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Variation of Aflatoxin Level in Different Poultry Feeds Used in Different Poultry Farms of Bangladesh Round the Year

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Abstract: As quality of feed is the main determinant factor in successful poultry farming attempts were made to evaluate the quality of feeds used in the farms of Bangladesh in respect of aflatoxin. Feed samples were collected from two layer farms, four big hatcheries and six broiler farms located in different areas of Bangladesh and the collection was continued for twelve months. In feed samples collected during the twelve months, aflatoxin content was found to vary from 0 to 98 ppb. Only aflatoxin B1 was detected in the feed samples and the percentage of incidence was at a maximum in the months of June, July and August, while and from November to February, no aflatoxin was detected in any feed. Higher amount of aflatoxin accumulation in the poultry feed was noticed in rainy season. A positive correlation (0.814) was found to exist between the moisture and aflatoxin contents of the feed. Feed used in the breeder farm was superior to those of broiler and layer farms with respect to aflatoxin content. Poor quality maize could be considered as a source of aflatoxin contamination in poultry feed as it contained reasonable amount of the toxin (23 to 121 ppb) irrespective of the season. Storage condition as practiced in the farm and the left-over feed of the farm might not have contributed to the elevation of aflatoxin level in poultry feed.

Key words: Aflatoxin, poultry feed, poultry farms, moisture content

Introduction

The people of Bangladesh and other developing countries are undernourished. There is a great shortage of protein especially animal protein in our diet. By increasing the productivity of poultry meat and eggs, the existing gap between supply and demand of animal protein can be bridged. Poultry meat contributes around 37% of the total animal protein supply of Bangladesh (Ahmed and Islam, 1990).

Though at present only 15% of the total poultry products are coming from commercial farms, poultry industry has established its position as the fastest growing segment in the agricultural sector. Profitable poultry farming is a highly specialized enterprise in which a lot of factors may be responsible to offset the profit. Among these, quality of chicks and feeds is most important. Feed alone accounts for approximately 65-70% of the total cost of production of poultry meat and eggs.

Bangladesh is a warm and humid country. In rainy season, the humidity persists at around 90-95%, which is very favorable for the growth of fungus facilitating the accumulation of aflatoxin in the poultry feed.

Imported feed ingredients are often reported to be contaminated with the same toxin. Feeding practices followed by the poultry farms may also be responsible for the deposition of aflatoxin in the feed. Aflatoxin causes aflatoxicosis in poultry, which tremendously decreases feed conversion efficiency and growth rate of poultry. It reduces the number of egg production as well

as egg weight. Along with other pathological changes, liver and kidney are the most affected organs. In extreme situation it may cause the death of the poultry (Rao, 1981). Aflatoxin is reported to be a potent carcinogen. In 1998, more than 200 farms in Savar area were affected by aflatoxicosis. At that time more than 40% of birds died of this disease. In some farms death toll raised up to 70%. Considering the deleterious effect of aflatoxin on poultry farming, its variation in the feed in different seasons of the year needs to be monitored.

The present study reports the sources of feeds and feeding practices followed in different farms at different parts of Bangladesh, the variations in aflatoxin level in poultry feeds used in the farms and aflatoxin contents in the feed ingredients as well as in the left-over feeds.

Materials and Methods

Feed samples were collected from twelve different poultry farms located in different areas of Bangladesh. Four of them were situated at Mymensingh, five at Gazipur; two at Dhaka and one was selected from those at Chittagong. Precise information about the poultry farms is given in Table 1. Among them two were layer farms, four were big hatcheries and the rest were broiler-farms. Feed samples were collected twice a month at an interval of fifteen days between each successive collection.

