

THE FUNGI RECORDED IN IMPORTED FEED SAMPLES WITH REFERENCE TO CONTROL OF T-2 TOXICOSIS BY ANTIOXIDANT SUBSTANCES IN CHICKS

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SUMMARY

Fifty samples of imported animal feed were evaluated for fungi and their toxins contamination. Nine genera and eight species of mould were isolated. The genus *Mucor* and *Penicillium* (66% and 65%) were predominantly isolated, followed by *Aspergillus* (40%), *Fusarium* (20%) and *Cladosporium* (14%). Belonging to genus *Aspergillus*, *A. flavus* was frequently isolated (28%) but *F. graminearium* was obtained only from 10% of samples. Species of *Rhizopus*, *Scopulariopsis* and *Alternaria* were yielded from 4, 2 and 2% of samples respectively. T₂-toxin (member of trichothecene) was detected in 30% of these samples with the mean amount of 60 ppb and aflatoxin in 20% with the mean amount of 3,4 ppb, whereas ochratoxin A was gained from 14% of samples with mean level of 2.2 ppb, but Zearalenon

and Fumonisin B₁ toxins were found in 6 and 2% with mean level of 22 and 70 ppb respectively.

The induction of T-2 toxicosis in the broiler chickens and their elimination by dietary Vitamin E and/or selenium revealed that T-2 toxicosis significantly decreased the concentration of serum total protein, albumin and beta globulin and increased α and γ globulins. Levels of total lipids, triglycerides, cholesterol and copper were significantly increased and levels of zinc, vitamin A and E were decreased. The groups of chicks given vitamin E and/or selenium in the T-2 toxic diet showed general improvement in most of biochemical findings which were altered due to T-2 toxicosis, where the levels of α , β and γ globulin were increased and a significant decrease in levels of total lipid, triglycerides and cholesterol were obtained. Also, the treatment of toxicated chicks with selenium and/or Vit. E produced significant

alteration in levels of Vit. A and E, copper, zinc and sodium. The antioxidant effect of Vit. E and/or selenium reduced the toxic effect of T-2 toxin but not inhibit it.

INTRODUCTION

The increased population in the world requires a parallel raise in the production of food. Some countries as Egypt had to import many food and feeds. The recent researches reported that the majority of this food may carry the dangerous factors for human and animal health. Fungal contaminations and their toxins represents the most significant contaminant of these food (Aly, 1993; Debey et al., 1995 and Magnoli et al., 1999). Various members of *Fusarium* species isolated from imported feed produced mycotoxins under condition of stress (Mirocha and Christensen, 1977). The T-2 toxin (a member of trichothecene group) is considered the most significant mycotoxin of *Fusarium* species (Aziz et al., 1997 and Sohn et al., 1999). It is capable of killing cells by causing extensive damage to cellular membrane (Shokri et al., 2000). It had been found in many cereals, feed and vegetables (Bamburg et al., 1970). The toxic effects of T-2 toxin in poultry were expressed as reduced body weight gains (-26% at 3 weeks), oral lesions and increased weight of liver and pancreas (Kubena et al., 1994). The significant effect of this toxin is alteration in serum concentration of total protein, albumin, minerals and lipid profiles in broiler (Kubena et al., 1997). Previous re-

ports confirmed these results in chicken fed dietary 6 mg T-2 toxin /kg of feed for 21 day and found changes in serum protein, albumin, globulins, potassium, cholesterol, α -tocopherol, beta carotene and no change in vitamin A (Yu et al., 1997). However, the altered copper and zinc levels in serum are the causative factors of the biological damage (Ehud et al., 1983). Also, it was reported that there is a relationship between the decrease of T-2 toxin toxicity and the change in its metabolism in rats fed selenium (0.5 mg/kg) supplemented diet (Kravchenko et al., 1990). Recently the dietary use of selenium alone or in combination with vitamin E, provided protection against acute and chronic toxicosis, caused by T-2 toxin (Hochler and Marquardt, 1996 and Shokri et al., 2000).

Therefore, in this study the imported animal feeds were screened for fungal contamination and their toxins. Also, the most important *Fusarium* mycotoxin (T₂-toxin) was used for induction of toxicosis in broilers. The toxicated broilers were also administered antioxidant materials, including vitamin E and/or selenium. Serum biochemical changes were also studied in both treatments.

MATERIAL AND METHODS

- Feed samples: Fifty samples of imported animal feeds collected from imported lots were subjected to mycological and mycotoxicological examination.

- **Laboratory animals:** Fifty chicks of one day old were reared in stainless steel chick battery and allowed to consume feed, light and water ad libitum for the first two weeks before experimental work.
- **Antioxidant agents:** To modulate the effect of T₂-toxin, antioxidant agents were prepared freshly and added to the drinking water. These agents were selenium as sodium selenite which was used as 4 mg/L and vitamin E as dl- α -tocopherol acetate (15%), which was provided by ADWIA Pharmaceutical Company and was used as 1000 mg/L.

