

## Mycotoxin detoxification of broiler feed by a mycotoxin binder

Reza Soleimani<sup>1\*</sup>, Omid Pourzargham Faradonbeh<sup>2</sup> and Hossein Bagheri<sup>2</sup>

<sup>1</sup>Department of Animal Science, Chaloos Branch, Islamic Azad University, Chaloos, Iran

<sup>2</sup>Department of Agriculture, Chaloos Branch, Islamic Azad University, Chaloos, Iran

### Abstract

In the present work, an *in vitro* model was tested for evaluation of an adsorbent to ameliorate the toxic effects of mycotoxins by quantifying free mycotoxins through thin layer chromatography analysis. *In vitro* binding ability of a commercial binder (0.2%) on aflatoxin B1 (AF) (500ng/g), ochratoxin A (OA) (1µg/g) and T-2 toxin (T-2) (2µg/g), when present alone or in combination at pH 4.5 and 6.5 in the diets. Binder showed significantly ( $P<0.05$ ) higher binding for AF (94.71%), whereas those recorded for T-2 (74.28%) and OA (63.13%) were moderate. Binding of each toxin decreased as the number of toxins in feed increased. Higher binding ability for binder was noticed at 6.5 pH of the medium.

**Keywords:** Aflatoxin; ochratoxin A; T-2 toxin; binder; *in vitro*

---

**To cite this article:** Soleimani R, OP Faradonbeh and H Bagheri, 2011. Mycotoxin detoxification of commercial broiler feed by a mycotoxin binder. Res. Opin. Anim. Vet. Sci., 1(12), 778-780.

---

### Introduction

Animal feed ingredients and compounded feed, by virtue of their high vital nutrients and moisture contents generously support the multiplication of moulds at all stages of the food chain, i.e., production, harvesting, handling, processing and storage. Many of these moulds produce toxic metabolites known as mycotoxins. Aflatoxin (AF), Ochratoxin A (OA) and T-2 toxin (T-2), are secondary metabolites of *Aspergillus flavus*/*Aspergillus parasiticus*, *Aspergillus ochraceus* and *Fusarium sporotrichoides*, respectively and are commonly encountered in animal feed stuffs. These mycotoxins when consumed in combination may show greater negative effects on well being and productivity of broiler chicken than when consumed alone (Raju and Devegowda, 2002). Many approaches have been tried to counteract mycotoxicosis in poultry operations including chemical, nutritional and biological methods. Though some of them have proved effective on some mycotoxins, the search is still on for a natural, cost effective and field applicable solution for this problem in poultry.

Herbal components like turmeric (*Curcuma longa*), garlic (*Allium sativum*) and green algae (*Spirulina plantensis*) have been shown to counteract mycotoxins and they also act as potential antioxidants. Moreover, because of their non-toxicity, the use of these agents for preventing effects of aflatoxins in chicken has been studied (Fanelli et al., 1985).

The present trial was conducted to study the *in vitro* binding ability of a commercial binder, a toxin binder which is claimed to be degrades peroxides, amides and lacto rings of non-polar toxins and prevent DNA adduct formation and cellular damages by preventing oxidation of toxins. Thus the present study was designed to study the *in vitro* binding efficiency of commercial mycotoxin binder.

### Materials and Methods

Mycotoxin binding efficacy of herbal binder (HB) was evaluated in toxin-contaminated feed under simulated *in situ* gastrointestinal (GI) tract environment of chicken. The composition of binder was as follows:

---

\*Corresponding author: Reza Soleimani, Department of Animal Science, Chaloos Branch, Islamic Azad University, Chaloos, Iran. Email: reza\_soleymani61@yahoo.com

- a) Minerals (extra purified clay containing diatomaceous earth mineral),
- b) Antioxidants (curcuminoids extracted from turmeric) and
- c) Enzymes (epoxidase and esterase).

### Experimental design

Aflatoxin B<sub>1</sub> (500 ppb), OA (1 ppm) and T-2 (2 ppm) were studied individually and in combination with and without binder (0.2%) (Table 1). Each of these treatments was tested at pH levels of 4.5 and 6.5, simulating the toxin adsorption activity of binder as in the fore and mid portion of GI tract of chicken in triplicates.

