STUDIES ON THE ROLE OF TRADE CATTLE IN THE TRANSMISSION OF BRUCELLOSIS IN KARAGWE DISTRICT, TANZANIA

V.P.S. Kiputa[1], S.I. Kimera[2]* and P.N. Wambura[3]

[1]Department of Agriculture and Livestock Development, Karagwe District Council, P.O. Box 20, Karagwe, Kagera; [2]Department of Veterinary Medicine and Public Health, Faculty of Veterinary Medicine, Sokoine University of Agriculture, P.O. Box 3021, Morogoro; [3]Department of Microbiology and Parasitology, Faculty of Veterinary Medicine, Sokoine University of Agriculture. P.O. Box 3019, Morogoro, Tanzania

SUMMARY

The role of trade cattle in the transmission of brucellosis was evaluated by determining the prevalence of *Brucella* antibodies in the traditional herd and in trade cattle. Rose Bengal Plate Test (RBPT), and serum agglutination test (SAT) were used to assay 162 and 56 serum samples collected from traditional herds and trade stock respectively. Results showed that RBPT detected 17.9% [95% (CI 17.4 to 18.4)] and SAT (10.5%) [95% (CI 10.1 to 10.8)] *Brucella abortus* seropositive cattle in the traditional herds. Based on SAT, the seropositivity was significantly higher in adult cattle (14.3%) [95% (CI 13,5 to 15.1)] than in young cattle (5.1%) [95% (CI 4.5 to 5.7)] ($p<0.05$). About 21% (n=56) of the cows in the trade stock were *Brucella* seropositive. This was significantly higher than in the traditional herd ($p<0.05$). Assessment of risk and awareness of livestock keepers and tradesmen on brucellosis was done by interviewing 123 livestock keepers and 37 tradesmen. Awareness of livestock owners on clinical signs, transmission and control methods for brucellosis was, 72.4%, 42.3% and 20.3% respectively. Abortions (48%) and poor disposal of foetal membranes and aborted foetuses (62%) were the risk factors for transmission of brucellosis among and between the traditional herds. Culling criteria based on poor fertility, lack of health certification, destination and fate of animals were the possible risk factors for the transmission of brucellosis by trade cattle. Low awareness on the zoonotic nature of brucellosis (21%) and consumption of raw milk in 13.8% of the families were the risk factors for the transmission of the disease to livestock keepers. It was concluded that the seroprevalence of brucellosis in trade stock in Karagwe district is high and hence the risk of its transmission through trade animals.

---

* To whom correspondence should be addressed. E-mail: vsilyamgoda@yahoo.com

INTRODUCTION

Brucellosis is one of the important bacterial diseases affecting both animals and man worldwide (Henk and Sally, 2004). The disease is also referred to as Bang's disease or contagious abortion in cattle, and ram epididymitis in sheep. The disease affects other domestic animals such as goats, pigs, dogs, horses, camels and water buffaloes. Wild animals such as buffaloes, wildebeests and zebra contract brucellosis as well (Susan and Asa, 1998; Fyumagwa, 2004). *Brucella* has also been isolated from marine mammals such as bottlenose dolphins, common seals, harbour tortoises, mink whales and otters (Betsy *et al*., 2000; Jahans *et al*., 1998).

The disease causes losses resulting from abortion, sterility and decrease in milk production. Other losses are as a result of increased inter-calving periods, loss of calves, deaths occurring due to acute metritis following retention of fetal membranes, and the cost of culling and replacement of valuable stock (Blood and Radostitis, 1989). In humans, the disease causes chronic painful illness known as undulant fever, Gibraltar fever or Mediterranean fever (Susan and Asa, 1998). When people are infected, treatment and hospitalization cost, impaired manpower caused by absence at work and sometimes loss of life, are the major losses attributed to brucellosis (Calmero *et al*., 1989). Brucellosis therefore continues to exert devastating effects on perpetuating poverty among farming and consumer communities because health of both man and animal population is important for economic development, prosperity and stability.

Infertility and low productivity in animals which are sequels of *Brucella* infection are also common criteria for culling in both traditional and dairy herds (Susan and Asa, 1998). Culled animals are sold as trade stock, this fastens the transmission of brucellosis since trade cattle are legally transported to various areas within and outside the country (Minja, 2002).

