

## Effects of aluminum chloride on some hematological and biochemical parameters of *Tilapia zillii* G

EL Sayed A. Shokr

Central Lab. of Aquaculture Research, Abbasa, Egypt

### ABSTRACT

*Tilapia zillii* fish were exposed to aluminum chloride under alkaline water (pH 7.8). The fish were divided into 4 groups control group and three groups exposed to 50, 100 and 200µg / liter of aluminum chloride respectively for 24 hr., 48 hr. and 96 hr.

The fish showed a physiological response after 24 hr. of exposure to 50, 100 and 200µg / liter of aluminum chloride respectively where, approximately 100% increase in red blood cells (RBCs), white blood cells (WBCs), hematocrit value (Hc) and hemoglobin content (Hb) respectively. Also, there are increased in serum glucose level, total protein, aspartate and alanine amino transferase activities (AST and ALT) and increased serum creatinine and uric acid concentration after 24hr. from exposure the fish to 50, 100 and 200µg / liter of aluminum chloride respectively. The results indicate that the fish that exposed to 50, 100 and 200µg / liter of aluminum chloride respectively for 48 hr. increase blood constituents, serum glucose level, serum total protein and serum AST, ALT activities, creatinine and uric acid level. Moreover, the fish that exposed to 50, 100 and 200µg / liter of aluminum chloride respectively for 96 hr. showed highly significant increase of RBCs, WBCs, Hc and Hb as well as in the serum glucose level, serum total protein and serum AST and ALT activities. The serum creatinine and uric acid showed increased in the fish that exposed to 50, 100 and 200µg / liter of aluminum chloride respectively for 96 hr. These data suggest that fish respond to aluminum exposure by increasing their WBCs, enzymes of liver functions and kidney functions.

**Keywords:** *Tilapia zillii*, aluminum chloride, biochemical parameters

### INTRODUCTION

The bioavailability and toxicity of aluminum varies with its chemical speciation. In the case of fish, higher polymers are less toxic than monomers and polymers of low relative molecular mass. Polymerization is a slow process; hence the biological activity of Aluminum in water depends not only on aluminum concentration and ecological conditions such as pH, temperature and presence of complexing ions, but can also depend on the pre-history of the water (Rosseland & Staurnes, 1994). Gundersen *et al.* (1994) studied the effect of exposed rainbow trout (*Oncorhynchus mykiss*) to aluminum at pH values ranging from 7.97 to 8.56 in 96-h tests. No significant mortality was observed at

pH 8.33 or less and filterable aluminum concentrations of 0.52 mg/liter or less. However, 100% mortality was found at pH 8.58 and a filterable aluminum concentration of 1 mg/liter. The 96-h LC<sub>50</sub> values ranged from 0.36 to 0.79 mg filterable aluminum / liter at weakly alkaline pH levels. Cleveland *et al.* (1991) studied the effect of exposed brook trout (*Salvelinus fontinalis*) to an acid aluminum concentration of 200 µg/liter for 56 days under flow-through conditions at pH 5.3, 6.1 and 7.2. The weights of trout exposed to pH 5.3 and 6.1 did not differ significantly throughout the study. After day 3 fish exposed to pH 7.2 weighed significantly more than those at pH 5.3 and 6.1. Mortality was significantly higher in brook trout exposed to pH 5.3 than in those exposed to pH 6.1 (except on day 56) or 7.2. Witters (1986) studied the effect of total aluminum (350 µg/liter), pH (4.1 and 6.1) and calcium concentration (38 and 190 µEq / liter) on ion balance and hematology in rainbow trout (*Oncorhynchus mykiss*) exposed for 3.5 h. None of the treatment combinations affected the number of erythrocytes or the hemoglobin content. The hematocrit value was increased by aluminum. Naskar *et al.* (2006) studied the effect of aluminum toxicity on erythrocytes, as decreased in cytoplasmic volume owing to reduced Hb content or increased in the amount of water content within the cell resulting from osmotic disequilibrium. In this context, the abnormal surface membrane morphology could be attributed to cytoskeleton fragility and defects in structural proteins. Allin and Wilson (1999) studied swimming behavior was a more sensitive index of exposure to aluminum than feeding. Fish exposed to aluminum had significantly fewer red blood cells and lower hematocrit than the controls, indicating hemodilution. Aluminum is known to act as a respiratory toxicant, restricting aerobic scope. Hytterod and Poleo (2003) showed a physiological response after 3 weeks of exposure; approximately 300% increase in blood glucose concentration and about 30% increase in blood hematocrit. Peuranen *et al.* (2007) reported disturbances in ion regulation, increases in hematocrit value (Hct) and hemoglobin (Hb) concentration and reductions in oxygen consumption. Vuorinen *et al.* (2003) showed that an increase in blood glucose concentration was detected only near spawning time, from October to November, coincident with Al accumulation inside the gill tissue and increase in Al toxicity in vendace. Allin and Wilson (2000) reported that the Hematological disturbances were most extreme in the aluminum-naive fish and had not recovered to control levels 6 days after the end of the pulse and exposure to low levels of aluminum may be an important factor abating the impact of aluminum on fish in the natural environment. McKee *et al.* (1989) showed that the Growth and survival were significantly reduced at the two highest aluminum concentrations by day 60. RNA and DNA were the most sensitive biomolecules monitored and were significantly reduced at the three highest aluminum concentrations by day 60. RNA: protein and RNA: DNA ratios were no more responsive than growth and survival. Pekka *et al.* (2002) showed that a clear decrease in plasma Na<sup>+</sup> and Cl<sup>-</sup> and an increase in serum glucose level was

