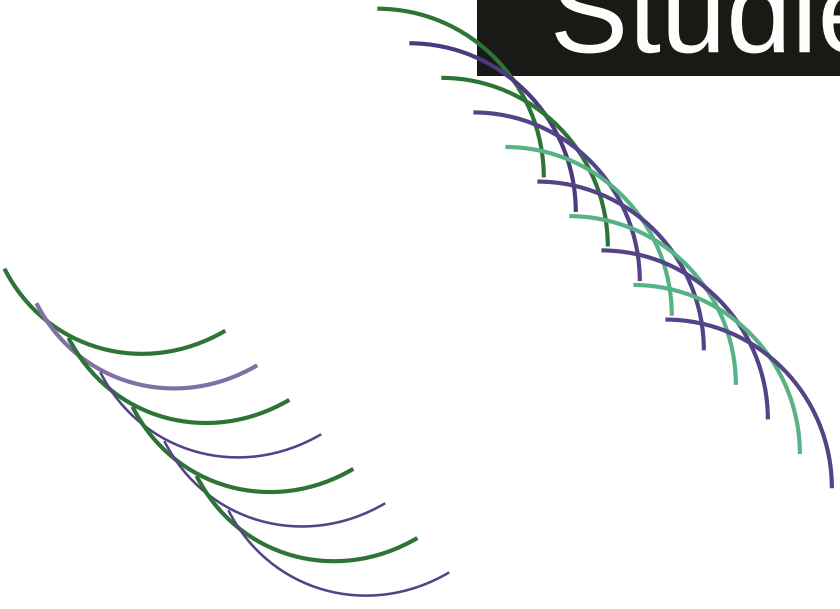
Volume 1 Issue 1 December 2015

ISSN: 2412-2505

Malawi Journal of Agriculture, Natural Resources & Development Studies



The interactive effect of water temperature and salinity on yolk absorption rate, growth and larval survival of African catfish *Clarias gariepinus* (Burchell 1822)

Ssenfuma R [1]*, Kassam D [2], Gondwe T N [2], Mtethiwa A. H [2] and Sikawa D [2]

1. Wild life and Aquatic Anima resources department, Makerere University, P. O. Box 7062, Kampala, Uganda
2. Aquaculture and Fisheries Science Department, Lilongwe University of Agriculture and Natural resources, P.O. Box 219, Lilongwe, Malawi. *Author to who correspondence can be addressed: ssenfumarobert@gmail.com

Abstract

The study was undertaken to determine the interactive effect of water temperature and salinity on yolk absorption rate, growth and survival of *C. gariepinus* larvae. Fertilized eggs, from broodstock reared for commercial seed production were incubated in 1000ml glass flasks and received one of the nine temperature (25, 28, and 31°C) and salinity (0, 3 and 6ppt) combinations in triplicates. The experiment was kept viable until all the yolk was fully disorbed. Data collected included yolk absorption rate and period, average rate of growth in length and larval survival of *C. gariepinus,* while water temperature was monitored periodically. The results showed that: Both temperature and salinity and their interaction had a significant effect on yolk absorption rate and period, average rate of growth in length and larval survival of *C. gariepinus*. A temperature and salinity combination of 28-31°C and 0ppt was found to be suitable for nursing *C. gariepinus* larvae. This is evidenced by having the least yolk absorption rate and period, highest rate of growth in length and highest survival of *C. gariepinus* larvae. Hence this temperature-salinity combination is recommended for the improvement of catfish seed production which is a major problem in catfish industry.

