

EFFECT OF ADDING GARLIC AND TURMERIC IN THE DIET CONTAMINATED WITH AFLATOXIN B1 ON PERFORMANCE OF GROWTHAND DIFFERENT BLOOD INDICATORS OF COMMON CARP FISH (*CYPRINUS CARPIO* L.)

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Abstract

The present study was conducted to investigate the effect of adding different levels of garlic and turmeric in AFB1 contaminated diets on 210 young Cyprinuse carpio L. (total weight between 45 to 50g). The fish were randomly distributed over seven treatments of 10 fish per aquarium and three replicates. The first treatment T1 represented the control diet (commercial diet consisting of 27% protein without additives), the second treatment T2 (Aflatoxin 15% of 99 ppm concentration) and the third treatment T3 consisting of (1% garlic concentration and 15% concentration of 99 ppm Aflatoxin), the fourth treatment T4 (1% garlic, 1% turmeric and 15% concentration of 99 ppm Aflatoxin), the fifth treatment T5 (1% curcuma and 15% concentration of 99 ppm Aflatoxin), the sixth treatment T6 (1% garlic, 2% turmeric and 15% concentration of 99 ppm Aflatoxin) and finally the seventh treatment T7 (1% garlic, 3% turmeric and 15% concentration of 99 ppm Aflatoxin). At the end of the experiment, the following results were obtained: The highest daily weight recorded an increase to 0.10 g for T4 treated fish, compared to the rest of the treatments, while the T2 fish recorded the lowest daily increase of weight reaching 0.04 g, compared to the fluctuation of the daily weight increase of the rest of treatments. There was an increase of (P<0.05) in the qualitative growth of fish weight of T4 and the control of 1.8 and 1.5 values respectively and for the qualitative growth of the total length of fish for the same treatments reached 0.47 and 0.41 respectively. Whereas the noticed qualitative decrease (P<0.05) of weight and length growth of the T3 fish reached 1.09 and 0.05 respectively. Regarding the food conversion factor, T4, control and T3 treated are the best qualitatively (P<0.5), compared to the other treatments, with the values 1.25, 1.28 and 1.39 respectively. It was noticed that the number of red and white blood cells fluctuated during the study. There was a significant increase (P<0.05) of the number of red blood cells in T2 treated fish reached 2.6 cells / mm compared with a decrease in the number of T3, T4 and control with similar values of 1.2, 1.3 and 1.4 cells / mm3 respectively. The highest qualitative increase (P<0.05) of white blood cell in T4, T2 and T5 treatment fish was 11.3, 10.8 and 10.6 cells / mm3, respectively, compared with the stability of the number in the other treatments despite the slight changes. T4 and control fish recorded the lowest values reaching 8.8 and 8.9 cells/mm 3. Fish fed on T2 diet showed a qualitative increase (P<0.05) of blood concentration to 39.6% and a decrease in T3, T4 and control fish to 27.5%, 25.7% and 28.7%. Also, blood hemoglobin concentration values recorded a qualitative increase (P \leq 0.05) for T2 at 15.9 g/dL, whereas the values for T4 and control fish decreased at 8.8 and 8.6 g/dL respectively. The total protein values recorded an increase (P<0.05) T4 and control fishes of 16.6 and 17.8 mg/mL respectively, compared with T2 fish recoding the lowest values of 11.3 mg/L. Albumin concentration recoded a qualitative increase (P<0.05) for the treated T4 and control fish reached 4.8 and 4.9 mg / mL, the lowest results were in T2 reaching 1.2 mg/mL.

Key words: garlic, turmeric, aflatoxin B1

Introduction

Fish is an important commercial food in the developed countries of the world, it is known that fish meat is very important for human nutrition in terms of health, as it is one of the most important economic goods traded in developed countries, and farming began to spread in the water bodies around the world to supply human food consumed It is necessary to control this type of aquaculture and develop its functions to meet the expected future increase of hunger and greed in the world (Muharram, 2004). Food The use of industrial food is traditional in aquaculture, due to the extent of its utilization in aquaculture and the abundance of capital. On the economic considerations of these farms (Al-ashaab *et* *al.*, 2017). The use of these industrial foods exhibit many different problems in the production of fish, moreover, the use of these foods may result in many poisonous fungi that produce dangerous metabolites called fungal toxins (Abdelhamid, 2004). Aflatoxins are one of the most contaminated fungal toxins in food, feed and their various components. They are a common global problem in fish breeding, especially in hot and humid areas. Low productivity (Mahfouz and Sherif, 2015).

