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The Ability of the Red Tilapia (*Oreochromis* sp.) to Tolerate Different Concentrations of Unionized Ammonia and Water Salinity Levels

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ABSTRACT

The study aimed to evaluate the ability of the red tilapia (*Oreochromis* sp.) to withstand varying unionized ammonia and water salinity levels on growth performance, feed utilization, and physiological state. Three distinct unionized ammonia concentrations of 0.15, 0.20, and 0.25mg/ l and two different levels of water salinity of 25 and 30ppt were assigned in six treatments (T1 through T6), each with three replicates. Eighteen aquariums held a total of 180 red tilapia distributed equally among them. Ten fish were placed at random in these aquariums, with an average initially body weight of 19.42± 0.04g and an initial body length of 8.48± 0.51cm. The experimental fish were fed commercial feed (30% crude protein) at a rate of 3% of their total biomass three times a day, at 08:00, 13:00, and 18:00, for 8 weeks. Blood samples were taken from five fish in each aquarium at the end of the trail to measure the hematological and biochemical parameters. The findings show that the interaction between unionized ammonia and the water salinity had no discernible effect on the majority of growth and feed utilization indices. WBCs, platelets, and MCHC levels were highest in T5, T3, and T4, respectively. T1 had the highest globulin value and urea contents, while T2 recorded the greatest values of liver enzymes ALT and AST. The current study suggests that different unionized ammonia levels affected fish health and quality, but water salinity decreased these effects slightly and allowed the red tilapia to grow well in high salinity water.

INTRODUCTION

Both inland and coastal resources abound in Egypt. The red tilapia, which can withstand high salinity and be raised in seawater, may therefore be an excellent choice for aquaculture **(Hassanen *et al.*, 2014)**. Egypt's fish farming industry faces several difficulties. Among these problems is water pollution from both organic and inorganic chemical sources, which poses a serious risk to the existence of aquatic life **(Saeed & Shaker, 2008)**. Ammonia contamination results in poor water quality, which kills most tropical organisms. Moreover, ammonia poisoning affects many aquatic animals, including fish **(Harris *et al.*, 1998)**.

In addition, fish may be negatively impacted by unionized ammonia (NH_3), ionized ammonia (NH_4^+), and harmful metabolites like nitrite and chloramines that are produced from it, as well as by water acidification **(De silva *et al.*, 2013)**. In general, NH_3 concentrations should be kept below 0.05mg l^{-1} for long-term exposure since freshwater fish are more tolerant of ammonia toxicity than saltwater fish, and warm water species are typically more tolerant than cold water fish **(Timmons *et al.*, 2002)**. Under healthy conditions, the kidney's contribution to the whole-body excretion of ammonia in freshwater fish is negligible, usually accounting for less than 20% of the total nitrogen eliminated, with the remaining >80% being expelled through the gills **(Zimmer *et al.*, 2014).** Nonetheless, in certain species, renal ammonia excretion is markedly elevated by metabolic acidosis **(Wood *et al.*, 1999)**.

Studies have indicated that raising the salinity of the water can reduce the harmful effects of NH_3 and nitrite on fish **(Sampaio *et al.*, 2002)**. In several fish species, salinity has been demonstrated to have a positive impact on growth rate and to be useful in reducing the toxicity of unionized ammonia. According to earlier research, turbot, *Scophthalmus maximus*, grew more quickly when the salt content was lowered **(Imsland *et al.*, 2001)**.

According to **Alkobaby and Hassanien (2007)**, tilapia is resistant to elevated ammonia concentrations. According to **Ogbonna and Chinomso (2010)**, toxic unionized ammonia can kill fish in a matter of days at levels as low as 0.6mg/ l. Prolonged exposure to harmful unionized ammonia concentrations as low as 0.06mg/ l can harm fish's gills and kidneys, stunt their growth, potentially damage their brains and reduce their ability to carry oxygen. Overdoses of ammonia can act as a harmful agent in aquatic animals by reducing growth, causing tissue erosion and degeneration, suppressing the immune system and increasing mortality **(Li *et al.*, 2014)**. In this work, we aimed to elucidate the impact of unionized ammonia and water salinity on the red tilapia (*Oreochromis* sp.) growth performance, feed utilization, and physiological state.

MATERIALS AND METHODS

Experimental design

The Mariculture Research Center (MRC), Faculty of Environmental Agricultural Sciences, Arish University, North Sinai, Egypt is the site of the current study. The MRC provided the red tilapia under study during March and May of 2022. Before the experiment began, the fish were given fifteen days as acclimatization period. During the acclimation period, fish were fed commercial feed (30% crude protein) at a rate of 3% of their total biomass, divided into 3 equal meals at 8:00, 13:00, and 18:00 hours. After this duration, a total of 180 fish representing the six treatments with three replicates, with an average initial body weight of 19.42± 0.04g and initial body length of 8.48± 0.51cm, were arranged at random in eighteen aquariums of 60 ×40 ×25cm and holding a total of 60 liters of water each. Three replicates of each treatment were conducted, representing

different concentrations of unionized ammonia (0.15, 0.20, 0.25mg/ L) combined with water salinity values of either 25 or 30ppt. The treatments included 0.15mg/ L unionized ammonia (UIA) with 30ppt water salinity (T1), 0.20mg/ L UIA with 30ppt water salinity (T2), 0.25mg/ L UIA with 30ppt water salinity (T3), 0.15mg/ L UIA with 25ppt water salinity (T4), 0.20mg/ L UIA with 25ppt water salinity (T5), and 0.25mg/ L UIA with 25ppt water salinity (T6).

Every day, ten liters of water with previously adjusted unionized ammonia and water salinity concentrations were replaced to each tank. The photoperiod was kept at a cycle of 12 hours D: 12 hours L. According to **Emerson *et al*. (1975)**, unionized ammonia concentrations were achieved by progressively adding NH_4Cl solution into the water to raise the NH_3-N concentration. This was expressed as (total ammonia × percentage of unionized ammonia at the pH and temperature in ammonia relationship table) ÷ 100. In the current experiment, water salinity was obtained from 35ppt artesian well water. To lower the salinity levels, fresh water was introduced to the water and diluted. Every day, the number of dead fish was counted in order to estimate the survival rate. Each aquarium's fish west product was taken out before the daily morning feeding. Every week, fish were collected from their aquariums, weighed, and the amount of feed was modified to account for variations in body weight over the trial. All fish were netted from each treatment aquarium at the end of the eight-week trial, weighed, measured to estimate growth parameters and feed utilization. Blood samples were taken then fish were dissected for physiological status and histological examination.

Water quality parameters

Every day at 12:00pm, before the second feeding, water quality indicators were measured. A mercury thermometer was used to record the water's temperature every day. The dissolved oxygen level was measured using a YSI model 56 oxygen meter (YSI Company, Yellow Springs Instrument, Yellow Springs, Ohio, USA). Total ammonia levels were measured twice a week using a DREL 2000 spectrophotometer (Hach Company, Loveland, CO, USA). A pH meter (Orion pH meter, Abilene, Texas, USA) was used to estimate the pH. Throughout the trial, the water's temperature remained constant at $22^oC \pm 1.0$, its pH remained constant at 7.9± 0.1, and its dissolved oxygen level remained constant at 7.99± 0.55mgl^{-1}.

Growth performance and feed utilization

The following equations were used to measure the growth performance and feed utilization selected parameters. Weight gain (WG) = final body weight (W2) – initial body weight (W1). ADWG = WG /t. Gain% = (WG/W1) x 100. Gain in length (GL) = final length- initial length. Condition factor (K) = $(W/L^3) \times 100$, where W is weight of fish in grams and L is total length of fish in cm. Specific growth rate (SGR) = (LnW2 – LnW1)/ t × 100, where Ln is the natural logarithm, W1 is initial body weight and W2 is the final body weight in grams and "t" is the experimental period in days (56 days). Feed conversion ratio (FCR) = feed intake (g)/ weight gain (g). Feed efficiency (FE %) =

weight gain (g)/feed intake (g). Protein efficiency ratio (PER) = weight gain (g)/ ingested protein (g).

Blood sampling for hematological and biochemical parameters

Five fish from each treatment group had their blood drawn at the end of the trial. The blood samples were divided into two sections. The first section was collected into clean tubes with anticoagulant for hematological parameters. In dry, sterile centrifuge tubes, the second section of the blood samples were drawn without the use of an anticoagulant. After the blood samples were placed in a benchtop digital centrifuge and spun at 3000rpm for 15 minutes, sera were separated. After the blood samples were drawn, all hematological and biochemical tests were conducted right away, without keeping the sample for a long time.

Hematological parameters

White blood cells (WBCs) and red blood cells (RBCs) counts were calculated using the **Dacie and Lewis (1991)** method. **Drabkin and Austin (1932)** method was used to calculate the hemoglobin concentration (g/dl^{-1}). In accordance with **Sorrell-Raschi and Tomasic (1998),** the hematocrit (Hct) value was determined. **Brecher *et al.* (1953)** method was used to calculate the platelet count. According to **Seiverd (1964)**, the mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) were calculated.

