

## LIVER AND KIDNEY DYSFUNCTIONS AMONG BRUCELLA REACTANT FRIESIAN COWS IN EL-MINYA PROVINCE, EGYPT

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### SUMMARY

The present study was carried on 50 Friesian female cows (3-5 years old) in El-Minya Province, Egypt. Serological investigation of cattle for brucellosis revealed that, 68, 56, 60, 48 and 56% of the tested cows were brucella reactant to the Buffered Acidified Plate Antigen Test (BAPAT), Rose Bengal Plate Test (RBPT), Serum Agglutination Test (SAT), Rivanol Test (RT) and Complement Fixation Test (CFT), respectively.

Biochemical analysis of the tested cows' sera showed that brucella reactant cows had a significant increase in serum Alanine Amino Transferase (ALT), Aspartate Amino Transferase (AST), Alkaline Phosphatase (AP), Urea and Creatinine levels. Significant increase of serum total proteins, globulin, lipids, cholesterol, triacylglycerol, free fatty acids and the high, low and very low density lipoproteins were also detected. On the

other hand significant decrease in serum albumin, A/G ratio, phospholipids, calcium, inorganic phosphorus, magnesium, sodium and potassium levels were shown among the brucella reactant Friesian cows compared to the non-reactant controls.

It could be concluded that, in addition to the hazard of distributing brucella infection, brucella reactant cows under study suffered hepatic and renal dysfunction that altered their serum constitution, which in turn implies itself on the animal health and they should be eradicated from the producing cattle herd as soon as possible.

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### INTRODUCTION

Brucellosis is considered a worldwide disease of economical importance that affects animal productivity and reproduction. Although most of the reports about cattle brucellosis dealt with the

serological testing and their comparative diagnostic efficacy, few of them dealt with the clinical chemistry profile of the brucella reactant cattle.

Using the BAPAT, RBPT, SAT and RT, Kadry, (1996) detected cattle brucellosis in 0.99%, 0.94% 0.86% and 0.87% respectively, out of the examined 2330 cattle from different localities at El-Sharkia Province. Ahmed and Nada, (1993) detected a significant increase in serum alkaline phosphatase and serum globulin values together with a significant decrease in serum albumin level among the brucella reactant camels. Ghazy, et al., (2000) reported that, a significant increase in serum total protein, globulin, bilirubin, indirect bilirubin and urea levels together with a significant decrease in serum albumin level were detected among the brucella reactant horses of different age and sex.

The present study was carried out to provide an additional biochemical diagnostic data among the brucella reactant cows with special reference to the liver and kidney dysfunction that might accompany the brucellosis syndrome in affected cattle.

## MATERIAL and METHODS

Serum samples were collected from 50 Friesian cows, in the high production age (3-5 years old), for the serological and biochemical studies.

### I- Serological Examination:

Buffered Acidified Plate Antigen Test (BAPAT), Rose Bengal Plate Test (RBPT), Serum Agglutination Test (SAT), Rivanol Test (RT) and Complement Fixation Test (CFT), were carried out according to the methods described by Alton, et al. (1988).

### II- Biochemical Examination:

According to the results obtained by the serum agglutination test (SAT), the tested cows were grouped into four groups: 20 SAT brucella non-reactant cows, 8 (1/20), 10 (1/40) and 12 (1/80) SAT brucella reactant Friesian cows.

The serum samples were collected and analyzed biochemically for the total protein, albumin, globulins, amino-transferases (ALT and AST), alkaline phosphatase (AP), urea and creatinine according to the methods described by, Hoffmann and Richterrich, (1979), Doumas, et al., (1971), Reitman and Frankel (1957), Kilichiling and Freiburg (1951), Tabacco, (1979) and Husdan and Rapport, (1968), respectively.

The lipogram: total lipids, total cholesterol, triacylglycerol, free fatty acids, phospholipids, and the high density lipoproteins (HDL), low density lipoproteins (LDL), very low density lipoproteins (VLDL) were measured in serum samples according to the methods described by Knight, et al. (1972), Watson, (1960), Fossati and Principe (1982), Schuster, (1979), Zilvermit and Davies

(1950) and Lopez-Virella, et al., (1977), respectively.

