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A Quantitative Risk Assessment of Human Exposure to Brucellosis Through the Consumption of Contaminated Raw Cow Milk in Arusha, Tanzania

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Abstract

The study aimed at assessing the risk of exposure to brucellosis through the consumption of cow milk in the Arusha region, Tanzania. Primary data related to milk consumption was collected through a structured questionnaire from 400 cattle farmers. Data was directly coded and entered into IBM SPSS version 20 and analysed for frequencies and descriptive statistics. Stochastic Monte Carlo simulation in @ risk Software (Version 8.1) platform was used to estimate the risk of human exposure to brucellosis through the consumption of contaminated milk.

Results revealed that 96.5% of the population consumed milk in three (3) portions: morning, afternoon, and night. More than 70% of the people in the area consume 500ml-1000ml of milk daily. People in rural settings (71%) reported consuming raw milk, compared to 10% of people in urban setting.

The probability of getting infected with Brucella through the consumption of raw milk was estimated at 0.64 (95%CI 0.333-0.861). The model also predicted the number of people likely to get infected with Brucella in Arusha region in a one-year consumption period to be 1,084,358 (95%CI: 565,000-1458,000), out of 1,694,310 people following consumption of contaminated raw milk. The risk of exposure was estimated to be high when dairy cows were infected with Brucella at the farm and when the milk portions were consumed raw.

The risk of human exposure to the Brucella pathogen is high. To reduce the risk of human exposure, there is a need to create awareness about brucellosis in the study communities concerning how the disease is transmitted to humans, its associated effects, and the preventive and control measures. Further studies are required to assess the risk of exposure to brucellosis through other pathways such as the consumption of soft cheese and contact with cattle.

Keywords: *Brucellosis, quantitative risk assessment, raw milk consumption, Tanzania*

1.0 Introduction

Brucellosis is a neglected zoonotic disease of public health and economic significance in most developing countries (El-wahab *et al*., 2020). Clinical signs of brucellosis are not specific, causing difficulties in its clinical diagnosis (Ducrotoy *et al.*, 2017). Abortion cases are prevalent manifestations of bovine brucellosis, which is also common in other reproduction-related infections such as leptospirosis, listeriosis, Q fever, bovine viral diarrhoea, trichomoniasis, mycotic abortion and neosporosis (Dereje *et al*., 2018). In humans, the disease presents as an acute to chronic illness characterised by intermittent fever, generalised and influenza-like syndrome (Franc *et al*., 2018). Other constitutional symptoms include joint pains, fatigue, and muscle ache that vary with the stage of infection and body system affected (Bodenham *et al*., 2020; Chota *et al*., 2016; Muturi *et al*., 2018), loss of appetite, muscular pain, lumbar pain, weight loss, hepatomegaly, splenomegaly and arthritis (Kunda *et al*., 2007).

Management-related factors such as the source of replacement stock, grazing strategy, breeding system, interaction with wildlife and herd size are significant risk factors for bovine brucellosis (Muma et al., 2007). Raw milk consumption, unpastunpasteurised products and assisting parturition without protective attire, direct contacts with aborted fetuses and slaughter practices are high-risk practices for this zoonosis (Akakpo *et al.*, 2010; Musallam *et al.,* 2019).

The quality of raw milk is important for humans and its technological processing on products, but milk may contain pathogenic microorganisms that can seriously affect consumers' health (Hanuš *et al*., 2021). Due to the shedding of the brucellae in milk, Brucella-contaminated raw milk and unpasteurised products are the most important vehicles of human infection and a considerable public health risk even in non-endemic countries (Jansen *et al*., 2020). Most *Brucella* species can survive in fresh milk for up to 5 days at 4°C and up to 9 days at − 20°C (Bayramoglu *et al.,* 2019). Studies conducted in Kenya, which is near Arusha, discovered that the prevalence of the pathogen in raw milk at the animal level, considering samples from individual animals, was 2.4% (95% confidence interval (CI) 1.1-4.5) (Wainaina *et al*., 2020).

The brucellosis prevalence in cow milk in Tanzania was estimated to be around 8% (95% CI 6.5–10.2) (Alonso *et al*., 2016). In countries like Syria, Iraq, Tanzania, and Uganda, brucellosis still spreads as an endemic zoonotic disease without adequate control approaches in dairy animals, making a realistic prevalence estimation difficult. According to studies conducted in endemic regions, there is a high prevalence of *Brucella* species contamination in raw milk (16.97%) compared with cheese (7.10%) (Dadar *et al*., 2020).

Bovine brucellosis has been reported in all districts of the Arusha region for so many years (Karimuribo *et al.,* 2007). In most rural areas in Arusha, there are pastoral societies whose diet entirely depends on milk and meat. In these communities, it is reported that, to a large extent, milk is consumed raw (Njarui *et al.*, 2011). This consumption behaviour is likely to increase the risk of acquiring various foodborne zoonotic diseases, including brucellosis. Previous studies have reported frequent consumption of raw milk in the Maasai community (Gibney & Burstyn, 1980; Gidel *et al.*, 1976; Melubo, 2020). It has been reported that raw cow milk is important in brucellosis transmission to humans (Akakpo *et al.*, 2010; Bouley *et al.*, 2012; Muma *et al.*, 2013; Muturi *et al.*, 2018), especially in the areas where the disease is endemic in animals like Tanzania (Karimuribo *et al.*, 2007). Currently, there is no information about the risk of people being exposed to Brucellosis in Tanzania through the consumption of raw cow milk. Therefore, this study aimed at quantitatively assessing the risk of exposure to Brucella species through the consumption of raw cow milk in Tanzania.

2.0 Materials and Methods

2.1 Study Design

This was a quantitative risk modelling study based on the Codex Alimentarius Commission (CAC) risk analysis framework, which involves four distinct steps including hazard identification, hazard characterisation, exposure assessment and risk characterisation. The study utilised data from the consumption survey and secondary data from the literature search to come up with input parameters to feed into the model. A Quantitative Risk Assessment Model (QRA) was developed starting from the farm to exposure of humans to Brucella at the point of consumption following the conceptual model pathways described in Figure 1.