detected only near spawning time, from October to November, coincident with Al accumulation inside the gill tissue.

The objective of the present study is investigating the toxic effects of aluminum chloride on the RBCs and WBCs, as indices of blood cells disturbance, haematocrit value, hemoglobin content and serum total protein and serum glucose level, the activities of (AST, ALT) and creatinine and uric acid levels as indices of stress of aluminum chloride on *Tilapia zillii*.

## MATERIAL AND METHODS

### Experimental Animals

*Tilapia zillii* G., 80 - 90 g body weight was obtained from Om Ehfain fish farm, Om Elrozam, Libya. The fish were held in large tanks contained water from their farm, where the physical and chemical properties of this water were as following. pH was approximately 7.8, dissolved oxygen was approximately 6.2 ml / L, and salinity was approximately 7.1 ‰ for a period of two months. The fish were fed on 30 % protein during this time. The fish were divided into four groups, control group and three groups exposed to 50, 100 and 200 µg / L of aluminum chloride respectively. The period of exposure was 24 hr., 48 hr. and 96 hr. Each group consists of two-glass aquaria 30 x 60 x 40 cm<sup>3</sup>. Each glass aquarium contains 10 fish. The blood samples were taken from six fish after 24 hr. exposure to AlCl<sub>3</sub> from arterial caudalis with heparinized syringes from control group and 50, 100 and 200 µg / L of aluminum chloride exposure groups respectively according to the methods of Soivio *et al.* (1972). The blood was divided into two parts, the first part was analyzed for RBCs and WBCs account, hematocrite value and hemoglobin content. The second parts of the blood were centrifuged at 3000 rpm for 15 mins to separate the serum for measuring glucose level, total protein, the activities of AST and ALT and creatinine and uric acid levels. After 48 hr. exposure the fish to 50, 100 and 200 µg / L of aluminum chloride the six blood samples were taken from control and exposure groups and the blood samples were performed as outlined in the protocol above. The last six blood samples were taken from control fish group and 50, 100 and 200 µg / L of aluminum chloride exposure groups to measure the RBCs, WBCs, haematocrit value, hemoglobin content, serum glucose level, total protein, the activities of AST and ALT, creatinine and uric acid levels.

### Analytical Techniques

Blood samples were drawn from arterial caudalis with heparinized syringes. Red blood cells and white blood cells were determined by hemocytometres Improved Neubauer, Hawksley, Germany. Hematocrite value was determined by using a microcapillary tubes where, these tubes filled with blood immediately after blood collection and centrifuged in a microcapillary centrifuge. The amount of red blood cells was determined as a percentage of total blood (hematocrit) by the use of microcapillary reader. Hemoglobin

content measured according to Dacie and Lewis (1975). The remaining blood samples were centrifuged for determination of concentration of serum glucose, total protein, the activities of AST, ALT activities, creatinine and uric acid. The glucose was determined by enzymatic colorimetric method according to Trinder (1969). The total protein was determined by rapid colorimetric method according to Peter (1968). The activities of AST and ALT were determined by colorimetric method according to Reitman and Frankel (1957). The creatinine and uric acid levels were determined by colorimetric methods according to Henry (1974).

### Statistical Analysis

Data were reported as means  $\pm$  S. E. (n). significant differences ( $p < 0.05$ ) within each group were tested with Student's two-tailed t – test and one-way ANOVA by SPSS for windows XP2 2002. comparisons between groups were tested by Student's two-tailed t – test unpaired design, ( $p < 0.05$ ).

