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STUDY ON THE SEROPREVALENCE OF SMALL RUMINANT BRUCELLOSIS IN THREE DISTRICTS OF SOMALILAND

BY

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ABSTRACT

This study was carried out in three different localities in Somaliland, The overall seroprevalence of brucellosis was (1.27%) in small ruminant (1.13%) and (1.35%) in sheep and goat, respectively by the screening test RBPT, while by using CFT, the overall seroprevalence was 0.8%, in sheep 0.72% and 0.85%. in goat. Moreover,observing the age grouping 2-4 years old was the highest in prevalence of ovine brucellosis (1.79%), while by confirmation using CFT the higher seroprevalence was in age group 2 – 4 year as it reached (1.15%) in sheep while in goat the higher seroprevalence was also in age group 2-4 year (1.21%). And there was a limited variation in different localities for seroprevalence of ovine and caprine brucellosis.

Despite of low prevalence rate of small ruminant brucellosis it is a potential hazard both for animals and human in the study area due to the traditional habits so, sheep and goat producers and consumers should be informed about the characteristics of brucellosis as well as the control measures, in order to prevent the uncontrolled circulation of the disease, and reduce the risk of human brucellosis.

Key words: Brucellosis, Ovine and caprine, Seroprevalence,

INTRODUCTION

Brucellosis is an infectious bacterial disease caused by microorganism of the genus Brucella and affecting numbers of animal species**. Hirsh and Zee (1999)** *Brucella* are facultative intracellular, Gram-negative coccobacilli that lack capsules, flagellae, and endospores. The genus *Brucella* comprises a group of closely related bacteria. The species *B. melitensis* (which infects sheep and goats), *B. suis* (swine), and *B. abortus* (cattle) cause significant economic losses for animal owners and severe human disease. *Brucella* spp. Are also a focus of interest as they are categorized as biological agents due to their high contagiousness and their impact on human and animal health **Alton et al, (1988).**

Generally speaking*, Brucella* spp. can survive for long periods in dust, dung, water, slurry, aborted fetuses, soil, meat, and dairy products. As the infectious dose is very low, infections are an occupational risk for farmers, veterinarians, abattoir workers, laboratory personnel, and others who work in contact with animals and consume their products **Smits and Cutler (2004)**

Brucellosis is considered by the Food and Agriculture Organisation (FAO), the World Health Organisation (WHO), and the Office International des Epizooties (OIE) as one of the most widespread zoonoses in the world **Schelling et al, (2003)**

Brucellosis is prevalent in most countries and is an imprtant zoonosis of serious public health problems **Alton et al, (1988)** and Infection is transmitted to humans through direct contact with the infected animals or by consuming infected milk or fresh cheese **Seleem et al, (2010)**

Economic losses due to brucellosis are due to abortion, retained fetal membranes and to a lesser extent, orchitis and epididymitis and infection of the accessory sex glands in males**Tomaso (2010)** .The classic principles of prevention and control which are based up on protection of healthy flocks, minimizing transmission by sanitary measures and use of test and slaughter method. **Kolar (1984).**

In serological diagnosis of Brucellosis, the Rose Bengale test was developed more than 20 years ago for the diagnosis of bovine brucellosis. **Blasco et al, *(*1994)** added that Rose Bengal test is internationally recommended for the screening of brucellosis in small ruminants (Joint FAO/WHO expert committee on Brucellosis, 1986; **Garin-Bastuji and Blasco, (1997)**. The Complement fixation test is the most widely used test for the serological confirmation of

brucellosis in animals. despite its complexity and the heterogeneity of the techniques used in different countries, there is agreement that this test is effective for the serological diagnosis of brucellosis in sheep and goats **MacMillan, (1990).**

The present work was carried out to study the seroprevalence of small ruminant brucellosis in the study area in Somaliland as a reason of the traditional managemental system and the consumption habits of feeding raw animal products, and to clarify the risk factors of small ruminant brucellosis.

