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# STUDIES ON MASTITIS IN GOATS



BY  
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## THESIS

Submitted in partial fulfilment of the  
requirement for the degree

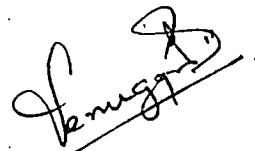
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**1978**

DECLARATION

I hereby declare that this thesis entitled 'STUDIES ON MASTITIS IN GOATS' is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title of any other University or Society.

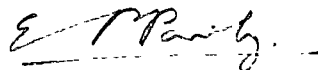


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CERTIFICATE

Certified that this thesis entitled 'STUDIES ON MASTITIS IN GOATS' is a record of research work done independently by Sri. Venugopal, K. under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to him.



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# INTRODUCTION

## INTRODUCTION

The domestic goat occupies an important position among the dairy animals. Exploitation of goats as a source of milk for human beings dates from antiquity. India stands first among the countries of the world in goat population (Food and Agriculture Organisation, 1972). According to the Livestock Census (1972), there are about 63 million goats in the country constituting about 19 per cent of the total world goat population. The report of the National Commission on Agriculture (1976) reveals that during 1971-72, goats produced about 6.75 lakh tonnes of milk constituting about three per cent of the total milk production of the country.

With the modern methods of selective breeding, better feeding and efficient management, an average doe producing only about 50-60 kg of milk per lactation, is gradually changing to a high milk producer. However, with this high production, there has been an increase in the prevalence and severity of the diseases of the udder. The importance of the diseases of the udder in dairy industry needs no special mention. The highly specialised mammary tissue is susceptible to abnormal conditions and infections by virtue of its location and activity. Considering the importance of udder diseases, it has become imperative to evolve suitable methods of disease diagnosis, therapy and control, to prevent loss to the farmer due to mortality and lowered production.



Mastitis is a disease complex having different causes, different degrees of intensity, with variations in duration and residual effects (Schalm and Woods, 1953). There is a change in the quantity and quality of milk. The disease in goats is more prone to become gangrenous than in cattle, which may even endanger the life of the animal.

Since goat's milk is consumed raw, especially in rural areas, the condition of the udder is particularly important from the public health point of view. The public health significance of goat mastitis is emphasized by the report of fatal staphylococcal intoxication from goat's milk (Need et al. 1943), excretion of Tuberculous organism (Mohan, 1950) and Brucella melitensis (Mathur, 1967) through goat's milk.

During the past few decades considerable work has been done on different aspects of bovine mastitis in India. Even though mastitis in goats is an important problem, the research work done is scanty in our country. In Kerala, much work has not been done so far on mastitis of goats.

There are numerous reports on the various aspects of somatic cells in milk and their importance in bovine mastitis. However, considerable knowledge is lacking in the number of cells in the normal and abnormal milk, their importance in disease diagnosis and correlation with California mastitis test (CMT) pertaining to caprine species.

Bacterial resistance to antibiotics is the principal obstacle to their successful therapeutic use (World Health Organisation, 1961). Due to the indiscriminate use of antibiotics for mastitis and other bacterial diseases both in human and Veterinary medicine many antibiotic resistant strains have emerged. This necessitates the choosing of the most effective antibiotic in a particular infection. In vitro antibiotic sensitivity test provide a very useful tool for assessing the possible effectiveness of the antibiotics against a particular micro-organism. Unless the results of antibiotic sensitivity testing are not taken into account, the treatment becomes empirical and often fails. The antibiotic sensitivity test has special application in treating cases of mastitis.

The present investigation is directed towards the study of following aspects:

1. To gauge the incidence of clinical and subclinical mastitis in goats.
2. To identify the causative agents involved in goat mastitis and to find out whether there is any correlation between the agents involved and the symptoms.
3. To evaluate the somatic cell count in milk samples of goats, in relation to California Mastitis Test, Whiteside Test and Teepol Mastitis Test.

4. To assess the sensitivity of bacterial isolates against chemotherapeutic agents and employ the results obtained for the treatment wherever possible.

# REVIEW OF LITERATURE

## REVIEW OF LITERATURE

Published literature on various aspects of goat mastitis are relatively few in number. Brief descriptions of the disease have been given in many text books (Little and Plastringe, 1946; Heidrich and Renk, 1967; Jubb and Kennedy, 1970; Schalm et al. 1971).

### Incidence

Earliest report on goat mastitis was by Nocard and Mollereau in 1887, and they were able to produce mastitis in a goat by intramammary inoculation of a culture of Streptococci of bovine origin. There were reports of outbreaks of caprine mastitis in 1911 with 12 to 25 per cent mortality in South West Africa and in 1935 in Italy where 62.5 per cent of the goats were affected (Heidrich and Renk, 1967).

In India, the first report of mastitis among goats would appear to be that of Dawa in 1940, who reported two outbreaks of gangrenous mastitis and later Gopalakrishnan (1940) reported two cases of caprine mastitis from Uttar Pradesh. Yakub (1942) described an outbreak of mastitis in goats in the Government Livestock Farm, Hissar, in which 98 animals developed the disease and 22 of them died. Bryan (1942) isolated bacterial organisms from 15 goats out of 619 examined. Viswanathan (1943) reported cases of goat mastitis from Malabar district. Chatupale (1948-49)

noticed mastitis in 30 among 120 goats from Daroda. Mohan (1950) was able to isolate Tuberculous organisms from the milk of two goats. Mukherjee and Das (1957) were able to isolate pathogenic bacteria from 94.5 per cent of the samples of milk collected from clinical cases of caprine mastitis in West Bengal.

The incidence of clinical and subclinical mastitis among goats at the Indigenous Goat Breeding Unit, Hissar were 9.4 per cent and 45.5 per cent respectively. Out of the 100 halves affected with clinical mastitis 46.3 per cent involved the left half and 53.7 per cent right (Kalra et al. 1962).

Out of 31,200 sheep and goats in Nicosia and Kyrena districts in Cyprus, 3,226 had mastitis of which 2818 were gangrenous. This caused death of 1135, loss of one half in 1346 and loss of both halves in 337 animals (Petris, 1963). Farrag and Oof (1966) noticed 22.0 per cent incidence of mastitis amongst a flock of 140 goats in U.A.R. In Bulgaria, Bozhilov et al. (1967) reported among 487 goats, 94 cases of gangrenous mastitis of which 23 died. Bacteriological examination of 150 milk samples from normal goats revealed microorganisms in 29.3 per cent of the samples (Panduranga Rao and Seetharaman, 1967). Ibrahim (1968) reported the isolation of pathogenic bacteria from 119 clinical and 175 subclinical cases of goat mastitis. Bozhilov et al. (1970) reported the isolation

of 141 strains of bacteria from 187 samples of udder tissue or milk collected from dead, sick or convalescent goats. In a French study involving 463 goats, eight per cent harboured pathogenic bacteria, 53 per cent had non-pathogenic organisms and remaining samples gave negative results (Roguinaky et al. 1971). Rosses (1972) isolated bacterial organisms from 59 out of 198 goat milk samples, although only seven goats showed clinical mastitis. Nag (1975) examined nine milk samples from cases of mastitis in goats and isolated bacterial organisms from seven cases. Mahendranath (1976) reported an incidence of 14.3 per cent of goat mastitis in Hyderabad. Roguinaky (1977) could isolate bacterial organisms from 61.8 per cent of sub-clinical cases and 86 per cent of clinical cases of mastitis in goats.

### Etiology

All organisms which cause mastitis in cattle can produce mastitis in goats also (Heidrich and Renk, 1967). Jubb and Kennedy (1970) reported more than 50 species of bacteria and 20 species of yeast-like fungi as the etiological agents of mastitis.

An organism tentatively types as Micrococcus caseolyticus was isolated from a case of goat mastitis which on intramammary inoculation into goats produced death (Anon, 1929-54). Kaplan (1944) considered Staphylococcus aureus as the main cause of mastitis in goats in U.S.A. Out of 109 samples of goat milk,

Stableforth (1949) could isolate Staphylococci, Streptococci and Coliforms from 34, 10 and 15 cases respectively.

Derbyshire (1958) artificially produced mastitis in goats by inoculation of viable cultures of Staph. aureus. Mukherjee and Lahiri (1960) in a study to note the bacterial flora of normal udders of healthy goats, found that 49 per cent of lactiferous sinuses and 25 per cent of mammary glands harboured Staphylococci or Streptococci. Of the Staphylococci, ten per cent was formed of Staph. aureus and the remaining, Staph. albus. Kalra et al. (1962) found that the chief etiological agents of clinical mastitis in goats were Staphylococci (80%) and Streptococci (16%). In subclinical infections, these organisms occurred in 58 and 33 per cent cases respectively.

In an extensive study conducted by Butozan and Mihajlovic (1963) on two lakhs of goats in Yugoslavia, Staph. aureus was involved in 81 per cent of gangrenous mastitis recorded. Petric (1963) showed that gangrenous mastitis was mainly due to Staph. albus although Escherichia coli and Streptococci were important. The organisms involved in non-gangrenous mastitis were Diplococci, Staphylococci, Streptococci and Corynebacterium pyogenes.

Inoculation of Staph. aureus strain S 63231 into the mammary gland of goats resulted in mastitis varying in intensity from mild inflammation to acute gangrenous type which caused death of four goats (Fujikura, 1966).



Bozhilov et al. (1967) gave a report of gangrenous mastitis in Bulgaria caused by Staph. aureus. Panduranga Rao and Seetharaman (1967) isolated 105 Staphylococci including 12 coagulase positive ones from the milk of healthy udders of goats. Bajleri (1968) isolated 30 strains of Staphylococci (including 22 Staph. aureus), one each of Streptococci and Esch. coli from cows and goats with mastitis. Ibrahim (1968) noted the percentage of incidence of various organisms in clinical and subclinical cases of mastitis in goats as Staph. aureus (33.6 and 36.6), coagulase negative Staphylococcus (3.4 and 32), Corynebacteria (10.1 and 8), Coliforms (5.9 and 6.3), Streptococci (5.9 and 3.4), Mycoplasma (10.1 and 0) and mixed infections (23.5 and 12.7).

Bozhilov (1970) made a study to note the etiology of goat mastitis and found that 80.9 per cent of the isolates were Staphylococci. Out of these, 49.1 per cent were albus type, 39.5 aureus and 11.4 citreus. In the subclinical infection, Staphylococci formed 72.6 per cent. Of these, 65.9 per cent were albus, 32.6 per cent aureus and 1.5 per cent citreus. In mixed infections, Staphylococci were isolated along with Esch. coli from three cases, with Diplococcus lanceolatus from nine cases and with Streptococcus dysgalactiae from eight cases.

Rosses (1972) isolated 35 strains of Staph. aureus, 12 strains of Staph. epidermidis and 12 strains of Micrococci from 198 goat milk samples.

Plommet (1974) reported that non-haemolytic Staphylococci are not pathogenic and hence do not often cause obvious clinical mastitis. But Guss (1975) noted that they may cause severe irritation to the udder tissue and such udders reveal swelling and hypersensitivity.

Nag (1975) examined nine cases of goat mastitis and isolated three strains of Staph. aureus along with two strains each of Streptococci and Corynebacterium. Lalithakunjamma (1976) in her study of pathological conditions of the mammary glands, isolated Staphylococci in majority of clinical and subclinical cases of goat mastitis. Streptococci and Coliforms were also isolated from some cases.

Rogunsky (1977) stated that haemolytic Staph. aureus was the most potent pathogen in many cases of goat mastitis. On a study on 15 caprine strains of Staph. aureus, he was able to show that they belong to the biotype C along with bovine and ovine strains but are serologically distinct.

The other organism important in mastitis is Streptococcus. The common species are Str. agalactiae, Str. uberis and Str. dysgalactiae.

Bryan (1942) examined 619 goat milk samples and he could isolate Str. agalactiae (D-Lancefield) from ten goats of which nine had chronic mastitis and five goats revealed Staph. aureus infection also. A further study conducted by him involving 300

goats revealed Streptococci in 2.3 per cent cases.

Pattison and Holman (1951) carried out extensive studies on experimental Streptococcal mastitis in goats and found that it simulated bovine Streptococcal mastitis. Pattison and Smith (1953) showed that histological changes in the mammary tissue of goats inoculated with Str. dysgalactiae resembled those produced by Str. agalactiae.

In a study to note the microflora of normal goat milk, Panduranga Rao and Setharaman (1967) examined 150 milk samples and isolated Str. agalactiae from four per cent of cases. In addition, they reported the occurrence of Achromobacter species, Alkaligenes, Escherichia, Pseudomonas, Paracolonobacterium and Brucella.

Among 50 goats, Neebakken (1975) noticed a herd problem of chronic mastitis caused by Str. zooepidemicus, showing symptoms of mammary atrophy, induration and abscessation.

According to Smith and Roguinsky (1977), the various species of Streptococci associated with mastitis in goats were Str. agalactiae, Str. uberis, Str. dysgalactiae and Str. zooepidemicus.

Naik (1948) indicated that C. pyogenes isolated from peritoneal exudates of goats were capable of producing mastitis.

Experimental inoculation of C. pyogenes into goat mammary gland showed varying reactions in lactating and non-lactating mammary glands. In the lactating gland, the reaction was only mild while in the non-lactating gland a progressive severe reaction was noticed (Jain and Sharma, 1964). The inoculation of C. pyogenes toxin into healthy non-lactating udder of goats showed heavy leukocytic infiltration into the mammary tissue (Jain, 1964).

In a survey of mastitis in goats, Farrag and Oof (1966) found that the organisms isolated in the order of prevalence were C. pyogenes and C. ovis, Staph. aureus, Str. agalactiae, Esch. coli and Pseudomonas aeruginosa. Ibrahim (1968) isolated Corynebacterium species from 10.1 per cent of clinical and eight per cent of subclinical mastitis. Mixed infections of Corynebacterium and Esch. coli were noticed in eight cases of goat mastitis by Bozhilov (1970).

Herak et al. (1961) reported a case of mastitis in a goat caused by Klebsiella species which did not respond to any treatment. Among the various micro-organisms isolated by Farrag and Oof (1966), Esch. coli was isolated from seven per cent of cases. Panduranga Rao and Seetharaman (1967) isolated Coliforms from milk of normal animals. An unusual outbreak of caprine mastitis in Mathura involving 15 lactating goats was reported by Adinarayanan and Singh (1968) caused by Klebsiella pneumoniae which responded to intramammary antibiotic treatment.

Ibrahim (1969), in a study to note the bacteria involved in goat mastitis, isolated Coliforms from 5.9 per cent of clinical and 6.3 per cent of subclinical mastitis.

Pseudomonas aeruginosa is also capable of producing mastitis in goats which may be acute, purulent, sometimes progressing to gangrene and death of the goat (Toizde, 1954; Lepper and Mathews, 1966). Pseudomonas was isolated from two per cent of milk of goats with clinical mastitis by Farrag and Gof (1966) and 1.3 per cent of subclinical cases by Panduranga Rao and Seetharaman (1967).

Brucella species though do not usually cause open clinical mastitis, they have been isolated from udders of goats. Heidrich and Renk (1967) reported the isolation of B. melitensis and B. abortus from the milk of apparently healthy goats without any evidence of clinical mastitis. In India, Mathur (1967) examined 11,647 milk samples of goats and isolated 39 strains of Brucella. Distinct signs of brucellosis were exhibited by the owners of goats from which B. melitensis were isolated.

