SOME BIOCHEMICAL CHANGES IN BROILER CHICKENS DURING AFLATOXICOSIS AND THEIR REDUCTION BY USING ACTIVATED CHARCOAL

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Abstract

This study was conducted to follow up of broiler chickens during aflatoxicosis by feeding them with dietary different concentrations of aflatoxin B1 (AFB1) for a period of four weeks from 0 to 28 days and their subsequent recovery two weeks after withdrawal of contaminated food, in addition to evaluate the effectiveness of charcoal in alleviating aflatoxicosis. One hundred and twenty broiler chickens of one day old were divided into six groups each of twenty chickens. The 1st group received a standard ration and kept as control group. The 2nd and 3rd groups received a diet contaminated with 0.5 mg and 1 mg AFB1 / kg ration, respectively. The 4th group received standard ration + 0.5 gm charcoal / kg ration. The 5th and 6th groups received diet contaminated with 0.5 and 1 mg AFB1/kg ration, respectively + 0.5 gm charcoal /kg ration for four weeks then all groups received standard ration for another two weeks as a clearance period. Chickens in all groups were weighed weekly and feed consumption for each group was calculated. Blood and tissue samples were collected from each group at the end of 2nd, 4th and 6th week.

Results of the experiment revealed that AFB1 in both doses induced dose-dependent reduction in body weight and feed consumption. Inclusion of activated charcoal in the diet of chickens received AFB1-contaminated ration resulted in moderate improvement in body weight and feed consumption of the 5th group (0.5mg AFB1/kg ration), while, no benefits were seen within chicks of the 6th group received higher level of AFB1 (1mg AFB1/kg ration).

Compared with the control group AFB1 elicited a significant dose-dependent increase in the activity of AP, GGT, LDH enzymes and uric acid with a significant decrease in cholesterol, triglycerides, calcium and inorganic phosphorus levels. Addition of activated charcoal

induced a significant improvement in enzymes, calcium, phosphorus and uric acid only in the 5th group.

Regarding the histopathological results, examination of the internal organs, sections revealed typical lesions of aflatoxicosis as the intensity and the persistence of the lesions related to the dose of the AFB1. Addition of activated charcoal to AFB1-contaminated ration moderately decreased the severity of the pathological lesions.

The body weight of the chickens, feed consumption and most of the studied biochemical parameters were improved during the clearance period and some of them returned to almost their normal level two weeks after AFB1 withdrawal.

These findings suggest that the AFB1 caused many alterations in the growth performance, and the biochemical parameters, which are confirmed by many pathological changes in the internal organs of the chickens. Addition of activated charcoal to AFB1-contaminated ration was marginally effective in alleviating some of the toxic effects associated with aflatoxicosis, but was of little benefits when highly contaminated ration with AFB1 was fed to growing broiler chickens.

INTRODUCTION

The contamination of animal feed with mycotoxins represents a worldwide problem for farmers. These toxins originate from molds whose growth on living and stored plants is almost unavoidable particularly under moist conditions. Mycotoxin-containing feed can cause serious diseases in farm animals resulting in suffering and even death and thus can cause substantial economic losses (Alexander *et al.*, 2001).

Aflatoxins (AF) are secondary metabolites produced by strain *Aspergillus flavus* and *Aspergillus parasiticus* and occur as natural contaminants of poultry feeds (Edds and Bortell, 1983). Four types of AF are produced: AFB1, AFB2, AFG1 and AFG2. Avian species especially chickens, duckling and Turkey poults are most susceptible to AFB1 toxicity . Signs of aflatoxicosis in poultry include anaemia, inhibition of the immune function, hepatotoxicosis, mutagenesis, teratogenesis, carcenogenesis, anorexia, haemorrhage, poor food utilization, decreased weight gain and susceptibility to environmental and microbial stresses. For these reasons, AF are a potential threat to poultry health and can cause severe economic losses in the poultry industries (Edds and Bortell, 1983).

Removing performed AF from contaminated foodstuffs remains a major problem. Large scale, practical and cost-effective methods for detoxifying AF-containing foodstuffs are not currently available. A variety of physical, chemical and biological methods for detoxifying AF have been employed with limited success . One approach has been to use non-nutritive adsorbing materials in the diet in order to bind AF and reduce its absorption from the gastrointestinal tract (Jindal *et al.*, 1994).

The use of activated charcoal as an oral antidote for the treatment of poisonings is well established. Charcoal acts as insoluble carrier that non-specifically adsorbs molecules, thereby preventing their absorption (Edrington *et al.*, 1996).

The objects of our research were to extend the description of aflatoxicosis in broiler chickens and to follow the recovery of birds after aflatoxin removal from the diet concerning the changes in the biochemical parameters and the pathological findings in the internal organs in order to find the most sensitive and useful measures for diagnosis. This investigation was also done to evaluate the prophylactic effectiveness of activated charcoal when included in a diet containing AF and fed to growing broiler chickens.

MATERIALS AND METHODS

1- Experimental chickens

One hundred and twenty chickens, one-day-old commercial Hubbard broiler chickens were used. The chickens were reared under standard hygienic conditions and fed on a balanced commercial ration. All chickens were vaccinated against Newcastle disease at 7 and 18 day old and Gumboro disease at 21 day old .