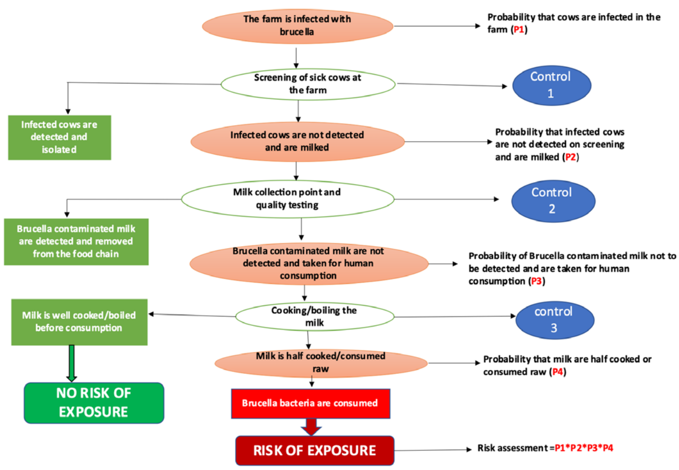

Figure 1: *The Conceptual Model Pathways from the farm to Folk (consumption)*

2.2 Study Area for the Consumption Survey

The study was conducted in two districts (Monduli and Longido) of Arusha region in Tanzania, as shown in Figure 2. The region was selected due to the high number of cattle in Tanzania, which is estimated at 1,373,839 (Livestock Population Census of 2014). The area is also estimated to have a human population of approximately 1.7 million, according to the population census of 2012 (Levira & Todd, 2017). More than 80% of the land in Arusha region is occupied by the Maasai community, which is famous for keeping livestock. The Maasai community is also known for maintaining the traditional culture. Their cultural food is mainly animal blood, meat and milk (Melubo, 2020). Also, the area is the tourism hub in Tanzania because it is surrounded by wildlife habitats such as national parks, game reserves, and conservation areas, all potential reservoirs for the disease. The pastoral societies in this region are known for consuming raw milk, despite brucellosis disease being reported in the area for many years.

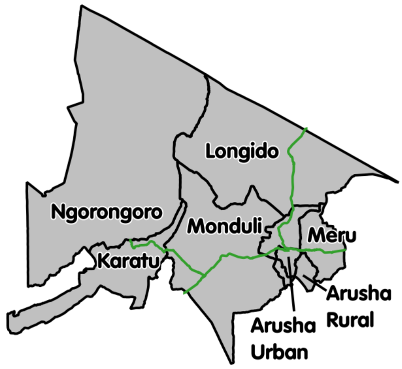

Figure 2: *A Map of Arusha Region, Tanzania* (Source Google map)

2.3 Instruments for Data Collection

A pretested structured questionnaire was used to collect primary data. Secondary data was collected using a checklist guided by quantitative risk assessment model questions or parameters in line with the Codex Alimentarius Commission food safety risk assessment framework.

2.3.1 Secondary Data

The literature search was conducted on electronic databases including Google Scholar, PubMed and Mendeley. The grey literature included reports from government institutions and non-governmental organisations obtained online using a Google Search Engine. Key terms included 'Brucellosis', 'raw milk consumption', 'quantitative risk assessment', 'consumption patterns', 'serving portions', and 'retail contamination'.

2.3.2 Primary Data

A structured household questionnaire addressed the knowledge gap about milk consumption patterns. Four hundred (400) samples were conveniently sampled from two districts of Arusha, namely; Monduli (176 samples) and Longido (224 samples). The respondents were categorised into two groups, those who stay in rural and urban areas.

The sample size of 400 was based on a statistical calculation by using the formula:

$$n = Z^2pq / E^2$$

Where: n= Required sample size; Z is Z-score from the Z table; E is the desired level of precision (margin of error); P is the estimated proportion of the population Q=1-p, and p=0.5. Assuming the Z-score was 1.96 at a 95% confidence level and a precision of 5%, a sample size of 384 was estimated. Since the study intended to get samples from ten (10) streets (from three different towns) and ten (10) villages in each district, the sample size was increased to 400, in which 176 samples were collected from Monduli District and 224 from Longido District.

2.4 The Quantitative Risk Assessment Process

The quantitative risk assessment was conducted following the quantitative risk assessment steps guided by Miller *et al*., (1993):

a) State the question and the scope of the risk assessment.
b) Identify the hazard of interest.
c) Develop a scenario tree that outlines the pathway of expected events and all the failures which could occur, culminating in the occurrence of the identified hazard.
d) Label the scenario tree and assign units.
e) Gather and document evidence (Data collection and management).
f) Assign values to the branches of the scenario tree.
g) Perform the calculations to summarise the likelihood of the hazard occurring.
h) Consider risk management options.

2.4.1 Risk Question

What is the risk of human exposure to *brucellosis* through the consumption of brucella-contaminated raw cow milk in among people of Arusha region in Tanzania?

2.4.2 Scope of the Risk Assessment

The risk assessment was limited to assessing the risk of people being exposed to brucellosis through consuming brucella-contaminated raw cow milk in Arusha. The study did not characterise the prevalence of different brucella pathogens.

2.4.3 Scenario Trees

The risk assessment model was described using information that could answer the risk question, 'What is the risk of exposure to brucella for people in the Arusha region through consuming brucella-contaminated raw cow milk?' A total of four probabilities were considered through which milk consumers could be exposed to Brucella pathogens in the milk food chain. These are.

i. The probability that the cows are infected at the farm (Figure 3);
ii. The probability that infected cows are not detected on screening and are milked (Figure 4);
iii. The probability that Brucella contaminated milk is not detected on quality control testing at collection points and is taken for human consumption (Figure 5); and
iv. The probability that milk is consumed half-cooked or raw (Figure 6).

The probability of exposure to Brucella was obtained as a product of all the four above-mentioned probabilities.

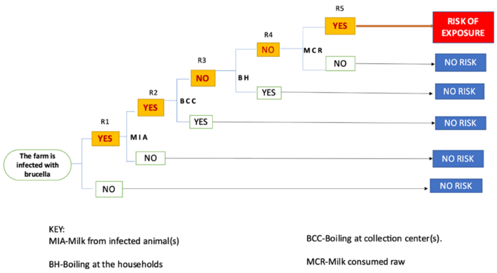

Figure 3: *The Scenario/risk Pathways for Exposure Assessment to Brucella through Milk Consumption*

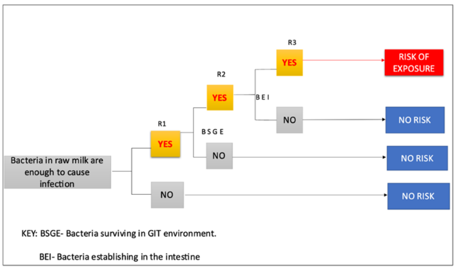

Figure 4: *The Scenario/risk Pathways for Brucella Hazard Characterisation*

2.5 Data Management and Analysis

Information from the questionnaire survey was directly coded and entered into *IBM SPSS® Statistics Version 21* for analysis using frequencies for nominal and ordinal variables and descriptive statistics for scale variables. The analysed were entered into *@ risk®* Software Version 8.1 embedded into a Microsoft Excel® spreadsheet containing four (4) attributes, the parameter number, Input parameter description, source of the information and their probability/proportion required. The obtained information was entered into a specific row and column with their minimum, most likely, and maximum probability values, or proportions with a respective probability distribution for continuous and discrete data. Risk Pert and Risk Uniform were the only function distribution used for continuous variables, while Risk Poisson, Risk Binomial and Risk Uniform were used for discrete variables.

