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Effect of Different Salinity Levels of Lake Qaroun Water on Growth Performance, Feed Utilization and Histological Changes on Liver and Gills of Rabbitfish Juvenile (*Siganus rivulatus*)

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Abstract : This study was carried out to determine the optimum salinity level on growth of rabbit fish juvenile (*Siganus rivulatus*(and clear the effect of pollution in Lake Qaroun water on its liver and gills. This work was begun at 15/8/2015 and ended 12/11/2015 for 90 days. The average initial weight (W_1) of investigated juvenile was 0.948±0.124 g. This trial was consisted of four treatments for four of different water salinity levels from Lake Qaroun. The first treatment, juvenile was reared in Lake Qaroun water have salinity (33 part per thousand ppt), second (25 ppt), third (15 ppt) and fourth (5 ppt). The juvenile was fed twice daily on diet have 36.44% CP, the feeding rate was 5% of fish body weight, the water exchange rate was 500 liter every two days and juvenile were stocked at 40 fish/ m^3 (120 fish/ pond). The results showed that, the highest final weight (W_2), total weight gain (TG) and average daily gain (ADG) were found of juvenile that reared in Qaroun Lake water(first treatment)and these parameters did not significantly differ with the other treatments. Also, the best feed conversion ratio (FCR) and feed conversion efficiency (FCE) were found with juvenile that reared in Qaroun Lake water (first treatment), followed by the treatment with 25ppt. whereas, histological studies was evident that severe damage of hepatocytes and gills with both high and low salinity was found as result of Lake Qaroun pollution and fish resistance of osmoregulatory stress in low salinity water. But osmoregulatory stress was more negatively effect on growth rate than Lake Qaroun water condition.
Key words: Rabbitfish, Lake Qaroun, Salinity, Histological and Pollution

Introduction

Rabbitfish *Siganus rivulatus* is a potential candidate for warm water marine aquaculture diversification[1, 2].Rabbitfish belong to the genus Siganus of the family siganidae[3]. Siganids are herbivorous marine and brackishwater fishes that are found throughout the indo west pacific[4] and the more common species are the objects of traditional subsistence and commercial fisheries throughout this region. There has been interest in the culture of these fishes in ponds or cages in several areas[5].

Rabbitfish high tolerances to environmental factors, tolerance to rough handling and crowding so can be stocked at very high density, fast growth and possibility to obtain their seeds from the wild or by artificial propagation[6].

Rabbitfish are considered to be excellent food fish in many parts of the world especially in the eastern Mediterranean and indo-pacific regions[1].

Egypt production of rabbitfish was about 1363 ton in 2014, Mediterranean Sea took part in 822 ton production, Red Sea (466 ton) and lakes (75 ton) according to[7]. In previous years, Lake Qaroun was developed by rabbitfish fry and the first production appeared in 2010 and reached this production about (1 ton), a maximum rabbitfish production of Lake Qaroun about 5 ton obtained in 2011, while rabbitfish production of Lake Qaroun in 2014 was 2 ton according to[7].

Lake Qaroun is a closed saline basin lying in the lowest northwest part of El-Fayoum depression-Egypt, between longitudes 30º 24' and 30º 50' E and latitudes 29º24' and 29º33' N. It has an elongated rectangular shape with average dimensions 45 km length, 5.7 km width and 4.2 m depth in average[8].

The salinity refers to the quantity of solids per gram, dissolved per kilogram of water[9]. Salinity affects various physiological processes in aquatic animals such as metabolism, osmoregulation and biorhythm[10].

Salinity is an important environmental variable for estuarine organisms and its many important physiological and ecological effects have been reviewed by[11]. Because salinity is variable in estuarine regions and osmoregulation is an energy demanding process, certain ambient salinities might help maximize growth and/or reproduction by decreasing osmoregulatory energy expenditure. Thus, the ability of estuarine fishes to deal with fluctuating salinity constitutes an important factor when trying to maximize fish growth and reproduction in captivity[12]. In general, fish at the extremes of their salinity tolerance range often exceed their ability to osmoregulate. The changes in salinity that are outside of optimal salinity range cause disturbance in physiological processes in fish. This leads to the higher production of cortisol levels in the blood[13]. The plasma cortisol level was also determined in the juveniles reared in different salinities.

Also, Lake Qaroun is a closed ecosystem, and as a result of extensive evaporation of water, the accumulation of chemical pollutants (heavy metals, pesticides and other pollutants) is expected to increase annually in all its components (e.g. water and fish) and change their quality and affect their aquatic life[14].The increasing pollution of water resources in Lake Qaroun and the consequent effects on aquatic environment and human health is an issue of great concern.

Previous studies reported that Lake Qaroun components are polluted with heavy metals[14]and with a wide variety of pesticides (e.g. lindane, aldrin, some DDT analogues, malathion) [15.]

Also, it was reported that the drainage waters discharged into the lake are high in solids, nutrients, pesticides, heavy metals and organics[16]. Moreover, a remarkable increase in the bacterial indicators of sewage pollution (total coliforms, faecal coliforms and faecal streptococci) in the lake was recorded[17].

This study aimed to determine the optimum salinity level of rabbitfish juvenile growth and clear the effect of Lake Qaroun water pollution on liver and gills of rabbitfish juvenile after rearing period for 90 days under different water salinity levels.

Materials and methods

Rabbitfish (*Siganus rivulatus*) juveniles were obtained from (Mediterranean Sea) Maadia region-Behaira Governorate- Egypt and translated to Fish Research Station in Shakshouk, Fayoum Governorate, National Institute of Oceanography and Fisheries (NIOF), Egypt for about 5hr. at August, 2015.The average initial weight (W_1) for these juveniles was 0.948 g ± 0.124 and an average initial length was 3.97 cm ± 0.200.

Fish acclimatization and feeding

Fish were acclimatized to be adapted to water salinity of Lake Qaroun level, 25ppt, 15ppt and 5ppt according to the treatments for one week (Lake Qaroun water were diluted by fresh water to 25, 15 and 5ppt) before size sorting and removal of large and small fish. One artificial diet was formulated by hand and used in this study, the diet formulated to be almost containing 36% crude protein (Table, 1). Feed was offered by hand.

