Acta Tropica 187 (2018) 99–107

Contents lists available at ScienceDirect

Acta Tropica

journal homepage: www.elsevier.com/locate/actatropica

Herd-level risk factors associated with *Brucella* sero-positivity in cattle, and perception and behaviours on the disease control among agro-pastoralists in Tanzania

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ARTICLE INFO

Keywords:
Brucellosis
Community-based control
Item count technique
Tanzania
Willingness to pay for vaccination

ABSTRACT

Brucellosis is endemic in Tanzania, particularly in agro-pastoral areas. This study investigated the herd-level sero-prevalence and risk factors for *Brucella* sero-positivity in cattle, and perception and behaviours associated with brucellosis control among agro-pastoralists in Morogoro Region, Tanzania. A cross-sectional study involving herd milk diagnosis by indirect enzyme-linked immunosorbent assay and questionnaire survey was conducted in 124 farms. Questions included potential risk factors, knowledge of brucellosis, willingness-to-pay for cattle vaccination, and item count technique (ICT) for selling behaviour of cows that experienced abortion. Risk factor analysis for *Brucella* sero-positivity in cattle and analysis of factors associated with willingness-to-pay were conducted using classical tests and generalised linear models. Most farmers had little knowledge about brucellosis (disease name: 13.7%, symptoms: 3.2%, transmission from cattle to human: 2.4%, and *Brucella* vaccine: 2.4%). The proportion of *Brucella* sero-positive herd was 44.4% (55/124, 95%CI: 35.5–53.5). No risk factors for *Brucella* sero-positivity were identified; however, using a veterinary service was identified as a preventive factor (OR = 0.39, 95%CI: 0.18–0.84, $p = 0.02$). For scenarios of vaccinating all cattle and only calves, 59.7% and 89.5% of farmers were willing to pay for vaccination, respectively. Being a Maasai tribe member was a hesitating factor for vaccinating all cattle (OR = 0.39, 95%CI: 0.19–0.83, $p = 0.01$) and using a veterinary service was an encouraging factor for vaccinating calves (OR = 4.0, 95%CI: 1.2–13.0, $p = 0.02$). The ICT found that 45.1% of farmers sold cows that experienced abortion. This estimate was not statistically different from that obtained by direct questioning (34.1%, SE = 7.5%, binomial p value = 0.27, factor score = 1.32), suggesting that farmers did not hesitate to sell such cows. The Maasai conducted more risky behaviours for human infection such as drinking raw milk ($p = 0.06$) or blood ($p < 0.01$) and helping delivery with bare hands ($p = 0.03$) than other tribes. Community-based brucellosis control programmes with calf vaccination may be feasible in the study areas. A One Health approach including the promotion of health education and expansion of veterinary services is crucial for disease control.

1. Introduction

Brucellosis is a worldwide zoonotic disease caused by several species of the genus *Brucella*. Although many developed countries have eradicated the disease, it is endemic in some regions such as Latin America, Mediterranean, Middle East, Africa, and Asia, causing large economic losses due to the problems of livestock production and human health (Corbel et al., 2006). In domesticated animals, the disease is characterised by abortion, infertility in adult animals, and reduced milk yields. Although most infected cows will only abort once, they remain a source of infection during subsequent normal calvings (Goodwin and Pascual, 2016). In humans, clinical symptoms include fever, headache, weakness, malaise, arthralgia, and other less common clinical manifestations (Sauret and Vilissova, 2002). *Brucella* species have also been detected from a variety of wildlife, such as bison (*Bison bison*), red deer (*Cervus elaphus*), feral swine, wild boar (*Sus scrofa*), African buffalo

Abbreviations: CI, confidence interval; GLM, generalised linear model; ICT, item count technique; IELISA, indirect enzyme-linked immunosorbent assay; OR, odds ratio; SE, standard error

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https://doi.org/10.1016/j.actatropica.2018.07.010
Received 18 January 2018; Received in revised form 12 July 2018; Accepted 12 July 2018
Available online 01 August 2018

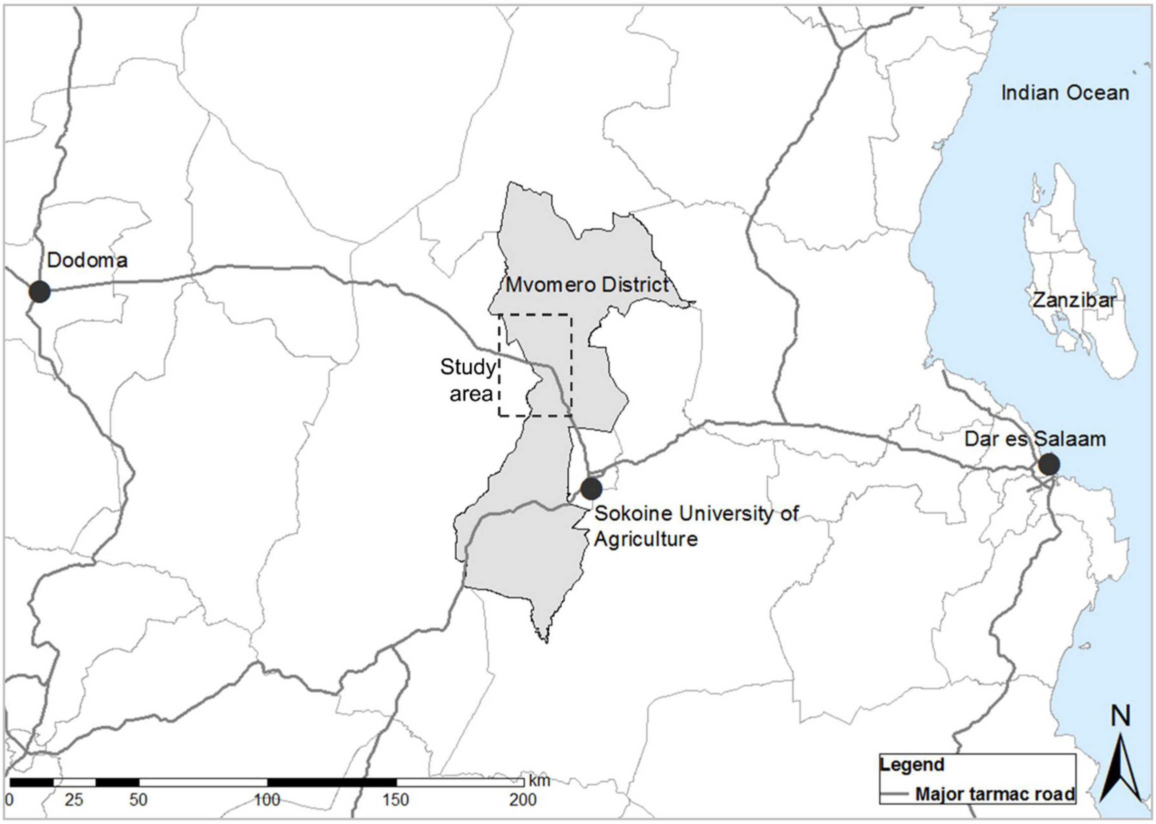

Fig. 1. Map showing Mvomero District in Morogoro Region, Tanzania. A square with dotted lines shows the study area (See Fig. 2).

