

## **EFFECT OF (MYCOFIX® PLUS) AND AFLATOXIN ON HEALTH AND PERFORMANCE OF BROILER CHICKENS**

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### **ABSTRACT**

AN experiment conducted at the department of veterinary public health, college of veterinary medicine, Mosul-Iraq, during September to October 2009 to study the effect of Mycofix ® plus (MY) on broiler health performance during aflatoxicosis. Eighty, one- day- old commercial male broiler chicks (Ross 308), were distributed to 4 dietary treatment groups with 2 replicates of 10 chicks each. Birds were reared for 35 days. All birds were fed on diet with or without aflatoxin (AF), and with or without (MY) in feed. The treatment groups were as follow: G1 (0 part per million (ppm) AF without MY); g2 (2.5 ppm AF without MY ); g3 (0 ppm AF & 0.25% MY ); G4 (2.5 ppm AF & 0.25 % MY ). Body weights and feed intake were recorded weekly. At 35 of bird's age, five birds were randomly selected from each group for estimation of WBCs, RBCs, HB and ESR; glucose, total protein, cholesterol, and triglycerides; serum levels of AST,ALT and ALP enzymes; antibody titers and CV% against Newcastle disease (ND) vaccine; relative weights of bursa of Fabricious , thymus, spleen, proventriculus, gizzard, kidney, liver, heart and pancreas, in addition to recording liver lesions. Results show that MY was effective in ameliorating the negative effect of AF on growth performance (body weight gain ,feed consumption and feed conversion ratio); on relative weights of affected internal body organs and the score lesion of liver; on blood picture (WCS, HB and glucose); serum enzymes (AST, ALT and ALP); antibody titer and CV% against ND vaccine in comparison to AF contaminated feed and control groups.

## INTRODUCTION

Aflatoxins (AF), the toxic secondary metabolites produced by *Aspergillus flavus*, *Aspergillus parasiticus* and *Aspergillus nominus*, are of major concern in the poultry production. AF metabolites are stable and fairly resistant compounds to degradation (Edds,1997). These metabolites are usually produced during the growth of the above mentioned fungi on certain foods and feedstuffs under favorable conditions of moisture, temperature and aeration (Dragan, *et al.*, 2010). Their toxicity depends on several factors including their concentrations, duration of exposure, species, sex, age, and health status of animal (Akande *et al.*, 2006). Contamination of feed with AF causes aflatoxicosis in poultry which is characterized by reduced feed intake, decreased weight gain, poor feed utilization (Al- Sadi *et al.*,2000; Bennett *et al.*, 2003; Clanek *et al.*, 1997; Dalvi and Ademoyero, 1984), increased mortality (Giambrone *et al.*, 1985), changing in the relative weight of internal organs (Smith and Hamilton, 2001), affecting blood profile, blood biochemical's and blood enzymes (Al-Jubory *et al.*, 2001; Oguz *et al.*, 2000a), inducing pathological conditions in liver and other organs (Espada *et al.*, 1992). AF can also lead impairment of immunity, which is able to enhance susceptibility to some environmental and infectious agents (Campbell *et al.*, 1983; Chang and Hamilton, 1982) and Severe economic losses have been reported in the poultry industry due to aflatoxicosis (Sharline *et al.*, 1980; Johri and Sadagopan, 1989).

Pre and post-harvest contamination can be reduced by using appropriate agricultural practices. However, the contamination is often unavoidable and still remains a serious problem associated with many important agricultural commodities, which emphasizes the need for a suitable process to inactivate the toxin. Besides the preventive management, several approaches have been employed including physical, chemical biological treatments and solvent extraction to detoxify AF in contaminated feeds and feedstuffs (Doyle *et al.*, 1982). All these methods cannot be used in practical feed manufacturing, because of the limitation of the nutrients decomposition, non availability of commercial methods and their residual effects. The increasing number of reports on detoxification of AF in poultry feed using different techniques has given rise to a demand for practical and economical detoxification procedures. Since the beginning of 1990s, the

adsorbent-based studies have also been reported to be effective in removing AF from contaminated feed and minimize the toxicity of AF in poultry. Among several adsorbents commercially available in the market, Zeolites (Miazzo *et al.*, 2000), bentonites (Ibrahim *et al.*, 2000; Pasha *et al.*, 2007,2008) and clinoptilolite (CLI), (Oguz *et al.*, 2000a,2003; Oguz and Kurtoglu , 2000b), and a new promising adsorbent, Mycofix® plus, were preferred because of their high binding capacities for AF and their reducing effect on AF-absorption from the gastrointestinal tract. Mycofix® plus was effectively used by many authors in alleviating T-2 toxicosis (Omar, 2010), and impairment of infectious bursal disease antibodies in broilers (Jargees ,2007).

For understanding the effect of Mycofix on aflatoxin detoxification, a study was conducted to investigate its effect on broiler health and performance during aflatoxicosis.