MATERIALS AND METHODS

Area and study population

Three districts of Somaliland (north of Somalia) were included in this study, Hargeysa, Burao, and Borama. The traditional habits of animal products consumption as raw milk, the traditionally made cheese and yoghurt in these areas were predominant, which in turn considered as the highly risky conditions for brucellosis. The flocks of sheep and goat in these districts were varied in their numbers from fifty up to six hundred per flock, but all of them participate the same regimen of grazing and availability of source of water, as they depend mainly on the shallow wells and small ponds of water, and there is no, source of surface water , the migratory nature of these flocks from place to place, rendered them in contact with other flocks and sometimes with wild animals which facilitate diseases transmission, specially when sharing the source of water and the grazing areas.

Study design:

A cross-sectional study was carried out on 9892 sheep and 18657 goat between April 2011 and September 2011, the animals were randomly selected from different age, and localities,

Sample Collection and Handling:

About 10 ml of blood was collected from the jugular vein of each sheep and goats using plain vacutainer tube (Becton Dickson, UK). The blood was allowed to clot for 1-2 hours at room temperature, stored horizontally overnight at 4^{o}c, and then the serum was separated from the clot by centrifugation at 2000 - 3000 rpm for 10-15 minutes. Then the separated serum was labeled and kept under refrigeration (-20^{o}c) until tested.

Serological tests:

Rose Bengal Plate Test.

All sera samples were screened using RBPT antigen (CZV.SPAIN B.No.102437). The test procedure recommended by **Alton et al (1988)** was followed. Briefly, 25 µL of RBPT antigen and 25 µL of the test serum were placed alongside on the plate, and then mixed thoroughly by a tooth pick or glass rod, The plate was shaken for 4 minutes by electric rocker and the degree of agglutination reactions was recorded.The sample was classified positive if any agglutination was observed and negative if no agglutination was noted.

Complement fixation test

All sera which tested positive by the RBPT were retested using CFT for further confirmation. Standard *B. abortus antigen* for CFT was used to detect the presence of anti-brucella antibodies in the sera. The test antigen obtained from veterinary serum and vaccine research institute, and the CFT was done at Brucella unit in Central Laboratory evaluation for Veterinary Biologics, Abbasia, Cairo. Sera with strong reaction, more than 75% fixation of complement (3+) at a dilution of 1:5 and at least 50% fixation of complement 2% at a dilution of 1 : 10 and at dilution of 1 : 20 were classified as positive (**Alton , *et al*., 1975; OIE, 2004**).

RESULTS

Table (1): Seroprevalence of small ruminant brucellosis by species of animals in Three Districts of Somaliland.

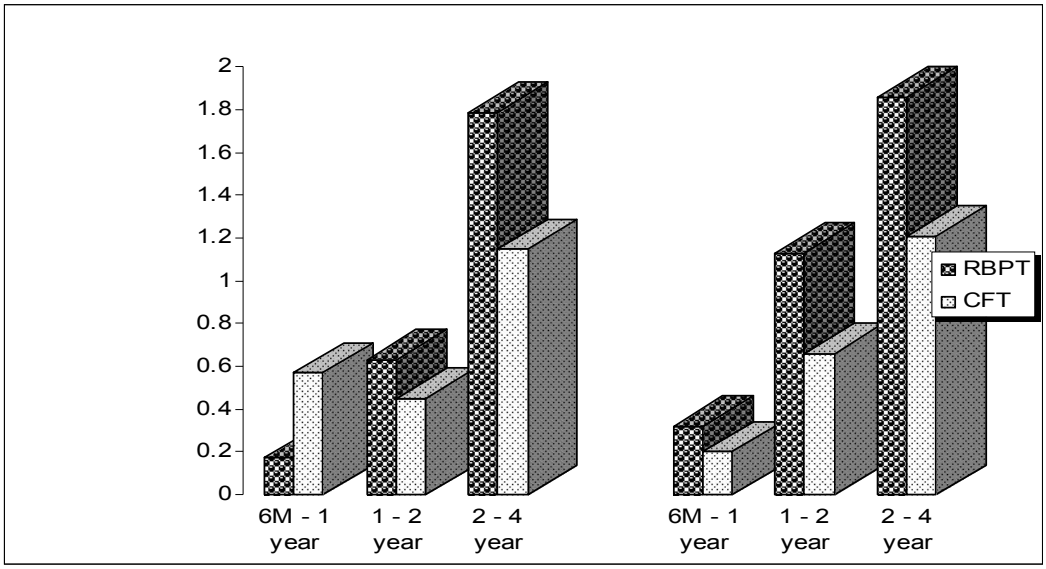
Animal species	Number of sera tested	RBPT positive	CFT positive
Sheep	9892	111 (1.13%)	72 (0.72%)
Goat	18659	253 (1.35%)	159 (0.85%)
Total	28551	364 (1.27%)	231 (0.8%)

