

SOME ECO-PHYSIOLOGICAL RESPONSES OF MALE NILE TILAPIA (*OREOCHROMIS NILOTICUS*) TO THERMAL AND SALINITY CONDITIONS

ZEINAB; A. NAGDY*, S. A. ABD-ELMAGEED*, M. M. SHAFIE
AND M. A. SALEM****

* Central Laboratory for Aquaculture Research-Agriculture Research Center- Agriculture Ministry

**Agriculture Faculty-Cairo University

Received: 13. 1. 2004.

Accepted: 10. 4. 2004.

SUMMARY

Two trials were conducted in Animal Physiology Lab., Faculty of Agriculture; Cairo University from May to November, 1999 to quantify and evaluate the growth performance of male Nile Tilapia (*Oreochromis niloticus*) affected by physiological responses to thermal and salinity conditions in aquaria. The experiments were conducted either on fry (for 30 days) or fingerlings (for 42 days). The experimental study incorporated six treatments (T1-T6) between two temperature degrees (28.59, 33°C for fry and 23.91, 33°C for fingerlings) and three salinity levels fresh brackish and sea water (0.1, 15 and 30 ppt, respectively). Both dissolved oxygen (DO) and pH did not reach critical values for both fry and fingerlings. The daily weight gain of fry was greater (0.145 g/fish/day) in fresh water than in brackish water (0.041 g/fish/day). The lowest daily gain was in

seawater (0.037 g/fish/day), the differences were significant ($p < 0.05$). After 30 days from starting the experiment, the daily weight gain of fingerlings showed equal means (0.1955 and 0.196 g/fish/day) in both fresh water and brackish water, compared to half value in seawater (0.096 g/fish/day). The fish could not tolerate the high temperature and/or concentrated salinity (seawater 30 ppt) further than 30 days. It was obvious that the fish chemical constitutions were not affected by temperature or salinity as well as their interactions. The concentrations of Na⁺, K⁺, P⁻⁻⁻ and Cl⁻ in blood plasma were, 477.47, 40.44, 0.88 and 249.09 mg/dl, respectively, under fresh water (0.1 ppt salinity). In the sea water (30 ppt salinity) the values were 490.97, 34.37, 1.24 and 300.5 mg/dl, while in brackish water were 456.45, 31.99, 1.31 and 337.59, respectively. The ions (Na⁺, K⁺ and P⁻⁻⁻) besides Ca⁺⁺ and Mg⁺⁺ in the fish tissue (dried body) were 22.95, 0.21,

3.36, 41.18 and 2.27 mg/g respectively in fresh water (0.1 ppt), while in seawater (30 ppt) the values were 18.1, 0.19, 3.38, 42.09 and 2.31 mg/g respectively. Fish in seawater treatments had the highest concentration of aldosterone hormone in blood plasma with the average of 0.55 pg/ml, while fresh and brackish water treatments induced lower and similar concentrations of aldosterone, 0.46 and 0.48 pg/ml, respectively after 30 days.

INTRODUCTION

Fish farming was limited primarily to fresh and brackish water of low salinity, however, their competition with agriculture together with programs of lagoon development led to the expansion of tilapia culture in brackish or even seawater (Payne, 1983). The limited freshwater resources in Egypt and the Egyptian laws restricting the use of freshwater in aquaculture projects (Egyptian law No.124, 1982) have encouraged sea or brackish water aquaculture. Anyhow these waters have variable conditions of salinity and temperature which are apt to affect-positively or negatively the performance of fish. There are trials to establish aquaculture of Tilapia (*Oreochromis niloticus*) fish in many regions in Egypt (i.e. Sahl El-tinah, Sinay). There is a need to test the water environmental conditions, which may affect performance of this species, in such an area. The physiology of salinity tolerance in Tilapia has been reviewed by Prunet and Bornancin,

(1989). Many species maintained good euryhalinity in African estuaries and coastal water as reported by Stickney (1986). Laboratory investigations were initiated to evaluate some morphological and eco-physiological parameters of the adaptive response to hypertonic media of one tilapia (*O. niloticus*) species of economic importance (Avella et al., 1993).

The ultimate goal of the present study was to detect, experimentally, the physiological response of sex reversal male Nile tilapia (*O. niloticus*) fry and fingerlings to environmental stresses of high salinity and hot condition as a prediction to the effect of pollution changes in natural water in the Nile and channels. The experimental work aimed at two objectives: growth performance of the fry under these stressful factors, growth performance and physiological reactions of fingerlings to those environmental conditions.

MATERIALS AND METHODS

The study was carried out on Nile tilapia fish (*O. niloticus*) in the Animal Physiology Lab., Animal Production Department, Faculty of Agriculture; Cairo University during 1999. The experimental work was extended from May to November as fixed by the availability of fry and fingerlings of Nile tilapia, from the hatchery of Faculty of Agriculture, Ain Shams Univ., Egypt. Eighteen (90-L) glass aquaria, with 60-L water volume were used, each aquarium was aerated with one air stone (Air

Pump) and one thermo-automatic-heater (the Rena F).

Experimental Design

a) Study of fry response

Sex reversal male Nile tilapia (*O. niloticus*) fry (0.43-0.73 g/fish)- which were collected from the females mouths and transported to the laboratory, after consumption of yolk sac the artificial feed was administrated to this fry, every one kg of this feed was treated with 60 mg of 17- α methyltestosterone and offered to the fry at a rate of 20% from the fry biomass four times a day for 21 days- were used to study the growth performance of fry under six treatments comprising three levels of water salinity (0.1, 15 and 30 ppt) combined with two levels of temperature (room water temperature, $28.59 \pm 0.43^{\circ}\text{C}$ and controlled $33 \pm 1^{\circ}\text{C}$). The treatments remained for 30 days, with stocking density of fifteen fry/ aquarium, in three replicates for each treatment.

b) Study of fingerlings response

Fingerlings of the same species (11.60 ± 0.42 g/fingerling) with 15 fish/aquaria stocking density were used to study the growth performance and some eco-physiological reactions, for 42 days under six conditions (three replicates for each treatment) of salinity and temperatures. Three levels of water salinity (0.1, 15 and 30 ppt) as used for fry study combined with two levels of temperature (23.91 ± 0.49 and $33 \pm 1^{\circ}\text{C}$).

