An Over View of Canine Dermatophytosis

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Author's contribution

The sole author designed, analysed, interpreted and prepared the manuscript.

Review Article

ABSTRACT

Dermatophytosis is a superficial infection of the keratinized layers of the skin and its appendages (hair, feathers, horns) and is caused by keratinophilic and keratinolytic genera such as *Microsporum*, *Trichophyton* and *Epidermophyton*. In dogs, nearly 70% of cases are caused by *Microsporum canis*, 20% by *M. gypseum*, and 10% by *Trichophyton mentagrophytes*. The Wood's lamp test is of diagnostic importance for the establishment of a tentative diagnosis of dermatophytosis in dogs. This overview will forecast more light on different aspects of this disease.

Keywords: Canine; dermatophytosis; epidemiology; transmission; diagnosis; treatment.

1. INTRODUCTION

Dermatophytoses are the most common fungal infections in dogs [1,2]. The dermatophytes have a high affinity for keratin, an important component of fur, skin and nails, which are the primary sites of fungal infection [3]. Clinical presentations of dermatophytic lesions include multifocal alopecia, mild or intense pruritus and round scaly lesions with erythematous and scaly borders [4]. According to their natural reservoir, dermatophytes are classified as anthropophilic, zoophilic or geophilic [5]. Several reports have stated that *Microsporum canis*, a typical zoophilic species, is the most common dermatophyte isolated from dogs and cats worldwide [6,4,2,7].

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2. EPIDEMIOLOGY

The epidemiology of the dermatophytes is related to the presence of suitable environment [8]. These fungi are classified according to their habitat in anthropophilic, geophilic, and zoophilic. The prevalence of dermatophytoses in dogs range from 4% to 10% however, few studies show a higher prevalence [9]. Young dogs aged between 6-18 months have high occurrence (46.67%) and susceptibility to infestation, compared to those pets above one and half year old (31.11%) and those in the higher age group of over three years (22.22%) [10]. No influence of sex on occurrence of canine dermatophytosis was observed by Cabanes et al. [11]. However, Singathia et al. [10] and Bhardwaj et al. [12] reported higher incidence in male than female (73.33%) and (26.67%) respectively. [13] Beside age, risk factors include poor nutrition, number of animals, poor management, and lack of an adequate quarantine period for infected pets [14]. As for breed, Yorkshire terriers had a statistically significant higher incidence of dermatophytoses, especially that caused by *M. canis* (46.4%), than other breeds. The disease was reported during pre-monsoon and monsoon seasons extending from June to October. The prevalence of the disease increase depends on geo-climatic, growth and distribution of pathogenic fungal elements. The infection was severe and more prevalent in dogs maintained indoor using desert coolers and/or air conditioners to combat impact of prevailing hot surrounding during summer and monsoon months (April to June). Higher humidity is a main factor for faster multiplication and propagation of fungal elements [15]. An increased prevalence of *M. canis* was reported in the fall /winter season while *M. gpseum* showed higher prevalence in the spring and summer [16].

2.1 Predisposing Factors to Dermato-phytosis

1- Young age (first 2 years of life)
2- Immunosuppression (including immunosuppressive treatment)
3- Other diseases
4- Nutritional deficits (especially proteins and vitamin A)
5- High temperature and high humidity
6- Skin trauma resulting from increased moisture,
7- Injury by ectoparasites or scratches due to pruritus
8- Playing or aggressive behaviour, clipping, etc.
9- Poor hygiene
10- Overcrowding in catteries

3. TRANSMISSION

Most cases of ringworm are spread by direct contact with infected animals or indirect with contaminated objects such as furniture or grooming tools. Broken hairs with associated spores are main sources for spreading of the ringworm. Contact does not always result in infection. As infection establishment depends on the fungal species and certain host factors, such as age, health, condition of exposed skin surfaces, grooming behavior, and nutrition.