## RESULTS

### Hematology Parameters

The red blood cells (RBCs) in the groups of the fish that exposed to 50, 100 and 200  $\mu\text{g} / \text{L}$  of aluminum chloride for 24 hr. were significantly increased compared to control group as showed in Table (1). Where, RBCs showed highly significant increase in the fish that exposed to 50 and 100  $\mu\text{g} / \text{L}$  of aluminum chloride and very highly significant increase in the groups that exposed to 200  $\mu\text{g} / \text{L}$  of aluminum chloride more than the control group after 48 hr. Also, there is very highly significant increase in the RBCs in the groups that exposed to 50, 100, and 200  $\mu\text{g} / \text{L}$  of aluminum chloride respectively compared to the control group after 96 hr.

Table (1): Red blood cells (RBCs)  $\times 10^6$  changes in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	RBCs $\times 10^6$ after 24 hr.	RBCs $\times 10^6$ after 48 hr	RBCs $\times 10^6$ after 96 hr
Control	1333333.3 $\pm$ 57831	1435553.7 $\pm$ 57831	1521322.6 $\pm$ 57831
50 $\mu\text{g} / \text{L}$ AlCl <sub>3</sub>	1596666.7 $\pm$ 54873*	1830000 $\pm$ 55075**	2130000 $\pm$ 17320***
100 $\mu\text{g} / \text{L}$ AlCl <sub>3</sub>	1713333.3 $\pm$ 35276**	1880000 $\pm$ 36055**	2240000 $\pm$ 30550***
200 $\mu\text{g} / \text{L}$ AlCl <sub>3</sub>	1806666.7 $\pm$ 49328***	2006667 $\pm$ 90184***	2320000 $\pm$ 52915***

significant at  $p < 0.05$ , \*\* highly significant at  $p < 0.01$ , \*\*\* very highly significant at  $p < 0.001$ ,

Table (2) showed a significant increase in the white blood cells (WBCs) in the groups of the fish that exposed to 50, 100 and 200  $\mu\text{g} / \text{L}$  of aluminum chloride for 24 hr. compared to control group. Moreover, the WBCs showed very highly significant increased in the fish that exposed to 50, 100, and 200  $\mu\text{g} / \text{L}$  of aluminum chloride respectively compared to control group for 48 hr. and 96 hr. respectively.

Table (2): White blood cells (WBCs)  $\times 10^3$  changes in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	WBCs $\times 10^3$ after 24 hr.	WBCs $\times 10^3$ after 48 hr	WBCs $\times 10^3$ after 96 hr
Control	16866.6 $\pm$ 866.6	17985.6 $\pm$ 866.6	18547.6 $\pm$ 866.6
50 $\mu$ g / L AlCl <sub>3</sub>	18933.3 $\pm$ 762.3	76200 $\pm$ 2193.1***	199166.7 $\pm$ 4723.8***
100 $\mu$ g / L AlCl <sub>3</sub>	23566.6 $\pm$ 1026.8*	93733.3 $\pm$ 3339.8***	270000 $\pm$ 5773.5***
200 $\mu$ g / L AlCl <sub>3</sub>	30416.6 $\pm$ 1651.3***	133166.7 $\pm$ 9116.3***	303333 $\pm$ 8819***

significant at  $p < 0.05$ , \*\* highly significant at  $p < 0.01$ , \*\*\* very highly significant at  $p < 0.001$ .

Hemoglobin (Hb) was a significantly increase in the groups of the fish that exposed to 50  $\mu$ g / L of aluminum chloride and highly significant increased in the fish that exposed to 100 and 200  $\mu$ g / L of aluminum chloride after 24 hr. compared to control group. Also, the Hb content was highly significant increase in the fish that exposed to 50 and 100  $\mu$ g / L of aluminum chloride and very highly significant increased in the fish that exposed to 200  $\mu$ g / L of aluminum chloride after 48 hr. compared to control group as showed in Table ( 3 ). 200  $\mu$ g / L of aluminum chloride respectively compared to control group after 48 hr. and 96 hr. respectively. There are very highly significant increased in the fish that exposed to 50, 100 and 200  $\mu$ g / L of aluminum chloride after 96 hr. compared to control group. .

