The toxic effects of ammonia on growth rate and some blood parameters in common carp (Cyprinus carpio L.)

Qussai Salih Jumaa*, Khalid Ahmed Hadi Al-Saeedy** and Wasan Srahin Obid**

Department of Pathology- College of Veterinary Medicine/ University of Tikrit
**Department of Physiology, Pharmacology and Biochemistry-
College of Veterinary Medicine/ University of Tikrit

Abstract

This study was performed to know the effect of ammonia on the blood picture and the weight of common carp fish. 50 fish were used at the age of 6 months and were divided into two groups. Each group included 25 fish. The first group represented the treatment group with ammonia at a concentration of 1.0 mg/L, and the second group represented control group. It is grown in plastic basins with dimensions of 1.5 × 2 meters and continued treatment for four weeks. It was observed that the weight in the ammonium treatment group had the lowest significant value (p≤0.05) compared to the control group. It was also observed in this study that the ammonia group had the lowest significant values in (white blood cells, red blood cells, hemoglobin concentration and Packed cell volume) compared with control group. As for the biochemical parameters, it was observed that the Glucose concentration and each of the enzymes Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST) had the highest value in the group treated with ammonia compared with the control group while the total protein recorded the lowest significant value in the group treated with ammonia Compared to control group. We conclude from this study that increasing the concentration of ammonia in the aquatic environment has a negative impact on fish breeding and health.

Key words: common carp, ammonia poisoning

Email: kh55550000@gmail.com
Introduction

Ammonia is one of the most dangerous nitrogen residues, especially in the aquatic environment. Ammonia is produced by the destruction of amino acids, pyurins and primidines. Ammonia is found in the aquatic environment in two main forms: ammonia (NH4) and non-ionized ammonia (NH3). Ammonia affects the aquatic environment by causing an imbalance in the acidic function of the aquatic medium and causing it to be increased by increasing the concentration of hydrogen due to the increase in the level of non-heterogeneous ammonia. (1) Non-heterogeneous ammonia is more toxic in the toxicity of ammonia than the organisms present in the water medium. This is due to their ability to spread in the gills membranes of these organisms (2) The increase in ammonia level causes a decrease in the level of growth of living organisms, inhibition of immunity and high rate of damage in organisms that spread in the aquatic environment through their toxic effect To the blood and tissues in those organisms. In addition to the non-heterogeneous ammonia cause nerve poisoning, oxidative stress, excess movement of the organism and the convulsions (3) the toxic effect of ammonia depends on several factors including temperature, acid function and oxygen ratio. Among the most important environmental signs of this effect is the heat as it is one of the most important criteria that affect the toxicity of ammonia. The researchers noted (4) that the high temperature leads to increased toxicity of ammonia, which adversely affects the living organisms in the aquatic environment, Indicates that increased temperature causes an increase in propagation ability, bioavailability and chemical reactions occurring within the water medium (5). Exposure to any poisonous substance in the water medium can have adverse effects on growth and production and an increase in the rate of fish losses. Among these widespread toxicants is ammonia, both ionic and non-ionic, which is a major cause of the increase in the rate of damage. A key indicator of fish poisoning is the rate of growth and weight gain (6). Studies have shown that exposure to toxic substances leads to physiological changes in blood standards, since these parameters are very sensitive to any change in the aquatic environment in which organisms live. These changes can determine the effect of toxic substances in the water medium (7). In a study carried out by the researcher (8) he noted that the poisoning of ammonia causes a decrease in all blood standards has been attributed to the poisoning of these compounds lead to anemia and the analysis of red blood cells.

Materials and Methods

- **Experimental animals**: 50 Common carp (*Cyprinus carpio*) were used at 6 months old, weighing between 500-580 g, obtained from the breeding fields of Al-Soera city, raised in plastic basins with dimensions 1.5 × 2 m in chlorine-free water, and left For a week for the purpose of acclimatization and to ensure that they are free of diseases with the provision of oxygen by special pumps for this purpose and the pH was set at 7.5 and at a temperature of 18-20 ºC. The experiment lasted for two months since 2/3/2016 to 5/5/2016.

