SERO - EPIDEMIOLOGY OF SHEEP BRUCELLOSIS IN BAGHDAD.

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ABSTRACT

A total of 531 serum samples were subjected to Rose Bengal test (RBT) , serum agglutination test (SAT) and indirect flurocent antibody test (IFAT) and ELISA.

The incidence of brucellosis was found to be 55.76%, 47.64% ,55.76% , and 58.70% ,on basis ofRBT.SAT.IFATand ELISA respectively . The agreement between the RBT and SAT was 85.47% and between RBT and IFAT was 100% ingiving apositive result. Incase of ELISA the positive results was higher and we recommended using it as the defenitive test for detection of brucellosis .

INTRODUCTION

Brucellosis is one of the zoonotic diseases and itis a problem of both public health and economic importance in several countries of the world , and is caused by bacteria of the genus Brucella which infect all domestic animals as well as human beings. It causes abortion and infertility in animals(10,12) and undulent fever in man (3) .In Iraq it is an enzootic disease(2)and there is afew studies on the incidence of brucellosis in sheep have been carried out (20,17,16).

There are many serological tests used for diagnosis like RBT ,SAT (5,6) but they may give false negative or false positive (9) especially SAT which cannot differentiate classes of immunoglubulines (7). It is not always possible to isolate the causal organasime from infected animals , so the serological tests play a major role in the routine diagnosis of brucellosis (5). Now anew technique was used to prevent many problems occur during many serological tests .This technique is ELISA which have the higher sensitivity to diagnosis of brucellosis (6,2,8) .

The present study was conducted to find the incidence of brucellosis in sheep in three animal floks in Baghdad by using four serological tests , Rose Bengal test (RBT), serum tube agglutination tests (SAT), Indirect flurocent antibody test (IFAT) and Enzyme -Linked immunosorbent assay (ELISA).

MATERIAL AND METHODS

Studies were performed on (531) blood samples collected from sheepin three separated flocks with history of abortion in Baghdad . Serum samples were tested for the presence ofbrucella antibodies by RBT according to (18).All positive serum samples was tested by SAT as described by (5). IFAT alsoCarried out as described by (1), and ELISA by using antigen produced by Diasorin company and IgA indicator that contain Brucalla spp . The method Was done according to Diasorin company instructions.

RESULTS

1- Rose bengal test (RBT):- A total of J^JI) blood samples were collected from three flocks examined by this test to detect the presence of brucella antibodies .The results revealed 296 , (55.76%) positive reactors out of total number of the test samples as shown in table - 1 -.While the number and percentage of positive reactors serum in the test flocks 120 (58.53%) 80 (51.47%) and 96 (56.47%). In first ,second and third flocks respectively .The remaining 235 (44.34%) were negative to RBT.

2- Serum tube agglutination test (SAT):- All positive reactors serum samples in RBT were examined by SAT .

The results as shown in table -2- revealed 75,37,45,63,33 positive reactors in titer 1/10 ,1/20, 1/40 ,1/80 , 1/160 respectively while 43 positive reactors sample in RBT were negative in SAT . The agreement between the RBT and SAT was (85.47%) in giving positive results.

3- Indirect Immuno -Fluroscent antibody test: -The same positive reactors serum samples in RBT were examined by IFAT.

The result as shown in table -2- revealed 28,51,51,50,116 positively reactor in titer 1/16,1/32,1/64,1/128 and 1/256 respectively . The agreement between RBT.and IFAT was(100%) in giving a positive result.

4-All positive and negative samples in <u>RBT</u> were examined by ELISA .

The results show that 29 sample from those which were negative in RBT give positive reaction and 15 sample of those with positive RBT were negative in ELISA .

The reading 0.208 was the limit between positive result in ELISA .
The agreement between IFAT and ELISA was 100% .

DISCUSSION

The incidence of brucellosis in sheep in the examined flocks was 55.76% as screened by the RBT. This result varies significantly from those reported by (20,17,16,3) who found that the incidence of bmcellosis in sheep were 0.1% ,1.1% ,0.93% and 7.91% respectively .

The high incidence of brucellosis (55.76%) in this study was also reported by previous studies in different provinces of Iraq (12).While (14) have noticed an incidence averaged out to 20% in aborted and non- aborted goats in different provines of Iraq .Of 296 sera positive to RBT 253 were positive to the SAT giving an agreement of (85.47%) between the two tests .This result was compatible with result reported previously by several worker (16,3). (18) whom inoculated six sheep with B.meltensis strain H 38 and showed that RBT identified infected animis in earlier stage than the SAT and was often positive when the SAT was negative or inconclusive . There was amarked drop in the agglutination test to nil in 34 weeks after challenge . While the RBT was still showing positive reaction .Concerning IFAT results obtained during this study it was shown that all positive reactors sera in RBT was positive in IFAT , giving an agreement 100% between the two tests and high number of serum samples (116) giving hightiter 1/256 .