Key words; *Clarias gariepinus*, Yolk absorption period, rate of growth, survival, temperature, salinity

Introduction

In Africa, full aquaculture potential of African catfish, *Clarias gariepinus*, has not yet been realized despite its possession of many qualities that make it suitable for commercial production. To a large extent, the main constraint facing its commercial culture in the continent has been the lack of adequate and reliable supply of quality fingerlings for stocking purposes (Rasowo et al., 2007). This has been attributed to its characteristic gonadal seasonal cycle i.e. gravid females may be found in freshwater from October (spring) until water temperatures drop in March/ April (autumn) (Britz, 1991). It does not show any parental care, its inability to spawn naturally in captivity (Rasowo et al., 2007) and the low survival of its hatchlings in earthen ponds (De Graaf and Janssen, 1996). Considering that this species cannot naturally spawn in captivity, the current production is mainly based on induced breeding techniques and for this reason induced breeding techniques have been perfected, adequately described and are routinely practiced in many hatcheries (Hogendoorn, 1979; Hogendoorn and Vismans, 1980). However the shortage of seed for stocking ponds continues to persist in Africa (Rasowo et al., 2007). It is apparent that management protocols covering egg production, egg hatching, and particularly production techniques that enhance fry and fingerling survival need to be further studied and refined to ensure a sufficient supply of quality catfish seed.

Temperature and salinity are the two most important environmental factors affecting fish hatchery production (Aktas, 2004). Such abiotic factors influence fish eggs and larval physiology and they have a direct effect on growth and survival of fish (Holliday, 1969). However, information on effect of water temperature, salinity, and their combination is available only from fingerling to adult stages but not available on the yolk-sac larvae stage. Therefore this study aimed at unveiling how such factors affect growth, yolk absorption and survival of yolk-sac, a precursor of the subsequent stages necessary to address the current shortage of catfish fingerlings.

Methods and Materials

Study site

The study was carried out at the National Aquaculture Center at Domasi, Zomba District, in Malawi. The timing was between September and December 2008, which is the natural breeding season of *C. gariepinus*.

Brood-stock management and hormonal treatment

The mature females and males of *C. gariepinus* broodfish were selected from a stock maintained for commercial breeding program at the National Aquaculture Center at Domasi (Malawi), with individual weights ranging between 500 and 700g. All broodfish were selected using external morphological characteristics; female broodfish which were selected had soft and distended abdomen from which matured eggs, based on their greenish coloration and singular occurrence, were stripped by gentle application of pressure in accordance to Janssen (1987). Male brood fish were selected if they possessed elongated and reddish pointed urino-genital papillae. Fifteen females and 10 males were selected. Males and females were kept separately in concrete tanks measuring 10 x 8 x 1.2 m. The broodstock were acclimatized in their new environment (concrete tanks) for 7 days at mean temperature of 28 ± 2°C and normal photoperiodic regimes with water pH around 7.1±0.1 The broodfish were fed on formulated pellets (35% crude protein) twice per day (7 am and 5 pm) at 5% of total fish biomass.

Prior to hormone injection, a total of four female and four male broodfish were randomly seined out from the tanks and kept singly in aerated 50L aquaria, with 25 litres of aerated water for 12 h. The randomly-selected females were measured in terms of weights and total lengths (TL) of 580, 660, 650 and 500g, and 47.5, 50.0, 49.5 and 58.0 cm, respectively, and males had weights and total lengths of 600, 650, 680 and 700g and 46.5, 40.0, 48.5 and 65.0 cm, respectively.

Broodfish were injected with ovaprim©, a synthetic analogue of gonadotropin releasing agent (Syndel Laboratories, Canada), between 6 and 7 pm. Ovaprim was administered in liquid form at 0.5 ml/kg body weight of

female fish. Each male was injected with half of the dose of the female in accordance to Legendre, 1986; Haniffa and Sridhar, 2002. The injected fish were returned and kept separately into their respective 50L aquaria for 12h, and water temperature was maintained at 28°C using thermostatically controlled water heaters.

