Journal of the Hellenic Veterinary Medical Society

Vol 74, No 2 (2023)

The The Efficacy of an Engineered Biocarbon in Young Broiler Chicken During an Aflatoxin Exposure

HO Bayir, C Ritz, J Fowler

doi: 10.12681/jhvms.30080

To cite this article:

Bayir, H., Ritz, C., & Fowler, J. (2023). The The Efficacy of an Engineered Biocarbon in Young Broiler Chicken During an Aflatoxin Exposure . *Journal of the Hellenic Veterinary Medical Society*, *74*(2), 5697–5704. https://doi.org/10.12681/jhvms.30080 (Original work published July 5, 2023)

J HELLENIC VET MED SOC 2023, 74 (2): 5697-5704
ΠΕΚΕ 2023, 74 (2): 5697-5704

Research article
Ερευνητικό άρθρο

The Efficacy of an Engineered Biocarbon in Young Broiler Chicken During an Aflatoxin Exposure

H.O. Bayir[1], C. Ritz[2], J. Fowler[2]

[1]*Ministry of Agriculture and Forestry of the Republic of Turkey*

[2]*The University of Georgia, Department of Poultry Science, Athens, GA*

ABSTRACT: This study was conducted to evaluate the aflatoxin B_1 (AFB_1) adsorption capacity of carbonaceous biomass (Bio-C) in vitro and the efficacy of a Bio-C in mitigating the effects of a 0.5 mg/kg dose of aflatoxins (AF) *in vivo*. *In vitro* AFB_1 affinity of Bio-C was determined by using different AFB_1 concentration-containing solutions, and these concentrations were measured by UV-visible spectrophotometry. In *in vivo* test, a total of 192 Cobb-500 male broilers were obtained on the day of hatch and randomly allocated to one of 32 treatment pens (6 birds/pen). Birds were fed a broiler starter mash diet containing either 0 or 0.5 mg/kg AF, with or without 0.4% of Bio-C, resulting in 4 treatments arranged as complete randomized design with 2×2 full-factorial. On day 21, three birds from each pen were killed by cervical dislocation and the liver, kidney, and spleen were removed and weighed for relative organ weight assessment. Our data indicate that Bio-C adsorption capacity ranged from 29% to 70%, with an average of 44%. *In vivo* results showed that the performance of birds receiving 0.5 mg/kg AF was not significantly different compared to the control group at any point during the 21-day trial. No main effects were seen on the performance parameters by the inclusion of Bio-C in the diet. There was also no AF/Bio-C interaction. The relative weights of the liver, kidney, and spleen were not significantly different whether birds were fed 0.5 mg/kg AF or 0.4% Bio-C. In conclusion, supplementation of Bio-C does not affect broiler performance when provided in diets either free of AF or containing up to 0.5 mg/kg.

Keywords: aflatoxin; biocarbon; binder; broilers

Corresponding Author:
H.O. Bayir, Tarim ve Orman Bakanligi, Universiteler Mah. Cankaya/Ankara,Turkey
E-mail address: haciomer.bayir@tarimorman.gov.tr

Date of initial submission: 29-03-2022
Date of acceptance: 20-11-2022

INTRODUCTION

Crops, such as corn, sorghum, rice, and wheat are major feed sources for poultry worldwide. The risk of these crops being contaminated with mycotoxins has drawn great attention due to the hazardous implications that mycotoxins have and the negative economic impact that they cause on the poultry industry (Rustom, 1997; Manafi, 2012). Aflatoxins (AF) are highly toxic and carcinogenic fungal metabolites produced by the *Aspergillus flavus* and *Aspergillus parasiticus* species. Although the forms of AF can range from AFB_1, AFB_2, $AFG_{1,}$ and AFG_2, the most commonly produced and most toxic is AFB_1. There are no animal species that are immune to the detrimental effects of these toxins once they have contaminated the feed. When exposed, AF induces "aflatoxicosis", a syndrome that results in decreased feed intake and weight gain, immunosuppression, reduced egg production, increased relative liver and kidney weights, impaired serum biochemistry, and eventually mortality (Lee et al., 2012; Eraslan et al., 2006; Jones et al., 1982). Therefore, efforts have been made and varied systems have been developed to detoxify grains and animal feeds that have been contaminated with AF. Once produced, there is no effective way to eliminate AF from the feeds, instead, efforts have been made to minimize their effects through binding or sequestration (Phillips et al., 1988; Oguz et al., 2000).