The disease is reported worldwide except in few countries. In Africa, it has been reported in many countries with a prevalence of 14.1-28.1% in Zambia (Muma *et al*., 2006), 5.8% in Nigeria (Cadmus *et al*., 2006), 8% in Burkina Faso (Coulibaly and Yameogo, 2000), 12% and 14.5-16% in Algeria and Sudan, respectively (Anon, 1998), 2.2% in Botswana (Bankuzi *et al*., 1993), 3.3 % in Central Republic of Africa (Nakoune *et al*., 2004), and 41% in Togo (Domingo, 2000). In the neighbouring countries of Uganda and Rwanda, Faye *et al*. (2005) and Kabagambe *et al*. (1988) reported seroprevalences of 14.8-16.8% and 25.7%, respectively.

Brucellosis was reported for the first time in Tanzania in 1927 in Arusha (Mahlau and Hammond, 1962 sited by Kitally, 1984) and since then, the disease has been reported in different parts of the country. Kitalyi (1984) reported 5.13% *Brucella* seroprevalence in the Central zone of Tanzania and Swai (1997) reported the seroprevalence of 7.6-22.1% in Coastal zone of Tanzania. Three percent seroprevalence was reported by Minja (2002) in Hanang district. Swai *et al*. (2005) reported the seroprevalence of 12.2% in Moshi and the findings by Mapunda (2006) showed that 16.2% of the cattle in Mbarali district were seropositive for brucellosis. The disease thus appears to be endemic in cattle in both dairy and pastoral communities with an average prevalence of 11% (Fyumagwa, 2004).

The Tanzanian government guidelines on control of brucellosis are not binding and have been implemented only in some commercial ranches and in small dairy farms (Kitalyi, 1984; Minga and Balemba, 1990; Mkonyi *et al*., 2004, Swai, 1997). Control of brucellosis in the traditional herd, pastoral and agro pastoral systems has been neglected. Yet, these are the major suppliers of trade animals and animal products (Kitalyi, 1984). This suggests that, in traditional herds, the prevalence of brucellosis may be in the increase or may be checked by factors associated with culling of animals that show clinical signs particularly of low reproductive performances and low productivity (Swai, 1997).

Animals are traded through livestock markets where minimal or non existent health inspection services are carried out (Kitalyi, 1994; Mkonyi *et al*., 2004). Unfortunately, animals infected with *Brucella* normally do not show obvious symptoms such as fever, unthriftness or obvious illness. Thus, animals purchased at livestock markets may appear normal even when they are infected. Such animals are sources of infections especially when purchased as replacement (Nielsen and Gall, 1994).

In Karagwe district, livestock trade has increased from 4,503 in 2005/2006 to 8,158 in 2006/2007. The animals purchased are transported within the country and to the neighbouring countries such as Burundi and Uganda. This poses a secondary pattern of disease transmission to all those destinations. This study therefore, was designed to estimate the level of brucellosis in the traditional herds in Karagwe district and evaluate the role of trade cattle in transmission of the disease.

MATERIALS AND METHODS

Study area

The study was conducted in Karagwe district, Kagera Region, Tanzania. The area lies between 1^0 10’ and 1^0 55’S and 31^0 00’ and 31^0 15’E at an altitude of 1500 m above sea level. The annual rainfall ranges between 800 - 1,200 mm and has an average temperature of 26^{o}C. This study covered eight villages in six wards which are a major source of trade cattle sold to livestock traders in Karagwe district.

Sampling techniques

Purposive sampling of villages and livestock markets was done to fit the objective of this study. Villages were selected based on a known record of number of cattle sold. Then herds were selected randomly from the list of known livestock keepers in the respective villages. Systematic random selection was employed to identify cattle from which blood samples were taken. Trade animals were purposively selected where only adult females (parity ≥ 1) were chosen.

A sample of livestock keepers to be interviewed for awareness and risk factors was obtained from list of known livestock keepers in the villages, where simple random sampling method was applied. For cattle traders, all tradesmen found at the livestock market place during the period of this study (December 2006 - March 2007) were interviewed.