Isolation and identification of fungi:-

Finely ground 10 grams of each feed samples were added to 90 ml of physiological saline then vigorously shaken. A one ml of each was poured to a Petri dish (in duplicate) before addition of Sabouroud's dextrose agar medium (15 ml to each dish) (Refai, 1979 and Baily and Scott, 1986). After incubation at 26°C for 3-5 days, the individual colony was briefly identified according to the morphology and microscopical characters (Conant et al., 1954; Raper and Fennell, 1965 and Booth, 1971).

Detection of mycotoxins in feeds:-

Measurement of mycotoxins in feeds was applied according to the method reported by Howell and Tylor (1981) using thin layer chromatography.

T₂ toxin production in feed:-

A strain of *F. graminearum* which was isolated from samples of this work was used for production of T₂ toxin. A flask containing 100 grams of finely ground corn and 40-50 ml of distilled water was mixed and autoclaved at 121°C for one hour. On each of 2 successive days after each autoclaving the flask were shaken to prevent cooking of yellow corn. It was inoculated with *F. graminearum* (*F. roseum*) and incubated for 4 weeks at 25-28°C, then for 2 weeks at 8-10°C. After incubation, the corn was removed from flasks, dried, ground and 50 grams of each was subjected to T₂ toxin extraction (Wyllie and Morchous, 1978).

Extraction of T-2 toxin from experimentally infected yellow corn by *F. graminearum*:

100 ml methanol: water (55 : 45) was added to 25 grams of infected yellow corn in erylen myer flask and vigorous shaking of contents was conducted for 1 hour and filtered. The mycotoxin was extracted and purified from the filtrate according to the method of Howell and Tylor (1981).

Experimental attenuation of T-2 toxicosis by selenium and vitamin E (Kubena et al., 1997): Fifty broiler chicken of one day old were randomly divided into five groups (each of ten chicken). The groups of chicken were treated as following (Table, 1):

Table (1): Experimental design showing treatments of broiler chicks with T₂, selenium and vitamin E individually or in combination.

Treatment Groups	Normal healthy feed	T2 toxi 5 ppm in diet	Selenium (4mg/L) in drinking water	Vitamin E (1000 mg/L) in drinking water
G1	+	-	-	-
G2	-	+	-	-
G3	-	+	+	-
G4	-	+	-	+
G5	-	+	+	+

The period of feeding is extended to 4 weeks and the chicks were sacrificed twice, the first after 2 weeks from beginning and the other at the end of the experiment.

Sampling:

Blood samples were individually collected, immediately after slaughtering, in dry clean centrifuge tubes. Samples were left to clot at room temperature for about 2 hours, stored overnight in a refrigerator at 4°C and centrifugated at 3000 rpm for 15 minutes. Serum samples were drawn in dry clean-capped tubes and kept in deep freeze. These samples were subjected to biochemical analysis.

Biochemical analysis:

Serum samples were subjected to biochemical analysis, Electrophoretic separation of serum proteins was done according to the method of Ornstein (1964); the concentration of serum total proteins was measured according to Sonnen-wirth and Jarete (1980). Determination of the levels of

serum total lipids, cholesterol and triglyceride were carried out according to Knight et al. (1972), Waston (1960) and Wahlefeld (1974), respectively. The major electrolyte sodium and potassium concentrations on serum were estimated by flame photometry as described by Henry et al. (1974). Serum copper and zinc concentrations were estimated by atomic absorption spectrophotometry (Pekin Elmer model 2380, USA) according to Joseph and Roger (1976).

RESULTS AND DISCUSSION

The intensive animal production to overcome the request of enormous population for protein sources resulted in the importation of food and feeds and this becomes critical problem.

In this paper the mycological examination of imported animals feeds revealed the high incidence of *Mucor* (66%) and *Penicillium* sp. (56%), followed by *Aspergillus* (40%), *Fusaiurum* (20%)

and *Cladosporium* (14%). The *Aspergillus flavus* was the predominant isolate of genus *Aspergillus* (28%), followed by *A. candidus* (8%), *A. ochraceus* (4%) and *A. niger* (4%), whereas *F. gramin-*

earum was isolated from 10% of samples; *F. solani* and *F. moniliforme* were isolated from (4% and 12%) of samples.

Table (2) Prevalence of moulds in imported animal feeds.

Species of moulds	Prevalence of moulds		
	No. of +ve sample	No. of -ve sample	% of +ve samples
<i>Mucor</i> sp.	33	17	66
<i>Penicillium</i> sp.	28	22	56
<i>Aspergillus</i> sp.	20	30	40
<i>A. flavus</i>	14	36	28
<i>A. candidus</i>	4	46	8
<i>A. ochraceus</i>	2	48	4
<i>A. niger</i>	2	48	4
<i>Fusarium</i> sp.	10	40	20
<i>F. graminearum</i>	5	45	10
<i>F. solani</i>	2	48	4
<i>F. poae</i>	2	48	4
<i>F. moniliforme</i>	1	49	12
<i>Cladosporium</i> sp.	7	43	14
<i>Rhizopus</i> sp.	2	48	4
<i>Scopulariopsis</i> sp.	2	48	4
<i>Alternaria</i> sp.	1	49	2

Fifty samples of feed were examined.

