Simultaneous infections of *Microsporum audouinii* and *Fusarium oxysporum* in Buffaloes

P.I. Ganesan[1*] N. Shweta[2] & C. Archana[3]

[1]Department of Veterinary Public Health, [2]Department of Animal Nutrition, [3]Department of Veterinary Pharmacology & Toxicology School of Veterinary and Animal Sciences, Centurion University of Technology and Management, Paralakhemundi, Odisha, India
[*]Corresponding author e-mail: drpiganesan@gmail.com

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Abstract

A study was carried out to assess the prevalence of dermatophytes infections in buffaloes at a dairy farm in Paralakhemundi, Odisha State accommodating 10 Holstein-Friesian cows, 12 Murrah buffaloes, and 6 Murrah calves of varying age groups. Several Murrah buffaloes and calves showed extensive skin lesions distributed across their bodies. In response, a clinical examination was conducted on all buffaloes and their calves, followed by the collection and analysis of skin scrapings to identify the underlying causative agents. Adult buffaloes and calves exhibited clinical signs including hair loss, grayish patches, alopecia, and skin scaling. Lesions typically appeared as scaly, hairless patches located predominantly around the neck, chest, face, dewlap, lower abdominal region, and the area between the jaws. Laboratory examination of the collected samples confirmed the presence of *Microsporum audouinii* and *Fusarium oxysporum*. This section of the study also explores the potential risk factors contributing to the occurrence of these fungal pathogens in the skin samples of the affected Murrah buffaloes.

Key-words: Buffaloes; concurrent infection; Microsporum; Fusarium oxysporum

Introduction

Buffaloe is important livestock of rural community in developing countries providing milk, meat and hide (Alshdaifat et al., 2024). It is robust and climate resilient livestock, but prone to certain diseases (Bhoi et al., 2021). Dermatophytosis is a transmissible fungal infection caused by a group of fungi collectively known as dermatophytes, which commonly inhabit living tissues and soil. Dermatophytes are categorized based on their preferred hosts as anthropophilic, zoophilic and geophilic species. Both zoophilic and geophilic species are capable of transmitting infections from animals to humans, and vice versa. Zoophilic species, such as *Microsporum canis*, are known to infect both animals and humans and have also been identified in ruminants. Fungal infections are frequently observed in healthy and immunocompetent hosts due to the ubiquitous nature of fungi in the environment (Kohler *et al.,* 2015; Gnat *et al.,* 2020). Both opportunistic and primary fungal pathogens have been detected in low numbers in animals and humans and are often associated with recurring infections (Fisher *et al.,* 2012; Bishnoi *et al.,* 2018; Shenoy & Jayaraman, 2019). Among the anthropophilic dermatophytes, well-documented species include *Trichophyton rubrum*, *T. tonsurans*, *T. violaceum*, *T. schoenleinii*, *Microsporum audouinii*, and *Epidermophyton floccosum*. *Microsporum audouinii* infection in buffalo calves and co-infection with *Mucor* species have been reported in Rajasthan (Sravani & Ganesan, 2024). In light of these earlier findings, the present investigation was undertaken at an organized dairy farm located in the Gajapati district of Odisha to identify the fungal pathogens responsible for dermatological lesions in the buffalo population.

Materials and Methods

Location of the study area and work place

The organized dairy farm in Paralakhemundi, Odisha State housed 10 cows, 12 Murrah buffaloes, and 6 Murrah calves of various age groups which were maintained under similar nutritional and management conditions. The work was carried out in the department of Veterinary Public Health, School of Veterinary and Animal Sciences, Centurion University of Technology and Management, Paralakhemundi campus, Odisha state.

Clinical examination of the affected buffaloes and their calves

Clinical examination of the adult buffaloes and calves exhibited hair loss, grayish patches, alopecia, and skin scaling. Hence clinical diagnosis of the affected animals was performed to know the etiological agents for dermatophytosis.

Laboratory procedure of the cutaneous fungal lesions for culture and staining technique

Media preparation:

Sabouraud Dextrose Agar (SDA) is a specialized selective media for identifying pathogens from skin, hair, and nails. Media was prepared by suspending 65 g of SDA in one liter of distilled water and allowed to dissolve completely. After sterilization in autoclave, media was allowed to cool for 45-50°C and poured into Petri dishes and allowed for solidification.

Inoculation of skin specimens:

The collected skin materials were implanted into the SDA agar under aseptic conditions by streaking and allowed for 37°C incubation for 1-4 weeks. The fungal morphology was examined using Lacto-phenol cotton blue staining, following the method described by Ellis *et al.* (2013).