Feed collection was begun from December 2000 and continued till November 2001 and a composite

Table 1: Precise information about the poultry farms under investigation

Farm No	Location	Type of farm	No. of birds	Source of feed
1	Mymensingh	Broiler	500	RF
2	Mymensingh	Broiler	1000	RF / SP
3	Mymensingh	Layer	500	RF
4	Mymensingh	Breeder	10,000	RF
5	Gazipur	Breeder	10,00,000	SP
6	Chittagong	Broiler	1000	RF
7	Gazipur	Broiler	5000	RF
8	Gazipur	Breeder	10,000	RF
9	Dhaka	Layer	1000	RS
10	Dhaka	Broiler	800	RS
11	Gazipur	Breeder	100,000	SP
12	Gazipur	Broiler	2,000	RF

RF= Renowned feed-mill, SP= Self produced, RS= Retailer shop

sampling was made in each occasion. Considering the individual farms and date of collection, each collected sample was endorsed with a code number, The samples were kept in refrigerator (-22°C) till subjected to quantification of aflatoxin. The moisture content of the feed sample was done in Biochemistry Laboratory of Bangladesh Agricultural University, Mymensingh.

The incidence of aflatoxin was reported to be associated with specific foodstuff such as peanut, maize or rice etc. Considering the wide use of maize in poultry feed, whole maize was collected from the retailer shop and analyzed for aflatoxin content. The left-over was collected from the poultry farm for aflatoxin quantification.

The feed samples were ground by using micro grinder (A-10 Analytical mill. Tekmar. Germany) and was passed through 100 mesh sieve. Dry matter was determined according to AOAC, 1990.

Determination of Aflatoxin from Feed Sample: Aflatoxin was determined in toxicology laboratory of the Institute of Food Science and Technology (IFST), Bangladesh Council of Scientific and Industrial Research (BCSIR), Dhaka. Extraction was done according to method of Romer (1980). Twenty-five grams of ground feed were taken in a blender and 125 ml of acetone and water (106:19) was added with it. It was blended for 2 minutes in high speed and was filtered. The filtrate was collected and 1.5 g of CuCO_3 was added with it. In another conical flask 85 ml of 0.2 M NaOH and 15 ml of FeCl_3 solution (66.6g/l H_2O) was added where slurry was formed. That slurry was added with the filtrate containing CuCO_3 and was again filtered. Seventy-five milliliters of filtrate, 75 ml of 0.03% H_2SO_4 and 5 ml of chloroform were taken in a separatory funnel and were shaken vigorously. The chloroform layer was separated and was washed with 50 ml [KOH(2.24g/l) + KCl (6g/l)] solution. Two milliliter of chloroform was collected in a vial and was dried up. Thus the extraction was completed.

Aflatoxin was quantitated according to the method of Coker *et al.* (1988). A silica gel (60) coated 20 x 20 cm²

TLC (Thin Layer Chromatography) plate was used for separation of aflatoxin. (Layer thickness 0.2 mm.; E. Merck Darmstadt, Germany). The extract collected and dried up in the vial was dissolved in 50 μl of chloroform. The plate was spotted by using micropipette. Aflatoxin standard (B1, B2, G1 and G2) was spotted on TLC plate. The sample was also spotted as 1, 2, 3, 4 and 5 μl . The plate was developed in chloroform acetone (9:1) solution for 20 minutes. The running of chromatogram was one-dimensional and ascending. After development the plate was dried for 5 minutes at 60°C. It was then placed under UV lamp (360 nm) and the quantity of aflatoxin was assayed by comparing with standard aflatoxin.

Results and Discussion

Analytical data of the feed samples collected from different poultry farms, round the year, are given successively as per collection sequence in Table 2.

It was observed that the dry matter (%) of the feed varied from farm to farm (Fig. 1) as well as month to month (Fig. 2). The variations among different farms and months were statistically significant ($p < 0.001$). It was observed that the dry matter (%) was lowest from June to July and then progressively increased. And in dry season (from mid October to December) it remained highest.

