Egypt. J. Vet. Sci. **Vol. 53,** No. 4, pp. 573-581 (2022)

Egyptian Journal of Veterinary Sciences
https://ejvs.journals.ekb.eg/

Effect of Garlic on Blood Parameters in Thermal Stressed Common Carp Fish (*Cyprinus Carpio* L)

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ONE of the most important environmental factors that affects fish is the impact of excess heat. The consumption of herbal immuno-stimulants in aquaculture to improve immune system activity and fish tolerance to infections has become widespread. This study was conducted to determine the protective effect of garlic on fish exposed to heat stress. A total of 60 common carp fish (*Cyprinus Carpio*) with a mean weight of 100±10g were used. Fish were randomly divided into six groups of 10 fish in each and placed in six glass aquariums (40×40×80) cm, (G1) was exposed to heat stress at 30°C for a week. (G2) was exposed in normal water temperature 25°C for a week. (G3) was exposed to heat stress and treated with garlic by stomach tube in a dose of 1.63g/ 100g of body weight at the same time. (G4) was exposed to heat stress in the first week and then treated with garlic in the second week. (G5) was kept at 25 °C and treated with garlic. (G6) was treated with garlic in first week then exposed to heat stress in the second week. Results have shown that there were a significant increase in HB concentration, and PCV, heterophil, glucose level and total protein $P<0.05$ in groups exposed to heat stress. Although the results observed that the stress index had a significant decrease in the G1. This study achieved that garlic has a role to reduce the effect of heat stress on blood parameters. The best results were observed in the third and sixth groups.

Keywords: Garlic, Heat Stress, Blood Parameters, *Cyprinus Carpio.*

Introduction

Fish are ectothermic or poikilothermic aquatic animals, their body temperature is equivalent to their water temperature, with profound effects on their physiology, metabolism and behavior [1], and can change the natural animal homeostasis [2].

Fish are stressed under natural and environmental conditions, as any change in environmental conditions can be considered a source of stress that triggers a variety of responses, which can be measured as changes in hormone concentrations or basic substance concentrations in the plasma, changes in the size and number of blood cells, or functional changes in vital organs such as gills, kidneys and intestines [3]

The influence of extra heat and exposure of the surface layers of fresh water to it is one of the most important environmental elements that affect fish. In summer water bodies (whether natural or as a result of human activity) may be exposed to an increase in water temperature of 4 degrees, and at times as high as 10 degrees, producing heat stress in fish [4]. As the quantity of water oxygen is inversely related to the water temperature, this will result in a drop in its concentration [5], although disturbance in metabolic processes

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(*Received* 11/07/2022; *accepted* 15/08/2022)
DOI. 10.21608/EJVS.2022.149872.1365

resulted from that increase in temperature [6]. Stress in fish may be caused by various factors, e.g. change of water temperature [7]. Generally, heat stress might influence fish directly as fish died or indirectly by inhibiting immunity and hence allowing pathogens invasion and disease occurrence [8].

Hematological and serum components analysis provide information that is useful in the process of diagnosing, and treatment of metabolic disorders and diseases in fish [9].When the ambient temperature rises, the water oxygen content drops and the metabolic rate rises, resulting in an increase in tissue oxygen consumption. Fish adjust to changes in environmental temperature and hypoxia by elevating their total hemoglobin concentration, as a result of the increased metabolic rate [10]. Furthermore, hematological measures such as hematocrit and hemoglobin can be used to monitor fish health management in a variety of situations, including stress exposure [11].

In current years, due to increased pathogens resistance to synthetic medications, worldwide use of medical plants has increased significantly [12]. Garlic (*Allium sativum*) which belongs to the Alliaceae family is one of the most significant native plants and has important therapeutics use [13]. Garlic is high in minerals including iron, sodium, potassium, iodine and phosphorus, as well as vitamins like (A and C) which have antioxidant properties [14].

Garlic has antimicrobial, antifungal, antistress, and anti carcinogenic effects, and is also used to improve nutritional indices, immunological and growth stimulants, antioxidant, and blood pressure balance [15]. Garlic utilization has been proven in certain studies to increase cytokine synthesis, macrophage and lymphocyte activity, and hence improves and stimulates the immune system [16].

