

## **USING DIETARY GLUTATHIONE, ALUMINOSILICATE AND TAFLA AS AN ATTEMPT TO PREVENT AFLATOXICOSIS IN LAYING HENS**

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### **ABSTRACT**

An experiment was conducted to evaluate effect of dietary tripeptide glutamate (reduced glutathione (GSH)) as antioxidant, tafla (TF), and hydrated sodium calcium aluminosilicate (HSCAS) as sorbent materials to prevent aflatoxicosis in laying hens. A total number of 371 (350 laying hens+21 cocks) thirty-wk's old El-Salam chickens were randomly divided into 7 groups; each group included 5 replicates of 10 hens each and reared in metallic batteries. The remaining 21 cocks were also divided into 7 groups of 3 cocks each and housed separately for semen collection. Birds were fed practical corn-soybean meal basal diet with or without 1 ppm aflatoxin B<sub>1</sub> (AFB<sub>1</sub>) alone or plus either 5ppm GSH, 0.6% TF, 0.5% HSCAS, 0.6% TF+5ppm GSH or 0.5% HSCAS+ 5ppm GSH to form 7 diets fed from 30 to 38 wks old. Results show that contamination of basal diet with 1 ppm AFB<sub>1</sub> for 8 wks decreased (P<.01) feed intake (25.1%), egg production (42.8%), egg weight (22.3%), shell thickness (32.6%), fertility (21.9%), hatchability of fertile eggs (20%), economic efficiency (EE,38.5%), liver vitamin A (29.1%), blood hemoglobin (35.6%), serum albumin (68%) and total lipids (51%), increased relative liver weight (138.8%), liver lipids (141.9%), blood total leucocytes (WBC's) (28%) and lymphocytes (27.2%) counts, serum enzymatic activities of AST (64%) and ALT (69%), and deposited AFB<sub>1</sub> residues in livers (68 ng/g), egg yolk (52 ng/g) and muscles (36 ng/g) compared to the controls. Adding TF or HSCAS separately into AF diet recorded similar protection effects averaged 45-56% against aflatoxicosis for the studied traits. Including GSH alone into the AF diet resulted in a little protective effects against AF diet for the all studied traits, except AST and ALT activities that showed a significant protective effect (20-28%). However, GSH together with sorbent materials significantly negated the adverse effects of AF diet for all studied traits and improved EE by 81% for TF+GSH and 75% for HSCAS+GSH compared to AF-diet alone. There was mortality only in the both two groups fed basal diet with AFB<sub>1</sub> alone (10%) and AFB<sub>1</sub>+GSH (6%). The present study revealed that TF (available product) presented similar safety protective effect for studied traits and EE as HSCAS (import product). Adding GSH as antioxidant together with TF or HSCAS, to AFB<sub>1</sub> contaminated diet significantly negated aflatoxicosis in the laying hens.

**Keywords:** Glutathione, aluminosilicate aflatoxin B<sub>1</sub>, laying chickens, tafla, residues.

### **INTRODUCTION**

Aflatoxin has elicited greatest public health concern of all mycotoxins because of its widespread occurrence in several grains as corn which comprises 50-60% of poultry diets (Phillips *et al.*,1988), in addition to the role of aflatoxins in the etiology of hepatocellular carcinoma that has been proved (Wild *et al.*,1990). The LD<sub>50</sub> values for AF (mg/kg body weight) were 6.5-16.5 in several chicken strains (Smith and Hamilton, 1970 and El-Samra, 1991). Depression by about 6-30% of chick growth (Edrington *et al.*,1997; Genedy *et*