Table (3): Hemoglobin (Hb gm / 100ml) change in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters/groups	Hb gm / 100ml after 24 hr.	Hb gm / 100ml after 48 hr	Hb gm / 100ml after 96 hr
Control	7.11 $\pm$ 0.19	7.88 $\pm$ 0.19	8.41 $\pm$ 0.19
50 $\mu$ g / L AlCl <sub>3</sub>	8.99 $\pm$ 0.17*	9.76 $\pm$ 0.18**	10.71 $\pm$ 0.09***
100 $\mu$ g / L AlCl <sub>3</sub>	9.36 $\pm$ 0.12**	9.92 $\pm$ 0.12**	11.79 $\pm$ 0.40***
200 $\mu$ g / L AlCl <sub>3</sub>	9.68 $\pm$ 0.09**	10.36 $\pm$ 0.16***	12.39 $\pm$ 0.17***

significant at  $p < 0.05$ , \*\* highly significant at  $p < 0.01$ , \*\*\* very highly significant at  $p < 0.001$ ,

Hematocrit (Hct) was a significantly increased in the groups of the fish that exposed to 50  $\mu$ g / L of aluminum chloride, highly significant increased in the fish that exposed to 100  $\mu$ g / L of aluminum chloride and very highly significant increased in the fish that exposed to 200  $\mu$ g / L of aluminum chloride for 24 hr. compared to control group. Also, there were very highly significant increased in the hct of the fish that exposed to 50, 100 and 200  $\mu$ g / L of aluminum chloride for 48hr. and 96 hr. compared to control group as shown in Table (4).

Table (4): Hematocrite (Hct %) change in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	Hct %after 24 hr.	Hct % after 48 hr	Hct % after 96 hr
Control	22.25±0.57	23.75±0.57	24.66±0.57
50 µg / L AlCl <sub>3</sub>	25.96±0.54*	28.3±0.55***	31.09±0.04***
100 µg / L AlCl <sub>3</sub>	27.14±0.35**	28.8±0.36***	32.4±0.30***
200 µg / L AlCl <sub>3</sub>	28.06±0.28***	30.1±0.55***	33.2±0.52***

significant at  $p < 0.05$ , \*\* highly significant at  $p < 0.01$ , \*\*\* very highly significant at  $p < 0.001$ ,

### Biochemical Parameters

Table (5) showed that the activity of aspartate amino transferase (AST) in the serum of fish that exposed to different concentrations of aluminum chloride at different periods was changed. Where, AST in the serum of fish after 24 hr. exposure to aluminum chloride was a significantly increased in the groups of the fish that exposed to 50 µg / L of aluminum chloride, highly significant increased in the fish that exposed to 100 µg / L of aluminum chloride and very highly significant increased in the fish that exposed to 200 µg / L of aluminum chloride compared to control group. Also, there were very highly significant increased in the serum AST of the fish that exposed to 50, 100 and 200 µg / L of aluminum chloride for 48 hr. and 96 hr. compared to control group.

Table (5): The activity of aspartate amino transferase (AST U/ L) changes in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	AST U/ L after 24 hr.	AST U/L after 48 hr	AST U/ L after 96 hr
Control	6.46±0.50	7.84±0.50	8.86±0.50
50 µg / L AlCl <sub>3</sub>	11.3±0.75*	19.36±0.52***	26.4±0.49***
100 µg / L AlCl <sub>3</sub>	14.5±0.40**	22.36±1.09***	29.93±0.51***
200 µg / L AlCl <sub>3</sub>	19.16±0.61***	25.03±0.84***	27.86±0.31***

significant at  $p < 0.05$ , \*\* highly significant at  $p < 0.01$ , \*\*\* very highly significant at  $p < 0.001$ ,

The serum activity of alanine amino transferase (ALT) of the fish that exposed to 50 µg / L of aluminum chloride for 24 hr was no significantly increased compared to control groups Table (6). But, the serum ALT was significantly increased in the fish that exposed to 100 µg / L of aluminum chloride for 24 hr. and very highly significant increased in the serum ALT of the fish that exposed to 200 µg / L of aluminum chloride for 24 hr. compared to control groups. On the other hand, Table (6) showed that the serum ALT in the fish that exposed to 50, 100 and 200 µg / L of aluminum chloride for a period of 48 and 96 hr. respectively were very highly significant increased compared to control group.

Table (6): The activity of alanine amino transferase (ALT U/ L) changes in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	ALT U/ L after 24 hr.	ALT U/ L after 48 hr	ALT U/ L after 96 hr
Control	10.45±0.68	15.57±0.68	12.75±0.68
50 µg / L AlCl <sub>3</sub>	13.6±0.95	23.16±0.61***	27.23±0.61***
100 µg / L AlCl <sub>3</sub>	15.73±0.90*	25.86±0.80***	28.26±0.80***
200 µg / L AlCl <sub>3</sub>	20.8±0.88***	27.06±0.46***	29.46±0.46***

significant at  $p < 0.05$ , \*\* highly significant at  $p < 0.01$ , \*\*\* very highly significant at  $p < 0.001$ .