Fertilization and preparation of test solutions

Ovulated females were stripped the following morning after injection at room temperature (25°C) to collect eggs. The ovulated eggs oozed out by slight thumb pressure onto the plastic bowl. The fish were stripped until traces of blood were observed which signified that the ovaries were empty. For male gametes, mature males were sacrificed (killed) and sperm sacs were collected, then incisions were made on the sperm sacs following Viveiros (2002). Milt was squeezed and spread over the eggs then mixed thoroughly with a soft clean feather. To this, 0.6% saline solution was added and further agitated for few seconds. Spermatozoa from one mature male were used to fertilize eggs stripped from three females while keeping the eggs from different females separately. The process from stripping to fertilization took about three minutes to accomplish. Three concentrations of the common salt, NaCl, (0, 3, and 6 g/l) were prepared by dissolving these three amounts of salt in a liter of natural freshwater used by National Aquaculture Center for breeding catfish, in order to obtain the required salinities, for all the tests on salinity tolerance of fertilized eggs and Yolk-sac larva. The mixture was tested with a salinometer, to confirm the salinities of the test solutions.

Egg incubation, Yolk absorption rate and period, rate of growth and survival

At incubation, a 3x3 factorial design was used where three temperatures levels (25, 28, and 31°C) and three salinity levels (0, 3 and 6ppt) levels each with three replicates were set. Immediately after fertilization, eggs were transferred into 1000ml-capacity beakers (100 eggs per each beaker), beakers were filled to 800ml mark with the test media. Aerators were placed in each water bath and in each beaker to ensure temperature homogeneity and oxygen supply respectively. At the end of hatching, the hatched larvae were left in the salinities and temperatures of incubation. Yolk absorption rate was determined by measuring the size (length and height) of the yolk sac of 27 larvae, 9 from each replicate using an ocular micrometer mounted on a light microscope daily until the yolks were fully absorbed in accordance to (Molokwu and Okpokwasili, 2002). The average rate of daily yolk absorption were then determined using the formula by Borode and Akin–James (2005):

$$\sum(nI\text{-}nF)/t$$

Where:

nI = initial yolk size per

day, nF = final yolk size per

day, t = rearing period

The yolk-sac period began from the end of the hatching period until when 50% of the larvae fully absorbed

their Yolk-sac. This was determined by visual observation and the time taken was recorded. Mortality was determined by counting and by recording on a daily basis the number of dead larvae until the Yolks were fully absorbed. Percentage survival was determined by the following formula according to Radonic et al. (2007)

$$(\%)\ \text{Survival} = \frac{\text{Number of live larvae at the end of Yolk absorption}}{\text{Number of live larvae after hatching}} \times 100$$

The rate of growth (GR) was determined by measuring the length of 27 larvae, 9 from each replicate using an ocular micrometer mounted on a light microscope. The average rate of growth in length in relation to the various salt and temperature was calculated using the formula by Borode and Akin-James (2005):

$$\sum(Lf\text{ -}Li)/t$$

Where:

Lf = final daily length of larvae, Li

= initial daily length of larvae t =

rearing period.

Other water quality parameters

The following Water parameters were monitored on a daily basis. Temperature was measured using a mercury in glass thermometer, pH were read using a pH meter model 191 CP-20 digital, Dissolved Oxygen (DO) level were maintained with RESUN LP- 100 low noise air pump while values were measured using the oxygen meter YS1 model 51B. Salinity values were monitored using portable refractometer salinometer.

Data analysis

Two-way analysis of variance after normality tests for ANOVA was used to analyse the data. When treatment means were found to be significant ($P<0.05$), multiple comparisons among means were done using Scheffe's test. Both ANOVA and Scheffe's tests were done using SAS.

Results

Effect of temperature on Yolk-sac larvae

The average rate of Yolk absorption per day increased with increasing temperature at all the three incubation salinities. However there was no significant differences ($P<0.05$) at 0 and 6ppt but there were significant differences ($P<0.05$), at 3ppt. The Yolk absorption period decreased with increasing temperature at 0ppt but it increased with increasing temperature at 6ppt and significant differences ($P<0.05$) were observed. On the other hand, the lowest Yolk absorption period was observed at 3ppt when temperature was at 28°C. However, there was no significant differences ($P<0.05$) at all the three incubation temperatures. The average rate of growth in length per day increased with increasing temperature but there were no significant differences ($P<0.05$) at 0ppt and 3ppt. Highest average rate of growth in length per day was at 6ppt and 31°C. The values were not significantly different ($P<0.05$) from that which were obtained at 28°C but significantly different ($P<0.05$) from that which was obtained at 25°C. In freshwater the survival percentage increased with increasing temperature and there were no significant differences ($P<0.05$) while in salty water,

it decreased with increasing temperature and significant differences were observed (Table 1).