(*Syncerus caffer*), and lion (*Panthera leo*), which may act as reservoirs for livestock and human infection (Assenga et al., 2015; Corbel et al., 2006; Godfroid et al., 2013b; Madsen and Anderson, 1995). The sources of infection for animals include aborted foetus, placenta, milk, and semen from infected animals (Corbel et al., 2006). The most common sources of human infection are unheated livestock products (Busch and Parker, 1972). Contact among livestock, wildlife, and human is common for pastoral and agro-pastoral farmers in Tanzania (Assenga et al., 2015). In addition, risky behaviours for human infection such as eating raw meat and drinking raw milk have been observed among some farmers (John et al., 2010).

Zoonoses can be controlled most efficiently and surely by tackling animal reservoirs. As control strategies, biosecurity at the farm level, test-and-slaughter programmes, and immunisation have been demonstrated as notable tools for brucellosis control in livestock (Pérez-Sancho et al., 2015). However, these control measures may conflict with the customs of affected communities, such as pastoralists and extensive agropastoralists, and may be challenging due to the high cost for surveillance, slaughter of infected animals, and general compensation in developing countries (Marcotty et al., 2009).

In terms of immunisation, *Brucella abortus* vaccines have been successfully used worldwide for bovine brucellosis control for decades. In sub-Saharan Africa, national brucellosis control programmes involving vaccination were performed in southern African countries. However, outside of southern Africa, vaccination was rarely conducted and if done, it was a makeshift effort rather than a coordinated national programme (McDermott and Arimi, 2002). One of the reasons of underuse of vaccines in developing countries is lack of public resources. Considering the circumstances, this study seeks an opportunity for a community-based disease control strategy wherein cattle farmers pay for *Brucella* vaccination by themselves, which would be sustainable if accepted.

Another concern about the maintenance and transmission of bovine brucellosis is cattle trade. Selling out cattle that experienced abortion may occur due to moral hazards; thus, investigation into the perception and behaviour of cattle keepers would provide useful information in planning brucellosis control strategies. Abortion is not only caused by *B. abortus* in cattle but also by several other pathogens such as *Neospora caninum*, bovine viral diarrhoea virus, bovine herpesvirus type 1, and *Leptospira interrogans* (Asmare et al., 2013; Fraser, 1991). However, previous reports have shown a significant association between abortion and infection with *B. abortus* in eastern and southern Africa (Asakura et al., 2017; Makita et al., 2011; Megersa et al., 2011; Muma et al., 2006), supporting the hypothesis that selling out cattle that experienced abortion may be a risk factor for disease spread.

In this study, we used the item count technique (ICT), introduced by Droitcour et al. (2004), to understand the behaviour of selling out cows that experienced abortion. The ICT is an indirect questioning technique used to estimate the proportion of people who have engaged in a sensitive behaviour. Estimation using the ICT is expected to be higher than that from conventional direct questioning. For example, Tsuchiya et al. (2007) reported that the ICT indicated significantly more shoplifting activities compared with direct questioning. In animal health, Randrianantoandro et al. (2015) studied illegal sales of African swine fever-infected pork in Madagascar using the ICT.

The objective of this study was to investigate the herd-level risk factors associated with *Brucella* sero-positivity in cattle and perception and behaviours related to its control, including willingness to pay for vaccination, among agro-pastoralists in Morogoro Region, Tanzania, where brucellosis is endemic (James, 2013).

2. Methods

2.1. Study site

This study was conducted in Mvomero District in Morogoro Region, Tanzania (Fig. 1). The target population was agro-pastoralists in Mvomero District. Two villages in Dakawa ward and four villages in Mvomero ward were selected from the district, based on the typical agro-pastoral system representative of the district, availability of the lists of cattle farms and the reachable distance from Morogoro municipality, where Sokoine University of Agriculture is located. The district is located between latitudes 8°0′ and 10°0′S and between longitudes 37°0′ and 28°22′E. The altitude of the district is between 380 and 1,520 m above sea level, providing a suitable climate for a variety of tropical and subtropical crops. The district receives a bimodal type of rainfall with peaks in April and December for long and short rains, respectively, while May to October remains relatively dry (Mkoma and Mjemah, 2011). The average rainfall amounts to 1200 mm per annum with variations from 800 to 2000 mm (Movek, 2008). The district's economy depends on agriculture, mainly from crop production. The livestock types found in the district comprise cattle, goats, sheep, pigs, donkeys, and birds. Most cattle are indigenous breeds raised with a semi-extensive or extensive system, and a few improved dairy cattle are reared mainly with an intensive system. Most of the cattle farmers in the study areas are agro-pastoralists who basically keep and graze their cattle herds independently without using communal grazing with other herds. Here a herd is defined as a group of cattle kept together and owned by an owner. As an exception, sometimes communal grazing is conducted especially in the case that the owners are family members, relatives or neighbours. On the other hand, some cattle owners who keep a large number of cattle separate their cattle into several herds. Generally in a family, men play roles in grazing livestock and in business decision, while women in milking regarding livestock husbandry in the area. However, some Maasai married women have ownership of livestock and make a business decision.

2.2. Study design and sample size estimation

A cross-sectional study involving herd milk sampling and a structured interview using a questionnaire was designed. Although this study had multiple purposes, the farm sample size was calculated by the following formula for estimating herd-level prevalence (Thrusfield, 2005):

$$n = \frac{1.96^{2} * P_{\exp}(1 - P_{\exp})}{d^{2}} \tag{1}$$

where n is the required sample size based on an infinite population, $P_{\exp}$ is the expected prevalence, and d is desired absolute precision. We set $P_{\exp}$ as 0.529 based on results from our previous study [18] and d as 0.05. The calculated sample size was 383. However, in the case of small populations, the required sample size, n_{adj}, given by the following formula can be adopted:

$$n_{adj} = \frac{N \times n}{N + n} \tag{2}$$

where n, obtained from Eq. (1), is the sample size and N is the number of cattle farms based on the lists of farms, which was 170. As a result, n_{adj} was calculated as 118. Farms to be sampled were proportionally allocated to the villages according to the numbers of cattle farms within them, and study farms were selected from the lists by random sampling using runif() function in statistical software R. After a recruitment process by the veterinary officers based on the random sampling, 124 farms were included in our study.