Blood sample and information collection

Blood samples for estimation of *Brucella* seroprevalence were collected from jugular veins using sterile needles and sterile plain vacutainer serum tubes. The labelled tubes were then placed in slanting positions in containers to avoid haemolysis and to assist in serum separation. The samples were stored at room temperature overnight to allow for clotting to take place. The sera obtained were carefully decanted into sterile cryogenic vials and were frozen in a deep freezer (-20°C) before testing was done at Sokoine University of Agriculture Microbiology laboratory.

The information on livestock keepers' awareness and risk factors and tradesmen's awareness, destination and fate of purchased animals were colleted using semi structured pre-tested questionnaires. The stakeholders were interviewed using Kiswahili language.

The record of numbers, categories of animals sold in the livestock markets and their destination were obtained from the District Agriculture and Livestock Development office. Livestock market registers and livestock movement permit books were the source of these information.

Determination of sample size

The sample size for estimation of *Brucella* seroprevalence was calculated according to Dohoo *et al*., (2004), where the required sample size for estimation of disease prevalence is given by:

$$n = \frac{z^2 \times p(1-p)N}{L^2(N-1) + z^2 p(1-p)}$$

Where
- N = size of a population
- n = required sample size
- p= known or estimated prevalence
- z = value of the standardized variate at 95% confidence level
- L = significant level or level of precision.

According to this formula 145 samples were required when N = 10,450, p = 0.11 and L = 0.05.
One member from each of the 123 (27%) house holds in the study area was interviewed for awareness and risk factors. All the 37 regular livestock tradesmen who were

involved in livestock marketing during the study period were also interviewed for awareness, destination and fate of the animals they purchase.

Serological tests

Rose Bengal plate test (RBPT)

Serum samples were tested using Rose Bengal Plate Test (RBPT) according to Brinley-Morgan *et al.* (1978). Briefly, 30 µl of standard *Brucella abortus* RBPT Antigen (Veterinary Laboratories Agency, New Haw Addlessone, Surrey KT15 3NB, UK) was placed on a transparent glass tile and alongside this, the same volume of serum sample was also placed. Using a stick applicator, the antigen and serum were thoroughly mixed and then gently swilled by hand for four minutes. The results were read by observation of evident granulation (agglutination) which was regarded as positive while samples that appeared clear without agglutination were recorded as negative.

Serum agglutination Test (SAT)

A five tube approach was adopted, whereby 0.9 ml of phenol saline was added to all the five tubes. 0.1 ml of a test serum was added to the first tube to make a 1:10 dilution of the test serum. After thorough mixing, 0.5 ml was transferred into the second tube repeating this serial dilution for the rest of the tubes. The 0.5 ml drawn from the last tube was discarded. Then 0.5 ml of standard *Brucella abortus* agglutination antigen suspension (Onderstepoot biological Products, RSA) was added to each tube making 1:20, 1:40, 1:80, 1:160 and 1:320 dilutions. The tubes were incubated at 56^0C for 30 minutes in a water bath then transferred and incubated at 37^oC for twenty hours. The results were observed by assessing the degree of agglutination as compared to the positive and negative controls. The serum with agglutination (clear solution) at 1:80 or above was considered positive.

Data analysis

The information obtained from questionnaires (livestock keepers and livestock traders), livestock market records and serological test results were analyzed for descriptive parameters; percentages, Chi squares as well as p values using the Statistical Package for Social Science (SPSS 11.5). Tables and graphics were generated using Microsoft Excel 2003®.

RESULTS

Brucella Seroprevalence.

One hundred and eighty one blood samples were collected from randomly selected different categories of cattle, out of which 163 were analysed. The results for *Brucella abortus* seroprevalence in different categories of cattle are shown in Figure 1. It can be noted here that cows had the higher *Brucella abortus* seroprevalence than other categories. There was a statistically significant difference ($p<0.05$, X^2 = 3.880, df 1) between the seroprevalence in adults (females and males) 14.3% [95% (CI. 13.5 to 15.1)] and in young animals 5.1% [95% (CI 4.5 to 5.7)]. It can also be noted that, the highest seroprevalence was observed in trade stock (adult females of parity ≥ 1) which was significantly higher than that of traditional herd ($P<0.05$, $X^2 = 4.3143$, df. 1).