There are previous studies on the effect of the heat stress on the different parameters at carp fish and other fish species [1,5,17], and others deal with effect of garlic on blood parameter and different parameter at carp fish and other fish species [14-16]. But this study have been done to determine the effect of garlic on blood parameter of carp fish exposed to thermal stress through measurement of hemoglobin HB, differential cell count DLC, packed cell volume PCV, glucose, and serum total protein.

Material and Methods

Fish

A total of 60 fish of common carp *Cyprinus Carpio* with mean weight 100±10g were bridged from local privet pond in Duhok city and kept for acclimation in glass aquarium with de chlorinated tap water and provided with air pump heater and thermostat for one week in the laboratory of veterinary medicine of Duhok university before experiment time, the water changed daily, and the water temperature was maintained at 25±2 °C, with pH 7.8.

Garlic

Dose of garlic: 5 g garlic extract / kg basal diet according to Karimi Pashaki et al. [16], which equal to 1.63g/ 100g of body weight [18].

Experimental Design

The fish is randomly divided to six groups 10 fish in each and treated as the follow:

- The first group (G1) was exposed to heat stress 30°C for seven days (control group for the heat stress)
- The second group (G2) was exposed to normal water temperature 25°C.
- The third group (G3) was exposed to heat stress 30°C and treated with garlic (by stomach tube in a dose of 1.63g/ 100g of body weight) at the same time for seven days
- The fourth group (G4) was exposed to heat stress 30°C for one week then treated with garlic by stomach tube in a dose of 1.63g/ 100g of body weight for the next week,
- The fifth group (G5) was exposed to normal water temperature and treated with garlic in a dose of 1.63g/ 100g of body weight by stomach tube for one week.
- The six group (G6) was treated with garlic in a dose of 1.63g/ 100g of body weight by stomach tube for one week and then exposed to heat stress 30°C for the next week.

Blood Samples Collection

Blood samples were quickly collected from the caudal vein of fish, one part of blood samples was saved in tubes contain anticoagulant Heparin for hematological parameters and the second part of blood was saved in non-anticoagulant tubes then centrifuged at a speed of 3000 rpm for 15 minutes to separate serum; then serum was stored

at -18 ^{0}C for total protein, albumin, globulin, and glucose concentration.

Blood Component Analysis
Hemoglobin Concentration

Drabkin hemoglobin- kit was used, According to the equation:

Hemoglobin in (g/dl) = A sample * 36.77[19].

Packed Cells Volume (PCV) according to Microhematocrit Method [20].

Differential Leukocyte Count (DLC)

Blood smear fixed with absolute methanol and then stained with Giemsa stain. The differential leukocyte count is estimated from examination of the stained blood smears in which numbers of different types of leukocytes are calculated per 100 cells in numerous fields of a smear [21].

Stress Index According To the Equation

Stress index = Heterophil / Lymphocyte * 100 [22].

Glucose Concentration

Glucose assay kit by Biolabo manufacturer; France**.**

According to equation: $\frac{Abs(Assay)}{Abs\ (standard)}$ * standard concentration [23].

Serum Total Protein

Total protein assay kit by Biolabo manufacturer; France.

According to equation: $\frac{Abs(Assay)}{Abs\ (standard)}$ *Standard concentration. [24].

Serum Albumin

Serum albumin assay kit by Biolabo manufacturer; France.

According to equation: $\frac{Abs(Assay)}{Abs\ (standard)}$ * standard concentration. [25].

Serum Total Globulin

It was calculated by subtract serum total albumin from total serum protein according to Coles [26].

Statistical Analysis

The use of Analysis of variance (ANOVA), applied with (Minitab program) [27].

Results

Hemoglobin Concentration

There was an increase in Hb concentration and significant difference between the first group (G1) in which fish exposed to heat stress 30 ºC for 7 days, and all the others groups involving the third group G3 which exposed to heat stress for 7 days and treated with garlic at the same time, and the fourth group G4 which treated with garlic for one week then exposed to heat stress second week and G6 which exposed to heat stress for 7 days and then treated with garlic in the second week.

