

## STUDY OF ADSORPTION POTENTIAL OF YEAST SLUDGE AGAINST AFLATOXINS IN BROILER CHICKS

I. Hashmi, T. N. Pasha, M. A. Jabbar, M. Akram\* and A. S. Hashmi\*\*

Department of Animal Nutrition \*Department of Poultry Production \*\*Department of Physiology & Biochemistry  
University of Veterinary & Animal Sciences, Lahore

### ABSTRACT

A study was conducted to investigate the adsorption potential of yeast sludge against aflatoxins produced in broiler feed. Productive, clinical and immune aspects of broilers fed on aflatoxins with yeast sludge in diet were also studied. Two hundred and forty broiler chicks of (one week age) were fed experimental diets for five weeks. In the present study the sludge has been used @ 1% of feed to reduce the toxic effects of aflatoxins through its adsorption potential. Three levels of aflatoxins (100, 200 and 300 ppb) along with two levels of yeast sludge (0 and 1%) were supplemented in commercial diet. The results showed that the feed intake, weight gain, feed conversion ratio were non-significantly ( $P>0.05$ ) different among birds provided yeast sludge supplemented diet and birds which were not supplemented with yeast sludge. However it was observed that supplementation of yeast sludge increased the level of serum total protein, serum albumin and antibody titer of ND, and reduced the activity of alanine transaminase in serum, aflatoxins concentration in liver and mortality. It was found that 1% yeast sludge worked effectively at 100 and 200 ppb aflatoxins level, but at 300 ppb aflatoxins level the concentration of yeast sludge was not adequate to detoxify the highest level of aflatoxins effectively. However an apparent improvement has been observed. It was also found that if 100 ml yeast sludge contains 8.96 gm yeast cells, it contains 0.26% mannan oligosaccharide, which is the principal compound that bind the aflatoxins.

**Key words:** Aflatoxin, yeast sludge, adsorption

### INTRODUCTION

Rapidly increasing human population and gradually decreasing food resources in under developed countries is making the situation dangerous. The human population in this country is increasing at the rate of 2.9% annually (Anonymous, 1997) which seeks rapid intensification of the food resources. Therefore, there is stressing demand to exploit potential resources of food, better storage facilities and saving the agriculture commodities from spoilage and toxic metabolites. The spoilage of food during storage is a problem, which not only deteriorates the nutrients but also affects the health of human beings and animals. Production of mycotoxins in spoiled food or feed, reduced the availability of nutrients, weight gain, feed conversion ratio and increased the mortality (Huff, 1988).

Mycotoxins include aflatoxins, Zearalenones, Trichothecenes and Ochratoxins which are toxic in nature, secondary metabolites of lower molecular weight produced by naturally occurring fungi (Chu, 1992). The Aflatoxins are highly toxic for man and animals and are the strongest natural poisons. Aflatoxins and other Mycotoxins are heat-stable toxins produced by fungi (Cheeke, 1998). These toxins can be a serious problem in grain stored under unfavorable conditions. In field conditions during storage fungi like *Fusarium* (*Graminearum*, *Culmorum*, *Calviceps purpurea*) on grains produce Zearalenone, Fumonisin, Trichothecene

while during storage fungi like, *Aspergillus flavus*, *Aspergillus parasiticus* and *Penicillium verrucosum* produce Aflatoxins B1, B2, G1, G2 and ochratoxins. However, a rapid explosion in the biotechnology has opened an avenue for biological means of detoxification. Ceigler *et al.* (1966) screened various fungi, bacteria and yeast for their ability to detoxify aflatoxins. Yeast cells can influence health of animal by adsorbing toxins and pathogenic microorganism (Dawson, 1993). In a study it was reported that Modified glucomannan (MGM) had the ability to adsorb more than 70% of the aflatoxins within 30 minutes and 90% within 90 minutes (Murthy and Devegowda, 2004). So, there is a need to utilize the yeast sludge, which is produced as a byproduct of alcohol industry in bulk quantity in Pakistan. The fermented sludge is a good source of yeast and can be used for adsorbing the aflatoxins and other toxic material. This sludge is waste product of distillery industry and normally goes waste. If this sludge is used in the poultry feed it can be very useful in improving the feed efficiency of the birds.