M. canis has been cultured not only from infected animals but also from dust, heating vents and furnace filters. An infected cat is normally the source of *M. canis* while *Trichophyton* infections are usually acquired by contact with the reservoir, generally rodents. Geophilic fungi inhabit the rich soil and may be acquired from the environment. Spores attach to the epidermis and germinate to produce hyphae that invade stratum corneum and hair. The incubation period from beginning of the infection to the onset of the skin lesions is normally seven to 14 days [17].

3.1 Clinical Features of Dermatophytosis

Canine dermatophytosis is characterized by typical round alopecic lesions and brittle hairs. Single or multi- focal scaly crusted lesions were observed. Local or widespread folliculitis may with or without foruncolosis was reported. Other clinical signs include dry seborrhoea, focal or multi-focal crusted dermatitis with well-defned erythematous margins, kerion, onychomycosis and/or paronychia. Signs and symptoms mainly depend on host-fungus interaction. Infection with *M. canis* in dogs usually presents more marked inflammation than in cats. Vesicles and pustules may also be seen. In later stages, the area is often covered by a crust and the edges swollen. *M. gypseum* or *T. mentagrophytes* infection often causes kerion (localized severe inflammation with swollen, boggy skin oozing pus) it is often associated with secondary bacterial infection. These infections frequently develop on the face and limbs of hunting dogs that spend a lot of time outdoors in contact with the ground. Onychomycosis is very rare in dogs and usually caused by *M. gypseum* or *T. mentagrophytes*. The nail becomes brittle, loses its shape with a developing of periungual inflammation [18].

Fig. 1. Alopecic lesion and erythematous left forelimb (yellow circle)

Fig. 2. Skin lesions covering almost the entire head

Fig. 3. Nodules on the muzzle of a dog

Fig. 4. Painful nodular lesion on the dog digit

Fig. 5. Dermatophyte kerion in a dog
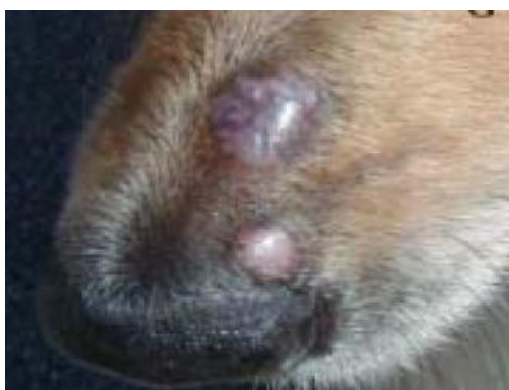

Fig. 6. Onychomycosis in a dog: nail and nail bed involvement

4. ETIOLOGY

Over 20 different species of dermatophytes have been reported to cause clinical disease in dogs. However the most commonly isolated pathogens are *Microsporum canis*, *M. gypseum* and *Trichophyton mentagrophytes* [19-21]. [22] isolated *M. nanum, M. vangreuseghemii, T. ajelloi, T. terrestre*. *M. persicolor* was isolated by [23]. In Southern Brazil *M. canis* var. *distortum* was reported [24]. As a case report [25] isolated *Trichophyton mentagrophytes* var. *erinacei* from a 5-year-old male mongrel dog. *T. tonsurans* which is a classic anthropophilic species usually isolated from human dermatophytosis has been isolated from a 2-year-old female dog [26] *T. rubrum* var. *raubitschekii* has been isolated for the first time from an 11-year-old male Yorkshire terrier dog [27], while *Epidermophyton floccosum* has been isolated from dogs in Norway [28] and United State [29].

5. DIAGNOSIS OF DERMATOPHYTOSIS

The diagnosis of dermatophytosis is based on clinical history, physical examination, and various diagnostic tests including Wood's light interposition, direct microscopic examination of the hairs and/or crusts, fungal culture and biopsy [30-32]. Despite the large number of diagnostic methods available, none of them is completely efficient [33].

