EVALUATION OF HYGIENIC PRACTICES AND ESTABLISHMENT OF CRITICAL CONTROL POINTS AND RAW MILK QUALITY IN THE SMALLHOLDER SUPPLY CHAIN OF NAKURU AND NYANDARUA COUNTY, KENYA

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DECLARATION AND APPROVAL

DECLARATION

I hereby declare that this is my original work and has not been presented for examination in this or any other institution.

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APPROVAL

This thesis has been presented for examination with our approval as University Supervisors

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DEDICATION

I dedicate my dissertation work to God almighty, my family and many friends.

This work is dedicated to my daughters Lucy and Peninah, who are very special and have never left my side, for their enormous support even when my studies seemed an uphill task. A special feeling of gratitude goes to my loving parents, Paul and Peninah Ndungu, my brothers and sisters for moral support throughout writing this thesis and my life in general. Their words of encouragement and push for tenacity ring in my ears.

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ABSTRACT

The milk marketed in Kenya has been reported to be of poor quality and does not meet national and international standards due to high bacterial load, high somatic cell count, adulteration and antibiotic residues. The study investigates the raw milk quality, adherence to hygienic code of practice and identifies the critical control points for improved raw milk quality in the smallholder supply chain. Compliance to hygienic code of practice was assessed using questionnaires. The factors that could contribute to raw milk quality deterioration were identified through observations using the hazard analysis critical control point principles. To establish the raw milk quality, physico-chemical and microbiological tests were carried out. Statistical analysis for laboratory experimentation involved analysis of variance and means were separated using least significance difference whenever the sampling level effect was significant $P \leq 0.05$. The results indicated low conformance to the hygienic code of practice including ineffective hand washing procedure before milking, use of plastic containers in milk handling, unawareness of the food safety concerns related to antibiotic residues in milk, delayed milk delivery and use of reusable udder cloth. The critical control points identified included milking, bulking milk in fifty liters can, transportation, the reception platform and the cooling tank. A quality control plan for the smallholder supply chain was developed. The means separation indicated that the average total plate count was 6.72×10^8 and 1.37×10^7 cfu/ml for Ngorika and Olenguruone respectively while the coliform count was an average of 3.18×10^6 cfu/ml and 1.34×10^5 in Ngorika and Olenguruone respectively. The antibiotic residues analysis was conducted using Delvo test and the positive results were 35% and 54% for Ngorika and Olenguruone respectively. Somatic cell count was analyzed using California Mastitis Test and 65% and 55% of the samples analyzed were within a range of 150-500 somatic cells count/1,000 ml. Water adulteration was analyzed using cryoscopy and 23.8% and 36.8% were positive for Ngorika and Olenguruone, respectively. There were no significant $P \leq 0.05$ correlation between total plate count and the resazurin test at route level for both locations. The study verified non-compliance to the hygienic code of practice and nonconformance of raw milk quality to the Kenyan standards. The study however developed the critical control plan which if adopted could guide on the corrective action at every node in the collection chain and improve the quality of raw milk in Kenya.

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LIST OF ABBREVIATIONS

AFNOR	Association French Normalization Organization Regulation
AOAC	Association of Official Analytical Chemists
CBE	Collection and Bulking Enterprises
CIP	Cleaning In Place
CC	Coliform Count
CCP	Critical Control Points
CFU	Colony Forming Units
DNA	Deoxyribonucleic Acid
EAS	East Africa Standards
GAP	Good Agricultural Practices
GDP	Gross Domestic Product
HACCP	Hazard Analysis Critical Control Point
HTST	High Temperature Short Time
IDF	International Dairy Federation
KAVES	Kenya Agricultural Value Chain Enterprises
KDB	Kenya Dairy Board
KEBS	Kenya Bureau of Standards
KMDP	Kenya Market-led Dairy Programme
LA	Lactic Acid
MoLD	Ministry of Livestock Development
MQT&T	Milk Quality Tracking and Tracing
MSNF	Milk Solids Not Fat
QBMPS	Quality Based Milk Payment System
RT	Resazurin Test
SCC	Somatic Cell Count
SOP	Standard Operating Procedures
TPC	Total Plate Count
USAID	United States Agency for International Development
PHD	Public Health Division
PRP	Pre Requisite Programs

CHAPTER ONE

INTRODUCTION

1.1 Background of the study

Kenya's dairy industry, the single largest livestock production sub-sector contributes 14% of the agricultural gross domestic product (GDP) and 3.5% of the total GDP (Corné *et al.,* 2016). According to Muriuki (2011), the dairy production systems differ in their sizes of operation, level of management and use of inputs and therefore can be classified as large, medium or small scale. Smallholder dairy farmers dominate the industry at the production level with more than 1 million smallholder dairy farmers, accounting for more than 70% of the total milk production. The smallholders practice zero and semi zero grazing in 3 to 5 acres of land and have about 2 to 5 cattle, each yielding an average of 5kgs of milk per cow per day.

Milk quality along the dairy value chain is characterized by poor on-farm milking hygiene and storage, high somatic cell counts, prolonged periods of time between milking at farm level and cooling plants level inadequate testing or no testing at all, use of unqualified and poorly trained milk graders and transporters, adulteration of milk and lack of milk traceability among the milk suppliers (Rademaker *et al*, 2015). The high cost of milk testing equipment, lack of proper skills on the use of the equipment, lack of milk quality management capacity and institutional gaps present a major hindrance to quality control and assurance (MoALF, 2013). According to Muriuki (2011), health risks are issues of concern due to the large amount of milk that is marketed unprocessed, and to weak monitoring of the market. The main public health concern is the potential risk of zoonotic diseases such as brucellosis and tuberculosis (TB), while drug residues even in the processed milk channel are also alarming. The greatest limitations in the whole raw milk value chain are proper ways to maintain cold collection due to the high investment costs demanded (Orregård, 2013). According to (MoLF, 2013), primary milk marketing faces infrastructure bottlenecks which can be attributed to poor road networks and lack of sufficient cooling and storage facilities. Consequently, 3% of total milk produced during flush periods would convert to losses. According to Kurwijila (2006) and Lee *et al.,* (2016), milk needs to be cooled within 2-3 hours from milking. However, since milk collection is conducted only once in the morning, evening milk is particularly of poor microbial quality when received by processors and hawkers the following morning. Orregård (2013) showed that, additive factors

like unhygienic milking and handling practices, results in poor raw milk quality. Plastic jerry cans are impossible to clean and are often used for transporting milk by most motor bike transporters. This result in a less hygienic handling compared with the use of aluminum cans whose only limitation is the initial acquisition cost. The Collection and Bulking Enterprises (CBEs) also lack a milk quality policy that guides the implementation and enforcement of milk procurement and milk quality assurance procedures (Rademaker *et al.,* 2015).

The regulatory institutions are constrained by lack of resources in terms of personnel and equipment to enforce safety regulations. Therefore, enforcing and monitoring the standards appropriately becomes a great challenge to them (Muriuki, 2011). This is especially due to the large numbers of farmers involved in the sector. Thus, a vacuum is created for the farmers to switch between buyers of raw informal milk especially when quality becomes an issue with the formal sector. (Corné *et al.,* 2016) indicated that the majority of milk that currently reaches consumers, both from informal and formal agents, is below Kenyan national standards. However, KEBS has adopted a Code of Practice (CAC/RCP 57-2004) for milk production to assist farmers in producing hygienic milk.

Enhancing raw milk quality should involve continuous monitoring and follow-ups at different points of milk handling and not only at the factory gate. The result is that, milk production at the farm level is done hygienically, from healthy animals and in the right environment (Omondi and Meinderts, 2009). Muriuki (2011), described the critical quality control challenges in line with milk bulking as adulteration, high bacterial load, presence of anti-microbial residues and zoonotic diseases while traceability is valuable particularly for the export market. This particularly affects the informal sector but also relatively in the formal sector (Orregård, 2013). According to a study in the retail chain (Mwangi *et al.* 2000), 58% and 82% of raw milk samples did not meet national standards for coliform and total plate counts, respectively. The same author observed that, adulteration with water had an estimate of 13% of all the collected retail samples. Thus production of high quality products, from low quality milk, that conform to the international standards specifications is a challenge. This study was designed to assess microbiological and physico-chemical quality of raw milk from two smallholder dairy farmers at four sampling levels in comparison to the KEBS requirements, adherence to hygienic code of practice and identify the critical control points for improved raw milk quality.

1.2 Statement of the problem

The dairy industry in Kenya is facing a lot of challenges in meeting raw milk quality standards specification. Farm practices by most small holder dairy farmers' do not comply with the Kenyan code of hygienic practices for production, handling and distribution of milk and milk products. This is due to a number of factors ranging from poor milk production and handling hygiene, poor road network and warm milk collection. The large numbers of milk suppliers (1.8 million) mostly smallholder farmers hinder effective inspection and regulation of their daily operations by the KDB. To address the raw milk quality problem the industry is facing, KDB which is mandated to regulate, promote and develop dairy industry has proposed partnerships to develop and implement the concept of a Quality Based Milk Payment System (QBMPS). Consequently, a successful conduct of this study will form the basis for the implementation of Hazard Analysis Critical Control Points (HACCP) principles to improve milk quality and suggest some quality analysis test that can be used for QBMPS.

1.3 Objectives

1.3.1 General Objective

To enhance the raw milk quality in the small holder supply chain in the Kenyan dairy sector.

1.3.2 Specific Objectives

1. To assess the compliance level of small scale dairy farmer to the hygienic practices in production, handling, storage and transportation of raw milk.
2. To develop a quality control plan using critical control points in raw milk value chain for smallholder dairy farmers.
3. To monitor, record and establish the quality of raw milk from collection points to the processing plant.

1.4 Research questions/hypothesis

1. Are milk production, handling, storage and transportation by smallholder dairy farmers complying with the code of hygienic practices?
2. Are there quality control plans in the raw milk value chain for the smallholder dairy farmers?
3. The quality of raw milk from the collection point to the processing plant for smallholder dairy farmer does not meet the set KEBS requirements.

1.5 Justification

Despite the fact that the Kenya Dairy Board has often advised the stake holders in dairy sector to take responsibility of regulating the industry and to pilot quality based milk payment system, none of them has embarked on this quality device to enhance raw milk quality in the smallholder supply chain. Historical data from Happy Cow Ltd. suggests major challenges in total plate count, coliform count, somatic cell count and antibiotic residue in the raw milk that are way above the KEBS standards. The results are reflected in the processing of yoghurt and cheese that became a challenge in meeting the international Standards. The thermal destruction process is logarithmic and bacteria are killed at a rate that is proportional to the number of bacteria present in the raw milk initially before processing. High Temperature Short Time (HTST) pasteurization is expected to accomplish five log cycle reductions of the microbial load. The actual reduction achieved depends on the equipment, flow rate, composition of product and the kind of bacteria. Systems must therefore be put in place that justifies the quality of raw milk delivered, its costs and at the same time identify responsible parties for the delivered raw milk quality along the chain. This will motivate most farmers to improve their hygienic practices and avoid vices such as adulteration.

CHAPTER TWO
LITERATURE REVIEW

2.1 Dairy industry development in Kenya

Market-oriented dairying in Kenya started nearly a century ago. This was after the introduction of dairy cattle breeds by European settlers from their native countries. They occupied the most agriculturally productive areas in the central parts of the Rift Valley and Central Provinces. In 1954, Crossbred cattle dairy production by Africans began after a colonial policy paper allowed them to engage in commercial agriculture (Muriuki, 2001). After independence, the dairy herd had increased to about 400,000 exotic animals including their crosses with the local breeds. Many foreign settlers chose to leave the country and therefore they sold their farms to indigenous Kenyans or to the government. Consequently, many of these farms were rapidly sold to African smallholders (Omore *et al.,* 1999). Due to decline of dairy cattle population in large-scale farms, the government decided to promote dairying. This included subsidized and efficient clinical services, providing artificial insemination services, restructuring the operations of the main formal output market and the removal of quotas that stipulated minimum milk deliveries (Kurwijila and Bennett, 2011). These efforts immensely supported the rapid growth of the smallholder dairy systems that dominate the industry at the production level. Each farmer has 3 to 5 acres of land and about two to five head of cattle yielding about 5 kg of milk per cow per day. Milk sales are low, at less than 10kg per day and use of inputs is low, though it varies depending on community traditions and the level of market orientation (Muriuki, 2011).

The Kenyan dairy sector contributes approximately 3.5% and 19% to the country's national and agricultural GDP respectively, offering over 900,000 jobs along the value chain hence, emerging as one of the largest dairy industries in sub-Saharan Africa (Muriuki, 2011). According to Orregård, (2013), KDB had indicated that the total cow's milk production by 2010 had reached an estimate of 4 .6 million tons. Ministry of Livestock Development (MoLD) and the KDB predicted an increase in milk supply by at least 4.5% per annum consecutively for four years from an estimated 4.2 to 5 billion liters by 2014 (Kabui, 2012). According to Gachango *et al.* (2014), adoption of new technologies by farmers has subsequently led to increased milk production. Nevertheless, marketing challenges overwhelm the farmers resulting into post-harvest losses if not forced consumption. Market rejections have been associated with poor handling in the whole raw milk value chain and prolonged time taken to reach markets especially

during the wet season, when production is high and roads are impassable. This results to post-harvest milk losses which are estimated to be more than 6% of the total production mainly due to spillage, lack of market, and rejection at the market (Muriuki, 2011). To enable farmers to acquire credit and inputs and sell their produce such as milk dairy co-operatives societies and other farmer groups such as self-help groups (SHG) are formed. About 3.34% constitutes the dairy societies (co-operatives) out of the total societies and unions formed in Kenya (Muriuki, 2003). Dairy cooperatives have in the past significantly contributed to the development of the smallholder milk marketing and provision of farm inputs and services at a relatively lower cost (Omiti and Muma, 2000).

2.2 Dairy value chain in Kenya

The formal milk trade is the market segment licensed by Kenya Dairy Board (KDB) contrary to the informal trading which is unlicensed. Informal markets control an estimated 70 percent of the total milk marketed in Kenya (MoALF, 2013). This informal sector is important and is driven by among other factors the traditional preferences for fresh raw milk and its relatively lower cost. Raw milk markets offer both higher prices to producers and lower prices to consumers but with several challenges relating to quality control and standards, and the associated health and safety concerns (Wambugu *et al.,* 2011). Other players in milk marketing include informal traders, distributors and retailers. The existence of informal trade results from a combination of the formal system's failure or inefficiency, consumer preferences, and price differences between raw and processed milk (Muriuki, 2011). Tremendous growth in the informal sector was realized after milk market liberalization in 1992 which comprised of small scale operators dealing in marketing of raw milk including direct sales to consumers, hawked milk sold by mobile traders, shops/kiosks, and co-operative societies (Muriuki, 2011; Wambugu *et al.,* 2011) and recently milk bars. Therefore, the dairy industry plays a vital role in food security and enhances the livelihoods of all its stakeholders in Kenya (Bebe, 2003).

Despite this high volume of production and the extensive formal marketing network in Kenya, estimates show that currently approximately 85-90% of marketed milk is not processed or packaged, but instead is bought by the consumer in raw form (EADD, (2008). The factors driving the continued importance of the informal market are traditional preferences for fresh raw milk which is boiled before consumption and unwillingness to pay the costs of processing and

packaging. By avoiding pasteurizing and packaging costs, raw milk markets offer both higher prices to producers and lower prices to consumers (Kurwijila, 2002). Growing consumption and latent capacity had led to a fragmented value chain, with numerous players vying for profits along the chain. Despite the competition, farm gate prices are largely indistinguishable between the formal and informal chains. The informal market has one main advantage over formal in that the farmers are being paid immediately for their milk while in the formal chain, farmers can wait up to a month or longer before they receive their payment. Therefore, smallholder farmers who are largely facing immediate cash flow problems, the informal market provides an advantage (EADD, 2008). Weakness can however arise from the small scale of milk output, 10kg per farm per day (Bebe, 2003) especially in terms of quality, the poor rural infrastructures and reliance on rainfall for production and the poor milk markets. As with all food safety standards worldwide, milk safety requires monitoring from production to consumption.