2.3. Field survey

A field survey was conducted from September to October 2016. Herd milk was sampled from milk can or bulk tank and was collected in Falcon tubes and brought to the laboratory in Sokoine University of Agriculture. Milk samples were stored in a freezer at −20 °C until diagnostic testing.

Information on farm owner, farm characteristics, the animals kept, and willingness-to-pay for *Brucella* vaccine was collected using a structured questionnaire written in English that had been pre-tested with cattle farmers. Translation of the questions into the national language, Swahili, was validated in advance by cross-checking with Swahili-speaking individuals, and the questionnaire was administered by face-to-face interviews in Swahili. In cases when the owners were absent, their family members or employees involved in cattle raising answered the questionnaire instead, except for the question on willingness to pay for vaccination, which was asked of the owners by telephone. Farmers were asked questions related to knowledge about brucellosis prior to the explanation of brucellosis. Willingness to pay for vaccination was surveyed after the explanation of brucellosis and the vaccine. The vaccine price was set as 3000 Tanzania shilling (approximately 1.3 USD at the time of writing) per shot in this study, taking into account pricing information obtained from a veterinary officer who owned his own veterinary medicine shop.

2.4. Diagnostic test

All herd milk samples were tested in duplicate using an indirect enzyme-linked immunosorbent assay (IELISA) (Boehringer Ingelheim Svanova, Uppsala, Sweden). IELISA was performed following the manufacturers' instructions at Sokoine University of Agriculture. For IELISA, the optical density was measured at 450 nm using an ELISA plate reader, Multiskan RC version 6.0 (Thermo Labsystems, Helsinki, Finland). A farm with a positive milk IELISA result was regarded as an infected farm.

2.5. Statistical analysis

The sensitivity and specificity of milk IELISA for individual milk samples was assumed to be 99.6% and 99.1%, respectively (Vanzini et al., 1998). However, in this study, herd milk samples were tested. As no information was available for herd-level sensitivity and specificity of milk IELISA, this study used the apparent prevalence, and the 95% confidence interval (CI) was calculated.

For univariable risk factor analyses, the Wilcoxon rank-sum test was performed for count and score data. Pearson's chi-squared test with Yates' continuity correction was performed for binary and categorical data, and Fisher's exact test was used when at least one cell included an expected frequency of less than 5. In addition, education and knowledge scores were established based on the level of education of the farmers and the number of knowledge items on brucellosis the farmers knew (farmers' education level and knowledge items are shown in Table 2).

In multivariable analysis, a generalised linear model (GLM) with binomial errors was used, selecting IELISA results as response variables, and variables with p values < 0.2 in univariable analyses were considered as explanatory variables. Collinearity was evaluated for all combinations of these explanatory variables with a cut-off correlation = 0.9; no collinearity was found among these variables. Backward stepwise simplification was conducted using the likelihood ratio test.

To identify factors associated with willingness to pay for vaccination, univariable and multivariable analyses were performed using the same method and procedure for risk factor analysis of brucellosis. However, the status of *Brucella* sero-positivity was excluded from the analysis because farmers did not know the test results at the time of interview. Associations between conduct of risky behaviours for human brucellosis infection and *Brucella* sero-positivity in cattle and between conduct of risky behaviours and tribes were analysed using Pearson's chi-squared test. Fisher's exact test was used in cases in which at least one cell included an expected frequency of less than 5. Statistical analyses were performed using the computer software R version 3.3.2 (R Core Team, 2016). Statistical significance was considered to exist at p values < 0.05.

2.6. The item count technique (ICT)

There are two types of ICT, the single list and double list. The double list technique was performed in this study since it cuts the variance of the estimate in half, consequently providing a more accurate estimate (Droitcour et al., 2004). Two baseline lists, X and Y, that each contained different items were prepared so that the key item was presented to all respondents. The key item was 'sell out cattle that experienced abortion to cattle markets'.

Baseline list X was as follows:

(1) Use bulls for breeding
(2) Drink raw milk
(3) Ask a veterinary officer when cattle have fever
(4) Send cattle for grazing

Baseline list Y was as follows:

(1) Use chemical insecticides on cattle to kill ticks
(2) Feed commercial concentrates to cattle
(3) Sell manure to others
(4) Have a biogas plant

Table 1 shows subsamples used in this study. The respondents were told that the questionnaire was anonymous in order to yield honest answers, and they were asked to report the number of items in each list that were true for them without mentioning which ones. According to a published guideline (Dalton and Wimbush, 1994), at least 40–50 respondents are needed for each subsample to assure stability and accuracy of the estimate. Thus, the total of 124 farmers was equally divided into three subsamples, and farmers were randomly assigned to a subsample.

The proportion of farmers engaged in the key item as assessed using the ICT was calculated by the following formula:

$$p_1 = \frac{1}{2}[(\overline{X}_{5A} - \overline{X}_{4B}) + (\overline{Y}_{5B} - \overline{Y}_{4A})] \quad (3)$$

where p_1 is the proportion of farmers who sell out cattle that experienced abortion, $\overline{X}_{5A}$ is the mean number of items on 'baseline list X plus key item' engaged in by Subsample A, $\overline{X}_{4B}$ is the mean number of items on 'baseline list X' engaged in Subsample B, $\overline{Y}_{5B}$ is the mean number of items on 'baseline list Y plus key item' engaged in by Subsample B, and $\overline{Y}_{4A}$ is the mean number of items on 'baseline list Y' engaged in by Subsample A. The variance of the estimate was calculated using the formula explained by Droitcour et al. (2004). The estimation from a direct question, p_2,was obtained from the following equation:

$$p_2 = \frac{n_y}{N_{dq}} \quad (4)$$

where n_y is the number of 'yes' responses and N_{dq} is the number of respondents of the direct question, which is the sample size of Subsample C. If p_1 is significantly higher than p_2, then human behaviour in the key item is considered to be sensitive.