These chickens were equally divided into 6 groups each of twenty. The 1st group received a standard ration and kept as control group. The 2nd and 3rd groups received a diet contaminated with 0.5 mg and 1 mg AFB1 / kg ration, respectively. The 4th group received standard ration + 0.5 gm charcoal / kg ration. The 5th and 6th groups received diet contaminated with 0.5 and 1 mg AFB1/kg ration, respectively + 0.5 gm charcoal/kg ration for four weeks, then all groups received standard ration for

another two weeks as a clearance period. Five chickens from each group were sacrificed for histopathological study after collection of blood samples via heart puncture at the end of 2nd, 4th and 6th week of the experiment.

2- Aflatoxin

Aflatoxin was produced by growing *Aspergillus flavus* (standard toxigenic strain) on corn meal according to the method of Merwe *et al.* (1963). Identification and quantitative estimation of aflatoxin presented in corn meal were done by thin layer chromatography . The prepared corn meal containing aflatoxin was mixed with ration to provide a final concentration of 0.5 and 1mg AFB1/ kg ration.

3- Neo-carbotrina

Manufactured by Arab drug company, Cairo A.R.E.

Composition:

Each tablet contains:

Diiodohydroxyquinoline	0.125 g
Phthalylsulphathiazole	0.125 g
Activated Charcoal	0.100 g
Extract belladonna	0.010 g

According to the severity of aflatoxicosis the appropriate dosage is recommended as it ranged from 0.2-0.5 g charcoal/ kg ration (Jindal *et al.*, 1994).

4- Body weight and feed consumption

Chickens in all groups were weighed weekly, feed consumption for each group was calculated daily during the experimental period, and mortality rates of each group were recorded.

5- Blood sampling

Five ml of blood were collected from 5 birds from each group via heart puncture. The blood samples were collected into centrifuge tubes, left to clot at room temperature and then centrifuged at 3000 r.p.m. for 5 minutes. The separated sera were subjected to biochemical analysis.

6- Biochemical studies

Serum Alkaline phosphatase (AP) activity was determined according to Rec (1972). Gammaglutamyl transferase (GGT) activity was measured according to Szaz (1969). Lactic dehydrogenase (LDH) was estimated according to Burd and Usategui-Gomez (1973). Serum cholesterol concentration was measured according to Finley *et al.* (1978). Serum triglycerides concentration was measured according to Wohlefeld (1974). Serum calcium and inorganic phosphorus were determined according to Tietz (1970) and Young *et al.* (1975), respectively. Serum uric acid was measured according to the method described by Fossati (1980).

7- Histopathological studies

The sacrificed chickens were subjected to post-mortem examination, specimens were collected from liver, kidney and heart, then fixed in 10% neutral formalin and embedded in paraffin wax. Sections of five microns thickness were prepared, stained by haematoxylin & eosin and examined microscopically (Lillie and Fulman, 1976).

8- Statistical analysis

The data obtained from this investigation were statistically analysed by student's "t" test according to Snedecor and Cochran (1980).

RESULTS AND DISCUSSION

Mycotoxins, particularly AF have been reported to produce severe economic losses and health problems in the poultry industry. Diagnosis is rather difficult and medical treatment may be almost impossible in aflatoxicosis cases of poultry. The most prevalent indicator of chronic aflatoxicosis is reduced growth rate. In the present study, feeding 0.5mg and 1 mg AFB1/kg ration for four weeks elicited dose-dependent decrease in body weight, weight gain and feed consumption as shown in Tables 1&2. The same results were also reported by Dalvi and McGowan (1984), Aravind *et al.* (2003) and Kim *et al.* (2003).

The exact mechanism by which AF impairs growth is unknown, but it is probably multifactorial, involving disturbances in carbohydrate, lipid and protein metabolism,

metabolic interaction with the toxin and disturbance in hormones. Additionally, poor appetite and reduced feed intake may partially account for reduced performance (Edrington *et al.*, 1994). Recently, Nasr-El-Deen (2002) added that, the recorded loss in body weight may be a reflection of reduced feed intake or decreased utilization and metabolism of food stuff due to intestinal and hepatic lesions.

When activated charcoal was included in the diet of chickens received 0.5 mg AFB1/kg ration, it elicited a marked improvement in body weight than those chickens received the same dose of AF alone and achieved body gains that were not significantly different from controls when examined over the experimental period. However, activated charcoal failed to improve the body weight in the 6^{th} group which received higher level of AF (1 mg AFB1/kg ration) as their body weights were still significantly less than those of the control and improved only during the last two weeks of the experiment after withdrawal of the toxins from the ration. Feed intake and body weight gain were slightly improved by the addition of activated charcoal compared with chickens fed AFB1 alone as shown in Table 2. Dalvi and McGowan (1984), Jindal *et al.* (1994) and Nasr-El-Deen (2002) reported a trend in improvement in body weight and feed intake of chickens when activated charcoal was added to AF-contaminated diet. However, Kubena *et al.* (1990) and Edrington *et al.* (1996) examined activated charcoal for the ability to alleviate chronic aflatoxicosis in growing broilers and turkey poults and reported no improvement in chick or poult performance. The reason of these conflicting results may be due to differences in type or physical characteristics of the activated charcoal, duration of feeding, composition of basal diet, concentration of AF fed and poultry species.

