Genetic parameters for reproductive traits in female Nile tilapia (*Oreochromis niloticus*): II. Fecundity and fertility

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ABSTRACT

Harvest weight is the main trait in Nile tilapia (*Oreochromis niloticus*) breeding programmes. The effects of selection for harvest weight on female reproductive traits are unknown. In this paper we estimate genetic parameters for reproductive traits and their correlation with harvest weight using females from the generation 12 of the GIFT strain of Nile tilapia in the Mekong Delta of Viet Nam. Spawning records were obtained from single pair mating as well as group mating experiments. The traits were categorised into two groups: fecundity-related traits and fertility-related traits. Fecundity traits were: number of eggs (NEGG), relative fecundity as the ratio of number of eggs to female spawning weight (RFEC), egg weight (EGGW) and egg diameter (EGGD); fertility traits were: number of fertilised eggs (FEGG), number of hatched eggs (HAT), number of swim-up fry (SWUP), and fertilisation rate (FER, in %). Heritability estimates for fecundity traits were low, ranging from 0.05 to 0.08. Heritability estimates for fertility traits were also low, ranging from 0.06 to 0.12. Genetic correlations for HW with NEGG and TEGGW were positive (0.51 and 0.42, respectively), while correlations for HW with RFEC, EGGW, and EGGD were negative (−0.72, −0.48, and −0.50, respectively). The same trend was observed for body weight at spawning (SPW), but genetic correlations between SPW and fecundity traits were higher than those between HW and fecundity traits. Genetic correlations between HW and fertility traits were all moderate to high (0.46 to 0.69), except for FER (0.15 ± 0.24). Genetic correlations between SPW and fertility traits were even higher (0.69 to 0.93). We conclude that both HW and SPW have favourable genetic correlations with NEGG, RFEC, and SWUP, which are the desired characteristics for Nile tilapia seed production. Selection for HW does not affect these traits. However, Nile tilapia females selected for large HW tend to produce smaller eggs. We recommend monitoring the phenotypic and/or genetic trend in this trait, as smaller eggs might, on the longer term, lead to lower fry survival.

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1. Introduction

The Genetic Improvement of Farmed Tilapia (GIFT) project has been conducted for ten years (1988–1997) to realise genetic improvement in Nile tilapia (*Oreochromis niloticus*) (Bentsen et al., 2012; Gjedrem, 2012). Thereafter, the Nile tilapia strain of GIFT origin has been further selected, mainly by the WorldFish Center, until now (Ponzoni et al., 2011). Harvest weight has been the main trait of interest, with genetic gains ranging from 10 to 15% per generation over six generations (Ponzoni et al., 2011; Thodesen et al., 2011). In addition to harvest weight, genetic parameters of other traits have been studied in different strains of GIFT origin, including body dimension (Nguyen et al., 2007), fillet yield (Nguyen et al., 2010; Thodesen et al., 2012), flesh composition (Ponzoni et al., 2011), and shape (Trọng et al., 2013). In other

Nile tilapia strains, genetic parameters of body dimension, gut length, visceral weight/index, and low-temperature tolerance were studied by Charo-Karisa et al. (2005, 2007), and fillet yield by Rutten et al. (2005). Recently, genetic parameters for harvest weight, pond survival and cold water tolerance of blue tilapia (*Oreochromis aureus*) have been reported by Thodesen et al. (2013).

Reproductive traits in fish are usually expressed as fecundity-related traits: number of eggs, egg weight, egg diameter, and egg volume (Gjerde, 1986). Genetic parameters for fecundity-related traits in fish have been studied mainly in salmonid species. In coho salmon (*Oncorhyncus kisutch*), heritability and genetic correlation between number of eggs, egg weight, spawn days, harvest weight, female prespawning and post-spawning weight were reported by Gall and Neira (2004). In wild brown trout (*Salmo trutta*), descriptive statistics of fecundity (expressed as number of eggs in the ovary), and egg diameter were reported by L'Abee-Lund and Hindar (1990) and by Hao and Chen (2009). In a random mating population of rainbow trout (*Oncorhynchus mykiss*), heritability estimate was 0.15 for female post-spawning weight, 0.32 for egg number and 0.28 for egg size (Gall and Huang,

1988). In selected populations of rainbow trout, heritability estimate was low (0.14) for spawning weight, but medium to high (0.52–0.65) for spawning date, egg size, number of eggs, egg volume, and for fertility-hatchability (Su et al., 1997).

