

Original Article : Open Access

Evaluation of antifungal potential of *Cassia tora* L. ointment in canine mycotic dermatitis

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Article Info

Article history

Received 19 April 2026

Revised 29 May 2026

Accepted 30 May 2026

Published Online 30 June 2026

Keywords

Cassia tora L.

Canine

Mycotic dermatitis

Antifungal

Dermatophytosis

Herbal ointment

Abstract

This study was conducted to assess the antifungal efficacy of *Cassia tora* L. oil-based ointment in the management of canine mycotic dermatitis. A total of 195 dogs with dermatological disorders were screened during the study period from August 2025 to January 2026, mycotic dermatitis was confirmed by techniques such as skin scraping examination, direct microscopy, and fungal culture. Among these, 20 dogs, confirmed with fungal dermatitis were selected and divided into two groups (n=10). Group T2 was treated with *C. tora* ointment (10%) along with supportive therapy for 28 days. Mycological examination revealed *Microsporum* spp. (42.5%) as the most prevalent pathogen with *Trichophyton* spp. (30%), *Malassezia* spp. (10%), *Aspergillus* spp. (7.5%), *Penicillium* spp. (5%), and *Candida* spp. (5%). Clinical manifestations included pruritus (72.5%), rough hair coat (65%), alopecia (57.5%), scaling (42.5%), crusting (30%), and erythema (20%). *In vitro* antifungal sensitivity testing indicated moderate antifungal activity of *C. tora* ointment. Therapeutic evaluation revealed a 50% recovery rate by day 28. Hemato-biochemical parameters remained largely within physiological limits. The findings suggested that *C. tora* ointment possesses moderate antifungal activity and may serve as a safe and economical topical therapeutic option.

1. Introduction

Dogs (*Canis familiaris*) have long been associated with humans as companion animals and play important roles in emotional and social well-being (Hart and Hart, 1988; Pachauri *et al.*, 1999). Dermatological conditions represent among the most frequently encountered clinical problems in small animal practice, accounting for approximately 12-75% of cases (Scott *et al.*, 2001; Sindha *et al.*, 2015). Among these, mycotic dermatitis is an important infectious skin disease in dogs, often marked by chronicity and frequent recurrence (Chermette *et al.*, 2008). Dermatophytes, including *Microsporum canis*, *Microsporum gypseum*, and *Trichophyton mentagrophytes*, are the principal etiological agents, capable of degrading keratin and invading superficial tissues (Ajello, 1953; Copetti *et al.*, 2006; Putignani *et al.*, 2010). Opportunistic fungi and yeasts, including *Malassezia pachydermatis*, *Aspergillus*, and *Candida* species, also contribute to disease occurrence under favourable conditions (Aujla *et al.*, 2000; Cafarchia *et al.*, 2004; Muller *et al.*, 1983). The occurrence of fungal dermatitis is influenced by environmental parameters, including

humidity, temperature, and hygiene, particularly in tropical regions (Nichita and Marcu, 2010). Diagnosis is typically based on microscopic examination and fungal culture techniques (Shalaby *et al.*, 2016).

Conventional antifungal agents, including azoles and allylamines, are effective but associated with limitations such as adverse effects, recurrence, resistance, and high cost (Ameen *et al.*, 2010). These drawbacks have led to increasing interest in plant-based therapeutic alternatives.

C. tora, commonly known as takala or chakramarda, is traditionally used for treating cutaneous infections, including ringworm, eczema, and pruritus (Srivastava *et al.*, 2017; Shadab *et al.*, 2019). It contains bioactive anthraquinone compounds, including chrysophanol, emodin, physcion, and rhein, along with aloe-emodin, which exhibit potent antifungal activity (Kim *et al.*, 2004; Telrandhe and Gunde, 2022). Previous *in vitro* studies have demonstrated its inhibitory effects against dermatophytes, including species of the genera *Microsporum* and *Trichophyton* (Phongpaichit *et al.*, 2004; Chankana *et al.*, 2017).

