



# Genotype by environment interaction for harvest weight, growth rate and shape between monosex and mixed sex Nile tilapia (*Oreochromis niloticus*)



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## ABSTRACT

In Kenya, Nile tilapia (*Oreochromis niloticus*) is mostly grown in ponds. To avoid excessive reproduction and stunted growth, fingerlings are treated with methyl-testosterone to make all-male populations (monosex). For a national breeding programme that aims to provide genetically improved broodstock to hatcheries that supply monosex fry to smallholder pond farmers, it is important to assess the genetic correlation ( $r_g$ ) for traits between the mixed sex breeding candidates from the breeding nucleus and monosex production fish. The purpose of the study was to estimate genetic parameters for harvest weight (HW), daily growth coefficient (DGC) and body shape and investigate genotype by environment interaction ( $G \times E$ ) for these traits between mixed sex and monosex populations. Forty-eight sires and 76 dams from the  $F_2$  generation of a local *O. niloticus* strain, kept at Sagana Aquaculture Research Station, Kenya were used to produce 76 full-sib families. Mixed sex fry (3 days old) from each full sib family were divided into two groups of 50 individuals each. One group (monosex) was fed a diet treated with methyl-testosterone to induce sex reversal while the other group (mixed sex) was reared on a control diet. After hapa rearing, tagging and weighing, fish were randomly divided and stocked in six earthen ponds, three for mixed sex and three for monosex fish. After 5 months, fish were harvested, photographed and weighed. Genetic parameter estimates for HW, DGC, and shape were obtained on 2105 fish. Heritability estimates for HW, DGC and shape were  $0.21 \pm 0.03$ ,  $0.26 \pm 0.04$  and  $0.12 \pm 0.03$  for mixed sex respectively. Genetic correlations for HW between monosex and mixed sex was  $0.74 \pm 0.14$ , suggesting low  $G \times E$ . The corresponding  $r_g$  for DGC and shape were lower;  $0.59 \pm 0.10$ , and  $-0.19 \pm 0.11$ , respectively, denoting the presence of  $G \times E$ . It is concluded that  $G \times E$  between the mixed sex nucleus and monosex production fish is important, and that a breeding programme for Nile tilapia needs to include production performance from monosex siblings.

### Statement of relevance

First study that reports estimates for Genotype by treatment interaction between hormone treated monosex Nile tilapia and mixed sex Nile tilapia, and discusses the consequences for nucleus breeding programmes.

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## 1. Introduction

Worldwide, Nile tilapia (*Oreochromis niloticus*), and its hybrids are the most cultivated and widely farmed fish species, ranking second only after carps (Charo-Karisa et al., 2006). In 2013, the production of Nile tilapia was estimated to be over 4.5 million tons (World Bank, 2013). Culture of tilapia primarily takes place in cages and ponds (Tsadik and Bart, 2007). A major concern for tilapia pond culture systems is the reduction of growth rate at the onset of sexual maturity and excessive reproduction leading to overpopulation (Damien et al.,

2003). Monosex tilapia culture that constitutes males only can be employed to control reproductive activity, to attenuate growth inhibiting effects of interactions between the sexes and to increase production because males grow faster than the female in these species (Pham et al., 1998; Phelps and Popma, 2000; Damien et al., 2003). As a consequence of these effects, the economic feed conversions are generally more favourable in monosex male populations (Lone and Ridha, 1993).

Selective breeding remains the main driving force for development of resource efficient, sustainable and increased productivity in any live-stock species. For most breeding programmes, selection of breeding candidates takes place in a well-controlled “nucleus” environment whereas the rearing of production individuals takes place in more

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heterogeneous environments. Genotype by environment interaction ( $G \times E$ ) can occur due to differences in the sensitivity of individuals to the nucleus and production environments. In the presence of  $G \times E$ , re-ranking of individuals and heterogeneity of genetic variance makes genetic improvement obtained by selecting individuals in one environment to differ with other environments (Lynch and Walsh, 1998; Kolmodin et al., 2002; Sae-Lim et al., 2014).

The magnitude and importance of  $G \times E$  in aquaculture vary depending on the production environments. In Nile tilapia, most  $G \times E$  studies have focussed on comparison of production systems such as ponds and cages, using harvest weight as the trait of interest (Khaw et al., 2009; Bentsen et al., 2012). Results so far show that  $G \times E$  is probably not biologically important as indicated by relatively high genetic correlations ranging from 0.73 to 0.99 (Eknath et al., 2007; Thodesen et al., 2011; Trong et al., 2013a, 2013b; Khaw, 2015). Surprisingly, there are no estimates of genetic correlations between monosex and mixed sex Nile tilapia.

In Kenya, Nile tilapia farming is practised by small scale farmers and is largely characterized by low inputs and diverse farming conditions in terms of income level and market objective (Omasaki et al., accepted for publication). The predominant production system is earthen ponds using monosex male tilapia. For a national breeding programme that aims to provide genetically improved Nile tilapia brood-stock to hatcheries that supply monosex fry to smallholder pond farmers, genetic parameters for monosex and mixed sex are needed. The purpose of this study therefore, was first, to estimate the genetic parameters for harvest weight, daily growth coefficient and body shape traits in the mixed sex nucleus, and secondly; to investigate the magnitude of genotype by environment interaction between mixed sex and monosex populations for these traits.