### Production and quantification of mycotoxins

AF, OA and T-2 were produced employing solid substrate fermentation as per the methods of Shotwell et al. (1969), Trenk et al. (1975) and Burmeister et al. (1971), respectively. The respective fungal cultures used were *A. parasiticus* MTCC 1894 (Source: Microbial Type Culture Collection and Gene Bank, IMT, Chandigarh, 160 036, India), *A. ochraceus* NRRL 3174 (Source: National Center for Agricultural Utilization Research, USDA, Peoria, Illinois 64604, USA) and *Fusarium sporotrichoides* MTCC 1894 (Source: Microbial Type Culture Collection and Gene Bank, IMT, Chandigarh, 160 036, India).

Mycotoxin content of the culture material was determined by thin layer chromatography as per AOAC, (1995) Rukmini and Bhat (1978) for AF and OA and Romer et al. (1978) for T-2.

### Experimental procedure

Compounded broiler feed weighing 25 g, was taken in a 250 ml erlenmeyer flask and the required quantity of culture material was added to arrive at the desired level of toxin. Binder was added at 0.2 % level to these flasks whereas the feed in control flasks of the

respective treatment was left untreated. Citric acid-sodium phosphate buffer (100 ml) of the desired pH (4.5/6.5) was added to each flask and the contents were incubated at 37°C for 3 h, then the contents were filtered and the residue was dried at 37°C for 2 h. The respective toxin was extracted from the residue and quantified.

The percentage difference in the toxin content between the beginning and at the end of trials in binder treated and control flasks were calculated. Percent binding of each toxin in different treatments was determined as follows by subtracting the per cent difference in toxin content of the control flasks from that of the treated flasks in the respective treatment.

$$\text{Per cent toxin adsorption} = \left[ \frac{B_T - E_T}{B_T} \times 100 \right] - \left[ \frac{B_C - E_C}{B_C} \times 100 \right]$$

Where

B<sub>T</sub> = toxin content in the treated flasks at the beginning

B<sub>C</sub> = toxin content in the control flasks at the beginning

E<sub>T</sub> = toxin content in the treated flask at the end

E<sub>C</sub> = toxin content in the treated flask at the beginning

### Results

The per cent binding of AF, OA and T-2 by binder, either alone or in combination are presented in Table 1. Significant (P<0.05) differences were noted in binding of different mycotoxins among the different dietary treatments. Among the diets containing the individual toxins, a significantly higher binding was recorded with AF than T-2 and OA.

At pH 4.5, the highest binding percentage of mycotoxins was noticed for AF (90.68%), whereas, the lowest binding ability was recorded for OA (61.73%) with diet containing 0.2 per cent binder. In the combination treatments, the highest binding ability was

**Table 1: Per cent mycotoxin binding *in vitro* by a commercial binder (0.2 %) at two levels in different dietary treatments**

Treatments	AF			OA			T-2		
	pH			pH			pH		
Mycotoxins	4.5	6.5	X	4.5	6.5	X	4.5	6.5	X
AF	90.68	94.71	91.94 <sup>a</sup>	-	-	-	-	-	-
OA	-	-	-	61.73	63.13	62.28 <sup>e</sup>	-	-	-
T-2	-	-	-	-	-	-	71.33	74.28	72.00 <sup>e</sup>
AF+OA	62.30	65.80	63.79 <sup>b</sup>	33.70	34.00	33.80 <sup>g</sup>	-	-	-
AF+T-2	43.24	44.67	43.95 <sup>c</sup>	-	-	-	46.14	46.39	46.76 <sup>f</sup>
OA+T-2	-	-	-	34.08	34.05	34.56 <sup>g</sup>	45.27	47.48	46.87 <sup>f</sup>
AF+OA+T-2	33.47	32.43	33.60 <sup>d</sup>	5.33	6.26	5.48 <sup>h</sup>	9.10	9.70	9.40 <sup>h</sup>
Average for each mycotoxin			58.23 <sup>x</sup>			34.14 <sup>y</sup>			43.75 <sup>z</sup>
SEM			3.54			1.94			2.06

<sup>a-h</sup> Means of different mycotoxins in each treatment, bearing common superscript, do not differ significantly (P<0.05); <sup>x-z</sup> Pooled means of each mycotoxin among the various treatments, bearing different superscripts differ significantly (P<0.05); AF: Aflatoxin B<sub>1</sub> 0.5 ppm, OA: Ochratoxin A 1 ppm, T-2: T-2 toxin 2 ppm.

noticed for AF+OA (62.30%) and lowest binding ability was recorded AF+OA+T-2 (5.33%) at the pH of 4.5.

