

**AWARENESS OF HEALTH RISKS AS A RESULT OF CONSUMPTION OF RAW
MILK IN ARUSHA CITY AND MERU DISTRICT, TANZANIA**

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**A DISSERTATION SUBMITTED IN PARTIAL FULFILMENT OF THE
REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE IN PUBLIC
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ABSTRACT

A cross-sectional study was conducted in Arusha city and Meru District Council, Tanzania, from October to December 2012, to assess the awareness of smallholder dairy farmers, milk vendors and milk retailers to milk quality and health risks associated with raw milk consumption. A total of 105 respondents were interviewed and milk samples collected for laboratory analysis. Laboratory assessment included physical and microbial quality using standard procedures and antibiotic residues using Delvotest. Questionnaire results indicated high level of awareness that consumption of raw milk could predispose consumers to health hazards. Among the health problems specified included tuberculosis and brucellosis. Nevertheless, majority of respondents used raw unboiled soured milk as fermented milk for sale. Plastic containers were commonly used for storage and transportation of milk. Milk pH below 6.6 was 35.2% and specific gravity below 1.028 g/ml was 13.3%. Mean Total Viable Count (TVC) of milk from vendors was higher than that from retailers and smallholder dairy farmers. Generally, 64.8% of milk samples assessed had higher TVC than the maximum recommended level of 2.0×10^5 cfu/ml (East Africa Community standards). Commonly isolated bacteria were *Staphylococcus aureus*, *E.coli*, *Pseudomonas* spp. and *Corynebacterium* spp. All smallholder dairy farmers were aware of drug residues in milk and 97.1% complied with drug withdrawal periods. This possibly led to all milk samples analysed to be free from antibiotic residues. It is concluded that the level of awareness to milk quality is high, although practices associated with milking and post harvest handling practices along the value chain predisposes milk to microbial contamination. It is therefore recommended that public education should be given to all stakeholders in dairy industry on milking and post harvest handling of milk to minimize the likely losses due to rejection of spoiled milk and milk-borne dangers which may occur due to consumption of contaminated milk.

DECLARATION

I, JUMA NGASALA BUKUKU, do hereby declare to the Senate of Sokoine University of Agriculture that this dissertation is my own original work done within the period of registration and that it has neither been submitted nor being concurrently submitted in any other institution.

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DEDICATION

This work is dedicated to my wife Alice Ezekiel Mapuga, and our children Richard, Jeremiah and Emmanuel for their patience during the period of my study.

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LIST ABBREVIATIONS AND SYMBOLS

Abbreviation	Descriptive meaning
%	Percent
&	and
<	Less than
>	Greater than
µg/ml	microgram per millitre
®	Registered trade mark
AIDS	Acquired immunodeficiency syndrome
CAC	Codex Alimentarius Commission
CBPP	Contagious bovine pleuropneumonia
CI	Confidence interval
CFU/ml	Colony forming unit per millitre
EAC	East African Community
ECF	East Coast Fever
et al	and others
FAO	Food and Agriculture Organization for the United Nations
FMD	Foot and Mouth Disease
g	gram
g/ml	gram per millitre
HIV	Human immune-deficiency virus
IDF	International Dairy Federation
ISO	International Organization for Standardization
MRL	Maximum residual limit

°C	Degrees Celsius
pH	Hydrogen ion concentration
RVF	Rift Valley Fever
R	Retailer
SDF	Smallholder dairy farmers
SG	Specific gravity
StdEv	Standard deviation
Spp	Species
SUA	Sokoine University of Agriculture
SV	Street vendor
TSHZ	Tanzania short horned Zebu
UHT	Ultra High Temperature
UK	United Kingdom
VIC	Veterinary investigation centre
TAMPA	Tanzania Milk Processors Association
TAMPRODA	Tanzania Milk Producers Association
TB	Tuberculosis
TBS	Tanzania Bureau of Standards
TDB	Tanzania Dairy Board
TFDA	Tanzania Food and Drugs Authority
WHO	World Health Organization

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background Information

Raw milk is a yellowish-white non-transparent fluid secreted by animals to feed their young (Afzal *et al.*, 2011; Pandey and Voskuil, 2011). For mammals milk is meant to be the first and the only food for the offspring as is almost a complete food (Pandey and Voskuil, 2011). Almost 87% of milk is composed of water and the remaining part comprises total solids contained in a balanced form and digestible elements for building and maintaining the human and animal body. Other milk ingredients include immunoglobulins which protect the newly born against a number of diseases (Pandey and Voskuil, 2011). The solid content of milk is a sum of fat, protein, lactose and minerals (Afzal *et al.*, 2011). Milk contains various properties, which make it easy to convert into different milk products or to use it as an ingredient for other food items. Through milk properties various human cultures have their own traditional ways of using milk and preparing different milk products (Pandey and Voskuil, 2011). However milk compositions have wide variation within species of animals, breed (Pandey and Voskuil, 2011). There also variation in milk composition within species, between breeds and between individual animal in a breed. The composition may also change from day to day depending on the feed and climate and during milking where the first milk differs from the last drops (Pandey and Voskuil, 2011).

Fresh milk contains pleasant soft and sweet taste and carries no bad smell (Pandey and Voskuil, 2011). The off flavours and or odours in milk may be caused by strong flavoured feedstuff like poor quality silage, strong smelling nearby plants like the wild onion or garlic, cow-barn flavour from dung. Others potential sources of off flavours in milk include; rancid flavours- which are caused by excessive agitation of milk during collection

and/ or transportation, high acidity flavours, oxidized flavours, from contact with container or exposure to sunlight and flavours from the use of chlorine, fly sprays or medications (Pandey and Voskuil, 2011). Adulteration by addition of water or other solids to milk may also change the milk flavour by introducing chemical and microbial health contaminants (Omore *et al.*, 2002).

Indeed, milk adulteration is the potential source of milk contaminant which may have direct effect to milk through shortening shelf life. However, adulterated milk has reduced nutritional and processing quality, palatability and market value (Omore *et al.*, 2002). The common material added for milk adulteration are water, starch solutions, industrial alkalis, and nitrite which may facilitate deterioration of the quality of end products, losses due to condemnation and a risk to consumers' safety (Mansour *et al.*, 2012). However, the most common form of milk adulteration has been adding water by unscrupulous and unfaithful farm workers, vendors or farm households (Afzal *et al.*, 2011). The added water may be contaminated with faeces, microorganisms, harmful chemicals and poisonous substances which ultimately contaminate the milk (Afzal *et al.*, 2011).

Good quality milk has normal chemical composition, completely free from harmful bacteria and harmful toxic substances, free from sediments and extraneous substances, which have lower degree of titratable acidity, of good flavour, adequate in preserving quality, and low in bacterial counts (Hossain *et al.*, 2011). High-quality milk apart from containing low bacterial count also contains a low number of somatic cells and is free of human pathogens and antibiotic residues (Oliver *et al.*, 2009).

When from a healthy animal contains only few bacteria (normal flora), but because of its high water activity, moderate pH 6.4 - 6.8 and adequate supply of nutrient makes it an

excellent medium for microbial growth (Omore *et al.*, 2005), and good carrier for transmission of diseases to human (Kivaria *et al.*, 2006). Microbial contamination in raw milk may originate from primary sources like infected animal, dirty udder and teats and secondary sources like, the milker's hands, milking utensils, soils, water, feeds or air (Mohamed and Alhagaz, 2011). The most frequent cause of high bacteria count is poor cleaning of milking systems (Mohamed and Alhagaz, 2011). Cows with mastitis (streptococcal and coliform), soiled cows and failure to cool milk rapidly to $<4.48^{\circ}\text{C}$ after milking can also contribute to high bacteria count in raw milk (Oliver *et al.*, 2009).

Some of the microbial contaminants are responsible for milk spoilage while others are pathogenic with potential health effects to cause milk-borne diseases (Kivaria *et al.*, 2006). Pathogenic bacteria contaminants pose serious threat to human health, and constitute to about 90% of all dairy related diseases (Donkor *et al.*, 2007a). The common raw milk pathogenic bacteria contaminants include: *Brucella abortus*, *Mycobacterium bovis*, *Campylobacter* spp., *Coxiella burnetii*, *Leptospira* spp., *Listeria monocytogene*, *Yersinia enterocolytica*, Shiga toxin producing *Escherichia coli*, *Staphylococcus aureus*, *Salmonella* spp., and *Clostridium* spp. (Koo, 2008).

Apart from raw milk being potential carrier of pathogenic microorganisms, it can also cause health risk of the consumers due to antimicrobial residues (Kivaria *et al.*, 2006; Nonga *et al.*, 2009). Antibiotic residues are small amounts of drugs or their active metabolites which remain in milk after treating cows (Syt, 2008). Problems associated with antibiotic residues in milk include the risk of allergic reactions after consumption by penicillin-sensitized persons, increased resistance of pathogens towards antibiotics, and inhibition of bacterial starter cultures used in dairy production (Kurwijila *et al.*, 2006). The concerns arise mainly from the possibility that antibiotic-resistant bacteria may be

transferred from animals to humans, through contact, contaminated environment or milk (Syit, 2008). The presence of residues may be a result of failure to observe the mandatory withdrawal periods, illegal or extra-label use of drugs and incorrect dosage levels (Syit, 2008).

To protect the public against milk-borne infections it is important to screen milk which is informally taken to the market. Milk contamination can be reduced through effective good hygienic practices from farm level to the final consumer (CAC, 2004). The lack of awareness of milk-borne infections in many developing countries and consumption of raw milk predispose farmers, farm workers and their family members and consumers at risk of getting infections (Mosalagae *et al.*, 2011).

In Tanzania, about 80-90% of households in urban centres purchase raw milk from street vendors or via home delivery with little control of microbial contamination (Msangi, 2006; Kurwijila *et al.*, 2009). The milk quality assessment is neither done after milk production at the households nor is it done before selling to consumers (Msangi, 2006). This poses high health risks since cows may have zoonotic diseases and may be undergoing treatment with antibiotic drugs. A study by Kurwijila *et al.* (2009) reported lack of awareness of the farmers, vendors and retailer as the reason for not assessing milk quality. As far as legislation is concerned, there are no existing regulations in Tanzania, which control the business of raw milk (Msangi, 2006).

Furthermore, raw milk business in Tanzania is on the increase in most of the major cities and towns (Msangi, 2006; Kurwijila *et al.*, 2009). However, over 90% of the consumed raw milk in urban areas is sold through the vending market by direct sales and small vendors (Kurwijila *et al.*, 2009); with questionable quality control (Msangi, 2006).

Nevertheless, limited studies (Swai and Schoonman, 2011; Kivaria *et al.*, 2006; Kurwijila, and Henriksen, 1998) have been conducted on milk quality in terms of levels of milk contamination, antimicrobial residuals and milk adulterations in most of the urban areas including Arusha city where the milk business is booming (Arusha city is the headquarter of the East African Community (EAC) and destination of a large number of tourists from all over the world. Outbreak of any serious disease through milk or other sources can jeopardize tourism industry in the country. It was therefore the aim of this study to investigate the quality of raw milk in Arusha city and Meru District with a view of controlling pathogenic microorganisms found in milk in the study area.

1.2 Objectives

1.2.1 Main objective

The overall objective of this study was to determine quality of raw milk and assess the awareness of some dairy stakeholders of health risks associated with consumption of raw milk in Arusha city and Meru District Council.

1.2.2 Specific objectives

- i) To assess levels of awareness of smallholder dairy farmers, milk retailers and milk vendors on milk quality and possible hazards associated with consumption of raw milk.
- ii) To determine microbial quality of milk produced and sold in the study areas
- iii) To determine physical quality of milk produced and sold in the study areas
- iv) To determine levels of antimicrobial drugs residues in raw milk from cattle in the study area

1.3 Research Questions

- i) Are smallholders dairy farmers, milk vendors and retailers aware of the health risk associated with unsafe milk production?
- ii) What are the physical and microbiological characteristics of raw milk?
- iii) Do cattle keepers observe withdrawal period from application of veterinary drug?
- iv) Are there antimicrobial residues in milk from smallholders dairy farmers?

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 General Overview of the Dairy Industry in Tanzania

The livestock industry in Tanzania largely depends on cattle which are estimated to be at 18.8 million (Kurwijila *et al.*, 2006; Njombe *et al.*, 2009) and are the main livestock species used for milk and meat production. The cattle industry in Tanzania is dominated by indigenous animals, the Tanzania short horned Zebu (TSHZ) which constitute (98%) of the total cattle population (Swai and Karimuribo, 2011). The TSHZ are mostly kept by pastoralists and agro- pastoralists. The TSHZ are characterised by small body size and poor milk production, a lactating cow produces less than 3 litres of milk per day (Swai and Karimuribo, 2011). The animals rarely get veterinary attention and are always exposed to animal diseases of different kinds (Kurwijila *et al.*, 2009). However, dairy cattle or improved dairy breed found in the country are crosses of Friesian, Jersey and Ayrshire (Njombe *et al.*, 2009). They constitute less than 2% of the cattle population in Tanzania and are mostly kept in urban and peri-urban areas under intensive system (zero grazing). The annual milk production in Tanzania is estimated at 1150 million litres (Kurwijila *et al.*, 2009), about 70% of the annual produce comes from the traditional sector (indigenous cows), where as the improved dairy cattle contributes about 30% (Kurwijila *et al.*, 2009; Njombe *et al.*, 2009; Swai and Karimuribo, 2011). Seventy two percent of annual milk production is consumed at farm level and 28% is marketed (Kurwijila *et al.*, 2009).

Studies show that Arusha region have 110 000 dairy cattle and the second region after Kilimanjaro region with large number of dairy cattle in the country (Swai and Karimuribo, 2011). Arusha city and Arumeru are the places having high number of dairy cattle kept by smallholder farmers in the region. The animal produces on average of 8 litres of milk per

day. However, because of tropical conditions and poor animal husbandry, dairy cattle in smallholder farms suffer a lot of infectious and non infectious diseases. Tick- and tsetse - borne diseases are the common cattle diseases which contribute substantial losses to farmers. However, mastitis, both clinical and subclinical forms has also been significantly causing losses to farmers (Karimuribo *et al.*, 2005). These necessitate farmers to abuse veterinary drugs and sometimes even without observing the drug withdrawal periods. This may pose danger to consumers of food emanating from animals like milk and meat.

Milk produced by smallholder dairy farmers is mainly used for business that aims at making profit. It is estimated that 90.5% of the milk from smallholder is sold through informal market (direct sales, hawkers and small vendors). The formal market channel represents only 9.5% of the marketed milk (Kurwijila *et al.*, 2009). Informal milk marketing is mainly practiced by vendors or hawkers who collect milk from their own cattle and other households to sell directly to retailers (kiosks, restaurants) and consumers. This marketing has the positive advantage that it provides employment and income to many milk hawkers who are involved in the milk chain and making their living. However, the milk may pose health risks to consumers since it is not inspected for quality and safety. This may be due poor organization of dairy sector in the country and lack of regulation enforcement by Tanzania Food and Drug Authority (TFDA) on routine milk quality assessment which gives a loophole to farmers, vendors and retailers to sell uninspected milk.

2.2 Safety and Quality of Raw Milk

The safety of raw milk like any other food is of a worldwide concern where various efforts are directed, since it is associated with food-borne diseases (Grace and Baker, 2009). Food quality is the assurance that food is acceptable for human consumption according to

intended use (CAC, 2004). Food quality includes all other attributes that influence a product's value to the consumer, this includes positive attributes such as the origin, colour, flavour, texture and processing method of the food and negative attributes such as spoilage, contamination with filth, discoloration and off-odours (FAO and WHO, 2002). On the other hand food safety describes handling, preparation, and storage of food in a ways that prevent food-borne illness. It includes a number of routines that should be followed to avoid potentially severe health hazards (FAO and WHO, 2002). Food safety refers to all those hazards, whether chronic or acute, that may make food injurious to the health of the consumer (FAO and WHO, 2002). Consumers strive to get clean, wholesome and nutritious food that is produced in sound and sanitary way and free from pathogens. To achieve this consumer need, production of quality milk is required (Khan *et al.*, 2008). Quality milk therefore is milk which is free from pathogenic bacteria and harmful toxic substances such as antibiotics/antimicrobials and other chemical residues like acaricides, free from sediment and extraneous substances, of good flavour, with normal composition and acidity, adequate in keeping quality and low in bacterial counts (Khan *et al.*, 2008). Milk safety and quality is the combination of physical, chemical and microbiological qualities of milk. To achieve food safety and quality is important to adherence to hygienic practices throughout the chain of food production (CAC, 2004).

