Wales Swansea in 1991 and a pooled spawning of these fish provided fry for transfer to the Philippines. Whilst the relative growth performance of this strain compared to others in a mixed sex environment was poor we felt that the high levels of genetic variation seen from allozyme studies (Macaranas *et al.*, 1993) indicated this would be a good base strain for a selection programme. Furthermore (Capili, 1995) demonstrated that Kenyan strains have high levels of sexual dimorphism and have relatively superior performance in all male environments

Kenya-Baringo (4): This strain originates from Lake Baringo in Kenya and is claimed by (Trewavas, 1983) to represent another sub-species *O. n. baringoensis*. It was first collected in 1975 with a supplementary collection in 1982 with stock transferred to Baobab farm in Kenya. Samples were transferred from there to the University of Wales Swansea in 1987 with progeny from a pooled spawning of several males and females transferred to the Philippines in 1991. The rationale for choosing this strain is similar to those stated for Kenya-Turkana.

BFAR strain (5): This strain is considered to be a genetically variable stock compared to other strains existing in the Philippines although its genetic composition is not documented. The strain is one that the Philippine Bureau of Fisheries and Aquatic Resources disseminates to government institutions and the private sector under a national breeding programme developed in collaboration with ICLARM and is derived from the GIFT tilapia.

The rotational mating scheme

The breeding programme was initiated by first selecting for combining ability for sex ratio. Twenty females were randomly selected from stock of each strain and stocked for progeny testing with YY males of the Egypt-Swansea strain. The female from each strain that produced the highest proportion of males in these progeny tests (100% male in all cases) were selected. These females were then crossed with randomly selected males from one of the other strains (male from Strain 1 with female from Strain 2 etc.) in the first round of rotational mating to produce the first families grown on within family selection. In the second generation the male from Family 1 were crossed with female from Family 3 and

similarly for all the families. The selected male from Family 1 was crossed with selected female from Family 4 in the third generation.

The selection procedure

Two selected lines were developed from the progeny resulting from the first rotational mating, a high line and a low line selecting for the fastest and slowest growth rate respectively, with five families in each line. With each pair mating of selected males and females 300 fry (newly swimming fry) from each pair in each strain/family were randomly selected for on growing. These 300 fry were reared in 150 cm diameter concrete tanks until they attained an average weight of 0.5 g. At this stage the number was reduced to 100 by selecting the individual fish with weights closest to the mean (collimation). The remaining 100 fingerlings were then on grown for a further 16 weeks in the same tank. Tanks were used for the grow-out phases due to the logistical and resource constraints of growing on numerous families in alternative environments such as earthen ponds, which might have been preferable environments, being more representative of typical aquaculture environments in the Philippines. During the grow-out period the fish (beginning at fry stage) were fed *ad libitum* with commercially available tilapia diets, starting with fry mash and later with crumble and pellet form. After 16 weeks, all the fish in each group were individually weighed and the biggest six males and ten females in each group in high line and the smallest six males and ten females in the low line were selected. All the selected fish were then PIT tagged for identification.

At the first selection (i.e. after grow out of five inter-strain crosses) in concrete tanks, the separate high and low lines were created. Within each family the biggest and smallest males and females were selected to constitute the high line and low line, respectively. After this point two lines, with five families per line, were maintained separately following the same selection procedure outlined above. Initial attempts were made to keep the production of each selected generation of each family and of each line synchronized. However, this did not prove possible due to the different times of spawning of the individual selected females in each line and also due to the much slower growth rates in the low line which resulted in delayed sexual maturation in the selected females. As a result the two lines came out of synchrony by up to six months. Families within each line were produced within a period of two months.

Realised heritability was estimated based on regression of response against cumulative selection differential according to Falconer and Mackay, (1996).