6. PHYSICAL EXAMINATION

In cases of suspected dermatophytosis examination involves palpation of the skin for lesions that might not otherwise be found, examination should be done in room light to identify affected areas by using a strong beam flashlight, which is particularly helpful for revealing lesions that are 'washed out' by room light [34].

7. WOOD'S LAMP EXAMINATION

Wood's lamp examination is a useful technique. About 50% of *M. canis* strains produce metabolites which fluoresce an apple-green colour when examined by the lamp. Positive hairs are excellent specimens for microscopically examination and culture. Wood's lamp examination should be used routinely when dermatophytosis is suspected. However, some *M. canis* strains and all of the *Trichophyton* spp. of veterinary importance don't produce fluorescence [34].

8. DIRECT EXAMINATION

Hairs and scales can be mounted in potassium hydroxide (KOH) of varying concentrations [35-37]. Infected hairs appear pale, wide and filamentous compared with normal hairs when microscopically examined at x4 or x10 magnification. Arthrospores can be visible on high magnification (x40). Positive result of KOH direct test can lead to positive cultures, which are considered as the gold standard. Method of calcofluor white as an alternative to KOH as it binds specifically to the fungal cell wall and fluoresces strongly can be used when viewed under a fluorescence microscope, the sensitivity of calcofluor white compared to KOH was found
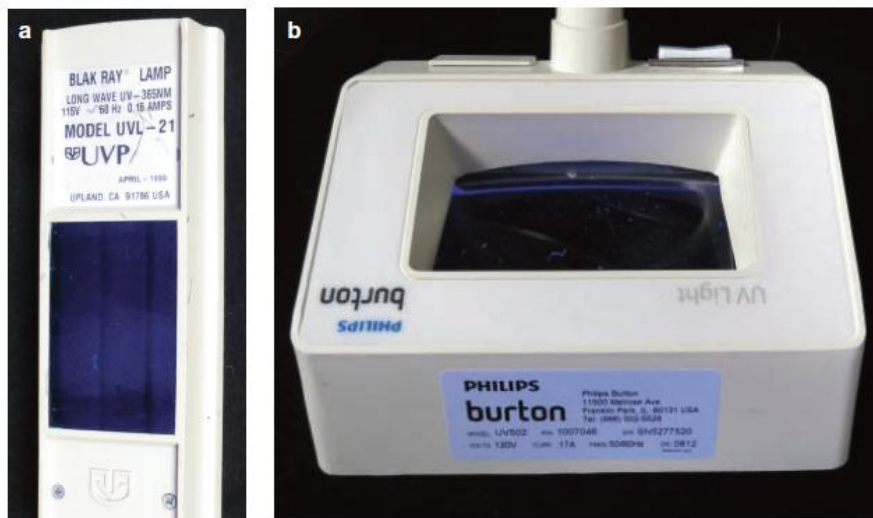

Fig. 7. Wood's lamps. (a) Small compact model and (b) model with built-in magnification

Fig. 8. Wood's lamps examination of the hair *in vivo*

76% and 39% respectively [38], however some studies reported no difference in positive predictive value when using calcofluor compared to KOH [39,40].

Fig. 9. Arrows show fluorescing hairs

9. SAMPLING TECHNIQUES

9.1 Hair Pluck

To collect samples for dermatophyte culture, sterile hemostat to pluck hairs from around the

