# \\ PREVALENCE OF DAIRY GOAT MASTITIS IN CENTRAL KENYA HIGHLANDS //

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A THESIS SUBMITTED TO THE DEPARTMENT OF CLINICAL STUDIES IN PARTIAL FULFILLMENT FOR THE DEGREE OF MASTER OF SCIENCE IN CLINICAL STUDIES.

### **DECLARATION**

I DECLARE THAT THIS IS MY ORIGINAL WORK AND HAS NOT BEEN PRESENTED IN ANY OTHER UNIVERSITY FOR EXAMINATION

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#### LIST OF ABBREVIATIONS

KARI Kenya Agricultural Research Institute

ODA Overseas Development Authority.

MALDM Ministry of Agriculture, Livestock Development and Marketing

FAO Food and Agriculture Organisation.

ISLP Integrated Small Livestock Project.

Ksh. Kenya shillings

CMT California Mastitis Test

IgG Immunoglobulin G

IgA Immunoglobulin A

IgM Immunoglobulin M

Ml Millilitre

Nagase N-acetyl-β-D-glucosaminidase

I.U. International units

KG Kilogramme

Mg Milligramme

Mm Millimitre

Ha Hectare

CMT Milk indicator test

μg Microgramme

AV PROD Average production

F1 First generation offspring

F2 Second generation offspring

DLC Direct leucocyte counts

DLC1 Direct leucocyte count (1st Sampling right half)

DLC2 Direct leucocyte count (1st Sampling left half)

DLCB1 Direct leucocyte count (2<sup>nd</sup> Sampling right half)

DLCB2 Direct leucocyte count (2<sup>nd</sup> Sampling left half)

DLCC1 Direct leucocyte count (3<sup>rd</sup> Sampling right half)

DLCC2 Direct leucocyte count (3<sup>rd</sup> Sampling left half)

CPS Coagulase positive Staphylococcus.

CNS Coagulase negative Staphylococcus.

Gok Government of Kenya

Gtz German Technical Cooperation

#### ABSTRACT

One hundred and thirty does in seven dairy goat farmer groups in Nyeri district (Central Province) were examined to determine the prevalence and aetiology of mastitis and risk factors associated with it. The goats, which were registered with the Kenya studbook, comprised of a mixed population of Saanen, Toggenburg, Small East African and German Alpine crosses.

The selected does, most (91%), of which were under zerograzing system were sampled over a three-month period. During each visit breed, kidding dates, stage of lactation, parity, litter size, milk yield and purpose of milk were recorded. A total of 260 samples were obtained from 130 lactating does and tested for mastitis using bacteriological and cytological techniques (indirect and direct somatic cell count).

The overall prevalence of infected glands was 39% while that of infected does was 58%. The prevalence of subclinical mastitis in the various dairy goat farmer groups ranged from 22% to 76% with groups in Mathira division having significantly (p<0.05) higher prevalence rates than the other divisions.

Various bacterial isolates were found with the most frequent ones being *Staphylococcus* spp (78%) and *Micrococcus* spp (16%). Of the *Staphylococcus* spp isolated, 71% were the coagulase negative while 29% were coagulase positive.

Poor milking hygiene i.e.not washing the udder before milking, not applying milking salve before milking and not dipping teats after milking, significantly (p<0.05) influenced the prevalence of mastitis. Similarly, does that were housed in temporary earthen floor enclosures had significantly higher (p<0.02) mastitis prevalence rates than those on raised slatted houses. These findings indicate that particular attention should be given to the design of dairy goat housing and milking hygiene which were found to be significantly (p<0.02) associated with the level of subclinical mastitis in the farms. It is suggested that preventive measures, including improvement of housing structures and application of good milking hygiene, be instituted before the disease takes root in the dairy goat flocks.

In this study, when direct leucocyte count was used as the true indicator of infection,
California mastitis test score of 3 was found to be highly sensitive and specific in detecting
mastitis in does. Bacteriology, however, was found to be highly sensitive (>75%) but mildly
specific (50%). Thus the mere isolation of microorganisms from milk samples may not be a
true indicator of infection. Due to its high sensitivity and specificity, direct leucocyte count
is the best test to be used in confirming mastitis diagnosis but it is not practical under field
conditions. As an alternative a CMT score of 3 may be used as confirmatory of mastitis
during surveillance of the disease in dairy goat flocks.

#### CHAPTER 1

#### 1.0 INTRODUCTION

Kenya has an estimated 11.5 million goats which are mainly in the southern and northern rangelands in mixed herds with sheep, cattle and camels (Kenya Agricultural Research Institute/Overseas Development Authority manual, 1996). The highest number of goats is found in the Rift valley, followed by Eastern, Coast, North Eastern, Nyanza, Central, Western and Nairobi province. The distribution of milk goats, however, differs from that of the total number of goats as the highest number is found in Central province, followed by Rift Valley, Eastern, Coast, Nairobi and lastly Nyanza province (Ministry of Agriculture Livestock Development and Marketing Annual Report 1995). In central Kenya, the dairy goat population is highest in Nyeri district with 84,800 followed by Muranga with 75,700 goats (KARI/ODA I, 1996).

The production objectives of goat farmers depend on individual community's needs, environment, availability of other resources including cattle, sheep and camels. Irrespective of the production objective, goat milk is especially important where or when cow milk production is low due to either the dry season or due to scarcity of land and fodder (KARI/ODA, 1996). Average milk offtake from the indigenous goats and the Kenyan Toggenberg is put at 50kg and 521 kg respectively for 240 days lactation period. In 1993 the estimated value of milk produced by dairy goats in both the highlands and range lands was Ksh.1, 480,640 million which represented 4.4% of the total milk sale value in the same year (KARI/ODA report, 1996).

In the highlands, the high human population density, rapid population growth and the land tenure system has led to extensive land fragmentation which has reduced the on farm animal feed resource base to household waste, crop residues, weeds and shrubs. In addition, it has been shown that the goat has a more efficient digestive system that converts this type of poor quality feed to high value animal protein (milk and meat) than sheep or cattle (Holmes-Pegler, 1964; Semenye and Hutchroft, 1992). It is also apparent that keeping 2-3 dairy goats can increase the productivity of the smallholder farmers and provide a small but all year round supplement of milk protein in the farmer's diet (Kinuthia, 1997).

With these production advantages, the Kenya and German Governments embarked on a project to upgrade local goats through cross breeding under the Integrated Small Livestock Programme (ISLP). This project which targets smallscale farmers in the central highlands involves crossing of the local goat breeds with the purebred German alpine.

The upgrading of the local goat has led to a change of management practice, including the adoption of more intensive methods of goat rearing. These changes would inevitably be expected to lead to a greater risk of disease including mastitis (Manser, 1986). Mastitis in goats, as a management disease has not been given a lot of attention in Kenya probably because, the dairy goat industry until recently did not receive adequate attention. With the current attention on this industry and the realization of the social and economic benefits of the dairy goat, this potentially important disease (i.e. mastitis) needs to be studied to avoid the losses that are associated with the disease in cattle (Jenzen, 1970; Njenga, Munyua, Karioki, Gachuiri, Wahome and Kiiniya, 1987).

Prevention of mastitis is more critical in dairy goats than in dairy cows due to the fact that milk from goats is often used or consumed without pasteurization by infants, recovering

patients and chronically ill people (Samuel, 1977; Blood, Radostits and Hederson, 1989). The latter is especially important in that several of the organisms involved in goats mastitis including Staphylococcus spp, *Coxiella burnetti*, *Toxoplasma gondii* and Brucella spp are zoonotic and may therefore cause diseases in susceptible owners or consumers (Samuel, 1977; Lyord, 1982).

The economic importance of mastitis which includes losses in milk resulting from reduced or no milk production, discarded milk, loss of udder function, cost of drugs, veterinary services and labor is well understood and documented in the cow (Jenzen, 1970; Kirk, Huffman and Anderson, 1980; Njenga *et al*, 1987). In a setting where goats are reared for milk production, similar factors will be considered and may be of great significance (Gross, Pollack, Anderson and Torell, 1978). In addition, losses may also be calculated in terms of poor growth rate and weight gain in kids (Oguchukwu, 1983). Peracute cases may lead to death of a doe while subclinical mastitis can reduce milk production by as much as 25% (Samuel, 1977; Oguchukwu, 1983).

In Kenya, a preliminary study by Maina, Munyua, Mutiga and Thaiya (1993) examined 13,950 Small East African and Gala goats in 14 districts of Kenya and concluded that mastitis might be a disease of considerable importance in the goat. These authors recommended a comprehensive survey of the disease.

# 1.1 Objectives

The objectives of this study were:

1. To determine the prevalence and etiology of mastitis in selected dairy goat flocks in Kenya's Central highlands (Nyeri District) and the risk factors associated with it.

2.	To compare the prevalence of mastitis between the milked dairy goats and the non-
	milked dairy goats.

3. To determine the possible economic importance of mastitis.

#### **CHAPTER 2**

### 2.0 LITERATURE REVIEW

#### 2.1 Anatomy of goat's udder

The goat udder consists of two long conical glands, each with a single large teat that ideally should be directed forward with each gland being referred to as a half or a side. Six to nine ducts join to form the teat cistern, with clear separation between the gland and the teat. At the end of the large teat cistern of each gland is a single streak canal (Mary and Michael, 1977).

### 2.2 Anomalies of goat udder

#### 2.2.1 Congenital anomalies

Supernumerary teats, which are common in does, may be found either cranial or caudal to the normal teat at the base. These should either be removed surgically or cauterized when the goats are young (Mary and Michael, 1977).

Double teat openings, which are congenital, are occasionally seen in milking does and should be closed or cauterized as early as possible as they result in a lot of milk wastage during milking (Mary and Michael, 1977).

Asymmetrical glands and blind teats are often encountered in milking does, and while they may be congenital, in most cases they are acquired or as a result of chronic mastitis. Teat

sphincter may also be too tight to allow normal milking. This may occur as a result of a congenital defect or damage to immature teat by abrasion or suckling. Removal of some tissue from the streak canal with 20 gauge 1 inch needle may correct this defect (Mary and Michael, 1977).

Long pendulous udders, which occur in some does due to the loosening of suspensory ligaments are often inherited and occur in a particular lineage.

Precocious udders have been seen in doelings that have not been bred or suckled especially those borne from high producing dams. However, the production in these unbred doelings is low and they should be observed for any complications until lactation ceases (Mary and Michael, 1977).

#### 2.2.2 Acquired conditions affecting the udder skin

Epizootic contagious ecthyma results in papulo-reticular lesions on the udder and the teats of affected does. Nursing does may get infected from the kids or from the environment through abraded udder lesions. Although does are known to recover within 3-4 weeks from contagious ecythema, vaccination in affected flocks and application of udder ointments have been shown to have some beneficial effects. Infected does should be milked last and the milker should wear gloves, to prevent human infection or spread to other goats. Chlorhexidine teat dips are considered effective in controlling spread and failure to treat these lesions may lead to mastitis (Mary and Michael, 1977; Blood *et al.*, 1989).

Caprine papillomatoses, though rare, may affect goat udders and is best treated by surgical removal of the papillomas if they are few. If, however, they are many it is advisable that the

doe be culled. Other viral conditions that may affect goat udder are goat pox and foot and mouth disease (Samuel, 1977).

Udder impetigo is a staphylococcal infection on skin and hair follicle of the udder leading to purulent necrotic lesions characterized by pinhead to pea-sized pustules especially at the base of teats, which may spread to other parts of the udder. The affected doe should be milked last to avoid spread of the disease while a topical antibiotic cream is being applied.

Furunculosis is a similar but more severe form of infection, characterized by hot reddened swellings on the tip of the teats caudally, which may rupture to release a foul smelling fluid. These lesions are treated by lancing and flushing with a disinfectant (Mary and Michael, 1977).

Chorioptic and sarcoptic mange may affect the udder and also other parts of the body causing constant rubbing of the udder on hard objects resulting in bruises, which predisposes the doe to mastitis. Skin scrapings of affected animals reveal mites which can be treated by applying acaricides topically and sulphur ointments for secondary bacterial infection (Samuel, 1977).

Biting flies cause erythema, edema and small crusts of dried blood on the udder and teat surface. In most cases housing the animals and application of fly repellents alleviates the problem (Mary and Michael, 1977; Samuel, 1977).

Other conditions occasionally seen in does include sun-burn, especially those with hairless distended udders are turned to pasture suddenly. Photosensitization either primary or secondary to liver disease may also cause reddening or necrosis of udder skin. Treatment

usually includes housing the goats, removal of photosensitizing plants from diets and supportive therapy in case of liver disease (Blood *et al.*, 1989).

#### 2.2.3 Udder oedema

Marked udder oedema, which in most cases is physiological, occasionally occurs in high producing does shortly before or after parturition. Limiting the use of sodium, potassium, as well as high-energy foodstuffs such as cornmeal in the dry period is helpful. Hot fomentation, frequent stripping and diuretics given once or twice daily for upto 3 days are effective in treating the condition (Lyord, 1982).

#### 2.2.4 'Hard udders'

This is an uncommon condition in which the udder is covered with a loose skin, is extremely hard and produces very little milk. It usually occurs in does first kidding and continues into subsequent kiddings. The etiology is unknown but is thought to be due to either nutrition, allergy, genetic and/or mycoplasmal mastitis (Mary and Michael, 1977). Various combinations of treatment including tetracyclines, diuretics, oxytocin and prostangladins have been tried in the management of the condition with little or no success. In most cases the condition usually regresses spontaneously within two weeks but if this does not occur, culling of the doe should be considered (Lyord, 1982).

#### 2.2.5 Cysts

Cysts of the udder in goats have been reported in India and Britain. They are of variable sizes and contain either milk, serous or cloudy fluid. These cysts either resolve spontaneously or can be broken down by massage (Lyord, 1982).