But there was no significant difference in Hb concentration between the G2 which exposed to normal temperature and G4 and G5 which were exposed to normal temperature and then treated with garlic.

Additionally there was a significant difference between the G2 and the G3 and G6 which there was no significant difference between them but there was increase in Hb concentration as shown in Table (1).

Packed Cell Volume (PCV)

Table 1 show that there was an increase in PCV and significant difference between the First group (G1), in which fish exposed to heat stress 30 ºC, and all the others groups including group (G3) which exposed to heat stress and treated with garlic at the same time, G4, which treated with garlic then exposed to heat stress, and G6 which exposed to heat stress and then treated with garlic.

But there was no significant difference in PCV between G2 which exposed to normal temperature and G5 which exposed to normal temperature and then treated with garlic.

Although there was no significant difference in PCV between G3, which exposed to heat stress and treated with garlic at the same time, and G6, which exposed to heat stress and then treated with garlic.

Stress Index

The results of stress index in Table (1) show that there was a decrease in Stress Index and significant difference between G1, in which fish exposed to heat stress 30 ºC, and all the others groups including G3, which exposed to heat stress and treated with garlic at the same time, G4, which treated with garlic then exposed to heat stress and G6 which exposed to heat stress and then treated with garlic.

But there was no significant difference in Stress Index between the G2 which exposed to normal temperature and group G3 which exposed to heat stress and treated with garlic at the same time, and the G4 which treated with garlic then

exposed to heat stress, and G6, which exposed to heat stress and then treated with garlic.

Also there was a noticeable variation between G5 which exposed to normal temperature and then treated with garlic and all the others groups.

Differential Leukocyte Count

As shown in Table 2 there was an increase in Heterophil number and a significant difference of (G1), in which fish exposed to heat stress 30 ºC, and G4 which treated with garlic then exposed to heat stress compared with other groups. While the differences among other groups were not significant.

Glucose Level

There was an increase in the glucose level and a significant difference between G1, in which fish exposed to heat stress 30 ^{0}C, and all the others groups including G3 which exposed to heat stress and treated with garlic at the same time. G4, which treated with garlic then exposed to heat stress and G6 which exposed to heat stress and then treated with garlic.

But there was no remarkable difference in glucose level between G2, which exposed to normal temperature the G5 which exposed to normal temperature and then treated with garlic. Besides, there was no indicative difference between G3, G4 and G6 as shown in Table (3)

Total Protein Level

There was an increase in total protein level and a significant difference between G1, in which fish exposed to heat stress 30 ^{0}C, and all the others groups including G3 which exposed to heat stress

TABLE 1. Effect of heat stress and garlic on (hemoglobin, PCV, stress index) of common carp fish (Cyprinus carpio).

Group	Hemoglobin (g/dl)	PCV (%)	Stress index
G1	20.20±1.77^{a}	33.00±0.81^{a}	0.030±0.006^{b}
G2	9.65±0.62^{d}	26.40±1.43^{c}	0.083±0.01^{ab}
G3	12.63±3.11^{bc}	28.60±2.71^{bc}	0.093±0.02^{ab}
G4	11.03±2.68^{cd}	31.00±1.58^{ab}	0.0129±0.01^{ab}
G5	9.66±0.83^{d}	27.40±3.06^{c}	0.147±0.22^{a}
G6	14.06±3.24^{b}	28.90±4.15^{bc}	0.083±0.01^{ab}

Means with the same letter are not significantly different (Fisher LSD, P < 0.05). ± denotes standard deviation of the mean total of each variable at each site (n=10).