At pH 6.5, highest binding percentage of mycotoxins was recorded for AF (94.71%) and lowest binding percentage was noticed for OA (63.13%) with diet containing 0.2 per cent binder. In the combination treatments, the highest binding ability was noticed for AF+OA (65.80%) and lowest binding ability was recorded for AF+OA+T-2 (6.26%) for the pH of 6.5.

## Discussion

The information on exact mechanism of action of this binder is scanty but the binding of mycotoxins could be attributed to the presence of diatomaceous earth in the binder. The diatomaceous earth is a powerful natural adsorbent and it might adsorb the toxins effectively through their polar ends of toxin.

Curcumin induces drug metabolizing enzymes like glutathione-S-transferase. Induction of enzymes results in efficient detoxification of cytotoxic or carcinogenic compounds (Shalini and Srinivas, 1987; Soni et al., 1992). Vitamin E and C are the main antioxidants that inhibit free radical damages in biological systems but curcumin removes free radicals by producing a stable radical and thus its molecules act as shuttle of scavenger continuously removing the radicals (Fanelli et al., 1985). This effect may be influenced by the nature of functional atomic groups, present on the mycotoxin molecule. However, the ability of the toxin binder to bind mycotoxins depends on pH, molecular arrangement and its geographic region of origin (Vieira, 2003). Thus it could be construed that binder has broad-spectrum efficacy against these mycotoxins and selectively inactivate certain mycotoxin molecules effectively.

From the results of this *in vitro* study, it could be concluded that the binder has high ability to adsorb AF from the feed. The highest binding percentage was 94.71 per cent individually and 65.80 per cent in combination with toxins. Binding of each toxin decreased as the number of toxins in feed increased. The medium pH showed higher binding ability for this binder.

## References

- AOAC, 1995. Official Methods of Analysis. 16th Ed., Association of Official Analytical Chemists, Washington, D.C.
- Burmeister, H.R. 1971. T-2 toxin production by *Fusarium tricinctum* on solid substrate. *Applied Microbiology*, 21: 739.
- Fanelli, C., Fabbri, A.A., Picretti, S., Finotti, S. and Passi, S. 1985. Effect of different antioxidants and free radical scavengers on aflatoxin production. *Mycotoxin Research*, 65-69.
- Raju, M.V.L.N. and Devegowda, G. 2002. Esterified glucomannan in broiler chicken diets contaminated with aflatoxin, ochratoxin and T-2 toxin: Evaluation of its binding ability (*in vitro*) and Efficacy as immunomodulator. *Asian-Australian Journal of Animal Science*, 15: 1051.
- Romer, T.R., Boiling, T.M. and MacDonald, J.L. 1978. Gas-liquid chromatographic determination of T-2 toxin and diacetoxyscirpenol in corn and mixed feeds. *Journal of Association of Official Analytical Chemists*, 61: 801.
- Rukmini, C. and Bhat, R.V. 1978. Occurrence of T-2 toxin in *Fusarium* infested sorghum from India. *Journal of Agricultural Food Chemistry*, 26: 647.
- Salini, V.K. and Srinivas, L. 1987. Anti-mutagenic and anti-carcinogenic action of turmeric (*Curcuma longa*). *Journal of Nutrition and Growth Cancer*, 4: 82-89.
- Shotwell, O.L., Hesseltine, C.W. and Goulden, M.L. 1969. Note on the natural occurrence of ochratoxin. *Journal of Association of Official Analytical Chemists*, 52: 81-83.
- Soni, K.B., Rajan, A. and Kuttan, R. 1992. Reversal of aflatoxin induced liver damage by turmeric and curcumin. *Cancer Letters*, 66: 115-121.
- Trenk, H.L., Butz, M.E. and Chu, F.S. 1975. Production of ochratoxin in different cereal products by *Aspergillus ochraceus*. *Applied Microbiology*, 21: 1032.
- Vieira, S.L. 2003. Nutritional implication of mould development in feed stuffs and alternatives to reduce the mycotoxin problem in poultry feeds. *World's Poultry Science Journal*, 59: 111.