Effectivenesss of some Feed Additives for Detoxification of Aflatoxin B1 in Laying Hens Sherif, Kh. E.<sup>1</sup>; R. A. Hasan<sup>2</sup>; M. R. El-Gogary<sup>1</sup> and Rdwa F. S. A. Ismail<sup>1</sup>

<sup>1</sup> Poultry Production Dept., Fac. of Agric., Mans. University

<sup>2</sup> Animal production research institute, Agriculture research center, ministry of agricultural, Dokki, Egypt.



### **ABSTRACT**

An experiment was undertaken to study the influence of dietary Aaflatoxin B1 (AFB1) on layer performance and the efficacy of some non-nutritive feed additives such as chemical Hydrated sodium calcium Alumino Silicat (HSCAS), prebiotic Mannanoligosaccharides (MOS) and lactobacillus acidophilus based probiotic (Biotop) to eliminate the adverse effects of AFB1 on productive and reproductive performance, egg quality and some blood constituents of local laying hens. A total number of 150 Dokki-4 hens plus 30 Dokki-4 cocks 28 weeks old, were divided into 5 groups of 3 replicates (10 hen +1 cock) and housed in floor pens. The remaining 15 cocks were also divided into 5 groups of 3 cocks each and reared separately in wire-cages for semen evaluation and fed the same treated diets. Birds were allotted on the following treatments basal diet ( without any additives), basal diet contaminated with 1 ppm aflatoxin B1/kg diet (AFB1-diet), AFB1-diet+5g HSCAS/kg diet, AFB1-diet+1gMos/kg diet and AFB1-diet+1g Biotop/kg diet. All experimental groups were fed on the experimental diets for 8 weeks as treated period, then they were fed free toxin diet for 4 weeks as a recovery period. Criteria of response were productive traits, egg quality, reproductive traits, semen evaluation, plasma biochemical constituents, AFB1 residue of egg yolk, New castle disease virus(NDV) antibodies titer and Protection percentage. The obtained results could be summarized as follows: The three AFB1-detoxifying agents applied significantly ameliorated the deleterious effect of AFB1, on final body weight and body weight gain. Addition with HSCAS, Mos or Biotop increased egg number/hen, average egg weight and egg mass. The best feed conversion ratio was obtained with the control group and dietary supplementation of HSCAS, Mos, or Biotop to AFB1diet improved (FCR). Feed additives supplementations to AFB1-diet increased yolk weight, shell weight %, shell thickness, yolk color and yolk cholesterol. No significant difference due to treatments among albumen weight %, yolk index %, Haugh unit % and egg shape index %. At the end of the recovery period all egg quality parameters were recovered in groups fed on AFB1-diets during treated period except those which fed on AFB1-diet alone without feed additives for shell-thickness and yolk cholesterol content, where lower than the control and other treatments. The addition AFB1-detoxifying agents significantly improved semen volume, sperm concentration, motility rate, sperm abnormality and dead sperm. Hens fed diets supplemented with HSCAS, Mos or Biotop recorded higher values of fertility, hatchability and chick weight at hatch and lower values of chick abnormality compared to AFB1-diet. After recovery period alterations caused by AFB1-diet were negated for fertility, hatchability and chick weight at hatch but group previously fed AFB1-diet alone without any additives still recorded significantly higher value of chick abnormality compared with control and other treatments. Addition of HSCAS, Mos and Biotop significantly reduced the severity of AFB1 effects by increasing plasma total protein, albumin, globulin, Calcium and phosphorus and decreasing plasma AST, ALT, ALP, Creatinine, cholesterol and uric acid compared to AFB1-diet. AT the end of recovery period after the withdrawal of the contaminated feed, all groups fed the aflatoxicated diet with HSCAS, Mos or Biotop were recovered for all blood parameters except AST, ALT, cholesterol and creatinine. Addition of HSCAS, Mos or Biotop to the aflatoxin contaminated diet significantly ameliorated the harmful effect of aflatoxin on immune response to NDV in all examined samples for hen serm, egg yolk and post-hatch chick. Addition of feed additives (HSCAS, Mos or Biotop) significantly decreased the level of AFB1 residues in egg yolk compared to those fed AFB1-diet alone. After 4 weeks of recovery period, there were no residues in egg yolk. Conclusively, Feeding AFB1 contaminated diet (1 ppm Aflatoxin B1) resulted significant reduction in productive and reproductive performance of Dokki - 4 laying hens. Addition of HSCAS, Mos or Biotop to the aflatoxin contaminated diets significantly ameliorated the harmful effect of aflatoxin and can be recommended as antitoxin for detoxification of AFB1in diets of laying hens. Four weeks recovery period was quite good to improve the laying performance.

**Keywords:** Aflatoxin B1, Hydrated sodium calcium aluminosilicate, Mannan oligosaccharides, probiotic, productive and reproductive performance of laying hens,

## INTRODUCTION

Aflatoxin, a class of mycotoxins which is ubiquitous in nature and continually encountered in feed ingredients (Manafi, et al., 2009). Aflatoxin is a secondary toxic metabolite produced by the fungi Aspergillus flavus and Aspergillus parasiticus (Smith et al., 1995). Aflatoxin B1, is the most toxic among all types of mycotoxins (Sweeney and Dobsm , 1998) as it induces severe economic losses such as immunosuppression, poor growth and feed conversion , increased mortality, decreased egg production, leg problems, liver damage and carcass condemnations (Soliman et al., 2008 and Yarru et al., 2009). Added to that, potential mycotoxin residues were detected in tissues and eggs of birds (Pandey and Chauhan , 2007) and become particularly important as potential hazard for human health .

In poultry, the addition of adsorbents (eg. Hydrated sodium calcium aluminosilicate (HSCAS) to AF contaminated diets significantly reduces the adverse effects of the toxin on chicken performance (Leeson *et al.*, 1995). Adsorbents have been used for years to deactivated AF by immobilization in gastrointestinal tract.

However, for other mycotoxins these so called "mycotoxin binder" have limitations. Biotechnological progress has opened new avenues for tackling this problem. Mannan oligosaccharides (MOS) derived from cells wall of Saccharomyces Cerevisiae, initially used as a performance promoter in the early 1990's was found to have beneficial effect on weight gain and immune response in broilers exposed to AF (Stanley et al, 1993). Mannan oligosaccharides (MOS) also showed considerably high binding ability (80 to 90%) with AF (Mahesh and Devegowda, 1996) and it has been preferred for detoxification of AF in poultry species.

Therefore, microorganisms capable of degradation or biotransforming mycotoxin in the GIT (Schatzmayr 2008).

This study was conducted to show the impact of AFB1 in layer and the efficacy of some non-nutritive feed additives such as chemical (HSCAS), prebiotic (MOS) and probiotic (Biotop) to eliminate the adverse effects of AFB1 on productive and reproductive performance, egg quality and some blood constituents of local laying hens.

## MATERIALS AND METHODS

This study was carried out at Sakha poultry research station, Animal production research institute, Agricultural research center, ministry of agriculture, Dokki, Egypt.

## **Experimental design:**

The present work was designed to investigate the effectiveness of some feed additive for detoxification of aflatoxin contaminated diets. The experiment lasted 12 weeks from 28 weeks of age, 8weeks treated period and 4 weeks recovery period. Three feed additive for detoxification of aflatoxin contaminated diets were used; Hydrated sodium calcium Alumino Silicat (HSCAS)5g/kg diet Mannanoligosaccharides (MOS)1g/kg diet and lactobacillus acidophilus based probiotic (Biotop)1g/kg diet. A total number of 150 Dokki-4 hens plus 30 Dokki-4 cocks 28 weeks old, were leg banded and divided into 5 groups of 3 replicates (10 hen +1 cock) and housed in floor pens .The remaining 15 cocks were also divided into 5 groups of 3 cocks each and reared separately in wire-cages for semen evaluation and fed the same treated diets. All hens were kept under the same managerial and environmental conditions. The experimental mash and tap water were offered ad libitum. Hens received 17 hours of light daily (natural and artificial light ). Birds were allotted on the following treatments: basal diet (without any additives and served as control, Table 1), basal diet contaminated with 1 ppm Aflatoxin B1/kg diet (AFB1-diet), AFB1-diet+5g HSCAS/kg diet, AFB1-diet+1g Mos/kg diet and AFB1-diet+ 1g Biotop/kg diet.

All experimental groups were fed on the experimental diets for 8 weeks as treated period, then they were fed free toxin diets for 4 weeks as a recovery period, to study the withdrawal time required for bringing back the flock to normal production.

## Criteria of response

Productive traits [live body weight, feed consumption, feed conversion (g feed to g egg), egg production and egg mass (g/hen/day)]. Egg quality traits [egg weight (g), yolk weight and its percentage, yolk index, albumen weight and its percentage, Haugh units, shell weight and its percentage and shell thickness (mm)]. Reproductive traits [fertility (%), hatchability and abnormality (%)]. Semen evaluation [semen volume (ml), sperm concentration (billion/ml), mass activity (%), abnormality rate (%) and died (%)]. Plasma biochemical constituents [plasma total protein (g/dl), albumin concentration (g/dl), globulin concentration , AST and ALT, ALP, creatinine , uric acid, cholesterol and

plasma calcium and phosphorus ]. AFB1 residue of egg yolk, New castle disease virus (NDV) antibodies titer and protection percentage.

Table 1. Chemical composition and calculated analysis of the basal experimental diet.

Ingredients	%
Yellow corn	64.84
Soybean meal (44%)	24.60
Limestone	7.60
Di-calcium phosphate	1.70
Sodium chloride	0.30
Vit.&Min. Mixture*	0.30
DL-Methionine	0.06
Clean sand	0.60
Calculated va	ılues**
Metabolizable energy (Kcal/kg)	2723
Crude Protein, %	16.43
Calcium, %	3.30
Available phosphate, %	0.46
Lysine, %	0.88
Methionine, %	0.45
Meth. + Cys., %	0.62
Determined analyses (	dry matter basis)
Dry matter, %	89.51
Crude Protein, %	16.55
Ether extract, %	2.66
Crude fiber, %	3.20
Aflatoxin B1, ppb	5.00

\*Supplied per kg of diet: Vit.A, 10000 IU, D<sub>3</sub>, 2000 IU, Vit.E, 10mg, Vit.K<sub>3</sub>, 1mg, Vit.B<sub>1</sub>, 1mg, Vit.B<sub>2</sub>, 5mg, Vit.B<sub>6</sub>, 1.5mg, Vit.B<sub>12</sub>, 10mcg, Niacin, 30mg, Pantothenic acid, 10mg, Folic acid, 1mg, Biotin, 50ug, Choline, 260mg, Copper, 4mg, Iron, 30mg, Manganese, 60mg, Zinc, 50mg, Iodine, 1.3mg, Selenium, 0.1mg and Cobalt, 0.1mg.