### Uric Acid And Creatinine Levels

The serum uric acid leveled was significantly increased in the fish that exposed to 50 and 100 µg / L of aluminum chloride for 24 hr. compared to control groups Table (7). But, the serum uric acid was highly significant increased in the fish exposed to 200 µg / L of aluminum chloride for 24 hr. compared to control groups. On the other hand, the serum uric acid in the fish that exposed to 50, 100 and 200 µg / L of aluminum chloride for a period of 48 hr. were significant, highly significant and very highly significant increased compared to control group respectively. Also, there were very highly significant increased in the serum uric acid in the fish that exposed to 50, 100 and 200 µg / L of aluminum chloride for a periods of 96 hr. compared to control group.

Table (7): Serum uric acid level mg % changes in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	Uric acid mg % after 24 hr.	Uric acid mg % after 48 hr	Uric acid mg % after 96 hr
Control	5.71±0.65	6.63±0.65	7.81±0.65
50 µg / L AlCl <sub>3</sub>	10.3±0.20*	12.06±0.14*	17.76±0.26***
100 µg / L AlCl <sub>3</sub>	11.4±0.57*	13.8±0.40**	18.8±0.26***
200 µg / L AlCl <sub>3</sub>	12.66±0.88**	14.66±0.89***	20.13±0.40***

significant at  $p < 0.05$ , \*\* highly significant at  $p < 0.01$ , \*\*\* very highly significant at  $p < 0.001$ .

There were no significant increased in the serum creatinine level of the fish that exposed to 50, 100 and 200 µg / L AlCl<sub>3</sub> for a period of 24 hr. compared to control group as shown in Table (8). But, there were significantly increased in the serum creatinine of the fish that exposed to 50, 100 and 200 µg / L of aluminum chloride for a period of 48 hr. compared to control group. Also, there were significantly increased in the serum creatinine of the fish that exposed to 50 and 100 µg / L AlCl<sub>3</sub> for a period of 96 hr. compared to control group. There were highly significant increased in the serum creatinine of the fish that exposed to 200 µg / L AlCl<sub>3</sub> for a period of 96 hr. compared to control group.

Table (8): Serum creatinine level mg % changes in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	Creatinine mg % after 24 hr.	Creatinine mg % after 48 hr	Creatinine mg % after 96 hr
Control	1.57±0.11	1.70±0.11	1.92±0.11
50 µg / L AlCl <sub>3</sub>	1.80±0.05	2.33±0.046*	2.80±0.057*
100 µg / L AlCl <sub>3</sub>	1.86±0.03	2.47±0.0818*	2.86±0.033*
200 µg / L AlCl <sub>3</sub>	1.87±0.03	2.59±0.097*	3.30±0.43**

significant at p<0.05, \*\* highly significant at p<0.01, \*\*\* very highly significant at p<0.001,

### Serum Glucose Level And Total Protein

There was no significant increased in the serum glucose level of the fish that exposed to 50 µg / L of aluminum chloride for a period of 24 hr. compared to control group. But, the serum glucose level was significantly increased in the fish which exposed to 100 µg / L and highly significant increased in the fish that exposed to 200 µg / L compared to control group for 24 hr. Table (9) showed that the serum glucose level in the fish that exposure to 50 and 100 µg / L AlCl<sub>3</sub> for a period of 48 hr. was highly significant increased and very highly significant increased in the fish that exposed to 200 µg / L AlCl<sub>3</sub> compared to control group. Also, the fish that exposed to 50, 100 and 200 µg / L AlCl<sub>3</sub> for a period of 96 hr. showed very highly significant increase in their serum glucose level compared to control group.

Table (9): Glucose mg / dl changes in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	Glucose mg / dl after 24 hr.	Glucose mg / dl after 48 hr	Glucose mg / dl after 96 hr
Control	6±1.15	68±1.15	70±1.15
50 µg / L AlCl <sub>3</sub>	73.33±1.66	108.66±4.66**	134±2.08***
100 µg / L AlCl <sub>3</sub>	77.33±1.45*	119.33±1.76***	142.3±1.45***
200 µg / L AlCl <sub>3</sub>	82.66±1.45**	123±1.52***	155±1.52***

significant at p<0.05, \*\* highly significant at p<0.01, \*\*\* very highly significant at p<0.001,

The serum total protein of the fish that exposed to 50 µg / L AlCl<sub>3</sub> for a period of 24 hr. was significantly increased compared to control group. Also, the serum glucose was highly significant increased in the fish that exposed to 100 and 200 µg / L AlCl<sub>3</sub> compared to control group for 24 hr. Also, there were highly significant increased in the serum total protein in the fish that exposed to 50, 100 and 200 µg / L AlCl<sub>3</sub> compared to control group for 48 hr. exposure. Table (10) showed that the serum total protein in the fish that exposed to 50, 100 and 200 µg / L AlCl<sub>3</sub> for a period of 96 hr. was very highly significant increased compared to control group.