- **Collecting of blood samples and extract the serum**: The blood was collected from the tail vein of the fish by means of a 5 cm³ plastic syringe and divided into two parts. The first was to be kept in tubes contain the anticoagulant substance (EDTA) for the purpose of the blood tests; the second was to be kept in glass tubes free of anticoagulation, the tubes were Place in a slant form for 30 minutes at room temperature, then place in a centrifuge at 3000 rpm for 3 minutes, then the serum pulled, and placed in sterile plastic tubes, and keep in the freezer until tests time.
- **Biochemical tests:**
  - Estimation of the efficiency of the enzyme Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST): The efficacy of these enzymes was estimated using the kit prepared by (RANDOX) company.
  - Determination of total protein level in serum: Determination of the total protein level using a supplied kit according to Biuret method, which is dependent on method (9).
  - Determination of serum glucose level: The Measurement of serum glucose using Syrbo's kit (Syrian).

- **Statistical analysis:** The results were analyzed using the SPSS program for values representing the rate and the standard error; the data were analyzing using ANOVA Analysis of variance One Way. The differences between the groups were determined using the Duncan multiple range test, at a probability level (P≤0.05).

**Results**

The current results showed that mean body weight of ammonia treated group were decreased as compared with control group, also, the hematological parameters including RBCs, WBC, HB and PCV were decreased as compared with control group. The presented results of AST, ALT and serum glucose were also increased in ammonia treated group in comparison with control group, while the total protein appeared to be significantly decreased as compared with control group.

**Table (1) Effects of ammonia on growth rate and some blood parameters in common carp (Cyprinus carpio L.)**

| Groups                  | Control group M±SD | Ammonia treatment group M±SD |
|-------------------------|--------------------|------------------------------|
| Fish weight (gm)        | 637.65±18.21 a     | 487.98±20.67 b               |
| RBC (×10^6 cell/ml³)    | 2.8±0.23 a         | 0.13±1.9 b                   |
| WBC (×10^3 cell/ml³)    | 27.5±0.45 a        | 23.8±0.21 b                  |
| Hb (gm/ml)              | 11.3±0.12 a        | 9.7±0.11 b                   |
| PCV (%)                 | 32.4±1.45 a        | 28.6±1.83 b                  |
| AST activity            | 112.43±1.45 b      | 142.43±1.83 a                |
| ALT activity            | 12.32±0.21 b       | 16.76±0.65 a                 |
| Total protein (gm./100cm³) | 2.77±0.21 a     | 2.12±0.22 b                 |
| Serum glucose (gm./100ml) | 55.67±1.67 b      | 62.76±1.65 a                |

Different letters in the columns refers to significant differences at (p≤0.05).

**Discussion**

In this study, it was observed that ammonia poisoning led to a decrease in the weight of fish in the treated group compared to control group. The results obtained from this study agreed with researcher (10) in his study that poisoning leads to a decrease in fish growth due to disorders in the body's vital organs, including hormones and enzymes caused by poisoning. The researcher (11) has proved in his study that ammonia poisoning leads to inhibition of growth rates of organisms in the aquatic environment and this is what we observed in our current study. The results obtained in the researcher (12), who explained the growth disorders in his experimental study, concluded that the large amount of ATP that the body is supposed to use for growth is used to get rid of toxic substances in the body. While the researcher (13) illustrate the decline in the rate of growth caused the rise in temperature in the aquatic environment in which the organism lives, as poisoning leads to an increase in the temperature of that environment. The blood parameters (white blood cells, red blood cells, hemoglobin concentration and packed cell volume) were the lowest in the ammonia treatment group, compared to the control group. The results were consistent with the researcher's (14) Which attributed the decrease in the blood film to poisoning in the aquatic environment, as poisoning
changes the nature of the biochemical blood, which drives the fish to take a larger amount of oxygen, which leads to raise the level of methemoglobin resulting from the collapse of gills because of these toxic substances. The researcher observed a significant reduction in the blood film of the fish exposed to ammonia experimentally. This is explained by the fact that the reduction of red blood cells and the rate of hemoglobin to the lack or absence of hormone stimulating the formation of red blood cell (the erythropoietin), while the reduction of WBC is due to the oxidative stress caused by these toxic substances on the organism and also indicated that the low level of packed cell volume (PCV) resulting from the decrease in the number of red blood cells. In terms of biochemical parameters (total protein, glucose, AST and ALT enzymes) in this study, the total protein was found to be decrease significantly in the ammonia treatment group. The results were consistent with the finding of the researcher (15), who explained the decrease in the level of proteins to the oxidative stress associated with the poisoning which affects the level of Total protein in the body. In the same study it was found that ammonia poisoning caused an increase in the level of glucose and this we found in our current study. The researcher explained that the high level of glucose is due to the glycogenolytic degradation of catecholamines in response to oxidative stress due to poisoning. In addition, AST and ALT were among the most important indicators of damage in the kidney and liver recorded the highest significant value in the group exposed to toxicity compared with the control group. The results obtained were agree with the fined of (16) who explained the damage caused to these tissues due to the high ambient temperature and the spread of toxic substances in the water medium.

