# SOME SERUM BIOCHEMICAL, HORMONAL AND PROTEIN ELECTROPHORETICAL STUDIES ON SHEEP AND GOATS SUFFERING FROM MYCOTOXICOSIS AND /OR BRUCELLOSIS

ABD EL FATTAH SH. M.1, A.D. HELAL2 AND F.E. SHEHATA2

1 National Research Centre

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#### Abstract

A total of 156 serum samples were collected from adult ewes and goats (78 animals per each species) for serological identification of brucellosis and mycotoxicosis through a study in Kaleubia province. Forty-seven feed-mixtures (concentrates), and forty-three Maize samples were collected from localities of brucella infected sheep and goat. The feed samples were used for extraction and measurement of different mycotoxins. The mycotoxins were identified and measured in serum of brucella infected and non-infected animals. One month after parturition or abortion, twenty animals from each species were divided into four equal groups as follows: the first group was the control animals, the second group was non-infected but naturally mycotoxin poisoned animals, the third group was brucella infected animals and the fourth group was both brucella infected and mycotoxin poisoned animals. The serum biochemical, hormonal and protein electrophoretical studies were carried out. Results revealed that Aflatoxin-B1 (AFB1), Ochratoxin-A (OA) and Fumonisin-B1 (FB1) mycotoxins were identified and measured in feed, but AFB1 and OA could only be identified in the serum samples. The mycotoxins were detected in 82.6 % of brucella infected animals, and in 10% of non-infected animals. In either mycotoxin poisoned and /or brucella infected animals, there was a significant decrease in serum prolactin levels. In non-infected animals, the mycotoxicosis induced non-significant increase of the the progesterone hormone. The triiodothyronine (T3) and the thyroxin (T4) hormones were significantly increased in serum of most ewes and goat groups. The mycotoxicosis induced significant hyperalpha-globulinemia and hypo-beta - globulinemia in non infected animals than control. Mycotoxicosis induced hypogamma globulinemia in goats. In most cases, mycotoxicosis and/or brucellosis induced hyperbilirubinemia, hyperlipedemia and significant increase of ALT-enzyme activity than control. In many cases, the mycotoxicosis, unless induced significant biochemical changes alone, induced such significant changes specially with brucellosis. Based on our study, we could suggest that the mycotoxicosis may perform a predisposing stress immunosuppressive factor for brucellosis. Also, mycotoxicosis and/or brucellosis may induce serum biochemical endocrinological, metabolic, hepatic and reproductive disturbances in ewes and goats.

<sup>&</sup>lt;sup>2</sup> Animal Health Research Institute, Agricultural Research centre, Ministry of Agriculture, Dokki- Giza - Egypt

#### INTRODUCTION

Mycotoxins are fungal metabolites that are toxic when consumed by animals and human beings. The toxins can accumulate in maturing corn, cereals, soybeans, sorghum, peanuts and other food and feed crops in the field and in grain during transportation and storage under conditions favourable for the growth of mycotoxin-producing fungi (Miller, 1995). Diseases in animals and human beings resulting from the consumption of mycotoxins are called mycotoxicosis. Among the mycotoxins identified, five mycotoxins of special importance are recognized: aflatoxins, deoxynivalenol, zearalenone, fumonisins and ochratoxins. Aflatoxin-B1 is the most toxic of the all known fungal metabolites, as it is immunotoxic and mutagenic in both mammals and birds. The immunosuppressive effect of aflatoxins was recorded by Dietert et al. (1985).

The *Brucella* organism is Gramnegative facultative intracellular bacterium capable of surviving and replicating in phagocytic leucocytes and epithelial cells. Brucellosis is an important contagious disease infecting animals and human and causes great losses in animal production allover the world (Radostits *et al. 2000*).

There is little data about the role of mycotoxins in brucella infected animals. Hence, the objective of the present study is to recognize the relationships between naturally induced mycotoxicosis and the brucellosis under the Egyptian environment, aiming to understanding the effects and the role of mycotoxicosis in spreading the infectious diseases among the Egyptian livestocks.

# **MATERIALS AND METHODS**

## **Animals**

The serum samples used for serological diagnosis of brucellosis in sheep and goat were taken from aborted animals which showed the clinical signs of brucellosis, and could be differentially diagnosed from other diseases causing abortion based on the clinical signs according to Blood *et al.* (1979).

