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## Research Article

# Prevalence of Brucellosis among High Risk Groups in Northern State, Sudan

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### Abstract

Aim this study was to determine the prevalence of brucellosis in various occupations in Northern State, Sudan, during 2012. A total of 753 serum samples (from 21 veterinarians, 39 meat inspectors, 407 abattoir workers and 286 animal handlers) in different localities were collected and screened for brucella antibodies by using Rose Bengal Plate Test (RBPT), Serum Agglutination Test (SAT) and 2 Mercaptoethanol Test (2MET). Then information's of occupation, age, sex, kind of work were recorded. Seroprevalence of brucellosis in veterinarians, meat inspectors, abattoir workers and animal handlers was 9.5%, 15.3%, 24.4% and 26.5% respectively. The variations in antibodies detected by RBPT, SAT and 2 ME test in the high risk individuals. Study documents the serological evidence of Br. abortus infection in high risk groups in Northern State particularly in Sudan, where RBPT can be used successfully in hospitals as screening test.

**Keywords:** *Brucellosis, High Risk, Northern Sudan*

### Introduction

Brucellosis is highly common bacterial Zoonosis worldwide, represents a threat to several domestic and wildlife animals as well as humans [1]. It is essentially a disease of animals caused by bacteria of the genus *Brucella*, with human accidental host [2]. The disease is usually transmitted to human by consumption of unpasteurized milk and dairy products or by direct contact with infected animals or both. In addition brucellosis is currently thought to be a possible biological weapon as it is highly contagious and air born transmission of the agent [3]. In Sudan, disease in man was reported in 1904 in Berber area in the Northern State of Sudan [4] and followed by another incident in the Blue Nile State [5]. The first isolation of *Br. abortus* from an infected person was reported by [6]. 311 human cases, distributed in nine Provinces in Sudan was reported by [7]. In Sudan particularly in Northern State, brucellosis is prevalent in Dongola, Daba and Marawe. The objective of this study was carried out to investigate the prevalence of this infectious zoonotic disease in high risk groups in Northern State by using Rose Bengal Plate Test (RBPT), Serum Agglutination Test (SAT) and 2 Mercaptoethanol Test (2 MET).

### Method

#### Area of the study

The study was carried in Northern State which is one of the biggest States in the Sudan, about 348,796 Km<sup>2</sup>, bordered by Egypt in the North, Libya and North Darfur in the West, Nahr Elneel in the East, and Khartoum and Northern Kordofan in the South. The population of the Northern State is about 699,065, while the population of animals is about 3,150,000 distributed in seven localities namely: Wadi Halfa, Dalgo, Al burgage, Dongla, Goled, Dabba and Marawe location. The Northern State is mainly an agricultural State (dates, wheat and beans). The veterinary services facilities depend on veterinary hospitals in all parts of the State as well as dispensaries and primary veterinary care units. Health services in the study area are delivered through district health net-work, consisting of a local hospital and a few health centers in the Cities. The study was focused on high risk groups namely: veterinarians, meat inspectors, abattoir workers and animal handlers.

#### Sample collection

A total of 753 serum samples (from 21 veterinarians, 39 meat inspectors, 407 abattoir workers and 286 animal handlers) in different localities were collected. A five ml of venous blood samples were withdrawn by a technician in Department of Microbiology, University of Dongola from each person by using sterile disposable syringes. Each blood sample was left to clot with minimum shaking and transported to the laboratory in an ice box, where it was refrigerated overnight, centrifuged for clarifying. The clear serum obtained from each person was stored at -20°C until used [8] Alton et al. (1975). For ethical considerations, the collection was done from volunteers and with the permission of Ministry of Health, the Northern State and all the participants were informed about the study and agreed to contribute in the study.

#### Rose Bengal Plate Test

The human serum samples were tested by RBPT and then samples were examined with SAT and re-examined with the test

2 ME.

#### Serum Agglutination Test

The SAT test was performed as described [8] by Alton et al. (1988).using commercial Brucella abortus antigen (Himedia. India) and the serum samples diluted to 1:120 - 1: 1280 each and incubated with the antigen for 24h at 37°C. The agglutination reactions were read by indirect light against a white background and the results were scored as follows: O = no agglutination (negative), 1+ = 25% agglutination, 2+ = 50% agglutination, 3+ =75% agglutination and 4+ =100% agglutination.