Furthermore, the plant exhibits anti-inflammatory, hepatoprotective, antidiabetic, antihypertensive, and antimutagenic activities, as reported in various pharmacological studies (Sarwa *et al.*, 2014; Bhandirge *et al.*, 2016). The presence of protease inhibitors in seed extracts contributes to their antimicrobial and antifungal efficacy by

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inhibiting pathogen-associated enzymatic activity (Tripathi *et al.*, 2011). Traditional medicinal systems have extensively utilized *C. tora* in the treatment of dermatological conditions such as ringworm, eczema, and pruritus, supporting its therapeutic relevance in skin diseases. Additionally, antioxidant activity associated with fatty acids and phenolic constituents further enhances its role in disease

management (Zhang *et al.*, 2007). These findings collectively indicate that *C. tora* possesses multitarget pharmacological potential, making it a valuable candidate for antifungal and dermatological applications. Accordingly, this study aimed to assess the antifungal effectiveness of *C. tora ointment* in canine mycotic dermatitis under clinical conditions.

Table 1: Phytochemical constituents of *C. tora* (Dighe *et al.*, 2009)

Phytochemical class	Constituents identified	Source/plant part
Anthraquinones (major active compound)	Emodin, chrysophanol, physcion, rhein, aloe-emodin, auranthio-obtusin, obtusin	Seeds, leaves
Naphthopyrones and glycosides	Naphthopyrone glycosides, anthraquinone glycosides	Seeds
Flavonoids	Karanjin-like flavonoids, phenolic flavonoids	Leaves, seeds
Phenolic compounds	Polyphenols, phenolic acids	Whole plant
Fatty acids (Volatile oil)	Oleic, linoleic, hexadecanoic, octadecanoic fatty acids	Seeds
Proteins and enzymatic inhibitors	Protease inhibitors	Seeds
Alkaloids	Trace alkaloidal compounds	Whole plant
Saponins	Saponin derivatives	Leaves
Triterpenoids	Triterpenoid compounds	Leaves
Carbohydrates and glycosides	Sugars, glycosidic compounds	Whole plant

2. Materials and Methods

2.1 Plant identification

The plant material (*C. tora*) used in the study was taxonomically authenticated by a qualified botanist at Yashvantrao Chavan Institute of Science, Satara (Affiliated to Shivaji University, Kolhapur), Maharashtra, India (Accession Number; BMB001).

2.2 Preparation of *C. tora* antifungal ointment

The antifungal ointment was prepared by the fusion method using simple ointment as the base. The ointment base was formulated accurately by weighing hard paraffin (10 g), wool fat (lanolin) (5 g), and white soft paraffin (85 g). These ingredients were melted together in a water bath maintained at 60-70°C with continuous stirring until a uniform molten mixture was obtained. The molten mass was allowed to cool gradually with gentle stirring to ensure uniform consistency and to prevent phase partition.

C. tora seed oil was purchased from a trusted ayurvedic drug store and stored in airtight containers at room temperature until further use. The required quantity of *C. tora oil* was gently warmed in a water bath at 40-50°C. Salicylic acid was then added to the warmed oil phase and stirred continuously until complete dissolution was achieved. Separately, the pre-prepared ointment base was softened by mild heating. The oil-salicylic acid mixture was gradually incorporated into the softened base with continuous stirring to ensure homogeneous dispersion of the active ingredients throughout the formulation.

For the preparation of *C. tora* ointment, a concentration of 10% (w/w) of *C. tora* seed oil was incorporated into the ointment base. Salicylic acid (2%, w/w) was added as a keratolytic agent to enhance antifungal efficacy and facilitate penetration of the active constituents into keratinized tissues (Beale, 2000; Foil, 1998).

The prepared ointment was allowed to cool to room temperature while stirring continuously to maintain homogeneity and prevent component separation. The final product was transferred into clean, sterile, airtight plastic containers, properly labelled, and kept in a cool, dry environment, protected from direct sunlight until further usage. The formulation was evaluated for physicochemical properties, including appearance, homogeneity, pH, and spreadability, prior to *in vivo* application to ensure stability and suitability for therapeutic use.