2.3 Bacteriological Quality

More than 90% of all reported cases of dairy related illness are of bacterial origin (Lingathurai and Vellathurai, 2010). The reasons for poor bacteriological quality of milk are due to unhygienic standard used in the production of milk including: unclean udder and teats which contributes the presence of coliform from various sources like manure, soil, feed, personnel and water; unhygienic milking procedures or equipments, and or use of water with inferior microbiological quality for cleaning, utensils and animals as well as

poor milk storage conditions (Khan *et al.*, 2011). However the existence of coliform bacteria may not necessarily indicate a direct faecal contamination of milk, but more precisely as an indicator of poor hygienic and sanitary practices during milking and further handling (Lingathurai and Vellathurai, 2010). Poor bacteriological quality of milk normally results from poor practicing of hygienic principles at the farms, which includes poor handling and transportation of milk. The principles include immediate cooling and storage of raw milk in proper container or equipment (Khan *et al.*, 2011). Storage of milk in equipments that is poorly designed or improper material, causes difficulties in cleaning and potential for fouling, this represent a source of psychotropic and some extent thermophilic bacteria, as well as the destructive enzymes (Kivaria *et al.*, 2006; Lingathurai and Vellathurai, 2010; Khan *et al.*, 2011). The environment in and around milking premises and the milking practices determine to a great extent the level of contamination of the milk (Pandey and Voskuil, 2011).

The Commonly isolated bacteria in contaminated milk include; *Staphylococcus* spp., *Campylobacter jejuni*, *Salmonella* spp., *E. coli* (EHEC and ETEC), *Listeria monocytogenes*, *Mycobacterium tuberculosis*, *Mycobacterium bovis*, *Brucella abortus*, *Coxiella burnetii*, *Yersinia enterocolitica*, *Leptospira* spp., *Clostridium* spp., *Klebsiella* spp., *Proteus* spp., *Enterobacter* spp., *Bacillus* spp., *Corynebacterium ulcerans*, *Pseudomonas aeruginosa* and *Streptococcus* spp. (Yirsaw, 2004; Donkor *et al.*, 2007a; Koo, 2008; Oliver *et al.*, 2009; Knutson *et al.*, 2010). Most of these bacteria are potentially pathogenic to human and others are known to cause milk spoilage (Donkor *et al.*, 2007a)

2.4 Chemical Changes in Raw Milk

During milk fermentation, micro-organisms produce several compounds; in particular they convert α - or β -lactose into D or L lactic acids. The increase of D and L lactic acids content during time is responsible of a pH decrease from milk pH (~ 6.8) to a value below 'caseins' isoelectric point (~ 4.6) (Hossain *et al.*, 2011).

Both acidity and pH of milk are used as indicators of quality (Hossain *et al.*, 2011). Milk acidity is expressed as percentage lactic acid because lactic acid is the principal acid produced by fermentation after milk is drawn from the udder. Fresh milk does not contain an appreciable amount of lactic acid. Therefore an increase in acidity and decrease of pH is a rough measure of its age and bacterial activity (Hossain *et al.*, 2011). Within a short time after milking, the acidity increases and pH decrease due to bacterial activity (Hossain *et al.*, 2011). The degree of bacterial contamination and the temperature at which the milk is kept are the principle factors which influence acid formation and decrease of pH. Therefore, the amount of acid depends on the cleanliness of production and the temperature at which milk is kept (Hossain *et al.*, 2011). Acidity affects quality of milk by altering its taste. For example, when acidity reaches about 0.3%, the sour taste of milk becomes sensible, at 0.4% acidity milk is clearly sour, and at 0.6% it precipitates at normal temperature, whereas at acidity over 0.9%, moulds start to develop (Hossain *et al.*, 2011).

2.5 Retail Business of Raw Milk

The consumption of milk is higher in urban areas than in rural areas, thus the retail business of raw milk is much more common in urban centres (Kurwijila and Henriksen, 1998). The retail milk business is highly associated with the informal marketing system, where producers supply their surplus production to their neighbours, vendor and/or in

local markets, mainly in the form of liquid milk (Yilma, 2012). The milk vendors in particular have been reported to be the main player in this system in many African countries (Kurwijila and Henriksen, 1998). They collect milk from the farmer and sell to households, kiosk and restaurant in the urban centres. A study conducted in Dar es Salaam city, Tanzania (Kivaria *et al.*, 2006) indicated that smallholder dairy farmer supply about 90% of the milk consumed in the city, however, 74% of all milk was marketed as raw milk through informal channels by vendors. In this system, the quality of milk and milk products is generally poor (Kivaria *et al.*, 2006). The main reasons are attributed to the prevailing situation where producers have limited knowledge of dairy product handling, coupled with the inadequacy of infrastructure such as electricity, transport and clean water in the production areas (Yilma, 2012; Kivaria *et al.*, 2006). Normally as is the case for most developing countries there are no operational legislation set to prohibit retail raw (unprocessed) milk business (Msangi, 2006). According to Swai and Schoonman, (2011) retail milk business by street vendors is highly associated with high microbial contamination which is caused by unhygienic handling, use of plastic containers and adulteration. The most common form of milk adulteration has been the addition of water to the milk (Mansour *et al.*, 2012). Poor quality of milk handled in informal market, lack of legislative instrument and the habit of people preferring to consume raw milk, predispose consumers to high health hazards (Kurwijila and Henriksen, 1998; Yilma, 2012; Kivaria *et al.*, 2006).

2.6 Antibiotic Residues

In veterinary practice, antibiotics are utilized for therapeutic and prophylactic purposes primarily to treat diseases and to prevent infection (Goffová, *et al.*, 2012). Antibiotics are also used at sub therapeutic levels to increase feed efficiency, promote growth and prevent diseases (Syt, 2008; Khaskheli *et al.*, 2008). The frequent use of antibiotics may result in

drug residues that can be found at different concentration levels in products from animal origin, such as milk and meat (Khaskheli *et al.*, 2008). The presence of residues above the maximum prescribed level is of public health concern (Khaskheli *et al.*, 2008). Antibiotic residues are small amounts of veterinary drugs or their active metabolites which remain in milk after treating various animal diseases like mastitis (Syit, 2008; Movassagh and Karami, 2010; Goffová *et al.*, 2012). The presence of antibiotic residues may be caused by failure to observe the required withdrawal periods, illegal use of drugs and incorrect dosage levels and route of administration (Akinwumi *et al.*, 2012). Other factors leading to the occurrence of antibiotic residues in animal products include poor records of treatment, failure to identify treated animals, lack of advice on withdrawal periods, off-label use of antibiotics, availability of antibiotics to lay persons over-the counter, extended usage or excessive dosages of antibiotics, non-existence of restrictive legislation or their inadequate enforcement and lack of consumer awareness about the human health hazards associated with antibiotic residues in the food (Akinwumi *et al.*, 2012).

Several studies on antibiotic residues in milk have been conducted in Tanzania (Karimuribo *et al.*, 2005; Kivaria *et al.*, 2006; Mdegela *et al.*, 2009). The general overview of the results from different areas of studies indicated that the issue of antibiotic residues in milk in many smallholder dairy farms in the respective areas was not a big problem.

However in another study by Kurwijila *et al.* (2006) a significant positive result was found showing that there was a problem of following withdrawal periods. These results indicated that antibiotic residue in milk varies from place to place, and therefore a general conclusion on the level of antibiotic residues in Tanzania may be uncertain. On the other hand, it has been reported from several studies from various places in Kenya (Shitandi, 2004; Kang'ethe *et al.*, 2005; Omoro *et al.*, 2005) that there is high problem of antibiotic

residue in milk. The attributed reasons for high prevalence of antibiotic residues include farm-level abuse of drugs and the market-level practices which introduce antimicrobials to milk as a means of preservation.

2.7 Potential Effects of Antibiotic Residues to Human Being

The antibiotic residues when taken above the maximum residue limit (MRL) can result into potential health effects to the human being. The effects include the occurrence of resistant strains of bacteria in humans, toxicity effects of the drug, allergic reactions (hypersensitivity reactions) in sensitized persons and inhibition of starter cultures in production of cultured milk products such as yogurt and also the manufacture of cheese (Movassagh and Karami, 2010; Kaya and Filazi, 2010).

Other effect includes intestinal dysbiosis (Goffová *et al.*, 2012). Also some drugs or their metabolites possess carcinogenic potential e.g. meat preserved with sodium nitrate and contains sulphamethazine residues, may develop a triazine complex that has a considerable carcinogenic potential (Goffová *et al.*, 2012). Prolonged ingestion of tetracycline in food has detrimental effects on teeth and bones in growing children. Some reports have also associated drug residues to destroy useful microflora of gastrointestinal tract, especially in children leading to enteritis problems (Goffová *et al.*, 2012).

2.8 Commonly Used Antibiotics in Dairy Cattle

Antibiotic are found in different groups which are available for treatment of infected livestock. The most common groups include the beta-lactams, Sulphonamides, Aminoglycosides, Macrolides, Tetracyclines and Chloramphenicol (Omore *et al.*, 2002; Movassagh and Karami, 2010; Pecou and Diserens, 2011). These antibiotics may be used singly or at times in combination when treating dairy cattle. Several studies have been

done on commonly used antibiotics in livestock in different countries. In Tanzania studies have been conducted by Mbando, (2004); Nonga *et al.* (2009); Midenge, (2011); Katakweba *et al.* (2012). The general conclusion from these studies indicated that there are rampant and indiscriminate uses of antibiotics among the Livestock keepers in Tanzania.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Area and Duration

The study was conducted in Arusha city and Meru District Council in Arusha Region, from October to December 2012. Arusha Region is laying in Latitude 3° 22' 0" South and longitude 36° 41' 0" East, in the northern part of Tanzania (Fig. 1). The region is bordered with Kenya on the Northern, Kilimanjaro region on the East, Manyara and Singida Regions on the South, Simiyu and Mara Regions on the west. Arusha Region has seven district councils that include Arusha Municipal, Meru, Arusha, Monduli, Longido, Karatu and Ngorongoro Districts (Fig. 1). According to the 2002 national census, Arusha Region had a population of 1 288 088 people while Arusha city had 281 608 and Arumeru District before division into Meru and Arusha district councils had 514 651 people. Arusha District council occupies the western part of Arusha city and Meru District council occupies the eastern part.

Arusha city and Meru District were selected as study areas because they both have large number of raw milk street vendors and smallholder dairy farmers. Meru District Council consist of 17 wards, however Nkoanrua and Siela Sing'isi wards were purposively selected to be the study areas because they have large number of smallholder dairy farmers. According to ward's livestock officers records, Nkoanrua ward have 4900 smallholder dairy farmers and Siela Sing'isi have 5900 smallholder dairy farmers. Each smallholder dairy farmer has cattle number ranging from one to five. Ambureni – Moivaro village in Nkoanrua ward and Sing'isi village in Siela Sing'isi ward in Meru district Council were selected as study villages for smallholder dairy farmers since milk produced

from these areas is supplied to different location of Arusha City. Arusha City is the focal point where the milk vendors and retailers sell the milk.

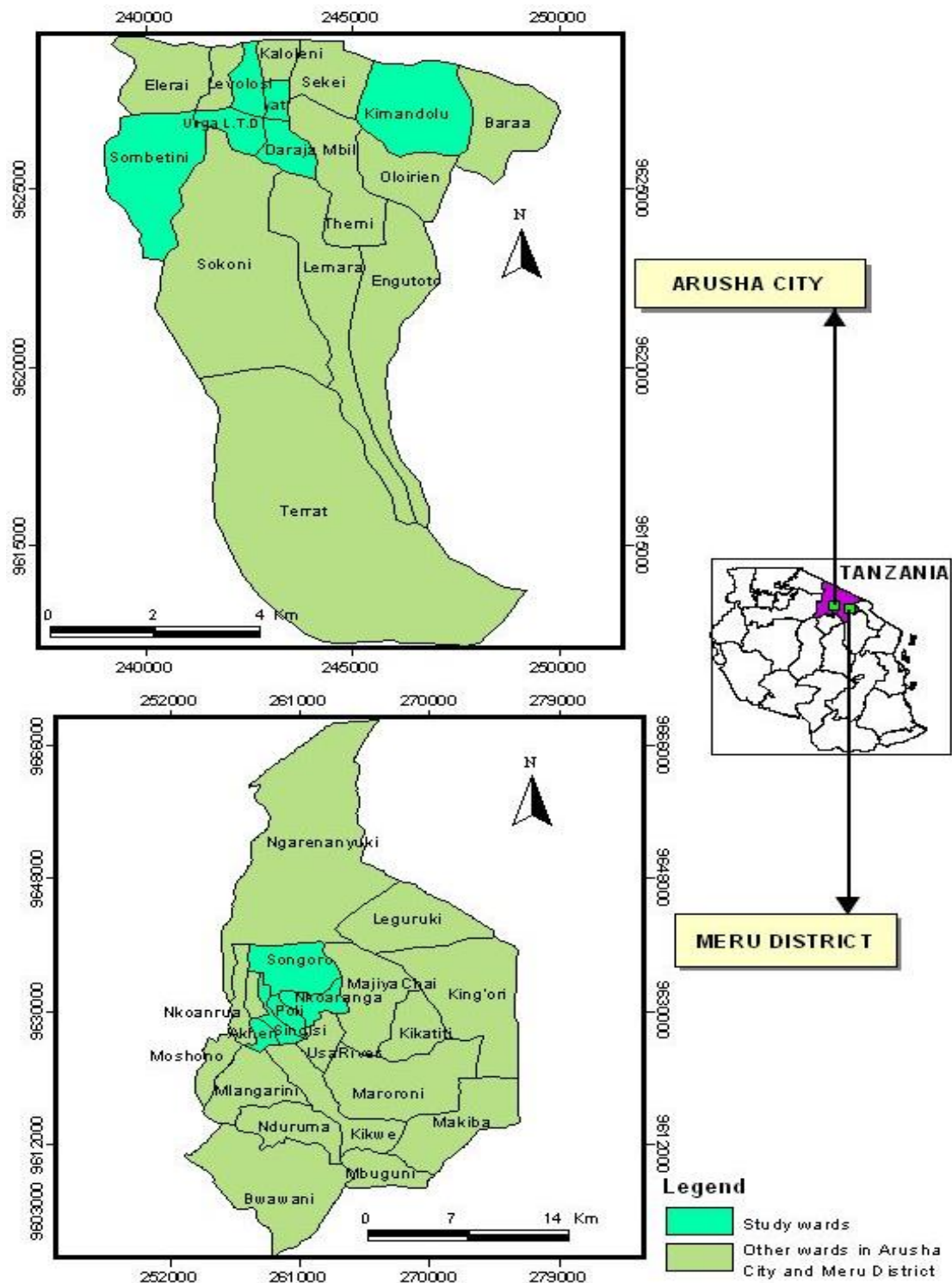


Figure 1: A map of Arusha City and Meru District showing study wards. Insert is Arusha region in the map of Tanzania that shows different regions.

3.2 Study Design

A cross-sectional study design was used. Smallholder dairy farmers, milk vendors and milk retailers were visited once and the questionnaires were administered to randomly selected study participants of randomly selected representative sample of the population from both Arusha city and Meru District Council. From the selected sample of respondents of the population, milk samples were collected for preliminary organoleptic assessment of physical quality of raw milk and laboratory analysis.