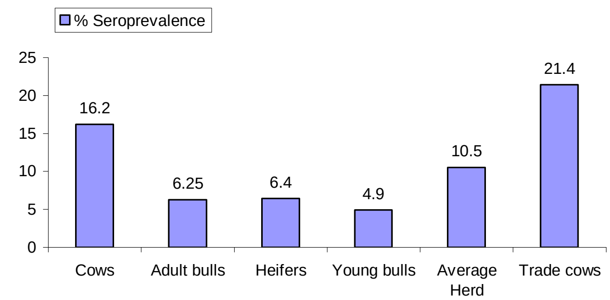

Figure 1: Brucella seroprevalence in different age groups and categories of cattle, Karagwe district

Livestock keepers' awareness

Most of the interviewed livestock keepers (n=123) had heard of brucellosis (84% and about 72% had a knowledge of clinical picture of the disease. However, the means of transmission (42%) and control methods (20%) were not known to livestock keepers. Majority of the livestock keepers were not even aware that brucellosis is a zoonosis (21%) and a similar proportion was also not taking any precaution such as careful handling of aborted foetuses, newly born calves and placentas. Members of some households were consuming either fresh or fermented raw milk. The results of awareness assessment are as summarized in Table 1.

Table 1: Awareness among livestock keepers on brucellosis: clinical signs, transmission, control, zoonosis and precautions at herd level

Awareness of livestock keepers on	Number of respondents (n=123)	Percent of +ve respondents	C I (95%)
Brucellosis	103	83.7	77.1-90.2
Clinical signs	89	72.4	64.5-80.3
Mode of transmission	52	42.3	33.5-51.0
Control measures	25	20.3	13.1-27.4
Zoonosis	26	21.1	13.9-28.3

Key: +ve = Positive
CI = Confidence interval

Risk factors

Abortion and retained placenta were reported to be a problem in some herds, with abortion occurring more frequently than retained placenta. In majority of the herds foetal membranes and aborted foetuses were carelessly disposed off in the surroundings; were either just left to rot or to be eaten by stray dogs. In most herds, culling animals was based mainly on low production, low reproductive performance and chronic diseases. Majority of the livestock keepers (97.6 %) were selling culled animals in livestock markets. The proportions and confidence intervals for major risk factors are summarized in Table 2.

Table 2. Risk factors for transmission of brucellosis at herd level

Risk factors	Respondents n = 123	Percent of +ve respondents	CI (95%)
Abortion problems	59	48	39.1-56.8
Retained placenta	37	30.1	21.9-38.2
Placenta/ fetus disposal*	77	62.6	54.0-71.1
Consumes raw milk	17	13.8	7.7-19.9
Take precautions	26	21.1	13.9-28.3
Criteria for culling+	100	81.3	74.4-88.2

* Improper disposal (Throwing in the surrounding or left to be eaten by dogs)
+ Infertility and low production

A total of 12,661 cattle were recorded to have been sold in livestock markets for the period of 2005-2007. About 47% of trade cattle were adult females (Parity ≥ 1) and 29% were breeding/adult bulls (>3 years), which were either too old to be used for breeding or infertile. The rest were young bulls (1-3 years old) (16%) and heifers of the same age category (6%). The proportions are as shown in Figure 2.

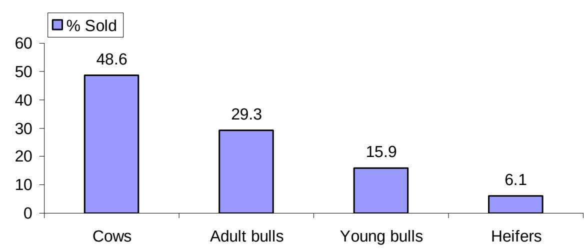

Figure 2. Cattle categories and their proportions in trade stock sold 2005-2007 n= 12,661

When the seroprevalences in different categories of cattle (Figure 1) and proportions of animals in trade stock (Figure 2) are used to calculate the expected and actual seroprevalence in trade stock, the results are as shown in Figure 3. There is no significant difference between the expected seroprevalence in trade stock and that in traditional herd. However, the actual seroprevalence in the trade animals based on the seroprevalence in the purchased adult females was significantly higher than that at the herd level ($p < 0.5$).

