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Oxidative Stress and Blood Parameters Affected by Carbaryl Insecticide in *Cyprinus carpio*

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ABSTRACT

This study was conducted to investigate the effects of the carbaryl insecticide on blood parameters and oxidative stress in *Cyprinus carpio*. A total of 60 fish were randomly assigned to three groups: the first group served as the control; the second group was placed in tanks containing water with a carbaryl concentration of 5mg per liter; and the third group was raised in tanks with a carbaryl concentration of 7.5 mg per liter. The fish were monitored for 14 days. The results showed a decrease in RBCs, hemoglobin, and PCV, along with an increase in WBC count in the experimental groups compared to the control group. Additionally, the study found a significant decrease in GSH and a significant increase in MDA in all experimental groups compared to the control group. The findings suggest that carbaryl has harmful effects on fish, as evidenced by changes in blood parameters and antioxidant indicators.

INTRODUCTION

One of the most significant challenges facing living organisms is the widespread use of pesticides to control pests that affect plants and animals. While pesticides offer benefits, they also pose significant risks to human and animal health, as well as the environment **(Bloom & Brandt, 2018)**. The carbamate group of insecticides includes the chemical compound 1-naphthyl-N-methylcarbamate, which appears as a colorless to pale brown crystalline powder. Common carbamate insecticides on the market include Sevin, Adios, Carbamic, Dinabon, Hexaphene, and Panam **(Bresciani *et al.*, 2020)**. Carbamate insecticides are divided into two types: the first type, monomethyl carbamates, includes pesticides such as Carbaryl, Temik, Ficam, and Zectran, while the second type, dimethyl carbamates, includes Dimethilan, Pirimicarb, and Isolan **(Rajagopalan *et al.*, 2023)**. The primary action of pesticides in this group is to inhibit the enzyme choline acetyltransferase in the nervous system. These pesticides form a complex with the enzyme, but the enzyme-carbamate complex is usually unstable. As a result, the enzyme regains its activity more quickly than with organophosphates (OP), making carbamate pesticides less dangerous than OP pesticides in human exposure **(Hanchate *et al.*, 2023)**.

Carbaryl, or Sevin, is widely used globally and in Iraq, particularly in large quantities. It is effective on contact and targets houseflies, mites, and aphids. Carbaryl is available in two formulations: a dusting powder with a 10% concentration and a wettable powder with an 85% concentration **(Mahmood *et al.*, 2023)**. These pesticides can reach fish tanks either through the air or through polluted water. While several studies have investigated the effects of pesticides on laboratory animals, our current study aimed to assess the impact of carbaryl insecticide on the health of common carp by examining blood parameters and oxidative stress profiles **(Rajagopalan *et al.*, 2023; Nasrabadi *et al.*, 2024)**.

MATERIALS AND METHODS

Clinical samples collection

The current study was conducted on private fish tanks in Al-Saouira City from July to December 2023.

Fish

A total of 60 *Cyprinus carpio* fish, with an average weight of 200-300 g, were used in the study.

Study groups

The fish were randomly divided into three groups (20 fish per group), with each group placed in separate tanks as follows:

- The first group served as the control group.
- The second group was raised in tanks containing water with a carbaryl insecticide concentration of 5mg per liter.
- The third group was raised in tanks containing water with a carbaryl insecticide concentration of 7.5mg per liter.

The fish were monitored for 14 days. Following two weeks of exposure to carbaryl insecticide, the fish were euthanized, and blood samples were collected **(Al-Fluj *et al.*, 2016; Kirschbaum & Formicki, 2020)**.

Blood collection

Blood was collected using a sterile syringe from the venous ventricle and were placed in EDTA tubes for blood analysis.

Blood picture

Blood parameters were assessed according to **Bloom and Brandt (2018)**.

Liver samples

Liver samples were collected to measure oxidative stress indicators.

Oxidative stress indicators

a) Glutathione (GSH) in the liver was measured according to **Jumma *et al*. (2022)** and **Al-Niemi *et al.* (2023)**.
b) Malondialdehyde (MDA) in the liver was measured according to **Rohani *et al.* (2022)**.

Statistical analysis

SPSS software was used to analyze the results. Values representing the mean and standard error were calculated. Data were analyzed using one-way ANOVA, and differences between groups were determined using Duncan's multiple-range test at a probability level of $P \leq 0.05$.