Feeding chickens with diet contaminated with 0.5 and 1mg AFB1/kg ration for 4 weeks, resulted in 10% and 25% mortalities, respectively (as recorded in Table 2). Addition of activated charcoal inhibited the mortalities among chickens received 0.5mg AFB1 while, reduced the mortalities to 10% in chickens given higher level of AFB1 (1 mg) compared to 25% in chickens fed the same dose of AFB1 alone. All mortalities occurred during the toxification period and no mortalities were recorded during clearance period in all groups. Nearly, similar results were obtained by Bonna *et al.* (1991) who studied the efficacy of activated charcoal in reducing the toxicity of dietary

aflatoxin to mink and reported that addition of charcoal to the diet containing 102 ppb AFB1 reduced mortality and increased the survival time of the mink.

Serum enzymatic evaluation revealed a significant rise in alkaline phosphatase, gammaglutamyl dehydrogenase and lactate dehydrogenase activities throughout the aflatoxin-feeding period (Table 3). It is well established that the elevation in AP may be due to the effect of toxin on bile duct and bone as a result of hypocalcaemia. Our results were also consistent with those reported by Jindal *et al.* (1994), Nasr-El-Deen (2002) and Yousef *et al.* (2003).

GGT is an enzyme for which an increase in activity indicates biliary cholestasis and hyperplasia in bile ducts (Kramer, 1989). This elevation coincided with the degree of hyperplasia of bile ducts observed in the liver of chickens. GGT increase was dependent on dose, time and severity of liver lesions. When the aflatoxin was withdrawn from the diet, hyperplasia of bile ducts and GGT levels decreased. Similar results were also reported by Fernandez *et al.* (1994), Aravind *et al.* (2003) and Kim *et al.* (2003).

Serum total LDH levels are the results of isoenzymes from different sources. In birds, the high levels were found in heart, liver and kidneys (Coles, 1986). LDH levels were elevated gradually with dose intake of AF and with the hepatomegaly observed at chickens necropsy. Fernandez *et al.* (1994) believed that the increase in LDH levels was due to an isoenzyme of hepatic origin. Similar results were also recorded by Kim *et al.* (2003) who found a significant increase in LDH activity in laying hens after feeding with a diet contaminated with 500 ppb of AFB1.

The changes in concentration of serum enzymes indicate that continued administration of aflatoxin caused severe cellular damage leading to increase cellular permeability, which in turn results in the release of enzymes into the serum (Shukla and Pachauri, 1995).

In this study, we recorded a significant decrease in serum cholesterol and triglycerides in chicken given both doses of AF-contaminated diet (Table 4) which is consistent with the general reduction of lipogenesis , impaired lipid transport and

specific inhibition of hepatic cholesterol biosynthesis by AF (Donaldson *et al.* 1972). These results were supported by the findings of Shukla & Pachauri (1995) and Raju & Devegowda (2000) who found a significant decrease in cholesterol level in broiler chickens after feeding of 0.3 mg AFB1/kg ration for 35 days and suggested that aflatoxin disrupted lipid biosynthesis and transport, due to hepatic damage.

The decrease in calcium and phosphorus concentration observed in this study, as shown in Table 5, has several practical implications. The low calcium level was a sensitive indicator of intoxication. Also, serum phosphorus levels decreased in chicken and were found to be related to the dose and time of exposure. A renal lesion was observed during the intoxication period, where calcium and phosphorus may have been lost. Glahn *et al.* (1991) found that aflatoxin in broiler chicken caused severe renal lesion, which affected calcium and phosphorus metabolism. They added that aflatoxin might directly alter the renal, intestine and parathyroid regulation of calcium and inorganic phosphorus. Hypocalcaemia observed in this work was in agreement with that found by Fernandez *et al.* (1994), Shukla & Pachauri (1995), Nasr-El-Deen (2002) and Kim *et al.* (2003). Also, the significant decrease in serum inorganic phosphorus was reported in other studies (Shukla & Pachauri 1995, Edrington *et al.*, 1997, and Kim *et al.*, 2003).

Uric acid is the primary catabolic product of protein and non-protein nitrogen in birds. Hyperurecemia in birds occurs with starvation, gout, massive tissue destruction and renal diseases (Coles, 1986). In the present study, AFB1 in both doses induced a significant increase in uric acid along the experimental period. These results agreed with those reported by Dawoud *et al.* (2002) and Nasr El-Deen (2002) who reported that, the significant increase in uric acid suggests that kidney function was severely impaired.