3.3 Study Population

The Population under study was considered to be heterogeneous comprising of varied gender and age groups. From the population, a representative randomly selected number of milk vendors, retailer and smallholder dairy farmers both in Arusha City and in Meru District Council were considered in the study. The inclusion criteria were; Smallholder dairy farmers both women and men who keep from one to five dairy cattle, or willingness to participate in the study and able to give information and accessibility of the place during data collection. The exclusion criteria included; unwillingness to participate in the study, unable to give information asked and living in inaccessible areas especially for smallholder dairy farmers. Also those who had no time to for interviews were excluded.

3.4 Sample Size Determination

A formula by Kothari (2004) for unknown population (i.e. $n = Z^2SD^2/e^2$) was used to calculate the study sample size. Where Z, is the estimated standard variant at 95% CI and considered the point of the normal distribution corresponding to the level of significance (Z=1.96). SD, the estimated standard deviation of the study sample and was considered to be 0.15 or 15% and the estimated error 'e' was considered at 0.05 or 5%.

The sample size 'n' was calculated as:

$$\left[\frac{1.96^2 (0.15)^2}{(0.05)^2} \right] \times 3 = 105$$

The 105 respondents applied for three categories of data (i.e. milk vendors, milk retailers and smallholder dairy farmers). Out of this calculated sample size, 35 questionnaires were administered to milk vendors, 35 questionnaires to milk retailers and the other 35 questionnaires to smallholder dairy farmers. Each category of respondents had a separate set of questionnaire. Generally the questionnaires were meant to assess the respondent's awareness on general issues with regard to milk quality, hypothesised possible health risk factors associated with consumption of raw milk and antibiotic residues. Simultaneously, 105 samples of raw milk were collected for laboratory analysis of pH, specific gravity, alcohol test, physical dirtiness, microbial quality and qualitative and quantitative determination of antibiotic residues.

3.5 Ethical Consideration

Research permit was provided by the Vice Chancellor Sokoine University Agriculture (Appendix 1) and permission letters were obtained from Executive Directors of Arusha City Council and Meru District Council (Appendix 2). Verbal consent was obtained from each of the heads of households of smallholder dairy farmers, vendors and retailers after explaining the purpose and importance of the study prior to commencement of interviews and sampling. Participation in the study was on voluntary basis. All the information collected from the participants and the laboratory results obtained after milk sample analysis were kept under the custody of the researcher as confidential and the study participants were anonymized.

3.6 Plan for Data Collection

3.6.1 Recruitment of research assistants

Two research assistants were recruited; one livestock field assistant officer who had a certificate in animal health was recruited to assist the researcher in data collection. The research assistant had good communication skills and experience on interaction with smallholder dairy farmers, milk vendors and retailers. The second research assistant had a diploma in laboratory technology. Inclusion of research assistant helped in maximizing trust of respondents, interviewer- interviewee interaction and facilitation of laboratory work. The research assistants were briefed on the objectives of the study, data collection process, target respondents, selection criteria, approach during interview, understanding questions, elaborating why and how each question was asked, assuring the respondents confidentiality, how to record responses from interviewees, laboratory analysis procedures and data collection and recording.

3.6.2 Sampling plan for milk vendors, retailers and smallholder dairy farmers

A multistage sampling technique was employed to obtain the milk vendors, milk retailers and smallholder dairy farmers as follows:

Stage I: This involved selection of study wards within Arusha City and Meru District council. Arusha City consists of 19 wards of which 6 were selected. Selection criterion was based on the availability of large number of both retailers and vendors particularly those which are within the city centres. Meru District Council consists of 17 wards which were listed down in order of number of smallholder dairy farmers and purposive sampling was done to select 2 wards with many smallholders' dairy farmers.

Stage II: Selection of streets and villages in the selected wards was done by simple random sampling. This was done only for milk retailers and smallholder dairy farmers.

For retailers, the number of street selected for each ward depended on concentration of business activities in a particular ward, where as for smallholder dairy farmers, selection of street within a village depended on the number of the farmers in a particular street. Selection of respondents within a street was random and based on the respondent's willingness to participate in the study. For milk vendors, the researcher and research assistant visited various selling locations and streets, using good communication skills and assurance of confidentiality, participants who were ready to participate were immediately enrolled for questionnaire and sample collection.

3.6.3 Pretesting of data collection tools

Pretesting of questionnaires was done in order to test the clarity, sequence of the questions and estimate the duration for each questionnaire. A total of five respondents were interviewed in each category (i.e. milk vendors, milk retailers and smallholder dairy farmers). After testing of the questionnaires, they were revised and arranged in a better chronology. The revised version of the questionnaires that was used in the study was translated into 'Kiswahili', the national language understood by majority of Tanzanians

3.7 Sociological Data Collection Techniques and Tools

Sociological data were collected from respondents (milk vendors, milk retailer and smallholder dairy farmers) during street and household visits by interviews using questionnaires with both closed and open-ended structured questions (Appendix 3). The face -to- face interview targeted all the selected respondents. Each cohort of respondents had a specific kind of questionnaires aimed to collect specific information. For the smallholder dairy farmers, the aim was to obtain information regarding general health status of cattle; disease management and control; milk production, handling, storage, explore the extent of existence of health problems acquired through drinking of raw milk,

issues of antibiotic residues and compliance to drug withdrawal period. For the milk vendors, the information collected included, sources of milk, transportation, challenges of milk spoilage and rejection by retailers, explore the extent of awareness of milk quality and hypothesised health risks associated with consumption of raw milk. Morales similar information was also collected from the milk retailers in Arusha City. The questionnaires were administered by a researcher and assisted by research assistants.

3.8 Milk Sampling for Laboratory Analysis

Milk sampling was done after questionnaire interview was complete. Samples were collected directly from the storage containers used by milk vendors and retailers during street visit. At least 250 ml of milk samples was collected and put into a sterile bottle and stored in a cool box with ice blocks during the field work. For case of smallholder dairy farmers, samples were collected from the milking containers. All the collected samples were being transported to the laboratory at Arusha Veterinary Investigation Centre (VIC) for analysis within six to eight hours of collection.

3.9 Laboratory Analysis

3.9.1 Physical dirt

Milk samples were filtered using a clean white cloth and assessed for physical dirt/contaminants (Lore *et al.*, 2006). Any debris remained on the white cloth; the milk was regarded as dirty.

3.9.2 Organoleptic assessment of milk

Smell of milk was assessed using sensory organs immediately after the respondent opened the lid of the container (EACs, 2006; Pandey and Voskuil, 2011), while the colour was

examined visually by putting well stirred milk in the clean glass container as described by Kurwijila *et al.* (2009).

3.9.3 Milk pH

Determination of pH of milk was done using electronic pH-meter at 20°C. Before taking the measurements, a pH-meter was calibrated using two standard buffer solutions at pH 7.0 and pH 4.0 (ISO 7218:2007). About 100 ml of milk was put into a clean measuring cylinder and then a pH-meter was dipped up to the mark. After the pH-meter had stabilized the reading was directly recorded.

3.9.4 Milk clot

Milk clot was assessed by alcohol test. A volume of 5 ml of well stirred milk sample was mixed with equal volume of 70% alcohol (ethanol) in a clean test tube. The mixture was then mixed by inverting the test tube several times and visual examination of presence or absence of milk clots was done on the sides of the test tube as described by Kurwijila *et al.* (2009).

3.9.5 Specific gravity

Specific gravity was measured by use of a lactometer at standardized temperature of 20°C (Kurwijila *et al.*, 2009). Milk temperature was adjusted using a water bath set at 20°C and checked by hand thermometer. Then by using a clean 50 ml measuring cylinder, milk was poured to three quarter of the cylinder. Then, carefully the lactometer was immersed and left to float freely, more milk sample was added to fill the remaining part of the cylinder. Before taking lactometer reading, change of milk temperature was rechecked by using a hand thermometer. Then, both lactometer and thermometer readings were recorded. Any change of temperature from 20°C led to correction of the lactometer reading, whereby for

every degree change above 20°C, 0.2 units were added to lactometer reading and for every degree change below 20°C, 0.2 units were subtracted from the lactometer reading (Kurwijila *et al.*, 2009; Pandey and Voskuil, 2011). A formula $SG = 1 + L/1000$, where L is corrected lactometer reading was used to calculate the Specific Gravity (SG) of the milk (Kurwijila *et al.*, 2009).

3.9.6 Total viable count (TVC)

Total viable count (TVC) also known as horizontal method for enumeration of microorganism or colony count technique at 37°C was applied (TZS 118:2007).

Apparatus and material used: Glass Petri dishes of 90 to 100 mm in diameter, pipettes tips of 1 ml capacity, pipettes of 10 ml capacity, test tubes of more than 10 ml capacity, glass flask of 1000 ml capacity, glass measuring cylinder 1000 ml capacity, incubator and nutrient agar and MacConkey agar powder (Oxoid[®] Ltd., Basingstoke, U.K.) as growth media.

Preparation: All glassware was thoroughly washed and dry sterilized in an oven at 120°C. Media preparation was done according to manufacturers' instruction (Oxoid[®] Ltd., Basingstoke, U.K.) where 14 gm of nutrient agar powder was measured using electronic weighing balance, and then put into a flask of 1000 ml containing pre measured 500 ml of distilled water. The content was stirred, shaken and boiled until the agar was completely dissolved. The media was then sterilized in an autoclave at 121°C for 15 minutes (TZS 118:2007). The media was then cooled to 45°C at room temperature ready for pouring 15 to 20 ml onto Petri dishes.

Sample preparation and incubation: Determination of level of bacterial contamination for each category was prior done using five samples. Ten- fold serial dilution from 10^{-1} to 10^{-10} in sterile normal saline solution was done, using disposable pipettes. One (1 ml) of the milk sample was added into 9 ml of sterile normal saline (10^{-1} dilution). Then, 1 ml of the dilution was transferred into a second tube containing 9 ml of normal saline (10^{-2} dilution); the procedure was repeated for further dilutions to 10^{-10} dilution as shown in Fig. 2.

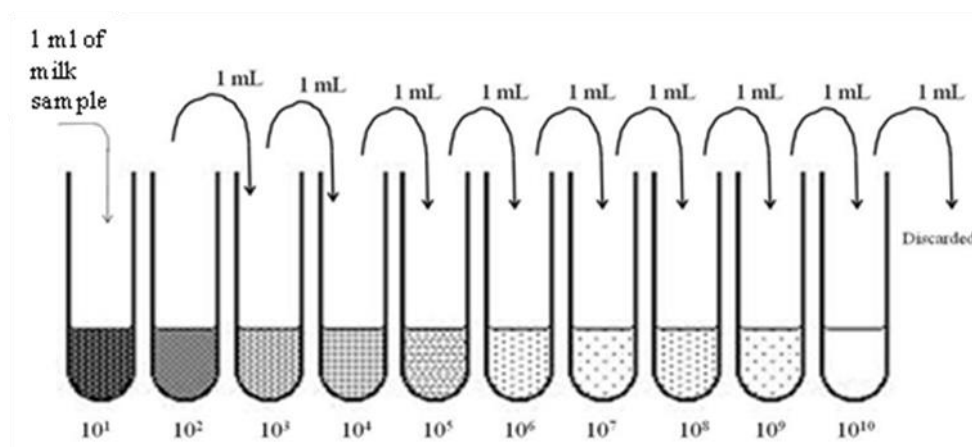


Figure 2: Serial dilutions of milk samples in 10 tubes containing 9 ml of normal saline each

From each dilution, 1 ml was placed on a sterile Petri dish followed by the addition of 15-20 ml of cooled to 45°C nutrient agar onto the dishes (ISO/FDIS 8261 (E), 2001). The sample and agar in a Petri dish were gently shaken by rotating to mix and left to solidify by leaving the Petri dishes standing on a cool horizontal bench. After solidification the Petri dishes were inverted and placed in the incubator at $37^{\circ}\text{C} \pm 1^{\circ}\text{C}$ under aerobic condition for $24 \text{ h} \pm 3 \text{ hours}$. The time from preparation of sample to pouring of media into dishes did not exceed 45 minutes.

From the results it was learnt that, for milk vendors and milk retailers, critical dilution of 10^{-4} to 10^{-6} were best for countable range of 30 - 300 colony forming units per plate

(cfu/plate), similarly critical dilution of 10^{-2} to 10^{-4} was best for samples from smallholder dairy farmers. Then the rest of the samples were ten-fold diluted from 10^{-1} to 10^{-6} for samples from vendors and retailers. Parallel with the test samples, controls were also prepared which involved Petri-dish containing sterile normal saline as test sample. The controls were used to check sterility of the diluents (normal saline), media and the environment as source of contamination.

Counting of bacterial colonies

After the incubation period bacterial colon count on the plates was done with the aid of portable magnifying lens, colonies in the culture plate were countered by using colony counter. Three critical dilutions per each sample were counted. A plate was divided into quarters, using a pointer under subdued light colonies were counted. Large, pinpoint and spread bacterial colonies were counted but spread colonies were counted as single colony. Two consecutive plates with 30 to 300 colony forming units (cfu/ml) were considered for record (Khan *et al.*, 2008; Hossain *et al.*, 2011).

Expression of results

The countable bacterial colonies from two consecutive plates of each sample were converted into colony forming units per millilitre (cfu/ml) using a formula $N = \sum C/v * 1.1 * d$, where N- number of bacterial colonies counted, C-sum of colonies counted in two successful dilutions, v- volume of sample and d- dilution in the first plate counted (ISO 7218:2007(E)).

Identification of common bacteria in cultures

Common bacteria were identified through their colony morphology and use of MacConkey media as differential media. During colony counting the most occurring

colonies were Gram stained for micromorphology examination under the light microscope. Standard biochemical tests were performed as a further means of identification of common bacteria (Carter and Wise, 2004).

3.9.7 Assessment of antimicrobial residues in milk samples by use of delvotest

Antimicrobial residues in milk were assessed qualitatively by using Delvo SP[®] Kit (SP mini kit; Delft, the Netherlands) which uses *Bacillus stearothermophilus var. calidolactis* as the test bacteria. The method was carried out according to manufacturer's instructions, using two standard control samples: Positive control sample used Gentamycin antibiotic (Laprovect[®], Indre Et Loire, France). This was prepared by using 10 mg of the (standard 1% Gentamycin antibiotic) into 10 ml of sterile distilled water forming a concentration of 100 µg/ml as stock solution. Then 0.1 ml of stock solution was added into 10 ml of UHT milk to a concentration of 1 µg/ml which was used as positive control (Goffová *et al.*, 2012). Fresh UHT milk bought from the shop was used as negative control.

Five samples were used to pre-test the Delvo SP[®] Kit for which two were collected from known antibiotic treated dairy animal and three from none antibiotic treated animals. A volume of 0.1 ml of each sample and the controls were added into the Delvo SP[®] Kit ampoules, and then incubated at 64°C±0.5°C for 3 hours. A colour change of the pH indicator from blue-violet to yellow indicated a negative result as the test bacteria strain grew (Fig. 3). Complete purple (blue-violet) colour of at least two third of the gel indicated a positive result to antimicrobial residues since they inhibit the growth of the tested strain, *Bacillus stearothermophilus var. calidolactis* (Fig. 3). Each time the test was run together with the controls.

