

PREVALENCE OF MYCOTOXINS IN POULTRY FEEDS AND FEED INGREDIENTS IN PUNJAB (PAKISTAN)

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ABSTRACT

A total of 100 samples, 50 each of layer starter and broiler starter feeds manufactured by 10 different commercial feed mills were collected to assess contamination of aflatoxin B₂ and Ochratoxin A in poultry feeds and feed ingredients in Punjab. 440 samples of different poultry feed ingredients including cereal grains and animal and vegetable protein sources were also collected from all over Punjab and were analyzed for Ochratoxin A (OA) content. The mean concentration of aflatoxin B₂ in layer and broiler starter rations was observed to range from 10.80 (± 3.16) to 39.20 (± 3.67) $\mu\text{g Kg}^{-1}$. Out of 100 feed samples tested, 40 contained higher than the maximum tolerance level of 20 $\mu\text{g Kg}^{-1}$ aflatoxin B₂. Out of the 440, 19.32 percent (n=85) were contaminated with OA. The occurrence of OA was found to be higher in maize (40%), sun flower (30%), wheat (28%), corn gluten (25%) and barley (24%). OA detected in rice, sorghum, peanut meal, soybean meal, guar meal, corn gluten and rice polish was found to be 20 percent. The findings further showed that maize had maximum mean OA concentration of 112.20 (± 22.69) $\mu\text{g Kg}^{-1}$, followed by 59.43 (± 22.32) $\mu\text{g Kg}^{-1}$ in wheat, 50.33 (± 13.79) $\mu\text{g Kg}^{-1}$ in sunflower meal, 49.20 (± 10.23) $\mu\text{g Kg}^{-1}$ in peanut meal and 39.14 (± 15.06) $\mu\text{g Kg}^{-1}$ in rice polishing. Corn gluten meal, sorghum, barley, cotton seed meal and rapeseed meal contained mean OA concentration ranging from 30 to 36 $\mu\text{g Kg}^{-1}$. The OA content ranging from 10 to 16.67 $\mu\text{g Kg}^{-1}$ was detected in samples of wheat bran, fish meal, blood meal and meat meal. The aflatoxin B₂ and OA contamination of poultry feeds and feed ingredients having deleterious effects seems to pose a serious threat for local poultry farming sector which calls for regular testing and surveillance of poultry feeds and adoption of necessary remedial measures.

Key words: Poultry feeds, feed ingredients, aflatoxin B₂, Ochratoxin A.

INTRODUCTION

Aflatoxins B₁ and B₂ have been detected as contaminants of grain crops before harvest, between harvesting and drying in storage and after processing and manufacturing (Cast, 1989). Occurrence of aflatoxins in poultry and animal feed stuffs is quite common in many countries (Dalcero *et al.*, 1998). Mycotoxins can contaminate the feed thereby affecting animal and human health (Bintvihok *et al.*, 2003). Mycotoxicosis has been implicated to influence quality of poultry feeds. Aflatoxins, a group of closely related, biologically active hepatotoxic mycotoxins are produced by strains of *Aspergillus flavus* and *Aspergillus parasiticus* (Edds and Bortell, 1983). Aflatoxins have elicited the greatest public health concern of all the mycotoxins because of their widespread occurrence in several feed grains (Phillips *et al.*, 1988). Poultry is one of the most sensitive groups of livestock to aflatoxicosis (Edds and Bortell, 1983). Profitability of poultry production can be greatly affected due to the frequency of feed contamination and the detrimental effects of the aflatoxins on performance of chickens (Huff *et al.*, 1988).

Occurrence of aflatoxicosis in poultry and animal feed stuffs is quite common (Dalcero *et al.*, 1998). Aflatoxins B₁ and B₂ ranging from 100 to 320 $\mu\text{g Kg}^{-1}$ in chick starter and broiler starter poultry rations have also been reported from Pakistan (Asghar, 1985). Forty percent of commercial poultry feeds produced in Punjab were found contaminated with aflatoxin (Rizvi *et al.*, 1990). Rehman (2005) observed severe depression in growth rate and immune suppression in broilers due to aflatoxicosis. Natural toxins produced by molds or fungus have threatened the quality and safety of food and have caused severe losses to poultry industry in recent times (Saeed *et al.*, 2009).