The ability of charcoal to reduce the biochemical alterations caused by different concentrations of aflatoxin was evaluated in our investigation. The results revealed that the addition of charcoal to the AF-free diet did not alter the biochemical parameters compared to control. Incorporation of charcoal in AF-contaminates ration elicited an improvement in the enzymatic activity, cholesterol and triglycerides levels and

corrected the alteration of calcium, phosphorus and uric acid among chickens received 0.5mg AFB1/kg ration compared with chickens fed AF alone. Meanwhile, charcoal failed to improve the studied biochemical parameters in chickens fed high level of AF as they still significantly differ than those of control one. In another reports, Kubena *et al.* (1990) and Edrington *et al.* (1996) reported no improvement in GGT or LDH activity nor in cholesterol or triglycerides when charcoal was added to AF-contaminated diet in broiler chickens. Jindal *et al.* (1994) reported a significant improvement in calcium and phosphorus level when activated charcoal was added to the diet compared with chickens fed with AF-contaminated diet alone. In our opinion, the clear differences between the ability of charcoal in alleviating the toxic effects of AFB1 may be due to that charcoal failed to adsorb all the amount of aflatoxin and there were free parts which caused these alterations in performance and biochemical parameters which became more marked with higher levels of AFB1 fed to the chickens.

During the clearance period, most of the studied parameters returned to their normal levels two weeks after removing the contaminated ration among the chickens consumed low level of AFB1, while, some parameters still significantly altered among chickens received higher dose of the aflatoxin. Chen *et al.* (1985) proved that, four days after withdrawal of the aflatoxin-contaminated ration, there were no detectable amounts of aflatoxin in any tissues of broiler chickens. They also added that broiler chickens rapidly clear aflatoxins from their tissues once they are transferred to an aflatoxin-free diet. Fernandez *et al.* (1994) reported that the reduced levels of cholesterol and triglycerides which observed during aflatoxicosis in chickens were significantly increased during the clearance period while, the reduced levels of calcium and phosphorus did not return to control values until 8 days after removal of the aflatoxin from the diet.

The biochemical changes occurred during aflatoxicosis in the present work were confirmed by the histopathological changes in the internal organs, which were found to be related to the concentration of the aflatoxin ingested. Macroscopically, the livers of broiler chickens of the 3rd group after 4 weeks were yellowish, friable and enlarged in size. Moreover, haemorrhages on- skeletal muscles, hydropericardium and enlarged kidney were also reported. Such findings were also observed by Rosa *et al.* (2001).

Microscopically, the lesions were dose and time dependent and ranged from mild lesions in the second group to more severe lesions in the third group which received higher level of aflatoxins. Biliary duct hyperplasia was detected only in birds exposed to 1 mg AFB1/kg ration. The same was observed with respect to the lesions in the kidney and heart, which were more evident with the highest dose of AFB1.

Livers of the third group at the end of the fourth week showed portal area highly infiltrated with leukocytic cells and hyperplasia of the bile duct (Fig. 1), vaculation of hepatic cells and fatty changes (Fig. 2), in addition to thickened capsule (Fig. 3). The detection of the newly formed bile ductules was similar to that reported by Kelly (1985) who suggested that the hyperplasia of the bile ducts is an attempt to regenerate hepatic parenchyma when the paranchymal cells have lost their capacity to regenerate themselves.

Kidneys of the third group showed oedema between the renal tubules (Fig. 4), congested blood vessels besides haemorrhage between renal tubules (Fig. 5) and degenerated cells of the renal tubules (Fig. 6).

The hearts of the same group showed oedema (Fig. 7) and leukocytic infiltration between the degenerated muscle fibers (Fig. 8).

Our results were confirmed by Tag El-Deen (1997) and Kim *et al.* (2003) who described the histopathological lesions of chickens during aflatoxicosis. They reported vaculated hepatocytes, bile hyperplasia with aggregation of inflammatory cells. The heart revealed dispersion of cardiac muscle fiber with oedema and focal areas of degenerated muscle fibers. The kidney showed degeneration and necrosis of renal tubules, with leukocytic infiltration. Addition of activated charcoal to AF-contaminated diet in the 5 th and 6th groups moderately decrease the number of affected broilers, the incidence and the severity of the pathological lesion.

After withdrawal of the ration containing aflatoxins, the gross lesions became mild to moderate or even disappeared two weeks after removing of the toxin according to the dose of the toxin consumed by the bird. Chen *et al.* (1985) studied the

pathological changes in broiler chickens during toxication with AFB1 for 35 days and their recovery on 1. 2, 4, and 8 days after replacing the contaminated diet. They reported that after withdrawal of the aflatoxin, all apparent gross lesions of aflatoxicosis disappeared, with no evidence of any lesions 8 days after replacing the contaminated diet.

It could be concluded that AFB1 caused dose-dependent alterations in the growth performance and the biochemical parameters, which are confirmed by many pathological changes in the internal organs of the broiler chickens. Most of these alterations were significantly improved two weeks after aflatoxin withdrawal. Addition of activated charcoal to AFB1-contaminated ration was marginally effective in alleviating some of the toxic effects associated with aflatoxicosis, but was of little benefits when highly contaminated ration with AFB1 was fed to growing broiler chickens.

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Table 1. body weight / gm / chicken post administration of aflatoxin (AF) and charcoal (Cc) and after clearance period, as compared with control group (mean values ± SE).