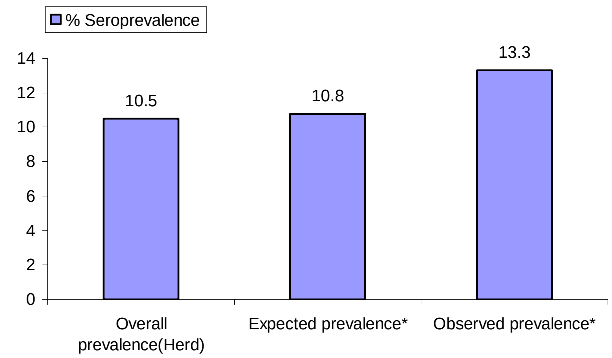

Figure 3. Expected and observed seroprevalences in trade as compared to the prevalence in general herd. Note * Trade cattle

The destinations of purchased cattle during the period (2005 to 2007) are summarized in Table 3. Most cattle (76%) were bought and transported outside the district. About 52% of these were transported within the country and 23% were sold outside the country particularly to Burundi. The remaining cattle (24%) were sold in the district, of which two thirds were for slaughter (16%) and the rest were for breeding purpose (8%)

Table 3: Distribution of cattle purchased by their destination and fate of affected animals 2005- 2007

Destination / fate of animals	**Number of cattle sold**	**Percent of the purchased**
Cattle sold within the district		
For breeding	1,026	8.10
Slaughtered	2,052	16.21
Total sold within the District	3,078	24.31
Cattle sold outside the district		
Within country	6,631	52.37
Outside country	2,953	23.32
Total sold outside the district	9,583	75.69
Total sold during 2005-2007	12,661	100.00

DISCUSSION

The observed seroprevalence of brucellosis is more or less similar to those reported in the northern highlands and Eastern zone of Tanzania (Swai, 1997, Swai *et al*., 2005). Lower seroprevalences (3.2%-5.1%) were reported in the Central zone and in Hanang district, Arusha region (Kitalyi, 1984; Minja, 2002). Higher (16.2%) seroprevalence is reported in some areas in Southern highlands (Mbarali district) as reported by Mapunda (2006). Such a high seroprevalence observed in this study can be associated to production systems, ignorance of livestock keepers that reflect the general management at herd level and inadequate veterinary services.

Pastoral and agro-pastoral systems allow for use of common grazing areas and free movement of cattle from one area to another, which bring into contact the infected and non infected herds. Knowledge on the disease and its mode of transmission is very important in the control of diseases including brucellosis (Kozukeev *et al*., 2006). Despite moderate awareness among livestock keepers and tradesmen on brucellosis, there was low awareness on other important areas such as the mode of transmission and disease control measures. This is reflected by the low proportion of livestock keepers who take control measures to prevent the disease from spreading between animals. Moreover, the area is rather wet for about six months in a year (September – December and February-May). This favours the survival and extended period of infectivity of *Brucella abortus* in the environment. In wet seasons, *Brucella* organisms can survive in the environment for a long time with infectivity persisting for up to 100 days. These become sources of infection to grazing animals (Minja 2002). The aborted foetuses and foetal membranes from infected cows which are carelessly disposed off in the environment have high concentration of *Brucella* organisms. These organisms being in favourable conditions are the sources of new infections in the herds (Susan and Asa, 1998; Olivier, 2002). These are factors that may maintain the disease at rather high prevalence in Karagwe district.

Culling and slaughter of infected animals are among recommended methods of brucellosis control (Mkonyi *et al*., 2004). Most of the livestock keepers interviewed in this study culled their animals on the basis of chronic diseases, decreased production and infertility which might include some of the animals infected with *Brucella abortus*. This may explain the higher *Brucella abortus* seroprevalence in the trade stock than in the general herd. However, the fate of trade cattle transported to other regions of Tanzania and those exported to other countries is not clearly known. Moreover, the long distances and the duration these animals stay close together under stress of transportation without adequate food and water may encourage further spread of the disease to the healthy trade animals (Cadmus *et al*., 2006).

This may suggest higher *Brucella abortus* seroprevalence in trade animals at the destination than that observed before transportation.

