ESTIMATES OF THE SEROPREVELANCE AND THE ASSOCIATED RISK FACTORS OF BRUCELLOSIS IN SHEEP AND GOATS IN BENADIR REGION OF SOMALIA

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A THESIS SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR MASTER OF SCIENCE IN VETERINARY EPIDEMIOLOGY AND ECONOMICS (MVEE) DEGREE OF THE UNIVERSITY OF NAIROBI.

UNIVERSITY OF NAIROBI

Department of Public Health, Pharmacology and Toxicology

2018
DECLARATION
This proposal is my original work and has not been presented for a degree in any other university.

Signature: ___________________________ Date: ___________________________

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DEDICATION

I dedicate this thesis to Allah my creator and the master, and to both my parents Fadumo Osman Ali and Sheikh Ali Jimale for educating me tirelessly up to the level I reached. Thanks dad and mum.
ACKNOWLEDGEMENTS

First, I would like to express my thanks to my creator (Allah), who has no partnership to his creations and nothing is possible without HIS help. If all the trees on earth were pens and the ocean (were ink), with seven oceans behind it to add to its (supply), yet would not the words of Allah be exhausted (in the writing): for Allah is exalted in Power, full of Wisdom; and prayers and peace be upon Mohamed His servant and messenger. I owe a deep debt of gratitude to the University of Nairobi for giving me an opportunity to undertake this study.

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# ACRONYMS AND ABREVIATIONS

<table>
<thead>
<tr>
<th>Acronym</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>ASAL</td>
<td>Arid and Semi-Arid Lands</td>
</tr>
<tr>
<td>BENELPA</td>
<td>Benadir Livestock Professional association</td>
</tr>
<tr>
<td>CELISA</td>
<td>Competitive Enzyme Linked Immunosorbent Assay</td>
</tr>
<tr>
<td>CFT</td>
<td>Complement Fixation Test</td>
</tr>
<tr>
<td>DNA</td>
<td>Deoxyribose Nucleic Acid</td>
</tr>
<tr>
<td>FAO</td>
<td>Food and Agriculture Organization</td>
</tr>
<tr>
<td>FEWSNET</td>
<td>Famine Early Warning Systems Network</td>
</tr>
<tr>
<td>FRS</td>
<td>Federal of Republic of Somalia</td>
</tr>
<tr>
<td>GDP</td>
<td>Gross Domestic Product</td>
</tr>
<tr>
<td>ILRI</td>
<td>International Livestock Research Institute</td>
</tr>
<tr>
<td>LPS</td>
<td>Lipopolysaccharide</td>
</tr>
<tr>
<td>MoLFR</td>
<td>Ministry of Livestock Forestry and Range</td>
</tr>
<tr>
<td>OD</td>
<td>Optical density</td>
</tr>
<tr>
<td>OIE</td>
<td>World Organization for Animal Health</td>
</tr>
<tr>
<td>OPD</td>
<td>Ophenylendiamine</td>
</tr>
<tr>
<td>PAHO</td>
<td>Pan American Health Organization</td>
</tr>
<tr>
<td>PCR</td>
<td>Polymerase Chain Reaction</td>
</tr>
<tr>
<td>RBPT</td>
<td>Rose Bengal Plate Test</td>
</tr>
<tr>
<td>SANCO</td>
<td>Sante et Consommateurs</td>
</tr>
<tr>
<td>UNDP</td>
<td>United Nations Development Programme</td>
</tr>
<tr>
<td>WHO</td>
<td>World Health Organization</td>
</tr>
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</table>
ABSTRACT

A cross sectional study was conducted from September 2017 to June 2018 to estimate the seroprevalence of brucellosis in sheep and goats and determine the associated risk factors in Benadir region of Somalia. A total of 400 sheep and goats over a half year of age with no past history of vaccination against brucellosis were systematically sampled until the required sample size was achieved. Serum samples were collected and tested using competitive enzyme-linked immunosorbent assay(C-ELISA) for brucella antibodies.

A total of 18 out of 400 serum samples tested positive for brucella antibodies equivalent to an overall seroprevalence of 4.5%. The seroprevalence was higher in goats (4.9%) than in sheep (3.6%). This difference was not statistically significant. Similarly, the seroprevalence was slightly higher in males (5.4%) than in females (4.2%). The difference was not statistically significant. There were no differences in seroprevalence according to the sampling sites (Sub-districts).

The risk factors identified for testing positive to brucella antibodies in both univariate and multivariate analysis were production system and source of replacement sheep and goats. Sheep and goats on extensive production system were 1.4 times (OR: 1.4) more likely to test positive to brucellosis antibodies relative to those on semi intensive production system. Similarly households that sourced replacement stock from outside their herds were 1.2 times (OR: 1.2) more likely to have animals in their herds testing positive. There was no evidence of confounding in this study.
In conclusion, this study has established the endemic status of brucellosis in the Benadir region of Somalia. There is a need to institute control measures of brucellosis in the region through vaccination, conducting regular sero-surveys and those animals that test positive removed from the herd. To avoid human exposure to brucella, sound educational programmes on proper disposal of aborted fetuses and membranes, and avoiding assisting animals giving births would be essential.
CHAPTER ONE

1.1 INTRODUCTION

Livestock production systems possess about 30 per cent of the planet's terrestrial surface area with a significance of at least $1.4 trillion hence are a significant worldwide resource (Steinfeld et al., 2006). Livestock sector is a source of employment to at least 1.3 billion people in the world in its various long market value chains. It also directly supports the source of income of 600 million poor small scale farmers in the developing world (Thornton et al., 2006). Keeping livestock is significant risk decline approach for pro-poor, and poor communities, they are source of essential nutrients and are also used for traction of small scale crop farms. Animal meat, milk and other products provide at least 33% of proteins globally and 17% of kilocalorie consumption, even though there is large difference in the intake of animal products between rich and developing countries, with rich countries consuming more products than developing countries (Rosegrant et al., 2009). Systems of livestock production have both beneficial and negative outcome on the public health, social value, natural resource base and financial development (World Bank, 2009). Currently, keeping domesticated animal sector is one of the sectors early developing enterprise as an the agricultural subsectors in poor countries. It is part of agricultural Gross Domestic Product (GDP), it is rapidly expanding driven by higher demand for animal products as result of through urbanization and population growth with parallel income growth in poor countries (Delgado, 2005).
Goats and sheep are notably adaptable to a wide range of environmental conditions in tropical livestock production systems in Africa where they contribute significantly to the national economy (ILRI, 2006). In Somalia, sheep and goats are the major type of livestock in the country to be estimated at 27.1 million out of the entire livestock populations estimated at 39.5 million (FAO, 2015a). In 2015 Somalia exported approximately 5.3 million animals (around 5 million goats and sheep; 300,000 cattle and 7,000 camels, valued at 384 million Dollars (USD). This was a significant proportion of the foreign earnings (FAO, 2015b). Livestock sector dominates the economy of Somalia and it is estimated to contribute 40% of the Somalia's Gross Domestic Product (FAO, 2015b).The sector continues to grow and the growth is attributed to export-focused interventions, geographical proximity to export markets, good prices and the readily available markets in the Middle East.

