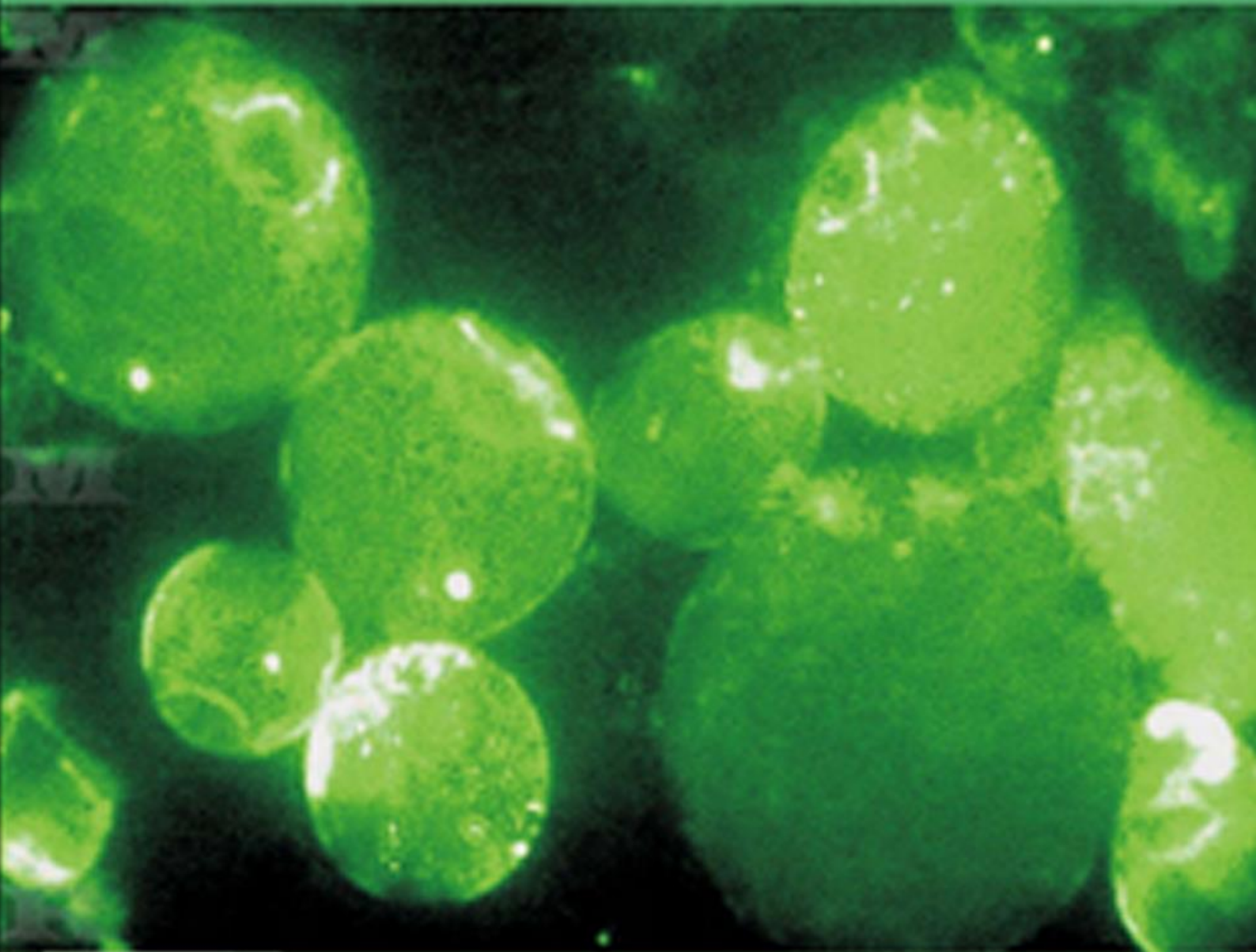


Molecular Microbiology Research

ISSN 1927-5595 (Online)



MMR 2015, Vol.5

<http://mmr.biopublisher.ca>

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Sophia Publishing Group

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Clinical Bovine Fungal Mastitis in Organized Dairy Farm

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Molecular Microbiology Research, 2015, Vol.5, No.5 doi: 10.5376/mmr.2015.05.0005