periphery of a newly formed skin lesion will be used, however recently medicated lesions must not be collected. During sampling damaged hair as well as hair in areas of active crusting should be selected [20].
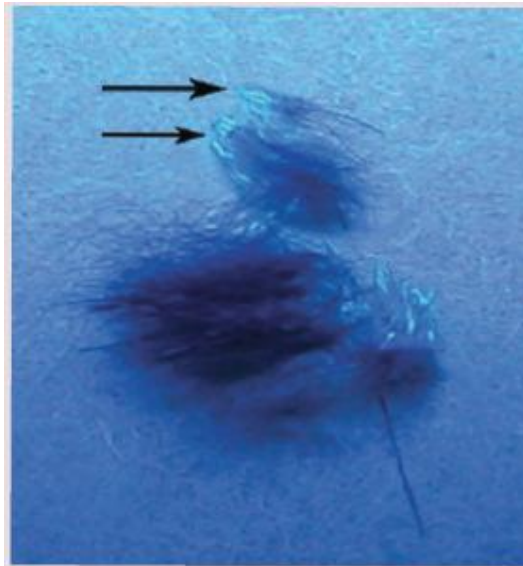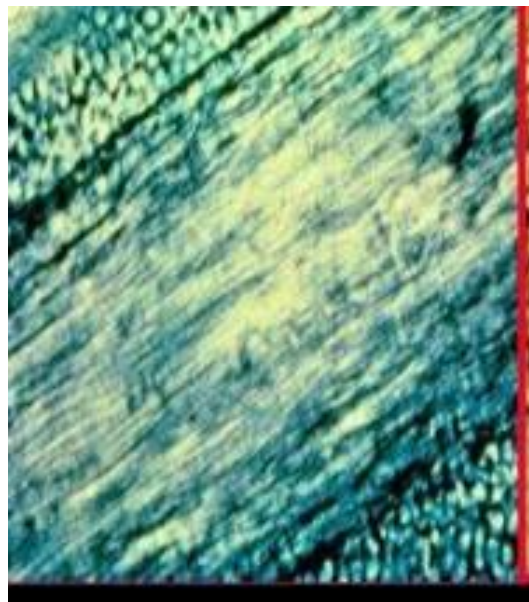

Fig. 10. Ectothrix spores of *M. canis*

9.2 Tooth Brush

Mackenzie brush technique is ideal to collect sample compared to hair plucks because the

latter can miss infected hairs and epithelium. With this technique, a new toothbrush is removed from its packaging and is rubbed gently over the suspect area, including the skin and haired margins of alopecic or scaly lesions. Brushing should be started with the unaffected area, followed by the affected lesions in order to avoid spreading of spores to un affected areas, which will also help in avoiding the losing of spores from affected areas following that the tooth brush bristles should be embedded gently into the fungal culture media. The brush technique is the most common procedure because it is simple, atraumatic, economical and fast.

The Mackenzie brush technique is also found helpful for collecting samples from asymptomatic carriers and animals undergoing antifungal treatment in which their skin lesions were clinically resolved. In these cases, areas with prior lesions have to be more focused beside entire body [41]. It is recommended to brush for one minute or to brush the length of the animal 10 times. In animals undergoing antifungal therapy, repeat cultures every two or three weeks, and continue treatment until two negative culture results are obtained.

9.3 Sticky Tape Technique

It is a rarely described but potentially useful technique. In this technique, a 4 cm length of tape is pressed over lesions and then pressed to the surface of a fungal culture plate. The result of lesion culture is equal whether using the brush technique or sticky tape technique; although the sticky tape technique appeared to be more sensitive [41].

9.4 Fungal Culture

Fungal culture is considered the 'gold standard' for diagnosis [42]. Sabouraud's dextrose agar (SDA) containing cycloheximide, penicillin and streptomycin were used in most diagnostic laboratories. Plates should be incubated at 25ºC for 5 weeks. Dermatophytes test media (DTM) is recommended as the best media for isolation of dermatophytes because the presence of the red color indicated positive result, this can help in early identification of highly suspected cultures [43]. The isolates should be examined macroscopically and microscopically after staining with lactophenol cotton blue using wet mount technique [44].

