

## **SEROLOGICAL TESTS AND BIOCHEMICAL PROFILES IN CAMELS INFECTED WITH BRUCELLOSIS.**

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### **SUMMARY**

Blood samples were collected from 80 camels kept in closed farm (Camel production unit - Animal production Institute and others), another 72 blood samples were collected from camels kept in close contact with cattle and other small ruminant from different areas in Gize governorate. As well as 94 blood samples were collected from imported camel from Sudan in camel market.

Our data showed that the highest percent of positive reactors was observed in the imported camels (Sudanese camel) in large herd (8.50 to 11.70% (9.50%, 10.60%, 9.50%, 8.51%, 9.57% and 11.70 to 8.50% for RBPT, BAPA, Riv, SAT, MET and DIA respectively), Camels in contact with other animals (6.94 to 11.10% (8.30%, 9.40%, 8.30%, 6.94%, 8.33% and 8.30 to 11.10% for RBPT, BAPA, Riv, SAT, MET and DIA, respectively) and Camels in closed farms (0.00 to 2.50% (1.25%, 2.50%, 0.00%, 1.25%, 0.00% and 1.25 to 5.00% for RBPT, BAPA, Riv, SAT, MET and

DIA, respectively). The results of sensitivity and specificity of DIA revealed that DIA using n.lauroylsarcosin extract is more specific than DIA whole bacterial antigen.

The sera of infected camels with brucella (either camels contact with animals or imported camels) showed elevated levels in each of the GGT, LDH, ALP, AST, ALT, total protein, albumin, glucose, urea, uric acid and creatinine. The sera of imported camels infected with brucellosis were characterized by increased levels of protein bands with molecular weights 29.83 -30.11, 45.95-46.27 kDa, with increase of 34.64, 35.29, 74.67, 87.74, 98.96, 99.75, 104.62, 110.57 , 115.54, 132.63, 134.12, 138.69, 140.25 kDa protein bands in both camels contact with animals and imported camels infected with brucellosis. Protein bands 181.31-183.34 and 214.36 KDa were apparent in camels contact with animal's sera infected with brucellosis and protein bands 189.59 and 231.79 were present in imported camel's sera infected with brucellosis especially in 1/320 antibody sera. The

LDH and ALP iso-enzymes had a characteristic profile in brucellosis. Our conclusions that imported camels infected brucellosis followed by camels contact with animals infected with brucellosis had more serious biochemical discordance. The results give us an index to diagnosis of brucellosis in the imported camel. The incidence percent of brucellosis in camels in closed farm in dedicate importance, good prognosis mean and diagnosis tools to detect and eliminate the infected camels, and aid in epidemiological controls of the disease.

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## INTRODUCTION

*Brucella* specie is a facultative, intracellular pathogenic bacterium that causes brucellosis, and is in a zoonosis affecting mammalian species. *Brucella* entry into cells is highly enhanced by opsonization (Cazevieuille et al, 2000).

The camel has been a symbol of stability for pastoralists in the arid zones of the world (Abbas et al, 2000). Kiel and Khan, (1989) suggested that the epidemiology of brucellosis in camels in a country or region was complicated by importation of living animals with higher prevalence of brucellosis than in the local animals and humans across national border. Three main camel populations were found in Egypt. Local breed in country-side contact with other animals includes cattle, buffalo, sheep and goats, Imported camels through EL-Aribaniy road and farm camels which are new camel categories for camel productions. These factors are appropriate where camels are kept (Abbas et al, 2000).

In Egypt, owing to the low prevalence of brucellosis in camel as compared to the other livestock animals and that the differences between the test conditions were not uniform and that the data and conclusions were not simultaneously obtained, thus it is of utmost importance to conduct surveys involving adequate number, similar rearing conditions of camels. There is a real need to conduct of surveys to know the informant of camel brucellosis based on adequate camel's samples raised under similar conditions to compare the risk of brucellosis in camels when kept close to other animals (Palling et al, 1988).

The aim of this work was therefore devoted to investigate and compare different serological and biochemical tests among different main camel populations in Egypt to scrutinize and evaluate the reason, the source, the defectiveness and the eccentricity of camel brucellosis as well as advice for control measures against the infection in camels.

## MATERIAL AND METHODS

Blood samples were collected to separate sera from three groups: the 1<sup>st</sup> group comprised of 80 camels kept in closed farm as Camel production unit - Animal production institute) and other private farms this group were recorded with no previous report of abortions or illness and samples were withdrawn from jugular vein. The 2<sup>nd</sup> group comprised of 72 blood samples was collected from camels kept in close contact with cattle and other small ruminant from different areas in Giza governorate during slaughter in El-Warak slaughter house. The 3<sup>rd</sup> group included of 94 blood

samples was collected from imported camel from Sudan in camel market in Giza during slaughter in EL-Basatine Slaughter house. The male and female camels with various ages were apparently healthy and were free from tuberculoses, trypanosome, Para-tuberculoses, infectious hepatitis, liver flukes and hydrated cysts.

### 1. Conventional Serological tests:

The camel sera was examined for brucellosis using the following tests:

1. Buffered acidified plate antigen test (BAPA): as described by Alton et al, (1988), using antigen obtained from Veterinary Serum and Vaccine Research Institute, Ames, Iowa, USA.
2. Rose Bengal plate test (RBPT), was done according to Alton and Jones, (1967) and Blood et al, (1983); using antigen obtained from Veterinary Serum and Vaccine Research Institute, Abbassia, Cairo, Egypt.
3. Rivanol test, according to the techniques described by Alton et al, (1988); using antigen and Rivanol solution supplied by Animal and Plant Health Inspections services, National Veterinary Laboratories, Ames, Iowa, USA.
4. Serum agglutination test (SAT), as described by Alton et al (1988) using antigen obtained from Serum and Vaccine Research Institute, Abbassia, and Cairo, Egypt.
5. Mercapto-ethanol test (MET) was done as recommended by FAO/WHO committee of brucellosis in 1984 (FAO/WHO/OID, 1986).

### Preparation of antigens for Dot-immunobinding assay:

Whole bacterial antigen (Watari et al, 2002):

*Brucella abortus* strains 544 cells were grown in brucella broth and bacterial cells were harvested by centrifugations and washed ones with distilled water then inactivated with formalin (0.5% final conc.) adjusted to optical density 1.5 at 600nm.

### n. Lauroy Lsarcosin extracted antigen (Erden-ebaatar et al, 2003):

n.. Lauroy Lsarcosin (0.5% final conc.) was added to bacterial suspensions and the cells were incubated at room temperature for 30 min with shaking. The bacterial suspensions was centrifuged and filtrated and the supernatant was transferred to a new centrifuge tube and used as the antigen. The protein concentration of antigens was checked by Bio-Rad protein assay.

### 6. Dot-immunobinding assay (DIA): as described by Halling and Koster (2001):

Whole bacteria or n.lauroylsarcosin antigens (50:1) was applied on nitrocellulose membrane 0.45 um porosity (Bio-Rad laboratories) and incubated at room temperature for 30 min. Each well was blocked for 30 min with 100 ml of tris buffered saline (TBS) containing 0.1% bovine serum albumin (BSA) and 0.05% Tween 20. The wells were then washed 2 times with 100 ml of TBS containing 0.3% Tween 20.

Twofold dilutions from 1/40 to 1/10240 of camel serum were applied to wells and incubated for 30 min. Nitrocellulose membrane were washed three times with the same washing buffer then 100 ml of horseradish per-oxidase labeled protein G (diluted 1 :1000) was added and incubated at room temperature for 30 min.

The membrane was then washed again three times and the color was developed by adding 4-chloro-1-naphthol in diethyleneglycol and distilled water. The reaction was stopped with double ionized water. The titer was designated as the highest dilution gave a darker signal than that of control negative serum.

**2. Biochemical examinations:** The positive sera for brucellosis of camels raised in farms 1<sup>st</sup> group were deserted from our calculations of biochemical analysis because of two reasons a low number (one only) and one class of antibody titer 1/80; the rest considered as control. The rest samples from 2<sup>nd</sup> and 3<sup>rd</sup> groups were divided according to the antibody titer into three main categories 1/80 1/160 and 1/320. The selected positive samples were examined by two serological tests and Dot test. The serum samples were stored at -20°C for further biochemical analysis.