The two species of Pasteurella, viz., P. multocida and P. hemolytica can produce mastitis which may be acute or chronic (Schalm et al. 1971). Bagadi and Razig (1976) reported clinical mastitis in goats caused by P. mastitidis with marked swelling of the udder and supramammary lymph nodes.

Tuberculous mastitis of the caprine udder has been reported by Mohan (1950). He could isolate Mycobacterium tuberculosis from milk of two goats. Tuberculosis of the caprine udder usually occurs during the course of generalised infections. After an initial edema, the udder becomes firm, irregular and painless lumps appear in the glandular tissue. Milk is initially watery, but later turns purulent (Heidrich and Renk, 1967).

Heidrich and Renk (1967) reported Mycoplasmal mastitis, a serious entity in some parts of the world. Ibrahim (1968) isolated 12 strains of Mycoplasma, along with one strain each of C. pseudotuberculosis and Nocardia species. Perreau et al. (1972) could isolate a Mycoplasma from a goat with mastitis and arthritis. The organism was typed as Mycoplasma mycoides var capri. Gourlay et al. (1973) produced experimental latent mastitis in lactating goats by inoculating T-mycoplasma isolated from urogenital tract into the mammary glands. Perreau (1974) reported that mycoplasma producing 'Contagious agalactiae' in goats has been reclassified as Mycoplasma capricolum.

Clinical mastitis was produced in three goats by intramammary inoculation of a local strain of Mycoplasma agalactiae subspecies bovis. The symptoms were fever, reduction in milk yield with purulent inflammation and necrosis of duct epithelium (Ojo and Ikede, 1976).

Nocardia asteroides, an organism found usually in the soil, water, air and herbage, may occasionally cause mastitis in animals. Dafaala and Charib (1958) reported a case of caprine mastitis caused by Nocardia asteroides. The udder was swollen, hard and milk was whey-like in consistency. Animal showed systemic reaction and had a temperature of 106°F. Sharma and Iyer (1974) made a study of the pathology of chronic lesions in goat mammary gland. They observed multiple nodular abscesses containing yellowish grey viscous pus replacing mammary parenchyma. Nocardia asteroides could be isolated from these lesions.

A variety of fungi can also produce mastitis in goats. Ainsworth and Austick (1955) reported a case of mycetoma in a goat's udder. The right half showed large plate like nodular lesions which was composed of regularly radiating hyphae. The organism resembled Aspergillus fumigatus. A case of mycotic mastitis in a goat characterized by nodular swellings was reported by Lepper (1964). The organism resembled A. fumigatus.

Galli and Socci (1969) experimentally produced mastitis in goats by inoculating Cryptococcus albidus or C. neoformans of bovine mastitis origin. There was hardening of the gland, reduction in milk yield and milk had a yellowish sticky appearance.

In a study to note the prevalence of mycotic mastitis in Haryana, Monga and Kalra (1971) found that 1.8 per cent of the diseased halves of goats examined had mycotic infection. The

fungi frequently isolated were C. neoformans, Candida albicans, C. krusei, C. parapsilosis and Saccharomyces species.

Satishkumar and Dhillon (1975) reported subclinical fungal mastitis among six per cent of goats. The important species were Candida, Cryptococcus, Geotrichum, Aspergillus, Rhodotorula glutinis, Penicillium, Rhizopus and Alternaria species.

### Cells in Milk

Literature available on the cytology of goat's milk and the role of cells, either in physiological processes or in mastitis, are relatively few.

Prescott and Breed (1910) introduced a new method of cell counting after spreading the milk on a glass slide.

Smith et al. (1954) reported that there was significant increase in neutrophil count and reduction in the milk yield in the unvaccinated control group of goats following intramammary challenge with Str. dysgalactiae strain 419.

On comparative study of milk of different species, Okada (1960) observed an average of 7,50,000 cells per ml of goat milk with 69.3 per cent neutrophils, 21.3 per cent lymphoid cells and 0.4 per cent epithelial cells.

In a single case of mycotic mastitis reported by Lepper (1964), the neutrophil counts were  $10^6$  per ml in both halves.



There was heavy leukocyte infiltration within six hours after inoculation of C. pyogenes culture into mammary glands of goats (Jain, 1964).

Cullen (1966) reported that in mastitis, in addition to increased cell count of milk, there was alteration in the lymphocyte-neutrophil ratio.

In experimental P. aeruginosa mastitis in goats, there was increase in milk cell count upto  $10^7$  per ml in three hours after inoculation (Lepper and Matthews, 1966).

All vaccinated experimental and unvaccinated control goats showed a cell count more than  $10^6$  following intramammary challenge with BB strain of Staph. aureus (Lepper, 1967).

Rahman and Range (1972) observed high leukocytic counts associated with hemolytic and coagulase positive Staphylococci in goat milk.

There were high milk cell counts following experimental subclinical mastitis produced by T-mycoplasma in goats (Gourlay et al. 1973).

Inoculation of Staphylococcal polytoxin into mammary glands of goats in four doses resulted in an increase in cell counts from 3.25 lakhs per ml to 9.95 lakhs, 6.81 lakhs, 5.87 lakhs and 17.86 lakhs after each inoculation (Benda and Rysanek, 1974).

The somatic cell counts of milk from cases of caprine mastitis caused by B. mastitidis were more than two million (Bagadi and Razig, 1976).

Somatic cell counts and their correlation  
with California Mastitis Test (CMT)

Schalm and Noorlander (1957) introduced the CMT, a new practical method to detect mastitis. CMT enables to produce a visible effect that can be scored numerically, with reference to somatic cells in milk. Many workers have worked out the correlation between CMT (and its modifications) and somatic cell counts in cows milk (Daniel et al. 1966; Luedcke et al. 1967; Sharma and Rajani, 1969; Bendse and Nair, 1970; Schalm et al. 1971).

Pattison et al. (1950) in a study of experimental Str. agalactiae infection in goats found that the average normal leukocytic counts of goats with Whiteside score zero were  $7.2 \times 10^3$  per ml. After infection with Str. agalactiae S-13, the Whiteside reaction were strongly positive for five days after which it subsided.

Fujikura (1966) during the studies on experimental Staphylococcal mastitis in goats found that leukocytic counts increased considerably as shown by modified CMT.

Ziv et al. (1968) made a study to note the effectiveness of CMT as a measure of somatic cells in ewe's milk. Samples

with CMT scores less than 1 had cell counts less than five lakhs and those above 1 had cell counts above five lakhs.

Schalm et al. (1971) subjected 140 goat milk samples to CMT and somatic cell count to study their correlation. The mean cell counts for CMT scores zero, trace, 1, 2 and 3 were 68,000, 2,68,000, 8,00,000, 25,60,000 and above 10,000,000 per ml.

Pilev (1973) studied the correlation of Fernberg test scores with somatic cell counts in subclinical mastitis in ewes. The somatic cell counts for test scores zero, 1+, 2+ and 3+ were less than 1 lakh, 1.47 lakhs, 3.02 lakhs and 17.63 lakhs per ml respectively.

#### Antibiotic Susceptibility

World Health Organization Expert Committee on Antibiotics (1961) has classified the antibiotic sensitivity testing into diffusion and dilution methods. The diffusion method has become the most accepted procedure because of the simplicity and rapidity (Anderson, 1970).

The literature available regarding the antibiotic sensitivity of the micro-organisms causing goat mastitis are very limited.

Ovejero et al. (1960) made a study on the bacteriology and antibiotic sensitivity of ten strains of Staphylococci isolated from cases of ovine and caprine mastitis. All the strains

were sensitive to Chloramphenicol, Tetracyclines and Erythromycin, while many were resistant to Streptomycin.

*Klebsiella* species isolated from goat mastitis were most sensitive to Streptomycin, followed by Terramycin, Chloromycetin, Chlorotetracyclines but were resistant to Penicillin and Sulphathiazol (Merak et al. 1961).

Krishnamurthy and Makholia (1963) found 27 per cent resistance to Penicillin out of 111 strains of different organisms isolated from domestic animals. Out of 49 isolates of Staphylococci of bovine udder 14.3 per cent were resistant to Penicillin.

Krzyzanowski et al. (1965) tested 200 strains of different organisms of bovine udder origin for antibiotic sensitivity. The resistance percentages were Penicillin (66.5), Neomycin (50), Erythromycin (48.4), Aureomycin (47.4), Tetracyclin (26.1), Terramycin (26) and Streptomycin (23). None were resistant to Chloramphenicol.

Swarbrick (1966) successfully used Erythromycin parenterally in acute or per-acute bovine mastitis caused by Streptococci or Staphylococci.

Farrag and Cof (1967) tested the effectiveness of various antibiotics on micro-organisms isolated from cases of bovine or caprine mastitis. Chloramphenicol and Tetracyclines gave the

best results. Penicillin and Erythromycin gave good inhibitory effects upon *Corynebacterium* species, Staphylococci and Streptococci. Streptomycin was effective on *Esch. coli* and *Pseudomonas*. In the study of Bejleri (1968) all Staphylococci and Streptococci isolated from cases of cattle and goat mastitis were sensitive to Penicillin and Tetracyclines but were resistant to Sulphathiazol.

In bovine mastitis, intramammary Erythromycin was very effective in eliminating *Staph. aureus* from 76.1 per cent of 67 quarters, *Str.agalactiae* from 92.3 per cent of 26 quarters and *Str. uberis* from 80.8 per cent of 9 quarters (Schultz, 1968).

Out of the one hundred strains of organisms of bovine udder origin, the number of strains that were resistant to various antibiotics were Penicillin (72), Erythromycin (69), Synermycin (62), Terramycin (42), Streptomycin (30) and Chloromycetin (8) (Ramachandrarao and Naidu, 1969).

Panduranga Rao et al. (1969) evaluated the antibiotic susceptibility of 82 isolates of Streptococci of bovine udder origin. Chloromycetin (100%), Penicillin (76.83%), Terramycin (56%), Streptomycin (42.6%) and Aureomycin (30.49%) were observed to be in the decreasing order of efficacy.

Spais and Giantzlis (1970) reported that Erythromycin or Spectinomycin when given parenterally for three days in

goats from two flocks reduced the incidence of 'Contagious agalactias' by 50 to 60 per cent.

Sharma et al. (1971) studied the in vitro sensitivity of *Staphylococcus*, *Streptococcus* and *Corynebacterium* isolated from bovine udder and their resistance to Penicillin were 33.3, 62.5 and 100 per cent respectively; but Tetracyclines were very effective in all cases.

Farzaliev et al. (1974) found that Chlorotetracyclines and Tetracyclines were most effective in 'Contagious agalactias' and confirmed the in vitro observations by effectively treating 1026 sheep and goats with these antibiotics.

The percentages of sensitivity to various antibiotics of 135 strains of mammary isolates were Penicillin (59.3), Furazolidon (76.2), Furaltadon (72.1), Oxytetracycline (65.2), Chloramphenicol (59.3), Chlorotetracycline (46.6), Neomycin (41.4) and Streptomycin (27) (Patra et al. 1974).

The resistance percentages of 621 strains of Staphylococcus aureus from animal sources were Penicillin (55), Streptomycin (25), Tetracycline (15), Methicillin (15), Neomycin (12.5), Chloramphenicol (7.5), Erythromycin (5) and Lincomycin (5) (Riberstein et al. 1974).

Staph. aureus strain isolated from a case of goat mastitis was sensitive to Chloramphenicol, Tetracycline, Neomycin,

Bacitracin and Streptomycin while it was resistant to Penicillin and Sulphonamide (Nag, 1975).

In the study by Jhala (1976) Neomycin and Chloramphenicol gave best results followed by Streptomycin, Terramycin, Bacitracin, Polymyxin B, and Erythromycin. All the organisms tested were Penicillin resistant.

Kohli (1975) reported that encouraging results could be obtained with the I.D.P.L. Mastitis formula (Indian Drugs and Pharmaceuticals Ltd.), containing Ampicillin, Grescofulvin and Prednisolone in goat mastitis. This drug was effective in completely curing mastitis in eight out of eleven goats.

## **MATERIALS AND METHODS**



## MATERIALS AND METHODS

The materials for the study were collected from—

- i) Kerala Agricultural University Goat Farm, Mannuthy;
- ii) The Goat Unit of the All India Co-ordinated Research Project on Goats for Milk Production, Mannuthy;
- iii) Veterinary Hospitals, Mannuthy and Trichur of the Kerala Agricultural University;
- iv) Government Veterinary Hospitals in and around Trichur.

A total of 435 milk samples from goats of different breeds and age group were examined during the present investigation. These included samples from 56 clinical cases and 379 apparently normal animals. Following tests were carried out:

- 1) Mastaid Test (Glaxo Laboratories)
- 2) Whiteside Test
- 3) Teepol Mastitis Test
- 4) Somatic cell counting
- 5) Cultural examination
- 6) Antibiotic Sensitivity Test

### Collection of milk samples

The milk samples were collected in sterile containers with aseptic precautions. For this the teats were washed well

with water and mopped dry with a clean cloth. The test tip was then cleansed with a swab dipped in 70 per cent alcohol. The first streams of milk were collected directly into the container, without touching the sides. During collection, the vial was canted to avoid, as far as possible, the entrance of dust, skin scales and hairs.

At the time of collecting the milk samples from clinical cases, the history of the case, nature of inflammation, condition of the animal, systemic reaction, if any, nature of milk etc., were noted.

#### Mastaid Test

The CMT reagent used consisted of an anionic surface-active agent and the indicator bromocresol purple. The reagent when mixed with mastitic milk produces a visible reaction that can be scored numerically. The reagent reacts with the Deoxy ribonucleic acid (DNA) released from the cells and causes the visible reaction thus determining the approximate number of leukocytes in milk.

Mastaid Test is a modification of CMT. The Mastaid reagent (Glaxo) used produces a visible reaction with mastitic milk.

The test was performed as follows:-

Approximately three ml of milk was collected in receptacles of the plastic paddle supplied with the testing solution.

Equal volume of the reagent was added and mixed well by slow circular movements for about ten seconds. The results were recorded as--

Negative	(-)	No precipitate
Trace	(T)	A slight slime forms with a tendency to disappear.
Weak	(+)	A distinct slime, but not a gel.
Distinct positive	(++)	Thick mixture with a precipitation and gel formation.
Strong positive	(+++)	A gel formed, causes the surface of the mixture to become convex.

During this study 373 samples of apparently normal milk and 47 mastitis milk from goats were subjected to mastid testing and the scores were noted.

#### Whiteside Test

In this test also the principle involved is the same as for CMT. The sodium hydroxide in the test reagent when mixed with milk causes the rupture of leukocytes and the DNA thus released combines with it forming a gel which is scored depending on the viscosity.

During this study, the modified whiteside test described by Murphy and Hansen (1941) was used. For performing the test, a glass plate, etched with vertical and horizontal lines, to provide 1.5 inch squares was used. About 5 drops of milk were

placed in the centre of each square and one drop of a four per cent sodium hydroxide solution was added. The mixture was then stirred vigorously with an applicator stick spreading it over a circular spot of about 3 cm diameter for about 20 seconds. The results were recorded as—

- (-) Mixture remains opaque and free of particles.
  - (±) No apparent reaction, but finely dispersed particles seen.
  - (+) A definite thickening occurs during stirring but do not adhere to the stick.
  - +) There is immediate thickening on stirring and finally separates into clear whey and thread like whorls.
  - (+++)
  - (++++)
- A tenacious mass forms immediately on stirring and adheres to the stick and finally separates into whey.
- A tenacious coagulum with no tendency to break down into whey.

