



Evaluation of Serological Tests Used for Diagnosis of brucellosis in Goats intended for exportation in Sudan

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ABSTRACT

A total of 1500 sera samples were collected from Goats from Suakin Veterinary Quarantine for exportation. All sera samples were screened for antibodies against *Brucella* by Rose Bengal Plate Test (RBPT) as screening test the test was done in quarantine lab in Suakin. All positive sera which were 180/1500 (12%) were further retested by three tests and the passivity was as follows Buffered Acidified Plate Antigen (BAPA) 135/180 (75%), Serum Agglutination Test (SAT) 36/180 (20%) and Indirect Enzyme Linked Immunosorbent Assay (I-ELISA) 46/180 (25.6%). The statistical analysis-using kappa showed good relationship between RBPT, BABA, SAT and I-ELIS

Introduction:

Brucellosis is a zoonotic infection caused by the bacterial of genus *Brucella* which was first recognized by David Bruce (Bruce, 1887). The disease is an old one that has been known by various names, including Mediterranean fever, Malta fever, gastric remittent fever, and undulant fever. Humans are accidental hosts, but brucellosis continues to be a major public health concern worldwide and is the most common zoonotic infection, (WHO, 2020).

Brucellosis is an important zoonotic bacterial disease widespread in the world, (Tian, *et al*, 2020). It has high public health significance and may poses threat to all human as diseases may transmit through consumption of raw and under cooked milk and milk products (Schelling *et al.*, 2003).

The signs of brucellosis in goats are similar to those in cattle. The disease is prevalent in most countries where goats are a significant part of the animal industry, and milk is a common source of human brucellosis in many countries. The causal agent is *Brucella melitensis*. Infection occurs primarily through ingestion of the organisms, (Nicoletti, 2013).

Sudan exports camels, goats, sheep and cattle to many countries especially to Saudi Arabia and other Arabic Gulf countries (Anon, 2011). These exported numbers influence with epidemics that emerge spontaneously. The number of tested animals in different species for Brucellosis depends on this exportation movement.

This aim of this study was evaluation of the occurrence of brucellosis by RBPT in Goat indented for exportation in Port Sudan Quarantine, also conduction of comparative study among different serological tests commonly used for detection of brucellosis in exported animals, Conformation of *Brucella anti bodis* detected in RBPT positive sera by use of (BAPA, SAT and I-ELISA) and Comparative analysis for different test (RBPT, BAPA, SAT and I- ELISA) used for Brucella diagnosis

Materials and Methods:

Study area

The present work was performed in Suakin quarantine meager in Port Sudan locality, Red Sea State, Sudan in the period from 2018 to 2020

Serum samples:

A total of 1500 sera samples were collected aseptically from the jugular vein of goat into sterile vacutainer tubes conveyed immediately to the laboratory and allowed to stand at upright position at room temperature. The separated sera were transferred to sterile eppendorf tubes and stored at -20°C until used.

Serological tests

All sera samples were screened for antibodies against *brucella* by RBPT as screening test, the test was done in quarantine meager lab in Suakin. All positive serum were further retested by BAPA, SAT. And I-ELISA

All antigens used were obtained from the Department of Brucella, Central Veterinary Research Laboratory, Soba (CVRL, Soba). Sudan.

Rose Bengal Plate test (RBPT):

The antigen was used as described by OIE manual (2016).

Serum Agglutination Test (SAT):

It is a quantitative test used to detect of immune globulins classes. The test was carried out according to the method used by OIE manual (2016)

Buffered Acidified Plate Antigen (BAPA)

The test was used as described by OIE manual (2016)

Indirect ELISA (I-ELISA)

Indirect ELISA for small ruminant (Boehringer Ingelheim Svanova Kitts) was used according to the manufacturer.

Statistical Analysis

Data were statistically analyzed by using Statistical Package for Social Science SPSS (2008). The agreement between serological tests was calculated using kappa analysis.

Results

Result of screening test in goat by RBPT showed that 180 samples were positive out of 1500 samples (12%), these positive samples were retested by BAPA, SAT and I-ELISA. BAPA test showed that 135 samples were positive (75%), 45 samples negative (25%), tested by SAT the result gave 36 samples were positive (20%) and 144 samples were negative (80 %), the result of I-ELISA test showed that 46 samples were positive (25.6%) and 136 samples were negative (74.4%).