One of the promising and cost-effective ways to ameliorate the toxicity of AF found in the animal diet is the addition of non-nutritive adsorbent materials. One such feed additive is carbonized biomass (biocarbon or charcoal) (Patil et al., 2014; Galvanoet al., 2001; Jindal et al., 1994). Biocarbon (Bio-C), which has a high porosity and high internal surface area, typically refers to the carbonaceous residue of wood, cellulose, coconut shells, or other by-products left after heating the organic matter in the absence of oxygen (Ioannidou et al., 2007). This odorless and tasteless black powder has been used in previous studies and shown to ameliorate the harmful effects of AF (Jindal et al., 1994; Dalvi and McGowan, 1984; Ramos et al., 1996; Rao and Chopra, 2001; Kana et al., 2010). Therefore, the objective of this study to evaluate the efficacy of an engineered Bio-C when added into the feed for protection against the toxicity of AF in young broilers.

MATERIALS AND METHODS

The experiment was conducted at the University of Georgia Poultry Research Center, with the research protocol approved by the Institutional Animal Use and Care Committee.

An experimental biocarbon was manufactured from Pine wood common to the southern U.S. The material was pyrolized at 400C° and ground to a powder.

In vitro adsorption test:

The procedure as described by Grant and Phillips (1998) was used, with some minor changes: a working solution was prepared at a concentration of 88 mg/kg AFB_1 (Sigma-Aldrich Inc. St. Louis, Missouri, USA) in acetonitrile and then diluted with deionized water to the treatment concentrations of 0.4, 1.6, 3.2, 6.4, 9.6 and 16 mg. A 0 mg blank solution was also prepared using deionized water. The concentration of the treatment solutions was verified by measuring the absorbance of the 365 nm AFB_1 peak in a scan (200-800 nm wavelength) with a UV-visible- spectrophotometer.

To each of the 5 mL of AFB_1 test solution tubes (with the concentrations 0.0, 0.4, 1.6, 3.2, 6.4, 9.6 and 16 mg), 10 mg of the Bio-C was added (0.2%). The concentrations were obtained by dilution of the working solution with deionized water. Each of the samples in this assay was prepared and analyzed in duplicate. After 24 hours of shaking at 200 motions/min on an orbital shaker, the samples were centrifuged at 51,000 g for 30 min and the amount of absorbed AFB_1 was determined, measuring the AFB_1 absorbance of the supernatant at 365 nm (in water-acetonitrile solvent) with UV-visible spectrophotometry. Also, to construct a standard curve, and AFB_1- solution at the concentrations of 0, 0.4, 1.6, 3.2, 6.4, 9.6, and 16 mg were measured.

In vivo study:

The experimental design consisted of 4 treatments, arranged as a 2×2 factorial: T1 (control with no AF and no Bio-C), T2 (0.4% Bio-C), T3 (0.5 mg AF/kg of diet), and T4 (0.5 mg AF/kg of diet plus 0.4% Bio-C), with eight replicates per treatment. A total of 192 Cobb-500 male chicks were obtained on day-of-hatch and randomly distributed among treatment pens (6 birds/pen). The birds were fed a standard broiler starter diet in mash form for 21 days, and all birds were allowed *ad libitum* access to feed and water. Live performance data were recorded at 0, 7, 14, and 21 days of age, and mortality was recorded daily. At the end of the experiment, three birds from each pen were randomly selected and killed by cervical dislocation, and the liver, kidney, and spleen were removed and weighed for relative organ weight assessment.