Table (3) Levels of mycotoxins in imported feeds.

Mycotoxins	Levels of mycotoxins (ppb)					
	+ve sample	-ve sample	% of +ve	Max.	Min.	Mean
Aflatoxins	10	40	20	15	1.5	3.4
Ochratoxins	7	43	14	3.2	1.0	2
T-2	15	35	30	50	2.0	60
Zearalenone	3	47	6	30	10	22
Fumonisin B1	1	49	2	70	70	70

Table (4-a): Mean values of total serum proteins and electrophoretic pattern in broiler chicks treated with T2, selenium and vit. E.

Groups		Control	T-2 (toxin)	T-2 + selenium	T-2 + Vitamin E	T-2 + (Vitamin E + selenium)
T. protein (gm %)	15 d	3.88 ± 0.04	3.71 ± 0.04**	3.76 ± 0.02*	3.69 ± 0.06*	3.84 ± 0.02
	30 d	4.33 ± 0.03	4.20 ± 0.03**	4.22 ± 0.03*	4.22 ± 0.03*	4.24 ± 0.03*
Albumin (gm %)	15 d	1.34 ± 0.04	1.06 ± 0.06***	1.1 ± 0.07**	1.11 ± 0.09*	1.18 ± 0.06*
	30 d	1.49 ± 0.06	1.11 ± 0.06***	1.13 ± 0.07**	1.20 ± 0.06**	1.26 ± 0.08*
T-Alpha (gm %)	15 d	0.89 ± 0.04	0.99 ± 0.03*	1.00 ± 0.04	0.96 ± 0.03	0.98 ± 0.08
	30 d	0.98 ± 0.04	1.18 ± 0.05**	1.20 ± 0.06**	1.18 ± 0.06**	1.14 ± 0.05*
T- Beta (gm %)	15 d	0.56 ± 0.01	0.42 ± 0.03***	0.44 ± 0.04**	0.43 ± 0.04**	0.46 ± 0.03**
	30 d	0.67 ± 0.02	0.49 ± 0.04***	0.50 ± 0.04**	0.51 ± 0.05**	0.54 ± 0.03**
T-Gamma (gm %)	15 d	1.09 ± 0.04	1.24 ± 0.04*	1.22 ± 0.06*	1.19 ± 0.05	1.22 ± 0.09
	30 d	1.19 ± 0.05	1.42 ± 0.02***	1.39 ± 0.03**	1.37 ± 0.04**	1.30 ± 0.06
T- globulin (gm %)	15 d	2.54 ± 0.06	2.65 ± 0.10**	2.66 ± 0.11*	2.58 ± 0.10	2.66 ± 0.14
	30 d	2.84 ± 0.08	3.09 ± 0.08***	3.09 ± 0.07***	3.06 ± 0.14**	2.98 ± 0.12*
A/G ratio	15 d	0.52 ± 0.03	0.40 ± 0.01***	0.41 ± 0.01**	0.43 ± 0.02	0.44 ± 0.01**
	30 d	0.52 ± 0.03	0.35 ± 0.02****	0.37 ± 0.02***	0.39 ± 0.02***	0.43 ± 0.02**

* Significant at P < 0.05 using t-test.

** Significant at P < 0.01 using t-test.

*** Significant at P < 0.001 using t-test.

Table (4-b): Mean values of total serum proteins fractions in broiler chicks fed with T₂-toxin, selenium and vit. E

Groups		Control	T-2	T-2 + selenium	T-2 + Vitamin E	T-2 + (Vitamin E + selenium)
Alpa ₁ (gm/dl)	15 d	0.21 ± 0.03	0.48 ± 0.06***	0.45 ± 0.08*	0.38 ± 0.06*	0.42 ± 0.07*
	30 d	0.23 ± 0.03	0.58 ± 0.06****	0.61 ± 0.09**	0.58 ± 0.07***	0.51 ± 0.06***
Alpa ₂ (gm/dl)	15 d	0.68 ± 0.03	0.51 ± 0.05****	0.55 ± 0.04*	0.58 ± 0.03*	0.56 ± 0.02**
	30 d	0.75 ± 0.04	0.60 ± 0.02**	0.59 ± 0.03**	0.60 ± 0.02**	0.63 ± 0.03*
Beta ₁ (gm/dl)	15 d	0.20 ± 0.02	0.19 ± 0.01	0.18 ± 0.01	0.16 ± 0.02	0.14 ± 0.02*
	30 d	0.26 ± 0.02	0.21 ± 0.01*	0.20 ± 0.02*	0.19 ± 0.02*	0.21 ± 0.01*
Beta ₂ (gm/dl)	15 d	0.36 ± 0.03	0.23 ± 0.02***	0.26 ± 0.01**	0.27 ± 0.02*	0.32 ± 0.01
	30 d	0.40 ± 0.02	0.28 ± 0.02***	0.30 ± 0.02**	0.32 ± 0.02**	0.33 ± 0.03*
Gamma ₁ (gm/dl)	15 d	0.71 ± 0.03	0.54 ± 0.04**	0.63 ± 0.02*	0.65 ± 0.04	0.64 ± 0.03
	30 d	0.80 ± 0.04	0.65 ± 0.02**	0.69 ± 0.04*	0.68 ± 0.03*	0.74 ± 0.01
Gamma ₂ (gm/dl)	15 d	0.38 ± 0.04	0.70 ± 0.06***	0.59 ± 0.05**	0.54 ± 0.06*	0.58 ± 0.04**
	30 d	0.39 ± 0.04	0.77 ± 0.08***	0.70 ± 0.07***	0.69 ± 0.07***	0.56 ± 0.05*

* Significant at P < 0.05 using t-test.