Table (1) Ingredients and a proximate chemical analysis of the experimental diet

Ingredients	**(g/100 g)**
Fish meal (72%CP)	22
Extruded full fat Soybean meal (37% CP)	43
Wheat bran fine	28
Fish oil	4
Super yeast	1
Starch	1.7
Vit. & Min. & premix	0.3
Total	100
Chemical analysis % on Dry matter basis	
Moisture (M)	6.94
Dry matter (DM)	93.06
Crude protein (CP)	36.44
Ether extract (EE)	13.78
Crude fiber (CF)	3.10
Nitrogen free extract (NFE)	39.02
Ash	7.66
Gross energy (GE, Kcal/g)*	5.09

Notice: -Chemical analysis was determined according to[18] and NFE was calculated by difference.
* Calculated according to[19].

Experimental ponds and trial design

This trial began at 15/8/2015; ponds that used in this work were made from concrete, eight concrete ponds were used in this work. The dimensions of each pond were 2.3 m length 1.6 m width and 1.25 m height and the water volume of each pond was 3 m^3. Also, eight small concrete ponds (m^3) above experimental ponds were used to blend or mix salt water with fresh water before water exchange operation.

This trial consisted of four treatments including four of different water salinity. the first treatment: fish reared in water salt of Lake Qaroun (33 ppt), the second treatment: fish were reared in 25 ppt, the third treatment: fish were reared in 15 ppt and the fourth treatment: fish were reared in 5ppt. Fish fed twice daily on diet (36.44% CP) (Table, 1), feeding rate was 5% biomass, the water exchange rate was 500 liter every two day and fish were stocked at 40 fish/m^3 (120 fish/pond). This trial ended 12/11/2015, (90 days)

Running water system in experimental units.

The system contained on water pump, sand filter unit and two large tanks (10000 liter/tank) used to store the water at a point between the water source (Lake Qaroun water) and experimental units. The water pump was raising the water from water source to the sand filter unit then to the large tanks and hence to experimental units.

Aeration system in experimental units.

The system contained on Blower connected to a network of plastic pipes, this pipes transport the air to each experimental unit, the air was controlled by tap of each pond or tank and the air diffusers was used to distribute of air in all experimental unit trends.

Water quality:

Water quality of water source (Lake Qaroun water)

The water quality of water source was measured after arriving to experimental units, water temperature, pH, salinity, electrical conductivity (EC), total suspended solids (T.S.S), some of elements and some of heavy metals, microbiologic count (Table, 2) throughout the experimental period.

Water quality in the experimental units (ponds).

The water quality of the indoor ponds laboratory (in the experimental units) were measured of each treatment. Temperature, pH, salinity and EC were measured daily at 1pm by centigrade thermometer, Orion digital pH meter model 201, Refractometer (VITAL Sine SR-6, China), Conductivity meter model (YSI.SCT-33). Dissolved oxygen (DO) was measured every week by oxygen meter (Cole Parmer model 5946).Nitrite, Nitrate and total ammonia were measured every two weeks by the chemical methods according to[20, 21].

The metals such as magnesium, potassium, manganese, phosphorus, zinc, copper, iron, nickel, boron, cadmium and lead were measured by using Inductively Coupled Plasma Emission Spectrometer (ICP) (ICAP-6300 Duo). Microbial count conducted according to standard methods for the examination of water and waste wáter[22] (Table, 3).

Table (2) Water quality in Lake Qaroun, Average water temperature, water pH, water EC, salinity, water T.S.S, Some of elements, some of heavy metals and microbiologic account during the period from 25/7/2015 to 26/11/2015.

Parameters	Measurement	Parameters	Measurement
Temperature, °C	26.10	Sulfate (SO_4), µg/l	913000.00
pH	8.25	Potassium (K), µg/l	298000.00
EC*, mS/cm*	39.9	Phosphorus (P), µg/l	142.50
Salinity, (ppt)	33.300	Boron (B), µg/l	2660.00
T.S.S, µg/l	228000.00	Iron (Fe), µg/l	97.00
Chloride (Cl), µg/l	11879000.400	Lead (Pb), µg/l	2.50
Calcium (Ca), µg/l	441000.00	Nickel (Ni), µg/l	6.50
Sodium (Na), µg/l	7035000.00	Cadmium (Cd), µg/l	4.10
Magnesium (Mg), µg/l	301000.00	Zinc (Zn), µg/l	52.00
Carbonate (CO_3), µg/l	24000.00	Manganese (Mn), µg/l	8.00
Bicarbonate (HCO_3), µg/l	256000.30	Copper (Cu), µg/l	9.00
Microbial Count			
Parameters		**Measurement**	
total coliforms		290 per 100 ml	
fecal coliforms		240 per100 ml	
fecal streptococci		460 per100 ml	

*, mS/cm, millisiemens/centimeter.

Table (3) Mean (±SE) of water quality parameters in the experimental units

Items	Treatments				SED*
	Lake Qaroun Salinity	25ppt	15ppt	5ppt	
Temperature (°C)	26.116±0.250	25.924±0.235	26.005±0.232	25.883±0.242	0.353
pH	8.184±0.121^a	8.062±0.138^b	8.106±0.165^b	8.244±0.206^a	0.022
Salinity, ‰	33.790±0.278^a	24.050±0.297^b	14.630±0.279^c	5.550±0.082^d	0.044
EC, mS/cm*	47.300±0.129^a	38.110±0.096^b	28.005±0.632^c	11.060±0.013^d	0.710
DO, mg/l	7.875±1.12b	7.230±0.170^d	7.730±0.470^c	7.985±0.215^a	0.014
Nitrite, mg/l	0.079±0.125^b	0.050±0.016^c	0.119±0.040^a	0.027±0.005^d	0.007
Nitrate, mg/l	0.120±0.012^c	0.394±0.297^b	0.537±0.135^a	0.116±0.035^d	0.001
Total ammonia, mg/l	0.527±0.160^a	0.387±0.121^b	0.319±0.114^c	0.275±0.078^d	0.001

(a, b, c and d) Average in the same row having different superscripts significantly different at ($P \leq 0.05$).
*, mS/cm, millisiemens/centimeter.
**, SED is the standard error of difference.

Measurements of growth performance and some of the internal organs

Final condition index (CI_2), Total weight gain (TG), average daily gain (ADG), Relative growth rate (RGR), specific growth rate (SGR), survival rate (SR), hepatosomatic index (HSI) and viscerosomatic index (VSI).