AF for the treatment diets was produced by the inoculation of *Aspergillus flavus* on rice as described by Shotwell and Hesseltine (1966). The rice was dried and ground into a powder form, diluted with corn meal to a concentration of 0.5 mg/kg, and then added to the diets to provide the desired AF concentration.

Statistical analysis

Data were analyzed as a 2×2 full-factorial for AF level, Bio-C inclusion, and their interaction using the GLM procedures of SPSS. Significant means ($p \leq 0.05$) were separated using Duncan's Multiple Range Tests. Data are presented as means ± SEM. Further, to confirm the effect of exposure to aflatoxin over time, the interaction between binder and aflatoxin with weeks of age was analyzed.

RESULTS

In vitro adsorption test:

The adsorbed amount of AFB_1 over the increasing test doses is shown in Figure 1. The results were fit to a quadratic-ascending broken-line model with an R^2 value of 0.91. At the 0.4 mg/kg dose, Bio-C adsorbed 29% of the AFB_1 in solution. At 1.6 mg/kg, 70% of the AFB_1 in the solution was adsorbed by the Bio-C. At 3.2, 6.4, 9.6 and 16 mg/kg dose, Bio-C adsorbed 61%, 35%, 40% and 30% of the AFB_1 in solution, respectively. The broken-line model showed a peak adsorption capacity for the 0.2% Bio-C in a solution of 4.93 mg/kg AFB_1. Overall, Bio-C adsorbed an average of 44% of AFB_1 in a solution of the range of doses (0.4, 1.6, 3.2, 6.4, 9.6, and 16 mg/kg).

In vivo study:

Table 2 shows growth performance parameters of birds fed 0 or 0.5 mg/kg AF contaminated diets with or without Bio-C supplementation on days 0 to 7. BW, WG, and FCR showed a small difference in birds fed a diet containing 0.5 mg/kg AF compared to the 0 mg control group, but no statistical difference was seen. The addition of Bio-C at 0.4 % of the diet showed an improvement in FI, as indicated by the AF × Bio-C interaction ($p=0.07$). Even though there was a slight increase in FI with the inclusion of the Bio-C, the results were significant only at a p-value of 0.07. There was no significant difference in mortality rate among treatments, but the highest mortality was seen in 0.5 mg/kg AF containing group a rate of 6.25 %.

Table 1. Ingredient and nutrient composition of the experiment (as-fed basis)

Item	Amount
Ingredient (% of diet)	
Corn, grain	57.47
Soybean meal, 48% CP	35.25
Soybean oil	2.63
Limestone	1.59
Dicalcium phosphate	1.39
Salt	0.49
DL-methionine	0.21
L-Lysine-HCI	0.14
Vitamin premix[1]	0.35
Mineral premix[2]	0.08
Calculated composition	
ME, kcal/kg	3050.00
CP, %	21.41
Crude fat, %	4.86
Ca, %	0.95
Available P, %	0.45
Lys, %	1.19
Met, %	0.53

[1]Supplied per kilogram of diet: vitamin A, 5511 IU; vitamin D3, 1102 ICU; Vitamin E, 11.02 IU; vitamin B12, 0.01 mg; Biotin, 0.11 mg; Menadione, 1.1 mg; Thiamine, 2.21 mg; Riboflavin, 4.41 mg; d-Pantothenic Acid, 11.02 mg; Vitamin B6, 2.21 mg; Niacin, 44.09 mg; Folic Acid, 0.55 mg; Choline, 191.36 mg.
[2]Supplied per kilogram of diet: Mn, 107.2 mg; Zn, 85.6 mg; Mg, 21.44 mg; Fe, 21.04; Cu, 3.2 mg; I, 0.8 mg; Se, 0.32 mg.