### MATERIALS AND METHODS

Two hundred and forty day-old male unvaccinated broiler chicks were obtained from a commercial hatchery. Individually weighed chicks were divided at random into 24 units, each containing 10 chicks each. Commercial feed was used and

supplemented according to experimental design. The experimental design consisted of 6 dietary treatments: 1) Basal diet plus 100ppb aflatoxins (AF); 2) Basal diet 100 ppb aflatoxins plus 1% yeast sludge (YS); 3) Basal diet with 200 ppb (AF); 4) In basal diet 200 ppb (AF) plus 1% (YS), 5) In basal diet 300 ppb (AF); 6) In basal diet 300 ppb (AF) plus 1% (YS). These 6 diets were assigned randomly to the chicks in a way that there were four replicates on each diet. Aflatoxins were produced from *Aspergillus parasiticus* NRRL 2999 culture through fermentation of rice by the method of Shotwell *et al.* (1996). Successfully fermented rice was then steamed to kill the fungus and dried and ground to a fine powder. The AF content in the rice powder was analyzed and measured with the help of ELIZA. The rice powder was incorporated into the basal diet to provide the required amount of AF level in the diet.

When the chicks reached at one week of age the feeding trial was started and at 6<sup>th</sup> week of age feeding trial was terminated. At the end of experiment two birds were selected randomly from each replicate for blood analysis. Feed intake, weight gain, feed conversion ratio and mortality were observed, while serum total protein, serum albumin, Alanine Transaminase (ALT), Hemagglutination Inhibition titer of ND and aflatoxins concentration in liver were measured. Serum albumin and ALT were assayed through commercial (Randox) kits, while serum total proteins was measured through Biuret method, (Henry *et al.*, 1957) and aflatoxins concentration were measured through spectrophotometrically (Vladimir, 1984). Hemagglutination Inhibition titer of ND was estimated through (Allan and Gouch, 1974) method. The data obtained were statistically analyzed through analysis of variance technique (Steel and Torrie, 1982) using completely randomize design and differences in means were compared through DMR Test (Duncan 1955). Differences were considered to be significant based on the 5% level of probability.

## RESULTS AND DISCUSSION

The results showed that 1% yeast sludge did not improve the feed intake, weight gain and feed conversion ratio at 100, 200 & 300 ppb AF level. Level of serum total protein, serum albumin and alanine transaminase was significantly improved by the supplementation of 1% YS, at 100, 200 & 300 ppb aflatoxins level. However, antibody titer against ND, aflatoxins concentration in liver and mortality showed also improvement in case of 1% YS supplementations at 100, 200 & 300 ppb aflatoxins level.

The present study was in line with the findings of Oguz and Parlat (2004). They observed that supplementation of Mannan oligosaccharide in the chick ration had significantly reduced the adverse effects of aflatoxicosis. Talay *et al.* (2004) also reported that the mortality was reduced with the supplementation of Mannan oligosaccharide. Elizabeth *et al.* (2003) reported no improvement in weight gain, feed intake and FCR while improvement in HI titer of ND on the supplementation of *Saccharomyces cerevisiae* (SCE) and Devegowda (1994) also reported the reduced activity of Alanine transaminase (ALT) in groups of chicks which were fed on diet supplemented with YS 1026.