Sheep and goats are a source of food and income, and in some households they are kept for prestige. Table 1.1. shows the distribution of livestock in the various regions of Somalia. Goats and sheep predominates in all regions of the country.
Table 1.1: Numbers, types and distribution of livestock in various regions of Somalia

<table>
<thead>
<tr>
<th>Region</th>
<th>Number of Camel</th>
<th>Number of Cattle</th>
<th>Number of Goats</th>
<th>Number of Sheep</th>
<th>Total Livestock Population Estimate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Awdal</td>
<td>396,890</td>
<td>65,696</td>
<td>2,332,466</td>
<td>1,088,945</td>
<td>3,883,997</td>
</tr>
<tr>
<td>Woqooyi Galbeed</td>
<td>564,659</td>
<td>96,567</td>
<td>2,745,465</td>
<td>1,139,224</td>
<td>4,545,915</td>
</tr>
<tr>
<td>Togdheer</td>
<td>496,815</td>
<td>5,018</td>
<td>1,952,918</td>
<td>582,689</td>
<td>3,037,440</td>
</tr>
<tr>
<td>Sool</td>
<td>236,260</td>
<td>0</td>
<td>1,541,657</td>
<td>1,267,790</td>
<td>3,045,707</td>
</tr>
<tr>
<td>Sanaag</td>
<td>233,942</td>
<td>0</td>
<td>1,959,593</td>
<td>1,217,801</td>
<td>3,555,266</td>
</tr>
<tr>
<td>Bari</td>
<td>86,649</td>
<td>0</td>
<td>1,496,383</td>
<td>745,638</td>
<td>2,328,670</td>
</tr>
<tr>
<td>Nugaal</td>
<td>377,872</td>
<td>0</td>
<td>1,959,593</td>
<td>1,217,801</td>
<td>3,555,266</td>
</tr>
<tr>
<td>Mudug</td>
<td>437,672</td>
<td>13,070</td>
<td>2,057,841</td>
<td>881,057</td>
<td>3,389,640</td>
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<tr>
<td>Galgaduud</td>
<td>461,495</td>
<td>33,978</td>
<td>2,031,000</td>
<td>850,953</td>
<td>3,377,426</td>
</tr>
<tr>
<td>Hiraan</td>
<td>638,935</td>
<td>347,044</td>
<td>1,995,619</td>
<td>680,917</td>
<td>3,662,515</td>
</tr>
<tr>
<td>Shabelle (Middle)</td>
<td>156,138</td>
<td>185,540</td>
<td>1,099,778</td>
<td>521,759</td>
<td>1,963,215</td>
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<tr>
<td>Shabelle (Lower)</td>
<td>286,770</td>
<td>535,447</td>
<td>981,022</td>
<td>464,193</td>
<td>2,267,432</td>
</tr>
<tr>
<td>Bay</td>
<td>361,562</td>
<td>800,964</td>
<td>1,171,477</td>
<td>117,007</td>
<td>2,451,010</td>
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<tr>
<td>Bakool</td>
<td>617,905</td>
<td>369,601</td>
<td>1,459,008</td>
<td>408,830</td>
<td>2,855,344</td>
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<tr>
<td>Gedo</td>
<td>770,894</td>
<td>336,629</td>
<td>1,825,849</td>
<td>750,202</td>
<td>3,683,574</td>
</tr>
<tr>
<td>Juba (Middle)</td>
<td>165,335</td>
<td>520,175</td>
<td>478,247</td>
<td>393,329</td>
<td>1,557,086</td>
</tr>
<tr>
<td>Juba (Lower)</td>
<td>322,042</td>
<td>620,654</td>
<td>732,224</td>
<td>492,673</td>
<td>2,167,593</td>
</tr>
<tr>
<td>Benadir region</td>
<td>148000</td>
<td>250000</td>
<td>74526</td>
<td>22264</td>
<td>494,790</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>6,759,835</strong></td>
<td><strong>4,180,383</strong></td>
<td><strong>28,777,905</strong></td>
<td><strong>13,670,174</strong></td>
<td><strong>53,388,297</strong></td>
</tr>
</tbody>
</table>

Small ruminants’ production is hindered by some constraints that include: low genetic development, poor management skills and prevalence of common diseases such as bacterial, viral and parasitic infestation (Kusiluka and Kambarage, 1996). When these diseases occur, they impact negatively on the economy of countries.

Brucellosis is one such bacterial infection caused by a bacterium of the genus Brucella. It is the most common disease that affects both man and animal in the world (Pappas et al., 2006). Bacterial diseases are known to be one of the major causes of substantial economic losses in the livestock sector particularly in pastoralist areas of the Horn of Africa (ILRI, 2006).

Brucellosis is the second most important zoonotic disease after rabies as ranked by the World Animal Health Organization (OIE), Food and Agriculture Organization (FAO) and World Health Organization (WHO), and causes an estimated half a million human cases annually in the world (Schelling et al., 2003; Pappas et al., 2006). The disease affects almost all wild mammal species as well as domestic animals threat of cross-transmission among sheep, goats, camels and other species (Ghanem et al., 2009).

Brucellosis is associated with considerable loss of productivity, high morbidity and tremendous economic losses in livestock production (Pappas et al., 2006; McDermott, et al., 2013). The disease causes reduction or complete loss of milk production, infertility and sterility. It is also a serious threat to human health (Young, 1991; Adams, 2002; Castellano-Estrada et al., 2004; Ehizibolo et al., 2011). Infection due to brucellosis is transmitted direct contact between animals to humans mainly through infected blood, placenta, fetuses and uterine secretions. It is also transmitted indirectly by use (consumption) of contaminated raw animal products, particularly
meat and milk. Globally brucellosis is difficult to diagnose in the laboratory due to lack of awareness by medical professionals who do not give much consideration and because it has clinical flu-like symptoms, it is highly under-reported (Corbel, 2006). It is placed under neglected diseases although it is widely distributed in Africa especially in sub-Saharan African countries (Grace et al., 2012). Brucellosis is reported to have the high incidence rates in areas there is high where intermingling of livestock occurs (McDermott and Arimi, 2002). The extent and effects of brucellosis in Somalia remains unknown but the problem is likely to be great particularly because of the large numbers of livestock and pastoral production systems practiced.

1.2 Objectives

1.2.1 Overall objective

To estimate the seroprevalence of brucellosis in sheep and goats and determine the associated risk factors in Benadir region of Somalia.

1.3 Specific Objectives

I. To estimate the seroprevalence of brucellosis in sheep and goats in Benadir region of Somalia

II. To determine the risk factors of brucellosis infection in sheep and goats in Benadir region of Somalia.
CHAPTER TWO
LITERATURE REVIEW

2.1 Livestock production in Somalia

The main types of livestock production systems in Somalia are pastoral, semi pastoral and agro pastoral. They contribute significantly to household in cash income (Gryseels, 1988; Ellis and Bahiigwa, 2003; Karugia et al., 2006; Little et al., 2008; Nega et al., 2009). Assets of households come from direct income from animal and animal products, while underprivileged households slightly maintain their daily livelihood income from livestock markets (Markakis, 2004; Liu, 2007a; IFAD, 2009; FEWSNET, 2010a; FEWSNET, 2010b; Pica-Ciamarra et al., 2010b).

About 43 million people in the Horn of Africa keep animals as a source of income, food, draught power, hauling services, prestige and manure (Thornton et al., 2002; World Bank, 2010). The demand for livestock and their products in the Eastern Africa (EA) region continues to increase with growth in human population (FAO, 2004). Somalia is considered to be highly dependent on livestock with 21 out of a possible 29 food economy zones under pastoral and agro-pastoral livestock production systems. Indeed, 75% of the human population depends on livestock for subsistence and livelihood.

2.2 Constraints of sheep and goat production in Somalia

Livestock in Somalia is kept under pastoralist (Central and Northern regions) or agro-pastoralist production systems (in the Southern and Western regions). Camels, dairy cattle, sheep and goats are the principle domesticated animals kept in Somalia (Ombui et al., 2014). The arid and semi-arid (ASAL) climate in Somalia can support rangeland pasture grasses, shrubs and acacia trees
used by livestock. Production system of livestock depends significantly on the efficiency of rangelands and availability of water and pasture. However, rangeland productivity is affected by environmental degradation due to soil erosion, deforestation, charcoal burning, and overgrazing. In addition, Somalia frequently suffers a period of prolonged droughts that usually lead to death of livestock due to lack of feed and pasture (Ombui et al., 2014).

Livestock productivity is affected by occurrence of livestock diseases including *Brucella melitensis* in small ruminants. Opportunities for small scale livestock producers in Somalia who do not have financial and technical support is increasingly constrained by the fact that the Middle East and Somali export enterprises demand for quality animals and animal products. These producers are faced with challenges such as traditional nature of management of livestock (small scale in nature) and lack of consistency in trade and markets (ILRI, 2005). Others challenges faced by the livestock producers include lack of knowledge of animal and product quality, use of drugs inadvertently without regard for residue potentials in products and lack or poor control programs for emerging and re-emerging transboundary animal diseases (ILRI, 2005).