The following biochemical parameters were determined: total protein, Henry,(1969); glucose, Trinder,(1969); Albumin using bromocresol green method, Dourmas et al., (1972); GGT Mayer, (1973); LDH, Cabaud, (1958); total ALT and AST, Reitman & Frankel, (1957); ALP, EDKC, (1972); urea nitrogen, Hillel and Cook,(1971); uric acid, Caraway, (1966); creatinine, Bartels et al,(1972). The electrophoresis pattern for protein using protein standard (range from 20-250 kDa).Bio-

Rad, USA using Laemmli methods (Laemmli 1970); LDH-iso-enzyme, (Davis, 1964) and ALP iso-enzyme, (Shaw and Prasad,1970)

### 3. Statistical analysis:

Estimation of the relative sensitivity, relative specificity and estimated false positive was carried out according to (Crawford and Hidalgo, 1977):

Relative sensitivity =  $TP / TP + FN$ .

Relative specificity =  $TN / TN + FP$ .

Estimated false positive =  $FP / FP + TN$ .

TP (true positive): those confirmed as being positive by other 2 or more tests.

TN (true negative): those confirmed as being negative by other 2 or more tests.

FP (false positive): those confirmed as being positive by other one or two tests.

FN (false negative): those confirmed as being positive by other one or two tests.

The statistical analysis of the results was performed using T-test (student t test) (Farver, 1989).

## RESULTS

The numbers of positive reactors to each serological test in camel groups are shown in table (1). Regarding to the 1st group, out of 80 collected serum samples only one sample (1.25%) was positive for RBPT and doubtful for SAT as well as 2 samples (2.5%) were positive to BAPA. All examined samples were negative for Riv and MET.

Table 1: Number of positive reactor to each serological test in examined camels.

Group	Sampled No.	RB	B.A.P.A	Riv.	SAT		MET	
					±	+	±	+
1 <sup>st</sup> group	1 (1.25%)	2 (2.50%)	-	-	1 (1.25%)	-	-	-
2 <sup>nd</sup> group	6 (8.33%)	7 (9.72%)	6 (8.33%)	1 (1.38%)	4 (5.55%)	1 (1.39%)	5 (6.94%)	
3 <sup>rd</sup> group	9 (9.58%)	10 (10.64%)	9 (9.58%)	2 (2.13%)	6 (6.49%)	26 (2.13%)	7 (7.45%)	
Total (246)	16 (6.50%)	19 (7.72%)	15 (6.10%)	4 (1.60%)	10 (4.07%)	3 (1.20%)	12 (4.90%)	
				14 (5.70%)		15 (6.10%)		

Regarding to the 2<sup>nd</sup> group (72 serum samples), 6 samples (8.3%) were positive for RBPT, 7 samples (9.7%) were positive to B.A.P.A, 6 samples (8.3%) for Riv. Moreover one sample (1.3%) was doubtful for SAT and MET. Four (5.8%) and five (6.9%) samples were positive for SAT and MET respectively.

Third Group had apprehensive profile, the percent of positive samples for RBPT, B.A.P.A, Riv, SAT and MET were 9.5%, 10.6%, 9.5%, 6.4% positive and 2.1% suspects and 7.4% positive and 2.1% suspicious respectively.

The total incidence of brucellosis in examined camels using RBPT, B.A.P.A, Riv, SAT and MET, were 6.5%, 7.7%, 6.1%, 5.7% and 6.1%, respectively.

In DIA using whole bacterial antigen 5% (4 out of 80) of serum samples in the 1<sup>st</sup> group were positive with low antibody titre (1/80), from 72 serum

samples collected in 2<sup>nd</sup> group, 8 (11.10%) gave positive results with antibody titre ranged from 1/80 to 1/640.

When using n lauroyl Isarcosin extracts as antigen, only one sample from 1<sup>st</sup> group was positive with antibody titer 1/80, while 5 (8.3%) serum samples from 2<sup>nd</sup> group were positive with antibody titer ranged from 1/80 to 1/640. Regarding to 94 of 3<sup>rd</sup> group, 8 (8.5%) were positive with antibody titer 1/80 to 1/1280.

Sensitivity % (table 3) for Rb, B.A.P.A, Riv, TAT, MET and DIA using whole bacterial antigen or n. lauroyl Isarcosin extracts equals to 93.33%, 100%, 86.67%, 92.85%, 86.82%, 100% and 100% respectively. While specificity % equal to 99.13%, 97.89%, 99.13%, 99.57%, 99.13% 96.12% and 99.57%. Estimated false positive were 0.86%, 1.5%, 0.86%, 0.34%, 0.86%, 3.88% and 0.43% respectively.

Table (2): Detection of antibody titers in the serum of examined camels by Dot immuno-binding assay using whole bacterial or n-lauroylsarcosin extract:

Groups	Serological no. 23	Positive sera using DOT immuno-binding assay										
		Whole bacterial antigen					n-lauroylsarcosin					
		1/80	1/160	1/320	1/640	1/1280	Total +ve	1/80	1/60	1/320	1/640	1/1280
1 <sup>st</sup> group	4 (5.00%)	-	-	-	-	4 (5.00%)	1 (1.25%)	-	-	-	-	1 (1.25%)
2 <sup>nd</sup> group	2 (2.80%)	2 (2.80%)	1 (1.40%)	2 (2.80%)	-	2 (11.1%)	1 (1.40%)	3 (4.20%)	1 (1.40%)	1 (1.40%)	-	6 (8.30%)
3 <sup>rd</sup> group	2 (2.10%)	4 (4.20%)	2 (2.10%)	3 (3.20%)	-	11 (11.7%)	1 (1.00%)	2 (2.00%)	1 (1.00%)	2 (2.00%)	2 (2.00%)	8 (8.50%)
total (246)	7 (2.80%)	7 (2.80%)	3 (1.20%)	5 (2.00%)	-	23 (9.30%)	3 (1.20%)	5 (2.00%)	2 (0.80%)	3 (1.20%)	2 (0.80%)	15 (6.00%)

Table (3): Sensitivity %, specificity % and estimated false positive % for each test in camel serum samples:

Criteria	RBPT	BAPA	Riv	SAT	MET	DIA using W	DIA using n-l
Sensitivity %	93.33%	100%	86.67%	92.85%	86.87%	100%	100%
Specificity %	99.13%	97.84%	99.13%	99.57%	99.13%	96.12%	99.57%
Estimated false positive	0.86%	1.50%	0.86%	0.43%	0.86%	3.88%	0.43%

Table (4): Serum biochemistry profile in control and brucellosis infected camels (according to camel sources and antibody titer)

Items	1 <sup>st</sup> Group	2 <sup>nd</sup> Group			3 <sup>rd</sup> Group		
		1/80	1/160	1/320	1/80	1/60	1/320
AST (U/L)	8.76 ±0.77	14.86 ±1.33	8.07 ±0.86	9.83 ±0.99	21.43 ±1.86	13.97 ±0.18	8.38 ±0.39
ALT (U/L)	18.12 ±0.98	51.26 ±3.18	33.89 ±4.26	30.14 ±4.13	31.02 ±4.23	28.36 ±1.08	39.33 ±2.19
ALP (U/L)	65.72 ±0.89	128.76 ±5.97	98.01 ±0.67	141.08 ±3.28	103.36 ±7.67	103.87 ±0.48	97.64 ±3.69
AST (U/L)	24.72 ±0.24	72.92 ±2.13	55.49 ±0.44	97.14 ±0.33	72.34 ±4.58	56.44 ±0.27	63.57 ±0.28
ALT (U/L)	10.91 ±0.46	33.24 ±0.40	31.61 ±0.12	36.19 ±0.31	49.39 ±5.01	31.90 ±0.08	41.30 ±1.07

Mean ± Standard error

\*, \*\*, \*\*\* significant difference at probability at P<0.05, 0.01 and 0.001.