A total of 156 animals (78 animals per each species) from adult ewes and goats (1 month after parturition or abortion), were collected from different private farms in Kaleubia province through a study lasting for 2 years. The serum samples were obtained from all animals of the farm, and the Buffer Acidified Plate Antigen (BAPA) test was used for primary diagnosis of brucella infected animals.

## Feed samples

Feed samples were collected at different periods from farms that showed abortions, where forty-seven feed-mixture samples (concentrates) and forty-three

maize samples were collected from the feed of the brucella infected and non-infected animals. The collected feed samples (each feed sample collected from different sites of feed packs) were used for detecting the different mycotoxins using Thin Layer Chromatography (TLC) and determined fluorodensitometrically according to AOAC (1980).

# Serological diagnosis of brucellosis

Serum samples were taken from 46 of brucella infected ewes and goats (1 month after abortion, 23 from each species) and other 110 serum samples from non-infected animals (1 month after parturition, 55 from each species).

The Buffer acidified plate antigen test (BAPAT) was done according to the procedure of the National Vet. Serv. Lab. (NVSL), Ames, Iowa, USA (Anon, 1984). The Tube agglutination test (TAT) was done, but with a modification to start with a dilution of 1/10, using the European assay and the Rivanol test (Rev. T) according to the method described by Alton *et al.* (1988).

#### Mycotoxins standards

The standards of AFB1, OA, FB1 were obtained from Sigma Co. (U.S.A).

# Mycotoxin analysis

The Myctoxin analysis was carried out according to AOAC (1980) as following

Aflatoxin analysis: Feed and blood samples were extracted and the aflatoxin  $B_1$  was determined fluorodensitometrically.

Ochratoxin A (OA) analysis: The OA amounts in feed were extracted, and then, determined fluorodensitometrically. The same method of extraction was used to measure the OA concentrations in the samples of blood serum. Prior to extraction, diluted serum samples were acidified with HCl / MgCl $_2$  solution. OA was quantitatively extracted into chloroform. The chloroform extract was separated from the aqueous serum by centrifugation. Each chloroform extract was washed once with water to remove the dissolved acid, then, dried under a steam of nitrogen gas. The dried residues were reconstituted in chloroform and the OA concentrations were determined fluorodensitometrically.

Fumonisin  $B_1$  analysis: Samples of feed and blood serum were extracted and fumonisin  $B_1$  was determined using the high performance liquid chromatography (HPLC).

# The tested groups of animals

Twenty animals were divided into four equal groups (5 animals per each group) from each ewe and goat species as in the following Table  $\bf 1$ .

Table 1. The groups of tested ewes and goats .

Table 1.	The groups of tested ewes and godes :
Group	Tested animal groups (in each species)
No.	
1	The normal control animals (1 month after normal parturition, free from brucella or mycotoxins).
2	Mycotoxin poisoned animals, free from infections, 1 month after parturition.
3	Brucella infected animals (1 month after abortion)
4	Brucella infected and mycotoxin poisoned animals (1 month after abortion)

#### Estimation of some serum hormones and some biochemical constituents

The Radioimmuno-assay method used for determination of some serum hormones (Challis *et al.* 1973), the polyacrylamide Gel Electropheresis Technique (Gordon, 1980) was used for fractionation of serum protein, and some serum biochemical constituents, spectrophotometrically determined as: the concentrations of total protein (Doumas *et al.* 1971) total lipids (Schmit, 1964) and total bilirubins (Jendrassiki *et al.* 1938) and the activity of Alanine aminotransferase enzyme (Reitman and Frankel, 1957).

The obtained data were statistically analysed using F-test through the analysis of variance (ANOVA), and the Student's t-test (Snedecor and Cochran, 1969).

# RESULTS

# Mycotoxins in feed

In feed mixture, the percentage of mycotoxin infected samples was 57.45%, while, in maize was 58.14%. The mycotoxins which could be detected in animal feed were: aflatoxin  $B_1$  (AFB1), ochratoxin A (OA) and fumonisin- $B_1$  (FB1), their values are shown in Table 2 .

# The Incidence and levels of serum mycotoxins

The percentage of ewes and goats positive to mycotoxins was 82.61% of the total brucella infected animals compared to 10% in the non-infected animal species (Table 3). The serum mycotoxins which were detected in ewes were: Aflatoxin B<sub>1</sub> (48.5  $\pm$  6.073 ng/ml) and Ochratoxin –A (116  $\pm$  5.43 ng/ml) and in goats were : Aflatoxin B<sub>1</sub> (94.6  $\pm$  17.4 ng/ml) and Ochratoxin –A (137.8  $\pm$  10.9 ng/ml), and the serum mycotoxins in goat were significantly increased than those of sheep, as recorded in Table 4.