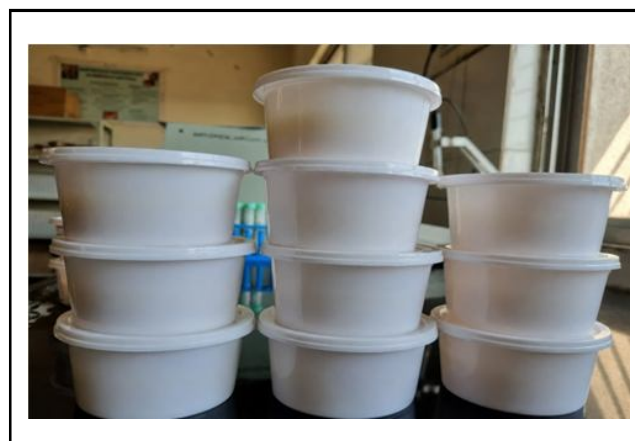


Figure 1: Bulk batch prepared *C. tora* antifungal ointment formulation.

2.3 Selection of animals

Dogs of different age groups, breeds, and sexes showing clinical manifestations including pruritus, alopecia, erythema, and scaling, presented to the Veterinary Clinical Complex at Krantisinh Nana Patil College of Veterinary Science, Shirwal, were evaluated. The diagnosis of mycotic dermatitis was confirmed by skin scraping

examination, direct microscopy, and fungal culture (Kumar *et al.*, 2002; Shalaby *et al.*, 2016). These methods are considered standard diagnostic tools in veterinary mycology (Kumar *et al.*, 2002; David *et al.*, 2004). A total of 20 dogs confirmed with mycotic dermatitis were enrolled in the study and randomly assigned to treatment groups.



Figure 2: Prepared *C. tora* antifungal ointment formulation.

2.4 Experimental design

A total of 195 dogs with dermatological disorders were screened during the study period from August 2025 to January 2026. Mycotic dermatitis was confirmed by techniques such as skin scraping examination, direct microscopy, and fungal culture. Among these 20 dogs, confirmed with fungal dermatitis were selected and randomly allocated into two groups (n=10), with 10 affected dogs in each group. Treatment consisting of topical application of Ketoconazole (2%) and *C. tora* ointment (10%) to groups T₁ and T₂, respectively till day 28 of the study. Along with this treatment, all the dogs were receiving other supporting treatments, including liver tonic @ 5 ml orally B.I.D. and multivitamin supplementation comprising Omega-3 and Omega-6 fatty acids @ 5 ml orally B.I.D. for a period of four weeks.

2.5 Clinical evaluation

Clinical parameters assessed included improvement in clinical signs such as pruritus, alopecia, scaling, crusting, erythema, and hair coat condition, along with hematological and biochemical parameters on day 0 and day 28.

2.6 Hematobiochemical analysis

Blood samples were obtained from the cephalic vein on Day 0 and Day 28. EDTA samples were used for hematological analysis, comprising haemoglobin (Hb), packed cell volume (PCV), erythrocyte (RBC) count, platelet count, total leucocyte count (TLC), and differential leucocyte count (DLC). Serum was isolated by centrifuging at 3000 rpm for 10 min and subsequently analysed using a semi-automated biochemistry analyser with commercial kits to estimate ALT, AST, ALP, total protein, albumin, and globulin. Statistical analysis was carried out using independent sample t-tests, with differences regarded at statistically significant at *p*<0.05.

3. Results

Occurrence of mycotic dermatitis in the present study, accounted for 21.02% of dermatological cases observed in 195 dogs.

3.1 Etiological agents

Of 41 dogs diagnosed with mycotic dermatitis based on clinical and microscopic examination, 40 were cultured for definitive identification of fungal etiological agents. Direct microscopic evaluation with KOH and lactophenol cotton blue preparations demonstrated the presence of fungal structures including septate hyphae, arthrospores, and conidia, with 74% of samples yielding positive results. Mycological culturing on sabouraud’s dextrose agar (SDA) and dermatophyte test medium (DTM) yielded fungal growth in 90% of cases, highlighting the importance of combined diagnostic methods.

Among the isolates, *Microsporum* spp. was the most prevalent (42.5%), followed by *Trichophyton* spp. (30.0%). Yeast organisms such as *Malassezia* spp. (10.0%) and *Candida* spp. (5.0%) were also identified, while non-dermatophyte fungi including *Aspergillus* spp. (7.5%) and *Penicillium* spp. (5.0%) were detected in fewer cases. *Epidermophyton* spp. was not observed in any sample.