The binomial test named *Twobinom* (LaBrie and Earleywine, 2000; Wilcox, 1996) was used for the comparison of p_1 and p_2. The factor score was calculated by dividing the estimation from the ICT by that from direct questioning, therefore showing the efficiency of the ICT compared with direct questioning.

Table 1
Questionnaire and sample size for each subsample of the ICT.

	Subsample A	Subsample B	Subsample C
Questionnaire	Baseline X + key item Baseline Y	Baseline X Baseline Y + key item	Direct question
Sample size	41	41	42

Table 2
Characteristics of the study farms.

Categories	Response (n = 124 farms)	Percentage
Characteristics of owner		
Tribe: Maasai	60	48.3
Male-owned farm	107	86.3
Level of education		
No education	59	47.6
Primary	46	37.1
Secondary	14	11.3
Diploma	1	0.8
University	4	3.2
Knowledge on brucellosis		
Name of the disease	17	13.7
Symptoms	4	3.2
Transmission from cattle to human	3	2.4
Brucella vaccine	3	2.4
Characteristics of farming		
Grazing system: semi/free grazing	123	99.2
Conducting agriculture	105	84.7
Cattle herded with goats or sheep	94	75.8
Breeding system		
Own bull	114	91.9
Bull from other farms	9	7.3
Artificial insemination	2	1.6
Purchase of cattle in the past year	67	54.0
Abortion of cattle in the past year	68	54.8
Contact with other livestock herd	113	91.1
Contact with wild animals	17	13.7
History of the use of any kind of vaccine	41	33.1
History of the use of *Brucella* vaccine	0	0
Using a veterinary service	84	67.7

3. Results

3.1. Characteristics of the study farms

Table 2 shows the socio-economical characteristics, knowledge about bovine brucellosis, and farming style of cattle owners. Only one farm operated a zero-grazing system; the others used semi/free grazing. One farm used both its own bull and artificial insemination for cattle breeding.

3.2. Herd-level prevalence

Fig. 2 shows the geographical distributions of brucellosis positive and negative herds in the study area. The apparent herd-level prevalence was 44.4% (55/124, 95%CI: 35.5–53.5).

3.3. Risk factors for brucellosis

Tables 3 and 4 show the results of univariable analyses for brucellosis for binary data and count and score data, respectively. Significant variables were using a veterinary service (p = 0.03) as a preventive factor and larger herd size (p = 0.049) as a risk factor.

In the multivariable analysis, the final model included one preventive factor, using a veterinary service [odds ratio (OR) = 0.39, 95%CI: 0.18–0.84, p = 0.02].

3.4. Willingness-to-pay for vaccination

Approximately 59.7% and 89.5% of farms were willing to pay for *Brucella* vaccine for all cattle and newborn calves, respectively. In the univariable analyses for vaccinating all cattle option, the Maasai

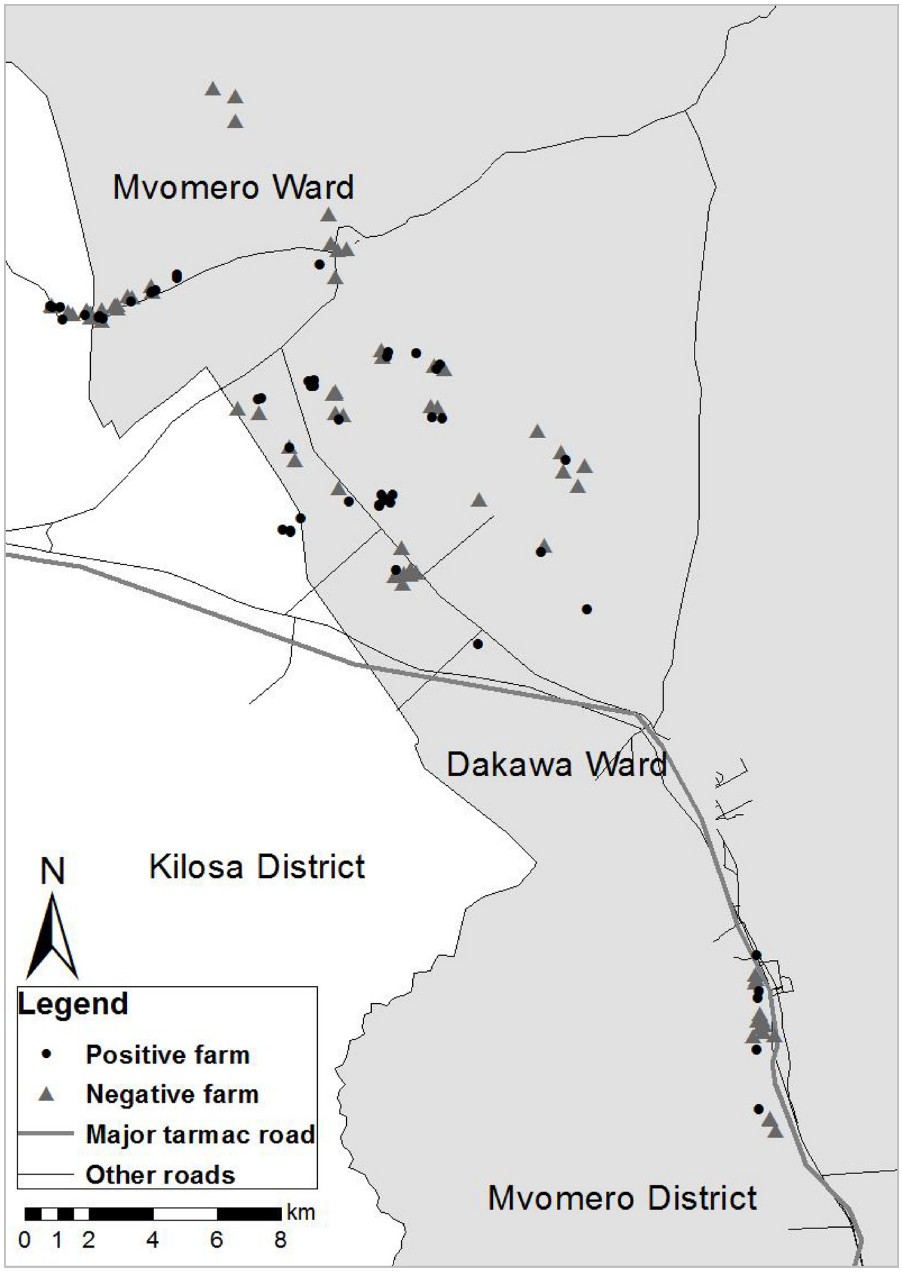

Fig. 2. The distributions of brucellosis positive and negative herds in Dakawa and Mvomero Wards.

significantly refused to pay for vaccination compared with other tribes (p = 0.02, Table 5), and the number of cattle of accepted farmers was significantly lower than that of refused farmers ($p < 0.01$, Table 6). In addition, education and knowledge scores were significantly higher in the group that accepted vaccination than the group that refused (p = 0.02 and 0.01, respectively, Table 6). Out of these factors, tribe was the single significant factor in the multivariable analysis for all cattle vaccination (Maasai: OR = 0.39, 95%CI: 0.19–0.83, p = 0.01). For vaccinating calves option, a significant factor in the univariable analysis was using a veterinary service (p = 0.03, Table 5), which was also the single significant factor in the multivariable analysis (OR = 4.0, 95%CI: 1.2–13.0, p = 0.02).

