PREVALENCE, DIAGNOSIS AND THERAPEUTIC MANAGEMENT OF MALASSEZIA INFECTION IN DOG AND CAT

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PREVALENCE, DIAGNOSIS AND THERAPEUTIC MANAGEMENT OF MALASSEZIA INFECTION IN DOG AND CAT

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Statement of Author

I, Sayma Mahmud Moon, certify unequivocally that I have performed all the tasks detailed in this report. The data was gathered from Field, books, national and international periodicals, and other sources. All citations have been properly acknowledged. Consequently, I am solely responsible for collecting, manipulating, preserving, and publishing all data compiled in this report.

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The Author

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Abstract

Malassezia yeasts are superficial normal commensals and opportunistic pathogens of the ear and skin for many warm-blooded animals. The aim of the study was to analyze the prevalence of Malassezia infections with or without bacterial infections in dogs and cats as well as the efficacy of treatment protocol. A study of 203 Malassezia infection cases with or without bacterial infection in dogs and cats was carried out over a 2-years period at teaching and training pet hospital and research centre, Dhaka. Malassezia infection was isolated from 60% dogs and 40% cats. Males were found more prevalent than females. About 45.81% more infections were confirmed in adult age (3-6 years) than any other age group. Local dog breeds were the rich number for occurrence of Malassezia dermatitis and in case of cat, the infected rates recorded more in exotic Persian cat breeds. Malassezia infection rate was higher in ear pinnae and canals (27.59%) followed by ventral trunk (23.15%), dorsal trunk (14.78%), generalized dermatitis (14.29%), arm pits (6.90%), neck region (5.42%), perenial region (4.43%) and interdigital area (3.45%), respectively. Free roaming patients developed Malassezia compared to indoor patients. Cytological examination revealed small footprint or pear shaped Malassezia spores under microscope as well as bacteria in mixed infection cases and considered as reliable diagnosis. Meanwhile, bacterial infection along with Malassezia was isolated from 57% patients and otitis externa encountered from 33% patients. Topical therapy of antiseptic, antibiotic and antifungal based preparation provided. And a range of systemic treatment was indicated only in severe disease processes. Treatment protocol was effective instead of those who had chronic infection.

Keywords: Malassezia infection; bacterial infection; otitis externa; cytological examination; dogs; cats.

Chapter 1: Introduction

The lipophilic yeasts of the genus Malassezia are very much familiar to affecting the skin and mucosa of warm-blooded vertebrates (Bond et al., 2020; Bajwa, 2023; Sierra et al., 2000; Theelen et al., 2018). Malassezia is considered as an opportunistic pathogen under certain conditions and causes dermatitis and otitis externa in dogs and cats. About 18 Malassezia species currently described and 11 have been detected in cats and nine in dogs by culture and using molecular methods (Hobi et al., 2022).

Malassezia have been associated with a variety of pathological conditions in animal's causes otitis externa, dermatitis and occasionally other clinical presentations such as paronychia and keratitis in dogs and cats (Morris, 1999; Machado et al., 2011; Ledbetter and Starr, 2015; Bajwa, 2017; Guillot and Bond, 2020; Spatz and Richard, 2020; Hobi et al., 2022). Not only yeasts belonging to Malassezia genus but also bacteria, mainly Staphylococcus spp., Streptococcus spp., Proteus spp., Pseudomonas spp., Escherichia coli, are frequently involved in the occurrence of otitis externa and dermatitis in carnivores (Rosser, 2004; Zur et al., 2011; Bugden, 2013).

Malassezia dermatitis e lesions are usually first seen on the abdominal skin with severe inflammation characterized by erythematous and/or greasy lesions, especially when lesions involve intertriginous areas and then the lesions may spread to the entire abdomen, the axilla and the inguinal region (Larsson et al., 1988). Hyperpigmentation and lichenification are frequently found in animals with chronic disease conditions. In diseased ears, however, Malassezia can be readily found alone or associated with bacteria that show erythematous vertical ear canals and pinnae with varying degrees of lichenification and scaling, accompanied by a yellow or brownish creamy discharge.