Modified whiteside test was done on 103 apparently normal goat milk samples.

#### Teepol Mastitis Test

Because of the difficulty in the procurement of alkyl aryl sulfates and sulphonates of sodium and potassium, CMT could not be made use of regularly. Hence a modified CMT reagent was tried by Sharma and Rajani (1969) by substituting the alkyl aryl salts with the detergent 'Teepol' an easily

available shell product. The test reagent had the following composition:-

Sodium hydroxide	1.5 g
Teepol	0.5 ml
Bromthymol Blue	0.01 g
Distilled water	100 ml

The test was performed with the above reagent in plastic paddle. The procedure for doing the test and the interpretation of results were same as for Mastaid test.

This test was done on 103 apparently normal milk samples of goats.

#### Somatic cell counting

Somatic cells in milk represent an inflammatory process in the udder. Somatic cell counts of milk are quite satisfactory as a screening test for mastitis.

The method employed for making the cell counts was the one described by Prescott and Breed, 1910. Here 0.01 ml of milk from a well mixed sample was pipetted on to a clean grease-free glass slide and spread evenly over an area of  $1 \text{ cm}^2$  with the help of a square template of  $1 \text{ cm}^2$  area. Two such smears were made for each sample of milk. The smears were then dried in air.

The staining was done with modified Broadhurst-Paley stain (Schalm et al. 1971) which was prepared as follows:-

Dissolved 1.5 g of methylene blue in 250 ml of hot 70 per cent ethyl alcohol. Ten ml of saturated alcoholic basic fuchsin solution and 5 ml of aniline was added, while continuously shaking the solution and keeping it warm. To this mixture 15 ml of diluted sulphuric acid was added mixed well and filtered. To every 100 ml of the filtrate 50 ml of hot distilled water was added and shaken well. The filtered stain was kept in a glass stoppered bottle in a refrigerator.

For making the somatic cell counts the microscope was calibrated. For this, the diameter of the microscopic field through an oil immersion objective was measured with a stage micrometer. From this, the area of the field was calculated. The microscope used in the present study had a diameter of 0.016 cm and area  $1/5000 \text{ cm}^2$ . Since 0.01 ml of milk is spread over an area of  $1 \text{ cm}^2$ , the possible number of such fields would be 5000. The milk volume represented in each field would be  $1/5000 \times 1/100$  or  $1/500,000 \text{ ml}$ . On this basis each cell in a field when taken at random would be 500,000 cells/ml of milk. This was the microscopic factor and from this, working factor was calculated by dividing with the number of fields counted. In this study a total of 50 fields were counted and the working factor was 10,000. The total number of cells counted was

multiplied with this working factor to obtain the number of cells per ml of milk.

The dried smear was stained as follows:-

1. The slide was immersed in xylene for two minutes and dried.
2. Kept in 95 per cent ethyl alcohol for 2 to 5 minutes and dried.
3. Immersed in Broadhurst-Paley stain for 5 seconds, rinsed gently in water and dried.

Broadhurst-Paley staining method has been used as a triple step process consisting of defatting, fixing and staining. In a stained smear, milk solids are stained pink, polymorphonuclear leukocytes deep blue and bacteria deep or light blue.

For cell counting the smear was examined under oil immersion and cells in 25 fields of each smear were counted. The total number of cells in 50 fields were noted and multiplied with the working factor to obtain the total number of cells per ml of milk.

In the present study 351 milk samples from goats were subjected to somatic cell counting.

#### Cultural examination

Milk from both clinical and subclinical cases of mastitis were subjected to cultural examination. The detection of

subclinical mastitis was carried out by the mastaid reagent. The samples that were positive to this were subjected to cultural examination after incubating at 37°C for 24 hours. The incubated samples were streaked on blood agar plates for isolating bacterial organisms (Merchant and Paeker, 1971) and Sabouraud's dextrose agar for fungal organisms (Davies, 1957). The blood agar plates were incubated at 37°C for 24 to 48 hours and Sabouraud's dextrose agar at room temperature upto a period of one month. Pure cultures were made from the specific colonies on blood agar slants and identified by their morphological cultural and biochemical characters as described by Cowan and Steel (1974).

The typing of Staphylococci was done by observing the catalase reaction, pigmentation of the colony, coagulase test with rabbit plasma, acid production on mannitol salt agar and Voges-Proskauer reaction.

Cultural examination was also conducted on 37 milk samples of goat which gave a negative mastaid reaction, to ascertain whether they were bacteriologically sterile.

The milk samples from clinical cases also were subjected to cultural examination for the detection of bacterial and fungal organisms. The procedures followed were the same as in the case of subclinical mastitis.

During the present study 37 normal milk samples (Mastaid



negative), 158 samples from subclinical cases and 56 samples from clinical cases were examined.

### Antibiotic Sensitivity Test

World Health Organization Expert Committee on Antibiotics, 1961 has described two types of antibiotic sensitivity tests - the diffusion and dilution methods. The method employed for the present study was the filter paper disc agar diffusion method.

#### Preparation of the disc

The method described by Cruickshank et al. (1975) was followed. Paper discs having 6 mm diameter were punched from Whatman No.1 filter paper and dispensed in batches of 100 in clean cork stoppered vials, which were then sterilized by dry heat at 140°C for 60 minutes. Solutions of antibiotics were prepared so that 1 ml contains 100 times the amount of antibiotic required in the disc. To one vial of 100 discs, 1 ml of the solution was added mixed well and was assumed that each disc will contain approximately 0.01 ml. The vials were then stored in wet condition in the refrigerator.

In this study 58 bacterial isolates from clinical cases of mastitis of goats were tested for their antibiotic susceptibility. The antimicrobial agents tried were Ampicillin, Chloramphenicol, Erythromycin, Kanamycin, Nitrofurazon, Penicillin, Streptomycin, Sulphadiazin and Terramycin. The concentration of

the drug in each disc of different antibiotics were made according to the standards described by Blair et al. (1970).

With a view to obtain the results of the sensitivity testing for application in clinical cases, the test was first done on primary culture itself. For this, sterile swabs were well soaked in the milk and streaked over blood agar medium uniformly. It was allowed to dry for 30 minutes. The discs were then placed on the medium suitably spaced, with the help of flamed forceps, and gently pressed.

With pure cultures, the sensitivity testing was done on nutrient agar plates. For Streptococcus species, which required an enrichment medium, blood agar plate was used even for pure culture sensitivity testing. For doing the test with pure cultures, a 24 hour old broth culture prepared from the blood agar slant was used as the inoculum. The technique was same as for the primary culture.

The plates were incubated at 37°C for 24 hours and the results were read. The diameter of the visible zone of inhibition around the disc was measured with a pair of calipers, the diameter of the disc was also included in the measurement (Plate XIII). The findings were recorded and interpreted adopting the guidelines of Blair et al. (1970). (Appendix-2).

The results of the sensitivity testing, done on primary culture which were available the next day, were taken as the

guideline for treating the cases.

In addition, the udders from three cases, were examined for histological changes. These included two cases from which glands were removed and one from an animal dead of gangrenous mastitis. The specimens were dissected and examined for gross changes. Small pieces of tissues from different portions of the udder were collected and preserved in 10 per cent formalin. The tissues were then processed by the paraffin embedding method and sections five microns thick were taken. The sections were stained with haematoxylin and eosin for histopathological examination.

## RESULTS

## RESULTS

A total of 435 milk samples of goats were examined during the study. This included milk samples from 56 clinical cases and 379 apparently normal animals.

Bacterial organisms were isolated from all the clinical cases examined. The major pathogen isolated from clinical cases was Staph. aureus which constituted 52 per cent. The other pathogens in the order of prevalence were Str. agalactiae, Staph. epidermidis, C. pyogenes, Esch. coli, Ent. aerogenes, K. pneumoniae and P. aeruginosa. Mixed infections of Staph. aureus and Str. agalactiae were noticed in two cases. None of the samples revealed fungal organisms.

The results of the bacteriological examination of milk samples from clinical cases are given in Table 1.

The symptoms of mastitis in goats were highly variable. Staph. aureus in more than 90 per cent of the cases produced acute mastitis. Gangrenous changes of varying degree were noticed in eight acute cases. In these cases, there was toxæmia and severe systemic reaction with elevation of temperature to 40 to 41.5°C, rapid heart rate (100-120 per minute) laboured breathing, complete anorexia, profound depression, muscular weakness and animals were usually recumbent. Local reactions

in the udder included gross swelling and hardness, pain on palpation with edema of the region. The initial red colour of the skin changed to blue involving the teats and udder and the affected portions were cold to touch. A line of demarcation was seen separating the living from the dead tissue. The skin could be easily peeled off. Later, a blackish colour developed and the gangrenous tissue was moist. The secretion was a blood-stained serous fluid without flakes or clots. Out of the eight cases of gangrenous mastitis, five were fatal (Plates I and II).

Histologically, the tissue showed focal areas of necrosis, dense infiltration with neutrophils and plasma cells, causing extensive destruction of parenchyma and acinar tissue. In most of the acini, desquamated cells were seen filling the lumen. Blood vessels were severely engorged and many of them showed thrombosis. Isolated foci of suppuration were also evident scattered in the parenchyma. Numerous dense staining corpora amylacea were also seen (Plates III and IV).

Acute mastitis due to Staph. aureus in 19 cases were not gangrenous. In such cases, the affected half was enlarged, warm and painful to touch. Generally systemic reaction was not observed. But five animals showed slight reaction with temperature elevated to 40 to 42°C and partial anorexia. The secretion in most cases were watery, straw coloured and contained flakes and pus.

In one case there was extensive suppuration with abscess formation in the gland and pus was draining out through a fistula at the base of the test. Histologically, the tissue revealed multiple foci of suppuration, scattered in the acinar tissue. There was extensive interlobular fibrosis causing atrophy of the lobules. Focal areas showed structures resembling 'Pseudo-actino body' and corpora amylacea (Plate V).

Coagulase negative Staph. epidermidis isolated from nine per cent of the cases, produced subacute mastitis in majority of the cases (80%). Here the affected half showed slight hardness and warmth. The secretion was normal but the quantity was reduced. There was no accompanying systemic effects.

Mastitis due to Str.agalactiae was 14 per cent and was of chronic nature in 75 per cent of cases. In these cases, the affected half showed marked induration without any cardinal signs of inflammation (Plate VI). There was reduction in the milk yield and the milk in many cases was watery with clots. One case of Streptococcal mastitis was of acute nature with the udder hot and painful and milk straw coloured. The animal was recumbent.

The Coliform (Bach. coli, Ent. aerogenes and E. pneumoniae) mastitis was usually of an acute type without showing gangrenous changes. Systemic reactions and symptoms of toxæmia with temperature of 40 to 41.5°C, anorexia and muscular weakness were observed. Local reactions included enlargement of the half,

hardness and secretion was a scanty serous fluid. In the present study Coliform mastitis was recorded in 14 per cent of the cases.

Mastitis caused by C. pyogenes (5%) were usually of an acute suppurative type, with yellow purulent secretion. In one case, C. pyogenes produced gangrenous changes in the skin of the udder and teats, of both halves. The lesion on the left involved deeper tissues and the initial moist gangrene turned dry. The similar lesion on right half was more superficial and sloughed off after treatment (Plates VII, VIII, IX and X).

Microscopic examination of the dry scab of the skin showed focal areas of dermal necrosis and associated with this areas of acanthosis and hyperkeratosis were seen. There was deposition of melanin pigments in focal areas. Dermal vessels were tortuous and severely engorged (Plate XI).

The section of mammary gland from this case did not show much gross changes. But microscopically, multiple foci of supuration and necrosis were seen. Interstitial tissue was diffusely edematous and was infiltrated with neutrophils. Regenerated and desquamated cells were seen filling the lumen of the acini. Some of the acini had been converted into foci of supuration without any evidence of acinar tissue (Plate XII).

A single case of chronic mastitis by P. aeruginosa was also noticed.



The correlation between bacterial isolates and the symptoms of mastitis are given in Table 2.

Examination of 276 milk samples from apparently normal goats revealed 47 per cent incidence of subclinical mastitis. The results of the examination of these samples are incorporated in Table 3.

Staph. epidermidis was the major bacteria encountered in subclinical mastitis constituting 53 per cent; Staph. aureus (30%), Str.agalactiae (10%), C. pyogenes (5%) and Esch. coli (2%) were also isolated.

The percentage distribution of bacterial species in subclinical mastitis are shown in Fig. 1.

Of the 56 clinical cases, the right and left halves were affected in 76.8 per cent and 23.2 per cent respectively. In subclinical mastitis, 47.6 per cent involved the right half and 52.3 per cent the left half.

A total of 317 samples of goat's milk were subjected to CMT. The percentage distribution of CMT scores Negative, Trace, +, ++ and +++ were 33, 24, 30, 7 and 6 respectively (Table 4).

Of the 76 samples that were culturally negative, eight per cent gave a CMT score of +, 43 per cent Trace and 49 per cent negative. The milk samples from which Staph. aureus was isolated

gave a CMT score of + in 60 per cent, ++ in 16 per cent and +++ in 24 per cent. Staph. epidermidis gave Trace in 50 per cent, + in 44 per cent and ++ in six per cent. More than 30 per cent of milk samples, from which Str. agalactiae, C. pyogenes and Coliforms were isolated, gave scores more than +.

The results of the examination of milk by CMT and its correlation with bacterial isolates are given in Table 4.

A total of 276 samples were subjected to Mastaid test and the number of samples giving different scores were Negative (111), Trace (74), + (86) and ++ (5).

Of the 74 samples with Trace score, 41 were culturally positive. Only three of the 86 samples giving + score were culturally negative. Thus, Mastaid reagent proved to be 96.5 per cent efficient in detecting subclinical mastitis in goats.

In order to ascertain the normal range of cells in milk, 37 milk samples, which were both CMT and bacteriologically negative, were subjected to somatic cell counting. The mean counts of cells in the normal milk samples were 0.9 lakhs per ml. The corresponding mean somatic cell counts of milk samples with CMT scores Trace, +, ++ and +++ were 3.2, 6.5, 17.4 and 229.6 lakhs per ml. respectively.

Details of the somatic cell counting and their correlation with CMT scores are given in Table 5.

It may be mentioned that the milk sample of a goat with acute mastitis due to Int. aerogenes gave a CMT score of +++ and the corresponding somatic cell count was  $31.8 \times 10^6$  per ml (Plate XIV).

For detecting subclinical mastitis, comparative efficacy Mastaid Test, Whiteside Test and Teepol Mastitis Test were carried out on 103 milk samples from apparently normal goats. The cell counts were grouped into 3 ranges as (i) below 5 lakhs (ii) 5 to 10 lakhs and (iii) above 10 lakhs and the number of samples in each of these ranges giving the different scores were noted.

It was observed that all the three tests were equally efficient in negative samples (i.e. cell counts below 5 lakhs per ml). However, to detect subclinical cases (5-10 lakhs), Whiteside Test and Teepol Mastitis tests failed in 29 and 70 per cent of samples respectively, while Mastaid Test was 100 per cent efficacious. For detecting samples having cell counts of 10 lakhs and above, Mastaid Test was found to be efficient in 90 per cent, Whiteside Test in 54 per cent and Teepol Mastitis Test in 63 per cent.