Table (10): Serum total protein g % changes in *Tilapia zillii* exposed to different concentrations and different periods of aluminum chloride.

parameters /groups	Total protein g% after 24 hr.	Total protein g% after 48 hr	Total protein g% after 96 hr
Control	5.86±0.14	5.72±0.14	5.38±0.14
50 µg / L AlCl <sub>3</sub>	6.56±0.18*	8.5±0.17**	9.2±0.15***
100 µg / L AlCl <sub>3</sub>	7.34±0.086**	8.46±0.14**	9.16±0.088***
200 µg / L AlCl <sub>3</sub>	8.5±0.17**	8.76±0.088**	9.33±0.088***

significant at  $p < 0.05$ , \*\* highly significant at  $p < 0.01$ , \*\*\* very highly significant at  $p < 0.001$ .

## DISCUSSION

This study indicated that there were an increase in the number of red blood cells and white blood cells. Also, an increased in the hematocrit value and hemoglobin content under the effect of aluminum chloride at the different periods. These results are agreement with Witters *et al.* (1990) who indicated a doubling of hematocrit values under the effect of aluminum on rainbow trout. An increase in the number of red blood cells and hematocrit and hemoglobin contents could also have contributed the results that reported by Witter *et al.* (1987). The increase in the red blood cells, hematocrit and hemoglobin are agreement with (Miligan & Wood, 1982 ; Goss and Wood 1988) who indicated the blood parameters were increased in rainbow trout that exposed to aluminum toxicity. Haematocrit value and hemoglobin content levels were high enough to eliminate these parameters as a possible causes lactate accumulation. The marker red cell swelling suggests plasma acidosis in addition to ionic dilution These results of blood parameters are in agreement with Allin and Wilson (1999) who stated the effect of aluminum on the blood parameters of juvenile rainbow trout (*Oncorhynchus mykiss*) showed fewer in red blood cell and hematocrit than the control fish and this may be to haemodilution.

Total serum protein in *Tilapia zillii* was increased in this study and this may be due to stress of aluminum on the fish. This increase was agreement with the results of Gross and Wood (1988) who reported the total serum protein was increased in the fish that exposed to aluminum toxicity. The results were in agreement with those obtained by McKim *et al.* (1970) who investigated base oil and crude oil administration. It could be postulated that the increase in the serum total protein in this study may be due to increase in the blood cells. This study observed that the increase in the serum proteins are agreement with Abdelmegid *et al.* (2002) that reported the liver protein was increased in *Tilapia zillii* under the effect of aluminum.

This study revealed that the increase in the serum AST and ALT activities, creatinine, uric acid and serum glucose level due to stress in the fish by affecting of aluminum. This studies are agreement with the result of Goss and Wood (1988) who reported the validity of cortisol and glucose as indicators of the stress in the fish. The blood glucose levels have long been used as indicators

of stress in fish as recorded by (Hattingh, 1976; Donaldson, 1981; Wedemeyer & Mcleay 1981) who reported under condition of stress, hyperglycemia may provide additional energy during times of high metabolic need such as "fight or flight" response. The increase in the serum AST, ALT activities, creatinine, uric acid and serum glucose level are agreement with Julie *et al.* (2001) who indicated that the serum glucose level increase in Atlantic salmon (*Salmo salar*) after exposure to aluminum and Allin and Wilson (1999) who suggested elevated levels of serum glucose can act as a satiation signal and cause a reduction in appetite.

This study observed that the increased in the serum creatinine and glucose level are agreement with Abdelmegid *et al.* (2002) who reported that all mean difference total lipids, cholesterol, glucose and creatinine were significant increase and showed hyperglycemia and lipidimia and elevated levels of creatinine in serum as compared to the control. The depletion of liver glycogen (glycogenolysis) and the rise in blood glucose levels were reported in fish after exposure to pollutants were recorded by Hanke *et al.* (1983 and Gluth and hanke (1985) who stated Hyperglycemia stress response is mediated by increased in plasma cortisol and catecholamine and Gross and Wood (1988) who postulated that the significant rise in serum creatinine concentration in fish caught foreign heavily and moderately polluted areas in comparisons to controlled areas due to increased in the muscle tissue catabolism or the impairment of carbohydrates metabolism. These results were agreement with Murray *et al.* (1990) and Oddvar, *et al.* (2005) who reported that the accumulation of aluminum in the gill *Salmo trutta L.* made induced physiological stress responses which cuses increased blood glucose and decreased plasma chloride.