** Significant at P < 0.01 using t-test.

*** Significant at P < 0.001 using t-test.

Table (5) Mean values of serum total lipid, triglycerid and cholesterol of broiler chicks with T₂, selenium and Vit. E.

		Control	T-2 (toxin)	T-2 + selenium	T-2 + Vitamin E	T-2 + (Vitamin E + selenium)
T. lipid (mg%)	15 d	509.3 ± 15.81	620.5 ± 16.96**	598.00 ± 24.70**	584.00 ± 22.5*	546.67 ± 20.14
	30 d	552.0 ± 35.03	390.0 ± 20.11***	421.33 ± 18.74**	457.33 ± 38.87*	490.20 ± 20.94
Triglycerides (mg%)	15 d	59.72 ± 4.50	81.20 ± 5.71**	69.24 ± 1.48*	71.19 ± 4.60*	64.84 ± 3.50**
	30 d	65.12 ± 6.85	34.76 ± 4.85***	38.36 ± 3.18***	37.64 ± 3.91**	40.74 ± 4.31
Cholesterol (mg%)	15 d	158.79 ± 4.70	173.96 ± 5.37*	160.25 ± 4.50	170.19 ± 3.30*	170.14 ± 3.44*
	30 d	158.79 ± 4.70	130.07 ± 7.92**	140.92 ± 6.37*	150.62 ± 4.41*	149.10 ± 6.13

Table (6) Mean values of serum vitamin A and E of broiler chicks with T2, selenium and Vit. E.

		Control	T-2	T-2 + selenium	T-2 + Vitamin E	T-2 + (Vitamin E + selenium)
Vit. A (µg/dl)	15 d	34.83 ± 2.21	22.41 ± 3.80**	24.12 ± 3.63*	25.6 ± 3.73*	26.56 ± 3.41*
	30 d	35.61 ± 4.01	18.72 ± 2.11***	20.20 ± 3.80*	26.1 ± 7.94*	23.60 ± 3.22*
Vit. E (µg/dl)	15 d	230.12 ± 20.10	161.0 ± 6.07**	165.32 ± 10.61*	186.46 ± 2.77*	181.13 ± 8.30*
	30 d	350.19 ± 29.41	200.03 ± 6.59***	230.17 ± 11.94***	257.73 ± 8.73**	244.58 ± 9.23***

* Significant at P < 0.05 using t-test.

** Significant at P < 0.01 using t-test.

*** Significant at P < 0.001 using t-test.

Table (7) Mean values of serum copper, zinc, sodium and potassium of broiler chicks with T2, selenium and Vit. E.

		Control	T-2 (toxin)	T-2 + selenium	T-2 + Vitamin E	T-2 + (Vitamin E + selenium)
Copper (µg/dl)	15 d	87.20 ± 4.00	104.67 ± 6.24*	97.67 ± 5.90	98.40 ± 4.31*	91.39 ± 9.21
	30 d	97.7 ± 6.81	121.80 ± 8.10*	119.71 ± 6.71*	116.50 ± 5.60*	109.6 ± 6.24
Zinc (µg/dl)	15 d	140.52 ± 6.61	91.00 ± 9.82***	109.69 ± 11.71*	94.00 ± 13.0**	101.70 ± 9.30
	30 d	151.80 ± 5.31	118.00 ± 9.71**	128.31 ± 7.24*	122.2 ± 8.67**	123.1 ± 11.16*
Sodium (mEq/L)	15 d	137.67 ± 5.36	167.23 ± 5.12**	157.33 ± 4.95*	156.0 ± 6.45*	151.00 ± 4.59*
	30 d	162.20 ± 5.70	136.00 ± 6.81	141.25 ± 7.49*	146.61 ± 6.01*	141.20 ± 5.21*
Potassium (mEq/L)	15 d	5.28 ± 0.35	6.42 ± 0.22**	6.21 ± 0.16*	5.98 ± 0.32	6.00 ± 0.48
	30 d	5.51 ± 0.23	4.57 ± 0.12***	4.82 ± 0.23*	5.00 ± 0.15*	4.98 ± 0.17*

* Significant at P < 0.05 using t-test.

** Significant at P < 0.01 using t-test.

*** Significant at P < 0.001 using t-test.

Other genera of moulds belong to *Rhizopus*, *Scopulariopsis* and *Alternaria* sp. were obtained in low frequency (4, 4 and 2%) respectively (Table, 2). Dajani et al. (1990) in Jordan, El-Zawahry et al. (1991) in Saudi Arabia and Adebajo et al. (1994) in Nigeria; Pettersson et al. (1995) in Sweden and Desjardins et al. (2000) from Himalaya mountains recovered the fungal contamination in cereals and feed stuffs.