References
1. Randall, D. J. & Tsui, T. K. (2002). Ammonia toxicity in fish. Mar. Pollut. Bull., 45(1-12): 17-23.
2. Sinha, A. K.; Liew, H. J.; Diricx, M.; Blust, R. & De Book, G. (2012). The interactive effects of ammonia exposure, nutritional status and exercise on metabolic and physiological responses in gold fish (Carassius auratus L.). Aquat Toxicol., 109: 33-46.
3. Li, M.; Yu, N.; Qin, J. G.; Li, E.; Du, Z. & Chen, L. (2014). Effects of ammonia stress, dietary linseed oil and Edwardsiella ictaluri challenge on juvenile darkbarbel catfish Pelteobagrus vachelli. Fish Shellfish Immunol., 38(1): 158-165.
4. Lemarie, G.; Dosdat, A.; Coves, D.; Dutto, G.; Gasset, E. & Person-Le Ruyet, J. (2004). Effect of chronic ammonia exposure on growth of European seabass (Dicentrarchus labrax) juveniles. Aquaculture, 229(1-4): 479-491.
5. Barbieri, E. & Bondioli, A. C. V. (2015). Acute toxicity of ammonia in Pacu fish (Piaractus mesopotamicus, Holmberg, 1887) at different temperatures levels. Aquacult. Res., 46 (3): 565–571.
6. Patra, R. W.; Chapman, J. C.; Lim, R. P.; Gehrke, P. C. & Sunderam, R. M. (2015). Interactions between water temperature and contaminant toxicity to freshwater fish. Environ. Toxicol. Chem., 34(8):1809-1017.
7. Kim, J. H. & Kang, J. C. (2015). The lead accumulation and hematological findings in juvenile rock fish Sebastes schlegelii exposed to the dietary lead (II) concentrations. Ecotoxicol. Environ. Saf., 115: 33-39.
8. El-Shafai, S. A.; El-Gohary, F. A.; Nasr, F. A.; der Steen, N. P. V. & Gijzen, H. J. (2004). Chronic ammonia toxicity to duckweed-fed tilapia (Oreochromis niloticus). Aquaculture, 232(1-4): 117-127.
9. Kim, J. H. & Kang, J. C. (2014). The selenium accumulation and its effect on growth, and haematological parameters in red sea bream, Pagrus major, exposed to waterborne selenium. Ecotoxicol. Environ. Saf., 104:96-102.
10. Ajani, F.; Olukunle, O. A. & Agbede, S. A. (2007). Hormonal and Haematological Responses of Clarias gariepinus (Burchell 1822) to Nitrite Toxicity. J. Fisheries Int., 2(1): 48-53.
11. Tietz, N. W. (1995). Text book of clinical chemistry. 3rd Ed. C. A. Curtis, E. R. Silverman L. M. Christensen R. H. PP. 523- 524.
12. Erickson, R. J.; Mount, D. R.; Highland, T. L.; Hockett, J. R.; Leonard, E. N.; Mattson, V. R.; Dawson, T. D. & Lott, K. G. (2010). Effects of copper, cadmium, lead, and arsenic in a live diet on juvenile fish growth. Can. J. Fish. Aquat. Sci., 67(11): 1816-1826.
13. Clearwater, S. J.; Farag, A. M. & Meyer, J. S. (2002). Bioavailability and toxicity of dietborne copper and zinc to fish. Comp. Biochem. Physiol. C Toxicol. Pharmacol., 132(3):269-313.
14. Vosyliene, M. Z. & Kazlauskiene, N. (2004). Comparative studies of sublethal effects of ammonia on rainbow trout (Oncorhynchus Mykiss) at different stages of its development. Acta Zoologica Lituanica, 14(1):13-18.
15. Carvalho, C. S. & Fernandes, M. N. (2006). Effect of temperature on copper toxicity and hematological responses in the neotropical fish Prochilodus scrofa at low and high pH. Aquaculture, 251(1): 109-117.
16. Thangam, Y.; Perumayee, M.; Jayaprakash, S.; Umavathi, S. & Basheer, S. K. (2014). Studies of ammonia toxicity on haematological parameters to freshwater fish cyprinus carpio (Common carp). Int. J. Curr. Microbiol. App. Sci., 3(12): 535-542.