2.5.1 Input Parameters for the Exposure Assessment

These calculations involved inputs associated with the cow milk chain from farm to folk (consumption). The input parameters used for this assessment were the amount of milk consumed per day per person, the amount (portions) of milk consumed in the study area in one year, the probability that the cows are infected at the farm, the probability that the infected cows are not detected on screening and are milked, probability of Brucella contaminated milk not detected during quality testing at the collection point and are taken for human consumption and the probability that milk is consumed raw or half-cooked.

2.5.2 Input Parameters for Hazard Characterisation

This involved establishing the dose-response relationship in the milk portion, considering the number of microorganisms consumed in various milk preparations with their respective effects. For this assessment, the input parameters used were the following: colony forming unit (cfu) per gram of the contaminated product, cfu surviving when milk is prepared done, cfu surviving when milk is prepared half done, cfu surviving when milk is prepared raw. Others were the probability of getting an infection from consuming prepared done milk, the probability of infection from consuming prepared half-done milk, the probability of infection from consuming prepared raw milk and the infectious dose at which half of the exposed population gets infected (infectious dose 50) for Brucella pathogens.

2.5.3 Input Parameters for Risk Characterisation

A Monte Carlo simulation was conducted to model the risk of exposure to Brucella species through milk consumption. In this case, the number of people exposed to Brucella and the number infected per year through milk consumption in the Arusha region were estimated. In this study, risk characterisation used the following input parameters; the total population of Arusha region, the population which consumes milk in Arusha, the proportion of a portion prepared raw, the proportion of a portion prepared half-done, the proportion of a portion prepared done, probability of exposure along the milk food chain. Other inputs are the number of people who can get infected from consuming prepared half-done milk, the number of people who can get infected from consuming prepared raw milk and the number of people infected who are likely to get ill in each population shown in Table 1.

Table 1: Probability Distribution of Input Parameters

Paramete	Input parameter descriptions	Source of informatio	Probability distribution	Level of confidenc
n1	Population size	Tanzania Bureau of Standard	RiskNormal (1694310)	1694310
n2	Milk consumed per day/person(L)	Survey	RiskPert (0.5;1;2)	0,619-1,652
n3	Amount (portions) of milk consumed in the study area in one year	Survey	RiskUniform (865792410;1298680000)	1080000000
p1	Probability that the cows are infected at the farm	(Chota et al., 2016; Id et al., 2021; Karimuribo et al., 2007; Li et al., 2021; Mathew et al., 2015; Ukita et al., 2021)	RiskPert (0.65;0.588;0.152)	0.4803
p2	Probability that the infected cows are not detected on screening and are milked	(Bricker et al., 2003; Fosgate et al., 2002)	RiskPert (0.28;0.11;0.068)	0.1313
p3	Probability of Brucella contaminated milk not detected during quality testing at collection point and are taken for human consumption	(Dadar et al., 2020; Waiswa et al., 2010)	Riskuniform (0.127;0.169)	0.14785
p4	Probability that milk is consumed raw or half-cooked	(Kai & Aotearoa, 2009; Kouamé-Sina et al., 2012)	Riskpert (0.516;0.42;0.349)	0.424
n4	Number of colonies forming unity (cfu) per gram contaminated product	(Hamdy & Amin, 2002; Kaden et al., 2018)	RiskPoisson (100;100000)	50050
n7	Number of colons forming unity surviving when milk is prepared done	(M. Corbel, 2006; Dadar et al., 2019; Davies & Casey, 1973; DUMUȚA-CODRE et al., 2010; Makita et al., 2012)	RiskPoisson (0)	0
n8	Number of colons forming unity surviving when milk is prepared half done	(Celebi et al., 2013; Méndez-González et al., 2011)	RiskUniform (0.35;0.5)	0.4
n9	Number of colons forming unity surviving when milk is prepared raw	(Makita et al., 2012; Saber Marouf et al., 2021)	RiskPoisson (1)	1
p5	Probability of getting infection from consuming prepared done milk	Product of n7*p10	RiskBinomial (0;0.6)	0
p6	Probability of getting infection from consuming prepared half-done milk	Product of p9*n8	RiskBinomial (1;0.4)	0.4
p7	Probability of getting infection from consuming raw milk	Product of p8*n9	RiskBinomial (0.64;1)	0.64
n10	The infectious dose at which the half of the exposed population gets infected (infectious dose 50)	(Teske et al., 2011)	RiskUniform (94;1885)	989.5
p8	Proportional of a portion of raw milk	Survey, (Njarui et al., 2011; Prakashbabu et al., 2020; Rock et al., 2016)	RiskPert (0.1;0.71;0.9)	0.64
p9	Proportional of a portion prepared half done	Survey	RiskUniform (0;2)	1
p10	Proportional of a portion prepared done	Survey (Pappas et al., 2006; Rock et al., 2016)	RiskPert (0.27;0.68;0.9)	0.64833
n12	Number of people who can get infected from consuming prepared half-done milk	(Mangen Otte et al., 2002; Pappas et al., 2006)	RiskUniform (34000;1459000)	677724
n13	Number of people who can get infected from consuming raw milk	(Ngasala et al., 2015; Njarui et al., 2011; Rock et al., 2016)	RiskUniform (565000;1458000)	1084358
n14	Number of people infected who are likely to get ill in a given population.	(Spink, 1954)	RiskUniform (96404;269147)	189762.7

3.0 Results

3.1 Demographic Information

Table 2 provides data on demographic information for the primary data. There were 305 males and 95 females in the study, representing 76.3% and 23.8%, respectively. Many respondents had secondary (44.8%) and primary (27%) levels of education. About 7.3% had never attended any level of education, but they could understand and respond to the questions in Kiswahili language.