Table 2: Identification of fungal agent in dogs

Fungus	Number of infected animals	Percentage (%)
<i>Microsporum</i> spp.	17	42.5
<i>Trichophyton</i> spp.	12	30.0
<i>Malassezia</i> spp.	4	10.0
<i>Candida</i> spp.	2	5.0
<i>Aspergillus</i> spp.	3	7.5
<i>Penicillium</i> spp.	2	5.0
Total	40	100

Dermatophytes (*Microsporum* and *Trichophyton*) constituted the major fungal pathogens, indicating their predominant role in canine mycotic dermatitis. However, the lower prevalence of *Malassezia* spp. compared to earlier reports suggests geographical and population-based variation in fungal distribution. The occurrence of fungal isolates during the current study is shown in table 2. These findings indicated *Microsporum canis* as the predominant dermatophyte affecting dogs.

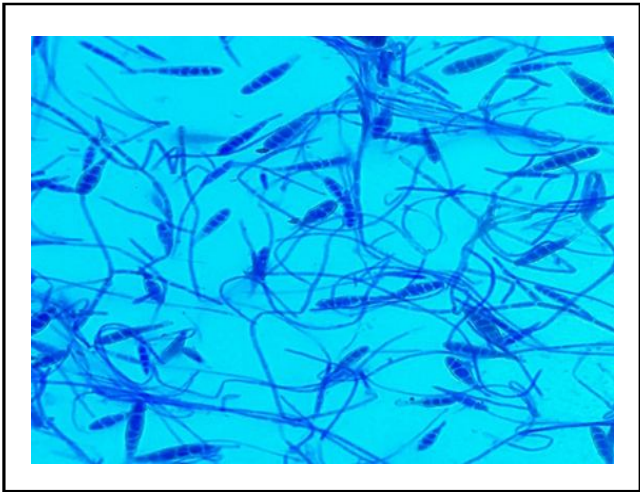


Figure 3: *Microsporum* spp. demonstrating septate hyphae and spores. under lactophenol cotton blue stain (40x).

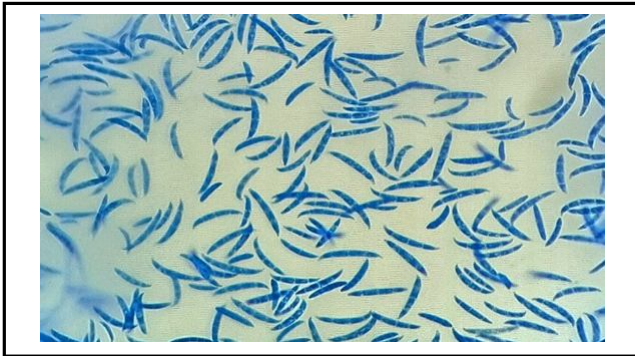


Figure 4: Macroconidia of *Trichophyton* spp. exhibiting distinct septa under lactophenol cotton blue stain (40x).

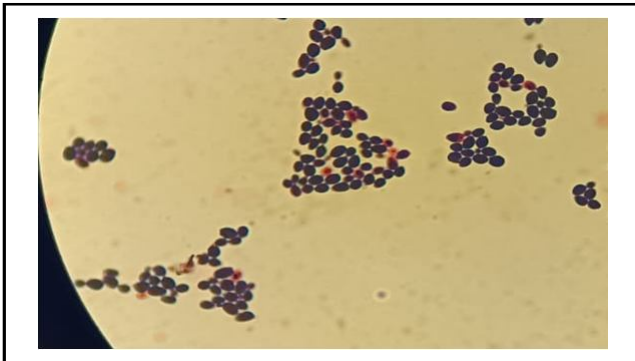


Figure 5: *Candida* spp. showing typical budding yeast cells under gram staining.