The titer of the sample was taken as the highest dilution in which that gave 50% agglutination (2+). All samples that had a titer of  $\geq 1: 160$  were considered positive.

#### Mercaptoethanol Test (2ME)

This test was performed as described by [9] Diaz et al. (1976) as follows: Each serum sample was added to four test tube in a rack as follows 0.08, 0.04, 0.02, 0.01ml. 1ml of 0.1M solution of 2ME was added to each tube and mixed by shaking the rack.

One milliliter of Brucella tube test antigen (without phenol) was added to give final dilutions of 1:25, 1:50, 1:100, and 1:200, respectively. After overnight incubation at 37°C, the tubes were read for agglutination similarly as the SAT.

## Results

High risk groups (veterinarians, meat inspectors, abattoir workers and animal handlers)

Table 1 indicated that 753 individuals of the high risk groups examined by the RBPT 14(18.8%) were positive. Using the SAT 183 individuals had  $\geq 1:160$  titers resulting in 13.9% prevalence.

The sero-prevalence of brucellosis in veterinarians, meat inspectors, abattoir workers and animal handlers was 9.5%, 15.3%, 24.4% and 26.5% respectively. This results are indicated the high prevalence (26.5%) was found among animal handlers.

The situation of brucellosis in the Northern State (SUDAN)

The disease in animals was first reported in Wadi Halfa locality in cattle, sheep and goats[11] (Abdalla, 1966). Since that time, the disease was not investigated in Northern State. The incidence of human brucellosis in the Northern State during 2005-2010 increased from 45 cases / 10,000 people in 2005 to 62 cases/ 10,000 people in 2008, with a peak of (82) cases/10,000 inhabitants in 2007.

## Discussion

Generally, in humans, brucellosis is usually caused by Br. melitensis which is associated with occupational exposure or consumption of dairy products [10], followed by Br. abortus and Br. suis. The disease can manifest itself as three different clinical symptoms, which are classified according to the duration of symptoms: acute (initial 2 months), sub-acute (2-12 months), and chronic (>12 months). The disease may be asymptomatic, subclinical and focal or is present with complications relapses and re-infections [11]. In Sudan the main species of concern are Br. melitensis and Br. abortus which are common strains in man and cause severe and prolonged disease with a risk of disability [12]. Laboratories in Hospitals in rural- Sub- Saharan Africa, have limited capacity for diagnosis of brucellosis. Brucellosis is commonly diagnosed after failure to respond to malaria, typhoid or tuberculosis treatment [13]. In current study, it was observed that serological diagnosis was only conducted in districts or designated district hospitals. In the Northern State, local clinics use the TAT but additional tests such as the cELISA and 2 ME are used. These facilities are not easily accessible to the majority of people in rural areas of the State, due to their geographical location and poor infrastructure. From public health view point, brucellosis is considered to be an occupational disease that mainly affects slaughter-house workers, butchers and veterinarians [14]. The present study focused on screening of the disease in the high risk-groups and it was the first study conducted in the area.

#### Veterinarian group

The prevalence of brucellosis in veterinarians (9.5%) is low compared to that reported by [15], who found that prevalence of brucellosis in veterinary workers in Kassala area is 14.7%, a prevalence of brucellosis in India (Delhi) for veterinarians is 57.14% [15](Kumar et al. 1997), which is higher than that reported by [16], who found that prevalence of brucellosis for veterinarians in Saudi Arabia is 5.4% and prevalence of brucellosis for veterinarians in India is 6.1% [11].

#### Abattoir workers group

The prevalence of brucellosis for abattoir workers in Khartoum State is relatively higher (24.3%) than that reported by [17], who reported prevalence of brucellosis for abattoir workers in Omdurman is 10%. The prevalence of brucellosis for abattoir workers in Kassala State is 9% [18]. However, the results obtained were closer to those from values of Arabian and African countries; 23.6% in Egypt; 22% in Saudi Arabia; 23.9% in Jordan and 24.1% in Lebanon [19] and 21.6% in Chad; 21.3% in Djibouti; 20.6% in Nigeria; 23.4% in Ethiopia and 25% in South Africa.