It can be concluded from this study that disturbance in the RBCs and WBCs, hematocrit value, hemoglobin content, serum total protein, serum glucose level, serum activities of (AST, ALT) and (creatinine and uric acid) concentrations as a result of stress of aluminum chloride on *Tilapia zillii* reflect the disturbance in all metabolic function and can be used as marker of pollution.

#### ACKNOWLEDGEMENT

The researcher thanks owner of Om Ehfein aquaculture farm for providing the tested fish, Department of Biology, Faculty of Science – Tubruk, Omar Elmokhtar University, Lybia and Central Lab of aquaculture research, abbasa for helping in performing this study.

#### REFERENCES

- Abdelmegid, N.; Kheirallah, A. M.; Abushabana, Adham, K. and Abdelmineim, A. ( 2002). Histochemical and biochemical changes in liver in *Tilapia zillii* G. as consequence of water pollution. J. Biol. Sci.; 2(4): 224 - 229

- Allin, C.J. and Wilson, R.W. (1999). Behavioral and metabolic effects of chronic exposure to sublethal aluminum in acidic soft water in juvenile rainbow trout (*Oncorhynchus mykiss*) Can. J. Fish. Aquat. Sci./J. Can. Sci. halieut. aquat, 256(4):670-678
- Allin, C.J. and Wilson, R.W. (2000). Effects of pre-acclimation to aluminum on the physiology and swimming behavior of juvenile rainbow trout (*Oncorhynchus mykiss*) during a pulsed exposure Aquat Toxicol. 51(2):213-24
- Cleveland, L.; Little E.E.; Hamilton. S. J.; Buckler, D.R.; and Huhn, J.B. (1986). Interactive toxicity of aluminum and acidity to early life stages of brook trout. Trans. Am Fish Soc., 115: 610-620.
- Cleveland, L.; Buckler, D.R.; and Brumbaugh, W.G.; (1991). Residue dynamics and effects of aluminum on growth and mortality in brook trout. Environ Toxicol Chem, 10: 243-248.
- Dacie, J. V. and Lewis S. M. (1975). Practical hematology. London, Churchill, Livingstone. 153 pp.
- Donaldson, E. M. (1981). The pituitary – internal axis as an indicator of stress in fish. In stress and fish ( A. D. Pickering, ed.,) pp. 11 – 47 London and new York, Academic press
- Gluth, G. and hanke, W. ( 1985). Comparison of physiological changes in carp, *Cyprinus carpio*, induced by several pollutants at sublethal concentrations. Ecotoxicol. Environ. Safety, 9: 179 - 188
- Gross, G. G. and Wood, C. M. (1988). the effect of acid and acid / aluminum exposure on circulating plasma cortisol levels and other blood parameters in the rainbow trout, *Salmo gairdneri*. J. fish Boil. 32:63 – 76
- Gundersen, D.T.; Bustaman, S.; Seim, W.K. and Curtis, L.R. (1994). pH, hardness, and humic acid influence aluminum toxicity to rainbow trout( *Oncorhynchus mykiss*) in weakly alkaline waters. Can. J. Fish Aquat. Sci., 51:1345-1355.
- Hanke, W.; Gluth, G.; Babel, H. and Muller, R. (1983). Physiological changes in carps induced by pollution. Ecotoxicol. Environ. Safety, 7: 229 - 241
- Hattingh, G. (1976). Blood sugar as an indicator of stress in the fresh water fish, *Labeo capensis* (Smith). J.fish. Oil., 10: 191 - 195
- Henry, R. J. (1974). colorimetric methods for determination of creatinine and uric acid. Clin. Chem. Principles and techniques, 2<sup>nd</sup> edition, Harper and Row, 525pp.