Fig. 11. Endothrix invasion of hair

Fig. 12. Hair plucking for culture sample

Fig. 13. Tooth brush *t*o collect samples for dermatophyte culture

Fig. 14. Sticky tape technique

In addition to the steps mentioned above, pigment production on corn meal agar, urease activity on urea agar base, growth at 37ºC on SDA *in vitro* and hair perforation tests are used for identification of dermatophytes [45,46]. Moreover, biochemical test have also been employed to differentiate *Trichophyton* spp using series Trichophyton agar from 1 to 7 which enriched with urea, thiamine, histidine, nicotinic acid and inositol, the isolates are subcultured in this media [47].
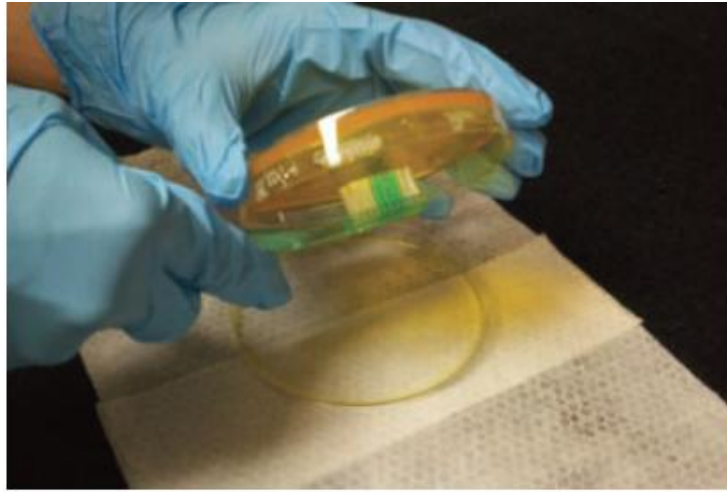
Fig. 15. Inoculating plates 'upside down' over a disinfectant wipe minimizes contamination

Fig. 16. *Microsporum canis* culture, macroscopic colony

Fig. 17. *Microsporum canis* microscopic observation in lactophenol cotton blue

Fig. 18. *Microsporum gypseum* culture, macroscopic colony

Fig. 19. *Microsporum gypseum* microscopic observation in lactophenol cotton blue

9.5 Molecular Diagnosis

Conventional diagnostic methods are time-consuming because it might take up to 4 weeks or longer to give the final results [48]. Furthermore, morphological identification may be confusing due to polymorphism of dermatophytes [49]. During the last decade, a wide variety of molecular techniques has become available as possible alternatives for routine identification of fungi in clinical microbiology laboratories [50,51]. The first and second internal transcribed spacers (ITS-1 and ITS-2, respectively) of nuclear ribosomal DNA and a part of the chitin synthase gene (designated p *chs*- 1) have showed particular promise as markers for the specific identification of dermatophytes [52]. Specific primer pairs were designed for the selective amplification of ITS _ (_ ITS-1 _ 5.8S rRNA gene _ ITS-2) or p *chs*- 1 of dermatophyte

species (i.e., *M. canis, M. gypseum, T. terrestre* and *T. mentagrophytes*) which are frequently isolated from the hair of animals , but not of other fungi associated with the coats [53]. One-step and nested PCRs used for identification of canine dermatophytes gave positive result for dermatophytes culture but were negative for other fungi isolates as shown in (Fig. 23) [54]. RFLP –PCR was used for identification of *M. canis* [55].

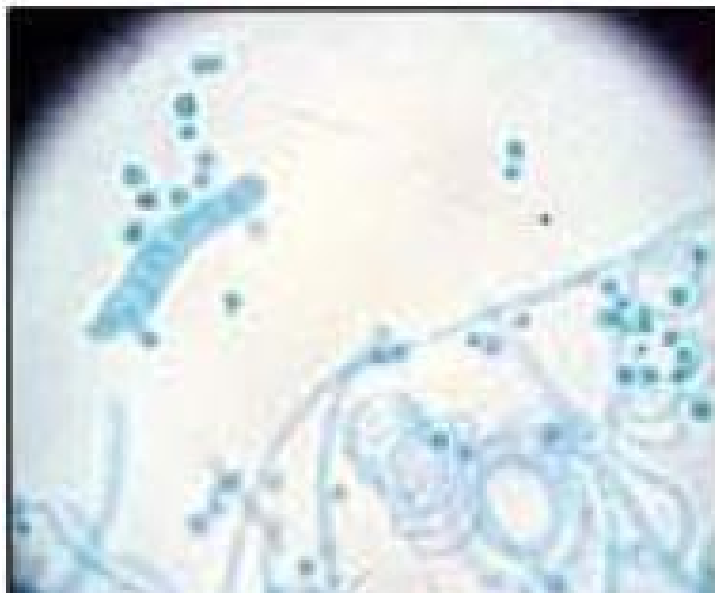**Fig. 20. *Trichophyton mentagrophytes* var. mentagrophytes culture, macroscopic colony**