3.5. Behaviour of selling cows that experienced abortion

Based on the ICT, 45.1% [standard error (SE) = 7.4%] of farmers sold cattle with a history of abortion to the cattle market. This estimate was not statistically different from that obtained by direct questioning (34.1%, SE = 7.5%, binomial p value = 0.27, factor score = 1.32). Thus, farmers sold out cattle with a history of abortion without hesitation.

3.6. Human risks for brucellosis

Table 7 shows the result of the univariable analyses of the risky behaviours for brucellosis infection in humans for binary data. No significant association was observed between *Brucella* sero-positivity in cattle and risky behaviours. However, the Maasai significantly conducted risky behaviours compared with other tribes (drinking raw milk: p = 0.06, drinking blood: $p < 0.01$, using gloves to help delivery: p = 0.03, Table 8).

Table 3
Univariable risk factor analysis on binary response.

Factors	Response	Infected herds	Healthy herds	Prevalence (%)	Test statistics	*p*-value
Presence of goats or sheep	Yes	39	55	41.5	x^2 = 0.86, df = 1	0.35
	No	16	14	53.3		
Contact with other livestock	Yes	53	60	46.9	Fisher's exact test	0.11
	No	2	9	18.2		
Contact with wildlife	Yes	7	10	41.2	Fisher's exact test	1
	No	48	59	44.9		
Abortion of cattle in the past year	Yes	32	36	47.1	x^2 = 0.24, df = 1	0.63
	No	23	33	41.1		
Using a veterinary service	Yes	31	53	36.9	x^2 = 4.96, df = 1	0.03
	No	24	16	60.0		
Public vet treatment	Yes	13	25	34.2	x^2 = 1.73, df = 1	0.19
	No	42	44	48.8		
Owners' sex	Male	49	58	45.8	x^2 = 0.30, df = 1	0.58
	Female	6	11	35.3		
Tribe	Maasai	30	30	50.0	x^2 = 1.09, df = 1	0.30
	Others	25	39	39.1		

Table 4
Univariable risk factor analysis for brucellosis on count and score data.

Factor	Infected herds (2.5 and 97.5th percentiles)	Healthy herds (2.5 and 97.5th percentiles)	*p*-value
Number of cattle	50 (8–219)	30 (4–230)	0.049
Number of cattle purchased in the past year	2 (0–16)	1 (0–33)	0.88
Age of owner	42 (20–75)	45 (21–71)	0.41
Education score (0: no education; 1: primary; 2: secondary; 3: diploma; 4: university)	1 (0–3)	1 (0–4)	0.84
Knowledge score: 0–4	0 (0–1)	0 (0–3)	0.07

4. Discussion

This study was performed from diversified perspectives towards brucellosis control focusing on agro-pastoral areas in Tanzania. In this study, the result of *Brucella* sero-positivity in cattle revealed that the disease is endemic in the study area, as suggested by other studies conducted in the same district (Asakura et al., 2017; Temba, 2012).

Multivariable risk factor analysis indicated using a veterinary service as a preventive factor of *Brucella* sero-positivity in cattle. Other studies mentioned that the lower prevalence of bovine brucellosis in herds under the supervision of a veterinarian is likely due to improved monitoring and preventive health measures for the disease such as proper disposal of aborted materials and hygienic procedures (Al-Majali et al., 2009; de Alencar Mota et al., 2016; Kaoud et al., 2010). It is well known that delivering adequate animal health services contributes to a low incidence of diseases. Veterinary extension is considered to play a key role in zoonosis mitigation through education of sanitary procedures and rearing measures for livestock farmers, raising awareness of the diseases.

Vaccination plays a major role in brucellosis control in endemic areas. National brucellosis control programmes involving vaccination have been performed in many countries (FAO, 2015; McDermott and Arimi, 2002; McDermott et al., 2013). However, to our knowledge, there were no reports about a community-based *Brucella* vaccination strategy in which farmers pay the cost themselves. Serological testing with slaughter is also the main method for disease control. However, it is not always cost-effective in cattle (FAO, 2015) and compensation for slaughtered cattle is difficult for developing countries where financial resources are scarce. Considering the limited resources and endemic situation of brucellosis in Tanzania (Assenga et al., 2015; Jiwa et al., 1996; Swai and Schoonman, 2010; Weinhäupl et al., 2000), we studied willingness to pay for *Brucella* vaccination by farmers themselves as a community-based intervention.

In general, brucellosis mass vaccination is targeted at the entire cattle population of the target area. However, vaccination of adult cows causes infertility and shedding of *Brucella* bacteria in milk (Chand et al., 2015; Kunda et al., 2007). Thus, this study investigated the cattle farmers' willingness to pay for vaccination in two scenarios: all cattle vaccination and calf vaccination. The results clarified that around 90% of farmers would be willing to pay for vaccination for calves, while around 60% of them agreed to pay for all cattle, indicating the feasibility of a calf vaccination programme. In addition, although the question of willingness to pay for calf vaccination assumed to include both sexes, vaccination is performed only for female calves when implemented, as vaccination in male calves may result in testicular infection and infertility (Dorneles et al., 2015). Thus, the acceptance rate of a vaccination programme targeting only female calves is expected to be even higher than that observed in our study. Similar to the analysis

Table 5
Univariable analysis for the factors associated with willingness to pay for *Brucella* vaccination on binary response.