Ochratoxin A (OA), a nephrotoxic mycotoxin chiefly produced by *Penicillium viridicatum* and *Aspergillus ochraceus* contaminate a wide variety of cereals and feedstuffs. Contamination of cereals, animal feed stuffs and mixed diets has been reported from many countries (Dwivedi and Burns, 1986). In Pakistan, OA has also been reported in poultry feeds and feed ingredients (Zafar *et al.*, 2001; Hanif *et al.*, 2006). Effects of OA in broiler chickens are both severe and diverse (Van der Merwe *et al.*, 1965) and include depression in body weight gain and feed conversion efficiency (Rehman, 2005). Field cases of suspected mycotoxicosis

including OA in laying birds (Page *et al.*, 1980) with depression in growth rate, enlargement of the kidneys, liver, crop, proventriculus and gizzard and regression of bursa of Fabricius have been reported (Dwivedi and Burns, 1986). Toxicogenic effects of OA with continuous dietary concentration up to $0.0004 \mu\text{gKg}^{-1}$ for one year in White Leghorn layers include decreased egg production (Page *et al.*, 1980), depression in body weight gain and feed conversion ratio and increased mortality rate (Dwivedi and Burns, 1986). A significant decrease in feed intake, body weight and egg mass production was observed in OA treated layer breeders (Hassan *et al.*, 2010).

The present study was conducted with the objectives of assessing aflatoxin B₂ contamination of poultry feeds (broiler starter and layer starter) and Ochratoxin A contamination of poultry feed ingredients in Punjab.

MATERIALS AND METHODS

This study was conducted at University of Veterinary and Animal Sciences (UVAS), Lahore, in collaboration with Poultry Research Institute (PRI), Punjab, Rawalpindi. A total of 100 feed samples, 50 each from layer starter and broiler starter feeds prepared by 10 different commercial feed mills were collected and tested for aflatoxin B₂ content. Besides, 440 samples of different poultry feed ingredients including cereals, animal and vegetable protein sources were also collected from Punjab and tested. These samples were packed in separate polythene bags and marked individually for identification. The particulars of the feed mills and poultry farms and type of feeds and feed ingredients were recorded on a specially designed Performa. The samples thus obtained were tested for quantitative determination of aflatoxin B₂ and Ochratoxin A contents through thin layer chromatography using methods of Association of the Official Analytical Chemists (1990) as described by Farida *et al.* (2001). The findings were interchangeably confirmed to note any error and deviations in the estimations. The levels of aflatoxin B₂ and Ochratoxin A so determined in poultry rations and poultry feed ingredients were subjected to statistical analysis to find out mean value and standard deviation in each feed sample (Steel *et al.*, 1996).

RESULTS AND DISCUSSION

The means of aflatoxin B₂ in 100 samples of feed ranged from 10.80 (± 2.16) to 39.20 (± 23.67) ppb (Table 1). The mean aflatoxin B₂ content in layer and broiler starter rations ranged from 10.80 to 39.20 and 11.00 to 31.40 μgKg^{-1} , respectively. 40 percent feed samples contained aflatoxin B₂ content higher than the

maximum tolerance level of $20 \mu\text{gKg}^{-1}$ in poultry feed (Bhatti *et al.*, 2001). Shaaban *et al.* (1988) detected aflatoxin B₂ in 36.60 percent feed samples with concentration of aflatoxin B₂ ranging from 3.60 to 53.0 μgKg^{-1} in the positive samples. Sahin and Sari (2001) reported aflatoxin B₂ incidence in 1.98 percent poultry feed samples. Ewaidah (1988) observed that 38 percent feed samples were above the FDA guidelines of $20 \mu\text{gKg}^{-1}$ aflatoxin and aflatoxin B₁ and B₂ concentration ranged from 5.4 to 121.2 μgKg^{-1} . The high levels of mycotoxin in different feed ingredients affect the quality of the finished feed (Sun *et al.*, 2006). In this way mycotoxin contaminates the feed and affects the animal health and ultimately the human health (Bintvihok *et al.*, 2003).