The Hazard Analysis Critical Control Points (HACCP) process is now a widely accepted methodology in risk analysis for industrially processed foods. HACCP is an improved system compared to the traditional sampling and testing of quality control. It is a proactive procedure in prevention which reduces the risk of processing and selling unsafe products, it is also a cost-effective program which is useful in milk production and processing (Keski and Gulsunoglu, 2012). It is essential to identify the areas in the operation where threats are posed by these hazards, their Critical Control Points (CCP), and develop Standard Operating Procedures (SOPs) to prevent issues from arising at these points (Karakök, 2007). According to Mwangi *et al.* (2000), HACCP identifies the points in a process that are hazardous, their risk factors and potential level of risk so that critical control points for corrective action can be implemented. He further explains that, application of HACCP is a major challenge in developing countries including Kenya, where food markets are mostly informal. His findings concurred with Muriuki *et al.* (2003), who recommended that an analysis of Critical Control Points (CCPs) in the Kenya milk marketing channel is necessary.

Dairy value chains link the actors and activities involved in delivering milk and milk products to the final consumer where with each activity, the product increases in value. It involves production, transport, processing, packaging and storage. The activities require inputs including financing and raw materials which are employed to add value and to transport dairy products to consumers. Every actor of the chain should give the product the maximum added value at the

minimum possible costs (Muriuki, 2011) at the same time ensuring hygienic handling. Value chain analysis is essential to an understanding of markets, their relationships, the participation of different actors and the critical constraints that limit the growth of lives to production and consequently the competitiveness of smallholder farmers. Setting up an efficient, hygienic and economic dairy chain is a serious challenge in many developing countries including Kenya. Among the reasons for this are; difficulties in establishing a viable milk collection and transport system because of the small quantities of milk produced per farm, seasonality of the milk supply, poor transport infrastructure, deficiency of technology and knowledge in milk collection and processing, poor quality of the raw milk, distances from production sites to processing units and also to consumers and difficulties in establishing cooling facilities (FAO, 2004).

Normally, milk needs to be cooled within 2-3 hours from milking. The main characteristic of the supply chain milk is the poor cold chain which lowers the quality of processed milk and prevents processors from producing long life products that need the high quality raw material. Since milk collection is conducted only in the morning, evening milk in particular is of poor quality when received by processors and hawkers the following morning (EADD, 2008). Milk handling equipment is one of the most significant sources of microbial contamination in milk. Moreover, if equipment is inadequately cleaned and milk residues are left on wet surfaces which will result in microbial growth, and could contaminate the milk. Plastic jerry cans are impossible to clean and are often used for transporting milk by most motor bike transporters in Kenya. This result in a less hygienic handling compared to the use of aluminum cans whose only limitation is the acquisition cost (Orregård, 2013). High numbers of bacteria in raw milk is indicative of poor handling practices/operations and inadequate maintenance of the cold chain (Irwin and Foreman, 2012). Milk borne hazards such as antibiotics and excessive load of bacteria can enter the milk chain at many points along the market depending on handling and ethical attributes of the actors along the chain (Muriuki *et al.*, 2003).

2.3 Raw milk quality

2.3.1 Raw milk quality in Kenya

Quality milk means, milk which has normal chemical composition and is completely free from harmful bacteria and harmful toxic substances, free from sediment and extraneous substances, has lower degree of titratable acidity, it's of good flavor, adequate in keeping quality and low in

bacterial counts (Gurmessa *et al.*, 2015). The nutritional composition and other features like high water activity make milk an outstanding medium for bacterial growth. Dairying has a huge potential of turning around the economy in Kenya especially in the rural areas but is besieged with hurdles particularly poor quality of milk and hygiene along the supply chain both in formal and informal milk market. Thus, Kenyan milk does not meet the Kenyan national quality standards due to bacterial growth even before farmers sell their milk (Chepkoech, 2010). It's associated with high coliform counts because of the use of plastic instead of metal containers (Mwangi *et al*., 2000). Equally important is the milk holding temperature and length of time milk is stored before testing and processing that allow bacterial growth. All these factors will influence the total bacteria count and the types of bacteria present in raw bulk tank milk (Murphy and Boor, 2000). Thus, milk and milk products can be important sources of food borne pathogens (Oliver *et al*., 2005).

The main challenge in the Kenyan dairy industry is the lack of sufficient enforcement on quality controls to ensure that farmers, milk traders and processors maintained the specified standards of quality (Kamau, 2009). Adulteration of milk with water which is very common in Kenya not only causes dilution of milk reducing the milk solids but it also involves, the risk of introducing contaminants into the milk further decreasing its microbial quality and lowering the density of the milk. By Adding water, the milk´s microbial quality could be compromised, which could result in a health hazard (Orregård, 2013). The scientific literature shows very clearly that a high Somatic Cell Count (SCC) is associated with higher incidences of antibiotic residues in milk (Oliver *et al*., 2005). Since it is a perishable food, milk should always be delivered in a pure, hygienic and unadulterated condition. According to Orregård (2013) the biggest problem in Kenya is the lack of hygiene, not only on the side of the farmers, but also on the hawkers and even processors. If the basic requirements of hygiene would be observed, the quality of raw milk reaching the market would be certain. The problem with the use of plastic jerry cans is that it has a small opening, which makes it impossible to clean. Continued use of the plastic jerry cans for milk storage and transport leads to accumulation of dirt, a major cause of milk contamination (Kamau, 2009).

According to Orregård (2013), one sample result reached a remarkable $2.1x10^7$cfu/ml total plate count which is more than 10 times higher than the limit for acceptable milk according to Kenya Bureau Standards (KEBS). Additionally, about 9% of the samples contained antibiotic residues

for the small scale agents. Along the value chain, 2% of the agents indicated that they used hydrogen peroxide as a way of preserving milk. Therefore, much of the milk being processed does not comply with the standard and therefore in theory the processors could be prosecuted for use of non-conforming raw milk (Irwin and foreman, 2012). Remarkably, 70% of pasteurized samples did not meet national standards for bacterial counts (Mwangi *et al.*, 2000). According to Kamau (2009), in some samples, the amount of bacteria was 281 times higher than the minimum allowed and none of the milk samples passed the standards as specified by the KEBS and therefore unfit for human consumption.

According to FOSS analytical (2005), the quality of the raw milk supply can be influenced via the payment systems where premiums and deductions are based to create the incentive to upgrade the quality of raw milk. The same study indicated that 82% of the countries participating in the quality-based milk payment study had reached their objectives through payment systems. In Poland year 1996, almost 30% of the milk supplied was below 400,000 cfu/ml unlike in 1995 where only 8% managed. Similarly, a 20% drop was observed from 85% of the suppliers delivering milk with a bacteria count above 3,000,000 cfu/ml. Denmark (in 1972), when the first class bacteria limit was changed from 400,000 to 200,000 cfu/ml. This change led to a drop in percentage of farmers in the first class from 80% to 65% and took six months before 80% of the farmers could supply first class milk again. Currently in Kenya, increase in volumes and market share are prioritized than milk quality assurance. According to Draaiyer *et al.* (2009), to improve the safety and hygienic quality of the milk, a payment system based on hygienic quality may be introduced. Quality-based milk payment system can instill responsibility on the farmers to ensure production and delivery of quality raw milk. According to Swai and Schoonman (2011) study, there was the need to implement good hygiene practices and effective monitoring from production through the delivery chain to the consumer. Accordingly, production of high quality raw milk, consumer's satisfaction and reduced public health concern are the most certain results among many more advantages.

2.3.2 Milk constituents

Milk is the normal, clean and fresh lacteal secretion extracted from the udder of a healthy cow, properly fed and kept, but excluding that obtained during the first seven days after parturition (EAS, 2007). Milk is synthesized by cells within the mammary gland and is virtually sterile

when secreted into the alveoli of the udder. Beyond this stage of milk production, bacterial contamination can generally occur from within the udder, outside the udder, and from the surface of equipment used for milk handling and storage. Cow health, environment, milking procedures and equipment sanitation can influence the level of microbial contamination of raw milk (Murphy and Boor, 2000). Milk and milk products play a vital role in building a healthy society and it contributes to one third of the world's intake of animal protein since it's considered as high quality protein (Bashir *et al*., 2013). There are several factors that lead to variation in milk constituents. The level of variation could be enormous depending on the individual animal, its breed, stage of lactation, age, health status, management practices of the herd and environmental conditions. On average milk composition can be tabulated as;

Table 1: Composition of cow's milk

Main constituent	Range (%)	Mean (%)
Water	85.5 – 89.5	87.0
Total solids	10.5 – 14.5	13.0
Fat	2.5 – 6.0	4.0
Proteins	2.9 – 5.0	3.4
Lactose	3.6 – 5.5	4.8
Minerals	0.6 – 0.9	0.8

Source; (O'Mahony, 1988).

2.3.3 Microbial quality of raw milk

The KEBS specification has classified the microbial quality of raw cow milk in three grades. The lowest grade should have a maximum of 2,000,000 cfu/ml and 50,000 cfu/ml for total plate count and coliform count respectively (EAS, 2007). The microbial load and types found in milk shortly after milking are influenced by factors such as animal cleanness and health, equipment cleanness, season, ambient temperature, storage and personnel health/hygiene (Gemechu *et al.,* 2015). Microbial load in raw milk determines its quality. International Dairy Federation guidelines stipulates that production of milk having Standard Plate Count of 10^4 cfu/ml reflects good hygienic practices while high initial Standard Plate Count of more than 10^5 cfu/ml are

indications of poor production hygiene (Jain and Shrivastava, 2014). The dominant bacterial isolate is *Lactobacillus lactis*, a species of bacteria used in starter cultures and if left in favorable temperatures lead to fermentative spoilage (Kahuta, 2013). *Bacillus, Clostridium*, *Corynebacterium*, *Mycobacterium*, *Micrococcus*, *Arthobacter*, *Streptococcus*, *Staphylococus* and *Lactobacillus* with the exception of *Arthobacter* and *Lactobacillus* are some of the Gram-positive psychrotrophic bacteria isolated from raw milk (Samarzija *et al.,* 2012). Moreover, some species and strains of *Clostridium, Microbacterium, Micrococcus, Bacillus, Cornebacterium, Arthrobacter, Lactobacillus* and *Streptococcus* grow well at ambient temperatures, withstand pasteurization temperatures and proliferate at refrigeration temperatures leading to spoilage of well-preserved milk (kahuta, 2013). *Pseudomonas fluorescenes* is a psychrotrophs that produces hydrolytic enzyme in raw and pasteurized milk (Rajmohan's *et al.,* 2002). Microbial spoilage of raw milk can potentially occur from the metabolism of lactose, protein and fat. High nutrients available in dairy products provide a good media for the microbial growth (Dilbaghi and Sharma, 2007). Environmental contaminants represent a remarkable percentage of spoilage microflora and they contaminate the cow, equipment, water, and milker's hands.

2.4 Raw milk assessment methods

2.4.1 Platform tests

Platform tests or milk reception tests are carried out both at the milk collection centers and at the platform of dairy processing plant. The tests are usually rapid quality control tests, serving as a basis of accepting or rejecting milk (Draaiyer *et al.,* 2009). Application of platform tests does not directly involve laboratory analysis of raw milk samples. A suspected case calls for a sample of the milk being taken to the laboratory for further quality assessment; therefore they should be treated separately until its quality is verified and the decision made either to accept or reject. Importantly, the organoleptic test is critical since it involves analyzing the milk before it leaves its original container. The tests involve the milk quality being judged by the use of a person's senses (sight, smell, and touch) and no equipment is required hence the person carrying out the tests should be experienced for reliable results (O'Mahony, 1988). If adulteration is suspected in milk, lactometer test serves as a quick method to confirm. The alcohol test is used for rapid determination of an elevated acidity of milk. The test is carried out by mixing equal quantities of milk and ethanol solution. This result in coagulation of the milk proteins if the developed acidity

is high since alcohol is a dehydrating agent which indicates the milk protein instability hence the milk cannot withstand pasteurization (Pandey and Voskuil, 2011). Application of platform tests is subjective since it doesn't involve laboratory analysis. Therefore, all aspects of quality cannot be fully analyzed on the platform and a need to establish whether the milk is suitable for processing or rejection is vital.

2.4.2 Laboratory tests

These are objective tests and are therefore used to confirm the milk quality after the platform tests have been carried out (O'Mahony, 1988). Titratable acidity test measures the concentration of lactic acid in the milk using sodium hydroxide solution and phenolphthalein indicator to the milk by titration. Normal milk ranges are 0.14-0.18% therefore beyond 0.18%, the milk is of poor quality and below 0.14% could be adulteration with an alkaline. The Resazurin test also called dye-reduction tests is an indicator of the hygienic quality of milk. When bacteria grow in milk, they use up the oxygen present and the colour changes according to the amount of oxygen present. The time taken to change or reduce the colour of the dye provides a good indication of the bacteriological quality of milk. A pH meter measures the current produced and it depends on the potential difference between two electrodes when they are in contact with a test sample. The pH of the milk depends on the hydrogen ion concentration in the milk (Pandey and Voskuil, 2011). Milk and water exhibit different freezing points i.e. water has a freezing point of 0 °C, while normal milk has a freezing point of around -0.540 °C, mainly due to lactose and salts. According to Draaiyer *et al*. (2009), freezing point test can be used as a confirmatory test for adulteration with water. The same author indicates that hydrogen peroxides and formaldehyde as some of the commonly added preservatives and are health hazards. Their presence in milk makes it impossible to process the milk into fermented products. The California mastitis reagent consists of a detergent and a pH indicator. The extent of the reaction between the detergent and the deoxyribonucleic acid (DNA) of the cell nuclei is a measure of the number of the somatic cells in the milk. At a concentration of 150,000 to 200,000 cells per milliliter a precipitate begins to form hence thicker gels occur in samples with larger number of cells (Draaiyer *et al*., 2009).

2.4.3 Microbial analysis

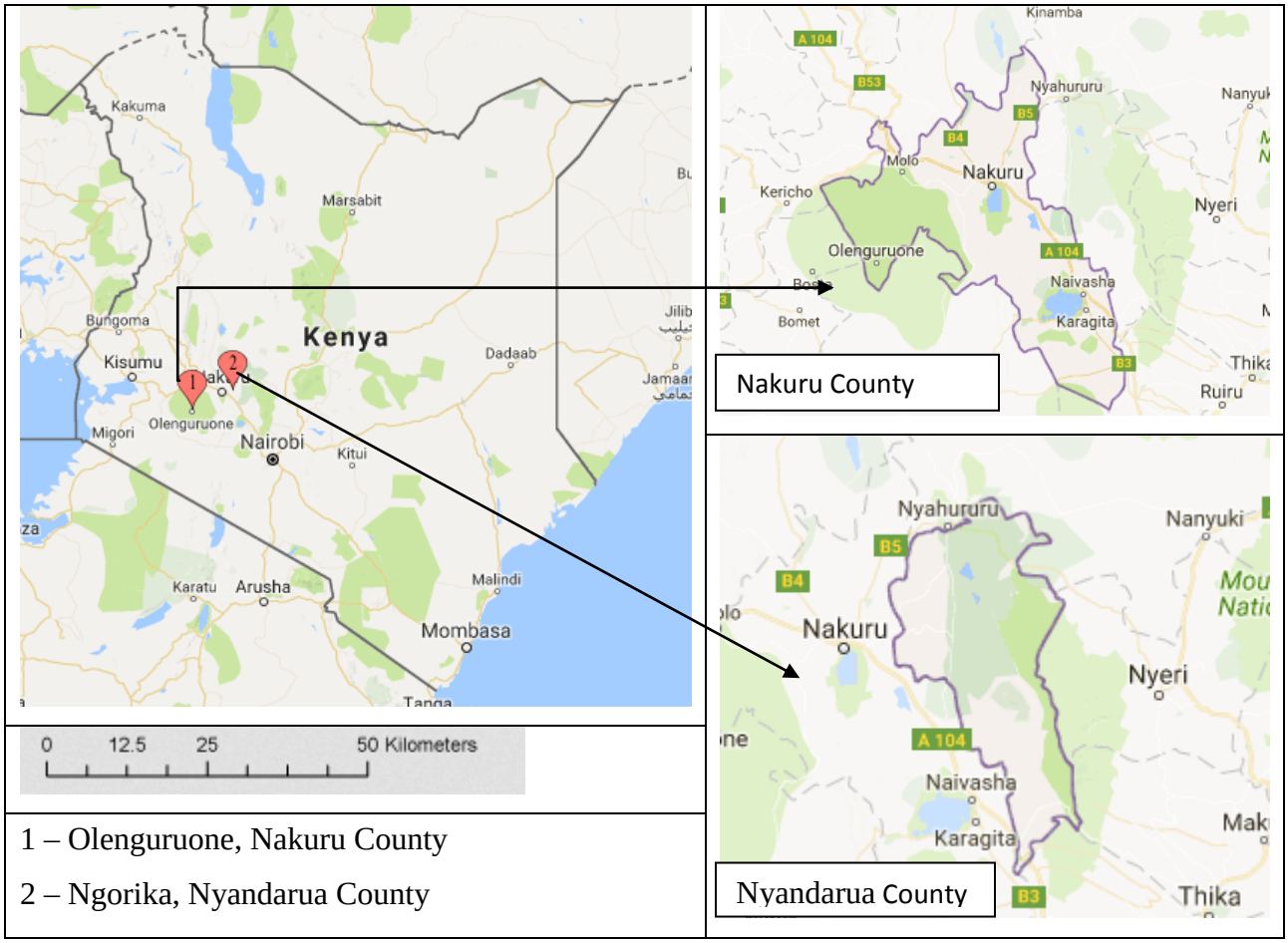
The traditional microbiological analyses in quality control, consumes a lot of time and immense laboratory work (Souza *et al.,* 2015). The 3M^{TM} Petrifilms plates are thin film, ready to use,

dehydrated versions of the conventional petri dish agar plate. They are well suited for quantitative tests because of their vast advantages over conventional agar plates including; in-built biochemical confirmation, ease of preparation and use coupled with the smaller space requirements (Silvia *et al*., 2005). They also have international recognition by Association of Analytical Communities (AOAC) and Association French Normalization Organization Regulation (AFNOR) (Souza *et al*., 2015; Barry, 2005). According to McCarron *et al.* (2009), for on-farm culture systems, use of 3M^{TM} Petrifilms was successful in detecting *Staph. Aureus* in clinical mastitis milk samples. Another study by De Sousa *et al.* (2005) showed that Petrifilms are suitable and convenient alternatives to the standard method of enumeration of aerobic flora in Crottin's goat's cheese. Furthermore, Souza *et al*. (2015) concluded that the results obtained from a related study verified positive correlation between conventional methods and petrifilm™ system for microbial analyses of mesophilic aerobics, total coliforms, *Escherichia coli*, *Staphylococcus aureus* and lactic acid bacteria in sheep milk.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study site