Table 1: The effect of temperature and salinity on average rate of Yolk absorption, Yolk absorption period, rate of growth and survival rate
(mean ±SE., n=9) of Clarias gariepinus larvae

Salinity (ppt)	Temperature (°C)	Average rate of yolk absorption per Day (mm3)	Yolk absorption period (h)	Average rate of growth in length per day (mm)	Survival (%)
0.00	25	0.997±0.110[a]	74.00±1.46[a]	0.894±0.0994[a]	90.67±1.15[a]
	28	1.013±0.112a	62.00±1.76[b]	0.675±0.0750[b]	79.67±1.52[b]
	31	1.050±0.116a	48.00±1.56[c]	0.668±0.0742[b]	54.67±4.16[c]
3.00	25	0.523±0.068a	80.00±3.46a	0.959±0.1065[a]	92.00±2.00[a]
	28	0.600±0.066b	74.00±2.46a	0.779±0.0866[b]	74.00±8.71[b]
	31	0.757±0.041c	76.00±3.06a	0.729±0.0809[b]	34.00±4.00[b]
6.00	25	0.305±0.033a	88.00±2.06b	1.011±0.1123[a]	93.00±1.00[a]
	28	0.360±0.040a	92.00±3.46a b	0.850±0.0945[b]	32.00±6.93[b]
	31	0.373±0.084a	96.00±0.56a	0.796±0.0884[a b]	08.67±1.15[c]

Means with the same superscript are not significantly different within columns (P>0.05)

Effect of salinity on Yolk sac-larvae

The average rate of Yolk absorption per day was significantly (P<0.05) highest in freshwater and it decreased progressively with increasing salinity at all the three incubation temperatures. Yolk absorption period was significantly (P<0.05) least in freshwater and it increased progressively with increasing salinity. The average rate of growth in length per day was significantly (P<0.05) highest in freshwater and increased with increasing salinity. Survival percentage was also significantly (P<0.05) highest in freshwater at all the three incubation temperatures and decreased with increasing salinity (Table 2).

Table 2: The effect of salinity on average rate of yolk absorption, yolk absorption period, rate of growth and survival rate (mean±SE n=9) of Clarias gariepinus larvae at a various temperatures.

Temperatures (°C)	Salinity (ppt)	Average rate of yolk absorption per Day (mm3)	Yolk absorption period (h)	Average rate of growth in length (mm)	Survival (%)
25	0.00	0.997±0.110[a]	74.00±1.46[a]	0.894±0.0994a	90.67±1.15[a]
	3.00	0.523±0.068[b]	62.00±1.76[b]	0.675±0.0750[b]	79.67±1.52[b]
	6.00	0.305±0.033[c]	48.00±1.56[c]	0.668±0.0742[b]	54.67±4.16[c]
28	0.00	1.013±0.112[a]	80.00±3.46[a]	0.959±0.1065[a]	92.00±2.00[a]
	3.00	0.600±0.066[b]	74.00±2.46[a]	0.779±0.0866[a]	74.00±8.71[b]
	6.00	0.373±0.041[c]	76.00±3.06[a]	0.729±0.0809[b]	34.00±4.00[b]
31	0.00	1.050±0.116[a]	88.00±2.06[b]	1.011±0.1123[a]	93.00±1.00[a]
	3.00	0.757±0.084[b]	92.00±3.46[ab]	0.850±0.0945[b]	32.00±6.93[b]
	6.00	0.360±0.040[c]	96.00±0.56[a]	0.796±0.0884[ab]	08.67±1.15[c]

Means with the same superscript are not significantly different within columns (P>0.05)