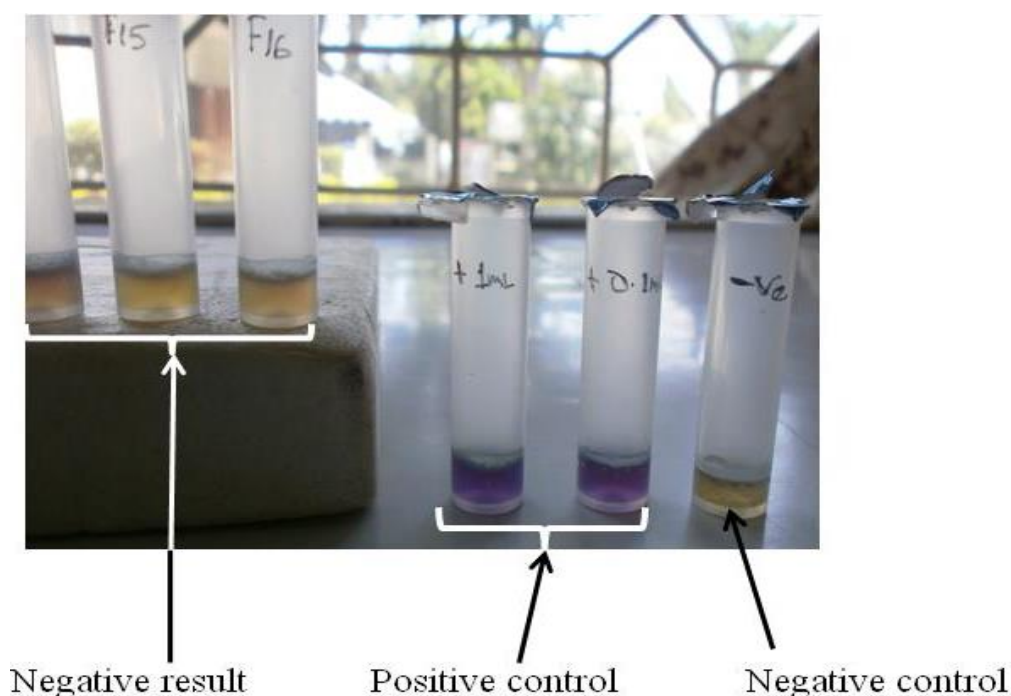


Figure 3: Delvotest SP[®] kit Controls and test results

Test samples

The required amount of ampoules were removed from the frame, and opened by punching the aluminium foil using a sharp blade and marked for sample identification. Fresh disposable pipettes were dipped 1 cm into samples to draw 0.1 ml and add straight into the agar medium gel.

Incubation: Temperature of the incubator was adjusted to $64^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$. The prepared ampoules were then put into the incubator with timer set to 3 hours. After incubation time, the colour of solid agar in the ampoules was observed. The colour change from purple (blue-violet) to yellow similar to the negative control was recorded as negative implying that there were no antibiotic residues and a no change in purple (blue-violet) colour similar to positive control was declared positive implying that there was inhibition by antibiotic residues (Fig. 3).

3.10 Data Analysis

Raw data established from organoleptic quality assessment of milk, questionnaires from respondents and laboratory analysis were entered and stored into Excel spread sheet. Proportions were computed using Epi- Info database (version 7 of 2012). Epi info database was also used to compute descriptive statistics for measure of central tendency and measure of variability. Continuous and proportions of categorical variables were computed and Chi-square used to compare for statistical significance at a critical probability of $p < 0.05$.

CHAPTER FOUR

4.0 RESULTS

This chapter comprise results from sociological survey using questionnaires and laboratory analyses based on the study objectives. Results are summarised using tables and figures. The chapter presents the findings for demographic characteristics of the respondent, awareness on milk quality, quality of milk and awareness of smallholder dairy farmers on veterinary drug uses.

4.1 Demographic Characteristics and Distribution of Respondents

4.1.1 Demographic characteristics of the study population

The study involved 35 participants from each category of milk vendors, milk retailers and smallholder dairy farmers, making a total of 105 respondents. Demographic information of the respondents is detailed in Table 1. The results shows that there were more males (97.1%) involved in milk vending business (walking around selling milk to individual, households, kiosks and restaurants) than females ($p = 0.0000$, $\chi^2 = 20.3546$). On the other hand, results showed there was no significant difference ($p = 0.2354$, $\chi^2 = 1.4042$) between female and males involved in retail (selling milk in kiosks or restaurants) business of milk. Significant different ($p = 0.0000$, $\chi^2 = 20.3386$) was observed for females being involved in smallholder dairy farming than men. On age groups also a significant difference ($p = 0.0001$, $\chi^2 = 17.6302$) was observed that more vendors were youth aged between 20 to 45 years than adults above 45 years. Furthermore, education wise results showed that majority of the milk vendors, (milk retailers and smallholder dairy farmers had primary school education.

4.1.2 Distribution of the respondents in the study wards and villages

Distribution of respondents (milk vendors, milk retailers and smallholder dairy farmers) in different wards and villages is presented in Table 1. All milk vendors were from Meru District in 6 wards and 10 villages. The wards included; Nkoarua, Akeri, Poli, Siela Sing'isi, Songolo and Nkoaranga. On the other hand milk retailers interviewed were from seven out of 19 wards of Arusha City which included; Darajambili, Kati, Kimandolu, Levulosi, Sombetini and Unga Ltd. Main categories of milk retail business were restaurants and kiosks. The smallholder dairy farmers who participated in the study were from two wards; Nkoanrua ward in Amburen Moivaro and Nguruma villages and Siela Sing'isi ward in Sing'isi village (Table 1).

Table 1: Demographic information of milk vendors, retailers and smallholder dairy farmers in Arusha City and Meru District Council (October - December, 2012)

Demographic information	Category	Number (%) of respondents		
		SDF	Milk vendors	Milk retailers
Gender	Female	27 (77.1)	1 (2.9)	20 (57.1)
	Male	8 (22.9)	34 (97.1)	15 (42.9)
	p- value	0.0000 *	0.0000 *	0.2354 ^{ns}
Education	Primary	33 (94.3)	32 (91.1)	21 (60.0)
	Secondary	2 (5.7)	8 (22.9)	12 (34.3)
	> Secondary	0 (0.0)	0 (0.0)	2 (5.7)
Age	<45	-	29 (82.9)	-
	>45	-	6 (17.1)	-
	P - value		0.0001 *	
Ward Distribution in Meru District	Nkoanrua	19 (54.3)	10 (28.6)	-
	Akeri	-	10 (28.6)	-
	Poli	-	3 (8.6)	-
	Siela Sing'isi	16 (45.7)	8 (22.9)	-
	Songolo	-	3 (8.6)	-
	Nkoaranga	-	2 (5.7)	-
Ward distribution in Arusha city	Daraja mbili	-	-	4 (11.4)
	Kati	-	-	2 (5.7)
	Kimandolu	-	-	10 (28.6)
	Levolosi	-	-	8 (22.9)
	Sombetini	-	-	3 (8.6)
	Unga Ltd	-	-	2 (5.7)

Key: *- Significant difference, ^{ns} - No significant difference, SDF= Smallholder dairy farmers

4.2 Awareness of Smallholder Dairy Farmers, Milk Retailers and Milk Vendors on Milk Quality

4.2.1 Awareness of vendors on milk quality

Awareness responses of vendors on milk quality are presented in Table 2. The major source of milk for the vendors was reported to be smallholder dairy farmers. Milk vendors reported that the quality of milk was mostly related to cleanliness of containers and milking practice at farm level. The vendors mostly sold milk to household, restaurants (54.2%, $n=19$) and some sold milk to customers at local open-air markets. All the

respondents admitted filtering milk using normal plastic filters during receiving milk from the farmers and selling to retailers. The common milk storage condition was room temperature and less dipping of containers with milk in cold water especially those who collected milk during the evening. The time estimated from milk collection to complete sale was between 5 to 8 hours. Plastic containers were commonly used by all vendors for collection, storage and transportation of milk to the city, the main means of transportation was through public town min buses, motorcycle and bicycle (Figure 4). After completion of milk selling and upon return home, empty containers were washed by soap and hot water and dried in inverted position. Collection of milk was done early morning and evening or both. The problem of milk spoilage was reported to be high and majority reported to use such spoiled milk as fermented milk which was again sold to consumers.

Table 2: Handling, storage and transportation of milk by 35 milk vendors in Arusha City and Meru District Council (October - December, 2012)

Parameter	Category	Number (%) of respondents
Source	SDF	34 (97.1)
	Own	1 (2.9)
Selling pints	HH	6 (17.1)
	HH & RS	19 (54.2)
	RS	4 (11.4)
	HH & KK	2 (5.7)
	RS & KK	1 (2.9)
	HH, KK & RS	2 (5.7)
	Others (School)	1 (2.9)
Milk filtration	Filter milk	35 (100)
Milk storage	Room temperature	28 (80.0)
	Deep in cold water	7 (20.0)
Milk collection time	Evening	7 (20.0)
	Early morning	24 (68.6)
	Evening & Morning	4 (11.4)
Milk spoilage	Yes	30 (85.7)
Use of spoiled milk	Feed family	8 (22.9)
	Dispose	2 (5.7)
	Ferment & sell	25 (71.4)
Transportation	Bicycle	2 (5.7)
	Motorcycle	2 (5.7)
	Public buses	30 (85.7)
	Others (walking)	1 (2.9)
Milk containers	Plastic container	35 (100)
Status of containers	Clean	11 (31.4)
	Dirt	24 (68.6)
Container cleaning	Soap & hot water	35 (100)
Hand washing	Soap & hot water	35 (100)

Key: SDF= Smallholder dairy farmers, HH= Household, RS= Restaurant, KK= Kiosk



Plate 1: Different means of milk transportation by milk vendors in Arusha City and Meru District Council (October - December, 2012). A and B using public mini bus, C using motorcycle and D loading onto a bicycle.

4.2.2 Awareness of milk retailers on milk quality

Awareness of retailer on milk quality is shown in Table 3. The main sources of milk for retailers were reported to be from street vendors and smallholder dairy farmers. The milk retailers also reported that the quality of milk was based on cleanliness of the milk sources, handling and transportation to the final consumers or retailers. Nearly half of the respondents reported to receive spoiled milk, of which majority sold as fermented milk and some disposed it.

It was further found that the common method for determination of good milk was through boiling, visual and smells examination. Milk received by retailers was mainly used for tea

making or used as fresh boiled milk to drink. Majority of the respondents fermented raw milk for sale while few respondents reported to use boiled milk for fermentation.

Table 3: Handling of milk by 35 retailers in Arusha City (October – December, 2012)

Variable	Category	Number (%) of respondents
Source of milk	SV	22 (62.9)
	SDF	7 (20.0)
	SV & SDF	4 (11.4)
	Own	2 (5.7)
Milk spoilage	Yes	16 (45.7)
	No	19 (54.3)
Determination of milk quality	Boil	23 (65.7)
	Smell	3 (8.6)
	Visual	7 (20.0)
	Boil & smell	1 (2.9)
	Boil & taste	1 (2.9)
Use of spoiled milk	Dispose	10 (29.4)
	Ferment & sell	25 (70.6)
Use of raw milk	Tea making	3 (8.6)
	Fresh & ferment	3 (8.6)
	Tea & fresh	19 (54.3)
	Tea & ferment	1 (2.9)
	Tea, fresh & Ferment	9 (25.7)
Treatment before fermentation	Boil	14 (38.5)
	None	21 (61.5)

Key: SV= Street vendors, SDF=Smallholder dairy farmers

4.2.3 Awareness of smallholder dairy farmers on milk quality

The common type of cattle husbandry practiced was zero grazing (97.1 %, $n=34$) in closed animal house (Table 4). A number of cattle diseases were mentioned by respondent. However, it was pointed out that the leading diseases affecting cattle were Worms, Trypanosomosis, respiratory diseases and Anaplasmosis (Fig. 4). On the other hand, the ward livestock officer reported that the leading cattle diseases in the study area were Anaplasmosis, Mastitis, Trypanosomosis, and Pneumonia. Treatment for the diseases in the area was done both by private livestock practitioners and ward livestock extension officers.

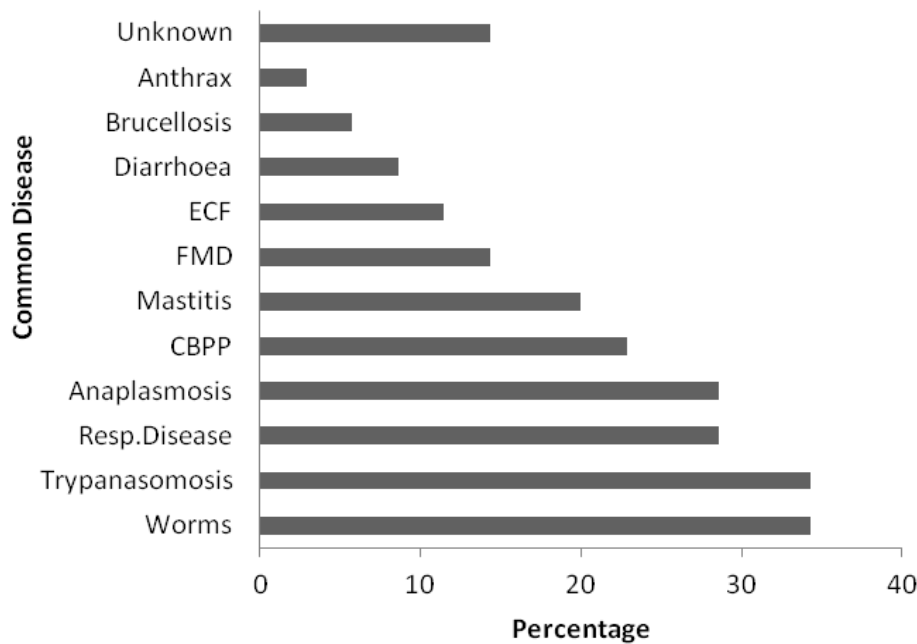


Figure 4: Common diseases affecting dairy cattle in Arusha City and Meru District Council (October - December, 2012)

Key: FMD= Foot and Mouth Disease, ECF= East Coast Fever, CBPP= Contagious Bovine Pleuropneumonia, Resp. Disease = Respiratory diseases

4.2.4 Milk production

Milk production practices by smallholder dairy farmers are presented in Table 4. It was found that milking was done in the same animal house used for feeding and general stay and that milking practice was done by hands. All the respondents admitted to use plastic containers to collect milk during milking. Majority of respondents used hot water and soap for washing containers and for washing their hands before milking. The udder was reported to be washed using warm water and was not dried by towel. It was indicated that all the produced milk was filtered and stored in plastic containers under room temperature before selling or other home uses.

Table 4: Milk production practices by smallholder dairy farmers in Arusha City and Meru District Council, (October - December, 2012)

Practice	Category	Number (%) of respondents
Milking place	Inside animal house	34 (97.1)
	Outside animal house	1 (2.9)
Type of utensils	Plastic container	35 (100)
Cleaning of utensils	Soap and cold water	1 (2.9)
	Soap and hot water	34 (97.1)
Hand washing	Soap and cold water	4 (11.4)
	Soap and hot water	31 (88.6)
Udder washing	Cold water	1 (2.9)
	Warm water	34 (97.1)

4.2.5 Awareness of the respondents to health risks associated with consumption of raw milk

Results showed there was no significant difference ($p > 0.05$) on awareness among respondents; milk vendors 33 (94.3%), milk retailers 32 (91.4%) and smallholder dairy farmers 34 (97.1%) that consumption of raw milk could result into health effects. Figure 5 shows diseases that were reported by respondents to be associated with consumption of raw milk. Tuberculosis and brucellosis were the two common diseases mentioned by majority of the respondents. Interestingly, milk vendors (54.3%) also named worm infestations as one of the diseases which can be infected to humans through consumption of raw milk. On the other hand, 40% ($n = 14$) of milk vendors, 35.3% ($n = 13$) of milk retailers and 6.1% ($n = 3$) of smallholder dairy farmers could not mention any disease condition known to be associated with consumption of raw milk. Majority of the respondents from all categories; milk vendors (97.1%, $n = 34$), milk retailers (88.6%, $n = 31$) and smallholders dairy farmers (94.3%, $n = 33$) reported that milk-borne diseases associated by consumption of raw milk could be prevented through boiling of milk.