1 – Olenguruone, Nakuru County

2 – Ngorika, Nyandarua County

Figure 1. Map of study site

The study was carried out at New Ngorika Milk Producers Limited (Ngorika) in Nyandarua County, Olenguruone Dairy Cooperative Society (Olenguruone) in Nakuru County. The two societies were well established smallholder dairy farmer's cooperatives which supplied milk to Happy Cow Ltd. located in Nakuru County. For both collecting and bulking enterprises (CBEs), milk from individual farmers was collected and bulked into milk-cans while warm and transported to the cooling plant. Milk collection points were not well established and therefore milk collection took an average of 6 hours.

Ngorika Settlement is an area in Central Province (Central), Kenya (Africa) with the region font code of Africa/Middle East. It is located at an elevation of 2,190 meters above sea level and its coordinates are 0°19'60" S and 36°13'60" E in Degrees Minutes Seconds. Ngorika had 600 active

members who delivered approximately 3,500 liters of milk per day from the 6 routes. The mode of milk transportation consisted trucks, tractors with trailers, donkeys and motor bikes. Milk was collected in the morning with some farmers offering their evening milk separately along the routes.

Olenguruone Settlement Area is an area in Rift Valley Province (Rift Valley), Kenya (Africa) with the region font code of Africa/Middle East. It is located at an elevation of 2,478 meters above sea level. Its coordinates are 0°37'0" S and 35°37'60" E in Degrees Minutes Seconds. Olenguruone had a total number of 1,600 active members with an average 6,000 liters of milk per day. It consisted of three cooling plants with a maximum capacity of 5,950, 1,650 and 3,200 liters for the main cooling plant located at Olenguruone town and other two satellite cooling plants at Kaplamai and Kiptangich respectively. This study was carried out at the main cooling plant as a pilot. Milk collection transport mode included motor bikes, vehicles, donkey carts and individual farmers.

Laboratory tests were carried out at the Happy Cow Ltd. laboratory and Egerton University Department of Dairy and Food Science and Technology for compositional analysis.

3.2 Experimental design

Questionnaires were used to depict the implementation of the code of hygienic practices in milk handling by the farmers, transporters, CBEs and the processor. This was done through issuing of questionnaires and the questions were answered through direct interaction with the respondents for accuracy of information. To develop the quality control system, observation were done to allow identification of the various shortcomings that could contribute to quality deterioration. In laboratory experimentation, a randomized complete block design with three replications was employed. The CBE were used as the blocks while the various levels of sampling (Can, Route, cooler, tanker) were used as treatments. The parameters of interest included; total plate count, coliform count, antibiotics residues, added water and somatic cell count.

3.3 Population of the study and data collection

The target population of the study was 2,200 farmers where 600 farmers were from Ngorika and 1,600 farmers from Olenguruone. Simple randomization procedure was used in sample selection of the farmers in the identified populations. In order to generate the required sample units, the determination of sampling frame was essential. The sample size of the study was 234 and 310

from Ngorika and Olenguruone respectively. This was according to sample size determination table by Krejcie and Morgan (1970), Appendix 1, at an alpha level 0.05 and a t-value of 1.96.
The questionnaires were developed, pre-tested before being administered to the selected individuals in the study. The researcher in person visited the CBEs, issued the questionnaires to the sampled farmers which enhanced the process of data collection as all the selected respondents were reached on time. During the distribution of the questionnaires, the purpose of the research was explained to the groups involved. This study was carried out in the month of June and July, 2015.

3.4 Hazard analysis

Quality deterioration factors were identified by observation of activities in the collection chain. The HACCP principles were employed to identify the quality deteriorating factors along the value chain as the hazards. Table 2 was used as a key in determining the level of risk at Happy Cow Ltd. HACCP documentation. The decision made at a certain level of risk was classified in three stages. They included; 1 to 2 where the impact was termed as negligible, 3 to 4 where the impact was referred as minor and 6 to 9 where the impact was major. The first two stages were defined as the pre- requisite programs while the last stage was defined as critical control points. According to Codex Alimentarius (2003), a decision tree was used to classify the factors as either prerequisite programs or critical control points. This was to facilitate a quality control system that would curb quality deterioration at all levels in the dairy value chain. To carry out the risk assessment and determine whether a quality deteriorating factor is a CCP or a PRP, the control level and the likelihood of occurrence were multiplied which enabled classification of the factors (Table 2). Where a factor's likelihood of occurrence and level of control was severe, the factor was referred to as a CCP. Likewise, any value above 6 indicated that the factor is a CCP and needs a corrective action.

Table 2: The control level and likelihood of occurrence in risk assessment

Likelihood of occurrence	Level of control			
		1 (Low)	2 (Moderate)	3(Severe)
	1(Low)	1	2	3
	2 (Moderate)	2	4	6
	3(High)	3	6	9

Source: Happy Cow Limited ISO documents

3.5 Laboratory analysis

3.5.1 Milk sampling and preparation

Milk sampling was conducted in June and July 2015. The organoleptic, alcohol and lactometer tests were done on each and every farmer's milk before acceptance and bulking together in a can. The milk in an aluminum can was then stirred using a plunger before sampling while the plastic jerry can was shaken well. A sample was drawn with a sanitized sampling dipper and transferred in a well labeled sterile sampling bottle. The sampling bottles were coded as per CBE/ route/ farmers can number/sampling date. At the can level, the samples were collected during early morning at the respective farmer's delivery points before transportation. The route level samples were collected at the cooling plant reception platform, the cooler level samples were collected immediately after the cooler filled up while the tanker level samples were collected at the processing factory reception platform.

At the CBE, the number of samples to be analyzed was determined by the platform reception tests i.e. upon acceptance of a milk can, the sample was accepted for analysis. The platform tests included organoleptic, alcohol, lactometer, acidity, 10 min resazurin and peroxide. A total of 119 and 189 samples were collected in Olenguruone and Ngorika respectively for TPC, CC, Somatic cell count, titratable acidity, resazurin test and compositional analysis. A total of 106 and 111 samples were analyzed on freezing point depression while 36 and 38 composite samples were analyzed for antibiotic residues from Olenguruone and Ngorika respectively. The samples were immediately placed in a cool box containing chilled water and ice packs hence cooling the samples to below 3^0C.

The maximum time taken from CBEs to the lab was 1hour and 3hours for Ngorika and Olenguruone, respectively. The samples were analyzed in the laboratory immediately after arrival.

3.5.2 Microbiological analysis

3.5.2.1 Total plate and coliform counts

Microbiological analysis for Total Plate Count and Coliform Count was done according to AOAC (2005) methods 986.33 using 3M^{TM} petrifilms plates without modifications. The diluent was prepared prior to the microbial analysis. This was done by dissolving 15 g peptone powder in 1000 ml distilled water followed by sterilizing using an autoclave (serial no. 125-0365) at 121^0C for 15min. Serial dilutions were done up to 10^{-6} where 10^{-4}, 10^{-5} and 10^{-6} were considered for plating in a sterile environment. For each plate, 1ml of the sample was pipetted and placed on a petrifilms plate using micropipette (Huawei serial no. 144226). Plating for coliform count was done for dilution 10^{-4} and 10^{-5} while total plate count plating was for dilution 10^{-5} and 10^{-6}. After plating, the samples were placed in the incubator (serial no. 14070247 and 14070240) at 32 ^{0}C, stacking them to a maximum of 20 pieces and incubated for 24hours and 48hours for coliform count and total plate count respectively. Colony counting was done using 3M plate reader (serial no. 03726) where the total counts were recorded automatically in an excel sheet in a computer software. An empty plate was placed under the same conditions as the sample plates for observation (control).

3.5.2.2 The 10-minute Resazurin test

The resazurin test was done as per Draaiyer *et al.* (2009) where a resazurin tablet was completely dissolved in 50 ml of sterile distilled water according to the manufacturer's instructions. One milliliter of the resulting solution was added into 10 ml of the milk sample in a test tube, mixed and then incubated at 37^0C in a water bath (serial no. 1407) for 10min. The samples were then read using a Lovibond comparator from a good source of light for colour change and numerical score value ranging from 1-6, assigned. A milk sample without the resazurin dye was similarly treated and used as the control in the comparator. Samples with comparator disc readings ranging from 4-6 were acceptable based on the (EAS, 2007) on milk quality.

3.5.3 Physico-chemical tests

3.5.3.1 Acceptance tests

The chemical tests including organoleptic, alcohol, lactometer, titratable acidity and freezing point were done according to EAS (2007) and Draaiyer *et al.* (2009). The acceptance tests were carried out at the farm level including organoleptic, lactometer and alcohol to either reject or accept the milk before bulking in a can composite. Organoleptic test involved colour observation, odour and taints smelling after stirring using a plunger and feeling the can for temperature detection. Alcohol test was done using an alcohol gun where 2ml of milk and an equal amount ethanol (at 76% concentration) were mixed in a transparent petri dish and clots observed. For lactometer test, the sample was put in a measuring cylinder and the lacto-density meter calibrated and confirmed by KEBS placed slowly into the milk until it floats freely. The temperature and the lactometer readings were taken and recorded.

3.5.3.2 Titratable acidity

The titratable acidity test was done as per Draaiyer *et al.* (2009). This was measured by titration whereby 3-4 drops of 0.5% phenolphthalein were added in 9ml of milk sample in a beaker on a white tile and titrated against 0.1equivalents/litre NaOH with constant shaking of the milk until a permanent colour change (pink) was observed. By recording the volume of base used and the volume of the milk sample, the amount of lactic acid developed was calculated and expressed as a percentage.

3.5.3.3 The antibiotic residues test

Antibiotic residues analysis involved only the bulk samples at the routes and cooler/ tanker levels due to the cost of the analytical method applied. The presence of the antibiotics residues (penicillin, cephalosporins, oxy tetracycline, gentamicin, streptomycin etc.) was detected using the Delvo test according to DSM, (2011). Delvo test is easy to use and covers the broadest spectrum of antibiotic residues in the industry. Moreover, it's reliable and accurate with detection levels closest to maximum residue levels and safe tolerance levels (Hillerton *et al.*, 1999). For Delvo SPNT ampules, 0.15 ml of the milk sample was added to the ampule and incubated for 3 hours at 64^{o}C in a Delvo incubator (serial no. 39031197) to observe colour changes. The Delvo test (BLF) involved use of ampules together with strips. The incubator was set at 64^{0}C and the ampules with 0.15 ml milk sample inserted in it for 2min. The milk sample

was swirled again before inserting the strips in the ampules for 3 min. The results were read, interpreted and recorded.

3.5.3.4 Somatic cell count

The somatic cell count was done on all the samples using California Mastitis Test (CMT) according to Mellenberger and Roth (2000). An equal amount of commercial CMT reagent was added to each cup and a gentle circular motion applied to the mixture in a horizontal plane and a positive gelling reaction occurred in 10 seconds with the positive samples. The gel formation and colour changes was observed and compared with the colour and viscosity comparison table and the results recorded.

Table 3: Colour and viscosity comparison for somatic cell count per 1000 ml

CMT	Interpretation	Cell count/1000ml
Negative	Liquid without gel	0 - 200
Traces	Light gell by transparence, will disappear after 10 seconds	150 - 500
1	Visible light gel by transparence, persistence	400 - 1000
2	Visible gel, viscous filament, adhesion cup	800 - 5000
3	Strong gel like the white of egg	>5000

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3.5.3.5 The freezing point test

The freezing point determination was carried out to assess adulteration incidences. According to Draaiyer *et al.* (2009), when milk is adulterated with water or other materials, the freezing point of milk change from its normal value (-0.54ºC) causing a detectable elevation. This was done using a cryoscope (serial no. 7150-02262) according to the manufacturers operating instructions. After calibrations required by the equipment were completed, 2.5ml of the milk sample was put in the sample vial and placed at the measuring point. The start measure of the machine was selected and the results presented on the display of the machine as well as its printed paper.

3.5.4 Compositional analysis

3.5.4.1 Butter fat test

The fat content was analyzed in the department of Dairy and Food Science Egerton University. Rose Gottlieb method according to AOAC (2000) Official Method 905.02 with modification (using the soxhlet method to evaporate the ethers) was employed. The procedure involved treating a sample of milk with ammonia to dissolve the precipitates and ethanol to precipitate the proteins. Fat extraction was done using the mixed ethers (petroleum ether and diethyl ether) before evaporation and weighing of the residue.

Homogenous sample of milk (10 ml) was accurately weighed into an extraction tube and 1.25ml of ammonium hydroxide (sp. gr. 0.8974) was added, stoppered and mixed thoroughly, each time releasing pressure by un-stoppering the tube. Absolute ethanol (10ml) was then added to the sample, mixed thoroughly and 25 ml diethyl ether (peroxide free) added and shaken thoroughly each time releasing pressure for 1 min and 25ml of petroleum ether (boiling range 40–60^{0}C) was added and again vigorously shaken for half a minute taking care of pressure build up. The sample was let to stand still until upper liquid which was clear then decanted carefully to remove the ether layer in a pre-weighed flat bottomed flask ensuring that none of the aqueous layer pours out off with ether layer. The extraction tube was washed with equal parts of the two ethers and washings added to flat bottomed flask.

The extraction was repeated twice each time using 15 ml of diethyl ether and the petroleum ether as prescribed earlier. The solvent was evaporated completely in soxhlet method and finally dried in an oven. It was then placed in a desiccator to cool. The flat bottled flask was weighed and then petroleum ether was added to dissolve the fat and decanted taking care to leave any sediment in the flask. It was then dried in the oven and weighed as before. Correct weight of extracted fat was determined by deducting the blank test results carried out to determine accuracy of the reagents used.

The % fat in milk was calculated using the following formula;

%Fat= Weight of fat/Sample weight x 100.

3.5.4.2 Total solids

The Richmond's formula was used to calculate the total solids and solids not fat present in the milk samples.

Total Solids % = CLR/4 + 1.21 F + 0.14

SNF % = CLR/4 + 0.21 F + 0.14

Where, CLR is the Corrected lactometer Reading and F is the Fat content in milk at a temperature of 20^0C.

3.5.5 Statistical analysis

The questionnaires were first edited and coded to ensure completeness and accuracy. The Statistical Package for the Social Sciences (SPSS) version 22 was used to generate descriptive statistics for the survey data.