Table 2: The Demographic Information of Respondents to the Questionnaire Survey (n=400)

Characteristics		Frequency	Percentage
Gender	Male	305	76.2
	Female	95	23.8
Level of education	None	29	7.25
	Primary	108	27
	secondary	179	44.75
	Tertiary	83	20.75
	Others	1	0.25
Number of people living in a household	1-2	54	13.5
	3-12	339	84.75
	13-18	7	1.75
Meals prepared with milk per day	1-2	13	3.25
	3-4	387	96.75
Source of milk	Own cattle	270	67.5
	Neighbor`s	33	8.3
	Local market	95	23.8
	others	2	0.4
Amount of milk consumed in a day/ person	<0.5	24	6
	0.5-1	291	72.8
	1-2	68	17
	>2	17	4.2

3.2 Milk Consumption Information

3.2.1 Number of Milk Meal Servings per day

The majority (96.5%) of the respondents had three serving portions of milk meals per day, which included, tea, lunch, and dinner. About 3.3% of the respondents had 1 to 2 meals made of milk in form of lunch and dinner per day as shown in Table 2.

3.2.2 Methods of Preparing Milk Meals

About 90% of the respondents in urban areas consumed their milk well-cooked (fully done), and 10% consumed raw milk in their households. Among the rural respondents, 27% consumed their milk meals well cooked, while 71 % consumed it raw. About 2% of the respondents partially cooked their milk before consumption. This was also referred to as half-cooked, which was interpreted as just warming milk as shown in Table 3.

Table 3: Milk Preparation Methods in Rural and Urban Setup.

MILK PREPARATION (%)				
		Raw	Half-done	Done
Area of residence	Town	10	0	90
	Village	71	2	27

3.2.3 Source of Milk

Table 2 also shows that most (67.5%) of the respondents consumed milk from their cattle, while 23.8% purchased it from local markets around their areas. About 8.3% of all the respondents got or purchased their milk from their neighbours, while 0.5% of all the respondents got their milk from other sources apart from the mentioned.

3.2.4 Size of Milk Portion Per Day

Most of the respondents (72%), consumed 0.5-1L of milk in a day, with 17% consuming 1-2L in a day. Those who consumed milk less than 0.5L were 6%, while 6% consumed greater than 2L per day.

3.3 Consumer Exposure Assessment

This section gives the route source of exposure to the bacteria of genus Brucella in individuals who consume milk. The pathway considered is from farm to consumption at households' level. This includes the results of the household survey questionnaire survey on serving portions, preparation methods and consumption patterns.

3.3.1 Case Definition

The population of people in Arusha region, which is used in this model, was estimated to be 1, 694,310 according to the national census of 2012 (Tanzania National Bureau of Statistics 2012). The study considered Arusha because it is the region with a large number of cattle in the country and the presence of Maasai people whose food entirely depends on milk and meat. The pathogen of interest is a bacterium of genus Brucella, while the food is milk, and a portion was 500ml, because it is the minimum amount consumed by most people. For a person to get ill, he must consume raw or partially cooked milk, which is contaminated with at least the minimum dose of the pathogen (Brucella). The consumption period considered in this study is one year (365 days), and then assessment of the number of people who will get ill in this consumption period of one year.

3.3.2 Serving Portions and Consumption Pattern

Milk consumption input parameters were entered into a Monte Carlo simulation model in @risk. The defined outputs were simulated at 1000 iterations and predicted the average milk consumption per person per day in Arusha found to be 1.08L (95%CI 0.62-1.68).

3.3.3 Kitchen Preparation

In this study kitchen preparation method meant how milk was prepared before consumption in the kitchen or at households, that is, whether raw, half-done, or well-done. The Monte Carlo simulation in @risk after simulation of kitchen preparation data at 5000 iterations predicted the estimation of the average proportion of milk portions prepared done to be 0.648 (95%CI 0.411-0.846). At the same time, the model predicted the average proportion of raw milk to be 0.64 (95%CI 0.333-0.861).

3.3.4 Probabilities for Risk Pathways

The conceptual model pathways of bacteria species from farm to consumption (**Figure 3**) show several probabilities that can lead people to be exposed to Brucella pathogens and their likely control points. The input parameters for the four probabilities are shown in **Table 1**. The parameters were entered into the Monte Carlo simulation model in @risk, and their outputs were defined accordingly and simulated at 100,000 iterations to predict the probability of each output. The results show that the probability that cows are infected at the farm is 0.48 (95% CI 0.29-0.62) as shown in **Figure 4**. The probability that infected cows are not detected on screening and are milked was predicted to be 0.30 (95% CI 0.27-0.32). The probability that Brucella contaminated milk was not detected and was taken for human consumption was predicted to be 0.15 (95% CI 0.13-0.17). The probability that milk was consumed raw was predicted to be 0.424 (95% 0.37-0.49).

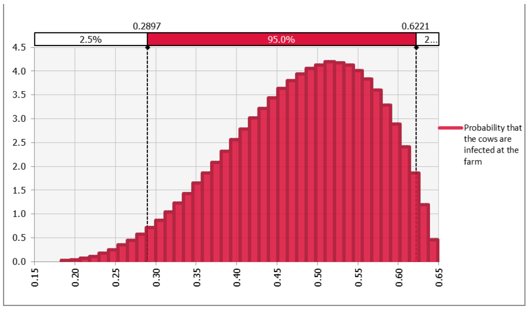

Figure 4: *The probability distribution that the cows are infected at the farm*

The model also estimated the risk of exposure from farm to consumption, which is the product of all possible exposure probabilities along the milk food chain. The product was defined as output and simulated once at 100,000 iterations. The probability of exposure was estimated to be 0.0089 (95% CI 0.0058-0.012), as shown in **Figure 5**.

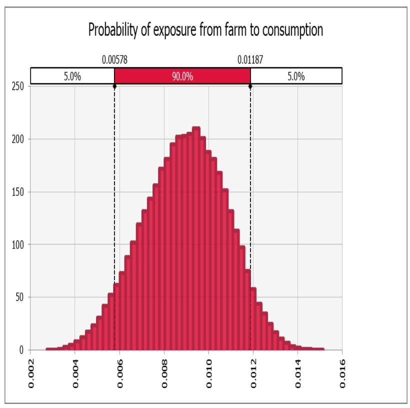

Figure 5: *Probability distribution of human exposure from farm to consumption*

3.4 Risk Characterisation

3.4.1 Probability of Infection

The probability of one getting infected with brucellosis following consumption of milk after various preparation was also calculated in the model based on the input parameters shown in **1**. The results were entered into the @Risk Software risk model estimated using Monte Carlo simulation, and the defined output was simulated at 100,000 iterations. After simulation, the model predicted the probability of getting an infection with Brucella through the consumption of prepared half-done milk, to be 0.4 (95%CI 0.32-0.48), as shown in **Figure 6.** The probability of one getting brucella infection following consumption of raw milk was predicted to be 0.64 (95% CI 0.54-0.73). The model estimated no probability of a person getting infected with Brucella through consuming milk prepared well done.