Fig. 21. *Trichophyton mentagrophytes* var. mentagrophytes microscopic observation in lactophenol cotton blue

Fig. 22. Culture of *T.rubrum* in DTM (dermatophyte test medium)

9.6 Serodiagnosis

An enzyme-linked immunosorbant assay (ELISA) for diagnosis of canine dermatophytosis was developed by [56]. A whole fungal extract antigen was obtained from an isolate of *Microsporum canis* which have been cultured on a liquid medium isolated from a cat with patches of alopecia. The test has good sensitivity (83.3%) and high specificity (95.2%) but some dogs retained positive titres after elimination of infection. The sensitivity is high compared to that of direct microscopic hair examination and similar to that of fungal culture with DTM (dermatophyte test medium).

9.7 Skin Biopsy

Skin biopsy for diagnosis of canine dermatophytosis was reported only for kerion reactions and granulomatous infections because cultures are often negative, in this case the species caused the infection can not be known. haematoxylin and eosin staining (H&E) may or may not identify dermatophytes and special stains such as periodic acid Schiff (PAS) and Grocott methenamine silver (GMS) are needed [30]. Histologically the lesion is characterized as a nest of ruptured hair follicles replaced by suppurative to pyogranulomatous inflammation sometimes with eosinophils oriented around hair fragments that contain fungal hyphae and are surrounded by fungal spores.

9.8 Treatment

Optimal therapy of dermatophytosis requires a combination of topical antifungal therapy, concurrent systemic antifungal therapy and environmental decontamination. The treatment should be continued until two consecutive negative cultures (at weekly or bi-weekly intervals) are obtained [57,58]. Topical treatments speed the resolution of clinical lesions and may help in preventing zoonotic contagion. Systemic therapies that have prolonged residual activity in the skin and hair provide the most effective treatments.
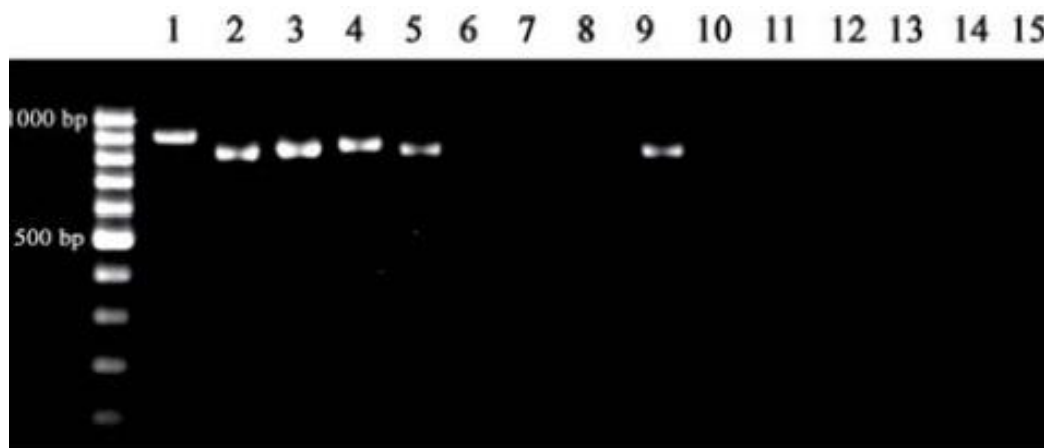