These parameters were calculated according to the following equations:

($CI_{2,\ g/cm^3}$) = (W_2/ $L^3{}_2$) × 100 whereas L_2: is the final length of fish in cm, TG, g = final weight (W_2) – initial weight (W_1), ADG, g/day = average weight gain, g / experimental period, day, RGR, % = [(W_2 - W_1) / W_1] × 100, SGR, % /day = [(ln W_2-ln W_1)/t] × 100 whereas ln: is the natural log. And t: is the time in days, SR% = (Number of fish at end/ Number of fish at start) × 100, (HSI, %) = (liver weight/body weight) ×100 and (VSI, %) = (weight of viscera and associated fat tissue/body weight) ×100.

Measurements of feed utilization efficiency

Feed intake g/ fish (FI), feed conversion ratio (FCR), feed conversion efficiency (FCE), protein efficiency ratio (PER), protein productive value (PPV), energy efficiency ratio (EER), energy productive value (EPV) and lipid retention (LR).

These parameters were calculated according to the following equations:

FI, g/fish feed intake during the trial period/ the final number of fish for this trial, FCR = feed intake, g / weight gain, g., FCE, % = (weight gain, g./ feed intake, g) × 100, PER= Weight gain, g/ Protein intake, g., PPV, % = (Retained protein, g/ Protein intake, g) × 100, EER = Weight gain, g/ Energy intake, Kcal, EPV, % = (Retained Energy, Kcal/ Energy intake, Kcal) × 100, LR, % = (Retained lipid, g/ lipid intake, g) × 100.

Chemical analysis of feeds and whole fish body

The conversional chemical analysis of diet and whole body fish samples were carried out as described by[18] and Gross energy (GE) was estimated for formulated diets the factors 5.64, 9.44 and 4.11 Kcal/g for CP, EE and carbohydrates respectively were used[19], for fish 5.5 and 9.5 Kcal/g for protein and fat respectively[23].

Histopathological examination

Samples were fixed in alcoholic Bouin's solution for 48 hours. The specimens were dehydrated in ascending concentrations of ethyl alcohol, cleared in xylol and embedded in paraffin wax. Sagittal sections were cut at 4 to 6 microns and at least 10 slides from each region were prepared. These were stained with Harri's haematoxylin and subsequently counter stain with eosin (H&E). Finally, the slides were microscopically examined to identify the histo-pathological features, then photographed and described.

Statistical analysis:

The analysis of variance and LSD of Duncan Waller were used to compare treatment means. Data were analyzed using stat graphic package software[24]SPSS Inc. Released 2007. SPSS for Windows, Version 16.0. Level of significant was 0.05.

Results and discussion

Growth performance

Results of growth performance parameters of rabbitfish juveniles which were reared in different water salinity were shown in Table (4). The statistical analysis appeared significant differences between the treatments in L_2, CI_2, W_2, TG, ADG, RGR, and SGR. The fish were reared at 25 ppt salinity level had the highest L_2 followed by Lake Qaroun salinity but the lowest L_2achieved with the 15 and 5 ppt salinity levels. The lowest CI_2 was obtained with 25 ppt while the other treatments did not significantly differ in CI_2. The highest W_2, TG and ADG were achieved by the fish were reared at Lake Qaroun salinity and this parameters did not significantly differ with the other treatments. But 25 ppt salinity level was the better in these parameters than 5ppt and 15ppt salinity levels.

The highest RGR was achieved by Lake Qaroun salinity followed by 25ppt and 5ppt salinity levels. While, the 15ppt was the lowest.

SGR did not significantly differ between the treatments but the fish were reared at Lake Qaroun salinity were the best in SGR followed by the 25ppt and 5ppt, while the 15 ppt level had the lowest SGR

The result also showed no significant differences were found between the treatments in survival rate (SR) but 25ppt salinity level had the best SR followed by Lake Qaroun salinity and 15 ppt while 5ppt was the lowest. This result differed with[25]found that, survival of *S. rivulatus* was 100 % when fish were reared at10–35 ppt salinities and also[26] found that, survival of *S. guttatus* was 100 % when fish were reared at 5–30% salinities.

No significant differences were found between the treatments in HSI. In relation to VSI, it significantly differed between the treatments, the 5ppt had the highest VSI followed by Lake Qaroun salinity and 25ppt but the 15ppt salinity level was the lowest.

The results revealed that, the fish were reared at Lake Qaroun was the best in growth performance parameters and the 25ppt salinity levels was the best survival rate. It can be observed that, a decrease of salinity negatively affected on growth performance parameters and survival rate thus referring a preference of the fish for high salinity. This is in agreement with[1] who reported, rabbitfish *Siganus rivulatus* is a potential candidate for warm water marine aquaculture diversification.[25]reported that, *S. rivulatus* salinity preferences are not documented, but this species is well established in the Indian Ocean where salinity is approximately 35 ppt, in the eastern Mediterranean where salinity is between 37 and 39 ppt, and in the red sea where salinity is between 39 and 41 ppt and indicated that, the rabbitfish *Siganus rivulatus* perform better at 35 ppt than at other salinities. In the same trend,[27] found that, final body weight and SGR of juvenile Lebranch mullet *Mugilliza* were significantly affected by water salinity. Whereas, they increased with increasing salinity, being significantly higher in salinity 24‰ than in fresh water ($p<0.05$).

Table (4) Effect of water salinity level on growth performance of rabbitfish (*Siganus rivulatus*) juvenile.

Items	Treatments				SED*
	Lake Qaroun salinity	25ppt	15ppt	5ppt	
Initial weight (w_1), g	0.948	0.948	0.948	0.948	-
Final length (L_2), cm	7.78^b	7.98^a	7.47^c	7.47^c	0.014
Final condition index (CI_2), gcm^{-3}	1.22^a	1.04^b	1.23^a	1.25^a	0.031
Final weight (W_2), g	5.75^a	5.30^b	5.15^b	5.22^b	0.077
Total weight gain (TG), g	4.80^a	4.36^b	4.20^b	4.28^b	0.083
Average daily gain (ADG), g/day	0.053^a	0.048^b	0.046^b	0.47^b	0.001
Relative growth rate (RGR), %	510.63^a	463.82^b	446.80^d	455.30^c	0.018
Specific growth rate (SGR/day, %)	2.01	1.92	1.89	1.90	0.070
Survival rate (SR, %)	86.25	87.5	68.75	50.00	17.681
Some of the internal organs parameters					
Hepatosomatic index (HSI, %)	1.55	1.70	1.64	1.64	0.070
Viscerosomatic index (VSI, %)	19.40^b	17.47^c	16.27^d	19.64^a	0.070

(a b, c and d) Average in the same row having different superscripts significantly different at level ($P\leq0.05$).
*, SED is the standard error of difference.