## Statistical analysis:

Data were statistically analyzed by one-way analysis of variance using SAS (2006) statistical software package. Significant difference among means of treatments was detected at 0.05%.

## RESULTS AND DISCUSSION

# Effect of aflatoxin with feed additives on body weight and productive performance:

Averages of egg number/hen/period (En/h/P), egg weight (Ew), egg mass/hen/Period (Em/h/P), feed intake (FI/h/d), feed conversion ratio (FCR) (gram feed required for gram egg) and body weight (BW) and body weight gain (BWG) during the treated and recovery period are illustrated in Tables (2 and 3) for Dokki-4 hens.

Table 2. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on productive performance of Dokki - 4 hen strain during treated period (0 - 8 weeks).

				T-4-1		E				
	Initial	Final	Body	Total	Average	Egg	Feed	FCR	egg	Mantalita
Treatments	body	body	weight	egg	egg	mass	intake	gm feed/	production	Mortality
	weight	weight	change	number	weight	(hen	g/h/d	gm egg	%	number
	(0 wk)	(8 wk)	(0-8)	(hen/period)	(g)	/period)	9 4	5 v88	, •	
Control	1425	1460 <sup>a</sup>	$35.00^{a}$	$37.00^{a}$	$47.30^{a}$	$31.25^{a}$	102.6 <sup>a</sup>	$3.28^{c}$	$66.07^{a}$	-
AFB1-diet	1425	1435 <sup>c</sup>	$10.00^{c}$	$32.00^{\circ}$	$43.50^{c}$	$24.86^{c}$	99.00°	$4.00^{a}$	57.14 <sup>c</sup>	2
AFB1+HSCAS	1428	1448 <sup>b</sup>	$20.00^{\rm b}$	$35.80^{b}$	$46.80^{b}$	$30.00^{ab}$	$101.0^{ab}$	$3.36^{b}$	63.93 <sup>b</sup>	-
AFB1+Mos	1425	$1450^{b}$	$25.00^{b}$	$35.20^{b}$	$46.96^{b}$	$29.52^{b}$	$100.6^{b}$	$3.41^{b}$	62.86 <sup>b</sup>	-
AFB1+Biotop	1420	$1440^{b}$	$18.00^{b}$	$35.00^{b}$	$46.00^{b}$	$28.75^{b}$	$100.8^{b}$	$3.51^{b}$	$62.50^{b}$	-
SEM§	5.42	8.36	3.76	0.52	0.421	0.631	0.835	0.080	0.926	-
Sioni	NS	*	*	*	*	*	*	*	*	*

a-b-C: For each of the main effects, means in the same column with different superscripts differ significantly (P≤0.05).

<sup>\*\*</sup>According to Egyptian Feed Composition Tables (2001)

<sup>§:</sup> Standard error of the means. NS: Not significant; \*: Significant at P≤0.05.

Table 3. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on productive per	formance of
Dokki - 4 hen strain during recovery period (9 - 12 weeks).	

Treatments	Initial body weight (9 wk)	Final body weight (12 wk)	Body weight change (9-12)	Total egg number (hen /period)	Average egg weight (g)	Egg mass (hen /period)	Feed intake g/h/d	FCR gm feed/ gm egg	egg production %	Mortality number
Control	1460 <sup>a</sup>	1470 <sup>a</sup>	$10.0^{c}$	18.8 <sup>a</sup>	$47.50^{a}$	31.89 <sup>a</sup>	102	$3.09^{b}$	67.1 <sup>a</sup>	-
AFB1-diet	1435 <sup>c</sup>	1460 <sup>b</sup>	$25.0^{a}$	$17.0^{\rm b}$	$46.06^{b}$	$27.97^{c}$	100.6	$3.60^{a}$	60.71°	-
AFB1+HSCAS	1448 <sup>b</sup>	1465 <sup>ab</sup>	$17.0^{\rm b}$	$18.70^{a}$	$47.00^{a}$	31.39 <sup>ab</sup>	101.5	$3.23^{\rm b}$	$66.78^{ab}$	-
AFB1+Mos	1450 <sup>b</sup>	$1460^{b}$	$10.0^{c}$	$18.70^{a}$	$47.08^{a}$	31.44 <sup>ab</sup>	100.8	$3.21^{b}$	$66.78^{ab}$	-
AFB1+Biotop	$1440^{\rm b}$	1463 <sup>ab</sup>	$23.0^{a}$	$18.40^{a}$	$47.00^{a}$	$30.89^{b}$	101.2	$3.28^{b}$	65.71 <sup>b</sup>	-
SEM§	8.36	3.53	0.360	0.350	0.621	0.432	0.880	0.176	0.583	
Signi.	*	*	*	*	*	*	N.S	*	*	

a-b-C: For each of the main effects, means in the same column with different superscripts differ significantly (P≤0.05). §: Standard error of the means. NS: Not significant; \*: Significant at P≤0.05.

The results showed that the group of hens fed AFB1-diet recorded significantly lower final body weight and body weight gain values at the end of treated period compared with the control and other treatments. The effect of feed additives on alleviation of the toxic severity of aflatoxin diets on final body weight and weight gain during treated period is clear. Generally, it could be seen that the three AF-detoxifying agents applied HSCAS, Mos or Biotop significantly ameliorated the deleterious effect of AFB1, on final body weight and body weight gain (by 0.91 and 100.0%, 1.05 and 150.0% and 0.35 and 80.0%) respectively. However the three agents significantly varied (p < 0.05) regarding the their protective efficacy. In this respect the protection percentage due to HSCAS inclusion was about (52.0 and 40.0%), Mos (60.0 and 60.0%) and for Biotop, (20.0 and 32.0%) respectively. These results agree with Qota et al, (2005) and Ali et al, (2006) who found significantly redaction in final body weight and body weight gain values due to fed contaminated diet.

Results revealed that the control group laid on the average significantly (p < 0.05) more eggs per hen during the treated period compared to the aflatoxin treated birds with or without HSCAS, Mos or Biotop supplementation. Results showed also that egg number/hen/period (En/h/p) was reduced significantly (p < 0.05) with 1 ppm AFB1, by 13.51%, however addition with HSCAS, Mos or Biotop improved En by 11.88%, 10.0% and 9.38% respectively compared with birds fed AFB1 supplemented diets and gave protection by 76.0%, 64.0% and 60.0% respectively at the end of treated period. The same trend was observed in average egg weight where reduced with AFB1 - diet by 8.03% compared with the control group, however supplementation with HSCAS, Mos or Biotop improved EW by 7.59%, 7.95% and 5.75% respectively compared with birds fed AFB1 supplemented diets and gave protection by 86.84%, 91.05% and 65.79% respectively, thus the control group fed on diets free of aflatoxin without supplementation laid on the average significantly (p < 0.05) heavier egg compared to the aflatoxin treated birds.

Results in Table (2) showed that egg mass laid per hen during the treated period for Dokki - 4 hen strain was decreased significantly (p < 0.05) with aflatoxinB1 in the diet by 20.45% compared to the control group and the decrease was more pronounced in hens fed on diets containing aflatoxin without HSCAS, Mos or Biotop. The addition of HSCAS, Mos or Biotop improve egg mass by (20.68,18.74 and 15.65%) respectively and can protect the

adverse effect of aflatoxin on egg mass by 80.44%, 72.93% or 60.88% respectively.

As presented in Table (2) average of amounts of feed intake (FI/h/d)) during the treated period for Dokki - 4 hens show that hens received diets contaminated with aflatoxin consumed (p < 0.05) lower feed intake by 3.51% during the treated period compared to the control group. Results revealed also in general that HSCAS, Mos or Biotop supplementation to AFB1-diets improve feed intake by (2.02,1.62 and1.82) respectively and gave protection for(FI/h/d)) by 55.56%, 44.44% or 50.0% respectively.

Average of feed conversion ratio (FCR) calculated as gm of feed required for production of one g of eggs (Table 2) show that the best feed conversion ratio (FCR) was obtained with the control group and increased (p < 0.05) with AFB1-diet by 21.95%. While, dietary supplementation of HSCAS, Mos, or Biotop to AFB1-diet improved (FCR) by (16.0 14.75 and 12.25 ) respectively and caused a protection against the adverse effect of aflatoxin for FCR by (88.89%, 81.94% or 68.06%) respectively.

Results of mortality numbers of Dokki - 4 laying hens during treated period as affected with aflatoxinB1 and dietary treatments (Table 2) show that mortality numbers increased in the contaminated diet with aflatoxin compared the other groups which exhibited no mortalities during the treated period.

In general our results indicate that aflatoxin decreased the egg number, average egg weight, egg mass, feed intake, increase mortality number and impair( FCR) in Dokki - 4 laying hens and addition of HSCAS, Mos, or Biotop decreases the adverse effects of the aflatoxin. These results are in agree with the finding of those obtained by Zaghini *et al.*(2005), Ali *et al.*(2006), Pandey and Chauhan (2007) and Verma *et al.*(2004) who observed that egg production and egg weight significantly decreased by aflatoxin contaminated diets. However, kubena *et al.*(1999) and Sahin and Sehu (2007) observed that contaminated diets with AF decreased significantly hen body weight.

Also, Abd EL-Hamid *et al.* (1992), Chowdhury and Smith (2004) and Zhao *et al.* (2010) found that AF-diets impaired feed conversion ratio in hens and laying Japanese quail

During the recovery period (4 weeks), all Dokki - 4 laying hens fed diets without aflatoxin averages of egg number laid per hen per period improved but were still significantly (p < 0.05) lower than that of the control group. The same trend was observed with the average egg

weight (g) where the control hens laid heavier eggs compared to those received previously the aflatoxin contaminated diets. These results, indicate that egg mass laid per hen per period was improved during the recovery period and the improvement was more pronounced in groups received diets with further supplementation with HSCAS, Mos or Biotop. Average of amount of feed intake / hen / day during the recovery period for groups previously treated with AFB1 were approximately similar to the control group. Results revealed also that supplementation with HSCAS, Mos or Biotop improved FCR in groups fed on diets contaminated with aflatoxin B1 (1 ppm/kg) during the treated period.