Results of comparative efficacy of these tests are given in Table 6.

In vitro antibiotic sensitivity testing carried out on

50 bacterial strains isolated from clinical caprine mastitis, showed that Chloramphenicol was effective in 84 per cent of the cases. The percentage of efficacy of other antibiotics in the order were Erythromycin (78), Ampicillin (71), Streptomycin (69), Penicillin (67), Terramycin (67), Nitrofurazon (45), Kanamycin (19) and Sulphadiazin (16) (Fig. 2).

By noting the antibiotic sensitivity of individual bacteria it was found that Staph. aureus had lowest number of resistant isolates for Erythromycin and Chloramphenicol while highest resistance was encountered for Sulphadiazin, Nitrofurazon and Kanamycin. Moderate number of resistant strains existed for Terramycin, Penicillin, Ampicillin and Streptomycin.

Str. agalactiae and Staph. epidermidis showed very good susceptibility for Penicillin, Ampicillin, Erythromycin and Streptomycin, moderate for Chloramphenicol and Terramycin and least for Nitrofurazon, Kanamycin and Sulphadiazin.

C. pyogenes isolates were highly sensitive to Penicillin, Ampicillin, Chloramphenicol, moderately to Terramycin and least to other antibiotics.

Coliforms showed maximum sensitivity for Chloramphenicol, Sulphadiazin and Kanamycin, moderate sensitivity for Terramycin, Nitrofurazon and Streptomycin. E. aeruginosa strain isolated was susceptible to Chloramphenicol, Sulphadiazin and Kanamycin.

Details of the in vitro antibiotic sensitivity of current bacterial isolates are given in Table 7 and plotted in Fig. 3.

Treatment was carried out on the basis of in vitro drug sensitivity in 40 selected clinical cases presented to the clinics attached to the College. The isolates from the above cases included Staph. aureus (21), Staph. epidermidis (5), Str.agalactiae (5), C. pyogenes (2), Ent. aerogenes (3), Bach. coli (2), K. pneumoniae (1) and P. aeruginosa (1).

Of the 21 Staph. aureus isolates eight were sensitive to Penicillin. In these cases, Pendistrin-SH (Squibb)<sup>1</sup> was used in five and Dicrysticin-S (Squibb)<sup>2</sup> in three. The six Streptomycin sensitive isolates were treated with Pendistrin-SH and Dicrysticin-S in three cases each. Out of six Terramycin sensitive isolates, three were treated with Oxystecilin (Squibb)<sup>3</sup> and three with Mastalone (Pfizer)<sup>4</sup>. Out of the Chloramphenicol sensitive isolates, one was treated with Vetycotine(TCP)<sup>5</sup>.

Among the three animals that died during treatment, two had gangrenous mastitis with systemic reaction at the time of presentation. These animals had been treated with Pendistrin-SH and Vetycotine. The other goat, which died during treatment with Dicrysticin-S, was showing systemic reaction with toxæmia when it was presented. One case turned chronic and in another the owner did not turn up.

Staph. epidermidis in three cases showed Penicillin sensitivity and were treated with Dicrysticin-S in two and Crys-4 (Squibb)<sup>6</sup> in one. One isolate sensitive to Streptomycin was treated with Dicrysticin-S and in another sensitive to Terramycin, Mastalone was used. The one case treated with Mastalone turned chronic, while all other cases were cured.

Of the five Str. agalactiae isolates the two, sensitive to Streptomycin, were treated with Dicrysticin-S and Penicillin-SI; two Terramycin sensitive ones, with Oxysteclin. The isolate which showed sensitivity to Chloramphenicol was treated with Vetycetine.

All these cases were chronic, with marked reduction in the milk yield, at the time of presentation. After treatment, there was improvement in the milk yield, though the chronicity persisted.

Of the two C. pyogenes isolates, one was sensitive to Penicillin and was treated and cured with Dicrysticin-S. The other isolate which showed sensitivity to Chloramphenicol was treated with Vetycetine. This case showed gangrenous changes on the skin of the teats. The left teat showed extensive gangrenous changes which necessitated the removal of the half subsequently, while the right teat showing only slight gangrenous changes healed.

Out of the three Ent. aerogenes isolates, two showed sensitivity to Terramycin and were treated with Mastalone and the other sensitive to Chloramphenicol with Vetycetino. All these cases were cured.

The one isolate of Esch. coli, sensitive to Chloramphenicol was successfully treated with Vetycetino. The other one sensitive to Terramycin treated with Mastalone did not turn up.

Each of the single isolates of K. pneumoniae and P. aeruginosa sensitive to Chloramphenicol were successfully treated with Vetycetino.

- 
1. Procaine Penicillin B. Vet. C., Streptomycin Sulphate B. Vet. Sulphamerazine U.S.P., Hydrocortisone acetate B. Vet. C.
  2. Procaine Penicillin, G., Penicillin, G. Sodium., Streptomycin Sulphate.
  3. Oxytetracycline dihydrate injection B. Vet. C.
  4. Oxytetracycline Hydrochloride I.P., Oleandomycin., Neomycin Sulphate I.P., Prednisolone I.P.
  5. Chloramphenicol I.P.
  6. Procaine Penicillin, G., Penicillin G Sodium.

Table 1. Results of the bacteriological examination of milk from 56 cases of clinical Mastitis in goats.

Sl. No.	Bacterial organisms	No. of isolates	Percentage
1	<i>Staphylococcus aureus</i>	29	51.79
2	<i>Streptococcus agalactiae</i>	8	14.29
3	<i>Staphylococcus epidermidis</i>	5	8.93
4	<i>Corynebacterium pyogenes</i>	3	5.36
5	<i>Escherichia coli</i>	3	5.36
6	<i>Enterobacter aerogenes</i>	3	5.36
7	<i>Klebsiella pneumoniae</i>	2	3.57
8	<i>Staphylococcus aureus</i> and <i>Streptococcus agalactiae</i>	2	3.57
9	<i>Pseudomonas aeruginosa</i>	1	1.78
Total		56	



Table 2. Correlation of bacterial isolates with symptoms of mastitis.

Sl. No.	Bacterial organisms	No. of isolates	Acute*		Sub-acute**	Chronic***
			Gangrenous	Non-gangrenous		
1	Staphylococcus aureus	29	8	19	1	
2	Streptococcus agalactiae	8	-	1	1	
3	Staphylococcus epidermidis	5	-	-	4	
4	Corynebacterium pyogenes	3	1	2	-	
5	Escherichia coli	3	-	3	-	
6	Enterobacter aerogenes	3	-	2	-	
7	Klebsiella pneumoniae	2	-	-	1	
8	Staphylococcus aureus and Streptococcus agalactiae	2	-	-	-	
9	Pseudomonas aeruginosa	1	-	-	-	

\* Udder hard, hot, painful, gangrene may or may not be seen, secretion watery or purulent; with systemic disturbances.

\*\* Slight hardness, warmth and pain, reduction in milk with flakes or clots; No systemic signs.

\*\*\*Very hard, no warmth and pain, reduction in milk yield with few clots and flakes.

Table 3. Results of the bacteriological examination of 276 milk samples from apparently normal udders of goats.

Sl. No.	Bacterial organisms	No. of isolates	Percentage
1	No bacterial organism	147	53.26
2	Staphylococcus epidermidis	69	25.00
3	Staphylococcus aureus	33	13.77
4	Streptococcus agalactiae	13	4.71
5	Corynebacterium pyogenes	6	2.17
6	Escherichia coli	3	1.09
Total		276	

Table 4. CMT scores on 317 samples of goat's milk and the percentage distribution of bacterial isolates in 251 selected milk samples.

	No. of samples	Percentage distribution by CMT scores				
		Negative	Trace	+	++	+++
CMT Reaction	317	33.44	23.34	30.28	6.62	6.32
No bacteria	76	48.68	43.43	7.09	-	-
Staphylococcus aureus	62	-	-	59.68	16.13	24.19
Staphylococcus epidermidis	74	-	50.00	44.59	5.41	-
Streptococcus agalactiae	23	8.69	4.35	52.17	30.44	4.35
Corynebacterium pyogenes	8	-	12.50	62.50	12.50	12.50
Coliforms	8	-	12.50	50.00	-	37.50

Table 3. CMT scores of 240 milk samples of goat correlated with somatic cell counts.

CMT score	No. of samples	Somatic cell counts per ml ( $10^6$ )		
		Range	Median	Mean
0	37	0.03 - 0.33	0.085	0.09 ± 0.06
Trace	74	0.17 - 0.495	0.31	0.32 ± 0.10
+	96	0.41 - 1.18	0.61	0.65 ± 0.13
++	21	0.91 - 2.46	1.82	1.74 ± 0.42
+++	20	12.5 - 31.8	24.35	22.96 ± 4.90

Table 6. Comparative efficacy of Mastaid Test, Whiteside Test and Teepol Mastitis Test in detecting subclinical mastitis of 103 goat milk samples.

Name of the test	Somatic cell counts/ml	No. of samples	Test scores			
			0	+	++	+++
Mastaid Test	Below 5 lakhs	68	65	3	-	-
	5-10 lakhs	24	-	23	1	-
	Above 10 lakhs	11	-	1	10	-
Whiteside Test	Below 5 lakhs	68	66	2	-	-
	5-10 lakhs	24	7	15	2	-
	Above 10 lakhs	11	-	5	6	-
Teepol Mastitis Test	Below 5 lakhs	68	68	-	-	-
	5-10 lakhs	24	17	4	3	-
	Above 10 lakhs	11	1	3	5	2



170053

Table 7. Results of the in vitro Antibiotic Sensitivity Testing of 53 bacterial isolates from clinical cases of Goat Mastitis.

Sl. No.	Bacterial organisms	No. of isolates	Percentages of sensitivity								
			Ampi-cillin	Chlor-amphe-nicol	Eryth-romy-cin	Kana-mycin	Nitro-fura-zon	Peni-cillin	Strep-tomycin	Sulpha-diazin	Terra-mycin
1	Staphylococcus aureus	31	74.19	83.87	87.10	6.45	41.91	67.74	67.74	3.22	74.19
2	Streptococcus agalactiae	10	100.00	70.00	100.00	-	40.00	100.00	90.00	10.00	70.00
3	Staphylococcus epidermidis	5	100.00	80.00	100.00	-	40.00	100.00	100.00	20.00	60.00
4	Corynebacterium pyogenes	3	100.00	100.00	33.30	33.30	33.30	100.00	-	-	66.67
5	Escherichia coli	3	-	100.00	33.30	100.00	66.67	-	66.67	100.00	66.67
6	Enterobacter aerogenes	3	-	100.00	33.30	66.67	100.00	-	66.67	33.30	66.67
7	Klebsiella pneumoniae	2	-	100.00	-	100.00	50.00	-	50.00	100.00	-
8	Pseudomonas aeruginosa	1	-	100.00	-	100.00	-	-	-	100.00	-

Fig.1

PERCENTAGE DISTRIBUTION OF 128  
BACTERIAL ISOLATES FROM APPARENTLY  
NORMAL HALVES OF GOAT'S UDDER

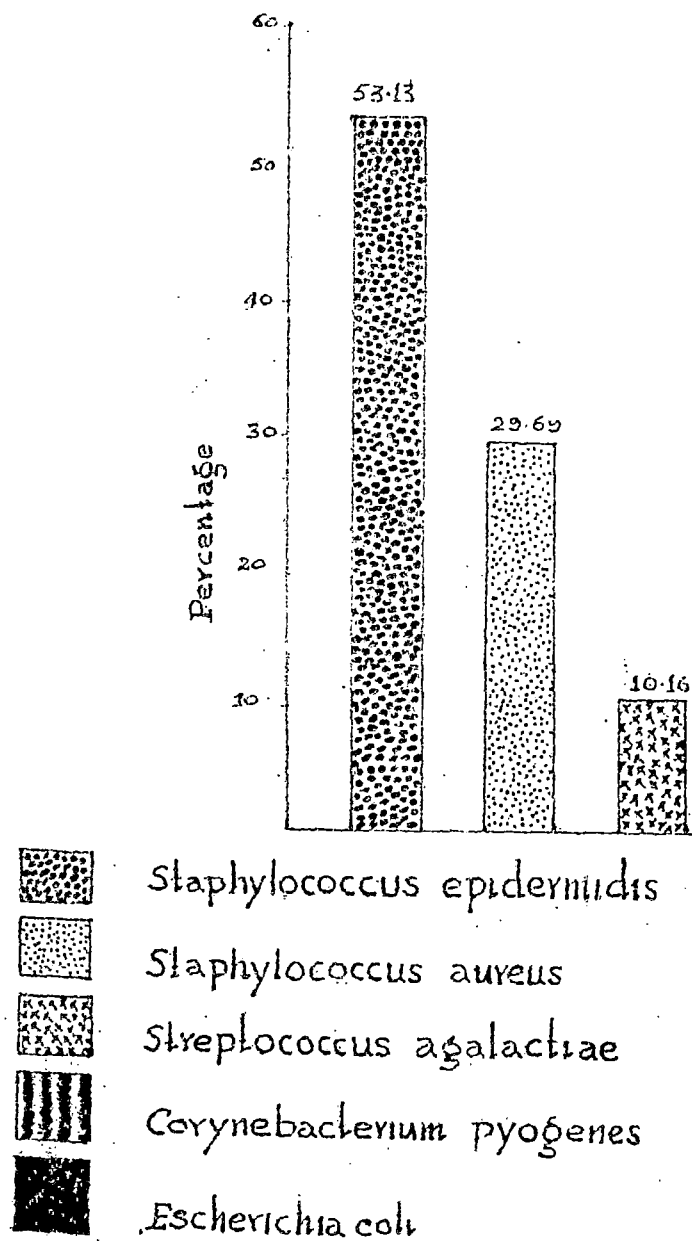
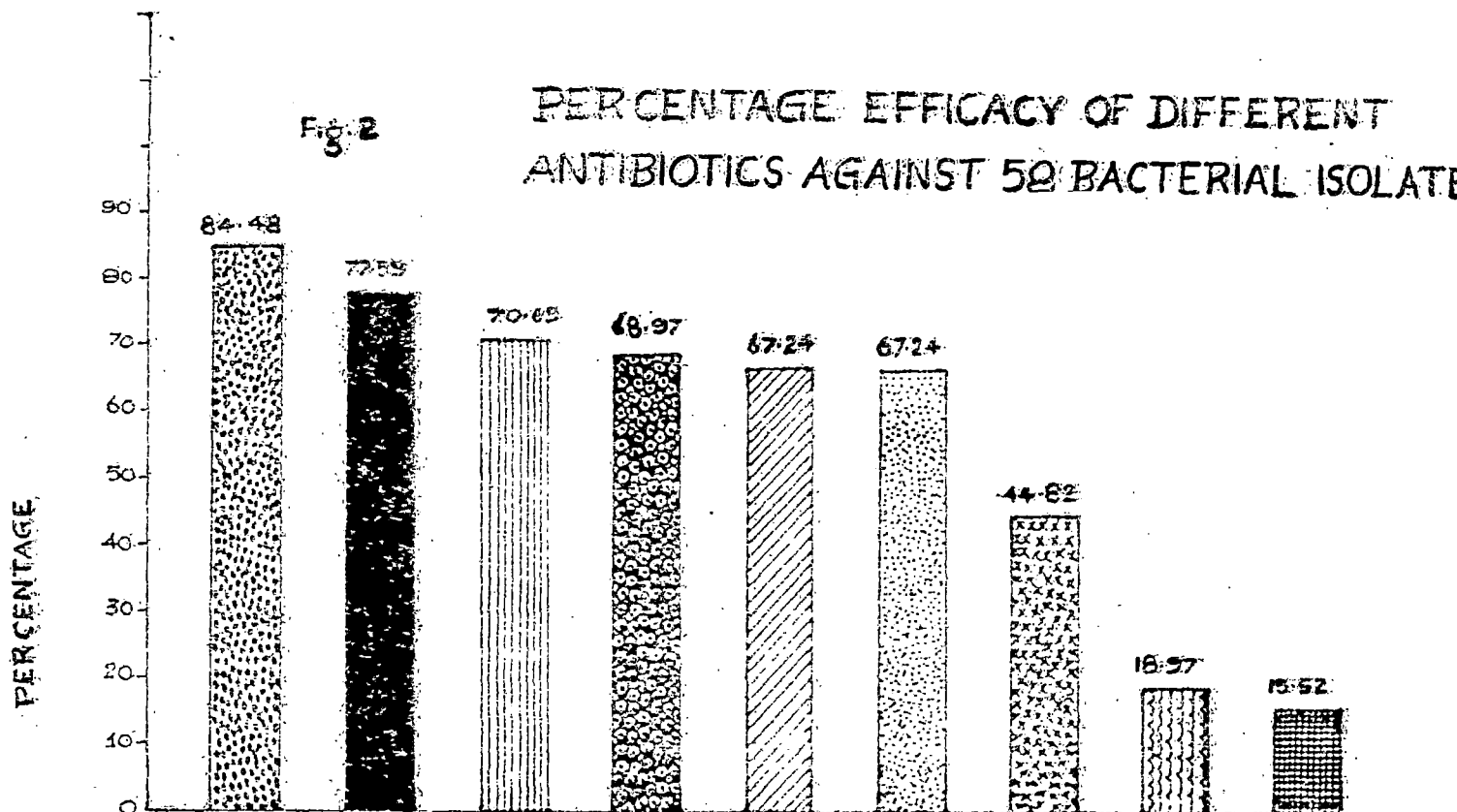