Maintenance of water conditions was conducted by replacing the aquaria waters with similar waters twice a week. The temperature and salinity has been measured and adjusted daily using the salinity meter (YSI 35). The dissolved oxygen and pH were measured weekly using Dissolved Oxygen meter (YSI 57) and Kit-system analysis (HACH, 1982), respectively.

Fish feed contained 40% protein was offered at an initial rate of 10 % (fry study), the protein percentage decreased gradually with increasing fish growth till 25% (fingerlings study) which fed at 5% of body weight. The feed was delivered two times a day. Feeding rates were recalculated and adjusted weekly on basis of the change in fish weight as assessed by fish samples. Fish were sampled every week to determine the fish weight and to study the growth.

The fry were acclimated to the aquaria water conditions for two weeks before recording the data on the studied traits. The fingerlings responses were recorded spontaneously at the start of the experiment. Survival Assessment (%), average Daily Weight Gain (DWG g/fish/day), Specific Growth Rate (SGR), Head and Gill weights percentages to body weight were assessed for the two ages, fry and fingerlings.

In blood plasma for the fingerlings after 30 and 42 days of experimentation some electrolytes were measured. Sodium (Na^+) and Potassium

(K⁺) concentrations were measured using kits of bio-analytics (palm City, USA) at wave lengths of 550 and 420 nm; respectively; Chloride (Cl⁻) using kit of Thyocynate method (Amposta, Spain) at wave length 450 nm; Inorganic Phosphorus (P⁻⁻⁻) level using inorganic phosphorus reagent set (UV) method (point Scientific Inc., Michigan, USA) at wave length of 340 nm; the spectrophotometer (model 1201 Milton Roy, USA) was used in these measurements as well as Aldosterone concentrations using immuno technique by Immuno Tech. kits (France) with the 1275 Mini-Gama Countr (USA).

Major electrolytes; Sodium (Na⁺), Potassium (K⁺), Magnesium (Mg⁺⁺), Calcium (Ca⁺⁺), and Inorganic Phosphorus (P⁻⁻⁻) levels were obtained after 30 and 42 days from starting of the experimental period in fingerlings dry body. The determinations were carried out by the technical methods as mentioned above for blood plasma. The extraction methods were according to AOAC (1990).

Statistical analysis

All data were subjected to analysis of variance by using general linear model (GLM) procedure of SAS (1996) for personal computer by two way-analysis of variance. The model used was as follow:

$$Y_{ijk} = M + S_i + T_j + (ST)_{ij} + E_{ijk}$$

Where,

Y is the observations,

M is the overall mean,

S is the effect of salinity where i = 1, 2 and 3,

T is effect of temperature where j = 1 and 2,

ST is the effect of interaction between salinity and temperature,

E is the error value.

Duncan's test least square means (LSM) were used to identify significant differences between treatments according to SAS (1996).

RESULTS AND DISCUSSIONS

Water DO and pH

It is clear from table (1) that the changes in both dissolved oxygen (DO) and pH in the present study were minute and satisfy the biological requirements for the *O. niloticus* species (fry and fingerlings). Accordingly these two elements will be of no effect on the responses of fry and fingerlings to the temperature and salinity conditions. The responses of DO to temperature and salinity agree with Boyd (1990) statement that the solubility of DO decreases as temperature and salinity increase. Swingle (1969) considered that concentrations of dissolved oxygen below 5.0 mg/l are undesirable in ponds for practical purposes. The pH values in this study never reached critically high or low levels as denoted by Boyd (1979 and 1990). Water with pH ranging from about 6.5 to 9.0 was most suitable for fish production as stated by Ellis (1937). These responses to salinity agree

Table (1): Dissolved oxygen (DO mg/l) and pH values of water under the different conditions of salinity (ppt) and temperature (°C) levels for cultured male *O. niloticus* fry and fingerlings after 30 days of treatment (Mean \pm SE).

Treatment	DO (mg/l)		pH	
	Fry	Fingerlings	Fry	Fingerlings
T 1 (28.59 °C* , 0.1 ppt)	5.84 ^A \pm 0.19	5.00 ^A \pm 0.47	7.73 ^B \pm 0.12	7.48 ^B \pm 0.10
T2 (33°C, 0.1 ppt)	5.4 ^A \pm 0.18	2.50 ^C \pm 0.44	7.48 B ^A \pm 0.09	7.43A ^B \pm 0.13
T3 (28.59°C*, 15 ppt)	5.69 ^A \pm 0.17	3.72 ^B \pm 0.22	8.23 ^A \pm 0.12	7.62A ^B \pm 0.10
T4 (33°C*, 15 ppt)	4.73 ^B \pm 0.27	3.59 ^B \pm 0.19	0.23 ^A \pm 0.13	7.76 ^A \pm 0.10
T5 (28.59°C, 30 ppt)	4.75 ^B \pm 0.11	4.73 ^A \pm 0.27	8.14 ^A \pm 0.07	7.71 ^A \pm 0.10
T6 (33°C, 30ppt)	4.59 ^B \pm 0.26	3.12 ^B \pm 0.27	8.16 ^B \pm 0.07	7.71 ^A \pm 0.13

* The temperature level of T1, T3 and T5 was 23.91°C for fingerlings instead of 28.59 for fry

A, B, C. Values-having different superscript are significantly (P<0.05 different)

with Boyd (1990) statement of increased pH due to the high concentration of alkali metal, usually sodium.