# The Serological diagnosis of brucellosis in ewes and goats

The specific antibody titer of the brucella infected goats was significantly higher than that of brucella infected ewes as serologically dermined by both tube agglutination test and Rivanol test (Table 4).

# **Serum hormones**

The serum hormone levels in the ewes and goats are shown in Table 5 as following:

#### 1. Prolactin hormone

The prolactin hormone was significantly decreased in all treated ewes and goat groups than control. Mycotoxins induced significant increase of prolactin hormone in brucella infected goats than that of only brucella infected goats.

# 2. Progesterone hormone

The levels of progesterone hormone in the two brucella infected groups (with or without mycotoxicosis) were significantly increased in sheep and goats than controls. The mycotoxicosis induced non significant increase of the progesterone hormone in non-infected ewes and goats than control.

## 3.Triiodothyronin (T3)hormone

The two groups infected with brucella (with or without mycotoxicosis) of ewes and goats showed significant increase of T3 levels than controls. The mycotoxicosis induced non-significant increase of T3 level in non-infected ewes than control.

## 4.Thyroxin (T4)hormone

The ewes and goats with mycotoxicosis and  $\!\!/$  or brucellosis revealed significant increase of T4 levels than that of control animals.

# Serum protein electrophoresis

The different serum protein fractions of ewes and goats are shown in Table 6 as following:

- **1-Alpha** ( $\alpha$ ) globulin fractions: The  $\alpha$ -globulins showed significant increase in all ewes and goat groups than control animals (except the brucella infected with mycotoxin poisoned group of ewes where  $\alpha$  globulin non significantly changed).
- **2-Beta** ( $\beta$ ) **Globulins:** The beta globulins were significantly decreased in all groups of goats than control, but, the ewes did not show any significant changes of the beta globulins.
- 3-Gamma (γ) Globulins: The gamma globulins were significantly decreased in mycotoxin poisoned non-infected goats and decreased non-significantly in ewes than control. However, gammaglobulins were significantly increased in brucella infected goats and ewes than control animals.

- 4-Total Globulins: The total globulins were significantly increased in all ewes and goat groups than control (except in mycotoxin poisoned goat where the change of total globulins was non significant).
- 5-Albumin: The albumin of ewes did not show any significant changes, but, it increased significantly in brucella infected goats and decreased significantly by mycotoxin poisoned goats either brucella infected or non infected than control.

# Some Serum Biochemical Constituents

The serum biochemical constituents of ewes and goats are shown in Table 7 as following:

- 1-ALT enzyme activity: Mycotoxins induced significant increase of ALT enzyme activity in all ewes and goat groups than control (except in mycotoxin poisoned goats, where the increase was non significant).
- 2- Total bilirubin: Mycotoxicosis and / or brucellosis induced significant increase of total bilirubin in all ewes and goat groups than control.
- 3-Total lipids: The total lipids were significantly increased in all ewes and goat groups (either suffering from mycotoxicosis and/or brucellosis) than control.
- 4-Total proteins: The total proteins were non-significantly changed by all groups of ewes and goats (except in the brucella infected ewes and goats, where it significantly increased than control).

Table 2. The concentrations of mycotoxins in feed samples which were used for feeding brucella infected animals in Kaleubia Province.

Feed sample	Total No. of tested feed samples	Total No. of contaminated samples	Percent of contaminated samples to totalexamined.	Mycotoxins in feed		
				Aflatoxin B <sub>1</sub> (AFB <sub>1</sub> ) mg/kg. Feed	Ochratoxin A (OA) mg/kg.feed	Fumonisin B <sub>1</sub> (FB <sub>1</sub> ) mg/kg.feed
Concentrates (Feed mixture)	47	27	57.45%	0.624±0.118	0.143±0.072	0.125±0.05
Maize	43	25	58.14%	0.355±0.053	0.231±0.120	0.274±0.084
Total samples	90	52	57.78%			
Overall means ± SE	-			0.490 ± 0.095	0.187 ± 0.022	0.200 ± 0.053

Table 3. The incidence of serum mycotoxins in 136 (from 156) serum samples of brucella infected and non-infected ewes and goats.