al., 1999 and Qota, 2003), impairment of feed efficiency (Kubena et al., 1995; Qota, 1999 and Qota et al., 2005) and higher mortality rate (Abdelhamid et al., 1995<sup>a</sup>; Qota, 2003 and Ali et al., 2006) by 0.5-4ppm AF contaminated diet caused very high economic loses. Inhibition of metabolism and immunity system by 0.75-2ppm AF contaminated diet caused increasing liver fat (60% of dry weight) and liver size (2-3 times) and liver damage (Smith et al., 1993 and Abd El-Hamid et al., 1992). The same authors reported also that AFB<sub>1</sub> inhibits DNA synthesis in the liver and possibly prevents proteins synthesis. The HSCAS at 0.5% in the diets has been shown to reduce aflatoxicosis in chicken (Scheideler, 1993; Qota et al., 2005 and Ali et al., 2006). The HSCAS binds AFB<sub>1</sub> *in vitro* (Phillips et al., 1988 and Scheideler, 1993). Thus, the efficacy of sorbent materials as HSCAS or Tafla probably lies in their ability to bind AF in the intestine, rendering the toxin unavailable for absorption (Southern et al., 1994). Ingestion of HSCAS to broilers does not improve skin pigmentation (Brake, 1987). The sorbent additives have raised questions about their effects on minerals and vitamins status, although Chung and Baker (1990) with P, Chung et al. (1990) with riboflavin and Southern et al. (1994) and Qota (2003) with Ca and P, have reported that HSCAS does not impair the nutrient utilization. Glutathione (GSH) is a tripeptide which found almost in its reduced form. It contains an unusual peptide linkage between the amino group of cystine and the carboxyl group of the glutamate. It is an antioxidant, protects cells from toxins such as free radicals during the tissue-damaging peroxidation process and increases enzymatic detoxification in the liver (Wattenberg, 1976). Epoxidation of the 2,3-double bond has been emphasized as a metabolic activation step and recent results indicate that the 2,3-epoxide is a reactive metabolite responsible for reaction with cellular macromolecules, as nucleic acids, and thus may well be the ultimate carcinogen (Swenson et al., 1977). Most damaging epoxidation form is AFB<sub>1</sub> epoxide. The present study was designed to evaluate the effect of TF, HSCAS and/or GSH to prevent aflatoxicosis in laying hens.

## **MATERIALS AND METHODS**

The present study was conducted at Sakha Animal Production Research Station and Laboratories, APRI, ARC, Egypt during Feb.-May 2007 to study the effect of sorbent materials as TF or HSCAS and antioxidants as GSH on laying performance during aflatoxicosis. A total number of 371 (350 hens+21 cocks) thirty-wk's old El-Salam (Nicolas×Mamourah) chickens were divided into 7 similar ( $BW=1450\pm23$  g) experimental groups (5 replicates of 10 hens each) and reared in metallic batteries. The remaining 25 cocks were also divided into 7 groups of 3 cocks each and housed separately for semen collection. Basal diet was formulated to cover nutrient requirements (Table 1) according to Egyptian Feed Composition Tables (2001). Birds were fed basal diet (control) without or with 1ppm AFB<sub>1</sub> alone (AF-diet) or plus either 5ppm GSH (AF+GSH), 0.6% Tafla (AF+TF), 0.5% HSCAS (AF+AS), 0.6% TF+5ppm GSH (AF+TF+GSH) or 0.5% HSCAS+ 5ppm GSH (AF+AS+ GSH) to form 7 experimental diets fed from 30 to 38 wk's old. The AF was produced

via fermentation of rice by *Aspergillus parasiticus* NRRL 2999 as described by Shotwell *et al.* (1966) and modified by West *et al.* (1973). Fermented rice was autoclaved, dried, ground and analyzed for its AFB<sub>1</sub> content (Nabney and Nesbitt, 1965) as modified by Wiseman *et al.* (1967). The AFB<sub>1</sub> in the rice powder was extracted by chloroform then incorporated into basal diet and confirmed by HPLC to provide the desired level of 1 ppm. The GSH was provided from Sigma-Aldrich Quimica S.A. Madrid 28100, Spain. The HSCAS was purchased from Integrated World Enterprises Co.. Tafla (available natural substance) was washed, grounded to a fine powder. Chemical analysis for HSCAS and tafla (Table 1) were done (AOAC, 1990). Feed intake, egg number and egg weight were recorded weekly.