Survivability

It is clear from the present results that 0.1 ppt salinity and 28.59°C temperature (T1) were the best conditions for *Tilapia* fry culture, compared to the other experimental conditions (Table, 2). By these circumstances the fish showed 93.33% survival (that means only 6.77% mortality). The data denote greater effect of salinity changes than temperature changes. It is clear that survival percentage of fry was affected by changes in salinity and its interaction with temperature level but not by changes in temperature. Opposite to fry the

fingerlings showed thermal effect on survival percentage, perhaps to wider temperature level than with fry, however less than salinity effect, with interaction only at the highest temperature (33°C).

These results agree with Likongwe et al. (1996), who reported a decreased in survival percentage when salinity increased, the highest survival percentage was 95.5% at the interaction between 32°C and 0.1 ppt salinity, and the lowest survival percentage was 44.4% at the interaction between 28°C and 16 ppt salinity, the differences were significant (p<0.05). Nour et al. (1996) stated that the survival rate of *O. niloticus* reached 77.5% in covered cement ponds (19.8°C) and 52.5% in

Table (2): Least square means (LSM) analyses of variance of male *O. niloticus* fry and fingerlings traits as affected by thermal and salinity conditions and their interactions, after 30 days of treatments.

Factor	Survival (%)						Daily weight gain DWG (g/fis/d)						Specific growth rate SGR					
	Fry		Fingerlings		Fry		Fingerlings		Fry		Fingerlings		Fry		Fingerlings			
	LSM	SE	LSM	SE	LSM	SE	LSM	SE	LSM	SE	LSM	SE	LSM	SE	LSM	SE		
<u>Temp. (T, °C):</u>	ns		*				**		ns				*		ns			
Room temp.	69.78	5.85	81.48	7.35			0.09	0.01	0.160	0.02			4.97	0.31	1.13	0.14		
Hot temp. (33 °C)	61.56	5.85	50.37	7.35			0.06	0.01	0.170	0.02			3.72	0.31	1.16	0.14		
<u>Sal. (S, ppt):</u>	**		**				**		*				**		*			
0.1	92.17	7.16	90.00	9.00			0.15	0.01	0.196	0.03			5.88	0.38	1.38	0.17		
15	69.00	7.16	70.00	9.00			0.04	0.01	0.196	0.03			3.64	0.38	1.31	0.17		
30	35.83	7.16	37.78	9.00			0.04	0.01	0.095	0.03			3.71	0.38	0.73	0.17		
<u>Room temp. (T1)*S:</u>	*		ns				**		ns				*		ns			
T1*0.1	93.33	8.38	91.11	9.16			0.18	0.01	0.190	0.04			6.42	0.43	1.38	0.24		
T1*15	75.67	8.38	91.11	9.16			0.05	0.01	0.210	0.04			4.07	0.43	1.42	0.24		
T1*30	40.33	8.38	62.22	9.16			0.05	0.01	0.070	0.04			4.41	0.43	0.57	0.24		
<u>Hot temp. (33 °C=T2)*S:</u>	*		*				**		ns				ns		ns			
T2*0.1	91.00	11.61	88.89	15.50			0.11	0.01	0.198	0.04			5.33	0.60	1.37	0.23		
T2*15	62.33	11.61	48.89	15.50			0.03	0.01	0.179	0.04			2.84	0.60	1.21	0.23		
T2*30	31.33	11.61	13.34	15.50			0.03	0.01	0.119	0.04			3.00	0.60	0.89	0.23		

Note: Room temperature level was 28.59 °C during fry study while it was 23.91 °C during fingerlings study.

* = significant (p<0.05), ** =highly significant (P<0.01), ns = not significant.

uncovered cement ponds (17.8°C) in winter season in Egypt. Ibrahim (2000) concluded that, *O. niloticus* had 100% survival percentage in salinity up to 16 ppt with slight reduction in survival percentage at 17 ppt. An increase in mortality percentage occurred between 17 and 20 ppt, while no individuals could survive after direct transferring to 20 ppt.

Growth Traits

The fish attained considerable final weight in that experimental period (30 days), denoting fast growth as daily weight gain (DWG) and specific growth rate (SGR) (Table, 2).

El-Eraky et al. (1988) concluded that the DWG of *O. niloticus* fingerlings (initial weight ranged 27.2-27.8 g/fish) cultured for 9 weeks increased from 0.05 to 0.16 g/fish/day with increasing the water temperature from 16 to 32°C in controlled system. The specific growth rate is one of the indices for evaluating salinity tolerance in fish (Ibrahim, 2000). He reported that the SGR of *O. niloticus* fingerlings (initial weight ranged from 8.55 to 9.55 g/fish, and final weight ranged from 17.93 to 21.77 g/fish) reared for 60 days under different salinities 0.1, 10 and 17 ppt was 0.57, 0.63 and 0.53, respectively.

Fry and fingerlings showed a significant response in DWG and SGR only to salinity (Table, 2). In general, the results showed that *O. niloticus* is generally tolerant to a wide range of environmen-

tal conditions, of salinity and temperature. However it grows best in fresh and brackish waters while in sea water (30 ppt) its growth decreases as mentioned above. These results agree with those of Payne (1983) and Assem (1995).

Likongwe et al. (1996) studied the effects of water temperature, salinity, and their interactions on growth of *O. niloticus* fry. They found that specific growth rates increased with increasing water temperature from 24°C to 28°C and 32°C, the respective daily increasing percentage were 2.03, 2.6 and 2.62. At lower temperature all salinity levels higher than 8 ppt was found to depressed SGR (2.41 and 2.18 %/day in 12 and 16 ppt, respectively) while it was 2.55 and 2.51%/day at 0.1 and 8 ppt. The authors concluded that salinity modified temperature effects on growth performance at 0.1, 8 and 16 ppt. Growth performance was the highest at 32 °C at all salinities except at 12 ppt, where the highest growth was observed at 28°C. Morsy (1994) noticed that there was a tendency to decrease in DWG by increasing the salinity for *O. niloticus*, *O. aureus*, and their hybrid. *O. niloticus* showed the lowest weight gain. Suresh and Kwei-Lin (1993) stated that a range of 10-20 ppt salinity is optimal for growth of tilapia species. Also, Watanabe et al. (1985) reported that the normal growth and reproduction are constrained by increasing salinity. Verdegem et al. (1997) found that the fry (initial mean weight 0.75g) cultured for 105 days, in the brackish water (19 ppt) grew slower than those in fresh water,

the final mean body weights were 104 and 115 g/fish, respectively.