Selection for sex ratio (in progeny tests with YY males) was carried out in the selection of females for the initial crosses, as described above. Sex ratio selection was also carried out in the combination with the first and third generation of within family selection for growth in the high line only. After selecting the biggest six males and ten females, the selected females were progeny tested by crossing to YY males to determine which females produce the highest percent males in progeny (with sample sizes in excess of 50). It was not feasible to select for sex ratio in the low line due to shortage of time. It was also not of commercial importance to select for sex ratio in this line as it served only as a control. As a

result, the females selected in the high line were not necessarily the heaviest reducing slightly the selection differential in this line compared to the low line.

It was also not feasible to select males in the high line families for sex ratio due to the shortage of XY or YY females in the Egypt-Swansea strain that would be required for the progeny testing. No selection for sex ratio was carried out in the second generation to hasten the selection for growth rate.

Growth evaluations after three generations of selection

After three generations of selection for growth rate and two generations of selection for sex ratio, growth evaluation trials were carried out to determine the cumulative response to selection. The ten biggest females and six biggest males in each family in the high line and the ten lightest females and six lightest males fish in each family in the low line (i.e. the preliminary fourth generation selected fish) were pooled, within lines, to produce progeny for growth evaluations using rotational mating between the families. Similar aged progeny from these crosses were pooled to provide the mixed sex fish for growth trials. These fish were nursed separately up to the age and size (approximately 5g) at which they could be marked by clipping of one of the paired fins (pectoral or pelvic). After marking, the fish were stocked in four different on-station environments at FAC as follows:

Earthen ponds (200 m²): High and low selected fish were stocked communally with the "original" intra strain GMT stocked as an internal reference control. Fish were stocked at three fish per m². Ponds were fertilized with organic and inorganic fertilizers for the first two months and were fed at 3% of biomass daily, with a commercially available pelleted diet up to harvest. Three replicate ponds were stocked.

Circular concrete tanks (1.5 m diameter): This represented the environment in which the fish were selected. Three replicates tanks were stocked for the high and low line fish with 60 fingerlings stocked per tank. Fish were fed twice a day with standard rates of commercially available tilapia feeds based on 5% biomass of fish in each tank. All the fish were sampled every two weeks to adjust feeding rates. Continuous flow of clean water was maintained at a rate of approximately one liter per minute.

Cages in pond (4 m²): Three replicates cages were stocked for the high and low line fish with 60 fingerlings stocked per cage. Fish were fed twice a day with standard rates of commercially available tilapia feeds based on 5% biomass of fish in each cage. All the fish were sampled every two weeks to adjust feeding rates.

Intensive circular tank (8 m diameter): This represented an intensive culture system with high line, low line and "original" intra-strain GMT stocked communally in a single 8 m diameter concrete tank with continuous water flow and aeration. A total of 6,000 fingerlings (plus 10% allowance for mortality) were stocked per tank with 1,839 high line, 2,006 low line and 2,919 GMT. The minor differences in stocking number were due to limited availability of genotypes of some fingerlings. The stocked fish were fed *ad libitum* with a commercially available floating pellet.

Analysis of variance on harvest weight was used to determine growth differences between strains within each environment. Chi-square contingency tests were used to compare sex ratios.

A similar set of trials, which will be reported in full elsewhere (Abucay and Mair, in preparation), were established to compare the growth performance of GMT produced by crossing females from the selected high line with YY males from the Egypt-Swansea strain with alternative GMT genotypes.

Results

Selection for sex ratio

Table 1 summarises the data from progeny testing of females in the base strains, first and third generation selected families in the high line. Sex ratios, with average sample sizes of 46-80 fish, adequate for estimating sex ratios, were surprisingly high in the progeny testing of females from the base strains, averaging 97.6% male. Mean sex ratios were lower at 92.2% in the first generation (weight) selected females but higher again in the third generation (96.9% male).

Table 1. Summary of data on which selection for sex ratio was based in the base (females from the five strains), first generation and third generation weight selected females. Sex ratios are derived from crosses of females (from ten females selected based on weight in first and third generations) with YY males of the Egypt-Swansea strain.