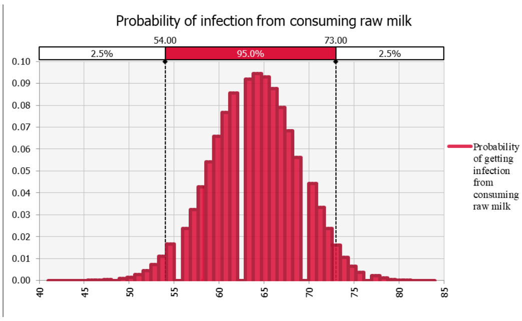

Figure 6: *Probability distribution of getting infection from consuming raw milk*

3.4.2 Infection and Illness

In this study, the infectious dose for Brucella humans was estimated to range from 10-100 microorganisms (De Figueiredo *et al.*, 2015; Grützke *et al.*, 2021; Kaden *et al.*, 2018). Since *Brucella melitensis* is among the species, which have been confirmed to cause infections in humans (Galińska *et al.*, 2013), the model assumed the infectious dose in which half of the exposed population will get ill (ID50) for *Brucella melitensis* from the human vaccine trial in which the minimum dose is 94cfu and the maximum dose considered to be 1885cfu (Teske *et al.*, 2011) to be considered as an infectious dose 50 for the Brucella species. After that, the input parameters were put in a Monte Carlo simulation model, and the outputs were defined. The infection and illness outputs were simulated at 100,000 iterations. The model predicted the number of people likely to get infected with Brucella Brucellan the Arusha region in a one-year consumption period of 677,678 (95%CI 33,000-1460,000) following consumption of prepared half-done milk, as shown in **7**. The number of people likely to get infections from consuming raw milk in one year was estimated to be 1,084,358 people (95%CI 565,000-1458,000), as shown in **8**.

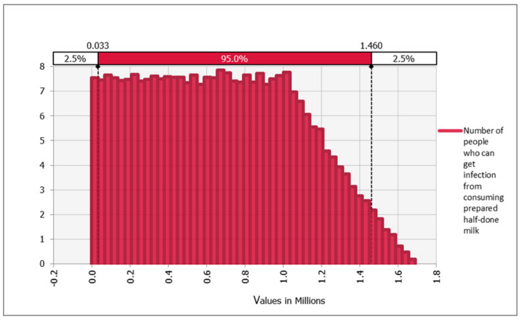

Figure 7: *Distribution of the number of people who can get infection from consuming prepared half-done milk*

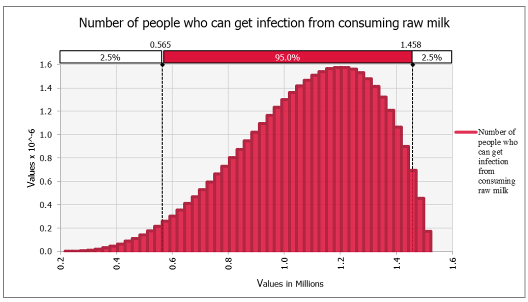

Figure 8: *Distribution of the number of people who can get infection from consuming prepared raw milk*

Spink (1954) reported that not all people who get Brucella infections are likely to develop and show clinical signs; instead, only 15-20% of infected people have that chance. All these possibilities were entered into the Monte Carlo simulation model in @risk, and the output was defined as the number of Brucella-infected people who are likely to get ill. This was simulated at 100,000 iterations. The model predicted the number of people who are likely to get ill after being infected with *Brucella* to be 189,760 (95%CI 96,696-269,804) as shown in **Figure 9.** The number of people who are infected with Brucella through consumption of different preparation; raw, half-done, and done milk portions together with the number of people who are likely to develop clinical signs are summarised in Figure 10.

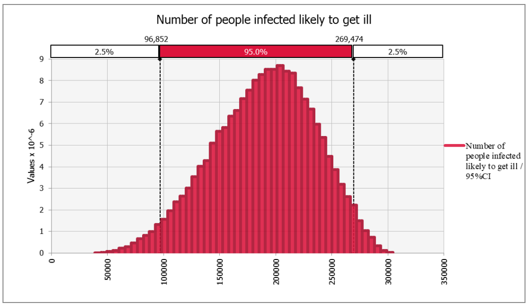

Figure 9: *The distribution of the number of people infected with Brucella pathogens who are likely to get ill*

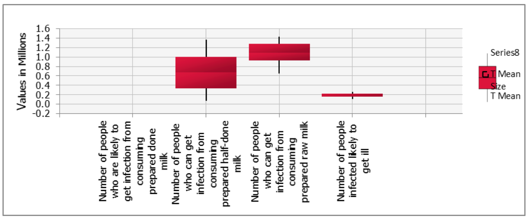

Figure 10: *Box and whisker plots of the number of people who can get infection through consumption of various preparation of milk*

3.4.3 Sensitivity Analysis

The model also estimated the influence of each probability and preparation factor on the risk of exposure through a sensitivity analysis. The results on a Tornado graph in **Figure 11** show that 83% of the risk of exposure was contributed by the probability that cows were infected at the farm, 36% the contaminated milk was not detected and is taken for human consumption, 31% probability that milk was consumed raw, and 22% the probability that infected cows were not detected on the screening and are milked.

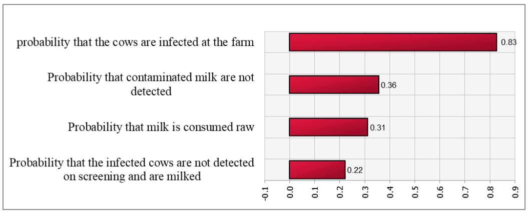

Figure 11: *A Tornado graph ranking the factors that facilitated the risk of exposure*

The Tornado graph shows a 100% chance of the consumers getting exposed to Brucella when milk is prepared raw, 3% when milk is prepared half-done, and no chance when milk is prepared well done as shown in **Figure 12**.

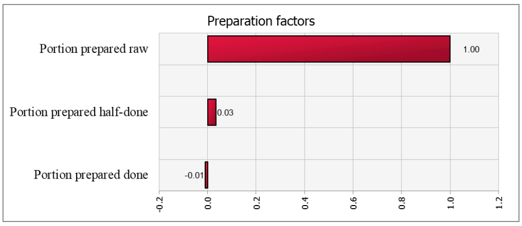

Figure 12: *The ranking of the preparation factors*

4.0 Discussion

This study was conducted to assess the risk of developing brucellosis through the consumption of brucella-contaminated raw milk in the Arusha region of Tanzania. The key questions were to find out the milk consumption scenario among people in the Arusha region and the risk of exposure to Brucella species through the consumption of raw milk.