TABLE 2. Effect of heat stress and garlic on differential leukocyte (%) count of common carp fish (*Cyprinus carpio*)

Group	Heterophils	Lymphocytes	Monocytes	Eosinophils	Basophils
G1	11.80±1.3^{a}	85.60±1.5^{bc}	0.00±0.0^{b}	0.80±0.4^{c}	1.80±0.4^{a}
G2	7.00±1.7^{b}	89.20±3.4^{a}	0.50±0.5^{a}	1.40±0.6^{a}	1.80±1.0^{a}
G3	8.20±2.0^{b}	88.50±2.9^{ab}	0.30±0.4^{ab}	1.00±0.0^{bc}	2.1±0.8^{a}
G4	11.00±1.0^{a}	85.00±2.1^{c}	0.20±0.4^{ab}	1.40±0.5^{ab}	2.4±1.3^{a}
G5	6.90±0.9^{b}	89.50±1.6^{a}	0.20±0.4^{ab}	1.00±0.0^{bc}	2.0±0.8^{a}
G6	7.40±1.7^{b}	88.70±3.4^{a}	0.20±0.4^{ab}	1.00±0.0^{bc}	2.7±2.4^{a}

Means with the same letter are not significantly different (Fisher LSD, P < 0.05). ± denotes standard deviation of the mean total of each variable at each site (n=10).

and treated with garlic at the same time, G4, which treated with garlic then exposed to heat stress, and G6 which exposed to heat stress and then treated with garlic. But there was no significant difference in total protein level between other groups as shown in Table (3).

Serum Albumin Level

As shown in Table 3 a there was an increase in serum albumin level a and significant difference between G1, in which fish exposed to heat stress 30 ^{0}C, and all the others groups including G3, which exposed to heat stress and treated with garlic at the same time, G4, which treated with garlic then exposed to heat stress, and G6 which exposed to heat stress and then treated with garlic. But there was no significant difference in serum albumin level between G2, which exposed to normal temperature, G4 and G5 which exposed to normal temperature and then treated with garlic.

Serum Globulin Level

The result of serum globulin shows that there was an increase in globulin level and a significant difference between G1, in which fish exposed to heat stress 30 ^{0}C, and all the others groups including G3, which exposed to heat stress and treated with garlic at the same time, G4 which treated with garlic then exposed to heat stress, and G6, which exposed to heat stress, and then treated with garlic. But there was no important difference in globulin level between other groups as shown in Table (3) .

Discussion

Hematological alterations are often used to evaluate the influence of environmental, dietary, or pathological stress on the body's state and to determine the body's health [28]. It was noted from the results of this study that exposure of fish to heat stress led to hematological changes represented by an increase in the hemoglobin concentration of groups exposed to heat stress compared with the control group, this could be as a result to increase of oxygen demand [29-30]. These results are in agreement with Bozorgnia et al. [31] that mentioned an increase in temperatures of the aquatic environments impacts and increase in the number of red blood cells, resulting in an increase in hemoglobin level in fish. Similar result was recorded by Docan et al. [32] who studied the effect of thermal stress on the blood parameters of trout *(Oncorhynchus mykiss)* that mentioned an increase in hemoglobin concentration and the packed cell volume PCV related with an increase in the temperature of the aquatic environment, although the increase in the PCV is associated with blood viscosity being considered adaptation mechanism, because of the extra cardiac attempt required in order to pump more viscous blood.

The increase of red blood cells count by the treatment with garlic is related with the effect of garlic on rise of glutathione intensity in red blood cell [33]. On the other hand, the garlic extract treatment improves the erythropoiesis as shown by the significant rise in the RBCs and Hb concentration, this enhancement in erythropoiesis may be associated to the improvement of antioxidant activity of this extract on RBCs (34). This findings agrees with results of many authors [14, 35] they established that administration of garlic caused significant rises in all blood

TABLE 3. Effect of heat stress and garlic on (glucose, total protein and serum albumin) of common carp (*Cyprinus carpio*)

Groups	Glucose (G\dl)	Total protein (G\dl)	Serum albumin (G/dl)	Serum globulin (G/dl)
G1	108.7±20.0^a	11.38±1.25^a	3.66±0.58^a	7.72±0.67^a
G2	52.84±9.98^c	2.06±0.45^b	0.82±0.12^c	1.24±0.33^b
G3	85.88±15.13^b	3.00±0.42^b	2.06±0.61^b	0.94±0.19^b
G4	91.31±13.55^b	3.16±0.55^b	0.73±0.31^c	2.43±0.24^b
G5	56.08±15.50^c	2.71±0.42^b	0.79±0.12^c	1.92±0.3^b
G6	92.49±14.83^{ab}	2.98±0.45^b	0.78±0.18^c	2.2±0.27^b

Means with the same letter are not significantly different (Fisher LSD, $P < 0.05$). ± denotes standard deviation of the mean total of each variable at each site(n=10).

parameters (red blood cell count, hemoglobin concentration and hematocrit level) in treated fish.