Groups	Tested Animals		No. of positive animals with any mycotoxins			Percent of positive animals with mycotoxins to the total No.			
	Ewes	Goats	Total	Ewes	Goat	Total	Ewes %	Goat %	Mean %
Brucella infected animals	23	23	46	17	21	38	73.91%	91.30%	82.61%
Non- infected animals	45	45	90	4	5	9	8.89%	11.11%	10%
Total tested animals	68	68	136	21	26	47	30.88%	38.24%	34.56%

Table 4. The serum levels of mycotoxins and the specific antibody titers of brucella infected ewes and goats (as measured by Tube Agglutination and Rivanol tests).

Groups of Animals	Serum levels of mycotoxins			Tube	Aggl. Test	Rivanol test	
	Aflatoxin B <sub>1</sub> (AFB1) (ng/ml)	Ochratoxin A (OA) (ng/ml)	Fumonisin B <sub>1</sub> (FB <sub>1</sub> ) (ng/ml)	Titer ranges (n=5)	Log10 – value of the reciprocal titers	Titer ranges (n =5)	Log10- values of the reciporcal titers
Control ewes	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 <u>+</u> 0.00
Brucella infected Ewes (n = 5)	48.5 ± 6.073	116 ± 5.43	0.00 ± 0.00	1/20-1/40	1.482 ± 0.066	1/25- 1/100	1.699 ± 0.085
Control Goats	0.00 ± 0.00	0.00 ± 0.00	0.00 <u>+</u> 0.00	0.00 <u>+</u> 0.00	0.00 <u>+</u> 0.00	0.00 ± 0.00	0.00 <u>+</u> 0.00
Bruceila infecteds Goats (n = 5)	94.6* ± 17.4	137.8 NS ± 10.9	0.00 NS ± 0.00	1/80-1/160	2.482* ± 0.066	1/100- 1/400	2.301* ± 0.085

<sup>\* =</sup> Significant change between means of mycotoxins and serological tests of ewes and goats at (P  $\leq$  0.05). NS = Non-significant change.

Table 5. Effects of mycotoxins (Aflatoxin  $B_1$  and Ochratoxin-A) and/or Brucellosis on some serum hormones of ewes and goats.

Hormones		Prolactin	Progesterone	Triiodothyronin	Thyroxin (T4)
		hormone	hormone	(T3) hormone	hormone
		(M.I.U./ml)	(ng./ml)	(n mol/L.)	(n mol./L.)
Species	Groups				
Sheep	Control	13.600 A	1.900 A	3.30 A	68.20 A
(ewes)	(Normal)	±	±	±	±
	, ,	1.397	0.291	0.316	3.162
	Mycotoxins	6.44 B	3.300 A	4.34 A	102.26 B
	Poisoned	±	±	±	±
		0.487	0.713	0.213	4.545
	Brucella	5.32 B	22.90 B	5.80 B	87.40 B
	infected	±	±	±	±
		1.075	1.649	0.906	5.433
	Brucella	7.30 B	19.40 B	7.50 B	99.80 B
	infected and	<u>±</u>	±	±	±
	mycotoxin	0.822	1.581	0.949	6.093
	poisoned				
	LSD	2.419	3.703	1.816	13.192
	$(P \le 0.05)$				
	Control	12.20 A	1.100 A	4.30 A	63.20 A
Goat	(Normal)	±	±	±	±
		1.291	0.316	0.646	3.162
	Mycotoxins	5.94 B	1.80 A	6.100 B	114.50 B
	poisoned	±	±	±	±
		0.671	0.293	0.782	7.376
	Brucella	3.86 C	19.90 B	7.40 B	88.70 C
	infected	±	±	±	±
		0.481	2.746	1.122	4.817
W. W.	Brucella	6.30 B	16.10 B	10.50 C	112.70 B
	infected and	±	±	±	±
	mycotoxin	0.721	1.122	1.358	4.033
	poisoned				
	LSD	1.443	4.015	1.319	7.182
	$(P \le 0.05)$				
	, - ,		1		

# N.B.:

<sup>1-</sup> The different capital litters in columns pointed to presence of significant change between means (at P  $\leq$  0.05).

<sup>2-</sup> LSD = Least Significant Difference between means (at P  $\leq$  0.05).