#### 2.2.6 Udder wounds

Udder wounds result from barbed wires, goring horns, dogbites or bites from suckling kids. They are especially common in goats on pasture with barbed wire fences. Dog bites frequently cause deep jagged wounds on tethered goats. Any udder or teat wound should be examined fully to determine the extent of damage and a decision made on form of treatment. The wound can either be treated as an open wound or, in case of large lacerations involving the full skin be sutured and the doe given 500-1500 I.U. antitetanus toxoid (Mary and Michael, 1977; Blood *et al.*, 1989).

Udder contusions with or without break of the skin may occur in goats with highly pendulous udders especially if made to jump over obstacles and this may lead to excretion of bloody milk (Mary and Michael, 1977). Bloody milk, however, is not always indicative of pathological condition as it may be observed in freshening does. However, this disappears spontaneously within five days (Lyord, 1982).

# 2.3 Qualities of goat milk

Goat milk is sweet, nourishing and may have some medicinal value, hence it is beneficial for consumption. It is not so apt to curdle as the cows milk due to its lower acidity and higher

buffering capacity, which makes it very suitable for human consumption. Nutritionally it is superior to cows milk as it has high phosphate content, smaller fat globules, proteins in a more finely divided state and less coagulum (Samuel, 1977; Lyord, 1982; Devendra and Burns, 1983; Wilson, 1991).

Goat milk may sometimes have an 'off flavor' due to either mastitis, bacterial contamination during or after milking, ketosis or shrubs and twigs smell in the feed (Holmes -Pegler, 1964; Lyord, 1982).

In some countries such as Ireland, Scotland, Middle East and Near East, special value is attached to goat milk as it is converted to butter and cheese. Due to its high phosphate content goat milk is very useful to vegetarians (Mackenzie, 1970; Lyord, 1982).

### 2.4 Mastitis in goats

#### 2.4.1 Definitions

Mastitis is broadly defined as inflammation of the mammary gland that alters its structure and function regardless of the cause. The disease is characterized by physical, chemical and cultural changes in milk and pathological changes in the udder. Injury to any part of the mammary tissue may induce an inflammatory response or mastitis, however major disease of the udder is that which is associated with microbial infection (Jain, 1979; Shearer, 1992).

The clinical signs of mastitis are similar in all animals and depend on the severity of the condition (Winkler and Roberts, 1986). The signs are in reality an expression of the host defense intended to destroy the invading organisms and make way for return to normal

function In affected animals mastitis is a complex condition due to the great variation in its causes, pathogenesis, intensity, duration, residual effects, immunity, therapy and eradication. The internal environment of the normal mammary gland is ideally sterile, although non-pathogenic bacteria are frequently isolated (Jain, 1979).

Mastitis starts with entry of pathogenic bacteria via streak canal into the interior of the gland and if the environment is favorable for survival then these bacteria multiply. The resulting byproducts of bacterial growth and metabolism irritate the delicate mammary tissue and induce an inflammatory reaction (Jain, 1979; Lyord, 1982; Shearer, 1992).

The severity of mastitis primarily is determined by the nature of the invading pathogen and the natural mechanisms of resistance available to the animal at that moment, the current stress placed on the mammary gland, milking practices, integrity of the teat sphincter, trauma and the environment (Jain, 1979). In general it is characterized by swelling, heat, pain or sensitivity and congestion of tissues (Samuel, 1977; Winkler and Robert, 1986).

#### 2.4.2 Forms of mastitis

Mastitis can be classified as either clinical or subclinical depending on whether there are obvious clinical signs or not. Clinical mastitis exhibits the characteristic features of inflammation seen as swelling, heat, redness, pain and disturbed function. This is further classified as peracute mastitis which apart from exhibiting all the signs of inflammation, also show systemic signs of fever, depression, anorexia and shivering. Acute mastitis on the other hand is milder than peracute and is characterized by fever and mild depression along with other signs of inflammation. In subacute or subclinical mastitis, the cardinal signs of inflammation are less pronounced and systemic signs are absent. Chronic mastitis is observed

when the inflammatory process persists for months. The udder may remain subclinically infected indefinitely, sometimes resulting in frequent eruptions of acute disease (Winkler and Roberts, 1986; Blood *et al.*, 1989). Existence of a pathogen within the mammary gland without any evidence of mastitis is referred to as latent infection and such glands seem to be very sensitive to trauma and infection (Jain, 1979).

Clinical mastitis is characterized by very low prevalence in most flocks. A survey of clinical mastitis in 900 goats selected from seven Government Nigerian goat flocks revealed a prevalence rate of 10% (Ameh, Addo, Adekeye and Gyang, 1993), while Kalra, Sharma and Dhuda, (1962) reported a prevalence of 9.4% in 400 Indian dairy goats.

Subclinical mastitis however has a higher prevalence and contributes to most losses in milk production. In Britain a survey of some 400 goats revealed a prevalence rate of 36% (Manser, 1986). Contreras, Correlas, Sierra and Marco, (1995) found a lower prevalence in Spain of 18%, while Boscos, Stefanakis, Alexopoulus and Samartzi, (1996) found a prevalence range of between 19-36% in Greek goats.

# 2.4.3 Predisposing factors to mastitis

# 2.4.3.1 Anatomy of the goat udder

The location of the udder at the inguinal canal is in itself a predisposing factor since it easily gets contaminated and can be traumatized when the animal lies down. Large pendulous udders with weak support and poor teat conformation are also highly predisposed to trauma of the udder and the teats. Long teats in particular have higher chances of contamination and infection while very short teats, require a lot of pulling during milking, predisposing the doe to

teat canal injury and infection (Jain, 1979; Winkler and Robert, 1986). Wear and tear associated with an increased lactational age reduces tightness of teat sphincter through fibrosis and thereby exposing the teat canal to increased infections (Jain, 1979).

# 2.4.3.2 Environment

Occurrence of mastitis is enhanced by the management factors that favor spread of the infective agents among the susceptible animals and, weaken the natural resistance of teat orifice to bacterial invasion (Jain, 1979). The immediate environment of the dairy animal is rich in mammary pathogens and poses a great threat to udder health. Cold weather leads to crowding of animals together which may lead to cross infection, while wet conditions enhance growth and transport of microorganisms from external surfaces. Animal beddings and especially saw dust and straw are a rich source of microorganisms, especially coliforms. The barns and the pens should therefore be cleaned regularly and preferably emptied on daily basis (Samuel, 1977).

The equipment used during milking including teat cups, buckets and towels may serve as a medium of infection from one animal to another. Milkers' hands especially if not cleaned between milking may pass infection from infected does to non-infected does (Samuel, 1977; Jain, 1979).

# 2.4.3.3 Trauma and infection

Udder and teat injury is the most important single stressor which lowers resistance to mastitis (Jain, 1979). Goats especially those on pasture easily get injured while jumping over fences or

climbing on walls and shrubs (Winkler and Robert, 1986). Wounds resulting from bites and poor milking techniques expose the animals to udder infection if not attended to in time. This is risky since the incidence of intrammamary infection has been shown to increase with the number of organisms on teat skin (Jain, 1977; Samuel, 1977; Blood *et al.*, 1989).

# 2.4.4 Aetiology of mastitis in does

Several species of bacteria and other organisms including fungi and viruses have been associated with mastitis in goats (Samuel, 1977; Blood *et al.*, 1989; Shearer, 1992). However individual animals vary in susceptibility, extent, types and duration of infection. Although some mammary pathogens can be found in the environment of the animals, the most important reservoir of the organism is the animal itself i.e. the infected doe (Jain, 1979). A characteristic common to all these organisms is the ability to colonize the streak canal through which they gain entry to the gland (Shearer, 1992).

Staphylococcus aureus has been found to be the most important mammary gland pathogen associated with mastitis in dairy goats in most countries (Lyord, 1982; Manser, 1986; Maina et al., 1993; Ameh et al., 1993; Contreras et al., 1995). In particular Lyord, (1982) found hemolytic Staphylococcus aureus as the most common organism in goat mastitis in Britain followed by non-hemolytic Staphylococcus spp and a variety of Streptococcus spp.

In Spain, 71% of isolates consisted of Staphylococcal spp, 12% Actinomyces pyogenes, 3% Coliforms spp, 9% Mycoplasmas spp, 2%, Pasteurella spp, and Streptococcus spp, 1% (Contreras et al., 1994). In Nigerian goat flocks, Ameh et al., (1993) found that 35% of clinical mastitis resulted from Staphylococcus spp. 19% from coliforms and 8% from Streptococcus spp. Staphylococcus spp were also found to be the most common organisms associated with mastitis in Kenyan flocks (Maina et al., 1993). Other organisms isolated include Bacillus spp, Acinetobactor spp, Proteus spp, and Micrococcus spp (Ameh et al., 1994, Maina et al., 1993; Egwu, Zaria, Onyeyili, Ambali, Adamu and Birdling, 1994; Munyua, 1998).

## 2.4.4.1. Staphylococcal mastitis

Staphylococcal mastitis occurs in almost all domestic species including cattle, sheep, pigs and goats and it is the most important udder pathogen (Blood *et al.*, 1989, Shearer, 1992). Coagulase positive staphylococcus (*S.aureus*) has been incriminated as the most common isolate in clinical mastitis in dairy goats (Lyord, 1982; Ryan and Greenwood, 1990; Dasgupta Chanda, Chowdhury, and Bhuin, 1993; Deinhofer and Pernthanei, 1995; Egwu *et al.*, 1993; Contreras *et al.*, 1995). The principal reservoirs of this organism are the udder, teat skin and milk of infected glands. The infection is spread rapidly during milking from one animal to another (Jain, 1979; Blood *et al.*, 1989). This organism has the capacity to penetrate tissues, producing deep seated foci hence reducing the efficacy of intramammary infusions in their eradication (Jain, 1979).

This species generally produces subclinical and chronic mastitis but may also cause peracute mastitis which may lead to gangrene of the udder (Jain, 1979, Blood *et al.*, 1989). Acute gangrenous mastitis has a sudden onset and a short course. It is characterized by severe depression, swelling, pain in affected gland and yellow clotted secretion. In goats gangrenous mastitis is characterized by sudden onset, presence of watery, dark red secretion. This may be accompanied by gas bubbles especially when there is secondary infection with gas forming organisms such as *Clostridium* spp. The udder appears, hyperemic edematous with progressive discoloration of the distal parts. Beta and K- toxins produced by these Staphylococcus organisms are thought to be involved in the pathogenesis of this form of mastitis (Jain, 1979; Mukhtart, Abu-Samra, Esanousi, Abdalla, Gameel, Abdel Aziz, Abbaj, Ibrahim and Indris, 1988). Death may result immediately or after several days, and in some cases, recovery has been seen with final sloughing of the necrotic tissue (Shearer, 1992).

Coagulase negative Staphylococcus causes insidious progressive mastitis in dairy goats and is of major importance in some flocks. These are almost always isolated from the milk samples of subclinically affected goats. Early cases may only show slight thickness and tenderness at the base of the gland near its attachment to the body wall. The milk usually shows no gross abnormality but direct leukocyte count or the California Mastitis Test (CMT) reveals abnormally high cell content. Due to the difficulty in detection and the insidious course, this form of mastitis is often recognized when the affected gland is already destroyed (Samuel, 1977; Jain, 1979; Blood *et al.*, 1989). Periodic bacteriological examination, segregation and treatment of infected animals could reduce incidence of staphylococcal mastitis (Jain, 1979).

# 2.4.4.2. Streptococcal mastitis

The most important *Streptococcus* spp in the dairy cattle industry are *S. agalactiae*, *S. agalactiae* and *S. uberis* with the former being more prevalent. These three species are also occasionally isolated from infected goat udders (Shearer, 1992).

Streptococcus agalactiae is very important as a causative organism of chronic mastitis in dairy cows. Since it is an obligate parasite of the udder the infection is aggravated by incomplete milking. The incidence of the disease in infected herds has been observed to increase with lactational age of the animals. Appropriate antibiotic therapy and management easily eradicates this species in dairy cattle (Jain, 1979). In dairy, goats *S. agalactiae* infections often result in chronic mastitis but it is neither widespread, common nor as easily spread as in dairy cows (Samuel, 1977; Shearer, 1992).

Streptococcus uberis and S. dysagalactiae are commonly encountered in bacteriological culture of goat milk but they are rarely involved in clinical cases (Samuel, 1977). These organisms are not obligate udder pathogens but they can survive for long periods in the environment of the animal. The infections caused by these organisms are non contagious and commonly non-clinical (Jain, 1979).

Clinical cases of streptococcal mastitis are characterized by swelling and tenderness of udder. The udder secretion is watery and contains small white flakes. During the early stage of an acute attack, the animal has a body temperature of 104-105°F. It is common to find fibrous udders after repeated attacks by *Streptococcus* spp (Samuel, 1977).

Some researchers have found very low rates (1-2%) of intramammary infections due to Streptococcus spp in dairy goats (Hunter, 1984; Contreras *et al.*, 1995) while others have just mentioned them as minor isolates in goat mastitis (Ameh *et al.*, 1993; Maina *et al.*, 1993; Egwu *et al.*, 1994; Munyua, 1998).

# 2.4.4.3 Coliform mastitis

Coliform mastitis is generally referred to as an environmental disease due to the fact that the causative organisms are mainly transferred from the environment to the animals through the bedding and manure (Jain, 1979, Blood et al., 1989). Teat end injury and routine dry period udder treatment tends to increase the incidence of coliform mastitis (Samuel, 1977; Jain, 1979; Lyord, 1982). However, this form of mastitis is relatively uncommon in all species probably due to the relatively high susceptibility of most coliforms to the humoral and cellular factors in milk (Jain, 1979). In goats, this form of mastitis is caused by *Escherichia coli, Klebsiella* spp, or *Enterobacter aerogenes* and occurs mainly in early lactation. These organisms have been isolated from goat milk samples in. Kenya, (Maina et al., 1993) Nigeria, (Egwu et al., 1994; Ameh et al., 1993), Britain (Lyord, 1982), and Spain (Contrera et al., 1995). The organisms are known to produce endotoxins which may lead to death of the animal inspite of treatment (Jain, 1979; Shearer, 1992).