Received: 21 Jul., 2015

Accepted: 21 Sep., 2015

Published: 10 Oct., 2015

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Preferred citation for this article:

Ghodasara S.N. and Gajbhiye P.U., 2015, Clinical Bovine Fungal Mastitis in Organized Dairy Farm, Molecular Microbiology Research, Vol.5, No.5 1-3 (doi: [10.5376/mmr.2015.05.0005](https://doi.org/10.5376/mmr.2015.05.0005))

Abstract A study was conducted to find out the incidence of various mastitis causing organism in cattle of organized dairy farm. Out of total 44 clinical mastitis samples 13.64% were found to have fungal mastitis and 77.26% were found to have bacterial mastitis. The yeast and yeast like fungi isolated were *candida spp.* and *Aspergillus spp.* Good hygiene, sanitation and managerial practices of farm animal and workers and judicious use of antibiotics will lower incidence of bovine mycotic mastitis.

Keywords Bovine; Fungal mastitis; Dairy farm; Managerial practices

1 Introduction

Mastitis is caused by multi etiological agent includes bacteria, mycoplasmas, viruses, fungi and algae (Kivaria and Noordhizen, 2007; Wellenberg et al., 2002). According to literature data, fungal infections account for 2%–13% of all cases of mastitis in cows (Krukowski et al., 2006; Krukowski, et al., 2000). Sometimes, their incidence is much higher or they are enzootic. It is widely prevalent in organized as well as unorganized dairy herds and associated with a significant loss of milk yield resulting in increased costs of production and treatment, which also deteriorated quality of milk and milk products (Arshad et al., 1998). Amongst fungal mastitis *Candida* is the most common species isolated from cases of mastitis in bovines (Radostitis, 1995). Many a times mycotic mastitis is unnoticed by clinician in first attempt of treatment and administration of antibiotics may aggravate fungal mastitis as some of the antibiotics like penicillin and tetracycline act as a source of nitrogen for various species of fungi (Meek, 1981). There for treatment of fungal mastitis is a challenge as many of these fungi do not respond to the antibiotics rather they use some of the antibiotic like tetracycline as their source of energy (Tarfarosh and Purohit, 2008). Due to this reason most of the mastitis cases remains incurable and source of infection for other adjoining animals.

Keeping in view, present work is undertaken to evaluate a prevalence of fungal mastitis in cattle at organized dairy farm (Cattle Breeding Farm, JAU, Junagadh).

2 Result

Out of total 44 samples examined, 6 samples yielded fungal agents, 34 samples were positive for bacteria and the remaining 4 were negative for fungal as well as bacterial agents (Table 1).

In this study, the incidence of fungal agents in clinical cases of mastitis was 13.64% (6). Out of 6 isolates of fungi, 2 isolates yielded smooth white or yellowish, cottony colonies having resemblance with *Candida spp.* (Figure 1). Upon microscopic examination these isolates showed oval shaped budding cells and were tentatively identified as *Candida spp.* Remaining 4 isolates revealed rapid growing colonies with greenish or black pigmentation, showing resemblance with

Table 1 Per cent distribution of bacterial and fungal isolates from clinical mastitis in Gir cattle

Number of samples positive for	Number	Percentage
Fungi (Yeast)	6	13.64%
Bacteria	34	77.26%
No growth	4	9.1%
Both (Bacteria and fungi)	0	0%
Total	44	100%



Figure 1 white or yellowish, cottony colonies having resemblance with *Candida spp.*

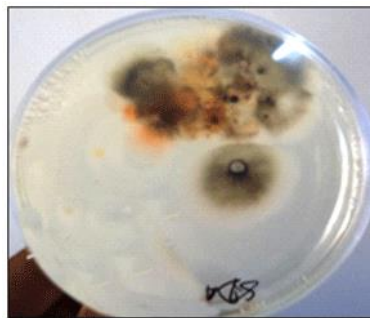


Figure 2 Greenish or black pigmentation, showing resemblance with *Aspergillus spp.*

Aspergillus spp. (Figure 2) and were revealed non-septate conidiophore with black spore heads.