Table (5): Serum biochemical profile in control and brucellosis infected camels (according to camel sources and antibody titer):

Items	1 <sup>st</sup> Group	2 <sup>nd</sup> Group			2 <sup>nd</sup> Group		
		1/80	1/160	1/320	1/80	1/60	1/320
T. protein (g/dl)	5.77 ±0.19	5.65 ±0.22	5.57 ±0.17	5.49 ±0.20	5.08 ±0.20	5.66 ±0.29	5.91 ±0.25
Albumin (g/dl)	4.57 ±0.14	5.35 ±0.12	4.43 ±0.15	5.43 ±0.24	3.44 ±0.12	5.49 ±0.17	4.43 ±0.32
Glucose (mg/dl)	63.84 ±2.55	67.33 ±4.54	101.38 ±2.19	67.89 ±4.06	65.84 ±2.90	120.21 ±2.26	78.55 ±6.74
Urea (mg/dl)	27.71 ±0.42	37.07 ±3.36	30.71 ±0.70	36.41 ±6.75	29.12 ±0.91	30.28 ±0.64	30.53 ±4.94
Uric acid (mg/dl)	2.35 ±0.13	5.06 ±0.40	2.41 ±0.05	2.41 ±0.05	3.49 ±0.17	2.93 ±0.06	2.08 ±0.03
creatinine (mg/dl)	1.14 ±0.07	2.60 ±0.40	2.53 ±0.05	2.57 ±0.05	2.63 ±0.17	2.44 ±0.06	1.14 ±0.03

Mean ± Standard error.

\*, \*\*, \*\*\* significant difference at probability at P<0.05, 0.01 and 0.001.

Table (3): Serum protein profile in control and brucellosis infected camels (according to camel sources and antibody titer) (using mlab spol elkhaymahrouf)

1 <sup>st</sup> Group	Camels infected with brucellosis										
	2 <sup>nd</sup> Group		1/2160		1/320		1/80		3 <sup>rd</sup> Group		
Mod. w. %	Conc.	Mod. w. %	Conc.	Mod. w. %	Conc.	Mod. w. %	Conc.	Mod. w. %	Conc.	Mod. w. %	Conc.
(34.64) 8.60 20.16	0.18 20.02	(33.29) 10.3 20.01	0.56 20.04	(34.64) 11.00 20.73	0.53 20.03	(29.83) 8.76 20.32	0.26 20.01	(34.31) 9.43 20.62	0.59 20.04	(30.11) 4.96 20.28	0.33 20.02
(48.29) 3.13 20.21	0.21 20.01	(33.29) 10.3 20.05	0.56 20.04	(35.62) 3.24 20.15	0.25 20.01	(36.29) 9.67 20.64	0.43 20.02	(43.95) 9.27 20.61	0.59 20.03	(46.27) 11.30 20.74	0.74 20.05
(49.61) 21.85 21.44	1.50 20.09	(48.58) 9.54 20.63	0.52 20.03	(50.66) 6.78 20.45	0.30 20.02	(62.86) 1.11 20.07	0.07 20.01				
(60.71) 1.72 20.11	0.12 20.01			(56.23) 3.16 20.21	0.07 20.01						

Molecular weight: Mod. w. : KDa  
 % = equals 100  
 Conc. (from total level)  
 Mean ± Standard error (1/4)





Table (6): Serum protein profile in control and brucellosis infected camels (according to camel sources and antibody titer) (using slab gel electrophoresis).

1 <sup>st</sup> Group	Camels infected with brucellosis											
	2 <sup>nd</sup> Group						3 <sup>rd</sup> Group					
	1/80		1/160		1/320		1/80		1/60		1/320	
Mol. w. %	Conc.	Mol. w. %	Conc.	Mol. w. %	Conc.	Mol. w. %	Conc.	Mol. w. %	Conc.	Mol. w. %	Conc.	
(34.64) 5.60 ±0.36	0.38 ±0.02	(35.29) 10.3 ±0.68	0.56 ±0.04	(34.64) 11.00 ±0.73	0.53 ±0.03	(29.83) 5.76 ±0.32	0.26 ±0.01	(34.31) 9.43 ±0.62	0.59 ±0.04	(30.11) 4.96 ±0.28	0.33 ±0.02	
(35.29) 3.13 ±0.21	0.21 ±0.01	(35.62) 5.24 ±0.35	0.25 ±0.01	(36.29) 9.67 ±0.64	0.43 ±0.02	(45.95) 9.27 ±0.61	0.59 ±0.03	(46.27) 11.30 ±0.74	0.74 ±0.05			
(49.61) 21.85 ±1.44	1.50 ±0.09	(48.58) 9.54 ±0.63	0.52 ±0.03	(50.66) 6.78 ±0.45	0.30 ±0.02	(62.86) 1.11 ±0.07	0.07 ±0.01					
(60.71) 1.72 ±0.11	0.12 ±0.01	(56.23) 3.16 ±0.21	0.07 ±0.01									

Molecular weight: Mol. w.: KDa

% = equiva (10)

Conc. (From total level)

Mean ± Standard error (1/4)

Table (6): Serum protein profile in control and brucellosis infected camels (according to camel sources and antibody titer) (using slab gel electrophoresis (continued)):

1 <sup>st</sup> Group	Camels infected with brucellosis												
	2 <sup>nd</sup> Group					3 <sup>rd</sup> Group							
	1/80	1/160	1/320	1/80	1/320	1/60	1/320	1/80	1/60	1/320			
(66.63) 6.80 ±0.29	0.47 ±0.02	(65.54) 9.57 ±0.42	0.52 ±0.02	(66) 5.62 ±0.24	0.29 ±0.01	(65.09) 6.78 ±0.29	0.30 ±0.01	(68.55) 2.75 ±0.12	0.18 ±0.01				
(68.55) 13.30 ±0.58	0.91 ±0.04												
(74.67) 2.19 ±0.09	0.15 ±0.01	(80.56) 0.36 ±0.01	0.02 ±0.01	(75.38) 9.52 ±0.42	0.45 ±0.02	(71.21) 24.40 ±1.07	1.17 ±0.05	(73.96) 6.78 ±0.30	0.30 ±0.01	(77.57) 5.23 ±0.23	0.33 ±0.01	(79.04) 2.86 ±0.12	0.19 ±0.01
(87.74) 2.83 ±0.21	0.19 ±0.01			(86.09) 8.23 ±0.61	0.39 ±0.02	(88.58) 2.44 ±0.18	0.12 ±0.01	(84.47) 6.77 ±0.50	0.30 ±0.02	(87.74) 7.99 ±0.59	0.51 ±0.04	(86.09) 4.73 ±0.35	0.31 ±0.02
(100.94) 3.66 ±0.08	0.25 ±0.01			(98.96) 11.10 ±0.82	0.52 ±0.03	(98.96) 2.63 ±0.19	0.13 ±0.01	(99.75) 5.57 ±0.41	0.25 ±0.01	(99.35) 17.20 ±1.27	1.09 ±0.08	(99.35) 6.17 ±0.45	0.40 ±0.03
(105.03) 4.66 ±0.10	0.32 ±0.01	(104.20) 7.58 ±0.17	0.41 ±0.01	(104.62) 11.10 ±0.24	0.52 ±0.01	(104.62) 6.56 ±0.14	0.31 ±0.01	(103.38) 4.97 ±0.11	0.22 ±0.01	(104.20) 7.42 ±0.16	0.47 ±0.01	(103.38) 5.56 ±0.12	0.36 ±0.01
(107.57) 8.70 ±0.19	0.60 ±0.01							(108.43) 7.47 ±0.16	0.33 ±0.01				

Molecular weight: Mol. w.: KDa  
% = equals 100.  
Conc.: (From total level)  
Mean ± Standard error. (2/4)

Table (6). Serum protein profile in control and brucellosis infected camels (according to camel sources and antibody titer) (using slab gel electrophoresis (continued)):