It appeared from the table 2 that feed from most of the farms contained aflatoxin in the months of June, July and August. In feed samples only B1 was detected though the samples were subjected to analysis for B1, B2, G1 and G2. Considering the number of farm infected, the percentage of incidence in those months were 75, 100 and 91% respectively. In the month of September and October the rates of incidence declined gradually and were recorded as 66 and 41% respectively. From November till February no aflatoxin was detected in any of the feed. From March onward the rate of incidence gradually increased again (Fig. 3). A positive correlation ($r = 0.814$) (Fig. 5) was found between moisture

Khan et al.: Variation of Aflatoxin Level in Different Poultry Feeds

Table 2: Aflatoxin content (ppb) of poultry feeds collected from different farms round the year

Months	F01	F02	F03	F04	F05	F06	F07	F08	F09	F10	F11	F12	Af in % of farms
Dec	0	0	0	0	0	0	0	0	0	0	0	0	0
Jan	0	0	0	0	0	0	0	0	0	0	0	0	0
Feb	0	0	0	0	0	0	0	0	0	0	0	0	0
Mar	0	0	0	0	0	0	0	0	0	15	0	0	8
Apr	3	0	0	14	0	23	25	0	0	24	0	0	41
May	23	34	32	25	32	34	0	0	17	56	0	0	66
Jun	34	54	45	36	0	54	95	0	56	41	0	76	75
Jul	98	40	86	47	23	81	33	76	85	81	42	78	100
Aug	82	31	42	12	14	24	48	12	54	21	0	51	91
Sep	54	14	10	0	0	10	23	0	24	35	0	12	66
Oct	21	14	0	0	0	0	18	0	17	14	0	0	41
Nov	0	0	0	0	0	0	0	0	0	0	0	0	0
Avr	26.25	15.58	17.92	11.17	5.75	18.83	20.17	7.33	21.08	23.92	3.5	18.08	

Each figure is the average of two feed samples collected in the same month

Table 3: Aflatoxin content in supplied feed and in respective left- over

Code No. of sample	Aflatoxin in fresh sample (ppb)	Aflatoxin in left-over (ppb)	Code No. of sample	Aflatoxin in fresh sample (ppb)	Aflatoxin in left-over (ppb)
F01-06	0	0	F06-11	34	30
F01-13	34	34	F06-17	30	32
F02-19	28	25	F07-23	18	20
F03-07	0	0	F08-17	24	28
F03-15	70	76	F09-12	34	34
F04-11	20	26	F09-19	30	32
F04-17	20	18	F10-15	41	52
F05-07	0	0	F11-18	0	0
F05-16	20	22	F12-04	0	0
F05-22	0	0	F12-22	0	0

Table 4: Dry matter and aflatoxin content in apparently good quality (GQ) and poor quality (PQ) whole maize

		Months								
		Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov
Dry matter (%)	GQ	91.2	90.3	91.6	90.6	89.9	90.1	91.2	91.8	91.5
	PQ	86.6	83.4	84.8	85.2	81.3	83.9	85.8	86.2	87.8
Aflatoxin (ppb)	GQ	0	0	0	0	0	0	0	0	0
	PQ	23	98	85	47	121	104	82	47	56

percentage of the feed and the level of aflatoxin detected. A positive correlation ($r = 0.71$) (Fig. 6) was also found between rainfall and aflatoxin. In 2001, the values of correlation between rainfall and aflatoxin were found to be 0.63, 0.66 and 0.76 at Mymensingh (Fig. 7), Dhaka (Fig. 8) and Chittagong (Fig. 9) area respectively. The elevation of aflatoxin level of poultry feed in rainy season was also reported earlier (Dawlatana, 1998). A devastating result of aflatoxicosis was observed in 1998 and it is interesting to note that the rainfall in that year was also remarkably higher than that of year 2001 (Fig. 10). Aflatoxin was detected in the samples varied from as low

as 3 ppb to 98 ppb (Table 2). Not more the presence but the level of toxin rendered a feed dubious for its use as poultry feed. According to EEC and Bureau of India standard the permissible levels are 20 ppb and 50 ppb respectively (Kurkure and Kalorey, 2001). In Bangladesh perspective, as mentioned in Khamar Bichitra and Poultry Nirdeshica by Rhone Poulenc, the permissible level has been raised to 50 ppb broiler for and 100 ppb for layer. So, if the permissible level of EEC is taken into account, the quality of most of the feed samples collected during the period from May 2001 to August 2001 was quite unacceptable. According to Bureau of India standard and