Fig. 2

### PERCENTAGE EFFICACY OF DIFFERENT ANTIBIOTICS AGAINST 50 BACTERIAL ISOLATES












- |   |  |  |
|---|--|--|
|  Chloramphenicol |  Streptomycin |  Nitrofurazon |
|  Erythromycin    |  Penicillin   |  Kanamycin    |
|  Ampicillin      |  Terramycin   |  Sulphadiazin |



Fig-3 ANTIBIOTIC SUSCEPTIBILITY OF DIFFERENT BACTERIAL ISOLATES

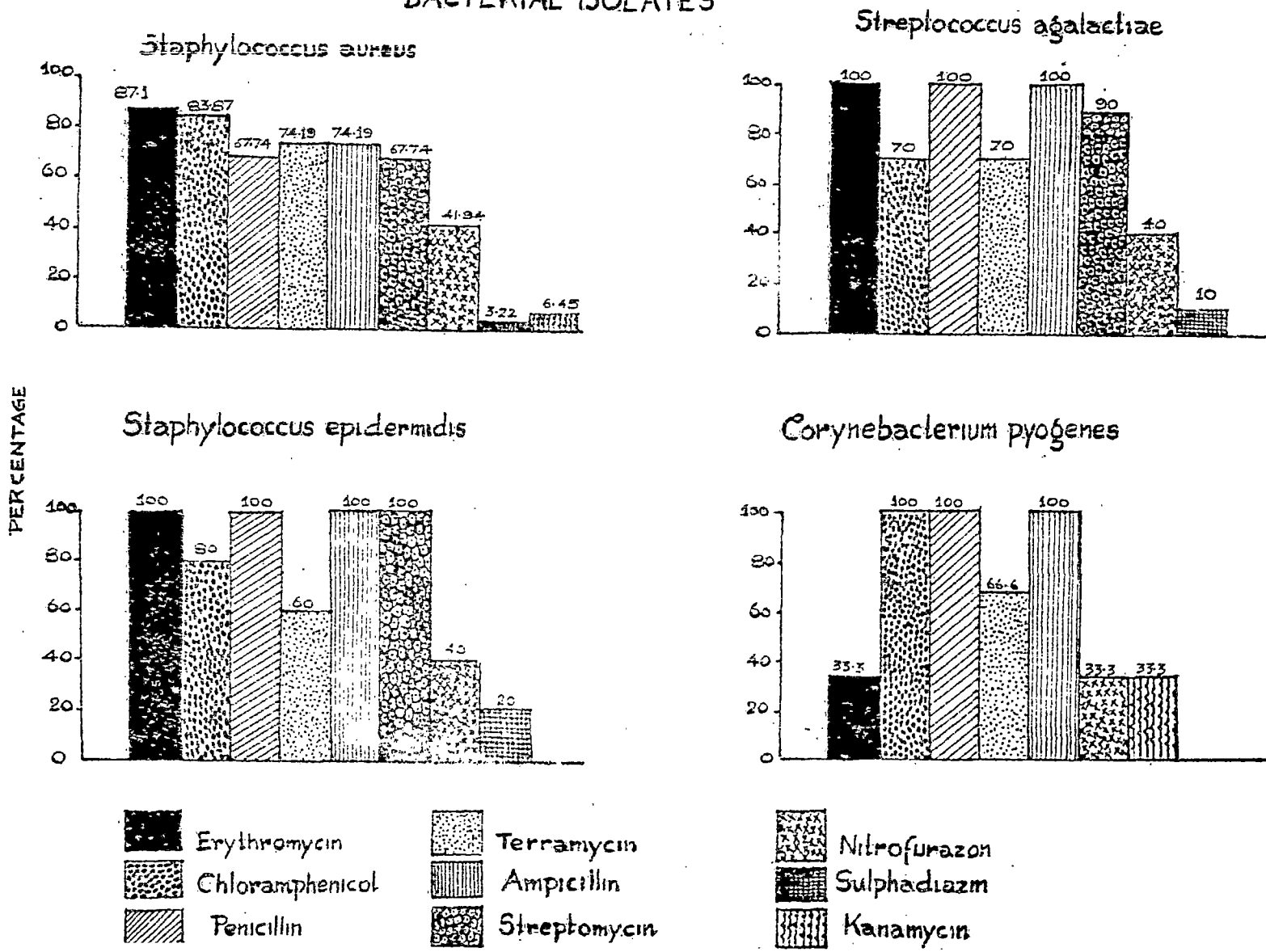
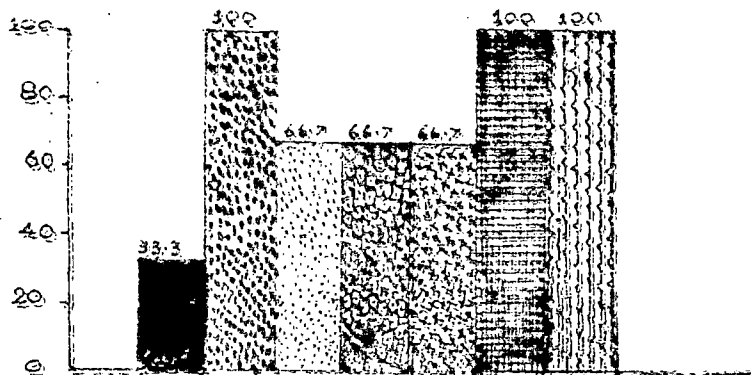


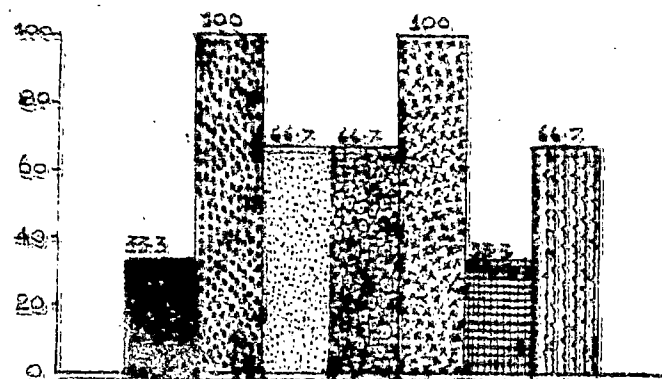
Fig 3 (cont'd)