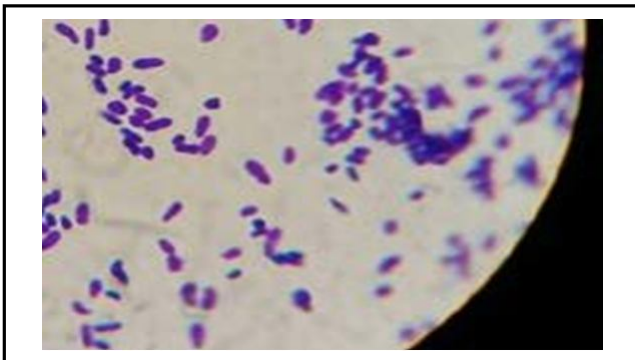


Figure 6: *Malassezia* spp. displaying characteristic budding yeast cells under gram stain.

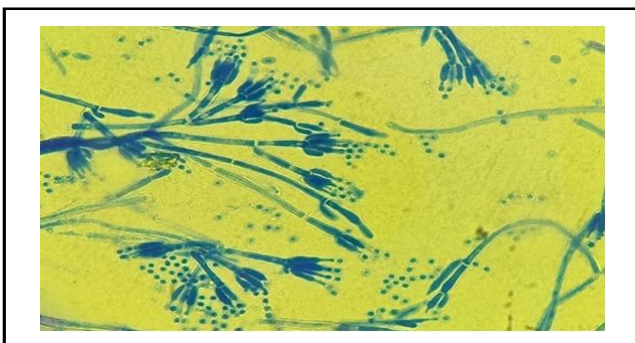


Figure 7: *Penicillium* spp. exhibiting branched conidiophores with chains of conidia under lactophenol cotton blue stain.

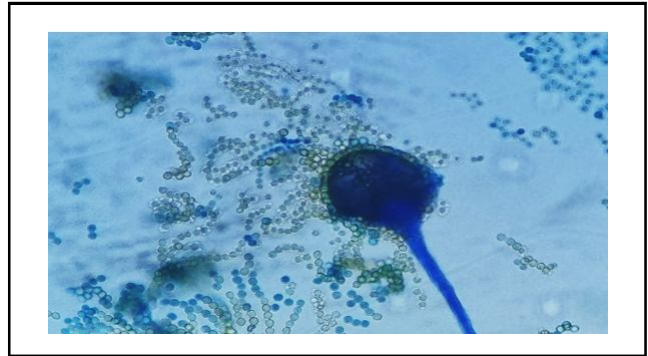


Figure 8: *Aspergillus* spp. showing characteristic conidiophore, vesicle, and conidia.

3.2 In vitro antifungal evaluation

The *C. tora* ointment demonstrated moderate antifungal activity compared with ketoconazole. The inhibition zones (mm) presented in table 3. This activity is attributed to anthraquinones, which disrupt fungal cell membranes and inhibit fungal enzymes (Kim *et al.*, 2004; Tripathi *et al.*, 2011). In this study *Candida* spp. and *Malassezia* spp. showed least inhibition whereas other species were greatly inhibited by *C. tora* ointment.

Table 3: Sensitivity pattern of fungal isolates against therapeutic agents (Diameter of the zone inhibition)

Organism	Zone of inhibition for 2% ketoconazole (mm)	Zone of inhibition for <i>C. tora</i> ointment (mm)
<i>Microsporum</i> spp.	17	12
<i>Trichophyton</i> spp.	20	13
<i>Malassezia</i> spp.	12	5
<i>Candida</i> spp.	10	4
<i>Aspergillus</i> spp.	16	10
<i>Penicillium</i> spp.	18	11

3.3 Clinical findings

Common clinical signs included pruritus, alopecia, scaling, crusting, and erythema, which are characteristic features of dermatophytosis (Medleau and Ristic, 1992; Beale, 2000). Generalized lesions were more common than localized lesions, indicating widespread infection.



Figure 9: Inflamed skin with mild papular eruptions.



Figure 10: Chronic inflammatory lesion exhibiting hyperpigmentation and early lichenification.

3.4 Hematobiochemical findings

Haematological evaluation revealed that haemoglobin (Hb), packed cell volume (PCV), total red blood cell count (TEC), and platelet count did not differ significantly between healthy and infected dogs and remained within physiological limits throughout the study, indicating that mycotic dermatitis did not markedly affect erythrocytic parameters or thrombocyte dynamics. The data on

Hematobiochemical parameter values are presented in table 4 for days 0 and 28 of the study. Following treatment, a mild, non-significant improvement in these parameters was observed, suggesting hematological safety of the therapeutic interventions.