## **MATERIALS AND METHODS**

### **Birds and diet:**

Eighty, one-day-old commercial male broiler chicks (Ross 308), were randomly distributed into 4 dietary treatment groups with 2 replicates of 10 chicks each. Birds were reared for a period of 35 days in batteries with continuous lighting and good ventilation. All birds were fed on diet with or without either aflatoxin (AF) or Mycofix® plus (MY). The ingredient composition is contained 22.0 % crude protein and 3060 kcal/kg metabolizable energy for the starter diet ; 20 % crude protein and 3145 kcal/kg metabolizable energy for grower diet and 18.0% crude protein and 3200 kcal/kg metabolizable energy for finisher diet based on lesson and summers (1997). Feed and water were available on *ad libitum* basis. All the birds were vaccinated against Newcastle Disease in drinking water with B<sub>1</sub> vaccine at one day and with Lasota strain at 21 days .( Nagy, 1999)

### **Experimental design:**

The experimental design consists of four dietary treatment groups; G1 (0 ppm AF & 0 MY); G2 (2.5 ppm AF & 0 MY); 3G (0 ppm AF & 0.25 % of MY); G4 (2.5 ppm AF & 0.25 % of MY).

**Aflatoxin production and analysis:**

Aflatoxins were produced by the inoculation of *A. parasiticus* NRRL 2999 (Kindly provided from the college of Agriculture and forestry, Mosul University) on rice in 1/2-liter Erlenmeyer flasks as described by (Shotwell *et al.*, 1966). Fifty ml distilled water were added to the rice, and the mixture was allowed to stand for 5 minutes with frequent shaking. The flasks were tightly plugged with cotton and autoclaved at 121°C at 15 (pounds per square inches) (psi) for 15 min and cooled at room temperature. They were then inoculated with 3ml spore suspension in a sterile environment, placed on an orbital shaker at 60 rpm and incubated at 28 °C. On 7-d the flasks were again autoclaved at 121°C at 15 psi for 15 min, and placed in a hot oven at 60 °C for 24hr till all the moisture was removed. The AF containing rice was grinded to powdered form and was quantitatively evaluated using thin layer chromatography (TLC) technique according to (Thomas *et al.*, 1975), using the formula:

$$\text{AF mg/kg} = \frac{\text{DM X 106}}{\text{E X 200 X 10 X L}}$$

Where D = optical density(optical density at 326 nm-optical density at 420 nm).

M= molecular weight.

E= absorbance factor.

L cell thickness (cuvette) in cm.

The AF containing rice was mixed in feed according to the calculation to get the desirable level of aflatoxin ( 2.5 ppm) in the feed. The prepared experimental diets were analyzed again using TLC technique to confirm the AF levels.

**detoxification of AF:**

A commercially available Mycofix® plus was added to the feed at the recommended dose rate of 0.25 % (Treatment groups 2&4).

**Sampling:**

Body weights and feed intake per group was recorded weekly. At 35 of bird's age, blood samples (3 ml) were collected from wing vein of randomly selected five birds per replicate for the calculation of WBCs, RBCs, HB, ESR, according to Coles *et al.*, (1986). Blood serum was separated for estimation of total protein, glucose, triglycerides cholesterol ALT, AST and ALP according to instructions of commercial kits from BIOLABO SA, France, by spectrophotometer (JENWAY 6300). After blood collection, birds were humanely killed and liver, kidney, heart, proventriculus, gizzard, pancreas, bursa of Fabricius, thymus and spleen were removed and weighed. Liver lesions were recorded. Antibody titers to Newcastle disease (ND), was determined using Enzyme linked immune-sorbent assay (ELISA) as described by (Nagy, 1999).

**Statistical analysis:**

The results (group means) were subjected to analysis of variance (ANOVA). Means were subjected to Duncan's test and statistical significance was accepted at  $P \leq 0.05$  (Bruing and Kintz, 1977)

## **RESULTS**

**Growth performance:**

Body weight of broiler chicks through 5 weeks of experimental period show significant differences between experimental treatment groups (Table 1). There was a significant ( $p < 0.05$ ) reduction in body weight in G2, when chicks fed 2.5 ppm AF, compared with control group G1, G3 and G4. The reduction was noticed from the first week and persist through the following four weeks of rearing broilers. Addition of Mycofix® plus, in feed at a rate of 0.25 % (G4), was effective in ameliorating the negative effect of AF on weight gain compared to G2, and by restoring the body weight to that of control one at 35 days of age. Numerical better body weight in G3 was obtained, after addition of Mycofix® alone to the basal diet than that of the control

group. The total body weight gain through 35 days of rearing period represents in (Figure 1). From figure it is evident that the gain was highest in both G1 and G3, in which birds fed AF free diet, or when amended with 0.25 % of Mycofix® plus alone. In the second order was G4, in which AF effect was counteracted by Mycofix® plus addition in feed. The least gain weight was noticed in G2, in which chicks fed AF only.