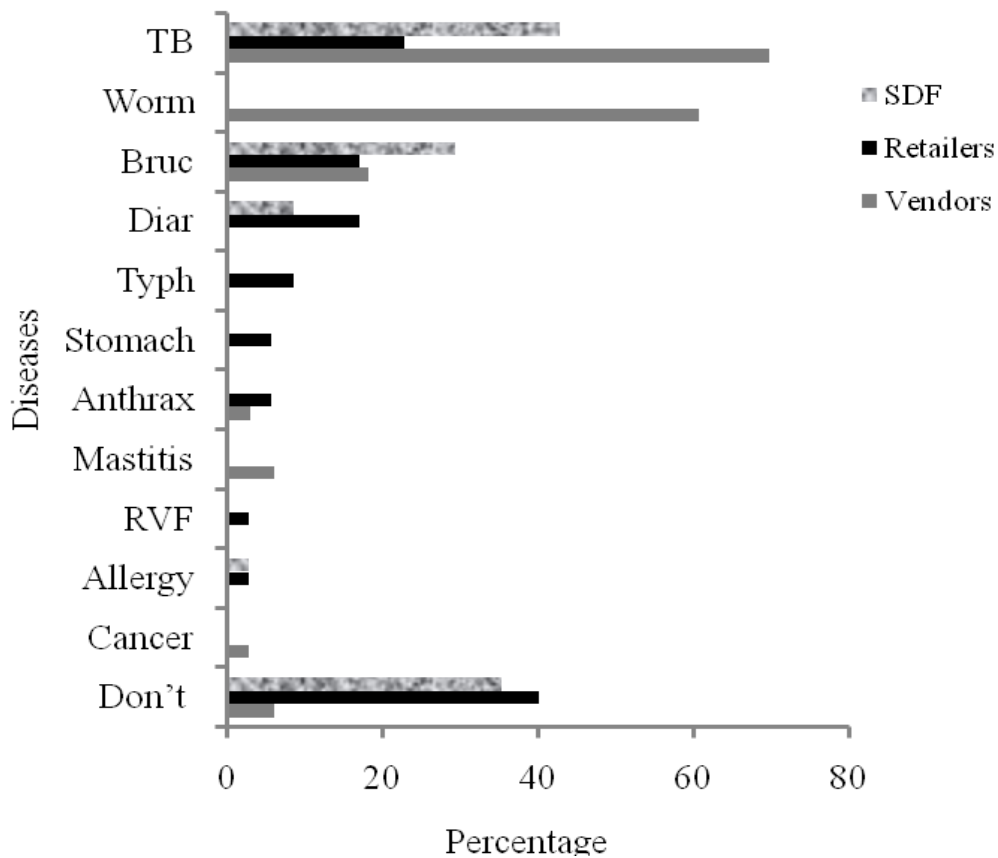


Figure 5: Milk-borne diseases thought to be transmitted through consumption of raw milk in Arusha City and Meru District Council (October – December, 2012)

Key: RVF= Rift valley fever, Typh = Typhoid fever, Diar = Diarrhoea, Bruc= Brucellosis, TB= Tuberculosis, Don't = don't know

4.2.6 Awareness on signs of milk-borne diseases associated with consumption of raw milk

The signs of diseases associated with consumption of raw milk are presented in Figure 6. Coughing was reported by 34.3% ($n = 12$) of smallholders dairy farmers, 25.7% ($n = 9$) of milk vendors and 11.4% ($n = 4$) of milk retailers as the common sign associated with tuberculosis in humans. On the other hand many retailers (71.4 %, $n = 25$), vendors (71.4%, $n = 25$) and smallholders dairy farmers (48.6%, $n = 17$), had no knowledge on the signs of most of the mentioned diseases associated with consumption of raw milk.

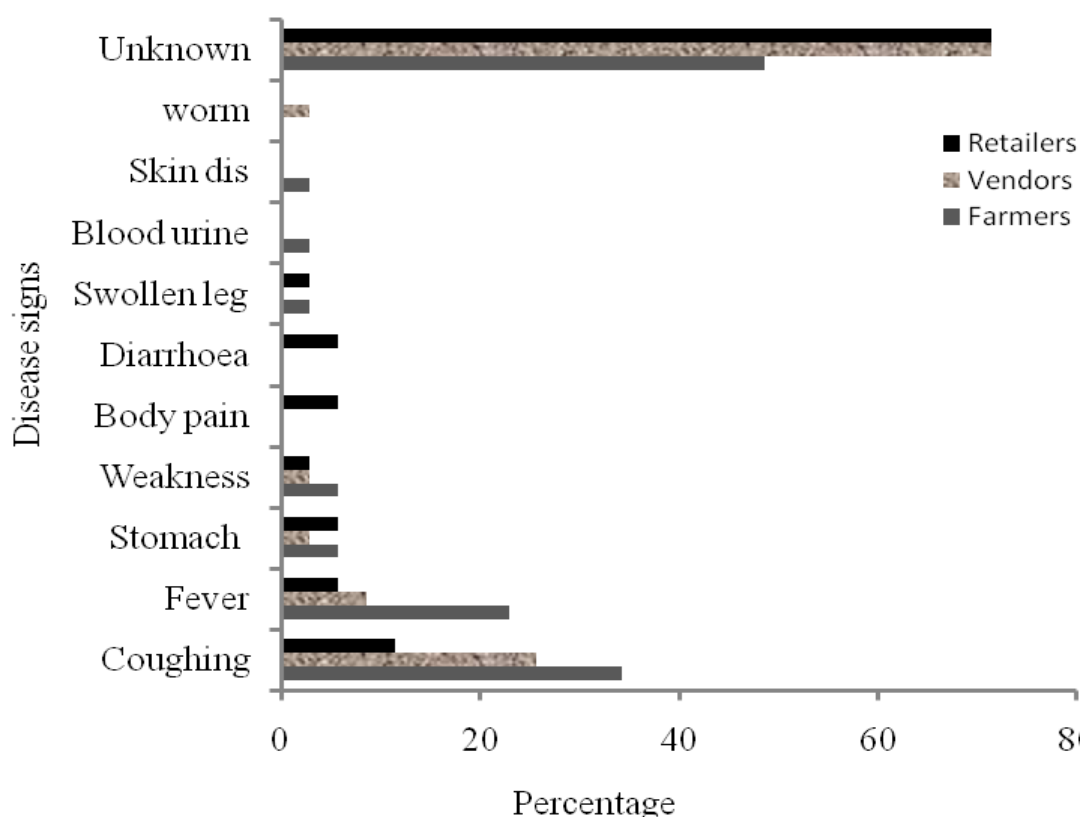


Figure 6: Signs of diseases associated with raw milk consumption in Arusha City and Meru District Council (October - December, 2012)

4.3 Quality of Milk Produced and Sold in the Study Area

4.3.1 Organoleptic characteristics

The quality of milk as defined based on organoleptic characteristics (colour, smell, physical dirt and clotting) are summarised in Table 5. Results show that most milk from smallholder dairy farmers, milk vendors, and milk retailers had yellowish white colour. Except for milk from retailers, where up to 42.9% ($n= 15$) of the samples had fermented smell, the rest of categories had relatively sweet milky smell. Few samples from all categories had suspended physical particles. Samples from retailers showed high level of clotting to alcohol test compared to milk sample from vendor and farmers. This indicated increased milk acidity.

Table 5: Organoleptic properties of milk from vendors, retailers, smallholder dairy farmer in Arusha City and Meru District Council, Tanzania (October – December, 2012)

Parameter assessed	Category	Number (%) of samples		
		Vendor	Retailer	SDF
Colour	Yellowish white	34 (97.1)	27 (77.1)	33 (94.3)
	Creamy	1 (2.9)	10 (28.1)	2 (5.7)
Smell	Normal	31 (88.6)	20 (57.1)	32 (91.1)
	Fermented	2 (2.7)	15 (42.9)	1 (2.9)
	Abnormal	2 (5.7)	0 (0.0)	2 (5.7)
Physical dirt	Yes	9 (25.7)	4 (11.4)	8 (22.9)
	No	26 (74.3)	31 (88.6)	27 (77.1)
Milk clotting	Yes	10 (28.6)	16 (45.7)	2 (5.7)
	No	25 (71.4)	19 (54.3)	33 (94.3)

Key: SDF = Smallholder dairy farmers

4.3.2 Specific gravity (SG) and PH

The Specific gravity (SG) and pH of milk from the three categories are summarized in Table 6. Results showed that more samples from milk retailers and milk vendors had pH below minimum standard pH of 6.6 than milk from smallholders. On the other hand quite small number of samples from vendors and from smallholder dairy farmers had pH above the upper standard of pH 6.8 for cow milk. All the milk samples which had lower pH than the recommended minimum value clotted on alcohol test. The overall pH range of the milk handled for all categories was 6.0 - 6.9, out of which 34.3% was lower than the recommended pH of 6.6 for cow milk as given by East African Community standard.

Furthermore results showed the mean SG of milk samples was 1.032 ± 0.03 g/ml for smallholder dairy farmers, 1.028 ± 0.03 g/ml for vendors and 1.029 ± 0.01 g/ml for retailers. The overall SG range of milk handled for all categories was 1.0168 – 1.0394 g/ml, out of which 13.3% of the samples had SG below standard of 1.028 g/ml as given by East African Community standard. Based on individual category, 37.1% milk from vendors, 22.9% from retailers and 2.9% from smallholders had SG below standard. Few

samples, of milk from smallholder dairy farmers and from retailers showed SG above the maximum standard of 1.036 g/ml.

4.3.3 Microbiological quality

Microbiological qualities of milk from the three categories are summarized in Table 6. Results showed that the mean total viable count (TVC) of raw milk handled by street vendors was significantly ($p < 0.05$) higher than that of raw milk from retailers and smallholder dairy farmers. It was further indicated that 22.9% ($n=8$) of milk from smallholder dairy farmers, all milk from vendors and 71.6% ($n= 25$) of milk from retailers had TVC above the maximum standard of 2.0×10^5 cfu/ml for raw milk. Furthermore, the results indicated the overall (64.8% , $n= 68$) of all milk handled by the three categories, had higher TVC than the maximum recommended of 2.0×10^5 cfu/ml as given by East Africa Community standards.

Table 6: Mean, range and Standard deviation for physical assessment of milk and bacterial viable count in Arusha City and Meru District Council (October - December, 2012)

Parameter	Measure	Categories (n =35)			Overall (n =105)
assessed		SDF	Vendors	Retailers	
pH	Range	6.5 - 6.9	6.3 - 6.9	6.0 -6.7	
	Mean	6.7	6.7	6.4	
	Median	6.7	6.7	6.4	
	StdEv	0.094	0.187	0.205	
	< 6.6 (%)	2 (5.7)	13 (34.3)	22 (62.9)	34.3
	> 6.8 (%)	1 (2.9)	3 (8.9)	0 (0.0)	
SG (g/ml)	Range	1.027-1.039	1.018-1.033	1.017-1.037	
	Mean	1.032	1.028	1.029	
	Median	1.032	1.028	1.031	
	StdEv	0.003	0.003	0.005	
	< 1.028 g/l (%)	1 (2.9)	13 (37.1)	8 (22.9)	13.3
	> 1.036 g/l (%)	2 (5.7)	0	1 (2.9)	
TVC (cfu/ml)	Range	2.7x10 ³ - 2.9x10 ⁶	3.9x10 ⁵ -2.9x10 ⁸	0.0 - 4.2x10 ⁷	
	Mean	2.7 x10 ⁴	1.5x10 ⁷	5.4x10 ⁶	
	Median	1.2x10 ⁴	8.4x10 ⁵	8.4x10 ⁵	
	>2.0x10 ⁵ cfu/ml (%)	22.9	100	71.6	64.8

P < 0.05, Comparison of milk sample from any of the two categories

Key: SDF- Smallholder dairy farmers, SV - Street Vendors, R – Retailers, TVC - Total Viable count, SG - Specific gravity, StdEv- standard deviation

4.3.4 Common isolated bacteria from milk samples

Bacteriological milk assessment revealed a number of common bacteria including *E.coli*, and *Staphylococcus aureus* followed by *Corynebacterium* spp. and *Pseudomonas* spp., fungal infections was observed but not characterised in this study. The results showed that there were no significant variations of the common bacterial and fungal infection obtained in all categories of vendors, retailers and smallholder dairy farmers.

4.4 Awareness of Smallholder Dairy Farmers on Veterinary Drug Uses

4.4.1 Veterinary drugs used for animal treatment in smallholder dairy farms

Of the interviewed smallholder dairy farmers (28.6%, $n = 10$) reported to use different types of veterinary drugs to treat diseases in their animals as shown in Table 7. The commonly reported drugs were Oxytetracycline, Sulphonamides, Gentamycin injection and Kanamycin intra-mammary infusion. On the other hand, over 71.4%, ($n=25$) of the respondents were not aware of the types of drugs used for treatment of their animals. Most of the drugs reported by the respondents were being administered by livestock extension officers except for the few like Kanamycin intra-mammary infusion.

Table 7: The commonly used veterinary drugs by smallholder dairy farmers in Arusha City and Meru Distric Council, (October - December, 2012)

Name of drug	Number (%) of respondents
Penstreptomycin	0 (0.0)
Oxytetracycline	6 (17.1)
Sulphonamide	1 (2.9)
Kanamycin intra-mammary infusion	1 (2.9)
Gentamycin	2 (5.7)
Tyrosine	0 (0.0)

4.4.2 Awareness on veterinary drug residues to smallholder dairy farmers

The general awareness of veterinary drug residues in milk are summarized in Table 8. Of the interviewed respondents, all were aware on drug residue in milk. However, 97.1% ($n=34$) reported to comply with withdrawal periods for a period of less than a week. Furthermore, it was reported that most of the smallholder dairy farmers disposed the milk collected from animals under treatment, while others fed the milk to calves, dogs and cats. Moreover, 57.1% of the respondents were unaware of possible health hazards to consumers which are associated with veterinary drug residues in raw milk.

Table 8: General awareness of farmers on antibiotic residues in Arusha City and Meru District Council (October - December, 2012)

Parameter	Category	Number (%) of respondents
Milk contain drug residue	Yes	35 (100)
Sell milk with drug residue	Yes	1 (2.9)
	No	34 (97.1)
Withdrawal period	< week	32 (91.4)
	> week	2 (5.7)
	No	1 (2.9)
Use milk with drug residues	Dispose	23 (65.7)
	Feed calves, dogs & cats	11 (31.9)
	Feed family	1 (2.9)
Follow withdrawal period	Yes	34 (97.1)
	No	1 (2.9)
Knowledge on effects of drug residues	Yes	32 (92.4)
	No	3 (7.6)
Hazards associated with drug residue	Toxicity	8 (22.9)
	Allergic reaction	6 (17.1)
	Cancer	1 (2.9)
	Don't know	20 (57.1)

4.4.3 Antibiotic residues based on Delvotest

A total of 35 samples from the smallholder dairy farmers were qualitatively screened for antimicrobial residues using Delvo SP® kit. The results indicated that all 35 milk samples analysed were negative for antibiotic residues, indicating that the milk was free from the detectable concentrations of antibiotic residues.

CHAPTER FIVE

5.0 DISCUSSION

The overall objective of this study was to determine the quality of raw milk and assess the awareness of stakeholders to health risks associated with consumption of raw milk in Arusha City and Meru District Council. Generally, it was found that the level of awareness among smallholder dairy farmers, milk vendors and milk retailers on the health risk associated with raw milk consumption was high although what was practiced along the milk value chain predisposed milk to contamination. Organoleptic and physical quality analysis revealed that the milk was generally of poor quality. Bacteriologically, high TVC was encountered in high number of samples which was over and above the recommended East Africa Community standards (EACs, 2006). The predominant bacteria were *S. aureus* and *E.coli*. Encouragingly, all the milk samples analysed were free from antibiotic residues.

5.1 Awareness on Milk Quality and Risks Associated with Consumption of Raw Milk

Raw milk is known to be associated with health hazards like pathogenic bacteria which cause milk-borne diseases such as tuberculosis, brucellosis, typhoid fever, among others as was indicated by the respondents during this study. The findings from the present study showed majority of the respondents were aware that consumption of raw milk may predispose consumers to health hazards. However, the findings of the current study differ from previous studies by Karimuribo *et al.* (2005) and Kivaria *et al.* (2006) who reported smaller percentage of people who were aware that consumption of raw milk could result to health hazard, and that majority of people consume raw milk despite the fact that such studies went down to the level of household. Such low levels of awareness have also been reported elsewhere by Mosalagae *et al.* (2011) and Tebug *et al.* (2011). The high

awareness observed during this study could probably be due to the work done by livestock extension officers who were reported to work closely with the smallholder dairy farmers. Meanwhile, it was also associated with hearing from friends, other people or the media that raw milk may be sources of health problems to people as also reported by Kilango *et al.* (2012).