The diagnosis of Malassezia dermatitis primarily relied on clinical signs, presence of elevated numbers of yeast organisms in lesioned skin and a clinical and mycological response to antifungal therapy. Although in veterinary practice, antifungal susceptibility testing is not widely carried out but some reported work has been noted against antifungal resistance of Malassezia organisms and resistance mechanisms involving efflux pumps, fungal wall sterol metabolism, and biofilm formation are recognized (Uchida et al., 1994; Figueredo et al., 2013; Cafarchia et al., 2015; Bumroongthai et al., 2016; Iatta et al., 2017; Kim et al., 2018; Peano et al., 2020; Hobi et al., 2022). Malassezia organisms can easily be identified on cytological preparations of

affected skin or otic discharge as small “peanuts” or “foot-prints” shaped organisms with a diameter of around 3–8 μm (Bond et al., 2020; Bajwa, 2023). The tape strip technique for Malassezia diagnosis is convenient and reliable: by this method a piece of clear adhesive tape is pressed onto the surface of the skin, to collect stratum corneum cells and any superficial microbes. Culture or DNA-based techniques diagnostic methods are typically reserved for research purposes (Bond et al., 2020).

Treatment option for Malassezia infection includes application of a topical antiseptic and/or antifungal preparation. Antifungal drugs combined with antibiotics and a glucocorticoid, reflecting the need to control concurrent bacterial infection and reduce inflammation and proliferative pathologic changes. Systemic azole antifungal therapy (principally clotrimazole, miconazole, ketoconazole or posaconazole, nystatin or terbinafine) is reserved for severe disease or for chronic cases not responding to topical treatment alone (Bond et al., 2020; Guillot and Bond, 2020; Hobi et al., 2022; Bajwa, 2023). Therapeutic management of any underlying diseases is critical to prevent relapse (Bond et al., 2020; Hobi et al., 2022; Bajwa, 2023).

Although Malassezia organisms are predominantly adapted to animals, nosocomial systemic infection has been reported in humans (Welbel et al., 1994). Transmission of Malassezia infection occurs to immunocompromised neonatal patients by health care workers who own pets (Chang et al., 1998).

Skin and otic infection due to Malassezia species with or without bacterial infection in dogs and cats are identified with upward trend frequency in veterinary practice in Dhaka city, Bangladesh. The present study was designed to show the statistical overview of Malassezia infection of dogs and cats in defined areas as well as clinical presentation, isolation and its successful management.

Objectives:

The sole purpose of the study was to determine the factors associated with Malassezia Dermatitis. By the study, clinician and owner will know the proper management steps to prevent Malassezia Dermatitis. Clinicians can know which breeds are at high risk of malassezia dermatitis, which will help them to make appropriate diagnosis and specific treatment.

Chapter 2: Materials and methods

2.1 Study area and duration of study:

The detailed data records of Malassezia affected dogs and cats with or without bacterial infection treated at the teaching and training pet hospital and research center, CVASU, Bangladesh were analyzed for two years (January, 2022 to December, 2023). A total of 203 Malassezia cases of dogs and cats which were exposed to medical treatment were evaluated in this study.

2.2 Study design:

The recorded Malassezia affected dogs and cat's cases were classified into different categories according to species, breed, age, source of animals, affected parts of the body and with or without bacterial dermatitis. On the other hand, the samples from affected parts by scrapping or using sterile cotton buds were collected to make a smear on the slide for cytological study.

2.3 Cytological study:

Sample smears fixated with methanol were stained with Giemsa Stain for detection of *Malassezia* spores with or without bacteria in affected body parts. After that, the stained slide examined under 10x and 100x objects magnification of light microscopy.

2.4 Treatment protocol:

Specific treatment was given to the patients based on clinical findings as well as cytological study.

2.5 Data analysis:

Data obtained from the study was inserted to the Microsoft Excel-2007 and then transferred to the statistical software STATA-11 for calculating the percentage of different variables.

Chapter 3: Results and Discussion

Total 203 samples were recorded with symptoms of scratching of affected parts, shaking of head, erythema, blackish discoloration of skin and bad odor which were linked to Malassezia dermatitis in dogs and cats. A summary of the information regarding Malassezia infection for dogs and cats included in the study is presented in bellow tables and charts.