	Base population	**First generation**	**Third generation**
No. of females tested	76	29	39
No. of fry tested	4,639	1,443	6,366
Mean no (± s.d.)	61.04 ± 28.88	46.3 ± 16.8	81.60 ± 27.9
Mean % male ± s.d.	97.59 ± 5.46	92.21 ± 13.56	96.93 ± 4.76
Minimum	75.00	55.56	81.42

No data are presently available for sex ratios produced by crosses of low line females with YY males, which would serve as a control to determine the response to selection for sex ratio.

Selection for growth rate

Table 2 summarises the parameters used in the selection for growth in the five families in the high and low lines. In general the selection intensities for the males (mean of 2.23 and 1.64 in the high and low lines respectively) were higher than for the females (1.93 and 1.42 in high and low lines). This differential in the high line can be explained to some extent by the selection for sex ratio that took place in the females in the first and third generation, in which case the selected females was not necessarily the largest in the family.

Selection indices for the low line were also lower than those for the high line reflecting the greater extremes of weight on the heavy side of the weight distribution.

Estimates of heritability based on regression of response on cumulative selection differential were -0.08 and 0.21 for the high line males and females and 0.30 and 0.40 for the low line males and females, respectively.

Table 2. Mean (from five families) weight, selection differentials and selection indices (standardized selection differentials) for three generations of high and low within family selection for 16-week growth in Nile tilapia.

Line/ Generation	Sex	Mean weight in 5 families (g)	Mean weight of selected fish (g)	Selection differential	Selection intensity
High Line	Male	55.68	85.74	30.06	1.93
Generation 1	Female	34.74	48.66	11.11	1.59
High Line	Male	54.23	76.66	2.43	2.42
Generation 2	Female	40.15	59.52	9.37	2.60
High Line	Male	51.41	78.55	7.14	2.33
Generation 3	Female	41.89	55.87	3.99	1.61
Low Line	Male	55.68	24.16	-1.52	2.01
Generation 1	Female	34.71	20.20	-4.54	1.72
Low Line	Male	43.46	16.92	-6.54	1.72
Generation 2	Female	25.96	12.10	-3.86	1.49
Low Line	Male	38.21	23.10	-5.11	1.20
Generation 3	Female	23.30	14.55	-8.74	1.06