There are few studies on reproductive traits in Nile tilapia. Phenotypically, fecundity (calculated as the ratio of number of eggs to weight of ovary) varied more with body length (11–32 cm) ($r = 0.860$) and body weight (15–600 g) ($r = 0.806$) than with age (1–3 years) ($r = 0.604$) (Babiker and Ibrahim, 1979). Genetic parameters for gonadosomatic index (GSI) in Nile tilapia (mean harvest weight 70.0–104.4 g) were estimated by Charo-Karisa et al. (2007). Heritability of GSI was 0.25 for females and 0.03 for males. The authors also estimated genetic correlation between harvest weight and GSI which was 0.27 for females and 0.01 for males. The GIFT strain has been selected for harvest weight, first at ICLARM in the Philippines (Bentsen et al., 2012) and thereafter at the WorldFish Center in Penang, Malaysia (Ponzoni et al., 2011). However, to our knowledge, the genetics of female reproductive traits in GIFT Nile tilapia have not been studied.

In this paper we investigate the effect of selection for harvest weight on reproductive traits for female Nile tilapia. The traits are categorised into two groups: fecundity-related traits (fecundity traits) and fertility-related traits (fertility traits). We first report heritability for fecundity traits: number of eggs, relative fecundity as the ratio of number of eggs to female spawning weight, egg weight, total weight of eggs per female, and egg diameter. Fertility traits are the number of fertilised eggs, number of hatched eggs (hatchling), number of swim-up fry, and fertilisation rate. Secondly, we report the genetic and phenotypic correlations between the two groups of reproductive traits and body weight at harvest and at spawning.

2. Materials and methods

2.1. Broodstock

Details for the source and rearing of the G12 broodstock are provided in the accompanying paper (Trọng et al., 2013b). Briefly, broodstock were from the Research Institute for Aquaculture No. 2 (RIA2) strain, which originated from the GIFT generation 10 strain obtained from the WorldFish Center, Penang, Malaysia (Ponzoni et al., 2010). The RIA2 GIFT strain has been selected, using the GIFT technology, for improved harvest weight since 2007 to date (Trọng et al., 2013a). Broodstock were randomly selected females from generation 12 (G12), except for females in the FAM experiment which had been selected on breeding value for harvest weight.

2.2. Experiments

Fecundity and fertility records were available from three experiments that differed in the mating design, namely 'Family' (FAM), 'Multiple males, multiple females' (MM) and 'Single male, multiple females' (SM). Briefly, MM was a group mating design with groups of 7 males and 15 females in each broodstock tank. The SM mating design consisted of groups of one male with 10 females in each tank. Details of MM and SM experiments are described in the accompanying paper (Trọng et al., 2013b). The aim of FAM experiment was to produce families for generation 13. The design followed the GIFT protocol described by the WorldFish Center (WorldFish Center, 2004). In this experiment, one female and one male were stocked into a $2.0 \times 1.5 \times 1.0$ m spawning hapa (mesh size 1 mm). After the female spawned, the same male was mated with a second female to produce a second half-sib family. Checks for spawning were conducted 4 days after stocking, and again at 8 days. If the female did not spawn, the pair was replaced. The experiment was conducted from September 2010 through January 2011. In total, 114 records of spawns were collected from 104 unique females that successfully mated with 50 males.