Average of feed conversion ratio (FCR) during the recovery period ranged between 3.09 to 3.60 g feed required for production one g eggs with significant differences among the treatment groups. Mortality during the recovery period were zero% for the control and all treatments groups. These results indicate in general that a four weeks aflatoxin recovery period was quite good to improve the laying performance and to reduce mortality number in Dokki - 4 laying hens and the improvement was more pronounced in hens fed on diet supplemented with HSCAS, Mos, or Biotop. These results are in agreement with those reported by Ali et al. (2006), Vicente et al.(2007) and Bozkurt et al.(2012) who found that, adding HSCAS, Mos or Biotop to aflatoxin contaminated diets alleviated the adverse effects of aflatoxicosis on chicken body weight, egg number (egg number/hen/day), egg mass (g/h/d), feed intake (FI/h/d), egg production, FCR and reduced mortality numbers during all studied periods.

The depression in productive performance upon feeding aflatoxin could be attributed to reduced protein synthesis as reported by Verma *et al.* (2002), increased lipid excretion in droppings, impaired nutrient absorption and reduced pancreatic digestive enzyme production as reported by Osborne and Hamilton (1981) and reduced appetite by Sharlin *et al.* (1980). *Hasan et al.* (2000) stated that the toxicity of aflatoxin was characterized by reduction in body weight gain as aflatoxin interfere with normal metabolic pathway through the inhibition of protein synthesis and enzyme system that is involved in carbohydrate metabolism and energy release.

Feed consumption in broilers fed on aflatoxin diet was significantly (p< 0.05) decreased and this is suggestive of reduced appetite during aflatoxicosis as a protection mechanism (Rauber *et al*, 2007). Zhao *et al*. (2010) reported that aflatoxin B1 decreased feed intake.

Also, (Santin *et al*,2003) noted that dietary Mos (0.2 to 2g/kg) was an effective agent for detoxification of AF in broilers. The role of Mos in AF detoxification were attributed to have selective binding capacity for molecules to modulate the immune response by providing nutrients to beneficial gut flora and to improve production (Fritts and Waldroup,2003 and Santin *et al*,2003).

Probiotic play their protective effect through production of antimicrobial compounds, reduction of gut PH by stimulating the lactic acid producing micro flora, competition for binding of receptor sites that pathogens occupy, liver function improvement, stimulation of immunemodulatory cells and competition with pathogens for available nutrients. lactobacilli are important for the maintenance of the intestinal microflora, protection from pathogens including many harmful bacteria, viruses and fungi, relief of constipation, and immune stimulation.

Probiotics alter gastrointestinal PH and flora to favor an increased activity of intestinal enzymes and digestibility of nutrients, which may result in increased feed consumption and be reflected as change in metabolic profile (Vicente *et al.*(2007) and Kalavathy *et al.*(2008).

### Effect of aflatoxin with feed additives on egg quality:

Data of egg quality at the end of treated and recovery periods for Dokki - 4 hens are presented in Tables (4 and 5). These results showed that at the end of treated period yolk weight, shell weight %, shell-thickness, yolk color and yolk cholesterol were decreased by 2.18, 12.39% 21.67%, 37.1% and 26.93% respectively, compared to the control group. Results revealed also in general that feed additives supplementations to diet with AFB1, increased these parameters (yolk weight, shell weight %, shellthickness, yolk color and yolk cholesterol) by1.29%, 12.20%, 17.33%, 39.25% and 21.05%, 1.20%, 11.71%, 20.0%, 42.11% and 25.99%, and 1.57%, 12.98%, 20.0%, 38.16% and 25.0% for HSCAS, Mos And Biotop respectively. In this respect the protection percentages due to HSCAS inclusion were (57.75%, 86.21%, 62.65%, 66.54 and 57.10%), Mos (53.52%, 82.76%, 72.29%, 71.38 and 70.51%) and for Biotop (70.42%, 91.72%, 72.29%, 64.68 and 67.83%) respectively. These results indicated that the decreasing of some egg quality parameters was due to aflatoxin affect on the hens by decreasing egg weight and egg mass. These results agreement with those obtained by Verma et al. (2004) who found a reduction in egg weight and shell-thickness for groups of hens fed a diet contains 1-2mg/kg aflatoxin B1.

Table 4. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on some egg quality traits of Dokki - 4 hen strain at the end of treated period.

DUKK	a Then stre	ann at the ch	u or treater	i periou.					
Treatments	Yolk	Albumen	Shell	Shell	Yolk Index	Haugh	Egg shape	Yolk	Yolk cholesterol
Treatments	Weight (%)	Weight (%)	Weight(%)	thickness(mm)	(%)	unit	index (%)	color	(mg/g fresh)
Control	32.50	55.8	$11.70^{a}$	$0.383^{a}$	48.00	80.12	78.6	$7.25^{a}$	13.85 <sup>a</sup>
AFB1-diet	31.79	57.96	$10.25^{c}$	$0.300^{c}$	47.05	79.20	78.0	4.56 <sup>c</sup>	$10.12^{c}$
AFB1+HSCAS	32.20	56.30	$11.50^{b}$	$0.352^{b}$	47.90	79.80	79.0	$6.35^{b}$	12.25 <sup>b</sup>
AFB1+Mos	32.17	56.38	11.45 <sup>b</sup>	$0.360^{b}$	47.86	80.00	78.8	$6.48^{b}$	12.75 <sup>b</sup>
AFB1+Biotop	32.29	56.13	11.58 <sup>b</sup>	$0.360^{b}$	47.90	80.05	78.6	$6.30^{b}$	12.65 <sup>b</sup>
SEM§	0.309	0.462	0.198	0.002	0.631	3.36	3.46	0.002	0.06
Signi.	N.S	N.S	*	*	N.S	N.S	N.S	*	*

a-b-C: For each of the main effects, means in the same column with different superscripts differ significantly ( $P \le 0.05$ ).

<sup>§:</sup> Standard error of the means. NS: Not significant; \*: Significant at P≤0.05.

Table 5. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on some egg quality traits of Dokki - 4 hen strain at the end of recovery period.

Treatments	Yolk	Albumen	Shell weight	Shell	Yolk	Haugh	Egg shape	Yolk	Yolk cholesterol
Treatments	Weight (%)	Weight (%)	(%)	Thickness (mm)	Index (%)	unit	Index (%)	color	(mg/g fresh)
Control	32.60	55.55	11.85	0.384 <sup>a</sup>	48.12	80.05	78.5	7.23	13.90 <sup>a</sup>
AFB1-diet	32.80	56.20	11.00	$0.361^{\rm b}$	48.06	80.00	78.3	7.00	12.85 <sup>c</sup>
AFB1+HSCAS	32.09	56.30	11.61	$0.379^{a}$	48.10	80.08	78.8	7.10	13.13 <sup>ab</sup>
AFB1+Mos	32.10	56.36	11.54	$0.376^{a}$	48.10	80.00	78.9	7.00	13.06 <sup>ab</sup>
AFB1+Biotop	32.10	56.28	11.62	$0.380^{a}$	48.12	80.06	78.8	7.20	12.96 <sup>b</sup>
SEM§	0.425	1.361	2.05	0.002	0.831	2.521	3.621	0.136	0.160
Signi.	N.S	N.S	N.S	*	N.S	N.S	N.S	N.S	*

a-b-C: For each of the main effects, means in the same column with different superscripts differ significantly ( $P \le 0.05$ ). §: Standard error of the means. NS: Not significant; \*: Significant at  $P \le 0.05$ .

The hen fed AFB1-diet recorded shell weight value significantly lower than those fed control diet. The same trend had been found in shell-thickness and these results agree with those obtained by Zaghini *et al.* (2005) who found that the shell weight significantly reduced by aflatoxin. There was a significant differences between different treatments in yolk color and hens fed AFB1-diet recorded the lowest value. The variation in color parameters might be connected to the AFB1 interference with lipid metabolism, carotenoid absorption or deposition in yolk Genedy *et al.* (1999) and Zaghini *et al.* (2005).

Our results revealed thate there were no significant difference due to treatments amonge albumen weight %, yolk index %, Haugh unit % and egg shape index %.

At the end of the recovery period the results showed that all egg quality parameters were recovered (no significant differences Table 5) in groups fed on AFB1-diets during treated period except those which fed on AFB1, (1ppm/kg diet) alone without feed additives for shell-thickness and yolk cholesterol content, where lower than control and other treatments.

Our results revealed that prebiotic (MOS) improved egg quality traits. The beneficial effect on the egg shell quality parameters induced by MOS may be related to the prebiotic influence on the metabolic activity of the beneficial bacteria colonized in the intestinal lumen of hens which positively influenced mineral absorption rate, especially those of Ca and Mg. It can be suggested that the improvement in egg shell quality in this study might be resulted from the increased mineral absorption.

Biotop added to feed for laying hens contributed to an improvement in egg shell quality. Li et al. (2006) found that dried Bacillus subtilis cultures increased egg shell thickness. Panda et al.(2008) also demonstrated that shell quality parameters improved in response to the dietary inclusion of probiotic bacteria (Lactobacillus sporogenes). Abdelgader et al. (2013) reported an increase in eggshell thickness and shell weight as a percentage of total egg weight in laying hens fed dietary Bacillus subtilis. According to Aghaei et al.(2010), Mikulski et al.(2012) and Youssef et al.(2013), probiotic exerted a beneficial influence on eggshell thickness. This beneficial effect may be attributed to a favorable environment in the gastrointestinal tract resulting from the administration of probiotics to birds (Panda et al., 2008 and Mikulski et al., 2012). Probiotics bacteria increase the rate of fermentation and the production of short-chain fatty acid, which reduces the luminal PH (Scholz - Ahrens et al.,2007). Short chain fatty acids stimulate intestinal epithelial cell proliferation and villus height (Garcia et al.2007), which increase absorption efficiency (Scholz-Ahrens *et al.*, 2007). As a result, more nutrients, including calcium, can be assimilated, thus improving egg shell quality. There were significant differences in yolk cholesterol (Table 5). The hens fed AFB1-diet recorded value of yolk cholesterol which was found to be lower than the other treatments and control, these may be as a result to the inhibition of cholesterol biosynthesis.