Khan *et al.*: Variation of Aflatoxin Level in Different Poultry Feeds

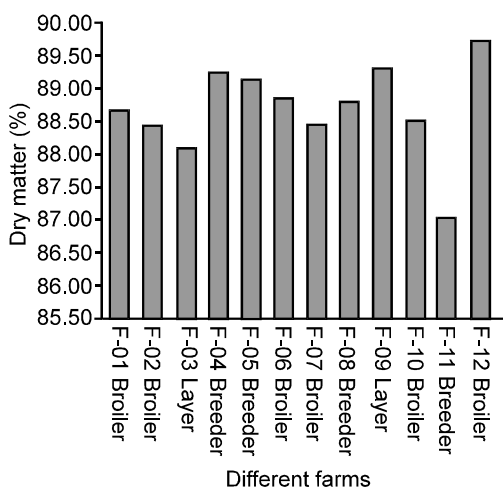


Fig. 1: Variation of average dry matter (%) of poultry feeds collected from different farms

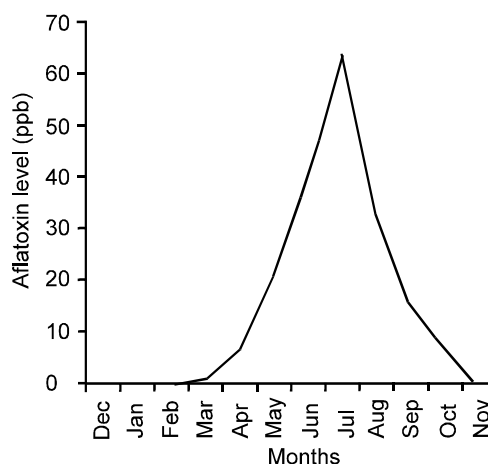


Fig. 3: Variation of average aflatoxin level of poultry feeds found in different months

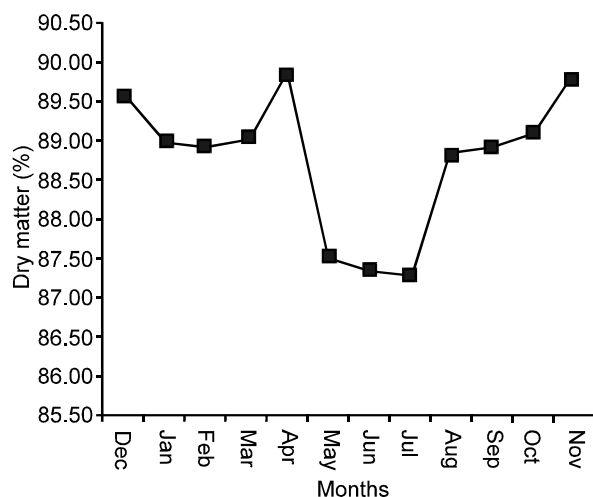


Fig. 2: Variation of dry matter (%) of poultry feeds in different months

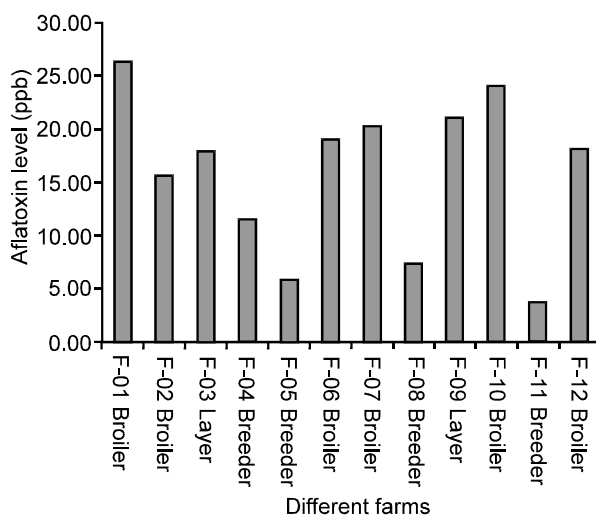


Fig. 4: Variation of average aflatoxin level (ppb) of poultry feeds collected from different farms

according to Bangladesh standard only a few of them appeared to be risky. It was also observed that compared with other types of farm, the breeding farm had used superior quality feed with respect to aflatoxin (Fig. 4). Farm-5, farm-8 and farm-11 were breeder farm and the reported rate of incidence of aflatoxin in those farms were thrice, twice and once in a year respectively (Table 2). The rate of incidence was much higher in other types of farm. The breeder farms had their own feed-mill, which might enable them to produce better quality feed. Farm-4 was also a breeder farm but the feed quality was not as good as the others because the feed was procured from elsewhere.