Fig. 23. PCR amplifi cation of ITS _ from genomic DNA samples carried out using primers DMTF18SF1 and DMTF28SR1 *Microsporum canis*, *M. fulvum*, *M. gypseum*, *Trichophyton interdigitale* (zoophilic), *T. terrestre* (lanes 1 – 5), species of *Alternaria*, *Aspergillus*, *Cladosporium* (lanes 6 – 8), *Chrysosporium* (lane 9), *Malassezia*, *Mucor*, *Penicillium*, *Rhizopus*, *Scopularopsis* (lanes 10 – 14) and no-DNA control (lane 15). Amplicons were sized by comparison with a 100 bp ladder (Gene Ruler, MBI Fermentas)
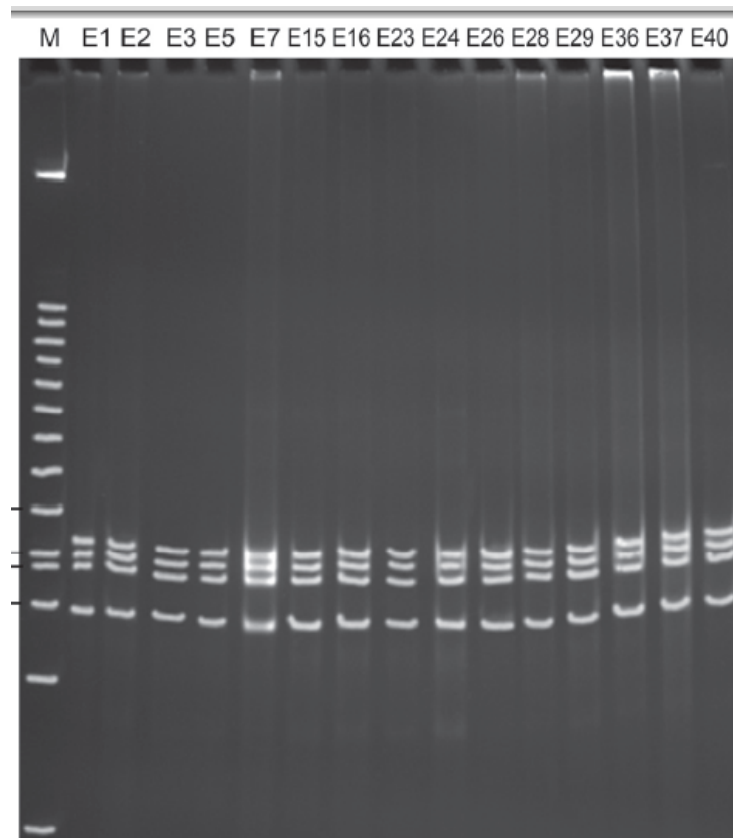
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Fig. 24. Polyacrilamide-gel electrophoresis of PCR products of *M.canis* isolates digested with *HinfI* restriction enzyme. The ITS1-ITS4 sets of primers were used to amplify ribosomal DNA including internal transcribed spacers (ITS)
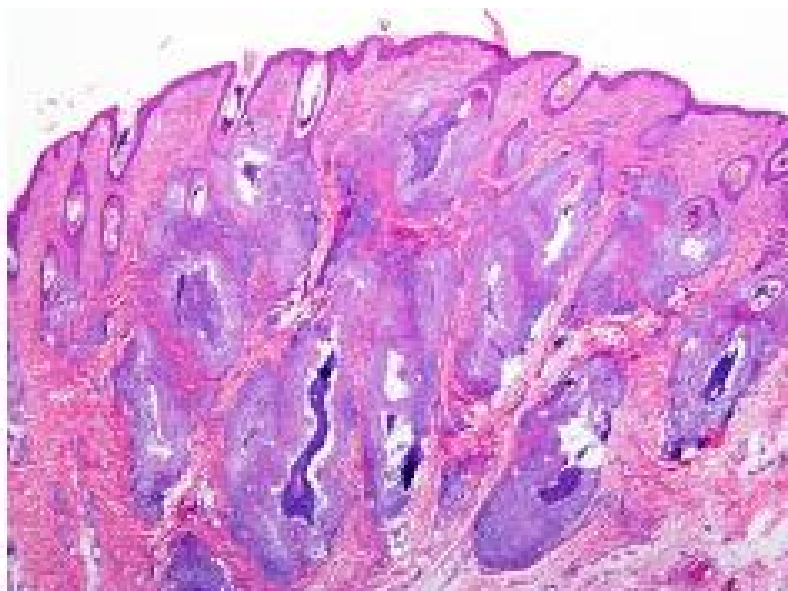

