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Aetiology of Mastitis in Nili-Ravi Buffaloes of Pakistan

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Mastitis is an acute, sub-acute or chronic infectious, and occasionally contagious disease of udder, invariably caused by bacteria. It results in progressive damage to the glandular tissue, gradual decrease in milk secretion and eventual depreciation of the animal.

It is one of the major hazards faced by persons dealing in milch animals. Economic losses suffered by dairy industry are colossal. Importance of this malady becomes evident from the voluminous literature being put out in the form of books and periodicals. Much more will be written in times to come and many a bacteriologist will devote attention to this scourge, because no effective control or preventive mechanism could so far be perfected.

Most of the information in this regards, however, relates to mastitis in the cow, and there is not much material available about the bacteriological picture of mastitis in the buffalo (*Bubalis bubalis*). According to an estimate buffalo population in Pakistan is about 10.0 million heads and is rapidly increasing. It is the major source of milk supply and in urban areas the buffalo has almost completely replaced the cow (ABDUL WAHID, 1973).

In and around the city of Lahore, there is a large population of "Gujjars", a clan who have for generations been raising milch animals and selling milk. Buffalo herds varying in size, from 10 to 50 or so animals per herd, are found scattered all over the town and thus an unbroken supply of fresh milk is maintained, right at the door-steps of every consumer. General husbandry practices have not suffered much change, since the time first milch animals were domesticated. The only visible improvement appears to have been in the realm of therapeutic and prophylactic facilities which have shown an impact on the improvement of health and production.

In case prompt and adequate Veterinary aid does not become available on the spot, the sick animals are brought to the Veterinary Hospital of the College of Animal Husbandry in the town. Cases suspected of suffering from infectious disease are refered to the Infectious Diseases Ward of the hospital. A preliminary report on aetiology of mastitis in buffaloes examined in the hospital is presented here.

Review of literature

Staphylococcus aureus, Streptococcus agalactiae, St. dysgalactiae, St. uberis, Escherichia coli, Corynebacterium pyogenes, and Pseudomonas aeroginosa, etc. have been described as the usual cause of mastitis in buffaloes (Mohan, 1945; Priestley & Artioli, 1945; Wahby & Hilmy, 1946; Narayanan & Iya, 1953, etc.).

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In domestic buffaloes in India, Dhanda and Sethi (1962) reported 30.5% cases due to Staphylococcus aureus; 7.9% by St. agalactiae, 11.3% by St. dysgalactiae, 17.2% by St. ubris, 4.0% by St. pyogenes, 4.6% by St. zooepidemicus, and 10.6% by St. equisimilus. Kalra and Dhanda (1964) also reported Staphylococci to be the commonest cause of mastitis in this animal, while St. agalactiae and St. dysgalactiae were next in importance as aetiological agents. Out of 1080 buffaloes examined in Egypt, Said and Malek (1968) recorded 135 cases due to Staphylococci, 96 due to St. agalactiae, 37 due to St. dysgalactiae, 33 due to E. coli and 3 due to Corynebacterium.

EL-GINDY et al. (1964) observed *St. agalactiae* to be the most common cause of mastitis in Cattle and Buffaloes in Egypt. According to them Corynebacteria and Staphylococci were next in importance. Higher incidence of streptococcal mastitis was also reported from Pakistan by AIJAZUL HAQ and MAJEED (1965) after examining 22 buffaloes.

Material and Method

During a period of one year, between 1965-66, milk samples were obtained from domestic buffaloes, received at the Hospital for the treatment of mastitis. Samples were collected from cases which had not received any treatments. Attempt was always made to collect about 10.0 ml of milk from individual quarters in separate sterile sample bottles, under as strict aseptic precautions as possible.

A total number of 740 samples were thus collected from 194 cases. Immediately after obtaining the samples, they were taken to the adjoining laboratory for processing. Blood agar was used for primary isolation of all organisms except Streptococci. Sodium azide crystal violet blood agar base containing 5.0 % defibrinated ox blood was used for primary isolation and subsequent culturing of the Streptococci.

Staphylococcus medium No. 110 was used for maintaining and studying the colony characters of Staphylococci. EMB agar served to separate the lactose fermenters from the non-fermenters. Selective and differentiating media were used where necessary. Rabbit plasma was utilized for coagulation study.

Fresh milk smears were made from each sample and stained with Newmann's stain for the presence of bacteria. After incubation at 37.5 °C for 24 hours, the milk was spread into smears and stained with Gram's system. A loopful of gravity cream from each sample was streaked on Sodium Azide Crystal Violet Blood Agar. Usual bacteriological techniques of culturing, sub-culturing and identification were utilised.