Factors	Response	Vaccinate all cattle				Vaccinate calves			
		Accept	Not accept	Accept (%)	*p*-value	Accept	Not accept	Accept (%)	*p*-value
Abortion of cattle in the past year	Yes	39	29	57.4	0.69	60	8	88.2	0.83
	No	35	21	62.5		51	5	91.1	
Using a veterinary service	Yes	53	31	63.1	0.35	79	5	94.0	0.03
	No	21	19	52.5		32	8	80.0	
Tribe of the owner	Maasai	29	31	48.3	0.02	52	8	86.7	0.48
	Others	45	19	70.3		59	5	92.2	

Table 6
Univariable analysis for the factors associated with willingness to pay for *Brucella* vaccination on count and score data.

Factor	Vaccinate all cattle			Vaccinate calves		
	Accept (2.5–97.5th)	Not accept (2.5–97.5th)	*p*-value	Accept (2.5–97.5th)	Not accept (2.5–97.5th)	*p*-value
Number of cattle	30 (3–203)	50 (12–234)	< 0.01	45 (4–218)	30 (12–229)	0.54
Age of owner	45 (21–70)	42 (20–76)	0.39	43 (21–75)	50 (28–69)	0.43
Education score: 0–4	1 (0–3.2)	0 (0–3.6)	0.02	1 (0–3)	0 (0–3)	0.13
Knowledge score: 0–4	0 (0–2.4)	0 (0–0.8)	0.01	0 (0–2.5)	0 (0–0.7)	0.49

Table 7
Proportions of infected and non-infected herd households conducting risky behaviours for human infections with *Brucella*.

Factors	Response	Infected herds	Healthy herds	Percentage	*p*-value
Drinking raw milk	Yes	33	38	46.5	0.71
	No	22	31	41.5	
Drinking blood	Yes	18	20	47.4	0.80
	No	37	49	43.0	
Using gloves to help delivery	Yes	5	8	38.5	0.88
	No	50	61	45.0	

Table 8
Proportions of Maasai and other tribe households conducting risky behaviours for human infections with *Brucella*.

Factors	Maasai ($n = 60$)	Percentage	Other tribes ($n = 64$)	Percentage	*p*-value
Drinking raw milk	40	66.7	31	48.4	0.06
Drinking blood	38	63.3	0	0	< 0.01
Using gloves to help delivery	2	3.3	11	17.2	0.03

of preventive factors of *Brucella* sero-positivity in cattle, the multivariable analysis for the factors associated with willingness to pay for vaccination showed that veterinarians were motivators for farmers to accept calf vaccination. A previous report also showed that advice from veterinarians was a promoting factor for farmers to have an intention towards zoonotic disease control (Ellis-Iversen et al., 2010). For the all cattle vaccination option, being a Maasai tribe member was a hesitating factor. This may be due to their traditional culture, which is very different from modern culture. Although most farmers including the Maasai agreed to calf vaccination, calf vaccination may have been temporarily accepted by the Maasai because a very limited number of cattle are supposed to be vaccinated initially. Since brucellosis and *Brucella* vaccine were new concepts for most of the farmers at the interview, the Maasai may be sceptical about using the vaccine.

Cattle play an important role in farmer's livelihood. Cattle were assets and were kept as a bank account for the studied farmers, and the livestock assets are sold when cash is necessary for such as school fees, ceremony, illness or injury, according to informal interviews during the survey of this study. Moreover, many farmers pay attention to livestock diseases because the disease status affect the owner's reputation and livestock price. This situation is likely to be associated with high willingness to pay for vaccination, regardless of the difficulty for farmers to perceive the loss caused by brucellosis due to the nonspecific symptoms of the disease both in cattle and human. In terms of benefit of vaccination, a study on brucellosis conducted in Mongolia has shown high benefit-to-cost ratio of mass vaccination for cattle and small ruminants for society: 3.2 (95%CI: 2.27–4.37) (Roth et al., 2003). This kind of study would be helpful for farmers to make decision for vaccination.

This study investigated human behaviour of selling aborted cows using the ICT. The ICT found that around half of the study farms sold out cows that experienced abortion without hesitation. The farmers in this study sell their cattle at the Mkongeni cattle market, where most adult cattle traded are brought to Dar es Salaam for slaughter and meat consumption (personal communication with district livestock officers). Selling out infected cattle will decrease the brucellosis within-herd prevalence. However, farmer-to-farmer trades of cattle were also observed in the market, and selling out cows that experienced abortion at the market may contribute to the spread of brucellosis to other farms. Although abortion in cattle can occur for several different reasons (as explained in Section 1), abortion is strongly associated with bovine brucellosis in endemic areas (Asakura et al., 2017; Makita et al., 2011; Megersa et al., 2011; Muma et al., 2006). Therefore, suggesting farmers to sell out cows that experienced abortion for slaughter, not for raising in other farms, and admitting meat consumption of slaughtered cattle might be practical control methods of brucellosis in developing countries, as long as farmers agree to participate.

Our study found poor knowledge about brucellosis among farmers in the study area, whereas other studies conducted in similar settings reported that agro-pastoralism was associated with high knowledge of zoonoses such as brucellosis (Kansiime et al., 2014) and pulmonary tuberculosis (Gele et al., 2009; Legesse et al., 2010). The farmers in the study area had few opportunities to learn about the disease (personal communication), thus promotion of health education is required.

In terms of the risk of human infection, the Maasai tended to conduct risky behaviours. This is due to their cultural background and traditional habits; thus, it may be difficult to change these customs. The Maasai had previously regarded education as less important and the education level of Maasai was significantly lower than that of other tribes ($p < 0.01$). However, the Tanzania government is currently providing a premium on education and the number of Maasai children who go to school has increased in the study area (personal communication with Maasai participants). For this reason, the probability of success in changing risky behaviours against human infection by health education programmes may increase.

This study was conducted with the representative agro-pastoral cattle keepers in Mvomero District; however, the findings may have limitation in extrapolating to the other agro-pastoral areas in Tanzania due to different composition of tribes and climate. Among animal brucellosis, we studied only bovine brucellosis. Although research focusing on human and small ruminants has been conducted in Tanzania (Assenga et al., 2015; Kunda et al., 2007; Swai and Schoonman, 2009), the information is still limited. Moreover, isolation and identification of *Brucella* species have not been performed in more than five decades in Tanzania (Assenga et al., 2015; Godfroid et al., 2013a). Recently, the Tanzania government has selected brucellosis as one of the prioritised zoonotic diseases for the country, confirming the plan to conduct One Health surveillance for both humans and animals for rapid and effective response to improve current public health situations (National Workshop on Prioritization of Zoonotic diseases, 2017). By using the knowledge generated in this study, supplemented by such current research opportunities, a feasible plan for community-based control of brucellosis may become available in Tanzania in the near future.