There was no significant difference in the feed consumption between G1, G3 and G4. A significant ( $p < 0.05$ ) reduction in the feed consumption was found when AF contaminate broiler diet with 2.5 ppm AF. A reflection of feed consumption and body weight gain in the four treatment groups was noticed in the results of feed conversion ratio. The lowest feed consumption and feed conversion ratio were reported in G2, in which chicks fed AF compared to all other groups (G1, G3 and G4). Addition of Mycofix® plus alone to feed (G3) had not negative effect on the studied growth performance parameters (Table 2).

**Liver lesions:**

The effect of AF and Mycofix® plus on liver in broilers at 35 days of age is presented in table 3. From table, it is evident that AF had a significant negative effect on the liver parenchyma of broiler chicks in treatment group 2, when compared with that of control group, by changing liver color from mahogany (Figure 2), to that which characterized by enlarged muddy or even to yellowish discoloration, with friable consistency and sub capsular hemorrhages (Figure 3). The addition of Mycofix® plus to the diet of broilers in treatment group 4, was effective in restoring the normal red brown liver color to that of chicks in treatment group 1 (Figure 4).

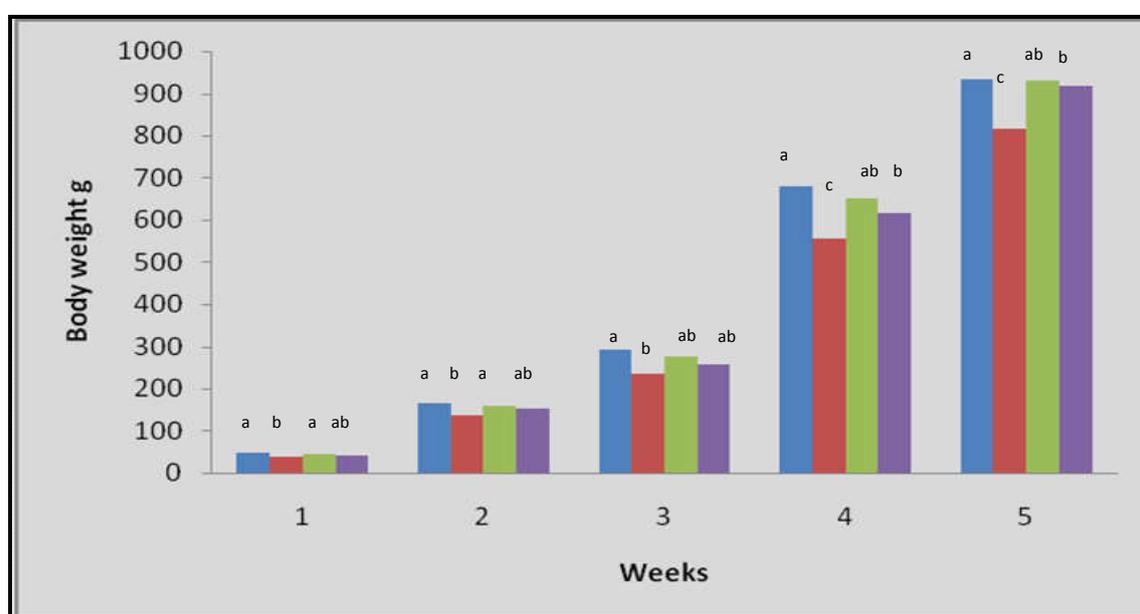
**Table (1): Effect of dietary AF with and without the addition of Mycofix® plus on BW in broiler chickens<sup>1</sup>**

Treatment	Dietary AF or Mycofix		Days of exposure to the experimental diets					
	AF ppm	MY %	0	7	14	21	28	35
G1	0	0	43.0± 0.9 <sup>a</sup>	89.6± 3.8 <sup>a</sup>	254.7± 16.2 <sup>a</sup>	547.4± 18.2 <sup>a</sup>	1128.3± 24.6 <sup>a</sup>	2061± 28.6 <sup>a</sup>

G2	2.5	0	43.4± 0.5 <sup>a</sup>	82.6± 5.0 <sup>b</sup>	218.4± 24.8 <sup>b</sup>	453.8± 44.4 <sup>b</sup>	1008.4± 51.0 <sup>c</sup>	1824.5± 57.0 <sup>c</sup>
G3	0	0.25	43.8± 0.7 <sup>a</sup>	88.1± 2.8 <sup>a</sup>	246.1 ±16.6 <sup>a</sup>	521.0± 21.8 <sup>ab</sup>	1171.2± 26.8 <sup>ab</sup>	2101.6± 43.0 <sup>a</sup>
G4	2.5	0.25	43.2± 0.3 <sup>a</sup>	84.5± 3.8 <sup>ab</sup>	236.9± 21.6 <sup>ab</sup>	495.4± 22.4 <sup>ab</sup>	1110.0± 56.8 <sup>b</sup>	2027.5± 71.2 <sup>ab</sup>

a,bMeans within a column lacking a common superscript differ (P <0.05).

<sup>1</sup>Values are means ± SEM of two replicates per treatment group (20 birds).