Aflatoxin AFB1 is considered the main pollutant for feed aquatic organisms, it is also the main cause of the death of a number of different fish species and increase susceptibility to disease, and low productivity in addition to the remaining effects of these toxins in the meat of these fish, as it leads to many economic losses and cause poisoning to human consumers Her and the animal (Abdelhamid *et al.*, 2004).

To this end, many researches and studies are designed to control the presence of these toxins and reduce the risks of them food content to reach the socalled safe limits of different animal species using many natural, chemical, biological and physical methods (Abdelhamid, 2004). Many medicinal plants and herbs have been evaluated to reduce the harmful effects of aflatoxin B1 in fish such as garlic Allium sativum and Curcuma longa (A-bayati and Raaed, 2017). In protecting cells from oxidative stress and damage to DNA (Carmia, 2001 Balu; et al., 2006; Hatcher et al., 2008; Sashidha, Sujatha, 2010; Chattopadhyay et al., 2014). The present study aims to evaluate the defensive and therapeutic properties of turmeric and garlic when they are added as fortified additives in common carp diet (Cyprinus carpio) of the metabolism of aflatoxin AFB1 by studying the growth indicators of fish and studying the blood properties of fish.

Materials and Methods

This study was carried out in the Fish Nutrition Laboratory, Fish Department, Animal and Fisheries Center, Agricultural Research Department, Ministry of Science and Technology in Zafaraniya south of Baghdad. It used 250 common carp (*Cyprinus carpio* L.) with a total length ranging from 14.3 to 14.8 cm and a total weight of 45 to 50 g/fish. For the purpose of studying the effect of garlic and turmeric on the growth and blood traits of common carp fish by adding them to aflatoxin-contaminated diets B1, 210 fish were randomly selected and distributed to 21 glass basins of $(30 \times 60 \times 30)$ cm pre-prepared with water and oxygen. 10 fish per aquarium and a total of 30 fish per treatment.

Growth indicators

Measurements were made for every two weeks, as the total length of the fish was measured according to Philipose *et al.*, (2013).

Blood standards

Toy image tests were reportedly carried out (Blaxhall and Daisey, 1973): the total number of red blood cells. RBC. Total number of white blood cells (WBC), PCV%, hemoglobin concentration (Hb) according to researcher (Carneiro *et al.*, 2007).

Statistical analysis

Transactions according to Duncan Polynomial Test (Al - Aqili and Al - Shayeb, 1998).

Results and Discussion

Growth indicators

Effect of adding different levels of garlic and turmeric powder to the contaminated diets of AFB1 Aflatoxin on fish weight.

It is clear by the table 1 that there was fluctuation in fish weight, the treatment fish T2 shows significant increase (p<0.05) in its weight at the meddle of treatment with average 5, 19gm compared with other treatments, during the continuing of treatment till the end the TA treatment shows the highest significant increasing (p<0.05) in fish weights at a total final weight reached to 52, 0gm. The table 4 results indicate that fish of treatment 4 have been recorded as a highest total significant increasing (p<0.05) at an average 11, 5 gm during whole time of treatment at a daily average weight 0, 19 gm/day, compared with other treatments whereas there was clear significant decreasing of fish treatment T2 total weight at an average 3.8 gm and total daily increasing weight reached to 0.04 gm/day.

The results of table 1 show that common carp fish in resent study shows the highest daily weight increasing at the end of T4 treatment fish experiment after feeding with AFB1 Aflatoxin contaminated diet and supported with 1% garlic and 1% turmeric compared with other treatments, whereas the fish of treatment T2 which feed with AFB1 aflatoxin contaminated diet show lowers daily weight at the end of treatment.