The high incidence of *Penicillium*, *Mucor* and *Fusarium* sp. in imported feeds could be explained by the trial of use refrigeration by the exporting countries to reduce the microbial growth in commodities and food. But the fridge temperature prevent the growth of some mould and allow the generation of other fungi as *Penicillium*, *Mucor* and *Fusarium* species which were reported recently to be able to produce dangerous mycotoxins and mycotoxicosis during keeping in fridge (Nikulin et al., 1996) and Jimenez et al. (1996) and this cause losses among animals (Sanchis et al., 1986). Generally the presence of different moulds in imported feed can also be sustained to various climatic conditions, temperature, humidity and handling during transportation from country to other which will expose these feed for over contamination. Gareis et al. (1986); Muller (1989) and Millano and Lopez (1991) detected the same observations in their studies.

During detection of mycotoxins in the same feed sample of this study, a *Fusarium* toxin (T₂) was

obtained in high frequency (30%) with mean amount of 60 ppb followed by aflatoxin (20%) with the mean amount of 3.4 ppb and ochratoxin (14%) with the mean level of 2.2 ppb. Whereas Zearlenone and Fumonisin B₁ were obtained in (6 and 2%) from imported feeds and levels of 22 and 70 ppb respectively (Table, 3).

It was previously reported that the *Fusarium* toxin was obtained in high incidence in food which was preserved at low temperature (L'Vovo et al., 1993) in Russia, Kazakhstan and Uzbekistan regions. So, the use of refrigeration in addition to other preservative measures to avoid obvious contamination in imported feeds by the producing countries leads to the formation of dangerous *Fusarium* toxin (as T₂). Significant effects on health, immune status and milk productions of animal vigorously occurred due to mycosis and mycotoxicosis (Wu et al., 1991 and V'anyl et al., 1994). Also, human consumed such these contaminated food were adversely affected by dangerous disease (Vasanthi and Bhat, 1998; Li et al., 1999 and Wang et al., 2000). Therefore, T₂ toxicosis was conducted with a trial for its elimination by Vit. E and/or selenium in feeds and food. In the present study as indicated in table (4-a), there were significant decrease in concentrations of serum total protein, albumin and beta globulins together with A/G ratio. Otherwise, there was a significant increase in alpha, gamma and total globulins in treated groups with T₂ toxin compared with controls. Similar observations were recorded in broil-

ers by Edrington et al. (1997) who attributed the decrease in serum protein to decreased efficiency of feed utilization and inhibition of protein synthesis as a result of damage in DNA caused by T-2 toxin. The decreased albumin, beta-globulin and A/G ratio were in agreement with Harvey et al. (1994).

T-2 toxin was associated with other alterations in serum protein fractions (Table, 4-b). These include a significant increase in alpha and gamma₂ and a significant decrease in alpha₂, beta₁, 2 and gamma₁ globulins in both experiments. These results might be attributed to the liver inflammation and immunosuppressive effect of T-2 toxin (Pang et al., 1987).

From Table (4 a) and (4 b) it is suggested that vitamin E and selenium alone or in combination modulate the immune response and increase humoral immunity (Hassan et al., 2001). This appeared in the first experiment and a little effect was noticed in the second experiment (Tables, 4-a and b).

The data in Table (5) indicated that chickens exposed to T-2 toxin had an alterations in the concentration of serum lipids profile. The levels of serum total lipids, triglycerides and cholesterol were significantly increased in 1st experiment, whereas, a significant decrease was noticed in 2nd experiment. Results of this experiment agreed with reports of Kubena et al. (1996). Also, they

found that T-2 toxin was increased in the relative weight of liver. This increase in liver weight might be associated with alterations in lipids metabolism which resulted in impaired lipids transport. The supplementation of vitamin E and selenium alone or in combination decreased hepatic cell damage caused by T-2 toxin (Atróshi et al., 1997).

The treatment of chickens with T-2 toxin (Table, 6) reduced significantly the serum vitamin A and E concentrations, compared with control. The treatment of T-2 toxin exposed chicken with vitamin E and selenium alone or in-combination did not have a modifying influence on vit. A reduction. These results were confirmed by Hochler and Marquardt (1996).

The current study (Table, 7) showed a significant increase in serum copper of chicken at 15 and 30 days post-treatment with T₂, vit. E. and T₂, T₂ + selenium and T₂ Vit. E, respectively. Meanwhile, the levels of serum zinc were decreased significantly at 15 and 30 day post-treatment in all groups except T₂ + vit. E + selenium group.

These results can be attributed to the oxidative damage caused by T-2 toxin, which affected concentration of superoxide dismutase (SOD) (Atróshi et al., 1997). This inactivation in (SOD) was accompanied by an increased in concentrations of Cu and decrease in concentration of Zn (Margalioth et al., 1983).