Table 6. Effects of mycotoxins (Aflatoxin  $B_1$  and Ochratoxin-A) and/or Brucellosis on the different electrophoretically separated serum protein fractions of ewes and goats.

	f . V	Alpha (α)	Beta (β) Globulins	Gamma (γ ) Globulins	Total Globulins	Albumin (g./dl.)	Total protein
Protein	fractions	Globulins		(g.dl.)	(g./dl.)	(g./ui.)	(g./dl.)
	T C	(g/dl.)	(g/dl.)	(g.al.)	(g./ui.)		(g./ui.)
Species	Groups		0.007.1	1.005.4	2 (00 4	2 002 4	E 6024
Sheep		0.757 A	0.837 A	1.095 A	2.689 A	3.003 A	5.692A
(ewes)	Control	±	±	±	±	_ ± _	±
	(Normal)	0.059	0.156	0.082	0.231	0.515	0.070
		1.414 B	0.459 A	0.944 A	2.817 B	2.805 A	5.622 A
	Mycotoxins	±	±	±	±	±	±
	poisoned	0.141	0.032	0.102	0.145	0.173	0.328
		1.217 B	0.537 A	1.576 B	3.33 C	3.125 A	6.455 B
	Brucella	±	±	±	±	±	±
	infected	0.151	0.028	0.243	0.156	0.351	0.343
	Brucella	0.752 A	0.726 A	1.526 B	3.004 D	2.496 A	5.50 A
	infected. &	±	±	±	土	±	±
	mycotoxin poisoned	0.084	0.097	0.228	0.266	0.276	0.447
	LSD (P < 0.05)	0.205	N.S.	0.377	0.055	N.S.	0.640
Goat	( 5.55)	0.327 A	1.203A	1.178 A	2.708 A	2.754A	5.462A
Couc	Control	±	±	±	±	±	±
	(Normal)	0.036	0.102	0.117	0.150	0.246	0.667
- 100	Mycotoxins	1.574 B	0.338 B	0.795 B	2.707 A	2.139 B	4.846 A
	poisoned	±	±	±	±	±	±
		0.402	0.029	0.107	0.127	0.282	0.296
	1	1.221 BC	0.699 C	1.815 C	3.735 B	3.188 C	6.923 B
	Brucella	±	±	±	±	±	±
	infected	0.141	0.035	0.127	0.134	0.324	0.282
	Brucella	0.87 C	0.958 D	1.392 D	3.22 C	2.318 B	5.538 A
	infected. &	±	±	±	±	±	±
	mycotoxin	0.144	0.054	0.156	0.196	0.326	0.493
	poisoned	0.177	0.037	0.130	0.150	0.020	050
	LSD	0.370	0.164	0.163	0.216	0.346	0.709
	$(P \le 0.05)$			para de la companya d			

**N.B. :** 1- The different capital litters in columns pointed to presence of significant change between means (at P  $\leq$  0.05). 2- LSD = Least Significant Difference between means (at P  $\leq$  0.05). 3- N.S. = Non – Significant changes between means.

Table 7. Effects of mycotoxins (Aflatoxin B<sub>1</sub> and Cochratoxin-A) and / or Brucellosis on

Serum constituents		ALT enzyme activity (U/dl)	Total Bilirubin (mg/dl)	Total lipids conc. (g./dl.)	Total proteins conc. (g/dl)
Species	Groups				
Sheep (ewes)	Control (Normal)	7.99±0.632 A	1.069±0.127 A	26.487±1.203 A	5.692±0.070 A
	Mycotoxins poisoned	10.32±0.626 B	2.060±0,220 B	34.944±1.514 B	5.622± 0.328 A
	Brucella infected	10.61±0.361 B	1.772±0.252 C	34.141±1.525 B	6.455±0.343 B
	Brucella infected & mycotoxin poisoned	13.28±1.020 C	2.154±0.301 B	38.985±1.621 C	5.50±0.447 A
	LSD (P ≤ 0.05)	1.138	0.287	1.389	0.640
Goat	Control (Normal)	9.60±0.354 A	1.346±0.123 A	30.337±0.981 A	5.462±0.667 A
	Mycotoxins poisoned	10.15±0.668 A	1.842±0.261 B	33.117±0.822 B	4.846±0.296 A
	Brucella infected	12.64±699 B	2.135±0.2958 BC	33.915±1.718 B	6.923±0.282 B
	Brucella infected & mycotoxin poisoned	14.19±1.318 C	2.485±0.262 C	36.2555±0.447 C	5.538±0.493 A
	LSD (P ≤ 0.05)	1.545	0.363	1.684	0.709

# N.B. :

- 1- The different capital litters in columns pointed to presence of significant change between means (at P  $\leq$  0.05).
- 2- LSD = Least Significant Difference between means (at  $P \le 0.05$ ).