### ANTIBIOTIC SUSCEPTIBILITY OF DIFFERENT BACTERIAL ISOLATES

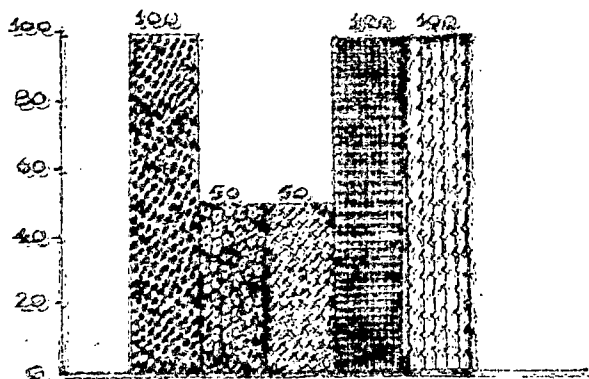
*Escherichia coli*



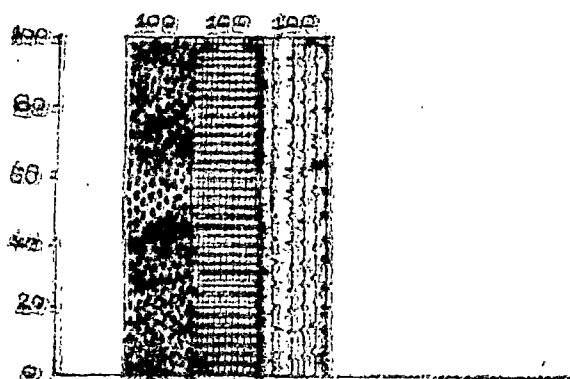
*Enterobacter aerogenes*



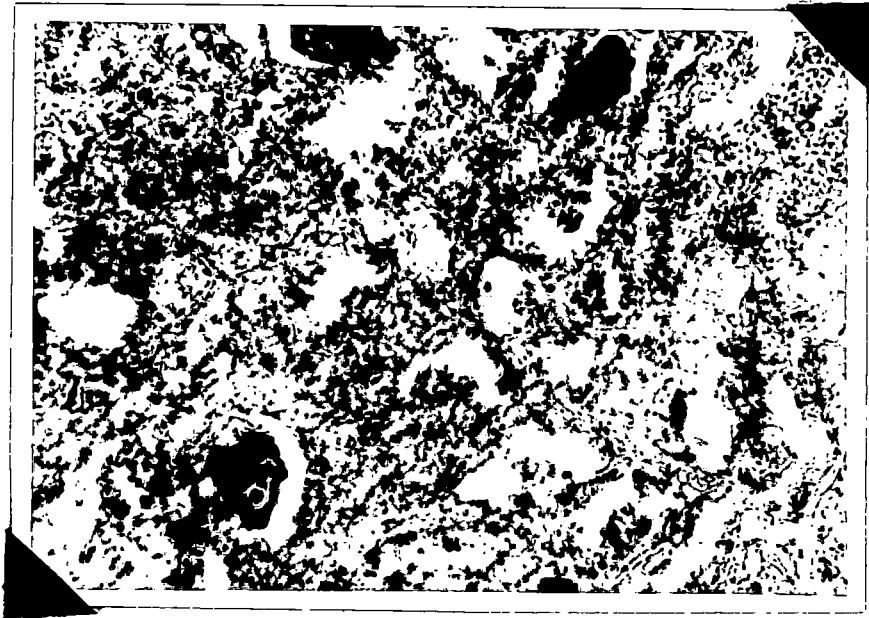
*Klebsiella pneumoniae*

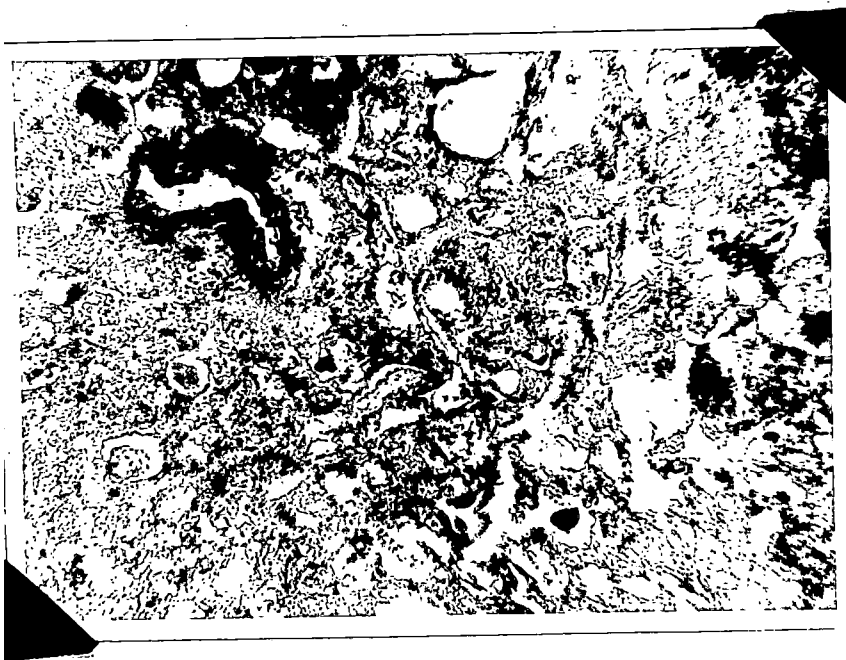


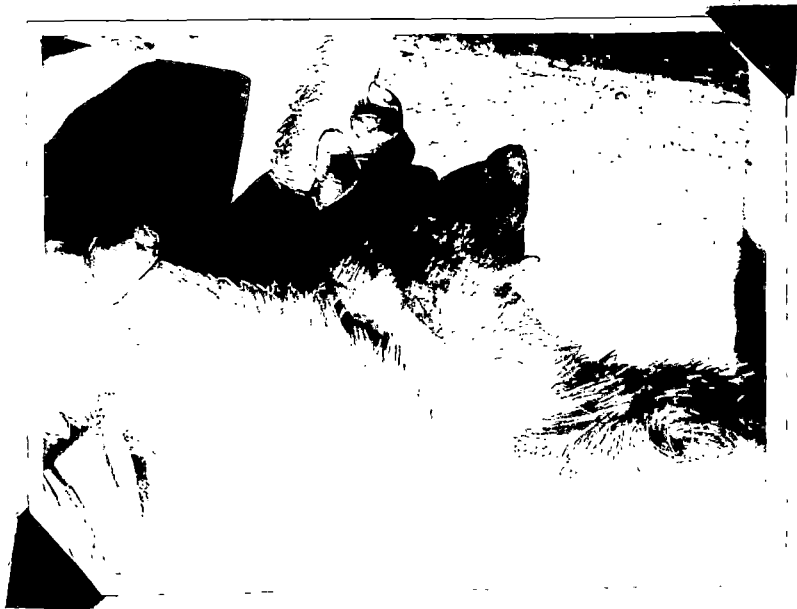
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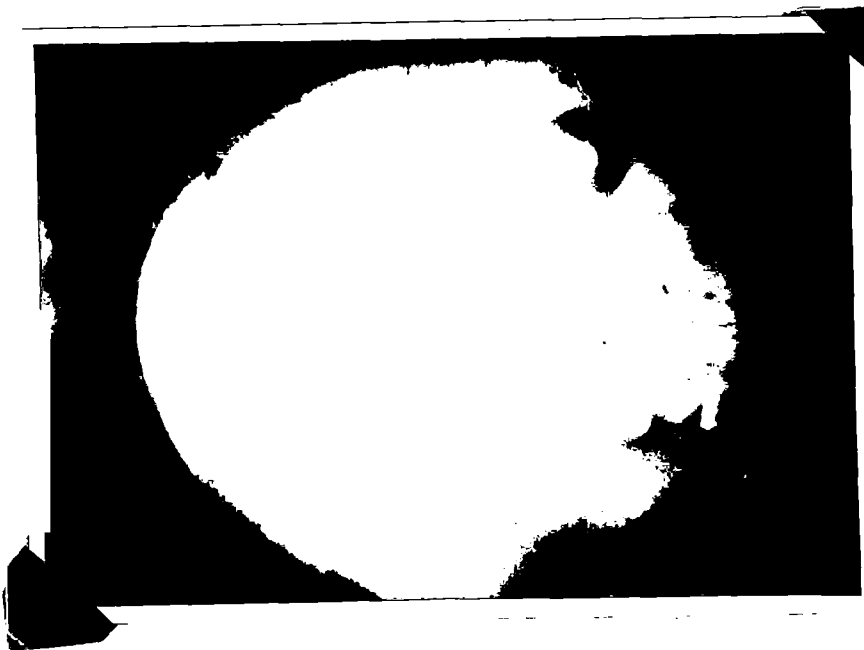
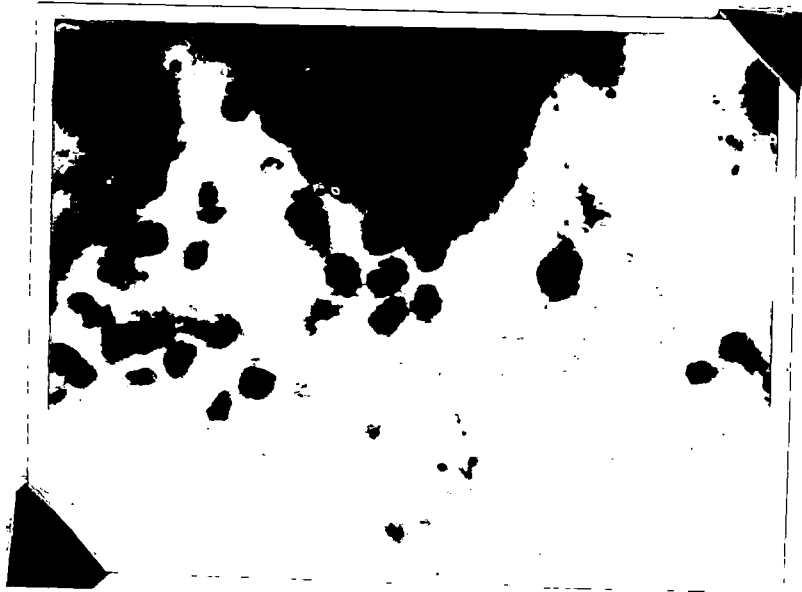














## **DISCUSSION**

## DISCUSSION

A total of 435 milk samples from goats were examined during the present study. Out of these, 56 were from clinical cases of mastitis and 379 from apparently normal udders.

The present work was mostly confined to the hospital cases and a correct assessment of the incidence of the disease in relation to goat population could not be made. Bacteria could be isolated from all the samples from clinical cases. Staph. aureus was the predominant etiological agent in clinical mastitis of goats, constituting upto 52 per cent. It was interesting to note that these organisms were present in pure culture in the large majority of the samples. The higher prevalence of Staph. aureus in caprine mastitis had been observed by Kaplan (1944), Mukherjee and Das (1957) and Kalra et al. (1962).

Staph. aureus mastitis in cattle is usually chronic although acute and peracute cases are not uncommon. However in goats, the commonest form observed was the acute mastitis some of them showing a tendency to become gangrenous. More than 90 per cent of Staph. aureus mastitis in goats examined during this study were acute, the rest being subacute and chronic. Among the acute cases, eight showed gangrenous changes of different degree and five of them were fatal. The gangrenous changes that occur in Staphylococcal mastitis is due to alphatoxin

which causes prolonged vasospasm leading to ischaemia, stagnant type of cyanosis and necrosis (Brown and Scherer, 1958). Mastitis paralleling the natural disease can be produced by infusing the udder with the Staphylococcal alphatoxin. Strains differ in their toxigenicity, but this is not necessarily related to pathogenicity. One strain which produced a gangrenous mastitis on some occasions, produced only a mild disease on other occasions (Jubb and Kennedy, 1970).

Development of gangrene could well be prevented by vaccination which will produce a high antitoxin titre in the serum which could neutralize the alphatoxin activity (Fujikura, 1966). Roguinsky (1977) reported that caprine strains of Staphylococci are serologically distinct from bovine and ovine strains although they all belong to the biotype C. The high incidence of gangrenous mastitis in goats may be due to the fact that these strains are more toxigenic.

In gangrenous mastitis observed during this study there was swelling of the gland; the skin on the teat and base of the skin were cold and moist. Bluish discolouration of these areas was more distinct in goats with light skin colour (Plates I & II). The epithelium of these portions could be easily peeled off. The secretion in most cases were watery and dark red. In all cases, there were accompanying systemic disturbances following the absorption of toxins.

Histological sections revealed focal areas of necrosis, dense infiltration with neutrophils and plasma cells causing extensive destruction of parenchyma and acinar tissue. The injury produced by the toxin to ductal epithelium results in the release of chemotactic substances from the cells which attract leukocytes. Severe engorgement of blood vessels were seen with thrombosis in many of them.

Schalm et al. (1971) reported that the thrombosis of large veins is responsible for the moist nature of gangrene with constant dripping of blood tinged serum from teat and skin around the base of the teat.

Isolated foci of suppuration were also evident scattered in the parenchyma with numerous dense bluish staining masses of corpora amylacea (Plates III & IV). Jubb and Kennedy (1970) reported the occurrence of large number of such bodies in mammary glands affected with mastitis.

In one case of acute mastitis without any tendency of gangrenous changes, there was abscess formation and pus was draining out through a fistula at the base of the teat. Histologically, the tissue revealed multiple foci of suppuration, scattered in the acinar tissue. There was extensive interlobular fibrosis causing atrophy of the lobules (Plate V).

Fatal cases of Staphylococcal intoxication from raw goat

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milk had been reported by Keed et al. (1943). This was due to an enterotoxin produced by the organism (Schalm et al. 1971). The high incidence of Staph. aureus mastitis in goats is of serious public health significance.

Staph. aureus was isolated from 30 per cent of sub-clinical infections also. In such cases, although animals did not show any symptoms, it will have far reaching effects. At any time, when the resistance of the animal goes down they may multiply and produce acute mastitis. In addition, these animals are a potential danger to other animals.

Coagulase negative, non-haemolytic Staph. epidermidis formed the major cause of subclinical mastitis in goats. They are only mildly pathogenic and usually do not cause obvious clinical mastitis. They do not produce any true toxin and are more susceptible to leukocytic enzymes (Schalm et al. 1971). However, they cause irritation to the udder which may lead to microscopic lesions with increased leukocytic infiltration. In such cases, the CMT score and somatic cell counts were high. The milk samples from which Staph. epidermidis was isolated gave a CMT score of + in 45 per cent cases and Trace in 50 per cent of cases.

Although it is stated that clinical cases of mastitis by Staph. epidermidis are of rare occurrence, five such cases were observed during the present study. But all these cases

were mild without any systemic reaction and gangrenous changes.

Staph. epidermidis colonize in the streak canal and skin surface in preference to milk. Some of these strains produce an antibiotic substance inhibitory to Staph. aureus and hence they appear to have the advantage that they resist colonization of Staph. aureus at least in the teat canal and skin (Edwards and Jones, 1966). From this point of view, it may appear beneficial to retain the bacterial flora of Staph. epidermidis in the udder tissue. But the fact that they rarely produce clinical mastitis, as is seen by the present study, may necessitate their elimination.

Str. agalactiae was isolated from 14 per cent of clinical and ten per cent of subclinical infections. Among the clinical cases 75 per cent were chronic and rest were acute or subacute. In chronic cases, the udder showed induration of varying degree, with apparently no systemic disturbances. There was reduction in the milk yield. In few cases, the milk was thin and watery with clots and flakes, while in others, there was no appreciable changes in the milk. The organisms usually live in the milk channels and the irritation of the tissue results from a toxin or accumulated lactic acid (Schalm et al. 1971). This leads to exudation of leukocytes and blood plasma factors into the alveoli and ducts and milk gives a high CMT score and somatic cell count. In this study, 52 per cent of the milk samples

from which Str. agalactiae was isolated gave a CMT score of +.

Chronic mastitis are of particular importance as they interfere with the economic production of milk. Apart from this, there are dangers of transmission of infection to other animals and occasional flare-ups to the acute form.

C. pyogenes usually causes acute suppurative mastitis. All the three cases encountered during the present study were of this type. In one case, there was moist gangrene on the skin of the teat of both halves with edema and bluish discoloration of the affected part (Plates VII & VIII). The lesion on the right half involved the superficial layers of the skin at the tip of the teat, while in the left it extended from the tip to the base of the teat, with involvement of deeper tissues of the udder. After treatment, the scab formed on the tip of the right teat sloughed off and the wound healed up, while in the left, scab formation occurred on the entire teat and the teat had shrunk. This necessitated the removal of the left half.

Schalm et al. (1971) reported that a bluish discoloration of the skin especially around the base of the teat occurred in a case of mastitis due to Corynebacterium in cattle.

The necrotic changes on the skin observed in the present study may probably be due to a dermonecrotxin produced by C. pyogenes as suggested by Merchant and Packer (1971).

Histological examination of the gangrenous portion of the skin showed focal areas of dermal necrosis associated with areas of acanthosis and hyperkeratosis. Jain (1965) observed thickening of epidermal layer on teats, with degeneration and squamation of the cisternal epithelium and fibrotic proliferation of subepithelial tissue of experimental C. pyogenes mastitis in goats. Although the section of the mammary gland did not reveal any gross changes, microscopically there were multiple foci of suppuration and necrosis. The interstitial tissue was diffusely edematous and was infiltrated with neutrophils. Some of the acini had been converted into foci of suppuration (Plates XI & XII). Jain (1964) had also observed, on histopathological examination of the udder from experimental infection with C. pyogenes, acute suppurative mastitis with infiltration of neutrophils and plasma cells. Abscess formation and necrosis were followed by fibrous tissue proliferation leading to pressure atrophy of lobules.

C. pyogenes caused two per cent of subclinical mastitis, and those milk samples gave a high somatic cell count and CMF score. Ibrahim (1968) isolated corynebacterium species from eight per cent of subclinical cases of mastitis in goats.

Coliform mastitis was not uncommon in goats. Out of 56 clinical cases, Coliforms were isolated from 14 per cent of samples. These included five per cent each by Esch. coli



and Ent. aerogenes and four per cent by K. pneumoniae. Majority (62.5%) of these were acute mastitis with pronounced systemic involvement and the remaining (37.5%) were subacute and chronic. In such cases the entire gland was tense and showed marked enlargement. The secretion was scanty and serous in nature. The systemic signs in Coliform mastitis has been ascribed to endotoxaemia rather than bacteriaemia (Schalm et al. 1971). All the Esch. coli and two Ent. aerogenes isolates produced acute mastitis.

Esch. coli was isolated from three cases of subclinical mastitis also. Ibrahim (1968) isolated Coliform in 5.9 per cent of clinical and 6.3 per cent of subclinical mastitis in goats.

Coliforms after entry into the udder undergo fast multiplication, producing acute inflammation, causing infiltration of leukocytes in large numbers. When this occurs, the organisms disappear because they are inhibited by leukocytes and other products of inflammation. The view that leukocytes play a prominent role in the control of Coliform mastitis is supported by the observation of unrestricted multiplication of Coliforms in leukopenic cows. Coliforms fail to establish in a slightly irritated udder. As long as the more common mastitis pathogens are available to invade the mammary gland which stimulate leukocytosis, Coliforms will not establish there (Schalm et al. 1971). This may be the probable reason for the low incidence of Coliform

mastitis, in spite of the fact that they are widespread in the environment.

A case of chronic mastitis caused by E. aeruginosa was observed, wherein the affected half was hard to touch and milk was thin and watery. There was no systemic disturbances and the condition responded to Chloramphenicol therapy.

The CMT scores of 317 goat milk samples were—

—	33.44	per cent
Trace	23.34	"
+	30.28	"
++	6.62	"
+++	6.32	"

Out of the 76 bacteriologically sterile goat milk samples, eight per cent gave false positive score of +. The somatic cell counts in these cases exceeded five lakhs per ml milk.

The 62 samples from which Staph. aureus was isolated gave a CMT score + and above. The percentage distribution of the CMT scores were + (60), ++ (16) and +++ (24).

Of the 74 Staph. epidermidis isolates, CMT scores of + and ++ were shown by 44 and 6 per cent of the samples respectively. The remaining samples gave only a Trace score. Str. agalactiae in 87 per cent of cases gave a CMT score of + and

above, and the remaining 13 per cent gave scores less than +. In these cases the somatic cells in the milk were also low. This may perhaps be due to reduced inflammatory process, as the condition moves towards chronicity. Pattison et al. (1950) showed that in experimental Str. agalactiae mastitis in goats, the milk was strongly positive for the Whiteside test for the first five days and thereafter became negative although milk was culturally positive.

In 87.5 per cent of cases from which C. pyogenes was isolated, gave CMT scores + and above and 12.5 per cent gave Trace score. Similar results were obtained with Coliform mastitis (Table 4).

Generally, in acute cases of mastitis, the CMT scores were +++.

Subclinical mastitis in goats was detected with Mastaid Test. The distribution of different scores of 276 samples were Negative (111), Trace (74), + (86) and ++ (5). Although only 41 samples with Trace scores revealed bacterial organisms, only three out of 86 subclinical cases with Mastaid score +, were negative. Thus this test showed 96.5 per cent efficacy in detecting subclinical mastitis.

Mastaid Reagent (Glaxo) is intended primarily for the

detection of subclinical mastitis in cattle. The efficacy of this reagent was proved in cattle by Seore and Khande (1972). But no work seems to have been done in goats with this reagent for detecting subclinical mastitis as evident from published literature. In the present study it was observed that Mastaid Reagent can effectively be used in goats to detect subclinical mastitis.

A total of 103 milk samples were subjected to Mastaid Test, Whiteside Test and Teepol Mastitis Test. In 68 samples with a cell count of below 5 lakhs, all the three tests gave negative reaction. The remaining 35 samples, with cell counts of more than 5 lakhs, Mastaid Test was positive in all, Whiteside Test in 28 and Teepol Mastitis Test in 17. From these observations, it was concluded that Mastaid Test is highly efficient in the detection of subclinical mastitis in goats (100%), the next in the order was Whiteside Test (80%) and the least Teepol Mastitis Test (49%).