1 <sup>st</sup> Group	Camels infected with brucellosis												
	2 <sup>nd</sup> Group				3 <sup>rd</sup> Group								
	1/80	1/160	1/320	1/80	1/60	1/320	1/80	1/60	1/320	1/80	1/60	1/320	
(110.16) 2.74 ±0.12	0.19 ±0.01	(115.54) 27.90 ±1.25	1.52 ±0.07	(116) 29.20 ±1.28	1.37 ±0.06	(110.16) 4.99 ±0.22	0.24 ±0.01	(115.54) 7.11 ±0.31	0.32 ±0.01	(110.6) 24.10 ±1.06	1.53 ±0.07	(107.57) 5.89 ±0.26	0.38 ±0.02
(132.63) 3.45 ±0.19	0.37 ±0.01	(134.12) 29.80 ±1.01	1.62 ±0.06	(131.16) 5.21 ±0.18	0.24 ±0.01	(135.63) 34.90 ±0.66	±1.67	(138.69) 28.80 ±0.98	1.29 ±0.04	(140.25) 4.65 ±0.15	0.29 ±0.01	(132.63) 33.60 ±1.14	2.18 ±0.07
(138.69) 3.45 ±0.10	0.21 ±0.01			(138.81) 4.76 ±0.16	0.22 ±0.01			(138.69) 28.80 ±0.98	1.29 ±0.04	(140.25) 4.65 ±0.15	0.29 ±0.01		
(158.81) 3.95 ±0.20	0.41 ±0.01												
(158.57) 2.25 ±0.07	0.15 ±0.01					(165.82) 5.22 ±0.17	0.25 ±0.01					(163.97) 13.2 ±0.45	0.86 ±0.03
(177.30) 1.78 ±0.06	0.12 ±0.01									(167.68) 5.11 ±0.17	0.32 ±0.17		

Molecular weight: Mol. w.: KDa  
 S<sub>2</sub> = equals 100  
 Conc. (From total level)  
 Mean ± Standard error. (3/4)

Table (6): Serum protein profile in control and brucellosis infected camels (according to camel sources and antibody titer) (using slab gel electrophoresis (continued):

1 <sup>st</sup> Group	Camels infected with brucellosis														
	2 <sup>nd</sup> Group				3 <sup>rd</sup> Group										
	1/80	1/160	1/320	1/80	1/60	1/320									
	*181.31 4.96 ±0.27	0.27 ±0.01	(183.34) 4.76 ±0.26	0.22 ±0.01	(181.31) 4.70 ±0.26	0.22 ±0.01	(189.59) 10.50 ±0.58	0.47 ±0.02	(189.59) 8.49 ±0.47	0.54 ±0.03	(198.25) 7.59 ±0.41	0.49 ±0.03			
	(193.87) 3.00 ±0.17	0.21 ±0.01													
207.30 ±0.07	0.09 ±0.01														
		(214.36) 2.77 ±0.15	0.13 ±0.01	(214.36) 3.16 ±0.17	0.15 ±0.01						(231.79) 1.39 ±0.07	0.09 ±0.01			
Total 100	6.85 ±0.54	100	5.44 ±0.33	100	4.67 ±0.38	*	100	5.78 ±0.61	100	4.47 ±0.68	**	100	6.32 ±1.16	100	6.50 ±1.22

Molecular weight: Mol. w.: KDa  
% = equals 100  
Conc. (From total level)  
Mean ± Standard error (4/4)

Table 10. From 1984 to 1990, people in control and knowledge infected a group for a group and another group of people.

Group	1984		1985		1986		1987		1988		1989		1990	
	Control	Knowledge	Control	Knowledge	Control	Knowledge	Control	Knowledge	Control	Knowledge	Control	Knowledge	Control	Knowledge
Group 1	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 2	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 3	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 4	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 5	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 6	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 7	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 8	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 9	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Group 10	100	100	100	100	100	100	100	100	100	100	100	100	100	100

Table 10. From 1984 to 1990, people in control and knowledge infected a group for a group and another group of people.

Table (8): Serum ALP iso-enzyme profile in control and brucellosis infected camels (according to camel sources and antibody titer) (using slab gel electrophoresis):

Items	1 <sup>st</sup> Group		Camels infected with brucellosis											
	%	Conc.	2 <sup>nd</sup> Group						3 <sup>rd</sup> Group					
			1/80		1/160		1/320		1/80		1/60		1/320	
ALP-1	30.00 ±4.40	19.48 ±2.17	44.90 ±2.63	38.65 ±0.63	29.20 ±3.44	28.59 ±3.85	25.80 ±1.33	25.19 ±5.11	35.80 ±2.55	40.82 1.08	41.10 ±3.87	41.93 ±3.70	30.40 ±2.65	42.89 ±2.22
ALP-2	34.85 ±0.75	22.88 ±2.24	28.60 ±2.37	24.62 ±2.01	40.70 ±4.30	39.85 ±3.91	35.80 ±1.21	34.96 ±0.83	36.80 ±1.58	41.96 ±2.66	25.80 ±2.10	26.32 ±3.81	31.60 ±2.78	44.58 ±2.33
ALP-3	35.15 ±3.65	23.19 ±3.18	26.50 ±0.03	22.81 ±2.43	30.10 ±0.87	29.47 ±1.24	38.40 ±0.12	37.49 ±4.65	27.40 ±0.45	31.28 ±2.21	33.10 ±1.73	33.77 ±2.47	38.00 ±0.23	53.61 ±4.30
Total	100	65.57 ±1.50	100	86.08 ±8.96	100	97.91 ±1.18	100	97.65 ±8.54	100	114.01 ±0.68	100	102.01 ±2.37	100	141.08 ±5.07

Mean ± Standard error.  
\*, \*\*, \*\*\* significant difference at probability at P<0.05, 0.01 and 0.001.

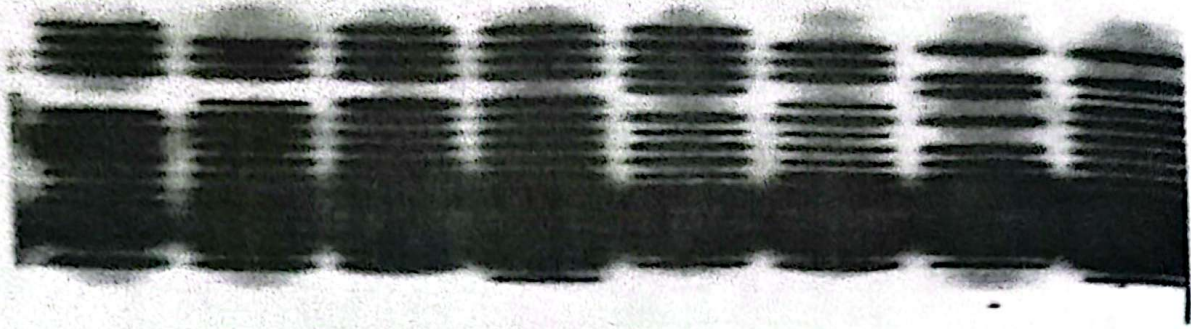


Fig 1: Serum protein profile of control and brucellosis infected camels.

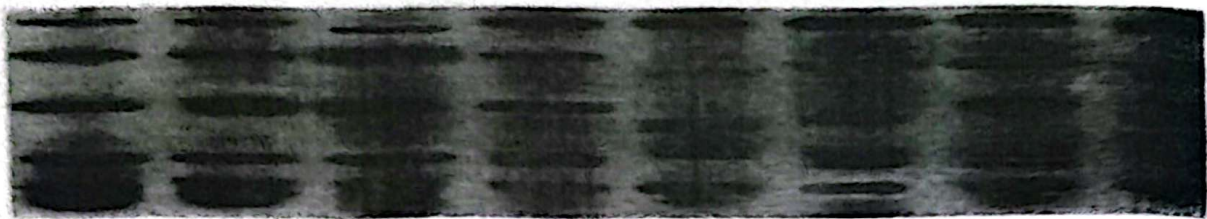


Fig 2: Serum LDH iso-enzyme profile of control and brucellosis infected camels.



Fig 3: Serum ALP-iso-enzyme profile of control and brucellosis infected camels.



Table 4 demonstrated that the GGT, LDH, ALP, AST, ALT levels were high significant increased ( $P<0.001$ ) in both either camels contact with animals or imported camels. The incidence of enzyme increases was in proportional with antibody titer in most tested enzymes in descending order (1/80, 1/160 and 1/320) except for LDH levels. The GGT level was increased only in camels sera of 1/80 antibody titer for brucellosis and decreased in 1/160 in imported camels sera.