**Table 1. Composition of the laying hen basal diet, HSCAS and Tafla.**

Ingredient	%	Composition <sup>3</sup>	HSCAS (%)	Tafla (%)
Yellow corn	64.84	Silica	64.70	59.80
Soybean meal, 44%	24.60	Aluminum	15.50	17.20
Dicalcium phosphate	1.70	Iron	1.75	2.30
Limestone	7.60	Calcium	1.26	1.90
NaCl	0.30	Potassium	1.80	2.30
Vit. + Min. Mix. <sup>1</sup>	0.30	Sodium	2.55	2.90
DL-Methionine	0.06	Magnesium	1.54	1.90
Clean sand	0.60	Moisture	10.56	10.32
<b>Calculated values<sup>2</sup>:</b>		Price, LE/kg	15.0	0.50
ME, Kcal/Kg	2723			
Lysine, %	0.88	<sup>1</sup> Vitamins +Minerals Mixture provided /kg of diet: 6000 IU vit A, 2000 ICU cholecalciferol, 10 IU vit E, 2.5 mg vit K <sub>3</sub> , 2.5 mg riboflavin, 12 mg nicotinic acid, 10 mg Ca pantothenate, 300 mg choline chloride, 4 µg cyanocobalamin, 5 mg pyridoxine, 3 mg thiamine, 0.5 mg folic acid, 0.2 mg biotin, Trace mineral (mg/kg of diet): 40 Mn, 40 Zn, 40 Fe, 4 Cu, 0.2 Se, 0.5 I.		
Meth. + Cys., %	0.62			
Av. Phosphorus, %	0.46			
Calcium, %	3.30			
<b>Determined analyses<sup>3</sup>:</b>		<sup>2</sup> According to Egyptian Feed Composition Tables (2001).		
Dry matter, %	89.51			
Crude protein, %	16.55			
Ether extract, %	2.66			
Aflatoxin B <sub>1</sub> , ppb	6.0			
Price, LE/Kg	1.55	<sup>3</sup> According to AOAC (1990).		

Shell thickness using Micrometer were estimated 3 times (18-days intervals) using 50 eggs/group/time and the yolk was separated for analysis. Hens were artificially inseminated once a week. Eggs laid were collected daily, stored for 7days at 15°C and 70% RH then incubated to estimate fertility and hatchability of fertile eggs. At the end of the experiment, 5 hens /group were slaughtered for tissues analyses. Liver lipids were extracted (Folch *et al.*, 1957). Liver vit. A content was estimated (Thompson *et al.*, 1971). Also, AFB<sub>1</sub> residues in fresh meat (breast:thigh, 1:1), liver and egg yolk were measured (Stubblefield *et al.*, 1982). Economic efficiency was calculated based on feed cost. Blood hemoglobin (Kampen and Zijlestra, 1961), leucocytes (WBC's) and lymphocyte counts (Wintrobe, 1969), serum albumin (Doumas *et al.*, 1977), total lipids (Chabrol and Charonnat, 1973) and alanine aminotransferase (ALT) and aspartate aminotransferase (AST) enzymatic activities (Reitman and Frankel, 1957) were measured using commercial kits. Data were statistically analyzed using one-way ANOVA of GLM procedure of

Statistical Analysis Software (SAS, 1994). Before analysis, all percentages were subjected to logarithmic transformation ( $\log_{10}x+1$ ) to approximate normal distribution. Significant differences among treatment means were ( $p \leq 0.05$ ) separated by Duncan's new multiple range test (Duncan, 1955).

## RESULTS AND DISCUSSIONS

### Productive and reproductive performance and economic efficiency:

Data presented in Table (2) showed that there were similar trends for treatments effect on the studied traits. It is clear that AF-diet ( $P \leq 0.05$ ) impaired feed intake (25.1%), egg production (42.8%), egg weight (22.3%), shell thickness (32.6%) eggs fertility (21.9%), hatchability of fertile eggs (20%) and economic efficiency (EE) (38.5%) compared to the controls. Inclusion of 0.6% TF or 0.5% HSCAS separately with AF-diet recorded similar protection effects ( $P \leq 0.05$ ) averaged 47-56% for productive and reproductive traits against aflatoxicosis. Adding 5 ppm GSH alone with AF-diet had little protective effects on laying performance traits studied. While GSH with TF or with HSCAS supplemented to AF-diet significantly prevented aflatoxicosis as assessed by performance traits (Table 2). Supplementing sorbent materials plus GSH with AF-diet improved EE by 81% for TF+GSH and 74.9% for HSCAS+GSH compared to AF-diet alone. There was mortality only in the both two groups fed basal diet with AFB<sub>1</sub> alone (10%) and AFB<sub>1</sub>+GSH (6%). Many authors used 1-3 ppm AF (Edrington *et al.*, 1997; Genedy *et al.*, 1999 and Ali *et al.*, 2006) with different chicken strains and showed similar deterioration in laying performance traits by AF-diet.