#### Effect of alfatoxin with feed additives on semen traits:

Data for semen traits of Dokki - 4 cocks at the end of treated period as affected by dietary treatments are shown in Table (6). Results indicated that control group had significantly (p < 0.05) improvement in semen volume (ml), sperm concentration (billion/ml<sup>3</sup>), motility rate (%), sperm abnormality (%) and dead sperm (%) compared to the other treatment groups for Dokki - 4 cocks. Feeding AFB1-diet without feed additives decreased semen volume by 33.02%, sperm concentration by 38.46%, mass motility by 15.29% and increased sperm abnormality by 40.85% and dead sperm by 64.71% compared to the control group. The addition of the three AF-detoxifying agents applied HSCAS, Mos or Biotop significantly improved semen volume (ml), sperm concentration (billion/ml<sup>3</sup>), motility rate (%), sperm abnormality (%) and dead sperm (%) (by 30.81,39.58,12.17,18.8 and 17.14%, 32.7,36.67,11.81,20.0 and 24.86% and 34.6,33.33,11.88,17.8 and 28.57%) for the three agents HSCAS, Mos or Biotop respectively. However, the addition of the three agents HSCAS, Mos or Biotop to aflatoxic-diets gave protection for semen volume by 62.5, 66.35, and 70.19%, for sperm concentration by 63.33, 58.67 and 53.33%, for mass motality by 67.38, 65.38 and 65.77%, for sperm abnormality by 64.83,68.97 and 61.38% and for dead sperm by 43.64,63.27 and 72.73% respectively. From these results, it seems that the severity of AFB1 effects on semen characteristics was decreased by adding the studied additives to AFB1-diets.

Feeding AFB1-diet caused degeneration and a decrease in germinal epithelial cells, disruption in spermatogenesis which might totally or partially suppress spermatogenesis, cause abnormality in spermatozoa and atrophy in testes.

The results of semen quality are in agreement with those previously reported by Sharlin *et al.* (1981) who reported decreased semen volum for mature White Leghorn males fed with 20ppm aflatoxin for five weeks. The observed improvement in sperm concentration in the feed additives fed males, reported herein, might have been due to enhanced availability of nutrients facilitated by more

efficient nutrient absorption at the gastrointestinal tract caused improvement in maturation of sperms in Mos-fed birds (Shashidhara and Devegowda, 2003).

Also, Muthiah (1996) reported that the percentage of sperm abnormality increased when aflatoxin B1 was included in the diet of breeder cocks. Furthermore, several works have reported that sperm count and percent of live sperm were affected by feeding aflatoxin contaminated diet with different levels (Manafi *et al.*(2009) ). Furthermore, several works

have reported improvement in maturation of sperms in Mosfed birds (Shashidhara and Devegowda, 2003).

After 4 weeks recovery period Table (6), alterations caused by AfB1-diet were negated for semen characteristics except males fed previously AFB1-diet alone without any additives still recorded significantly lower values of semen volume, sperm concentration and mass motility and higher values of abnormality and dead sperms compared with control and other treatments.

Table 6. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on semen quality of Dokki - 4 cock at the end of treated and recovery periods.

		Tı	reated period			Recovery period						
<b>Treatments</b>	Volume	Concent.	Mass	Abnormality	dead	Volume	Concent.	Mass	Abnormality	dead		
	(ml)	(Bilion/ml)	Motility %	%	%	(ml)	(Bilion/ml)	Motility %	%	%		
Control	$0.630^{a}$	$3.90^{a}$	85.0 <sup>a</sup>	$7.10^{c}$	4.25 <sup>c</sup>	0.615 <sup>a</sup>	3.82 <sup>a</sup>	$85.00^{a}$	$6.00^{\rm b}$	$4.10^{b}$		
AFB1-diet	$0.422^{c}$	$2.40^{c}$	$72.0^{c}$	$10.00^{a}$	$7.00^{a}$	$0.562^{b}$	$3.15^{b}$	80.15 <sup>b</sup>	8.33 <sup>a</sup>	$5.80^{a}$		
AFB1+HSCAS	$0.552^{b}$	$3.35^{b}$	$80.76^{\rm b}$	8.12 <sup>b</sup>	$5.80^{b}$	$0.610^{ab}$	3.55 <sup>ab</sup>	84.12 <sup>ab</sup>	$6.36^{ab}$	$4.46^{ab}$		
AFB1+Mos	$0.560^{\rm b}$	$3.28^{b}$	$80.50^{b}$	$8.00^{b}$	$5.26^{b}$	$0.608^{ab}$	$3.60^{ab}$	83.88 <sup>ab</sup>	$6.40^{ab}$	$4.38^{ab}$		
AFB1+Biotop	$0.568^{b}$	$3.20^{\rm b}$	80.55 <sup>b</sup>	$8.22^{b}$	$5.00^{b}$	$0.600^{ab}$	3.58 <sup>ab</sup>	$84.00^{ab}$	$6.30^{ab}$	$4.48^{ab}$		
SEM§	0.001	0.27	1.39	0.23	0.42	0.006	0.22	1.28	0.58	0.41		
Signi.	*	*	*	*	*	*	*	*	*	*		

a-b-C: For each of the main effects, means in the same column with different superscripts differ significantly (P≤0.05).

# Effect of aflatoxin with feed additives on fertility and hatchability of eggs

Results for reproductive traits of Dokki - 4 strain as affected by treatments during treated period are presented in Table (7).

Results revealed that feeding the AFB1-diet decreased fertility by (9.42%), hatchability by (23.26%) and chick weight at hatch by (2.86%) and increased abnormalities by (167.58%) compare to the control group. These results may attributed to low concentration of AFB1 transferred into the fertilized eggs might be the cause of serious problems. These results agree with those obtained by Jayakumar *et al.* (1988) and Tiwari *et al.*(1989) who found that AF-dietary exposure resulted in embryonic mortality and reduction in hatchability compared to control.

It is clear from these results that the reduction in reproductive traits especially fertility may be due to impaired semen quality in this study while increasing total death of embryo during hatch period may explain the reduction in hatchability of fertile eggs. These results are in agreement with those obtained by Johri *et al.* (1990) who found reproductive traits impaired by aflatoxicated diets for different chicken strains.

Concerning to the effect of dietary supplementation with aflatoxin Diet, higher values of fertility (7.83%,8.64% and 7.77%), hatchability (21.44%,23.03% and 21.36) and chick weight at hatch (2.24%,1.76% and 1.71) and lower values of chick abnormality (54.45%,51.82% and 55.04%) were found for hens fed diets supplemented with HSCAS, Mos or Biotop compared to AFB1-diet. This increase may be attributed to significant improvement of semen quality. Also, hatchability percentages were significantly (P< 0.05) higher in HSCAS, Mos or Biptop diets compared to the AFB1-diet group. Such increase may depend on egg shell thickness improvement in treated groups.

There were protection against AFB1 on fertility by (75.28,83.15 and 74.72%), on hatchability by (70.75,67.0 and 70.5%),on chick weight at hatch by (76.0,60.0 and 58.0%) and abnormality by (86.95,82.75 and 87.88%) for HSCAS, Mos or Biotop respectively.

Table 7. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on Fertility and hatchability of Dokki - 4 hen strain at the end of treated and recovery periods.

		Tr	eated period			Reco	very period	
Treatments	Fertility	Hatchability	Chick	Chick weight	Fertility	Hatchability	Chick	Chick weight
	%	%	Abnormality %	at hatch (g)	%	%	Abnormality%	at hatch (g)
Control	94.5°	$86.00^{a}$	2.56 <sup>c</sup>	$35.00^{a}$	94.2 <sup>a</sup>	$87.00^{a}$	1.25 <sup>c</sup>	35.50
AFB1-diet	85.6°	$66.00^{c}$	$6.85^{a}$	$34.00^{b}$	$90.50^{b}$	80.52 <sup>b</sup>	$3.20^{a}$	34.96
AFB1+HSCAS	$92.30^{b}$	80.15 <sup>b</sup>	$3.12^{b}$	$34.76^{ab}$	$93.86^{a}$	$86.17^{ab}$	1.86 <sup>b</sup>	35.41
AFB1+Mos	$93.00^{b}$	$81.20^{b}$	$3.30^{b}$	$34.60^{ab}$	$93.50^{a}$	85.20 <sup>ab</sup>	1.90 <sup>b</sup>	35.26
AFB1+Biotop	$92.25^{b}$	$80.10^{b}$	$3.08^{b}$	34.58 <sup>ab</sup>	$93.70^{a}$	85.10 <sup>ab</sup>	$2.00^{b}$	35.30
SEM§	0.621	2.06	0.93	0.162	0.521	2.851	0.005	3.00
Signi.	*	*	*	*	*	*	*	N.S

a, b, c Means with no common superscripts within each column are significantly different (p < 0.05)

These results approach with those reported by Shashidhara and Devegowda (2003) who reported that the MOS influence fertility and hatchability in older breeder female by improving egg shell quality and sperm production in male breeders. After 4 weeks recovery

period (Table 7), alterations caused by AfB1-diet were negated for fertility, hatchability and chick weight at hatch but group previously fed AFB1-diet alone without any additives still recorded significantly higher value of chick abnormality compared with control and other treatments.

<sup>§:</sup> Standard error of the means. NS: Not significant; \*: Significant at P≤0.05.

### Effect of aflatoxin with additives on blood parameters

Results for blood parameters of Dokki - 4 strain at the end of treated and recovery periods as affected by treatments are presented in Table (8 and 9).

## Plasma total protein, albumin and globulin concentration:-

Table (8) showed that, at the end of treated period, experimental group fed on aflatoxinB1- diet significantly (p < 0.05) decreased in plasma total protein, albumin and globulin of Dokki - 4 hens by (27.18,28.57 and 26.05%) respectively, compared with the control group. However, the experimental groups of birds received diet with HSCAS, Mos or Biotop the decrease in plasma total protein, albumin and globulin concentration were declined for birds suffered from aflatoxinB1 at 1ppm/kg diet by (31.71%,36.67% and 27.83% for HSCAS, 31.22%,

37.78% and 26.09% for Mos and 31.71%, 34.44% and 29.57% for Biotop respectively).

The addition of HSCAS, Mos or Biotop to AFB-diet gave protection for plasma total protein by (84.97%,83.66% and 84.97%), albumin by (91.67%,94.44% and 86.11%) and for globulin by (79.01%,74.07% or 83.95%) respectively, at the end of treated period.

Such effect may be due to the metabolism of aflatoxin in the liver, where it interferes with protein synthesis and RNA production, resulting in decreasing albumin and globulin, this resulted from the damage caused by aflatoxin in the liver.

These findings are in agreement with Staynley *et al.*(2004) and Sahin and Sehu (2007) observed that serum total protein decreased and this is due to the harmful effect of aflatoxin.

Table 8. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on some blood constituents of Dokki - 4 hen strain at the end of treated period.