Table 5 revealed the following observations, the total protein levels did not changed in 2<sup>nd</sup> group (camels contact with other animals infected with brucellosis) but high significant decrease in 3<sup>rd</sup> group (imported camels infected with brucellosis) with 1/80 and 1/160 antibody titer was found. The albumin level was high significant increased ( $P<0.001$ ) in 2<sup>nd</sup> group, especially with 1/80 and 1/320 antibody brucellosis titer. While, in 3<sup>rd</sup> group, albumin level was high significant decreased ( $P<0.001$ ) in camels sera (with 1/80 antibody titer) and non-statistical decreased in sera with 1/320 antibody titer. Glucose level was shown with high significant increased in 2<sup>nd</sup> group with 1/160 antibody titer only, but elevated in 3<sup>rd</sup> group. Urea, uric acid and creatinine levels were high significant increased in both 2<sup>nd</sup> and 3<sup>rd</sup> groups in the three different antibody categories (1/80, 1/160 and 1/360) with more commonness and severity in the later.

Table 6 and Fig1 that the sera of group 3 infected with brucellosis were characterized by an increase with low molecular weights protein bands of 29.83 -30.11 kDa, especially with 1/80, 1/320 an-

tibody titer. The levels of 34.64, 35.29 and 36.29 kDa sera of both 2<sup>nd</sup> and 3<sup>rd</sup> group of camels infected with brucellosis with 1/80, 1/160 and 1/320 antibody titer were increased as compared with control sera (1<sup>st</sup> group). The 3<sup>rd</sup> group sera of imported camels infected with brucellosis characterized by appearance of proteins of molecular weights ranged between 45.95-46.27 kDa. There was a decrease in the protein bands with molecular weight 66.63 in both camels groups compared with control sera. There was increase in conc. and % of 74.67, 87.74, 98.96, 99.75, 104.62, 110.57, 115.54, 132.63, 134.12, 138.69, 140.25 kDa protein bands in both groups (2<sup>nd</sup> and 3<sup>rd</sup> groups) infected with brucellosis with the three classified serological categories. Protein bands of 181.31-183.34 and 214.36 kDa in 2<sup>nd</sup> group sera infected with brucellosis and protein bands of 189.59 and 231.79 kDa in 3<sup>rd</sup> group sera infected with brucellosis especially in 1/320 antibody sera were shown.

The LDH-1, LDH-2, LDH-3, LDH-4 levels were high significant increased ( $P<0.001$ ), in (both % and conc.), in 2<sup>nd</sup> group and 3<sup>rd</sup> group with the three categories 1/80, 1/160 and 1/320 antibody titer (Table 7 and Fig 2). The LDH-5 level was high significant decreased ( $P<0.001$ ), in (both % and conc.), in both 2<sup>nd</sup> group and 3<sup>rd</sup> group with three categories 1/80, 1/160 and 1/320 antibody titer (table 7 and Fig 2).

The ALP-1, ALP-3 level were high significant increased ( $P<0.001$ ) in (both % and conc.) in 3<sup>rd</sup> group with the three categories 1/80, 1/160 and 1/320 antibody titer. The ALP-2 level was high sig-

nificant decreased ( $P < 0.001$ ) in (both % and conc.) in 2<sup>nd</sup> group and 3<sup>rd</sup> group with three categories 1/80, 1/160 and 1/320 antibody titer (table 8 and Fig3).

## DISCUSSION

The magnitude of brucellosis sero-prevalence in camels is based on serological surveys by a variety of procedures (table 1). In this study different serological tests were carried out to show a prevalence of the diseases among 3 groups of camels. The first group was kept in closed herds, the second group was kept in close contact with other animal species in country side and third group was imported camels from Sudan.

The results obtained in this study as summarized in table (1) revealed that among all examined camels (246) the percentage of reactors were 6.50%, 7.7%, 6.10%, 5.70% and 6.1% for RBPT, BAPA, Riv, SAT and MET respectively. The high percentage of RBPT and BAPA suggests the efficiency of these tests as screening tests for detection of recent and chronic infections of camel's brucellosis (Yagoub et al, 1990). The higher percentage of RBPT and BAPA as compared to SAT explained the basis that SAT may miss some infected animals especially in the chronic stage of the disease (Morgan, 1967). The equal number of MET and Riv reactors due to both tests detect the same immunoglobulin class Ig G (Alton et al., 1988).

Our results showed that the highest percent of positive reactors was observed in imported camels (Sudan's camel) in large herd followed by camels

in contact with other animals then followed by camels in closed farms (table 1). Camels of the first group were kept in closed farm under good hygienic conditions; this might explain the lower incidence of positive reactor among this group. Therefore, we can conclude the possibility of spread of brucellosis between different animal species and this might explain the higher incidence of the disease in the second group. This result agrees with Hashim et al, (1987) who reported that contact between camels and small ruminants was incriminated in transmission of brucellosis to camels. Also, Radwan et al, (1995) suggested that lateral transmission of the disease between different animal species can occur and play a serious role in spread of the disease. Abbas et al, (2000) demonstrated that camel pastoralists invariably keep relatively large flocks of sheep and goats along side the camels. Also, larger herds provide more chances of contact between animals leading to more chance of infection (Sulaiman, 1987).

In DIA using whole bacterial antigen, the incidence of positive cases was recorded as 9.30% with antibody titer ranged from 1/80 to 1/640. While the incidence of positive cases in camel using n-lauroylsarcosin extract as antigen was 6% with antibody titer ranged from 1/80 to 1/1280. DIA has an advantage over ELISA in case of soluble protein antigen that has low affinity to adhere to polystyrene plates or those antigens that could be altered in configuration when absorbed to the ELISA plates (Kumar et al, 1985).

In this study, the relative sensitivity, specificity and estimated false positive of different employed serological tests and DIA have been calculated as

shown in table (2). It is important to note that SAT has been shown with lower diagnostic sensitivity when compared to RBPT and BAPA (Alton et al, 1988). RBPT and BAPA are thus comparable and greater analytical sensitivity especially in detection of Ig G1 (Wright and Nielsen, 1990). In Riv and MET, the agglutination activity of Ig M have been removed (thus improving diagnostic specificity) while the promo tom of Ig G1, reactivity led to improved diagnostic specificity (Neilson et al, 1984).

Also, the results of sensitivity and specificity of DIA revealed that DIA using n.lauroylsarcosin extract is more specific than DIA whole bacterial antigen. Also, this result supported the previous investigation of Erdenebaatar et al, (2003) who suggested that an ELISA with n-laureylsarcosin extract is useful for differentiated between natural brucella to infected animals from brucella immunized animals.

Initially the brucellosis incidence percent among camels of closed farm may dedicate out the importance of an easy and good prognoses and diagnosis tools to eliminate the infected camels to get rid of affected camels, in epidemiology controlling of the disease.

The sera of infected camels with brucella (either camels contact with animals or imported) (Table 4) showed an elevation in each of the GGT, LDH, ALP, AST, ALT levels ( $P < 0.001$ ). Some of these enzymes were considered as liver specific enzymes as GGT, ALP, LDH and ALT. This indicate that in camels liver is the main pretentious organ in consequence of the brucellosis in camels.

However, we can not diffract between the consequences of brucella in camels groups either camels contact with animals or imported camels. But, the observed remark increase of enzymes levels which were in proportional to the presence of antibody titer in all tested enzymes in ascending order (1/80, 1/160 and 1/320) except for LDH levels is a good index for the incidence of the disease. The GGT level was increased only in camels sera of 1/80 antibody titer for brucellosis or even decreased in 1/160 in imported camels sera. The main unchanged GGT levels in brucellosis infections may reflect the importance of this enzyme in amino acid biosynthesis (Martin et al, 1985) this observation can be also confirmed by the unchanged total protein levels.