**Table 2. Productive and reproductive performance of El-salam laying hens fed dietary treatments from 30 to 38 wk's old.**

Dietary treatment <sup>1</sup>	Feed intake (g/b/d)	Egg prod. (%)	Egg wt. (g)	Shell thick. (mm)	Fert-ility (%)	Hatchabi- lity (%)	EE* (%)
Control	112.2 <sup>a</sup>	66.8 <sup>a</sup>	53.0 <sup>a</sup>	0.371 <sup>a</sup>	89.6 <sup>a</sup>	84.0 <sup>a</sup>	64.33 <sup>a</sup>
AF-diet	84.1 <sup>c</sup>	38.2 <sup>c</sup>	41.2 <sup>c</sup>	0.250 <sup>c</sup>	70.0 <sup>c</sup>	67.2 <sup>c</sup>	39.58 <sup>e</sup>
AF+GSH	92.0 <sup>bc</sup>	45.4 <sup>bc</sup>	44.1 <sup>bc</sup>	0.301 <sup>bc</sup>	74.7 <sup>bc</sup>	71.4 <sup>bc</sup>	45.36 <sup>d</sup>
AF+AS	99.7 <sup>b</sup>	52.3 <sup>b</sup>	47.0 <sup>b</sup>	0.324 <sup>b</sup>	79.9 <sup>b</sup>	75.4 <sup>b</sup>	52.73 <sup>c</sup>
AF+TF	100.1 <sup>b</sup>	52.6 <sup>b</sup>	46.9 <sup>b</sup>	0.310 <sup>b</sup>	80.3 <sup>b</sup>	74.6 <sup>b</sup>	53.61 <sup>c</sup>
AF+AS+GSH	106.2 <sup>ab</sup>	59.8 <sup>ab</sup>	50.1 <sup>ab</sup>	0.352 <sup>ab</sup>	84.6 <sup>ab</sup>	79.8 <sup>ab</sup>	58.11 <sup>b</sup>
AF+TF+GSH	106.1 <sup>ab</sup>	60.1 <sup>ab</sup>	49.9 <sup>ab</sup>	0.343 <sup>ab</sup>	85.0 <sup>ab</sup>	79.6 <sup>ab</sup>	59.62 <sup>b</sup>
SEM	1.97	0.842	1.08	0.005	1.14	2.14	0.471
P value	0.007	0.002	0.004	0.003	0.006	0.005	0.014

<sup>a-d</sup> Values followed by different letters within columns are significantly different ( $P \leq 0.05$ ).

<sup>1</sup>AF=1ppm Aflatoxin B1, TF=0.6% Tafla, AS=0.5% Hydrated sodium calcium aluminosilicate (HSCAS), GSH=5ppm Tripeptide glutamate (Glutathione). Values are means of 5 determinations.

\*EE (Economic efficiency)= [Total revenue (number of newly healthy hatched chicks  $\times$  its price (1.15 LE) + (useless eggs for incubation  $\times$  its price (0.30 LE)) per hen – Total feed cost (Total feed intake  $\times$  its price, LE/hen)  $\div$  Total feed cost]  $\times 100$ .

Lack of essential nutrients such as minerals and vitamins, as a result of feed intake decrease, and inhibition of metabolism and immunity system by aflatoxicosis may explain the present impairments of egg production, shell thickness and reproductive traits as those showed by Smith and Hamilton (1970). Regarding sorbent materials protection, the present results confirmed

those of Genedy *et al.* (1999) and Ali *et al.* (2006). They showed, by working on different chicken strains, that adding 0.5% HSCAS to basal diet contaminated with AF did diminished aflatoxicosis impact on productive and reproductive traits by about 50-60%. Tafla and HSCAS had similar ( $p \leq 0.05$ ) protective effect against aflatoxicosis as they contain similar composition (Table 1). They sorbed AFB<sub>1</sub> selectively during the digestive process, which rendered most of the AF unavailable for absorption from the gastrointestinal tract as those reported by Huff *et al.* (1992), Kubena *et al.* (1993) and Qota (2003).