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Figure (1): Effect of dietary AF with and without the addition of Mycofix® plus on BWG in broiler chicken.<sup>1</sup>

**Table (2): Performance of broiler chickens fed diets contaminated with AF with and without the addition of Mycofix<sup>1</sup>**

Treatment	Dietary DAS or Mycofix		Gain (g) 1 to 35 d		Feed intake (g)		Feed:gain ratio
	AF ppm	MY %	Mean ±SEM	% of control	Mean ±SEM	% of control	g/g
G1	0	0	933.2± 14.3 a	100	2141.9± 51.8 <sup>a</sup>	100	2.29± 0.02 a
G2	2.5	0	816.1± 28.5 c	87.45	2038.5± 56.7 <sup>b</sup>	95.17	2.49± 0.04 b
G3	0	0.25	930.4 ±21.5 a	99.69	2181.4± 66.7 <sup>a</sup>	101.81	2.33± 0.03 ab
G4	2.5	0.25	917.5 ±24.2 ab	98.31	2128± 86.5 <sup>a</sup>	99.35	2.32± 0.05 ab

a–cMeans within a column lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means ±SEM of two replicates per treatment group (20 birds).

**Table (3): Effect of MY on liver lesions of broiler chicks fed diets containing 2.5 ppm AF**

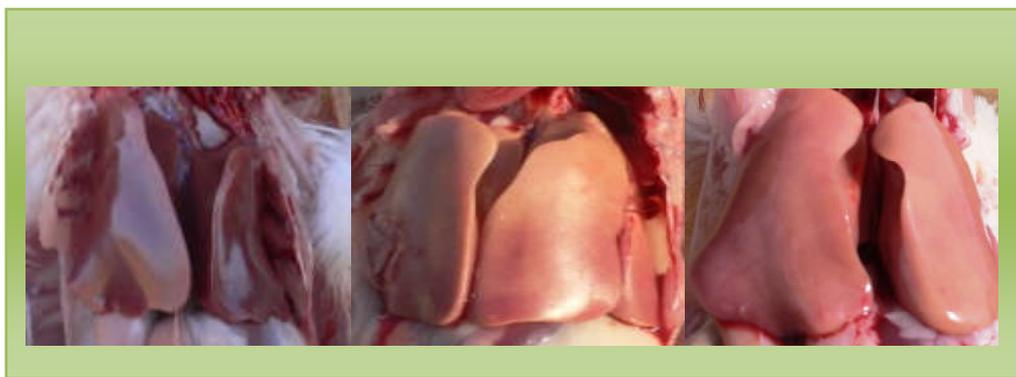
Treatments	Liver score lesions		
	No. with lesion)	Score	%
G1	5(5)	-*	0%
G2	5(1)	+	20%
	5(2)	++	40%
	5(2)	+++	40%
G3	5(5)	-	0%
G4	5(3)	-	0%
	5(2)	+	20%

\*: no lesion; +:congested liver;+ +: fatty change in liver with muddy or yellowish discoloration;

+++: liver with yellowish discoloration and sub capsular hemorrhage.

**Relative internal organs weight:**

At 35 days of broilers age, Liver, kidney, heart and spleen relative weights were significantly ( $p \leq 0.05$ ) enlarged, while bursa of Fabricious and thymus were decreased with no change in the weight of proventriculus ,gizzard and pancreas when AF was added to the broiler diet in G2, compared with G1. Amending broiler diets with the detoxifying Mycofix ® plus (G4), was effective in ameliorating( $p \leq 0.05$ ) the negative AF effect on these organs , when compared with G2. Addition of Mycofix ® plus alone (G3) had no negative effect on the above mentioned organs.(Tables 4,5 &6).



**Figure 2: Liver of broiler in treatment 1, fed no AF or. Mycofix ® plus. Normal, red or mahogany liver of broilers at 35 days of age.**

**Figure 3: Liver of broiler in treatment 2, fed AF , Extreme pale , yellowish greasy liver (fatty change), large and fragile of broilers at 35 days of age**

**Figure 4: Liver of broiler in treatment 4, fed AF and Mycofix ® plus Normal, red brown liver of broilers at 35 days of age.**

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Table (4): Effect of MY on relative liver, heart and kidney weights of broiler chicks fed diets containing 2.5 ppm AF <sup>1</sup>

Treatment	Dietary AF or Mycofix		Liver	Heart	Kidney
	AF ppm	MY %			
G1	0	0	2.50± 0.25 <sup>a</sup>	0.59±0.014 <sup>a</sup>	0.60 ±0.14 <sup>a</sup>
G2	2.5	0	3.99± 0.70 <sup>c</sup>	0.82± 0.005 <sup>b</sup>	1.00± 0.05 <sup>b</sup>
G3	0	0.25	3.05±0.74 <sup>ab</sup>	0.67 ±0.008 <sup>ab</sup>	0.62 ±0.11 <sup>a</sup>
G4	2.5	0.25	3.23±0.90 <sup>ab</sup>	0.70± 0.09 <sup>ab</sup>	0.71 ±0.05 <sup>a</sup>

a–cMeans within a column lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means ± SEM of two replicates per treatment group (20 birds).