The specific economic losses related to diseases in Somalia include loss of milk, infertility and sterility, cost of vaccines and dropped value of animal culled due to disease (Bale, 1991). The primary effects diseases are associated with concomitant reduction in production and reproductive systems, as well as serious risk to human health (Young, 1991; Adams, 2002; Santellano-Estrada et al., 2004; Ehizibolo et al., 2011). One of the reproductive diseases which impacts negatively on the production of livestock is brucellosis which causes reproductive losses in sexually mature domestic animals (Radostits et al., 1994). In males, it is characterized by
epididymitis and orchitis and in females, it is recognized by stillbirths, late-term abortions, infertility, placentitis, weak calves and reduced milk yield (McDermott et al., 2013).

The infected animals excrete the microorganisms through milk and uterine discharges (Radiostits et al., 1994). The disease is passed essentially from animals to human mainly by direct contact with uterine secretions, blood and placenta and infected fetuses. It is also transmitted indirectly through consumption of contaminated animal products predominantly under-cooked meat, unpasteurized milk and soft cheese, as well as inhalation of airborne agents (Yagupsky and Baron, 2005). Globally, brucellosis is recognized as well-known laboratory acquired infection, and therefore an occupational hazard for people working in the livestock sector. Human-to-human transmission has been documented in very rare cases (WHO, 2013).

2.3 Brucellosis

Brucellosis is an infectious, contagious and internationally important zoonotic disease in which Brucella is the causative bacterial agent. It causes economic losses that impact on sheep and goat production and health in developing countries (Dijkhuizen et al., 1995; Bernues et al., 1997). It is a disease associated with substantial illness that can lead to increased rates of extemporaneous abortions in livestock and also in humans.

2.3.1 Epidemiology of brucellosis

2.3.1.1 Aetiology

The Brucella organism is intracellular coccobacillary, Gram-negative, non motile and non spore forming pathogen (Wernery, 2014). Currently, six species of Brucella organism are identified: B. ovis (sheep), B. suis (swine), B. abortus (affecting mainly cattle), B. canis (dog), B. neotomae
(desert rats), *B. melitensis* (sheep and goats). A couple of new species including: *B. microti* (isolated from common voles), *B. ceti* (cetaceans), *B. pinnipedialis* (pinnipeds) and *B. inopinata* (isolated from a human patient) has been described but the classification Subcommittee on Brucella has not then as distinct species approved (Blasco, 2010). Under the light microscope Brucella organisms appear as short rods measuring about 0.5 - 0.7 and 0.6 - 1.5 μm long. They resist discoloration by weak acids thus they stain acid fast when modified Ziehl Nelsen (ZN) technique used for staining (Alton *et al.*, 1988).

### 2.3.1.2 Occurrence and Distribution

Brucellosis occurs globally, but most developed countries have controlled the disease through routine screening of susceptible production animals and carrying out of mass vaccinations (Corbel, 1997; Maloney and Fraser, 2004). Clinical form of the disease is still common in the Asia, Middle East, Africa, the Mediterranean Basin, Central and South America and the Caribbean (Corbel, 1997; Godfroid *et al.*, 2005). The main sources of the disease in human are sheep and goats (Young, 1995). The use of bacterial biotyping is important for tracing sources of infection and the field usually biogroup 1 and 3 are common (Corbel, 1989a). *B. abortus* biogroup 1 has a high prevalence among large farms populated by indigenous or imported animals. However, biogroup 3 is isolated in higher frequency in small herds and in nomadic or semi-nomadic herds where these herds are kept together with indigenous animals (FAO, 2003).

Humans acquire brucellosis is through intake of contaminated milk and milk products and from contact with sick animals or infected carcasses during slaughter. While sheep and goats are the major hosts of *B. melitensis* infection, there is growing evidence of emergence of this serotype in
camels and cattle populations (FAO, 2010). *Brucella ovis* mainly occurs in sheep and is of little significance to humans (WHO, 2006). Transmission route of *B. melitensis* in (sheep and goats is not different to that of cattle however; venereal transmission may play a major role in the occurrence of the disease. According to Cloeckaert *et al.*, (2001, 2003), all the Brucella serotypes mentioned in the sections above are not host specific, and may infect other animal species in one way or another under appropriate conditions.

The epidemiology of brucellosis in animals depends on the host species affected; for instance the disease in cattle, usually *B. abortus* is the causative agent. However, *B. suis* and *B. melitensis* can also affect cattle and the transmission is similar to that of *B. abortus*. Brucella organism are usually transmitted through contact typically from an aborted animal (and its foetus, afterbirth) to susceptible hosts (McDermott *et al.*, 2010). Factors related to the host agent, the environment and management practices determine the extent of exposure spread and maintenance of brucellosis in a geographical area (Godfroid 2002). Furthermore, *B. melitensis* serotype is one of the most important causes of abortions in sheep and goats. *B. melitensis* persists in Middle East and Mediterranean countries. Brucellosis is globally distributed and continues to pose a threat despite efforts to control and eradicate it from food animal populations (Mantur *et al.*, 2007).

**2.3.1.3 Transmission**

Natural transmission of brucellosis is by ingestion of Brucella bacteria that are present in large numbers in fetal membranes, milk, contaminated feedstuffs, aborted fetus and postpartriuient uterine and vaginal discharges of infected animals especially during the time of parturition (Basjuni *et al.*, 1998; Mangen *et al.*, 2002). Small ruminants act as reservoir of *B. melitensis* and are an infection threat to large ruminants including camels through prolonged contact. The
chance of transmission is higher during abortion and parturition when bacterial load is at its peak and contamination occurs (Dafni et al., 1991).

Inhalation, conjunctival route and intra uterine semen deposition during artificial insemination have also been documented as potential entry points of the pathogen. Brucellosis may be mainly acute for almost half of the cases in a herd with an incubation period of 2 to 3 weeks. While the onset in the other half is slow and progressive, with signs and symptoms developing over quite a long period from the time of infection (OIE, 2005).

Brucella organisms enter the host through mucus membranes of the conjunctiva, wound or intact skin (Limbrinke et al., 2008, Kalin 2008). The disease in small ruminants affects sexually mature animals; the predilection sites being the reproductive tracts of males (testes) and females (pregnant uterus). This is thought to be as result of the fact that female sex hormones and erythritol, which take part in the stimulation of the growth and multiplication of Brucella organisms increase with age and sexual maturity of animals (Radostits et al., 2000). Congenital (utero) infections affect limited numbers of lambs, kids and calves born of infected dams. Large quantities of \textit{Brucella melitensis} in lambs and kids are acquired through ingestion of infected colostrum (Grillo et al., 1997). The mixing of colostrum from different dams to feed new born animals can therefore transmit the disease into this age group (FAO 2006).

Spread of brucellosis infection is due to movement of infected animals to where susceptible animals are disease free herds. Contact of infected herds to non-infected susceptible herds occurs at water points where livestock mingle together without restrictions. Transmission of brucellosis to humans occurs, following direct contact with blood, tissues, urine, aborted foetuses or placentas and vaginal discharges of infected animals (FAO, 2003). Food-borne infection occurs following consumption of raw milk and other dairy products from infected animals. Eating raw
meat from infected carcasses rarely causes brucellosis in humans (FAO, 2003). There is also a potential for exposure of the disease to the public by contacting animals during lambing assistance and handling aborted fetus (Pal, 2007; Hadush and Pal, 2013).

2.3.1.4 Mode of infection

The main route of infection of the *Brucella* organism is through the mucous membranes of the conjunctiva, oropharynx, and upper respiratory tract (Moreno *et al.*, 2002). Other likely routes of infection include the mucous membranes of the female and male reproductive tracts. After gaining entrance to the body, the organisms encounters and attacks the cellular defenses of the host, and then move via the lymph channels move to the nearest lymph node.

The outcome of this encounter depends on the animal species infected, immune status of the host, age, pregnancy status and the virulence of invading *Brucella* organism (Sanco, 2001). The organism overcomes the body immune system and thereafter bacteremia is established. The organism can be detected after ten to twenty days and persists from thirty days to more than two months. Bacteremia often leads to the invasion of the uterus if the animal is pregnant. Infection also develops in various lymph nodes and organs, mostly in the udder and sometimes in the spleen.