#### Liver status and hematological traits:

Contaminating the basal diet with 1 ppm AFB<sub>1</sub> significantly ( $P \leq 0.05$ ) increased relative liver weight (138.8%), liver lipids (141.9%), blood total leucocytes (WBC's) (28%) and lymphocytes (27.2%), decreased liver vitamin A (29.1%), and hemoglobin (Hb) contents (35.1%) compared to the controls (Table 3). Supplementing 0.6% TF or 0.5% HSCAS separately to the AF diet resulted in a significant protection against aflatoxicosis by about 51.0, 50.7, 50.1, 53.3, 48.8 and 51.8% for liver weight, liver lipids, liver vitamin A, WBC's, lymphocytes and Hb, respectively, compared to the controls. Insignificant protective effects were recorded for liver status and hematological traits by adding GSH alone to the AF diet. Supplementing GSH with TF or HSCAS into the AF diet significantly negated AF effects on the liver and hematological traits (Table 3). The present results confirmed those of Abdelhamid *et al.* (1995<sup>a</sup>) with chickens and Qota (2003) with turkey who reported similar alterations in liver status by 0.5-2.5 ppm AF diets. Increasing liver weight in the present study may be due to increase the accumulation of fat as a result of interference of AF with lipid metabolism as reported by Smith and Hamilton (1970). In the same manner, Abd El-Hamid *et al.* (1992) reported that aflatoxicosis impaired fat transport which could attribute to inhibited RNA synthesis that caused a marked increase in liver fat content. Decrease liver vitamin A content caused by the AF diet may be due to maldigestion and malabsorption.

**Table 3. Liver status and hematological traits of El-salam laying hens fed dietary treatments from 30 to 38 wk's old.**

Dietary treatment <sup>1</sup>	Liver wt. (%)	Liver lipids (%)	Liver vit. A ( $\mu\text{g/g}$ )	Leuco-cytes $10^3/\text{mm}^3$	Lymphocytes $10^3/\text{mm}^3$	Hemo-globin mg/100ml
Control	3.12 <sup>c</sup>	5.35 <sup>c</sup>	21.47 <sup>a</sup>	21.4 <sup>c</sup>	14.83 <sup>c</sup>	12.61 <sup>a</sup>
AF-diet	7.45 <sup>a</sup>	12.94 <sup>a</sup>	15.22 <sup>c</sup>	27.4 <sup>a</sup>	18.87 <sup>a</sup>	8.19 <sup>c</sup>
AF+GSH	6.33 <sup>ab</sup>	11.02 <sup>ab</sup>	16.79 <sup>bc</sup>	25.9 <sup>ab</sup>	17.80 <sup>ab</sup>	9.21 <sup>bc</sup>
AF+AS	5.31 <sup>b</sup>	9.18 <sup>b</sup>	18.36 <sup>b</sup>	24.5 <sup>b</sup>	16.78 <sup>b</sup>	10.31 <sup>b</sup>
AF+TF	5.34 <sup>b</sup>	9.22 <sup>b</sup>	18.34 <sup>b</sup>	24.6 <sup>b</sup>	16.82 <sup>b</sup>	10.33 <sup>b</sup>
AF+AS+GSH	4.22 <sup>bc</sup>	7.33 <sup>bc</sup>	19.90 <sup>bc</sup>	23.0 <sup>bc</sup>	15.77 <sup>bc</sup>	11.51 <sup>ab</sup>
AF+TF+GSH	4.21 <sup>bc</sup>	7.36 <sup>bc</sup>	19.94 <sup>bc</sup>	22.9 <sup>bc</sup>	15.81 <sup>bc</sup>	11.53 <sup>ab</sup>
SEM	0.194	0.251	0.873	3.214	2.14	0.308
P value	0.001	0.003	0.006	0.006	0.005	0.011

<sup>a-d</sup> Values followed by different letters within columns are significantly different ( $P \leq 0.05$ ).

<sup>1</sup>AF=1ppm Aflatoxin B1, TF=0.6% Tafla, AS=0.5% Hydrated sodium calcium aluminosilicate (HSCAS), GSH=5ppm Glutathione (Tripeptide glutamate). Values are means of 5 determinations.