Growth evaluation trials

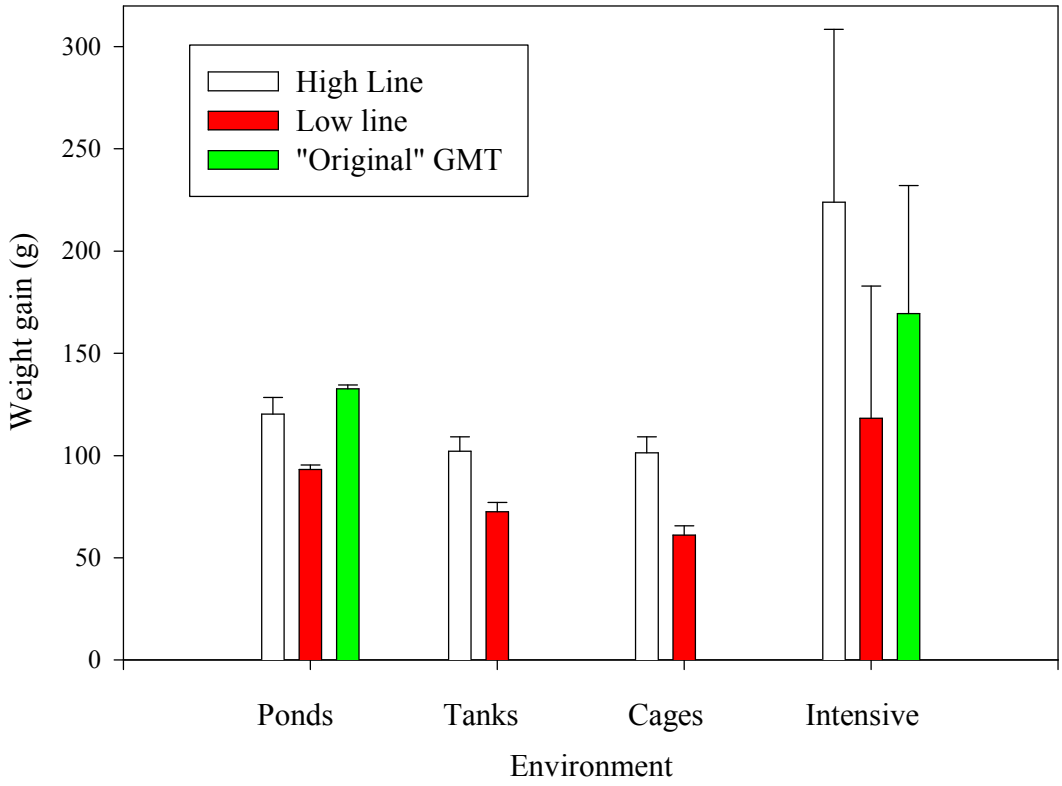
The growth evaluation trials revealed highly significant differences between the high and low selected lines in all the environments. The results from the culture performance trials are summarised in Table 3. The difference in the growth rates of high and low line fish (expressed as a % of the low line weight gain) was 37% in extensive ponds, 41% in concrete tanks, 66% in cages- in-ponds and 102% in intensive tanks, all of these differences being highly significant ($P<0.01$). The relative differences in weight between the high and low lines was greater for the females than for the males reflecting the higher levels of heritability for growth in females. In the two environments in which the "original" GMT were added as internal reference controls, there were significant differences in initial weight (GMT were smaller) which may have impacted on the relative growth of these fish in communal stocking due to size based interactions between the genotypes. In the extensively managed earthen ponds the mean weight of GMT was not significantly different to the selected high line despite the significantly higher proportion of males (80% compared to only 35%). In the case of the intensively managed tanks, the high line had significantly greater weight gain

than the GMT with similar differences in sex ratio. There were no significant differences in the survival of the genotypes during grow-out in any of the environments. An alternative rough method for calculation of realised heritability based on standardized selection responses seen in the growth trials and standardized cumulative selection differentials produced estimates of h^2 ranging from 0.06 to 0.11 in the high line and 0.06 to 0.2 in the low line with higher levels estimates for female growth.

Table 3. Summary of data from a series of culture performance trials comparing the high and low selected lines (mixed sex), following three generations of divergent selection for growth rate, in four different culture environments (values are given as means +/- standard deviation; P represents levels of probability at which statistical tests were significant; ns – not significant).