Results

Morphological description

Microsporum audouinii* and *F.oxysporum

The skin scrapings collected from the ailing animals subjected to culture studies using SDA revealed *M.auduoinii*.(**Fig.1**) The colonies are flat, cottony with greyish-white, spindle-shaped macroconidia and clavate microconidia with external walls. The morphology of the culture and tests with lactophenol cottonblue and rice grain color pigmentation studies for species wise identification of *M.auduoinii* carried out by rice culture exposure method which differentiated *M.canis* non-sporulating strain. The *F.oxysporum* colonies produced both micro and macroconidia. Macroconidia are two to several celled fusiform to sickle-shaped, elongated apical cell pedicellate basal cell. Microconidia are one or two celled smaller than macroconidia, straight or curved **(Fig.2**) (Ellis *et al*., 2013).

Fig. 1 ***M. auduoinii***

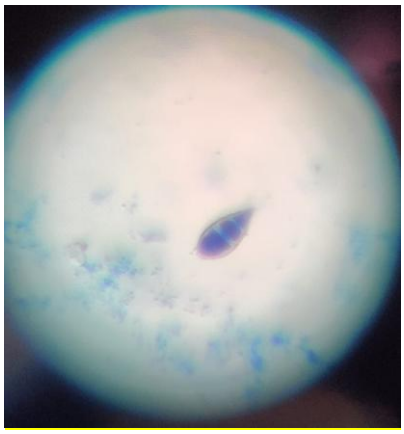

Fig. 2 ***F. oxysporum***

Discussion

Medical investigation of the buffaloes

Current research focused on buffaloes and their calves exhibiting clinical symptoms such as alopecia, scaling, hair loss, and greyish-white lesions. According to Sravani and Ganesan (2024), skin lesions in buffaloes were commonly located on the neck, chest, face, dewlap, lower abdomen, and inter-maxillary areas. In alignment with these observations, animals in the present study displayed similar signs, including white patches, scaling, and hair loss.

Etiological agents and their association for skin disorders

Emenuga and Oyeka (2013) isolated *Trichophyton verrucosum*, *Trichophyton mentagrophytes*, *Microsporum gypseum*, *Candida albicans*, *Fusarium solani*, and *Aspergillus* species from skin samples of sheep and goats. Gawaz and Weisel (2018) highlighted the co-occurrence of yeast-like fungi and dermatophytes within the same lesion. Zhang *et al.* (2020) reported that the shifts in microbial community occurs due to piled up dairy manure. It was reported that the prevalence of highly pathogenic communities in fecal samples in dairy breeding farm occurs and the pathogenic genera significantly increased in manure pits, and these included *Bacillus, Alternaria* and *Fusarium* species. *Fusarium oxysporum* (F.O) is a fungus infect animals, including cows. Mycotoxins produced by these organisms suppress the innate immune pathways in both plants and animals and it is attributed to low milk production. (Nag *et al.,* 2022). Abd El Haleem *et al.* (2018) reported the prevalence of *Fusarium oxysporum* in buffalo's milk and their products. Various strains of *Fusarium oxysporum* both pathogenic and non-pathogenic forms infect both invertebrates and vertebrates (Debourgogne *et al.,* 2016). While *F. oxysporum* is primarily recognized as a plant pathogen, it can also infect animals. The buffaloes infected with *F.oxysporum* suffered with the clinical signs of necrosis followed by gangrene of the dependent parts and reduced milk production and draught power. These studies revealed the prevalence of *Fusarium oxysporum* as a co-pathogen along with other dermatophytes which indicated the opportunistic pathogen status of these organisms in the affected domestic animal population.

Attributed risk factors associated for skin disorders

The attributed risk factors associated for such infection from opportunistic pathogen status to pathogenic status included compromised immune status of the affected animal population, damage to the structural integrity of the cutaneous system of the infected animal population, level of mycotoxins developed by the organisms to suppress the innate immune pathways in animals and plants, level of transfer of pathogenicity related traits from non-pathogenic to pathogenic *F. oxysporum* among species and the level of contamination of the plant sources. Contamination of bedding materials within the dairy shed, a persistent population of buffaloes carrying dermatophyte and fungal infections, continual fungal contamination of the shed environment, and a year-round climate characterized by alternating hot and cold conditions, which favored fungal growth and persistence. This study was carried out at a dairy farm situated in R. Sitapur village, Paralakhemundi, Odisha region with harsh climatic conditions that may favor the persistence and adaptation of dermatophytes and other fungal organisms in animals. The results observed in this study align with these environmental influences.

Conclusions

Clinical evaluation, along with culture and staining of skin lesions in buffaloes, confirmed infections caused by *Microsporum audouinii* and *Fusarium oxysporum*. The warm and humid conditions within the dairy environment likely facilitated the spread and persistence of these fungal pathogens. Further research is necessary to characterize these organisms at the molecular level and to investigate host-related factors contributing to the development of such dermatological conditions in livestock.

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