Mineral concentrations of serum calcium, inorganic phosphorus, and magnesium levels of the collected sera were carried on according to the methods described by Glinder and King, (1972), Kilichling and Freiberg, (1951), Neil and Nelly, (1956), respectively. Sodium and potassium serum levels were determined using the Corning 410C flame photometer according to Oser, (1979).

Statistical analysis of the obtained data for the mean, standard error and t testing for significant differences between serum values of the brucella

SAT non-reactant controls and the reactant cows were carried out according to Snedecor and Cochran (1976).

## RESULTS

Table (1), represents the percentage of brucella reactant and non-reactant Friesian cows in El-Minya Province according to the used serological tests.

Tables (2), (3), (4) and (5) present the biochemical data obtained from serum analysis of the brucella SAT non-reactant and the SAT reactant Friesian cows in El-Minya Province, Egypt.

Table (1): Results of Serological Examination for Cattle Brucellosis Using Different Serological Tests.

Serological Tests	Examined Friesian Cows (n = 50)		
	% (-) ive	% (+) ive	% (±)
Buffered Acidified Plate Antigen Test (BAPAT).	32	68	--
Rose Bengal Plate Test (RBPT).	44	56	--
Serum Agglutination Test (SAT).	40	44	16
Rivanol Test (RT).	52	48	--
Complement Fixation Test (CFT)	44	52	4

% (-) ive = Percentage of Brucella non-reactant cows.

% (+) ive = Percentage of the Brucella reactant cows.

% (±) = Percentage of suspiciously reacting cows.

Table (2): Serum Liver and Kidney Function Tests among Brucella SAT Non-reactant and the Reactant Friesian Cows in El-Minya Province.

Test Parameters	Negative Reactors (n = 20)	Reactants at 1/20 (n = 8)	Reactants at 1/40 (n = 10)	Reactants at 1/80 (n = 12)
ALT (U/L)	25.80 ± 0.75	28.38 ± 1.02	35.30*** ± 1.33	36.58*** ± 0.22
AST (U/L)	33.30 ± 1.11	40.25** ± 1.90	45.00*** ± 1.03	55.42*** ± 1.44
AP (U/L)	2.03 ± 0.05	2.13 ± 0.07	2.39*** ± 0.05	2.46*** ± 0.06
Urea (mg/dl)	26.81 ± 1.13	41.43*** ± 0.54	46.71*** ± 1.22	49.34*** ± 0.91
Creatinine (mg/l)	16.90 ± 0.03	17.20* ± 0.16	17.50*** ± 0.08	17.65*** ± 0.08

SAT = Serum Agglutination Test. ALT = Alanine Amino Transferase.  
 AST = Aspartate Amino Transferase. AP = Alkaline Phosphatase.  
 \* = Significant at P< 0.05 \*\* = Highly significant at P< 0.01  
 \*\*\* = Very highly significant at P< 0.001

Table (3): Serum Proteinogram Values among Brucella SAT Non-reactant and the Reactant Friesian Cows in El-Minya Province.

Test Parameters	Negative Reactors (n = 20)	Reactants at 1/20 (n = 8)	Reactants at 1/40 (n = 10)	Reactants at 1/80 (n = 12)
Total Protein (g / dl)	6.90 ± 0.11	7.45** ± 0.15	7.56** ± 0.18	7.78*** ± 0.15
Albumin (g / dl)	3.27 ± 0.05	3.00 ± 0.18	2.76*** ± 0.11	3.00** ± 0.08
Total Globulin (g / dl)	3.63 ± 0.13	4.45** ± 0.10	4.80*** ± 0.10	4.78*** ± 0.22
A / G Ratio	0.90 ± 0.02	0.67*** ± 0.04	0.58*** ± 0.04	0.64*** ± 0.04

SAT = Serum Agglutination Test.  
 A / G Ratio = Albumin / Globulin Ratio.  
 \*\* = Highly significant at P< 0.01  
 \*\*\* = Very highly significant at P< 0.001.

Table (4): Serum Lipogram Values among Brucella SAT Non-reactant and the Reactant Friesian Cows in El-Minya Province.