In addition to, [28]affirmed that, Florida red tilapia cultured at 36 ppt. And the optimum salinity for growth is 30 ppt for the Amarillo snapper, *Lutjanus argentiventris*[29].

The results confirmed that, *Siganus rivulatus* achieved the best growth performance at the highest salinity (Lake Qaroun salinity) this may be due to Na+K+-ATPase activity (NKA) was lowest in fish reared in Lake Qaroun salinity and higher in fish reared at salinities lower and, consequently to a higher energy expenditure associated with osmoregulation. Whereas, the osmoregulatory energy demand is usually directly correlated to gill NKA or the lower growth observed in rabbitfish maintained in low salinity seems to be related to a higher energy cost associated with osmoregulation under this environmental condition. This was in agreement with[25, 27] and this similar to[30] further observed a low density of chloride cells in branchial epithelium and the lowest branchial Na+-K+-ATPase activity in 50% seawater, suggesting that food energy was spared for body growth rather than osmoregulation. Moreover, in optimum salinity, less energy spend for osmoregulation and thus, it can be conserved for the metabolic process, thus increasing the growth of fish[31]. Any salinity range that is out of the optimal level will lead to an increase in energy.

On the other hand, [32]found that salinity did not affect the growth of *Argyrosomus japonicas* juvenile, the size of individuals was 9.4 g weight and the salinity treatments was 5-45 practical salinity unit (psu). The final weight and the specific growth rate of juvenile *Siganus guttatus* were significantly greater at 10 % than fish in all the treatments.

These results were shown in table (4) indicated that, the fish reared at 5, 15 and 25ppt salinity levels did not differ in W_2, TG and ADG. This confirmed that, rabbitfish can life and were possibility reared at brackish water as it was reported by[4] but, the growth performance of rabbitfish increased with an increase of salinity level over 25 ppt.

From these results it can be found that, insignificant differences between the fish were reared in 5 ppt and 15 ppt in w_2, TG, ADG and SGR, however, the growth performance values was higher with 5ppt than 15 ppt. Also, the 15 ppt salinity level had the lowest value of growth performance, this may be attributed to water NO_2 and NO_3 of 15 ppt was relatively higher than 5ppt and there a strong effect of medium salinity on gill NKA activity[33, 34]. Also a decrease of survival rate for fish were reared at 5ppt led to reduce of fish number at this treatment, this may be resulted in improve of growth performance, according to[35].

On the contrary, many of researches were conducted on other fish species and affirmed that, the optimum salinity level for growth was 15 ppt, such as[36] who reported that, the optimum salinity for Hybrid tiger grouper (TGGG) juvenile rearing was in the range of 10 ppt to 20 ppt. Also, [37]suggest that, growth of Sparussarba enhancement at 15 ppt.

In general, rabbitfish juvenile can be reared at 5 ppt, 15 ppt , 25 ppt and Lake Qaroun salinity but the high salinity level (Lake Qaroun Salinity) was the best compared with the other salinity levels in growth performance and survival rate. In addition to, rabbitfish *Siganus rivulatus* is a euryhaline herbivorous fish[4]. Whereas, euryhaline fish can live in a wide range of environmental salinities due to their ability to synthesize new salt transporting proteins as they move from salt to fresh water and vice versa[38].

Feed utilization efficiency

The feed utilization efficiency of rabbitfish juvenile were reared under different water salinity were shown in Table (5) There were significant differences between the treatments in all feed utilization parameters. The highest FI was achieved by the fish were reared at 15 ppt salinity level followed by 5 ppt. While, the lowest FI was achieved by the fish were reared at 25ppt and Lake Qaroun salinity level. The best FCR and FCE were achieved with fish were reared at Lake Qaroun salinity level, followed by 25ppt and 5ppt. while, the worst FCR and FCE were achieved with 15 ppt salinity level.

The fish were reared at Lake Qaroun salinity had the highest PER, PPV, EER, EPV and LR followed by 25 ppt in comparison with the fish were reared at 5 ppt and 15ppt salinity level. EER, EPV and LR values were higher with 5 ppt than 15ppt but PPV was the lower with 15 ppt than 5 ppt. This indicates that, Lake Qaroun salinity and 25 ppt are suitable for culturing *Siganus rivulatus*. Low salinity may supress stress and resulting in poor the feed efficiency.

These results were in similar with [39]who found that, the lowest FCR value of tilapia hybrid was achieved by 32ppt salinity level. They interpreted this result and reported may be partially explained by the assumption that growth of euryhaline teleosts is greatest at salinities near the iso-osmotic level. And also, [40]found that, the lowest FCR value of juvenile Black Sea Bass was obtained by 30 ppt salinity level. Moreover, [41]revealed that, inland saline groundwater of high salinity can be profitably utilized for the culture of milkfish. Highest growth and feed utilization were observed when the fish were cultured at 25‰ salinity.

Table (5) Effect of water salinity level on feed utilization efficiency of rabbitfish (*Siganus rivulatus*) juvenile.

Items	Treatments				SED*
	Lake Qaroun salinity	25ppt	15ppt	5ppt	
Feed intake (FI), g/ fish	12.93[c]	12.87[c]	16.98[a]	16.20[b]	0.070
Feed conversion ratio (FCR)	2.69[d]	2.95[c]	4.01[a]	3.78[b]	0.014
Feed conversion efficiency (FCE, %)	37.12[a]	33.87[b]	24.73[d]	26.41[c]	0.024
Protein utilization					
Protein efficiency ratio (PER)	1.01[a]	0.92[b]	0.68[c]	0.72[c]	0.0176
Protein productive value (PPV, %)	45.62[a]	36.11[b]	33.19[c]	26.500[d]	0.070
Energy utilization					
Energy efficiency ratio (EER, g/Kcal)	0.072[a]	0.066[b]	0.048[c]	0.051[c]	0.002
Energy productive value (EPV, %)	45.05[a]	43.00[b]	33.63[d]	36.25[c]	0.031
Lipid utilization					
Lipid retention (LR, %)	105.05[a]	121.02[b]	94.43[d]	100.46[c]	0.044

(a b, c and d) Average in the same row having different superscripts significantly different at level ($P \leq 0.05$).