# **DISCUSSION**

In the present study, the mycotoxins which could be detected in the feed mixture and maize used in feeding of brucella infected animals were: aflatoxin  $-B_1$  (AFB<sub>1</sub>), ochratoxin - A (OA) and fumonisin-B<sub>1</sub> (FB<sub>1</sub>), but the AFB<sub>1</sub> and OA could only be detected in blood of goats and ewes, so that the toxicity of mycotoxins in Brucella infected ewes or goats were due to the mixed toxicity of both AFB<sub>1</sub> and OA. The fumonisin B<sub>1</sub> which could not be detected in serum may reflect its possible degradation in rumen of animals according to CAST (1989).

In the current study, the mycotoxins were detected in 82.6% of the brucella infected ewes and goats, while, the mycotoxins were detected only in 10% of the total non-infected animals. This indicates that the mycotoxicosis may play a predisposing factor for accepting and spreading the brucella organism or other infecting agents. This suggestion could be recognized in the present study as the mycotoxins are immunosuppressors, due to their induction of hypogammaglobulinemia in non-infected goats (significantly) and sheep (non – significantly). The aflatoxicosis may induce depression of the cell –mediated immune respone ( Dietert *et al.* 1985). The mycotoxins could increase the disease incidence and reduce production efficiency in cattle due to suppression of the immune system (CAST,1989).

Mycotoxicosis induced significant hyperalphaglobulinemia in non-infected ewes and goats. The elevated levels of some alphaglobulins have been reported with the toxicity of some chemicals (Kaneko, 1989),

Beta  $(\beta)$ globulins were significantly decreased in mycotoxin poisoned or in brucella infected goats than control. On the other side, the  $\beta$ -globulins were significantly increased in brucella infected and mycotoxin poisoned goats animals than only brucella infected goats. The increased  $\beta$ -globulin level may be attributed to some immunoglobulins transferred to the region of the betaglobulin fractions in response to acute inflammatory diseases, autoimmune disease, hemolytic anaemia and iron deficiency (Kaneko, 1989).

Either mycotoxin poisoned or brucella infected animals induced significant decrease in prolactin hormone in ewes and goats than control in the current study. Similar study revealed significant decrease of serum prolactin by mycotoxicosis in rats by Manning (1987) who attributed the prolactin decrease by mycotoxicosis to the reduction of rough endoplasmic reticulum, golgi apparatus, ribosomes and secretory granules in the prolactin secreting cells. The major effects of prolactin hormone on the follicle stimulating hormone (FSH) and luteinizing hormone (LH) secretion appear to be exerted by inhibiting the secretion of gonadotropin releasing hormone (GNRH), so that there was a reverse relationship between prolactin level and the levels of FSH and LH hormones, and consequently, the progesterone and estrogen ovarian hormones (Cheung, 1983). The progesterone hormone was non-significantly increased by mycotoxicosis in ewes and goats of the present study. Similar results in beef cows with mycotoxicosis were recorded by Burke and Roie (2002) who mentioned that neither serum progesterone or estradiol nor corpus luteum diameter changed by mycotoxicosis induced by the ingestion of fungus infected fescue grass. The symptoms

of mycotoxicosis may be non-specific as reduced milk production, subnormal reproduction, increased abortion or embryonic mortalities, silent heats, irregular estrous cycles, expression of estrous in pregnant cows and decreased conception rates due to their induction of endocrine and neuroendocrine changes in cows suffering from acute mycotoxicosis (CAST, 1989).

In most cases, the serum triiodothyronin (T3) and the thyroxin (T4) hormones were significantly increased in serum of goats and ewes groups suffering from mycotoxicosis and/or brucellosis than control animals. The hyperthyroidism may induce toxic goitre which is mainfested by increased metabolic reactions and loss of weight (Georgieva, 1989). The mycotoxins induce alterations in nutrient contents, and the normal absorption and metabolism in cattle (CAST, 1989).

The present data revealed that , either mycotoxicosis and/or brucellosis induced significant increase in total lipids in ewes and goats than control animals. The hyperlipidemia may be induced because of the interference with lipid metabolism or with some xenobiotics (Kaneko, 1989).

In most cases, the serum total bilirubin concentration and the alanine aminotransferase (ALT) enzyme activity were significantly increased in brucella infected and/or mycotoxin poisoned ewes and goats than control animals, and this indicates presence of certain degree of liver dysfunction (induced by mycotoxicosis and/or brucellosis) in these animals according to Kachman and Moss (1976).