Aflatoxins decontamination procedures had been focused on degrading, destroying, inactivating or removing AF by physical, chemical and biological methods. Recently, the researchers have directed towards the effective biological elimination process for AF. In this context, live yeast (SCE) was used in controlling the severity of AF and provided significant improvements. The beneficial effects of SCE have been attributed to mannan oligosaccharide (MOS) and the researches then extracted this complex sugar from the cell wall of SCE and modified (Aravind *et al.*, 2003)

**Table-1: Adsorption effect of 1% yeast sludge in broiler chicks fed which was contaminated with (100, 200 and 300 ppb) aflatoxins at 7 to 42 days of age.**

Treatments	Avg. Feed Intake/bird	Avg. Weight Gain/bird	Avg. FCR/bird	Serum Total Protein	Serum Albumin	Alanine Trans-aminase	HI titer of ND	AF conc. In liver (P<0.10)	Mortality
	gm	gm		%	%	U/I		Mg/100gm	%
100AF	2526.45 <sup>b</sup>	1340.62 <sup>c</sup>	1.9 <sup>a</sup>	3.06 <sup>bc</sup>	1.44 <sup>bcd</sup>	49.8 <sup>b</sup>	3.416	4.75 <sup>abc</sup>	25
100AF+1% YS	2493.44 <sup>b</sup>	1340.06 <sup>c</sup>	1.86 <sup>a</sup>	3.84 <sup>d</sup>	1.80 <sup>d</sup>	40.48 <sup>a</sup>	3.583	4.36 <sup>a</sup>	20
200AF	2493.00 <sup>b</sup>	1229.44 <sup>c</sup>	2.03 <sup>a</sup>	2.46 <sup>ab</sup>	1.00 <sup>ab</sup>	60.53 <sup>c</sup>	3.166	5.00 <sup>bcd</sup>	27.5
200AF+1% YS	2397.75 <sup>b</sup>	1207.24 <sup>bc</sup>	1.99 <sup>a</sup>	3.53 <sup>d</sup>	1.57 <sup>cd</sup>	49.05 <sup>b</sup>	3.333	4.51 <sup>ab</sup>	17.5
300AF	1865.46 <sup>a</sup>	941.15 <sup>a</sup>	1.97 <sup>a</sup>	1.98 <sup>a</sup>	0.60 <sup>a</sup>	68.8 <sup>d</sup>	2.50	5.44 <sup>d</sup>	47.5
300AF+1% YS	2030.04 <sup>a</sup>	1049.33 <sup>ab</sup>	1.94 <sup>a</sup>	2.94 <sup>bc</sup>	1.20 <sup>bc</sup>	56.93 <sup>c</sup>	3.00	5.17 <sup>cd</sup>	30

\* = Different superscripts on means show significant difference (P<0.05)

The role of MOS in AF detoxification was attributed to have selective binding capacity for AF molecules, to modulate the immune response. It is hypothesized that esterified glucomannan (EG) might trap the AF molecule in its glucomannan matrix and prevent toxin adsorption from the gastrointestinal tract (Raju and Devegowda, 2000). The single addition of MOS to an AF free diet did not cause considerable negative changes compared to controls. This has also supported that MOS was inert and non-toxic in terms of biochemical-hematological parameters.

In conclusion; it was noted that the 1% yeast sludge which has limited amount (0.26%) of Mannan oligosaccharide showed a little improvement. A significant improvement was noted in serum biochemical values (serum total protein, serum albumin and ALT activity) but not in immune aspects (HI titer of ND, aflatoxins concentration in liver) and productive parameters (feed intake, weight gain and FCR) and mortality percentage. It was observed that 1% yeast sludge act as toxin binder effectively at 100 and 200 ppb aflatoxins level, but its efficiency was reduced at 300 ppb aflatoxins level. So, it was observed that higher levels of yeast sludge would effectively improve the aflatoxicosis condition. However there is a need for improving the adsorption potential of yeast sludge through different processes.