Table (5): Effect of MY on relative proventriculus, gizzard and pancreas weights of broiler chicks fed diets containing 2.5 ppm AF <sup>1</sup>

Treatment	Dietary AF or Mycofix		Proventriculus	Gizzard	Pancreas
	AF ppm	MY %			
G1	0	0	2.69± 0.58 <sup>a</sup>	0.62±0.09 <sup>ab</sup>	0.39 ±0.06 <sup>a</sup>
G2	2.5	0	2.62± 0.61 <sup>a</sup>	0.85± 0.01 <sup>b</sup>	0.61± 0.04 <sup>ab</sup>

G3	0	0.25	2.60±0.53 <sup>a</sup>	0.55 ±0.05 <sup>a</sup>	0.42 ±0.06 <sup>a</sup>
G4	2.5	0.25	2.70±0.49 <sup>a</sup>	0.68± 0.07 <sup>ab</sup>	0.40 ±0.03 <sup>a</sup>

a–c Means within a column lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means  $\pm$  SEM of two replicates per treatment group (20 birds)

**Table (6): Effect of MY on relative organ weights of broiler chicks fed diets containing 2.5 ppm AF<sup>1</sup>**

Treatment	Dietary AF or Mycofix		Thymus	Bursa of Fabricious	Spleen
	AF ppm	MY %			
G1	0	0	0.13± 0.003 <sup>a</sup>	0.30±0.007 <sup>a</sup>	0.18 ±0.005 <sup>a</sup>
G2	2.5	0	0.08± 0.001 <sup>b</sup>	0.24± 0.004 <sup>b</sup>	0.25± 0.001 <sup>b</sup>
G3	0	0.25	0.12±0.002 <sup>ab</sup>	0.29 ±0.007 <sup>a</sup>	0.18 ±0.003 <sup>a</sup>
G4	2.5	0.25	0.11±0.004 <sup>ab</sup>	0.27± 0.007 <sup>ab</sup>	0.16 ±0.002 <sup>a</sup>

a–c Means within a column lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means  $\pm$  SEM of two replicates per treatment group (20 birds).

**Blood profile:**

The effect of feeding diet contaminated with AF to broiler chickens at a rate of 2.5 ppm (G2) was responsible for significant ( $p \leq 0.05$ ) increase in the WBCs, but in the opposite manner was lowered erythrocytes, hemoglobin and erythrocyte sedimentation rate, when compared with the control group (G1). Amending the AF contaminated diet with Mycofix ® plus (G4), was effective in counteracting ( $p \leq 0.05$ ) the negative effect of

AF in the above mentioned blood parameters compared with AF alone (G2).The addition of Mycofix ® plus alone (G3) had no negative effect on blood profile (Table 7).

**Table (7): Effect of MY on blood parameters of broiler chicks fed diets containing 2.5 ppm AF <sup>1</sup>**

Treatment	Dietary AF or Mycofix		WBCs (10 <sup>3</sup> /mm <sup>3</sup> )	RBCs (10 <sup>6</sup> /mm <sup>3</sup> )	HB (g/100ml)	ESR %
	AF ppm	MY %				
G1	0	0	25.80± 1.78 a	2.43 ±0.03 a	30.40± 1.85 a	19.40± 1.59 a
G2	2.5	0	35.40± 2.00 c	2.13 ±0.01 b	11.60 ±2.07 c	15.76± 0.65 b
G3	0	0.25	24.20± 2.28 a	2.27± 0.02 ab	25.80 ±1.94 ab	18.9 ±0.58 a
G4	2.5	0.25	28.00 ±7.03 ab	2.26 ±0.03 ab	23.00 ±1.00 ab	17.48± 0.17 ab

a–c Means within a column lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means ± SEM of two replicates per treatment group (20 birds)

**Blood biochemistry:**

Biochemical parameters of broilers at 35 days of age after feeding diet contaminated with AF to broiler chickens at a rate of 2.5 ppm (G2) was responsible for significant ( $p \leq 0.05$ ) decrease in the total protein, glucose, triglycerides and cholesterol, when compared with the control group (G1). Addition of Mycofix ® plus (G4) to the AF contaminated diet, was effective in ameliorating ( $p \leq 0.05$ ) the negative effect of AF in the above mentioned blood biochemical parameters compared with AF alone (G2).The addition of Mycofix ® plus alone (G3) had no negative effect on blood profile (Table 8).

**Serum enzymes:**

Serum enzymes of broilers fed AF at a rate of 2.5 ppm (G2) was responsible for significant ( $p \leq 0.05$ ) increase in ALT, AST and ALP enzymes, when compared with the control group (G1). Inclusion of Mycofix<sup>®</sup> plus (G4), to the AF contaminated diet was effective in alleviating ( $p \leq 0.05$ ) the negative effect of AF in the above mentioned serum enzymes compared with AF alone (G2). The addition of Mycofix<sup>®</sup> plus alone (G3) had no negative effect on blood profile (Table 9).