Treatments	Total	Albumin	Globulin	AST	ALT	ALP	Cholest.	Creatinine	Uric acid	Ca	Ph
Treatments	Protein (g/dl)	(g/dl)	(g/dl)	(U/L)	(U/L)	(U/L)	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)
Control	5.63 <sup>a</sup>	2.52 <sup>a</sup>	3.11 <sup>a</sup>	42.50 <sup>c</sup>	10.48 <sup>c</sup>	23.60°	100.6 <sup>b</sup>	$0.526^{c}$	4.20°	12.20 <sup>a</sup>	6.15 <sup>a</sup>
AFB1-diet	$4.10^{c}$	$1.80^{c}$	$2.30^{c}$	85.62 <sup>a</sup>	$38.46^{a}$	51.46 <sup>a</sup>	$150.10^{a}$	1.215 <sup>a</sup>	$6.36^{a}$	$8.50^{c}$	$4.80^{c}$
AFB1+HSCAS	$5.40^{b}$	$2.46^{ab}$	$2.94^{ab}$	$51.25^{b}$	13.11 <sup>b</sup>	$30.60^{b}$	$110.6^{ab}$	$0.660^{b}$	$4.85^{b}$	$10.86^{b}$	$5.50^{b}$
AFB1+Mos	5.38 <sup>b</sup>	$2.48^{ab}$	$2.90^{\rm b}$	$50.00^{\rm b}$	$13.60^{\rm b}$	$29.10^{b}$	116.2 <sup>ab</sup>	$0.581^{b}$	$4.80^{b}$	$10.90^{\rm b}$	$5.60^{b}$
AFB1+Biotop	$5.40^{b}$	$2.42^{b}$	$2.98^{ab}$	$52.00^{b}$	$14.00^{b}$	29.65 <sup>b</sup>	113.8 <sup>ab</sup>	$0.592^{b}$	$4.70^{\rm b}$	$11.00^{b}$	5.76 <sup>b</sup>
SEM§	0.085	0.003	0.076	2.621	2.73	1.36	2.63	0.026	0.112	0.431	0.08
Signi.	*	*	*	*	*	*	*	*	*	*	*

a-b-C: For each of the main effects, means in the same column with different superscripts differ significantly ( $P \le 0.05$ ).

§: Standard error of the means. NS: Not significant; \*: Significant at P≤0.05.

### Transaminase (AST) and (ALT):

Data presented in Table (8) showed that, at the end of treated period, control group were normal values of AST and ALT. Results of the same Table revealed also that, there were significantly (p < 0.05) higher AST and ALT values at the end of treated period with hens fed on AFB1-diet alone without any additives by (101.46% and266.98%) respectively compared with the control group.

On the other hand, hens fed on AFB1-diets with HSCAS, Mos or Biotop had improvement in AST and ALT values of birds suffered from aflatoxin (1ppm/kg diet) by (40.14%, 41.60% and 39.27% for AST and 65.91%, 64.64% and 63.60% for ALT respectively) . This gave protection for AST and ALT by (79.71% and 90.60%) for HSCAS, by (82.61% and 88.85%) for Mos or by (77.97% and 87.42%) for Biotop respectively.

In this respect most increases in ALT activity are associated with hepatocellular damage and also, when cellular degeneration or distraction occurrences in liver cells. So, the increase of ALT level during aflatoxicosis may be due to the effect of aflatoxin on permeability of the liver cell which causes liver cell death and / or liver damage. These results are in agreement with those obtained by Attia *et al.* (1990) found that both AST and ALT were increased at 50 AFB1ppb/kg diet fed for broiler chicks. They indicated that even the low levels of aflatoxin contamination were destructive to the liver, So could increase AST and ALT levels.

#### Alkaline phosphates (ALP):

Alkaline phosphates determination is useful in the diagnosis of obstructive and degenerative hepatic disease, in addition to the consideration of ALP in relation to the

liver. So, increasing ALP was mainly due to the action of the toxin on the liver cells.

Table (8) showed that, treated groups fed on aflatoxicated diets without or with HSCAS, Mos or Biotop had higher (p < 0.05) concentration for ALP compared to those fed control group.

Results of the same Table revealed also that, birds fed AFB1-diet alone had increase in ALP value by 118.05% compared with the control group.

In the present study, the increasing of ALP activity could be a result of damage of liver cells and bile duct obstruction due to proliferation of its cells. This agreement with Mature *et al.*(2010) reported that serum alkaline phosphatase levels were significantly higher in old Ross 308 broiler breeders fed with aflatoxin contaminated diet than those of hens fed the uncontaminated diet.

It was noticed in this study, that treated groups fed on AFB1-diets with HSCAS, Mos or Biotop caused improvement in the ALP values of birds suffered from AFB1, at 1ppm/kg diet by (40.54%,43.45% and 42.38%) respectively. Also ,gave protection by (74.87%,80.26% and 78.28%) for HSCAS, Mos or Biotop respectively. These results are in agreement with those obtained by Abo-Norag *et al.* (1995) who found that dietary combination of 0.5% HSCAS plus 3.5 or 5 ppm AFB significantly diminished the harmful effect of aflatoxicosis for broiler chicks.

#### Plasma creatine and uric acid concentration:

Data presented in Table (8) indicated that, at the end of treated period, birds fed AFB1-diet without additives had significantly (p < 0.05) increase in plasma creatine and uric acid values by (130.99% and 51.43%) respectively, compared to the control group. Hens fed on AFB1-diets with

HSCAS, Mos or Biotop had improvement in plasma creatine and uric acid values of birds suffered from aflatoxin (1ppm/kg diet) by (45.68%, 52.18% and 51.28% for plasma creatine and 23.74%, 24.53% and 26.10% for uric acid respectively). This gave protection for plasma creatine and uric acid values by (80.55% and 69.91%) for HSCAS, by (92.02% and 72.22%) for Mos or by (90.42% and 76.85%) for Biotop respectively. This reults agreement with that of Denli *et al.*(2008) who reported that the increase of uric acid concentration are indicating to the severity of kidney affection and revealed that there was interference with protein metabolism.

### Plasma Calcium and phosphorus concentration:

Data presented in Table (8) show that there were significant differences between treatments in calcium and phosphorus levels. The hens fed AFB1-diet alone recorded lower values of calcium and phosphorus by 30.33% and 21.95% compared with the control group. Addition of HSCAS, Mos or Biotop significantly increased levels of calcium and phosphorus compared to hens fed AFB1-diet alone by (27.76%,28.24% and 29.41% for calcium and 14.58%, 16.67% and 20.0% for phosphorus, respectively). This gave protection by (63.78% and 51.85%), (64.86% and 59.26%) or (67.57% and 71.11%) for HSCAS, Mos, or Biotop , respectively. These results agree with those obtained by Stanley *et al.* (2004) who found that feeding aflatoxin at the rate of 3mg/kg to Cobb broiler breeder hens significantly reduce serum calcium and phosphorus levels.

## Plasma cholesterol concentration:

In this study, treated hens fed diet with AFB1 alone significantly increased Plasma cholesterol by (49.20%) compared with the control group. Addition of HSCAS, Mos, or Biotop significantly decreased plasma cholesterol compared to hens fed the AFB1-diet alone by

(26.32%, 22.58% and 24.18% respectively). This gave protection by (79.80%, 68.48% and 73.33%) for HSCAS, Mos, or Biotop, respectively. (Table 8). These results agree with those obtained by Stanely *et al.* (1993) dealing with broiler chicks found that addition of 5 ppm of AF significantly increased serum cholesterol. On the other hand Stanely *et al.* (2004) reported that feeding aflatoxin at the rate of 3mg/kg to Cobb broiler breeder hens showed a significant decrease in serum total cholesterol.

Generally , addition of HSCAS, Mos and Biotop were significantly (p< 0.05) effective in the protection against aflatoxin B1 by preventing its toxic effect, as was reflected by ameliorating the alterations in plasma biochemical parameters (increasing in plasma total protein, albumin , globulin, Calcium and phosphorus and decreasing in plasma AST,ALT,ALP, Creatinine, cholesterol and uric acid).

Sahin and Sehu (2007) and Soliman *et al.*(2008) demonstrated that the addition of HSCAS to the aflatoxin B1 contaminated diet significantly ameliorated the advers effect of aflatoxin. Supplementation of mannan oligo saccharides at 2gm/kg seems to be essential to block the carry over effect of aflatoxin as reported by Attia *et al.*(2016) who also added that diet supplemented with Lactobacillus acidophilus was the best. Many research has shown that probiotics may be able to lower the deleterious effects but this is still a debatable area of research (Kalavathy *et al.*, 2003).

Data presented in Table (9) revealed that, at the end of recovery period after the withdrawal of the contaminated feed, all groups fed the aflatoxicated diet (1 ppm/kg diet) with HSCAS, Mos or Biotop were recovered for all blood parameters except AST, ALT, cholesterol and creatinine. While the group fed previosly aflatoxicated diet alone (without any additives) contained effective for most blood parameters.

Table 9. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on some blood constituents of Dokki - 4 hen strain at the end of recovery period.

Treatments	Total	Albumin	Globulin	AST	ALT	ALP	Cholest.	Creatinine	Uric acid	Ca	Ph
Treatments	Protein (g/dl)	(g/dl)	(g/dl)	(U/L)	(U/L)	(U/L)	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)
Control	5.68 <sup>a</sup>	2.50	$3.18^{a}$	$42.46^{c}$	$10.60^{c}$	$24.00^{b}$	105.21 <sup>c</sup>	$0.586^{c}$	$4.16^{b}$	$12.17^{a}$	$6.80^{a}$
AFB1-diet	$5.08^{b}$	2.38	$2.70^{\rm b}$	$63.60^{a}$	$20.09^{a}$	$27.17^{a}$	130.68 <sup>a</sup>	$0.869^{a}$	5.13 <sup>a</sup>	$10.06^{b}$	5.13 <sup>b</sup>
AFB1+HSCAS		2.50	$3.03^{ab}$	$53.70^{\rm b}$	$12.31^{b}$	$25.15^{b}$	$110.60^{\rm b}$	$0.636^{b}$	$4.84^{ab}$	11.80 <sup>ab</sup>	5.96 <sup>ab</sup>
AFB1+Mos	5.52 <sup>ab</sup>	2.52	$3.00^{ab}$	$52.36^{b}$	$13.06^{b}$	$24.05^{b}$	109.25 <sup>b</sup>	$0.622^{b}$	$4.58^{ab}$	11.56 <sup>ab</sup>	$5.86^{ab}$
AFB1+Biotop	$5.60^{a}$	2.50	$3.10^{ab}$	53.46 <sup>b</sup>	$12.76^{b}$	$24.68^{b}$	$109.00^{\rm b}$	$0.626^{b}$	$4.76^{ab}$	11.73 <sup>ab</sup>	$6.03^{ab}$
SEM§	0.261	0.131	0.025	2.621	1.621	0.150	3.621	0.082	0.216	0.635	0.486
Signi.	*	N.S	*	*	*	*	*	*	*	*	*

a-b-C: For each of the main effects, means in the same column with different superscripts differ significantly ( $P \le 0.05$ ).