Klebsiella spp causes an extremely severe reaction in the udder which is accompanied by a very high body temperature. Only small amount of very clear yellow fluid may be removable from the udder. Very few animals survive the infection, and those which survive do not show any evidence of the attack in the mammary tissue or in production potential (Samuel, 1977; Blood *et al.*, 1989).

Escherichia coli is found in the lower intestinal tract of most domestic animals and humans. Mastitis caused by this organism commonly occurs when teat ends are injured and the animals are kept in filthy wet bedding, walk through mud or using water contaminated with manure as udder wash (Samuel, 1977). Acute E. coli cases are characterized by severe depression, cold swelling of the affected gland and small amounts of bloody secretion from the udder. Lowered blood calcium with 'milk fever like symptoms' are characteristic of this condition. Prognosis is poor and surgical removal of the gland is recommended in most cases (Samuel, 1977).

#### 2.4.4.4. Other bacterial causes of mastitis

Actinomyces pyogenes and A. ovis are not rare in dairy goats. A. pyogenes in particular, commonly causes thickened milk secretion and development of multiple abscesses within the udder while A. ovis usually produce hard encapsulated abscesses of the supramamary lymph glands, reduction in milk volume, and watery milk (Samuel, 1977). Affected animals should be culled from the herd as they respond poorly to treatment (Samuel, 1977). Researchers working on mastitis in dairy goats have isolated Actinomyces spp in mammary glands of does with or without clinical mastitis (Ameh et al., 1993; Maina et al., 1993; Egwu et al., 1994; Contreras et al., 1995; Munyua, 1998). The reported prevalence in healthy udders is however low compared to affected udders (Ameh et al., 1993; Egwu et al., 1994).

Proteus spp and Pseudomonas spp have been isolated in mastitic milk samples from goats. It is, however, rare and occurs as sporadic cases after intramammary contamination with materials containing these organisms. They are common in the environment of cattle and they

cause mastitis which result in high mortality rates in all animals (Mary and Michael, 1977; Blood et al., 1989; Ameh et al., 1993; Dasgupta et al., 1993).

Pasteurella spp were isolated in 5% and 3% of the milk samples from goats with mastitis in Nigeria and Kenya respectively (Ameh et al., 1993; Maina et al., 1993). These organisms have also been found in mammary glands of non-clinically affected goats in Spain (Contreras et al., 1995).

Pasteurella spp commonly causes mastitis in ewes and a comparatively rare peracute gangrenous mastitis in goats (Blood et al., 1989). It is usually caused by P. hemolytica and infection is thought to occur through injuries to teats caused by over vigorous sucking by big lambs, or kids. The cause of mastitis due to this bacterium is usually systemic with involvement of the udder (Blood et al., 1989). Other minor bacterial isolates include Listeria monocytoges which may cause mastitis in rare cases (Bourry, Cochrad, and Poutrel 1997).

# 2.4.4.5 Mycoplasmal mastitis

Mycoplasma spp has been isolated in cases of goat clinical mastitis by several researchers (Blikslager and Anderson, 1992; Contreras et al., 1995; Upadhaya, Rao, Misra, and Kar, 1992). The condition in affected animals is characterized by sudden onset, involvement of all quarters, severe swelling of the udder, a decrease in production and a gross abnormality of milk (Blood et al., 1989).

# 2.4.4.6 Fungal mastitis

*Cryptococcus neoformans* which is a yeast causes mastitis in cattle, buffaloes and experimentally in goats. *Cryptococcus spp* inoculation into the glands of 10 goats resulted in mastitis in all of them but there was no spread to the un-inoculated udder half

(Sigh, Gupta, Rana and Jand, 1994). Mastitis may be acute with severe swelling of the gland and the supramammary lymphnodes, severe fall in milk yield and appearance of viscid mucoid gray white secretion. It is of public health importance especially where milk is taken without pasteurization since it causes human cryptococcoses (Blood et al., 1989). Other fungal organisms isolated from goat mastitis samples include Aspergillus niger and Candida albicans (Upandhya et al., 1992).

# 2.5 Defence mechanism of the mammary gland.

The immune system of mammary gland supplies both specific and non-specific defenses to the newborn and to itself. Non specific defense mechanisms relate to the anatomical structure of the gland, integrity of teat canal, humoral factors, phagocytic cells and food supplements while specific mechanisms relate to the release of species specific immunoglobulins (Semiento and Soler, 1995).

#### 2.5.1 Physical defences

#### 2.5.1.1 Teat canal

The teat canal is a narrow passage in the teat, being narrower at the tip and wider at the base and this limits the entry of microorganisms. Other features include the lining of the teat canal, which consists of stratified squamous epithelia similar to the skin of the teat. This epithelium continuously undergoes keratinization to form sebum like material, which covers the lining of the canal. This material has long chain fatty acids that have a bacteriostatic effect on certain bacteria like *S. agalactiae* (Jain, 1979). The canal is also surrounded by a true sphincter of smooth muscle fibers which function in maintaining tight closure of the canal (Jain, 1979).

# 2.5.2 Humoral defences

# 2.5.2.1 Milk factors

Normal and mastitic milk contain humoral and cellular factors that inhibit bacterial growth (Jain, 1979). These are derived from blood serum or made locally by cells of lymphocyte system and plasma cell series situated close to the glandular epithelium (Jain, 1979; Lascelles. 1979). Lactoferrin in milk has been found to be bacteriostatic *in vitro* for a variety of organisms due to its iron-chelating activity. It is found in normal milk, neutrophils, dry period udder secretion and mastitic milk with a higher concentration in the latter (Jain, 1979).

Antibody and complement system in milk and colostrum are bactericidal to coliform and staphylococcal organisms and activity of this system increases with severity of mastitis (Lescelles, 1979). The major antibody in the milk and colostrum of the ruminant is IgG1 derived from blood and transferred selectively relative to IgG2 to the milk secretion. In acute inflammation this selective transfer of IgG1 is suppressed and IgG2 is able to get to the udder together with serum albumin. In inflammation, local production of antibodies especially IgA and IgM do occur. These antibodies are capable of attaching to invading organisms to elicit the release of opsonin, a substance that facilitates phagocytosis (Lescelles, 1979; Semiento and Soler 1995).

# 2.5.3 Cellular defences

Normal milk contains an estimated 1 x 10<sup>3</sup> leukocytes and somatic cells/ml, majority of which are macrophages (Semiento and Soler, 1995). Neutrophils enter the mammary gland during mastitis and are mainly involved in engulfing the invading pathogens (Jain, 1979).

# 2.6 Diagnosis of mastitis in goats

# 2.6.1 Physical examination

Diagnosis of clinical mastitis (observable) is easy and one mainly relies on physical examination of the udder and udder secretions to appreciate the changes in consistency, temperature, nature and volume of the milk. To observe the changes, milk is usually drawn into a strip cup and examined.

Clinical mastitis can be defined as subacute (mildly clinical) when only minor alterations in milk are visible. Clots and flakes may be observed in the affected halves and also slight swelling and tenderness. In acute cases, physical changes observed include swelling of the udder, tenderness on palpation, pain on touching (the animal resents being touched), udder feels hot, reduced secretion or none, secretions may also have clots, may be watery or blood stained (Samuel, 1977; Shearer, 1992).

In peracute cases, systemic changes may also be seen as rise in temperature, depression and anorexia (Samuel, 1977; Blood *et al.*, 1989; Shearer, 1992).

# 2.6.2 Indirect tests

These tests are important in subclinical cases, which are characterized by absence of gross changes in the udder and slight gross changes in volume and quality of milk (Maisi, 1990). In this situation the indirect tests of mastitis have to be applied for diagnosis. Tests carried out include California mastitis test (CMT), whiteside test and somatic cell counts either by direct microscopy or by electronic methods (Coulter counter or Fluoro-opto electronic cell countinguising Fossomatic machine) all of which estimate the somatic cells counts in the milk to confirm diagnosis.

These indirect tests of determining the somatic cell counts are greatly affected by other particles (cytoplasm particles) present in the goat milk which are not present in the bovine milk hence giving high readings even with normal milk in the goat (Mary and Michael, 1977; Dulin, Paape, Schulze, and Weinland, 1983; Maisi, 1990b). Direct microscopic leukocyte count although time consuming has been found to be the most accurate and sensitive test in diagnosing mastitis (Upadhaya and Rao, 1993). The use of coulter counter and the Fossomatic cell counts despite being simpler in determining the mammary gland infection status is in doubt due to the fact that they are influenced by other factors including breed, parity and stage of lactation. It is therefore recommended that high scores are confirmed by direct microscopy or bacteriological examinations (Boscos *et al.*, 1996). Schalm, Caroll and Jain, (1971) gave the following as an interpretation of CMT in goat milk (Table 2.1).

Table 2. 1: CMT reactions and their interpretation of goat milk as compared to cow milk.

Reaction	Changes	Mean of neutrophil count		
Renetion		Cattle	Goat	
0	No reaction	25,000	68,000	
Trace	Slight slime which tends to disappear on swirling	125,000	268,000	
+1	Distinct slime without gel	475,000	800,000	
+2	Immediate gel forms moves as a mass during swirling	1,900,000	2,560,000	
+3	Gel develops as a convex surface and adheres to bottom of the cup	>5,000,000	>10,000,000	

Uninfected goat milk may give a CMT reaction of trace or +1, but +2, +3 should be important indicators.

Serological diagnosis of mastitis caused by some bacterial species is also possible and in particular *Listeria monocytogenes* using enzyme linked immunosorbent assay (ELISA) (Bourry *et al.*, 1997).

Analysis of physiological changes in the milk does evaluate pathologic changes in goat milk. These include concentration of Lactose, Chloride, N-Acetyl-β-D-glucosaminedase (NAgase), antitrypsin, total lipids and cholesterol (Mary and Michael, 1977; Maisi, 1990b; Upadhaya and Rao, 1993). Upadhaya and Rao (1993) set a CMT score of +1 together with any two or three of the following threshold values as accurate indicators of subclinical mastitis in goats (Table 2.2).

Table 2.2: Threshold values for parameters used in mastitis diagnosis in goats.

Parameter	CMT +1 0.63x10 <sup>6</sup> /ml		
Leukocyte count (Direct)			
Lactose	4.33%		
Chloride	0.109%		

Maisi and Riipinen (1992) observed CMT score of  $\pm 1$  or  $\pm 2$  throughout lactation in healthy udder except during colostral period when the scores could go upto  $\pm 3$ ,  $\pm 5$  throughout lactation in infected halves. Thus it was recommended a CMT score of  $\pm 4$  to indicate infection and a CMT of  $\pm 3$  as suspicious. Subclinical staphylococcal infections were associated with significant elevation NAgase. NAgase concentration of healthy goat milk is  $6.0 \pm 1.5$  while that of infected milk is  $10.3 \pm 6.3$  (Maisi and Riipinen, 1992).

Somatic cell count is widely applied and is either directly counted or electronically counted using a coulter counter and Fossomatic equipment counter in which a threshold of 1 x 10<sup>6</sup>/m can identify most infected halves (Hunter, 1984; Kalogridou, Vassiliadou, Manolkidis and Tsigoida, 1992). Mary and Michael (1977) suggested 5 x10<sup>5</sup>- 2 x 10<sup>6</sup> cells/ml to indicate weakly pathogenic and non-pathogenic organisms. Hunter (1984) reported that all the coagulase positive staphylococcus infected samples had a somatic cell count of 2 x 10<sup>6</sup> cells/ml. Due to lack of a definitive diagnosis for subclinical mastitis, most researchers apply combinations of tests.

#### 2.6.3 Culture examination

In cases which are positive on the above tests or in cases where the doe is physically exhibiting the signs, culturing of the milk samples is the final step to determine the organisms involved and to do drug sensitivity tests (Blood *et al.*, 1989).

# 2.7 Treatment and control of mastitis

Treatment of mastitis in does, is similar to that of cattle and is highly effective in managing infection and returning the milk to normal composition though not to previous levels of

production. Degree of success depends on type of causative agent, speed of commencement of treatment, severity of tissue infection and duration of infection. Treatment of mastitis in does can either be local or parental depending on the etiological agent(s) and sensitivity results, extent of tissue damage, duration and severity of infection, cost of drug(s) of choice, their availability and access to patient (Mary and Michael, 1977; Blood *et al.*, 1989; Semiento and Soler, 1995).

Parentral treatment is advisable in all cases of systemic reaction to prevent/control development of bacterimea or septicemia. Higher than normal doses are used to ensure enough concentration get to the udder). Drugs commonly used include penicillin 16500 LU/kg body weight oxytetracyclines at 10 mg/kg body weight, Tylosine/erythromycin at 12.5 mg/kg body weight and sulphadimidine 200mg/kg body weight (Blood *et al.*, 1989).

Udder infusions are convenient and efficient in treatment of mastitis since they ensure a good concentration of the drug in the udder. It is always important to evaluate the gland as much as possible before infusion to ensure a good diffusion of drugs. The minimum inhibition concentration of the drug should be maintained in the gland for at least 24 hours. Penicillin streptomycin combination has been shown to be very effective in streptococcal and staphylococcal mastitis (Samuel, 1977, Blood *et al.*, 1989).

Milking out the udder is effective in eliminating some of the infections and this can be aided by injecting 2 I.U of oxytocin followed by infusion of the affected gland with half the content of the tube of bovine mastitis medication (Shearer, 1992).

For chronic cases, and especially those caused by *S. aureus*, dry treatment is recommended and this is done either at the beginning or end of dry period. This practice offers a good control program for mastitis (Samuel, 1977; Blood *et al.*, 1989).

Adoption of supportive therapy such as fluid therapy and hot formentation will be dependent on the extent of systemic involvement, local edema and tissue damage. In cases of gangrenous mastitis, it is advisable that the udder is extirpated surgically and the doe culled (Samuel 1977).