Biochemical parameters

Glucose was calculated using **Tietz's (1986)** method. Serum uric acid was measured in accordance with **Young (1995)**. The method used to determine serum urea enzymatically was conducted following the method of **Patton and Crouch (1977)**. Serum creatinine was determined using **Henry's (1974)** technique. Albumin and total protein were calculated using **Doumas's (1975)** method. **Reitman and Frankel (1957)** method was used to determine the levels of aspartate aminotransferase (AST) and alanine aminotransferase (ALT).

Histological examination

Gills, kidneys, and livers were carefully removed from three to five fish per treatment at the end of the experiment, and they were preserved in Bouin's solution for roughly a day. After that, the specimens were stored in 70% ethyl alcohol. The samples underwent the customary xylene clearing and paraffin wax embedding processes. The sections of 4-6µ thickness were mounted on dry clean glass slides. According to **Pearse (1972)**, the sections were prepared and then stained with Harri's Haematoxylin and Eosin (Hx and E).

Ethical approval

The authors adhered to all relevant international, national, and/or institutional norms for the care and use of fish at Arish University's Animal Care and Ethical Committee (No: AGRI02).

Statistical analysis

SAS, version 6.03 **(Statistical Analysis System, 1996)** was used to evaluate all data. The individual impacts of the covariates and their interactions were analyzed using two-way ANOVA, and all differences were deemed significant at $P < 0.05$. The results are shown as means with pooled standard error (SE).

RESULTS

Growth performance and feed utilization

Table (1) displays the growth performance results of water salinity, unionized ammonia, and their combination in the red tilapia (*Oreochromis* sp.). The initial body weight (IBW) variations across treatments were not statistically significant ($P = 0.750$), suggesting that the experimental treatments were assigned at random at the beginning of the study. There was no significant difference ($P > 0.05$) observed in any of the growth performance measures when unionized ammonia (0.15, 0.20, and 0.25mg/ l) and water salinity (25 and 30ppt) interacted.

There was no significant difference ($P > 0.05$) in the survival rate (SR) ($P = 0.1028$) among the different treatments. Additionally, compared to other treatments, the SR of T2, T3, and T4 was higher (Table 1). Water salinity, unionized ammonia and their combination had no effect on feed efficiency (FE%), feed conversion ratio (FCR), feed intake (FI), or protein efficiency ratio (PER) and the differences were not statistically significant ($P > 0.05$) (Table 2).

Hematological parameters

Table (3) displays data of the red tilapia (*Oreochromis* sp.) treated with unionized ammonia and water salinity, including RBCs, WBCs, hemoglobin (Hb), hematocrit (Hct), platelets count (Plt), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC).

The interaction between unionized ammonia and water salinity had a negligible effect on RBCs and Hct ($P > 0.05$). However, WBCs showed a significant difference ($P = 0.0152$), with T5 recording the highest value and T1 the lowest. T3 had a significantly higher Hb (g/dL) value ($P = 0.0050$), whereas T5 recorded the lowest. Table 3 illustrates how analysis of variance revealed significant differences ($P < 0.05$) between treatments for Plt, MCH, and MCHC. There was no discernible difference in MCV ($P > 0.05$). Plt increased in T3 but decreased in T2. T5 recorded the highest MCH ($P = 0.0001$), whereas T4 recorded the lowest. T4 exhibited the highest MCHC ($P = 0.0001$), while T2 had the lowest MCHC value (Table 3).

Table 1. Growth performance parameters of red tilapia affected by unionized ammonia and water salinity for 56 days

Treatments	Water Salinity (ppt)	UIA (mg/l)	IBW (g fish^{-1})	Growth Performance						
				FBW (g fish^{-1})	WG (g fish^{-1})	ADWG	SGR (% day^{-1})	Survival rate %	GL (cm fish^{-1})	K
Individual treatment means*										
T1	**30**	**0.15**	**19.40**	**33.22**	**13.82**	**0.25**	**0.96**	**83.33^{ab}**	**3.18**	**2.17**
T2	**30**	**0.20**	**19.40**	**31.59**	**12.19**	**0.22**	**0.87**	**90.00^{ab}**	**2.98**	**2.17**
T3	**30**	**0.25**	**19.47**	**32.14**	**12.68**	**0.22**	**0.89**	**93.33^{a}**	**3.23**	**1.99**
T4	**25**	**0.15**	**19.47**	**33.02**	**13.55**	**0.24**	**0.94**	**86.67^{ab}**	**3.02**	**2.08**
T5	**25**	**0.20**	**19.40**	**32.95**	**13.55**	**0.24**	**0.94**	**83.33^{ab}**	**2.96**	**2.09**
T6	**25**	**0.25**	**19.40**	**33.92**	**14.52**	**0.26**	**1.00**	**80.00^{b}**	**3.36**	**2.12**
Pooled SE								**3.43**	**0.21**	
Means of the main effect**										
	30		**19.42**	**32.32**	**12.89**	**0.23**	**0.91**	**88.89**	**3.13**	**2.11**
	25		**19.42**	**32.30**	**13.87**	**0.25**	**0.96**	**83.33**	**3.11**	**2.10**
		0.15	**19.44**	**33.12**	**13.69**	**0.25**	**0.95**	**85.00**	**3.10**	**2.13**
		0.20	**19.40**	**32.27**	**12.87**	**0.23**	**0.91**	**86.67**	**2.97**	**2.13**
		0.25	**19.44**	**33.03**	**13.59**	**0.25**	**0.95**	**86.67**	**3.29**	**2.06**
ANOVA (p-value)										
Water Salinity			**0.4605**	**0.4604**	**0.4776**	**0.5258**	**0.4684**	**0.7991**	**0.9572**	**0.350**
UIA			**0.5301**	**0.6423**	**0.6362**	**0.5568**	**0.6459**	**0.2551**	**0.9393**	**0.459**
Water Salinity × UIA			**0.7500**	**0.5136**	**0.5373**	**0.5324**	**0.5430**	**0.1028**	**0.7096**	**0.855**

*Treatments means represent the average values of three aquaria per treatment. Duncan multiple range test was conducted for individual means only if there was a significant interaction (ANOVA: $P < 0.05$).

**Means followed by the same letter are not significantly different.

Table 2. Feed utilization parameters of the red tilapia during trail period (8 weeks)

Treatment	Water Salinity (ppt)	UIA (mg/l)	Feed Efficiency			
			FI	FCR	PER	FE%
Individual treatment means*						
T1	**30**	**0.15**	**61.35**	**4.44**	**0.75**	**22.53**
T2	**30**	**0.20**	**58.71**	**4.82**	**0.69**	**20.76**
T3	**30**	**0.25**	**58.87**	**4.64**	**0.72**	**21.52**
T4	**25**	**0.15**	**59.96**	**4.42**	**0.75**	**22.60**
T5	**25**	**0.20**	**60.34**	**4.46**	**0.75**	**22.42**
T6	**25**	**0.25**	**61.19**	**4.21**	**0.79**	**23.73**
Pooled SE						
Means of the main effect						
	30		**59.64**	**4.63**	**0.72**	**21.60**
	25		**60.53**	**4.36**	**0.76**	**22.92**
		0.15	**60.66**	**4.43**	**0.75**	**22.57**
		0.20	**59.57**	**4.64**	**0.72**	**21.59**
		0.25	**60.03**	**4.43**	**0.76**	**22.63**
ANOVA (*P-value*)						
Water Salinity			**0.3219**	**0.5731**	**0.5875**	**0.5622**
UIA			**0.9360**	**0.5745**	**0.5485**	**0.5552**
Water Salinity × UIA			**0.8235**	**0.4170**	**0.3659**	**0.3656**

*Treatments means represent the average values of three aquaria per treatment. Duncan multiple range test was conducted for individual means only if there was a significant interaction (ANOVA: $P < 0.05$).