On the other hand, total leucocyte count (TLC) and neutrophil percentage were significantly elevated in infected dogs at Day 0, indicating leukocytosis and neutrophilia associated with active fungal infection and inflammatory response. Following treatment, a statistically significant decrease ($p < 0.05$) in TLC and neutrophil levels was noted across both groups by Day 28, reflecting effective control of infection and resolution of inflammation. Lymphocyte, monocyte, eosinophil, and basophil percentages showed no significant variation, indicating that the infection primarily elicited a neutrophilic response rather than marked changes in other leukocyte populations.

Biochemical analysis revealed that serum enzymes such as ALT, AST, and ALP remained within normal physiological ranges and showed no significant variation before and after treatment, suggesting the absence of hepatic involvement and confirming the safety of the formulations. Total protein levels, although slightly lower in infected dogs initially, increased significantly following treatment, indicating improvement in nutritional status, immune response, and overall health.

Table 4: Comparative hematobiochemical changes in dogs with mycotic dermatitis (Day 0 vs Day 28)

Parameter	T1 (ketoconazole) Day 0	T1 (ketoconazole) Day 28	T2 (<i>C. tora</i>) Day 0	T2 (<i>C. tora</i>) Day 28
Hb (g/dl)	11.514 ± 0.109	11.626 ± 0.103	11.423 ± 0.114	11.520 ± 0.091
PCV (%)	42.519 ± 0.512	42.683 ± 0.372	41.695 ± 0.517	42.964 ± 0.708
TEC (×10 ⁶ /cu.mm)	5.446 ± 0.091	5.550 ± 0.048	5.308 ± 0.061	5.495 ± 0.063
TLC (×10 ³ /cu.mm)	18.657 ± 0.302 ^a	13.435 ± 0.438 ^b	18.128 ± 0.512 ^a	15.615 ± 0.445 ^b
Neutrophils (%)	75.84 ± 0.411 ^a	71.82 ± 0.430 ^b	77.38 ± 0.390 ^a	74.24 ± 0.537 ^b
Lymphocytes (%)	19.258 ± 0.498	19.432 ± 0.458	19.354 ± 0.857	19.531 ± 0.859
Platelets (×10 ³ /μl)	4.384 ± 0.165	4.588 ± 0.118	4.254 ± 0.265	4.414 ± 0.268
ALT (IU/L)	79.64 ± 1.733	79.64 ± 1.772	80.01 ± 1.479	80.28 ± 1.351
AST (IU/L)	103.42 ± 1.398	106.92 ± 1.407	104.02 ± 1.546	107.07 ± 1.489
ALP (IU/L)	98.59 ± 1.554	98.91 ± 1.568	98.68 ± 1.596	98.92 ± 1.684
Total protein (g/dl)	5.57 ± 0.058 ^a	7.88 ± 0.048 ^b	5.407 ± 0.053 ^a	6.307 ± 0.121 ^b

Different superscripts (a, b) within the same row denotes a statistically significant difference ($p < 0.05$).

3.5 Therapeutic evaluation

3.5.1 Topical ketoconazole treatment

Topical ketoconazole treatment demonstrated rapid and progressive clinical improvement in dogs affected with mycotic dermatitis over the period of 28 days. Severe clinical signs such as pruritus showed marked reduction by Day 14 and complete resolution by Day 28. Lesions including scales, crusts, kerion, papules, ulcers, and erosion resolved completely between Day 21 and Day 28, indicating strong antifungal efficacy.

Alopecia and rough hair coat exhibited gradual improvement; however, mild residual lesions persisted in a few animals at the end of the treatment period. Chronic lesions such as hyperpigmentation

and lichenification showed slower resolution, likely due to prolonged inflammation. No nodular lesions were observed throughout the study.

Of the 10 treated dogs, 7 (70%) achieved complete clinical recovery, with 2 by Day 21 and 5 by Day 28. The remaining 3 dogs showed incomplete response, possibly due to chronicity or deeper follicular involvement. Remaining dogs continued treatment with additional clinical aid.