Interactive effect of temperature and salinity on yolksac larvae

The highest average rate of yolk absorption per day was obtained at temperature-salinity combinations of 31°C and 0ppt, this was not significantly different (P<0.05) from those obtained at 28°C and 0ppt and 25°C and 0ppt, while the least mean average rate of yolk absorption per day was recorded at temperature-salinity combination of 25°C and 6ppt. The least yolk absorption period was obtained at temperature-salinity combination of 31°C and 0ppt, and the highest yolk absorption period was recorded at temperaturesalinity combination of 31°C and 6ppt. The highest average rate of growth in length per day was obtained at temperature-salinity combinations of 31°C and 0ppt, however, this was not significantly different (P>0.05) from those which were recorded at 28°C and 0ppt and 25°C and 0ppt. The highest survival rate was recorded at temperaturesalinity combination of 31°C and 0ppt, however this was not significantly different (P>0.05) from those which was recorded at 28°C and 0ppt, and 25°C and 0ppt, while the least survival rate of 8.67 was recorded at temperaturesalinity combination of 31°C and 6ppt.

Table 3: The interactive effect of temperature and salinity on average rate of yolk absorption, yolk absorption period, rate of growth and survival rate (mean±SE., n=9) of Clarias gariepinus larvae.

Temperatures (°C)	Salinity (ppt)	Average rate of yolk absorption per Day (mm3)	Yolk absorption period (h)	Average rate of growth in length (mm)	Survival (%)
25	0.00	0.997±0.110[a]	74.00±1.46[d]	0.894±0.0994[a]	90.67±1.15[a]
	3.00	0.523±0.068[b]	80.00±3.46[c]	0.675±0.0750[b]	79.67±1.52[b]
	6.00	0.305±0.033[c]	88.00±2.06[b]	0.668±0.0742[b]	54.67±4.16[c]
3.00	0.00	1.013±0.112[a]	62.00±1.76[e]	0.959±0.1065[a]	92.00±2.00[a]
	3.00	0.600±0.066[b]	74.00±2.46[d]	0.779±0.0866[b]	74.00±8.71[b]
	6.00	0.360±0.040[ef]	92.00±3.46[ab]	0.729±0.0809[b]	34.00±4.00[b]
6.00	0.00	1.050±0.116[a]	48.00±1.56[f]	1.011±0.1123[a]	93.00±1.00[a]
	3.00	0.757±0.084[b]	76.00±3.06[c]	0.796±0.0884[b]	32.00±6.93 [b]
	6.00	0.373±0.041[e]	96.00±0.56[a]	0.850±0.0945[b]	08.67±1.15[e]

Means with the same superscript are not significantly different within columns (P>0.05)

Discussion

Water Temperature affected the average rate of yolk absorption per day and yolk absorption period the average rate of yolk absorption per day increased with increasing temperature while the yolk absorption period decreases with increasing temperature. A similar trend was reported by Haylor and Mollor (1995) who incubated *C. gariepinus* eggs in freshwater and reported the yolk absorption period of 74.40-90.24h at temperature range of 24-26°C, 63.06h at 28°C and

48.96-55.20h at 30-32°C. Increased metabolism at higher temperature could have been responsible for the direct relationship between the average rate of yolk absorption per day and temperature and the inverse relationship between temperature and yolk absorption period (Nwosu and Hertzlohner, 2000).

At 3 and 6ppt, the yolk absorption period decreased with increasing temperature while the least average rates of yolk absorption per day was obtained at 28°C. However, this was not significantly different from those obtained at 25°C and 31°C (Table 2). The inverse relationship between yolk absorption period and temperature could be due to the high metabolic rates at high temperature as reported by Nwosu and Hertzlohner, (2000). While the results obtained for the average rate of yolk absorption per day suggests that salinity modifies the effect of temperature on yolk absorption per day.