The difference in serological response to the three tests may be due to that, the antibody classes mediating these tests may differ in sheep and the length of time that animal remain infected as a reservoir host, may involve different classes of antibody reacting in various tests (22) it must inevitable include some animals with negative serology and other with titers at every up to the maximum possible, this is due to the fact that after infection, the antibody titers in various serological tests will vary with the course of antibody production (5).

During the incubation period , the results of serological tests may be negativealthough infected animals may abort soon after wards .

The result of the SAT is either negative or inconclusive in the incubative stage or in the late chronic stage of brucellosis , while RBTseems to identify infected animals at an earlier stage than the SAT and is often positive when the SAT is negative or inconclusive but IFAT is positive (21,18).

This may be ascribed to the type and concentrarion of immuno – globuline induced in animals due to brucella infection , so IFAT was proven to detect both an initially induced antibodies early infection and alately induced antibody (Late stage of infection) in brucella infection . These results are compared with those reported by (3) , they used IFAT in diagnosis of human brucellosis and they concluded that IFAT are very reliable and sensitive test for diagnosis of brucellosis .

With regard to the incomplete agreement ofELISA and RBT test in this study we observed that this may occure due to the positive reaction with nonspecific antidobies which give false positive reaction (4) .The fales negative results may occure due to the low degree of sensitivity against immunoglobulin because the high sensitivity is against IgM with comparism to IgG(4,1).

In case of SAT and its little agreement with ELISA may be due to the early and late stages of infection that give fales negative in SAT (19,15). In this study we can conclud that RBT is a good serological test in case of survys but ELISA and IFAT were the most acqurate which make them the most dependable tests in diagnosis and special reserchs on brucellosis .SAT must be under more studies to evaluate its capability to used as a diagnosis tool in brucellosis .

Table -2- Show titer of positive serum with RBT-in tube agglutination test and Immuno - Fluoresent antibody technique .

TESTS											
No of samples	Titer in tube agglutination						Titer in Immuno Fluorescent antibody technique				
	10	20	40	80	160	Negative	16	32	64	128	256
120	84	8	12	30	10	12	11	19	14	24	52
80	7	10	23	12	13	15	10	8	19	16	27
96	20	19	10	21	10	16	7	24	18	10	37
296	75	37	45	63	33	43	28	51	51	50	116

Table -I- Show total number of serum samples and number of positive sera and their percentage in RBT .

Flocks	No of animal	No of positive animal	Percentage %	No of negative animal	Percentage%
1	205	120	58.53	85	41.47
2	156	80	51.27	76	48.73
3	170	96	56.47	74	43.53
Total	531	296	55.76	235	44.34

Table - 3 - Relation ships between result of RBT , SAT , IFAT and ELISA of using sheep sera .

No of sample examined	No of sample shawing positive reaction			
	RBT	SAT	IFAT	ELISA
531	296 55.76%	253 47.64%	296 55.76%	310 58.40

دراسة وبائية ومصلية عن البروسيلا في الأغنام في بغداد

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الخلاصة

لقد تم فحص ٥٣١ عينة مصل بواسطة اربعة اختبارات مصلية هي الوزبنجال (Rose Bengal test) تلازب المصل الانبوبي (tube agglutination test) واختبار الاستشعاع المناعي الغير مباشر (Indirect flurocent antibody test) ، والاليزا (ELIZA) .

ظهر ان مدى حدوث مرض البروسيللوسز هو ٥٥,٧٦% ،٤٧,٧٦% ، ٥٥,٧٦% ، ٥٨,٤٠% عند اجراء اختبارات الوزبنجال، تلازن المصل الانبوبي، اختبار الاستشعاع المناعي الغير مباشر والاليزا على التوالي، وكان التوافق بين اختبار الروزبنجال وتلازن المصل الانبوبي هو ٨٥,٤٧% وبين اختبار الروزبنجال واختبار الاستشعاع المناعي هو ١٠٠% من اعطاء نتيجة موجبة، بينما كانت نسبة الاصابة اعلى بحالة الاليزا مما يؤكد دقتها ويؤيد ان نوصي به كاختبار نوعي تاكيدي للاصابة.

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