Group	Treatment	1st week	2nd week	3rd week	4th week	5th week	6th week
1	Control	127.8 ± 5.7	331 ± 7.1	592.6 ± 10.7	881.8 ± 22.2	1195 ± 15.45	1522 ± 19.3
2	AF 0.5 mg	114.2* ± 5.1	309.6* ± 9.8	546.2* ± 18.2	804* ± 22.6	1112* ± 19.4	1472 ± 18.6
3	AF 1 mg	105.6** ± 7.2	288** ± 13.1	522.6** ± 19.8	778** ± 23.4	1102** ± 20.1	1430.4* ± 23.5
4	Cc 0.5 gm	126.2 ± 2.9	328 ± 4.3	598.6 ± 10.5	888.2 ± 13.6	1182 ± 18.2	1518 ± 9.2
5	AF 0.5 mg + Cc 0.5 gm	121.6 ± 3.5	323 ± 9.3	574 ± 10.9	846.4 ± 22.7	1152 ± 19.1	1488 ± 21.2
6	AF 1 mg + Cc 0.5 gm	113.2* ± 4.8	308* ± 11.2	550* ± 12.6	822* ± 12.1	1134 ± 18.6	1462 ± 19.2

* significantly different from control at P<0.05 ** Highly significant at P<0.01]

Table 2. Mortality %, body weight gain and feed consumption post administration of aflatoxin (AF) and charcoal (Cc) and after clearance period in broiler chickens. (n = 5).

parameter	G1 Control	G2 AF 0.5 mg	G3 AF 1 mg	G4 Cc 0.5 gm	G5 AF 0.5 mg + Cc 0.5 gm	G6 AF 1 mg + Cc 0.5 gm
Initial number of chickens	20	20	20	20	20	20
Mortality number	--	2	5	--	--	2
Mortality %	0	10%	25%	0	0	10%
Initial body weight (gm)	42.8 ± 0.6	43.6 ± 0.83	44.8 ± 0.9	43.2 ± 0.8	44.2 ± 1.1	43.6 ± 1.2
Body weight gain at the end of 4th week (gm)	839.0	760.0	733.2	845.0	808.2	778.4
Total feed consumption at 4th week (gm/chick)	1786	1662	1582	1771	1702	1608
Final weight gain at the end of 6th week (gm)	1479.2	1428.4	1385.6	1474.8	1443.8	1418.4
Total feed consumption at 6th week (gm/chick)	3520	3396	3326	3524	3556	3482

Table 3. Some enzymatic activity post administration of aflatoxin (AF) and charcoal (Cc) and after clearance period, compared to control (mean values ± SE). (n = 5).

Group	Treatment	2nd week			4th week			6th week		
		AP U/L	GGT U/L	LDH U/L	AP U/L	GGT U/L	LDH U/L	AP U/L	GGT U/L	LDH U/L
1	Control	49.2 ± 2.7	51.2 ± 1.85	69.8 ± 1.36	52.2 ± 2.4	50.4 ± 1.17	72.4 ± 1.9	50.4 ± 2.8	51.8 ± 2	71.8 ± 1.7
2	AF 0.5 mg	58.0* ± 1.4	57.4* ± 1.6	76.2* ± 1.8	60.8* ± 2.7	57.2* ± 1.8	78.6* ± 1.7	55.6 ± 1.85	54.2 ± 1.7	73.0 ± 1.7
3	AF 1 mg	59.2** ± 1.16	60.0** ± 1.79	77.4* ± 1.96	62.6** ± 1.9	61.8** ± 2.06	80.2* ± 1.96	60.8* ± 2.4	55.8 ± 1.43	75.0 ± 1.26
4	Cc 0.5 gm	48.0 ± 2.4	51.0 ± 1.18	71.0 ± 1.95	49.6 ± 2.7	50.0 ± 0.9	73.6 ± 1.4	51.2 ± 2.5	51.6 ± 1.6	72.4 ± 1.2
5	AF 0.5 mg + Cc 0.5 gm	54.0 ± 2.1	52.6 ± 0.98	73.0 ± 1.1	54.8 ± 3	53.2 ± 2.13	74.2 ± 1.7	53.8 ± 1.28	52.8 ± 1.85	72.8 ± 1.24
6	AF 1 mg + Cc 0.5 gm	56.4* ± 1.5	57.0* ± 1.67	75.0* ± 1.5	59.8* ± 2.1	56.8* ± 2.1	76.6 ± 1.25	58.4* ± 1.3	53.4 ± 1.96	74.4 ± 1.2

* significant at P<0.05

** highly significant at P<0.01

Table 4. Cholesterol and triglycerides levels post administration of aflatoxin (AF) and charcoal (Cc) and after clearance period compared with control (mean values ± SE). (n = 5).