Zilva and Pannall (1983) and Abdelhamid *et al.* (1995<sup>b</sup>) referred to presence of diseases or functional disorders in some organs, by aflatoxicosis, such as hepatitis, pancreatitis, nephrotic syndrome, anaemia, and carcinoma. The protection of sorbent materials against AF effects on liver and hematological traits was also observed by Kubena *et al.* (1993), Qota (2003) and Hassan (2006).

**Aflatoxin B<sub>1</sub> residues in fresh tissues and serum constituents:**

Results in Table (4) showed that birds fed basal diet contaminated with 1ppm AFB<sub>1</sub> for 8 wk's deposited AFB<sub>1</sub> residues by highest value in their fresh livers (68 ng/g) followed by egg yolk (52 ng/g) then muscles (36 ng/g), decreased serum albumin (68%) and total lipids (51%) and increased enzymatic activities of AST (64%) and ALT (69%). Adding TF or HSCAS to AF diet, similarly, reduced AFB<sub>1</sub> residues by 51, 48, and 50 % in the yolk, liver and meat; respectively, and diminished AF effects on albumin, lipids, AST and ALT by 56, 46, 49 and 51%; respectively. A significant alleviating effect for adding GSH separately to the AF diet was observed with AST and ALT. Moreover, adding GSH with TF or HSCAS to the AF diet significantly negated the adverse effect of the AF diet on serum constituents, and reduced the AFB<sub>1</sub> residues by 75, 74 and 75% in yolk, liver and meat; respectively. The present results confirmed those of Qota (2003), Ali *et al.* (2006) and Hassan (2006) who detected AFB<sub>1</sub> residues in tissues of birds fed contaminated diets. Increasing accumulation AFB<sub>1</sub> in the liver than other tissues, in the present study, was observed as showed by Rizk *et al.* (1993), Abdelhamid *et al.* (1995<sup>b</sup>) and Qota *et al.* (2005). Decreasing serum albumin and lipids, and increasing ALT and AST activities by aflatoxicosis were reported in many studies (Genedy *et al.*, 1999; Hassan, 2000 and Qota *et al.*, 2005). The protective effect of sorbent materials against AF diet for AFB<sub>1</sub> residues and serum constituents, in the present study, was observed also by Kubena *et al.* (1993), Genedy *et al.* (1999) and Qota *et al.* (2005).

**Table 4. Aflatoxin B<sub>1</sub> residues in fresh tissues and serum constituents of El-salam laying hens fed dietary treatments during 30-38 wk's old.**

Dietary treatment <sup>1</sup>	AFB <sub>1</sub> residues			Serum constituents			
	Yolk (ng/g)	Liver (ng/g)	Meat (ng/g)	Albumin g/100m	Lipid (g/l)	AST (IU/L)	ALT (IU/L)
Control	***	***	***	2.5 <sup>a</sup>	6.7 <sup>a</sup>	12.1 <sup>e</sup>	7.52 <sup>e</sup>
AF-diet	51.6 <sup>a</sup>	67.8 <sup>a</sup>	36.4 <sup>a</sup>	0.8 <sup>c</sup>	3.3 <sup>c</sup>	19.8 <sup>a</sup>	12.7 <sup>a</sup>
AF+GSH	37.2 <sup>ab</sup>	51.5 <sup>ab</sup>	27.3 <sup>ab</sup>	1.2 <sup>bc</sup>	4.2 <sup>bc</sup>	17.6 <sup>b</sup>	11.4 <sup>b</sup>
AF+AS	25.5 <sup>b</sup>	34.3 <sup>b</sup>	18.2 <sup>b</sup>	1.6 <sup>b</sup>	5.1 <sup>b</sup>	15.8 <sup>c</sup>	10.1 <sup>c</sup>
AF+TF	24.9 <sup>b</sup>	33.9 <sup>b</sup>	18.4 <sup>b</sup>	1.5 <sup>b</sup>	5.2 <sup>b</sup>	15.9 <sup>c</sup>	10.2 <sup>c</sup>
AF+AS+GSH	13.4 <sup>bcd</sup>	17.6 <sup>bcd</sup>	9.21 <sup>bcd</sup>	2.2 <sup>ab</sup>	6.2 <sup>ab</sup>	14.0 <sup>d</sup>	8.79 <sup>d</sup>
AF+TF+GSH	12.8 <sup>bcd</sup>	18.2 <sup>bcd</sup>	9.09 <sup>bcd</sup>	2.1 <sup>ab</sup>	6.0 <sup>ab</sup>	13.9 <sup>d</sup>	8.81 <sup>d</sup>
SEM	1.98	2.17	1.88	0.31	0.51	1.01	0.71
P value	0.003	0.001	0.004	0.002	0.01	0.01	0.01

<sup>a-d</sup> Values followed by different letters within columns are significantly different ( $P \leq 0.05$ ).