Table 3. Hematological indices of red tilapia after experimental period (8 weeks)

Treatment	Water salinity (ppt)	UIA (mg/l)	Hematological indices							
			RBCs $\times 10^6$	WBCs $\times 10^3$	Hb (g dl^{-1})	Hct %	Plt $\times 10^3$	MCV (fl)	MCH (pg)	MCHC (%)
Individual treatment means*										
T1	30	0.15	2.77	12.10^{c}	8.43^{b}	24.66	431.66^{b}	89.03	30.43^{a}	34.19$_{b}$
T2	30	0.20	3.27	19.40$^{a}_{b}$	9.97^{b}	29.83	368.66^{c}	91.22	30.49^{a}	33.42$_{c}$
T3	30	0.25	4.53	16.30^{b}	13.63^{a}	40.33	466.00^{a}	89.03	30.09$_{b}$	33.79^{b}
T4	25	0.15	2.90	32.40^{a}	6.70^{c}	13.50	463.00^{a}	46.55	23.10$_{c}$	49.63^{a}
T5	25	0.20	1.80	48.03^{a}	5.53^{c}	15.33	435.00^{b}	85.17	30.72$_{a}$	36.07^{a}
T6	25	0.25	3.43	14.90^{b}	10.33^{a}	30.67	433.33^{b}	89.42	30.12$_{b}$	33.68^{b}
Pooled SE										
Means of the main effect**										
	30		3.52	15.93	10.67	31.61	422.11	89.76	30.34	33.80
	25		2.71	31.78	7.52	19.83	443.78	73.71	27.98	39.79
		0.15	2.84	22.25	7.57	19.08	447.34	67.79	26.77	41.91
		0.20	2.54	33.72	7.75	22.58	401.83	88.19	30.61	34.75
		0.25	3.98	15.60	11.98	35.50	449666.67	89.23	30.11	33.74
ANOVA (*P-value*)										
Water Salinity			0.7664	0.3624	0.2399	0.3450	0.5268	0.3201	0.0001	1.0000
UIA			0.9527	0.1646	0.2609	0.8289	0.3089	0.2989	0.0000	0.2532
Water Salinity × UIA			0.6202	0.0152	0.0050	0.4077	0.0666	0.4274	0.0001	0.0001

*Treatments means represent the average values of three glass aquaria per treatment. Duncan multiple range test was conducted for individual means only if there was a significant interaction (ANOVA: $P < 0.05$).

**Means followed by the same letter are not significantly different.

Biochemical parameters

Table (4) displays the data about the levels of glucose (mg dl^{-1}), uric acid (mg dl^{-1}), urea (mg dl^{-1}) and creatinine (mg dl^{-1}) in the red tilapia (*Oreochromis* sp.) after they were treated with unionized ammonia under different water salinity. Unionized ammonia and water salinity had a substantial ($P = 0.0128$) impact on glucose level. T6 had the greatest glucose value, whilst T4 had the lowest glucose value. There were negligible variations ($P = 0.3552$) in serum uric acid among the various treatments. Moreover, there was a significant difference ($P < 0.05$) in urea concentration. T1 had the highest urea value ($P = 0.0176$), whereas T4 and T5 had the lowest values. Furthermore, T6 had the highest creatinine levels and T5 had the lowest one.

Total protein values (g dl^{-1}) did not change significantly ($P > 0.05$) among all treatments. The findings of the analysis of variance demonstrated that unionized ammonia, water salinity, and their combination had a significant ($P < 0.05$) impact on AST and ALT. T2 recorded the greatest ALT and AST levels. However, T1 recorded the lowest ALT and AST. There was minimal variation in albumin ($P = 0.9253$) between the

various treatments. Moreover, there was a significant difference in globulin ($P < 0.05$). T1 recorded the highest globulin value, whereas T5 recorded the lowest one. There were significant variations ($P < 0.05$) in the A/G ratio ($P = 0.0063$) across the various treatments. Moreover, T3 achieved the maximum A/G ratio. However, the A/G ratio's lowest levels were recorded in T1 (Table 5).

Table 4. Biochemical parameters of the red tilapia during experimental period (8 weeks)

Treatment	Water Salinity (ppt)	UIA (mg/l)	Biochemical Parameter			
			Glucose (mg dl^{-1})	Uric acid (mg dl^{-1})	Urea (mg dl^{-1})	Creatinine (mg dl^{-1})
Individual treatment means*						
T1	**30**	**0.15**	**226.00^{d}**	**2.73**	**21.50^{a}**	**0.77**
T2	**30**	**0.20**	**255.67^{b}**	**4.80**	**17.67^{b}**	**0.80**
T3	**30**	**0.25**	**265.33^{ab}**	**5.20**	**20.83^{a}**	**1.03**
T4	**25**	**0.15**	**190.33^{f}**	**4.83**	**13.00^{c}**	**2.17**
T5	**25**	**0.20**	**248.67^{c}**	**5.70**	**13.00^{c}**	**0.67**
T6	**25**	**0.25**	**294.67^{a}**	**2.37**	**15.00^{b}**	**3.30**
Pooled SE				**1.28**		
Means of the main effect**						
	30		**249.00**	**4.24**	**20.00**	**0.87**
	25		**244.56**	**4.30**	**13.67**	**2.05**
		0.15	**208.17**	**3.78**	**17.25**	**1.47**
		0.20	**252.17**	**5.25**	**15.34**	**0.74**
		0.25	**280.00**	**3.79**	**17.92**	**2.17**
ANOVA (*P-value*)						
Water Salinity			**0.0816**	**0.7254**	**0.2391**	**0.2757**
UIA			**0.7555**	**0.7546**	**0.1102**	**0.8671**
Water Salinity × UIA			**0.0128**	**0.3552**	**0.0176**	**0.4387**

*Treatments means represent the average values of three glass aquaria per treatment. Duncan multiple range test was conducted for individual means only if there was a significant interaction (ANOVA: $P < 0.05$).

**Means followed by the same letter are not significantly different.

Table 5. Selected liver function tests of red tilapia during experimental period (8 weeks)

Treatment	Water Salinity (ppt)	UIA (mg/l)	Liver Function Test					
			Total protein (g dl^{-1})	AST (U/l)	ALT (U/l)	Albumin (g dl^{-1})	Globulin (g dl^{-1})	A/G ratio
Individual treatment means*								
T1	**30**	**0.15**	**18.93**	**9.33^{d}**	**13.67^{c}**	**2.40**	**16.53^{a}**	**0.15^{d}**
T2	**30**	**0.20**	**7.90**	**42.33^{a}**	**20.00^{a}**	**2.40**	**5.50^{ab}**	**0.44^{c}**
T3	**30**	**0.25**	**9.23**	**16.33^{bc}**	**17.33^{b}**	**3.67**	**5.53^{ab}**	**0.66^{a}**
T4	**25**	**0.15**	**7.23**	**14.00^{c}**	**18.33^{a}**	**2.17**	**5.07^{b}**	**0.43^{b}**
T5	**25**	**0.20**	**6.20**	**18.00^{ab}**	**16.00^{b}**	**2.27**	**3.93^{c}**	**0.58^{a}**
T6	**25**	**0.25**	**6.50**	**23.33^{b}**	**15.67^{ab}**	**1.73**	**4.77^{b}**	**0.36^{c}**
Pooled SE								**0.08**
Means of the main effect**								
	30		**12.02**	**22.66**	**17.00**	**2.82**	**9.19**	**0.42**
	25		**6.64**	**18.44**	**16.67**	**2.06**	**4.59**	**0.46**
		0.15	**13.08**	**11.67**	**16.00**	**2.29**	**10.80**	**0.29**
		0.20	**7.05**	**30.17**	**18.00**	**2.34**	**4.72**	**0.51**
		0.25	**7.87**	**19.83**	**16.50**	**2.70**	**5.15**	**0.51**
ANOVA (*P-value*)								
Water Salinity			**0.0870**	**0.0674**	**0.1675**	**0.7963**	**0.0367**	**0.2995**
UIA			**0.1614**	**0.0316**	**0.1955**	**0.6840**	**0.1235**	**0.9549**
Water Salinity × UIA			**0.2412**	**0.0213**	**0.2113**	**0.9253**	**0.1023**	**0.0063**

Treatments means represent the average values of three glass aquaria per treatment. Duncan multiple range test was conducted for individual means only if there was a significant interaction (ANOVA: P* < 0.05).

**Means followed by the same letter are not significantly different.

Histological examination

The gills

After 56 days of treatment with unionized ammonia and water salinity, examination of the gill fish tissues of the red tilapia (*Oreochromis* sp.) revealed mild shortening, moderate congestion with moderately to markedly dilated capillaries, moderate blunting and shortening of lamellar with mild degenerative changes of lining cells with focal congestion, moderate shedding of lining cells, moderate degeneration and shedding of gill filaments with minimal congestion of capillaries, marked autolytic changes, and mild degenerative changes in lamellae lining cells (Fig. 1).