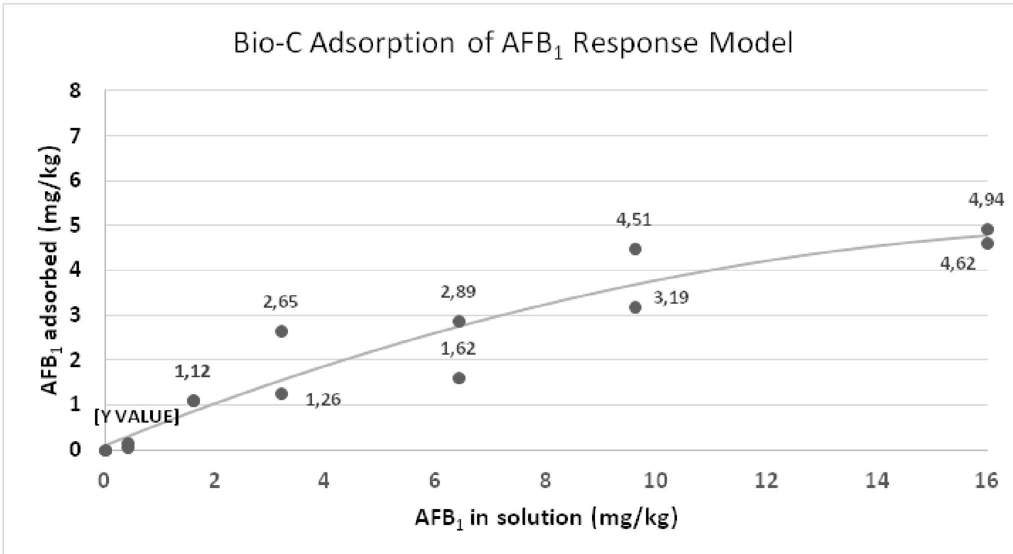

Figure 1. Broken-line (with a quadratic-ascending segment) model for the adsorbed amount of AFB_1 in solution over the increasing test doses

Table 2. Effect of Bio-C on growth performance parameters of birds fed with diets containing 0 or 0.5 mg AF per kg of diet (day 0 to 7)

	Variables				
Treatments	BW (g/bird)	WG (g/bird)	FCR	FI (g/bird)	Mortality (%)
AF (mg)					
[1]0	139±13	100±13	1.19±0.07	119±13	0.00±0.00
[2]0.5	132±22	92±18	1.21±0.09	111±17	6.25±4.38
Binder					
[3]Control	140±12	101±12	1.21±0.06	122±12	2.08±2.10
[4]Bio-C	146±9	107±9	1.18±0.04	126±10	0.00±0.00
P-Value					
AF	0.84	0.84	0.72	0.60	0.40
Bio-C	0.17	0.16	0.72	0.07	0.40
AF x Bio-C	0.22	0.19	0.39	0.23	0.10
[5]SEM	2.69	2.66	0.01	2.32	1.21

[1]0: Basal diet only (no AF and Bio-C), [2]0.5: Basal diet plus 0.5 mg/kg AF, [3]Control: Basal diet plus 0.4% Bio-C
[4]Bio-C: Basal diet plus 0.5 mg/kg AF and 0.4% Bio-C, [5]SEM: Standard error of mean, N: 48 birds/treatment (6 birds/replicate)

Table 3 shows growth performance parameters of birds fed 0 or 0.5 mg/kg AF contaminated diets with or without Bio-C supplementation on days 0 to 14. The presence of 0.5 mg/kg AF in the diet did not impair any of the performance parameters compared to the control group. Statistical analysis also indicated that the birds receiving 0.4% Bio-C in the diet showed no changes in any of the performance parameters. However, the mortality rate was higher in the 0.5 mg/kg AF containing group comparing the others.