Asghar (1985) analyzed and recorded aflatoxin contamination in more than 700 samples of various types of poultry feeds and reported aflatoxin B (both B₁ and B₂) content ranging from 100 to 320 μgKg^{-1} in chick starter and broiler starter rations. Rizvi *et al.* (1990) observed that 42 percent of the commercial poultry feeds produced in Punjab were contaminated with aflatoxin. Bhatti *et al.* (2001) reported that aflatoxin B₁ content in poultry feeds ranged from 13 to 78 μgKg^{-1} . Aflatoxin B₂ have been detected in poultry feed samples (Sahin and Sari, 2001; Shaaban *et al.*, 1988). Aflatoxins are frequent contaminants of harvested feeds and foods like rice, peanut, corn, soybean, sorghum and other grains stored under conditions of high humidity and temperature (Cavalheiro, 1981). Cereals can be highly susceptible to fungal growth when still in the field, during storage and in processing (Stoloff, 1976). Small grains (wheat, sorghum, oats, rye, barley and rice) unless abused in storage or after preparation, appear to be less susceptible to mycotoxin formation than are larger grains such as maize (Cavalheiro, 1981). In view of huge economic losses to the poultry sector due to aflatoxicosis, prevention and control of aflatoxicosis is of great significance. The best control of aflatoxicosis is prevention. Proper sanitation and prevention of fungal development during harvesting, storage and feeding of feed stuffs is vital. Since fungi need relatively high moisture to grow well, grain and poultry feed should be stored below 13 percent moisture (Cavalheiro, 1981).

Out of 440 samples of feed stuffs, 85 were found contaminated with OA showing an incidence of 19.32 percent (Table 2). The results showed that 40 percent (n=10) of maize, 30% of sun flower meal and corn gluten meal, 28% of wheat, 25% of corn gluten meal, 24% of barley, 20 % of rice, sorghum, peanut meal, soybean meal, guar meal, corn gluten and rice polish, 15 % of wheat bran, fish meal, cotton seed meal and rape seed meal, 12% of grams, 10% of blood meal and meat meal and 4% of millet were contaminated with OA. The results further indicated that maximum mean OA concentration in maize was found to be 111.20 μgKg^{-1} , followed by

wheat (59.43 μgKg^{-1}), sunflower (50.30 μgKg^{-1}) and peanut meal (49.20 μgKg^{-1}). The OA concentration in corn gluten meal, sorghum, barley, cotton seed meal, and rape seed meal ranged from 30 to 36 μgKg^{-1} . The OA content ranging from 10 to 22.75 μgKg^{-1} was detected in wheat bran, fish meal, blood meal, meat meal, guar meal and soybean meal, whereas, millet contained 5.0 μgKg^{-1} OA.

The findings of the present study showing higher concentrations of OA in maize and wheat are in agreement with the earlier findings of Shotwell *et al.* (1976) who reported occurrence of OA in corn and wheat as 150 and 115 μgKg^{-1} , respectively. Contamination of cereals, animal feed stuffs and mixed diets have been reported around the world (Dwivedi and Burns, 1986). Majority of earlier reports suggest frequent OA contamination of foods (Dwivedi and Burns, 1986). Buckle (1983) found OA in 12.8 percent of cereals even at low moisture content; barley and wheat in particular had a high concentration of OA. Presence of OA particularly in cereals with frequencies up to 14 percent has been indicated (Krogh, 1978). Field cases of Ochratoxicosis in broilers and layers with adverse effect on their performance have been reported (Page *et al.*, 1980; Doerr *et al.*, 1981). Ochratoxicosis A has been reported to reduce hatchability of fertile eggs with depressed progeny performance during their first 2 weeks of life (Choudhury *et al.* 1971). The detrimental effect of OA in young broilers with dietary concentration of 2 ppm

(Dwivedi and Burns, 1986 and Rehman, 2005) and in layers up to 4 ppm (Page *et al.*, 1980) has been indicated.