Theoretically, a healthy udder should not contain any somatic cell (Schalm et al. 1971). However, practically such an udder cannot be found and a certain number of somatic cells in milk are considered to be normal. To assess the normal range of cells in goat's milk, 37 samples which were CMT negative and bacteriologically sterile were subjected to somatic cell counting. The mean value was found to be  $0.09 \pm 0.06 \times 10^6$  per ml.

The cell counts of samples giving different CMT scores were also determined and these values are more or less in agreement with those made by Schalm et al. (1971) as shown below:

<u>CMT score</u>	<u>Mean somatic cell counts (x 10<sup>5</sup> per ml)</u>	
	<u>Present study</u>	<u>Schalm et al. (1971)</u>
0	0.9 ± 0.6	0.68
T	3.2 ± 1.0	2.68
+	6.5 ± 1.3	8.00
++	17.4 ± 4.2	25.60
+++	229.6 ± 49.0	More than 100.0

The range of somatic cells in goat milk samples giving different CMT scores showed that CMT is relatively an efficient indicator of the changes in leukocytic counts.

The observations made during this study showed that somatic cell counts below one lakh/ml represented more or less healthy glands. Counts of one to five lakhs cells per ml suggested moderate inflammatory changes in the gland due to the presence of non-pathogenic or mildly pathogenic organisms such as Staph. epidermidis. Counts above five lakh cells per ml indicated the presence of pathogenic bacteria like Staph. aureus producing a subclinical infection or moderately pathogenic bacteria like Str. agalactiae producing a clinical infection. Very high cell counts above ten million cells per ml probably indicated highly pathogenic bacteria like C. pyogenes, Staph. aureus

or Coliform producing an acute clinical disease. Generally, a cell count of 5 to 10 lakhs (CMT +) can be considered as subclinical infection. Since a high somatic cell count and CMT score indicates presence of leukocytes in milk which can also occur from non-infectious conditions, for making a confirmatory diagnosis of mastitis, this should be accompanied by cultural examination of milk.

In vitro antibiotic sensitivity testing provide a useful tool for assessing the possible effectiveness of antibiotics in vivo against microorganisms. The results of testing the overall effectiveness of the antibiotics against the 58 bacterial isolates, reveal that Chloramphenicol is the most effective antibiotic, followed by Erythromycin, Ampicillin, Streptomycin, Penicillin, Terramycin, Nitrofurazon, Kanamycin and sulphadiazin. Chloramphenicol gave the best results in similar studies by Ovejero et al. (1960); Krzyzanowski et al. (1965) and Jhala (1976).

In the present study it was observed that Chloramphenicol was 100 per cent effective against P. aeruginosa, K. pneumoniae, Ent. aerogenes, C. pyogenes and Bach. coli. Moderate resistance to Chloramphenicol was shown by Staph. aureus, Staph. epidermidis and Str. galactiae. Shaw et al. (1970) studied the mechanism of Chloramphenicol resistance in bacteria and

observed that it was due to an enzyme Chloramphenicol acetyltransferase of bacterial origin.

Although Chloramphenicol is a very valuable drug against many mastitis pathogens, British Veterinary Association (1976) strongly recommended the necessity for putting restrictions to its use in animals. This is because Chloramphenicol is the valuable therapeutic agent in human typhoid and the erratic use of this drug in animals may lead to the evolution of resistant strains of typhoid bacilli by transfer of resistant factors.

Erythromycin, the next in the order, was effective against all strains of Str. agalactiae, Staph. epidermidis and 87 per cent of Staph. aureus strains. Other bacterial isolates showed high percentage of resistance. Resistance to Erythromycin emerges in bacteria during serial cultural passages in the presence of antibiotics and it is believed to reside in cell free amino acid polymerization systems (Haight and Finland, 1952).

Penicillin and Ampicillin were 100 per cent effective against Str. agalactiae, Staph. epidermidis and C. pyogenes. But Staph. aureus showed resistance of 33 per cent for Penicillin and 26 per cent for Ampicillin. None of the gram negative organisms showed sensitivity to these antibiotics. The resistance of Staph. aureus to these antibiotics were due to the enzyme Penicillinase ( $\beta$ -lactamase) which breaks up the  $\beta$ -lactam ring of Penicillin making it inactive (Davis et al. 1973). Riberstein et al. (1974) reported that multiple drug resistance of

Staph. aureus are decreasing because of a restraint in the use of anti-microbial agents and by the introduction of semi-synthetic penicillin which are moderately resistant to the action of Penicillinase.

Streptomycin was highly effective against Staph. epidermidis and Str.agalactiae, moderately against Staph. aureus, Esch. coli and Ent. aerogenes. Bacterial resistance to Streptomycin is mediated through plasmids which causes destruction of these antibiotics with the help of enzymes (Davis et al. 1973).

Moderate sensitivity to Tetracyclines was shown by 1 bacterial isolates. However, K. pneumoniae and P. aeruginosa were totally resistant. This drug resistance to Tetracyclines is believed to be due to impermeability of the bacteria to these drugs (Laskin and Last, 1971).

Treatment was carried out on the basis of in vitro drug sensitivity in 40 selected clinical cases, caused by different organisms. A complete cure was obtained in 27 animals. Of the remaining 13 cases, seven turned chronic, three died and in one case a half had to be removed. In other two cases, the results were not available since the owner did not turn up.

All the three goats that died during treatment had mastitis due to Staph. aureus. Of these, two animals had gangrenous



changes in the udder with systemic reaction and toxæmia when it was presented for treatment. The other animal was showing systemic reaction with toxæmia at the time of commencement of treatment.

The alphatoxin is responsible for the development of a gangrenous form of Staph. aureus mastitis. The tissue changes occur very rapidly. The toxin produces ischaemia and thrombosis of large vessels preventing the parenterally administered drug to reach the site of action. Local intramammary infusion of the drug also will not give the desired effect because the organism multiply deep in the parenchyma where the drug has little accessibility. The living bacteria are often surrounded by dense fibrous tissue which makes the drug inaccessible to the organism (Derbyshire, 1958). These may be some of the reasons for the failure of treatment in spite of the fact that most specific drug was given.

Mastitis caused by Staph. epidermidis was only subacute, which reflects upon its low pathogenicity causing little tissue damage and the drug had more accessibility and hence produced cure.

All the Str. agalactiae mastitis were chronic with marked fibrosis. Even treatment with most specific drug did not give the desired effect in this condition except for slight increase in the milk yield. In chronic mastitis, the inflammation is

restricted to acinar and ductal epithelium. The inflammation later subsides in few days after infection and is replaced by connective tissue proliferations in the inter acinar space. This results in the step-wise loss of secretory function, increase in fibrosis and eventual atrophy (Blood and Henderson, 1974). Even if the organisms are removed naturally or by treatment, this fibrotic tissue persists.

In one case of C. pyogenes, it was necessary to remove one half of the gland. The organism was sensitive to Chloramphenicol and was treated with Vetycetine. There was moist gangrenous changes on skin of both teats, more extensive in the left. After treatment, the lesion turned dry and was about to slough off when the gland was removed. However, the minor gangrenous lesion sloughed off and wound healed. Other two cases showed acute suppurative mastitis and was cured completely.

Generally, Coliforms showed susceptibility to Chloramphenicol and Terramycin. Those cases were successfully treated with proprietary preparations containing these antibiotics. The single case of P. aeruginosa mastitis was treated successfully with Vetycetine, since it was Chloramphenicol susceptible.

From these observations it is inferred that the in vitro drug sensitivity is a pre-requisite for the rational treatment of mastitis.

# SUMMARY

## SUMMARY

A total of 435 milk samples from goats were examined during the present study. These included milk samples from 56 clinical cases of mastitis and 379 apparently normal udders.

In clinical mastitis, Staph. aureus was the predominant etiological agent isolated constituting 52 per cent. The percentages of other pathogens isolated were Str. agalactiae (14), Staph. epidermidis (9), C. pyogenes (5), Esch. coli (5), Ent. aerogenes (5), K. pneumoniae (4) and P. aeruginosa (2). Mixed infections with Staph. aureus and Str. agalactiae were observed in four per cent of cases.

In majority of the cases, Staph. aureus produced acute mastitis out of which eight cases showed gangrenous changes of varying degree. Histologically, the tissue sections from these cases revealed thrombosis of vessels, leukocytic infiltration and focal areas of necrosis with extensive destruction of acinar tissue. Of these, five cases were fatal. Staph. epidermidis produced subacute mastitis in majority of the cases. Str. agalactiae mastitis in 75 per cent were chronic with marked induration and drop in milk yield. C. pyogenes caused acute suppurative mastitis. Gangrenous changes were noticed on the skin of the teat in one case. Generally, Coliform mastitis were acute

with toxæmia and systemic disturbances. A chronic case of *B. aeruginosa* mastitis was encountered.

The percentage of subclinical mastitis among 276 milk samples from apparently normal goats was 47. Of these, *Staph. epidermidis* formed the major pathogen constituting 53 per cent. The percentages of occurrence of other bacteria were, *Staph. aureus* (30), *Str.agalactiae* (10), *C. pyogenes* (5) and *Esch. coli* (2).

A modified CMT was done using Mastaid reagent on 317 samples of milk. The percentage distribution of different scores were zero (33), Trace (23), + (30), ++ (7) and +++ (6). In most of the acute clinical mastitis the scores were +++ indicating high cell counts. Mastaid Test revealed 96.5 per cent efficacy in detecting subclinical mastitis in goats.

In order to assess the normal range of cells in milk, 37 CMT negative and bacteriologically sterile milk samples were subjected to somatic cell counting and the mean value was found to be 0.9 lakhs per ml. Similar values for the CMT scores Trace, +, ++ and +++ were 3.2, 6.5, 17.4 and 229.6 lakhs per ml respectively.

Counts below one lakh per ml represented normal udders and one to five lakhs per ml suggested slight inflammatory reaction. Generally, a cell count of five to ten lakhs (CMT +) could

be considered subclinical infection.

When a comparative study was conducted with Mastaid Test, Whiteside Test and Teepol Mastitis Test, it was observed that Mastaid reagent could only be relied upon in detecting subclinical mastitis.

In vitro drug sensitivity conducted on 58 bacterial isolates showed that Chloramphenicol had least resistant strains. The efficacy of other antibiotics in the decreasing order were Erythromycin, Ampicillin, Streptomycin, Penicillin, Terramycin, Nitrofurazon, Kanamycin and Sulphadiazin.

Chloramphenicol was 100 per cent effective against Coliforme, C. pyogenes and P. aeruginosa and moderately against Staph. aureus, Staph. epidermidis and Str. agalactiae. Erythromycin showed good inhibitory action against gram positive Cocci. Penicillin and Ampicillin gave inhibitory action against Str. agalactiae, Staph. epidermidis and C. pyogenes while Staph. aureus revealed moderate resistance. Streptomycin had good inhibitory action on Staph. epidermidis and Str. agalactiae; moderately so to Staph. aureus, Bach. coli and Ent. aerogenes. Moderate sensitivity to Tetracyclines was shown by many bacterial isolates. However, K. pneumoniae and P. aeruginosa were totally resistant.

Treatment was carried out on the basis of in vitro drug

sensitivity on 40 selected clinical cases. A complete cure was obtained in 27 cases. Of the remaining 13 cases, seven turned chronic, three died and in one case a half of the gland had to be removed. In the other two cases, results were not available since owner did not turn up. In the three fatal cases, there was toxæmia and gangrenous changes, prior to the commencement of treatment.

It was inferred that in vitro drug sensitivity is a pre-requisite for the rational treatment of mastitis.

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originals not consulted.

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# APPENDIX

Appendix I. Zone size interpretive chart used for disc sensitivity test.

Antibiotic/Chemotherapeutic Agent	Disc potency	Inhibition zone in millimetres		
		Resistant	Moderately sensitive	Sensitive
Ampicillin	10 mcg	20 or less	21 - 28	29 or more
Chloramphenicol	30 "	12 "	13 - 17	18 "
Erythromycin	15 "	13 "	14 - 17	18 "
Kanamycin	30 "	13 "	14 - 17	18 "
Nitrofurans	300 "	14 "	15 - 16	17 "
Penicillin	10 I.U.	20 "	21 - 28	29 "
Streptomycin	10 mcg	11 "	12 - 14	15 "
Sulphonamides	300 "	12 "	13 - 16	17 "
Tetracyclines	30 "	14 "	15 - 18	19 "

Appendix II (A)

Results of examination of 37 normal goat milk samples.

Sl. No.	CMT score	Cultural examination	Somatic cell count (x 10 <sup>6</sup> /ml)
1	Negative	Negative	0.09
2	"	"	0.19
3	"	"	0.06
4	"	"	0.085
5	"	"	0.09
6	"	"	0.38
7	"	"	0.09
8	"	"	0.11
9	"	"	0.05
10	"	"	0.095
11	"	"	0.065
12	"	"	0.09
13	"	"	0.18
14	"	"	0.03
15	"	"	0.05
16	"	"	0.045
17	"	"	0.05
18	"	"	0.11
19	"	"	0.05
20	"	"	0.07
21	"	"	0.055
22	"	"	0.095
23	"	"	0.075
24	"	"	0.095
25	"	"	0.065
26	"	"	0.11
27	"	"	0.045
28	"	"	0.095
29	"	"	0.075
30	"	"	0.095
31	"	"	0.1
32	"	"	0.095
33	"	"	0.085
34	"	"	0.08
35	"	"	0.04
36	"	"	0.035
37	"	"	0.11

Appendix II (b)

Results of examination of 74 goat milk samples with CMT -  
Trace score.

Sl. No.	CMT score	Cultural examination	Somatic cell count (x 10 <sup>6</sup> /ml)
1	Trace	Negative	0.39
2	"	Staphylococcus epidermidis	0.32
3	"	-do-	0.27
4	"	-do-	0.31
5	"	Negative	0.31
6	"	Negative	0.41
7	"	Staphylococcus epidermidis	0.495
8	"	Corynebacterium pyogenes	0.41
9	"	Streptococcus agalactiae	0.38
10	"	Negative	0.21
11	"	Negative	0.32
12	"	Negative	0.38
13	"	Negative	0.29
14	"	Negative	0.18
15	"	Staphylococcus epidermidis	0.39
16	"	Negative	0.39
17	"	Negative	0.24
18	"	Escherichia coli	0.39
19	"	Negative	0.19
20	"	Streptococcus agalactiae	0.48
21	"	Negative	0.19
22	"	Negative	0.49
23	"	Negative	0.19
24	"	Negative	0.30
25	"	Negative	0.19
26	"	Negative	0.38
27	"	Staphylococcus epidermidis	0.29
28	"	-do-	0.42
29	"	-do-	0.48
30	"	-do-	0.41
31	"	-do-	0.48
32	"	-do-	0.495
33	"	-do-	0.41
34	"	-do-	0.48
35	"	Negative	0.20
36	"	Negative	0.39
37	"	Negative	0.21
38	"	Staphylococcus epidermidis	0.30
39	"	-do-	0.24
40	"	-do-	0.27
41	"	-do-	0.31

Sl. No.	GHT score	Cultural examination	Somatic cell count (x 10 <sup>6</sup> /ml)
42	Trace	Negative	0.38
43	"	Negative	0.32
44	"	Negative	0.40
45	"	Negative	0.27
46	"	Negative	0.39
47	"	Negative	0.38
48	"	Negative	0.27
49	"	Staphylococcus epidermidis	0.17
50	"	-do-	0.32
51	"	-do-	0.41
52	"	-do-	0.31
53	"	-do-	0.325
54	"	-do-	0.42
55	"	-do-	0.405
56	"	-do-	0.18
57	"	-do-	0.28
58	"	-do-	0.19
59	"	-do-	0.39
60	"	-do-	0.30
61	"	Negative	0.31
62	"	Negative	0.18
63	"	Staphylococcus epidermidis	0.21
64	"	-do-	0.27
65	"	-do-	0.31
66	"	-do-	0.19
67	"	-do-	0.22
68	"	Negative	0.29
69	"	Staphylococcus epidermidis	0.40
70	"	-do-	0.22
71	"	-do-	0.30
72	"	Negative	0.31
73	"	Negative	0.21
74	"	Negative	0.495

## Appendix II (c)

Results of examination of 96 milk samples with GMT + score.