Furthermore, majority of the respondents used unboiled soured milk for making fermented milk for sale. This practice was likely to predispose milk consumers to health risks like raw milk. Similar finding have been reported by Omore *et al.* (2004). The factors that might have led to such practice could be due to little knowledge that fermentation of milk without prior boiling could predispose consumers to health hazards. Studies by Kurwijila (2006); Swai *et al.* (2010) and Kilango *et al.* (2012) reported that boiling of milk prior to consumption is the best approach to prevent milk-borne diseases especially in low income communities.

The common problem of milk spoilage especially handled by milk vendors and retailer as was observed during the current study could be due to delayed time before delivery or receipt and lack of cold chain. The finding indicated that the average time from milking to complete distribution while at room temperature ranged between 30 minutes to five or more hours. This gives time for the microbes to multiply in the milk and fasten the process of spoilage. Since no cooling facilities were available at farms or during transportation, a high temperature to raw milk implies that short shelf life to the milk. Storage temperature and time has also great effect on sensory properties of milk, pH and milk clotting (Murphy and Boor, 2010). Similar observation was reported by Karimuribo *et al.* (2005) and Majige (2007).

As part of hygienic measure to minimize contamination, respondents in the present study reported to use hot water and soap during hand and equipment cleaning. Other reports in Tanzania (Mdegela *et al.*, 2004) and Uganda (Byarugaba *et al.*, 2008) reported similar observations. However, the type of milk containers used was of poor quality as per Tanzania Bureau of Standards (TBS) and international (Codex) standards for milk handling. It was evident from the current study that the containers which were being used for milk storage, handling and transportation were of plastic type. Similar findings were also reported in previous studies in Tanzania (Kivaria *et al.*, 2006; Swai and Schoonman, 2011) and elsewhere (Omore *et al.*, 2002; Donkor *et al.*, 2007a; Oliver *et al.*, 2009; Mosalagae *et al.*, 2011). The Use of hot water and soap for cleaning the equipment is a common practice among individual at household level expected to remove milk remnants and sterilize the containers. However, based on the makeup and design of the containers (mainly used the closed ones with a small mouth as shown in Fig. 4), plastic containers are difficult to wash and sanitize especially at the corners and bottom. This may save as another potential source of microbial milk contamination. Poor quality of water used for washing and insufficient temperature may also jeopardize the expected container sterility. Comparable findings have also been reported elsewhere (Omore *et al.*, 2002; Khan *et al.*, 2011).

Physical dirt on raw milk were evidence of the extent to which visible insoluble matter has gained entrance to the milk and the extent to which such material has not been removed from milk (EACs, 2006; Pandey and Voskuil, 2011). All respondents in the current study reported to filter milk after milking, during selling and receiving. However, it was observed that the quality and type of milk filters used were poor (non-food grade plastic materials) and the pore size was large to the extent that smaller particles of sediments could pass through. According to Kurwijila *et al.* (2006) and (EACs, 2006), good raw

milk filters are white cotton piece of cloth and white lintine cotton discs respectively. Presence of dirty particles in milk further suggests contamination and downgrades the quality of milk. Also, dirty milk has a poor eye appeal to consumers and this may lead to lowered marketability and rejection for aesthetic reasons.

5.1.2 Physico-chemical quality of milk produced and sold in the study areas

The present study showed that most of the milk assessed in the current study had yellowish creamy white colour. Comparable studies in other countries (Khan *et al.*, 2008; Mubarack *et al.*, 2010) reported lower percentage of milk samples with yellowish white colour. This is the good news and shows that the milk is of normal quality in terms of colour (Khan *et al.*, 2008). Nevertheless, only 5.7% of the samples had abnormal smell especially samples from smallholder dairy farmers and milk vendors, which was mainly thought to be associated with poor milking hygiene, handling, storage and transportation.

Based on the results from alcohol test and pH, milk from milk vendors and milk retailer were relatively of poor quality. It was noted that, smallholder dairy farmers and milk vendors who were the main milk distributor handled milk at room temperature for long time. This practice enhances microbial activities which ultimately lead to increased milk acidity and hence clot on boiling. The findings were in line with other studies in Tanzania (Mdegela *et al.*, 2009) and elsewhere Mubarack *et al.* (2010). The pH of milk is a general measure of acidity. Low pH values indicate an acidification process caused mostly by bacterial spoilage (Pesta *et al.*, 2007). Milk acidity increases perceptibly within a short time after milking due to bacterial activity. The degree of bacterial contamination and the temperature at which the milk is kept are the key influencing factors of acid formation (Hossain *et al.*, 2011). On the other hand the amount of acid formation depends on the

cleanliness of the production and the temperature at which milk is stored (Hossain *et al.*, 2011).

The current study further showed significant number of samples from milk vendors and milk retailer, with specific gravity (SG) of milk below standard of 1.028 g/ml as given by East African community. Several factors are reported to influence SG of milk. These include breeds of cattle, stage of lactation, season of the year, feeds, health and physiological status of the animal (Pandey and Voskuil, 2011).

However, under normal circumstances, the low SG of milk in developing countries is associated with adulteration with water which is done mainly by the unscrupulous producers and middlemen with the purpose of increasing milk volume. This practice has side effects to the consumers' health, downgrading the milk and also shortening of shelflife of milk (Ali *et al.*, 2011). Low SG observed in the milk samples during the current study suggests that unfaithful milk dealer's maliciously added water to increase volume of milk. However, excessive milk shakes may result into milk aeration during transportation could also lower SG. Low SG for milk handled by milk vendor have also been reported by Kivaria *et al.* (2006); Swai and Schoonman (2011) and elsewhere (Omore *et al.*, 2002; Monsour *et al.*, 2012).

Generally, adulteration of milk by addition of water is reported as a potential source of contamination with microorganisms and harmful chemicals (Afzal *et al.*, 2011). Addition of water to milk may also reduce the nutritional and processing quality, palatability and marketing value of the milk which may lead to condemnation (Kivaria *et al.*, 2006). Therefore, deliberate disciplinary measures needed to be taken against such unfaithful milk dealers. In addition, there is a need of continued education to all people involved in

the milk value chain on how better quality milk can be achieved and its importance as far as the public health is concern.

5.1.3 Microbiological quality of milk

The quality of milk in the present study findings showed to be poor. The TVC values for milk handled by milk vendors and milk retailers were significantly higher ($p > 0.5$) than milk from smallholder dairy farmers. Based on the East African community standard (EACs, 2006) which has set a level of 2.0×10^5 cfu/ml as standard for acceptable microbiologically good milk, then milk from milk vendors and milk retailers was therefore of poor quality and could predispose the consumer to health hazards if consumed raw. The average value of TVC for smallholder was lower than the acceptable level (2.0×10^5 cfu/ml). Previous studies in Tanzania (Swai and Schoonman, 2011; Kilango *et al.*, 2012) have reported low TVC in milk associated with smallholders at farm level, as has also been reported from other studies (Gran *et al.*, 2002; Yirsaw, 2004; Khan *et al.*, 2008).

Bacterial load in milk indicates the degree level of hygiene practiced in the whole milk production process. Total bacterial count is a rough gauge to measure the quality of milk, herd health, efficacy of farm sanitation, milk handling and storage/transportation temperature (Fillimon *et al.*, 2011). Total bacterial counts further reflects the time elapsed since milking or the processing at ambient temperature while coliform bacteria like *E. coli* usually reflect faecal contamination due to poor hygiene. Bacterial contamination of raw milk can generally occur from three main sources; within the udder, outside the udder, and from the surface coming into contact with the milk. The high mean TVC values in milk from smallholder dairy farmers in the current study are linked with un-hygienic milking, milk handling, contamination from animal bedding, mixing of normal milk with the milk collected from the animal suffering from mastitis and lack of cold storage. However, high

bacteria count to the milk samples from vendors and retailers could be attributed to delayed delivered time. According to international regulations milk should be delivered and refrigerated within 3 hours (IDF 1990) after milking. However, lack of cold chain, long time for delivery, poor milk handling and transportation, and use of non-food grade plastic containers as observed to milk retailers and milk vendors could account for the high microbial load (Kivaria *et al.*, 2006; Donkor *et al.*, 2007b). The current study indicated that, out of the 105 milk sample analysed, 64.8 % had TVC values, above the acceptable level of 2.0×10^5 cfu/ml. Previous study (Kivaria *et al.*, 2006) reported 35.5 % of TVC above acceptable level, from milk selling points in Dar es Salaam city. Such finding strongly suggests a considerable proportion of milk consumers are at risk of milk-borne diseases and losses to farmers and other milk dealers due to rejection of spoiled milk.

The present study isolated various bacteria being dominated by *S. aureus* and *E.coli*. The findings are in agreement with the previous reports in Tanzania by Mdegela *et al.* (2004, 2005, 2009); Karimuribo *et al.* (2005) and Kivaria *et al.* (2006), and elsewhere by Yirsal, (2004); Donkor *et al.* (2007b); Byaruagaba *et al.* (2008); Mubarack *et al.* (2010) ; Sharma *et al.* (2012); Tebug *et al.* (2012) and Ayano *et al.* (2013). The possible causes of such bacterial contamination in milk are as stated in the previous paragraph. Many studies implicate *S. aureus* as the common mastitis causing organism in lactating cows (Karimuribo *et al.*, 2005; Kivaria *et al.*, 2006; Mdegela *et al.*, 2009). According to Kivaria and others (2006), *S. aureus* is the most important species likely to cause food poisoning through drinking contaminated milk. The bacteria are known to produce heat stable toxins. The main threat is from the fact that about 10% of mastitis staphylococci are known to be producers of enterotoxin. The toxins may be produced when *S. aureus* counts exceed 10^5 cfu/ml. Some reports have also associated *S.aureus* with gastroenteritis through

enterotoxins (Mubarack *et al.*, 2010). *Escherichia coli* O157:H7 can also cause diarrhoea in humans and consumption of raw milk is reported as among the important route of transmission (Kivaria *et al.*, 2006).

5.1.4 Awareness of smallholder dairy farmers on antimicrobial residue in raw milk

The present study showed that all smallholder dairy farmers were aware of the drug residue in milk. The high awareness of smallholders on drug residue could have been a result of the advice that was given by the veterinary practitioners and livestock extension officers who took a lead in delivery of veterinary services to dairy cattle. Similar study in Ethiopia (Syt, 2008) reported 78.4% of respondents were aware of drug residue in milk following treatment of sick dairy animal. Moreover, the findings of the current study indicated 97.1% of the respondents were aware of withdrawal periods. These findings were in line with other findings in Tanzania as have been reported by Katakweba *et al.* (2012) and Nonga *et al.* (2009), who both reported 90% awareness, while, Midenge, (2011) reported 72.2% awareness on drug withdrawal period. Similarly, Ombui (1994) reported that there was high awareness on withdrawal periods among farmers in Kiambu district, Kenya.

Compliance to withdrawal periods to antibiotics by smallholder dairy farmers in the study area was proved true when majority of the farmers admitted that they were told not to use milk or sell milk when the dairy cattle were under treatment and after treatment for a specific time period of around three to seven days depending on the type of drug used. On the other hand the presence of a livestock training institute (Tengeru Livestock Institute) around the area could be another reason for high awareness, following frequent field practical done by students in the area. Nevertheless, only 42.9% of smallholder dairy farmers were aware on the health effects associated with antibiotic residue in milk. The

result from this study concurs with other study (Midenge, 2011), and is in contrast to Syit, (2008); Sungura, (2010); Katakweba *et al.* (2012) and Widayati *et al.* (2012), who found high people's awareness on drug health effects. The reasons for the current study could be lack of information flow from the livestock extension officers to smallholder dairy farmers.

Qualitative analysis for antimicrobial residues using Delvotest SP® kit indicated that all the milk samples were negative for antimicrobial residues. This was an encouraging finding which suggests that farmers were following the withdrawal periods. Similar other studies (Karimuribo *et al.*, 2005; Kivaria *et al.*, 2006; Mdegela *et al.*, 2009) found lower levels of antimicrobial residues which were not significant for health effects almost similar to the current study. More results elsewhere (Omore *et al.*, 2002; Kang'ethe *et al.*, 2005; Syit, 2008; Movassagh and Karami, 2010; Kaya and Flazi, 2010; Addo *et al.*, 2011) have also reported low level of antibiotic residues. Differently, Kurwijila *et al.* (2006) reported high prevalence of antibiotic residues in a study conducted in Mwanza and Dar es salaam, Tanzania; similar to what was also reported in other countries by Shitandi, (2004) and Khaskheli *et al.* (2008). In Kenya, the problem of high prevalence of antibiotic residue in milk seems to be a threat to health of milk consumers (Shitandi, 2004; Kang'ethe *et al.*, 2005; Omore *et al.*, 2005).

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

From the findings of this study, it is therefore concluded that:

- i) The business of milk vendor is dominated by young males aged between 20 and 39 years, and that majority of them come from Meru District Council, which is the major source of milk sold in Arusha City.
- ii) There is high level of awareness that consumption of raw milk can predispose the consumer to health hazards, although what was practiced along the milk value chain predisposed milk to contamination.
- iii) Large proportion of people consume raw milk in the form of fermented milk
- iv) The quality of milk handled by vendors and retailers is of poor quality and hazardous for human consumption.
- v) Milk adulteration is practiced among the business agents particularly vendors and retailers.
- vi) Smallholder dairy farmers have high level of awareness on antibiotic drug residue following treatment of sick lactating cattle and hence obliged to observe the withdrawal period.
- vii) Drug residue in milk from smallholder dairy farmers is not a problem in the study area

6.2 Recommendations

Based on the findings from this study, the following are recommendations that could be done to quality of milk along the milk chain:

- i) Limited awareness on health risks associated with raw milk consumption amongst rural and urban communities needs to be addressed by various stakeholders like Tanzania Food and Drugs Authority (TFDA), Tanzania Dairy Board (TDB), Tanzania Milk Processing Association, (TAMPA), Tanzania Milk Producers Dairy Association (TAMPRODA), Tanzania Bureau of Standards (TBS) and Prime minister Office – Regional Authority and Local Government (PMO-RALG) health sector in order to safeguard community health.
- ii) Milk vendors play an important role in the milk business. However, there is a need for formation of Milk Vendors Association, which can organise them, recognise them and educate them on various areas related to general milk handling and hygiene.

REFERENCES

- Addo, K. K., Mensah, G. I., Aning, K. G., Nartey, N., Nipah, G. K., Bonsu, C., Akyeh, M. L. and Smits, H. L. (2011). Microbiological quality and antibiotic residue in informally marketed raw milk within the coastal savannah zone of Ghana. *Tropical Medicine and International Health* 16: 227 - 232.
- Afzal, A., Mahmood, M. S., Hussain, I. and Akhtar, M. (2011). Adulteration and microbiological quality of milk (A review). *Journal of Nutrition* 10: 1195 - 1202.
- Akinwumi, O. A., Olatoye, O. I., Odunsi, A. A., Omojola, B. A. and Rafiu, A. T. (2012). Effect of antibiotic residues on the physical quality of beef. In: *Proceeding of the 4th International Conference on Agriculture and Animal Science*. 2012, Oyo state, Nigeria. 10 - 776pp.
- Ali, A., Iftikhar, L., Mahmood, M. S. and Masood, A. (2011). Adulteration and microbiological quality of milk. *Pakistan Journal of Nutrition* 10: 119 - 202.
- Ayano, A. A., Hiriko, F., Simyalew, A. M. and Yohannes, A. (2013). Prevalence of subclinical mastitis in lactating cows in selected commercial farms of Holeta District, Ethiopia. *Journal of Veterinary Medicine and Animal Health* 5: 67 - 72.

Byarugaba, D. K., Nakavuma, J. L., Vaarst, M. and Laker, C. (2008). Mastitis occurrence and constraints to mastitis control in smallholder dairy farming system in Uganda. *Livestock Research for Rural Development*. Volume 20, Article #5. [http://www.lrrd.org/lrrd20/1/byar20005.htm] site visited on 04/06/2013.