Immune response to infection with *Brucella* organisms typically results in the induction of both cell-mediated and humoral immune responses, but the magnitude and duration of these responses is affected by various factors including the size of infecting inoculums, the virulence of the infecting strain, sex, immune status of the host and pregnancy (FAO/WHO, 1986).
2.3.1.5 Clinical signs

Brucellosis is a disease that affects many systems of the host animal, and may present with various clinical manifestations (Yetkina et al., 2006). The incubation period has been variously defined, *interalia* as the period between exposure and the onset of abortion in females. The length of the incubation period is affected by the size of the infective dose, sex and age of the host, stage of gestation and immunity of the infected animal. Clinical presentation of the disease also depends on many factors including breed, vaccination status and herd management factors such as flock size density (Garin-Bastuji et al., 1998). Ruminants generally abort only once in the mid-third gestation but re-infection of uterus in subsequent pregnancies occurs and results in shedding of the organisms in uterine fluids and retained fetal membranes (CSFP, 2007; Nela et al., 2010).

All *Brucella* organisms are related and result in lifelong chronic infection, and are usually found within the cells of milk glands and reproductive system leading to abortion and to an extent sterility of infected animals (PAHO & WHO 2001). Shedding of organisms in milk is frequent and cows develops permanent mastitis. Infections of Sheep with *B. melitensis* and *B. ovis* result in orchitis and epididymitis respectively (Seleem *et al.*, 2010). Arthritis is not common for sheep and goats affected by *B. melitensis*. For horses, infection is often symptomless and development of abscesses in bursae may be the only clinical sign. Camels infected by *B. melitensis* spread the pathogen in milk. This leads to common public health problem for countries populated with camel. Infected goats are source of infection to humans and shed the organism in their blood, urine and milk.
People infected with Brucella show symptoms of irregular fever, also referred to as undulant fever, which is usually misdiagnosed as drug-resistant malaria in tropical countries. Other signs and symptoms include fatigue, depression and joint pains (WHO, 2006).

2.4 Risk factors of Brucella infection (brucellosis)

Uncontrolled livestock movement plays a major role in the transmission of livestock diseases including brucellosis. During restocking farmers may purchase new animals from infected farms unknowingly which may introduce infection in their farms. This happens especially on farms which do not have well established biosecurity systems or measures (Baluka et al., 2013).

Drought is another important contributory factor in the transmission of brucellosis. For example, within pastoral communities, when there is drought farmers move their livestock over long distances in search of pasture and water (Baluka et al., 2013). The wildlife-domestic animal interface also plays a key role in disease transmission. During drought or in case there is plenty of pastures in national parks, pastoralists tend to encroach on the national parks which results in intermingling of wild animals and livestock in the grazing areas. This plays a very important role in the transmission of brucellosis from wild animals to livestock (Kabagambe et al., 2000; Baluka et al., 2013).

Mixed farming systems of different livestock is mainly practiced by the pastoral communities as a strategy to cope with shocks. The different livestock share water, pastures and housing which increases direct contact between animals hence facilitating transmission of brucellosis. The lack
of vaccination and immunization in the pastoral community is also major factor that perpetuates brucellosis transmission and outbreaks in the rural areas (Kabagambe et al., 2000).

2.5 Seroprevalence of Brucellosis in East African Countries

Brucellosis infection in sheep and goats has been shown to occur worldwide and prevalence of the disease is well studied in many regions. Brucellosis is endemic in many countries in Africa, for instance the sero-positivity of *Brucella melitensis* infection in goats in Eastern, Northern and Western parts of Nigeria has been estimated at 4.3-12.5% while 9.4-14.5%, of sheep in the same area have also been found to be *Brucella melitensis* seropositive (Cadmus et al., 2006; Tijjani et al., 2009; Kaltungo et al., 2013). In central Ethiopia about 1.5% of goats and 1.3% of sheep have been reported to be Brucella seropositive (Tekleye B. and Kasali O. B. (1990)), while in Tanzania the prevalence of Brucella in goats and sheep has been estimated at 4.3% and 2.2 %, respectively (Chota, A. C. et al., 2016). *Brucella melitensis* infections in goats have also been reported in Somalia, Kenya, Zambia, Malawi and South Africa (Lughano and Kambarage, 1996). There are similar figures for Kenya where a seroprevalence of brucellosis of 12% has been reported in the pastoral areas of Kajaido (Nakeel 2016) and Marsabit (Kahariri, 2018).

In Somalia previous seroprevalence of brucellosis estimates ranged from 2.8% to 5.6% (Flade and Hussein, 1979). In abattoir sero-survey of brucellosis in Kismayo estimates of Brucella seroprevalence ranged from 7.2% in sheep and 5.3% in goats (Andreani et al., 1982).


2.6 Diagnosis

The brucella organism can be recovered from a variety of materials and usually depends on the clinical signs presented in the animal (OIE, 2009). The most reliable and the only unique method for diagnosing animal brucellosis is isolation of Brucella organism (Alton et al., 1988). The placenta is the most infective and contains the greatest concentration of bacteria in animal while in humans the highest bacteria concentration is usually found in the lymph nodes, milk and blood (Poister et al., 2010). Other materials rich in the organisms include: spleen, lungs and stomach contents from aborted foetuses, semen, vaginal swabs, and arthritis or hygroma fluids from adult animals. Presumptive diagnosis of Brucella infection can be made by assessing specific cell-mediated and serological responses to Brucella antigens (Radostits et al., 2000).

Diagnosis, disease prevention and control of brucellosis especially in animals are often done at herd level. Some animals may have a long incubation period where they staying serologically negative for some time after infection. Thus, diagnoses of disease in one or more animals with may be enough evidence for the presence of the disease and meaning that serologically negative animals may be harboring the disease hence present a risk. 

There are two groups of diagnostic tests: those that detect an immune response to its antigens and those that demonstrate the presence of the organisms (Corbel, 2006). Currently, diagnosis of this zoonosis is based on serological and microbiological laboratory tests (Navarro, 2002).

2.6.1 Culture

Presumptive diagnosis is done by using subjective microscopic examination of modified Ziehl Neelsen stained smears of vaginal swabs, placentas and abomasum of aborted fetuses. Using
these stains Brucella stains red against a blue background although *Coxiella burnetii* and *chlamydophils abortus* may confuse the diagnosis (FAO 2003; Godfroid *et al.*, 2004). Lymph nodes, spleen, udder, uterus, epididymis and *testorone* are recommended samples for microscopic examination and culture from dead animals (FAO, 2003; FAO 2010). From animal carcasses, the preferred tissues for culture include mammary glands, supramammary, parotid, medial and internal iliac, retropharyngeal and prescapular lymph nodes and spleen (OIE, 2009; Ahmed *et al.*, 2010). Ideal samples from live animal are milk, blood and vaginal swabs because mammary gland is the target organ in small ruminants (Marin *et al.*, 1996).

2.6.2 Serological tests

2.6.2.1 Rose Bengal plate test (RBT)

Rose Bengal Test (RBT) is one of a group of tests known as the buffered Brucella antigen tests which rely on the principle that the ability of IgM antibodies to bind to antigen is markedly reduced at a low Ph. The RBT is a simple spot agglutination test where drops of stained antigen and serum are mixed on a plate and any resulting agglutination signifies a positive reaction. This serological test is good in disease identification but may be oversensitive for identification of infected and vaccinated animals (Corbel, 2006). The interpretation of the results is done according to the degree of agglutination observed (Nielsen and Dunkan, 1990). The overall sensitivity of the test is 92.9% in endemic areas, The drawbacks of RBT include: low sensitivity particularly in chronic cases, relatively low specificity in endemic areas and prozones make strongly positive sera appear negative (Díaz *et al.*, 2011).
2.6.2.2 Competitive Enzyme Linked Immunosorbent Assay (C-ELISA)

In C-ELISA, Brucella antigen is immobilized on the plate. Subsequently, the test serum and a monoclonal antibody directed against an epitope on the antigen are co-incubated. This anti-brucella monoclonal antibody is conjugated to an enzyme, the presence of which is detected when it binds to the antigen. This will only occur if there is no antibody in the serum sample which is bound preferentially (EC, 2001).