The stress index is ratio of heterophils to lymphocytes [22] as demonstrated in the current study showed that there was a decrease in stress Index of first group exposed to heat stress. But in groups that treated with garlic the stress index is raised which could because of the adding garlic to fish diets increased the red blood cell number, haemoglobin concentrations, hematocrit level, white blood cell, and thrombocytes [36], Also an increase in Immunological parameter is caused by the stress which lead to increase in the number of hetrophils than the normal range (the normal range of heterophils is 6-7% of total leucocyte number), in spite of their low number in the blood, but it rises when fish are exposed to Stressed conditions or when exposed to pathological condition, as for lymphocytes, they comprise a higher level of white blood cells (85-90%) [30].The findings of this study agrees with the result of [37-38] who reported that leucocyte counts improved significantly in juvenile hybrid tilapia fish fed 1% and 0.5% garlic supplemented food.

Significant hyperproteinemia was observed in the group exposed to heat stress as the heat stress affected physiology, behavior and metabolism of fish [1].High serum protein levels have been reported to be revealing of osmoregulatory dysfunctions, heamodilution, or damaging of tissue enclosing blood vessels[39], which agrees with the results of some investigators[35].

In fish fed diets comprising various sources of *Allium sativum*, glucose levels in blood serum decreased considerably. This circumstance was attached to enhancing of the antioxidant system in cells of pancreas to generate insulin. These results agree with of Metwally [40] who founded that feeding of *Tilapia nilotica* with garlic produced significant reduction of serum glucose concentrations.

Conclusion

This study concluded that garlic has a role to reduce the effect of heat stress on hemoglobin concentration, packed cell volume, Glucose level, stress Index. The best results were observed when the garlic was used with heat stress at the same time, and the usage of the garlic before exposure to heat stress.

Acknowledgment

We would like to thanks to all those who helped us to finish this study, especially College of Veterinary Medicine, University of Duhok.

Conflict of interest

The authors declare no conflict of interest.

Funding statement

No funding

References

1. Sébastien, A., Manuel, G. and Bastienm, B. Temperature increase and its effects on fish stress physiology in the context of global warming. *Journal of Fish Biology*, **98**(6), 1496-1508 (2021). https://doi.org/10.1111/jfb.14599.

2. Mariana, S. Alfons and Badr, G. Impact of heat stress on the immune response of fishes, *Journal of Survey in Fisheries Sciences*. **5**(2),149-159(2019). https://doi:10.18331/sfs2019.5.2.14.

3. Ahmed, S.M. and Jaffar, R.S. Effect of salt stress on energy usage and growth in grass carp Ctenopharyngodon idella (Valenciennes, 1844) and common carp (*Cyprinus carpio* L. juveniles). *IRAQI J. Aquac*., **10**(1),1-24 (2013). doi: 10.21276/ijaq.2013.10.1.1.

4. Radoslav, D., Aleksandar, I., Rajko, G.,Goran, T., Danijela, C. and Lolic, S. Effect of thermal stress of short duration on the red blood cell parameters of *Barbus balcanicus* Kotlik, Tsigenopulos, Rab, Berrebi, 2002. *Academic Journals. African Journal of Biotechnology,* **12**(18),2484-2491(2013).

5. Al-Hamdani, A. and Al-Tai, S. Effect of heat stress on Carassius auratus fish. *Basra Journal of Veterinary Research,* **8** (2): 20-25(2009).

6. Farrell, A.P. Cardiorespiratory performance during prolone swimming with salmonids: aperspective on temperature effects and potential analytical. *Journal of Biological Sciences,* **362**,201-203(2007).

