

# Biological Sciences Section

Pak. j. sci. ind. res., vol. 33, no.9, September 1990

## EFFECT OF AFLATOXIN ON HAEMOGLOBIN AND SERUM PROTEIN CONCENTRATION IN THREE STRAINS OF BROILER CHICKEN

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(Received September 12, 1989; revised August 22, 1990)

Addition of 250 µg aflatoxin B<sub>1</sub>/kg feed caused a decrease in haemoglobin content and serum protein levels in three strains of broiler chicken. The fall in haemoglobin level was of large magnitude, possibly because the haemoglobin synthesis is maintained preferentially at the expense of other proteins. The level of serum proteins, however, was much reduced (approximately 25%) during the second week of the study but improved slightly at the end of the third week. Possible reasons of these changes are discussed.

**Key words :** Aflatoxin, Haemoglobin, Serum proteins, Broiler strain.

### Introduction

Presence of aflatoxins in the poultry feed causes poor growth rates, poor feed conversion ratios, increased mortality and increased susceptibility to infectious agents [1]. In addition, protein nutrition is adversely affected causing increased demand for achieving the stipulated growth of poults [2]. The economic implications of such a condition are obvious considering the high cost of protein rich ingredients.

Considerable evidence is now available suggesting alteration of lipid and protein metabolism as a result of aflatoxicosis [3- 7], most published literature showing the effect of aflatoxin on serum protein and haemoglobin of broiler chicken at three weeks of age. Since variability in response to dietary aflatoxins has been previously reported in commercial broiler strains [8], this preliminary study was undertaken to ascertain the differential response in the haemoglobin and serum protein contents as a result of 250 µg/kg of aflatoxin in the feed. Studies were undertaken in three strains of broiler chicken at the end of 1, 2 and 3 weeks of age, to indicate changes from the onset of aflatoxicosis to third week of development.

### Materials and Methods

Three strains of healthy, unvaccinated day-old broiler chicks were obtained from the three different well established commercial hatcheries of Karachi. Each strain comprising of 40 chicks was marked with different colours and designated as "A", "B", "C". The chicks weighing between 30 to 40 gm and of mixed sex, were randomly divided into control group and those which were to receive toxic feed (treated groups). The birds were housed in litter-based pens under continuous illumination. Feed and water was provided ad-libitum. A pure culture of *Aspergillus flavus* NRRL 3357, obtained from North Regional Research Laboratory, Peoria, III, USA, was inoculated on broken rice to produce aflatoxin. This contaminated broken rice was incorporated in the feed as a source of toxin. Equal quantities of broken rice were included in both the

control and treated feeds (provided by a commercial feed miller). The protein content in the feed was 21.5%. Ten samples from both the feeds were analysed with CB method [9], quantified by comparison with standards (Standards were obtained from L. Leistner, Institute of Bacteriology 8650 Kulmbach, W. Germany), and confirmed by trifluoroacetic acid derivatives and by spraying 50% H<sub>2</sub>SO<sub>4</sub>. The control feed had no aflatoxin within detectable limit, while the toxic feed contained about 250 ± 15 µg aflatoxin/kg feed.

Three birds were randomly removed from each strain for both diet groups after intervals of 7, 14 and 21 days and sacrificed to collect the blood. Haemoglobin of each individual bird was used for photometric determinations of total serum protein. Diagnostic kits Merckotest 3317 and 3327 (obtained from E. Merck Darmstad, W. Germany) were used for the determination of haemoglobin and total serum protein respectively.

### Results and Discussion

**Haemoglobin.** Hemoglobin in the blood was lower in all the chicks receiving diet containing aflatoxins, as compared to control. After the first week of treatment, a decrease of 12.7, 18.8 and 36.9% of haemoglobin in strain 'A', 'B' and 'C' respectively was observed. Chicks of strain 'A', 'B' and 'C' suffered 28, 54.2 and 23.5% decreases respectively at the end of the second week and 25.5, 11.3 and 14.3% loss respectively at the end of the third week when the experiment was terminated.