In laboratory experimentation, Analysis of Variance (ANOVA) was used where PROC GLM procedure of the Statistical Analysis System version 9.1.3 (SAS, 2006) was employed. Means were separated using Least Significance Difference (LSD) whenever there was variability at $P \leq 0.05$ (Gacula, 1984).

The statistical model for the data analysis was;

$$\mathbf{Y_{ijkl} = \mu + \beta_j + T_k + \alpha_l + \beta T_{jk} + \beta\alpha_{jl} + T\alpha_{kl} + \beta T\alpha_{jkl} + \varepsilon_{ijkl}}$$

Where; Y_{ijkl} is the response variable for milk quality, μ is the overall mean, β_j is the j^{th} effect due to CBE blocking, T_k is the k^{th} effect due to treatment, α_l is the l^{th} effect due to time, βT_{jk} is the j^{th} CBE blocking effect on the k^{th} treatment, $\beta\alpha_{jl}$ is the j^{th} CBE blocking effect on the l^{th} time, $T\alpha_{kl}$ is the k^{th} treatment effect on the l^{th} time, $\beta T\alpha_{jkl}$ is the interaction of the j^{th} effect due to CBE blocking effect, the k^{th} effect due to treatment and the l^{th} effect due to time and ε_{ijkl} is the random error term.

CHAPTER FOUR

RESULTS AND DISCUSSIONS

4.1 Code of the hygienic practices at various levels in the dairy value chain

4.1.1 Dairy farmers and transporters hygienic practices

Results on dairy farmer's hygienic practices (Figure 2) indicated that cleaning of the cow shed by farmers was not prioritized. Hence, a period of one month elapsed before the cowshed was attended at a rate of 75% and 25% in Olenguruone and Ngorika, respectively. Farmers should maintain elaborate farm hygiene in the milking parlor and sheds to ensure clean milk production (Gietema, 2002). This will facilitate maintenance of a healthy herd. The study also realized that 49% and 51% of the farmers in Olenguruone and Ngorika respectively did not use detergent when washing their hands prior to milking. Cleaning of hands without detergents, as indicated by some farmers, can contribute to microbial contamination in milk. Milking management and hygiene protocol are important to milk quality because they minimize transmission of mastitis in farms. Nevertheless, 50.6% and 49.4% of the farmers in Olenguruone and Ngorika respectively, used a reusable udder cloth while milking their animals. The same udder cloth was used to dry their hands before milking. This compromises hygiene milking practices and may contribute to cross transmission of mastitis from an infected cow to a healthy cow.

After milking, 50% of the farmers held the milk on their farms to attend to other chores in both locations. Farmers took an additional 30 minutes to deliver their milk to transporters at 49.2% and 50.8% in Olenguruone and Ngorika respectively. This contributed to delays in milk delivery to the chilling plants in both CBEs. Further delays were observed during transportation where 60% and 40% of the transporters in Ngorika and Olenguruone respectively, took more than 2 hours to transport the milk from the farms to the CBEs cooling plants. Milk quality analysis tests were not carried out by the transporters before bulking the milk at the farm levels. This was because 60% of the milk handlers from Ngorika and 100% from Olenguruone had no basic training in milk handling and hygiene. The mixing of high and low quality milk from different suppliers without grading led to milk quality deterioration. During transportation, milk handling hygiene was rarely observed with at least 20% of the transporters from Ngorika failing to wash their hands before handling the milk along the routes. In Olenguruone, transporters cleaned their

hands before handling milk and many of them transported milk to the cooling plant using the farmer's containers.

Plastic containers are not ideal for milk handling since they are impossible to clean. However, the study found that 90.4 % and 49.6 % of the farmers in Olenguruone and Ngorika respectively, delivered their milk using plastic containers owing to their low cost, availability and convenience. According to Wafula *et al.* (2016) reduction of microbial load in plastic containers was difficult and could contribute highly to milk contamination. Therefore, dairy actors should be encouraged to use food grade plastic containers. The milk transport modes used included; donkeys, motor bikes, lorry, pickups, tractor and individual farmer deliveries. In Ngorika, milk transportation was done using aluminum cans and though cleaning was not assessed, effectiveness of cleaning is questionable as lack of portable water for rinsing was observed. In Olenguruone, all the transporters used plastic containers. Cleaning of the plastic jerry cans involved use of hot water and detergent although its effectiveness was not evaluated. According to Mwangi *et al.* (2000), use of plastic containers contribute to milk quality deterioration since they are impossible to clean especially around the handles that are not accessible during cleaning. According to Orregård (2013) study, on quality analysis of raw milk along the value chain of the informal milk market, use of aluminum cans is a more appropriate method of milk transportation unlike plastic containers. Use of plastic containers, lack of cooling before delivery and long duration in transportation favours quick bacterial multiplication (Swai and Schoonman, 2011).

Farmer's awareness concerning antibiotic residues in milk was found to be at 49.7% and 50.3% for Olenguruone and Ngorika respectively. Additionally, half of the farmers in both CBEs were not aware of the effects of antibiotic residues in milk quality, the withdrawal period required for various antibiotics and their effects on human health. These results compare to those of Orregård (2013), where farmers did not understand about antibiotic residues and their effect on milk quality. The same author concluded that, antibiotic residues in milk can be traced exclusively from the farms. Further findings from Aboge *et al*. (2000) indicate that, to eliminate the challenge of antibiotic residues in milk, care should be taken at both the farm and market level.

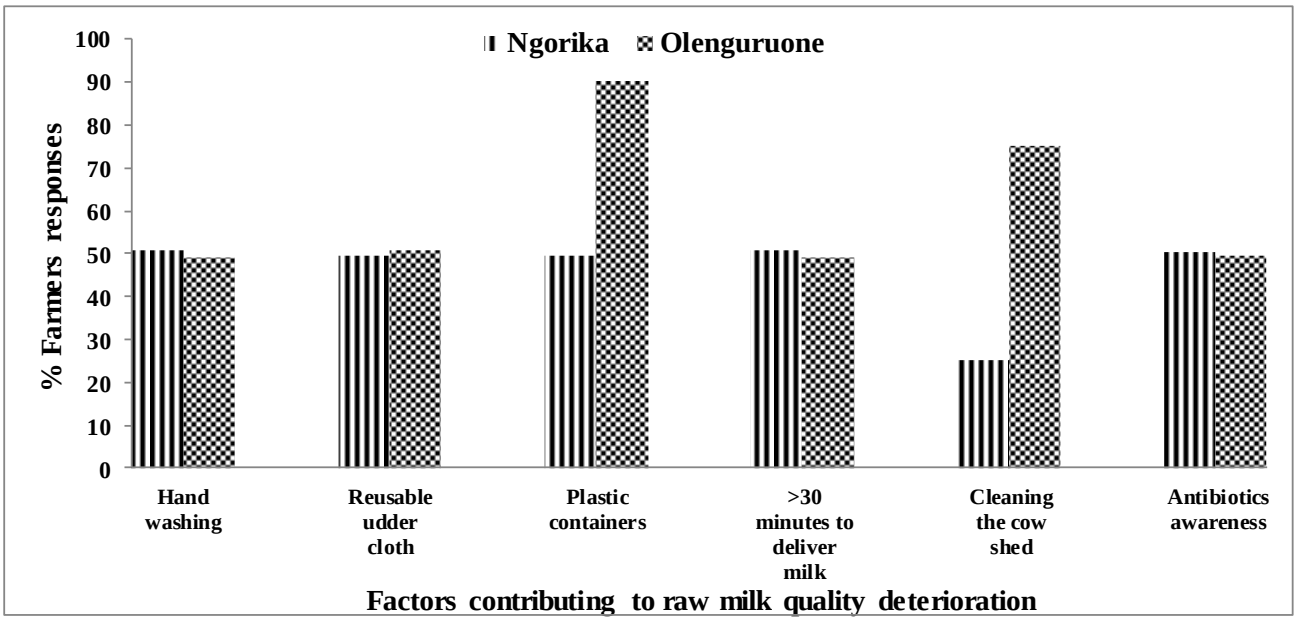


Figure 2: Dairy farmers practices contributing to raw milk quality deterioration observed at the farm level.

4.1.2 Milk handling and preservation at the collection and bulking enterprises

Personnel handling the milk at the cooling plants were qualified dairy technologist in both CBEs. Cleaning of the cooling tank was done immediately after emptying the milk to the tanker. This compares to a study done by Pandey and Voskuil, (2011) which recommends that, the cooler must be cleaned, disinfected and kept in good condition after each milk collection. Maintaining hygiene in Ngorika cooling plant premise was easy. On the other hand, Olenguruone cooling plant premise was a semi-permanent building with a rough floor which compromised on hygiene. Rain and borehole were the available sources of water in both CBEs. This water was not treated before use, a factor that could contribute to milk quality deterioration. Ideally, milk should be cooled within 2-3 hours after milking. Contrary, in both CBEs some milk delivery exceeded the recommended time which had a negative impact on milk quality. The cooling efficiency in both CBEs was a challenge. The coolers took more than 3 hours to cool the milk from 18^0C to 4^0C. The study found that monitoring of the cooler efficiency to prompt maintenance and repair was hardly done. For instance in Olenguruone, it was done after 3 months or during breakdowns. According to Pandey and Voskuil (2011), milk must be cooled immediately to minimize bacteria multiplication and should be protected from contamination during transportation and subsequent

storage. Poor quality milk cannot be improved by cooling at a later stage (Orregård, 2013), therefore there is a need to improve and hasten raw milk collection system and enhance the cooling efficiency.

4.1.3 Milk handling at the processor level

At the reception platform at Happy Cow Limited, quality control personnel cleaned exit where the milk was to be emptied before connecting the pipes. The quality control personnel were dairy technologists. A sample was then drawn from each compartment separately for quality analysis (%lactic acid, alcohol test, lactometer test, total plate count, 10 minutes resazurin test and antibiotic residue delvo test). There was no significant temperature variation observed in the milk after transportation from the CBEs. The tanker was cleaned immediately (full CIP) after emptying milk. The concentrations of the cleaning detergents used in the tankers were checked. Borehole water for general cleaning was treated with 3ppm chlorine while that used for sanitation was at 300ppm. This showed that the milk processor was careful on matters regarding milk quality and handling hygiene.

4.2 Identification of quality control critical control points

4.2.1 Characteristics of raw milk

During the field visits, eight steps were identified and listed in a flow diagram (Figure 3) to illustrate the occurrence of activities in the delivery of milk from the farm to the cooling plant. The steps involved three participants including farmers, transporters and graders. The farmer handles the milk from milking to the collection point where the transporter collects the milk, bulks and transports to the cooling plant. Subsequently, the milk is graded at the CBE platform and bulked in the cooler by the grader.

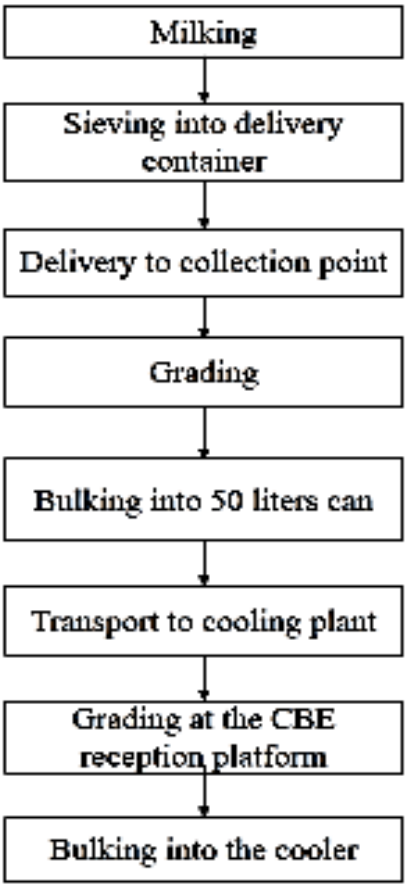


Figure 3: Flow diagram of steps for raw milk collection to cooling plant

Milk delivered at the collection chain had a lot of foreign material for instance cow dung, fir and organic matter. This could have originated from poor milking procedure and failure to sieve the milk before delivery. Presence of foreign material contributes to the increase in microbial contaminants, objectionable oduors and appearance. Failure to observe the withdrawal period after treating the animals at the farm level will allow introduction of antibiotic residues into the milk. Antibiotic residues in milk are a chemical hazard to milk consumers due the allergic reactions and development of antibiotic resistance in human. Adulteration with water and preservatives could be done by farmers/herders although during transportation, chances of adulteration are also likely. These malpractices could lead to milk safety and quality concerns. The factors affecting milk quality were examined and are reported in Table 4 including their workable corrective actions.

According to EAS (2007), milk shall contain not less than 3.25% milk fat and not less than 8.50% milk solids not fat. It should have a characteristic creamy-white colour, free from flavours, taints and objectionable matter. It should not clot on boiling and should test negative to the alcohol test. It should not contain added water, preservatives, or other added substances, no proportion of a natural constituent should be removed. The density should be within the range of 1.028 -1.036 g/ml at 20 ^{0}C and not more than 0.17% lactic acid. The freezing point depression should be within 0.525 and 0.550^{0}C and it shall conform to maximum limits of pesticides, antibiotic and veterinary drugs residues.

Table 4: List of possible factors contributing to quality deterioration at each step

Process	Description/Activities	Possible factors and their Sources	Control Measure
Milking	The cow is entered in the parlor and is restrained. Milking takes off.	**Physical:** Animal fur, dung, personal effects and dirt that may come with the milking procedure. **Chemical:** Antibiotics, milking jelly, H_2O_2, Somatic cell count. **Biological:** Bacterial load	Sensitizing farmers on, milking hygiene, withdrawal period and mastitis treatment.
Sieving the milk into the delivery cans	The farmer sieves the milk as it's transferred into the delivery container.	**Physical:** cleanliness of the sieve. **Chemical:** detergents residues **Biological:** microbial contamination	Sensitize the farmers on hygiene.
Transport to collection points	The milk is taken at the collection point.	**Biological:** microbial multiplication due to time lapse.	Sensitize the farmers.
Grading	The milk is graded at the collection point before bulking into 50 liters aluminum cans.	**Physical:** Introduced. **Chemical:** H_2O_2, antibiotics.	Proper grading, sensitize the farmers.
Bulking into 50 liters aluminum cans	Graded milk is collected into 50liters aluminum cans.	**Chemical:** detergents residues, antibiotics. **Biological:** microbial contamination	Proper rinsing of the aluminum cans before bulking.
Transport to the cooling center	The bulked milk is transferred to CBE.	Biological: multiplication of the microbes due to time lapse.	Sensitization of the transporters.
CBE reception plat form	The milk is graded again for acceptance or rejection.	**Biological:** microbial growth due to time lapse.	Sensitization of the quality control personnel at the reception.
Cooling tank	The milk is pumped into the cooling tank.	**Biological:** microbial multiplication. **Chemical:** detergents residues, antibiotics and adulterants due to bulking.	Sensitization of all the stakeholders in the value chain.

4.2.2. Identification of critical control points

The Critical Control Points (CCPs) were identified in line with HACCP principles concept using the deteriorating factors identified above in table 4. To categorize the factors as prerequisite program or CCP as in table 5, risk assessment was carried out where the likelihood and severity were considered. The microbial contamination, hydrogen peroxide, cleaning detergents residues, exhaust fumes, organic matter and antibiotic residues were identified as factors with high risk in milk quality deterioration. The decision made at a certain level of risk was determined by likelihood of occurrence and severity. Where negligible, or the impact was minor, it was controllable at that particular step and records kept. If the impact was severe, the factor was considered a CCP and therefore control factor was determined. Based on the identified CCP, the corrective actions were established that would ensure the safety and quality of the milk delivered to the CBEs.

According to ISO 22000:2005 food safety management system HACCP system has been recommended as one of the most effective ways of ensuring high quality and safe food. According to Mwangi *et al.* (2000), the HACCP system is a preventive approach that identifies the points in a process that are hazardous, their risk factors and potential level of risk so that critical control points for remedial action can be implemented. According to FAO/WHO (1998), risk is the likelihood of occurrence and it is a function of likelihood of occurrence and the control level (seriousness level).