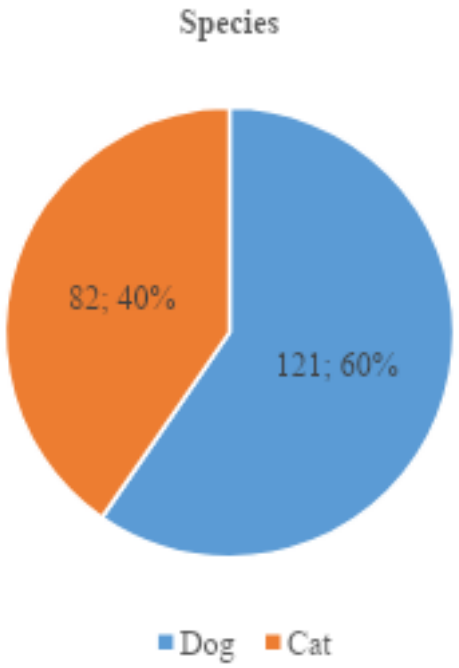

Fig-1: Occurrence of Malassezia infection according to species

According to our observed data, we found 60% of dogs and 40% of the cat's population having the Malassezia infection. Dogs constitute a generally high percentage of infection due to unfavorable environmental conditions in the case of outdoor ones. Some articles showed that dogs were the higher prevalence for Malassezia infection than in cats while other papers found cats were the superior percentage than in dogs (Cafarchia et al., 2005). Our data largely differ from those of Nguyễn et al., (2023) who have reported Malassezia infection in dogs is 91.7%, higher than 8.3% in cats. We collected data from dogs and cats that were not in good hygienic

conditions and also ear canal was not cleaned in the local area. So, data findings may vary widely to the other researcher findings.

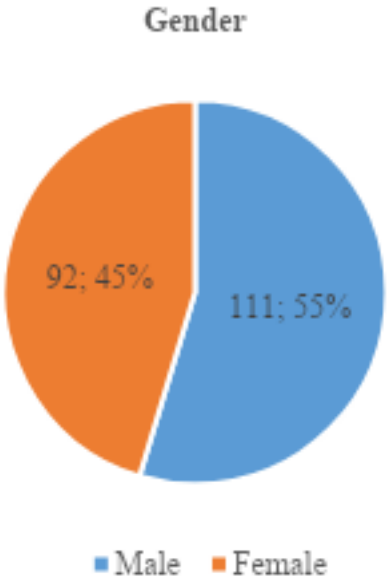

Fig-2: Occurrence of Malassezia infection according to gender

Malassezia infection accounted in males more than in females which were 55% and 45%, respectively. This finding are almost similar to the study of Nguyễn et al., (2023) and they reported 54.2% Malassezia infection in males and 45.8% in females. Another study, Dhoot et al. (2021) presented that Malassezia infection in males was 60% against 40% in females. Research carried out on molecular epidemiology of dermatophytosis in cats, dogs in the Kurdistan region of Iraq and by Jarjees & Issa (2022) and outcome was dermatophytosis in male cats was higher than in female cats, 47.6% compared with 41.4% with no statistical significance, while this percentage was the exact in male and female dogs. However, the prevalence of Malassezia infection between male and female were nearly the same that stated by Nardoni et al. (2004) in the study of occurrence of Malassezia species in healthy and dermatologically diseased dogs.

Table-1: Age wise occurrence of Malassezia infection in dogs and cats

Age	Frequency	Percentage (%)
Kitten/Puppy (0 - 6 months)	6	2.96
Junior (7 months - 2 years)	58	28.57

Adult (3 years - 6 years)	93	45.81
Mature (7 years – 10 years)	38	18.72
Senior (11 years – 14 years	7	3.45
Geriatric (15 years+)	1	0.49
Total	203	100