#### 2.8 Control of mastitis

Mastitis cannot be totally eliminated from the herds but the incidence can be held at a minimum. The major points in control of mastitis are good husbandry practices and sanitation. The pens, milking area and exercise area should be well drained and ventilated to provide a clean dry environment for the goats. The barns should have minimum trash, and barbed wire littering should be avoided in the barns and pasture area. Goats should be dehorned and have regular footcare thereby reducing potential for traumatic injury to the teats and udder. Goats with draining abscesses should be isolated and treated or removed from the herd (Shearer, 1992).

Milking procedures and hygiene are important in keeping the disease to a minimum level. Hair on udders and flanks should be clipped to avoid accumulation of dirt and excess moisture. Udder and teat should be washed with an appropriate preparation and dried before milking while the milkers hands should be kept clean and dry (Shearer, 1992).

The CMT should be performed on all lactating does monthly as this will help in detecting disease early (Blood *et al.*, 1989, Shearer, 1992). Intramammary therapy at drying off is also recommended as it eliminates existing infection and prevents establishment of existing infections. Half one tube of the standard bovine tube is sufficient (Shearer, 1992).

Mastitis control program for adoption should provide an economic advantage, be easy to understand and apply, fit in the management system employed and reduce occurrence of mastitis (Blood *et al.*, 1989).

A good control program aims at two major things namely

- i) Reducing duration of infection by treating all halves at drying off, treating clinical cases as they occur and culling chronic cases.
- Reducing new infection rates by dipping both teats in a suitable disinfectant after each milking, in case of machine milking, adequately servicing and maintaining milking machine, in case of hand milking, washing hands in between milking (Blood *et al.*, 1989).

#### CHAPTER 3

# 3.0 MATERIALS AND METHODS

# 3.1 Study area

This study was carried out in central Kenya highlands (Nyeri District) which is a high potential district that is densely populated. The area has a humid climate receiving an annual rainfall ranging from 700mm to 2000mm. The dense population of approximately 200 persons per square kilometre has led to land subdivision (fragmentation) to essentially uneconomical units of about 0.74 ha/household with each household holding an average of 5 persons (Kinuthia 1997).

# 3.2 Choice of group

Seven dairy goat groups were chosen for this study with the following considerations:-

- The group had to have been with the Intergrated Small Livestock Project (ISLP) project for 3-4 years and have 35-40 mature female goats (does), whether in milk or not.
- The group had to be within a range of at least 30-40 km from the Karatina Veterinary Investigations laboratory where the samples were to be processed to reduce logistical problems including immediate delivery of samples from the farms to the laboratory.
- The groups had to be accessible by road in wet and dry season.
- The farmers involved had to have some form of record keeping especially on goat identification, date of kidding, litter size, parity and service date.

v) The farmers nominated had to be willing to participate and cooperate in data collection throughout the study.

From a list of 26 ISLP registered groups, seven that fitted the set criteria were selected (Table 3.1).

Table 3.1: Selected farmer groups and their respective localities (division) and allocated codes.

Group code	Name	Division	No of lactating does
1	Kimumu	Othaya	15
2	Ngaini	Mathira	17
3	Miiri	11	17
4	Gathareini	11	13
5	Thiha	Mukurweini	31
6	Ngatha	- 11	23
7	Muruguru	Municipality	14

# 3.3 Sample size determination

To calculate the correct size of sample needed for determining the prevalence (P) of mastitis the following formula was used based on Thrushfield's (1990) recommendations.

$$n = \underbrace{P \times Q \times (1.96)^2}_{\text{(L)}^2}$$

Where P = an initial estimate of the prevalence here used as 10%. The prevalence in this case was estimated from studies carried out in other countries with almost similar practices as Kenya such as Nigeria and India i.e. (10.9%, 9.4% and 10%) (Egwu *et al.*, 1993; Ameh *et al.*, 1994; Dasgupta *et al.*, 1993) calculated as the arithmetic mean of these three.

Q = (100 - P). A confidence interval of 95% and an admissible absolute error (L) of 5% was used. Therefore based on the above formula, it was determined that at least 110 does were an adequate sample size. The seven groups purposefully selected for the study had 130 lactating does that were included in the study.

#### 3.4 Choice of animals

The does selected for inclusion were those that:

- i) Were participating in the ISLP dairy goat-upgrading program.
- ii) Were identifiable, had records including those of service, kidding date, litter size, parity and the level of crossing.
- iii) Were in milk (being milked) or suckling. All had to be >3 days post kidding to ensure no colostrum was sampled.

Based on these criteria, a total of one hundred and thirty lactating does were recruited for inclusion in the study.

# 3.5 Community sensitisation and mobilisation

There was an initial one-week orientation to the study area during which all the seven groups participating in the study were visited. Visits were first made to the divisional headquarters where information on the number of goats in milk, localities, and possible convenient day of visiting the groups was gathered from the dairy goat extension coordinators. Information on the production calendar and incidence of the disease was also gathered. Specific appointments were made when individual farms were to be visited for examination of the does and sample collection

# 3.6 Assessment of general farm management

The selected farms were visited monthly for 3 months to observe the general management procedures, types of housing, feeding regimes, milking procedures, frequency of emptying the pens and stocking rates. The housing was classified as either raised or earthen based on the type of construction. The general hygiene of the pens was graded as poor or good based on the frequency of emptying the pens. Milking hygiene was also graded as poor or good based on description of the milking procedure (whether a disinfectant was used during milking, the milker washed his hands before milking, or a towel was used). The results were recorded in a questionnaire as in Appendix 1.

# 3.7 Retrospective survey of mastitis

A field questionnaire was dispatched to all divisional co-ordinators of the project to determine the number of cases of mastitis recorded over the past one year. Details of the treatment instituted and response rate were also included as shown in Appendix 2.

#### 3.8 Examination of the animals

Each of the lactating does was subjected to a general physical examination to determine the general health status. Temperature, heart rate and respiratory rate were determined and the general demeanor observed. The general body condition score was graded as poor, fair or good. The mammary gland (udder) and the teats were examined for congenital abnormalities, physical injuries/defects, consistency and warmth.

#### 3.9 Sampling of the lactating does

All lactating does in the selected farms were sampled once every month for three months.

Does kidding during the study period were recruited and sampled along with those that had been previously recruited and sampled.

## 3.9.1 Sampling procedure

Does were restrained and sampled in their pens or in the open. Before each sampling, the teats were thoroughly cleaned with gauze soaked in 70% alcohol. The first steams of fore milk were discarded after which 10 ml of milk from each half were drawn into separate labelled bijoux bottles. The sterile bottle was held at an angle to the body wall of the doe to avoid falling hairs entering the bottle. These samples were kept in a cool box at 4 -7°C until taken to the laboratory the same day for analysis.

## 3.10 Laboratory sample processing

# 3.10.1 Mastitis indicator test (CMT) (Somatic cell count)

On arrival in the laboratory, 2 mls of each sample from each half were placed into each cup of CMT plate and 2 mls of the CMT was squirted into each of the cups. This reagent contains a surface acting detergent (sodium alkyl aryl sulphonate and a dye bromocresol purple). This reagent acts by precipitating the nucleus material of the cells in milk (leucocytes and somatic cells) resulting in gel formation whose thickness depends on the number of cells in milk. The paddle was rocked gently to allow proper mixing of the reagents and the sample. The results were read within 10 seconds and recorded as either nil, trace, +1, +2, +3 reaction. After each test the plate was washed and rinsed twice before the next set of samples were tested.

#### 3.10.2 Direct leukocyte count

The microscopic somatic cell count technique was used to determine the number of leukocytes in the samples. The cells in the milk samples were prepared and examined by the standard Gram staining techniques as outlined below (Coles, 1968; Manual for Veterinary Investigation Laboratories 1986).

- 1. After mixing the milk thouroughly to disperse the cream throughout the milk, a thin smear of milk sample from each half was prepared on a clean microscope slide over an area of 1 square centimetre using the standard 4mm bacteriological wire loop (0.01 ml) and was allowed to dry at room temperature for at least 30 minutes.
- 2. The smear was fixed by immersing the slide in equal parts of methanol and ether (deffating fixative) for 15 minutes.
- 3. Then the smear was drained and stained with the standard Grams stain before being examined under microscope at x 100. Twenty fields were examined and the number of leukocytes counted multiplied by 10<sup>5</sup> to give the value in every ml of milk for each half (Coles, 1968; Manual for Veterinary Investigation Laboratories, 1986).

## 3.10.3 Bacteriology

#### 3.10.3.1 Bacterial isolation and identification

Milk sample from each half was plated on sheep blood agar (Oxoid UK) in duplicates and McConkey agar (Oxoid UK) using the standard wire loop (0.01ml) which had been flamed and cooled. One sheep blood agar plate was incubated aerobically while the other was incubated anaerobically at 37°C overnight. The plates were examined after 24 hours and if no **DEVERSITY OF NAIROBI LIBORIA** 

growth was observed, they were reincubated for a further 24, 48, and 72 hours. When no growth was evident after 72 hours, the sample was regarded as bacteriologically negative. A doe was considered infected if five or more pure bacterial colonies were present on any of the plates (Manser, 1986). Where there were mixed growth, individual similar colonies numbering more than five were then subcultured onto the same media on which it grew until pure cultures were obtained. The pure culture isolates were then gram stained and observed under oil immersion for identification. The organisms were subsequently subjected to standard biochemical tests using characterization and differentiation media which included mannitol salt agar, citrate agar, urea agar and oxidative and fermentative media (Manual for Laboratory investigation techniques, 1986). Other tests carried out for differentiation included catalase test, and the coagulase test. The organisms isolated were identified to species levels where possible using Manual of Veterinary Laboratory techniques 1986.

# 3.10.3.2 Antibiotic sensitivity testing

Each of the organisms isolated was streaked uniformly onto nutrient agar plates. Oxoid multidisks (Oxford Ltd. London, England) were then placed on the agar surface before the plate was incubated for 24 hours at 37°c. The multidiscs contained ampicillin (25  $\mu$ g), streptomycin (10  $\mu$ g), tetracycline (25  $\mu$ g), chloramphenical (30  $\mu$ g) and sulphamethoxazole (200  $\mu$ g), cotrimoxazole (25  $\mu$ g), kanamycin (30  $\mu$ g), gentamycin (10 $\mu$ g). Resistance or sensitivity to each drug was determined by measuring the size of the zone of growth inhibition around the discs as recommended by the manufacturer.

# 3.1.1 Statistical analysis

3.1.2

Each of the variables was coded for ease of analysis and the data entered into a statistical analysis program (STATISTICS (SX.4.0). Descriptive statistics analysis using the same

program was undertaken on the categorical variables (breed, CMT, abnormalities, doe type, grazing, housing, milking hygiene, isolates, stage of lactation, milk purpose, number of kids and parity stage) for the three samplings. The prevalence of the various bacterial isolates for each sampling was also determined.

The overall infection status for individual doe was classified as either infected or non-infected.

Does were classified as infected if they were positive by any of the diagnostic tests used. Does with missing milk cultures were excluded from the data analysis for that sampling.

Using Pearson chi-square test for homogeneity, bivariate relationships between infection status and the various variables mentioned above was examined for the pooled data from the 7 farmer groups (130 does).

A logistic regression model was used to analyse more than two confounding risk factors of mastitis under investigation in this study. Since the data was in the form of discrete variables, introduction of indicator variables was found necessary with variables having more than two categories into one less than the actual number of categories. The data (predictor/risk factors and the dependent variable (mastitis) was then run through the regression model Appendix 3. From these results, variables with high p values were eliminated stepwise re-running the model after every elimination of a variable. This was repeated until the best model remained with variables with p values of less or equal to P<0.05. This was followed by a calculation of the odds ratio and their confidence intervals for the remaining variables Appendix 3.

#### **CHAPTER 4**

#### 4.0 RESULTS

#### 4.1. General farm characteristics

All the small holder farms recruited for the present study were located in Nyeri District. The farms, which measured ≤ 0.721 ha each had little or no grazing land and were registered with the Integrated Small Livestock Project, a German Technical co-operation (GTZ\Government of Kenya (GOK) Intergrated Small Livestock development project (ISLP) in central highlands.

#### 4.1.1. Housing

The recommended houses were timber structures with wood offcuts sides, raised slatted wooden floors and iron sheet roofs. It was observed that 115 (88%) does were housed in raised houses while 15 (12%) were housed in temporary structures with earthen floors (Fig 4.1 and 4.2 respectively). In this latter group of structures, remains of shrubs and grass used as feed for the does served also as the bedding. Adult goats were kept in twos per 3m² cubicle and had to be of the same sex especially and preferably littermates. There were separate pens for the adult females, males, and kids and each pen had an open yard where the goats could come out occasionally to bask and exercise. Each of these houses also had a provision for an in built feeding trough and a waterer. One out of the 130 farmers had constructed a milking crush for milking his does while in the other farms does were either milked outside or inside the pens.

The general hygiene in these farms differed from one farm to another. This depended on whether the does were in the raised houses or on the earthen floors. In raised houses cleaning the pens was easy as the farmers only emptied the floor under the raised floors while those with earthen floors had to clear the whole pen of the wet beddings. In this latter group of structures, no rainproof roof was present and sides were not covered. The frequency of cleaning the pens varied from farm to farm. Fifty farmers (39 %) indicating that they cleaned pens once a week, while 80 (61%) cleaned the pens once in two to four weeks.

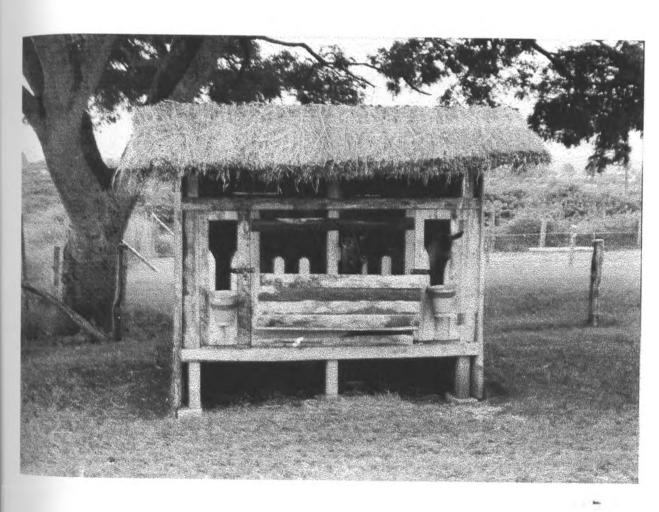


Fig 4.1: Modern housing structure recommended for the dairy goats.