2.6.2.3 Complement Fixation Test (CFT)

Complement fixation test (CFT) is a good but complex diagnostic and screening test that requires good laboratory facilities, equipments and well trained staff. It has a high sensitivity and specificity. If such prerequisites are available and the test is conducted regularly with care, it can produce valid results and it is also paramount to make titration of every test serum in light of the event of the prozone marvel whereby low weakening dilutions of some sera from infected animals don't fix complement. This is because of the nearness of high levels of non-complement fixing immune response (antibody) isotypes competing for fixing to the antigen (FDA, 1987). At higher dilutions these are weakened out and stable. Thus, positive samples are wasted in the event that they are just screened at a single dilution. In different cases, defiling microorganisms or different factors in serum tests settle or crush complement causing a reaction, even in the absence of antigen. Such “anti-complementary” reactions make the test void and a CFT result cannot be obtained (Corbel, 2006).
2.7 Treatment prevention and control

When brucellosis is detected in a flock/herd in a Region or Country, international veterinary restrictions may be imposed on animal movements and trade which results in huge economic losses. This is the reason why brucella control or eradication programs have been implemented worldwide in livestock (Godfroid et al., 2010). All cases of abortion must be thoroughly investigated by veterinarians, the cause identified and appropriate actions taken. Aborted fetuses and associated fetal membranes must be properly disposed and contaminated areas disinfected (WHO, 2006).

An assortment of antimicrobial medications have action in vitro against Brucella species, thus, the result of routine tests don't constantly associate with clinical viability. Therefore, beta-lactam anti-microbials and macrolide anti-infection agents, for example, penicillin, cephalosporin and erythromycin are related with inadmissibly high rates of backslide when used to treat human patients with brucellosis (WHO, 2006). In spite of the fact that more current macrolides, for example, azithromycin and clarithromycin are more dynamic in vitro than erythromycin, they are not better than current regimens for treatment of human brucellosis, and their role in therapy remains to be determined (WHO, 2006). The best treatment option in acute form of human brucellosis is mainly antibiotic therapy. The best outcomes come with the administration of the combinations of doxycycline and rifampicin for minimum of 6 weeks. Treatments should be prolonged or administered as prescribed in persistent forms (EC, 2001).
2.7.1: Prevention

The preventive methods for brucellosis include; careful selection of replacement animals, isolation of purchased replacements for at least 30 days, prevention of contacts between non-infected herds with those herds or flocks with unknown status (Alton, 1990). Programmes for control must locate the infection, contain it and eliminate infected animals by employing a testing scheme, i.e., test and slaughter (Radostits et al., 2000). Vaccination has been applied to control the spread of animal brucellosis but it does not eliminate infection and therefore constitutes a perpetual infection risk to consumers of raw animal products (Muendo et al., 2012). Prevention is more economical than control and eradication programs which are much more costly (Lopez et al., 2002).

Human brucellosis is normally counteracted by controlling infections in animals (Corbel, 2006). Pasteurization of milk products is a vital security measure where this disease is endemic. Unpasteurized dairy items and crude or undercooked animal products (counting bone marrow) ought not to be expended. Great cleanliness and defensive attire/gear are vital in forestalling wound related introduction. Caution ought to be taken to maintain a strategic distance from pollution of the skin, and also inward breath or unintentional ingestion of creatures while helping at a birth, carrying out a necropsy, or butchering animals for consumption. Specific care ought to be taken when dealing with aborted fetuses or its films and liquids. Dangerous agrarian practices, for example, smashing the umbilical string of infant domesticated animals with the teeth or cleaning prematurely delivered young animals should be avoided (OIE, 2009).
2.7.2 Control

Brucellosis is an infectious disease which has been controlled and eradicated in some countries in the world (Godfroid et al., 2004). In Sub-Saharan Africa, Animal Health Services delivered by the public sector have greatly decreased over the last 20 years due to various factors such as decreasing government budgets, particularly for operational costs of disease control. Thus, programs that require coordinated surveillance, information exchange and application of control measures are not implemented in many sub-Saharan countries (McDermott and Arimi 2002; Smits and Cutler, 2004).

Control programs for brucella include slaughter of infected animal through disposal after serological testing and vaccination of young and mature animals (Taleski et al., 2002). The indigenous knowledge and behavior of livestock owners must be considered if sustainable control programs are to be conducted (Pfeiffer et al., 1988, Tiongco et al.; 2012). Poor knowledge of the disease and the high risk practices with the existing absence of prevention and management programmes lead to continuous disease spread in both humans and animals.

Control of animal movements within territory and across international boundaries is essential to prevent spread of brucellosis among farms, regions and countries with different Brucellosis status. Movements must be permitted only between the areas with the same certified brucellosis status. For movement control to be effective, animal identification must be include brucellosis status in areas of origin using tags, brands and tattoos. Individual animal identification helps in quick identification of restricted animals. Imported animals must be certified brucellosis free before they are authorized entry (OIE, 2004; WHO 2006).
The control of brucellosis in animals is aimed at reducing impact of the disease in human health and the economy at large. If infected goats are left untreated, they usually recover within ten days to two weeks and remain carriers of the disease and cause very serious economic losses to farmers (Brisibe et al., 1996). Elimination of the illness from the population isn't the goal of control, and it is certain that some "acceptable level" of infection will stay in the population (WHO, 2006). Control programs have an undefined duration and should be kept up even after the "acceptable level" of infection has been reached, so that the infection does not occur again (WHO, 2006).
CHAPTER THREE
MATERIALS AND METHODS

3.1 Study area

The research was conducted in Benadir region of Somalia. It is to the east of Somalia (Indian Ocean) and extends from the Gulf of Aden in the north to the Juba River in the south (Fig.3.1). The capital city of Somali (Mogadishu) is within this region. Benadir is close to the equator and has a comparatively dry climate. The area covers a total land mass of 1657 km$^2$ and has human population of 2.5 million people. The area receives an annual rainfall of 427 mm with a maximum 430 mm occurring in the wet season in the month of April to July. Benadir region has 16 administrative districts. The livestock population is estimated to be cattle 250000, camels 148,000, goats 74,526, and sheep 22,264 (Source: FGS, 2017).

![Map of Somalia showing the location of Benadir region (Somalia map 2017)](image)

Figure 3.1 Map of Somalia showing the location of Benadir region (Somalia map 2017)
3.2. **Study population**

The study population were sheep and goats in three purposively selected districts of Benadir region. Sheep and goats 6 months or above were selected. The selected animals had no past history of vaccination against brucellosis.

3.3. **Sampling and data collection**

Three districts of Benadir region were purposively selected based on abundance of sheep and goats. Then within each selected district, 3 sub-districts were randomly selected to get a total of 9 sub-districts. For each selected sub district 9 households were randomly selected from a list compiled with the help of Benadir Livestock Professional Association (BENELPA). The households were systematically sampled until the required number of sheep and goats were achieved. Data was collected through administration of a questionnaire (Appendix 1.0), to the household heads via personal interviews. Data collected included herd sizes, production system, water sources and animal movements.

3.4 **Sample size determination**

Sample size was determined according to the formula in Dohoo *et al.*, (2003):

\[ n = \frac{z_{\alpha}^2 pq}{L^2} \]

Where, \( n \) is required sample size,

\( z_{\alpha} \) =the normal deviate that provides 95% confidence intervals (1.96)

\( p \) = *A priori* estimate of the prevalence of the disease

\( q=1-p \)

\( L \) =the allowable error of the estimate
A P value of 16% was adopted since it was the estimate obtained for Marsabit County in Kenya which has a similar production system as Benadir region (Kahariri, 2018). Thus,

\[ n = 1.96^2 \times 0.16 \times (1-0.16)/ (0.05)^2 \]

The total sample size was 206 animals.

3.5 Blood collection

Blood samples from sheep and goats were drawn by venipuncture using vacutainer tubes 8-10 mls. The blood was allowed to settle at room temperature for serum separation then transferred into sterile vials. The serum was kept at -20°C until ready for laboratory analysis at the directly Department of Public Health, Pharmacology and Toxicology, University of Nairobi.

