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Towards diversification of aquaculture in Ghana: Reproductive

response of Sarotherodon *melanotheron* to locally formulated fish feed

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Research

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ABSTRACT

Sarotherodon melanotheron has a great potential for culture in Ghana. Among these potentials is the ability of the species to dwell and reproduce under captivity. This research aims to assess the effect of different feed types on the reproductive of *S. melanotheron* under culture. The feed types were formulated to contain fishmeal, soya bean cake as well as combination of fishmeal and soya bean cake as sources of protein. Some reproductive response parameters such as fecundity, oocyte diameter, spawning frequency of *S. melanotheron* fed on local formulated feed types were evaluated against Raanan fish feed, a common commercial fish feed commonly used in Ghana, for a period of 180 days. Absolute fecundity ranged from 238 to 315 whiles relative fecundity ranged from 14.79 to 17.00. Egg size and spawning frequency ranged from 1.6 to 2.9 mm and 26.50 to 30.5 days respectively among the various feed treatments. Fecundity, egg size and spawning frequency were not significantly different (p > 0.05) among the different feed types. Based on the findings of this current research, we conclude that *S. melonatheron* broodfish can be maintained under suitable culture condition with on-farm locally formulated feeds fortified with the appropriate additives to cut down cost of production.

Key words: Sarotherodon melanotheron, reproductive performance, fecundity

The black-chinned tilapia, *Sarotherodon melanotheron*, is a fish species reported to be endemic and abundant along the coast of West Africa (Teugels and Falk 2000). In Ghana, it forms the major composition of all fish species caught in coastal lagoons and estuaries (Aheto, Okyere, Asare, Dzakpasu, & Wemegah, 2014), and dwells in fresh and sea waters (37 psu) (Gueye, Kantoussan & Mbaye, 2013). *S. melanotheron* has attracted some level of research interest due their great potential for both coastal and in-land aquaculture in Ghana (Anani, Ofori-Danson & Abban 2010).

Fish reproduction is an important quality of a fish that cannot be overlooked towards their commercial aquaculture production, since it is the only means to ensure continuous production. It is influenced by both environmental and nutritional factors: protein, lipids, carbohydrates, vitamins especially A, C and E and minerals (Quintero *et al.*, 2009). Brood stock nutrition is of paramount importance for better reproductive performance of farmed fish, as it has been reported to impact on fecundity, survival, egg size, larval quality and spawning frequency (Manissery *et al.*, 2001). With the increasing advocacy for commercial culture of *S. melanotheron*, studies on broodstock nutrition are essential.

Fish feed is reported to account for about 60-70% of the running expenses involved in operating an aquaculture enterprise (Obirikorang et al., 2015). High cost of fish feed is mostly attributed to fishmeal which is in short supply due to dwindling of wild capture fisheries, and competition for use by other animal production sectors. Broodfish requires high protein feed, which further increase the cost of feeding. Other alternative source of protein such as soya bean cake have been reported to generally yield good result when partially or completely replace fish meal in fish feed (Koumi, Atsé, Otchoumou, & Kouam, 2008). This study therefore sought to investigate how locally available protein ingredient such as soya bean can be explored to enhance the reproductive capacity of S. melanotheron broodfish. This can help to cut down production cost of production of S. melanotheron in Ghana.

2. MATERIALS AND METHODS

2.1. Study duration and site

The study was conducted over a period of 180 days on a fish farming facility located on a reservoir at Okyereko near Winneba in the Central Region of Ghana

2.2. Selection/ Procurement of ingredients

Fishmeal, soya bean cake, wheat bran, additives such as salt, vitamin A/D premix, broiler premix, methionine, lysine and palm oil were purchased from Ashaiman timber market in the Greater Accra Region of Ghana. These ingredients were selected based on their readily availability in Ghana. The ingredients were transported to University of Cape Coast and processed into feeds for use during the experiments.

2.3. Biochemical analyses of feed ingredients

The bio-chemical composition of the wheat bran, fishmeal and soya bean cake were determined following the protocol by Association of Official Analytical Chemists [AOAC] (1990). Percentage (%) dry matter (DM), (%) Crude protein (CP), (%) Ash, % Crude lipids (CL) also known as the Ether Extract (EE) of fat, % Crude fibre (CF) and % moisture were determined. Nitrogen-free extract (NFE) was computed using the formula: % NFE = (100 - % CP + % CF +% EE + % Ash). The experimental feeds were formulated based on the percentage crude protein composition of the ingredients. After formulation, the biochemical composition of the formulated experiment feeds as well as the control commercial feed were determined to ensure they meet the nutritional requirement of the fish under study.