The concentration of sodium and potassium showed significant increase in 1st treated group and a significant decrease in 2nd group, compared with control. Hazzele et al. (1992) observed same results. The alteration in serum levels of sodium and potassium were explained on account of the toxic effects of T-2 toxin which increases a relative weight of kidney caused by an impaired renal function which affected ion excretion (Glahn et al., 1989)

Supplementation of antioxidant to chicks induced marked effects, on serum copper, zinc, potassium and sodium. The differences in the results indicate that the antioxidant can be affected by certain receptor or enhance excretion of these mineral from proximal convoluted tubules of kidneys (Glahn et al., 1994).

The potency of protective action of antioxidants (as selenium and α -tocopherol) on certain biochemical analysis after toxicosis with T-2 toxin may be due to modulation of drug metabolism in liver (Roomi et al., 1998).

The T-2 toxin plays a role in the toxicity of chickens and had harmful effect on the serum biochemical parameter. The antioxidants (Vit. E or selenium) alone or in combination can be used to reduce these effect but not inhibit it as reported here.

From the foregoing results, it is clear that the importation of food carry more dangerous microbial and toxic factors which affect by many ways the productivity of animals and health of man. Elsewhere the use of antioxidant is a critical and rapid method for elimination of these contaminates from imported feed. But the effective way to avoid the danger effects of these toxins is the use of local feed and food in our country and prevent the importation as possible, thus keeping of animal wealth, quality of its productivity (meat, egg and milk) and safe of human health become under control.

REFERENCES

- Adebajo, L.O.; Idowu, A.A. and Adesanya, O.A. (1994): "Mycollora and mycotoxin production in Nigerian corn and corn based snakes." *Mycopathologia*, 126 (3): 183-192.
- Aly, M. (1993): "Toxigenic fungi and mycotoxins content in poultry feed stuff ingrediants." *J. of Basic Microbiol.*, 33 (2): 101-104.
- Atroschi, F.; Rizzo, A.; Biese, I.; Veljalainen, P.; Antilua, E. and Westermarck, T. (1997): "T-2 toxin-induced DNA damage in mouse livers: The effect of pretreatment with coenzyme Q10 and alpha-tocopherol." *Mol. Aspects Med.*, 18: 255-258.
- Aziz, N.H.; Attia, E.S. and Farag, S.A. (1997): "Effect of gamma-irradiation on the natural occurrence of Fusarium mycotoxins in wheat, flour and bread." *Nahrung*, 41 (1): 34-37.

- Baily, S.A. and Scott, A.D. (1986): "Diagnostic Microbiology." The C.V. Mosby Company St.Lois, Toronto.
- Bamburg, J.R.; Strong, F.M. and Smalley, E.B. (1970): "Toxins from moldy feed cereals." *J. Agric. Food Chem.*, 17: 443-448.
- Booth, C. (1971): "The genus *Fusarium*." Common Wealth Mycological Institute, Kew, Surrey, England.
- Conant, N.F.; Martin, D.S.; Smith, D.T.; Baker, R.D. and Callaway, J.L. (1954): "Manual of Clinical Mycology." Philadelphia W.B. Saunders Company.
- Dajani, N.F.; Natar, R.M.; Salhab, A.S. and Mohasneh, A. (1990): "Incidence of zearlenone and zearlenone producing fungi in cereal and animal feed stuffs in Jordan." *Arab. Gulf. J. Sci. Res.*, 8 (2): 119-127.
- Debey, M.C.; Trampel, D.W.; Richard, J.L.; Bundy, D.S.; Hoffman, L.J.; Meyer, V.M. and Cox, D.F. (1995): "Effect of environmental variables in turkey confinement houses on air born *Aspergillus* and mycoflora composition." *Poult. Sci.*, 74 (3): 763-771.
- Desjardins, A.E.; Manandkar, G.; Plantner, R.D.; Maragos, C.M.; Sheestha, K. and McCormick, S.P. (2000): "Occurrence of *Fusarium* species and mycotoxins in Nepalese maize and wheat and the effect of traditional processing methods on mycotoxin levels." *J. Agric. Food Chem.*, 48 (4): 13377-13383.
- Edrington, T.S.; Kubena, L.F.; Harvey, R.B. and Rottenghous (1997): "Influence of a superactivated charcoal on the toxic effects of aflatoxin and T-2 toxin in growing broiler." *Poult. Sci.*, 76: 1205-1211.
- Ehud, J.M.; Joseph, G.S. and Mordechai, C. (1983): "Copper and zinc levels in normal and malignant tissues." *Cancer*, 52: 868-872.
- El-Zawahry, Y.A.; Aziz, N.H. and El-Fouly, M.Z. (1991): "Incidence of toxin and pathogenic microorganism in different Egyptian and Saudi Arabian food commonalities and their decontamination by gamma irradiation." *Egypt. J. Microb.*, 26 (2): 267-282.
- Gareis, M.; Bauer, J. and Gedek, B. (1986): "Survey of agriculture commodities for trichothecene mycotoxin, zearlenone, ochratoxin A and aflatoxin B1: Results of four year study." *Proc. 2nd World Congress Food Born Infection Intoxic.*, Berlin (West) F.R.G., p. 893.
- Glahn, R.P.; Shapiro, R.S.; Veno, V.E.; Wideman, R.F. and Huff, W.E. (1989): "Effect of chronic ochratoxin A alone and in combination with citrinin on kidney function of single comb white Leghorn pullets." *Poult. Sci.*, 68: 1205-1212.
- Glahn, R.P.; Van Campen, D. and Dousa, T.P. (1994): "Aflatoxin B1 reduces Na⁺ - (P*i*) Co-transport in proximal renal epithelium: studies in opossum kidney (OK) cells." *Toxicology*, 92: 91-100.
- Harvey, R.B.; Kubena, L.F. and Elissalde, M.H. (1994): "Influence of vitamin E aflatoxicosis in growing swine." *Am. J. Vet. Res.*, 55: 572-577.
- Hassan, H.M.; Nasser, M.H.; Ramadan, A.A.; El-Sakran, M.N. and Mansour, U.M. (2001): "Effect of vitamin E and selenium as a dietary supplements on cellular and humoral, immune responses in horses." *Arab. Vet. Med. Congress*, 69: 155-164.
- Hazzele, F.M.; Gunter, W.; Marquardt, R.R. and Frohlich, A.A. (1992): "Beneficial effects of dietary ascorbic acid supplement on hens subjected to ochratoxin A toxicosis under normal and high ambient temperature." *Canad. J. Anim. Sci.*, 73 (1): 149-152.