Fig 4.2: Traditional temporary structures being used by some farmers to house the dairy goats.



Fig 4.3: Open yard part of the houses for exercise

## 4.1.2 Feeding/grazing

Most of the does 118 (91%) were zero-grazed while the rest 12 (9%) were either tethered and grazed along the roads or partly zero-grazed and partly tethered. Feeds consisted of shrubs and weeds from the farms, remains of the food from the house, napier grass, sweet potato vines and maize plant remains. The chopped feed (Napier grass, banana peels) was placed in the feeding troughs while sweet potato vines and shrubs were hang in the pens. One farmer indicated that he used dairy meal supplement during milking while others 20 of 130 (15%) offered maize as part of the daily ration. All farmers indicated they supplied water to the does within the pens in simple homemade waterers by. Seventy percent of the farmers supplied water ad libitum while 30 % indicated that they replenished the supply only when the containers were found dry.

# 4.1.3 Milking and milking hygiene

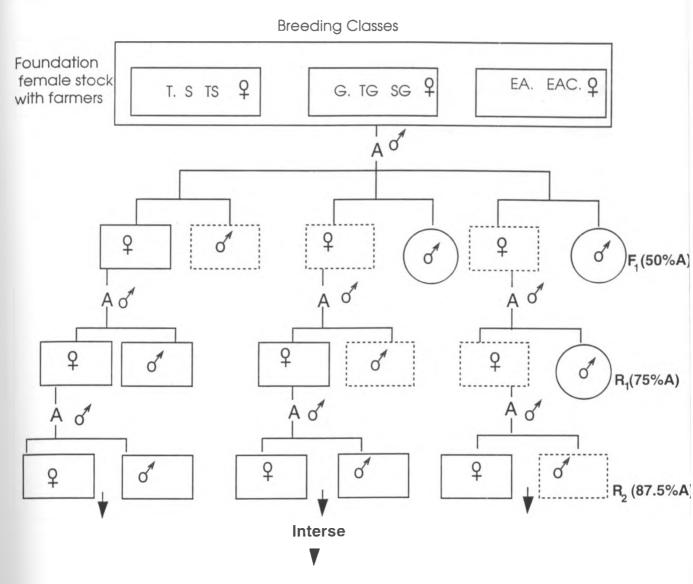
A total of 97 goats were being milked at the time of first sampling. Milking was done twice a day both in the morning and evening. Forty two of the does (43%) routinely had their udder halves washed with plain warm water before milking while 55 (57%) were washed with warm water and milking salve was applied before milking. All the farmers interviewed did not use disinfectant in form of teat dip before or after milking or performed any on-farm mastitis testing using a strip cup. As a policy, does were milked until a month to the expected date of kidding when the does were dried off.

#### 4.2 Doe characteristics

# 4.2.1 Breeds (Breeding chart)

Does that participated in the study were of various breeds including mixed crosses (Figure 4.4). All the lactating does in the choosen farms were miked, irrespective of their age. The does comprised of 46 (35 %) local crosses and 84 (65 %) German alpine crosses. The local crosses comprised of Toggenburg crosses, Galla crosses and the small east African goat crosses. The German Alpine crosses comprised of mainly (70%) F1 which are 50 % German Alpine and (30%) F2 which are 75% German alpine.

# **Breeding Plan for the Dairy Goat Development Programme**



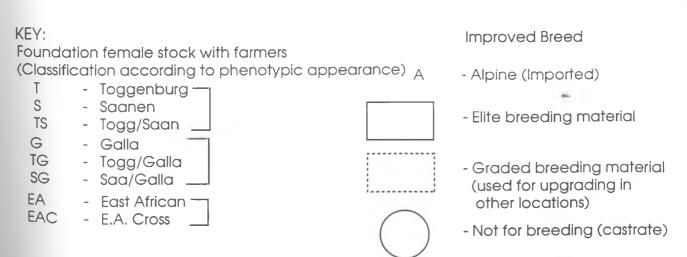


Figure 4.4: Breeding chart used by the integrated small livestock project (GTZ/GOK) in central Kenya highlands

# 4.2.2 Lactation and milk production

Out of the 130 does sampled, 46 (35 %) does were in early lactation and their average milk production was  $1.53\pm0.21$  litres/day, 40 (31 %) were in mid lactation with an average production of  $1.49\pm0.09$  litres/day while 44 (34 %) were in late lactation with an average production of  $1.61\pm0.12$  (Fig 4.5).

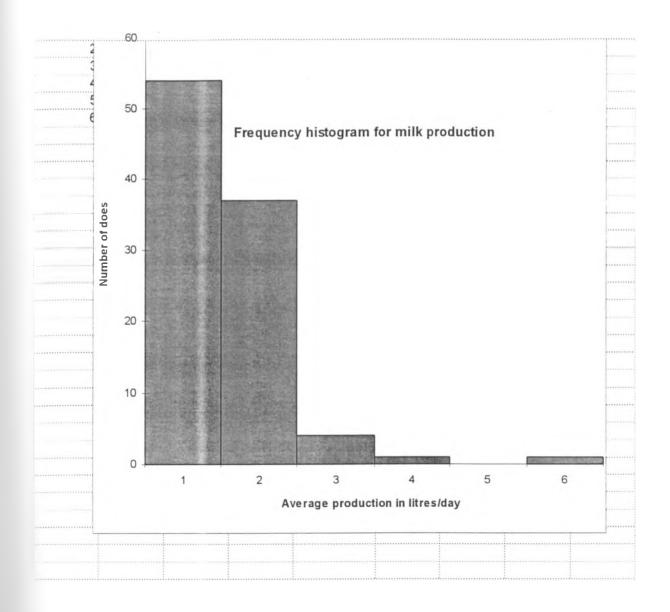


Figure 4.5: Distribution of average daily milk production of the selected does.

#### 4.1.2.3 Number of kids

Seventy-five (58 %) does had one kid, 51 (39 %) had twins, 2 (2 %) had 3 kids and 1 (1 %) had 5 kids but 2 had died. As a policy, kids were weaned at 80 days in all flocks and some farmers only started milking for home consumption weaning. Of the does sampled, one of the German alpine crosses was found to have a precocious udder without any kid.

## **4.2.3** Parity

Table 4.1 shows a summary of the parities of the does sampled. The doe with a zero parity was found to have a precocious udder and the farmer complained that the doe came on heat regularly but had never conceived despite being served by a fertile buck. It was apparent that higher numbers of does 44 % were the primiparas followed by those of two parities.

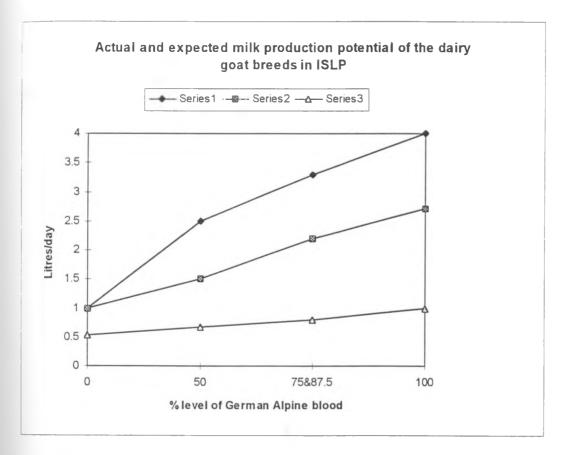
Table 4.1: Distribution of does in relation to parity.

Parity		Frequency	Percentage
	0	1	1
	1	57	44
	2	39	30
	3	17	13
	4	10	8
	5	5	4
	6	1	1
Total	21	130	100
Mean	3.5	21	

# 4.2.4 Purpose of the milk

Most of the does 99 (77%) were being raised for milk production. Of these, 78 (60%) of the does being milked without suckling while 22 (17%) were suckling and being milked for

The ISLP milk production annual report (1997-1998) showed that the actual milk yield being realized by the farmers with the local breeds is about 0.53 litres / day, F1 German Alpine crosses are producing 0.67 litres / day and, F2 crosses are producing 0.79 litres /day which is below the expected (Figure 4.6).



Key :Series 1-Expected milk yield

2-Assuming no suckling 3-Actual milk yield

Figure 4.6: Actual and expected milk production potential of the dairy goat breeds in ISLP area.

## 4.3 Physical examination results

Throughout the study period, all the does examined had the body temperatures, respiratory, heart rates within the expected ranges (Blood *et al.*, 1989). Seventy percent of the does were in good body condition while the rest (30 %) were in fair body condition. At the time of the study, no doe had any evidence of mastitis. However, physical examination revealed a number of abnormalities. Eight percent of these affected either of the halves while and 92% of the abnormalities affecting both halves (Table 4.2).

Table 4.2 Distribution of the various abnormalities in the halves of goats examined.

Abnormality	right(%)	left(%)
None	117(90 %)	117(90 %)
Supernumerary teats	3(2 %)	4(3 %)
Udder impetigo	2(2 %)	3(2 %)
Pendulous udder	2(2 %)	2(2 %)
Udder asymmetry	1(1%)	1(1%)

# 4.4 Survey of prevalence of mastitis

# 4.4.1 Field questionnaire results

The results obtained from the questionnaire (Appendix 2) indicated that 17 cases of mastitis had been reported in the previous 12 months. Of these, seven were in Nyeri, six in Maragwa, and twoeach in Kirinyaga and Embu each.

The most commonly observed clinical signs were clots in milk and swelling of the udder.

Other signs observed included pain, reduced milk production, firm udder, watery milk

production, blood stained milk and reduced feed intake. None of the does exhibited clinical signs of mastitis during the study.

#### 4.5 Laboratory findings

Milk samples collected were free from any discoloration, foreign particles or any physical abnormalities such as clots.

## 4.5.1 Mastitis indicator test (CMT) results

In the first sampling of all the 130 does, 49 (31%) had zero reading on both halves, 18 (15%) had a CMT score of trace on both halves, and 13 (10%) had a CMT of +1 on both halves. Seven had a CMT of +2 on both halves while only two had a CMT of ≥3 on both halves. Only 9/130 (7%) of the does had both halves CMT mastitic in the first sampling. In the second sampling 108 does were sampled out of which fifty-six (52 %) had nil reading on CMT plate on both halves, eight (7 %) had CMT of trace on both halves and 18 (17 %) had CMT of +1 on both halves. Five had a CMT of +2 on both halves while only 1 had a CMT of >+3 on both halves. Six (6) out 108 (6 %) of the does had significant readings both halves. In the 3rd sampling 77 does were sampled. Out of these 36 (47%) had nil reading on both halves, 10 (13%) had a reading of trace, 5 (6%) had a reading of +1, 3 (4%) had +2 reactions while only 1 (1%) had >+ 3 reaction. Four (5%) does therefore had paired significant readings.

The number of does with significant CMT readings (>2) on their left halves were 15% in first sampling, 10% in second sampling, and 9% in the third sampling as compared to 12% in first sampling, 7% in second sampling, and 6% in third sampling on their right halves (Table 4.3).

Table 4.3: CMT results for the udder halves for the three samplings.

Reading	Frequency(%)					
	Right (%)			Left(%)		
	1 <sup>st</sup>	2 <sup>nd</sup>	$3^{rd}$	1 st	2 <sup>nd</sup>	3 <sup>rd</sup>
Nil	65(50)	64(59)	44(57)	56(43)	62(57)	40(52)
Trace	26(20)	14(13)	16(21)	35(27)	16(15)	17(22)
+1	24(18)	23(21)	12(16)	20(15)	23(21)	13(17)
+2	11(9)	6(6)	4(5)	17(13)	6(6)	6(8)
≥+3	4(3)	1(1)	1(1)	2(2)	4(4)	1(1)

NB The numbers in bracket represent the percentage of the total number of does sampled during that sampling.

Does were classified as either CMT mastitic or non-mastitic based on a cut off score of +2 (Table 4.4). A chi-test revealed no significance (P<0.05) association between CMT status and the halve.

Table 4.4: Summary table of the proportions of CMT mastitic for both halves in the three samplings

Half	Number infected in each sampling			
Right	1 st	2nd	3rd	
Left	15/130* <sup>m</sup>	7/108 <sup>n</sup> *	5/77 <sup>n</sup> *	
			(X-)	
	19/130* <sup>m</sup>	10/108* m	7/77* <sup>m</sup>	

**Key:** \* Values on the same column with the same superscript were not significantly (p<0.05) different from each other.

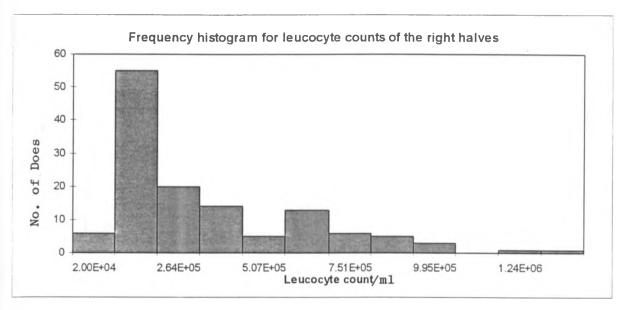
m, n: Values on the same row with different letters were significantly (p<0.05) different.

## 4.5.2 Direct leukocyte counts (DLC)

In this study, counts  $>8 \times 10^5$ /ml was considered as the cut off point. The direct leukocyte count ranged from  $2 \times 10^4$  -1.36 ×  $10^6$  with a mean of 2.754 ×  $10^5$  in the right half and 3.21 ×  $10^5$  in the left quarter in the first sampling. Using this criteria, the does that were found to be DLC mastitic were 11/130 (10%) and 16/130 (12%) in their right halves and left halves respectively (Figures 4.7 and 4.8).