Results

Etiological agents isolated are summarised in Fig. 1 along with their characteristics. Out of 740 samples examined, 326 (44.0 $^{0}/_{0}$) proved positive for *Staphylococcus aureus*. Alpha, Beta and Gamma haemolysis was shown by 1.6 $^{0}/_{0}$, 41.1 $^{0}/_{0}$ and 1.3 $^{0}/_{0}$ isolates respectively. Rabbit plasma was coagulated in one hour by 33.8 $^{0}/_{0}$, in three hours by 6.2 $^{0}/_{0}$ and in twenty four hours by 0.6 $^{0}/_{0}$ isolates. 43.2 $^{0}/_{0}$ isolates liquified gelatine in 72 to 120 hours.

Streptococci were isolated from 310 (41.9 %) samples, 30.0 % yielding St. agalactiae, 8.7 % St. dysgalactiae, and 3.2 % St. uberis. Alpha, narrow zone Beta, wide zone Beta and Gamma haemolysis was exhibited by 4.1 %, 4.1 %, 4.1 %, and 30.2 % isolates respectively.

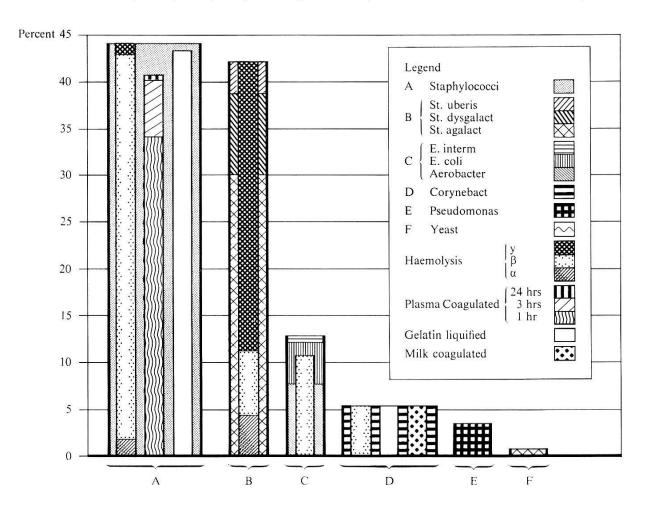
Coliform organisms were isolated from 92 (12.4%) samples. 10.8% isolates produced wide zone Beta haemolysis. On EMB agar all isolates produced typical

colonies with fish-eye centre and metallic sheen all over. $7.6^{\circ}/_{0}$ isolates were identified as *Aerobacter aerogenese*, $4.3^{\circ}/_{0}$ as *E. coli*, and $0.5^{\circ}/_{0}$ as *E. intermedium*.

Corynebacterium pyogenese was isolated from 38 (5.1%) samples. All isolates coagulated milk, liquified gelatine and exhibited narrow zone Beta haemolysis.

Pseudomonas aeruginosa was isolated from 24 $(3.2^{\circ}/_{\circ})$ samples giving luxurient growth on all media, colonies being translucent and spreading. Thick pellicle, turbidity and heavy sediment was produced in nutrient broth with bluish green coloration of the medium.

Three (0.4%) samples yielded yeast cells, which resembled Candida tropicalis.



Discussion

In the study it was observed that Staphylococcus was the most important organism involved in the causation of mastitis in buffaloes. Streptococci came next. Coliform organisms were not uncommonly met with, while Corynebacteria and Pseudomonas were also occasionally found. The findings agree in general with other workers investigating mastitis in buffaloes. The fact that infection due to Staphylococci exceeded that of the Streptococci could probably be ascribed to the reason that mastitis Streptococci are highly sensitive to commonly used antibiotics. As already mentioned use of intramammary antibiotics is quite easy in the town from where these buffaloes mostly came. This might have been a factor that helped to keep down the streptococcal infection, a probability that has been recognised in bovine mastitis. Boddle (1969) stated that at one time 80% of streptococcal infection in cows was due to *St. agalactiae*, but with the use of antibiotics the proportion has been markedly reduced. White and Jordan (1963) observed that with the widespread use of penicillin and improved control

measures, the indidence of contagious bovine mastitis due to *St. agalactiae* has greatly diminished. According to them among the samples from clinical cases in cows *St. agalactiae* mastitis was $44\,^{0}/_{0}$ in 1942–43, while the same dropped to $3.9\,^{0}/_{0}$ in 1956. Predominance of staphylococcal mastitis has been reported by BAGADI (1970) and LEE and FROST (1970) in cows as well.

While cows are comparatively neat animals, the mud wallowing habit of buffaloes offers ample opportunities for the infection to be picked up through teat canal. The udder is also comparatively pedulous with long teats. However, sphincters of buffalo teat carry more smooth muscles fibres and blood vessels than that of the cow, and thus provide a better barrier against infection (Krishnaswamy, 1965). Information, however, is not available regarding the relative susceptibility of buffaloes to mastitis as compared to cows.

Inspite of tight sphincters the teats are said to be "patent" and offer ease of milking. According to Dodd and Neave (1951) such teats are considered more susceptible to staphylococcal infection. Murphy et al. (1954) described an association between infection and the conformity of teat canal and anatomical variations in apex of teat. In view of such factors, a comparative study on mastitiscomplex in cows and buffaloes is indicated.

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