Anatomical features

The weight ratio "Head/Body" was the least under the T1 conditions (0.1 ppt salinity and 28.59°C) for fry but at higher salinity (15 ppt salinity and 23.91°C) for fingerlings 24.60% and 24.05% respectively), when compared to the other treatment conditions (Table, 3). This ratio increased with increasing temperature or salinity. However, the statistical LSM analysis did not show significant level, except for salinity on fry (Table, 3). This phenomenon needs exhaustive study with older ages (marketable fish) since it suggests less edible flesh under those stressful conditions.

Miller and Ballantine (1974) stated that the head/body weight % ranged from the smallest value (22.2%) in *O. mossabicus* to the largest value (32%) in *T. hornorus*. Morsy (1994) stated that the length of *O. niloticus* head increased slightly with increasing age, representing nearly 28% from the total body length.

It is obvious from table (3) that the heavily stressed fish (fry and fingerlings), under the higher salinity (30 ppt) and/or the highest temperature (33°C) had the greatest Gill: Body ratio in a struggle to maintain internal optimum physiological norms.

It is well known that the gills are the major system acting in two vital processes, respiration (O₂ and CO₂ exchange) and osmotic balance (water solutes) (Daoust et al., 1984; Galat et al., 1985; Norrgren et al., 1991 and Cataldi et al., 1995). Doyle and Gorecki (1961) stated that the number of chloride cell in gill lamella increase (expected to increase gills' weight) in response to high external Cl⁻ concentration, and this is a common, non pathological occurrence. Galat et al. (1985) studied the correlation coefficients (r) among lake water chemical constituents and histological changes of gill chloride cells hyperplasia of *Salmo clark henshaw*, living in lakes of different salinity and alkalinity. The (r) values were 0.362 with total dissolved salts and 0.355, 0.469, 0.154, 0.371 and 0.194 with Na⁺, K⁺, Ca⁺⁺, Mg⁺⁺ and Cl⁻, respectively. They reported that the incidence and severity of gill chloride cells hyperplasia increased as lake water total dissolved salts increased and appeared slightly more related to alkalinity (HCO₃⁻ + CO₃⁻⁻) than to Sodium Chloride.

It is clear that the alimentary tract: body weight percentage of fingerlings was affected severely by salinity and interaction with temperature, but not by temperature alone. The values were greater in brackish water especially with the high temperature, while it was smaller in sea water (Table, 4). Generally, the liver: body weight percentage was not affected by these conditions and interactions, except that at 23.91°C with salinity level 30 ppt. The values were higher in fresh wa-

Table (3): Least square means (LSM) analysis of varinace for the effect of salinity and temperature levels on some fry and fingerlings traits after 30 days of treatment.

Factor	Head:Body (%)				Gill:Body (%)			
	Fry		Fingerlings		Fry		Fingerlings	
	LSM	SE	LSM	SE	LSM	SE	LSM	SE
Temp. (T, °C):								
Room temp.	ns 27.29	1.60	ns 26.65	0.79	ns 7.13	0.41	ns 5.95	0.29
Hot temp. (33 °C)	31.19	1.60	26.76	0.79	7.37	0.41	6.00	0.29
Sal. (S, ppt):								
0.1	*	1.92	ns	0.97	**	0.50	ns	0.35
15	25.10	1.92	25.16	0.97	5.98	0.50	5.67	0.35
30	29.63	1.92	26.16	0.97	7.12	0.50	5.86	0.35
	32.98	1.92	28.80	0.97	8.65	0.50	6.39	0.35
Room temp. (T1)*S:								
T1*0.1	ns	2.20	ns	1.32	ns	0.77	ns	0.41
T1*15	24.60	2.20	26.18	1.32	5.53	0.77	5.77	0.41
T1*30	27.60	2.20	24.05	1.32	7.17	0.77	5.52	0.41
	29.67	2.20	29.72	1.32	8.70	0.77	6.55	0.41
Hot temp. (33 °C=T2)*S:								
T2*0.1	ns	3.20	ns	1.42	ns	0.65	ns	0.57
T2*15	25.60	3.20	24.14	1.42	6.43	0.65	5.57	0.57
T2*30	31.67	3.20	28.27	1.42	7.07	0.65	6.19	0.57
	36.30	3.20	27.88	1.42	8.60	0.65	6.24	0.57

Note: Room temperature level was 28.59 °C during fry study while it was 23.91 °C during fingerlings study.
 * = significant, ** =highly significant, ns = not significant.

ter and high temperature but lower in sea water (Table, 4).

The water ions necessary to compensate osmotic loss during the normal physiological processes (ion exchange and respiration) were found to be absorbed by the intestine through active absorption of mono-valent ions. This process begins when the ions concentrations were lower than that in the seawater (Cataldi et al., 1988). On the other hand the desalting of water in the anterior alimentary canal seemed to be an important process in the gastric function as stated by Holstein (1979). According to Kirsch et al. (1975) intestine's ions concentration, after passage through the esophagus, is comparable to that in the water.

General chemical body composition

Neither temperature nor salinity or their interactions had a significant effect on the body chemical constitution of fingerlings (Table, 5). However, raising water temperature decreased the moisture percentage in both fresh and brackish water while it has no effect in seawater. Increasing salinity to the level 15 ppt induced no considerable effect, further increase in salinity showed slight reduction in moisture content. Raising temperature was accompanied with decrease in protein content. Higher protein values were in seawater treatment than the others. Increased water temperature reduced the fat percentage and increased salinity reduced it. Increased both water temperature and salinity raised the ash content.