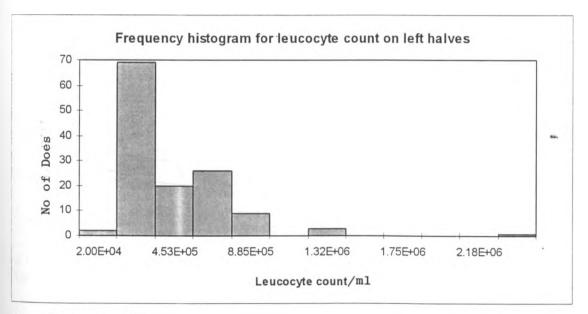
In the second sampling the leukocyte count ranged from  $2x10^4$ -  $1.26x10^6$  with the mean leukocyte count for the right quarter being  $2.28 \times 10^5$  while that of the left halve was  $2.30 \times 10^5$ . Using the above cut off point, 8/108 (7%) and 10/108 (9%) right halves and left halves respectively were found to be DLC mastitic (Figures 4.9and 4.10).

In the third sampling the leukocyte count ranged from 2x 10<sup>4</sup> - 1.24x 10<sup>6</sup>. The mean leukocyte count for the right half was 1.90 x10<sup>5</sup> while the left was 2.94 x 10<sup>5</sup>. Right halves were mastitic in 4/77 (5%) while left halves were DLC mastitic in 12/77 (15%) (Figures 4.11 and 4.12).



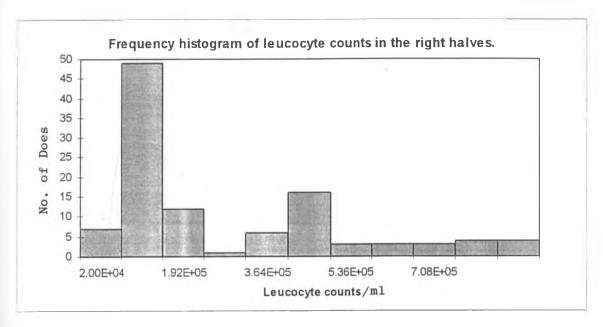
Key:  $E+X=10^X$ 

Figure 4.7: Frequency distribution of leukocyte counts for the right halves (1st Sampling).



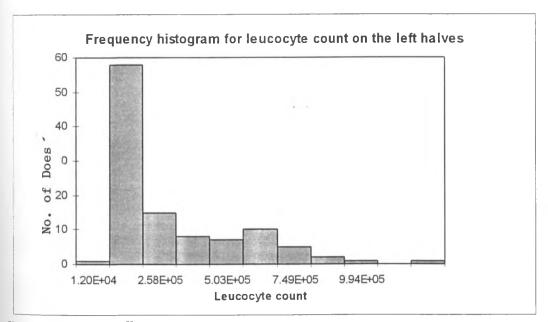
Key:  $E + X = 10^{X}$ 

Figure 4.8: Frequency distribution of leukocyte counts for left halves (1st Sampling).



Key:  $E+X=10^X$ 

Figure 4. 9: Frequency distribution of leukocyte counts for right halves (2nd Sampling)



Key:  $E + X = 10^X$ 

Figure 4.10: Frequency distribution of leukocyte counts for left halves (2nd Sampling).

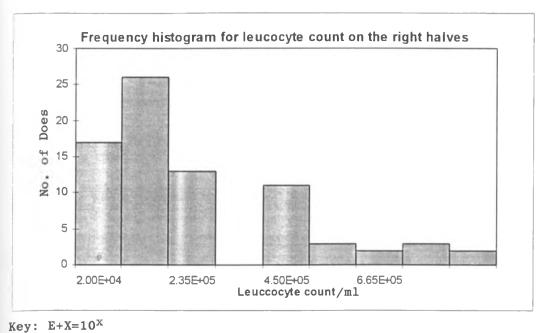
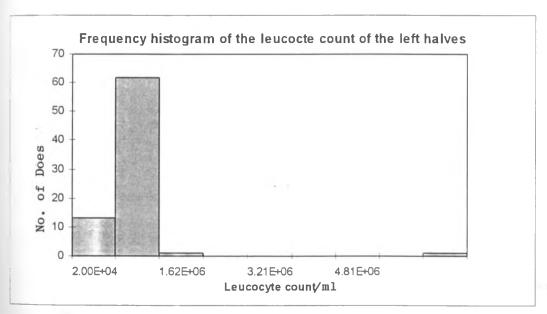


Figure 4.11: Frequency distribution of leukocyte counts for right halves (3rd Sampling)



Key:  $E + X = 10^X$ 

Figure 4.12: Frequency distribution of leukocyte counts for left halves (3rd Sampling).

Does that were DLC mastitic in either of the halves were compared to check for significant prevalence differences between the two halves (Table 4.5). In the first and second sampling no significant (p<0.05) differences were observed but in the third sampling significant (p<0.05) differences were observed.

Table 4. 5: Mastitic samples in the various samplings by DLC

Halve	Samp	Sampling						
	1 st	2 <sup>nd</sup>	3 <sup>rd</sup>					
Right	11*	8*	4*					
Left	16*	10*	12 <b>*</b>					
Total	130	108	77					

**Key:** \* Values on the same column with different superscripts significantly (P<0.05) differed from each other.

# 4.5.3 Bacteriology results

Various organisms were isolated from the milk samples. Most of the organisms were differentiated up to genus level except in cases where reagents for differentiation to species levels were available. Each of these organisms was either isolated in pure or mixed culture. Organisms isolated: included, coagulase positive *Staphylococcus aureus*, coagulase negative *Staphylococcus*, *Streptococcus* spp, *Micrococcus* spp, *Actinomyces* spp, *Acinetobactor* spp, *Achromobactor* spp and *Bacillus* spp (Table 4.6).

Table 4.6: Distribution of organisms isolated from the halves in the first sampling.

Type of organism isolated	1 <sup>st</sup> Sampli	ng	2 <sup>nd</sup> Sampli	ing	3 <sup>rd</sup> Sam	pling
	Freq.(%)	(R &L)	Freq.(%)	(R&L)	Freq.(%	(n) (R&L)
	Right	left	Right	Left	Right	Left
Nil	95(73)	80(62)	81(75)	81(69)	59(77)	57(74)
CPS (Coagulase Positive Staphylococcus)	e 6(5)	7(5)	7(7)	9(8)	5(6)	7(9)
CNS(Coagulase negative Staphylococcus)	e 15(12)	16(12)	12(11)	9(8)	9(12)	7(9)
Micrococcus spp	8(6)	13(10)	1(1)	4(4)	2(3)	4(5)
Acinetobactor spp	2(2)	1(1)	2(2)	2(2)	0(0)	2(3)
Streptococcus spp	1(1)	0(0)	0(0)	0(0)	0(0)	l(1)
Actinomyces pyogenes	0(0)	3(4)	1(1)	3(3)	2(3)	0(0)
Others spp	1(1)	3(4)	1(1)	2(2)	0(0)	0(0)
CNS + Micrococcus spp	1(1)	1(1)	1(1)	2(2)	0(0)	0(0)
Actinomyces + others	1(1)	1(1)	1(1)	0(0)	0(0)	0(0)
S. aureus and others	0(0)	1(1)	0(0)	0(0)	0(0)	0(0)
Acinetobactor + others	0(0)	2(2)	1(1)	1(1)	0(0)	0(0)

Key: R=Right half, L=Left half

In the first sampling, 73% and 63% of the samples from the right and left halves respectively had no growth (Table 4.6). Most of the infections were due to *Staphylococcus* spp as shown by 16% of 27% and 18% of 39% staphylococcal isolations in their right halves and left halves respectively. In both halves the majority of these were the coagulase negative *Staphylococcus* (12% of 27% and 12% of 18% in the right and left halve respectively). Infected left halves were more than the right, halves as in this case 39% of these had microorganisms compared to 27% in the right halves (Table 4.6).

In the second sampling, majority of these samples did not yield any microorganism (75%) (Table 4.6). Similarly, *Staphylococcus* spp were still the most prevalent organism, 18 % out of 25% isolates especially coagulase negative *Staphylococcus* (11%).

As in first sampling, third sampling also revealed low prevalence of intramammary infection in the right half (23%) compared to left halves (26%). Similarly, highest prevalence was for the *Staphylococcal* spp 18% out of 23% in the right halves and 18% out of 26% in the left halves (Table 4.6).

S. aureus was isolated from both halves in 3 out of 15 does (20%) but most (67%)of the organisms that were isolated from both halves were coagulase negative Staphylococcus. Microccoccus spp was isolated from both halves in 2 out of 15 (13%) while other isolates were only encountered in one half at any one time. In the second sampling, S. aureus was isolated from both halves in 9% of the does, coagulase negative Staphylococcus 14%, Micrococcus 2%, Actinomyces spp 1% while the others were isolated in 2% of the does. In the third sampling, S. aureus was isolated from both halves in 4 of the 77 (9%) does. Coagulase negative Staphylococcus was isolated in 3 does (4%), Micrococcus 1 of the 77 does (1%). None of the other organisms (Actinomyces spp and Acinetobactor spp) were found in both halves at any time. A summary of the infected does in the various sampling was compiled and subjected to a chi-test for significant differences in infection rates of the halves (Table 4.7).

Table 4.7: Summary of mastitic does (bacteriologically positive) in the three samplings.

Half	1 <sup>st</sup> Sampling	2 <sup>nd</sup> Sampling	3 <sup>rd</sup> Sampling
Right	35/130*	27/108*	18/77*
Left	40/130*	33/108*	20/77*
Total	130	130	108

**Key:** \* Proportions on the same column with similar superscripts were not significantly different (P<0.05) from each other.

# 4.6 Group infection results

Prevalence of mastitis differed significantly (p<0.05) from one group to another with a range of 22 % to 76 % (Table 4.8).

Table 4.8: A summary of does infected in the various farmer groups sampled.

Group code	Number of does	Number infected	Percentage (%) of infected does.
1	15	4	27
2	17	5	76
3	17	5	76
4	13	7	54
5	31	12	39
6	23	5	22
7	14	9	64

Group 5 and 6 were in Mukurwe-ini division and both had low mastitis prevalence rates compared to the groups (2,3 and 4) in Mathira division and group (7) in Municipality division. In general Mathira division had the highest mastitis prevalence rates.

### 4.7 Effect of various risk factors studied on infection status

Risk factors affecting the infection status of the udder under investigation included, type of doe (milking or suckling), purpose of milk (home consumption, kid consumption, both kid and home consumption), litter size, parity, milking hygiene, type of housing and breed. The prevalence rates of infection in the various categories in which the factors had been grouped were subjected to statistical analysis for significant differences between the categories (Appendix 3).

# 4.7.1 Type of doe on infection status

From the two groups the goats had been classified, (milking, non-milking) results of infected proportions were as shown (Table 4.9). Based on this classification, 43/96 (45%) of the milking does, had infection on one or both halves. In the suckling does (non-milking) 19/34 (56%) had organisms isolated from one or both halves (Table 4.9).

Table 4.9: A summary of the proportions of doe types affected.

Sampling	Infected proportions (%)				
	Milked	Suckling			
1st Sampling	43/96(45) <sup>m</sup>	19/34(56) <sup>n</sup>			
2nd Sampling	31/81(38) <sup>m</sup>	12/27(44) <sup>m</sup>			
3rd sampling	20/52(38) <sup>m</sup>	13/25(52) <sup>n</sup>			

**Key:** " Values on the same row with different superscripts were found to be significantly (P<0.05) different from each other on chi- test.

In the 2nd sampling 31/81(38 %) milked does had infection on one or both while in the suckling does 12/27 (44%) were infected in one or both halves (Table 4.9). In the third

sampling 20/52 (38%) milked does had infection while 13/25 (52%) of the suckling does were infected (Table 4.9). A chi test on these results revealed that infected proportions in the suckling category were significantly (P < 0.05) higher than in the milked category during first and third sampling (Table 4.9).

# 4.7.2 Effect of purpose of milk on infection status

Based on the eventual use to which the milk was put, does were grouped into those that produced milk for home consumption only, kid consumption only, and both kid and home consumption. The findings of the proportions infected in the various categories for the three samplings are shown (Table 4.10).

Table 4.10: A summary of the proportions affected in the three categories of the does.

Purpose of milk categories	Infected pro	portions (%)	
	1st sampling	2nd sampling	3rd Sampling
Kid consumption	15/29(51) <sup>m</sup>	12/24(50) <sup>m</sup>	6/15(40) <sup>m</sup>
Home consumption	38/79(48) <sup>m</sup>	24/63(38) <sup>m</sup>	19/50(38) <sup>m</sup>
Both	10/22(45) <sup>m</sup>	8/20(40) <sup>m</sup>	4/12(33) <sup>m</sup>

**Key** <sup>m</sup>; Values on the same column with the same superscript were not significantly (p<0.05) different on chi-test.

Out of the 29 exclusively suckling does in the first sampling, 15 does (52%) were infected in one or both halves. Of the 24 does sampled in this category during the second sampling, 12 (50%) were infected in one or both halves while 6 (40%) out of the 15 in the same category sampled in the third sampling were infected (Table 4.10).

Thirty-eight does out of 79 does in the second category (home consumption only) were found to be infected in one or both halves in the first sampling. In the second sampling 24/63 (38%) does in this category were infected, while in the third sampling 19/50 (38%) in the same category were infected (Table 4.10).

In the third category (both kid and home consumption), 10/22 (45%) does sampled were infected in one or both halves, and in the second sampling, 8/20 (40%) does sampled were infected. In the third sampling, 4/12 (33%) does sampled in this category were infected (Table 4.10). There were no significant (P<0.05) differences in the proportions affected by mastitis between the various categories of purpose of milk. On logistic regression purpose of milk was not found to affect the infection status of the doe significantly (P<0.05, R=0.04).