Higher *Brucella abortus* seroprevalence in trade cattle coupled with sub-standard slaughter premises and negligence in safety precaution during meat inspection can be a potent source of disease transmission. This implies greater occupational hazard to butchers and meat inspectors. Butchers and meat inspectors are exposed to materials such as blood, vaginal discharges, foetuses, urine and placentas from infected animals. They are thus, at higher risk of acquiring the disease through broken skin and aerosols (Cadmus *et al*., 2006).

The observed proportion of trade animal that are kept for breeding purpose in Karagwe is about 33.3%. This may have positive effects on the transmission of brucellosis especially so when the purchased infected animals are either pregnant, have recently aborted or calved (Corbel, 1997; Susan and Asa, 1998). Such animals usually appear normal and healthy when sold. In the absence of brucellosis testing before purchasing, infected animals are also traded and transported to various destinations within and outside Tanzania.

We conclude here that lack of knowledge among stakeholders and criteria for selection of animals for sale at herd level appear to be the factors attributable to the high prevalence among trade animals. The high seroprevalence in trade cattle, destination and fate of purchased cattle appear to influence the gross effect of the role of trade cattle in transmission of brucellosis. Livestock keepers should therefore be educated on the disease and its economic and public health importance.

ACKNOWLEDGEMENT

The authors wish to thank all who, in one way or another played a role in this investigation. Special appreciation need to be accorded to Mr. Mugusi of the Department of Veterinary Microbiology and Parasitology for his technical assistance in sample analysis.

REFERENCES

Anon. (1998). A perspective of brucellosis surveillance in North Africa and The Middle East. Studies in Algeria, Iran, Kuwait, Morocco, Oman, Sudan, and Syria. FAO consultancy mission report. January 1998.

Bankuzi, F.R., Zymbo, G.C.N. and Morris, S.D. (1993). A serological survey of bovine brucellosis in the Molopo District of Bophuthatswana. *J Afr Vet Assoc* **64,** 154-155.

Betsy, J.B., Darla, R.E., Alastair, P.M., Geoff, F. and Simon, B. (2000). Molecular Characterization of Brucella Strains Isolated from Marine Mammals. *J Clin Microbiol* **38,** 1258-1262.

Blood, O.C. and Radosistis, O.M. (1989). Brucellosis caused by *Brucella abortus* (Bang's disease) In: Veterinary medicine . A text of the diseases of cattle, sheep, pigs, goats and horses. Seventh edition. ELBS/Bailleare and Tindall. London.pp77-780.

Brinley-Morgan, W.J., Macknnon, D.J., Gill, K.P., Gowers, S.G. and Morris, P.I.W. (1998). Brucellosis diagnosis standard laboratory technique. 2nd Edition. New Haw, Weybridge; pp 7-17.

Cadmus, S.I.B., Ijagbone, I.F., Oputa, H.E. and Adesonkan, H.K. (2006). Serological survey of brucellosis in livestock animals and workers in Ibadan, Nigeria. *Afr J Biomed Res* **9,** 163-168.

Calmero, C.J.D., Cabrera, F.P.F., Hernandez, M.S., Reguera, C., Iglesias, J.M., Pinedo, S.A. and Caslilo, C.A.M. (1989). Social economic effect of human brucellosis. *Rev Clin Español* **185,** 459-69.

Colbel, M.J. (1997). Brucellosis: an overview. *Emerg Infec Dis* **3,** 213-221.

Coulibaly, N.D. and Yameogo, K.R. (2000). Prevalence and control of zoonotic diseases: Collaboration between public health and veterinary. Dohoos in Bukina Faso. *Acta Tropica* **76,** 53-57.

Dohoo, I. Martin, W. and Henrik, S. (2004). Veterinary Epidemiology Research. AV C Inc. Charllotetown . Prince Edward Island.Canada. pp 39-46.

Domingo, A.M. (2000). Current status of some zoonosis in Togo. *Acta Tropica* **76,** 65-69.

Faye, B., Castel, V. and Lesnoff, M. (2005). Tuberculosis and brucellosis prevalence survey on dairy cattle in Mbarara milk basin, Uganda. *Prevent Med* **67,** 267-281.