Table 5 : Raw milk quality risk assessment

Process	Factor		Likelihood (L)	Severity (S)	Significance	Control	Recommendation
Milking	Physical: Animal fur, dung, personal effects and dirt.		3	3	9	CCP	Sensitize farmer, milkers on clean milk production
	Chemical	H_2O_2	1	3	3	PRP	Reject milk with H_2O_2
		Antibiotics	3	3	9	CCP	Sensitize farmers on the withdrawal period.
	Biological	Somatic cell count.	3	3	9	CCP	Sensitize farmers on animal husbandry
		Microbial load	3	3	9	CCP	Clean milk production and delivery time
Sieving the milk into delivery cans	Physical: dirt from milk		3	1	3	PRP	Clean milk production
	Chemical: cleaning detergents		2	2	4	PRP	Proper rinsing of the milking equipment.
	Biological: microbial load		1	3	3	PRP	Proper cleaning of the equipment.
Transport to collection points	Chemical: H_2O_2, alkaline		2	3	6	CCP	Reject milk with alkaline and H_2O_2.
	Biological: microbial multiplication		2	3	6	CCP	Quick delivery to collection point
Grading	Physical: introduced		1	2	2	PRP	Hygiene
	Biological: contamination		3	3	9	CCP	Proper sanitation of grading equipment.
Bulking into 50 liters aluminum cans	Physical: introduced dirt		1	3	3	PRP	Training and extension
	Chemical: H_2O_2, antibiotics, cleaning detergents		3	3	9	CCP	Traceability
	Biological: microbial load, somatic cell count.		3	3	9	CCP	Quick delivery and good animal husbandry.
Transport to the cooling center	Chemical: H_2O_2, alkaline		2	3	6	CCP	Reject the milk with H_2O_2 and alkaline.
	Biological: microbial multiplication		3	3	9	CCP	Quick delivery to CBE
CBE reception plat form	Physical: introduced dirt		2	3	6	CCP	Proper hygiene
	Chemical: exhaust fumes		2	3	6	CCP	Sensitize transporters on GMPs
	Biological: microbial multiplication		3	3	9	CCP	Proper sensitization of the grading equipment.
Cooling tank	Chemical: cleaning detergents		2	3	6	CCP	Proper rinsing of the tanks.
	Biological: microbial multiplication		3	3	9	CCP	Proper maintenance of the cooler.

The decision tree in appendix 2, assisted in identifying critical control points as indicated in Table 6. This identified 6 out of 8 of the processes as CCP with a significance of 9. Poor milking procedures, the health of dairy cows and delayed milk delivery are factors under the jurisdiction of the farmers. As the recommendations in Table 5 outline, farmer's keenness to hygienic milking and prompt delivery of milk should be emphasized.

It was identified that due to low milk production in the farms, transporters had to bulk milk from 6 to 9 farms to fill one can. The mixing of the milk gave chance to mixing good quality and poor quality milk leading to quality deterioration of the bulk. Due to the modes of transport and poor road networks in the rural areas serving the two CBEs, there was delayed delivery of the milk from collection points to the cooling plant. There were chances of adulteration of the milk in transit by unscrupulous transporters. The last CCP was identified as the inefficiency of the cooler which would take long before cooling the milk which gave chance to further multiplication of microorganisms.

From the identified CCP for each process, measurable parameters to ascertain quality in the delivery chain were identified as outlined in the CCP plan in Table 6. The farmer has to deliver the milk promptly and can be evaluated by the temperature range of the delivered milk. This can be done with the background knowledge that, the faster the delivery the more the milk temperature will near the udder temperatures that range from 25^0C to 37^0C. The thermometer reading should be carried out at collection points and the farmer sensitized to adhere to prompt delivery practice. This will eliminate delays in the homes where farmers milk and first attend to other chores. At the transport to the collection point, there were neither measurable parameters nor any corrective action that would be concluded as a CCP.

To ensure bulking of quality milk at the collection points, milk should be tested on density using a lactometer, delivery temperature, protein stability using alcohol test. Milk that passes the above tests would be considered to be of good quality. To safeguard on quality, the milk should be rejected and records kept for periodic quality monitoring. Subsequently, all the milk from the transporters at the platform and any suspected milk should be subjected to advanced laboratory analysis by the quality controller at the cooling plant. Lastly, the cooler should effectively cool the milk from 25 to 4 degrees Celsius in the least time possible. To verify the efficiency at the cooling plant, the measurable parameters identified were microbial counts, adulteration, density,

temperature, detergent residues and protein stability. Monitoring procedures should give an indicator of the point where quality is bound to deteriorate, who, when, what and how to monitor. The records generated could act as a reference point for corrective actions.

Table 6: Determination of critical control points using the decision tree

Process	Factor	Significance	Question 1	Question 2	Question 3	Conclusion
Milking	Poor milking procedure, utensils, milking bucket, cow health	9	Yes	Yes	Yes	CCP
Transport to collection points	Delayed delivery	6	Yes	No	Yes	CCP
Bulking into 50 liters aluminum cans	Mixing of 6 to 9 farmer's milk increases chances of mixing good quality milk with poor quality milk.	9	Yes	No	Yes	CCP
Transport to the cooling center	Delayed delivery , adulterants	9	Yes	Yes	Yes	CCP
CBE reception plat form	Delays while grading and dirt from the surrounding. Exhaust fumes collected from delivery vehicles.	9	Yes	Yes	Yes	CCP
Cooling tank	Poor efficiency of the cooler	9	Yes	Yes	Yes	CCP

Table 7: Critical Control Points Plan

Process	Measurable parameter	Critical limit	Monitoring				Correction	Corrective action	Records	Verification
			Who	What	When	How				
Milking	Delivery temperature	25^0C -37^0C	Farmer	Temperature	At delivery	Thermometer reading	Sensitizing the farmer	Continued advise	Temperature recorded daily	Quality checks
Transport to collection point	N/A	N/A	Farmer	N/A	At delivery	N/A	Sensitizing the farmer	N/A	Acceptance or rejections	Quality checks
Grading and bulking	Temperature, density, protein stability	>28^0C 1.027-1.033g/ml, alcohol negative	Grader	Lactometer reading, alcohol test	Every day	Alcohol gun, Lactometer and thermometer	Sensitize farmers	Reject non-conforming milk	Temperature, alcohol tests results and lactometer readings recorded daily	Quality checks
CBE platform	Traces of lead in milk	N/A	Quality controller / grader	Presence of lead	When suspected	Advanced lab Analysis	N/A	Avoid grading while the motor mode of transport is running	Instances recorded	Quality checks
Cooling tank	Bacterial counts, adulteration, density, temperature, protein stability, detergents residues	Time from 25^0C to 4^0C	Quality controller	Cooler efficiency (time and temperature), cleaning effectiveness	Every day	Temperature, use of litmus paper, bacterial count, stop watch	Sensitize the quality controllers	Ensure proper cooling and faster grading, use litmus paper	Cooler efficiency records available	Quality checks

N/A –indicates not applicable at that factor/level.

Milk quality encompasses prevention on each step of production. Quality control systems aimed at the prevention of defects, rather than their detection. Quality control occurs at every step in the production, as a raw material on farm condition. The developed CCP compare with those developed by Keski and Gulsunoglu, (2012) who reported on possible hazards, control and orientation of raw milk although he went further to elaborate several CCP at the farm level. The biological, chemical and physical hazards pose food safety and quality risks in a milk production system. Pre-requisite programs are recommended and proven management procedures that help prevent low risk food safety problems from occurring and are the foundation of the HACCP study. Operational pre-requisite programs and risk analysis need to be established for the effective applicability of HACCP that determine physical, chemical and microbiological hazards in dairy industry. According to Torkar and Teger (2004), to achieve food safety and reduce risk, implementing the hazard analysis critical control points (HACCP) concept and quality assurance from the farm to the dairy plant should be considered. This study therefore agrees with (Karakök, 2007), who recommends that, it is paramount for every farm to determine and continuously control critical points of fresh milk production which will prevent possible hazards. The benefit of adhering to the CCPs leads to improved milk quality and enhances consumer confidence.

4.3 Raw milk quality along the smallholder collection chain

4.3.1 Microbial quality

The samples analyzed had higher total plate counts and coliform counts compared to KEBS standards. According to EAS (2007), bacteriological quality grade III for total plate and coliform counts are 2×10^6 and 1×10^3 cfu/ml, respectively. In Ngorika, the mean total plate count and coliform count per ml (Table 8) was not significantly different ($P \leq 0.05$) among the milk samples collected from the route composite, CBE cooler and processor tanker but it was significantly different ($P \leq 0.05$) at the can level. The means total plate count at the can level were not significantly different ($P \leq 0.05$) from the means at the cooler level. This could have been due to the dilution effect where good quality milk was mixed with low quality milk in the cooler. The means total plate count at the processor tanker level were significantly different ($P \leq 0.05$) from the can level. This could have been contributed by time lapse during transportation and cooling facilitating microbial multiplication.

Table 8: Means for total plate count cfu/ml, coliform counts cfu/ml, lactic acid and resazurin test for Ngorika samples

	Sampling levels			
Test	**Can**	**Route composite**	**CBE cooler**	**Processor tanker**
TPC	$2.49 \times 10^8 \pm 3.87^b$	$6.57 \times 10^8 \pm 5.89^a$	$5.104\ 1\times 10^8 \pm 2.85^{ab}$	$6.72 \times 10^8 \pm 3.24^a$
CC	$6.10 \times 10^5 \pm 8.16^b$	$1.74\times 10^6 \pm 5.14^a$	$8.97 \times 10^6 \pm 4.10^{ab}$	$1.41 \times 10^6 \pm 7.04^{ab}$
LA	0.15 ± 0.01^b	0.16 ± 0.01^a	0.15 ± 0.004^{ab}	0.15 ± 0.01^{ab}
RT	3.97 ± 1.21^a	3.65 ± 0.93^a	3.85 ± 0.56^a	4.11 ± 0.60^a

Means within a row marked with different letters are significantly different at ($P \leq 0.05$) for Total Plate Count (TPC), Coliform count (CC), Lactic Acid (LA) and Resazurin Test (RT).

Table 9: Means for total plate count cfu/ml, coliform counts cfu/ml, lactic acid and resazurin test for Olenguruone samples

	Sampling levels			
Test	**Can**	**Route composite**	**CBE cooler**	**Processor tanker**
TPC	$3.61 \times 10^6 \pm 3.81^b$	$4.67 \times 10^6 \pm 3.40^b$	$5.87\times 10^6 \pm 2.56^b$	$1.37 \times 10^7 \pm 3.82^a$
CC	$2.22 \times 10^4 \pm 6.37^a$	$5.99 \times 10^4 \pm 3.51^a$	$2.09 \times 10^5 \pm 7.33^a$	$2.46 \times 10^5 \pm 8.93^a$
LA	0.15 ± 0.02^a	0.15 ± 0.01^a	0.15 ± 0.01^a	0.15 ± 0.01^a
RT	4.82 ± 0.74^a	5.00 ± 0.59^a	5.00 ± 0.50^a	4.89 ± 0.00^a

Means within a row marked with different letters are significantly different at ($P \leq 0.05$) for Total Plate Count (TPC), Coliform count (CC), Lactic Acid (LA) and Resazurin Test (RT)

In Olenguruone, the means for TPC were significantly different ($P \leq 0.05$) at the processor tanker level compared to the can, composite route and cooler levels (Table 9). The means CC count cfu/ml for the can and route cooler and tanker sample were not significantly different ($P \leq 0.05$).

There was a significant increase ($P \leq 0.05$) in microbial population for TPC cfu/ml from the can to the tanker sampling levels for both locations (Table 8 and 9). This could have been majorly contributed by several factors including; the poor cooling efficiency of the coolers that took more than 3 hours to cool the milk to 4^0C, poor milking practices, use of the plastic containers in the collection of milk and inappropriate milk handling through the value chain. Additionally, delays in the collection routes were observed. In Ngorika, milk collection in the routes took 5 to 6 hours before the milk was delivered to the cooling plant. In addition, some farmers were not separating the morning and evening milk during delivery. Mixing of the two intakes and inappropriate can cleaning could have favoured microbial multiplication leading to the high microbial counts in the study. In Olenguruone, milk collection was majorly by use of motorbikes. They took an average of one hour to transport the milk to the cooling plant. It was also noted that evening milk was not supplied by the farmers from this CBE. Moreover, the morning milk delivered was warm as indicated by touching the farmer's cans. These differences in each CBE operations could explain the different microbial counts.

The dairy farmer's hygienic practices results indicated poor clean milk production at farm levels and poor milk handling hygiene during transportation. The identification of critical control points indicated that milk had foreign matter, a factor that could lead to milk quality deterioration. These could have contributed to the high TPC and CC indicated in microbial analysis in this study.

Warm collection in the dairy value supply chain creates an optimum environment for microbial growth consequently causing milk quality deterioration (Mwangi *et al.*, 2000: Orregård, 2013). According to Mwangi *et al.* (2000), this may be due the contribution of insufficient pre-milking udder preparation, insufficient cleaning of milk handling equipment, and use of poor quality water for cleaning, the storage time and lack of cold chain facility starting from the production site. According to Doyle *et al.* (2015), increases in TPC observed along the value chain may be due to several factors like contamination at the farm, storage and transport using improperly cleaned milk cans, and lack of controlled temperature during transportation. As reported by Van Kessel *et al.* (2004), the use of insufficient and poor quality water for cleaning of milk handling equipment can result in milk residues on equipment surfaces that provide nutrients for the growth and multiplication of bacteria that can then contaminate the milk. Murphy and Boor (2000) noted that ineffective cleaning, use of water without heat treatment and the absence of sanitizers tend to fasten growth of less heat resistant organisms. Similarly, mastitis infected cows can also contribute to high TPC.

Generally, the presence of coliforms in milk confirms that the milk has been contaminated with fecal materials and it's an indicator of poor sanitary conditions in the production and handling of the milk starting from production (Orregård, 2013). Accordingly, poor herd/farm hygiene, use of contaminated water, unsanitary milking practices, and use of improperly washed equipment for storage and distribution can all lead to elevated coliform count (CC) in raw milk (Gemechu *et al.,* 2015). The fact that high proportion (90%) of the milk samples taken from all levels had coliform counts way above the maximum limit of KEBS standards accepted for CC in grade III raw milk specification, it provides irrefutable evidence that the udder of the cows have been soiled with fecal materials and/or the udder was improperly washed; i.e., milk contamination in the study area happened starting from milking of the cows. In addition, the presence of coliform in an aseptically collected sample of raw milk shows the use of bacteriological low quality water, either for washing utensils or mixing in raw milk (Farhan and Salik, 2007). Apart from safety and public health concerns, high contaminations by coliforms results in off flavours in milk and reduce shelf life of dairy products (Reta and Addis, 2015: Kaindi *et al.,* 2011). Generally, the bacterial generation (doubling) time is between 10 to 15 minutes depending on the conditions. In this study, milk was transported while warm and in plastic jerry cans. Karuga (2009), explain that the plastic jerry cans could contribute to milk quality deterioration unlike the recommended aluminum containers that don't have adhesive properties and are easy to clean. According to Orregård (2013), aluminum cans allows better hygienic handling unlike plastic jerry cans.

Moreover, more than 3hrs, where natural lactoperoxidase enzyme could sustain the milk quality, were surpassed in some routes before refrigeration could take place. Cooling of milk is advocated to help in significantly reducing the multiplication of bacteria and in turn reduce spoilage (Kurwijila, 2006). According to Reta and Addis (2015), higher coliform and total plate counts observed in different study areas of the country could be due to contamination of raw milk samples either from the cows, the milker, milk container and the milking environment and transportation utensils. Similarly, adequate sanitary measures including proper handling of the milk, cow, personal hygiene, use of hygienic milking and processing equipment, improving milk handling practices should be taken seriously (Teklemariam and Estifanos, 2015). Omore *et al.* (2001) indicates that there is generally a high proportion of raw milk in the Kenyan market that does not achieve KEBS standards and probably, over 50 percent of the raw milk in the market would convert to losses if the standards were to be enforced. Lack of cold chain may be a major factor contributing to milk quality challenges.

4.3.2 Titratable acidity test

The means separation indicated non-significant difference ($P \leq 0.05$) at the can, cooler and tanker level unlike the route level in Ngorika (Table 8). The difference in acidity between the cooler level and the route level could have been contributed by the dilution factor as milk is being bulked together in the cooling tank from the routes. A different scenario was observed in Olenguruone where non-significant difference ($P \leq 0.05$) at the can, route, cooler and tanker level samples (Table 9). All the samples at the can, route, cooler and tanker levels were found to have acidity levels within the acceptable range of 0.16 ± 0.02. According to Salman and Hagar (2013) findings, raw milk acidity was in the range of 0.15-0.18% lactic acid implying that higher acidity in milk (beyond 0.18%) suggests a high microbial count in the milk.