Age wise occurrence of Malassezia infections in dogs and cats is highlighted in Table-1. Among the 203 infected animals in this study, the highest number of Malassezia infection found in adult dog and cats that constitutes about 45.81% followed by junior ages (28.57%), matures (18.72%), seniors (3.45%), kittens/puppies (2.96%) and in geriatric pets found only 0.49%, respectively. We observed the same theme of the study result in a study performed by Sudipa et al., (2021) with 40% of Malassezia infection in dogs above three years. The study of Girao et al., (2006) and Seetha et al., (2018) reported that the majority of patients with Malassezia infections were from 1-3 years (56%), in contrast we noticed the maximum percentage age range from 3-6 years. According to Nguyễn et al., (2023) Malassezia cases also found from 1-3 years, constituting 45.8%. Based on age range occurrence of Malassezia infection, we found relatively analogous with the study of Dhoot et al. (2021) who completed the study of prevalence of Malassezia infection in patients of Nagpur City reported that the percentage of Malassezia infection under one year was 0.39%, from 1 to 3 years was 3.92%, and gradually increased infection to 49.8% from 3 to 9 years old and 2.7% at the age above 12 years old.

Table-2: Breed wise occurrence of Malassezia infection in dogs

Dog breeds name	**Frequency**	**Percentage (%)**
Local	40	33.06
German Shepherd	24	19.83
Golden Retriever	16	13.22
Labrador	4	3.31
Lhasa Apso	10	8.26
Shitzu	3	2.48
Spitz	8	6.61
Beagle	4	3.31

Bull dog	1	0.83
Great Dane	2	1.65
Maltese	1	0.83
Pomeranian	1	0.83
Sorail	1	0.83
Mixed	6	4.96
Total	**121**	**100**

In the dogs, 14 breeds were involved in Malassezia infection. Local non-descriptive dogs were the most common (33.06%) followed by German Shepherd (19.83%), Golden Retriever (13.22%), Lhasa Apso (8.26%) and so on highlighted on tables. Our study findings suggest more or less in the findings of Seetha et al., (2018) that was Non-descriptive dogs were the higher percentage (55%), followed by Labrador Retriever and Spitz (13%), German Shepherd (11%) Chinese Pug (4%), Dalmatian (2%), Doberman and Great Dane (1%). The highest incidence of Malassezia infection among Non-descriptive dogs may be influenced due to the middle or low income people who were rearing Non-descriptive local dogs instead of expensive exotic breeds. There are certain dog breeds reported with an increased risk of Malassezia dermatitis and these are West Highland White Terriers, American Cocker Spaniels, Dachshunds, Boxer, Poodles, English Setters, Australian Silky Terriers, Shih Tzus and Basset Hounds (Bond et al., 2020; Guillot and Bond, 2020; Hobi et al., 2022; Bajwa, 2023). The opposite result from our study was observed in another study carried by Nguyễn et al., (2023), stating that the percentage of Malassezia infection in domestic breeds was 25%, lower than 75% in exotic animals. Malassezia infection can be found in dogs of any age, breed, and sex.

Table-3: Breed wise occurrence of Malassezia infection in cats

Cat breeds name	Frequency	Percentage (%)
Local	17	20.73
Persian	45	54.88
Scottish fold	6	7.32
British shorthair	4	4.88

Russian Blue	2	2.44
Mixed	8	9.76
Total	**82**	**100**

The detailed breed wise occurrence of Malassezia infection in cats is presented in Table. Based on the observed breed wise incidence, the study showed that highest percentage found in Persian cat breed (54.88%) followed by local breeds (20.73%), mixed breeds (9.76%), Scottish fold (7.32%), British shorthair (4.88%) and Russian blue (2.44%), respectively. Malassezia dermatitis in cats is not commonly seen like in dogs (Morris, 1999; Ordeix et al., 2007; Negre et al., 2009; Machado et al., 2011; Bajwa, 2017). Moriello et al. (2017) reported that 75% of Persian cats had just four cases diagnosed with dermatophytosis in comparison with other research about breed factors in dermatophytosis and Malassezia. However, some cat breeds were identified mainly Devon Rex and Sphynx have high burdens of colonizing Malassezia, predisposing them to oily seborrhoea and Malassezia-related skin diseases (Åhman et al., 2007; Ahman and Bergstrom, 2009).