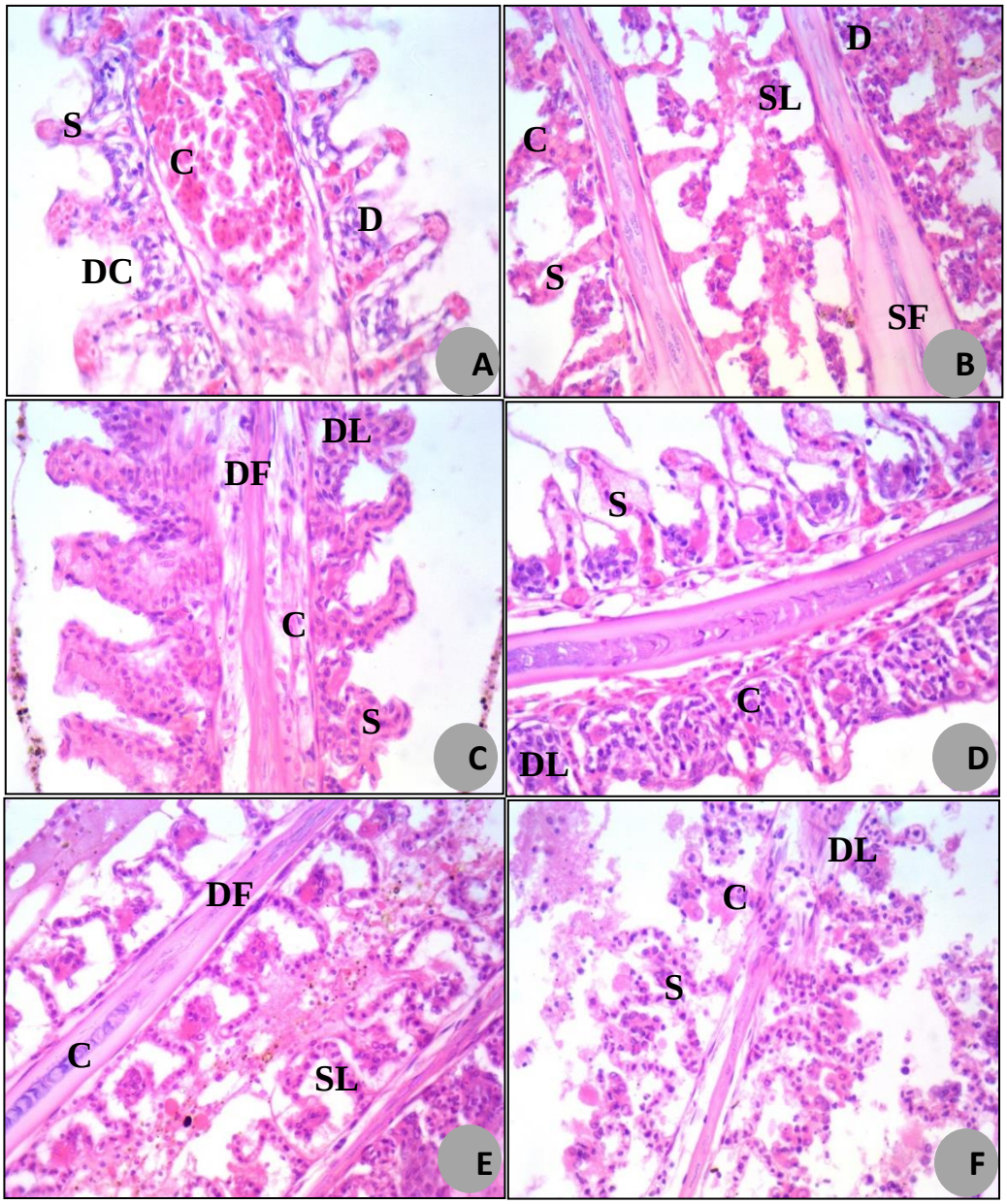

Fig. 1. Transverse sections of fish gill show mild shortening **(S)**, moderate congestion **(C)** with moderately to markedly dilated capillaries **(DC),** moderate shortening and blunting of lamellar with mild degenerative changes of lining cells **(D),** shedding of lining cells **(SL)**, shedding of gill filaments **(SF)**, foci showed moderate degeneration of lamellae **(DL)** and mild degeneration of filaments **(DF)**; T1, T2, T3, T4, T5 and T6 were A, B, C, D, E and F, respectively. H & E 400 X

The kidney

Examining the kidney tissues of the red tilapia (*Oreochromis* sp.) treated with unionized ammonia and salinity of the water revealed mild interstitial edema, focal mild inflammatory infiltrate, and moderate vacuolar degeneration of tubular epithelial cells; foci revealed

thickening of vessels, marked interstitial edema, and marked degeneration of tubules with moderate interstitial inflammatory infiltrate; marked congestion was observed, and a small number of glomeruli displayed focal shrinkage with moderate focal edema (Fig. 2).

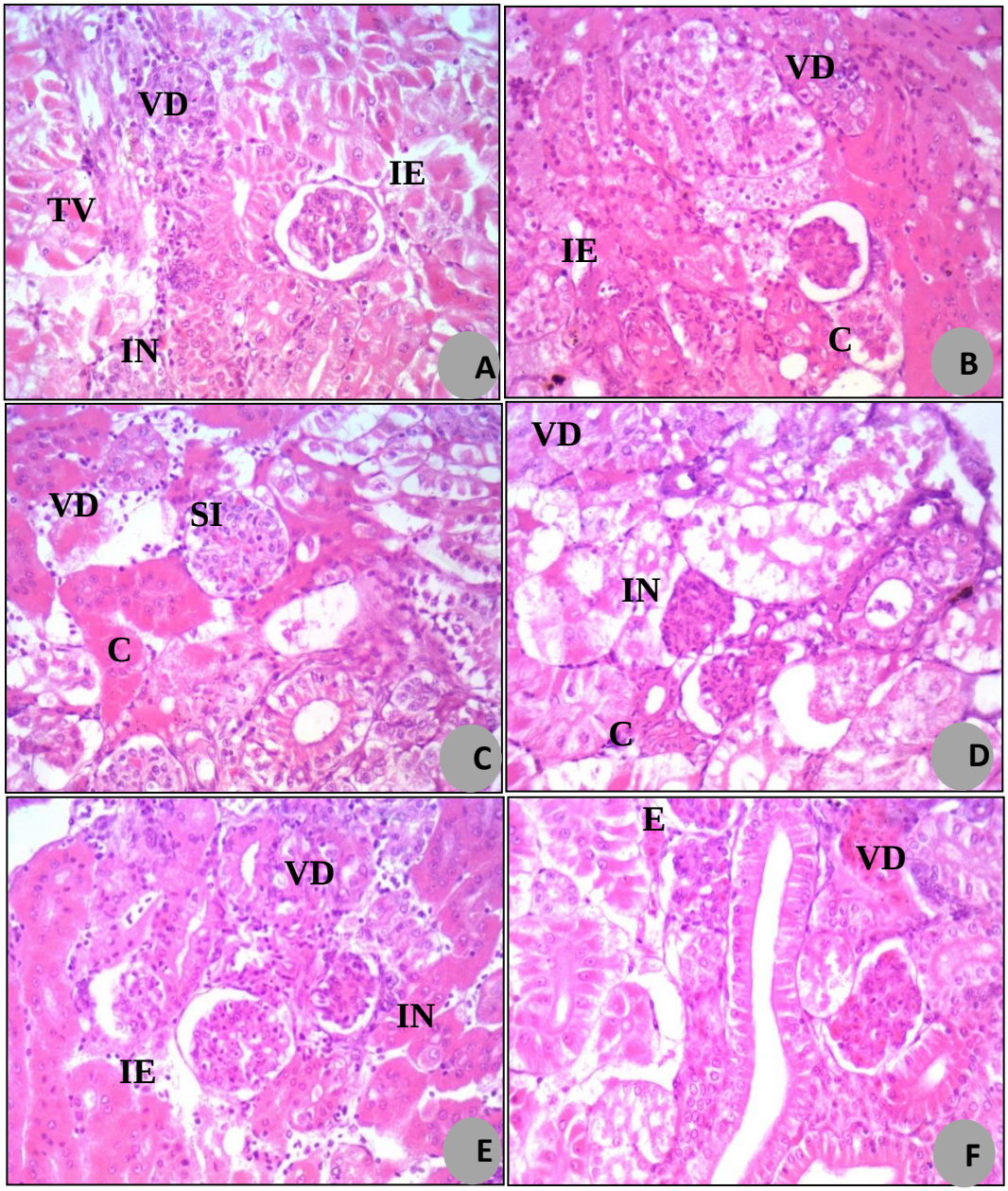

Fig. 2. Transverse sections of fish kidney show moderate vacuolar degeneration of tubular epithelial cells **(VD)** with moderate interstitial edema **(IE)**, focal mild inflammatory infiltrate **(IN)**, foci showed thickening of vessels **(TV)**, moderate vacuolar congestion **(C),** focal minimal edema **(E)** and few glomeruli showed focal shrinkage and moderate inflammatory infiltrate **(SI)**; T1, T2, T3, T4, T5 and T6 were A, B, C, D, E and F, respectively. H & E 400 X

The hepatopancreas

When the red tilapia (*Oreochromis* sp.) hepatopancreas tissues were examined after being treated with unionized ammonia and salinity of the water, they revealed significant steatosis of the hepatopancreas cells, a focal mild inflammatory infiltrate, moderate tubular epithelial cell vacuolar degeneration, mild congestion, necrosis of the hepatocytes, and vacuolation of the hepatopancreas cells (Fig. 3).