- Henry, R.J.; Cannon, D.C. and Winkelman, J.W. (1974): "Clinical Chemistry, Principles and Techniques." 2nd Edition, Harper and Row, p. 525.
- Hochler, D. and Marquardt, R.P. (1996): "Influence of vitamin E and C on the toxic effect of ochratoxin A and T-2 toxin in chicks." *Poult. Sci.*, 75: 1808-1815.
- Howell, H.V. and Taylor, P.W. (1981): "Determination of aflatoxins, ochratoxin A and zearalenone in mixed feeds with detection by TLC or HPLC." *J.A.O.A.C.*, 64 (6): 1356-1363.
- Jimenez, M.; M'a-nez, M. and Hern'andez, E. (1996): "Influence of water activity and temperature on the production of zearalenone in corn by three *Fusarium* species." *Int. J. Food Microbiol.*, 29 (2-3): 417-421.
- Joseph, A. and Roger, W.G. (1976): "Clinical Chemistry, Principles and Procedures." 4th Ed., Boston, pp. 168-179.
- Knight, J.A.; Anderson, S. and James, M.R. (1972): "Chemical basis of the sulphosphovanilin reaction for estimating total serum lipids." *J. Clin. Chem.*, 18: 199-210.
- Kravchenko, L.V.; Kuzmina, E.E.; Avenieva, L.I. and Kumpulainen, J.J. (1990): "Dietary selenium protects against acute toxicity of T-2 toxin in rats." *Food Addit. Contam.*, 7: 821-827.
- Kubena, L.F.; Edrington, T.S.; Buckley, S.A.; Phillips, T.D.; Rottinghaus, G.E and Casper, H.H. (1997): "Individual and combined effect of Fumonisin B1 present in *Fusarium moniliforme* culture material and T-2 toxin or deoxynivalenol in broiler chicks." *Poult. Sci.*, 76 (9): 1239-1247.
- Kubena, L.F.; Edrington, T.S.; Harvey, R.B.; Phillips, T.D.; Sarr, A.B. and Rottinghaus, G.E (1996): "Individual and combined effect of *Fusarium moniliforme* culture material and diacetoxyscirpenol or ochratoxin A in turkey poults." *Poult. Sci.*, 76 (2): 256-265.
- Kubena, L.F.; Smith, E.E.; Genettes, A.; Harvey, R.B.; Edrington, T.E.; Phillips, T.D. and Rottinghaus, G.E. (1994): "Individual and combined toxicity of T-2 toxin and Cyclopiazonic acid in broiler chicks." *Poult. Sci.*, 73: 1390.
- Li, Y.C.; Ledoux, D.R.; Bermudez, A.J.; Fritsche, K.L. and Rottinghaus, G.E. (1999): "Effects of fumonisin B1 on selected immune responses in broiler chicks." *Poult. Sci.*, 78 (9): 1275-1282.
- L'vova, L.S.; Orlova, N.L.U.; Kova, Z.k.; Omel'chenko and Remele, V.V. (1993): "Propagation of toxigenic fungi and mycotoxin in various grain." *Parikl. Niokhim. Mikrobiol.*, 29 (1): 70-79.
- Magnoli, C.E.; Saenz, M.A.; Chiacchiera, S.M. and Dalce-ro, A.M. (1999): "Natural occurrence of *Fusarium* species and fumonisin-production by toxigenic strains isolated from poultry feeds in Argentina." *Mycopathologia*, 145 (1): 35-41.
- Margalioth, E.J.; Schenker, J.G. and Chevion, M. (1983): "Copper and zinc levels in normal and malignant tissues." *Cancer*, 52: 868-872.
- Millano, G.D. and Lopez, T.A. (1991): "Influence of temperature on zearalenone production by regional strain of *F. graminearum* and *F. oxysporum* in culture." *Int. J. Food. Microbiol.*, 13 (4): 329-333.
- Mirocha, C.J. and Christensen, C.M. (1977): "Mycotoxicosis in human and animal helath." Cited by Rodricks et al. (1977) *Pork Forest South Pathatox*, 11-Illinois, pp. 345.