Table 4 describes the performance parameters of birds fed 0 or 0.5 mg/kg AF contaminated diets with or without Bio-C supplementation throughout the 21 days of age. Over the entire 21-day feeding period, birds fed the AF contaminated diet showed no statistical effects on their performance parameters. When 0.4% Bio-C was included in the diets, the performance parameters were also not significantly different among treatments (whether they contained 0 or 0.5 mg/kg AF). 0.5 mg/kg AF containing group showed the highest mortality rate among treatments even though there was no statistical difference.

The effects of experimental diets on relative organ weights are presented in Table 5 (grams per 100 g of body weight). The results found that relative liver, kidney, and spleen weights were not affected by dietary AF, and the inclusion of Bio-C in the diet did not show any effect on those values as indicated by the lack of AF × Binder interactions.

Table 3. Effect of Bio-C on growth performance parameters of birds fed with diets containing 0 or 0.5 mg AF per kg of diet (day 0 to 14)

	Variables				
Treatments	BW (g/bird)	WG (g/bird)	FCR	FI (g/bird)	Mortality (%)
AF (mg)					
[1]0	377±35	338±35	1.27±0.08	428±35	0.00±0.00
[2]0.5	376±45	336±45	1.24±0.04	418±46	6.25±4.38
Binder					
[3]Control	386±32	347±32	1.26±0.04	439±36	2.08±2.10
[4]Bio-C	396±24	357±24	1.25±0.05	447±22	0.00±0.00
P-Value					
AF	0.74	0.73	0.31	0.98	0.40
Bio-C	0.25	0.24	0.88	0.13	0.40
AF x Bio-C	0.66	0.63	0.55	0.46	0.10
[5]SEM	6.22	6.19	0.01	6.37	1.21

[1]0: Basal diet only (no AF and Bio-C), [2]0.5: Basal diet plus 0.5 mg/kg AF, [3]Control: Basal diet plus 0.4% Bio-C
[4]Bio-C: Basal diet plus 0.5 mg/kg AF and 0.4% Bio-C, [5]SEM: Standard error of mean, N: 48 birds/treatment (6 birds/replicate)

Table 4. Effect of Bio-C on growth performance parameters of birds fed with diets containing 0 or 0.5 mg AF per kg of diet (day 0 to 21)

	Variables				
Treatments	BW (g/bird)	WG (g/bird)	FCR	FI (g/bird)	Mortality (%)
AF (mg)					
[1]0	810±70	770±70	1.31±0.19	1001±66	0.00±0.00
[2]0.5	844±96	804±96	1.24±0.16	987±66	6.25±4.38
Binder					
[3]Control	854±48	815±48	1.25±0.14	1008±72	2.08±2.10
[4]Bio-C	868±82	828±82	1.26±0.16	1030±37	0.00±0.00
P-Value					
AF	0.37	0.38	0.55	0.86	0.40
Bio-C	0.21	0.21	0.72	0.27	0.40
AF x Bio-C	0.70	0.71	0.51	0.41	0.10
[5]SEM	13.47	13.45	0.03	10.93	1.21

[1]0: Basal diet only (no AF and Bio-C), [2]0.5: Basal diet plus 0.5 mg/kg AF, [3]Control: Basal diet plus 0.4% Bio-C
[4]Bio-C: Basal diet plus 0.5 mg/kg AF and 0.4% Bio-C, [5]SEM: Standard error of mean, N: 48 birds/treatment (6 birds/replicate)

Table 5. Effect of Bio-C as a feed additive on relative organ weights in AF exposed birds on day 21 (g/100 g of body weight)

	Variables		
Treatments	RLW	RKW	RSW
AF (mg)			
[1]0	2.51±0.16	0.55±0.05	0.11±0.02
[2]0.5	2.48±0.22	0.55±0.07	0.12±0.01
Binder			
[3]Control	2.46±0.27	0.54±0.04	0.12±0.01
[4]Bio-C	2.52±0.28	0.55±0.06	0.12±0.02
P-Value			
AF	0.89	0.74	0.66
Bio-C	0.91	0.79	1.00
AF x Bio-C	0.56	0.65	0.20
[5]SEM	0.04	0.01	<0.01