### **Immune response to NDV:**

The effects of experimental treatments on immune response of Dokki - 4 hen to NDV at the end of treated period are presented in Table (10). The determined values of antibodies titer against NDV showed that consuming AFB1 contaminated diet resulted significant reduction in antibody titers against NDV in either hen serum, egg yolk or post-hatch chick as compared to the control by (28.08%, 32.04% and 13.33%). Addition of HSCAS, Mos or Biotop to the aflatoxin contaminating diet significantly ameliorated the harmful effect of aflatoxin on immune response to NDV in all examined samples by (29.52%, 28.0% and 32.0%f0r hen serm,23.65%, 30.51% and 25.45% for egg yolk and 9.62%, 12.69% and 11.15% for post-hatch chick for HSCAS, Mos, or Biotop , respectively.

Results in Table (10) showed that antibodies titer against NDV of egg yolk showed the same trend of serum samples these results suggested that maybe we can use yolk to measure the antibodies titer and get real immune status which will be more easy and applicable since the eggs collection are more easily than collection of serum samples. Furthermore, the presented results showed that post-hatch chicks showed the same trend of immune response of hen, but values of antibodies titer against NDV were higher in egg yolk followed by hen serum and the lowest values recorded in post-hatch chicks of each treatment. Adding HSCAS, Mos and Biotop to contaminated diet decreased the severity of AFB1 effects on NDV antibodies and increased titer values

<sup>§:</sup> Standard error of the means. NS: Not significant; \*: Significant at P≤0.05.

compared with AFB1 group. These finding confirmed that reported with previous authors (Gupta *et al.*, 2003).

Enhancement of the humoral immune response after addition of binders like HSCAS in this investigation is in line with Ibrahim *et al.* (2000) who detected that addition of sodium bentonite binder was significantly effective in ameliorating the negative effect of aflatoxin.

Data presented in Table (10) revealed that, at the end of recovery period after the withdrawal of the contaminated feed, all groups fed the aflatoxicated diet (1 ppm/kg diet) with HSCAS, Mos or Biotop were recovered for values of antibodies titer against NDV. While, the group fed previosly aflatoxicated diet alone (without any additives) still lower than other treatments and control.

Table 10. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin - B1 on immune response to NDV in hen serum, yolk and progeny at a day of hatch of Dokki - 4 hens at the end of treated and recovery periods.

	•	Treated per	riod	Recovery period					
<b>Treatments</b>	Antil	bodies titer ag	Antibodies titer against NDV						
	Serum hen	Egg yolk	Post-hatch chick	Serum hen	Egg	yolk	Post-hatch chick		
Control	$7.30^{a}$	9.02 <sup>a</sup>	$6.00^{a}$	7.26 <sup>a</sup>	8.8	85°	6.10 <sup>a</sup>		
AFB1-diet	5.25°	6.13°	$5.20^{c}$	6.36 <sup>b</sup>	7.8	81 <sup>b</sup>	5.75 <sup>b</sup>		
AFB1+HSCAS	$6.80^{b}$	7.58 <sup>b</sup>	$5.70^{\rm b}$	$7.06^{a}$	8.3	36 <sup>a</sup>	$6.00^{a}$		
AFB1+Mos	$6.72^{b}$	$8.00^{b}$	$5.86^{ab}$	$7.00^{a}$	8.0	$00^{a}$	$6.08^{a}$		
AFB1+Biotop	6.93 <sup>b</sup>	7.69 <sup>b</sup>	5.78 <sup>ab</sup>	7.11 <sup>a</sup>	8.0	$08^{a}$	$6.00^{a}$		
SEM§	0.2631	0.3112	0.1080	0.2811	0	.3000	0.1112		
Signi.	*	*	*	*	:	*	*		

a-b-C: For each of the main effects, means in the same column with different superscripts differ significantly (P≤0.05). 8: Standard error of the means. NS: Not significant: \*: Significant at P<0.05.

### Aflatoxin residue in egg yolk:

Aflatoxin B1 residue which found in egg yolk of hens fed AFB1-diets without or with studied additives (HSCAS, Mos or Biotop) are presented in Table (11).

There was significant differences between experimental treatments, the hens fed AFB1-diet alone recorded the highest value (1.48 micro g/kg), while there were no residues in egg yolk of hens fed the control diet. In this respect, Oliveira *et al.* (2000) found that residues of AFB, were detected only in the eggs of hens given 500 micro g AFB,/kg feed and indicated that the feed: egg AFB, transmission ratio was approximately equals to 5000: 1.0. Oliveira *et al.* (2003) with laying Japanese quail, found that the previous ratio was 3333: 1 for diet containing 100 micro g AFB,/kg feed.However, Zaghini *et al.* (2005) reported that aflatoxin B1 residues was found in eggs of layer hens supplemented with diet containing 2.5ppm aflatoxin B1.

The feed additives (HSCAS, Mos or Biotop) significantly decreased the level of AFB1 residues in egg yolk compared to those fed AFB1-diet alone by (58.65%, 59.12% and 56.08%), respectively.

After 4 weeks of recovery period, there were no residues in egg yolk. These results are in agree with those obtained by Ali *et al.* (2006) found no residues in egg yolk after 4 weeks of recovery period for EL-Salam strain.

Similar results were obtained by Sehu *et al.* (2007) who concluded that HSCAS at 0.5% concentration could significantly and completely ameliorate the performance depressing effect of AFB, as silica binders have been shown to bind the toxins in the digestive tract making them unavailable for gut absorption and allowing the mycotoxin to pass harmlessly through the animals intestinal tract.

Saccharomyces cerevisiae bound as much as 77% of mycotoxins and modified mannan - oligosaccharides (derived from Saccharomyces cerevisiae cell) were found to bind up to 95% of mycotoxin Raju and Devegowda (2000).

In the current study, supplementing a contaminated diet with AFB1 with Biotop significantly ameliorated the toxic effects of AFB1 on layers performance.

The basic mechanism seem to be that lactobacillus casei and lactobacillus acidophilus germinated in animal

tract and secrete the active substance with degrades aflatoxin thus alleviating the effects of aflatoxicosis (Pizzolitto *et al.*, 2011) has been demonstrated that lactobacillus casei strain is able to bind 49.2% of the available aflatoxin in vitro after 4 h of incubation (Hernandez - Mendoza *et al.*, 2009).

Results from the present study demonstrated that the incorporation of Biotop, in hen diet was effective in alleviating of productive performance caused by exposure to AFB1. In general the positive effect of Biotop, additive tested on productive performance are in agreement with the results reported by several researchers (Nayebpor *et al.*, 2007and Mountzouris *et al.*, 2010).

In our study the protective effects of Biotop, against aflatoxin might be due to its capability of effecting a specific biotransformation of aflatoxin in animals intestinal tract for this the amount of aflatoxin absorbed by the intestinal tract is reduced (Yu Fan *et al.*, 2013).

Table 11. Effect of HSCAS, Mos or Biotop as detoxification agent for aflatoxin on AFB1 residue in egg yolk (micro g/kg) at the end of treated and recovery periods.

	Treated period	Recovery period
Treatments	AFB1 residue in	AFB1 residue in
	egg yolk	egg yolk
Control	0.00 <sup>d</sup>	0.00
AFB1 - diet	1.48 <sup>a</sup>	0.00
AFB1 - diet + HSCAS	0.612 °	0.00
AFB1 - diet + Mos	0.605 <sup>c</sup>	0.00
AFB1 - diet + Biotop	0.650 <sup>b</sup>	0.00
SEM§	0.05	0.00
Signi.	*	

a-b-C-d: For each of the main effects, means in the same column with different superscripts differ significantly (P≤0.05).

§: Standard error of the means. NS: Not significant;

\*: Significant at P≤0.05.

## Conclusively, the obtained results showed that:

1. Feeding AFB1 contaminated diet (1 ppm Aflatoxin B1) resulted significant reduction in productive and reproductive performance of Dokki-4 laying hens.

- Addition of HSCAS, Mos or Biotop to the aflatoxin contaminated diet significantly ameliorated the harmful effect of aflatoxin and can be recommended as antitoxin for detoxification of AFB1 in diets of laying hens.
- In general a four weeks aflatoxin recovery period was quite good to improve the laying performance and to reduce mortality number in Dokki-4 laying hens.

### REFERENCES

- Abd EL-Hamid, S., Shakshouk, A.G. Korshem, M., Manakhly, E.M. and Bekhiet, A.B. (1992). Effects of aflatoxin on broiler chickens. Egyptian, J. Poult. Sci., 12:443-469.
- Abdelqader, A., AL-Fata Ftah, A.R. and Dos, G. (2013). Effects of dietary Bacillus subtilis and inulin supplementation on performance, egg shell quality, intestinal morphology and micro flora composition of laying hens in the late phase of production. Anim. Feed Sci. Tech., 179:103-111.
- Abo-Norag, M., Edrington, T.S., Kubena, L.F. and Harvey, R.B. (1995). Influence of hydrated Sodium Calcium aloominosilicate and virginiamycin on aflatoxicosis in broiler chicks. Poult. SCi., 74:626-632.
- Aghaei, A., Tabatabaei, S, Chaji, M. and Nazari, M. (2010). Effects of dried whey (prebiotics) and probiotics on laying hens performance and intestinal flora. J. Anim. Vet. Adv., 9:1996-2000.
- Ali, M. N., E. M. A. Qota., R.A. Hassan and Abou-Elmaged (2006). Novel methods of detoxification of aflatoxin B, in contaminated local laying hen diets. Egypt. Poult. SCi., 26:911-940.
- Attia, A. N., Hegazy, S. M., Awad, Y.L. and Awadalla, S. A. (1990). Effect of aflatoxin contaminated and detoxified ration on liver and kidney function tests of poultry. Egypt. J. Vet. SCI., 27:97-108.
- Attia, Y.A., A.E. Abd Al-Hamid, H.F. Allakany, M.A. Al-Harthi & N.A. Mohamed (2016) Necessity of continuing of supplementation of non-nutritive feed additive during days 21–42 of age following 3 weeks of feeding aflatoxin to broiler chickens, Journal of Applied Animal Research, 44:1, 87-98, DOI: 10.1080/09712119.2015.1013964.
- Bozkurt M, Kucukyilmaz K, Catli AU, Cinar M, Bintas E and Coven F (2012). Performance, egg quality, and immune response of laying hens fed diets supplemented with mannan-oligosaccharide or an essential oil mixture under moderate and hot environmental conditions. Poultry Science, 91: 1379-1386.
- Chowdhury, S.R. & SMITH, T.K., (2004). Effects of feeding blends of grains naturally contaminated with *Fusarium* mycotoxins on performance and metabolism of laying hens. *Poult. Sci.*, 83: 1849–1856
- Denli, M., Blandon, J.C., Guynot, M.E., Salado, S. & Perez, J.F. (2008). Efficacy of a new ochratoxin-binding agent (OcraTox) to counteract the deleterious effects of ochratoxin A in laying hens. *Poult. Sci.*, 87: 2266-2272.