**Table (8): Effect of MY on blood biochemistry of broiler chicks fed diets containing 2.5 ppm AF<sup>1</sup>**

Treatment	Dietary AF or Mycofix		Total protein Mg/100 ml	Glucose Mg/100 ml	Triglycerides Mg/100 ml	Cholesterol Mg/100 ml
	AF ppm	MY %				
G1	0	0	3.34± 0.42 a	282.14 ±29.85 a	106.02± 13.72 a	217.14± 90.77 a
G2	2.5	0	2.35± 0.28 b	168.00 ±15.92 c	62.70 ±12.09 b	145.30± 30.18 b
G3	0	0.25	3.30± 0.22 a	259.58± 21.40 ab	101.34 ±13.05 a	198.66 ±63.15 a
G4	2.5	0.25	2.67 ±0.40 ab	244.20 ±36.44 ab	76.38 ±9.53 ab	178.88± 14.13 ab

a–cMeans within a column lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means ± SEM of two replicates per treatment group (20 birds)

**Table (9): Effect of MY on serum enzymes of broiler chicks fed diets containing 2.5 ppm AF<sup>1</sup>**

Treatment	Dietary AF or Mycofix		ALT (IU/L)	AST (IU/L)	ALP (IU/100ml)
	AF ppm	MY %			
G1	0	0	20.80 ±0.68 a	31.95 ±2.15 a	52.59± 4.03 <sup>a</sup>
G2	2.5	0	36.40± 1.22 b	55.18± 2.90 c	90.96 ±1.65 <sup>b</sup>
G3	0	0.25	20.80± 2.25 a	30.63± 6.74 a	54.78 ±2.93 <sup>a</sup>
G4	2.5	0.25	20.60 ±0.91 a	39.86 ±9.33 ab	63.53 ±1.87 <sup>a</sup>

a–cMeans within a column lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means  $\pm$  SEM of two replicates per treatment group(20 birds)

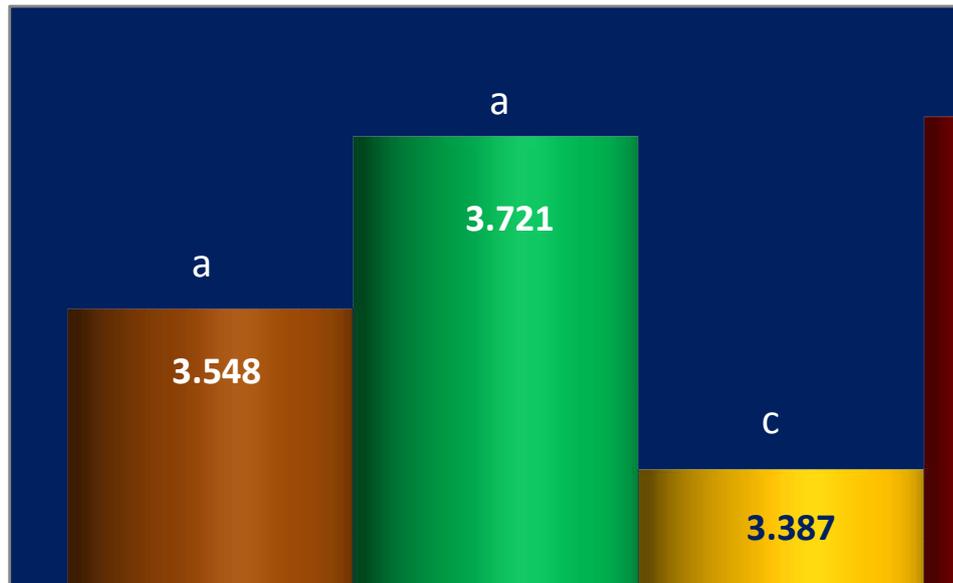
#### Newcastle antibody titer:

The means of antibody titer (HA) against Newcastle disease (ND) showed significant ( $P > 0.05$ ) difference between treatments when analyzed at 35 days of the trial (Figures 5&6 ). Aflatoxin had detrimental ( $p < 0.05$ ) effect in reduction of Log<sub>10</sub>ND ELISA mean antibody titer of 3.387 with % CV of 74.92, compared with the control treatment (G1), which had Log<sub>10</sub>ND titer of 3.741 with % CV of 36.69. A significant restoring of Log<sub>10</sub>ND antibody titer to those of control group (G1) was recorded by addition of Mycofix ® plus to the feed of broilers in (G4) to be 3.458 with % CV of 38.74 , compared with the AF fed T2 group.