*, SED is the standard error of difference.

On the other hand, the best feed efficiency of juvenile *Siganus guttatus* was achieved by 10 and 30 ppt salinity level compared with 5 and 20 ppt salinity level[26]. [36]indicated that, salinities of 30 ppt and 35 ppt negatively affected on FCR of Hybrid TGGG juveniles. Also, [37]found that protein efficiency ratios (PER) of sea bream cultured at 15 ppt were consistently higher than those at other salinities.

Whole body chemical composition and energy content

Whole body chemical composition and energy content of rabbitfish (*Siganus rivulatus*) juvenile at the beginning and the end of the experimental period are shown in Table (6) There were significant differences between the treatments at the end of the experimental period in dry matter (DM), crude protein (CP), ether extract (EE), ash and gross energy (GE).The fish were reared at Lake Qaroun salinity had the highest DM followed by 15 ppt and 25 but 5 ppt was the least. The highest CP was obtained with Lake Qaroun salinity followed by 25 ppt and 5ppt while 15ppt was the least. Unlike, the highest EE and GE were obtained by 5ppt and 15 ppt followed by 25ppt while Lake Qaroun salinity was the least.

Ash content was the highest with 15ppt followed by 25 ppt and it did not differ between Lake Qaroun salinity and 5ppt salinity level.

From these results cleared that, the highest CP content obtained with the fish were reared under the high salinities as Lake Qaroun salinity and 25 ppt salinity level. Vice versa, the high EE and GE obtained with the fish were reared under the low salinities as 5pp and 15 ppt salinity level.

Table (6) Effect of water salinity level on body chemical composition and energy content (On DM basis) of whole body rabbitfish (*Siganus rivulatus*) juvenile.

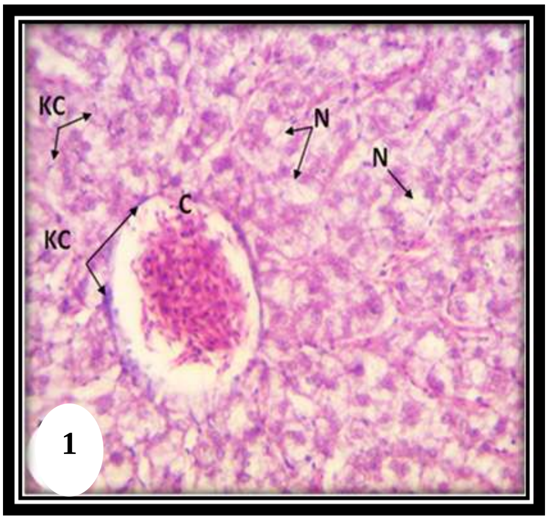
Items	Start	Treatments				SED*
		Lake Qaroun Salinity	25ppt	15ppt	5ppt	
Moisture (M, %)	81.48	69.54[d]	70.78[b]	70.14[c]	72.26[a]	0.014
Dry matter (DM, %)	18.52	30.46[a]	29.22[c]	29.86[b]	27.74[d]	0.018
Crude protein (CP, %)	62.02	47.60[a]	43.05[b]	40.00[d]	41.22[c]	0.077
Ether extract (EE, %)	11.78	34.64[c]	42.61[b]	45.08[a]	45.11[a]	0.0137
Ash, %	24.03	13.13[c]	13.72[b]	15.53[a]	13.04[c]	0.031
Gross energy (GE, Kcal/g)	4.53	5.90[c]	6.14[b]	6.48[a]	6.55[a]	0.031

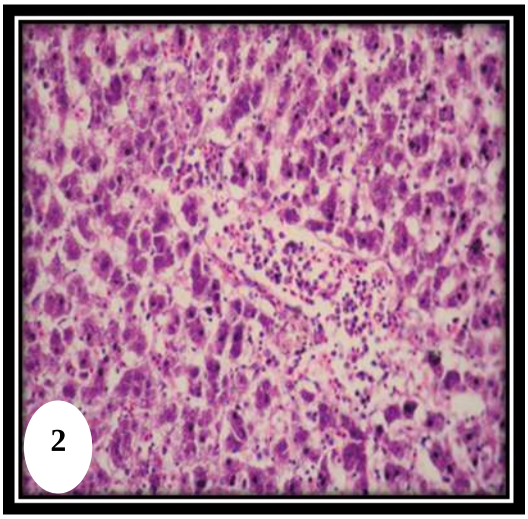
(a b, c and d) Average in the same row having different superscripts significantly different at level ($P \leq 0.05$).

*, SED is the standard error of difference

Histological study

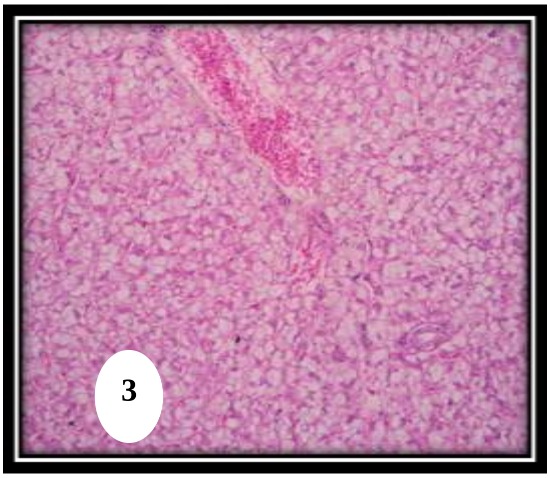
Liver histological





Photomicrograph (Photo.) 1: Liver of rabbitfish reared in low salinity (5ppt) showing: degenerated hepatocytes, focal area of necrosis and congested blood vessels in central vein, appearance of kuppfer cells around the central vein (H&E, X 400).