Carter, G. R. and Wise, D. J. (2004). *Essentials of Veterinary Bacteriology and Mycology*. Blackwell, Hoboken. 290pp.

Codex Alimentarius Commission (CAC) (2004). *Code of Hygienic Practice for Milk and Milk Products*. CAC/RCP 57. Rome, Italy. 98pp.

Donkor, E. S., Aning, K. G. and Quaye, J. (2007a). Bacterial contaminations of informally marketed raw milk in Ghana. *Ghana Medical Journal* 41: 58 - 60.

Donkor, E. S., Aning K. G., Omore , A., Nurah, G. K., Osafo, E. L. K. and Staal, S. (2007b). Risk factors in the hygienic quality of milk in Ghana. *The Open Food Science Journal* 1: 6 - 9.

East African Community (EAC) (2006). *East African Standard, Raw cow Milk Specification*. pp. 1 - 2.

Filimon, M. N., Borozin, A. B., Bordean, D. M., Popescu, R., Gotia, S. R., Verdes, D., Morariu, F. and Treitli, S. (2011). Quality assessment of raw milk and pasteurized milk using microbiological parameters. *Animal Science and Biotechnologies* 44: 412 - 416.

- Food and Agriculture Organisation (FAO)/World Health Organisation (WHO), (2002).
Pan-European conference. In: *Food Safety and Quality*, 25–28 February 2002,
Budapest, Hungary. pp. 200 – 210.
- Goffová, Z. S., Kozárová, I., Máte, D., Marcincák, S., Gondová, Z. and Sopková, D.
(2012). Comparison of detection sensitivity of five inhibition tests for the
screening of amino glycoside residue in fortified milk. *Journal of Food
Science* 30: 314 - 320.
- Grace, D., Baker, D. and Randolph, T. (2009). Innovative and participatory risk based
approach to assess milk safety in developing countries. A case study in north
east India. In: *Proceeding of the International Association of Agricultural and
Economists (IAAE) Conference*. (Edited by Gdrace, D. *et al.*), 17 - 22 August
2009, Beijing, China. 1 – 10 pp.
- Gran, H. M., Mutukumira, A. N., Wetlesen, A. and Narhus, J. A. (2002). Smallholder
dairy processing in Zimbabwe: Hygienic practices during milking and
microbiological quality of milk at the farm and on delivery. *Food Control* 13:
41 - 47.
- Hossain, T. J., Alam, M. K. and Sikdar, D. (2011). Chemical and microbiological quality
assessment of raw and processed liquid market milks of Bangladesh. *Journal
of Food Science and Technology* 5: 6 - 17.
- IDF (1990). *Handbook on Milk Collection in Warm Developing Countries*. IDF special,
Brussels, Belgium. 148pp.

International Standards Organisation (ISO) (2007). *Microbiology of Food and Animal Feeding Stuffs- General Requirements and Guidance for Microbiological Examination*. No.7218. pp. 8 - 15.

ISO/FDIS (2001). *Milk and Milk Products – General Guidance for the Preparation of Samples, Initial Suspensions and Decimal Dilutions for Microbiological Examination*. International Organization for Standardization, Geneva, Switzerland. No. 8261 (E).

Kang'ethe, E. K., Oboge, G. D., Arimi, S. M., Kanja, L. W., Omore, A. O. and McDermott, J. J. (2005). Investigation of the risk of consuming marketed milk with antimicrobial residue in Kenya. *Food Control* 16: 349 - 355.

Karimuribo, E. D., Lughano, J., Kusiluka, L. J. M., Mdegela, R. H., Angolwisye, M., Kapaga, A. M., Sindato, C., Kambarage, D. M. (2005). Studies on mastitis, milk quality and health risks associated with consumption of milk from pastoral herds in Dodoma and Morogoro regions, Tanzania. *Journal of Veterinary Science* 6: 213 - 221.

Katakweba, A. A. S., Mtambo, M. M. A., Olsen, J. E. and Muhairwa, A. P. (2012). Awareness of human health risks associated with the use of antibiotics among livestock keepers and factors that contribute to selection of antibiotic resistance bacteria within livestock in Tanzania. *Livestock Research for Rural Development* Volume 24, Article #170. [<http://www.lrrd.org/lrrd24/10/kata24170.htm>] site visited on 04/03/2013.

Kaya, E. S. and Filazi, A. (2010). Determination of antibiotic residue in milk samples.

Research Article 16: 31 - 35.

Khan, M. T. G., Zinnah, M. A., Siddique, M. P., Rashid, M. H. A., Islam, M. A. and

Choudhury, K. A. (2008). Physical and microbial qualities of raw milk collected from Bangladesh. *Bangladesh Journal of Veterinary Medicine* 6: 217 - 221.

Khan, A. A., Massod, F. A. and Bhat, B. A. (2011). Bacteriological quality and safety of

raw milk in Kashmir valley. *Wayamba Journal of Animal Science* 3: 2102 – 5789.

Khaskheli, M., Malik, S. R., Arain, M. A., Soomro, H. A. and Arain, H. H. (2008).

Detection of β -lactam antibiotic residue in market milk. *Pakistan Journal of Nutrition* 7: 682 - 685.

Kilango, K., Makita, K., Kurwijira, L. R. and Grace, D. (2012). Boiled milk, food safety

and the risk of exposure to milk borne pathogens in informal dairy markets in Tanzania. In: *Proceedings of the World Dairy Summit Conference*. (Edited by Kilango, K. *et al.*), 4 - 5 November 2012, Cape Town, South Africa. 1 – 13pp

Kivaria, F. M., Noordhuizen, J. P. T. M. and Kapaga, A. M. (2006). Evaluation of the

hygienic quality and associated public health hazards of raw milk marketed by smallholder dairy producers in the Dar es Salaam region, Tanzania. *Tropical Animal Health Production* 38: 185 - 94.

- Knutson, D. R., Currier, W.R., Ribera, L. and Goeringer, P. (2010). Asymmetry in raw milk safety perceptions and information: Implications for risk in fresh produce marketing and policy. In: *The Economics of Food, Food Choice and Health*. (Edited by Knutson, D.R. *et al.*), 15 - 17 September 2010, Freising, Germany. 1 – 22pp.
- Koo, I. (2008). A guide to milk-borne infectious disease. *Journal of Dairy Science* 84: 1 - 11.
- Kothari, C. R. (Eds.) (2004). *Research Methodology, Method and Techniques*. New age international publishers, India. 401pp.
- Kurwijila, L. R. and Henriksen, J. (1998). Milk supply to urban centres in Tanzania with particular reference to the City of Dar es Salaam. [<http://www.FAO.org>] site visited on 12 /5/2012.
- Kurwijila, R. L., Omore, A., Staal, S. and Mdoe, N. S. Y. (2006). Investigation of the risk of exposure to antimicrobial residue present in marketed milk in Tanzania. *Journal of Food Protection* 69: 2487 - 2492.
- Kurwijila, L. R. (2006). *Hygienic Milk Handling, Processing and Marketing: Reference Guide for Training and Certification of Small-Scale Milk Ttraders in Eastern Africa*. International Livestock Research Institute (ILRI), Nairobi, Kenya. 27pp.

Kurwijila, L. R., Omore, A. and Staal, S. (2009). *Dairy Sub Sector Development Strategy, East Africa Regional Initiatives in Value Chains*. Lesson from on- going R & D initiatives in Dairy value chains, Rural Livelihood Development Company. Version Board 17 - 04 - 2009. [www.rldc.co.tz/docs/rldcdairy] site visited on 24/3/2012.

Lingathurai, S. and Vellathurai, P. (2011). Bacteriological quality and safety of raw cow milk in Mandurai, south India. *Webmedcentral Microbiology* 1: 1 - 10.

Lore, T. A., Kurwijila, L. R. and Omore, A. (eds.) (2006). *Hygienic Milk Production. A Training Guide for Farm-level Workers and Milk Handlers in Eastern Africa*. International Livestock Research Institute (ILRI), Nairobi, Kenya. 13pp.

Majige, K. N. (2007). Improving milk handling and quality control- a way forward for increasing income to the community. The case of Serengeti dairy co operative society in Bunda district, Tanzania. Dissertation for Award of MSc Degree at The Southern New Hampshire University and The Open University of Tanzania, Dar es Salaam, Tanzania. 110pp.

Mansour, A. I. A., El-Loly, M. M. and Ahmed, O. R. (2012). A preliminary detection of physical and chemical properties, inhibitory substances and preservatives in raw milk. *Journal of Food Safety* 14: 93 - 103.

- Mdegela, H. R., Kusiluka, J. M. L., Kapanga, M. A., Karimuribo, D. E., Turuka, M. F., Bundala, A., Kivaria, F., Kabula, B., Manjurano, A., Loken, T. and Kambarage, M. D. (2004). Prevalence and determinants of mastitis and milk borne zoonoses in smallholder dairy farming sector in Kibaha and Morogoro districts in eastern Tanzania. *Journal of Veterinary Medicine B* 51: 123 - 128.
- Mdegela, R. H., Karimuribo, D. E., Kusiluka, L. J. M., Kabula, B., Manjurano, A., Kapaga, A. M. and Kambarage, D. M. (2005). Mastitis in smallholder dairy and pastoral cattle herds in the urban and peri-urban areas of the Dodoma municipality in central Tanzania. *Livestock Research for Rural Development* 17: 11.
- Mdegela, R. H., Ryoba, R., Karimuribo, E. D., Phiri, E. J., Løken, T., Reksen, O., Mtengeti, E., Urio, N. A. (2009). Prevalence of clinical and subclinical mastitis and quality of milk in smallholder dairy farms in Tanzania. *Journal of the South African Veterinary Association* 80: 163 - 168.
- Midenge, B. Y. (2011). Awareness on recommended veterinary drugs withdrawal period among small scale dairy cattle farmers in Kinondoni Municipality. Dissertation for Award of MSc Degree at Muhimbili University of Health and Allied Sciences, Dar es salaam, Tanzania. 90pp.
- Mmbando, L. M. G. (2004). Investigation of Oxytetracycline use and abuse: Determination of its residues in meat consumed in Dodoma and Morogoro Municipalities. Dissertation for Award of MVM Degree at Sokoine University of Agriculture, Morogoro, Tanzania. 140pp.

- Mohamed, O. M. A and Alhagaz F. M. M. (2011). The impact of applying some hygienic practices on raw milk quality in Khartoum state, Sudan. *Journal of Agriculture and Biological Sciences* 7: 169 - 173.
- Mosalagae, D., Pfukenyi, D. M. and Matope, G. (2011). Milk producer's awareness of milk-borne zoonoses in selected smallholder and commercial dairy farms of Zimbabwe. *Tropical Animal Health and Production* 43: 733 - 739.
- Movassagh, M. H. and Karami, A. R. (2010). Determination of antibiotics in bovine milk in Tabriz, Iran. *Global Veterinarian* 5(3): 195 - 197.
- Msangi, B. S. J. (Ed.) (2006). Value-chain analysis and socio-economic assessment of the dairy industry in Tanzania. In: *Proceeding of the Tanzania Dairy Board, The six National Dairy Development*. (Edited by UNDP/ Business care services/ Tetrapak), 2 June, 2006, Morogoro Hotel, Morogoro, Tanzania. 14 – 25pp.
- Mubarack, H. M., Doss, A., Dhanabalan, R. and Balachander, S. (2010). Microbial quality of raw milk samples collected from different villages of Coimbatore district, Tamilnadu, south India. *India Journal of Science and Technology* 1: 61 - 63.
- Murphy, S. C. and Boor, K. J. (2000). Trouble shooting sources of high bacteria counts in raw milk. *Dairy Food Environmental Sanitation* 20: 606 - 611.
- Njombe, A. P., Msanga, Y. N., Temba, A. E. and Tsoxo, M. (2009). Effort to increase improved dairy cattle in Tanzania. [www.mifugo.go.tz] site visited on 12/03/2013.

- Nonga, H. E., Mariki, M., Karimuribo, E. D. and Mdengela, R. H. (2009). Assessment of antimicrobial usage and antimicrobial residues in broiler chicken in Morogoro municipality, Tanzania. *Pakistan Journal of Nutrition* 8: 203 - 207.
- Oliver, S. P., Boor, J. K., Murphy, C. S. and Murinda, E. S. (2009). Food safety hazards Associated with consumption of raw milk. *Food-borne Pathogens and Disease* 6: 793 - 806.
- Ombui, J. N. (1994). Antibiotic residues in milk received by dairy cooperatives societies in Kiambu district, Kenya. *East African Medical Journal* 71: 628 - 630.
- Omore, A. O., Arimi, S. M., Kang'ethe, E. K., McDermott, J. J. and Staal, S. J. (2002). Analysis of milk-borne public health risks in milk markets in Kenya. In: *Proceeding of the Annual Symposium of the Animal Production Society of Kenya*, 9 - 10 May, 2002, Kari-nahrs, Naivasha. 21 – 30pp.
- Omore, A., Staal, S. J., Osafo, E. L. K., Kurwijila, R. L., Barton, D., Mdoe, N., Nurah, G. and Aning, G. (2004). *Market Mechanisms, Efficiency, Processing and Public Health risk in Peri –urban Dairy Product Market: Synthesis of findings from Ghana and Tanzania Final Report for LPP Project R7321*. 127pp.
- Omore, A., Lore, T., Staal, S., Kutwa, J., Ouma, R., Arimi, S. and Kangethe, E. (2005). *Addressing the Public Health and Quality Concerns Towards Marketed Milk in Kenya*. SDP Research and Development Report 3 Smallholder Dairy (R & D) Project. 44pp.

Pandey, G. S. and Voskuil, G. C. S. (2011). *Manual on Milk Safety, Quality and Hygiene*.

Golden Valley Agricultural Research Trust, Zambia. 52pp.

Pecou, A. and Diserens, J. (2011). Strategies for detecting antibiotic residue in milk. In:

Guidance on the Application of Screening and Confirmatory Methods in Integrated Dairy Chain Management. (Edited by SCAMAC – project group),

27 may 2011, Lyon, France. 1 – 17pp.

Pesta, M., Williams, P., Zampa, N., Garry, E. and Ouattara, G. (2007). The effects of raw

milk storage condition on freezing point, pH and impedance. *Advanced*

Instrument Inc. pp. 1 - 3. [<http://www.aicompanies.com>] site visited on 4/6/2013.

Sarkiyayi, S. and Sheshu. (2011). Effect of boiling and fermentation on the nutrient

composition of raw milk in Kaduna Metropolis, Nigeria. *Research Journal of Chemical Sciences* 1: 81 - 84.

Sharma, N., Jin Rho, G., Ho Hong, Y., Kang, T. Y., Lee, H. K., Young Hur, T. and Jeong,

D. K. (2012). Bovine mastitis: An Asian perspective. *Asian Journal of Animal and Veterinary Advances* 7: 454 - 476.

Shitandi, A. (2004). Risk factors and control strategies of antibiotic residues in milk at farm level in Kenya. Thesis for Award of PhD Degree at the Swedish

University of Agricultural Sciences, Uppsala, Sweden. 36pp.

- Sungura, K. H. (2010). Assessment of veterinary drug use and determination of antimicrobial residues in broiler chicken meat in urban district, Zanzibar, Tanzani. Dissertation for Award of MSc Degree at Sokoine University of Agriculture Morogoro, Tanzania. 62pp.
- Swai, E. S., Schoonman, L. and Daborn, C. J. (2010). Knowledge and attitude towards zoonoses among animal health workers and livestock keepers in Arusha and Tanga, Tanzania. *Tanzania Journal of Health Research* 12: 282 - 288.
- Swai, E. S. and Schoonman, L. (2011). Microbial quality and associated health risks of raw milk marketed in the Tanga region of Tanzania. *Tanzania Journal of Tropical Biomedicine* 11: 217-222.
- Swai, E. S. and Karimuribo, E. D. (2011). Smallholder dairy farming in Tanzania: Current profiles and prospects for development. *Outlook on Agriculture* 40: 21 - 27.
- Syit, A. D. (2008). Detection and determination of Oxytetracycline and penicillin antibiotic residue levels in Bovine bulk milk from Debrezeit and Nazareth dairy farms in Ethiopia. In: *Proceedings of the 1st International Technology, Education and Environment Conference African Society for Scientific Research*. (Edited by Human Resource Management Academic Research Society). May 2008, Addis Ababa, Ethiopia. 325 – 337pp.