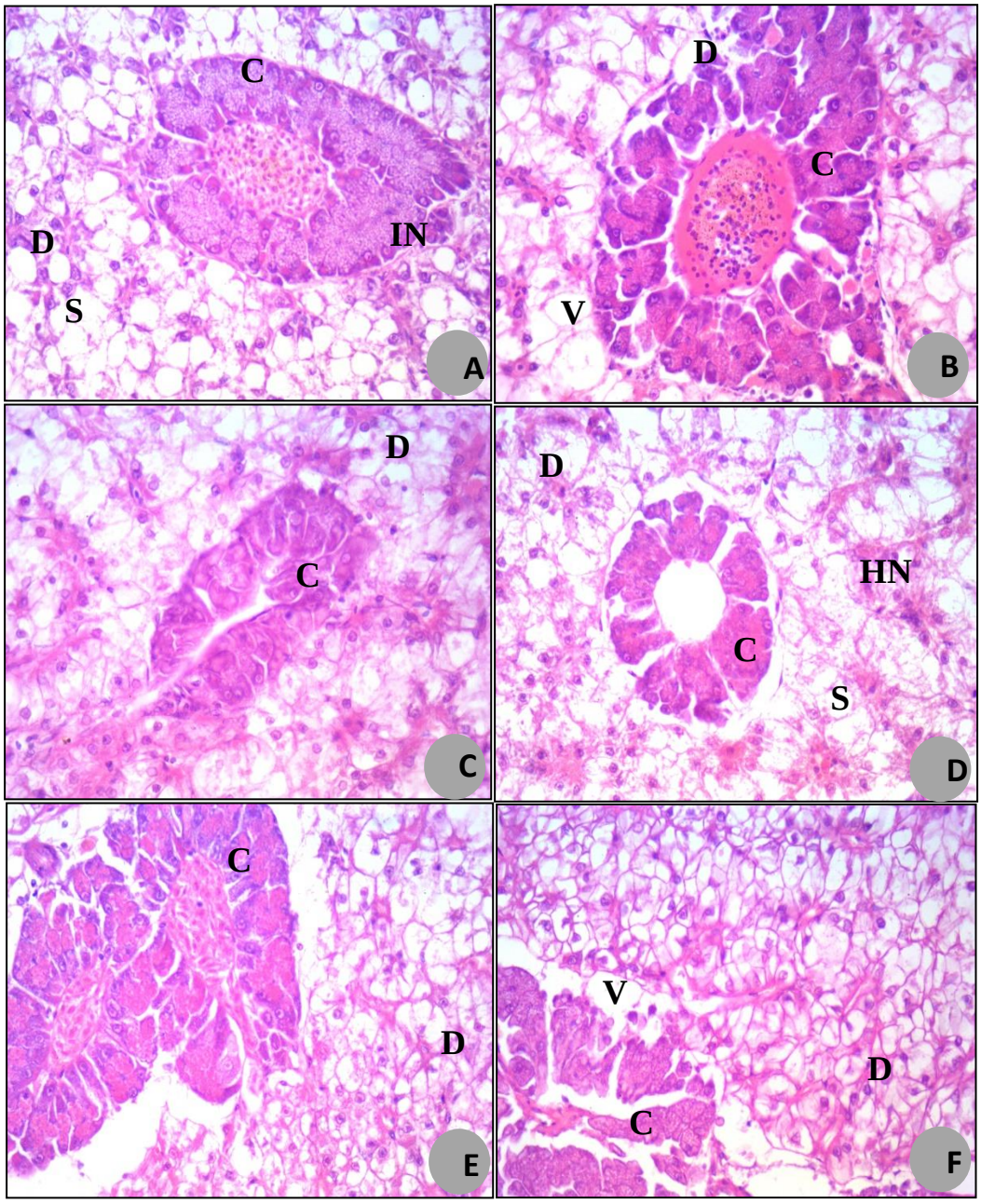

Fig. 3. Sections of fish hepatopancreas show marked steatosis of liver cells **(S)** with focal mild inflammatory infiltrate **(IN)**, moderate vacuolar degeneration of tubular epithelial cells **(D)**, mild congestion **(C)** and vacuolation of liver cells **(V)** with hepatocytes necrosis **(HN)**; T1, T2, T3,T4, T5 and T6 were A, B, C, D, E and F, respectively. H & E 400 X

DISCUSSION

Growth performance and feed utilization

Two ecological elements unique to the aquatic environment that have a big influence on the growth and survival of marine fish are unionized ammonia and water salinity. For growth, many young marine species favor intermediate salinities **(Kida & Ruddin, 2019)**. Numerous research on acute and chronic ammonia toxicity have also been conducted on several fish species **(Dosdat *et al.*, 2003)**. One of the numerous possibilities for the unionized ammonia and estimated water salinities is the genetic makeup of the evaluated tilapia strains, which includes the red tilapia (*Oreochromis* sp.). Since certain cells proliferate more readily in this species, osmotic control is remarkably effective **(Kaneko *et al.*, 2002)**. Perhaps a difference in the salinity of the water could reduce the toxicity of ammonia. Prior research on Atlantic salmon, *Salmo salar*, indicated that increasing the dissolved oxygen and salinity concentration in water could reduce the toxicity of ammonia **(Alabaster *et al.*, 1979)**.

While the current study found no significant differences in growth rates between 25 and 30ppt, other investigations, including **Mena *et al.* (2002)** found no significant differences between freshwater and 15ppt of water salinity which confirmed our findings. However, **Kida and Ruddin (2019)** concluded from their experiment on red tilapia that fish reared at 20 ppt salinity reached a higher average final weight compared to those reared at 15 ppt salinity. According to **Kida and Ruddin (2019)**, there was a substantial difference in the salinity treatment values, with the minimum condition factor value recorded for salinity of 20ppt and the greatest value recorded for salinity of 30ppt. Furthermore, **De Azevedo *et al.* (2015)** observed that water salinity levels had a substantial impact on the Nile tilapia's daily weight increase, feed conversion, and survival rate. At water salinities of 14 and 21gl^{-1}, these values were considerably lower. Regression analysis also showed that the daily weight growth, feed conversion, and survival rate followed a quadratic pattern with superior values of 5.62, 2.08, and 4.19gl^{-1} of water salinity, respectively.

Joel and Amajuoyi (2010) findings support the current findings, which indicate that ammonia has a tendency to harm the fish when its concentrations are above 0.2mg/ l. Even at low quantities, ammonia causes stress in fish, as demonstrated by the current study. However, the harmful effects of unionized ammonia are lessened when varying salinity levels of water are present. According to **Szumnski *et al.* (1982)** warm water fish could be treated with UIA-N at a concentration of 0.08mgl^{-1} without negative effects. **El-Sherif and El-Feky (2008)** reported that no fish mortality occurred in any of the experimental groups for the duration of the experiment when the fish were exposed to varying doses of UIA-N (0.01, 0.05, 0.1, 0.15, and 0.004mg/ l control). The current results are comparable to those of **Smith and Piper (1975)**, who noted that after 4 months, there was no change in the growth rate of trout exposed to 0.033mgl^{-1} UIA-N,

but after 6 and 12 months, there was a considerable drop. The maximum allowable hazardous concentration for growth, according to **Wajsbrot *et al.* (1993)** was 0.27–0.47mgl^{-1} UIA-N.

El-Sherif and El-Feky (2008) reported that the FCR attained in the fourth UIA-N concentration (0.15mg/ l) was significantly higher than that attained in the first ones (4.6 and 2.8, respectively), the current results, as shown in Table (2), are consistent with those obtained by **El-Sherif and El-Feky (2008)**, who reported that mean feed conversion ratio of the tilapia increased as UIA-N concentrations increased. According to **De Azevedo *et al.* (2015)**, daily feed intake was found to be 0.54g on average per day, with water salinity levels at 14 and 21gl^{-1} having no effect on the Nile tilapia. The other aspects of fish performance were impacted by the salinity levels of the water.

According to **Ercan *et al.* (2015)**, the European sea bass (*Dicentrarchus labrax*) fed *ad libitum* at 24‰ salinity throughout the November–January period had an FCR value of 3.67 in the first period, which was substantially greater than that of the other groups. The group that was given 2.5% of its weight at 18‰ salinity had the lowest value, 2.21, according to the calculations. Additionally, the group fed 2.5% of its weight at 18‰ salinity had the lowest FCR value, calculated to be 1.05, and the group fed *ad libitum* at 24‰ salinity had the highest value, calculated to be 2.63. This group's FCR value was significantly higher than that of the other groups at 10‰ salinity level in the November–January period. Moreover, both SGR and FCR fed *ad libitum* had significantly higher values than those fed 2.5% of its weight. That was the same only for FCR at 24‰ salinity level in November-January period **(Ercan *et al.*, 2015)**.

Atle *et al.* (2004) discovered that when UIA-N concentrations rose, the mean feed conversion ratio dropped. **El-Shafai *et al*. (2004)** also demonstrated that ammonia concentrations greater than 0.068mg l^{-1} UIA-N had an impact on feed conversion. They also found that there was no difference in feed conversion rates (FCR) between the control group (0.004mg l^{-1} UIA-N) and those exposed to 0.068mg l^{-1} UIA-N, with FCR of 1.5 and 1.6, respectively, and those exposed to 0.14 and 0.26mg l^{-1} UIA-N, respectively. Conversely, **John and Semra (2001)** found no impact on the growth or feed conversion ratio of the blue tilapia and channel catfish at 0.91mg l^{-1} UIA-N.