3.5.1 Laboratory analysis

Presence of Brucella antibodies was detected in serum samples using Competitive Enzyme linked Immunosorbent assay (c-ELISA) according to the manufacture’s recommendations (OIE, 2009).

3.5.2. Competitive-enzyme Linked Immunosorbent Assay (COMELISA) Test for brucellosis

C-ELISA kit was standardized for the diagnosis of brucellosis in small ruminants and humans. The ELISA plates were precoated with brucella antigen. Then test sera including positive and a negative control sera were added. The plates were incubated for an hour then washed using a wash solution. Antibodies if present in the sera would react with the antigen on the plate and would not be removed in the subsequent wash. Then conjugate Horse Reddish Peroxidase...
(HRPO) anti-melitensis IgG was added and allow to react for 1 hour. A substrate for the enzyme o-phenylenediamine dihydrochloride was added to the plate after the final wash. The reaction was allowed to proceed until there was colour development. The optical densities were read using a mini-reader at 492 nm. Absence of color development indicates that the sample tested was positive. A positive/negative cut-off was computed as 60% of the mean of optical density thickness of the 4 conjugate control wells. Any test giving OD equivalent to or underneath this value was viewed as being positive. The result was regarded valid if the mean of the 6 negative control wells was more than 0.7 (the ideal mean negative is 1), the mean of 6 positive control wells was under 0.100 figure 3.2 demonstrates a positive result.

![Figure 3.2: Plate Positive result of a COMELISA](image)

### 3.6: Data handling and analysis

Data were entered into MS Excel spreadsheet and exported to SPSS version 11 for statistical analysis. Brucella seroprevalence was concluded by dividing the number of seropositive samples by total number of sera tested. Descriptive statistics (means, proportions etc.) were also derived using SPSS. Tests of association of positivity and various potential risk factors were assessed both in univariate and multivariate analysis. A level of P value of 0.2 was used to select
variables in the univariate analysis. Those variables which turned significant were used in the multivariate logistic regression model, to identify risk factors for occurrence of brucellosis in sheep and goats.
CHAPTER FOUR

RESULTS

4.1 Livestock Populations

A total of 80 herds were visited in the course of the study. Over half (53%) of the pastoralists kept goats while 42% kept sheep and 4% kept cattle (Table 4.1).

Table 4.1: Distribution of species of livestock kept by households in Benadir region of Somalia

<table>
<thead>
<tr>
<th>Species</th>
<th>Numbers kept</th>
<th>Proportion (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goats</td>
<td>1914</td>
<td>53.09</td>
</tr>
<tr>
<td>Sheep</td>
<td>1543</td>
<td>42.80</td>
</tr>
<tr>
<td>Cattle</td>
<td>148</td>
<td>4.10</td>
</tr>
</tbody>
</table>

4.2 Management practices

Most households practiced individual grazing (82.5%) and watered animals by individual herds (85%) (Table 4.2). This was an indication that herds rarely mixed either in grazing fields or at watering points. The most common source of breeding males were their own bulls (48.8%) followed by neighbors bulls (35%, and communal bulls (16.3%). Over a half (60%) of surveyed households obtained their replacement stock from outside their herds and 40% from their own herds. An overwhelming majority (64%) of the households bought drugs and treated their sick animals, an indication that the delivery of veterinary services in Benadir region and indeed in the whole of Somalia is poorly developed. Almost all of the 80 households (94%) engaged in the dangerous practice of assisting their animals while calving thus exposing themselves to diseases.
such as Brucellosis. Half most of the households (50%) were on extensive production system, 31.25% semi intensive and 18.8% intensive.

Table 4.1: Management practices of livestock herds by the households in Benadir region of Somalia

<table>
<thead>
<tr>
<th>Factors</th>
<th>Level</th>
<th>Number practicing</th>
<th>Proportion (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grazing system</td>
<td>Individual grazing</td>
<td>66</td>
<td>82.5</td>
</tr>
<tr>
<td></td>
<td>Communal</td>
<td>14</td>
<td>17.5</td>
</tr>
<tr>
<td>Watering system</td>
<td>Individual</td>
<td>68</td>
<td>85</td>
</tr>
<tr>
<td></td>
<td>Communal</td>
<td>12</td>
<td>15</td>
</tr>
<tr>
<td>Management health/vet services</td>
<td>Consult Vet/CAHWs</td>
<td>29</td>
<td>36.3</td>
</tr>
<tr>
<td></td>
<td>self-treatment</td>
<td>51</td>
<td>63.8</td>
</tr>
<tr>
<td>Introduction of new animals</td>
<td>Yes</td>
<td>48</td>
<td>60</td>
</tr>
<tr>
<td></td>
<td>No</td>
<td>32</td>
<td>40</td>
</tr>
<tr>
<td>Assist delivery</td>
<td>Yes</td>
<td>75</td>
<td>93.8</td>
</tr>
<tr>
<td></td>
<td>No</td>
<td>5</td>
<td>6.3</td>
</tr>
<tr>
<td>Production system</td>
<td>Intensive</td>
<td>15</td>
<td>18.8</td>
</tr>
<tr>
<td></td>
<td>Extensive</td>
<td>40</td>
<td>50.0</td>
</tr>
<tr>
<td></td>
<td>Semi-intensive</td>
<td>25</td>
<td>31.25</td>
</tr>
</tbody>
</table>
4.3 Knowledge about animal brucellosis

A large proportion (96%) of the respondents reportedly knew of Brucellosis in small stock. The most frequently reported signs of brucellosis were abortion (50%) and retained placenta (15%). Aborted fetuses were not disposed of properly as 78% of the respondents said they threw them into the bush and 22% said they fed them to dogs.

4.4 Distribution of antibodies to brucella by species, district, sex and age in Benadir region of Somalia.

A total of 400 sheep (137 and goats 263) serum samples were tested for presence of Brucella antibodies using the C-ELISA test. Overall, 18 serum samples tested positive for a prevalence of 4.5% (Table 4.3). There was no statistical difference between the goat and sheep seroprevalence (4.9% vs 3.6, respectively). There was also variation in seroprevalence by the sub-districts, although the differences were not statistically significant. Similarly, the seroprevalence by sex and age were not statistically different (Table 4.3).
Table 4. 2: Distribution of seroprevalence proportion by selected variables in Benadir region of Somalia, 2017

<table>
<thead>
<tr>
<th>Variable</th>
<th>Levels</th>
<th>No. tested</th>
<th>No. positive</th>
<th>Proportion (%)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Species</td>
<td>Sheep</td>
<td>137</td>
<td>5</td>
<td>3.6</td>
<td>0.55</td>
</tr>
<tr>
<td></td>
<td>Goats</td>
<td>263</td>
<td>13</td>
<td>4.9</td>
<td></td>
</tr>
<tr>
<td>Sub-district</td>
<td>Barwaqo</td>
<td>48</td>
<td>6</td>
<td>12.5</td>
<td>0.06</td>
</tr>
<tr>
<td></td>
<td>Mingiska</td>
<td>43</td>
<td>0</td>
<td>(0)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Abagado</td>
<td>43</td>
<td>2</td>
<td>4.5</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Fooriloow</td>
<td>41</td>
<td>1</td>
<td>2.4</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Tiida</td>
<td>46</td>
<td>1</td>
<td>2.2</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Raderka</td>
<td>46</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Irshaad</td>
<td>45</td>
<td>3</td>
<td>6.7</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Xirkadhere</td>
<td>44</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Talefishinka</td>
<td>44</td>
<td>2</td>
<td>4.5</td>
<td></td>
</tr>
<tr>
<td>Sex</td>
<td>Male</td>
<td>112</td>
<td>6</td>
<td>5.3</td>
<td>0.60</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>288</td>
<td>12</td>
<td>4.1</td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td>Young(1-2yrs)</td>
<td>258</td>
<td>13</td>
<td>5.04</td>
<td>0.48</td>
</tr>
<tr>
<td></td>
<td>Old (&lt;2yrs)</td>
<td>142</td>
<td>5</td>
<td>3.52</td>
<td></td>
</tr>
</tbody>
</table>
4.5 Risk factors of Brucella infection in univariate and multivariate analysis