Water temperature affected the average rate of growth in length per day and survival of yolk sac larvae at a given salinity in that, at all the three incubation salinities (0-6ppt), the average rate of growth in length per day increased with increasing temperature. These results are in line with those reported by Britz and Hecht (1986) who observed better growth of *C.gariepinus* larvae with increasing temperature. Similar results were also reported by Haylor and Mollor (1995) that number of day-degrees decrease with increasing temperature until hatching, first feeding and yolk sac absorption in *C.gariepinus*. In the current study, at 0ppt, the survival of yolk sac larvae increased with increasing temperature. These results are in contrast with those by Nwosu and Hertzlohner (2000), who reported that survival of larvae of *Heterobranchus longifilis* reduced with increasing temperature. When they reared their larvae in fresh water; they obtained the highest survival of 67% at 25°C, 50% at 27°C, 33% at 29°C, and 13% at 32°C. However the results are in line with those by Fashina-Bombata and Busari (2003), who reported the highest survival of 93.7% when they reared *H. longifilis* at 27±0.5°C in fresh water. At 3 and 6ppt the survival of *C.gariepinus* yolk sac larvae decreased with increasing temperature, however the decrease was more pronounced at high temperature. These results are in line with those reported by Borode et al. (2002) who reported no significant difference when they reared *C.gariepinus* yolk sac larvae at temperatures of 26-27°C in salinities of 0, 2, 4, and 6ppt, however they did not show the exact figures for their results. Bioenergetic studies indicate a strong positive relationship between temperature and metabolism in fish (Wotton, 1995). Wotton, further noted that increased metabolic rate increases the levels of food intake in order to maintain growth and survival rates. In the current study, the observations show a positive relationship between the average rate of growth in length per day and survival of yolk sac larvae with temperature. This relationship could have been due to the increased metabolic rate which increased yolk utilization hence maintaining high growth and survival rates of *C.gariepinus* yolk sac larvae. The finding of this study therefore indicates that the survival of *C.gariepinus* yolk saclarvae increased with increasing temperature in freshwater.

Salinity affected average rates of yolk absorption per day and yolk absorption period at a given salinity in that, at all the three incubation temperatures (25-31°C). The average rate of yolk absorption per day decreased with increasing salinity, while the Yolk absorption period increased with increasing salinity from 0-6ppt. Similar results were reported by Borode and Akin-James (2005), who reported that the yolk absorption decreases progressively with increased salt concentration for the crosses of *C.gariepinus* x *H. longifilis*. They found out that yolk absorption rate was highest in the control, but it decreased with increased salinity. High rate of yolk absorption means small yolk size and low rate of yolk absorption means large yolk size. These results are in agreement with those reported by Borode et al. (2002), who reported that yolk size was highest at higher salt concentrations than at lower salt concentration. They recorded the yolk size of 0.3mm3 at 0ppt, 0.4mm3 at 2ppt, 0.3mm3 at 4ppt and 0.7mm3 at 6ppt. Rice (1990) explained that this inverse relationship could have been due to the decrease in metabolic rate at high salinities, He further reported that, factors which reduce metabolic rate like high salinity may be accompanied by a decrease in yolk consumption. Therefore in the present study, the inverse relationship between average rates of yolk absorption per day and yolk absorption period and salinity might have been due to the decreased metabolism at high salinities.

Salinity affected the rate of growth in length per day and larval survival in that, at all the three nursing

temperatures ranging from 25 to 31°C, the average rate of growth in length per day and larval survival decreased with increasing salinity. These results are in line with those reported by Borode and Akin-James (2005) who reported that mean body lengths and survival of the larvae of a hybrid C. gariepinus (f) x H. longifilis (m) decreased with increasing salt concentration. They further explained that this was as result of the predominance of *C. gariepinus* (f) over H. longifilis (m). *C. gariepinus* is a freshwater stenohaline fish known for increasing maintenance requirements at higher salinities and consequent weight loss which resulted into reduced growth and survival (Borode and Akin-James, 2005). These results are also in agreement with those reported by Britz and Hecht (1986) who reported decreasing growth rate with increasing salinity for *C. gariepinus* larvae. Furthermore the results obtained in this study agree with those reported by Fashina-Bombata and Busari (2003), who reported the highest survival of 93.75% at 0ppt when they reared yolk sac larvae of H. longifilis at 27±0.5°C in water at different salinities. They further reported that, the yolk sac-larvae which were hatched in water at high salinities and transferred in water at lower salinities highest survival of 93.75% at 0ppt. The finding of the current indicated a negative relationship between the average rate of growth in length per day and survival of yolk sac larvae with salinity. This could have been due to the characteristic nature of *C. gariepinus* as a freshwater stenohaline fish which increases maintenance requirements at higher salinities resulting into weight loss, reduced growth and survival (Borode and Akin-James, 2005).