Group	Treatment	2nd week		4th week		6th week	
		Cholesterol mg/dl	Triglycerides mg/dl	Cholesterol mg/dl	Triglycerides mg/dl	Cholesterol mg/dl	Triglycerides mg/dl
1	Control	126.8 ± 4.5	79.8 ± 1.9	133.2 ± 3.7	81.4 ± 1.5	134.0 ± 3.3	80.8 ± 2.9
2	AF 0.5 mg	108.4* ± 4.52	69.4* ± 2.9	113.4* ± 6.03	70.2* ± 3.3	122.6 ± 4.1	76.6 ± 3
3	AF 1 mg	89.0*** ± 4.2	64.2** ± 2.85	94*** ± 3.24	64.0** ± 3.5	107.0** ± 5.8	74.0 ± 1.8
4	Cc 0.5 gm	126.0 ± 2.9	78.0 ± 2	133.0 ± 2.2	82.0 ± 3.4	135.4 ± 2	81.0 ± 2.7
5	AF 0.5 mg + Cc 0.5 gm	112.0 ± 4.94	72.8 ± 3.5	122.6 ± 4.7	75.4 ± 3.1	128.8 ± 1.4	75.8 ± 1.3
6	AF 1 mg + Cc 0.5 gm	99.2** ± 5.7	67.4* ± 3.7	100.0** ± 6.4	68.6* ± 3.9	110.6* ± 7.72	74.2 ± 3.43

* significant at P<0.05 ** highly significant at P<0.01 *** very highly significant at P<0.001

Table 5. Calcium, phosphorus and uric acid levels post administration of aflatoxin (AF) and charcoal (Cc) and after clearance period,

Group	Treatment	2nd week			4th week			6th week		
		Calcium mg/dl	*Phosphorus* mg/dl	Uric acid mg/dl	Calcium mg/dl	*Phosphorus* mg/dl	Uric acid mg/dl	Calcium mg/dl	Phosphorus mg/dl	Uric acid mg/dl
1	Control	9.28 ± 0.22	4.56 ± 0.09	5.54 ± 0.13	9.08 ± 0.18	4.4 ± 0.06	5.66 ± 0.14	9.06 ± 0.15	4.26 ± 0.11	5.38 ± 0.18
2	AF 0.5 mg	8.22* ± 0.27	3.94* ± 0.22	6.16* ± 0.17	8.1* ± 0.27	3.72* ± 0.23	6.28* ± 0.19	8.42 ± 0.25	4.06 ± 0.12	5.68 ± 0.17
3	AF 1 mg	7.68** ± 0.27	3.6** ± 0.19	6.24* ± 0.23	7.62** ± 0.33	3.52** ± 0.2	6.54* ± 0.17	8.18* ± 0.29	3.76 ± 0.2	5.9* ± 0.13
4	Cc 0.5 gm	9.1 ± 0.17	4.52 ± 0.07	5.62 ± 0.15	9.06 ± 0.22	4.24 ± 0.18	5.54 ± 0.15	9.0 ± 0.14	4.16 ± 0.15	5.34 ± 0.18
5	AF 0.5 mg + Cc 0.5 gm	8.46 ± 0.44	4.12 ± 0.2	5.82 ± 0.17	8.38 ± 0.25	4.08 ± 0.18	5.84 ± 0.23	8.5 ± 0.25	4.06 ± 0.09	5.62 ± 0.22
6	AF 0.5 mg + Cc 0.5 gm	7.92* ± 0.36	3.78* ± 0.25	6.1* ± 0.18	7.98* ± 0.3	3.72* ± 0.24	6.18* ± 0.14	8.3* ± 0.22	3.98 ± 0.12	5.78 ± 0.27

compared with control (mean values ± SE). (n = 5).