**Serum proteins.** All the three commercial strains of broilers receiving aflatoxin in their ration, showed a fall in their serum protein levels during the period under study. Thus, at the end of the first week, strains 'A', 'B' and 'C' showed 26.1, 31.8 and 63.3% decrease in their serum protein levels respectively. At the end of the second week, these levels reduced drastically registering a fall of 91.3, 92.0 and 73.1% respectively. However, there was a slight improvement in the

TABLE 1. EFFECT OF DIETARY AFLATOXIN (250 µg/kg) ON HAEMOGLOBIN AND SERUM PROTEIN (POOLED SERUM OF 3 CHICKS IN EACH GROUP) OF 3 STRAINS OF BROILER CHICKENS.

Parameters	Week	Strain 'A'			Strain 'B'			Strain 'C'		
		Control	Treated	% Decrease	Control	Treated	% Decrease	Control	Treated	%Decrease
Haemoglobin g/100 ml	Ist	7.9± 7.5	6.9± 6.3	13	9.6± 8.8	7.8± 7.3	19	10.3± 8.8	6.5± 6.2	37
	2nd	8.9± 8.3	6.4± 5.7	28	10.7± 10.4	4.9± 5.6	54	8.9± 8.2	6.8± 6.4	24
	3rd	9.0± 9.0	6.7± 6.2	26	8.0± 8.0	7.1± 6.8	11	8.4± 7.8	7.2± 7.8	14
Total proteins g/100 ml.	Ist	2.3	1.7	26	2.2	1.5	32	3.0	1.1	63
	2nd	2.3	0.2	91	2.5	0.2	92	2.6	0.7	73
	3rd	2.0	1.4	30	2.3	1.1	50	2.5	1.3	48

± Standard error

level of serum proteins in all the three strains at the end of the third week.

The results show that all the three strains were sensitive to the effect of aflatoxin on haemoglobin and serum proteins. There was a decrease in the haemoglobin content of all the three strains from the first week to the third week. However, this decrease was not very marked as compared to the fall in the protein levels in the three strains which was very conspicuous during the second week but improved slightly during the third week.

Aflatoxin in chicken feed has been shown to be highly toxic when fed to young broiler chickens from 1 to 21 days of age, and has been known to cause decrease in body weight [10], decrease in packed cell volume [6] and inhibition in secretory proteins of the liver [3-5]. Furthermore, the protein level of a diet has a profound effect on the severity of the aflatoxicosis. If the protein level of the diet is increased, it exerts a protective effect against the aflatoxicosis [11]. In other words, the aflatoxins increase the protein requirements of the birds. These deleterious effects of aflatoxins are dose-dependent. Higher the dose of aflatoxin, greater the damaging effect on the animal [6,7].

A decrease in the serum protein levels in chickens exposed to dietary aflatoxin has been previously reported by a number of workers [12-14]. Present results showing a reduction in serum protein levels support the results of the earlier workers [3] who found the deleterious effects of aflatoxin to be dose dependent and species independent, Inhibitory effect of this toxin was also reported on serum protein [3]. The decrease in serum protein levels caused by aflatoxins was primarily due to reduced rate of synthesis rather than to an increase in the breakdown of protein [12].

A decrease in the haemoglobin content of the blood was observed in the chickens exposed to aflatoxin, although this decrease was not quite marked. A reduction in the haemoglobin and packed cell volume has previously been observed by other workers [6]. Similar adverse effects on blood constituents have also been reported in other avian species such as

turkeys [15] and ducklings [3]. Anemia was confirmed in chicken exposed to aflatoxicosis and it was suggested to be hemolytic in nature [6] but other workers could not find evidence to support the hemolytic anemia hypothesis [12], they suggested it was more like a protein deficiency anemia as reported earlier [16]. The haemoglobin synthesis and consequently the erythrocyte production is maintained preferentially at the expense of other body proteins [17]. This may possibly be the reason that in the present study a very marked fall in the haemoglobin values in the treated chickens was not observed, although the serum protein levels were drastically reduced during the second week.

Aflatoxin has been shown to bind to DNA [18,19] and to impair mRNA synthesis by selective inhibition of RNA polymerase [20,21] leading to inhibition of protein synthesis. Furthermore translation of export proteins such as plasma proteins is dependent in a close association of ribosomes with the endoplasmic reticulum [22,23]. Since aflatoxin can cause a disruption of this association [13], a decrease in plasma proteins of the chickens exposed to dietary aflatoxin can be explained on this basis.

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