Culture environment	Parameters	High line	Low line	"Original" GMT
Earthen pond	Area of pond	200 m^2	200 m^2	200 m^2
(Communal	Stocking number (fish)	600/pond	600/pond	600/pond
rearing using	Mean initial wt. P<0.01	4.80 ± 0.40 [a]	5.17 ± 0.45 [a]	3.60 ± 0.10 [b]
three ponds)	Mean wt. gain (F&M) P<0.001	120.16 ± 8.26 [a]	93.25 ± 2.15 [b]	132.65 ± 1.87 [a]
	Mean % survival ns	77.60 ± 3.22	77.50 ± 5.00	73.00 ± 5.77
	Mean % male P<0.001	35.00 ± 3.19 [a]	47.65 ± 2.09 [b]	80.22 ± 2.98 [c]
	Mean wt. gain (F) P<0.01	103.57 ± 7.67 [a]	66.56 ± 3.54 [c]	82.04 ± 4.98 [b]
	Mean wt. gain (M) P<0.01	151.27 ± 8.07 [a]	122.60 ± 3.52 [b]	145.04 ± 1.24 [a]
Small circular	Volume of tank	1 m^3	1 m^3	
Tank	Stocking number	60 fish/tank	60 fish/tank	
(Separate	Mean initial wt. ns	6.43 ± 0.06	5.50 ± 0.76	
rearing with	Mean wt. gain (F&M) P<0.01	102.10 ± 7.06 [a]	72.56 ± 4.48 [b]	
three	Mean % survival ns	87.78 ± 4.20	80.00 ± 6.01	
replicates)	Mean % male ns	44.29 ± 4.13	45.29 ± 5.39	
	Mean wt. gain (F) P<0.01	83.37 ± 7.16 [a]	54.76 ± 3.26 [b]	
	Mean wt. gain (M) P<0.01	127.03 ± 3.58 [a]	94.30 ± 5.18 [b]	
Cage-in	Size of cage	2m x 2m x 1m	2m x 2m x 1m	
-Pond	Stocking number	60 fish/cage	60 fish/cage	
(Separate	Mean initial wt. ns	6.32 ± 0.11	6.03 ± 0.28	
rearing with	Mean wt. gain (F&M) P<0.01	101.22 ± 7.99 [a]	61.14 ± 4.33 [b]	
three	Mean % survival ns	88.33 ± 1.67	81.11 ± 10.05	
replicates)	Mean % male ns	39.04 ± 7.86	42.74 ± 6.86	
	Mean wt. gain (F) P<0.001	94.78 ± 2.68 [a]	53.40 ± 5.24 [b]	
	Mean wt. gain (M) P<0.05	111.85 ± 16.95 [a]	71.70 ± 3.19 [b]	
Intensive	Volume of tank	60 m^3	60 m^3	60 m^3
Circular tank	Stocking number (line)	6764 (1839)	6764 (2006)	6764 (2919)
(Communal	Mean initial wt. wt P<0.001	3.91 ± 1.33 [a]	3.19 ± 1.55 [b]	1.32 ± 0.73 [c]
rearing using	Mean wt. gain (F&M) P<0.001	223.99 ± 84.55 [a]	118.21 ± 64.62 [c]	169.38 ± 62.60 [b]
one tank	Mean % survival ns	86.19	68.35	72.80
only)	Mean % male ns	39.50	55.36	87.39
	Mean wt. gain (F) P<0.001	194.73 ± 62.29 [a]	88.58 ± 45.16 [b]	130.17 ± 49.97 [c]
	Mean wt. gain (M) P<0.001	268.99 ± 93.98 [a]	142.01 ± 68.02 [b]	175.06 ± 62.22 [c]

Figure 1. Grouped bar chart showing relative weight gains of high line, low line and "original" GMT in culture performance trials, in four different culture environments, following three generations of divergent selection for growth.



Discussion and conclusions

The data reported indicate relative success for this breeding programme, combining within family selection for growth and selection for combining ability (with YY males) for sex ratio. Highly significant deviations in growth performance were observed between the high and low selected lines after three generations of selection, reflecting the high intensity of selection. Selection response and estimates of realised heritability were similar to those observed by other authors in comparable programmes (Jarimopas, 1990; Brzeski and Doyle, 1996). Furthermore, estimates of realised heritability were within the ranges observed in the comprehensive studies conducted under GIFT project, which also utilised a genetically variable "synthetic" base population comprising several strains (Eknath and Acosta, 1998).

However, from an applied perspective, the lower heritability estimates in the high line compared to the low line is less encouraging. The lower heritability estimates for the high line reflects the higher selection differentials in this line due to the longer tails on the heavy side of weight distributions and may not necessarily reflect a significantly lower response to up selection. Asymmetric response to divergent selection is common, as observed in channel

catfish by Bondari (1986), for numerous reasons including natural selection, scale-effects, genetic asymmetry and genes with large effects (Falconer and Mackay, 1996).