The severity consequence of the elevated enzymes associated with brucellosis is more pronounced in camels contact with animals infected with brucellosis more than in imported camels infected with brucellosis; this may be explained on the basis of the re-infections process. However, the increased incidence of brucellosis by serological test in the imported camels give attentions to the persist ant source of infections and brucella consequence especially with high titer (1/160 or more.) this indicated that in these categories the hepatic brucellosis takes a serious and sever directions owing to the asymptomatic character of this form (that localization by Brucella (hepatic brucellosis) Sisteron, (2002).

The increase of ALT in low response compared with the preceding AST, LDH and ALP in imported camels infected with brucellosis. Our results were in-agreement with EL-Sawalhy et al.,

(1996) who reported an increase in both ALT and AST. The explanations of our data indicated that the absence of one or more of regulating factors and mediators which controlling brucella infections (Ko et al., 2002), may explain the difference between the infected camels with brucellosis contact with other animals or imported camels.

However, the total protein levels did not changed in camels contact with other animals (table 5) but high significant decrease in imported camels especially with 1/80 and 1/160 antibody titer. Our data concerning local camels were in agreement with the results in Minia governorate of Thanaa, (1993) but disagree with (Thabet et al, 1993 and EL-Sawalhy et al., 1996). In the imported camels infected with Brucella, Brucella as suppressing factor beside other factors is responsible for the decreases in total protein level.

Albumin level in brucellosis infected camels was high significant increased ( $P < 0.001$ ) in camels contact with animals especially with 1/80 and 1/320 antibody brucellosis titer while in imported camels, significant decreased ( $P < 0.001$ ) in camels sera (with 1/80 antibody titer) and non-statistical decreased in sera with 1/320 antibody titer. This may be attributed partially, in the former group, to compensated the change in both fluid and trace minerals (copper and iron) metabolism (Thanaa, 1993) or due to leucocytic endogenous mediator, released during phagocytosis, stimulating liver mRNA for several amino acid transport (Thabet, 1993). In the later group of camels (imported), the albumin level decrease may be due to the changes in the kidney filtrations rate, and to the increase in sera beta-2 globulin levels which was

disturbed during kidney affections due to outer Brucella membrane (rough lipopolysaccharide) (Alonso-Urmeneta, 1998).

The significant increase of glucose level was found in camels sera contact with other animals with 1/160 antibody titer only as well as in imported camels with different antibody titer (1/80, 1/160 and 1/320). Our result doesn't agree with (Thanaa, 1993) in cattle and (Abu Damir et al., 1989 and Thabet, et al., 1993) in camels. The increase in sera glucose levels in infected camels can be connected with the increase of several enzymes in brucellosis involved glucose metabolism like glycosyltransferase, (Vermulpalii, 1999) erythrose phosphate dehydrogenase, malate dehydrogenase, hexokinase and CuZn superoxide dismutase (Corbel, 1997).

Alterations in liver protein metabolism affect kidney functions manifested by elevations of urea, uric acid and creatinine. All of them were increased in both camels contact and imported camels infected with brucellosis at the three different antibody categories (1/80, 1/160 and 1/360) with more commonness and severity in later. Due to several reasons increase in albumin fraction, increase adenine and guanine monophosphate inhibitors (Corbel, 1997), or due to a local recruitment due to Brucella contact phagocytes (Cazeville et al, 2000).

The protein profile of imported camels infected with brucellosis has more pronounced picture characterized by significant decrease in pre albumin, albumin and beta-1 globulin region ( $P < 0.001$ ) and high significant increased in both

alpha-1, alpha-2 globulin and beta-2 globulin level ( $P < 0.001$ ) in the three antibody categories (1/80, 1/160 and 1/320). In case of camels contact with animals infected with brucellosis the profile did not modify tremendously. The gamma -1 globulin level significant increased ( $P < 0.001$ ) in both either camels contact with other animals and imported camels infected with brucellosis in the three antibody categories (1/80, 1/160 and 1/320) while gamma-2 showed little changed. The changes associated with gamma-1 globulin can be connected with the increase of Ig A and Ig G levels in 85.6% patients infected with both acute and chronic brucellosis (Osoba et al, 2001 and Kutlu et al, 2003).

The protein had more complicated picture (table 6 and Fig 1) by using slab gels and molecular weight standards. However, it revealed several important points; the imported camels infected with brucellosis had more protein sera bands, the 1/320 antibody titer categories in both classes of examined camels had characteristic fractions. The source of these protein bands either from body reactions (Kittelberger et al., 1995) or as a result of the stress-induced proteins related to intracellular survival, or mediators to phagocyte functions (Debarh et al., 1995 and Alonso-Urmeneta, et al, 1998). The sera of imported camels infected with brucellosis were characterized by the increased levels of protein bands with molecular weights of 29.83 -30.11, 45.95-46.27 kDa. There was an increase in conc. and % of 34.64, 35.29, 74.67, 87.74, 98.96, 99.75, 104.62, 110.57, 115.54, 132.63, 134.12, 138.69, 140.25 kDa protein bands in both camels contact with animals and imported camels infected with brucellosis with all serologi-

cal categories. Comparably, there was a decrease in the protein bands of molecular weight 66.63 in both camels group compared with that control sera. Protein bands of 181.31-183.34 and 214.36 kDa in camels contact with animal's sera infected with brucellosis and protein bands of 189.59 and 231.79 in imported camel's sera infected with brucellosis especially in 1/320 antibody sera were observed.

The present data revealed an LDH-iso-enzyme finger print of brucellosis were not different among camels. These were associated with increase in total activity with significant increases of LDH-1, LDH-2, LDH-3, LDH-4 level ( $P < 0.001$ ) and decrease of The LDH-5 (table 7 and fig 2) (both % and conc.) in both camels contact with other animals and imported camels infected with brucellosis with three categories 1/80, 1/160 and 1/320 antibody titer. These manipulate of brucellosis indicated that the stress-protein released during the phagocytes which are influenced in several organs before/after brucella settled in the lymph and/or lymph node.

The results (table 8 and Fig 3) had shown in revealed that the ALP-1 and ALP-3 level were significant increased ( $P < 0.001$ ) in (both % and conc.) imported camels with three categories 1/80, 1/160 and 1/320 antibody titer.

**The present study, we can conclude that:**

1. The highest incidence of brucellosis in camels present in imported camels (Sudanese camels) followed by camels kept in contact with other animal and finely by the camels of farms, respectively.

1. The sera of infected camels with brucella (either camels contact with animals or imported) showed elevations levels of the GGT, LDH, ALP, AST, ALT, total protein, albumin, glucose, urea, uric acid and creatinine in both camels contact and imported camels infected with brucellosis at the three different antibody categories (1/60, 1/160 and 1/360).
2. The sera of imported camels infected with brucellosis are characterized by increased levels of protein bands with molecular weights 29.83 - 30.11 and 45.95-46.27 kDa. There was an increase in conc. and % of 34.64, 35.29, 74.67, 87.74, 98.96, 99.75, 104.62, 110.57, 115.54, 132.63, 134.12, 138.69, 140.25 kDa protein bands in both camels contact with animals and imported camels infected with brucellosis throughout the three classified serological categories.
3. Contrarily, There was a decrease in the protein bands of molecular weight 66.63 in both camel groups as compared with control sera.
4. Protein bands of 181.31-183.34 and 214.36 kDa in camels contact with animals sera infected with brucellosis and protein bands of 189.59 and 231.79 in imported camels sera infected with brucellosis especially in 1/320 antibody sera were present.
5. Characteristic changes of LDH and ALP isoenzyme were observed of brucellosis infected camels.
6. The imported camels infected brucellosis as

well as camels in contact with animals infected with brucellosis had biochemical changes related with to the products of protein metabolism and liver functions activity.

7. The brucellosis incidence percent in camels in closed farm may be easy to be good prognosis and diagnosed can be facilitate the action work to eliminate the infection in camels and get rid of the affected camels, as epidemiological control of the disease.