2.4. Feed preparation and formulation

The experimental feeds were formulated using Pearson's square method (Pillay, 1990). Consistent with previous study by Lim and Webster (2006), the formulations were done to contain 40 % crude protein. The feeds were formulated using different protein sources (plant source and animal source). Fishmeal and soya bean cake were used as the sources of protein to formulate three distinct types of feed: feed type 1 contained only fishmeal, feed type 2 had soya bean cake, and feed type 3 had (50-50 %) combination of fishmeal and soya bean cake as the sources of protein. Wheat bran was a common ingredient to all the feed types.

The ingredients were sieved to remove larger bones and stones which might cause pellet instability and affect the milling process. Top pan balance was used to measure the various ingredients in weight according to the proportion determined using the Pearson's square method. The ingredients were mixed in a large basin and the following additives: methionine, lysine, vitamin A/D premix, broiler premix added each consisting 0.5 % of the feed based on manufactures prescription. Salt (0.2 %) and palm oil (4.5 %) were also added. The pellets of different sizes based on development stages of the fish were then formed using a meat miner, sundried, bagged and store in a cool and dry place for use throughout the experiment period.

2.5. Experimental set-up

Sixteen (16) fine mesh hapas (size 1 mm) with each capacity of $(1m \times 1m \times 1m)$ were submerged in Okyereko dam on a cage platform with one quarter (1/4) above the water surface to prevent the fish from escaping.

Fecundity and egg size

Conditioned brood fish (average weight 3 ± 98 g) were stocked at a density of 10 female fish per metre square for the individuals used for fecundity and egg size determination. The individuals used for the spawning frequency were stocked at 2 fish per metre square at the sex ratio 1 male to 1 female. For each of the experiment, four treatments with triplicate were randomized. After stocking, the fish were allowed to acclimatize for two days before the feed trial began. The fish were reared till maturity and sampling was conducted biweekly to check whether there was a gonadal development based on the bulging of the belly early morning before they were fed. Individuals with very large bulge from each treatment were measured for length, weight using a digital balance (model: Tanita KD 200 and finally dissected to remove the gonad. Females with ripe ovaries were sorted after opening the abdomen. Ripe ovaries preservation and counting were done following the method by

Bagenal & Braum, (1978). Ripe ovaries were preserved in 10 % formalin for five days to harden the ova so that they could easily be separated for counting. Prior to counting of the eggs, the formalin was decanted, and the eggs were rinsed in water and weighed. The clumps of eggs were gently teased with a dissecting needle and all eggs counted to determine the fecundity of the fish. The whole count method was used due to the small number of eggs in the ovary.

Egg diameter was estimated following Shinkafi & Ipinjolu (2012). This was achieved using dissecting microscope. Twenty randomly selected eggs per female were measured along the two axes using a calibrated eyepiece micrometer under the microscope. The egg was viewed through the eyepiece. The stage on which the egg was mounted was adjusted till one side of the egg touched the circumference of the circular view under the eyepiece and then the reading on the scale was taken. Later, the egg was moved out of the circumference of the circular view and immediately the reading on the scale was again taken. The difference between the two readings gave the diameter of the fish egg. For every egg, the measurement was taken on both sides of the egg because fish egg is not round but oval.

Spawning frequency

With regards to spawning frequency, twelve males and twelve females were used for this aspect of the experiment. Two were paired in each separate hapas at a sex ratio of one male to one female for the spawning frequency experiment. Checks were done on weekly basis to see whether they had spawned or not. The days on which eggs were found in the mouth of the male were noted and the eggs taken from their mouth. Differences between the spawning days were noted to determine the spawning frequency.

2.6. Feeding regime

The fish were fed every day by hand-casting at 0800 GMT, 1000 GMT, 1200 GMT, 1400 GMT and 1600 GMT. They were fed at 10 % of their body weight throughout the experiment.

2.7. Physico-chemical parameters

Dissolved Oxygen, Temperature, Turbidity, Salinity and pH were monitored throughout the experimental period.

2.8. Analysis of experimental data

To compare the impact of the various feed types on the reproduction values of the fish, the data collected under the various feeding experiments were subjected to one-way analysis of variance (ANOVA) using minitab. The critical p- value was taken to be 0.05. Whenever a significant difference was found in any set of data, the Turkey post hoc test was used to determine which pairs of means were significantly different.

3. RESULTS

3.1. Bio-Chemical composition of feed ingredients

The proximate result of the ingredients, the percentage composition of the ingredients in the feeds as well as the proximate composition of the formulated feed are expressed on a dry matter basis and presented in table 1, 2 and 3 respectively.