Hematological indices of the red tilapia

Hematological indices, such as RBCs, WBCs, and Htc, are used to periodically check data on aquatic animals' nutritional condition and physiological reactions **(NRC, 2011)**. WBC count and infection severity are directly correlated **(Douglas *et al.*, 2010)**. The interaction between unionized ammonia and water salinity of the red tilapia (*Oreochromis* sp.) did not alter the mean corpuscular volume (MCV), hematocrit (Hct), or red blood cell count (RBCs), according to the current result displayed in Table (3). There were significant differences ($P< 0.05$) in the counts of white blood cells (WBCs), hemoglobin (Hb), platelets (Plt), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC).

The results are comparable to those of **De Azevedo *et al*. (2015)**, who discovered that blood parameters for hematocrit values, erythrocyte counts, and leukocyte counts were adequate for water salinity levels of 0 and 7g l^{-1}, indicating healthy fish. In contrast, the values obtained for water salinity levels of 14 and 21g l^{-1} were insufficient, indicating that the fish may have experienced stress due to the higher salinity levels.

Elarabany *et al.* (2017) reported that various water salinity levels had a noteworthy impact on a few estimated parameters. Their study findings demonstrated a reduction in the levels of hemoglobin, red blood cells, and hematocrit in the Nile tilapia treated groups at 8 and 12g/ l. According to **Sherif *et al*. (2013)**, the Nile tilapia fish exposed to 0.5mg l^{-1} experienced hemolytic anemia; when compared to other groups, the hemogram studies showed a substantial drop in RBCs, WBCs, Hb, Hct, MCV, MCH, and MCHC. After 14 days of exposure, fish subjected to 0.5mg l^{-1} of NH_3 were unable to return to normal levels. Additionally, it was clear that the Nile tilapia was anemic. **Atle *et al.* (2004)** and **El-Sherif and El-Feky (2008)** reported that the average Hct in the experimental groups declined as NH_3 concentrations increased.

The current findings were in conflict with those of **El-Sherif and El-Feky (2008)**, who demonstrated that as UIA-N concentrations in the Nile tilapia (*O. niloticus*) increased, the hematocrit value decreased for the UIA-N concentrations of 0.01, 0.05, 0.1, 0.15, and 0.004mg/ l, respectively. The average Hb content of *O. niloticus* fingerlings at the UIA-N varied, according to **El-Sherif and El-Feky (2008)** (0.05, 0.1, 0.15 and 0.004mg/ l). On the other hand, there were no notable ($P>$ 0.05) variations between the control (0.004mg/ l) and UIA-N concentration (0.01mg/ l). The hematopoietic tissue of the kidney, spleen, and liver produces teleost red blood cells. Ammonia stress can harm these organs to the point that erythrocyte production is reduced **(Das *et al.*, 2004)**.

Biochemical parameters

Fish exposed to water salinity (0.16%) did not exhibit any alteration in their glucose level, as demonstrated by **Kavya *et al*. (2015)**. The glucose level of the tilapia, *Sarotherodon melanotheron*, exposed to 9ppt salinity did not change. However, fish exposed to 18ppt for 72 hours had a relatively high glucose level and their liver tissue's glycogen level significantly decreased ($P<$ 0.05). This indicates that higher water salinity causes a high rate of glycogenolysis activity to meet high energy demands, which in turn results in lower liver glycogen levels.

The current glucose and urea results are comparable to those of **Sherif *et al*. (2013)**, who demonstrated that exposure to varying NH_3 concentrations considerably increased glucose levels and had a significant influence of NH_3. *O. niloticus* exposed to 0.5mg l^{-1} observed the significant maximum glucose level. **Sherif *et al*. (2013)** showed that elevated levels of NH_3 had a deleterious impact on the kidneys and gills based on creatinine and urea concentrations. The highest values recoded for creatinine and urea were 1.43 and 7.5mg/ dl, respectively, in *O. niloticus* treated with 0.5mgl^{-1}. Additionally, **Evans *et al.* (2006)** reported that fish blood glucose levels dramatically increased with higher NH_3 exposure duration and concentrations.

The current albumin and creatinine results are comparable to those reported by **Salah El-Deen (1999)**, who reported that *O. niloticus* exposed to 0.5mg l^{-1} had the highest recoded creatinine value of 1.43mg/ dl, suggesting that high levels of NH_3 had a negative impact on kidney function. The results obtained are comparable to those of **Sherif *et al.* (2013)**, who found that *O. niloticus* exposed to 0.5mg l^{-1} had the highest levels of liver enzymes, ALT and AST (11.7 and 42.3U/ l, respectively). These levels significantly decreased after the high level of NH_3 exposure was stopped (8.7 and 35.4U/ l, respectively). These findings are consistent with those of **Abbas (2006)**, who found that after 6 hours of exposure to 0.93mg l^{-1} NH_3-N at pH 7.5, the ALT and AST of *Cyprinus carpio* fingerlings drastically reduced, and they continued to increase for the remaining 7 days of the trial. According to **Niels *et al.* (1998)**, these elevations in liver enzymes might be a sign of tissue necrosis.

Histological examination

A number of researchers have published findings that differ from the current investigation. For example, **Sherif *et al.* (2013)** demonstrated that the largest pathological alterations in *O. niloticus* toxicity with ammonia occurred in the gills, liver, kidney, spleen, and muscles. The gills were congested, suggesting that *O. niloticus* was under stress. When compared to the group exposed to 0.5mg l^{-1} of NH_3, *O. niloticus* subjected to 0.05 and 0.1mg l^{-1} of NH_3 demonstrated normal condition. The gills of *O. niloticus* exposed to 0.5mg l^{-1} of NH_3 displayed blood vessel congestion, inflammatory cell infiltration, degeneration and necrosis in the epithelium lining the lamella with telangiectasis, hyperplasia (shaped like a bladder), and an increase in chloride cells in certain lamella and additional lamella showed tip fusion linked to an increase in chloride cell count **(Sherif *et al.*, 2013)**.

When fish are unable to expel the metabolic waste product, blood-ammonia levels rise and internal organs are harmed **(Thurston *et al.*, 1978; Daud *et al.*, 1988)**. This can result in chronic or sublethal exposure to UIA and tissue degeneration of the gills and kidneys. According to **Malik *et al.* (1986)**, lesions caused by sublethal ammonia exposure for 28 days were observed to change in the gill tissues of common carp (*Cyprinus carpio*). Ammonia exposure typically resulted in degenerative lesions, while chronic exposure mainly caused proliferative lesions.

According to **Sherif *et al.* (2013)**, *O. niloticus* exposed to 0.5mg l^{-1} experienced gill injury. These results were inconsistent with those reported by **El-Shebly and Gad (2011)**, who found that *O. niloticus* exposed to 0.1mg l^{-1} NH_3 had a slight pathological alteration in its gills, while two other groups of fish exposed to 0.2 and 0.4mg l^{-1} NH_3) displayed severe hyperplasia in their gills, including lifting of the epithelium lining, fusion between lamellae, and congestion in blood vessels.

According to **De Azevedo *et al.* (2015)**, the first treatment showed both mild changes, such as epithelial lifting and secondary lamellae fusion, and more severe changes, such as telangiectasia in a small number of secondary lamellae.

Small changes such epithelial lifting, secondary lamella fusion, and cell aggregation in the primary lamella were noted in water with a salinity of 7g l^{-1}. More severe changes included telangiectasia and aneurysms in a few secondary lamellae **(De Azevedo *et al.*, 2015)**. Slight changes, including bifurcation and fusion of secondary lamellae and epithelial lifting, were noted in water salinity of 14g l^{-1}. Severe changes, including telangiectasia and aneurysms, were more common than in earlier treatments. A higher number of minor changes, including epithelial lifting, secondary lamellae fusion, secondary lamellae structural changes, chloride cell hypertrophy, and primary lamella cell aggregation, were seen in water with a salinity of 21g l^{-1}. Among the severe changes, telangiectasia and aneurysms were noted **(De Azevedo *et al.*, 2015)**. Additionally, they demonstrated that since fish exposed to elevated metabolic ammonia are known to be more sensitive to bacterial gill diseases, the ensuing gill lesions may result in decreased oxygen transport across membranes and predispose fish to bacterial infections.

The current kidney tissue histology results are comparable to those reported by **Sherif *et al*. (2013)**, who demonstrated that the kidneys of fish given a high dose of ammonia (0.5mg l^{-1}) showed signs of renal blood vessel congestion, hemorrhage between renal tubules, edema in some glomeruli, shrinkage of the glomerular tuft in other glomeruli, degeneration and necrosis of the renal tubules' epithelial lining, and complete sloughing of the tubules' epithelium lining.