- Muller, H.M. (1989): "Measures for lessening the mycotoxin formation and concentration in feed." *D.T.W.Dtsch.Tierarztl. Wochenschr.*, 96 (7): 383-398.
- Nikulin, M.; Lappalainen, S.; Pasanen, A.L.; Laamanen, I.; Veijalainen, P.; Berg, S. and Hintikka, E.L. (1996): "Comparison of detection methods for trichothecenes produced by *F. sporotrichioides* on fodder and grains at different air humidities." *Nat. Toxins*, 4 (3): 117-121.
- Ornstein, L.C. (1964): "Disc electrophoresis. I. Background and Theory." *Ann. N.Y. Acad. Sci.*, 212: 321-332.
- Pang, V.F.; Lambert, R.J. and Felsburg, P.J. (1987): "Experimental T-2 toxicosis in swine following inhalation exposure: effects on pulmonary and systemic immunity and morphologic changes." *Toxicol. Pathol.*, 15: 308-319.
- Pettersson, H.; Hedman, R.; Engstrom, B.; Elwinger, K. and Fossum, O. (1995): "Nivalenol in Swedish cereals occurrence, production and toxicity towards chickens." *Food Addit. And Contam.*, 12 (3): 373-376.
- Raper, K.B. and Fennel, D.T. (1965): "The genus *Aspergillus*." Williams and Wilkins Co., Baltimore Maryland.
- Relai, M.K. (1979): "Manual of Food Quality. 4. Microbiological Analysis." Food and Agriculture Organization of the United Nations, Rome, FAO, P.P. F2 and 5D 32.
- Roomi, M.W.; Netke, S. and Tsao, C. (1998): "Modulation of drug metabolizing enzymes in guinea pig liver by high intakes of ascorbic acid." *Int. J. Vitam. Res.*, 68: 42-47.
- Sanchis, V.; Sala, N.; Palmes, A.; Santamina, P. and Burdaspal, P.A. (1986): "Occurrence of aflatoxin and aflatoxigenic moduls in food and feed in Spain." *J. Food Protect.*, 49 (6): 445-448.
- Shokri, F.; Heidari, N.; Gharagozloo, S. and Ghazi-khausari, M. (2000): "In vitro inhibitory effects of antioxidant on cytotoxicity of T-2 toxin." *Toxicol.*, 146 (2-3): 171-176.
- Sohn, H.B.; Seo, J.A. and Lee, Y.W. (1999): "Co-occurrence of *Fusarium* mycotoxins in mouldy and healthy corn from Korea." *Food Addit. Contam.*, 16 (4): 153-158.
- Sonnen wirth, A.C. and Jarete, L. (1980): "Gradwal's Clinical Laboratory Methods and Diagnosis." Vol. I, 8th Ed., pp. 258-259. The C.V. Mosby Co. St. Louis, Toronto, London.
- V'anyl, A.; Bata, A. and Kovacs, F. (1994): "Effect of T-2 on the egg yield and hatchability in geese." *Acta Vet. Hung.*, 42 (1): 79-85.
- Vasanthi, S. and Bhat, R.V. (1998): "Mycotoxins in foods- occurrence, health and economic significance and food control measures." *Indian J. Med. Res.*, 108 (H-D): 212-224.
- Wahlefeld, A.W. (1974): "Colourmetric determination of enzymatic triacyl glycol in serum. In *Methods of Enzymatic Analysis*." Vol. 5, M.M. Berneyer Ed., Academic Press, New York, pp. 1831-1835.
- Wang, H.; Wei, H.; Ma, J. and Luo, X. (2000): "The fumonisin B1 content in corn from North China, a high-risk area of oesophageal cancer." *J. Environ. Pathol. Toxicol. Oncol.*, 19 (1-2): 139-141.
- Waston, D. (1960): "Determination of serum cholesterol." *Clin. Chem. Acta*, 5: 637-645.
- Wu, W.; Cook, M.E. and Smalley, E.B. (1991): "Decreased immune response and increased incidence of tibial dyschondroplasia caused by *Fusaria* grown on sterile corn." *Poult. Sci.*, 70 (2): 293-301.

Wyllie, T.D. and Morehouse, L.G. (1978): "Mycotoxic fungi. Mycotoxins and Mycotoxicosis." Vol. 2, (Mycotoxicosis of domestic and laboratory animals, poultry and aquatic invertebrates and vertebrates). Marcell Dekker Inc., New York, pp. 750.

Yu, M.W.; Chiang, Y.C.; Lien, J.P. and Chen, C.J. (1997): "Plasma antioxidant vitamins, chronic hepatitis B virus infection and Urinary aflatoxin B1-DNA adducts in healthy males." *Carcinogenesis*, 18 (6): 1189-1194.