- Fritts CA, Waldroup PW. (2003) Evaluation of Bio-Mos © mannan oligosaccharides as a replacement for growth promoting antibiotics in diet for turkeys. International Journal of Poultry Science, 2. 1:19-22.
- Garcia, V., Catala Gregori, P., Hernandez, F., Megias, M.D. and Madrid, J. (2007). Effect or formic acid and plant extracts on growth, nutrient digestibility, intestine mucosa morphology, and meat yield of broilers. J. Appl. Roult. Res., 16: 555-562.
- Genedy, S.G.K., N. M. El-Naggar., N.S Isshak and E. M. A. Qota (1999). Effect of aflatoxins contaminating agents on performance, blood constituents and some tissues of local poultry strains. Egypt. Poult. SCi., 19:351-377.
- Gupta, K., Ramneek, B., and Singh, A. (2003). Immunomodulatory effects of aflatoxicosis and infectious bursal disease vaccination in broilers. Ind.Vet. J. 80:78-80.
- Hasan, A.A., R. Shahid, H.S. Tassawar and A. Iqbal, 2000. Effect of sodium bentonite as aflatoxin binder in broiler feeds containing fungal infected grains. Pak. J. Agri. Sci., 37(3-4):163-165.
- Hernandez-Mendoza, A., H. S. Garcia, and J. L. Steele. 2009 Screening of Lactocacillus casei strains for their ability to bind aflatoxin B-1 Food Chem. Toxicol. 47:1064-1068.
- Ibrahim, I.K., A.M. Shareef. And K.M.T.AL-Joubory (2000). Ameliorative effects of sodium bentonite on phagocytosis and Newcastle disease antibody formation in broiler chickens during aflatoxcosis. Res. Vet. Sci., 69 (2):119-122.
- Jayakumar, P.M., Valsala, K.V. & Rajan, A. (1988). Experimental aflatoxicosis in the duck with special reference to pathology of the testes. *J. Vet. Sci.*, 19: 122-128.
- Johri, T. S., Grawal, R. and Sadagopan, V. R. (1990). Effect of low dietary levels of aflatoxin on laying quails (Conturnix japonica) and their response to dietary modifications. Indian J. of Anim. SCi., 60(3):355-359
- Kalavathy R., Abdullah. N., Jalaludin S., Wong C.M.V.L. and Ho, Y.W. (2008). Effects of lactobacillus cultures and oxytetracycline on the growth performance and serum lipids of chickens. International J. of Poultry Sci., 7(4):385-389.
- Kalavathy, R.N., Abdullah, N., Jalaludin, S. and Ho, Y.W. (2003). Effects of lactobacillus cultures on growth performance, abdominal fat deposition, serum lipids and weight of organs of broiler chickens. Brit, Poult, Sci., 44:139-144.
- Kubena, L.F., Harvey, R.B., Buckley, S.A. Bailey, R.H. & Rottinghaus, G.E. (1999). Effects of long-term feeding of diets containing moniliformin, supplied by *Fusarium fujikuroi* culture material, and fumonisin, supplied by *Fusarium moniliforme* culture material, to laying hens. *Poult. Sci.*, 78: 1499-1505.
- Leeson., Diaz, G. J. and Summers, J. D. (1995). Poultry metabolic disorders and mycotoxins. Guelph. 1995.
- Li,L., Xu,C.L., Ju,C., Ma,Q., and Jin, Z.K. (2006). Effects of dried Bacillus subtilis culture on egg quality. Poult. Sci., 85:364-368.

- Mahesh, B. K. and Devegowda, G. (1996). Ability of aflatoxin binders to bind aflatoxin in contaminated poultry feed an in vitro study procedings of the 20<sup>th</sup> words Poultry Congress, New Delhi, 4:296.
- Manafi, M, B. Umakantha, H.D. Narayanaswamy and K.Mohan. (2009). Evaluation of high grade sodium bentonite on performance and immune status of broilers, fed with ochratoxin and aflatoxin. World. Myco. J, 2(4):435-440.
- Matur, E., Ergul, E., Akyazi, I., Eraslan, E. & Ciraklit, Z. T. (2010). The effects Saccharomyces cerevisiae extract on the weight of some organs, liver and pancreatic digestiveenzyme activity in breeder hens fed diets contaminated with aflatoxins. Poult. Sci.,89:2213-2220.
- Mikulski, D., Jankowski, J., Naczmanski, J., Mikulska, M. and Demey, V. (2012). Effects of dietary probiotic (Pediococcus acidilactici) supplementation on performance, nutrient digestibility, egg traits, egg yolk cholesterol, and fatty acid profile in laying hens. Poult. Sci., 91:2691-2700.
- Mountzouris K.C., P. Tsitrsikos, I. Palamidi, A. Arvaniti, M. Mohnl, G. Schatzmayr, and K. Fegeros. (2010). Effects of probiotic inclusion levels in broiler nutrition on growth performance, nutrient digestibility, plasma Immunoglobulins, and cecal micro flora composition. Poultry Science 89:58-67.
- Muthiah, J. (1996). Studies on the effect of aflatoxin B1 on reproduction performance of egg type breeders and their amelioration. Ph.D. thesis submitted to Tamil Nadu Veterinary and Animal Science University, Madras, India.
- Nayebpor, M., P. Farhomand, and A. Hashemi. 2007. Effects of different levels of direct fed microbial (PrimaLac) on growth performance and humora; immune response in broiler chickens. J. Anim. Vet. Adv. 6:1308-1313.
- Oliveira, C.A., E. Kobashigawa, T. Reis, L. Mestieri, R. Albuquerque and B. Correa. (2000). Aflatoxin B1 residues in eggs of laying hens fed a diet containing different levels of the mycotoxin. Food Addit. Contam., 17:459-462.
- Oliveira, C.A., J.Rosmaninho, A. Castro, T. Butkeraitis, A. Reia and B. Correa. (2003). Aflatoxin residues in eggs of laying Japanese quail after long-term administration of rations containing low levels of aflatoxin B. Food Addit. Contam., 20:648-653.
- Osborne, D.J. and P.B. Hamilton (1981). Steatorrhea during aflatoxicosis in chickens. Poult. Sci., 60:1398-1407.
- Panda, A.K., Rama, S.S., Raju, M.V and Sharma, S.S. (2008). Effect of probiotic (Lactobacillus sporogenes) feeding on egg production and quality, yolk cholesterol and humoral immune response of white leghorn layer breeders. J.Sci.Food Agric., 88:43-47.
- Pandey, L. and S.S. Chauhan (2007). Studies on production performance and toxin residues in tissues and eggs of layer chickens fed on diets with various concentration of aflatoxin AFB., Br. Poult. Sci., 48:713-727.

- Pizzolitto, R.P., D. J. Bueno, M. R. Armando, L.R. Cavaglieri, A.M. Dalcero, and M. A. Salvano. 2011. Binding of aflatoxin B1 to lactic acid bacteria and Saccharomyces cerevisiae in vitro: A useful model to determine the most efficient microorganism. Aflatoxins Biochemistry and Molecular Bio-Logy. Intech Publications, Croatia, PP.323-346.
- Qota, E. M. A., M. A. Ali., R. A. Hassan and M. K. Abou-Elmaged (2005). Detoxification of aflatoxin contaminated local chicken diets using Aluminosilicate Sodium Sulphate and peroxidase enzyme. 3<sup>rd</sup> International Poultry Conference 4-7 Apr. 2005 Hurgada, Egypt.
- Raju MVLN, Devegowda G. (2000). Influence of esterified - glucomannan of performance and organ morphology, serum biochemistry and hematology in broilers exposed to individual and combined mycotoxicotoxicosis aflatoxin, ochratocin and T2 toxin. Br. Poult. Sci, 41: 640- 650.
- Rauber, R.H., P. Dilkin, L.Z. Giacomini, C.A. Araujo de Almeida and C.A. Mallmann, 2007. Performance of turkey poults fed different doses of aflatoxins in the diet. Poultry Sci., 86:1620-1624.
- Şahin, T. and A Şehu.(2007). Effects of hydrated sodium calcium aluminosilicate (HSCAS) on aflatoxicosis in broilers. Arch.Geflügelk., 71 (2). S. 88–92, 2007, ISSN 0003-9098.
- Santin, E., Paulillo, A.C., Maiorka, A., Nakagui, L.S.O., Macari, M., Saliva, A.V.F. and Alessi A.C. (2003). Evaluation of Saccharomyces serevisiae cell wall to ameliorate the toxic effects of aflatoxin in broilers. International J. of Poult. SCi., 2:341.
- SAS Institute (2006). SAS/STAT User's Guide. Release 9.1. SAS Inst. Inc., Cary, NC.
- Schatzmayr, G. (2008). Latest Innovations in the deactivation of mycotoxins. WPC 2008, Brisbane 30 June 4 July 2008. Aust.
- Scholz Ahrens, K.E., Ade,P,Marten, B,Weber, P,Timm, W,Asil,and Gluer,C.C.(2007). Prebiotics, probiotics and synbiotics affect mineral absorption, bone mineral content and bone structure, J.Nutri., 137: 838-846.
- Sehu A, Ergun L, Cakir S, Ergun E, Cantekin Z, Sahin T, Essiz D, Sareyyupoglu B, Gurel Y, Yigit Y, (2007). HSCAS for reduction of AF in quails Dtsch Tier Wochenschr, 114:252-259.
- Sharlin, J.S., Howarth, B., Thompson, JR. F.N. & Wyatt, R.D. (1981). Decreased reproductive potential and reduced consumption in mature WL males fed aflatoxin. *Poult. Sci.*, 60: 2071-2708.
- Sharlin, J.S., Howarth, B.Jr. and Wyatt, R.D. (1980). Effects of dietary aflatoxin on serum characteristics of mature broiler breeder males. Poult. Sci., 59:1311.
- Shashidhara, R.G., and G. Devegowda. (2003). Effects of dietary mannan oligosaccharide on broiler breeder production traits and immunity. Poult. Sci., 82:1319-1325.
- Smith, J.E., G. Solomons, C. Lewis and J.G. Anderson, (1995). Role of mycotoxins in human and animal nutrition and health. Nat. Toxins, 3:187-192.