* significant at P<0.05

** highly significant at P<0.01

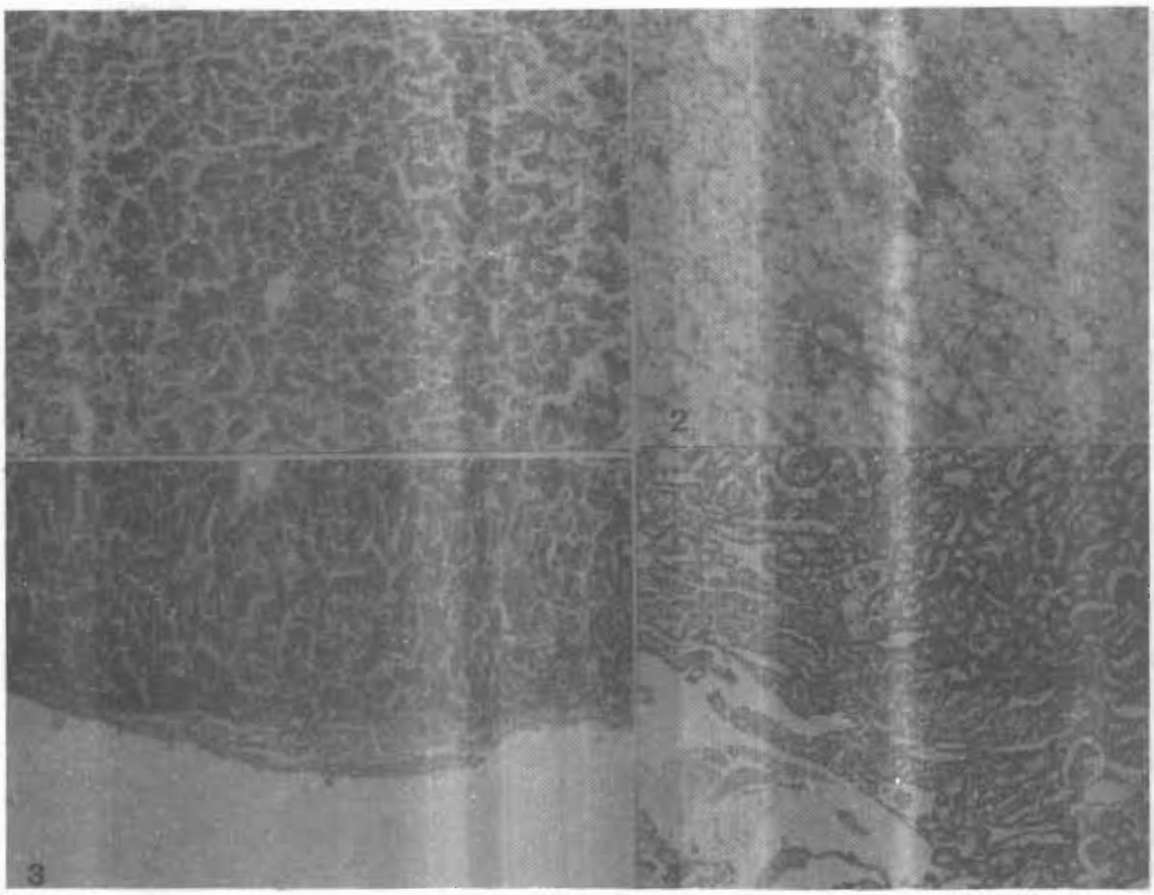

Fig. 1. liver of group (3) at the 4th week showing portal area infiltrated with leukocytic cells beside hyperplasia of bile duct. (H&E. X 300).

Fig. 2. liver of group (3) at the 4th week showing fatty changes. (H&E. X 300).

Fig. 3. liver of group (3) at the 4th week showing thickened capsule. (H&E. X 300).

Fig. 4. kidney of group (3) at the 4th week showing oedema between the renal tubules. (H&E. X 300).

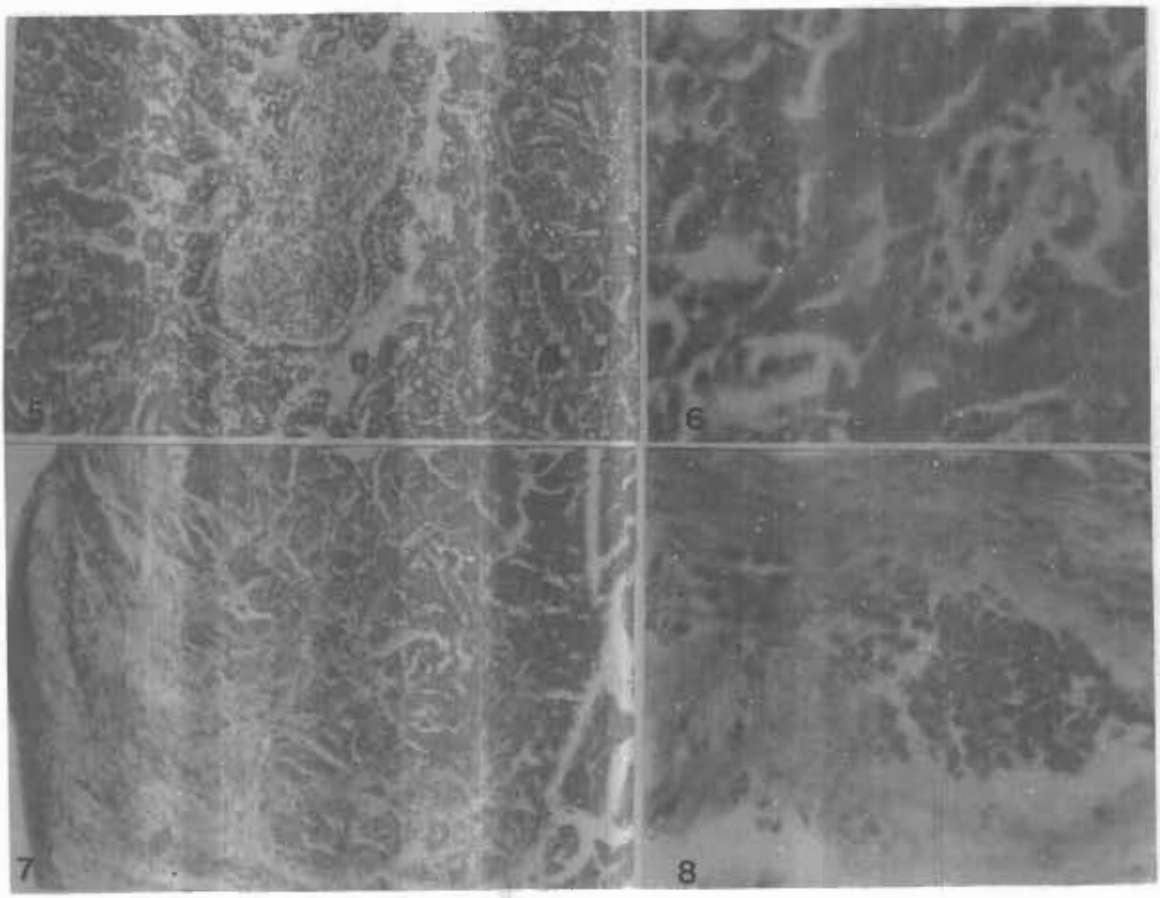

Fig. 5. kidney of group (3) at the 4th week showing congested blood vessels beside haemorrhage between renal tubules. (H&E. X 300).