3 Discussion

The incidence of mycotic infection in clinical cases of mastitis was 13.64% (6). Similarly, the same incidence rate of fungal mastitis (13%) were reported by Sukumar and James, 2012 and higher rates of incidence 29.27% and 34% were reported by Simaria and Dholakia, 1986 and Sudarwanto, 1987, respectively. The incidence of mastitis due to fungi have also been reported by various workers (Zaragoza et al., 2011; Ranjan et al., 2011; Lagneau et al., 1996; Singh et al., 1992; Shah et al., 1986; Sharma et al., 1977). Out of 6 isolates of fungi, 2 isolates yielded smooth cottony colonies with budding cells resemblance with *Candida spp.* of fungi as described by Pachauri, et al., 2013. Similarly, Sukumar and James (2012), (Gupta et al., 1981) and Simaria and Dholakia (1986), reported relatively more frequent isolation of *Candida tropicalis* from mastitis udder than any other species of *Candida*. Remaining 4 isolates having greenish or black pigmentation colonies with non-septate conidiophore resemblance with *Aspergillus spp.* which were also described by Pachauri, et al., 2013. Other researcher have also reported *Aspergillus spp.* as a one of the causative agent for bovine mastitis (Blowey and Edmondson, 2010; Stephan et al., 2000)

The incidence of fungal mastitis in this study may be due to unhygienic condition of the animal sheds and high humidity along with favorable environmental conditions supporting growth of fungal spores. Thereby that increases the chances of fungal spore to enter into the udder which provide suitable environment to these fungi for their growth (Elad et al., 1995; Williamson and Di Menna, 2007). Second probable

cause is extensive and indiscriminate use of antibiotics for treatment of mastitis, because some of the samples during the study were collected from the mastitic cases, which had been unresponsive to antibiotics treatment.

According to Loftsgard and Lindquist (1960), *Candida spp.* utilizes nitrogen from penicillin and tetracycline antibiotics. The use of such antibiotics encourages establishment of the infection by damaging the mammary epithelium. Similarly, during this study, it was observed that initially, the use of antibiotics worsened the condition of clinical mastitis. Chahota et al., (2001) documented that association of fungi (*G. candidum*) with mastitis is greater in those patients which have been subjected to prolonged irrational antibiotic therapy, as is evident in this case.

The incidence of mycotic mastitis may be due to development of antibiotic resistance in the bacteria which prolongs treatment and favoring growth of fungal spp. such as *Candida* and *Aspergillus* to infect as secondary invader, due to physiological change in udder and milk. The managerial practices adopted at Cattle Breeding farm, like discarding first few strips of milk on ground while milking of animals as well as during treatment of mastitic animals and reluctant to disinfect hand between milking by milkers may contributes as potent source of lateral transmission of infection in farm. The same were reported by Pachauri et al., 2013. Further detailed investigation is required in relation to pattern of antibiotic therapy advocated as well as managerial and hygienic practices of farm animals and workers.

4 Materials and Methods

Forty four milk samples were collected from cows showing clinical mastitis from cattle Breeding Farm,

Junagadh Agricultural University, Junagadh, Gujarat, India. The affected quarter milk was sampled aseptically and transported immediately at 4 °C to the laboratory. A heavy inoculum of thoroughly mixed mastitis milk was inoculated on 5% Ox Blood Agar (OBA) and Sabouraud Dextrose Agar (SDA) and incubated at 37 °C for 24-48 h and at room temperature for 2-7 days, respectively. The inoculum inoculated of SDA were incubated at room temperature and examined for growth every day (24, 48, 72 hrs up to week) after which plates showing no growth were considered as negative (Tarfarosh and Purohit, 2008). The fungal colonies were studied for their cultural and morphological characteristics. The morphological characteristics were noted after staining with Gram's and lacto phenol cotton blue stain. Colonies obtained on OBA were transferred to MacConkey Agar (MA), Eosin Methylene Blue Agar (EMB) and Nutrient Agar (NA) for further identification. Colonies were identified morphologically by Grams staining and Biochemical identification was done using 3% Potassium Hydroxide (3% KOH), catalase and oxidase test as per the methods described by Carter (1994) and Cowan and Steel (1974) (result not shown in this article).

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