- Tebug, S. F., Njunga, G. R., Mapemba, J. P., Chagunda, M. G. G. and Wiedemann S. (2011). Smallholder dairy farmers' awareness and risk behaviour for milk-borne zoonoses transmission in Northern Malawi. In: *Proceedings of Colloquium on Zoonoses and Neglected Infectious Diseases of Africa*. 1 - 4 November 2011, Johannesburg, South Africa. 1pp.
- Tebug, S. F., Kasulo V., Chikagwa-Malunga, S., Wiedemann S., Roberts, D. J. and Chagunda, M. G. G. (2012). Smallholder dairy production in Northern Malawi: production practices and constraints. *Tropical Animal Health Production* 44: 55 - 62.
- Tanzania Bureau of Standards (2007). *Microbiology of Food and Animal Feeding Stuffs- Method for Enumeration of Microorganisms - Colony Count Technique at 30 °C. Tanzania Standards Specification*. No 118:2007.
- Widayati, A., Suryawati, S., De Crespigny, C. and Hiller, J. E. (2012). Knowledge and beliefs about antibiotics among people in Yogyakarta City, Indonesia. *Antimicrobial Resistance and Infection Control* 1: 1 - 38.
- Yilma, Z. (2012). Microbial properties of Ethiopian marketed milk and milk products and associated critical points of contamination: An epidemiological perspective. *East Africa Dairy Development Programme* 15: 298 - 318.
- Yirsaw, W. A. (2004). Bacteriological quality of bovine milk in small holder dairy farms in Debre zeit, Ethiopia. Dissertation for Award of MSc Degree at Addis Ababa University, Ethiopia. 56pp.

APPENDICES

Appendix 1: Vice Chancellor's Letter



KIBALI CHA KUFANYA UTAFIGITI NCHINI TANZANIA
CHUO KIKUU CHA SOKOINE CHA KILIMO
OFISI YA MAKAMU WA MKUU WA CHUO
S.L.P. 3000, MOROGORO, TANZANIA
Simu: 023-2604523/2603511-4; Fax: 023-2604651, MOROGORO

Kumb. Zetu : SUA/ADM/R.1/8

Tarehe 13/9/2012

Mkurugenzi Mtendaji,
Halmashauri ya Wilaya ya Meru,
ARUSHA.

UTAFITI WA WAALIMU NA WANAFUNZI WA CHUO KIKUU

Madhumuni ya barua hii ni kumtambulisha kwako Mwanafunzi Juma Bukuku ambaye ni mwanafunzi wa mwaka wa pili wa shahada ya Uzamili MSc. (PHFS) katika Chuo Kikuu cha Sokoine cha Kilimo. Huyo hivi sasa yuko katika shughuli za utafiti.

Chuo Kikuu cha Sokoine cha Kilimo (SUA) kilianzishwa na Sheria ("Universities Act No.5 of 2005") na Hati Ridhia ("SUA Charter, 2007") ambayo ilianza kutumika Januari 1, 2007. Hati Ridhia ilichukua nafasi ya Sheria Na.6 ya mwaka 1984. Moja ya majukumu ya SUA ni kufanya tafiti mbalimbali na kutumia matokeo ya tafiti hizo. Kwa sababu hiyo, waalimu, wanafunzi na watafiti wa Chuo hufanya tafiti mbalimbali katika nyakati zinazostahili.

Ili kufanikisha utekelezaji wa tafiti hizo Makamu wa Mkuu wa Chuo SUA amepewa mamlaka ya kutoa vibali vya kufanya utafiti nchini kwa waalimu, wanafunzi na watafiti wake kwa niaba ya Serikali na Tume ya Sayansi na Teknolojia.

Hivyo basi tunaomba umptie Mtaalamu aliyetajwa hapo juu msaada atakaohitaji ili kufanikisha uchunguzi wake. Gharama za malazi na chakula chake pamoja na usafiri wake atalipia mwenyewe kutokana na fedha alizopewa na Chuo Kikuu. Msaada anaohitaji zaidi ni kuruhusiwa kuonana na viongozi na wananchi ili aweze kuzungumza nao na kuwauliza maswali aliyo nayo.

Kiini cha Utafiti wa mwanafunzi aliyetajwa hapo juu ni: "ASSESSMENT OF QUALITY OF RAW MILK".

Sehemu anazofanyia utafiti huo ni: Kata ya Nkoanrua.

Ikiwa kuna baadhi ya sehemu ambazo zinazuiliwa, ni wajibu wako kuzuia zisitembelewe.

Muda wa Utafiti huo ni kuanzia tarehe Oktoba, 2012 hadi Septemba, 2013.

Ikiwa utahitaji maelezo zaidi tafadhali wasiliana nami.

Wasalaam,

Prof. Gerald C. Monela

MAKAMU WA MKUU WA CHUO

Nakala: Mtafiti: Juma Bukuku

MAKAMU WA MKUU WA CHUO
CHUO KIKUU CHA SOKOINE CHA KILIMO
S. L. P. 3000
MOROGORO, TANZANIA

Appendix 2: District Directors permit

HALMASHAURI YA WILAYA YA MERU
(Barua zote za kiofisi zitumwe kwa Mkurugenzi Mtendaji)

Telegram: Meru
 Simu: (+ 255) 027 254-1112
 Faksi: (+ 255) 027 254-1112
 Barua pepe: merudc@yahoo.com



Ukumbi wa Wilaya,
 P.O. Box 462,
USA RIVER

MM/MER/N10/5VOL11/72

11/12/2012

AFISA MTENDAJI WA KATA
KATA NKOANRUA

YAH: KIBALI CHA KUFANYA UTAFITI MR. JUMA BUKUKU

Husika na mada tajwa hapo juu.

Napenda kukujulisha kuwa ndugu, JUMA BUKUKU kutoka chuo kikuu cha **Sokoine** cha kilimo Morogoro amepewa kibali cha kufanya utafiti wa "ASSESSMENT OF QUALITY OF RAW MILK" katika kata yako. Ninaomba umpe msaada unaostahili kwa kumruhusu kufanya shughuli hiyo muhimu kwake na Taifa kwa ujumla.

Kibali hiki kinaanza mwezi October 2012 hadi September 2013.
 Nategemea utampatia ushirikiano wa dhati.

B.P. MUSHI

Kny: **MKURUGENZI MTENDAJI**
HALMASHAURI YA WILAYA YA MERU

Ndugu-JUMA BUKUKU
RESEARCHER

Appendix 3: Questionnaires

A: Questionnaire for Cattle Keepers

Assessment of awareness on health risk of raw milk in Arusha city and Meru district

Date of interview.....Respondent's ID No.....

Interviewers Initials.....

A.1. General Information

1.1 Name of head of the household.....

1.2 Sex F/M.....

1.3 Village.....

1.4 Ward.....

1.5 District.....

1.6 Level of education: Primary (P).....Secondary (S).....Graduate
(G).....Professional (PR)

1.7 What is your main current occupation?

i) Civil servant.....ii) Engage in business.....iii) Self employed..... iv) Others
(specify).....

A.2 General Health status of cattle and uses of antibiotics

2.1 Type of animal keeping practiced

i) Free grazing (Fr)..... (ii) Zero grazing (ZG)..... (iii) Mixed (M).....

2.2 Housing (i) Open house (OH)..... (ii) Closed house (CH).....

2.3 Number of milking herds

2.4 How many dairy cattle do you keep.....

2.5 What are the common diseases associated with your cattle?

i) Mastitis.....(ii) Foot and mouth disease (FMD).....(iii) East coast
fever (ECF)..... iv) Respiratory disease..... (v) Unspecified diseases.....(vi
Diarrhoea.....vii) Helminthosis ... (viii) Trypanosomiasis(ix) Contagious
Bovine Pleuropneumonia..... (x) Others (specify).....

2.6 How do you overcome the disease problem to your animals?

i) Teat myself..... (ii) Call a livestock officer and/or veterinarian
to attend them..... (iii) Others (specify).....

A.3 Milk production

1. Milking place;

- i) Within the cows house (WCH)..... (ii) Outside cow house (OT).....

2. Type of utensils used for milking process.....

3. Utensils cleaned before milking

- i) Just with cold water (CW).....

- ii) Soap and cold water (SCW).....

- iii) Soap and hot water (SHW).....

4. Does the milker wash hands before milking Y/N

5. If Yes in question 4, how does the milker wash hands before milking?

- Just with cold water (CW)..... .ii) Soap and cold water (SCW)

- iii). Soap and hot water (SHW).....

6. Is the udder washed before milking Y/N.....

7. If yes how is the udder washed before milking

- i) Just with cold water (CW)..... (ii) Soap and cold water (SCW).....iii) Soap and hot water (SHW).....

8. What is the total milk production volume per dayLts

3.2 Handling, storage and Transportation

1. Is milk filtered after milking (Y/N)

2. If yes in question 1, what do you use for filtering (i) Use white clean cloth (WC).....(ii)other

3. How is milk stored (i) In plastic containers (PC).....(ii) Stainless steel containers (SSC).....(iii) Aluminium containers (AC).....(iv) Others (specify).....

4. How do the buyers transport the milk bought (i) using car (car) (ii) Bicycle (Bc) ... (iii) Motorcycle (MC) ... (iv) Others (specify)...

5. Where do you think the buyer send the milk bought

A.4 Awareness of risks associated with raw milk

1. Do you know that consumption of raw milk can cause human illness(Y/N).....?

2. If yes what are the common diseases caused through consumption of raw milk.....

3. What are the common clinical signs of hazards from raw milk consumptions.....?
4. How can you prevent such hazards.....

A.6 commonly used antibiotics & health risks of drug residues

1. What are the common diseases do you encounter to your herds?
 - i). Mastitis (MST)..... (ii) Foot and Mouth Disease (FMD).....
 - iii) Anaplasmosis (AN)..... (iv) Others.....
2. What type of antibiotic is given to your dairy cattle? (The researcher may ask/request to see if there are any empty bottles/packs)
 - i). Pen streptomycin..... ii) Tetracycline (OTC & CTC)..... iii). Sulphonamide
 - (v) Kanamycin intramammary infusion (vi) Tyrosine..... (vii) Gentamycin.....
 - (viii) Other (specify).....
3. Who administers drugs to your dairy cattle when they fall sick?
 - i). A livestock officer (LO)..... (ii) Other (specify).....

A.7 Awareness on recommended veterinary drugs withdrawal periods and drug residue effects

1. Do you sell milk immediately after last dose of cattle treatment?
 - i). Yes ☐ (ii). No ☐
2. If the answer in question 1 is **No**, why not selling milk immediately after last dose of treatment?
 - i) Observe veterinary drugs withdrawal periods..... ii) Milk contains veterinary drugs..... iii) Others (specify)
2. If the answer in question 1 is **No**; how long do you wait before starting to sell the milk from a cow under treatment?
 - i) Less than one week..... ii) More than one week.....
 - iii) Stop from selling as per drug manufacturer's recommendations....
 - iv) Other (specify).....
3. What do you do with milk from an animal that has just finished last dose of treatment?
 - i) Pour the milk..... (ii) Give them to pet animals like dogs and cats...
 - (iii) Other (specify).....
4. Do you know the drug withdrawal period? (*The researcher or research assistant should clarify the term*)

- (i). Yes..... (ii). No.....
5. If answered **Yes**, do you follow it? (i) Yes..... (ii) No.....
6. Is there any health effects if a person consumes milk products with drug/ antibiotics residues? i) Yes... (ii) No (iii) I don't know.....
7. If answered **Yes** in question 7 list down the health effects you know which may be caused by drugs /antibiotics residues in milk
- i) Allergic reactions to some sensitive individuals (AR) ... (ii) Direct toxic effects (DT)...
- (iii) Bacterial resistance to antibiotics (BR).....
- v) Cancer (C)..... (v) Others (specify).....

B: Questionnaires for Milk Vendors

Assessment of awareness on health risks of raw milk in Arusha City and Meru District

Date of interview.....Respondent's ID No.....

Interviewers Initials.....

1.B General information

1. Name of the vendor..... 2.Sex (M/F)
3. Age (Yrs)..... 4. Village
5. Ward.....6. District.....
7. Level of education:
- Primary (P)..... Secondary (S).....Graduate (G).....Professional PR)...
8. How long have you been vending raw milk?

B.2 Handling, Storage and transportation

1. Where do you buy milk (i) Local farmers (LF)(ii) Collection centre(CC) (iii) Larger farmers (LG).....
2. Where do you sell your milk (i) Households (HH)..... (ii) Kiosk (KK)... (iii) Restaurant (RS)... (iv) Processing industry (PI).... (v) Others...
3. Is milk filtered before buying; (i) Yes... (ii) No ... (iii) don't know...
4. When do you collect milk (i) evening (Ev)... (ii) Early Morning (EM)...
- 5.If evening how is milk stored;(i) Refrigerated (R),(ii)Room temperature (RT) (iii) Others
6. Have you ever faced problems of milk becoming spoiled? Yes/No
7. If Yes how often? i) Regularly...ii) Once...iii) Twice...iv) others.....

8. What do you do with the milk which is spoiled?

- i) I sell at a low price ... ii) I feed my family.....iii) I mix with fresh milk and sell..... (iv) I dispose it.....

9. How do you transport the milk bought; (i) Bicycle (B)..... (ii) Car (C)..... (iii) Motorcycle (MC).....(iv) Others

10. Container material for carrying milk (i) Plastic (PC).....(ii) Stainless steel (SSC).....(iii) Aluminium (AC)..... (iv) Other.....

11. Status of the container (the research to assess).....

12. How do you clean the containers

- (i) Just with cold water (CW).....(ii) Soap with cold water (SCW)...(iii) Soap with hot water (SHW).....(iv) Other (specify).....

B.3 Awareness of risks associated with raw milk consumption

1. Do you know that consumption of raw milk can cause human illness(Y/N).....?

2. If yes what are the common diseases caused through consumption of raw Milk.....

3. What are the common clinical signs of hazards from raw milk

Consumptions.....

4. How can you prevent such hazards.....?

C: Questionnaires for Milk Retailers

Assessment of awareness on health risk of raw milk in arusha City and Meru District

Date of interview.....Respondent's ID No.....

Interviewers Initials.....

1. C General information

1. Name of the retail business Kiosk/restaurant
2. Name of the owner.....2. Sex (M/F)
3. Name of the street the business is carried.....4. Ward.....
5. District/City.....
6. Level of education: Primary (P).....Secondary (S).....Graduate (G).....Professional (PR).....

C.2 Handling, Storage and transportation

1. Where do you obtain milk (i) Street vendors (SV) (ii) Direct from Small Farmers (SF)..... (iii) Others (specify).....
2. Who are your customers
3. Have you ever faced problems of milk becoming spoiled?
 - i). YES..... ii).NO.....
4. If Yes, how often? i) Regularly... (ii) Once... (iii) Twice... iv) Others....
5. If yes in question 3, how do you determine for good milk (non- spoiled) milk
 - (i) By visual (V)..... (ii) Smell (S)..... (iii) Taste (T)..... (iv) Colour (C) (v) Others (specify).....
6. What do you do with the milk which is spoiled?
 - i) I sell at a low price.....ii) I feed my family.....iii) I mix with fresh milk and sell..... iv) I dispose it
7. How do you use the milk (i) making tea drink..... (ii) Fermented milk..... (iii) Yoghurt.....(iv)Fresh milk drink.....
8. If make fermented milk or Yoghurt what processing method do you perform?
 - (i) Pasteurize..... (ii) Boil..... (iii) None.....

C.3 Awareness of risks associated with raw milk

1. Do you know that consumption of raw milk can cause human illness(Y/N).....?
2. If Yes what are the common diseases caused through consumption of Raw milk.....
3. What are the common clinical signs of hazards from raw milk consumptions.....?
4. How can you prevent such hazards