[1]0: Basal diet only (no AF and Bio-C), [2]0.5: Basal diet plus 0.5 mg/kg AF, [3]Control: Basal diet plus 0.4% Bio-C
[4]Bio-C: Basal diet plus 0.5 mg/kg AF and 0.4% Bio-C, [5]SEM: Standard error of mean, N: 48 birds/treatment (6 birds/replicate)

DISCUSSION

The adsorbed amount of AFB_1 over the increasing test doses was determined by in vitro adsorption test. In vitro affinity of Bio-C for AFB_1 ranged from 29 % to 70%. A peak adsorption capacity of 4.93 mg/kg AFB_1 in 0.2% Bio-C in solution. Overall, an average of 44% of AFB_1 in solution (0.4, 1.6, 3.2, 6.4, 9.6 and 16 mg/kg) was adsorbed. Galvano and Pietri (1996) evaluated the efficacy of 17 different activated carbons (ACs), such as exhausted olive residues, peach stones, almond shells, and commercial activated carbons, in binding AFB_1 in solution. The authors used 5 ml of a 0.004 mg/ml aqueous solution of AFB_1 and 2 mg of an AC and found that the adsorption abilities of these 17 ACs ranged from 44.47% to 99.82%.

One of the common indicators of AF exposure is depressed bird performance. The results of this experiment indicate that 0.5 mg/kg AF in the diet did not depress body weight or feed conversion after three weeks. Likewise, Verma et al. (2004) reported no significant effects on performance parameters when birds were fed 0.5 mg/kg AF contaminated diets for 7 weeks. However, the authors observed depressed body weights, reduced feed consumption, and impaired feed efficiency when birds were given 1 and 2 mg/kg of AF in the diets. Magnoli et al. (2011) fed birds with 0 and 0.1 mg/kg AF contaminated diets for 33 days and found no significant differences in performance parameters among treatments. Miller and Wyatt (1985) fed broiler chicks with 0, 1.25, 2.5, and 5 mg/kg AF containing diets for three weeks. The authors found that 1.25 and 2.5 mg/kg AF in the diets did not affect body weights, however, body weight was reduced by 5 mg/kg AF in the diets. Contrary to the results from the present experiment, Bintvihok and Kositcharoenkul (2006) found impaired growth performance in birds fed diets contaminated with 0.05 and 0.1 mg/kg AF. However, in that study, birds were fed AF-contaminated diets for six weeks. Moreover, Huff et al. (1986) fed broiler chickens with diets containing 0, 1.25, 2.5, and 5 mg/kg AF for three weeks and reported that growth performance was depressed only by 2.5 and 5 mg/kg AF in the diet compared to the control at the end of the study. In the current study, body weights between the 0 mg and the 0.5 mg treatment only varied by 4.2%. In most studies in the literature in which BW reduction was reported, the dose of AF in the feed was greater than the dose used in this study. However, the dose selected for this study was still 25x the FDA's advisory action level (0.02 mg/kg). This was done to better reflect a realistic contamination scenario. In addition, since body weight gain and feed intake did not significantly differ, feed conversion ratios were not significantly different among treatments. The inclusion of 0.4% Bio-C in the diet did not have a beneficial effect on growth performance when either added alone or in combination with AF-contaminated diets in the present study. Kubena (1990) and Edrington (1996) studied the efficacy of activated charcoal to ameliorate the toxic effect of AF (7.5 and 0.75 mg/kg, respectively) in broilers and turkeys and indicated no improvements in birds' performance at a 0.5% charcoal dose in the diets in both studies. However, an improvement has been observed in performance parameters of birds fed AF-contaminated diets with inclusion of activated charcoal by (Jindal et al., 1994; Ioannidou et al., 2007; Dalvi and McGowan, 1984), who fed doses of AF ranging from 0.5 to 10 mg/kg. The reason behind these conflicting results is likely due to differences in the duration of feeding, the composition of the feed, the dose and source of AF fed, particular species or strain of the birds, and bird age. Further, Bio-C (or, in some cases, activated charcoal) from different sources are known to have different absorption capacities or physical characteristics. So, the particulars of each product need to be taken into account.