Fig. 6. kidney of group (3) at the 4th week showing degenerated cells of the renal tubules. (H&E. X 300).

Fig. 7. heart of group (3) at the 4th week showing oedema between the degenerated muscle fibers. (H&E. X 300).

Fig. 8. heart of group (3) at the 4th week showing leukocytic infiltration between the degenerated muscle fibers. (H&E. X 300)

بعض التغيرات البيوكيميائية فى دجاج التسمين أثناء التسمم بالافلاتوكسين و محاولة التقليل منها باستخدام الفحم النباتى النشط

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أجـرى هـذا البحث لدراسة تأثير الافلاتوكسين على دجاج التسمين عند استخدام تركيزات مختلفة من الافلاتوكسين لمدة أربعة أسابيع من عمر يوم حتى ٢٨ يوماً و تقييم الفحم النباتى كمادة واقية من الافلاتوكسين و قد استخدم لهذا الغرض عدد ١٢٠ كـتكوتا عمـر يوم قسمت إلى ٦ مجاميع متساوية. المجموعة الأولى تركت بدون معاملة كمجموعة ضابطة للتجربة. المجموعة الثانـية و الثالـثة تم تغذيتها على عليقة تحتوى على الافلاتوكسين بجرعة مقدارها ٠,٥ مجم و ١ مجم / كجم عليقة على التوالى. المجموعـة الرابعة غذيت على عليقة خالية من الافلاتوكسين و مضافا إليها ٠,٥ جم فحم نباتي. المجموعة الخامسة و السادسة تم تغذيـتها علـى عليقة تحتوى على ٠,٥ مجم و ١ مجم من الافلاتوكسين + ٠,٥ جم فحم نباتى على التوالى لمدة أربعة أسابيع ثم غذيت جميع المجموعات على عليقة خالية من أي إضافات لمدة أسبوعين. تم وزن جميع الطيور فى كل المجاميع أسبوعيا كما تم حسـاب معـدل استهلاك الغذاء و الزيادة فى الوزن. كذلك تم جمع عينات من الدم والأنسجة من كل مجموعة فى نهاية الأسبوع الثانى و الرابع و السادس من بداية التجربة

أظهـرت النـتائج أن الافلاتوكسين احدث نقصا معنويا فى وزن الدجاج و معدل استهلاك الغذاء و كان هذا النقص اكثر وضـوحا فـى التركيز الأعلى من الافلاتوكسين. و كان لإضافة الفحم النباتى أثر إيجابى فى تحسن الوزن و استهلاك الغذاء فى المجموعة الخامسة فقط بينما لم يحدث تحسن معنوى فى المجموعة السادسة وظلت الأوزان اقل من المجموعة الضابطة للتجربة.

اظهـر تحليل السيرم أن الافلاتوكسين تسبب فى حدوث زيادة معنوية فى نشاط إنزيم الفوسفاتيز القلوى، الجاما جلوتاميل ترانسـفيراز و اللاكـتات ديهيدروجينـيز وكذلـك حمـض البوليك. و كان هذا مصحوبا بنقص معنوى فى نسبة الكوليسترول، الجلسـريدات الثلاثـية، الكالسيوم و الفوسفور. و قد كانت شدة هذه التغيرات تتناسب طرديا مع زيادة تركيز الافلاتوكسين. و قد احـدث الفحـم النـباتى تحسنا ملحوظا فى قياسات هذه العناصر فى المجموعة الخامسة بينما لم يحدث هذا التحسن فى المجموعة السادسة التي تم تغذيتها بالتركيز الأعلى من الافلاتوكسين.

وقـد كانت التغيرات الكيميائية فى الدم انعكاسا للتغيرات المرضية للأعضاء الداخلية حيث تسبب الافلاتوكسين فى حدوث تغـيرات تحللـية و تـنخر فى خلايا الكبد و الكلى و ألياف عضلة القلب و قد تفاوتت شدة هذه التغيرات الباثولوجية تبعا لتركيز الافلاتوكسين فى عليقة الدجاج.

كمـا أوضـحت نـتائج التجربة أن استبدال العلائق الملوثة بالافلاتوكسين بعلائق خالية منها لمدة أسبوعين احدث تحسنا ملحوظا فى وزن الدجاج، استهلاك الغذاء ومعظم العناصر البيوكيميائية التي تمت دراستها.

و مـن هـذه النـتائج نستخلص أن التسمم بالافلاتوكسين تسبب فى العديد من التغيرات البيوكيميائية و الباثولوجية و التي انعكسـت سـلبيا علـى الـوزن و معدل النمو فى الدجاج كما أن إضافة الفحم النباتي كان له تأثير إيجابي فى التقليل من الآثار الجانبية المصاحبة للافلاتوكسين و ليس منعها خاصة مع التركيزات العالية من الافلاتوكسين.