Table (4): Least square means (LSM) analysis of variance for the effect of salinity and temperature levels on some fingerlings traits after 30 days of treatment.

Factor	Alimentary Tract (%)		Liver (%)	
	LSM	SE	LSM	SE
Temp. (T, °C):	ns		ns	
Room temp.	5.37	0.37	2.04	0.32
Hot temp. (33 °C)	6.46	0.37	2.76	0.32
Sal. (S, ppt):	**		ns	
0.1	5.72	0.45	2.74	0.39
15	7.46	0.45	2.61	0.39
30	4.57	0.45	1.86	0.39
Room temp. (T1)*S:	*		*	
T1*0.1	5.10	0.41	2.61	0.22
T1*15	6.83	0.41	2.15	0.22
T1*30	4.16	0.41	1.38	0.22
Hot temp. (33 °C = T2)*S:	ns		ns	
T2*0.1	6.33	0.80	2.86	0.75
T2*15	8.08	0.80	3.07	0.75
T2*30	4.98	0.80	2.34	0.75

Note: Room temperature level was 28.59 °C during fry study while it was 23.91 °C during fingerlings study.

* = significant, ** = highly significant, ns = not significant.

Table (5): Least square means (LSM) analysis of variance of temperature and salinity levels on major components of the fingerlings body after 30 days of treatment.

Factor	Moisture %		Protein %		Fat %		Ash %	
	LSM	SE	LSM	SE	LSM	SE	LSM	SE
Temp. (T, °C):								
Room temp.	71.70	0.67	62.87	2.07	17.10	1.00	22.52	0.96
Hot temp. (33 °C)	70.56	0.67	61.26	2.24	15.87	1.00	24.66	0.96
	ns		ns		ns		ns	
Sal. (S, ppt):								
0.1	71.30	0.82	61.71	2.54	17.43	1.23	22.04	1.17
15	71.45	0.82	60.51	2.54	16.45	1.23	23.76	1.17
30	70.63	0.82	63.99	2.84	15.58	1.23	24.96	1.17
	ns		ns		ns		ns	
Room temp. (T1)*S:								
T1*0.1	72.61	0.66	61.40	4.46	18.01	1.96	21.25	1.62
T1*15	72.07	0.66	62.26	4.46	17.14	1.96	23.30	1.62
T1*30	70.42	0.66	64.95	4.46	16.17	1.96	23.01	1.62
	ns		ns		ns		ns	
Hot temp. (33 °C=T2)*S:								
T2*0.1	69.99	1.49	62.01	2.11	16.85	1.49	22.83	1.69
T2*15	70.83	1.49	58.77	2.11	15.76	1.49	24.22	1.69
T2*30	70.85	1.49	63.02	2.89	14.99	1.49	26.93	1.69
	ns		ns		ns		ns	

Note: Room temperature level was 28.59 °C during fry study while it was 23.91 °C during fingerlings study.
ns = not significant.

Several studies showed that total protein as a percentage of body weight tended to be relatively constant for a given species (Wandsvik and Jobling, 1982). Gill and Weatherley, 1984, stated that at low temperatures fish tend to have a slightly higher protein content than those at or near their optimum temperature. The percentage of edible protein seemed to vary with the species (Miller and Ballantine, 1974) and fish size (Hickling, 1968). Agouz (1997) found that protein percentage (on dry matter basis) ranged from 53.01 to 61.46 for whole body of male *O. niloticus* fed different diets varying in protein levels and cultured for 20 weeks, the fat percentage ranged from 8.64 to 23.98% for whole body of *O. niloticus* fed different levels of protein diet and cultured for 20 weeks, while the ash % was 17.16-30.40. Fine et al. (1996) reported that common carp *Cyprinus carpio* (45 g/fish) cultured under 26°C had fat% (on dry matter basis) ranging from 10.00 to 10.60%, while in lower temperature (17°C) the fat% ranged from 16.4-17.4%. The ash and protein percentages were not affected by these changes in temperature. Burel et al. (1996) found that the fat percentage (on wet weight basis) of turbot *Scophthalmus maximus* (initial weight 40 g) cultured for 83 days under different temperatures; 8, 11, 14, 17 and 20°C were 6.20, 6.03, 5.89, 5.72 and 4.7 %, respectively, these differences were significant ($P < 0.05$).

Electrolytes content in blood plasma

Table (6) shows the concentrations of the major

electrolytes in the blood plasma of fingerlings under the different treatments.

Sodium concentration was not affected by the temperature but, it was affected by water salinity. It was higher in freshwater and seawater than in brackish water.

No significant effects of salinity, temperature and their interactions were found in plasma Potassium concentrations; however the values were higher in fresh water than in brackish or sea water.

Plasma chloride concentrations were affected by temperature, but not by salinity and their interactions, however its concentrations were higher in brackish water and sea water than in fresh water, the differences were not significant ($p > 0.05$).

Plasma phosphorus concentrations were not affected neither by temperature nor by salinity but, by their interactions. At low temperature (23.9°C) the P^{---} concentration was higher in brackish water than in both fresh and sea water, while under the high temperature (33°C) the concentration of P^{---} increased with the increase of salinity, the highest level was observed in sea water (Table, 6).

Zaghloul (1997) reported that the serum Na^+ ranged from 326.46 to 406.92 mg/dl for fish (*O. niloticus*) collected in summer and from 298.87

Table (6): Least square means (LSM) analysis of variance for the effect of salinity and temperature levels on the concentrations of some electrolytes (mg/dl) in blood plasma of fingerlings after 30 days of treatment.