4.3.3 Resazurin test

Resazurin test uses phenolphthalein indicator resazurin to measure the bacteriological quality of milk. The majority of the organisms in milk are capable of reducing and decolorizing the resazurin dye. When bacteria grow in the milk they utilize oxygen, the rate of removal or reduction of oxygen is proportional to the keeping quality and consequently color disappearance (Draaiyer *et al.,* 2009). Thus, the time used to reduce the dye is taken as a measure of the number of organisms in milk. Although it is likely a measure of the total metabolic reactions proceeding at the cell surface of the bacteria. The Resazurin dye is more sensitive than the methylene blue and for this reason this test provides a rapid measure of the keeping quality of milk. The methylene blue reduction test has lost much of its popularity because of its low correlation with other bacterial procedures particularly in those samples which show extensive multiplication of the psychotropic species. There was no significant difference ($P \leq 0.05$) for 10 minutes resazurin test at the four sampling levels (Table 8 and 9) as indicated by the means separation.

Table 10: Correlation of some quality results evaluated in smallholder dairy farmers supply systems in Kenya

	TIME	ROUTE	TPC	CC	LA	RT
TIME	**1.00000**	**-0.74248*** **	**0.42845** **	**0.53512*** **	**0.22400**	**-0.47639** **
ROUTE		**1.00000**	**-0.46522*** **	**-0.47591*** **	**-0.17855**	**0.64902*** **
TPC			**1.00000**	**0.67682*** **	**-0.06786**	**-0.23962**
CC				**1.00000**	**0.02256**	**-0.21445**
LA					**1.00000**	**-0.34558***
RT						**1.00000**

****, **, *** indicates significance at P≤0.05, P ≤0.01 and P≤0.001 respectively.***

The correlation between 10min resazurin test and TPC was not significant ($P \leq 0.05$) at the route level (Table 10). According to EAS, (2007) raw milk specification, only 20.6% of samples were unacceptable based on this test contrary to the TBC where all the samples were way above grade III raw milk standards which is unacceptable. The study results agrees with the study carried out by Muliro *et al.* (2013) on quality assessment of raw camel milk using dye reduction tests that the resazurin test is not reliable as a measure of total viable bacterial count. According to Murphy and Boor (2000), a significant correlation ($P \leq 0.01$) between the total plate count and time used to deliver milk for cooling was observed. Moreover, Kurwijila *et al*. (1992) and Muliro *et al.* (2013), explains that, the TPC test has been reported to be generally accepted as the most accurate and informative method of testing the bacteriological quality of milk.

4.3.4 Somatic cell count

Most of the samples were in the range of 150-500 cells/1000 ml (Figure 4). This could be considered as an indication of presence of sub-clinical mastitis in the farm and could have contributed to the high bacterial load. In Ngorika, no sample was within the 0 to 200,000 SCC which reflects absence of somatic cells in milk. Thus, it can be concluded to have higher incidences of mastitis compared to Olenguruone. Somatic cell count should not exceed 300,000 per ml when tested in accordance with ISO 13366 procedure. The California Mastitis Test (CMT) has been used for more than 50-years and continues to be the most accurate

screening test for subclinical mastitis (Ruegg and Reinemann, 2002). The heavier the gel the higher the somatic cells in the milk and vice versa which indicates the leukocyte count (Quinn *et al.*, 1994). *Staphylococcus agalactiae* is known to be an occasional cause of high bacterial counts and subclinical mastitis problems and should be considered when both the SCC and TPC are high (Ruegg and Reinemann, 2002). Thus the higher somatic cell counts detected could have contributed to the high levels of the total plate counts observed.

Increased somatic cell numbers are positively correlated with concentrations of plasmin, a heat-stable protease, and of lipoprotein lipase in freshly produced milk (Barbano *et al.*, 2006). Activities of these enzymes can supplement those of bacterial hydrolases, hence shortening the time to spoilage. The major determinants of quantities of these enzymes in the milk supply are the initial cell numbers of psychrotrophic bacteria, their generation times, abilities to produce specific enzymes, and the time and temperature at which the milk is stored before processing (Ledenbach and Marshall, 2009). Several conditions must exist for lipolyzed flavor to develop from residual lipases in processed dairy foods, that is, large numbers (>10^6 cfu/ml) of lipase producers, stability of the enzyme to the thermal process, long-term storage and favorable conditions of temperature, pH, and water activity.

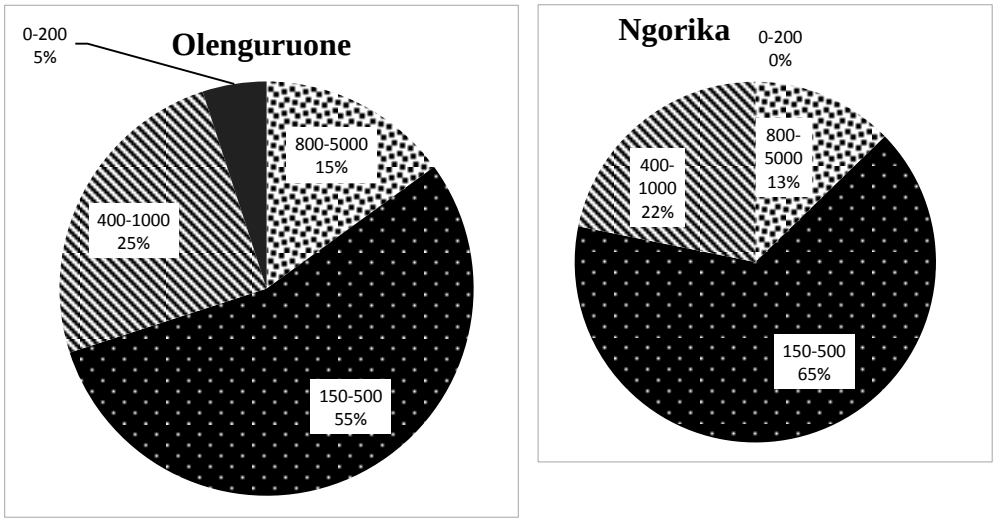


Figure 4: Somatic cell count per 1000 ml per collection and bulking enterprises.

4.3.5 Antibiotic residues

In total, 74 samples were analyzed in both CBE and out of these, 54% and 35% from Olenguruone and Ngorika respectively, were positive (Figure 5). The delvo test was carried out for the composite samples at the route and the cooler levels. The delvo test is easy to use

and covers the broadest spectrum of antibiotic residues in the industry. It is reliable and accurate with detection levels closest to maximum residue levels and safe tolerance levels (Hillerton *et al.*, (1999). This could have been due to the fact that the farmers were less aware of the withdrawal periods after treating their animals as indicated by the dairy farmer's practices. The results showed high incidences of β- lactams that indicates unawareness of the farmers with the withdrawal period or ignorance.

The level and duration of antibiotic diffusion into milk depends upon several factors including the particular antibiotic, its concentration and method of preparation (aqueous solution, nature of suspending medium). The method of preparation markedly influences retention and can affect adhesion of the antibiotic to equipment and pipelines (Mullan, 2003). Antibiotics in milk are a major concern due to the risk of allergic reactions, development of antibiotic resistant pathogen and inhibition of dairy starter cultures used to develop acid (e.g., lactic acid bacteria), which can result in the loss of significant amounts of product and milk (Popelka *et al.*, 2004). According to Orregård (2013), many farmers could not remember the last time they used antibiotics due to minimal or no interest on how the veterinarian treated the cow. Based on those facts, unclear statements were given, both regarding the frequency of treatment and the equipment used during treatment. This study corresponds to another study carried out by Aboge *et al.* (2000) on antimicrobial residues detected on marketed milk in Kenya. According to Shitandi (2004), eighteen percent (18%) of the samples from small-scale producers in his study area were significantly β-lactam positive significant ($P<0.001$). Other studies carried out within Kenya showed that many animal products on the Kenyan market have a high level of drug residues which is unacceptable (Muriuki, 2001; Odero, 2002). According to Gallagher (2015), consumption of food with antibiotic residues can lead to bacteria becoming completely resistant to treatment in human beings a situation reffered to as antibiotic apocalypse.

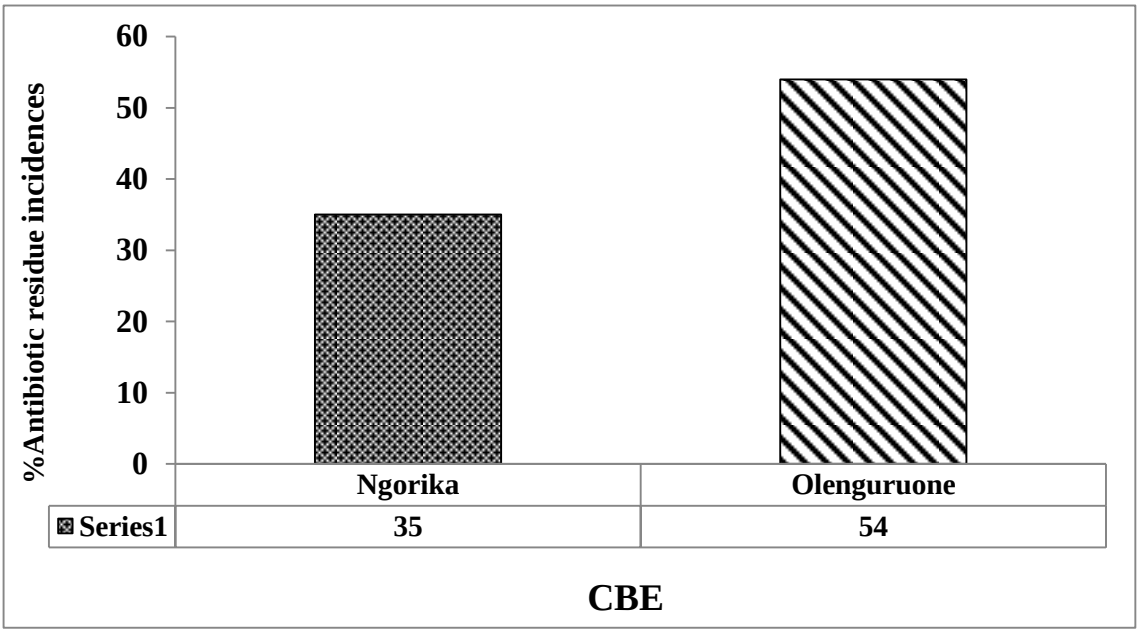


Figure 5: Antibiotic residues detected in percentage for both collection and bulking enterprises

4.3.6 Adulteration

Freezing point depression results indicated that adulteration incidences in Ngorika and Olenguruone were at 23.8% and 36.8%, respectively (Figure 6). Adulteration in Ngorika was lower than in Olenguruone. The farmers in Ngorika were penalized an equivalent amount of the current milk price for a full can if found practicing water adulteration. The penalties could have assisted in reducing this vice. Freezing point of milk is its most constant property. According to the Kenya Standard for raw milk (KS 05-1552), the freezing point of milk is approximately -0.545^0C but not less than -0.525^0C. Added water in milk can be detected by measuring its freezing point. The freezing point is slightly less than that of pure water and relatively constant. Typical milk generally has a freezing point below minus 0.542 degrees Hortvett. When water is added to milk, the freezing point increases approximately 0.005°H for every 1% water addition. Outside a range of -0.525 to -0.565°H should have a cause for investigation.

Adulteration by addition of water to milk may introduce chemical and microbial health hazards as well as reducing the nutritional and processing quality, palatability, and market value of the milk (Muriuki *et al.*, 2003). Added water can occur in milk due to both unintentional (e.g. poor system drainage) and intentional addition (Kurwijila, 2006). Intentional addition could be aggravated by because payment is based on quantity delivered. Added water could be from the roadside ditches with dissolved solids which carries bacterial

load, and the numbers will be a function of the source and cleanliness of the water (Leeuw, 2014). Draaiyer *et al.* (2009) demonstrated that lactometer cannot detect the water adulteration and therefore the freezing point determination confirms the lactometer test as a standard gauge for water adulteration. According to (KS 05-1552), density of milk at 20^0C shall be within a range of 1.026-1.032 g/ml. According to Nirwal *et al.* (2013), adulteration in milk is still in practice and the consumers must be more proactive against milk adulteration. It could alternatively be included in quality based payment system to discourage this vice.

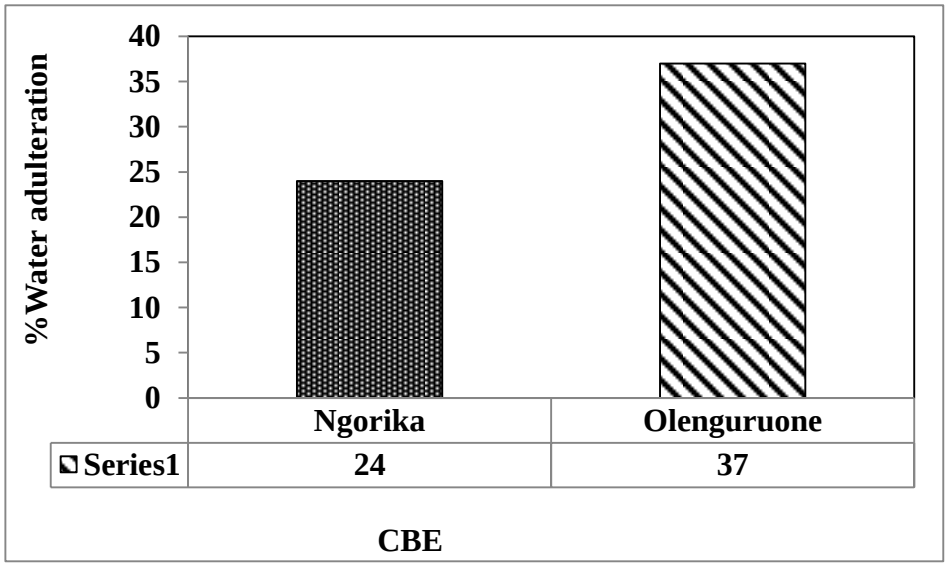


Figure 6: The average percent water adulteration incidences for both collection and bulking enterprises

4.3.7 Compositional analysis results

The analysis of butter fat was done according to AOAC (2000) Rose Gottlieb procedure. The compositional analysis for butter fat and total solids on average did not have difference in comparison with the KEBS standards. Samples that had butterfat content below specification for raw milk were 15.25% and 12.23% for Olenguruone and Ngorika respectively (Table 11). The butter fat content in milk generally influences the total solids in milk. The low total solids observed in Olenguruone could be due to the higher incidences of adulteration observed in this CBE. Table 11, illustrates an average percentage for butter fat content analysis and total solids.

Table 11: The average butter fat content and total solids content of milk for Ngorika and Olenguruone in comparison with KEBS standards

Parameter	Ngorika	Olenguruone	KEBS Standards
Butter Fat	3.6%	3.4%	3.25%
Total Solids	12.11%	11.62%	11.75%

CONCLUSION

1. The study showed that the code of hygiene practice was not observed in the small holder supply value chains.
2. A CCP plan was established for the small holder supply value chains.
3. The raw milk quality of the two CBEs did not conform to the KEBS standards in the total plate count, coliform count and somatic cell count.
4. Antibiotic residues and water adulteration incidences were prevalent.

RECOMMENDATION

1. Processors should assist the suppliers in developing strong extension services together with the ministry officials.
2. To improve the raw milk quality, the CCP plan developed should be adopted and reviewed after a period of time.
3. Rapid tests for antibiotic residues detection applicable at farm levels should be developed.
4. Penalties should be exerted to farmers practicing water adulteration.

REFERENCES

Aboge, G.O., Kang'ethe E.K., Arimi, S.M., Omore, A.O., McDermott, J.J., Kanja, L.W., Macharia, J.K., Nduhiu J.G. and Githua, A., (2000), Antimicrobial agents detected in marketed milk in Kenya. *Paper prepared for oral presentation at the 3rd All Africa Conference on Animal Agriculture*.

Alimentarius, C. (2003). Hazard analysis and critical control point (HACCP) system and guidelines for its application. *Annex to CAC/RCP*, 1-1969.

A.O.A.C. (2000). Rose gottlieb method of milk fat analysis. *Official method 905.02 Fat in milk* (17 th ed.). Gaithersburg, Md.: AOAC International.

A.O.A.C. (2005). Aerobic plate count in foods, dry rehydratable film, method 990.12. In *Official Methods of Analysis of AOAC International* (17th ed.). Gaithersburg, Md.: AOAC International.