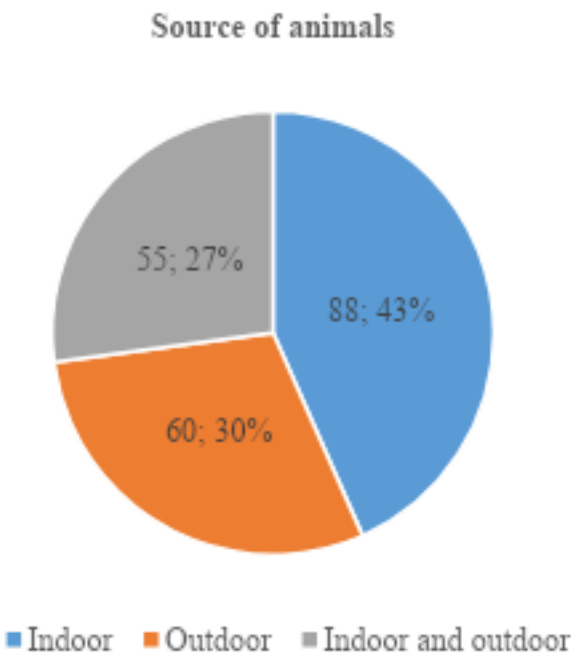

Fig-3: Occurrence of Malassezia infection according to patient's lifestyle

The patients were categorized in this study according to their lifestyle from where they came to hospital with Malassezia infection. We classified into 3 groups such as indoor patients, outdoor patients and both habitats. Among the sources of animals, the indoor lifestyle patients constituted the maximum percentage about 43%, then 30% found outdoor patients and finally 27% came both indoor and outdoor living patients. Animals living in the outside of the house have contact with unfavorable environmental conditions that may predispose the multiplication of Malassezia spore on the skin. Nardoni et al., (2005), explained that 130 of the 151 cats examined were living in a cattery or having contact with the outdoors, where direct transmission of Malassezia among animals is possible.

Table-4: Frequency of Malassezia isolation from different locations of patients

Affected locations	Frequency	Percentage (%)
Dorsal trunk	30	14.78
Ear pinnae and canals	56	27.59
Ventral trunk	47	23.15
Neck	11	5.42
Arm pits	14	6.90
Perenial region	9	4.43
Generalized dermatitis	29	14.29
Interdigital area	7	3.45
Total	**203**	**100**

Malassezia organism affects many parts of the body and causes alopecia, erythema, scales, crusts, greasiness and hyperpigmentation in chronic case and lichenification (Bond et al., 2020; Guillot and Bond, 2020; Hobi et al., 2022; Bajwa, 2023). This study recorded higher frequency in ear pinnae and canals (27.59%) followed by ventral trunk (23.15%), dorsal trunk (14.78%), generalized dermatitis (14.29%), arm pits (6.90%), neck region (5.42%), perenial region (4.43%) and interdigital area (3.45%), respectively. The findings of present study contradict the results of Seetha et al., (2018) who recorded higher Malassezia rate found in neck region (8) followed by dorsal and ventral trunk (6), ear pinnae, hind legs, prescrotal region and in cases of generalized

dermatitis (4), fore legs (3), perenial region and interdigital area (1). And also similar findings observed in the report of Yurayart et al., (2010), noted that maximun number of yeasts were counted from the neck region followed by ear canal, interdigital area and groin. In dogs, affected body areas of Malassezia infection includes pinnae, external ear canals, muzzle, ventral neck, ventral body sites, medial thighs and paws (Hobi et al., 2022; Bajwa, 2023). In most cats, frequently involved areas are the pinnae, face, chin, neck, limbs and ventral abdomen but in case of n Sphynx and Devon Rex breeds, the ventral neck, axillae, inguinal areas and paws are mainly affected (Hobi et al., 2022).

Fig-4: Ventral trunk of dog with Malassezia spp. overgrowth. Alopecia with erythema.

Fig-5: External ear canal of cat with Malassezia spp. overgrowth. External otitis with yellow or brown waxy-ceruminous secretion.