## REFERENCES

- Almas, B.G; Qasbi, A.A and AlHawari, A. (2000): Survey of camel husbandry in Qadisiyah region, Iraqi Journal Res. Adv. vet. med. exp. 55:285-293.
- Abu-Damir, H; Tag, E; Dar, H; Kenyon, L.I and Abu, I.F. (1989): Isolation of brucella abortus from experimentally infected dromedary camels in Sudan. A preliminary report. Vet. Res. Commun. 13: 413-416.
- Almas-Urmeneta, B; Marm, C; Arques, V.; Blasco, J.M; Diaz, R. and Meryon, I. (1998): Evaluation of immunoprecipitates and polysaccharides of different epitope structure in indirect enzyme-linked immunosorbent assay for diagnosis of brucellosis in small ruminant and cattle. Clin. Diagn. Lab. Immunol. 1998, Nov; 5 (5):749-54.
- Alam, G.D. and Jones, L. M. (1967): Laboratory techniques in brucellosis. Wild. Health Org. Tech. Report. Ser. No:55:1-69.
- Alam, G.D; Jones, L. M.; Arques, F.D and Vergey, J.M. (1988): Techniques for brucellosis laboratory manual national De La Recherche Agronomique 147. Rue De L'université, 75017 Paris.
- Borvik, G; Marm, C.; Bøks, M.; Dam, N. and Mery, I. (1972): Kinetic determinants of creatinine utilization in

- tainizations. *Clin. Acta.* 1972,37.:193-197.
- D.C.; Rodostits, M. and Hendensin, J.A. (1983): Serological methods. *Veterinary medicine* 6th edition. Baillier Tindall, London.
- ad. D.; Alan, B; Marin, C.; Bkico., M.; Dia, K. and Jory, L. (1958): colorimetric determination of LDH activity according to the Cabaud and Wroblewski method. *m. J. Clin. Path.* 1958, 30(2):234-254.
- way, W.G; Jon, M. M. and Meryer, J.M. (1966): Techniques for of uric acid determinations in alkaline medium. *Clin. Chem.* 1966,12.:16-18..
- vieille, C.; Porte, F., Bettache, S., Liautard, J.P. and Widada, J. (2000): Early events and implication of actin and annexin I associated structure in the phagocytes uptake of *Brucella suis* by the J-74A.1 murine cell line and human monocytes. *Microb. Pathog.* 28(6):343-2.
- el. MJ (1997): Brucellosis: an overview. *Emerg. Infect. Dis.* 1997, April:3(2):213-21.
- xford, R. P. and Hidalgo, R.J. (1977): Bovine brucellosis, international symposium texas A&A university press collage, station, london.
- is, B.J. (1964): the electrophoresis II. Method and applications to human serum proteins. *Annals, new york Academy of Sciences*, 121: 404-427.
- barh, H.S.; Cloeckaert, A.; Zygmunt, M.S. and Bubray G. (1995): Identifications of seroreactive *Brucella melitensis* cytosoluble proteins which discriminate between antibodies elicited by infection and Rev.1 vaccination in sheep. *Vet. Microbiol.* 1995 Apr; 44(1):37-48.
- imas, B.A.; Maco, A.J. and Heth, B.R. (1972): In standards Methodes of clinical methods. *Acad. Press N.Y.* 1972(7):175-180.
- «C Empfehlungen der Deutschen Gesellschaft für Klinische (1972): kinetic determinations of alkaline phosphatase activity as recommended by the German clinical society. *Z. Klin. Chem.. Biochem.* 1972,10:182.
- El-Sawalhy, A.A.; Montaser, A. M. and Rizk, L.G. (1996): Diagnostic and biochemical evaluations of camel brucellosis. 4<sup>th</sup> Sci. Proc. April3-6,1996. *Vet. Med. J. Giza.* Vol.44, No.2 (1996):332-329.
- Erdenebaatar, J.G; Byarsaikhan, B. ; Watarai, M.; Makino, S. and Shirahata, T. (2003): Enzyme linked Immunosorbent assay to differentiate the antibody responses of animals infected with brucella species from those of animals infected with *yesimia enterocolitica* 0:9. *Clin.Diag.Lab.Immunol.*(10)4:710-714.
- FAO/WHO/OID, (1986): Brucellosis epidemiology, serology and diagnosis. FAO/WHO committee of brucellosis in 1984, p 33-37, news letter. Geneva.
- Farver, B.T.,(1989): Concepts of normality in clinical biochemistry. In *clinical Biochemistry of domestic animals*, Kaneko, edit. P2-18 Academic Press. New York.
- Halling, S.M and Koster, N.A. (2001): Use of different extract of brucella abortus RB51 to detect serologic response in RB51 vaccinated cattle. *J.Vet.Diagn. Invest.* 13:408-412.
- Hashim, N.H.; Galil, G.A.; Hulaibi, M.A. and Al-Sleim, E.M. (1987): The incidence of brucellosis and species of brucella organisms isolated from animals Al Has, Saudia Arabia. *World Animal Rev.* 61:32-53.
- Henry, H. (1969): determination of total protein "Hawks clinical chemistry" Lang Press. P38-70. Hillelt, C.J. and Cook, L.G.H. (1971): enzymatic determinations of urea-urease-GLDH) method. *Clin. Chim. Acta.* 1971,36,33-54.
- Kiel, F.W and Khan, M. (1989): Brucellosis in Saudi Arabia. *Soc. Sci. Med.* 29:999-1001.
- Kittelberger, R.; Hilbink, F.; Hansen, M.F.; Ross, G.P. and Bruyn, J. (1995): Identifications and characterization of immunodominant antigens during the course of infection with *Brucella ovis*. *J.Vet. Diagn. Invest.* 1995 Apr; 7 (2):210-8.
- Ko, J.; Gendron-Fitzpatrick, A. and Splitter, G.A.

- (2002): susceptibility of INF regulatory factor-1 and IFN consensus sequence binding protein deficient mice to brucellosis. *J. Immunol.* 1:168(5):2433-40.
- Kultlu, S.S.; Celikbass, A. Ergonul, O.; Kutlu, M.; Aksaray, S. and Guverner, E.etc (2003): the value of immunoglobulin G avidity test for serologic diagnosis of brucellosis. *Mikrobiyol Bul.* 2003, Oct;37(4):261-7.
- Kumar, S.; Band, A.H., Samantaray, J.C.; Dany, N. and Talwar, G.P. (1985): A dot enzyme linked immunosorbent assay for detection of antibody against *Entamoeba histolytica*. *J. Immunol. Methods* 83:125-133.
- Laemmli, U.K. (1970): Electrophoretic separations of proteins. *Nature*, 227,680-684. Martin, W.D; Mayes, P. A. and Rodwell, V.W. (1985): "biosynthesis of amino acids" Harper's review of biochemistry. Lange medical publications, 19th ed, P267.
- Mayer, K. (1973): "Das medizinische Laboratorium", 18th ed, P26, 125-132, 172-180, 198-204.
- Morgan, W.J. (1967): The serological diagnosis of bovine brucellosis. *Vet. Rec.* 80(20)612-621.
- Neilson, K.; Hech, F.C.; Wagner, G.G.; Stiller, S.; Rosenbaum, B.; Pugh, R. and Flores, E. (1984): Comparative assessment of antibody isotypes to brucella abortus by primary and secondary binding assays. *Prev. Vet. Med.* 2,1:197-205.
- Osoba, A.O.; Balkhy, H. Memish, Z.; Khan, M.Y.; Al-Thagafi, S. and Al Shareef, A. (2003): Diagnostic value of brucellosis ELISA IgG and IgM in bacteremic and non-bacteremic patients with brucellosis. *J. Chromatography* 2001, Aug;69(8):4816-22.
- Palling, R.W.; Waghela, S., Macowan, K.J. and Health, B.R. (1988): The occurrence of infectious diseases in mixed farming of domesticated wild herbivores and live stock in Kenya II. Bacterial diseases. *J. Wild Dis.* 24 (2):308-312.
- Radwan, A.I.; Bekairi, S.I.; Mukayel, A.A.; Al-Bokmy, A.M. Prasad, P.V. Azar, F.N., and Coloyan, E.R. (1995): Control of brucella melitensis infection in large camel herd in Saudi Arabia using antibiotherapy and vaccination with Rev.1. *Vaccine Rev. Sci. Technol.* 14:719-732.
- Reitman, S. and Frankel, S. (1957): colorimetric determination of GOT and GPT activity according to the Reitman and Frankel method. *Am. J. Clin. Path.* 1957, 28(2):56-64.
- Shaw, C.R and Prasad, R. (1970): starch gel electrophoresis of enzymes - A compilation of recipes. *Biochem. Genet.* 4,4:297-320.
- Sisteron, O.A. J. Soucia, P. Chevalliera, E. Cuab and J. N Brunetona (2002): Hepatic abscess caused by Brucella: US, CT and MRI findings-Case report and review of the literature. A Service de Radiologie, Hopital l'Archet I; 151 Route de Saint-Antoine de GinestiÈre. -06202 Nice cedex 3, France b Service de MÈdicine Interne HÙpital l'Archet I, 151 Route de Saint-Antoine de GinestiÈre, F-06202, Nice cedex 3, France.
- Sulaiman, M.A. (1987): The prevalence of bovine brucellosis in khartoum and Gezira areas. M.V.Sc thesis . Khartoum, Sudan.
- Thabet, A.; Abd El-Fattah, A. M. and Manna, A.M.; Manna, A.M.; Sayed, A. M and Ibtisam, M.H. (1993): Prevalence of brucellosis among camels in New Vally governorate. 2<sup>nd</sup> Sci. Cong. Egyptians Society for camel diseases, 5-7 Dec, Assiut, Egypt. 290-296.
- Thanaa, Y.N. (1993): levels of some vitamins and trace elements in the blood of Brucella infected cattle in Matruh governorate. M.V.Sc. Thesis Assiut University.
- Trinder, P. (1969): enzymatic determinations of glucose. *Ann. Clin. Biochem.* 1969, (6):24-34.
- Watari, M.; Makino, S. I. and Shirahata, T.M. (2002): A essential virulence protein of brucella abortus, VirB10, requires an intact nucleoside triphosphate-binding domain. *Microbiol.* 148:1439-46.
- Wright, P.F. and Nielsen, K. (1990): Current and future trends in the diagnosis of brucellosis. *Vet. Med. J., Giza.* Vol.54, No.2(2006)