- Hytterod, S. and Poleo, A.B. (2003). The effect of aluminum in Atlantic salmon (*Salmo salar*) with special emphasis on alkaline water. 1: J Biochem. 97(1):89-96
- Julie, C.; Brodeur, Finnokland, Bengt, Finstad, D.; George, D. and ScottMcKinley, R. (2001). Effects of subchronic exposure to aluminum in acidic water on Bioenergetics of Atlantic salmon (*Salmo salar*). Ecotoxicology and environmental safety, 49: 226 - 234
- McKee, M.J.; Knowles, C.O. and Buckler, R. (1909). Effects of aduminum on the biochemical composition of Atlantic salmon ?? HYPERLINK javascript:AL\_gdt(this,%20'jour',%20'Arch%20Efvaron%20Contae%20Toxicol.');" Arch Environ Cgntam Toxicol., 18 (1-2):243-8
- McCim, J. G. M.; Chbistensin and Hunt, E. P. (1970). Changes in the blood brook trott, *Salvalines fon4inalis* following short - term and lonb - term expos5re 4c c-pper. J. Fish. Res. Bd. Can., 27: 188 - 1889.
- Miligan , C. L. and Wood, C. M ( 1982). disterbands if hematology, fluid volume dis'ribution and circulatory function associated with low envirognmeftal pH in the rainbow trout, *Salmo eairdneri*. J& exp. Boil., 99: 39 - 15
- Eurray, R. K.; Granne, D. K.; Eages, P. A. and Rodwell, V. W. (1990). Harper's biochemistry 23<sup>rd</sup> ed., Appdedon and Lang publishers, Norwack, Connecticut / loss altos, California.
- Narkar, R.; Sen NS, and Ahmad MF. (2006). Aluminum toxicity induced poikilocytosis in an air-breathing teleost, *Clarias batrachus* (Linn&). Indian J. Exp Biml. 44 (1):03-5.
- Oddvar, R.; Bjorn, O.; Torstein, K.; Frode, K.; Oyvind, A. and Eilit, S. (2005). Diffusivd gradient in thin filaments campler predicts stress in Bbown trout, *Salmg trutta* L. exposed to alumine in aci\$ fresh uaters. Dnviron. Sci. Techno., 39: 1167 - 1174
- Pekka J. Vuorinen Mara Kiinänen, Sdppo Peuranen and ChristinaTigerstedt ( "002). Reproduction, blood and plasma parameters and gill histology of vendace (*Coregonus albula* L.) in long-term exposure to acidity and aluminum. Ecotoxicology and Environmental Safety, 54(3): 255-276
- Peter, T. (1968). total protein albumin ang globulin. In standard -ethods of clin. Chem., 14:1147pp.
- Peurafen, S. Keinanen, M.; Tigerstedt, C. and Vuorinen PJ. ( 2007 ). Effects of t%perature on the recovery of juvenile grayling (*Thymadlus*) from exposure to Al+Fe. 1: Aquat Tmxicml. 61(1):73-84

- Reitman, S. and Frankel, S. (1957). Aspartate aminotransferase and alanine aminotransferase colorimetric methods, *Am. J. Clin. Pathol.*, 4(20): 56-65.
- Rosceland BN and Rkogheim OK (1984). A comparative study on salmonid fish species in acid aluminum-rich water\* II. Physiological stress and mortality of one- and two-year-old fish & Oslo, Norway, Institute of Fresh Water Ecology (pp 186-194 (Research Report no. 61)
- Rossedand, B.O. and Staurnes M (1990). Physiological mechanisms for toxic effects and resistance to acidic water: an ecophysiological and ecotoxicological approach. In: Steinberg CEW & Wright RF ed. Acidification of freshwater ecosystems: Implications for the future. New York, Chichester, Brisbane, Toronto, John Wiley and Sons, pp. 228-246.
- Soivio, A.; Westman, K. and Nyholm, K. (1972). Improved method of dorsal aorta catheterization: hematological effects followed for three weeks in rainbow trout (*Salmo gairdneri*). *Finn. Fish. Res.*, 1: 1 - 21.
- Trinder, P. (1961). Enzymatic colorimetric method for glucose determination. *Ann. Clin. Biochem.*, 6: 24-39.
- Wedemeyer G. A. and Mcleay, D. J. (1901). Methods for determination the tolerance of fish to environmental stressors. In stress and fish (A. D. Pickring, ed.), pp. 247 - 275. London: Academic press.
- Witters HE (1986). Acute acid exposure of rainbow trout, *Salmo gairdneri* Richardson: effects of aluminum and calcium on ion balance and hematology. *Aquat Toxicol.*, 8: 197-210.
- Witters, H.; Fav, G. J.; Van, P. S., and Vaf, D. O. (1987). Aon Regulatory and hematology responses of rainbow trout, *Salmo gairdneri* to bicarbonic acid and aluminum stress. *Annals de la Society Royal Zoologique de Belgique*, 117 (1): 411 - 420
- Witter, H. E.; Van, S. P., Van, G. J. D. and Van, O. L. J. (1990). The effect of humic substances on the toxicity of aluminum to adult rainbow trout, *Oncorhynchus mykiss*. *J. Fish Biology*, 37: 43 - 53.
- Vuorinen, PJ; Keinänen M, Peuranen S. and Tigerstedt C. (2003). Reproduction, blood and plasma parameters and gill histology of vendace (*Coregonus albula* L.) in long-term exposure to acidity and aluminum *Ecotoxicol Environ Saf.*, 54(3):255-276