The interactions of both temperature and salinity on average rate of yolk absorption per day and yolk absorption period revealed that, the highest average rate of yolk absorption per day and the least yolk absorption period were obtained at the highest incubation temperature (31°C) in freshwater. These results are in line with those reported by Borode et al, (2002), who nursed *C. gariepinus* larvae at different water salinities and reported that rate of yolk absorption was delayed in saline water than in fresh water. They found out that the rate of yolk

absorption was significantly faster in the control and 2ppt salt treatments, but slower in 4, 6, and 8ppt. This could have been as a result of the decrease in metabolic rate at high salinities (Rice, 1990) and the high metabolic rates at high temperature (Nwosu and Hertzlohner, 2000).

In a conclusion, this study showed that both temperature and salinity and their combinations had a significant effect on yolk absorption period, average rate of yolk absorption per day, average rate of growth in length and larval survival of *C. gariepinus*. The temperature and salinity combination of 31°C and 0ppt is better for the nursing of *C. gariepinus* as evidenced by having least yolk absorption period, high average rate of development in length and high survival of *C. gariepinus* larvae, therefore it is recommended that *C. gariepinus* larvae should be nursed at a temperaturesalinity combination of 31°C and 0ppt.

Acknowledgement

Special thanks goes to the National Aquaculture Centre (NAC) at Domasi for availing their institutional facility for this research and Icelandic International Development Agency (ICEIDA) for providing the first author with the postgraduate scholarship in Malawi.

References

Akitas, M. 2004. Combined effect of temperature and salinity on egg hatching rates and incubation time of Penaeus semisulcatus (Decapoda: Penaeidae). Israel journal. 2: 250-257

Borode, A.O. and Akin–James O. 2005. Effect of salinity level on embryonic development, hatchability and survival potential of reciprocal hybrid larvae of Clariid catfish (Clarias gariepinus x Heterobranchus bidorsalis). Journal of Food, Agriculture & Environment. 3(1): 243-248

Borode, A.O., Balogun, A.M. and Omoyeni, B.A. 2002. Effect of salinity on embryonic development, hatchability and growth of African catfish, Clarias gariepinus, eggs and larvae. Journal of Applied Aquaculture. 12(4): 89-93.

Britz P. J. 1991. The utility of hornoplastic pituitary glands for spawning induction of the African catfish (Clarias gariepnus) in commercial aquaculture in Africa. Water South Africa. 17: 237-241.

Britz, P.J. and Hecht, T. 1986. Temperature preferences and optimum temperature for growth of African catfish (Clarias gariepinus) larvae and post-larvae. Aquaculture. 63: 205-214.

De Graaf, G. & Janssen, H. 1996. Artificial reproduction and pond rearing of the African catfish Clarias gariepinus in sub Saharran Africa. Fisheries Technical Paper 362. Food and Agricultural Organisation of the United Nations. Rome, Italy

Fashina-Bombata and Busari A.N A.N. 2003. Influence of salinity on the developmental stages of African catfish Heterobranchus longifilis (Valenciennes, 1840). Aquaculture. 224: 213–222

Haniffa, M.A.K., and Sridhar, S. 2002. Induced spawning of spotted murrel Channa punctatus) and catfish Heteropneustes fossilis using human chorionic gonadotropin and synthetic hormone (Ovaprim). Vetinary Achievement. 72(1): 51 – 56.