The overall seroprevalence of brucellosis was 1.27% in small ruminant (1.13%) for sheep and (1.35%) for goat by the screening test RBPT **Table (1),** in which 111 and 253 Out of 9892 sheep and 18659 goat respectively in three different localities in the north of Somalia, was seropositive for brucellosis, while in confirmation using CFT, the overall seroprevalence was (0.8%) in small ruminant and (0.72%) and (0.85%) in sheep and goat, respectively.

Table (2): Serprevalence of small ruminant brucellosis according to age.

Risk factor	Total examined	Positive RBPT	Positive CFT
Sheep Age			
6 month up to 1year	1740	3 (0.17%)	1 (0.57%)
1 – 2 year	3316	21 (0.63%)	15 (0.45%)
2 – 4 year	4836	87 (1.79%)	56 (1.15%)
Goat Age			
6 month up to 1year	2483	8 (0.32%)	5 (0.20%)
1 – 2 year	7776	88 (1.13%)	52 (0.66%)
2 – 4 year	8400	157 (1.86%)	102 (1.21%)

Fig.(1): Serprevalence of small ruminant brucellosis according to age.

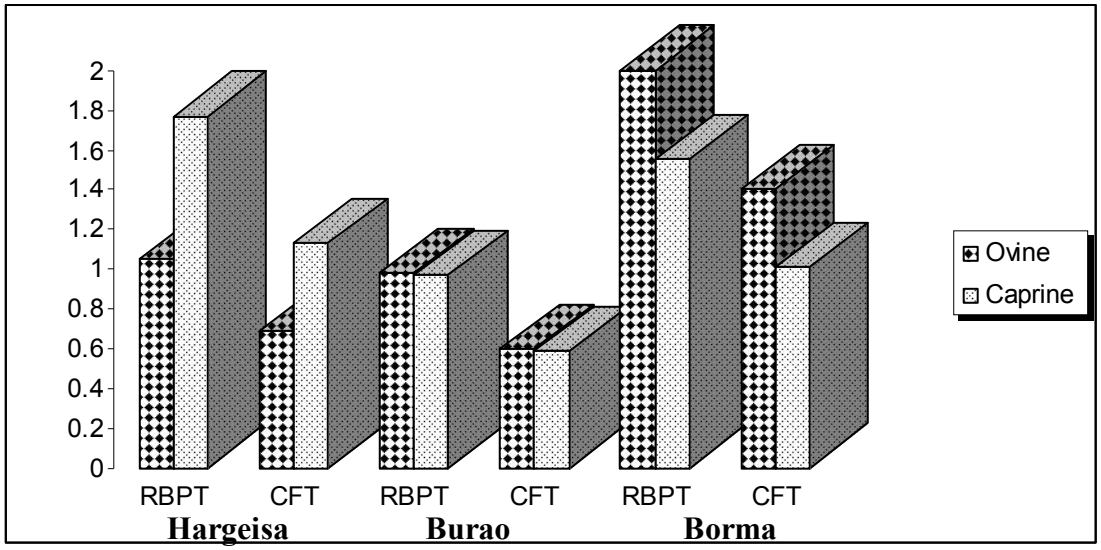


The age grouping 2-4 years old were the highest in prevalence of sheep brucellosis on which out of 4836 examined sheep 87 (1.79%) were positive for brucellosis, followed by group aging 1-2 years where 21 out of 3316 examined sheep were positive for brucellosis in a percent of (0.63%), and last the youngest group aging from 6 months up to 1 year showed that 3 sheep out of 1740 examined animals in percentage of (0.17%) were positive for sheep brucellosis while goat brucellosis, the age grouping 2-4 years old were the highest in prevalence of brucellosis as 157 out of 8400 (1.86%), followed by group aging 1-2 years as 88 out of 7776 (1.13%), and the group aging from 6 months and up to 1 year represented by 8 out of 2483 (0.32%) were positive Table (2) & Fig (1).while by confirmation using CFT the higher seroprevalence found in age group 2 – 4 years as it reached (1.15%) in sheep and in goat the higher seroprevalence was found in age group 2 – 4 years (1.21%).