ological methods. In Admsi, L.G.(ed.)Proceedings of the international symposium on advances in brucellosis. Texas A &M Univerisity collage station, T.X.

Yagoub, L. A; Mohamed, A. A. L. and Salim, M. O. (1990): Serological survey for brucella abortus antibody prevalence in the one humped camel (*Camelus dromedarius*) from Eastern Sudan Rev Elv. Med. Vet. Trop. 43: 167-171.

# الإختبارات السيرولوجيا والتغيرات الكيميائية الحيوية

## فى الجمال المصابة بالبروسيلة

ب. ج. ١٠. فهمى x وهدى م. زكى xx

قسم الكيمياء الحيوية وأمراض النقص الغذائى والسّموم \* وقسم البروسيلة xx

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لإجراء هذا البحث تم أخذ عينات دماء من ٨٠ من الجمال المرباه فى المزارع المختلفة (مزرعة الإنتاج الحيوانى ومزارع أخرى) بالإضافة إلى عينات من ٧٢ من الجمال التى تربي فى وجود حيوانات الزراعة الأخرى مثل الأبقار والجاموس وخلافه من مناطق عديدة فى محافظة الجيزة وعينات أخرى من ٩٤ من الجمال المستوردة من السودان لغرض الذبح فى أسواق الجمال.

وأظهرت النتائج أن أعلى النسب الإيجابية بالإختبارات السيرولوجيا كانت فى الجمال المستوردة وكانت تراوح بين ٨.٥٠٪ إلى ١١.٧٠٪ بإستخدام (إختبار بفر د أسيدفايد بليت أنتيجين (BAPA) وإختبار روس بنجال (RBPT) وإختبار روس بنجال (RBPT) وإختبار ريفانول (Rew.) وإختبار التلزن الدموى (SAT). وإختبار مركبتو - إيثانول (MET) وإختبار الإرتباط المناعى (٨.٣٠٪ و ٩.٤٠٪ و ٨.٣٠٪ و ٦.٩٤٪ و ٨.٣٣٪ و ٨.٣٠٪ إلى ١١.١٠٪) على الترتيب بإستخدام الإختبارات السابق ذكرها. يليها الجمال المرباه ومع الحيوانات الأخرى وكانت النسبة بين ٦.٩٤ إلى ١١.١٠٪ (٨.٣٠٪ و ٩.٤٠٪ و ٨.٣٠٪ و ٦.٩٤٪ و ٨.٣٣٪ و ٨.٣٠٪ إلى ١١.١٠٪) على الترتيب وأخيراً الجمال المرباه فى المزارع المختلفة وكانت النسب بين ١.٥٠٪ إلى ١١.٢٥٪ و ١٢.٥٠٪ و ١٠.٠٠٪ و ١١.٢٥٪ و ٧.٠٪ إلى ١٥٪). كما أظهر النتائج أهمية إستخدام إختبار الإرتباط المناعى النقطى (DIA) بإستخدام مادة ن-لايورولساركوسين عن إستخدام كل المضاد البكتيرى.

لم تختلف التغيرات الكيميائية الحيوية نتيجة الإصابة بالبروسيلة فى الجمال المستوردة عن تلك المرباه بين لحيوانات الأخرى ففى الحالتين لوحظ أرتفاع فى أنزيمات الأنزيم الناقل للجاما جلوناميك واللاكتيت دى هيدروجينيز الألكلين ترانسفيريز واللانزيم الناقل للالنين والأخر للأسيرتيت. وأيضاً إرتفاع فى عناصر البروتين الكلى والألبومين الجلوكوز واليوريا حمض البوليك والكرياتينين.

إختلفت الصورة عندما تم عمل الفصل الكهربائى لبروتين المصل حيث تميز مصل الجمال المصابة بالبروسيلة لمستوردة بظهور بروتينات لها وزن جزئى يتراوح بين ٢٩.٨٢ إلى ٢٠.١١ ومن ٤٥.٩٥ إلى ٤٦.٦٧ كيلو دلتون مع إرتفاع فى عسويات بروتينات لها أوزان ٣٦.٦٤ . ٣٥.٢٩ و ٧٤.٦٧ و ٨٧.٧٤ و ٩٨.٩٦ و ٤.٦٢ . ١٠.٤ أو ١٠.٥٧ أو ١١٠.٥٧ و ١١٥.٥٤ و ١٢٢.٠٢ و ١٢٨.٦٩ و ١٢٤.١٢ و ١٤٠.٢٥ كيلو دلتون فى نوعى الجمال محل الدراسة والمصابة بالبروسيلة

أما البروتينين ١٨١.٣١ - ١٨١.٣٤ و ٢١٤.٢٦ كيلو دلتون فقد ظهرت فى الجمال المرباه مع الحيوانات أخرى بينما ظهرت بروتينات ١٨٩.٥٩ و ٢٣١.٧٩ كيلو دلتون فى أمصل الجمال المستوردة فقط وخصوصاً فبا الأمصال الإيجابية ذات نوى ٢٢٠/١.

أظهرت النتائج صورة مميزة عند فصل كل من أنزيم الأكتين دي هيدروجينيز والألكلين فوسفاتيز كهرتابياً في حالة إصابة الجمال بالبروسبلا .

تستخلص من البحث النتائج التالية أن الجمال المسنودة بلبها الجمال المرباه مع الحيوانات الأخرى هي مصدر العدوى والإصابة بالبروسبلا للجمال والحيوانات الأخرى تشير النتائج أيضاً أن الصورة الكيمبائية الحيوية في الجمال قد تأخذ منحى وتأثير على الكبد ووظائفه والبناء البروتيني والأبيض له بصورة مختلفه عن المعروف في حالة الإصابة . الإصابة في الحيوانات الأخرى .

نسب الإيجابية المنخفضه في الجمال المرباه في المزارع إنما تشير إلى أهمية توافر الكشف السريع والتشخيص والإختبارات العملية الدقيقة في إكتشاف الإصابة مبكراً والتخلص من الجمال المصابة سريعاً كوسيله من الوسائل هي تنبع وبائية المرض والقضاء عليه .