The study observed that 6% consumed less than 500ml or 500g of milk a day while 4.2% consumed more than 2L or 2000g per day. The large population in Arusha, about 72.8%, consumed milk between 0.5-1L per day, whereas 17% of the population consumed between 1-2L per day per person. Following the simulation of the input parameters in a Monte Carlo simulation platform, the average milk consumption per day per person in the Arusha region was predicted to be 1.0834L (95% CI 0.618-1.6850). The value is slightly similar to that reported earlier by Njarui *et al*. (2011), which reported the average milk consumption per day per person in the Maasai community in Morogoro to range between 0.84-1.75L. This might be facilitated by the similar traditions of the Maasai people, whose diet depends much on meat and milk, as previous studies described it (Gibney & Burstyn, 1980; Gidel *et al.,* 1976; Melubo, 2020). The survey also showed that most people (99.5%) in the study area consumed milk daily, whereas the preparation methods varied depending on residency. This is contrary to the study done in Abidjan, Côte d'Ivoire by Kouamé-Sina *et al.* (2012), who reported that only 28% consumed milk every day. This difference might be caused by the affordability of milk in these areas. People who stay in town 90% consume prepared done milk (well-cooked/boiled) either during breakfast as tea, freshly boiled milk or mixed with other ingredients to make their food and 10% consume raw milk. According to the survey, people in the village settling 27% consumed their milk while well done, mostly during tea and traditional food. About 71 % consume raw milk in their households while 2% do partially cook their milk before consumption, which is referred to as half-done.

When all the input data about kitchen preparation were simulated by using a Monte Carlo simulation in @risk, it gave the estimation of the average proportion of milk portions that are prepared done to be 0.648 (95%CI 0.411-0.846) the proportion of the portions, which were prepared raw to be 0.64 (95%CI 0.333-0.861). This is in line with Kouamé-Sina *et al.* (2012), who reported that 51.6% of residents in urban and peri-urban consumed raw milk whereas 48.6% of residents in urban and peri-urban consumed boiled/pasteurised. Studies conducted in Kenya (which is near to Arusha) discovered that the prevalence of the pathogen in raw milk at the animal level (considering samples from individual animals) was 2.4% (95% confidence interval (CI) 1.1-4.5) (Wainaina *et al.*, 2020) but also it was reported that only 1 cfu/ml is enough to initiate the brucellosis infections (Bhankole, 2013). Therefore, there is a high risk of getting infections for the population who consumes more milk compared to the population who consumes less.

Different preparation of milk gives different implications on the transfer of diseases from the results obtained from Monte Carlo simulation. There was a high risk of exposure to Brucella infection, which was 0.64(95%CI 0.54-0.73) through consumption of prepared raw milk. These findings agree with Bhankole's (2013) report, which reported 0.63 as the risk of getting Brucella infection from consuming contaminated dairy products. This is because the contaminated Brucella bacteria in raw milk are active and have the potential to cause disease in comparison with other preparations. That is why the ingestion of fresh milk or other dairy products prepared from unheated/un-boiled milk is said to be the major source of Brucella infection in most communities all over the world, as it was described by Corbel *et al*. (2006), John *et al.* (2010), and by Massis *et al*., (2019). Other researchers described the consumption of raw milk as the leading risk factor for human brucellosis and are strongly conditioned by the geographical situation, which impacts the number of infected animals and the occurrence of Brucella contamination in dairy products (Dadar *et al*., 2019). Also, the model showed that the consumption of half-cooked/half-done milk also has a risk of getting brucellosis, which was described as the probability of getting infection to be 0.4 (95%CI 0.32-0.48). This might be because not all Brucella bacteria will die on the partial heating or boiling of the milk, making them have the potential for disease transmission. This agrees with the study conducted by Méndez-González *et al*. (2011), in which *Brucella melitensis* survived in goat milk in the combination of various temperatures and time. A similar observation was reported by Davies and Casey (1973) on *Brucella abortus* in which the viable cells count were visible after heating treatment of milk at 161-162°F (71.7°c) for 5 seconds. The model also assessed the risk of getting a disease for those who consumed prepared or well-cooked/boiled milk. It showed that there was no risk that a person can get infected with any Brucella species by consuming prepared well-done milk. This is because all bacteria are killed by heating or high-temperature pasteurisation. Corbel *et al*. (2006), reported that heating milk at a temperature between 80-85°c for several minutes destroys all Brucella bacteria. Also, a report from Davies and Casey (1973) on *Brucella abortus* in which after heat treatment of milk in 161-162°F (71.7°c) for 5 minutes, there was no observed viable *Brucella abortus* cell. Manyori *et al*. (2017), also demonstrated that the risk of developing salmonellosis in Zambia was low due to overcooking.

The factors that influence the risk of exposure from the farm to the time of consumption were ranked using a Tornado correlation coefficient, which showed that the high risk of exposure (83%) could be contributed to cows getting infected at the farm. This is in agreement with John *et al*. (2010), who reported on the cow's infection in the herd as an influential risk factor for human brucellosis. If the contaminated milk is not detected and is released for human consumption, there is a 36% chance of contributing to people's exposure to brucellosis. The risk of exposure at 31% can be influenced when milk is consumed raw, and there was a 22% chance of people being exposed to various Brucella species when the infected cows were not detected during screening and are milked at the farms. Also, on the preparation of portions, when someone consumed prepared raw milk, the chance of being exposed to various species of Brucella is as high as 100%. There was a 3% chance of exposure when consuming prepared half-done milk. When the milk portion is prepared well-done, there is no influence on the exposure to Brucella species. This is in line with Corbel (2006) and Davies and Casey (1973), who reported that high risk of exposure to Brucella when milk is consumed raw, reduces risk when partially boiled, and no risk when milk is well boiled before consumption

As it was previously described by Wainaina *et al.* (2020), that the prevalence of the pathogen (Brucella) in raw milk at the animal level (considering samples from individual animals) is 2.4% (95% confidence interval (CI) 1.1-4.5) but also it was reported by Bhankole (2013) that only 1 cfu/ml is enough to initiate the brucellosis infections. Basing on these findings, 71% of the population from the villages are at higher risk of getting the infection because they consume raw milk, while 10% of the town population is also at risk of getting brucellosis.

The conceptual model pathways of Brucella species from farm to consumption show four probabilities that could lead people to be exposed to Brucella species along the milk food chain from the farm to the table. These are the probability that cows are infected at the farm, the probability that infected cows are not detected on screening and are milked, the probability that Brucella contaminated milk is not detected and is taken for human consumption and the probability that milk is consumed raw.