Factor	Na		K		Cl		P	
	LSM	SE	LSM	SE	LSM	SE	LSM	SE
<u>Temp. (T, °C):</u>								
Room temp.	472.98	6.60	35.56	3.46	249.09	28.90	1.00	0.11
Hot temp. (33 °C)	476.92	7.13	35.65	3.74	342.37	28.90	1.29	0.10
	ns		ns		*		ns	
<u>Sal. (S, ppt):</u>								
0.1	477.46	8.09	40.44	4.24	249.09	36.64	0.88	0.13
15	456.44	8.09	32.00	4.24	337.59	32.77	1.31	0.14
30	490.96	8.04	34.37	4.74	300.50	36.64	1.24	0.13
	*		ns		ns		ns	
<u>Room temp. (T1)*S:</u>								
T1*0.1	471.24	9.26	39.78	6.14	211.07	63.31	0.95	0.19
T1*15	461.79	9.26	37.53	6.14	242.92	51.69	1.26	0.19
T1*30	485.92	9.26	29.36	6.14	293.28	51.69	0.79	0.19
	ns		ns		ns		ns	
<u>Hot temp. (33 °C)(T2)*S:</u>								
T2*0.1	483.68	13.59	41.10	5.82	287.11	40.29	0.81	0.17
T2*15	451.09	13.59	26.46	5.82	432.26	40.29	1.36	0.17
T2*30	496.00	16.64	39.37	7.13	307.73	49.35	1.70	0.20
	ns		ns		ns		*	

Note: Room temperature level was 28.59 °C during fry study while it was 23.91 °C during fingerlings study.
* = significant, ns= not significant.

to 315.87 mg/dl for fish collected in winter from different sites of river Nile in Egypt. The author reported that serum K^+ ranged from 12.90 to 13.71 mg/dl in summer and from 13.99 to 14.76 mg/dl in winter. Burel et al. (1996) found that the blood plasma Cl^- concentrations of turbot *Scophthalmus maximus* (initial weight 40 g/fish) cultured for 60 days under different temperatures; 8, 11, 14, 17 and 20°C were 503.74, 498.78, 498.43, 494.53 and 498.43 mg/dl, respectively with final weight 60.5-77.4 g/fish. In another experiment the same author reported in another experiment that the blood plasma chloride concentrations of the same species at final weigh range 299.1-363.4 g/fish cultured for 60 days under the same temperature were 492.40, 480.70, 489.56 and 476.09 mg/dl, respectively. However, the differences in both experiments were not significant.

In the present study the plasma concentrations of Na^+ , K^+ , Cl^- and P^{---} were 477.46, 40.44, 249.09 and 0.88 mg/dl, respectively, in the case of fresh water (0.1 ppt salinity). The plasma concentrations of the same electrolytes in the case of sea water (30 ppt salinity) were 490.96, 34.37, 300.50 and 1.24 mg/dl, respectively. It is clear that Na^+ , Cl^- and P^{---} increased in plasma of fish with increased water salinity, while K^+ decreased.

Holmes and Donaldson (1969) found that the concentrations of total dissolved electrolytes Na^+ , K^+ , Ca^{++} , Mg^{++} and Cl^- were 367.8, 1.1,

11.2, 3.6 and 400.6 mg/dl, respectively in the blood of *Oncorhynchus tshawytschus* (fresh water, spawning) in fresh water. The respective values in marine water were 389.7, 3.6, 6.6, 2.3 and 489.2 mg/dl, respectively. It is clear from these data that Na^+ , K^+ , and Cl^- increased with increasing salinity, while Ca^{++} and Mg^{++} decreased. Cataldi et al. (1995) found that the concentrations of Na^+ , K^+ and Cl^- (mg/dl) in blood plasma of Italian sturgeon *Acipenser naccari* (fresh water sp.) were 325.19, 14.27 and 466.52 mg/dl, respectively. The concentrations of the same electrolytes, mentioned above, in the same species acclimated to brackish water were 358.64, 11.73 and 579.61 mg/dl, respectively, and in that acclimated to seawater were 340.25, 13.29 and 607.08 mg/dl, respectively. This means successive increase in Cl^- concentrations with successive increase in salinity. The K^+ showed the least concentration in brackish water contrary to the Na^+ which was the highest in brackish water.

Table (7) shows the electrolytes concentrations in fingerlings tissue. The Na^+ concentration was similar in both fresh and seawater, but it was higher than in brackish water. Both K^+ and Ca^+ concentrations were not affected by salinity or temperature although affected by their interactions at the high temperature (33°C). No effect was found for salinity or temperature as well as their interactions on Mg^{++} and P^{---} concentrations. The concentrations of Na^+ , K^+ , Ca^{++} ,

Table (7): Least square (LSM) analysis of variance for the effect of salinity and temperature levels on the concentrations of some electrolytes in fingerlings body tissues (mg/g dried body) after 30 days of treatment.

Factor	Na		K		Ca		Mg		P	
	LSM	SE	LSM	SE	LSM	SE	LSM	SE	LSM	SE
Temp. (T, °C): Room temp. Hot temp. (33 °C)	ns		ns		ns		ns		ns	
	16.96	2.27	0.25	0.03	42.68	0.78	2.35	0.06	3.36	0.01
	17.21	2.93	0.16	0.03	42.30	0.78	2.33	0.06	3.37	0.01
Sal. (S. ppt): 0.1 15 30	ns		ns		ns		ns		ns	
	22.95	3.93	0.21	0.04	41.18	0.95	2.27	0.07	3.36	0.01
	10.20	2.78	0.22	0.04	44.20	0.95	2.44	0.07	3.38	0.01
	18.10	2.78	0.19	0.04	42.09	0.95	2.31	0.07	3.38	0.01
Room temp. (T1) T1*0.1 T1*15 T1*30	ns		ns		ns		ns		ns	
	25.11	4.69	0.28	0.07	40.37	1.86	2.20	0.13	3.35	0.01
	9.74	4.69	0.22	0.07	44.38	1.86	2.54	0.13	3.38	0.01
	16.02	4.69	0.23	0.07	43.29	1.86	2.30	0.13	3.37	0.01
Hot temp. (33 °C) T2*0.1 T2*15 T2*30	ns		**		**		ns		ns	
	20.78	4.07	0.13	0.01	41.98	0.42	2.35	0.05	3.36	0.01
	10.66	2.35	0.21	0.01	44.02	0.42	2.33	0.05	3.37	0.01
	20.18	2.35	0.14	0.01	40.89	0.42	2.31	0.05	3.38	0.01

Note: Room temperature level was 28.59 °C during fry study while it was 23.91 °C during fingerlings study.
** =highly significant (P<0.01), ns = not significant.