## DISCUSSION

The results from the present study are in agreement with other studies where significantly reduced body weights were observed when birds were exposed to dietary AF (400 ppb, 750 ppb). The depression in growth upon feeding AF was attributed to reduced protein and energy utilization (Dalvi and Ademoyero, 1984; Verma *et al.*, 2002) which impaired nutrient absorption and reduced pancreatic digestive enzyme production (Osborne and Hamilton, 1981) and consequently reduced appetite (Sharline *et al.*, 1980) and above, and these depression in body weight in toxin fed groups were reported to be dose dependent (Beura *et al.*, 1993). Similarly, significant depressions in body weight gain were also recorded in broilers given diets containing 1 and 2 mg/kg of AF (1000 to 2000 ppb) at 4 and 7 weeks of age and in a dose-dependent manner (Huff and Doerr, 1981; Nandakumar *et al.*, 1984; Johri and Majumdar, 1990). With the line of our results, reduced feed intake and poor feed efficiency in broilers has also been reported in birds fed diets containing AF at 2, 4 and 6 weeks of age when level of dietary AF was higher than 100 ppb (Nandakumar, 1984). These authors have suggested that the reduced appetite during aflatoxicosis could be due to impaired liver metabolism caused by the liver damage, as seen in our study by the significant increase in the liver score lesion (Table 2& Figure 3 ). It is likely that broiler chicks may highly respond to AF in diet as their physiological needs and capacity to absorb is higher compared to older birds. Gabal and Azzam, (1998), suggested that prolonged administration of AF at the low levels may cause relevant lesions in liver and renal tissues. Moreover, the metabolism of broilers seems to be more adapted to high concentrations of aflatoxin in the feed when administered to 35 d of age, when compared with data reported from similar experiments conducted with broilers aging 1 to 21 d and with other species such as turkey poults (Giambrone, 1985).

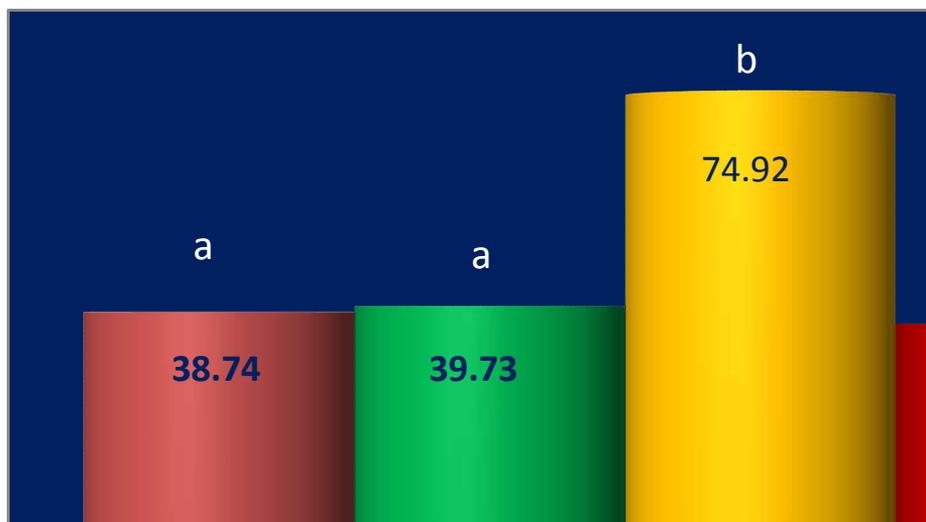
The effect of AF on the relative internal organs weight of broilers fed AF at a rate of 2.5 ppm by significant changes in liver, spleen, kidney, bursa of Fabricious, went in the same line with what was founded by (Ibrahim *et al.*, 1997). The increased liver size and its yellowish color and creasy consistency in broilers fed AF could be attributed disturbance in the formation of the proteins responsible for transportation of fat from the liver to other tissues especially low density



**Figure (5): Effect of MY on ND antibody titer of broiler chicks fed diets containing 2.5 ppm AF<sup>1</sup>**

a–c Means lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means of two replicates per treatment group (20 birds)



**Figure 6: Effect of MY on CV% OF ND antibody titre of broiler chicks fed diets containing 2.5 ppm AF<sup>1</sup>**

a–b Means lacking a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>Values are means of two replicates per treatment group (20 birds)

Lipoproteins”LDL”. It is well known that liver is the main target organ for the effect of AF even at low concentration (0.5 ppm) ( Bryden , *et al.*, 1979).

Hemolytic anemia caused by AF is one of the factors of heterophilia noticed in G2 birds and in the same time may be the principle factor in spleen enlargement of chicks fed AF toxin , which is agreed with (Jargees, 2007). The protective effect of MY against the negative AF effects on blood profile, biochemistry and enzymes is its role as a chelating agent in the sequestration of the AF through the gastrointestinal tract, preventing them from absorption, and so help in getting rid of these toxins from the body outside the body. Adsorption starts in the oral cavity during salivation and continues in stomach and gut. The fixed mycotoxin being unable to enter the blood and subsequently excreted in feces.

Immunosuppression induced by feeding AF is referred by many investigators (Ibrahim *et al.*, 2000), that results from involution of the lymphatic organs ( thymus and bursa of fabricious), and reduction in Log10 ND titers. The sensitivity of the immune system to mycotoxin-induced immunosuppression arises from the vulnerability of the continually proliferating and differentiating cells that participate in immune mediated activities and regulate the complex communication network between cellular and humeral components (Shivachandra *et al.*, 2003). AF was reported to inhibits the histological development and functional maturation of lymphoid organs .Morphological evidence to explain the immunosuppressive effects of AF was documented . in broiler chickens from 1 week to 7 weeks of feeding 20,40,60,80 and 100 ppb and the major signs were reduction in the weights of lymphoid organs including bursa of Fabricius, spleen and thymus (Arulmozhi and Varghese, 2011).