7. Ji, L., Jiang, K., Liu, M., Wang, B., Han, L., Zhang, M. and Wang, L. Low temperature stress on the hematological parameters and HSP gene expression in the turbot Scophthalmus maximus. *Chin. J. Oceanol. Limnol.*, **34**, 430–440(2016). https://doi.org/10.1007/s00343-016-4367-z

8. Zelikoff, J. T., Enane, N. A., Bowser, D., Squibb, K. S., and Frenkel, K. Development of fish peritoneal macrophages as a model for higher vertebrates in immunotoxicological studies: I. Characterization of trout macrophage morphological, functional, and biochemical properties. *Toxicological Sciences*, **16**, 576-89(1991). https://doi.org/10.1093/toxsci/16.3.576.

9. Jamalzadeh, H. R., and Ghomi, M. R. Hematological parameters of Caspian salmon Salmo trutta caspius associated with age and season. *Marine and Freshwater Behaviour and Physiology,* **42**(1), 81-87 (2009). https://doi.org/10.1080/10236240902771226.

10. Brix, O., Thorkildsen, S. and Colosimo, A. Temperature acclimation modulates the oxygen binding properties of the Atlantic Cod (*Gadus morhua* L.) genotypes Hbl* 1/1, Hbl* 1/2, and HbI*2/2- by changing the concentrations of their major hemoglobin components (results from growth studies at different temperatures). *Comparative Biochem. Physiol. A. Mol. Integr. Physiol.,* **138**(2),241-251 (2004). https://doi:10.1016/j.cbpb.2004.04.004.

11. Cheng, C. H., Ye, C. X., Guo, Z. X. and Wang, A. L. Immune and physiological responses of pufferfish (Takifugu obscurus) under cold stress. *Fish and Shellfish Immunology*, **64**,137–145 (2017). https://doi.org/10.1016/j.fsi.2017.03.003

12. Ghasemi Pirbalouti, A. Iranian medicinal and aromatic plants (2nd Edition). Islamic Azad University Publishers, Shahrekord, Iran (in Farsi). 16-23(2009).

13. Hyun Kim, J., Fridman, S., Borochov-Neori, H., Sinai, T. and Zilberg, D. Evaluating the use of garlic (*Allium sativum*) for the remedy of Cryptocaryon irritans in guppies (Poecilia reticulata). *Aquaculture Research*, **50**(1) 431-438(2019). https://doi.org/10.1111/are.13904

14. Farahi, A., Kasiri, M. Sudagar, M. Iraei, M. S. and Shahkolaei, M. D. Effect of garlic (*Allium sativum*) on growth factors, some hematological parameters and body compositions in rainbow trout (Oncorhynchus mykiss), *AACL Bioflux,* **3**(4), 317-323 (2010).

15. Fazlolahzadeh, F., Keramati, K., Nazifi, S., Shirian, S. and Seifi, S. Effect of garlic (*Allium sativum*) on hematological parameters and plasma activities of ALT and AST of Rainbow trout in temperature stresses, *Australian Journal of Basic and Applied Sciences*, **5**(9),84-90(2011).

16. Karimi Pashaki, A., Ghasemi, M., Zorriehzahra, M. J., Sharif Rohani, M. and Hosseini, S. M. Effects of dietary garlic (Allium sativum) extract on survival rate, blood and immune parameters changes and disease resistance of Common carp (*Cyprinus carpio* carpio Linnaeus, 1758) against Spring Viremia of Carp (SVC). *Iranian Journal of Fisheries Sciences*, **19**(3), 1024–1039(2020).

17. Saber Tagrid, H. Histological Adaptation to Thermal Changes in Gills of Common Carp Fishes *Cyprinus carpio* L. *Rafidain Journal of Science*, **22**(1), 46–55(2011). https://doi.org/10.33899/rjs.2011.3246.