Fig. 25. Multifocal areas of pyogranulomatous inflammation oriented around and replacing hair follicles
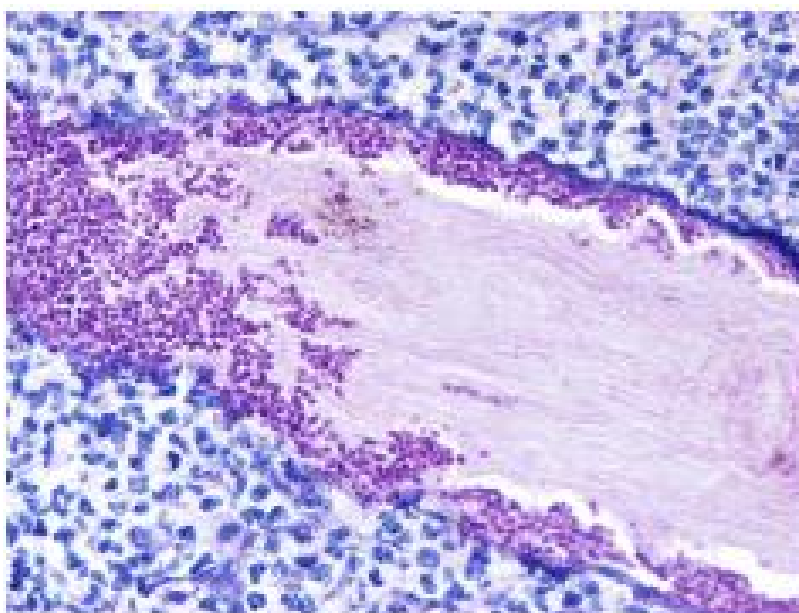
Fig. 26. Hair shaft with dermatophyte hyphae within the hair shaft and fungal spores surrounding the hair. The hair shaft is surrounded by neutrophils and macrophages

9.9 Topical Therapy

This kind of treatment is recommended for limited number of lesions, firstly hairs should be clipped all around lesions. Topical antifungal drugs are different in their efficacy. A whole body treatment with Lime-sulphur solution or a 0.2% enilconazole solution twice weekly have been found to be the best fungal growth inhibitor when compared to Chlorhexidine and povidone iodine solutions [59].

9.10 Systemic Therapy

All oral systematic antifungal drugs such as, griseofulvin, itraconazole and terbinafine are effective. Griseofulvin in doses of 30–50 mg/kg daily was recommended for weeks to months and should be continued for at least two weeks after clinical recovery. Imidazoles, like ketoconazole (10 mg/kg) and itraconazole (5 to 10 mg/kg) may also be used. Terbinafine (Lamisil) (10-30 mg/kg) is safer than ketoconazole or even itraconazole [60].

Treatment of kerion lesions in dogs was reported to be treated with miconazole, gentamicin (antibacterial agent to treat secondry infection), betametazone (to decrease inflammation) twice a day. The lesions will be recovered in 45 days [61].

10. CONCLUSION

Dermatophytoses are the most common fungal infections in dogs. Many studies were done considering different aspects of the disease (eg. epidemiology, clinical presentation and diagnosis, treatment, prevention, and control). Infected dog with dermatphytes can be a source of infection to human this can lead to public health problem.

COMPETING INTERESTS

Author has declared that no competing interests exist.

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