Test Parameters	Negative Reactors (n = 20)	Reactants at 1/20 (n = 8)	Reactants at 1/40 (n = 10)	Reactants at 1/80 (n = 12)
Total Lipids (mg / dl)	415.6 ± 13.8	480.5*** ± 10.0	537.0*** ± 13.3	560.0*** ± 11.0
Total Cholesterol (mg / dl)	148.5 ± 6.8	179.8* ± 9.8	203.5*** ± 6.0	211.3*** ± 7.9
Triacylglycerol (mg / dl)	43.1 ± 1.5	56.9*** ± 1.8	66.8*** ± 1.7	75.7*** ± 1.3
Free Fatty Acids (mg/ dl)	1.8 ± 0.12	2.2** ± 0.06	2.5*** ± 0.04	2.6*** ± 0.03
Phospholipids (mg/ dl)	220.0 ± 7.0	201.1 ± 12.8	183.8*** ± 5.9	187.6** ± 6.6
HDL (mg / dl)	30.7 ± 1.8	36.1 ± 2.3	42.2*** ± 2.1	42.9*** ± 2.6
LDL (mg / dl)	109.6 ± 2.3	153.6*** ± 6.4	131.4*** ± 2.2	132.6*** ± 1.8
VLDL (mg / dl)	8.6 ± 0.1	10.7*** ± 0.2	12.6*** ± 0.3	14.0*** ± 0.2

SAT = Serum Agglutination Test.

HDL = High Density Lipoproteins

LDL = Low Density Lipoproteins

VLDL = Very Low Density Lipoproteins

\* = Significant at P< 0.05

\*\* = Highly significant at P< 0.01

\*\*\* = Very highly significant at P< 0.001.

Table (5): Some Serum Mineral Values among Brucella SAT Non-reactant and the Reactant Friesian Cows in El-Minya Province.

Test Parameters	Negative Reactors (n = 20)	Reactants at 1/20 (n = 8)	Reactants at 1/40 (n = 10)	Reactants at 1/80 (n = 12)
Calcium (mg / dl)	8.88 ± 0.30	7.95* ± 0.13	7.96* ± 0.11	7.50*** ± 0.21
Inorganic Phosphorus (mg / dl)	9.45 ± 0.62	8.73 ± 0.50	8.12* ± 0.15	7.99* ± 0.19
Cal x Ph (Value)	83.92 ± 0.19	69.56*** ± 0.27	64.64*** ± 0.12	59.93*** ± 0.14
Magnesium (mg/ dl)	2.65 ± 0.06	2.12* ± 0.13	2.42** ± 0.03	2.13** ± 0.06
Sodium (m Eq / L)	125.3 ± 1.24	117.4*** ± 0.46	114.8** ± 1.11	114.8*** ± 0.45
Potassium (m Eq / L)	4.84 ± 0.45	2.93** ± 0.45	2.39** ± 0.57	2.40*** ± 0.13
Na / K (Ratio)	25.88 ± 1.07	40.06*** ± 0.53	48.03*** ± 0.68	47.81*** ± 1.80

SAT = Serum Agglutination Test.

\* = Significant at P<0.05

\*\* = Highly significant at P<0.01

\*\*\* = Very highly significant at P<0.001.

## DISCUSSION

The serological results obtained by the BAPAT and RBPT tests (table, 1) showed that, the incidence of brucella reactor Friesian cattle in El-Minya Province were 68 and 56%, respectively. These results revealed that BAPAT detects the highest number of positive reactors and this may be ascribed to the fact that the test is more sensitive in detecting IgM as well as IgG immunoglobulins (Stemshorn, et al., 1985). The higher number of BAPAT positive reactors (12% more than those detected by RBPT) could be attributed to

the higher amount of serum used in this test in addition to the fact that the lower pH 3.6 of the RBPT antigen allows less amount of IgM reaction (Alton, et al., 1976) and inhibits non specific reaction of serum agglutinins (Morgan, 1978).