Some of studies refer to using *Allium sativum* in diets of fish and cattle where it's using become well-known as a supporting and growth estimating supplier, increasing levels of its weights and high efficiency of metabolism and activating of immunity systems against various disease that attack *O. niloticus* fish (Diab *et al.*, 2002, Metwally 2002).

The studies of (Khalil *et al.*, 2001), (Diab *et al.*, 2002) and (Shalaby et al., 2006) show that allicin in garlic helps to improve digestion that leads to make benefit of energy to get perfect daily growth, whereas the best growth of O. niloticus fish when feed it with diets supported by garlic was 1, 5 to 3% which it is approximate to recent results, in spite of little concentrates that are used in this study was the most effect of fish growth. the effect of adding garlic to diets have various results because the instability of allicin which is related to differ age, kind, size of fish, experimental environment and the period of AFB1 Aflatoxin exposure (ALY et al., 2008) (Aly and Mohamed 2010). The human consumption of contaminated food with Aflatoxin is consider the great dangerous to the health in various parts of the world especially the Asian and African countries (Wild and Gong, 2010). Some studies refer to accumulating of poisons of Aflatoxin in the body of O. niloticus fish when it is exposure to different levels of AFB1 which leads to make harm to the liver cells in human body (Salem et al., 2009) (Han et al., 2010) (Salem et al., 2012) (Rahdi, 2018) whereas the study of (Leya et al., 2017) refer to increasing of weight and improving of growth activities of C. mrigala fish which are feed on different levels of turmeric where the higher daily weight increasing was recorded by percentage of 1, 5% of turmeric in diets feed of fish compared of other percentages, this was approximate to recent results. The results refer to ability of improving fish growing which are feed with garlic and turmeric contaminated diets like treatment T4 that fallowed by treatments T5 and T6 Compared to other trading diets.

The effect of adding different levels of garlic and turmeric powder to the contaminated diets with AFB1 aflatoxin on specific growth values and metabolism factor of fish

We can see in table 1 the value of specific growth

for total weight, length and metabolism of common carp fish which are feed on AFB1 aflatoxin contaminated diets and supported with adding different levels of garlic and turmeric powder all the experiment period where it is recording the higher significant increasing p<0, 05 at the end of experiment for both T4 treatment and control of specific growth to fish weight in average 1.8 and 1.5 consecutively whereas the lower value of specific growth for fish of treatment T2 with 1, 09 average compared with other treatment while the growth of specific length has clear significant increase p<0, 05 in treatments T4 and T3 with average values 0, 47 and 0, 41 respectively and clear lower of fish length in treatment T2 by average 0, 05 in compared with other treatments ,the fish of treatment T4, control and T3 was the best significantly p<0, 05 with average values 1.25, 1.28, and 1.39 respectively compared with other treatments.

The study of *Mahmood* 2008 showed that diets supported with 1%, 5% of garlic was the best significantly in what are related of specific growth for weight and total length of fish. It is accepted with recent study where is the improving of growth in different levels related to the effect of allicin in garlic which is enhances gastric plantation that leads to promote the digestion and make benefit of energy and improve growth in the best way (Khalil *et al.*, 2001). the study of (Mooraki *et al.*, 2018) reported that in spite of the important of adding various levels of turmeric powder 0.1%.

The study of Maniat *et al.*, (2016) showed that the effects of adding garlic powder vary according to the concentrations added to fish diets as well as the different types of fish, their environment, sex and sizes, and that the high concentrations of it may have a negative impact on the qualitative characteristics of fish growth. It is similar to the current study. The current results may indicate the importance of adding a mixture of garlic powder and turmeric in different proportions and their

 Table 1: Effect of adding different levels of garlic and turmeric powder to the contaminated diets of AFB1 Aflatoxin on fish weight(Average± standard error).