Means of antibody titer with (ELISA) against Newcastle disease (ND) showed differences ( $P>0.05$ ) between treatments when analyzed at 35 day of the trial (Figure 5). The presence of AF in the feed is reported to decrease vaccinal immunity and may therefore lead to the occurrence of disease even in properly vaccinated flocks (Ibrahim *et al.*, 2000; Sadeghi *et al.*, 2012). Aflatoxins have been associated to have immunosuppressive effect due to direct inhibition of protein synthesis, including those with specific functions such as immunoglobulin’s IgG, IgA, inhibition of migration of

macrophages, interference with the hemolytic activity complement, reduction in the number of lymphocytes through its toxic effect on the Bursa of Fabricius and impairment of cytokines formation by lymphocytes (Giambron *et al.*, 1978 a, b). In present study, significant difference ( $P>0.05$ ) in ELISA titres was observed when treatment with AF (treatment 2) was compared with all other treatment groups suggests that birds exposed to 2.5 ppm AF in diet did show signs of immunosuppression.

Mycofix® plus, is one of the adsorbent that can be added in poultry feed and is claimed to neutralize moderate levels of aflatoxin (up to 2500-3500 ppb) in poultry feed. The counteracting effect of AF by Mycofix® plus on the growth performance parameters, organs lesion scores, blood profile, biochemistry, and serum chemistry, relative internal organs of these the lymphoid organs and ND antibody ELISA titers. Adsorption starts in the oral cavity during salivation and continues in stomach and gut. The fixed mycotoxin being unable to enter the blood and subsequently excreted in faces. The beneficial effect of Mycofix® plus in ameliorating the negative effect of AF on ND antibody titers in this study is attributed to the excellent contents of this detoxifier.

## تأثير (mycofix® plus) وسم الافلا على صحة وأداء فروج اللحم

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### الخلاصة

أجريت هذه الدراسة في قسم الصحة العامة البيطرية ، كلية الطب البيطري /جامعة الموصل / العراق ، خلال المدة من ايلول إلى تشرين الاول ٢٠٠٩ لمعرفة تأثير المايكوفكس (MY) Mycofix® plus على الأداء الصحي والانتاجي لفروج اللحم أثناء تعرضه لسم الافلا ، اذ تم توزيع ذكور فروج اللحم (روس ٣٠٨) بعمر يوم واحد إلى ٤ مجاميع وبواقع مكررين (١٠ افراخ لكل منهما). تم تربية الافراخ لمدة ٣٥ يوماً وغذيت مع أو بدون سم الافلا (AF) او المايكوفكس (MY). وزعت مجاميع الافراخ على النحو التالي: المجموعة الاولى :صفر جزء في المليون سم الافلا (AF) وخالية من المايكوفكس ؛ المجموعة الثانية : 2.5 جزء في المليون سم الافلا (AF) وخالية من المايكوفكس ؛ المجموعة الثالثة : ٠.٢٥ % مايكوفكس (MY) وخالية من سم الافلا ؛ المجموعة الرابعة 2.5

جزء في المليون سم الافلا (AF) و ٠.٢٥ % مايكوفكس . تم تسجيل وزن الجسم واستهلاك العليقة أسبوعياً. وفي ٣٥ يوم من عمر الطيور تم اختيار خمس طيور عشوائياً من كل مكرر للمجاميع الاربعة وذلك لتقدير الاعداد الكلية لخلايا الدم الحمراء ، وخضاب الدم ( HB ) وسرعة ترسيب كريات الدم الحمراء ( ESR ) ، ومستوى الجلوكوز و البروتين الكلي و الكوليسترول و والدهون الثلاثية و مستويات الانزيمات AST ، ALT و ALP. و قياس معيار الأجسام المضادة ومعامل اختلافها ضد لقاح مرض نيوكاسل في مصل الدم . كما واخذت الأوزان النسبية لجراب فابريشيا ، الغدة الصعترية ، الطحال ، المعدة الغدية ، القانصة ، الكلى ، الكبد ، القلب والبنكرياس اضافة إلى تسجيل آفات الكبد. اشارت النتائج إلى أن المايكوفكس (MY) كان فعالاً في تقليل التأثير السلبي لسم الافلا على أداء النمو لافراخ فروج اللحم (زيادة وزن الجسم واستهلاك العليقة ونسبة تحويلها) وعلى الأوزان النسبية لأعضاء الجسم الداخلية المتأثرة و آفات الكبد وكذلك على صورة الدم ( WBCs ، HB والجلوكوز) وانزيمات المصل (AST، ALT و ALP) ومعيار الأجسام المضادة ومعامل اختلافها ضد لقاح مرض نيوكاسل مقارنة مع المجاميع المستهلكة لسم الافلا .

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