The statistical analysis using kappa showed good relationship between RBPT, BABA, SAT and I-ELISA. The results showed that all positive samples for RBPT were also found positive for BABA and 45 (25%) negative result, all positive samples for RBPT were positive for SAT 36 (20%) , 144 (80%) were negative for SAT and all positive samples for RBPT gave 46 (25.6%) positive result and 134 (74.4%) negative results for I- ELISA. Data are shown in the table (1) and figure (1) the result of statistical analysis are shown in table (2) and figure (2).

Table (1): Serological results according to different diagnostic techniques of *Brucella* in Goat

Test	Total sample-No	Positive	Negative	Infection Rate%
RBPT	1500	180	1320	12 %
BABA	180	135	45	75 %
SAT	180	36	144	20 %
I-ELISA	180	46	134	25.6 %

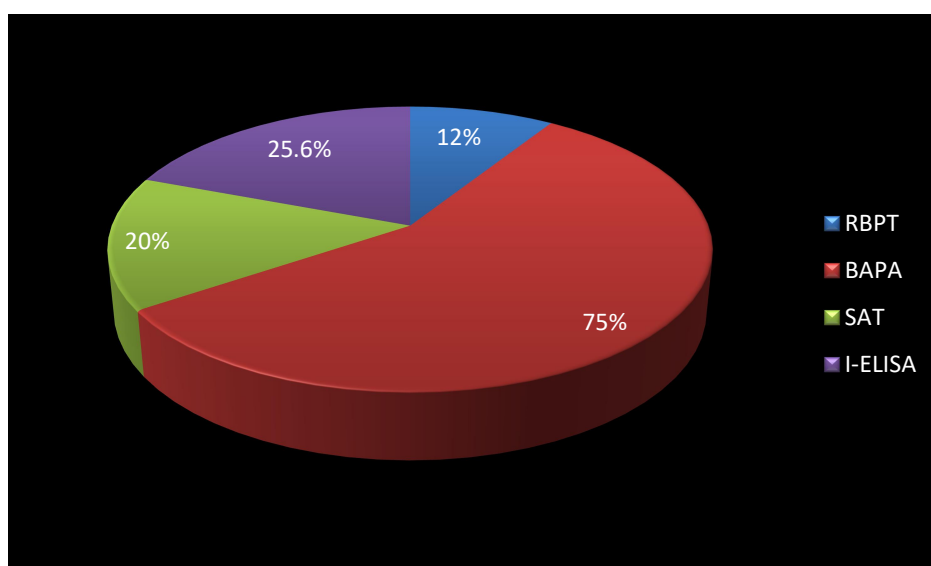


Fig (1): Occurrence of *Brucella* in Goat by using different diagnostic tests

Table (2): Cross tabulation between RBPT.BABA, SAT and I-ELISA tests

	RBPT		BABA		SAT		I_ ELISA	
	+ve	- ve	+ve	- ve	+ve	- ve	+ve	- ve
COUNT	135	45	36	144	46	134		
PERCENTAGE	75%	25 %	20%	80%	25.6%	74.4%		