Fyumagwa, R.D. (2004). Disease status in livestock–wildlife interface in Tanzania. Proceeding of the WDA-Africa and Middle East Section annual conference in Abu Dhabi, UAE, 11th-13th December 2004.

Henk, L.S. and Sally, J.C. (2004). Contribution of biotechnology to control and prevention of brucellosis in Africa. *Afr J Biotech* **3,** 631-637.

Jahans, K.L., Foster, G. and Broughton, E.S. (1998). Characterization of *Brucella* strain isolated from marine mammals. International Journal of veterinary microbiology.Volume 57 Article # 4: 371-381.

Kabagambe, J., Nshimiyimana, A., Muberuka, J. and Nyiligira, J. (1988). Les maladies enzootiques des bovines au Bugesera. *Bulletin Agricole du Rwanda* **21**, 159-168.

Kitalyi, J. (1984). Bovine brucellosis in government parasitatal and Ujamaa village dairy farms in Central zone of Tanzania. Assessment of control in some of the farms. Proceedings of 2nd Tanzania Veterinary Association Scientific conference. 4-6 December 1984. Arusha Tanzania pp. 172-183.

Kozukeev, T.B., Ajeilat, S., Maes, E. and Favolov, M. (2006). Risk factors for brucellosis in Leylek and Kadamjay District, Batken Oblast, Kyrgyzstan. In: Morbidity and Mortality Weekly **55** (Sup 01), 31-34.

Mahlau, E.A. and Hammond, J.A. (1962). Brucellosis survey of the Western areas of Tanganyika. *Bull Epiz Dis Afr* **10,** 511.

Mapunda, C.J. (2006). Evaluation of the microagglutination test for screening of brucellosis in cattle in Mbarali District, Tanzania. MPVM degree paper, Sokoine University of Agriculture, Morogoro, Tanzania.

Minga, U.M. and Balemba, O.B. (1990). ELISA, a new serodiagnostic test for bovine brucellosis in Tanzania. Preceedings of the 8th Tanzania veterinary Association scientific conference, pp 105-111.

Minja, K.S.G. (2002). Prevalence of brucellosis in indigenous cattle: Implication for human occupation groups in Hanang and Babati District, Tanzania. MVM degree dissertation, Sokoine University of Agriculture, Morogoro, Tanzania.

Mkonyi, P.A., Kimambo, E., Kime, A., Muhairwa, A. and Minde, G. (2004). Brucellosis surveillance in cattle at Kikulula heifer breeding farm, Karagwe district, Tanzania Preceedings of the 22nd Tanzania Veterinary Association scientific conference.

Muma, J.B., Samui, K.l., Siamudaala, V.M., Olaya, G., Munyembe, M.K., Mubita, C. and Skejerve, E. (2006). Prevalence of antibodies to *Brucella* spp and individual risk factors of infection in traditional cattle, goats and sheep reared in Livestock-wildlife interface areas of Zambia. *J Trop Anim Health* **38,** 195-206.

Nakoune, E., Debaere, O., Koumanda-Kotogne, F., Selekon, B., Samory, F. and Talarmin, A. (2004). Serological surveillance of brucellosis and Q fever in cattle in the Central African Republic. *Acta Tropica* **92,** 147-151.

Nielsen, K. and Gall, D. (1994). Review: Advances in diagnosis of bovine brucellosis use of Enzyme Immunoassays. *Gen Eng Biotech* **14,** 25-39.

Olivier, A.J. (2002). Brucellosis and Tuberculosis; Animal health for developing farmers. ARC – Onderstepoort Veterinary Institute.

Susan, E.A. and Asa, M. (1998). Brucellosis in large animals In: Merk veterinary manual. 8th edition. Merk and Co., Inc White house station, N.J., USA. Pp 998-1002.

Swai, E.M. (1997). Studies on the prevalence of bovine brucellosis and reproductive performance in small scale dairy cattle herds in Dar es Salaam and Morogoro Region. MVM degree dissertation, Sokoine University of Agriculture, Morogoro, Tanzania.

Swai, E.S., Mshanga, D., Sanka, N.P. and Marandu, N.H. (2005). Prevalence of bovine brucellosis in smallholder dairying area, Moshi, Tanzania. *Bull Anim Prod Afr* **53,** 97-105.