Generally, the probability of exposure from farm to consumption is estimated by taking the product of all the probabilities. It is sometimes, referred to as a probability of release. Individual probability was taken as an output defined in a Monte Carlo simulation during simulation, and the given probabilities were obtained. The overall probability of exposure was calculated to be 0.0089. Among all the four probabilities, the highest probability observed on the probability that cows are infected at the farm, which is 0.48. This is similar to findings from Mathew *et al*. (2015), who reported herd prevalence of 48% (95% CI 41-55). This similarity might be attributed to the similar highland climatic conditions of Mbeya and Arusha regions. The probability that milk was consumed raw was 0.42, slightly similar to the report from Kai and Aotearoa (2009), who reported the probability of 0.35 that milk was consumed raw. The lowest probability observed on the probability that Brucella-contaminated milk is not detected and is taken for human consumption was 0.1478, while the probability that infected cows are not detected on screening was 0.296.

After running and integrating the results from hazard identification, hazard characterisation and consumer exposure assessment, the Monte Carlo simulation predicted the number of people who are likely to get infected by various species of Brucella through the consumption of milk at an average consumption of 1.08L per day per person on different milk preparations. The model predicted that through consumption of half-done milk, many people are likely to be infected with various Brucella species in a year in both town and village settings. This number is high and has an economic impact on individuals, households, and national levels. On the other hand, the model predicted that 1,084,358 people (95%CI 565,000-1458,000) out of 1,694,310 would get infected with various species of Brucella through the consumption of prepared raw milk. Then, in most cases, brucellosis goes asymptomatic, and it is reported that at least, 15 to 20% of people who get infected with Brucella Brucellaere are likely to develop the disease and show clinical signs (Spink, 1954). Therefore, the model also predicted the number of brucellae-infected people who were likely to get ill and show the clinical signs to be 189,760 (95%CI 96,696-269,804) out of 1,084,358 who were likely to get infections in one year. Most of these people who are affected, especially in east Africa, are reported from the low economic societies, as was reported in previous studies by Wainaina *et al.* (2020), and Pappas *et al*., (2006).

References

Akakpo AJ, Têko-Agbo A, Koné P, et al. The impact of Brucellosis on the economy and public health in Africa. *Compendium of Technical Items Presented to the OIE World Assembly of Delegates or to OIE Regional Commissions, 2009, file:///E:*, 71–84; 2010.

Alonso S, Dohoo I, Lindahl J, Verdugo, C, Akuku I, Grace D. *Prevalence of tuberculosis, brucellosis and trypanosomiasis in cattle in Tanzania : a systematic review and meta-analysis*. *17*(1).https://doi.org/10.1017/S146625231600013X; 2016.

Bayramoglu G, Ozalp VC, Oztekin M, Arica MY. Rapid and label-free detection of Brucella melitensis in milk and milk products using an aptasensor. *Talanta*, *200* (March), 263–271. https://doi.org/10.1016/j.talanta; 2019.03.048.

Bhankole A. *Brucellosis risk assessment*. 29–31; 2013.

Bodenham RF, Lukambagire AS, Ashford RT, Buza JJ, Rubach MP, Sakasaka P, Shirima GM, Swai ES. Prevalence and speciation of brucellosis in febrile patients from a pastoralist community of Tanzania.*Scientific Reports*, 1–11. https://doi.org/10.1038/s41598-020-62849-4; 2020.

Chota AC, Magwisha HB, Stella B, Bunuma EK, Shirima GM, Mugambi JM, Omwenga SG, Wesonga HO, Mbatha P, Gathogo S, Complex V, Muguga C. Prevalence of Brucellosis in livestock and incidences in humans in East Africa. *African Crop Science Journal*, *24*(Supplement s1), 45–52. https://doi.org/http://dx.doi.org/10.4314/acsj.v24i1.5S; 2016.

Corbel M. et al. Brucellosis in humans and animals. *World Health Organisation 2006 All*, 89. https://doi.org/10.2105/AJPH.30.3.299; 2006.

Dadar M, Fakhri Y, Shahali Y, Mousavi Khaneghah A. Contamination of milk and dairy products by Brucella species: A global systematic review and meta-analysis. *Food Research International*, *128*, 108775. https://doi.org/10.1016/j.foodres.2019.108775; 2020.

Dadar M, Shahali Y, Whatmore AM. Human brucellosis caused by raw dairy products: A review on the occurrence, major risk factors and prevention. *International Journal of Food Microbiology*, *292* (November 2018), 39–47.https://doi.org/10.1016/j.ijfoodmicro.2018.12.009; 2019.

Davies G, Casey A. The survival of Brucella abortus in milk and milk products. *The British Veterinary Journal*, *129*(4), 345–353. https://doi.org/10.1016/S0007-1935(17)36436-9; 1973.

De Figueiredo P, Ficht TA, Rice-Ficht, A, Rossetti CA, Adams LG. Pathogenesis and immunobiology of brucellosis: Review of Brucella-host interactions. *American Journal of Pathology*, *185*(6), 1505–1517. https://doi.org/10.1016/j.ajpath.2015.03.003; 2015.

Dereje T, Benti D, Feyisa B, Abiy G. Review of common causes of abortion in dairy cattle in Ethiopia. *Journal of Veterinary Medicine and Animal Health*, *10*(1), 1–13. https://doi.org/10.5897/jvmah2017.0639; 2018.

Ducrotoy M, Bertu WJ, Matope G, Cadmus S, Conde-Álvarez R, Gusi AM, Welburn S, Ocholi R, Blasco JM, Moriyón I. Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. *Acta Tropica*, *165*, 179–193. https://doi.org/10.1016/j.actatropica.2015.10.023; 2017.

El-wahab EWA, Hegazy Y, El-tras WF, Mikheal A, Kabapy AF. *A multiple risk model for brucellosis at the human-animal interface in Egypt*. https://doi.org/10.1111/tbed.13295; 2020.

Franc KA, Krecek RC, Häsler BN. *Brucellosis remains a neglected disease in the developing world : a call for interdisciplinary action*. 1–9. https://doi.org/10.1186/s12889-017-5016-y; 2018.

Galińska EM, Zagórski J, Em G, Brucellosis ZJ. 233-238 in Humans-Etiology, Diagnostics, Clinical Forms. *Annals of Agricultural and Environmental Medicine*, *20*(2), 233–238. www.aaem.pl; 2013.

Gibney MJ, Burstyn PG. Milk, serum cholesterol, and the Maasai. A hypothesis. *Atherosclerosis*, *35*(3), 339–343.https://doi.org/10.1016 /0021-9150(80)90131-8; 1980.

Gidel R, Albert JP, Le Mao G, Retif M. [Epidemiology of human and animal brucellosis in western Africa. The results of six studies in the Ivory Coast, Upper Volta, and Nigeria]. *Developments in biological standardisation*, *31*, 187–200; 1976.