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Daily weight gain	Total weight gain	Total weight of fish (g)			Treatment
of fish (g/d)	of fish(g)	End of the	The center of	Beginning of	
		experiment	the experiment	the experiment	
$0.16 \pm 0.02b$	$9.4\pm0.8b$	$49.9 \pm 0.3b$	$47.3 \pm 1.7b$	5.40±1.9a	(control) T1
$0.04 \pm 0.02d$	$3.8 \pm 0.5 d$	$44.1 \pm 0.6d$	51.9±1.8a	$3.40 \pm 1.8a$	T2
$0.12 \pm 0.05c$	$7.2 \pm 0.4c$	$47.7 \pm 0.7c$	$44.4 \pm 1.9c$	$40.5 \pm 1.9a$	T3
$0.19 \pm 0.01a$	$11.5 \pm 0.2a$	52.0±0.6a	$44.5 \pm 1.9c$	$40.5 \pm 1.9a$	T4
$0.13 \pm 0.04c$	$7.5 \pm 0.7c$	$47.5 \pm 0.8c$	$45.4 \pm 1.7c$	$40.5 \pm 1.7a$	T5
$0.14 \pm 0.05c$	$8.2 \pm 0.7c$	$48.5\pm0.7c$	$44.8 \pm 1.8c$	$40.3 \pm 1.8a$	T6
$0.11 \pm 0.03c$	$6.7 \pm 0.6c$	$47.1 \pm 0.8c$	$46.4 \pm 1.5 bc$	$40.4 \pm 1.7a$	T7

*Averages with different characters within the column differ significantly between them (p<0.05).

positive impact on growth performance, especially those that feed on feed contaminated with Aflatoxins AFB1. Onchorhynchus mykiss (Nya and Austin, 2000; Farahi et al., 2010), Sterlet sturgeon (Lee et al., 2012) (2012), Epinephelus coioides (Guo et al., 2012), and Clarias gariepinus (Nwabueze et al., 2012). While the study of Mahmoud et al., (2014) showed that the addition of turmeric at different rates as supplements to fish diets did not show a significant effect in growth performance. Contamination of feed material varies according to mold types and by geographical area, humidity, temperature and sanitary conditions (Mohamed et al., 2017). The Radhi (2018) study showed that common carp fish exposed to feeds contaminated with aflatoxin toxins AFB1 showed a significant decrease in growth indicators for total weight and daily growth rate. AFB1 causes bleeding in tissues due to its effect on endothelial cells in the circulatory system. Endothelial cells appear to be sensitive to AFB1. Most pathological damage is observed in these cells (Mehrim et al., 2006). Which has an effect on the digestive system in fish (Applegate et al.,).

Blood standards

Red blood cells RBC and white WBC

Table 3 indicates the changes in fish red and white blood cell values for all treatments during the duration of the experiment. The highest fluctuation (P < 0.05) was observed in the number of red blood cells of T2 fish at the middle of the experiment at a rate of 2.2 cells/mm3 and at the end it reached 2.6 cells/mm3 compared to the decrease in the number of fish for all other nutritional treatments. T6 treatment at 1.2 cells/mm3. As shown in the same table for white blood cells, a significant increase (P<0.05) was observed in the number of cells at the middle of the experiment for T4, T2 and T5 fish at 11.3, 10.8 and 10.6 cells/mm3, respectively, compared to the steady numbers of the other treatments. Minor changes were recorded for the lowest values for fish-treated control at a rate of 8.6 cells/mm 3.

Mahmood's study (2008) showed a clear effect on the physiological traits of common carp C. When fed on diets fortified with different levels of powder, garlicfortified diets had a significant effect in increasing the number of red blood cells, while the number of white blood cells increased. In fish fed diets containing 5% garlic powder, compared to other experimental treatments. Marentek et al., (2013) study showed that when feeding tilapia (10.4 grams) on fortified diets of 2% garlic powder for 10 weeks, growth performance improved and macrophage cell activity increased, as well as an increase in Types of white blood cells compared to fish control group. The study of Martins et al., (2002) confirmed that the addition of garlic powder to the dietary diet of fish leads to clear changes in blood standards and vary depending on fish species and experimental conditions.