## 2. Materials and methods

### 2.1. The location of the breeding programme

Selective breeding programme for *O. niloticus* in Kenya is conducted at Sagana Aquaculture Research Station (National Breeding Centre for Kenya Freshwater Aquaculture). It was initiated in 2011 from a base population of locally available strains. Selection target is to improve harvest weight in low-input production ponds.

### 2.2. Selection of breeding candidates

Fifty males and 150 females from the 2nd generation were selected to become parents. Selection was based on their estimated breeding values for harvest weight. The selected individuals were conditioned in hapas ( $4 \times 2 \times 1$  m) separately by sex for one month. They were fed twice a day on a Kenyan local floating pelleted feed with 26% crude protein, at a feeding rate of 3% of body weight.

### 2.3. Production of fingerlings

The production of fingerlings took place from 22nd November 2013 to 28th February 2014. An earthen pond ( $1800 \text{ m}^2$ ) equipped with fifty breeding hapas ( $2 \times 1.5 \times 1$  m) was used for fry production. Three female fish were placed with one male fish in each hapa. Inspection of the breeding hapas was conducted after every five days to collect swim up fry and fertilized eggs. Every time a female fish spawned, it was immediately removed from the breeding hapa. Collected fertilized eggs were then taken to the hatchery and incubated until hatching. After hatching, fry was transferred to hapas ( $1 \times 1 \times 1$  m). Eggs and fry that died were removed from the hapas on a daily basis. In total, 76 full-sib families from 48 sires and 76 dams were produced.

### 2.4. Rearing of fingerlings and tagging

A hundred individuals per family were collected from three day old mixed sex juveniles of Nile tilapia and divided in two equal groups of 50 individuals. Each group was placed in a hapa ( $1 \times 1 \times 1$  m, mesh size 1 mm) suspended in a  $1800 \text{ m}^2$  earthen pond. One group was fed for 30 days with a diet (Skretting: 35% CP, 9% fat) containing 60 mg  $17\alpha$  methyltestosterone per kg feed ( $17\alpha$  MT; Sigma-Aldrich company, Netherlands) to induce sex reversal; the other group was reared on a control diet (Skretting: 35% CP, 9% fat).

After a rearing period of 3 months, 20 fingerlings, randomly chosen from each hapa-family, were anaesthetized using tricaine methane sulphate (MS222) at a concentration of 100 mg/L, weighed and tagged using Passive Integrated Transponder (PIT) tags (Pocket RFID readers chips, Dorset Identification, Netherland). All full sib individuals of the same family and treatment group were tagged at the same time and returned to the hapas before stocking. Due to differences in egg collection dates, fingerlings were tagged when they were 94–177 days (8.85–73.56 g) old. In total 3014 fingerlings were tagged (Table 1).

### 2.5. Grow-out and pond management

Before stocking, tagged fingerlings from each family were removed from the hapas and scanned using an electronic scanner to check the presence of the tags. Body weights and total body lengths were taken and recorded for all fingerlings before stocking. Each full sib treatment group of fingerlings was then randomly divided into three groups. Each group was assigned to one of three  $150 \text{ m}^2$  ponds. For monosex fingerlings, 3 ponds A, B and C were used while ponds D, E and F were used for mixed sex fingerlings. In total, a range of 454–524 fish were stocked in each pond at an average stocking density of 3 fish per  $\text{m}^2$ . All the grow-out earthen ponds used were located at Sagana Aquaculture Research Station. During the grow-out period, water temperature stayed relatively constant at 25–27 °C in all the ponds. Fish that died within the first week of stocking were replaced with newly tagged individuals from the same family. Fish were fed twice a day on a commercial floating pelleted feed (Skretting: 35% CP, 9% fat) following the recommendations of the manufacturer.

### 2.6. Records

Fish were harvested after approximately 5 months of growth, using the seine net. They were first sedated using tricaine methane sulphate (MS222) at a concentration of 100 mg/L. Sex was assigned by visually examining the urogenital papilla of the fish. Harvest body weight (HW) in grammes and digital pictures (lateral view) were taken and recorded as follows: each fish was scanned using an electronic scanner and put in a measuring board placed underneath a camera, which was suspended at a fixed height above the fish. A piece of paper was then placed on the fish with a unique number, identifying the pond (A, B, C, D, E, F) and the fish (1–524) correspondingly. ImageJ software (1.47 for windows) (Rasband, 2008) was used to measure

**Table 1**

Stocking age, number of fish at stocking and harvest, sex ratio at harvest and mean survival (%) at harvest for both monosex and mixed sex fish.

Treatment	Pond	Stocking age	Number stocked	Number harvested	% of males	Survival (%)
Mono-sex	A	97–180	454	350	94.0	77.1
	B		525	428	94.5	81.5
	C		524	404	95.3	77.1
Mixed sex	D	103–186	456	283	48.8	62.1
	E		531	347	46.9	65.4
	F		524	293	47.1	55.9

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