# 4.7.3 Relationship between litter size and prevalence of mastitis

The does were grouped into three categories namely one kid, twins and any with three or more kids. Thirty-five (47%) does of the 75 with one kid in the first sampling were infected on one or both halves while 25 of the 58 does (43%) does in the same category in the second sampling were infected. In the third sampling 17 out of 40 does in this category were infected (Table 4.11).

Table 4.11: Proportions of affected does in the various categories.

Category	Proportions affected						
	1st	2nd	3rd				
1 <sub>n=1</sub>	35/75(47)*	25/58(43)*	17/40(42.5)*				
2 <sub>n=2</sub>	25/51(49)*	18/46(39)*	13/35(37)*				
3 <sub>n≥3</sub>	3/3(100)#	0/3(0)#	1/2(50)#				

**Key:** \*#; Values with on the same column with different superscripts were significantly different from each other.

In the second category 25/51 (49%) of the does were infected in the first sampling and 18/46 (39%) were infected in the second sampling. In the third sampling 13/35 (37%) does sampled in this category were infected in either one or both halves (Table 4.11).

All the three does in the third category sampled in the first sampling were found to be infected. In the second sampling none of the three does sampled in this category was infected while in the third sampling only one of the two does in this category was infected (Table 4.11). The litter size did not significantly (P<0.05) affect the infection status (Table 4.11).

# 4.7.4 Effect of parity on infection status of the halves

The does were grouped into three categories according to the number of parities which included 1st parity (1), 2nd parity (2), third (3) and greater than or equal to fourth parity (4) (Table 4.12).

Table 4.12: Summary table of the proportions infected in the various stages of parities.

Parity	Proportions	affected	
	1st	2nd	3rd
1 <sub>n=1</sub>	26/56(46)*	16/46(35)**	14/33(42)*
2 <sub>n=2</sub>	18/39(46)*	13/32(41)*	7/23(30)* *
3 <sub>n=3</sub>	9/18(50)*	7/14(50)* *	5/12(42) *
4 <sub>n≥4</sub>	7/16(44)*	7/15(47)*	5/9(55)* *

**Key:** \*, \*\*; Values on the same column marked with two stars were significantly different from each other (p<0.05) on chi-test.

In category one, 26/56(46%) and 16/46 (38%) does in the 1st and second sampling respectively were infected on one or both halves. During the third sampling, 14/33 (42%) sampled in this category were infected (Table 4.12).

Of the 39 does in category two, 18(46 %) were found to be infected in the first sampling. In second and third sampling, 13/32(41%) and 7/23(30%) does respectively were infected in this category (Table 4.12).

Nine (50%) out of 18 does in category three in first sampling were infected while in second sampling, 7/14 (50%) does in the same category were found to be infected. In the third sampling infection was found in 5/12 (42%) in this category were infected (Table 4.12).

Seven (44%) out of 16 and seven (47%) out of 15 does in category four were found to be infected in the first and second sampling respectively. Five out of the nine does (56%) does in this category sampled during the third sampling were infected (Table 4.12). Despite the chi-test indication of some significant differences between some parities, logistic regression analysis showed otherwise (p<0.05) (Appendix 3).

# 4.7.5 Effect of housing on infection status

The housing floor type was classified as either slatted raised or earthen. In the first sampling 15/115 (44%) does housed in raised slatted floors were infected while in the second sampling 34/94 (36%) does sampled in this category were found to be infected. In the third sampling 23/69 (33%) does in the same category were also found to be infected (Table 4.13).

Twelve (80%) of the 15 does and 10/14 (70%) does housed on earthen floors were infected in the first sampling and second sampling respectively (Table 4.13). In the third sampling 7/8 (88%) does sampled in the same category were infected (Table 4.13). On logistic regression does housed on earthen floors had infection rates significantly

(P<0.02) higher than those reared in raised slatted pens the former being 9 times more prone to mastitis than the latter (Appendix 3, P<0.02, O.R=9).

Table 4.13: Proportions affected in the two types of housing.

Housing type	Proportions a	iffected	
	1st	2nd	3rd
Raised	51/115(44)*	34/94(36)*	23/69(33)*
Earthen	arthen 12/15(80) <sup>4</sup>		7/8(88) *

**Key:** \* \*; Values on the same column with different superscripts were found to be significantly (p<0.05) different from each other on chi-test.

# 4.7.6 Effect of stage of lactation on infection status

The does were grouped into three categories depending on the duration post kidding as follows;

1) Early lactation (3 days-3 months post partum), (2) Mid lactation (3 months -5 months post partum) and (3) late lactation (>5 months post partum).

Based on this classification, 20/46 (43%) does in early lactation were infected during the first sampling. In the second 16/41 (39%) and in the third sampling 6/27 (22%) does in this category were infected in one or both udder halves (Table 4.14).

In comparison 22/40 (55%) and 13/33 (39%) does in mid lactation in the first and second sampling, respectively, were found to be infected. In the third sampling eight (33%) out of the 24 does in mid lactation were infected (Table 4.14). In the third category, out of the 44 does sampled in first sampling, 21(48%) were found to be infected. In the same category, 17/34 (50%) and 16/26 (62%) does in second and third sampling respectively were infected (Table 4.14).

On logistic regression analysis, stage of lactation was found not to affect the infection status significantly (P<0.05) (Appendix 3).

Table 4.14: Proportions affected in the various stages of lactation.

Stage of lactation	Proportions a	ffected	
	1st sampling	2nd sampling	3rd sampling
1	20/46(43)**	16/41(39)*	9/27(33)*
2	22/40(55)**	13/33(39)*	8/24(33)*
3	21/44(48)**	17/34(50) **	16/26(61) **

**Key:** \*\*,; Values on the same column with different number of stars superscripts were significantly different from each other on chi-test.

# 4.7.7 Effect of milking hygiene on infection status

Based on the level of hygiene practiced at milking, 2 categories of milking hygiene were recognized. These were good hygiene (water and milking salve were used) and poor hygiene where only plain water was used (Table 4.15).

Table 4.15: Proportions affected in the two classes of milking hygiene.

Milking hygiene	Proportions affe	cted in the various sa	mpling
	1 st	2nd	3rd
Poor	26/47(55)*	23/42(60)*	17/30(56)*
Good	20/49(41)*	18/39(46)*	13/28(46)*

Key: \*\*; values on the same column with different superscripts were significantly (p<0.05) different from each other on chi-test.

In the does washed with plain water only, 23/47 (49%) and 15/42 (38%) does were found infected in the first and second sampling respectively. In the third sampling, 11 (37%) out of the 30 does were found to be infected in one or both of their udder halves (Table 4.15). In those that were milked hygienically, 20/49(41%) does were infected in the first sampling while 15/39 does were infected in the second sampling. In the third sampling 13/28 (46%) does sampled in this category were found to be infected (Table 4.15).

On logistic regression, does that were in the poor milking hygiene category had a significantly (p>0.02) higher risk of contracting intramammary infection than those which were in the good milking hygiene category. They were twice (O.R=2) as likely to contract mastitis than the former category (Appendix 3).

# 4.8 Drug sensitivity results

Drugs tested for sensitivity included gentamycin (Gen), streptomycin (Str), kanamycin (Kan), ampicillin (Amp), cotrimazole (Cot), tetracycline (Tet), chloramphenical (Chl), nalidixin (Nal), and sulpadimidine (Smx), nitrofurazone (Nit) (Table 4.16).

Table 4.16: Drug sensitivity results of the organisms isolated from the does.

	Organism	result	Smx	Chl	Cot	Tet	Amp	Gen	Kan	Str	Nit	Nal
	Cps <sub>n=16</sub>	†ve	1	2	6	11	13	12	8	3	0	1
		-ve	15	14	8	5	3	4	6	13	46	15
	Cns $_{n=22}$	†ve	1	8	4	21	19	19	8	10	3	4
		-ve	21	14	18	1	3	3	14	12	19	18
	Strept spp n=2	*ve	0	1	1	1	2	1	2	2	0	0
		-ve	2	1	1	1	0	1	0	0	2	2
	Actino spp <sub>n=6</sub>	†ve	0	3	1	4	3	5	2	1	0	0
		-ve	6	3	5	2	3	1	4	5	6	6
	Acine spp n=7	⁺ve	0	3	0	1	0	4	3	0	0	0
		-ve	7	4	7	6	7	3	4	7	7	7
	Micro spp <sub>n=20</sub>	⁺ve	0	11	5	16	18	13	5	14	3	2
	11	-ve	20	10	15	4	2	7	15	6	17	18
Ke	y: 've_ sensitive	-ve_Not	sensitive					4				

Cps- Coagulase positive Staphylococcus, Cns-Coagulase negative Staphylococcus, Strept -Streptococcus spp

Actino -Actinomyces spp, Acine -Acinetobactor spp, Micro -Micrococcus spp,

N=Total number of isolates tested

From the above results it is evident that *staphylococcus aureus* (cps) were highly, sensitive to ampicillin (81%) followed by gentamycin (75%) and tetracycline (68%). Coagulase negative staphylococci were also highly sensitive to the same drugs above 90%, 90%, and 100% ampicillin, gentamycin and tetracycline respectively. Similar sensitivity pattern was found for the other isolates

# 4.9 Economic importance of mastitis

Seventeen clinical cases had been reported over the previous one year and the survey revealed that economic losses occurred from a number of factors. These factors included the following: -

#### 4.9.1 Losses in milk

All seventeen mastitis cases reported from the field had a history of reduced milk production. In one case there was no milk produced from the affected half, which was subsequently lost permanently. Losses in milk production were also incurred through milk discarded due to either discoloration (yellowish or bloody), clots or withholding milk after treatment. In the latter case milk was discarded for 72 hours post treatment.

#### 4.9.2 Cost of treatment

From the field survey the treated cases responded after 1.5-2 tubes of infusions per half were used. In monetary terms this was estimated at Ksh 180/half. In some acute cases, systemic treatment was combined with intramammary treatment, which increased the cost, by a further Ksh 120/ animal. This figure is arrived on the basis of 1ml/ 10kg adult goat (40kg) given a

total of 4mls of Penicillin streptomycin (Norbrook,UK)<sup>R</sup> combination for 3 days @ Ksh. 10/ml. In some cases where samples had to be taken to the laboratory for further analysis the cost increased by Ksh.150/ half giving a total of Ksh 390/half. In unresponsive cases veterinary surgeons had to be consulted with at an extra charge of Ksh. 100/visit excluding the cost of new treatment regimen. This gives the total cost at least Ksh 490/half.

# 4.9.3 Herd replacement costs

In two of the cases reported in the field, the farmers were forced to cull the does due to chronic mastitis, which was unresponsive to treatment, and replace them with others at a cost. The culled does were sold for slaughter at "meat" prices of approximately Ksh. 60/kg live weight or Ksh 2500/goat as compared to the replacement cost of Ksh 6,000 per milking doe. This reflected a loss of Ksh. 7,000 for the two does culled.

# 4.9.4 Death of does and poor growth of kids

One of the farmers reported death of their doe after the animal developed severe toxic mastitis. Four other farmers reported that does which exhibited severe pain, became anorexic and lost weight during the course of the disease. This resulted in poor growth of kids as the affected does could not suckle them due to pain or a drop in milk production.

#### 5.0 DISCUSSION AND CONCLUSIONS

The prevalence of udder abnormalities on the studied goats was low (10%). Most (6%) were of the hereditary type and were mainly found on both halves. Mary and Michael (1977) recorded similar abnormalities in America.

The presence of bacteria in samples was observed to elevate the CMT scores and DLC counts in all the samples. The magnitude of elevation however, depended on the organisms isolated with *S. aureus* resulting in the highest CMT scores and the highest DLC counts. The present investigation reveals results similar to what has been observed in cows and ewes (Stefanakis *et al.*, 1995). These researchers found that the presence of bacteria resulted in higher cell counts both by CMT and direct cell counts. Dulin *et al.*, (1983); Hunter, (1984); and Boscos *et al.*, (1996) also found elevated cell counts in bacteriologically positive goat milk samples and the degree of elevation depended on the type of organism isolated.

Unlike Boscos *et al.*, (1996), this study did not establish any significant (P<0.05) differences in the mean CMT scores and DLC counts of different parities or stages of lactation.

The prevalence of infected does varied significantly (P< 0.05) from group to group (range 22%-76%). The variation in the prevalence of the disease may be explained partly by the difference in geographical locations. Groups in the same divisions (2,3,4), (5 and 6), 1, and 7 had similar disease pattern probably due to the similarity in the prevailing climatic conditions. Differences in the management practices and advice given by the various extension coordinators may have also influenced the differences in the prevalence rates. Mastitis prevalence rates in the flocks was found to vary with the period that the particular groups had been participating in the project with those groups which had been with the project longest (

groups 2,3,4) reporting higher prevalence rates (76%, 76%, 54% respectively). Several factors may account for this situation including increasing parity, milk production and average age of the does. It is imperative that farmers are trained on how to detect and control mastitis along side the upgrading program to reduce the high mastitis prevalence rate. One group (7) that had over 50% of the does suckling the kids without being milked for home consumption, also had very high prevalence rate (65%) probably due to there being little or no attention given to the udder health when the does are not being milked. There is a need therefore to tell farmers to continue examining the udder at regular intervals even when not milking the doe for home use. While the prevalence rates observed in this study are higher than those reported by Contreras *et al.*, (1995) in Spain, they are not alarming as they are similar to those observed by other researchers in Britain (Manser, 1986), America (East, Birnie and Farver, 1987) and Greece (Boscos *et al.*, 1996).

The half infection rate (39%) was significantly (p<0.03) lower than the doe infection rate (58%), which indicated that most infections affected single. There was a trend of left halves being more affected than right halves though not significantly. These results agree with those reported by Contreras *et al.*, (1995) in Spain and Boscos *et al.*, (1996) in Greece who found that the halve infection rate was significantly lower than the doe infection rates. In particular, Boscos *et al.*, (1996) found a significant hiher prevalence of bacteria in the left mammary gland than the right.