In the study of Mooraki et al., (2018) to evaluate the effect of turmeric powder added as a dietary supplement on Andinoacara rivulatus, the results showed that fish fed on a diet supported by 0.3% of turmeric powder recorded a significant increase in the number of red and white blood cells compared to the control group. Al-Bayati study indicated that there was no significant effects of garlic powder on the number of red blood cells of laying hens.

Arunkumar et al., (2016) recorded 15 days after feeding common carp on turmeric-fortified diets with an increase in the numbers of red and white blood cells, while the lowest numbers of cell types were recorded for the same fish after 45 feeding in the fortified feed at a concentration of 0.3 ppm. The present results indicate that there are obvious effects of adding garlic and turmeric powder in different proportions on fish fed with aflatoxins AFB1 toxins. The fluctuation in the numbers of red and white blood cells in fish may be due to immune physiological changes. The effect of garlic and turmeric

Table 2: The effect of adding different levels of garlic and turmeric powder to the contaminated diets with AFB1	aflatoxin on
specific growth values and metabolism factor of fish (Average± standard error).	

Food conversion		Specific growth of length		Specific weight growth		
End of the	The center of	End of the	The center of	End of the	The center of	Treatment
experiment	the experiment	experiment	the experiment	experiment	the experiment	
$1.28 \pm 0.09a$	$1.23 \pm 0.15a$	$0.38\pm0.02b$	$0.35 \pm 0.07a$	$1.5 \pm 0.21a$	$1.30 \pm 0.15b$	(control) T1
$1.61 \pm 0.05c$	$1.50 \pm 0.11c$	$0.05\pm0.02d$	$0.04 \pm 0.01d$	1.09±0.16c	$1.0 \pm 0.15d$	T2
$1.39 \pm 0.09a$	$1.33\pm0.05b$	0.41 ± 0.12 ab	$0.25\pm0.08b$	$1.40 \pm 0.23b$	$1.33 \pm 0.23b$	T3
$1.25 \pm 0.09a$	$1.20 \pm 0.11a$	$0.47 \pm 0.03a$	$0.41 \pm 0.05a$	$1.8 \pm 0.25a$	$1.65 \pm 0.1a$	T4
$1.45\pm0.04b$	$1.35 \pm 0.11b$	$0.20 \pm 0.12c$	$0.18 \pm 0.04c$	$1.22 \pm 0.12b$	$1.16 \pm 0.10c$	T5
1.47±0.06b	1.33±0.07b	0.22±0.09c	0.17±0.5c	1.23±0.11b	1.17±0.11c	T6
1.39 ± 0.05 ab	$1.32 \pm 0.03b$	$0.25 \pm 0.06c$	$0.22 \pm 0.06 bc$	$1.22 \pm 0.10b$	$1.18 \pm 0.15c$	T7

*Averages withdifferent characters within the column differ significantly between them (p<0.05).

WBC (cell × 10 ³ /Mm ³)		RBC (c		
End of the The center of		End of the The center		Treatment
experiment	the experiment	experiment	the experiment	
$8.8 \pm 0.22c$	8.6 ± 0.34b	$1.4 \pm 0.12b$	$1.3 \pm 0.12c$	(control)T1
$11.7 \pm 0.34a$	$10.8 \pm 0.35a$	$2.6 \pm 0.13a$	$2.2 \pm 0.12a$	T2
$9.8 \pm 0.35b$	$9.4 \pm 0.31b$	$1.2 \pm 0.13b$	$1.5 \pm 0.14b$	T3
$8.9 \pm 0.32c$	$11.3 \pm 0.35a$	$1.3 \pm 0.12b$	$1.4 \pm 0.14 bc$	T4
$9.8 \pm 0.29b$	$10.6 \pm 0.34a$	$1.15 \pm 0.11c$	1.05 ± 0.11 d	T5
$9.6 \pm 0.34b$	$9.9 \pm 0.33b$	$1.14 \pm 0.12c$	$1.2 \pm 0.13c$	T6
$9.5 \pm 0.34b$	$9.2 \pm 0.29b$	$1.16 \pm 0.12c$	$1.06 \pm 0.10d$	T7

Table 3: Rate rates Red blood cells RBC and white WBC For common carp fish in the current study (Average± standard error).