18. Mohammad, K. Laboratory guide in toxicology. 1st ed, 2000 pp. (2012).

19. Parrino, V., Cappello, T., Costa, G., Cannavà, C., Sanfilippo, M., Fazio, F. and Fasulo, S. Comparative study of haematology of two teleost fish (Mugil cephalus and Carassius auratus) from different environments and feeding habits. In *European Zoological Journal*, **85**(1), 193-199(2018). https://doi.org/10.1080/24750263.2018.1460694.

20. Coles, E.H. Veterinary Clinical Pathology. 4th Edition, W.B. Saunders Company, Philadelphia, 17-19(1986).

21. Kondera, B., Bojarski, K. Ługowska, B. Kot, M. and Witeska. E. Effects of oxytetracycline and gentamicin therapeutic doses on hematological, biochemical and hematopoietic parameters in Cyprinus carpio juveniles. *Animals,* **10** (12), 2278 (2020). https://doi.org/10.3390/ani10122278.

22. Bailey, M. The mucosal immune system: Recent developments and future directions in the pig. *Developmental and Comparative Immunology*, **33**(3), 375-383(2009). https://doi.org/10.1016/j.dci.2008.07.003.

23. Trinder, P. Determination of Glucose in Blood Using Glucose Oxidase with an Alternative Oxygen Acceptor. *Annals of Clinical Biochemistry: International Journal of Laboratory Medicine*, **6**(1):24-27(1969). https://doi.org/10.1177/000456326900600108.

24. Wootton L.I. Micro-analysis in medical biochemistry in micrometer. 4th ed. London: J&A Churchill, 1964. 264p.

25. Doumas, B.T., Biggs, H.G., Arends, R.L. and Pinto, P.V. Determination of Serum Albumin. *Stand. Methods Clin. Chem.,* **21**, 175–188(1972).

26. Coles, E.H. Veterinary Clinical Pathology; W.B. Saunders Company: Philadelphia, PA, USA; London, UK, p. xii+615(1974).

27. Bower, K. M. Analysis of variance (ANOVA) using MINITAB. *Scientific Computing and Instrumentation*, **17**, 64-65(2000).

28. Seibel, H., Baßmann, B. and Rebl, A. Blood Will Tell: What Hematological Analyses Can Reveal About Fish Welfare. *Front. Vet. Sci.,* **8**,616955(2021).https://doi:10.3389/fvets.2021.616955

29. Verma, A. K., Pal, A. K., Manush, S. M., Das, T., Dalvi, R. S., Chandrachoodan, P. P., Ravi, P. M. and Apte, S. K. Persistent sub-lethal chlorine exposure augments temperature induced immunosuppression in Cyprinus carpio advanced fingerlings. *Fish and Shellfish Immunology,* **22**(5),547-555(2007). https://doi.org/10.1016/j.fsi.2006.08.001.

30. Roberts, R. J. Fish Pathology: Fourth Edition. New york: John Wily and sons (2012). https://doi.org/10.1002/9781118222942.

31. Bozorgnia, A., Hosseinifard, M. and Alimohammadi, R. Acute Effects of Different Temperature in the Blood Parameters of Common Carp (*Cyprinus carpio*). *2nd International Conference on Environmental Science and Technology, IACSIT Press,* Singapore, **6**,52-55(2011).

32. Docan, A., Cristea, V. and Dediu, L. Influence of Thermal Stress on the Hematological Profile of Oncorhynchus Mykiss Held in Different Stocking Densities in Recirculating Aquaculture Systems. *AGRIS,* **55**, 267–272(2011).

33. Sheen, L. Y., Chen, H. W., Kung, Y. L., Liu, C. T. and Lii, C. K. Effects of garlic oil and its organosulfur compounds on the activities of hepatic drug-metabolizing and antioxidant enzymes in rats fed high- and low-fat diets. *Nutrition and Cancer*. **35**(2),160-166(1999).

34. Al-Azzawie, H. F. and Alhamdani, M. S. S. Hypoglycemic and antioxidant effect of oleuropein in alloxan-diabetic rabbits. *Life Sciences*, **78**, 1371–1377(2006). https://doi.org/10.1016/j.lfs.2005.07.029.