Barbano, D.M., Ma, Y., and Santos, M.V. (2006). Influence of raw milk quality on fluid milk shelf life 1, 2. Journal of dairy science, ***89***, E15-E19.

Barry, M. (2005). A brief introduction to microbiology and the use of 3M™ petrifilms.

Bashir, S., Awan, M.S., Khan, S.A., Rathore, H.A., Qureshi, M.A., and Kathu, Z.H. (2013). An evaluation of milk quality in and around Rawalakot Azad Kashmir. *African Journal of Food Science*, ***11***: 421-427.

Bebe, B.O. (2003). *Herd dynamics of smallholder dairy in the Kenya highlands*. Wageningen University. Biotechnology Books.

Chepkoech, T.B. (2010). Ministry of livestock development, Milk quality control and regulation in dairy production: a case of dairy producers in Kikuyu Division, Kabete District, Central Province-Kenya. *Poster presented at the Joint 3rd African Association of Agricultural Economists (AAAE) and 48th Agricultural Economists Association of South Africa (AEASA) Conference, Cape Town, South Africa.*

Corné J.R, Bebe B.O, Jan, L., Catherine K. and Charles T. (2016). Sustainable growth of the Kenyan dairy sector; A quick scan of robustness, reliability and resilience. Report 3R Kenya/WLR 979. http://dx.doi.org/10.18174/391018

De Sousa, G.B., Tamagnini, L.M., Gonzalez, R.D., and Budde, C.E. (2005). Evaluation of

Petrifilm™ method for enumerating aerobic bacteria in Crottin goat's cheese. *Revista Argentina de Microbiologia*, ***37***(4), 214.

DSM. (2011). Delvo test technical bulletin, DSM food specialties B.V., Netherlands. Accessed on 20th February 2016. http://www.agripress.be/_STUDIOEMMA_UPLOADS/downloads/Delvotest.pdf

Doyle, M.M., Garcia, S., Bahati, E., Karamuzi, D., Cullor, J.S. and Nandi, S. (2015). Microbiological analysis of raw milk in Rwanda. *African Journal of Food Science and Technology*, ***6***(5) 141-143.

Draaiyer, J., Brian, D., Antony, B. and Jerome, M. (2009). Milk testing and payment systems. A practical guide to assist milk producer groups. (FAO) *Food and Agricultural Organization. Rome*, Italy.

Dilbaghi, N., and Sharma, S. (2007). Food and Industrial Microbiology: Food spoilage, food infections and intoxications caused by microorganisms and methods for their detection. *Guru Jambheshwar University of Science and Technology, Hisar-Haryana, India*.

EADD, (2008). The Dairy Value Chain in Kenya. A report by Techno Serve Kenya for the East Africa Dairy Development Program August 2008.

Farhan, M., and Salik, S. (2007). Evaluation of bacteriological contamination in raw (un-processed) milk sold in different regions of Lahore (Pakistan). Journal of Agriculture and Social Sciences (Pakistan), ***3***(3): 104-106.

Food and Agriculture Organization (FAO) and International Dairy Federation (IDF) (2004). Dairy Development Newsletter. October 2004 – Issue No. 8

Food and Agriculture Organization and World health Organization FAO/WHO (1998). The application of risk communication to food standards and safety matters (Joint FAO/WHO Expert consultation).

Food and Agriculture Organization (FAO) (1997), Food Safety: Guidebook for the preparation of HACCP plans. Accessed on December 2015 http://www.haccpalliance.org/sub/haccpmodels/guidebook.pdf.

FOSS analytical, (2005). The key to high quality products lies in your raw milk supply. Influence the composition, volume and hygienic quality of your raw milk through payment systems. P/N 1025189, Issue 3 GB, NOV. 2005.

Gachango, F.G., Andersen, L.M., and Pedersen, S.M. (2014). Adoption of milk cooling technology among smallholder dairy farmers in Kenya. *Tropical animal health and production*, ***46***(1), 179-184.

Gacula, J., MC and Singh, J. (1984). *Statistical Methods in Food and Consumer Research*, 2nd edition. Pg. 335.

Gallagher, J. (2015). Antibiotic resistance: world on cusp of "post-antibiotic era". www.bbc.com/news/health-34857015. Accessed on November 20, 2015.

Gemechu, T., Beyene, F., and Eshetu, M. (2015). Physical and chemical quality of raw cow's milk produced and marketed in Shashemene Town, Southern Ethiopia. *ISABB Journal of Food and Agricultural Sciences*, ***5***(2), 7-13.

Gietema, B. (2002). *Modern dairy farming in warm climate zones*. STOAS. Vol. 3 Agromisa Foundation, Wageningen,

Gurmessa, T.D., Mitiku, E. and Alemayehu, R. (2015). Physico-chemical qualities of raw cow milk in Ethiopia. The case of Borana zone, Yabello District. *Global Journal of Dairy Farming and Milk Production.* ***3***(2) 086-091.

Hillerton, J.E., Halley, B.I., Neaves, P., and Rose, M.D. (1999). Detection of antimicrobial substances in individual cow and quarter milk samples using Delvotest microbial inhibitor tests. *Journal of dairy science*, ***82***(4), 704-711.

Irwin and Foreman, (2012). Quality Based Milk Payment Study. Kenya Dairy Sector for SNV KDMP Project, *The Friesian business development company*, Netherlands.
Istanbul, Turkey.

Jain, N. and Shrivastava, S., (2014). Quality Assurance of Marketed Raw Milk in Bhopal city, MP, India. *International Research Journal of Biological Sciences*, ***3***(11), 23-27

Kabui, K.K. (2012). Assessment of milk quality and the potential of a quality based payment system in smallholder farms in Limuru and Eldoret, Kenya. Master of Veterinary Public Health (MVPH) thesis. Nairobi, Kenya: University of Nairobi.

Kahuta, G. (2013). *Milk quality and on-farm factors leading to milk spoilage in Bugaaki Sub County, Kyenjojo District* (Doctoral dissertation, Makerere University).

Kaindi, D.W.M., Schelling, E., Wangoh, J., Imungi, J.K., Farah, Z., and Meile, L. (2011). Microbiological quality of raw camel milk across the Kenyan market chain. *Food 5. Global Science Books,* 79-83.

Kamau, P. (2009). The organic farmer. The magazine for sustainable agriculture in Kenya, Nr.

Karakök, S.G. (2007). Cow milk quality and critical control points on farm conditions. Hayvansal Üretim, ***48***(2), 55-59.

Karuga, S. (2009). Draft report on dairy chain analysis, Timau milk shed. Micro enterprise support program trust.

Kenya bureau of standards (KEBS) (2007). Kenya standards East African Standards (KS EAS). Raw cow milk specification, 1st edition, Arusha. Tanzania.

Kenya Bureau of Standards (KS 05-1552) (2000). Kenya Standard Code of Hygienic Practice for Production, Handling and Distribution of Milk and Milk Products. KS. 67. 020. Nairobi.

Keski, O.Y., and Gulsunoglu, Z. (2012). Quality Management Systems in Dairy Industry. International Conference on Industrial Engineering and Operations Management

Krejcie, R.V. and Morgan, D.W. (1970). Determining sample size for research activities. *Educational and Psychological Measurement, **30***, 607-610.

Kurwijila, L.R. (2002). Evolution of dairy policies for smallholder production and marketing in Tanzania. *Rangnekar D. and Thorpe W.,* Pg. 538.

Kurwijila, L.R. (2006). Hygienic milk handling, processing and marketing. Reference guide for training and certification of small-scale milk traders in East Africa.

Kurwijila, L.R. and Bennett, A. (2011). Dairy development institutions in East Africa: Lessons learned and options. *Rome: Food and Agriculture Organizations of the United Nations*.

Kurwijila, R.L., Hansen, K.K., Macha, I.E., Abdallah, K., and Kadigi, H.J.S. (1992). The bacteriological quality of milk from hand-and machine-milked dairy herds in Morogoro, Tanzania. African Livestock Research (ILCA), ***2***: 59-67.

Ledenbach, L.H., and Marshall, R.T. (2009). Microbiological spoilage of dairy products. In *Compendium of the microbiological spoilage of foods and beverages* (pp. 41-67). Springer New York.

Lee, A.P., Barbano, D.M., and Drake, M.A. (2016). Short communication: The effect of raw milk cooling on sensory perception and shelf life of high-temperature, short-time (HTST)–pasteurized skim milk. Journal of Dairy Science, 99(12), 9659-9667.

Leeuw, B. (2014). Improving food safety by introducing a quality based milk payment system with the help of enabling technologies. The Friesian dairy development company. A feasibility study report.

McCarron, J.L., Keefe, G.P., McKenna, S.L., Dohoo, I.R., and Poole, D.E. (2009). Evaluation of the University of Minnesota Tri-plate and 3M Petrifilms for the isolation of Staphylococcus aureus and Streptococcus species from clinically mastitic milk samples. *Journal of dairy science*, ***92***(10), 5326-5333.

Mellenberger, R., and Roth, C.J. (2000). California Mastitis Test (CMT). *Fact Sheet, Dept. of Animal Sciences, Michigan State University and Dept. of Dairy Science, University of Wisconsin-Madison*.

MoALF. (2013). Sessional paper no. 5 on the national dairy development policy. Towards a Competitive and Sustainable Dairy Industry for Economic Growth in the 21st Century and Beyond. Ministry of Agriculture, Livestock, Fisheries and Cooperatives, State department of livestock, Republic of Kenya.

Muliro, P.S, Shalo, P.L. and Kutima, P.M., (2013). Quality assessment of raw camel milk using dye reduction tests. *African journal of food science and technology, **4***(5). Pp 116-121.

Mullan, W.M.A. (2003). Inhibitors in milk. https://www.dairyscience.info/index.php/inhibitors-in-milk/51-inhibitors-in-milk .html. Accessed: 30 April, 2016.

Muriuki, H. Omore, A., Hooton N., Waithaka, M., Ouma, R., Staal, S.J., and Odhiambo, P. (2003). The policy environment in the Kenya dairy sub-sector: A review. *Smallholder Dairy (Research and Development) Project, Nairobi, Kenya*.

Muriuki, H.G. (2001). Smallholder dairy production and marketing in Kenya. In *Smallholder dairy production and marketing—Opportunities and constraints. Proceedings of a South–South workshop held at NDDB, Anand, India* (pp. 13-16).

Muriuki, H.G. (2003). Assessment of the level, type and value of post-harvest milk losses in Kenya. Results of a rapid appraisal for a national sub-sector assessment for FAO.

Muriuki, H.G. (2011). Dairy development in Kenya. *Food and Agricultural Organization, Rome*.

Murphy, S.C., and. Boor, K.J. (2000). "Sources and causes of high bacteria counts in raw milk: (An abbreviated review." *Dairy, Food and Environmental Sanitation, **20***(8) 1-4

Mwangi, A., Arimi, S.M., Mbugua, S., Kang'ethe, E.K., Omore, A.O., and McDermott, J.J. (2000). Assurance of marketed milk quality in Kenya. Paper presented at the Faculty of

Veterinary Medicine Biennial Scientific Conference, 30-31 August 2000, University of Nairobi, Kenya.

Nirwal, S., Pant, R., and Rai, N. (2013). Analysis of milk quality, adulteration and mastitis in milk samples collected from different regions of Dehradun. *International Journal of PharmTech Research*, **5**(2), 359-364.

Odero, M., (2002). Drug Residues in Animal Products. All Africa News Agency. 38. http://www.poultrytimesofindia.com/issues/2002/nov/index.htm. Accessed, 22 November 2013.

Oliver, J.D. (2005). The viable but non-culturable state in bacteria. *Journal of Microbiology*, **43**(1), 93-100.

O'Mahony, F. (1988). *Rural dairy technology: Experiences in Ethiopia* (No. 4). ILRI (aka ILCA and ILRAD).

Omiti, J. and Muma, M. **(**2000). *Policy and institutional strategies to commercialise the dairy sector in Kenya*. IPAR Occasional Paper No. 006/2000. Nairobi, Institute of Policy Analysis and Research (IPAR).

Omondi, S.P.W. and Meinderts, J. (2009). The status of good dairy farming practices on small scale farms in central highlands, Kenya.

Omore, A., Arimi, S., Kangethe, E., McDermott, J., Staal, S., Ouma, E., Odhiambo, J., Mwangi, A., Aboge, G., Koroti, E., and Koech, R. (2001). Assessing and Managing Milk-borne Health Risks for the Benefit of Consumers in Kenya. MoA/KARI/ILRI/UoN/KEMRI Collaborative Research Report, Smallholder Dairy (Research and Development) Project, Nairobi, Kenya.

Omore, A.O., Muriuki, H., Kenyanjui, M., Owango, M., and Staal, S. (1999). The Kenyan Dairy Sub-Sector: A Rapid Appraisal: Research Report of the MoA/KARI/ILRI Smallholder Dairy (R and D) Project. *International Livestock Research Institute. Nairobi (Kenya). 51pp*.

Orregård, M. (2013).Quality analysis of raw milk along the value chain of the informal milk market in Kiambu County, Kenya.

Pandey, G.S., and Voskuil, G.C.J. (2011). Manual on milk safety, quality and hygiene. *Golden Valley agricultural Research Trust, Zambia*, 52.

Popelka, P.E.T.E.R., Nagy, J.O.Z.E.F., Popelka, P.A.V.E.L., Marcincak, S., Rozanska, H., and Sokol, J.O.Z.E.F. (2004). Comparison of sensitivity of various screening assays and liquid chromatography technique for penicillin residue detection in milk. *Bulletin-Veterinary Institute in Pulawy*, ***48***(3), 273-276.

Quinn, J., Carter, E., Markey, B. and Carter, R. (1994). California mastitis test (CMT). In. *Clinical Veterinary Microbiology*. 1st ed. London: Wolfe publishing. 333-334.

Rademaker, I.F., Koech. R.K., Jansen, A., Van der Lee, J., (2015). Smallholder Dairy Value Chain Interventions; The Kenya Market-led Dairy Programme (KMDP) – Status Report. Wageningen UR (University and Research) Centre for Development Innovation. Report CDI-16-018. Wageningen.

Rajmohan, S. Dodd, C.E.R and Waites, W.M. (2002). Enzymes from isolates of pseudomonas fluorescenes involved in food spoilage. *Journal of applied microbiology*, ***93***(2), 205-213.

Reta, A.M. and Addis, A.H. (2015). Microbiological Quality Assessment of Raw and Pasteurized Milk. *International Journal of Food Science and Microbiology*, ***2***(6), 087-091

Ruegg, P.L., and Reinemann, D.J. (2002). Milk quality and mastitis tests. *Bovine practitioner*, **36**(1), 41-55.

Salman, A.M.A. and Hagar, E.M. (2013). Some Bacterial and Physical Quality of Pasteurized Milk in Khartoum. *Journal of Applied and Industrial Sciences, **1** (2): 30-37, ISSN: 2328-4595 (PRINT), ISSN: 2328-4609.*

Samarzija, D., Zamberlin, S., and Pogacic, T. (2012). Psychrotrophic bacteria and milk and dairy products quality. *Mljekarstvo*, ***62***(2), 77-95.

SAS Institute Inc. (2006). *Base SAS® 9.1.3 Procedures Guide* (2nd ed., Vol. 1, 2, 3 and4). Cary, NC: SAS Institute Inc.

Shitandi, A. (2004). *Risk factors and control strategies for antibiotic residues in milk at farm level in Kenya.* Doctoral dissertation. Vol. 3.

Silvia, B.O., Caraviello, D.Z., Rodrigues, A.C., and Ruegg, P.L. (2005). Evaluation of Petrifilms for the isolation of Staphylococcus aureus from milk samples. *Journal of dairy science*, ***88***(8), 3000-3008.

Souza, L.M.J.D., Costa, A.C., Nero, L.A., Couto, E.P., and Ferreira, M.D.A. (2015). Evaluation of Petrifilm™ system compared with traditional methodology in count of indicators of

sanitary-hygienic quality and pathogenic microorganisms in sheep milk. *Food Science and Technology (Campinas), **35***(2), 375-379.

Swai, E.S. and Schoonman, L. (2011). Microbial quality and associated health risks of raw milk marketed in the Tanga region of Tanzania. *Acian Pacific Journal of Tropical Biomedicine, **1***(3). 217-222.

Teklemariam, G. and Estifanos, H., (2015). Microbial Quality of Raw Cow Milk Collected from Farmers and Dairy Cooperatives of Haramaya District, Ethiopia. *An International Journal of Life Sciences and Chemistry, **32*** (1), 313-322.