The target organs for aflatoxicosis were the liver, kidney, and spleen in poultry. Our data indicate that the relative weights of those organs were not impaired by dietary AF, no improvement was seen by the addition of Bio-C in diets comparing the control. In contrast, Kubena (1990) fed birds with a 5 mg/kg AF-contaminated diet and reported increased relative weight of the liver and kidney. Further, Edrington (1996) fed turkey poults with diets contaminated with 0.75 mg AF and found decreased liver and increased kidney and spleen weight. However, both authors did not find any beneficial effect with the inclusion of activated charcoal into the diet at a 0.5% level. Also, in both of those studies, the concentration of AF in the diets was higher than the concentration used in the present experiment. It is possible that though the dose used here (of 0.5 mg) is 25-times the FDA action level for aflatoxins, the concentration was still low enough to not affect the relative organ weights or the growth of the birds to any statistically significant degree.

CONCLUSIONS

In conclusion, feeding a 0.5 mg/kg AF contaminated diet did not impair the performance of broilers, nor the relative organ weights, after three weeks. Further, the dietary supplementation of Bio-C (at 0.4%) had no beneficial effect when it was added either in the control diet or the diet containing AF over the rearing period. Based on the results of the present experiment and previous reports, the toxicity of AF on birds appears both doses- and time-dependent, and Bio-C shows a degree of variation in its ability to alleviate the toxic effects of AF in poultry.

ACKNOWLEDGEMENTS

This manuscript is produced from a master thesis entitled Evaluation of an Engineered Biocarbon in Broiler Production written by Haci Omer Bayir.

REFERENCES

Bintvihok A. and Kositcharoenkul S (2006) Effect of dietary calcium propionate on performance, hepatic enzyme activities and aflatoxin residues in broilers fed a diet containing low levels of aflatoxin B1. Toxicon 47(1):41-46.

Dalvi RR and McGowan C (1984) Experimental Induction of Chronic Aflatoxicosis in Chickens by Purified Aflatoxin B1 and Its Reversal by Activated Charcoal, Phenobarbital, and Reduced Glutathione. Poultry Science 63(3):485-491.

Edrington TS, Sarr AB, Kubena LF, Harvey RB, Phillips TD (1996) Hydrated sodium calcium aluminosilicate (HSCAS), acidic HSCAS, and activated charcoal reduce urinary excretion of aflatoxin M1 in turkey poults. Lack of effect by activated charcoal on aflatoxicosis. Toxicology Letters 89(2):115-122.

Eraslan G, Essiz D, Akdogan M, Karaoz E, Oncu M, Ozyildiz Z (2006) Efficacy of dietary sodium bentonite against subchronic exposure to dietary aflatoxin in broilers. Bulletin-Veterinary Institute In Pulawy 50(1):107.

Galvano F, Pietri A, Fallico B, Bertuzzi T, Scirè S, Galvano M, Maggiore R (1996) Activated carbons: in vitro affinity for aflatoxin B1 and relation of adsorption ability to physicochemical parameters. Journal of Food Protection 59(5):545-550.

Galvano F, Piva A, Ritieni A, Galvano G (2001) Dietary strategies to counteract the effects of mycotoxins: a review. Journal of Food Protection 64(1):120-131.

Grant PG, and Phillips TD (1998) Isothermal Adsorption of Aflatoxin B1 on HSCAS Clay. Journal of Agricultural and Food Chemistry 46(2):599-605.