Based on our study, it could be concluded that, ewes and goats naturally poisoned from the field environment with AFB1 and OA mycotoxins, may become ready for accepting brucellosis or perhaps other infectious diseases, because of the immunodepressant activities of these mycotoxins. Also, the mycotoxins and/or brucellosis induced disturbances in reproductive and thyroid hormones and serum biochemical constituents, which may lead to disturbances in the total reproductive function, liver function and normal metabolism. So, a controlling program should be investigated and suggested against the mycotoxin contaminations of the animal feeds to be advised to diminuting the mycotoxicosis in the Egyptian livestocks.

# REFERENCES

- Alton G.G., L.M Jones and D.E Pietz. 1988. Laboratory Techniques in brucellosis, 2<sup>nd</sup>, Edition WHO Monograph series No. 55, Genera Switzerland: 112-113.
- Anon 1984.Instructions for conducting brucellosis serological test, National Veterinary Services, Ames, Iowa, USA.
- AOAC (Association of Analytical Chemists). 1980. Official methods of analysis, 13<sup>th.,</sup> ed., Washington D.C., U.S.A.
- 4. Blood B.C., J.A Handerson and O.M. Radostits. 1979. Veterinary Medicine 5th Edition.
- Burke J.M. and R.W. Roie. 2002. Changes in ovarian function in mature beef cows grazing endophyte infected tall fescue. Therogenology, 57 (6): 1733 – 1742.
- CAST (Council for Agricultural Science and Technology). 1989. Mycotoxins: Economic and Health Risks. Task Force Report No 116, Ames, Iowa.
- 7. Challis J.R.G., I.J. Davies and K.J.P. 1973. Endocrinol., 96: 185.
- Cheung C.Y. 1983. Prolactin suppresses luteinizing hormone secretion and pituitary responses of LHRH by a direct action at the anterior pituitary. Endocrinol., 113: 632-638.
- Dietert R.R., M.M. Qureshi and S.E. Bloom.1985. Embryonic exposure to aflatoxin-B1: Mutagenicity and influence of development and immunity. Environ. Mutagen, 7: 715-725.
- Doumas B.T., W.A. Watson, and H.G. Bigs.1971. Kits (El-Nasr Co.) used for serum total protein determination. Clin. Chem. Acta, 31 (1): 87.
- Georgieva S.A. 1989. Essentials of physiology pp: 256, translated from Russian by Nicolai Lybimov, Mir publishers, Moscow.

- Gordon A.H. 1980. Electrophoresis of proteins in polyacrylamide and starch gels. In laboratory Techniques in Biochemistry and Molecular Biology. Elsevier North Holand Biochemical press, Amesterdam, pp: 213.
- Jendrassiki G.P. 1938. Verienfachte photometriche metheden zur bestimmung de blutbilirubins. Biochemical J., 2 (297): 81.
- Kachman J.F. and D.W. Moss .1976. Clinical biochemistry of domestic animals, Academic press, Inc.
- Kaneko J. 1989. Enzymes, In: Fundamentals of Clinical Chemistry, pp: 565-598., W.B. Saunders, philadelphia,
- Manning W.S.Jr. 1987. Fescue induced inhibition of prolactin secretion: development of a rat model. Dissertation, Abstracts, international, B. Sciences and Engennering, 47 (7): 2695 – 2696.
- Miller J.D. 1995. Fungi and mycotoxins in grains. Implications for stored product research. J. Stored Prod. Res., 31,1.
- Radostits, O.M.,C.C. Gay,D.C. Blood and K.W. Hincheliff. 2000. Veterinary Medicine "A text book of the diseases of cattle, sheep, pigs and horses" 9<sup>th</sup>., ed, W.B. Saunders Company Ltd.
- Reitman S. and S.Frankel. 1957. Kits (El-Nasr Co.) for determination of SGOT and SGPT. J. Clin. Path., 28: 56.
- Schmit J.M. 1964. Kits (El-Nasr Co.) for determination of serum total lipid concentration, thesis, lyon.
- 21. Snedecor G.W. and W.G. Cochran. 1969. Statistical Methods, 6<sup>th</sup>., ed. IOWA state University Press, Ames., IOWA.