Table (8): Blood plasma concentration of sodium (Na), potassium (K), chloride (Cl) and aldosterone hormone of fingerlings under different conditions of temperature and salinity after 30 days of treatment

Item	Fresh water (0.1 ppt)		Brackish water (15 ppt)		Sea water (30 ppt)	
	T1 (23.91 °C)	T2 (33 °C) Change %	T3 (23.91 °C)	T4 (33 °C) Change %	T5 (23.91 °C)	T6 (33 °C) Change %
Na ⁺ (mg/dl)	421.24	483.68 15.00	461.79	451.09 -2.00	485.92	496.00 2.00
K ⁺ (mg/dl)	39.78	41.10 3.00	37.52	26.46 -29.00	29.36	39.37 34.00
Cl ⁻ (mg/dl)	211.07	287.15 36.00	242.90	432.28 78.00	293.28	307.71 5.00
Aldos. (pg/ml)	0.34	0.57 68.00	0.56	0.39 -30.00	0.46	0.64 39.00

% Percentage of change (+ or -) in concentration of electrolytes and aldosterone due to increased temperature

Mg⁺⁺ and P⁻⁻⁻ in the fish tissue of male *O. niloticus* in fresh water (0.1 ppt) were 22.95, 0.21, 41.18, 2.27 3.36 mg/g (dried body), respectively. In sea water (30 ppt) the values were 18.10, 0.19, 42.09 and 2.31 and 3.38 mg/g (dried body), respectively. It is clear that there was a slight decrease in Na⁺ and K⁺ levels with increasing salinity, while Ca⁺⁺, Mg⁺⁺ and P⁻⁻⁻ levels were increased with increasing salinity.

Agouz (1997) reported that the Ca and P % in the whole body of male *O. niloticus* fish, fed on different diets varying in level of protein, at the end of the experimental period (20 week) (on dry matter basis) ranged from 0.06 to 0.13 % for calcium and from 0.03 to 0.06% for phosphorus.

Aldosterone / electrolyte relation

A preliminary assessment of the aldosterone concentration in relation with the major electrolytes (Na⁺, K⁺ and Cl⁻) in blood plasma of the fingerlings is presented in table (8). Generally it is clear from the table that plasma Na⁺ and Cl⁻ levels increased with increasing temperature or salinity. The aldosterone concentrations showed a trend of change by these environmental conditions almost equal to the % of changes in K⁺ concentration in case of brackish water and sea water only. In the case of brackish water the table shows drop in Na⁺, K⁺ and Aldosterone concentrations by (-2), (-29) and (-30)% opposite to enormous increase in Cl⁻ (+78%) due to increased temperature. These drops due to salinity of 15 ppt were not

found with lower or higher salinity, a result which needs further elucidation.

This study could be concluded that male Nile tilapia (*O. niloticus*) can tolerate considerable range of environmental conditions, such as salinity and temperature. Fish grew best in brackish water. In sea water its survivability dropped and its growth decreased. The eco-physiological response of tilapia is affected harshly by conditions hot temperature (33°C).

REFERENCES

- Agouz, H. M. A., 1997: Performance of fish fed diets varying in protein levels. M. Sc. Thesis, Agric. Fac. Cairo univ. Egypt.
- Assem, H., 1995: Optimum salinity for the growth of the teleost, *Oreochromis niloticus* confirmed by the saturation kinetic model for physiological response. J. Egypt Ger. Soc. Zool., 16: (A) 41-52.
- Association of Official Analytical Chemists (AOAC) 1990: Official Methods of Analysis. Association official agriculture chemists (15th ed). Washington D.C. USA.
- Avella, M., Berhaut, J. and Bornancin, M., 1993: Salinity tolerance of two tropical fishes, *Oreochromis aureus*, and *O. niloticus*, 1- Biochemical and morphological changes in the gill epithelium. J. Fish Biology, 42:243-254.
- Boyd, E. C., 1979: Water quality in warm water fish ponds. Alabama Agricultural Experimental Station, Auburn, Alabama, USA.