<sup>1</sup>AF=1ppm Aflatoxin B1, TF=0.6% Tafla, AS=0.5% Hydrated sodium calcium aluminosilicate (HSCAS), GSH=5 ppm Glutathione (Tripeptide glutamate).

Values are means of 5 determinations. \*\*\*=No detection of AFB<sub>1</sub>.

Few studies have been carried on glutathione as detoxification of AF. Role of GSH comes after the absorption of AF and during its metabolism process in the liver. It, as an antioxidant, protects cells from toxins such as free radicals during the tissue-damaging peroxidation process and increases enzymatic detoxification in the liver (Wattenberg, 1976). Ehrich *et al.* (1984) and Ehrich and Larsen (1983) proved that detoxification enzyme systems in chickens could be increased by the administration of the antioxidants. Hsieh (1982) found that primary hepatic metabolites of AFB<sub>1</sub> may subjected to cytoplasmic reductase system producing aflatoxicol or to liver microsomal oxidase system producing AF: Q<sub>1</sub>, M<sub>1</sub> and B<sub>1</sub>-epoxide. Except for AFB<sub>1</sub>-epoxide, all metabolites contain hydroxyl groups are transformed into a water-soluble conjugate and to facilitate excretion. The transient B<sub>1</sub>-epoxide can be conjugated by GSH to form another type of conjugate. A prospective action may be afforded by reaction of AFB<sub>1</sub> metabolite with GSH (Lotikar *et al.*, 1980). Presence of AFB<sub>1</sub>-GSH conjugate in the bile of AF-treated rats, and its formation *in vitro* in liver-derived subcellular fractions, has been reported (Dengen and Neumann, 1978; Moss *et al.*, 1983). Some nutrients increased the activity of GSH for detoxification of AF in birds tissue such as Se is used as a cofactor for Se dependent GSH peroxidase (SeGSHpx) which is important in detoxification of hydrogen-px and lipid hydro-px, and increase GSH-px activity (Combs, 1981). Only Se increased the activity of Se GSH-Px in all tissues (Combs, 1981; Nahm, 1995). Also, Se enhanced the formation of water-soluble conjugated forms of AF which promotes the clearance of the toxin (Gregory and Edds, 1984). In the same manner methionine is a more distal precursor of GSH (Veltmann *et al.*, 1983). Vitamin C affected GSH metabolism at low level (Kim and Combs, 1992). From the present study, it may be concluded that TF (available product) had a similar safety protective effect for studied traits and EE as HSCAS (import product). Using GSH as antioxidant with TF or HSCAS, as sorbent materials into AF diet significantly negated aflatoxicosis in the laying hens.