Among the other farms maximum incidence of aflatoxin (8 months in a year) was noticed in Farm-10 (Table 2) and maximum average value of aflatoxin (26.25 ppb)

was observed in Farm-1 (Table 2). In both the cases, the feeds were purchased from retailer shop. As the feed of farm-1 was produced by a renowned feed-mill, higher incidence of aflatoxin in the feed was undesirable. Use of aflatoxin contaminated feed ingredients or improper and too long storage of the feed either in mill or in shop might have deteriorated the quality of the feed. In case of farm-10, the retailer shop was not aware or sincere to maintain the quality of the feed.

Considering the humid condition in the poultry house it was assumed that the left-over feed might be a source for aflatoxin for the birds. But after analysis it was found that the aflatoxin content (Table 3) of the left-over was almost identical with that of respective supplied feed. It was also observed that the farm owner regularly cleaned the feeder and very little or no left-over feed remained in

Khan *et al.*: Variation of Aflatoxin Level in Different Poultry Feeds

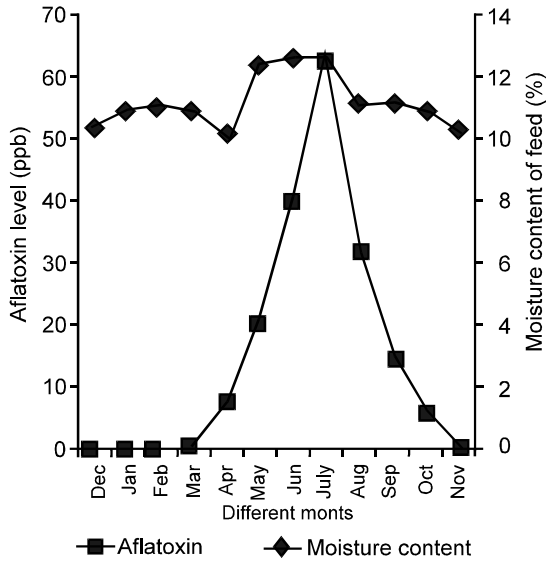


Fig. 5: Correlation between aflatoxin level and moisture content of feed

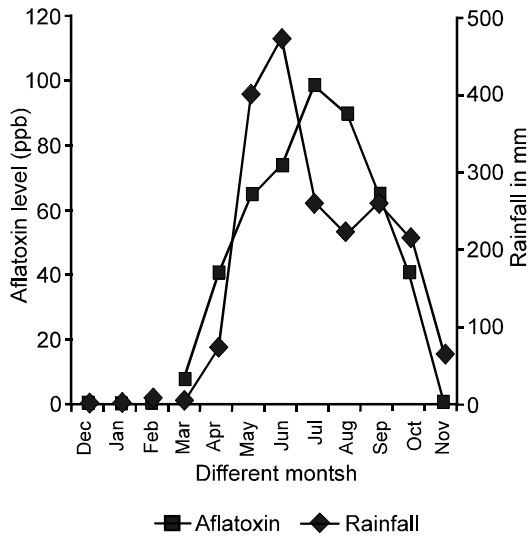


Fig. 6: Relation between rainfall and alfatoxin level in different months

the feeder. So, there was presumably no chance of aflatoxin contamination from feeder. Storage practice of the feed in the individual farm plays an important role in the production of aflatoxin. But it was found that no farm stored the feed for more than 15 days. The feeds were noticed to be contained in water-proof plastic sac. So, storage practices of the farms have little contribution in elevating aflatoxin level of a feed.