RESULTS AND DISCUSSION

Table (1) shows a decrease in RBC count, hemoglobin levels, and PCV, along with an increase in WBC count in the experimental groups compared to the control group. The decrease in RBC count, hemoglobin, and PCV suggests the toxic effects of carbaryl on fish red blood cells, a finding also reported by **Abbas *et al.* (2021)**. The increase in WBC count may be attributed to carbaryl's impact on the differentiation of lymphocytes **(Reddy *et al.*, 2021)**. On the other hand, the observed decrease in RBC count due to pesticide exposure indicates an anemic condition. Anemia may result from the death of red blood cells caused by reactive oxygen species (ROS), which are generated in substantial amounts due to the harmful effects of pesticides **(Mostakim *et al.*, 2018)**.

Erythropoietin, a crucial factor for stimulating red blood cell production, helps maintain normal circulating RBC levels. A low RBC count is associated with low erythropoietin levels, which can occur as a result of herbicide toxicity, such as that caused by Diuron **(Al-Fluj *et al*., 2016)**. The reduction in hemoglobin content in fish may result from either its degradation or decreased production **(Najem *et al.*, 2020; Al-Juhaish, 2025)**, impairing its ability to carry oxygen. Another harmful effect of pesticides is the disruption of the enzymatic system responsible for converting methemoglobin into hemoglobin, which can lead to methaemoglobin accumulation and hypoxia **(Mostakim *et al.*, 2015)**.

Blood tests can provide precise data on the impact of pesticides on environmental exposure, and in many cases, hematological parameters serve as significant early indicators of altered physiological states in fish resulting from stress, pollution, pesticide exposure, or infection. Key hematological measures in fish include total erythrocyte count (TEC), hemoglobin concentration, and packed cell volume (PCV) **(Jumma, 2024)**. Additionally, the total leukocyte count (TLC) is an important assay. An elevation in TLC often results from pesticide exposure. Leukocytosis, characterized by an elevated white blood cell count, may indicate a protective and compensatory response to the detrimental effects of pesticides on fish **(Alrudainy & Jumaa, 2016; Yousef *et al.*, 2023)**.

Table 1. Blood parameters (RBC, WBC count, Hb content and PCV value) in *Cyprinus carpio* exposed to different concentrations of carbaryl insecticide for 14 days

Parameter	Experimental groups		
	Control group	1^{st} group (5mg/L)	2^{nd} (7.5mg /L)
RBCs ($x10^6$ cell/mm^3)	1.44±0.01 A	1.29±0.06 a	1.18±0.02 b
WBCs ($x10^3$cell/mm^3)	9680± 823 C	11380± 612 b	12130± 857 b
Hemoglobin (g/100ml)	14.3± 1.01 A	12.1± 0.9 b	11.4± 1.1 b
PCV %	37.9±1.4 A	33.5± 1.1 b	31.1± 1.2 c

The various letters in the columns show significant differences at ($P \leq 0.05$).

Table 2. Results of GSH and MDA in *Cyprinus carpio* exposed to different concentrations of carbaryl insecticide for 14 days

Parameter	Experimental groups		
	Control group	1^{st}group (5mg/L)	2^{nd} (7.5mg /L)
GSH μ mol./ g	1.45± 0.009 A	0.89±0.01 b	0.63±0.012 c
MDA μ mol./ g	203.7±29.6 C	267.5± 31.7 b	289.1±31.9 a

The various letters in the columns show significant differences at ($P \leq 0.05$).

The results presented in Table (2) show a decrease in glutathione (GSH) levels in the treated groups, along with an increase in malondialdehyde (MDA) levels. This outcome may be due to functional abnormalities in cellular mitochondria induced by carbaryl, which also causes structural and functional alterations, leading to oxidative stress and nutritional deprivation in fish. These factors contribute to an elevation in lipid peroxidation products **(Nasrabadi *et al.*, 2023)**. Additionally, excessive lipid peroxidation may be linked to the overproduction of urine 8-iso-prostaglandin F2α (8-iso-PGF2α), which is known to significantly elevate lipid peroxidation levels **(Sun *et al.*, 2019)**.

This effect may result from the stimulation of fatty acyl-CoA oxidase and the initiation of fatty acid oxidation, leading to elevated hydrogen peroxide (H_2O_2) production. The increased H_2O_2 promotes lipid peroxidation by altering cellular membrane permeability, ultimately causing cellular damage **(Vaiyanan *et al.*, 2018)**. According to several studies, carbaryl induces a dose-dependent increase in cell death and oxidative stress by enhancing reactive oxygen species (ROS) formation, decreasing GSH levels, elevating MDA concentrations, and reducing antioxidant levels in fish exposed to pesticides **(Singh *et al.*, 2018; Yousef *et al.*, 2023)**.

CONCLUSION

A study on carbaryl, a common pesticide, in fish suggests that stress or toxicity can impact blood parameters and antioxidant levels, thereby affecting fish health and the aquatic environment.

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