Table (3): Seroprevalence of small ruminant brucellosis by locality of animals in Three Districts of Somaliland.

Species / Locality	HARGEISA			BURAO			BORAMA		
	No.	**+Ve RBPT**	**+Ve CFT**	**No.**	**+Ve RBTP**	**+Ve CFT**	**No.**	**+Ve RBTP**	**+Ve CFT**
Sheep	4363	46 (1.05)	30 (0.69%)	4479	44 (0.98)	27 (0.60%)	1050	21 (2.00)	15 (1.4%)
Goat	7611	135 (1.77)	86 (1.13%)	9174	89 (0.970)	54 (0.59%)	187٤	29 (1.5°)	19 (1.01%)
Total	**11974**	**181 (1.51%)**	**116 (0.96%)**	**13653**	**133 (0.97%)**	**81 (0.59%)**	**292٤**	**50 (1.70%)**	**34 (1.16%)**

Fig. (2): Seroprevalence of small ruminant brucellosis by locality of animals in Three Districts of Somaliland



According to the locality there was a limited variation in different localities as, Hargeisa the prevalence was (1.05%) 46 out of 4363 examined sheep, in Burao the prevalence was (0.98%) 44 out of 4479 examined sheep, and in Borama, the prevalence was (2.00%) 21 out of 1050 examined sheep while goat brucellosis in Hargeisa, the prevalence was (1.77%) 135 out of 7611 examined goat, in Burao the prevalence was (0.970%) 89 out of 9174 examined goat, and in Borama, the prevalence was (1.55%) 29 out of 1872 examined goat as shown in Table (3).

DISCUSSION

The overall seroprevalence of brucellosis in small ruminant was 1.27% and 0.8% by RBPT and CFT, respectively as shown in Table (1) regarding to sheep(1.13%) and (0.72%) by RBPT and CFT, respectively while in goat (1.35%) and (0.85%) by RBPT and CFT, respectively. in three different localities in the north of Somalia,

The seroprevalence reflected a past or present exposure to Brucella organisms, specially there was no, history of vaccination programme against brucellosis in the area of the study; this result was nearly similar to that of **MacKinnon (1963)** who reported that the seroprevalence was 1.5% in Somalia, that difference might be attributed to o the difference in the density and concentration of animal in this regions, Which facilitate the transmission of the disease.

These results nearly were similar to those obtained by **Blasco et al (1994),** the sensitivities found with the 140 animals from which B. melitensis was isolated were ELISA, 100%; DTH, 97.1%; RBT, 92.1%; and CFT, 88.6%. Those results put into question the value of RBT and CFT as screening and confirmatory tests for sheep brucellosis and at least indicate that their standardization should be modified. For 151 tested sheep from which B. melitensis was not isolated, the percentages of positive animals were ELISA, 100%; DTH, 94.0%; RBT, 57.6%; and CFT, 53.6%. All tests were negative for 100 tested sheep from Brucella-free flocks.

The influence of the age factor on prevalence of brucellosis revealed that was , age grouping 2-4 years old was the highest in prevalence of ovine brucellosis as out of 4836 examined sheep 87 (1.79%) were positive for brucellosis, followed by group aging 1-2 years where 21 out of 3316 examined sheep was positive for brucellosis in a percent of (0.63%), and last the youngest group aging from 6 months and up to 1 year showed low prevalence that 3 sheep out of 1740 examined animals (0.17%) was positive for sheep brucellosis while goat brucellosis, the age grouping 2-4 years old were the highest in prevalence of brucellosis as 157 out of 8400 (1.86%), followed by group aging 1-2 years as 88 out of 7776 (1.13%), and the group aging from 6 months up to 1 year as 8 out of 2483 (0.32%) were positive **Table (2) & Fig (1).** while by confirmation using CFT the higher seroprevalence found in age group 2 – 4year as it reach 1.15% in ovine and in caprine the higher seroprevalence foud in age group 2-4 year (1.21%). The high percent in the old sheep and goat was in agreement with **Mohammed et al (2010)** who reported the percent of (1.54%) in Ethiopia in sexually mature