According to **Benli *et al.* (2008)**, the Nile tilapia kidney tissues showed signs of hyperemia and glomerulonephritis following exposure to varying sublethal ammonia concentrations (0.07, 0.14, 0.35, and 0.71mg l^{-1} NH_3–N). Moreover, hyaline degradation in renal tubules and thrombus formation in renal blood arteries were observed by **El-Sherif and El-Feky (2008)**. The pathological alterations in renal tissue are linked to elevated levels of urea and creatinine in *O. niloticus* serum subjected to varying unionized ammonia concentrations.

The current liver tissue results are comparable to those reported by **Sherif *et al*. (2013)**, who found that the liver displayed severe vacuolar degenerative changes in hepatocytes, hemorrhage, focal infiltration with inflammatory cells, an increase in the melonomacrophage center, and the main hepatic lesions of *O. niloticus* exposed to 0.5mg l^{-1} of NH_3. Other hepatocytes showed necrosis. Fish subjected to varying NH_3 concentrations had significantly increased liver tissues, and the extent of this activity increase was positively correlated with the ammonia concentration **(Hegazi, 2011)**.

The hepatocyte damage in *O. niloticus* exposed to 0.05, 0.1, and 0.5mg l^{-1} NH_3 was confirmed by an increased level of liver enzyme in serum. The histopathological examination of the liver revealed congestion of blood vessels and sinusoids, focal infiltration with inflammatory cells, an increase in melanomacrophage center, vacuolar degeneration, and necrosis of the hepatocyte. The severity of pathological alteration varied depending on the dose. These findings were reported by **Thurston *et al.* (1984)**, **El-Shafai *et al.* (2004)** and **El-Sherif and El-Feky (2008)**.

CONCLUSION

According to the current study, increasing water salinity might reduce ammonia toxicity. The red tilapia exhibits notable histological alterations in addition to alterations in blood properties as a result of unionized ammonia concentration and water salinity levels. To guarantee high output in fish farms, the red tilapia should be farmed in low-unionized ammonia water while all other environmental parameters are kept at a safe level.

REFERENCES

Abbas, H.H. (2006). Acute toxicity of ammonia to common carp fingerlings *Cyprinus carpio* at different pH levels. Pakistan Journal of Biological Sciences, 9(12): 2215-2221.

Alabaster, J. S.; Shurben, D. G. and Knowles, G. (1979). The effect of dissolved oxygen and salinity on the toxicity of ammonia to smolts of salmon, *Salmo salar L*. Journal of Fish Biology, 15: 705-712.

Alkobaby, A. I. and Hassanien, H. A. (2007). Acute and chronic ammonia toxicity to Nile tilapia (*Oreochromis niloticus*) fingerlings. Egyptian Journal of Aquatic Research, 33(1): 455-465.

Atle, F.; Siikavuopio, S. I.; Saether, B. S. and Evensen, T.H. (2004). Effect of chronic ammonia exposure on growth in juvenile Atlantic cod. Aquaculture, 237: 179-189.

Benli, A.C.K.; Koksal, G. and Ozkul, A. (2008). Sublethal ammonia exposure of Nile tilapia (*Oreochromis niloticus* L.): Effects on gill, liver and kidney histology. Chemosphere, 72 (9): 1355-1358.

Brecher, G.; Schneiderman, M. and Cronkite, E.P. (1953). The reproducibility of the platelet count. American Journal of Clinical Pathology, 23:15-21.

Dacie, J.V. and Lewis, S.M. (1991): Evaluation of R.B. Cs in hematology. 7th edition, the English language book society and Churchill living stone. Pp.37-58.

Das, P. C.; Ayyappan, S.; Jena, J. K. and Das B.K. (2004). Acute toxicity of ammonia and its sublethal effects on selected haematological and enzymatic parameters of mrigal, *Cirrhinus mrigala* (Hamilton). Aquaculture Research, 35(2): 134-143.

Daud, S. K.; Hasbollah, D. and Law, A. T. (1988). Effects of unionized ammonia on red tilapia (*Oreochromis mossambicus* × *O. niloticus* hybrid) fry, In R.S.V. Pullin, T. Bhukaswan, K. Tonguihai and J. L. Maclean (eds.) The Second International Symposium on Tilapia in Aquaculture, 411-413.

De Azevedo, R. V.; De Oliveira, K. F.; Flores-Lopes, F.; Teixeira-Lanna, E. A.; Takishita, S. S. and Tavares-Braga, L. G. (2015). Responses of Nile tilapia to different levels of water salinity. Latin American journal of aquatic research, 43(5): 828-835.

De Silva, F. J. R.; Dos Santos Lima, F. R.; Do Vale, D. A. and Do Carmo, M. V. (2013). High levels of total ammonia nitrogen as NH_4^+ are stressful and harmful to

the growth of Nile tilapia juveniles. Acta Scientiarum. Biological Sciences, 35(4): 475-481.

Dosdat, A.; Ruyet, J.P.; Coves, D.; Dutto, G.; Gasset, E.; Le Roux, A. and Gilles, L. (2003). Effect of chronic exposure to ammonia on growth, food utilization and metabolism of the European sea bass (*Dicentrarchus labrax*). Aquatic Living Resources, 16: 509-520.

Douglas, J. W.; Jane, W.K. and Schalm, O.W. (2010). Schalm's Veterinary Hematology, 6th ed. Wiley-Blackwell, Ames, Iowa, pp. xxiii, 1206.

Doumas, B.T. (1975). Colorimetric determination of total protein in serum or plasma. Clinical Chemistry, 21(8):1159-1166.

Drabkin, D.L. and Austin, J.H. (1932). Spectrophotometric studies; spectrophotometric constant for common hemoglobin derivatives in human, dog and rabbit. Journal of Biological Chemistry, 98: 719-733.

Elarabany, N.; Bahnasawy, M.; Edrees, G. and Alkazagli, R. (2017). Effects of salinity on some haematological and biochemical parameters in Nile tilapia, *Oreochromis niloticus.* Agriculture, Forestry and Fisheries, 6: 200-205.

El-Shafai, S. A.; El-Gohary, F. A.; Nasr, F. A.; Van Der Steen, N. P. and Gijzen, H. J. (2004). Chronic ammonia toxicity to duckweed-fed tilapia (*Oreochromis niloticus*). Aquaculture, 232(1-4): 117-127.

El-Shebly, A. A. and Gad, H. A. M. (2011). Effect of chronic ammonia exposure on growth performance, serum growth hormone (GH) levels and gill histology of Nile tilapia *(Oreochromis niloticus*). Journal of Microbiology and Biotechnology Research, 1(4): 183-197.

El-Sherif, M. S. and El-Feky, A. M. (2008). Effect of ammonia on Nile Tilapia (*O. niloticus*) performance and some hematological and histological measures. In Eighth International Symposium on Tilapia in Aquaculture. Cairo, Egypt. 1: 513-530.

Emerson, K.; Russo, R. C.; Lund, R. E. and Thurston, R. V. (1975). Aqueous ammonia equilibrium calculations effects of pH and temperature. Journal of the Fisheries Research Board of Canada, (32): 2379-2383.

Ercan, E.; Agrali, N. and Tarkan, A. S. (2015). The effects of salinity, temperature and feed ratio on growth performance of European sea bass (*Dicentrarchus labrax* L., 1758) in the water obtained through reverse osmosis system and a natural river. Pakistan Journal of Zoology, 47(3): 625-633.

Evans, J. J.; Park, D. J.; Brill, G. C. and Klesius, P. H. (2006). Unionized Ammonia Exposure in Nile Tilapia: Toxicity, Stress Response and Susceptibility to *Streptococcus agalactiae*. North American Journal of Aquaculture, 68: 23-33.

Harris, J. O.; Maguire, G. B.; Edwards, S. and Hindrum, S. M. (1998). Effect of ammonia on the growth rate and oxygen consumption of juvenile greenlip abalone, *Haliotis laevigata* Donovan, 1808. Aquaculture, 160: 259-272.

Hassanen, G.D.; Salem, M.; Younes, M.I. and Abd Elnabi, H.E. (2014). Combined effects of water temperature and salinity on growth and feed utilization of juvenile red tilapia (*Oreochromis niloticus x O. aureus*). World Journal of Zoology, 9(1): 59-70.

Hegazi, M.M. (2011). Effect of chronic exposure to sublethal of ammonia concentrations on NADP+-dependent dehydrogenases of Nile tilapia liver. Egyptian Journal of Aquaculture Biology and Fisheries, 15(1): 15-28.

Henry, R.J. (1974). Clinical chemistry, principles and techniques, 2nd Edition, Harper and row, p.525.

Imsland, A. K.; Foss, A.; Gunnarsson, S.; Berntssen, M. H. G.; FitzGerald, R.; Bonga, S. W.; Ham, E. V.; Naevdal, G. and Stefansson, S. O. (2001). The interaction of temperature and salinity on growth and food conversion in juvenile turbot (*Scophthalmus maximus*). Aquaculture, 198: 353-367.

Joel, O. and Amajuoyi, C. S. (2010). Determination of the concentration of ammonia that could have lethal effect on fish pond. ARPN Journal of Engineering and Applied Science, 1-4.