In conclusion, this study showed that a One Health approach for joint planning and actions of community-based brucellosis intervention using calf vaccination and selling out cows experienced abortion, with the supports by the veterinary and public health authorities including animal health and health education, is feasible in Tanzania, as well as potentially in the other endemic countries.

Funding

This work was supported by the Ministry of Education, Culture, Sports, Science and Technology of Japan under the research project “Development of rapid diagnostic kits for infectious pathogens in industry animals and establishment of effective control methods through global analysis of transmission routes (grant number S1391001)” of “2013 Support grant for establishment of strategic research platform for private universities”.

Ethics approval and consent to participate

This study was approved by the Ethical Committee of the Graduate School of Dairy Science, Rakuno Gakuen University (approval number 16-3). Permission to conduct the study in Mvomero District was sought and granted by district executive officers (reference number MVDC/C.80/4 VOLII/126). Verbal informed consent was obtained from each participant after veterinary field officers explained the aim of the study.

Authors’ contribution

SA wrote the majority of the manuscript and coordinated the research and performed field surveys, laboratory diagnosis and data analysis. GM contributed to field surveys and laboratory diagnosis. RK contributed to design and coordination of the research. KM is the principal investigator of the research and contributed to study design, field surveys, supervising data analyses, and writing the manuscript. All authors read and approved the final manuscript.

Competing interests

The authors declare that they have no competing interests.

Acknowledgments

The authors would like to thank all farmers who participated in the survey. We also thank Mvomero district officials and veterinary, livestock and agriculture field officers for their cooperation during the implementation of this study.

References

Al-Majali, A.M., Talafha, A.Q., Ababneh, M.M., Ababneh, M.M., 2009. Seroprevalence and risk factors for bovine brucellosis in Jordan. J. Vet. Sci. 10, 61–65.
Asakura, S., Makingi, G., Kazwala, R., Makita, K., 2018. Brucellosis risk in urban and agro-pastoral areas in Tanzania. Ecohealth 15, 41–51.
Asmare, K., Regassa, F., Robertson, L.J., Martin, A.D., Skjerve, E., 2013. Reproductive disorders in relation to Neospora caninum, Brucella spp. and bovine viral diarrhoea virus serostatus in breeding and dairy farms of central and southern Ethiopia. Epidemiol. Infect. 141, 1772–1780.
Assenga, J.A., Matemba, L.E., Muller, S.K., Malakalinga, J.J., Kazwala, R.R., 2015. Epidemiology of Brucella infection in the human, livestock and wildlife interface in the Katavi-Rukwa ecosystem, Tanzania. BMC Vet. Res. 11, 189.
Busch, L.A., Parker, R.L., 1972. Brucellosis in the United States. J. Infect. Dis. 125, 289–294.
Chand, P., Chhabra, R., Nagra, J., 2015. Vaccination of adult animals with a reduced dose of Brucella abortus S19 vaccine to control brucellosis on dairy farms in endemic areas of India. Trop. Anim. Health Prod. 47, 29–35.
Corbel, M.J., Elberg, S.S., Cosivi, O., 2006. Brucellosis in Humans and Animals. World Health Organization, Geneva.
Dalton, D.R., Wimbush, J.C., 1994. Using the unmatched count technique (UCT) to estimate base rates for sensitive behaviour. Pers. Psychol. 47, 817–829.
de Alencar Mota, A.L.A., Ferreira, F., Ferreira Neto, J.S., Dias, R.A., Amaku, M., Hildebrand Grisi-Filho, J.H., Telles, E.O., Picao Goncalves, V.S., 2016. Large-scale study of herd-level risk factors for bovine brucellosis in Brazil. Acta Trop. 164, 226–232.
Dorneles, E.M., Sriranganathan, N., Lage, A.P., 2015. Recent advances in Brucella abortus vaccines. Vet. Res. 46, 76.
Droitcour, J., Caspar, R.A., Hubbard, M.L., Parsley, T.L., Visscher, W., Ezzati, T.M., 2004. Measurement Errors in Surveys: The Item Count Technique as a Method of Indirect Questioning: A Review of its Development and a Case Study Application.
Ellis-Iversen, J., Cook, A.J., Watson, E., Nielen, M., Larkin, L., Wooldridge, M., Hogeveen, H., 2010. Perceptions, circumstances and motivators that influence implementation of zoonotic control programs on cattle farms. Prev. Vet. Med. 93, 276–285.
FAO, 2015. Regional Workshop on Brucellosis Control in Central Asia and Eastern Europe. FAO Animal Production and Health Report 8.
Fraser, C.M., 1991. The Merck Veterinary Manual. Merck, Rahway, NJ.
Gele, A.A., Bjune, G., Abebe, F., 2009. Pastoralism and delay in diagnosis of TB in Ethiopia. BMC Publ. Health 9, 5.
Godfroid, J., Al Dahouk, S., Pappas, G., Roth, F., Matope, G., Muma, J., Marcotty, T., Pfeiffer, D., Skjerve, E., 2013a. A “one health” surveillance and control of brucellosis in developing countries: moving away from improvisation. Comp. Immunol. Microbiol. Infect. Dis. 36, 241–248.
Godfroid, J., Garin-Bastuji, B., Saegerman, C., Blasco, J.M., 2013b. Brucellosis in terrestrial wildlife. Rev. Sci. Tech. (Int. Off. Epizoot.) 32, 27–42.
Goodwin, Z.I., Pascual, D.W., 2016. Brucellosis vaccines for livestock. Vet. Immunol. Immunopathol. 181, 51–58.
James, L.W., 2013. Studies of Human Brucellosis in Mikumi–Selous Ecosystem, Morogoro, Tanzania. Sokoine University of Agriculture, Morogoro, Tanzania.
Jiwa, S.F.H., Kazwala, R.R., Tungaraza, R., Kimera, S.I., Kalaye, W.J., 1996. Bovine brucellosis serum agglutination test prevalence and breed disposition according to prevalent management systems in the Lake Victoria zone of Tanzania. Prev. Vet. Med. 26, 341–346.
John, K., Fitzpatrick, J., French, N., Kazwala, R., Kambarage, D., Mfinanga, G.S., MacMillan, A., Cleaveland, S., 2010. Quantifying risk factors for human brucellosis in rural northern Tanzania. PLoS One 5, e9968.
Kansiime, C., Mugisha, A., Makumbi, F., Mugisha, S., Rwego, I.B., Sempa, J., Kiwanuka, S.N., Asiimwe, B.B., Rutebemberwa, E., 2014. Knowledge and perceptions of brucellosis in the pastoral communities adjacent to Lake Mburo National Park, Uganda. BMC Publ. Health 14, 242.
Kaoud, H.A., Zaki, M.M., El-Dahshan, A.R., Nasr, S.A., 2010. Epidemiology of brucellosis among farm animals. Nat. Sci. 8.
Kunda, J., Fitzpatrick, J., Kazwala, R., French, N.P., Shirima, G., Macmillan, A., Kambarage, D., Bronsvoort, M., Cleaveland, S., 2007. Health-seeking behaviour of human brucellosis cases in rural Tanzania. BMC Publ. Health 7, 315.
LaBrie, J.W., Earleywine, M., 2000. Sexual risk behaviors and alcohol: higher base rates revealed using the unmatched-count technique. J. Sex Res. 37, 321–326.
Legesse, M., Ameni, G., Mamo, G., Medhin, G., Shawel, D., Bjune, G., Abebe, F., 2010. Knowledge and perception of pulmonary tuberculosis in pastoral communities in the middle and Lower Awash Valley of Afar region, Ethiopia. BMC Publ. Health 10, 187.
Madsen, M., Anderson, C.E., 1995. Serologic survey of Zimbabwean wildlife for brucellosis. J. Zoo Wildl. Med. 26, 240–245.
Makita, K., Fevre, E.M., Waiswa, C., Eisler, M.C., Thrusfield, M., Welburn, S.C., 2011. Herd prevalence of bovine brucellosis and analysis of risk factors in cattle in urban and peri-urban areas of the Kampala economic zone, Uganda. BMC Vet. Res. 7, 60.
Marcotty, T., Matthys, F., Godfroid, J., Rigouts, L., Ameni, G., Gey van Pittius, N., Kazwala, R., Muma, J., van Helden, P., Walravens, K., de Klerk, L.M., Geoghegan, C., Mbotha, D., Otte, M., Amenu, K., Abu Samra, N., Botha, C., Ekron, M., Jenkins, A., Jori, F., Kriek, N., McCrindle, C., Michel, A., Morar, D., Roger, F., Thys, E., van den Bossche, P., 2009. Zoonotic tuberculosis and brucellosis in Africa: neglected zoonoses or minor public-health issues? The outcomes of a multi-disciplinary workshop. Ann. Trop. Med. Parasitol. 103, 401–411.
McDermott, J.J., Arimi, S.M., 2002. Brucellosis in sub-Saharan Africa: epidemiology, control and impact. Vet. Microbiol. 90, 111–134.
McDermott, J., Grace, D., Zinsstag, J., 2013. Economics of brucellosis impact and control in low-income countries. Rev. Sci. Tech. (Int. Off. Epizoot.) 32, 249–261.
Megersa, B., Biffa, D., Abunna, F., Regassa, A., Godfroid, J., Skjerve, E., 2011. Seroprevalence of brucellosis and its contribution to abortion in cattle, camel, and goat kept under pastoral management in Borana, Ethiopia. Trop. Anim. Health Prod. 43, 651–656.
Mkoma, S.L., Mjemah, I.C., 2011. Influence of meteorology on ambient air quality in Morogoro, Tanzania. Int. J. Environ. Sci. 1, 1107–1115.
Movek, D.S., 2008. Small Farmer Productivity Through Increased Access to Draught Power Opportunities. Consultancy Report Stakeholder Mapping in Morogoro Region.
Muma, J.B., Samui, K.L., Siamudaala, V.M., Oloya, J., Matop, G., Omer, M.K., Munyeme, M., Mubita, C., Skjerve, 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. Trop. Anim. Health Prod. 38, 195–206.
Pérez-Sancho, Marta, García-Seco, Teresa, Domínguez, Lucas, Álvarez, Julio, 2015. In: Manal, Mohammad, Baddour (Eds.), Control of Animal Brucellosis—The Most Effective Tool to Prevent Human Brucellosis, Updates on Brucellosis. In Tech..
R Core Team, 2016. R: A Language and Environment for Statistical Computing.
Randrianantoandro, T.N., Kono, H., Kubota, S., 2015. Knowledge and behavior in an animal disease outbreak—evidence from the item count technique in a case of African swine fever in Madagascar. Prev. Vet. Med. 118, 483–487.
Roth, F., Zinsstag, J., Orkhon, D., Chimed-Ochir, G., Hutton, G., Cosivi, O., Carrin, G., Otte, J., 2003. Human health benefits from livestock vaccination for brucellosis: case study. Bull. World Health Organ. 81, 867–876.
Sauret, J.M., Vilissova, N., 2002. Human brucellosis. J. Am. Board Fam. Pract. 15, 401–406.
Swai, E.S., Schoonman, L., 2009. Human brucellosis: seroprevalence and risk factors related to high risk occupational groups in Tanga Municipality, Tanzania. Zoonoses Publ. Health 56, 183–187.