Further, the apparent greater response to selection in the females than in males provides less promise for the use of the selected line as a female line in a crossbreeding programme for GMT production. Nevertheless, preliminary results of crossbred GMT using the selected high line as the female parent, crossed to YY males of the Egypt-Swansea strain, show considerable increases in growth rates compared to the "original" intra-strain GMT (Abucay and Mair, unpublished data).

GMT sex ratio produced by the high line females in crosses with YY males (97.6% male) were comparable with sex ratios expected in intra strain crosses using the same YY males (Mair *et al.*, 1997a). Such proportions of males would be adequate for control of reproduction in most circumstances of commercial tilapia culture.

In conclusion, this intensive selection programme produced significant positive response to selection for growth rate, and possibly also for combining ability for GMT sex ratio. From a practical perspective it would appear to be worth continuing with this programme, both for production of faster growth mixed sex fish, and particularly, as was originally intended, as a female line for GMT. With a longer term perspective, significant loss of genetic variation and inbreeding is likely to occur after further rounds of rotational mating, with high selection intensity, whereby only a single male and a single female are selected in each family. It is thus recommended that the programme be modified through inclusion of more families and the selection and breeding of more males and females per family. Provision of a suitable control population, such as a heterozygous clone produced using isogenic gynogenetic lines, would facilitate this as the low line could be discontinued, freeing up resources for expansion of the high line. It is further recommended that selection of combining ability for GMT sex ratio be carried out but not necessarily every generation.

References

Abucay, J.S., Mair, G.C., Skibinski, D.O.F. & Beardmore, J.A. 1999. Environmental sex determination: the effect of temperature and salinity on sex ratio in *Oreochromis niloticus L. Aquaculture,* 173: 219-234.

Baroiller, J.F., Fostier, A., Caulty, A., Rognon, X. & Jalabert, B. 1996. Effects of high rearing temperatures on the sex ratio of progeny from sex reversed males of *Oreochromis niloticus.* pp. 246-256. *In*: Pullin, R.S.V*., et al.* (eds.). *The Third International Symposium on Tilapia in Aquaculture* (), International Center for Living Aquatic Resources Management (ICLARM), Centre de recherches océanologiques (CRO). Institut francais de recherches scientifique pour le développment en coopération (ORSTOM). Centre de coopération internationale en recherche agronomique pour le developpment/ Département d'élevage et de médecine vétérinaire (CIRAD-EMVT), Manila, Philippines.

Bolivar, R.B. & Newkirk, G.F. 2000. Response to selection for body weight of Nile tilapia (*Oreochromis niloticus*) in different culture environments. *In*: Fitzsimmons, K.,

Filho, J.C. (Eds.), Proceedings from the Fifth International Symposium on Tilapia Aquaculture. American Tilapia Association and ICLARM, pp. 12-23.

Bondari, K. 1986. Response of channel catfish to multi-factor and divergent selection of economic traits. *Aquaculture,* 57: 163-170.

Bondari, K., Dunham, R.A., Smitherman, R.O., Joyce, J.A. & Castillo, S. 1983. Response to bidirectional selection for body weight in blue tilapia. *In:* Fishelson, L. & Yaron, Z. (eds.), *Proceedings of the International Symposium on Tilapia in Aquaculture*, Tel Aviv University, Tel Aviv, Israel. pp. 302-312

Brzeski, V.J. & Doyle, R.W. 1996. A test of an on-farm selection procedure for tilapia growth in Indonesia. pp. 219-230. *In* Doyle, R.W.*, et al.* eds. *Genetics in Aquaculture V*, Elsevier Science, Amsterdam, Netherlands.

Capili, J.B. 1995. Growth and Sex Determination in the Nile Tilapia, *Oreochromis niloticus* (L.). Ph.D. Thesis, University of Wales Swansea, U.K. 271p.

Eknath, A.E. and Acosta, B.O. 1998. Genetic Improvement of Farmed Tilapia Project - Final Report (1988-1997). International Center for Living Aquatic Resources Management, Manila, Philippines. 75p.