Haylor, G.S. 1993. Controlled hatchery production of African catfish, Clarias gariepinus (Burchell): an overview. Aquaculture and Fisheries Management. 24: 245-252.

Haylor, G. S. and Mollah, M.F.A. 1995. Controlled hatchery production of African catfish, Clarias gariepinus: the influence of temperature on early development. Aquatic Living Resource. 8: 431-438

Hecht, T., and Appelbaum, S. 1988. Observations on intraspecific aggression and coeval sibling cannibalism by larval and juvenile Clarias gariepinus (Clariidae: Piscis) under controlled conditions. Journal of Zoology London. 214: 21-44.

Hocutt, C.H. 1989. Seasonal and diel behaviour of radio-tagged Clarias gariepinus in Lake Ngezi, Zimbabwe (Pisces:Clariidae). Journal of Zoology London. 219: 181-199.

Hogendoorn, H. 1979. controlled propagation of African catfish Clarias lazera. Reproductive Biology and field experiments. Aquaculture. 17: 323-333.

Hogendoorn, H., Hardeman, G.J., Vismans, M.M., Viveen, W.J.A.R., (1980). Controlled propagation of the labyrinthic catfish, Clarias lazera (C. and V.) for experimental purposes. In: Proceedings of the 7th ICLAS Symposium, Utrecht, 1979. Gustav Fisher, Stuttgart, pp. 363–371.

Holliday, F.G.T. 1969. Effect of salinity on the eggs and larvae of Teleosts. In: Hoar, W.S., Randall, D.J. (Eds.), Fish Physiology, vol. 1. Academic Press, New York, pp. 293–311.

Hossain, M.A.R., Beveridge, M.C. M., Haylor, G.S., 1998. The effects of density, light and shelter on the growth and survival of African catfish (Clarias gariepinus Burchell, 1822) fingerlings. Aquaculture. 160: 251-258.

Khwuanjai, H., Ward F.J., and Pornchai J. 1997. The effect of stocking density on yield, growth and mortality of African catfish (Clarias gariepinus Burchell (1822) cultured in cages. Aquaculture. 152: 67-76.

Legendre, M. 1986. Seasonal changes in sexual maturity and fecundity and human chorionic gonadotropin (HCG) induced breeding of the catfish Heterobranchus longifilis Val (Clariidae) reared in Ebrie lagoon (Ivory Coast). Aquaculture. 55: 201-213

Molokwu, C.N. and Okpokwasili, G.C. 2002. Effect of water hardness on egg hatchability and larval viability of Clarias gariepinus Kluwer. Aquaculture International. 10: 57–64,

Nwosu, F.M. and Hertzlohner S. 2000. Influence of temperature on egg hatching, growth and survival of larvae of Heterobranchus longifilis Val. 1840 (Teleostei: Clariidae). Journal of Applied Ichthyology. 16: 20-23

Radonic. M., Muller M.I., Lopez A.V., Bambill, G.A., Spinedi, M., and Boccanfuso, J.T., 2007. Improvement in flounder Paralichys orbignfarus controlled spawning in Argentina Ciencias Marinas. 33(2): 187-196.

Rasowo, J., Okoth, O. E., and Ngugi, C. C. 2007. Effects of formaldehyde, sodium chloride, potassium permanganate and hydrogen peroxide on hatch rate of African catfish Clarias gariepinus eggs. Aquaculture. 269: 271-277.

Tomasz, P., Czeslowa G.A.J, Henryle B. 1997. Usage of NaCl as a prophylactic medium for Egg incubation and for rearing of the African Catfish (Clarias gariepinus) at the stage of hatch and Fry. Aquaculture. Xxvii: 53-58.

Viveiros, A.T.M., Fessehaye Y., ter Veld M., Schulz R.W. and Komen J. 2002. Hand-stripping of semen and semen quality after maturational hormone treatments, in African catfish Clarias gariepinus. Aquaculture. 213: 373–386.

Wotton RS. 1995. Temperature and lake-outlet communities. Journal of Thermal Biology 20: 121– 25.