#### Meat product

The results of this study were differed from those reported by [20], who recorded that in Kassala State (Eastern Sudan), the prevalence of brucellosis for butchers, slaughter-house workers, milkers and cow attendants is 1%,.

#### Animal handlers

The prevalence of brucellosis for animal handlers were found to be the most affected among the occupational groups (26.5%). This might be due to the lack of awareness among this group compared with others and also, they were at high risk due to their close contact with their animals, day and night. Brucellosis is traditionally more prevalent in areas with intensive agricultural

activity [21].

The increase in human brucellosis in the study area, during 2005-2011 with a peak of 82 cases/100.000 inhabitants (patients admitted to hospitals) in 2007 and the high prevalence rate 24.3% among the high risk-categories such (veterinarians, meat inspectors, abattoir workers and animal handlers), suggest that, occupational exposure may not be the only hazard to human. Therefore, public health education, in addition to occupational hygiene should emphasize the importance of food hygiene. The prevalence of human brucellosis reported in the Northern State was probably much lower than the actual one as under-diagnosis and under reporting of cases are recognized problems in many developing countries [22]. It has been estimated that for each reported case, there at least 2 additional cases that are not reported or not diagnosed [23].

Therefore, the actual number of cases may be up to 3 times as many as the reported number. This can partly be explained by the absence of proper laboratory facilities, lack of awareness about the disease, poor co-operation and exchange of information between veterinary and health services. The age-group 20-50 years was the commonly affected with brucellosis in this study. Comparable findings have been reported from Egypt (mean age 23 years) Kuwait (mean age 34.4 years), Saudi Arabia (mean age 33.8 years) and Djibouti (mean age 31.6 years) [24]. Brucellosis is predominately an occupational disease [25]. It is expected that this age group would be the affected, having been exposed longer to risk factors related to their occupation. More males were found affected than females (3-750), this is in accord with studies conducted in Egypt, Kuwait, Saudi Arabia and India [26]. This sex distribution in the incidence of brucellosis infection may be because males are more involved in activities such as slaughter and handling of animals and carcasses, and as a consequence they are at greater risk of exposure to infection. The disease is more prevalent in males, which agrees with [22-24].

Other studies however, observed that the incidence of infection was greater in females than males (or roughly equal) where females milking the cows and small ruminants, and thus having a higher chance of contact and acquiring infection [27]. The clinical symptoms of brucellosis are usually severe, but may be confused with various diseases that require a different diagnostic approach. The economical and widely used laboratory tests in diagnosis of the disease are the agglutination tests TAT and RBPT. Among these tests, TAT still remains the reference test for the diagnosis of human brucellosis in many countries of the world [25]. The RBPT was found to be positive in 171 of 183 serum samples, whose SAT test showed titers  $\geq$

1:160. An agreement of 93.4% was found between the RBPT and SAT, indicating that RBPT is an easy to use and cost-effective screening test that can provide reliable results in a relatively short time. Similar results were obtained by [28, 29], who reported 95.7% and 96% agreement respectively between the RBPT and SAT tests. Other investigators reported 100% agreement with [30] (Mert et al. 2003 and [31] Hukie 2009). RBPT yields results within minutes where as a minimum of the 24 hours is needed for the SAT test. Therefore, RBPT can be used successfully in our hospitals as a screening test. Positive results can be confirmed by some other more specific tests, like TAT and ELISA[(Young,1995).

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Table1:Prevalence of brucellosis in occupational examined by the RBPT, SAT and 2 ME

Occupation	Total sample examined	Number positive according to:		
		RBPT	SAT ( $\geq$ 1:160)	2ME
1-Vetrinerans	21	2 (9.5%)	2 (9.5%)	1 (4.7%)
2-Meat-inspectors	39	6 (15.3%)	6 (15.3%)	4 (10.2%)
3-Abattoir workers	407	78 (19%)	99 (24.3%)	66 (16%)
4-Animal handlers	286	75 (26.2%)	76 (26.5%)	50 (17.4%)
Total	753	171 (22.7%)	183 (24.3%)	121 (16%)

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