Sl. No.	GMT score	Cultural examination	Somatic cell count ( $\times 10^6/\text{ml}$ )
1	+	Staphylococcus aureus	0.52
2	+	Staphylococcus epidermidis	0.61
3	+	-do-	0.58
4	+	-do-	0.58
5	+	-do-	0.44
6	+	Staphylococcus aureus	0.68
7	+	-do-	0.70
8	+	-do-	0.61
9	+	Streptococcus agalactiae	0.51
10	+	-do-	0.67
11	+	Staphylococcus epidermidis	0.91
12	+	-do-	0.72
13	+	-do-	0.83
14	+	-do-	0.61
15	+	-do-	0.61
16	+	Corynebacterium pyogenes	0.72
17	+	Staphylococcus epidermidis and Escherichia coli	0.79
18	+	Staphylococcus aureus	0.49
19	+	-do-	0.81
20	+	-do-	0.68
21	+	-do-	0.59
22	+	-do-	0.71
23	+	-do-	0.51
24	+	-do-	0.49
25	+	Streptococcus agalactiae	0.52
26	+	-do-	0.68
27	+	Negative	0.51
28	+	Corynebacterium pyogenes	0.59
29	+	Staphylococcus epidermidis and Streptococcus agalactiae	0.71
30	+	Streptococcus agalactiae	0.57
31	+	Staphylococcus aureus	0.68
32	+	-do-	0.72
33	+	-do-	0.53
34	+	Staphylococcus epidermidis	0.51
35	+	-do-	0.48
36	+	-do-	0.51
37	+	Staphylococcus aureus	0.68
38	+	-do-	0.72
39	+	-do-	0.61
40	+	-do-	0.61
41	+	-do-	0.62
42	+	-do-	0.58
43	+	-do-	0.50
44	+	-do-	0.67
45	+	-do-	0.52



Sl. No.	CMT score	Cultural examination	Somatic cell count (x 10 <sup>6</sup> /ml)
46	+	Staphylococcus epidermidis	0.51
47	+	-do-	0.49
48	+	-do-	0.51
49	+	-do-	0.59
50	+	-do-	0.59
51	+	Negative	0.48
52	+	Streptococcus agalactiae	0.61
53	+	Staphylococcus aureus	0.69
54	+	-do-	0.79
55	+	-do-	0.71
56	+	-do-	0.56
57	+	-do-	0.61
58	+	-do-	0.69
59	+	-do-	0.59
60	+	Corynebacterium pyogenes	0.61
61	+	-do-	0.79
62	+	Streptococcus agalactiae	0.59
63	+	-do-	0.72
64	+	Staphylococcus epidermidis	0.72
65	+	-do-	0.81
66	+	-do-	0.59
67	+	-do-	0.49
68	+	-do-	0.61
69	+	-do-	0.41
70	+	-do-	0.50
71	+	Corynebacterium pyogenes	0.58
72	+	Staphylococcus epidermidis and Escherichia coli	0.72
73	+	Streptococcus agalactiae	0.59
74	+	Staphylococcus aureus	0.68
75	+	-do-	0.90
76	+	-do-	0.72
77	+	-do-	0.91
78	+	-do-	0.61
79	+	Staphylococcus epidermidis	0.50
80	+	-do-	0.60
81	+	-do-	0.61
82	+	-do-	0.50
83	+	-do-	0.61
84	+	-do-	0.61
85	+	-do-	0.52
86	+	Negative	0.60
87*	+	Staphylococcus aureus and Streptococcus agalactiae	0.78
88*	+	Streptococcus agalactiae	0.69
89*	+	Escherichia coli	0.82
90*	+	Staphylococcus aureus	0.97
91*	+	Staphylococcus epidermidis	1.02
92*	+	Streptococcus agalactiae	0.62
93*	+	Staphylococcus epidermidis	1.18
94*	+	Enterobacter aerogenes	0.68
95*	+	Staphylococcus epidermidis	0.97
96*	+	Streptococcus agalactiae	1.02

\* Cl

ly positive cases.

## Appendix II (d)

Results of examination of 21 goat milk samples with CMT ++ score

Sl. No.	CMT score	Cultural examination	Somatic cell count (x 10 <sup>6</sup> /ml)
1	++	Staphylococcus aureus	1.18
2	++	-do-	0.91
3	++	-do-	0.92
4	++	Streptococcus agalactiae	1.32
5	++	-do-	1.41
6*	++	Staphylococcus aureus	1.82
7*	++	-do-	2.17
8*	++	Staphylococcus aureus and Streptococcus agalactiae	1.75
9*	++	Corynebacterium pyogenes	1.81
10*	++	Streptococcus agalactiae	1.95
11*	++	-do-	1.35
12*	++	-do-	1.89
13*	++	-do-	1.85
14*	++	Staphylococcus aureus	2.12
15*	++	-do-	2.46
16*	++	-do-	1.86
17*	++	Staphylococcus epidermidis	1.48
18*	++	-do-	1.81
19*	++	Staphylococcus aureus	1.93
20*	++	-do-	1.95
21*	++	-do-	2.45

\* Clinically positive cases.

Appendix II (e)

Results of examination of 20 milk samples\* with CMT +++ scores.

Sl. No.	CMT scores	Cultural examination	Somatic cell count ( $\times 10^6$ /ml)
1	+++	Staphylococcus aureus	19.0
2	+++	-do-	24.0
3	+++	-do-	24.2
4	+++	-do-	24.8
5	+++	-do-	25.2
6	+++	-do-	21.2
7	+++	Corynebacterium pyogenes	28.0
8	+++	Staphylococcus aureus	24.6
9	+++	-do-	18.2
10	+++	-do-	29.6
11	+++	-do-	19.2
12	+++	-do-	24.5
13	+++	Streptococcus agalactiae	12.5
14	+++	Enterobacter aerogenes	12.8
15	+++	Escherichia coli	20.1
16	+++	Staphylococcus aureus	27.0
17	+++	-do-	25.9
18	+++	-do-	20.9
19	+++	-do-	26.1
20	+++	Enterobacter aerogenes	31.8

\* Clinically positive cases.

Appendix III (a)

Reactions of *Staphylococcus aureus* cultures isolated from clinical mastitis of goats.

Characters	Number of the isolates														
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
1. Haemolysis	+	-	+	-	+	+	+	+	+	+	-	+	+	-	+
2. Acid fastness	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3. Gram reaction	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4. Shape	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
5. Spores	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6. Motility	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
7. Growth in air	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
8. Catalase	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
9. Voges-Proskauer	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
10. Pigmentation	+	+	+	+	+	+	+	+	-	-	+	+	+	+	+
11. Growth on Mannitol salt agar	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
12. Coagulase	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
13. Gelatin liquefaction	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
14. Lactose	+	+	+	+	+	+	+	+	+	-	+	-	+	+	+
15. Mannitol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
16. Glucose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
17. Sucrose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

+ Positive reaction; - Negative reaction; S - spherical.

(contd.....)

Characters	Number of the isolates															
	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31
1. Haemolysis	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+
2. Acid fastness	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3. Gram reaction	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4. Shape	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
5. Spores	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6. Motility	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
7. Growth in air	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
8. Catalase	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
9. Voges-Proskauer	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
10. Pigmentation	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
11. Growth on Mannitol salt agar	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
12. Coagulase	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
13. Gelatin liquefaction	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
14. Lactose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
15. Mannitol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
16. Glucose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
17. Sucrose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

(cont.)

Appendix III (b)

Reaction of *Streptococcus agalactiae* isolated from cases of goat mastitis.

Character	1	2	3	4	5	6	7	8	9	10
1. Haemolysis	B	B	B	B	B	B	B	B	B	B
2. Acid fastness	-	-	-	-	-	-	-	-	-	-
3. Gram reaction	+	+	+	+	+	+	+	+	+	+
4. Shape	S	S	S	S	S	S	S	S	S	S
5. Spores	-	-	-	-	-	-	-	-	-	-
6. Motility	-	-	-	-	-	-	-	-	-	-
7. Growth in air	+	+	+	+	+	+	+	+	+	+
8. Catalase	-	-	-	-	-	-	-	-	-	-
9. Litmus milk	AC	AC	AC	AC	AC	AC	AC	AC	AC	AC
10. Gelatin liquefaction	-	-	-	-	-	-	-	-	-	-
11. CAMP test	+	+	+	+	+	+	+	+	+	+
12. Growth on 10% Bile	+	+	+	+	+	+	+	+	+	+
13. Glucose	+	+	+	+	+	+	+	+	+	+
14. Lactose	+	-	+	+	-	-	-	+	+	+
15. Mannitol	-	-	-	-	-	-	-	-	-	-
16. Sucrose	+	+	+	+	+	+	+	+	+	+

AC = acid clot; B = beta haemolysis; S = spherical.

Appendix III (c)

Reactions of *Staphylococcus epidermidis* isolated from clinical cases of goat mastitis.

Character	1	2	3	4	5
1. Haemolysis	-	-	-	-	-
2. Acid fastness	-	-	-	-	-
3. Gram reaction	+	+	+	+	+
4. Shape	S	S	S	S	S
5. Spores	-	-	-	-	-
6. Motility	-	-	-	-	-
7. Growth in air	+	+	+	+	+
8. Catalase	+	+	+	+	+
9. Voges-Proskauer	-	-	-	-	-
10. Pigmentation	-	-	-	-	-
11. Growth on Mannitol salt agar	-	-	-	-	-
12. coagula	-	-	-	-	-
13. <del>lactose</del> liquefaction	-	-	-	-	-
15. Mannitol	-	-	-	-	-
16. Glucose	-	-	-	-	-
17. Sucrose	+	+	+	+	+
	+	+	+	+	+

S - spherical.

Appendix III (d)

Reactions of *Corynebacterium pyogenes* cultures isolated from goat mastitis.

Character	1*	2*	3*	4	5	6	7	8	9
1. Shape	R	R	R	R	R	R	R	R	R
2. Acid fastness	-	-	-	-	-	-	-	-	-
3. Gram reaction	+	+	+	+	+	+	+	+	+
4. Spores	-	-	-	-	-	-	-	-	-
5. Motility	-	-	-	-	-	-	-	-	-
6. Catalase	-	-	-	-	-	-	-	-	-
7. Haemolysis	+	+	+	+	+	+	+	+	+
8. Gelatin liquefaction	+	+	+	+	+	+	+	+	+
9. Metachromatic granules	-	-	-	-	-	-	-	-	-
10. Glucose	+	+	+	+	+	+	+	+	+
11. Lactose	+	+	+	+	+	+	+	+	+
12. Sucrose	-	+	+	+	+	+	-	-	-
13. Mannitol	-	-	-	-	-	-	-	-	-

\* Clinical positive cases;      R - rod.



Appendix III (c)

Reactions of Gram negative organisms isolated from goat's milk.

Characters	Escherichia coli						Enterobacter aerogenes			Klebsiella pneumoniae		Pseudomonas aeruginosa	
	1	2	3	4	5	6	1	2	3	1	2	1	
1. Acid fast	-	-	-	-	-	-	-	-	-	-	-	-	-
2. Gram reaction	-	-	-	-	-	-	-	-	-	-	-	-	-
3. Shape	R	R	R	R	R	R	R	R	R	R	R	R	R
4. Capsule	-	-	-	-	-	-	+	+	+	+	+	+	-
5. Motility	+	+	+	+	+	+	+	+	+	+	+	+	+
6. Growth in air	+	+	+	+	+	+	+	+	+	+	+	+	+
7. Catalase	+	+	+	+	+	+	+	+	+	+	+	+	+
8. H <sub>2</sub> S on TSI	-	-	-	-	-	-	-	-	-	-	-	-	-
9. Nitrate reduction	+	+	-	+	+	-	+	+	-	-	-	-	+
10. Urease	-	-	-	-	-	-	-	-	-	-	+	+	-
11. Gelatin liquefaction	-	-	-	-	-	-	-	-	-	-	+	+	+
12. Indole	+	+	+	+	+	+	-	-	-	-	-	-	-
13. Methyl Red	+	+	+	+	+	+	-	-	-	-	+	+	+
14. Voges-Proskauer	-	-	-	-	-	-	+	+	+	+	-	-	-
15. Citrate as a carbon source	-	-	-	-	-	-	+	+	+	+	+	+	-
16. Glucose	+	+	+	+	+	+	+	+	+	+	+	+	+
17. Lactose	+	+	+	+	+	+	+	+	+	+	+	+	-
18. Mannitol	+	+	+	+	+	+	+	+	+	+	+	+	+
19. Sucrose	-	-	-	-	-	+	+	+	+	+	+	+	+
20. Starch	-	-	-	-	-	+	+	+	+	+	+	+	+

(Green pigment)

# **STUDIES ON MASTITIS IN GOATS**

**BY**

**VENUGOPAL K.**

## **ABSTRACT OF A THESIS**

Submitted in partial fulfilment of the  
requirement for the degree

## **MASTER OF VETERINARY SCIENC**

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Department of Medicine

**COLLEGE OF VETERINARY AND ANIMAL SCIENCES  
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## ABSTRACT

In the present investigation, milk samples from 56 clinical and 379 apparently normal goats were examined. Staph. aureus was isolated from 52 per cent of clinical cases. The other organisms isolated were, Staph. epidermidis, Str. gas-actiae, E. pyococcus, B. pneumoniae, Esch. coli, Str. carnosus and B. aspergillus. The samples examined from apparently normal goats revealed an incidence of 47 per cent subclinical mastitis. Mastoid Test conducted on milk samples from apparently normal goats revealed that this test was 96.5 per cent efficient in detecting subclinical mastitis. From the somatic cell count performed on milk samples, it was observed that normal milk (SCC zero) had a mean cell count of 0.9 lakhs per ml. The cell counts of 5 to 10 lakhs (SCC +) represented subclinical mastitis. When a comparative study was conducted with Mastoid Test, Mastoid Test and Teepol Mastitis Test it was observed that Mastoid reagent could only be relied upon in detecting subclinical mastitis. Chloramphenicol was found to be the drug of choice in goat mastitis, as evidenced by the in vitro sensitivity tests. Cryomycin, Ampicillin, Terramycin and Penicillin were moderate efficient. Results of the treatment with sensitive drugs on 4 selected clinical cases have been discussed.