Eknath, A.E., Tayamen, M.M., Palada-deVera, M.S., Danting, J.C., Reyes, R.A., Dionisio, E.E., Capili, J.B., Bolivar, H.L., Abella, T.A., Circa, A.V., Bentsen, H.B., Gjerde, B., Gjedrem, T. & Pullin, R.S.V. 1993. Genetic improvement of farmed tilapias: the growth performance of eight strains of *Oreochromis niloticus* tested in different farm environments. *Aquaculture,* 111: 171-188.

Falconer, D.S. & Mackay, T.F.C. 1996. *Introduction to Quantitative Genetics,* Longman, England. 464p.

Hulata, G., Wohlfarth, G.W. & Halevy, A. 1986. Mass selection for growth rate in the Nile tilapia (*Oreochromis niloticus*). *Aquaculture,* 57: 177-184.

Jarimopas, P. 1990. Realized response of Thai red tilapia to 5 generation of size-specific selection for growth. pp. 519-522. *In:* Hirano, R. & Hanyu, I. (eds.). *The Second Asian Fisheries Forum (Proceedings of The Second Asian Fisheries Forum,Tokyo,Japan,17-22 April 1989)*.

Lester, L.J., Lawson, K.S., Abella, T.A. & Palada, M.S. 1989. Estimated heritability of sex ratio and sexual dimorphism in tilapia. *Aquaculture and Fisheries Management,* 20: 369-380.

Macaranas, J.M., Eknath, A.E., Agustin, L.Q., Velasco, R.R., Ablan, M.C.A. & Pullin, R.S.V. 1993. Genetic improvement of farmed tilapia: documentation and genetic characterization of strains. pp. 296. *In:* Gall, G.A.E. & Chen, H. (eds.). *Genetics in Aquaculture IV, Proceedings of the Fourth International Symposium on Genetics in Aquaculture*. Elsevier Science Publishers B. V., Amsterdam, Netherlands.

Mair, G.C., Abucay, J.S., Beardmore, J.A. & Skibinski, D.O.F. 1995. Growth performance trials of genetically male tilapia (GMT) derived from YY - males in *Oreochromis niloticus* L.: On station comparisons with mixed sex and sex reversed male populations. *Aquaculture,* 137: 313-322.

Mair, G.C., Abucay, J.S., Skibinski, D.O.F., Abella, T.A. & Beardmore, J.A. 1997a. Genetic manipulation of sex ratio for the large-scale production of all-male tilapia, *Oreochromis niloticus*. *Canadian Journal of Fisheries and Aquatic Science,* 54: 396-404.

Mair, G.C., Scott, A.G., Penman, D.J., Beardmore, J.A. & Skibinski, D.O.F. 1991 Sex determination in the genus *Oreochromis* 1. Sex reversal, gynogenesis and triploidy in *O. niloticus* (L). *Theoretical and Applied Genetics,* 82: 144-152.

Mair, G.C., Capili, J.B., Skibinski, D.O.F., Beardmore, J.A., Pascual, L.P., Abucay, J.S., Danting, J.C. & Reyes, R.A. 1997b. Sex ratios and growth performance of crossbred genetically male tilapia using YY males in *Oreochromis niloticus* L. pp. 262 - 271. *In*: Fitzsimmons, K. (ed). *Tilapia Aquaculture. Proceedings from the Fourth International Symposium on Tilapia in Aquaculture*). Vol. 1, Northeast Regional Agricultural Engineering Service (NRAES), New York.

Trewavas, E. 1983. *Tilapiine fishes of the genera Sarotherodon, Oreochromis and Danakilia,* Trustees of the British Museum, London. 583 p.

Uraiwan, S. & Doyle, R.W. 1986. Replicate variance and the choice of selection procedures for tilapia (*Oreochromis niloticus*) stock improvement in Thailand. *Aquaculture,* 57: 93-98.