Table 1. Proximate (mean \pm standard deviation) composition of
the ingredients used to prepare the feed

Type of constit- uent	Ingredient		
	Fishmeal	Soya bean cake	Wheat bran
% Crude protein	63.64 ± 0.43	58.96 ± 0.13	18.34 ± 0.24
% Dry matter	85.55 ± 0.49	88.90 ± 0.18	87.88 ± 0.16
% Fat / Oil	11.21 ± 0.17	2.28 ± 0.15	3.43 ± 0.16
% Ash	23.61 ± 0.38	6.72 ± 0.07	5.14 ± 0.06
% Crude fibre	0.25 ± 00	5.17 ± 0.08	9.53 ± 0.25
% Carbohydrate	1.29	26.87	63.56

Table 2: Combination of ingredients for achieving 40 % Crude

 protein in the formulated feed

Feed types				
Ingredient	1	2	3	4
% Fishmeal	40.6	-	25.3	-
% Soya bean Cake	-	45.3	25.3	-
% Wheat bran	52.7	47.5	42.2	-
% Methonine	0.5	0.5	0.5	-
% Lysine	0.5	0.5	0.5	-
% Vitamin A/ D premix	0.5	0.5	0.5	-
% Broiler premix	0.5	0.5	0.5	-
% Palm oil	4.5	4.5	4.5	-
% Salt	0.2	0.2	0.2	-
Total	100	100	100	-

 Table 3: Proximate composition of the formulated and the commercial feed

Feed types				
Bio- chemical	1	2	3	4
% Crude protein	38.48	38.72	39.23	40.9
% Lipid	10.93	10.25	8.24	3.70
% Fibre	6.27	7.16	6.05	4.85
% Ash	12.44	6.80	8.83	8.95
% Carbo- hydrate	34.21	28.53	35.24	38.09

The effects of the different feed treatment on absolute, relative fecundity, egg sizes and spawning frequency of the experimental fishes are shown in Figure 1 and Tables 4, 5 and 6 respectively. Absolute fecundity of broodfish fed the different feed types are presented in Figure 1. Feed type 4 gave the highest absolute fecundity (315.90 ± 32.63) followed by feed type 1 (249.10 ± 32.20) and feed type 2 (238.10 ± 18.63). The least absolute fecundity was recorded in feed type 3. There were no significant differences in the fecundity of the fish fed on the different kind of feeds (one–way ANOVA, F₃ = 2.15, P = 0.110).

Relative fecundity of fish fed the different feed types are presented in Table 4. Feed type 2 produced the highest relative fecundity (17.00 \pm 0.86) followed by feed type 3 (16.44 \pm 1.35). Feed types 4 and 1 produced relative fecundity of 14.79 \pm 0.95 and 14.95 \pm 1.26 respectively. However, there were no significant differences in the relative fecundity induced by the different feed types (one–way ANOVA, F3 = 0.94, P = 0.43).

The egg sizes for broodfish from the different feed treatments are presented in Table 5. Feed type 2 gave the highest mean egg size $(2.19 \pm 0.05 \text{ mm})$ followed by feed type 3 $(2.15 \pm 0.29 \text{ mm})$. Feed type 4 produced the least mean egg size of $1.67 \pm 0.05 \text{ mm}$. There were no significant differences in the egg sizes induced by the different feed types (one–way ANO-VA, F₃ = 1.71, P = 0.163).

Spawning frequency of broodfish fed different feed types are presented in Table 6. Feed type 2 produced the highest mean spawning frequency (30.50 ± 1.50) followed by feed types 1 and 3. Feed type 4 produced

the least mean spawning frequency (26.50 ± 3.50). There were no significant differences in the spawning intervals induced by the feed types (one-way ANO-VA, F₃ = 0.50, P = 0.701).

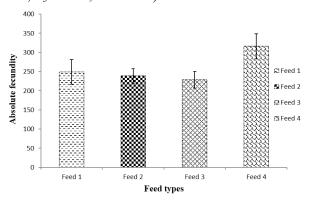


Figure 1: Absolute fecundity (mean ± 1 standard error) of broodfish fed different feed types

Table 4: Relative fecundity (mean ± 1 standard error) of broodfish fed different feed types

Feed types	Number of individual broodfish	Standard length (cm) Range	Mean relative fecundity
Feed 1	10	7.10 - 10.10	$14.95^{a}\pm1.26$
Feed 2	10	6.70 - 8.10	$17.00^{a}\pm0.86$
Feed 3	10	6.90 - 8.40	$16.44^{a} \pm 1.35$
Feed 4	10	7.40 - 10.10	$14.79^a\pm0.95$

Values with the same superscript are not significantly different (P > 0.05). N = 10 for each treatment.