Photo. 2: Liver of rabbitfish reared in low salinity (5ppt) showing disorganization of hepatocytes distortion of hepatic cords, individualization of hepatocytes, inflammatory cells infiltrations and interstitial edema (H&E, X 400).



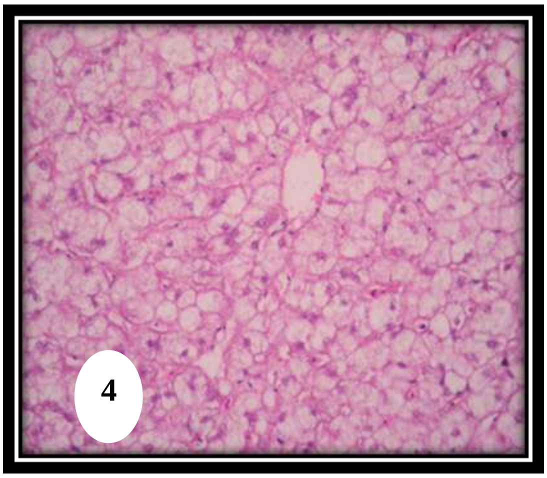
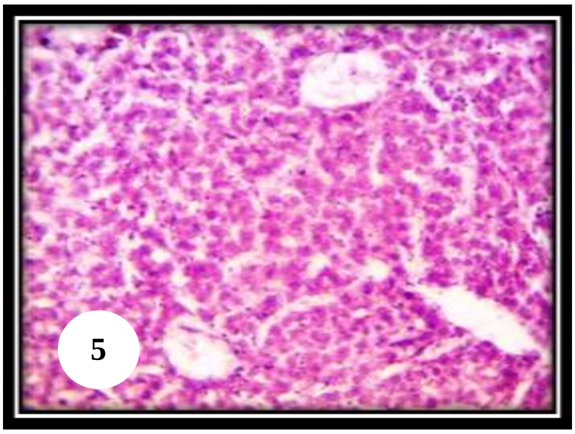


Photo.3 and 4: Liver of rabbitfish reared in salinity (15ppt) showing congested central vein, vacuolar degeneration of hepatocytes and dilated sinusoids with red blood cells (H&E, X400).



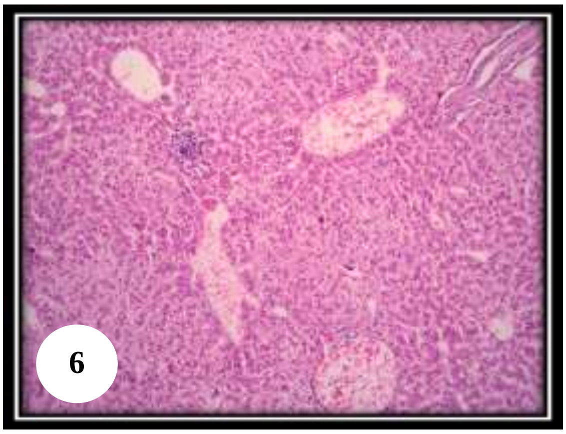
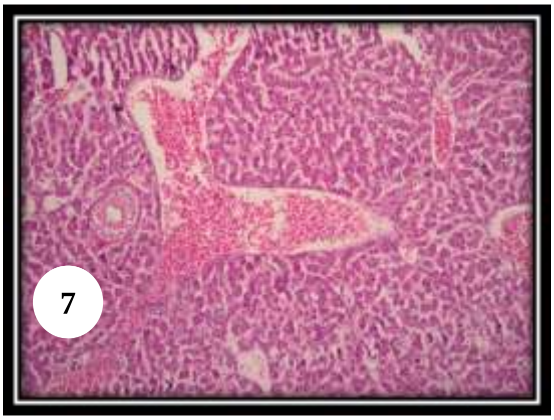


Photo. 5: A sagittal section in Liver of rabbitfish reared in salinity (25ppt) showing: severe damage of hepatocytes represented by focal necrosis. Also, eosinophilic cytoplasm with pyknotic nuclei (H&E, X400). Photo. 6: Liver of rabbitfish reared in salinity (25ppt) showing dilated central vein, focal necrosis of hepatocytes and infiltration of inflammatory cells (H&E, X200).



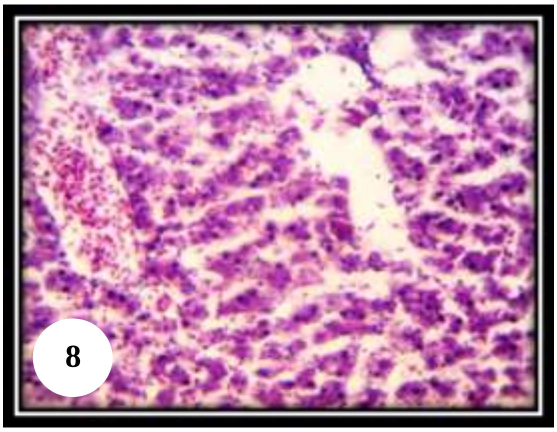


Photo.7: Liver of rabbitfish reared in Lake Qaroun salinity showing dilated center vein (C. V.) and vacuolar degeneration of some hepatocytes (H&E, X200).

Photo. 8: A sagittal section in Liver of rabbitfish reared in Lake Qaroun salinity showing: severe congestion and rupture of hepatocytes with multi necrotic area and infiltration of macrophages cells (H&E, X 400). (C: Congestion, KC: kuppfer cells, N: Necrotic area)

In the present study, liver tissue of rabbitfish reared in salinity 5 ppt and Lake Qaroun salinity was most harmful compared with other treatments. Whereas, liver tissue of fish reared in salinity 5 ppt showed degenerated hepatocytes, focal area of necrosis and congested blood vessels in central vein, individualization of hepatocytes, inflammatory cells infiltrations and interstitial edema. These signs cleared that, rabbitfish reared in low salinity suffered the stress of osmoregulation and this stress may have effect on liver tissue as reported by [42]who said that the impact of salinity on the liver tissue of juvenile chum salmon was high and the exposure to low salinity caused some liver cells to breakdown leading to serious vacuolization of the tissue.