35. Shalaby, A. M., Khattab, Y. A. and Abdel Rahman, A. M. Effects of garlic (*Allium sativum*) and chloramphenicol on growth performance, physiological parameters and survival of Nile tilapia (*Oreochromis niloticus*). *Journal of Venomous Animals and Toxins Including Tropical Diseases*, **12**(2), 172-201(2006). https://doi.org/10.1590/S1678-91992006000200003.

36. Martins, M. L., Moraes, F. R., Miyazaki, D. M. Y., Brum, C. D., Onaka, E. M., Fenerick, J. and Bozzo, F. R. Alternative treatment for Anacanthorus penilabiatus (Monogenea: Dactylogyridae) infection in cultivated pacu, Piaractus mesopotamicus (Osteichthyes: Characidae) in Brazil and its haematological effects. *Parasite*., **9**, 175-180(2002). https://doi.org/10.1051/parasite/2002092175.

37. Ndong, D. and fall, J. The effect of garlic (*Allium sativum*) on growth and immune responses of hybrid tilapia (Oreochromis niloticus x Oreochromis aureus). *Journal of Clinical Immunology and Immunopathology Research*, **3** (1),1–9 (2011).

38. Nwabueze, A. A. The Effect of Garlic (*Allium sativum*) on Growth and Haematological Parameters of *Clarias gariepinus* (Burchell, 1822). *Sustainable Agriculture Research*, **1**(2), 222-228(2012). https://doi.org/10.5539/sar.v1n2p222

39. Hille, S. A literature review of the blood chemistry of rainbow trout, Salmo gairdneri Rich, *J. Fish Biol.,* **20**, 535-569(1982). https://doi:10.1111/j.1095-8649.1982.tb03954.x.

40. Metwally, M. A. A. Effects of garlic (Allium sativum) on some antioxidant activities in tilapia nilotica (Oreochromis niloticus). *World Journal of Fish and Marine Sciences.* **1** (1),56–64(2009).

تأثير الثوم على بعض معاير الدم في أسماك الكارب (*Cyprinus Carpio* L) الشائع مجهده بالحراره

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استخدمت ٦٠ سمكة الكارب الشائع بمتوسط وزن ١٠٠± ١٠ غرام في هذه الدراسة. لغرض دراسة تأثير الإجهاد الحراري على مختلف مكونات الدم ودور الثوم في الحد من هذه الآثار. في المختبر تم تكييف الأسماك لمدة سبعة أيام. وتم المحافظة على درجة حرارة الماء في٢٥ درجة مئوية . ووضعت الأسماك في ستة أحواض زجاجية مقاس ٤٠ × ٤٠ × ٨٠ سم وتم تقسيمها إلى ست مجموعات. كل مجموعة تحتوي على ١٠سمكة .

عرضت الاسماك في المجموعة الأولى للإجهاد الحراري ٣٠درجة مئوية لمدة اسبوع. المجموعة الثانية كانت في درجة حرارة الماء العادية ٢٥ درجة مئوية لمدة اسبوع. المجموعة الثالثة عرضت الا سماك المجموعة للإجهاد الحراري مع اعطاء الثوم عن طريق أنبوب المعدة بجرعة ١,٦٣ جم\١٠٠ جم من وزن الجسم في نفس الوقت لمدة اسبوع. المجموعة الرابعة تعرضت الأسماك للإجهاد الحراري في الأسبوع الأول ثم جرعت الثوم في الأسبوع الثاني. المجموعة الخامسة حفظت عند ٢٥ درجة مئوية وجرعت االثوم بنفس الوقت ولمدة أسبوع ايضا. أسماك المجموعة السادسة جرعت االثوم في الأسبوع الأول ثم عرضت للإجهاد الحراري في الأسبوع الثاني.

أظهرت النتائج وجود زيادة معنوية في تركيز الهيموجلوبين ، حجم خلايا الدم المرصوصة، الجلوكوز ، البروتين الكلي في المجموعة الأولى المعرضة للإجهاد الحراري ، لكن مؤشر الإجهاد انخفض بشكل ملحوظ ، كما أدى الثوم إلى تحسين صورة الدم ، خاصة في المجموعتين الثالثة والسادسة.