Concerning the SAT, RT and CFT, the incidence were 60, 48 and 56% respectively. The obtained results showed that SAT was slightly more sensitive than the other two tests and this could be attributed to its greater ability in detecting IgM immunoglobulins (Alton, et al., 1976) while CFT is mainly an IgG<sub>1</sub> detector. The lower incidence

detected by the RT (48%) could be a matter of the IgM precipitation prior to the test procedure and so it detects mainly the IgG2 immunoglobulin (Alton, et al., 1988). However, CFT appeared to be the most efficient test used in the diagnosis of brucellosis (Wright and Nielsen, 1988) due to the good balance of its sensitivity and specificity of reaction in addition to its higher ability in detecting low concentrations of IgG<sub>1</sub> specifically in brucella infection (Nicoletti, 1969 and Stemshorn, et al., 1985).

The significantly increased serum ALT, AST, AP, Urea and Creatinine among brucella reactant Friesian cows in El-Minya Province (table, 2) are indicative of hepatic and renal changes (Kaneko, et al., 1997). This agreed with Ahmed and Nada, (1993) findings among camels and could be greatly explained by the hepatic and pathologic changes reported by Montaser, (1991).

The significantly increased serum total protein and globulin levels among brucella reactant cows (table, 3), agreed with Ahmed and Nada, (1993) among camels and Ghazy, et al., (2000) among horses. Such an increase could be attributed to the moderately decreased serum albumin levels together with the progressively increased immunoglobulins (Kaneko, et al., 1997) during the course of cattle brucellosis. The significantly decreased serum albumin levels among brucella reactant cows agreed with Ahmed and Nada, (1993) finding among brucella reactant camels and Ghazy, et

al., (2000) among horses. In fact, deficient synthesis of albumin occurs commonly in association with hepatic disease and hypoalbuminemia develops as a result of excessive protein breakdown, which occur in prolonged fever (Coles, 1986), and this could be greatly explained by the hepatopathological findings during brucellosis recorded by Montaser, (1991). The significantly decreased A / G ratio shown in table (3), was a result of the increased serum globulin and decreased serum albumin levels and agreed with Kaneko, et al., (1997) statement.

Significant increase in serum total lipids, total cholesterol, triacylglycerol, free fatty acids, together with significant increase of serum lipoproteins (HDL, LDL and VLDL) among brucella reactant Friesian cows in El-Minya Province were shown in table (4). These increased serum lipid values, could be attributed to the variety of changes that can occur in the levels of various lipid and lipoprotein metabolism (Kaneko, et al., 1997) and is greatly supported by the hepatic and renal pathological changes during brucellosis reported by Montaser, (1991). As triglycerides are formed in the liver through esterification of free fatty acids, they are mildly increased in acute hepatic injury and alterations in serum lipids and lipid metabolism have been found in several animal species during renal failure and hypercholesterolemia occurs in association with the nephrotic syndrome (Kaneko, et al., 1997). The significantly decreased phospholipids shown in table (4), could

be attributed to the decreased synthesis of phospholipids as the liver is the main source of phospholipid synthesis (Kaneko, et al., 1997) and is greatly supported by the hepatic pathologic findings during brucellosis syndrome reported by Montaser, (1991).

Table (5) presents a significant decrease in serum calcium, inorganic phosphorus, magnesium, sodium and potassium. The positive linear relationship that exists between total serum calcium and serum albumin levels (Coles, 1986), in addition to the fact that half of the total serum calcium exists in an albumin bound form and serum calcium level is decreased in hypoalbuminemia (Berry, et al., 1973), could greatly explain the significantly decreased serum calcium level among the brucella reactant Friesian dairy cows under study. The significantly decreased serum phosphorus, magnesium, sodium and potassium levels among cows under study, is a matter of renal tubular defects as the kidney is responsible for the control of plasma magnesium concentration and excessive renal sodium and potassium loss can occur with altered renal tubular function (Kaneko, et al., 1997). This is greatly supported by the renal pathological findings during brucellosis syndrome reported by Montaser, (1991).

In conclusion the present study showed that, in addition to the hazard of distributing brucella infection the brucella reactant cows suffered hepatic and renal dysfunction that reflects itself on some

serum constitution. This in turn implies itself on the animal health and these brucella reactant cows should be eradicated from the producing cattle herd as soon as possible.

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