Torkar, K.G., and Teger, S.G. (2004). The microbiological quality of some critical control points in the cheese production of individual Slovenian cheese-makers. *Acta agriculturae slovenica*, ***84***(1), 43-61.

Van Kessel, J.S., Karns, J.S., Gorski, L., McCluskey, B.J., and Perdue, M.L. (2004). Prevalence of Salmonellae, Listeria monocytogenes, and fecal coliforms in bulk tank milk on US dairies. *Journal of Dairy Science, **87***(9), 2822-2830.

Wafula, W. N., Matofari, W. J., Nduko, M. J., and Lamuka, P. (2016). Effectiveness of the sanitation regimes used by dairy actors to control microbial contamination of plastic jerry cans' surfaces. *International Journal of Food Contamination, **3***(1), 9.

Wambugu, S., Kirimi, L., and Opiyo, J. (2011). Productivity trends and performance of dairy farming in Kenya. *Tegemeo Institute of Agricultural Policy and Development.*

www.fao.org/agriculture/dairy-gateway/the-dairy-chain Accessed on 12 April 2017.

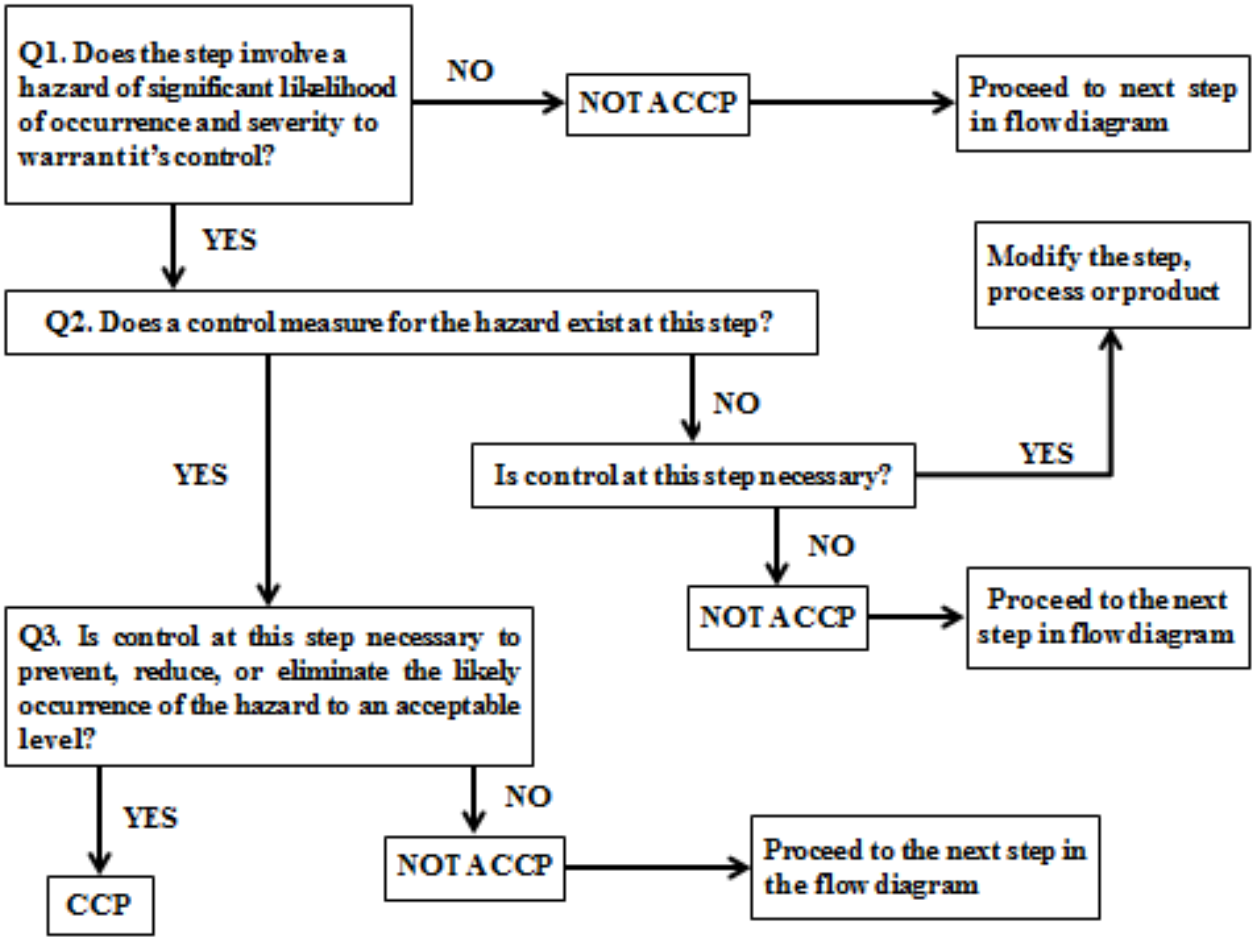
APPENDICES

Appendix 1. Krejcie and Morgan sample table for determination of sample size

Population Size	Sample Size	Population Size	Sample Size	Population Size	Sample Size	Population size	Sample size
10	10	150	108	460	210	2200	327
15	14	160	113	480	214	2400	331
20	19	170	118	500	217	2600	335
25	24	180	123	550	226	2800	338
30	28	190	127	600	234	3000	341
35	32	200	132	650	242	3500	346
40	36	210	136	700	248	4000	351
45	40	220	140	750	254	4500	354
50	44	230	144	800	260	5000	357
55	48	240	148	850	265	6000	361
60	52	250	152	900	269	7000	364
65	56	260	155	950	274	8000	367
70	59	270	159	1000	278	9000	368
75	63	280	162	1100	285	10000	370
80	66	290	165	1200	291	15000	375
85	70	300	169	1300	297	20000	377
90	73	320	175	1400	302	30000	379
95	76	340	181	1500	306	40000	380
100	80	360	186	1600	310	50000	381
110	86	380	191	1700	313	75000	382
120	92	400	196	1800	317	100000	384
130	97	420	201	1900	320		

140	103	440	205	2000	322

Appendix 2. The decision tree



Source: FAO, (1997), Guidebook for the preparation of HACCP plans.

Appendix 3. Dairy farmer practices questionnaire.

Date of interview.....

Route.....

Milk collection center number……

Hygiene information

1. Do you wash your hands before milking? (1)Yes…… (2)No……
   If you wash, what do you use?
   (1)Water alone; (2) Water + soap/disinfectant; (3) other (specify) _____
2. If you wash your hands, do you dry them before milking? (1) Yes….. (2) No……..
   If you dry your hands, what do you use?
   (1)Newsprint; (2) Disposable paper towels;
   (3) Re-usable cloth; (4) other, specify _____
3. Do you wash your cow's udder before milking? (1) Yes….. (2) No……..
   If yes, when do you wash it?
   (1) Cleaned before milking only (2) cleaned after milking only
   (3) Cleaned both before and after milking
4. If you clean the udder, what do you use
   (1) Udder cloth……. (2) Disposable towels……
   If the answer in udder cloth, do you use a separate one for each cow?
   (1) Yes…. (2) No……
5. If you use the udder cloth, how often do you wash it?
   (1) Daily……. (2)Weekly…… (3) Never…….
6. How do you wash the udder cloth?
   (1) With warm water…… (2) With warm boiled water…….
   (3) With cold unboiled water…… (4) With cold boiled water……..
7. Do you use a sanitizer when washing the udder cloth? (1)Yes…. (2)No……
   If yes, what type of sanitizer do you use?
   (1)Hypochlorite…….. (2) Iodophore …… (3) Other (specify)……..
8. Do you use milking cream? (1) Yes…….. (2) No…..……..
9. Do you use teat dipping after milking to prevent mastitis? (1) Yes…… (2) No ……

10. What type of milk container do you use?

(1)Plastic…… (2)Aluminum…… (3)Other……

11. How often do you wash the container?

(1)Before every use (2) After every use (3), before and after every use

12. How do you clean the container?

(1)With cold water alone…… (2)With hot water alone……

(3)With cold water and soap……. (4)With hot water and soap……

(5)With detergent and water……. (6)Others (specify)……

13. What is your source of water?

(1) Piped/ tap……. (2) River/ stream……. (3) Community ground pump…

(4) Roof catchment (rain water)…… (5) Private ground pump/well……

(6) Other (specify)……

14. How do you store the milk containers after cleaning?

(1) On rafts…… (2) Hanging them…….. (3) On the ground……

(4) Other (specify)……

15. Do you cool the milk before sale? (1) Yes…. (2) No……

16. How far is the milk collection center?.....

17. How long does it take you to transport the milk to the collection center?

(1) < 30 minutes…… (2) 30-45 min……. (3) 45- 60 min…… (4) > 1 hour…….

18. How often do you clean/ remove manure from the shed?

1= Daily 3= Monthly

2= Weekly 4= Others (specify)…….

19. Have you ever used antibiotics to treat your animals? (1) Yes…. (2) No……

If yes, are you aware of the antibiotic withdrawal period? (1) Yes…. (2) No……

If yes, did you observe this period? (1) Yes…. (2) No……

20. Are you aware of some of the compositional parameters in milk?

(1) Yes…. (2) No……

If yes, are you aware of how to influence the compositional quality of milk?

(1)Yes……. (2) No…… …….

21. Are you aware of how the quality of your milk compares to others?

(1)Yes…. (2) No……

If yes, is it (1) Above average (2) Average (3) Below average

22. Does your milk get spoilt before delivery?

(1) Yes…. (2) No……

If yes, how many times has it spoilt in the last week?

23. Has your milk been rejected by the cooperative in the last one month? (1)Yes…. (2)No……

If yes, why was it rejected?..... ……

1. Low fat 3. Abnormal colour 5. Failed Alcohol test
2. Low Density 4. Abnormal smell 6. Dirt Others

24. Do you do any milk test before delivering milk to the collection center?

Yes…. No……

If yes, which are these tests?..... ……

1. Alcohol test 3. Density Test
2. Clot on boiling test 4.Other (Specify)……..

Appendix 4. Milk transporter hygienic practices questionnaire.

Name ……

Date of interview.....

Route.....

CBE……

1. Do you handle milk directly in any way?

   Yes ……. No…….

   If yes do you wash your hands?

2. Do you wash your vehicle/ cart/motorbike?

   Yes …….. No……

   If yes how often?

3. Is your cart/vehicle/motorbike having a cover/shade for milk?

   Yes ….. No…..

4. Do you have any training on milk handling?

   Yes ….. No…...

5. How long does it take to collect and deliver milk to the CBE?

6. Have your milk been rejected?

   Yes …… No ……

   If yes, how often and which are these tests?

   1. Alcohol test 3. Density Test

   2. Clot on boiling test 4.Other (Specify)……

7. Do you carry other things when carrying milk?

   Yes …… No ……

   If yes how do you ensure milk quality is maintained?

8. Why do you use plastic containers?

9. Do you sanitize the containers before putting the milk?

Appendix 5. CBE hygienic practices questionnaire.

Date of interview.....

Name/responsibility……

CBE Name……..

1. How often do you clean the cooling tank?

   1. Immediately 2. Just before putting milk

2. How often do you check on the concentration of your detergents?

   1. Daily 2. Weekly 3. Monthly 4. Never

3. What is the source of water?

   1. Rain 2. River

   3. Borehole 4. Others specify

4. Do you experience milk rejects?

   Yes …… No …..

   If yes, how often and which are these tests?

   1. Alcohol test 3. Density Test

   2. Clot on boiling test 4.Other (Specify)……

5. The person managing milk quality is he/she trained in milk handling?

   Yes ….. No…..

   If yes to what level?

   1. Certificate 2. Diploma 3. Degree 4. Others specify

6. How often do you confirm the cooling efficiency of the cooler?

   1. Weekly 2. Monthly 3. When it breaks down

7. What time on average does the first and last transporter bring his/her milk?

   First: Am …….. Last: Am……. Pm……

Appendix 6. Processor hygienic practices questionnaire.

Date of interview

Name/ responsibility

1. When do you clean the tanker?

   1. Immediately 2. Just before putting milk

2. How often do you check on the concentration of your detergents?

   1. Daily 2. Weekly 3. Monthly 4. Never

3. What is the source of water?

   1. Rain 2. River

   3. Borehole 4. Other's specify……..

   How do you ensure its cleanliness and safety?

4. Do you experience milk rejects?

   Yes …... No …….

   If yes, how often and which are these tests?

   1. Alcohol test 3. Density Test

   2. Clot on boiling test 4.Other (Specify)……

5. The person managing milk quality is he/she trained in milk handling?

   Yes …… No …….

   If yes what level

   1. Certificate 2. Diploma 3. Degree 4. Others specify

6. How often do you confirm the efficiency of the tanker?

   1. Weekly 2. Monthly 3. When it breaks down

7. How long does it take you from the CBEs to the factory?

8. Does the quality of milk change from CBE to the factory?

   Yes…….. No……

   If yes, what changes are often and how do you deal with them?

Appendix 7. Research paper 1

Quality control of raw milk in the smallholder collection and bulking enterprises in Nakuru and Nyandarua Counties, Kenya

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Kenya has one of the largest dairy industries in sub-Saharan Africa. Most of the milk marketed by small scale farmers in Kenya has been reported to be of poor quality and does not meet national and international standards due to high bacterial load, high somatic cell count, adulteration and antibiotic residues. This study was designed to assess status of microbiological and physico-chemical quality of raw milk from two small holder dairy farmers at four sampling levels. Three hundred and eight raw milk samples were collected and analyzed along the value chain. Microbiological analysis for total bacterial count and coliform count was carried out using 3MTM Petrifilms plates. The average total bacterial and coliform counts Log10 per ml at the processing factory was 8.462 and 6.770 for Ngorika and Olenguruone, respectively. The antibiotic residues especially β- lactam was prevalent with 44.5% of all the analyzed samples being positive. Likewise, 60% of the samples had a range of 150,000 to 500,000 somatic cells/ml. Average water adulteration level for the two collecting and bulking enterprises was 30.3%. TVBC and CC should be used instead of resazurin while freezing point determination should be used for adulteration.

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Appendix 8: Research paper 2

Hygienic practices and critical control points along the milk collection chains in smallholder collection and bulking enterprises in Nakuru and Nyandarua Counties, Kenya

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Dairy value chains link the actors and the activities involved in delivering milk and milk products from production to the final consumer. In every activity, the product increases in value from production, transportation, processing, packaging and storage. The study was designed to evaluate some hygienic practices along the value chain and develop the quality control system (CCPs) in the smallholder supply chain in Nakuru and Nyandarua County, Kenya. To assess the level using critical control points of compliance to hygienic code of practice, the questionnaires were developed and pre-tested before being administered to the selected individuals in the study. Descriptive statistics was used to depict the implementation of the code of hygienic practices in milk handling by the farmers, transporters, collection and bulking enterprises (CBEs) and the processor. Among the various aspects investigated at farm level in this study was, hand washing before milking, use of reusable udder cloth while milking, use of plastic containers in milk delivery, time taken to deliver milk, cleaning of the cow shed and awareness of the antibiotic resides in milk and its effect. The results indicated poor conformance to the hygienic code of practice along the dairy value chain in the smallholder supply system. The various factors that could contribute to raw milk quality deterioration were identified as, the critical control points (CCPs) using the hazard analysis critical control points (HACCP) principles. Seven factors were identified at five critical points along the milk collection chains. The critical control points identified includes milking at the farm level, bulking milk in a fifty liters can at collection points, transportation, at the CBE platform and the cooling tank. The quality of raw cow's milk produced and marketed from the study areas was low.

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Appendix 9. ANOVA Table

Source	DF	TBC	CC	Lactic acid	RT	BF	Density	Temperature	pH
LOCATION	1	25.801***	63.113***	0.000^{ns}	24.579***	0.891*	0.000^{ns}	57.346^{ns}	28.637^{ns}
Route	13	1.209***	5.537***	0.001***	2.972***	0.900***	0.000^{ns}	111.928***	28.637*
Sample from	3	4.085***	9.010***	0.000^{ns}	2.069^{ns}	0.084^{ns}	0.004***	51.695^{ns}	28.637**
Error	141	0.349	1.269	0.000	0.988	0.147	0.000	28.637	0.041
R-Square		0.401	0.345	0.306	0.226	0.282	0.146	0.206	0.227
Coefficient of variation		7.237	17.594	7.089	23.337	10.856	1.971	48.764	3.067