Huff WE, Kubena LF, Harvey RB, Corrier DE, Mollenhauer HH (1986) Progression of Aflatoxicosis in Broiler Chickens. Poultry Science 65(10):1891-1899.

Ioannidou O and Zabaniotou A (2007) Agricultural residues as precursors for activated carbon production—a review. Renewable and Sustainable Energy Reviews 11(9):1966-2005.

Jindal N, Mahipal S, Mahajan N (1994) Toxicity of aflatoxin B1 in broiler chicks and its reduction by activated charcoal. Research in Veterinary Science 56(1):37-40.

Jones FT, Hagler WH, Hamilton PB (1982) Association of Low Levels of Aflatoxin in Feed with Productivity Losses in Commercial Broiler Operations1,2,3. Poultry Science 61(5):861-868.

Kana J, Teguia A, Tchoumboue J (2010) Effect of dietary plant charcoal from Canarium schweinfurthii Engl. and maize cob on aflatoxin B1 toxicosis in broiler chickens. Advances in Animal Biosciences 1(2):462.

Kubena LF, Harvey RB, Phillips TD, Corrier DE, Huff WE (1990) Diminution of Aflatoxicosis in Growing Chickens by the Dietary Addition of a Hydrated, Sodium Calcium Aluminosilicate1. Poultry Science 69(5):727-735.

Lee JT, Jessen KA, Beltran R, Starkl V, Schatzmayr G, Borutova R, Caldwell DJ (2012) Mycotoxin-contaminated diets and deactivating compound in laying hens: 1. Effects on performance characteristics and relative organ weight. Poultry Science 91(9):2089-2095.

Magnoli AP, Texeira M, Rosa CAR, Miazzo RD, Cavaglieri LR, Magnoli CE, Dalcero AM, Chiacchiera SM (2011) Sodium bentonite and monensin under chronic aflatoxicosis in broiler chickens. Poultry Science

90(2):352-357.
Manafi M (2012) Counteracting effect of high grade sodium bentonite during aflatoxicosis in broilers. Journal of Agricultural Science and Technology 14(3):539-547.
Miller BL and Wyatt RD (1985) Effect of Dietary Aflatoxin on the Uptake and Elimination of Chlortetracycline in Broiler Chicks. Poultry Science 64(9):1637-1643.
Odette L, Shotwell C, Hesseltine W, Stubblefield RD, Sorenson WG (1966) Production of aflatoxin on rice. Applied Microbiology 14(3):425-428.
Oguz H, Kurtoglu V, Coskun B (2000) Preventive efficacy of clinoptilolite in broilers during chronic aflatoxin (50 and 100 ppb) exposure. Research in Veterinary Science 69(2):197-201.
Patil RD, Sharma R, and Asrani RK (2014) Mycotoxicosis and its control in poultry: A review. Journal of Poultry Science and Technology 2(1):1-10.
Phillips TD, Kubena LF, Harvey RB, Taylor DR, Heidelbaugh ND (1988) Hydrated sodium calcium aluminosilicate: a high affinity sorbent for aflatoxin. Poultry Science 67(2):243-247.
Ramos AJ, Fink-Gremmels J, Hernandez E (1996) Prevention of toxic effects of mycotoxins by means of nonnutritive adsorbent compounds. Journal of Food Protection 59(6):631-641.
Rao SN, and Chopra R (2001) Influence of sodium bentonite and activated charcoal on aflatoxin M1 excretion in milk of goats. Small Ruminant Research 41(3):203-213.
Rustom IY (1997) Aflatoxin in food and feed: occurrence, legislation and inactivation by physical methods. Food Chemistry 59(1):57-67.
Verma J, Johri TS, Swain BK, Ameena S (2004) Effect of graded levels of aflatoxin, ochratoxin and their combinations on the performance and immune response of broilers. British Poultry Science 45(4):512