Liver tissue of fish were reared in salinity 25and Lake Qaroun salinity ppt showed dilated center vein, focal necrosis and infiltration of inflammatory cells and vacuolar degeneration of some hepatocytes. Also, severe congestion and rupture of hepatocytes with multi necrotic area and infiltration of macrophages cells or severe damage of hepatocytes represented by focal necrosis. These signs cleared that, rabbitfish were reared in water polluted by metal. The accumulation of metal ions in the liver may lead to hepatic lesions. Whereas, liver is the principal organ responsible for detoxification in vertebrates generally and in fish particularly[43]. And, [44]derived a link between exposure to heavy metals and lesions in liver. Similar conclusions were described by[45] in *Clarias gariepinus* after exposure to lead. Degeneration and necrosis of hepatocytes may be due to the cumulative effect of the metals and increase in their concentration in liver.

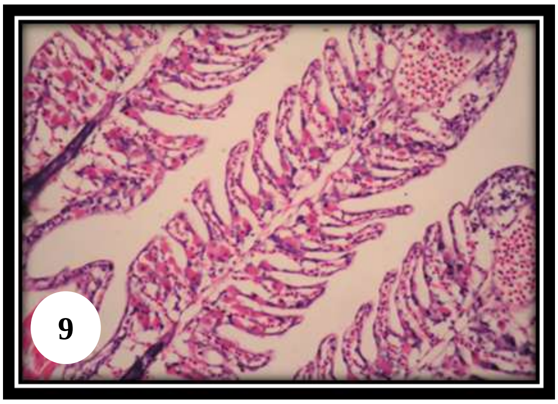
Moreover, the evident damage of the central vein, degeneration of liver tissues, and necrosis observed in samples from the polluted sites could be attributed to the accumulation and infiltration of neutrophils and lymphocytes as indicated by[46]**.** These histopathological changes suggest high metabolic activity in hepatocytes in response to the uptake of heavy metals[47].

[48]reported that, the deterioration in histoarchitecture observed in gill, liver, and kidney of both fish species collected from Lake Qaroun and fish farms around it was generally in accordance with the results of residual heavy metals, which suggests too slow defense mechanisms in these tissues to immobilize or eliminate heavy metals and shows the sensitivity of fish cells to metal exposure. This was confirmed by Previous studies reported that Lake Qaroun components are polluted with heavy metals[49], solid and nutrients[16]and with a wide variety of pesticides (e.g. lindane, aldrin, some DDT analogues malathion)[15]. Moreover, a remarkable increase in the bacterial indicators of sewage pollution (total coliform, faecal coliforms and faecal streptococci) in the lake was recorded[17].

These results agreed with [50]found that, Liver samples obtained from *Tilapia zillii* fish inhabiting the water of Lake Qaroun suffered from many pathological alterations. These results agreed also with those obtained by[51]who said that changes appeared in liver rabbitfish reared in Lake Qaroun may be due to microbial pollution and this is shown in Table (2). [50]reported that hemorrhage in liver tissue was caused by bacteria present in sewage water. [52, 53]added that, necrosis may be caused by bacteria and toxins secreted by micro-organism in sewage water.

There were mild changes appeared in rabbitfish liver which reared in salinity 15ppt this may be due to decreasing of heavy metal, hence decreasing of accumulation and elimination processes of metals ions in liver tissue .

Gills histological



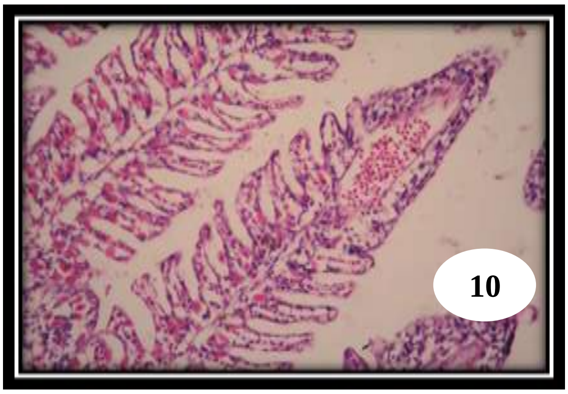
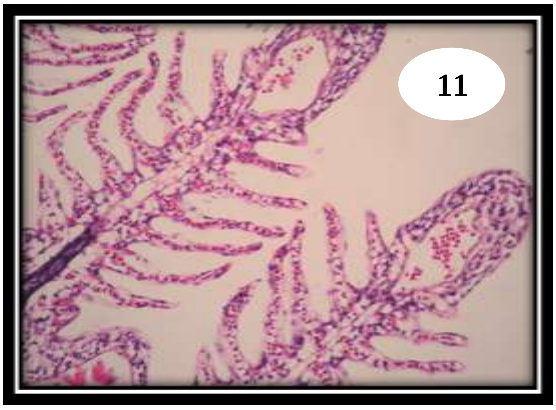


Photo. 9 and 10: Gills of rabbitfish reared in salinity (5ppt) showing congested blood vessels, proliferation of chloride cells and fusion of secondary lamellae on the proximal and distal ends (H&E, X400).



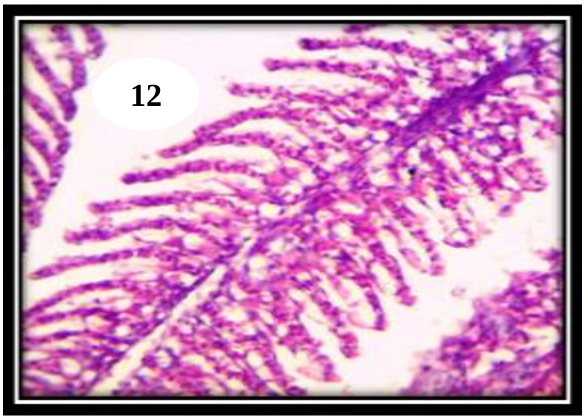


Photo.11: A sagittal section in gills of rabbitfish reared in salinity (15ppt) showing lifting epithelium with necrosis and degeneration of epithelial lining the secondary lamellae (H&E, X200).

Photo. 12: A sagittal section in gills of rabbitfish reared in salinity (15ppt): desquamation of the epithelium of primary gill filaments and sloughing of some secondary lamellae hyperplasia of epithelial

cells leading to decrease of interlamellar space and bending or curling of some secondary gill lamellae (H&E, X 400).