- Boyd, E. C., 1990: Water quality in ponds for aquaculture. Alabama Agricultural Experimental Station, Auburn Univ. Lowell T. Forbush, Director Auburn Univ. Alabama, USA.
- Burel, C., and Person-Le-Ruyet, J., Gaumet, F., LeRoux, A., Sèveke, and A. Boeuf, G., 1996: Effects of temperature on growth and metabolism in juvenile turbot. *J. fish Biol.*, 49:678-692.
- Cataldi, E.; Crosetti, D.; Conte, G.; Dovidio, D. and Cataudella, S., 1988: Morphological changes in the oesophageal epithelium during adaptation to salinities in *Oreochromis mossambicus*, *O. niloticus* and their hybrid. *J. Fish Biology* 32, 191-196.
- Cataldi, E.; Ciccotti, E.; Di Marco, P.; DiSanto, O.; Bronzi, P. and Cataudella, S., 1995: Acclimation trails of juvenile Italian sturgeon to different salinities: Morphophysiological descriptors. *J. Fish Biology* 47, 609-618.
- Daoust, P. Y.; Wobesor, G. and Newstead, J. D., 1984: Acute pathological effects of inorganic mercury and copper in gills of rainbow trout. *Vet Pathol.*, Vol., 21 (1)93-101.
- Doyle, W. and Gorecki, D., 1961: The so-called chloride cell of the fish gill. *Physiol. Zool*, 34, 81-85.
- Egyptian laws no. 124, 1982: Official newspaper at 25 August, 1983. volume (34) followed by the special law no. 124 in 1983 -for fish catch, aquatic living and fish farms- and its list (ministerial decision no. 303 in 1987 which published in El Wakkea El Massrya newspaper, at 28 April 1987, vol (99) p.2. The public organization for official presses, Cairo, 1993.
- El-Eraky, W.; Saleh, G.; El-Bouhy, Z. and Hassanein, E. L., 1988: Relation between water temperature and two levels of protein on growth fingerlings (*Tilapia nilotica*). *Zagazig Vet. J.* Vol. XVI No. 2(B) pp. 210-219.
- Ellis, M. M., 1937: Detection and measurement of stream pollution. U.S. Bureau of fish, Bull. 22:267-437.
- Fine, M.; Zilberg, D.; Cohen, Z.; Degani, G.; Moav, B. and Gertler, A. 1996: The effect of dietary protein level, water temperature and growth hormone administration on growth and metabolism in the common carp (*Cyprinus carpio*). *Comp. Biochem. Physiol.* Vol. 114A, No. 1, pp. 35-42.
- Galat, D. L.; Post, G.; Kefee, T. J. and Bouck, G. R., 1985: Histological changes in the gill, kidney and liver of Lohantan cutthroat trout, *Salmo clarki henshawi*, living in lakes of different salinity-alkalinity. *J. Fish. Biol.*, 533-552.
- Gill, H. H. and Weatherly, A. H., 1984: Protein, lipid and caloric contents of bluntnose minnow, *Pimephales notatus* Rafinesque, during growth at different temperature. *J. Fish Biology*, 25, 491-500.
- HACH, 1982: Hach Company, Fishries/Farming Manual Test Kit Model FF-2, Loveland, Colorado, USA.
- Hickling, C. F., 1968: The fish farming. Pergamon Press, London: 88 p. (Book).
- Holmes, W. N. and Donaldson, E. M. 1969: the body compartments and distribution of electrolytes. In *Fish Physiology*. Vol, I: Excretion, Ion Regulation and metabolism. (W.S. Hoar & D.J. Randall, eds.), pp. 1-89. New York: Academic Press.
- Holstein, B., 1979: Gastric acid secretion and drinking in the Atlantic cod (*Gadus morhua*) during acidic or hyper-osmotic perfusion of the intestine. *Acta Physiol. Scand.* 106, 257-265.

- Ibrahim, I. H., 2000: Genetic studies on some tilapia species under different salinity conditions. M. Sc. Thesis, Faculty of Agriculture, Ain Shams Univ.
- Kirsch, R.; Guiner, D. and Means, R., 1975: L'équilibre hydrique de l'anguille Européenne (*Anguilla anguilla* L.). Etude du rôle de l'oesophage dans l'utilisation de l'eau de boisson et étude de la perméabilité osmotique branchiale. *J. Physiol.*, Paris. 70, 605-626.
- Likongwe, J. S.; Stecko, T. D.; Stauffer, J. R. and Carline, R. F., 1996: Combined effects of water temperature and salinity on growth and feed utilization of juvenile Nile tilapia *Oreochromis niloticus* L. *Aquaculture*, 146:37-46.
- Miller, J. W. and Ballantine, D. F., 1974: Product quality of cultured fish. FAO. *Aquaculture Bull.*, 6, (4):8.
- Morsy, M. S., 1994: Effects of the crossing and some environmental factors on growth performance of Tilapia fish. Ph. D. Thesis. Faculty of Agriculture, Cairo Univ.
- Norrgrén, L.; Wicklund Glynn, A. and Malmberg, O. 1991: Accumulation and effects of aluminium in the minnow (*Phoxinus phoxinus* L.) at different pH levels. *J. Fish Biology* 39, 833-847.
- Nour, A. M., El-Ebiary, E. H.; Badawy, A. M. and Abo-Elwafa, A. M., 1996: The effect of over wintering and/or nutrition on growth performance, feed utilization and development of gonads of Tilapia sp. *Alex. J. Agric. Res.* 41(2):111-121.
- Payne, A. I., 1983: Estuarine and salt tolerance Tilapias. . pp 534-543 in proceeding, International Symposium on Tilapia in Aquaculture, Phalastein, Tel Aviv Univ.
- Prunet, p. and Bornancin, M., 1989: Physiology of salinity tolerance in tilapia: An uptake on basis and applied aspects. *Aquatic Living Resources* 2, 91-97.
- SAS, 1989-1996: Statistical Analysis System. The SAS system for Windows Release 6.12. Inc., Cary, NC, 27513, USA.
- Stickney, R. R., 1986: Tilapia tolerance of saline waters: a review. *Prog. Fish. Cult.*, 48(3):161-167.
- Suresh, A. V. and Kwei-Lin, C., 1993: Tilapia culture in saline water: a review. CRSP Research Reports Pond Dynamics/ Aquaculture Collaborative Research Support Program, Office of International Research and Development, Oregon State Univ., Snell Hall 400, Corvallis, Oregon 97331-1641 USA.
- Swingle, H. S., 1969: Methods of analysis for water. Organic matter and pond bottom soils used in fisheries research. Auburn Univ., Auburn, Alabama, USA. 119 pp.
- Verdegem, M. C. J.; Hillbrands, A. D. and Boon, J. H., 1997: Influence of salinity and dietary composition on blood parameters values of hybrid red tilapia, *Oreochromis niloticus* (Linnaeus) x *O. mossambicus* (Peters). *Aquaculture Research*, 28:453-459.
- Wandsvik, A. and Jobling, M., 1982: Observations on growth rates of arctic charr, *Salvelinus alpinus* reared at low temperature. *J. Fish Biology*, 20, 689-699.
- Watanabe, W. O., Kuo, C. M. and Huang, M. C., 1985: Salinity tolerance of Nile tilapia fry (*Oreochromis niloticus*) spawned and hatched at various salinities. *Aquaculture*, 48:159-176.
- Zaghloul, H. K., 1997: Studies on the effect of water pollution along different sites of the river Nile on the survival and production of some fresh water fishes. Ph. D. Thesis. Faculty of Science, Cairo Univ. Giza, Egypt.