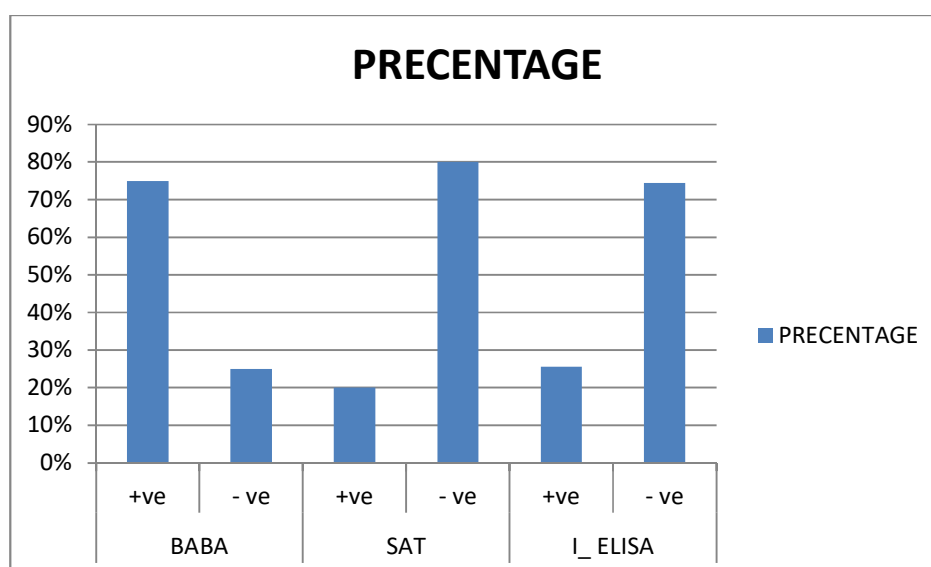


Fig (2): Cross tabulation between RBPT, BABA, SAT and I-ELISA tests

Discussion:

Sudan exports camels, goats, sheep and cattle to many countries especially to Saudi Arabia and other Arabic Gulf countries (Anon, 2011). These exportation processes affected with epidemics that emerge spontaneously. The present study was conducted to determine the occurrence of Brucellosis in goats intended for export by RBPT, Positive samples were retested by many confirmatory tests, BABA, SAT and I-ELISA (OIE, 2016). The importing countries re-examine the animals with RBPT test upon arrival and reject the consignment if the total positive cases exceeded 3%. The RBPT is a sensitive test and is recommended by the OIE (2016) for screening animals for export and control measures.

In Goat the results showed that the Prevalence of brucellosis by RBPT was (12%) (180/1500) this result is lower than that reported by Waffa *et al.*, (2016) and higher than Eman *et al.*, (2018) in Sudan and lower than Kaltungo *et al.*, (2013) in Nigeria who reported 38.9%, 11.4% and 25.8% In goat respectively.

In this study, the seroprevalence estimated by RBPT (180/1500) was confirmed by BABA, which revealed Prevalence of 75% (135/180), this is lower than Ghobashy *etal* (2009) and Montasser (2011) who were reported 100% using BABA test in Goat in Egypt.

In present study, all positive sera samples by RBPT (180/1500) were tested by SAT. The Prevalence was 20 % (36/180), this is higher than many reports from different parts of the country which were between 0.3% and 6.0% Ahmed (2004), Rayas (2004), Elnasry *et al.*, (2001), Samah, (2015) and Solafa *et al.*, (2014). On the other hand, it is higher than a report from the Northern part of Sudan, which was 16.3% (Zein, 2015).

In this study all positive sera samples by RBPT (180/1500) were tested by I-ELISA, the prevalence rate was 25.6 % (46/180) this result is higher than Solafa *et al.*, (2014) used I-ELISA who reported 18% in Sudan, and lower than Ahmed *et al.*, (2011) who were reported 66.7% using I-ELISA in Bangladesh. Statistical analysis showed that BABA test is more sensitive than SAT and I-ELISA in Goat. The differences in the prevalence reported between the countries are likely to be associated with different environmental and management conditions, the type of

study conducted and sample size. It can be concluded that Goat brucellosis according to serological diagnosis is prevailing in Swakin Quarantine, Red Sea State at a low rate, BAPA is considered as quick and effective tests for diagnosis and screening of brucellosis, BAPA test and RBPT techniques were recommended to be used for disease diagnosis in Sudan.

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References:

1. **Ahmed, M.M. (2004)** Studies on Animals' Brucellosis in Red Sea State, Sudan. Master Thesis, Faculty of Veterinary Medicine, University of Khartoum, Khartoum North, the Sudan.
2. **Anon, (2011)**. Reports of the general directory of quarantines and meat hygiene, Ministry of Animal resources, fisheries and land ranges. Sudan.
3. **Bruce, D., (1887)**. Note on the discovery of a micro-organism in Malta fever. Practitioner, 1887, 39:160-170.
3. **El-Ansary, H.E., Mohammed, A.B., Hamad, A.A. and Karom, O.A. (2001)** Brucellosis among animals and human contacts in Eastern Sudan. *Saudi Med. J.*, 7: 577-579.
4. 0916.
5. **Eman, M.A.M, Abdelhamid , A. M.E, Enaam, M. El-Sanousi, Hatim H.I, Saad, E.M.N, Mohamed, A.A. and Yassir A. S. (2018)**. Seroprevalence and risk factors of caprine brucellosis in Khartoum State, Sudan. *Veterinary World*, 11(4): 511-518.
6. **Ghobashy, H.M.M., I.A. Samaha, A.M. Montaser, M.K. El-Kholi and S.M. El-Gibaly, (2009)**. Sero- surveillance on Brucellosis among farm animals in some governorates in Egypt. *Egypt. J. Appl. Sci.*, 24.
7. **Kaltungo, B.Y, Saidu, S.N.A., Sackey, A.K.B. and Kazeem, H.M. (2013)** Serological Evidence of Brucellosis in Goats in Kaduna North Senatorial District of Kaduna State, Nigeria. Hindawi Publishing Corporation ISRN Veterinary Science Volume 2013, Article ID 963673, 6 pages
8. **Montasser ,A.M, Affi, M.M, El-Bayoumy, E.M, Abdul-Raouf, U.M and Mohamad ,H.A (2011)** Efficiency of Serological Tests for Detection of Brucellosis in Ruminant at South Provinces of Egypt, *Global Veterinaria* 6 (2): 156-161, 2011
9. **Muktaderul Ahmed, Ariful, M.D. Islam, M. Minara, K and Byeong- K. B (2011)** Evaluation of four Serological Tests for the detection of Brucellosis in goats and Cattle under the Field Condition of Bangladesh, *Asian Journal of biological Sciences* 4(5):477-482, 2011
10. **Nicoletti, P, (2013)**. Brucellosis in Goats. MSD Veterinary Manual, Merck and Co., Inc., Kenilworth, NJ, USA.
11. **OIE (2016)**. Manual of Diagnostic Tests and Vaccines for Terrestrial Animals, Chapter 2.1.4. Bovine Brucellosis. OIE, Paris, France
12. **Rayas, A.R. (2004)**, Studies on Caprine Brucellosis in Nyala area, South Darfur State, Master Thesis, Faculty of Veterinary Medicine, University of Khartoum, Khartoum North, the Sudan.
13. **Samah, M.A. (2015)**, Seroprevalence and Risk Factors of Caprine Brucellosis in El-Genaina, West Darfur, Sudan. Master Thesis, College of
14. **Schelling, E., Diguimbaye, C., Daoud, S., Nicolet J., Tanner, M. and**
15. **Solafa, Z.E., Angara, T.E., Elfadil, A.A., El-Sanousi, E.M. and Ibrahim, A.M. (2014)** Prevalence and risk factors of ruminants brucellosis in Jabel Aolia locality, Sudan. *Sudan J. Sci. Tech.*, 15: 60-72.

16. **SPSS (2008)** Statistical Package for the Social Sciences. Version 16 and 17 (Win/Mac/Linux), User's Guide SPSS Inc., Chicago.
17. **Tian, M., Song, M., Yin, Y., Lian, Z., Li, Z., Hu, H., Guan, X., Cai, Y., Ding, C., Wang, S., (2020).** Characterization of the main immunogenic proteins in *Brucella* infection for their application in diagnosis of brucellosis. *Comp. Immunol. Microbiol. Infect. Dis.* 2020, 70, 101462.
18. **Waffa A. Ahmed¹, Shaimaa A. Majeed¹, Ameer H. Abdul Ameer¹, Nawal D. Mahmmod¹, Nameer I. Saeed², Luma. Y. Hanaa³ (2016)** Sensitivity and Specificity of Various Serological Tests for Detection of *Brucella spp.* Infection in Male Goats and Sheep *Advances in Microbiology*, 2016, 6, 98-103.
19. **World Health Organization (WHO), (2020).** <https://www.who.int/news-room/fact-sheets/detail/brucellosis>
20. **Zein, A.M. (2015)** Short communication: Prevalence of brucellosis in farm animals in Northern State, Sudan. *Sudan J. Vet. Res.*, 30: 43-44.
21. **Zinsstag, J. (2003).** Brucellosis and Q-fever seroprevalences of nomadic pastoralists and their livestock in Chad. *Prev. Vet. Med.*, 4: 279-293.