2.3. Data collection

2.3.1. Tagging weight, harvest weight and spawning weigh

A detailed description of data collection (tagging weight and harvest weight) is given in the accompanying paper. Females from the FAM experiment were treated in the same way as females used for the MM and SM experiments. The age of the fish at the start of the experiments age ranged from 359 to 705 (MM and SM-1), 620–855 (FAM), and from 765 to 1001 (SM-2) days (Table 1). In total, 804 records were collected for harvest weight (HW) and 1181 records (including 377 repeated records) were collected for SPW. Body weight of spawned females (SPW) was recorded at the time of egg/fry collection. The spawning age (SA) was calculated from the date that the female was collected (from the mother's mouth) until the date she spawned.

2.3.2. Fecundity traits

At collection, for each batch of eggs, the stage of eggs or fry (STAGE) was identified by eye and was cross-checked later on by back-counting from the day that the eggs hatched. Egg stages were identified as 'egg-1' (1 day old), 'egg-2' (2 days old), and 'egg-3' (3 days old). In some cases, yolk sac fry were collected from the females. These were counted and recorded as 'fry' (NFRY). The total number of eggs per female (NEGG) or fry (NFRY) was counted for each spawn. Relative fecundity per female (RFEC) was calculated as $\frac{\text{NEGG}}{\text{SPW}}$.

Eggs were incubated in 0.5 litre plastic down-welling bottles with a constant flow-through of water. For each spawn, a sample of thirty eggs was measured, and the mean diameter for these thirty eggs was used in calculations. Total weight of eggs per female (TEGGW, in g) was calculated as $\frac{\text{EGGW}}{30} \times \text{NEGG}$, with EGGW being the weight of thirty eggs (in g). Egg diameter was measured for each single egg under microscope (Olympus SZX7) with $25\times$ magnification (EGGD, in mm).

2.3.3. Fertility traits

Total number of fertilised eggs per female (FEGG) was counted at 'egg-3' stage and fertilisation rate (FER, %) was calculated as $100\times \frac{\text{FEGG}}{\text{NEGG}}$. The numbers of newly hatched fry (HAT) were counted shortly after all eggs hatched. Hatchlings were then transferred to $30 \times 40 \times 5$ cm plastic trays and number of swim-up fry (SWUP) were counted after all fry had their yolk-sac completely absorbed.

2.4. Statistical analysis

Genetic parameters were estimated using ASReml version 3 (Gilmour et al., 2009) as described in the accompanying paper. Here, only records of spawned females are used ($N = 1181$). For harvest weight (HW), the following mixed animal model was used:

$$Y_{ijk} = \mu + \text{CL}_i + \beta_1 \text{TW}_{ijk} + \beta_2 \text{GT}_{ijk} + \beta_3 \text{GT}^2_{ijk} + \text{ANIMAL}_j + \text{DAM}_k + e_{ijk}, \quad (1)$$

where Y_{ijk} is the phenotypic value of HW for the kth fish; μ is the population mean; CL_i is the fixed effect of the ith collection date for the fish, calculated as the number of dates from January 1st 2008 until the date that the fish (egg/fry) was collected from the mother's mouth ($i = 1,\ldots, 23$ collection days); β_1 is the regression coefficient of the co-variable tagging weight, TW_{ijk}; β_2 is the regression coefficient of the co-variable grow-out time, GT_{ijk}; β_3 is the regression coefficient of the co-variable grow-out time squared, GT^2_{ijk}; ANIMAL_j is the random effect of the jth fish with $N(0, A\sigma_a^2)$ where $\mathbf{A}$ is the additive genetic relationship matrix among the animals and σ_a^2 is the additive genetic variance; DAM_k is the random environmental effect common to full-sibs with $N(0, I\sigma_c^2)$; e_{ijk} is the random residual term with $N(0, I\sigma_e^2)$ where $\mathbf{I}$ is the identity matrix and σ_e^2 is the residual variance.

For spawning weight (SPW), the following model was used:

$$Y_{ijk} = \mu + \text{CL}_i + \beta_1 \text{SA}_{ijk} + \beta_2 \text{SA}^2_{ijk} + \text{ANIMAL}_j + \text{PE}_j \pm \text{DAM}_k + e_{ijk}, \quad (2)$$