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Caprine mastitis due to combined infection of cryptococcus neoformans and coagulase-negative staphylococci (CNS)

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Abstract
Goat intramammary (mastitis) infections are a major threat to animal health, changing the composition of milk, reducing its hygienic value, affecting its processing qualities, and resulting in significant financial losses for dairy farmers today. The objective of the present study focuses on the isolation, identification of combined mastitis infection causes by *Cryptococcus neoformans* and Coagulase-negative staphylococci (CNS). The study was investigated by the clinical sample taken form infected animal under the treatment at TVCC, Mannuthy, Thrissur. *Cryptococcus neoformans* conformed through the mouse inoculation method. Staphylococcus is conformed by culture, biochemical tests and sugar fermentation tests.

Keywords: Cryptococcus neoformans, coagulase-negative staphylococci, goat, mastitis

Introduction
Mastitis in goats is a serious animal health issue that affects the composition, hygienic value and processing qualities of milk (Stuhr and Aulrich 2010) [10]. Many animals could be infected with cryptococcosis, that had been associated to pneumonia and mastitis in cattle and goats. Even though, *cryptococcus neoformans* had frequently been identified as spontaneous mastitis occurrences in dairy animals, particularly goats in India (Sharma, 1983) [10]. Staphylococci are also commonly isolated from intramammary infections (IMI), and as a result, they are regarded as the primary causative agents of goat mastitis (Bergonier *et al.*, 2003) [1]. Staphylococcus aureus is regarded as a prominent infectious pathogen within this category. CNS is a pathogenic agent that causes chronic, subclinical, and clinical mastitis as well as gangrenous mastitis, which frequently results in the loss of the afflicted gland. For these reasons, it must be regarded as a significant agent of caprine mastitis. The present study's focus is to isolation and identification of combination infections with CNS and *Cryptococcus neoformans* that induce mastitis.

Materials and Methods
A four year old goat with mastitis was presented to TVCC, Mannuthy, Thrissur during December 2022. Milk samples were taken for the investigation of the causative agents. The sample was cultured on brain-heart infusion agar (BHIA) and blood agar (BA), then incubated at 37º C for 24-48 hrs for the isolation of bacteria.
However, sample were aslo cultured in Sabouraud dextrose agar (SDA) and BA 37º C for 2-5 days anaerobically for the identification of yeasts. The colonies were identified using staining and biochemical methods (Koneman *et al*., 1983; Quinn *et al*., 1994) [6, 9]. Negative staining using Indian ink and mouse inoculation was carried out on the conformation of yeast.

Result and Discussion
In BHIA the organisms showed small, yellowish colonies and flat, moist and shiny mucoid colonies. In BA with normal conditions, colony showed beta-haemolytic and anaerobic BA had mucoid, flat non-haemolytic colony. On SDA organisms exhibit cream-colored smooth, mucoid, yeast-like colonies. This was the identification of the presence of bacteria and yeasts.
On gram staining, the bacterial cell is identified as gram-positive with a tiny, spherical, grape-like clustering coccus that is immobile.

Furthermore, other round cells with gram-positive granules, are dark-walled, and budding was observed.

The biochemical and sugar fermentation data of the bacteria revealed that *Coagulase-negative staphylococci (CNS).* For the conformation of yeast, experiment demonstrated in mouse inoculation intraperitoneally. Capsulated Cryptococcus neoformans were observed in the negative staining of the impression smear of heart.

CNS are primarily responsible for subclinical mastitis; less pathogenic than *Staphylococcus aureus* (*S. aureus*), it can nevertheless result in long-lasting clinical infections in the mammary gland. (Contreras *et al*., 2003; Bergonier *et al*., 2003; Contreras *et al*., 2007) [3, 1]. Staphylococcus species-related mastitis in goats is regarded as a reservoir for enterotoxins that pose a health concern to humans (Contreras *et al*., 2007; Podkowik *et al*., 2013) [4, 8]. Although many staphylococci species are capable of producing these enterotoxins, limited is understood about the CNS's capacity for enterotoxicity (Podkowik *et al*., 2013) [8].

Goats had been reported to be occasionally infected with cryptococcosis. Neural granulomas are among the clinical symptoms and meningoen- cephalitis and abortion (Blanchard and Filkins, 1992) [2] and sinusitis (Scott *et al*., 1974), pneumonia (Hilbert *et al*., 1980) [5]. Mastitis in dairy cattle is infrequently caused by *Cryptococcus neoformans*. There have been isolated reports of avian cryptococcosis (Malik *et al*., 2003) [7].

Conclusion

In this study, Coagulase-negative staphylococci (CNS), *Cryptococcus neoformans* was isolated form goat mastitis. Identification of CNS were carried out by biochemical and sugar fermentations tests. *Cryptococcus neoformans* was identified by negative staining carried out by using impression smear from mouse.

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Author's Contribution

Not available.

Conflict of Interest

Not available.

Financial Support

Not available.

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