بعض الدر اسات البيوكيميائية والهرمونية والفصل الكهربي لبروتين مصل النعاج والماعز التي تعانى من السموم الفطرية والبروسيلا

شعبان مصطفى عبد الفتاح ' علاء الدين هلال على موسى ' فوزى إبراهيم شحاتة '

١ المركز القومي للبحوث

٢ معهد بحوث صحة الحيوان – مركز البحوث الزراعية – وزارة الزراعة –الدقى – جيزة –مصر

تم فحص ١٥٦ عينة سيرم من النعاج والماعز البالغة (٧٨ عينة من كل من نوعى الحيوانات) لمرض البروسيللا والسموم الفطرية من خلال دراسة بمحافظة القليوبية، وتم فحص السموم الفطرية في ٤٧ عينة مغلوط علف، و٤٣ عينة ذرة من مواقع الحيوانات المصابة بالبروسيللا، وبعد شهر من الولادة أو الإجهاض تم تقسيم ٢٠ حيواناً إلى أربع مجموعات متساوية من كل من النعاج والماعز كالتالى: المجموعة الأولى مجموعة الحيوانات السليمة واستخدمت كضابط للتجربة، والمجموعة الثانية حيوانات غير مصابة بالمرض وبها سموم فطرية بالبروسيللا والمجموعة الرابعة حيوانات مصابة بالبروسيللا والمجموعة. الرابعة حيوانات مصابة بالبروسيللا وبها أيضا سموم فطرية بالسيرم، وقد تم عمل قياسات بيوكيميائية وهرمونية وعمل الفصل الكهربائي لبروتين السيرم في مجموعات النعاج والماعز

أوضحت نتائج الدراسة أن أنواع السموم الفطرية التى تم قياسها فى العلف هى: الأفلاتوكسين ب١ ، والأوكراتوكسين -أ والفيومونزين ب١، أما فى السيرم فقد تم الكشف عن الأفلاتوكسين ب١ والأوكراتوكسين أ فقط، وأن هذه السموم الفطرية تم اكتشافها فى سيرم ٢٠٨% من الحيوانات المصابة بالبروسيللا، بينما تم كشف السموم الفطرية فى سيرم ١٠% فقط من الحيوانات الغير مصابة بالمرض، وفى معظم مجموعات الحيوانات المختلفة فإن التسمم بالسموم الفطرية أو الإصابة بمرض البروسيللا أو كليهما قد أحدثا انخفاضاً معنوياً فى مستوى هرمون البرولاكتين فى كل من النعاج والماعز وزيادة معنوية فى مستوى هرمون الغدة الدرقية الـ (٦٦) ولى الحيوانات الغير مصابة بالبروسيلا فقد أحدثت السموم الفطرية زيادة غير معنوية فى مستوى هرمون البروجستيرون، كذلك أوضح الفصل الكهربي لبروتين مصل الحيوانات أن السموم الفطرية أحدثت فى معتوى البيتاجلوبيولينات فى الحيوانات أن الغير مصابة بالبروسيلا، وكذلك أحدثت السموم الفطرية نقصاً معنوياً فى مستوى البيتاجلوبيولينات فى المعارية نقصاً معنوياً فى مستوى البلوسيلا (مقارنة بالماعز المصابة بالبروسيلا (مقارنة بالماعز الماعز المصابة بالبروسيلا (مقارنة بالماعز الماعز المصابة بالبروسيلا (مقارنة بالماعز الماعز المصابة بالبروسيلا (مقارنة بالماعز المصابة بالبروسيلا (مقارنة بالماعز الماعز الما

وفى معظم الأحوال فقد أوضحت الفحوص البيوكيميائية أن السموم الفطرية أو الإصابة بالبروسيللا أو كليهما قد أحدثا زيادة معنوية فى تركيزات البيليروبين الكلى والدهون الكلية ونشاط إنزيم السلام، وفى كثير من الأحيان فقد أوضحت الدراسة أن السموم الفطرية تبدى نشاطاً سمياً معنوياً لكثر مع الإصابة بالبروسيللا.

ومن خلال هذه الدراسة فقد أمكن استنتاج أن السموم الفطرية يمكن أن تشكل عوامل مجهدة ومثبطة للمناعة وممهدة للإصابة بمرض البروسيللا وربما للإصابة بالأمراض الأخرى، وكذلك فإن كلا من السموم الفطرية أو البروسيللا أو كليهما معاً يمكن أن يحدثا اضطراباً كيميائياً وهرمونياً وفي الميتابوليزم وفي الوظائف التناسلية ووظائف الكبد في كل من النعاج والماعز .