Swai, E.S., Schoonman, L., 2010. The use of rose bengal plate test to asses cattle exposure to Brucella infection in traditional and smallholder dairy production systems of Tanga region of Tanzania. Vet. Med. Int. 2010.

Temba, P., 2012. Seroprevalence of Brucella Species Infections and Associated Risk Factors in Wildlife-Livestock Interface: A Case Study of Mikumi-Selous Ecosystem. M.Sc. Dissertation. Sokoine University of Agriculture, Tanzania.

Thrusfield, M., 2005. Veterinary Epidemiology, third edition. Blackwell Science ltd, Oxford, UK.

Tsuchiya, T., Hirai, Y., Ono, S., 2007. A study of the properties of the item count technique. Publ. Opin. Q. 71, 253–272.

Vanzini, V.R., Aguirre, N., Lugaresi, C.I., de Echaide, S.T., de Canavesio, V.G., Guglielmone, A.A., Marchesino, M.D., Nielsen, K., 1998. Evaluation of an indirect ELISA for the diagnosis of bovine brucellosis in milk and serum samples in dairy cattle in Argentina. Prev. Vet. Med. 36, 211–217.

Weinhäupl, I., Schöpf, K.C., Khaschabi, D., Kapaga, A.M., Msami, H.M., 2000. Investigations on the prevalence of bovine tuberculosis and brucellosis in dairy cattle in Dar es salaam Region and in zebu cattle in Lugoba area, Tanzania. Trop. Anim. Health Prod. 32, 147–154.

Wilcox, R.R., 1996. Statistics for the Social Sciences. Academic Press, San Diego, CA.