## REFERENCES

- Allan, W.H. and R.E. Gouch, (1974). A standard hemagglutination inhibition for ND (1) A comparison of macro and micro methods. *Vet. Rec.*, 15: 120-123.
- Anonymous, (1997) Economic Survey. 1996-97. Government of Pakistan, Finance Division, Economic Advisor Wing, Islamabad.
- Aravind, K.L., V.S. Patil, G. Devegowda, B. Umakantha and S.P. Ganpule, (2003) Efficacy of esterified glucomannan to counteract ycotoxicosis in naturally contaminated feed on performance and serum iochemical, hematological parameters in broilers. *Poultry Science*, 82:71-576.
- Ceigler, A., E, B.Linllehoj, R.E.Peterson and H.H.Hall. (1996) Microbial detoxification of aflatoxin. *Appl. Microbial*. 14(6)934-939.
- Cheeke, P.R. (1998). Natural toxicants in Feed, forages and Poisonous Plants. 2<sup>nd</sup> ed. Interstate publishers, Danville, IL.
- Chu, S. F. (1992). Recent progress of analytical techniques for mycotoxins in feed stuffs. *J.Amin. Sci.* 70: 3950-3963.
- Dawson, K.A. (1993). Current and future role of yeast culture in animal production: A review of research over the last seven years. In: *Biotechnology in the Feed Industry*, Proc. of Allteche's 9<sup>th</sup> annual symposium. Alltech Technical Publications, Nicholasville, Kentucky.
- Devegowda, G. (1994). Use of *Saccharomyces cerevisiae* cultures to feed. In proc. of 10<sup>th</sup> annual symposium on Biotechnology in the Feed Industry, Loughborough, Leicestershire, England.
- Duncan, K.A. (1955). Multiple range and multiple F test. *Biometrics*. 11: 1-42.
- Elizabeth, A. C. Paulillo; Al. Maiorka; L. S. O. Nakaghi and Marcos macari. 2003. Evaluation of the Efficacy of *Saccharomyces cerevisiae* Cell Wass to Ameliorate the Toxic Effects of Aflatoxin in Broilers. *International J. Poultry Sci.* 2 (5): 341-344.
- Henry, R.J., C. Sobel and S. Berkman. (1957). Determination of protein by the Biuret Reaction. *Anal Chem.* 29: 1491 – 1494.
- Huff, W.E., (1988). Mycotoxin: toxicity and control. US Feed Grains Council. Grain handling and quality workshop. New Delhi, India.
- Murthy, T.N.K., and G. Devegowda. (2004). Efficacy of Modified Glucomannan (Mycosorb) to adsorb aflatoxins B1 in gut condition of broiler chickens, Proc. of Alltech's 19<sup>th</sup> International Feed Industry Symposium, Istanbul.,m turkey. Page 470.
- Oguz, H. and S.S.Parlat. (2004). A study on organic protection of aflatoxicosis in Japanese quail (coturnix Coturnix japonical) by dietary addition of mannan oligosaccharide (Bio-MOS). Proc. of Alltech's 19<sup>th</sup> International Feed Industry Symposium, Istanbul., Turkey.
- Raju, M.V.L.N. and G. Devegowda. (2000). Influence of esterified-glucomannan on performance and organ morphology, serum biochemistry and hematology in broilers exposed to individual and combined mycotoxicosis (aflatoxins, ochratoxin and T-2 toxin). *British Poultry Science*, 41: 640-650.
- Shotwell, O.L., C.W.Hessltine, R.D.Stablefield and W.G.Soreson. (1966). Production of aflatoxins on rice. *Appl. Microbial*. 14:425-428.
- Steel, R.G.D. and J.H. torrie (1982). Principles and procedures of statistics. 2<sup>nd</sup> Ed. International student edition pp-137 and 187. McGraw Hill Koghakusha Ltd. London, U.K.
- Talay, U., M. Bilgig and P.Spring. (2004). effect of mannan oligosaccharide on broilers performance under field conditions in turkey. Proc. of Alltech's 19<sup>th</sup> International Feed Industry symposium, Istanbul. turkey. Page 522.
- Vladimir, B. (1984). Mycotoxin production, isolation, separation and purification. Elsevier, New York PP, 115-116.