When a comparison was made between the good (GQ) and the poor quality (PQ) whole maize with respect to their moisture and aflatoxin contents, poor quality maize was found to contain aflatoxin all the time (Table 3) irrespective of season. They also contained higher

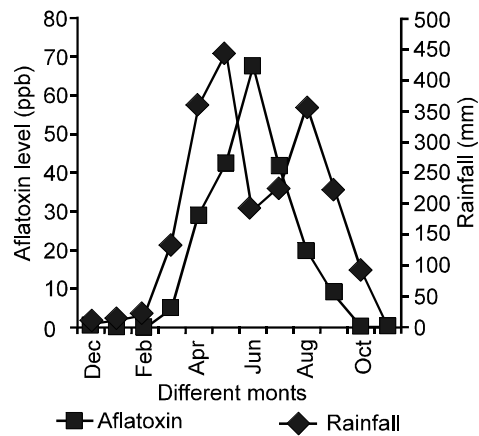


Fig. 7: Relation between rainfall and aflatoxin level (ppb) in Mymensingh

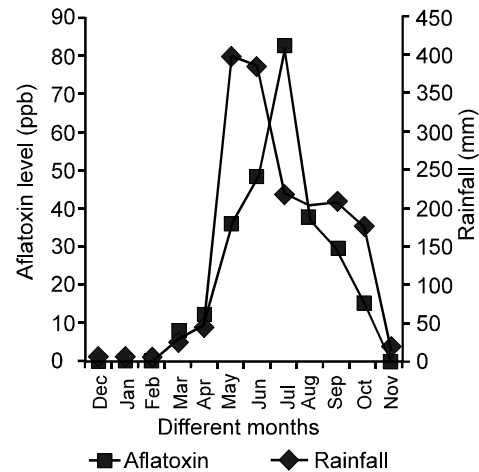


Fig. 8: Relation between rainfall and aflatoxin level (ppb) in Dhaka

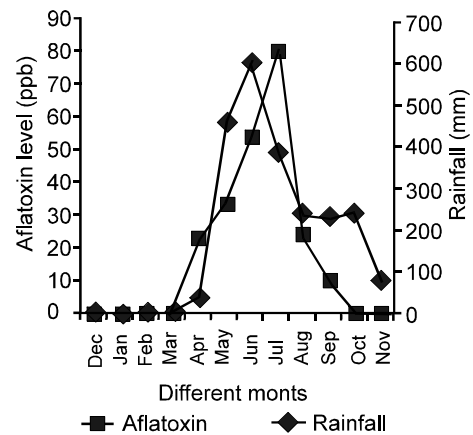


Fig. 9: Relation between rainfall and aflatoxin level (ppb) in Chittagong

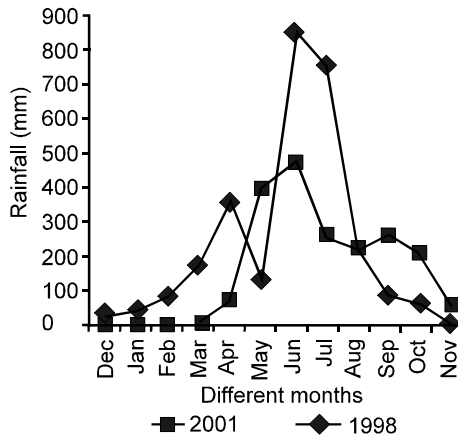


Fig. 10: Rainfall in 2001 and 1998

amount of moisture. In good quality maize, aflatoxin was not noticed even in wet season (June, July and August). On the basis of this study the following conclusion can be made. The quality of feed used in the breeder farms appeared to be better than those of broiler and layer farms. The feed produced by the renowned feed-mill are better than those from retailer shop. A seasonal variation is found in case of dry matter (%) of the feed. In wet season dry matter (%) of the feed decreases. A seasonal variation of aflatoxin content in the poultry feed has been observed. Storage practice and the left-over feed may not be considered as factors in the elevation of aflatoxin level of the feed. Feed ingredients such as maize may be considered as a great factor in the elevation of aflatoxin level of the feed.

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