*Averages with different characters within the column differ significantly between them (p<0.05).

Table 4: The rates of pH and hemoglobin values for common carp fish in
the present study (Ave rage \pm standard error).

Hb (g/Deciliter)		(% P		
End of the	The center of	End of the	The center of	Treatment
experiment	the experiment	experiment	the experiment	
8.6±0.5c	$8.9 \pm 0.5d$	$28.7 \pm 1.4 d$	$28.7 \pm 1.4c$	(control) T1
$15.9 \pm 0.7a$	$13.1 \pm 0.5a$	$39.6 \pm 1.4a$	38.6±1.4a	T2
$10.1 \pm 0.7b$	$10.9 \pm 0.6c$	$25.7 \pm 1.4 d$	22.3 ± 1.4 d	T3
$8.8\pm0.7c$	$10.3\pm0.4c$	$27.5 \pm 1.5 d$	$30.3 \pm 1.3c$	T4
$10.7 \pm 0.4b$	$11.8\pm0.5b$	$34.0\pm1.4b$	$35.7 \pm 1.3b$	T5
$10.9 \pm 0.6b$	$12.7\pm0.5b$	$31.6 \pm 1.8c$	$33.7 \pm 2.1 \text{bc}$	T6
$10.7 \pm 0.4b$	$11.8 \pm 0.7b$	$33.5 \pm 1.7b$	$35.0 \pm 1.8b$	T7

*Averages with different characters within the column differ significantly between them (p < 0.05).

may vary in different ratios individually or in mixture on changes in fish, the increase in red blood cells of T2treated fish does not stimulate the immune system and the effort of the fish is fed by contaminated diets. The present results may indicate the stability of the immune system of T4 fish by adding the levels of garlic powder (1%) and turmeric (1%) which reduced the effects of mycotoxins and therefore the powder mixture was the best in achieving relatively acceptable results.

PCV% and hemoglobin concentration

Table 4 indicates the changes in the values of fish accumulation and hemoglobin for all treatments during the duration of the experiment. The fish fed on the T2 diet showed a significant increase (P<0.05) for the concentration of blood in the middle and the end of the experiment by 38.6% and 39.6% compared to the other treatments. At the end of the experiment, the accumulated values of T7, T5 and T6 decreased by 34.0% and 33.5% and 31.6% respectively. The lowest values were recorded at the end of the experiment for T3 and T4 and control by 27.5% and 25.7%. 28.7%. Also, the hemoglobin concentration of fish blood (Table 10) showed a significant

increase (P<0.05) of values for T2 fish at the center and end of the experiment (13.1 and 15.9g/dl, respectively). While the values were fluctuating between high and low to the end of the experiment, they recorded lower values for hemoglobin for T3 fish at 10.1g/dL, T5 and T7 at 10.7g/dL each, then T6 at 10.9g/dL, while lower values were recorded. Values of control fishes and T4 at 8.6 and 8.8gm/dL Mooraki et al., (2018) indicated that there were no significant differences in the values of stacking and hemoglobin concentration in A. rivulatus when fed to different levels of turmeric powder individually despite an increase Slight in values of 0.3% of turmeric for blackberry compared to control fish. It is similar to the current results indicating the effective effect of garlic and turmeric mixture in different proportions. Current results may indicate an increase in the number of red blood cells and have a direct impact on the pH values. The present indications may be attributed to the effectiveness of the garlic and turmeric mixture in the diet T4, T6 and T7, which showed improvement in blood performance and stability at the end of the experiment compared to the contaminated T2 diet without additives. The improvement of fish blood standards may be due to different substances in the proportions

used in the current experiment, which work on the efficiency of the digestive system and play an important role in the disposal of food waste, and stimulate the intestines to absorb some minerals and vitamins, as it produces a substance that mimics the work of antibiotics that fight bacteria Harmful (Abboudi *et al.*, 2006).

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