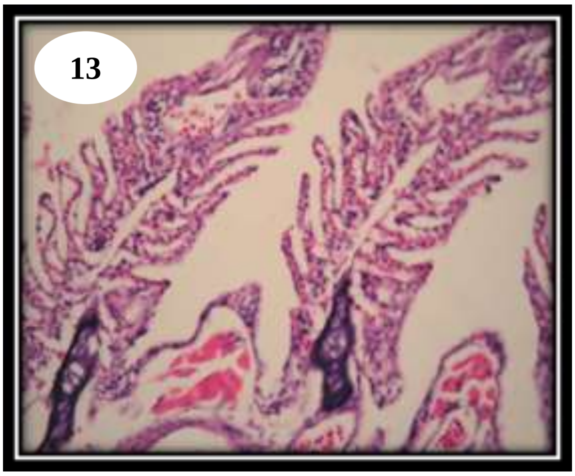


Photo. 13: A sagittal section in gills of rabbitfish reared in salinity (25ppt)showing epithelial lifting (H&E, X200).

Photo. 14: A section in gills of rabbitfish reared in salinity (25ppt) showing: hyperplasia of secondary lamellae leading to increase of interlamellar space (H&E, X 400).

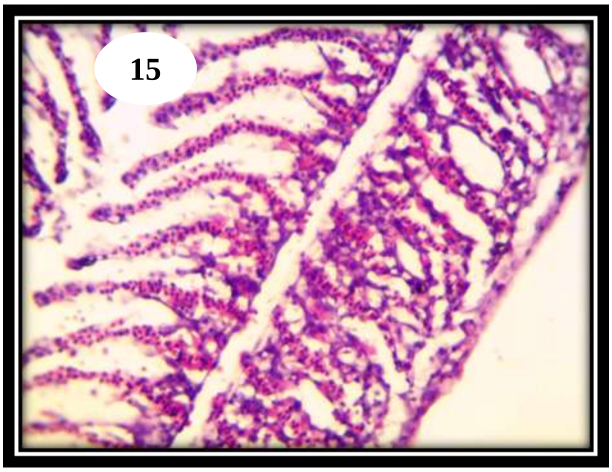


Photo. 15 and 16: A section in gills of rabbit fish reared in Lake Qaroun salinity showing: hyperplasia of secondary lamellae leading to decrease of interlamellar space, and severe congestion in primary filament, with infiltration of lymphocytes (H&E, X400). Also, gills showing fusion of secondary lamellae at one side and necrosis of secondary lamellae on the other side with inflammatory cell infiltrations (H&E, X400).

In the present study, gills of fish reared in salinity 5 ppt showed congested blood vessels, proliferation of chloride cells and fusion of secondary lamellae on the proximal and distal ends. It can be said that, the fish gills are directly connected to the external environmental they are quite sensitive to chemical and physical changes of water[54]. And regarding of rabbitfish are marine herbivores fish and it prefers the saline water condition so the signs which appeared on gills return to face of low salinity. This resulted in proliferation of chloride cells as shown in (Photo. 9 and 10). It can be noted that, The “chloride” or mitochondria-rich cells of the gill play a prominent role in teleostean fish‘s osmoregulation. Indeed, the permeability of this cell type is modified according to the salinity of the external environment: in fresh water, the chloride cells absorb a small quantity of electrolytes[55]whereas in salt water they are the site of a large ionic excretion[56]. Moreover, [57]reported that, the histological lesions in gills fish might arise also because of the stress such as acid base changes between two environments, in the same trend, [58]found that, anomalies observed in the gills do not reflect the impacts of a certain pollutant as these anomalies may be caused by irregularity in the ecosystems. Also, [59]stated that, morphological changes in gills represent adaptations that occur as a response to environment changes.

In view of Photo. (15 and 16) Sections in gills of rabbitfish reared in Lake Qaroun salinity also Photo. (13 and 14) sections in gills of rabbitfish reared in salinity 25 ppt. these figures shown that, hyperplasia of secondary lamellae leading to decrease of interlamellar space, and severe congestion in primary filament, with infiltration of lymphocytes. Also, gill showing fusion of secondary lamellae at one side and necrosis of secondary lamellae on the other side with inflammatory cell infiltrations and gills showing epithelial lifting. This signs as result of Lake Qaroun water pollution by metals or microbial or high level of ammonia and nitrite. This was in agreement with[50; 51]who revealed that, these malformations in gills may be due to increase of ammonia and heavy metals, pH change, oxygen depletion occurrence of bacteria, microorganisms and parasites with increasing in turbidity in Lake Qaroun. Also, [60]suggested that, the bacteria produce in extracellular hyperplasia inducing factor and they added that, these pathological changes may be a reaction to toxicants intake or an adaptive response to prevent the entry of the pollutants through the gill surface.

On the other hand, ammonia toxicity causes osmoregulatory imbalance kidney failure and suppressed excretion of endogenous ammonia, resulting in neurological and cytological failure. So, ammonia in water is an indicator of possible bacterial, sewage and animal waste pollution. Natural levels in ground and surface water are usually below 0.2 mg/l [61].

In the light of Table (2) it can be found that, total coliforms, fecal coliforms and fecal streptococci were 290, 240 and 460 /100 water also Table (3) cleared that, total ammonia in lake Qaroun, 25 ppt, 15ppt and 5 ppt were 0.527, 0.387, 0.319 and 0.27 mg/l respectively. Hence the bacteria and ammonia cause many problems of fish gills.

Photo. (11 and 12) sections in gills of rabbitfish reared in salinity 15 ppt showed the least harmful of gills compared with the others treatments. This harmful may be due to 15 ppt level salinity is unfit for gills to work also there a strong effect of medium salinity on gill NKA activity[34].

Infer from this study that, the effect of low salinity stress on growth performance was higher than the effect of Lake Qaroun water pollution stress. It can be said that, in spite of rabbitfish can life in wide range of salinity however it prefers the high salinity whereas its growth rate was better under Lake Qaroun water condition than the low salinity (5ppt).

Conclusion

Rabbitfish juvenile can be reared at 5 ppt, 15 ppt , 25 ppt and Lake Qaroun salinity but the high salinity level (Lake Qaroun Salinity) was the best compared with the other salinity levels in growth performance and survival rate. Also, histological study investigated that; there were severe damage of hepatocytes and gills with both high and low salinity. However, rabbitfish faster grown under Lake Qaroun water conditions than other were reared in low salinity water.

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