Cytological Examination

To diagnose Malassezia spore, an important fruitful, practical and rapid method is direct microscopy of tape strip, impression smear, cotton swab or skin scrape samples (Miller et al., 2012; Bond et al., 2020; Bajwa, 2023). Malassezia can be isolated clearly due to some typical features like uni-polar and often broad-based budding referred to as “foot print”, “babushka” or “peanut” shapes (Miller et al., 2012; Bond et al., 2020; Bajwa, 2023). For cytology study, the samples were collected from affected skin and ears of the patients. After preparation of the smear

(impression and swab smear), the slide was stained by Giemsa staining method. Then, the prepared slides were examined by 10x and 100x microscopic objects. Small foot print or pear shaped Malassezia spore isolated under microscope as well as bacteria also identified in mixed infection cases of this study. Karlapudi, (2017), diagnosed as Malassezia pachydermatis characterized by blue colored footprint shaped organisms when stained with diff quick method and also bacteria like Staphylococci, Pseudomonas and mixed infections from affected cases. Another study performed by Seetha et al., (2018), by Methylene blue staining, they detected peanut or footprint shaped budding yeast cells and presented that 26% cases were positive by impression smear, 34% cases were positive by adhesive tape and 40% cases were positive by skin swab smear. Cytological impression recognized nuclear streaming multiple Malassezia yeasts and a high number of coccoid bacteria from the affected parts (Hobi et al., 2024).

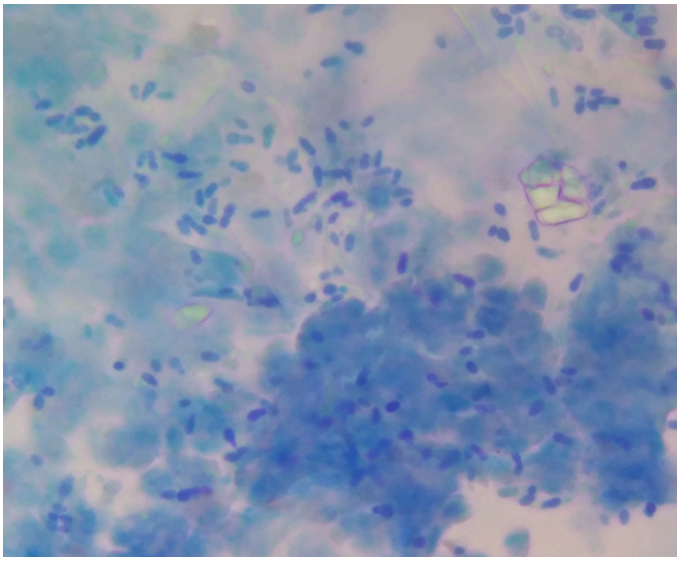

Fig-6: Cytological impression showing blue colored foot print or pear shaped Malassezia yeast and a high number of coccoid bacteria

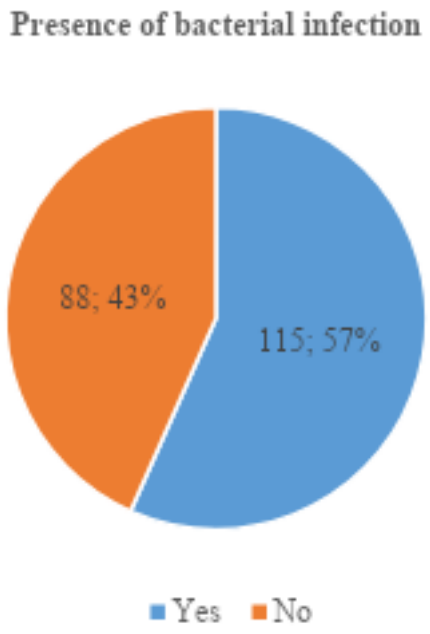

Fig-7: Occurrence of Malassezia dermatitis with or without bacterial infection

There are several commensal or environmental pathogens which are sometimes opportunistic pathogens. Under favorable conditions, along with Malassezia infection in skin they also aggravate the disease condition. In our study, we accounted for bacterial dermatitis in Malassezia infection after cytological examination of the sample and found that bacterial infection was present in 57% cases and rest 43% cases was free from bacterial dermatitis.