Grützke J, Gwida M, Deneke C, Brendebach H, Projahn M, Schattschneider A, Hofreuter D, El-Ashker M, Malorny B, Al Dahouk S. Direct identification and molecular characterization of zoonotic hazards in raw milk by metagenomics using brucella as a model pathogen. *Microbial Genomics*. https://doi.org/10.1099/MGEN.0.000552; 2021.

Hanuš O, Kučera J, Samková E, Němečková I, Čítek J, Kopec, T, Falta D, Nejeschlebová H, Rysová L, Klimešová M, Elich O. Raw cow milk protein stability under natural and technological conditions of environment by analysis of variance. *Foods*, *10*(9). https://doi.org/10.3390/foods10092017; 2021.

Jansen W, Demars A, Nicaise C, Godfroid J, de Bolle X, Reboul A, Al Dahouk S. Shedding of Brucella melitensis happens through milk macrophages in the murine model of infection. *Scientific Reports*, *10*(1), 1–10. https://doi.org/10.1038/s41598-020-65760-0; 2020.

John K, Fitzpatrick J, French N, Kazwala, R, Kambarage D, Mfinanga GS, MacMillan A, Cleaveland S. Quantifying risk factors for human brucellosis in Rural Northern Tanzania. *PLoS ONE*, *5*(4). https://doi.org/10.1371/journal.pone.0009968; 2010.

Kaden R, Ferrari S, Jinnerot T, Lindberg M, Wahab T, Lavander M. Brucella abortus: Determination of survival times and evaluation of methods for detection in several matrices. *BMC Infectious Diseases*, *18*(1), 1–6. https://doi.org/10.1186/s12879-018-3134-5; 2018.

Kai TMK, Aotearoa A. Microbiological Risk Assessment of Raw Cow Milk. *Fsanz*, *December*; 2009.

Karimuribo ED, Ngowi HA, Swai ES, Kambarage DM. Prevalence of brucellosis in crossbred and indigenous cattle in Tanzania. *Livestock Research for Rural Development*; 2007.

Kouamé-Sina SM, Makita K, Costard S, Grace D, Dadié A, Dje M, Bonfoh B. Hazard identification and exposure assessment for bacterial risk assessment of informally marketed milk in Abidjan, Côte d'Ivoire. *Food and Nutrition Bulletin*, *33*(4), 223–234. https://doi.org/10.1177/156482651203300402; 2012.

Kunda J, Fitzpatrick J, Kazwala R, French NP, Shirima G, MacMillan A, Kambarage D, Bronsvoort M, Cleaveland S. Health-seeking behaviour of human brucellosis cases in rural Tanzania. *BMC Public Health*, *7*. https://doi.org/10.1186/1471-2458-7-315; 2007.

Levira F, Todd G. Urban Health in Tanzania: Questioning the Urban Advantage. *Journal of Urban Health*, *94*(3), 437–449. https://doi.org/10.1007/s11524-017-0137-2; 2017.

Manyori CI, Mumba C, Muma JB, Mwale MM, Munyeme M, Bwanga EK, Häsler B, Rich KM, Skjerve E. Quantitative risk assessment of developing salmonellosis through consumption of beef in Lusaka Province, Zambia. *Food Control*. https://doi.org/10.1016/j.food cont.2016.10.027; 2017.

Massis F De, Zilli K, Di G, Id D, Nuvoloni R, Pelini S, Sacchini L, Alterio ND, Giannatale E Di. *Distribution of Brucella field strains isolated from livestock , wildlife populations , and humans in Italy from 2007 to 2015*. 1–16; 2019.

Mathew C, Stokstad M, Johansen TB, Klevar S, Mdegela RH, Mwamengele G, Michel P, Escobar L, Fretin D, Godfroid J. First isolation , identification, phenotypic and genotypic characterisation of Brucella abortus biovar 3 from dairy cattle in Tanzania. *BMC Veterinary Research*, 1–9.https://doi.org/10.1186/s12917-015 -0476-8; 2015.

Melubo K. *Why are wildlife on the Maasai doorsteps ? Insights from the Maasai of Tanzania*. https://doi.org/10.1177/1177180120947823; 2020.

Méndez-González KY, Hernández-Castro R, Carrillo-Casas EM, Monroy JF, López-Merino A, Suárez-Güemes F. Brucella melitensis survival during manufacture of ripened goat cheese at two temperatures. *Foodborne Pathogens and Disease*, *8*(12), 1257–1261. https://doi.org/10.1089/fpd.2011.0887; 2011.

Muma JB, Samui KL, Oloya J. *Risk factors for brucellosis in indigenous cattle reared in livestock – wildlife interface areas of Zambia*. *80*, 306–317. https://doi.org/10.1016/j.prevetmed.2007.03.003; 2007.

Musallam I, Prisca A, Yempabou D, Ngong CC, et al. Acta Tropica Brucellosis in dairy herds : A public health concern in the milk supply chains of West and Central Africa. *Acta Tropica*, *197*(May), 105042.https://doi.org/10.1016/j.actatropica. 2019.105042; 2019.

Muturi M, Bitek A, Mwatondo A, Osoro E, et al. Risk factors for human brucellosis among a pastoralist community in South - West. *BMC Research Notes*, 1–6. https://doi.org/10.1186/s13104-018-3961-x; 2018.

Njarui D, Gatheru M, Wambua J, Hospital LC, Nguluu S. *Consumption Patterns and Preference of Milk and Milk Products among Rural and Urban Consumers in Semi-Arid Kenya This article was downloaded by : On : 31 May 2011 Access details : Access Details : Free Access Publisher Routledge Ecology of Food and Nutrit*. *June 2016*. https://doi.org/10.1080/03670244.2011.568908; 2011.

Pappas G, Papadimitriou P, Akritidis N, Christou L, Tsianos EV. *The new global map of human brucellosis*. *6*(February), 91–99; 2006.

Spink W. *Family studies on Brucellosis*. 127–140; 1954.

Teske SS, Huang Y, Tamrakar SB, Bartrand TA, Weir MH, Haas CN. Animal and Human Dose-Response Models for Brucella Species. *Risk Analysis*. https://doi.org/10.1111/j.1539-6924.2011.01602.x; 2011.

Wainaina M, Aboge GO, Omwenga I, Ngaywa C, Ngwili N, Kiara H, Wamwere-Njoroge G, Bett B. Detection of Brucella spp. in raw milk from various livestock species raised under pastoral production systems in Isiolo and Marsabit Counties, northern Kenya. *Tropical Animal Health and Production*, *52*(6), 3537–3544. https://doi.org/10.1007/s11250-020-02389-1; 2020.