4.5.1 Univariate analysis

Of the individual four factors of species, age, sex and district none was associated with testing positive for Brucella antibodies in univariate analysis. The univariate analysis of herd level factors are displayed in table 4.4 of the 7 herd-level factors considered, only two were associated with testing positive to Brucella antibodies production system and source of replacement animals. Animals that were on extensive grazing were 1.4 times more likely to test positive relative to those on semi-intensive grazing (OR=1.4, P<0.05). Similarly, households that obtained replacement animals from outside the herd were 1.2 times more likely to have animals testing positive relative to those that maintained closed herds (OR= 1.23, P< 0.05). (Table 4.4)
Table 4.4: Univariate analysis of herd-level factors associated with testing positive to Brucella antibodies in sheep and goats in Benadir region of Somalia 2017.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Level</th>
<th>No. Respondents</th>
<th>No. testing Positive</th>
<th>Proportion (%)</th>
<th>95% CI</th>
<th>p-value</th>
<th>OR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Production System</td>
<td>Intensive</td>
<td>15</td>
<td>8</td>
<td>18.8</td>
<td>0.133-0.59</td>
<td>0.02</td>
<td>1.4</td>
</tr>
<tr>
<td></td>
<td>Extensive</td>
<td>40</td>
<td>6</td>
<td>50</td>
<td>-0.17-0.27</td>
<td>0.6</td>
<td>1.05</td>
</tr>
<tr>
<td></td>
<td>Semi</td>
<td>25</td>
<td>1</td>
<td>31.5</td>
<td>-0.11-0.36</td>
<td>0.3</td>
<td>1.13</td>
</tr>
<tr>
<td>Grazing pattern</td>
<td>Single herd</td>
<td>66</td>
<td>2</td>
<td>82.5</td>
<td>-0.17-0.27</td>
<td>0.6</td>
<td>1.05</td>
</tr>
<tr>
<td></td>
<td>Herds graze</td>
<td>14</td>
<td>13</td>
<td>17.5</td>
<td>-0.11-0.36</td>
<td>0.3</td>
<td>1.13</td>
</tr>
<tr>
<td>Watering</td>
<td>Together</td>
<td>68</td>
<td>14</td>
<td>20.6</td>
<td>0.04-0.37</td>
<td>0.01</td>
<td>1.23</td>
</tr>
<tr>
<td></td>
<td>Separate</td>
<td>12</td>
<td>1</td>
<td>8.3</td>
<td>-0.11-0.36</td>
<td>0.3</td>
<td>1.13</td>
</tr>
<tr>
<td>Replacement animals</td>
<td>Yes</td>
<td>48</td>
<td>13</td>
<td>27</td>
<td>0.22-0.19</td>
<td>0.89</td>
<td>0.98</td>
</tr>
<tr>
<td></td>
<td>No</td>
<td>32</td>
<td>2</td>
<td>6.2</td>
<td>0.04-0.37</td>
<td>0.01</td>
<td>1.23</td>
</tr>
<tr>
<td>Herd size</td>
<td>Small(0-25)</td>
<td>17</td>
<td>3</td>
<td>17.6</td>
<td>0.08-0.25</td>
<td>0.33</td>
<td>1.08</td>
</tr>
<tr>
<td></td>
<td>Large(&gt;25)</td>
<td>63</td>
<td>12</td>
<td>19</td>
<td>0.08-0.25</td>
<td>0.33</td>
<td>1.08</td>
</tr>
<tr>
<td>Breeding</td>
<td>Own buck</td>
<td>39</td>
<td>9</td>
<td>23</td>
<td>0.08-0.25</td>
<td>0.33</td>
<td>1.08</td>
</tr>
<tr>
<td></td>
<td>Communal buck</td>
<td>41</td>
<td>6</td>
<td>14.6</td>
<td>0.08-0.25</td>
<td>0.33</td>
<td>1.08</td>
</tr>
<tr>
<td>Handling of aborted fetus</td>
<td>Throw/bury</td>
<td>62</td>
<td>11</td>
<td>17.7</td>
<td>0.24-0.16</td>
<td>0.67</td>
<td>0.95</td>
</tr>
<tr>
<td></td>
<td>Give dogs</td>
<td>18</td>
<td>4</td>
<td>22.2</td>
<td>0.24-0.16</td>
<td>0.67</td>
<td>0.95</td>
</tr>
</tbody>
</table>
4.5.3 Multivariate analysis

Of the factors that were included in the final logistic regression model only one herd-level factor retained its significance, i.e. production system. The odds ratio in this analysis was not different (1.38) from the OR in the univariate analysis. This was an indication that there was no confounding by any of the variables measured or unmeasured. The factor ‘where you get replacement animals” lost its significance in the multivariate model, although the OR did not vary by a wide margin (1.2 vs 1.18).
CHAPTER FIVE: DISCUSSIONS

5.1 DISCUSSION

In African countries where pastoralism is practised, small ruminants play important roles in provision of milk, meat and increased household income through selling of animals and other products. Indeed they are useful as saving account where they meet immediate household demands such as food, school fees, etc. Thus, when a disease like brucellosis occurs it has very devastating effects on the economy and health of pastoralists, since it is zoonotic. Human exposure to brucellosis was not investigated in this study.

Seroprevalence estimates of brucellosis in sheep (3.6%) and goats (4.9%) were similar to estimates made in earlier studies in Somalia and in the east African region. Ghanem (2008) in seroprevalence study in northern Somalia documented seroprevalance estimates of 4% and 3.1% in sheep and 3.9% to 4.9% in goats. In abattoir survey in southern Somalia (Mogadishu and Kismayo) Andreani et al. (1982) made estimates of 7.2% in sheep and 5.3% in goats. Similar estimates of brucellosis seroprevalence may be attributed to the almost uniform management practices of small ruminants in pastoral areas. These practices would include keeping of large herds, keeping of animals in closed enclosures which encouraged close mixing of animals and uncontrolled breeding.

The prevalence estimates made in the current study were comparable to those made in similar studies in Kenya (6% in both sheep and goats) (Waghela, 1976 ; El-ansary et al., 2001).

In the Sudan seroprevalence estimates of 14.2% in sheep and 16.2% in goats were made while lower estimates in Eritrea of 1.4% in sheep and 3.8% in goats were made (Shimeles, 2008), in central Ethiopia estimates of 3.2% in sheep and 5.8% in goat were made (Ashenafi et al., 2007).
Other than the results from Sudan, the results from the current study and indeed from the region, shows that the seroprevalence of brucellosis was always low and that it was slightly higher in goats than in sheep. The slight differences may be attributed to the different sample sizes and different tests used with varying sensitivities and specificities. In addition, the sample may have lacked power to detect any differences in groups if those differences existed.

In all studies conducted in the region, seroprevalence of brucellosis was higher in goats than in sheep. This despite the fact that these animals are grazed together, watered together and kept together where horizontal transmission is a possibility. However, this observation may be explained by the fact that goats are more susceptible to Brucella infection than are sheep (Rodastits et al., 2000). In addition, goats are known to excrete Brucella organisms for longer periods than sheep, this reduces the probability of Brucella spread possible only if the two are raised separately sheep flocks.

Although seroprevalence of brucellosis was higher in females than in males, the difference was not statistically significant (P<0.06). This slight difference can be explained by the fact that the female goats have multiple mating partners and thus increase the risk of Brucella infection.

The risk factors for testing positive to Brucella organism in small stock in both univariate and multivariate analysis were livestock production system and where herders obtained their replacement animals. The risk of testing positive was higher in herds on extensive production systems. This is understandable because these systems are characterized by extensive movement of animals for long distances in search of pasture and water. This practice encourages mixing of animals from many sources thus increasing the risk of infection by Brucella. However, it was
surprising that the risk of testing positive to Brucella was more in intensively reared animals than in semi intensively reared ones. One would expect the risk to be more in semi-intensively reared animals as opposed to intensively reared ones due to minimal movement of animals in the latter system. This may have occurred due to improper definition of the production systems leading to misclassification of animals in the three production systems.