sheep, and agree with **Hill (1963)** who clarified that in Brucella infection, prevalence increases with age, probably because of greater exposure to infection. Moreover, sexually mature animals are more prone to the infection than sexually immature animals of either sex and with **CDC (2007)** which clarified that sex hormones and meso-erythritol (in male testicles and seminal vesicles) and erythritol in female allantoic fluid stimulate the growth and multiplication of Brucella organisms and tend to increase in concentration with age and sexual maturity.

Seroprevalence of sheep and goat brucellosis had a limited variation in different localities as, Hargeisa the prevalence was (1.05%) 46 out of 4363 examined sheep, in Burao the prevalence was (0.98%) 44 out of 4479 examined sheep, and in Borama, the prevalence was (2.00%) 21 out of 1050 examined sheep while caprine brucellosis Hargelsa the prevalence was (1.77%) 135 out of 7611 examined goat, in Burao the prevalence was (0.970%) 89 out of 9174 examined goat, and in Borama, the prevalence was (1.55%) 29 out of 1872 examined goat **Table (3)& Fig.(3),** the high seroprevalence in Borama may be largely attributed to the migratory nature of these flocks and their contact with other, potentially infected, sheep and goat during their movement and interaction of domestic and wild animals, which facilitate transmission of disease **MacPherson (1995)**

Despite of low prevalence rate of ovine and caprine brucellosis in this study,it is a potential hazard for both animals and human in the study area due to the traditional habits of consumption of raw or decooked animal products. The sheep and goat producers and consumers should be informed about the characteristics of the disease, and control measures, to prevent the uncontrolled circulation of the disease and reduce the risk of human brucellosis.

Insufficient preventive measures, the lack of adequate control as well as uncontrolled animal transportation and migration through “open” borders increased the risk that brucellosis will spread in some regions. New seroprevalence data are needed urgently to evaluate the current situation and for continuous monitoring of necessary control programs in the studied area.

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الملخص العربى

دراسة نسبه الانتشار المصلي لداء البروسيلا في المجترات الصغيرة في ثلاث مناطق من الصومال

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أجريت هذه الدراسة في ثلاثة مواقع مختلفة في أرض الصومال ، وكان الانتشار المصلي الشاملة لل داء البروسيلات (١,٢٧ ٪) في المجترات الصغيرة و (١,١٣ ٪) (و (١,٣٥ ٪) في الأغنام و الماعز على التوالي من قبل الفحص RBPT، في تأكيد استخدام الأموال وتمويل الإرهاب ، كان معدل الانتشار المصلي الكلي ٠,٨ ٪ ، في الأغنام بنسبة ٠,٧٢٪ و ٠,٨٥ ٪ .في الماعز . كانت تجمع سن ٤٠٢ سنة وهي أعلى نسبة في انتشار الحمى المالطية الأغنام (١,٧٩ ٪)، في حين قبل تأكيد استخدام الأموال وتمويل الإرهاب و الانتشار المصلي أعلى وجدت في الفئة العمرية ٤٠٢ سنوات حيث تصل إلى ١,١٥ ٪ في الأغنام بينما الماعز في الانتشار المصلي أعلى كما وجدت في الفئة العمرية ٤٠٢ سنة (١,٢١ ٪ . (وكان هناك تباين محدود في مناطق مختلفة ل معدل الانتشار المصلي من الأغنام و الماعز مرض البروسيلا . على الرغم من انخفاض معدل انتشار الأغنام و الماعز مرض البروسيلا هو الخطر المحتمل على حد سواء للحيوانات و الإنسان في منطقة الدراسة بسبب العادات التقليدية لذلك والأغنام و الماعز المنتجين و المستهلكين يجب أن تكون على علم خصائص الأمراض ومكافحتها التدابير ، لمنع تداول غير المنضبط من المرض، و الحد من مخاطر الحمى المالطية