Table 5: Mean egg sizes (mean ± 1 standard error) for broodfish fed different feed types

Feed types	Number of individual broodfish	Standard length (cm)Range	Mean egg sizes (mm)
Feed 1	10	7.10 - 10.10	$2.04^{a}\pm0.05$
Feed 2	10	6.70 - 8.10	$2.19^{a}\pm0.05$
Feed 3	10	6.90 - 8.40	$2.15^{a}\pm0.29$
Feed 4	10	7.40 - 10.10	$1.67^{a}\pm0.05$

Values with the same superscript are not significantly different (P > 0.05).

Table 6: Spawning frequency (days between spawning) of broodfish fed different feed types (mean ± 1 standard error)

Feed types	Number of indi- vidual broodfish	Standard length (cm) Range	Spawning intervals (days)
Feed 1	6	6.70 - 8.10	$27.00^{a}\pm3.00$
Feed 2	6	6.90 - 8.40	$30.50^{a}\pm1.50$
Feed 3	6	7.10 - 10.10	$27.50^a \pm 1.50$
Feed 4	6	7.40 - 10.10	$26.50^a\pm3.50$

Values with the same superscript are not significantly different (P > 0.05). N =6

Table 7: Summary of water quality parameters at the site during
the experimental period

Parameters	Minimum	Maximum	Mean ± standard error
Temperature (° C)	24.20	33.20	29.08 ± 0.29
DO (mg/l)	1.88	3.06	2.57 ± 0.03
pН	5.29	6.93	6.15 ± 0.03
Turbidity (ppm)	202.00	253.60	$218.28 \pm \\ 1.05$
Salinity (ppm)	194.40	243.40	210.58± 0.92

4. DISCUSSION

S. melanotheron was chosen for the study because it has been demonstrated that the species can be cultured (Eyeson 1983; Legendre *et al.* 1989; Anani *et al.* 2010), for example, in brackish water environments to augment the production of fish in coastal areas and its commercial production cannot be achieved without the understanding of its reproduction under culture conditions.

Consistent with previous studies by Boyd (2005) and Xie *et al.* (2010), the physico-chemical parameters recorded during the research were within the acceptable range for tilapia culture hence the impact are solely due to the feed types.

Fecundity can be defined as 'the number of ova that are likely to be laid by a fish during the spawning season' thus, it is a measure of the reproductive capacity of a female fish. Absolute fecundity of the females fed the different feed types ranged from 238 to 315 and showed no significant difference (P = 0.110) among the various treatments. The mean numbers of eggs produced falls within the range of number of eggs reported by Trewavas (1983) for *S. melanotheron*. Effect of the different feed types on both absolute and relative fecundity of the cultured fish was not significantly different among the various treatments.

Eyeson (1983) reported that in a confined environment, S. melanotheron can be sexually active within 4 to 6 months at a size as small as 4 to 5 cm standard length. In the current research spawning was observed from 4-6 months consistent with earlier observation by Eyeson, however standard lengths at first spawning in this study (7.2 cm) was higher than what Eyeson observed. This difference could be attributed to the differences in habitat conditions and strains. Eyeson worked on brackish water individuals whiles strains that inhabit fresh water were used in the current research. It is worth noting that spawning occurred first in fish fed feed type 4 (commercial feed). This is in-line with the findings of Nkhoma and Musuka (2014), who noticed early sexual maturation of Oreochromis niloticus when fed with commercial feed.

In this current study, mean egg sizes ranging from 1.6 to 2.9 mm (see Table 5) were recorded for the various feed treatments and there were no significant differences among the various treatments. Most of the egg sizes found were within the ranges found by Trewavas (1983) and Eyeson who identified eggs of *S. melanotheron* to have a diameter ranging from 2.0 to 3.5 mm and 2.0 to 2.5 mm respectively.

The spawning frequency (days between spawning) in this study ranged from 26.50 to 30.5 days for all the treatments. There was no significant difference among the different feed treatments. The average spawning frequencies recorded in this study were above the average of 22 days reported by Eyeson (1979).

5. CONCLUSION

In this study, all the feed types had similar influence on relative fecundity, absolute fecundity, egg sizes and spawning frequency. On the bases of the above observations, all the feed types are recommended for maintaining broodfish of *S. melanotheron* during production cycle under suitable environmental conditions.

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