John, A. H. and Semra, K. (2001). Effects of dial unionized ammonia fluctuation on juvenile hybrid striped bass, channel catfish, and blue tilapia. Aquaculture, 195:163-181.

Kaneko, T.; Shiraishi, K.; Katoh, F.; Hasegawa, S. and Hiroi, J. (2002). Chloride cells during early life stages of fish and their functional differentiation. Fisheries Science, 68: 1-9.

Kavya, K. S.; Kulkarni, R. S. and Jadesh, M. (2015). Some blood biochemical changes in response to saline exposure in the fresh water fish, *Notopterus notopterus* (Pallas). International Letters of Natural Sciences, 4.

Kida, A. D. and Ruddin, A. S. (2019). Long-term combined effects of salinity and temperature on the growth of juvenile red tilapia (*Oreochromis niloticus*). Journal of Critical Reviews, 7(2): 56-61.

Li, M.; Yu, N.; Qin, J.G.; Li, E.; Du, Z. and Chen, L. (2014). Effects of ammonia stress, dietary linseed oil and Edwardsiella ictaluri challenge on juvenile darkbarbel catfish *Pelteobagrus vachelli*. Fish and Shellfish Immunology, 38(1): 158–65.

Malik, E.S.; Gyore, K. and Olah, J. (1986). Effects of ammonia on gill tissues of common carp (*Cyprinus carpio* L.). Aquacult. Hung, 5: 97-105.

Mena, H. A.; Sumano, L. H. and Macias, Z. R. (2002). Effects of water salinity on the growth of hybrid red tilapia *O. niloticus* (Linnaeus) x *O. mossambicus* (Peters) cultured under controlled laboratory condition. Veterinaria México, 32(1): 39-48.

Niels, E. V.; Korsgaard, B. and Jenson, F. B. (1998). Isolated and combined exposure to ammonia and nitrite in rainbow trout *Oncorhynchus mykiss*: Effect on electrolyte status, blood respiratory properties and brain glutamine/glutamate concentrations. Aquatic Toxicology, 41: 325-342.

NRC, Nutrient Research Council (2011). Nutrient Requirements of Fish and Shrimp. National Academy Press, Washington: DC, USA.

Ogbonna, J. and Chinomso, A. (2010). Determination of the concentration of ammonia that could have lethal effect on fish pond. Journal of Engineering and Applied Sciences (Asian Research Publishing Network), 5: 1-5.

Patton, C.J. and Crouch, S.R. (1977). Urea colorimetric end point determination urease-berthelot reaction. Analytical Chemistry, (49): 464- 469.

Pearse, A.G. (1972). Histochemistry: theoretical and applied. Volume 1. Churchill Livingstone publisher, London.

Reitman, S. and Frankel, S. (1957). A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. American journal of clinical pathology, 28(1): 56-63.

Saeed, S. M. and Shaker, I. M. (2008). Assessment of heavy metals pollution in water sediments and their effect on *Oreochromis niloticus* in the northern Delta lakes, Egypt. 8th International symposium on Tilapia in Aquaculture, Pp. 475-490.

Salah El-Deen, M. A. (1999). Toxicology and physiological effects of ammonia on grass carp *Ctenopharyngodon idella* at different pH levels. Egyptian Journal of Zoology, 33: 219-235.

Sampaio, L. A.; Wasielesky, W. and Mirandafilh, K. C. (2002). Effect of salinity on acute toxicity of ammonia and nitrite to juvenile *Mugil platanus*. Bulletin of Environmental Contamination and Toxicology, 68: 668-674.

Seiverd, C.E. (1964). Hematology for Medical Technologists. Lea and Febiger, Philadelphia, p. 946.

Sherif, A. H.; Soliman, H. A. and Heba, H. E. (2013). Negative impacts of unionized ammonia (NH_3) on health status of cultured *Oreochromis niloticus.* Abbassa International Journal for Aquaculture, 6(2): 319-347.

Smith, C. E. and Piper, R. G. (1975). Lesions associated with chronic exposure to ammonia. The Pathology of Fishes. University of Wisconsin Press, Madison WI, Pages 497-514 in: W. E. Ribelin and G. Migaki, eds.

Sorrell-Raschi, L.A. and Tomasic, M. (1998). Evaluation of automated methods of measuring hemoglobin and hematocrit in horses. American journal of veterinary research, 59(12): 1519-1522.

Statistical Analysis System, (1996). SAS procedure Guide "Version 6.12 ed". SAS Institute Inc., Cray, NC, USA.

Szumnski, D. S.; Barton, D. A.; Puntam, H. D. and Polta, R. C. (1982). Evaluation of EPA unionized ammonia toxicity criteria. Journal of the Water Pollution Control Federation, 54: 282- 291.

Thurston R.V.; Russo R.C. and Smith C.E. (1978). Acute toxicity of ammonia and nitrite to cutthroat trout fry. Transactions of the American Fisheries Society, 107: 361-368.

Thurston, R. V.; Russo, R. C.; Lauedtke, R. T.; Smith, C. E.; Meyn, E. L.; Chakoumakos, C.; Wang, K. C. and Brown, C. J. D. (1984). Chronic toxicity of ammonia to rainbow trout. Transactions of the American Fisheries Society, 113:56–73.

Tietz, N.W. (1986): Textbook of clinical chemistry, Saunders, W.B. Co. London Philadelphia, p. 796.

Timmons, M. B.; Ebeling, J. M.; Wheaton, F. W.; Summerfelt, S. T. and Vinci, B. J. (2002). Recirculating Aquaculture Systems. p. NRAC Publication, No.01-002.

Wajsbrot, N.; Gasith, A.; Diamant, A. and Popper, D. M. (1993). Chronic toxicity of ammonia to juvenile gilthead seabream *Sparus aurata* and related histopathological effects. Journal of Fish Biology, 42: 321–328.

Wood, C. M.; Milligan, C. L. and Walsh, P. J. (1999). Renal responses of trout to chronic respiratory and metabolic acidosis and metabolic alkalosis. American Journal of Physiology Regulatory, Integrative and Comparative Physiology, 277: R482-R492.

Young, D.S. (1995): Effects of drugs on clinical lab. Tests, 4th Edition AACC press.

Zimmer, A. M.; Brauner, C. J. and Wood, C. M. (2014). Ammonia transport across the skin of adult rainbow trout (*Oncorhynchus mykiss*) exposed to high environmental ammonia (HEA). Journal of Comparative Physiology B, 184(1): 77-90.

ARABIC ABSTRACT

قدرة أسماك البلطي الأحمر (*Oreochromis* sp.) على تحمل تراكيزات مختلفة من الأمونيا غير المتأينة ومستويات مختلفة من ملوحة المياه

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إن قدرة أسماك البلطي الأحمر (*Oreochromis* sp.) على تحمل تركيزات الأمونيا غير المتأينة ومستويات ملوحة المياه المختلفة على أداء النمو، والاستفادة من الغذاء، والحالة الفسيولوجية هي المحور الرئيسي للدراسة. تم دراسة ثلاثة تراكيزات مختلفة من الأمونيا غير المتأينة 0.15، 0.20، 0.25 ملجم/لتر بالإضافة إلى مستويين مختلفين من ملوحة الماء 25 و30 جزء في الألف في كل من المعاملات الستة (من T1 إلى T6)، ولكل منها ثلاث مكررات. يحتوي ثمانية عشر حوضًا مائيًا على إجمالي 180 سمكة بلطي أحمر موزعة بالتساوي فيما بينها. حيث تم وضع عشرة أسماك بشكل عشوائي لكل حوض من هذه الأحواض، بمتوسط وزن جسم ابتدائي 19.42 ± 0.04 جم وطول جسم ابتدائي 8.48 ± 0.51 سم. تمت تغذية أسماك التجربة ثلاث مرات يوميًا، عند الساعة 08:00 و13:00 و18:00، بأعلاف تجارية (30% بروتين خام) بمعدل 3% من إجمالي كتلتها الحيوية لمدة 8 أسابيع. تم أخذ عينات دم من خمس أسماك مختارة عشوائياً من الأحواض الثلاثة لكل معاملة في نهاية التجربة بهدف قياس مؤشرات الدم والكيمياء الحيوية. أظهرت النتائج أن التفاعل بين الأمونيا غير المتأينة وملوحة المياه لم يكن له تأثير ملحوظ على غالبية مؤشرات النمو والاستفادة من الغذاء. فكانت مستويات كرات الدم البيضاء والصفائح الدموية وMCHC أعلى في المعاملات T5 وT3 وT4 على التوالي. وكانت المعاملة T1 أعلى في قيم الجلوبيولين وتركيزات اليوريا. وسجلت المعاملة T2 أكبر قيم لإنزيمات الكبد ALT وAST. كما تشير الدراسة الحالية إلى أن مستويات الأمونيا غير المتأينة المختلفة أثرت على صحة الأسماك ونوعيتها، لكن ملوحة المياه قللت من هذه التأثيرات قليلاً وسمحت لنمو البلطي الأحمر في مياه الآبار ذات الملوحة العالية.