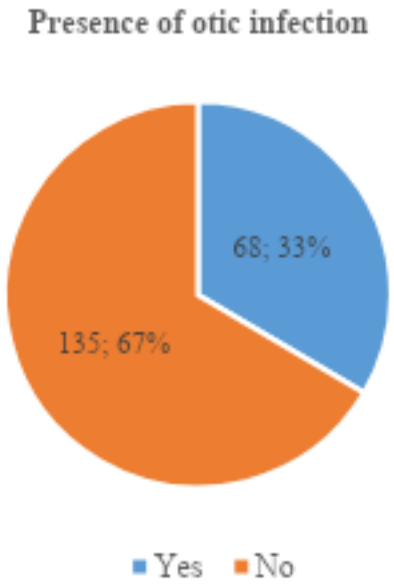

Fig-8: Frequency of otitis externa in Malassezia affected cases

One of the secondary causes of otitis externa is thought to be Malassezia species.The ideal condition for yeast overgrowth are created in in dog and cats with narrow ear canals or increased ear secretion. In the present study, the cytological result of 203 individuals revealed that Malassezia with otitis externa was detected in 68 cases, or 33% of total cases and 67% dog did not have otitis externa. In a study published in 2023, Nguyen et al., found that out of all cases, 15.38% of Malassezia cases had otitis externa. However, another study (Nardoni, S. et al., 2014) suggests a contradictory finding in which otitis externa was shown to be responsible for almost half of all instances of malassezia in both dogs (58.2%) and dogs (52.7%).

Chapter 4:Conclusion

The study provided an overview of the epidemiology, clinical presentation, and management of Malassezia infections in dogs and cats, with or without bacterial co-infections, in Dhaka, Bangladesh. Malassezia infections were more prevalent in dogs (60%) than in cats (40%), with males showing a higher incidence than females. The highest rate of infection was observed in adult animals (3-6 years), particularly in local dog breeds and Persian cats. The study confirmed that environmental factors, such as outdoor exposure, were associated with a higher incidence of Malassezia infections.The most commonly affected body parts were the ear pinnae and canals (27.59%), followed by the ventral trunk (23.15%).

Cytological examination using Giemsa-stained smears identified Malassezia spores, which were "footprint" or "peanut" shaped organisms, in infected animals, and bacterial co-infections were found in 57% of cases, with otitis externa identified in 33% of the patients. The treatment protocol, consisting of topical antiseptics, antifungal agents, and systemic therapy for severe cases, was generally effective, although chronic cases showed less favorable outcomes. This study contributes valuable data on the prevalence, clinical presentation, and management of Malassezia infections in companion animals in Dhaka, Bangladesh. Further studies need for improved diagnostic and treatment strategies in veterinary practice to address both Malassezia and its associated bacterial infections.

LIMITATION OF STUDY:

1. No molecular test like PCR was performed to diagnose Malassezia
2. We couldn't track the treatment response of the affected animals.

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The Author

BIOGRAPHY

Sayma Mahmud Moon, daughter of **MD. Chan Mahmud Monir** and **Shahnaj Parvin**, was born on 26 may, 2000 at Sagardighi, Ghatail, Tangail. She passed her Secondary School Certificate Examination from Sakhipur Pilot Girls High School, Sakhipur, Tangail in 2016 (GPA 5.00). Then she passed her Higher Secondary School certificate examination from Shahid Sayed Nazrul Islam College, Mymensingh in 2018 (GPA 4.83). Now she is completing her one-year long internship program for fulfilling the requirement of Doctor of Veterinary Medicine (DVM) degree in Chattogram Veterinary and Animal Sciences University, Chattogram, Bangladesh. During her internship period she received her clinical training on Veterinary Medicine from CVASU Lab Rotation, ShahedulAlamQuadery Teaching Veterinary Hospital (SAQTVH), PRTC, Teaching & Training Pet Hospital and Research Center (TTPHRC), Anowara UVH, Sakhipur UVH and Intensive short course of veterinary diagnostic pathology and reproduction care from Universitas Brawijaya, Indonesia ,UPM Malaysia etc.

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