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**استخدام الجلوتاثيون والالومنيوم سيليكات والطفه فى العلف كمحاوله لازاله التسمم بالافلاتوكسينات فى الدجاج البياض**  
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أجرى هذا البحث لتقييم كفاءة كل من الجلوتاثيون (ترابى بيتيد جلوتامات) كمضاد أكسدة والطفله والالومنيوم سيليكات كمواد ماصة بالعلف كمحاوله لازاله التسمم بالافلاتوكسينات فى الدجاج البياض وذلك بمحيطه بحوث الانتاج الحيواني بسخا- معهد بحوث الانتاج الحيواني باستخدام عدد (٣٢١) دجاجه بياضه + (٢١) ديك طائر عمر ٣٠ أسبوع من سلاله السلام مقسمه عشوائيا إلى ٧ مجموعات بكل منها ٥ مكررات تشمل المكرره ١٠ دجاجات رببت في بطارات سلك و ٢١ ديك المتبقيه قسمت أيضا إلى ٧ مجاميغ وكل مجموعة ٣ ديك تم تسكينها فرديا لجمع السائل المنوى وغذيت الطيور إما على علف الأساس بدون إضافات أو مع إضافه ١ جزء في المليون أفلاتوكسين ب، فقط أو بالإضافة إلى ٥ جزء في المليون جلوتاثيون أو ٦،٦% طفله أو ٥،٥% الالومنيوم سيليكات أو ٥ جزء في المليون جلوتاثيون + ٦،٦% طفله أو ٥ جزء في المليون جلوتاثيون + ٥،٥% الالومنيوم سيليكات لتكون في النهاية ٧ علاق غذيت عليها الطيور من عمر ٣٠ إلى ٣٨ أسبوع وأشارت النتائج المتحصل عليها إلى أن إضافه ١ جزء في المليون أفلاتوكسين ب، إلى علف الأساس لمدة ٨ أسابيع تسببت (بالمقارنة بالكتنرول) في نقص كل من كمية العلف المأكل (١٪) وإنماج البيض (٤٢،٨٪) وزن البيضة (٢٢،٣٪) وسمك القشرة (٣٢،٦٪) ونسبة البيض المخصب (٢١،٩٪) ونسبة التفريخ من البيض المخصب (٢٠٪) ونسبة الكبد من فيتامين أ (٢٩،١٪) وهيموجلوبين الدم (٣٥،٦٪) ومحتوى السيروم من كل من الألبومين (٦٨٪) والدهون الكلية (٥١٪) وأيضا زيادة كل من وزن الكبد (١٣٨،٨٪) ودهون الكبد (١٤١،٩٪) وعدد كرات الدم البيضاء (٢٨٪) وعدد الكرات الليمفائية (٢٧،٢٪) ونشاط إنزيمي AST و ALT (٦٤٪ و ٦٩٪ على التوالي) في السيروم كما أنها تسببت في احتجاز كمية من الأفلاتوكسين ب، في كل من انسجه الكبد (٦٨ نانوغرام/جرام) وصفار البيض (٥٢ نانوغرام/جرام) والعضلات الطازجة (٣٦ نانوغرام/جرام). كما أدت إضافه اي من الطفله أو الالومنيوم سيليكات بصورة منفردة إلى العلف الملوث بالافلاتوكسين ب، إلى حدوث تأثير واقى متقارب يتراوح من ٤٥ إلى ٥٦ % للصفات المدروسه والكافه الاقتصاديه وذلك بالمقارنة بالمجموعة الملوثه بدون إضافات، وسجلت إضافه الجلوتاثيون منفردة إلى العلف الملوث تأثيرا واقيا قليلا غير معنوي للصفات المدروسه ماعدا نشاط إنزيمي AST و ALT فقد كان التأثير الواقي (٢٨-٢٠٪) معنوا، أما إضافه الجلوتاثيون مع الطفله أو مع الالومنيوم سيليكات إلى العلف الملوث فقد سجلت أفضل التأثيرات وأزال التأثير الضار للأفلاتوكسين ب، جوهريا في كل الصفات المدروسه كما أنها حسنت الكفاءه الاقتصاديه بمقدار ٨١٪ (الطفله مع الجلوتاثيون) و ٧٥٪ (الالومنيوم سيليكات مع الجلوتاثيون) مقارنه مع العلف الملوث. نسبة النفوذ ١٠٪ في المجموعه المغذاه على علف ملوث بدون إضافات و ٦٪ في المجموعه ذات العلف الملوث مضافا إليه الجلوتاثيون أما باقى المجاميع لم تسجل اي نفوذ. وتخلص الدراسة إلى نقطتين أولهما أن الطفله (المتوفره في البيئه المحليه) لها تأثير أمن وواقي ضد الأفلاتوكسين ب، مساويا تماما في جميع الفياسات والكافه الاقتصاديه مع الالومنيوم سيليكات (المستوردة من الخارج) وثانيهما أن إضافه الجلوتاثيون كمضاد أكسدة مع الطفله أو الالومنيوم سيليكات على السواء كماده ماصة إلى العلف الملوث كانت أفضل الإضافات وأزال التأثير السام للأفلاتوكسين ب، في الدجاج البياض ويحتاج هذا الموضوع إلى مزيد من الدراسات لتأكيد هذه النتائج.