Household that had a management policy of procuring their replacement stock from outside their herds, were at increased risk of having their animals testing positive for brucellosis. It has been shown previously that this practice increases chances of introducing diseases particularly if the disease history of the source herds is unknown (Rodastitis et al., 2000).

There were minor differences recorded in the seroprevalence of brucellosis between young and adult sheep and goats. This may due to physiological and immune development or unbalanced samples according to the age group. In older animals the disease could become chronic resulting in antibody titre falling to undetectable levels thus giving rise to false negative results in the serological diagnosis of brucellosis (Godfroid et al., 2002; Tessaro et al., 2004). While young animals may be infected, they generally do not show any clinical signs but only weak and transient serological response. Conversely, susceptibility to brucellosis increases with sexual maturity and especially during pregnancy (E C., 2001). Most of the positive young animals in this study were in early puberty; hence the possibility of being infected. Although some of them may have been infected as kids through suckling infected dams, many may have been exposed through mating with infected bucks. Goats are known to reach puberty between 4 to 8 months of
age (Delgadillo et al., 2007) and because mating in free range is not controlled, they become sexually active during early puberty and are therefore exposed to Brucella infection.

A dangerous practice was observed where a huge proportion of respondents reportedly assisted calving animals and scattered aborted fetuses and membranes in the environment. This practice would enhance exposure to Brucella organism to humans. This aspect was not investigated in the current study. However, similar observations have been made in the Masai pastoral community in Kajiado (Nakeel, 2016) and in pastoral communities of Marsabit county in Kenya (Kahariri, 2018). There is a need to educate these communities on the proper handling of aborted fetuses.

In Conclusion, this study has shown that brucellosis is endemic in Benadir region of Somalia, and perhaps in other areas of Somalia where nomadic pastoralism is practised. There is need to institute control measures of brucellosis through vaccination education on control to the public and conducting sero-surveys and those animals testing positive culled.
CHAPTER SIX: CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

The following conclusions can be drawn from the study

i. The study established occurrence status of brucellosis in Benadir region of Somalia. Overall the seroprevalence of brucellosis in sheep and goats was 4.5%. Although seroprevalence in goats (4.9%) was slightly higher than in sheep (3.6%), the difference was not statistically significant. There were no statistical differences by age of animal, sex and species.

ii. The only variables that were statistically associated with testing positive for brucellosis in both univariate and multivariate logistic analysis were production system and source of replacement stock. Animal raised in extensive systems characterized by movement of animals over large areas were 1.4 times more likely to test positive to brucella relative to animals on semi intensive systems. Similarly households that procured their replacement stock from outside their herds were 1.2 times more likely to have animals testing positive to brucella relative to those who obtained them from their own herds.

iii. A dangerous practice for human exposure to brucella organisms was observed where most households reportedly assisted animals while giving birth as well as throwing aborted fetuses and membranes into the environment.
6.2 Recommendations
The following conclusions can be drawn from the study

i. The source of replacement stock should be from herds whose disease history is known.

ii. Mixing animals from different herds in the grazing fields and watering points should be kept at a minimum. This is to avoid cross-infection.

iii. Well designed and sound educational programs should be instituted especially on the practice of assisting female animals giving birth and disposal of aborted fetuses and membranes. This would reduce the risk of human exposure to brucella organisms.

iv. Brucellosis was shown to be endemic in Benadir region of Somalia. Thus, prevention and control measures should be instituted through proper surveillance, education programs to the public on control, vaccinations and conducting regular sero-surveys and those animals testing positive are culled.

v. Further studies are required especially on epidemiology and economic impact of brucellosis in Somalia. This will provide solid data that can be used to convince policy makers on the importance of the disease and thus provision of funds for control.
REFERENCES

Adams, L. G. (2002). The pathology of brucellosis reflects the outcome of the battle between
the host genome and the Brucella genome. Veterinary Microbiology, 90 (1-4): 553-561.

A. (2010). Pathological and molecular studies on mammary glands and supramammary

Brucellosis Laboratory. Institut National de la Recherche Agronomique, Paris, France.

Andreani, E., Prosperi, S., Salim, A. H., and Arush, A. M. (1982). Serological and
bacteriological investigation on brucellosis in domestic ruminants of the Somali
Democratic Republic. Revue d'elevage et de medecine veternaire des pays

on of brucellosis among small ruminants in the pastoral region of Afar, eastern Ethiopia. Revue
scientifique et technique, 26 (3): 731.

In Contribution to a symposium in honour of Prof. Saka Nuru, National Animal
Production Research Institute,(NAPRI), Zaria (pp. 15-26):

and tuberculosis eradication programme in table -1 Economic losses due to brucella
melitensis (per animal / annum in Rupees) in sheep and goats a mountain area of Spain.

*Pre-veterinary Medical journal*, 30: 137-149.


Corbel, Michael J. (2006). Brucellosis in humans and animals. World Health Organization


Dahoo I., Martin W., Stryhn H. (2003). *Veterinary Epidemiology. res.; AVC.*


FAO (2006), Brucellosis in humans and animals, Food and Agriculture Organisation, Rome, Italy.


FAO (2010), Brucella melitensis in Eurasia and the Middle East. FAO. Animal Production and Health Proceedings. No. 10. Rome, Italy.


FEWSNET (2010a) Tanzania Food Security Outlook, April to September 2010. FEWSNET, Uganda and Washington D.C.

FEWSNET (2010b) Uganda Food Security Outlook, April to September 2010. FEWSNET, Uganda and Washington D.C.


Gryseels, G. (1988, August). The role of livestock in the generation of smallholder farm income in two Vertisol areas of the central Ethiopian highlands. In Management of Vertisols in


enzyme-linked immunosorbent assay for the diagnosis of Brucella ovis in sheep’, *The Veterinary Record* 143, 390-394.


SANCO, (2001), Brucellosis in sheep and goats (Brucella melitensis), Health and Consumer Protection Directorate General, European Union.


APPENDICES

Appendix 1: Questionnaire for determining the presence of brucellosis in herd and assessing the risk factors in Benadir region of Somalia

Name of Interviewer: _________________________________________

Name of the Herder/livestock owner: ___________________________

Date of Interview: ___________________________

District: ________________________Sub location: _________________

CIRCLE

Livestock Species Owned


   No of male _____________    Number of female_______________
   No of male _____________    Number of female_______________

   Age: 1). 6 month -1 year  2). ≤2 years  3). 3-5 years

4. Others specify___________

5. Production system

   1). Intensive  2). Semi intensive  3). Extensive

6. Do your animals graze together with other animals?

   1). Yes  2). No

7. Do your animals drink water together with other animals

   1). Yes  2). No
8. Breeding system

9. How do you manage them when feeding, watering and sleeping?
   1). Together    2). Separately    3). If others specify

10. Do you know a disease called brucellosis?
    1). Yes    2). No

11. Have you ever seen the following signs in your herd?

12. Have you noticed infertility, abortions or retained placenta in your herd last two years?
    1). Yes    2). No

    If yes, specify
    1). How many Sheep ______
    2). How many Goats ______

13. At what stage of the pregnancy did the animals in all above aborted?
    1). First trimester    2). Second trimester    3). Third trimester
    No of sheep ______    No of goats ______

14. How do you handle aborted fetus
    1). Burn    2). Throw away    3). Give to dogs    4). Others specify

15. How do you dispose retained placenta
    1). Burn    2). Throw away    3). Give to dogs    4). Others specify

16. In case there is animal abortion or retained placenta what do you do?
    1). Leave them in the herd    2). Separate aborted animal from the herd

17. How do you manage health problems in your flock?
    1). Consult veterinarians / CAHWs    2). Buy drug and treat    3). Others specify

18. Have you introduced new animals in your farm in the last one year?
    1). Yes    2). No

    If yes, specify
    1). How many Sheep ______
2). How many Goats _____________

19. Do you know that brucellosis can affect human and animals?
   1). Yes                      2). No

21. Do you know that brucellosis is curable in human and animals?
   1). Yes                      2). No

22. Do you assist animals while of delivery calving?
   1). Yes                      2). No