- Soliman, E.K.; Hanan, A. Tag El-Din and Abeer, S. Abd El-Rahman, (2008). Effect of hydrated sodium calcium aluminosilicate on egg quality and serum biochemical parameters in table-egg Layers fed on aflatoxin contaminated ration. *Egypt. J. Comp. Path. & Clinic. Path. Vol. 21 No. 4 (December) 2008; 258 282.*
- Stanley, V.G., Winsman, M., Dunckley, C., Daley, M. Kruger, W.F., Sefton, A.E. & Hinton, A. (2004). Impact of yeast culture residue on the supplementation of dietary aflatoxin in performance of broiler breeder hens. J. Appl. Poult. Res., 13: 533-539.
- Stanley, V.G., Woldesenbet, S. and Hutchinson, D.H. (1993). The use of saccharomyces cerevisiae to suppress the effect of aflatoxicosis in briler chicks. Poult. Sci., 72:1867-1872.
- Sweeney, M.J. and A.D. Dobsom. (1998). Mycotoxin production by Aspergillus, fusarium and penicillium species. Inter. J. Food Microbiol, 43(3):141-158.
- Tiwari, R.P., Virdi, J.S., Gupta, L.K., Saini, S.S. & Vadehra, D.V. (1989). Development of chicks exposed to aflatoxin B1 during embryogenesis. *Indian J. Anim. Sci.*, 59: 1473-1478.
- Verma, J., B.K. Swain and T.S. Johri, (2002). Effect of various levels of aflatoxin and ochratoxin A and combinations thereof on protein and energy utilization on broilers. J. Sci. Food Agric., 82:1412-1417.
- Verma, J., T.S. Johri., B.K. Swain, and S. Ameena., (2004). Effect of graded levels of aflatoxin, ochratoxin and their combinations on the performance and immune response of broilers. Br. Poult. Sci. 45:512-518.

- Vicente JL, Avina L, Torres-Rodriguez A, Hargis B, Tellez G. (2007). Effects of a lactobacillus spp-based probiotic culture product on broiler chickens performance under commercial condition. Int J Poult Sci. 6(3):154–156.
- Yarru, L. P., R.S. Sttivari, D.R. Ledoux and G.E. Rottighaus (2009). Toxicological and gene expression analysis of the impact of aflatoxin B, on hepatic function of male chicks. Poult. Sci., 88:360-371.
- Youssef, A.W., Hassan, H.M., Ali, H.M. and Mohamed, M.A. (2013). Effects of probiotics, prebiotics and organic acids on layer performance and egg quality. Asian J. Poult. Sci., 7:65-74.
- Yu Fan, L. Zhao, Qiugang M. Xiaoying L. Huiqin S. Ting Zhou, J. Zhang, and J. Cheng. (2013). Effects of Bacillus subtilis ANSB060 on growth performance, meat quality and aflatoxin residues in broiler fed moldy peanut meal naturally contaminated with aflatoxins food and Chemical Toxicology. 59:748-753
- Zaghini, A., Martelli, G., Ronchada, P. & Rizzi, L. (2005). Mannanoligosaccharides and aflatoxin B1 and M1 residues in fed of laying hens. Effect on egg quality, aflatoxin B1 and M1 residue in eggs and aflatoxin B1 levels in liver. *Poult. Sci.*, 84: 825-832.
- Zhoa J, Shirley RB, Dibner JD Uraizee F, Officer M, Kitchekk M, Vazquez-Anon M, Knight CD, (2010). Comparison of HSCAS and yeast cell wall on counteracting aflatoxicosis in broiler chicks. Poult Sci., 89:2147-2156.

تأثير العلائق الملوثه بالافلاتوكسين على الاداء الانتاجى للدجاج البياض خليل الشحات شريف  $^1$ ، رضا على حسن  $^2$ ، محمد رافت الجوجرى  $^1$  و رضوى فوزى صديق  $^1$  قسم الدواجن — كلية الزراعة — جامعة المنصورة  $^2$  قسم تغنيه الدواجن مركز البحوث الانتاج الحيوانى  $^2$ 

أجريت هذه التجربة لدراسة تأثير العلف المحتوى على افلاتوكسين ب1 على الأداء الإنتاجي للدجاج البياض وكفاءة بعض الإضافات الغذائية مثل هيدريت صوديوم كالسيوم الومنيوسليكات، بريبيوتك (منان اوليجو سكرايد)، وكذلك بروبيوتك (بيوتوب) لتقليل التأثير الضار للافلاتوكسين ب1 على الأداء الإنتاجي والتناسلي وجودة البيض وبعض مكونات الدم على الدجاج البياض. استخدم في هذه الدراسة 150 دجاجة بالإضافة الى 30 ديك دقي 4 عمر 28 أسبوع قسمت الى خمس مجموعات من 3 ثلاث مكررات (10 دجاجات+ديك) سكنت في بيوت أرضية. الخمسة عشر ديك المتبقية قسمت الى خمس مجموعات كلّ منها 3 ديوك وسكنت فرديا في اقفاص معننية لتقييم السائل المنوي وتم تغنيّتها على نفس العلائق التجربيبية. تم توزيع الطيور على المعاملات التالية: المعاملة الاولى غذيت على العليقة الأساسية بدون اى إضافات و المعاملة الثانية غذيت على العليقة الأساسية ملوثة 1 جزء في المليون افلاتوكسين ب1و المعاملة الثالثة غذيت على العليقة الملوثة بالإضافة ألى 5 جرام /كيلو جرام عليقة من مادة هيدريت صوديوم كالسيوم الومنيو سليكك والمعاملة الرابعة غذيت على العليقة الملوثة +1 جرام منان اوليجو سكر ايد/كيلو جرام عليقة اما المعاملة الخامسة غذيت على العليقة الملوثة بالإضافة الى 1 جرام بيوتوب /كيلو جرام عليقة. جميع المجموعات التجريبية تم تغذيتها لمدة 8 أسابيع كفترة معاملة. بعد ذلك غذيت على عليقة خالية من الافلاتوكسين لمدة 4 أسابيع كفترة استشفاء وتم أخذ قياسات الأداء الإنتاجي(جُودة البيض - الأداء التناسلي- تقييم السائل المنوءي- بالإضافة الى بعض مكونات الدم – المتبقي من الإفلاتوكسين ب1 في البيض - وكذلك مقدار الأجسام المناعية ضد مرُض النَبُو كاسلُ - ونسبة الحماية) . وتتلخص أهم النتائج المتحصل عليها في الأتى: [- الإضافات الغذائية الثلاثة للعليقة الملوثة قالت معنويا من التأثير الصار للافلاتوكسين ب1 على وزن الجسم النهائي ومعدل التغير في وزن الجسم 💆 الإضافات الغذائية الثلاث للعليقة الملوثة حسنت من متوسط عدد البيض ووزن البيض وكتلته. 3- افضل نسبة تحويل غذاتي سجلت لمجموعةً الكونترول بينما الثلاث إضافات الغذائية للعليقة الملوثة حسنت معامل التحويل الغذائي. 4- الإضافات الغذائية للعليقة الملوثة أدت الى زيادة وزن الصفار، وزن القشرة وسمكها, لون الصفار ومحتواه من الكولسترول.5- ليس هناك تأثير معنوي للمعاملات بالنسبة لوزن البياض ودليل الصفار ووحدات (هلو) ودليل شكل البيضة في نهاية فترة الاستشفاء 6- جميع قياسات جودة البيض المتأثرة تم استشفائها ما عدا المجموعة التي تم تغذيتها على العليقة الملوثة فقط بدون إضافات غذائية بالنسبة لسمك القشرة ومحتوى الصفار من الكولسنرولّ ظلت اقل من الكوننرول وباقي المعاملات.7- الثلاث إضافات الغذائية حسنت معنويا من حجم القذفة ,وتركيز الاسبرمات وحركيتها ,ونسبة التّشوهات والميتُ. 8- الدَّجَاجاتُ المغذاة على الثَّلَاث إضافات غذائيَّة سجلت قيم أعلى للخصوبة والفقس ووزن الكتكوت عند الفقس وقيم اقل للكتاكيت الغير طبيعية بالنسبة للعليقة الملوثة 9- بعد قَرة الاستشفاء. التغيرات السلبية للتغذية على العليقة الملوثة، اختفتُ في نسبة الخصوبة والفقس ووزن الكتكوت عند الفقس، فيما عدا المجموعة التي تم تغذيتها مسبقا على العليقة الملوثة فقط بدون إضافات ظلت مسجلة معنويا قيم أعلى من الكتاكيت الغير طبيعية بالنسبة لمجموعة الكونترول والمعاملات التجريبية الأخرى. 10- الإضافات الغذائية الثلاث خفضت المعاناة من تأثيرات الافلاتوكسين ب 1 حيث أدت الى زيادة البروتين الكلى للبلازما والالبيومين والجلوبيولين والكالسيوم والفسفور وقالت من اسبارتك ترانس امينيز والالكالين فوسفلتيز والكرياتينين والكولسترول وحامض اليوريك بالمقارنة بالعليقة الملوئة 11\_ في نهاية فترة الاستشفاء وبعد إزالة العليقة الملوثة جميع المجموعات التي نتاولت الإضافات الثلاثة تم الاستشفاء لكل مقليس الدم بإستثناء اسبرتك ترانس امينيز, الانين ترنس امينيز كولسترول, والكريلتنين.12- الإضافات الغذائية قللت معنويا التأثير الضار الاستجابة المناعبة لمرض النيو كاسل في جميع العلائق المختبرة سواء من سيرم الدجاجات ٍأو صفار البيض وسيرم الكتاكيت الفاقسة. 13- الإضافات الغذائية أيضا خفضت بدرجة مخوية المنتقى من الافلاتوكسين ب1 في صفّل البيض بالمقارنة بالدجاجات التي غنيت على العليقة الملوثة بمفردها وبعد اربع أسلبيع من الاستشفاء لم يكن هناك منبقي من الافلاتوكسين ب1 في صفار البيض . نخلص من ذلك بأن1- التغذية على العليقة الملوثة بجزء واحد في المليون من الافلاتوكسين ب إ يخفض من الأداء الإنتاجي والتناه لدجاجات دقي 4 البياضة.2- الإضافات الغذائية المستخدمة للعلائق الملوثة يمكن التوصية بها لخفض التأثير الضار للأفلاتوكسين ب1 في علائق الدجاج البياض.3- أرّبعة أسابيع كفترة استشفائية تعتبر كافية لتحسين الأداء الإنتاجي للدجاج البياض والتخلص من الآثار السلبية للأفلاتوكسين ب1.