The presence of aflatoxin B₂ and OA in poultry feed ingredients and processed feeds having deleterious effects on performance of broilers and layers seems to pose a serious threat for local poultry farming sector which calls for regular testing and surveillance of poultry feeds for aflatoxicosis and OA contamination and adoption of necessary remedial measures.

Table-1: Concentration of Aflatoxin B₂ (μgkg^{-1}) with their standard errors detected in different poultry feeds samples

Commercial samples.	Samples (No.)	Mean concentration of aflatoxin B ₂	
		Layer starter	Broiler starter
A	10	17.40 \pm 6.46	11.0 \pm 4.58
B	10	18.20 \pm 13.86	11.4 \pm 5.12
C	10	14.80 \pm 4.02	20.0 \pm 8.33
D	10	17.40 \pm 8.01	19.20 \pm 13.10
E	10	10.80 \pm 2.16	*26.0 \pm 9.19
F	10	*23.0 \pm 7.38	*26.40 \pm 4.33
G	10	15.20 \pm 7.42	16.60 \pm 4.77
H	10	*29.40 \pm 13.61	*21.0 \pm 5.38
I	10	13.20 \pm 4.08	*22.40 \pm 9.76
J	10	*39.20 \pm 3.67	*31.40 \pm 10.40
Total	100	--	--

μgKg^{-1} = Micro gram per kilogram

*=Higher levels than maximum tolerance level of μgKg^{-1} (Bhatti *et al.*, 2001)

Table-2: Ochratoxin A Concentration (μgkg^{-1}) in different poultry feed ingredients

Ingredients.	Samples tested. (No)	Positive samples (No) (%contaminated).	OA concentration.		Mean OA concentration (Standard error).
			Min.	Max.	
Wheat	25	7(28)	25	98	59.43 (\pm 22.32)
Maize	25	10(40)	75	140	111.20 (\pm 22.69)
Rice	25	5(20)	15	40	27.40 (\pm 9.65)
Sorghum	25	5(20)	25	40	33.60 (\pm 5.46)
Barley	25	6(24)	15	112	30.33 (\pm 8.77)
Peanut meal	25	5(20)	30	58	49.20 (\pm 10.23)
Millet	25	1(4)	05	08	5.00 (\pm 0.00)
Grams	25	3(12)	10	15	12.67 (\pm 2.05)
Cotton seed meal	20	3(15)	20	42	32.33 (\pm 9.18)
Soybean meal	20	4(20)	10	33	22.75 (\pm 8.7)
Rape seed meal	20	3(15)	28	37	33.00 (\pm 3.74)
Sun-flower meal	20	6(30)	30	68	50.33 (\pm 13.79)
Guar meal	20	4(20)	15	30	21.75 (\pm 5.40)
Com gluten meal (60%)	20	5(25)	20	48	32.20 (\pm 10.24)
Com gluten meal (30%)	20	4(20)	20	56	36.00 (\pm 15.43)
Rice polishing	20	4(20)	25	57	39.14 (\pm 15.06)
Wheat bran	20	3(15)	05	10	11.67 (\pm 6.24)
Fish meal	20	3(15)	12	20	16.67 (\pm 3.40)
Blood meal	20	2(10)	10	18	14.00 (\pm 4.00)
Meat meal	20	2(10)	05	15	10.00 (\pm 5.00)
Total:-	440	85(19.31%)	--	--	--

Min = Minimum.

Max. = Maximum.

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