In agreement with Dulin *et al.*, (1983), the present study could not establish any association between the stage of lactation and prevalence of intramammary infections. This contrasts with the findings of East *et al.*, (1987) who found an association between the prevalence of intramammary infections and the stage of lactation.

Similar to the findings of East *et al.*, (1987), the present study did not find any association of the age (which can be deduced from the number of parities) with the infection status. This may have been a reflection of the sample size rather than a true pattern in the present study. This opinion is supported by the findings of Boscos *et al.*, (1996) and Dulin *et al.*, (1983) who found an association between the infection status and the doe parities.

Results of this study contrasts with the findings of East *et al.*, (1987) who concluded that there was an association between some particular breeds (Nubian) and high prevalence of mastitis. According to these workers, this association was not evident in Saanen, Toggenburgs and Alpines. In the present study there was no association between the breed (German alpine and local crosses) and the prevalence of mastitis. Thus it is possible that the association observed by East *et al.*, (1987) may have been a reflection of genetic predisposition which had been reported earlier by other workers (Ai rawi, Pollack and Laben, 1979).

In the present study, an association was observed between the type of doe (milking/suckling) and the infection status. Suckling does were found to have significantly (P>0.02) higher prevalence of intramammary infection than the milked does. This association could be explained by the fact that suckling does are rarely examined and with increasing milk production, the kids may not be able to empty the glands fully. In addition, infectious agents on the surface of the udder/teats may also gain entry through injuries inflicted by the suckling kids.

This study revealed a trend of increased prevalence rate of mastitis with increase in litter size. The increase was however not significant (P>0.05) and may have been a reflection of the very low sample size of does with litter size of more than three kids. Large litter size would be expected to increase the susceptibility of the udder to infection as it would lead to excessive stress to the mammary gland (Boscos *et al.*, 1996).

Poor housing and milking hygiene were highly associated (P<0.02) with the infection status in this study with does that were housed in raised floors having significantly (P<0.05) lower rates than those housed on earthen ones. This may be explained by the fact that dirty wet beddings, which were a common finding on earthen floors, tend to harbor a variety of infectious agents, which contaminate udders and teats. Wet beddings also serve as a nutritious media in which pathogenic organisms thrive and multiply and therefore serve as multiplication media for these organisms (Samuel, 1977; Jain, 1979). The situation may be compounded by poor milking hygiene which on its own significantly (P < 0.02) affected the infection status. These results reflect a similar influence of housing and milking hygiene on mastitis in dairy cattle. Most of the organisms associated with mastitis in dairy animals and especially in the goat are found freely in the environment (Blood et al., 1989). Of particular importance is the Staphylococcus spp which is found in large numbers on the human skin and on the skin of the goats and is also the most important pathogen in goat mastitis (Manser, 1986; Guss, 1992; Shearer, 1992; Maina et al., 1993; Munyua, 1998). In cases, therefore, where milking hygiene is poor high levels of subclinical infections would be expected.

The etiological agents associated with caprine intramammary infections are several ranging from bacterial, mycoplasma and fungi (Egwu *et al.*, 1993). In this study, Staphylococcus *spp* were the most prevalent (78%) followed by *Micrococcus* spp (16%). Among the Staphylococcus species coagulase negative strains were the most prevalent

(71%). Other organisms isolated included *Actinomyces* spp (2%), *Acinetobactor* spp (1%), and *Streptococcus* spp (1%). No coliforms or yeasts were detected in this study. The pattern of isolation of etiological agents in this study was similar to those of Contreras *et al.*, (1995) and Boscos *et al.*, (1996) but different from those reported by Ameh *et al.*, (1993) in Nigerian goats. These latter groups of workers reported comparatively higher prevalence of *Streptococcus* spp and *Actinomyces* spp than in this study. This may however, be as a result of presence of infected cattle around the environment of the does rather than the importance of the latter organisms in goat mastitis (Blood *et al.*, 1989).

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It is evident that intramammary infection mostly due to *Staphylococcus* spp is widespread in the studied region and although this form of mastitis may not be obviously important initially, it poses a great danger to dairy goat farmers. This is mainly due to its insidious nature that usually precedes the clinical form, its long duration, reduction in milk production and adverse effects on the milk quality. Of utmost importance is the fact that the affected doe serves as a reservoir of *Staphylococcus* spp that may be spread to other animals within the flocks (Shearer, 1992).

The economic losses arising from dairy goat mastitis in the studied areas are enormous and can make the whole enterprise unprofitable if no intervention measures are put in place. In the present study similar losses as those reported in the cattle including losses in milk, death of does, cost of therapy and laboratory services, and discarded milk were observed (Janzen, 1970; Njenga *et al.*, 1987).

This study revealed that CMT had high sensitivity (92%) and specificity (87%) based on leucocyte count as the true indicator of infection. In comparison bacteriology was found

to have very low sensitivity leading to the conclusion that the mere isolation of microorganisms does not necessarily indicate an infection. A CMT of 2 would therefore be regarded as suspicious in any routine survey of mastitis in the field.

The present study also revealed that most of the organisms associated with dairy goat mastitis were highly sensitive to ampicillin, gentamycin, tetracycline and also streptomycin. Blood *et al.*, (1989) records a similar sensitivity pattern wich means that these drugs are still effective in control of this disease in dairy goats. Most of the coagulase positive *staphylococcus* (81%) which are the main organisms associated with dairy goat mastitis were sensitive to ampicillin and thus this drug can be very useful in treatment of clinical cases. Gentamycin and tetracycline had also high sensitivity i.e. 75% and 69, % respectively.

Based on the findings of the present study, it was concluded that dairy goat farmers in Kenya should institute various mastitis control and prevention strategies. These measures include raised floors housing system for the milking does, dehorning and foot trimming on regular basis to avoid injury to each other, draining pens, exercise yards and maintenance of well ventilated pens all of which are known to predispose animals to mastitis (Shearer, 1992). Good milking procedures and hygiene should be emphasized including routine use of a teat and udder disinfectant (e.g iodophor®) and drying the udder and teats with a dry towel. It is also important that milkers are advised on their own personal cleanliness. This includes washing of their hands in between the does to avoid spreading infectious agents from one doe to another. Milkers should also ensure that their hands are properly dried before milking and use proper milking technique (squeezing method rather than stripping). Goats are also known to appreciate being gently handled during milking, resulting in easy milk letdown and therefore ease at

milking. Irregular milking times, overmilking and inadequate preparation all result in increased stress to the mammary gland and should therefore be avoided as it reduces the immunity of the gland (Guss, 1992). It would also be advisable to have teat dips applied after every milking to reduce colonization of bacteria at the teat orifice and therefore reduce the incidence of new intramammary infections (Guss, 1992). The coordinators in this present area should be well informed on the management practices required in a dairy goat production set up so that they can advise the farmers appropriately.

Farmers should be advised to have infected does treated as soon as practical to avoid the losses associated with chronic cases. They should also be advised to have infected does milked last and isolated from healthy does. In cases of unresponsive does the farmers should be advised to cull the animals since these serve as reservoirs of infections in the flock. Farmers need to be advised on factors to consider when selecting does for milk production including long pendulous udders, genetic predisposition and poor teat placement (e.g. facing downwards instead of tilting forwards). Flocks should be regularly monitored using the CMT for cases of subclinical infection as this can help in detecting early cases before they become clinical. It would also be advisable for farmers to have their does treated at drying off with a suitable dry cow intramammary infusion, at the rate of half a bovine tube per teat.

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# 7.0 APPENDICES

# Appendix 1. MASTITIS DATA COLLECTION SHEET

# FIELD DATA SHEET

Date			
Name of farmer	Group I D		
Division			
Doe /name/No	Date fo Kidding		
Breed(local,F1, R1, R2)			
No of kids	Parity no		
Type of grazing(zero grazing, Padd			
Type of housing(raised, earthen)_	Level o	f hygiene(Good,	fair,
poor)			
Size of the pen			
No of does/pen	Frequency of emptying pen_		
Milking procedure			
Milking hygiene(poor,good)			
			ber.
Physical examination results			
i)Body condition score(poor, fair, go			
Temp Heart rate	Respiratory rate		
ii)Udder examination/teats			
Physical defects/injuries			
Consistency			
Temperature		4	

# Production

Avearage milk production/day\_\_\_\_\_

Purpose of milk

a)Kid consumption b)Home consumption c)Both

# Appendix 2. RETROSPECTIVE MASTITIS DATA SHEET

# FIELD DATA SHEET

District					
Division					
Group I D					
Have you had any case of mastiti	is in the l	est one vear?			
	15 111 1110 16	ist one year:			
a)Yes					
b)No					
If yes, how many?					
ii) What were the signs?					
a)					
b)					
c)					
d)					
e)					
f)					
iii) Were any laboratory samples ta	ken?				
If yes, how much was charged on e	each samp	ole?	_Ksh/sample		
iv) What treatment	was	adminstered	and	for	how
long?		-			
v)Have any doe (s ) been culled du	e to mast	itis?			
vi)What are the common mastitis	s control	programmes be	ing instituted	in the	farms?

# **Appendix 3. LOGISTIC REGRESSION RESULTS**

STATISTIX 4.0

# UNWEIGHTED LOGISTIC REGRESSION OF ANYPOS

PREDICTOR						
VARIABLES	COEFFIC	CIENT	STD	ERROR	COEF/SE	E P
CONSTANT	0.42379	0.770	01	0.55	0.5821	
BREED	-0.73744	0.548	321	-1.35	0.1786	
HOUS	2.33827	1.1114	49	2.10	0.0354	
KIDS1	-0.42805	0.474	33	-0.90	0.3668	
LCTST1	0.19866	0.6282	24	0.32	0.7518	
LCTST2	-0.05670	0.519	87	-0.11	0.9132	
MLKHYG	0.95596	0.464	58	2.06	0.0396	
PARITY NO	-0.65433	0.50	555	-1.29	0.1956	
PURP2	0.00453	0.663	21	0.01	0.9945	

DEVIANCE 115.34 P-VALUE 0.0159

DEGREES OF FREEDOM 85

CASES INCLUDED 94 MISSING CASES 36

#### STATISTIX 4.0

# UNWEIGHTED LOGISTIC REGRESSION OF ANYPOS

PREDICTOR					
VARIABLES	COEFFI	CIENT	STD ERROR	COEF/SE	E P
CONSTANT	0.42713	0.5961	1 0.72	0.4737	
BREED	-0.73798	0.5425	6 -1.36	0.1738	
HOUS	2.33885	1.1081	4 2.11	0.0348	
KIDS1	-0.42772	0.4718	0 -0.91	0.3646	
LCTST1	0.19670	0.5594	5 0.35	0.7251	
LCTST2	-0.05675	0.5198	3 -0.11	0.9131	
MLKHYG	0.95654	0.4569	1 2.09	0.0363	
PARITY NO	-0.65349	0.4902	.5 -1.33	0.1825	

DEVIANCE 115.34 P-VALUE 0.0191

DEGREES OF FREEDOM 86

#### STATISTIX 4.0

### UNWEIGHTED LOGISTIC REGRESSION OF ANYPOS

#### **PREDICTOR**

VARIABLES	COEFFICI	ENT S	STD ERR	OR	COEF/SE		P
CONSTANT	0.47755		0.53533		0.89		0.3724
BREED	-0.78887	0.536	579 -	1.47	0.14	17	
HOUS	2.41273	1.104	76	2.18	0.02	90	
KIDS1	-0.46501	0.468	78 -	0.99	0.32	212	
MLKHYG	0.97403	0.4522	28	2.15	0.03	13	
PARITY_NO	-0.66278	0.4912	3	-1.35	0.1	773	

DEVIANCE 116.33 P-VALUE 0.0275

DEGREES OF FREEDOM 89

CASES INCLUDED 95 MISSING CASES 35

STATISTIX 4.0

#### UNWEIGHTED LOGISTIC REGRESSION OF ANYPOS

#### **PREDICTOR**

VARIABLES	COEFFICIENT	STD ERROR	COEF/S	E P
CONSTANT	0.15734	0.42383	0.37	0.7105
BREED	-0.67305	0.51713	-1.30	0.1931
HOUS	2.25892	1.08609	2.08	0.0375
MLKHYG	0.96714	0.44877	2.16	0.0312
PARITY_NO	-0.63012	0.48622	-1.30	0.1950

DEVIANCE 117.32 P-VALUE 0.0281

DEGREES OF FREEDOM 90

CASES INCLUDED 95 MISSING CASES 35

STATISTIX 40

# UNWEIGHTED LOGISTIC REGRESSION OF ANYPOS PREDICTOR

VARIABLES COEFFICIENT STD ERROR COEF/SE P

CONSTANT -0.18222 0.33096 -0.55 0.5819

BREED -0.41888 0.47079 -0.89 0.3736 HOUS 2.27165 1.08701 2.09 0.0366

MLKHYG 0.91276 0.44121 2.07 0.0386

DEVIANCE 119.04 P-VALUE 0.0259

DEGREES OF FREEDOM 91

#### CASES INCLUDED 95 MISSING CASES 35

### STATISTIX 4.0

### LOGISTIC REGRESSION ODDS RATIOS FOR ANYPOS

 PREDICTOR
 95% C.I.

 VARIABLES
 LOWER LICMT
 ODDS RATIO
 UPPER LICMT

 BREED
 0.26
 0.66
 1.66

 HOUS
 1.15
 9.70
 81.63

 MLKHYG
 1.05
 2.49
 5.92

#### STATISTIX 4.0

# VARIANCE-COVARIANCE MATRIX FOR COEFFICIENTS

CONSTANT BREED HOUS MLKHYG CONSTANT 0.10953 BREED -0.06212 0.22164

HOUS -0.03993 -0.08135 1.18160

MLKHYG -0.08497 -0.01861 0.03409 0.19467