The observed therapeutic results indicated effectiveness of topical ketoconazole in dermatophytosis. The drug acts by suppressing ergosterol synthesis *via* inhibition of lanosterol 14 α -demethylase, resulting in compromised fungal cell membrane integrity and ultimately cell death.

3.5.2 Therapeutic evaluation of (*C. tora*) semi-herbal ointment (10%)

Dogs treated with 10% *C. tora* ointment exhibited gradual but comparatively slower clinical improvement over the treatment period of 28 days. Reduction in lesions such as scales, crusts, and erosion was evident from Day 14 onwards, with complete resolution of these parameters by Day 28. However, clinical signs including pruritus, alopecia, kerion, erythema, and lichenification persisted in a proportion of dogs even at the end of the study, indicating a slower therapeutic response. Out of 10 treated dogs, 5 animals (50%) achieved complete clinical recovery, where three dogs recovering by Day 21 and the remaining two by Day 28. The remaining 5 dogs showed partial or inadequate response to therapy. Papules, ulcers, and nodules were not observed throughout the treatment period.

The observed moderate therapeutic efficacy of *C. tora* ointment demonstrates its antidermatophytic activity. The antifungal effect is primarily attributed to anthraquinone derivatives including emodin, chrysophanol, physcion, rhein, along with aloe-emodin, which disrupt fungal cell membrane integrity and inhibit growth. Additionally, protease inhibitory activity may reduce fungal virulence and tissue invasion.

Despite these properties, the comparatively lower recovery rate might be due to factors such as reduced penetration into keratinized tissues, variability in phytochemical concentration, and lower potency compared to synthetic antifungal agents. Although, the efficacy was lower than ketoconazole, the herbal ointment demonstrated significant clinical improvement, suggesting its potential as an alternative therapy.

Table 5: Therapeutic evaluation of topical ketoconazole preparation (group T1) against mycotic dermatitis in dogs (n = 10)

Clinical examination of skin	Days post treatment				
	0	7	14	21	28
Pruritus	++++ (7)	+++ (5)	++ (2)	+ (1)	-
Rough hair coat	+++ (6)	+++ (5)	++ (3)	++ (2)	++ (2)
Alopecia	++++ (5)	++++ (5)	+++ (4)	++ (2)	++ (1)
Scales	+++ (3)	++ (2)	++ (1)	-	-
Crusts	+++ (2)	++ (1)	+ (1)	-	-
Kerion	++ (2)	+ (1)	-	-	-
Erythema	+++ (3)	++ (2)	+ (1)	+ (1)	-
Papules	+ (1)	+ (1)	-	-	-
Hyperpigmentation	++ (4)	++ (3)	+ (2)	+	+
Ulcers	+ (1)	+ (1)	+	-	-
Lichenification	++ (2)	++ (2)	+ (1)	+	-
Nodules	-	-	-	-	-
Erosion	++ (2)	++	+	-	-

Table 6: Therapeutic evaluation of formulated (*C. tora*) semi-herbal ointment (10%) (Group T2) against mycotic dermatitis in dogs (n = 10)

Clinical examination of skin	Days post treatment				
	0	7	14	21	28
Pruritus	+++ (7)	+++ (5)	++ (4)	+ (3)	+ (2)
Rough hair coat	+++ (6)	+++ (5)	++ (3)	+ (2)	+ (1)
Alopecia	++++ (5)	+++ (4)	++ (4)	++ (2)	++ (1)
Scales	+++ (3)	++ (2)	+ (2)	+ (1)	-
Crusts	+++ (2)	++ (2)	+ (2)	+ (1)	-
Kerion	++ (3)	++ (2)	++ (1)	+ (1)	+
Erythema	+++ (2)	++ (2)	+ (1)	+ (1)	+
Papules	-	-	-	-	-
Hyperpigmentation	++ (4)	++ (3)	++ (2)	+ (2)	+ (1)
Ulcers	-	-	-	-	-
Lichenification	++ (3)	++ (2)	++ (1)	++ (2)	+
Nodules	-	-	-	-	-
Erosion	++ (3)	++ (2)	++ (2)	+ (1)	-



Figure 11: Before treatment: ventral cervical lesion characterized by erythema, alopecia, and hyperpigmentation.



Figure 12: After treatment: healing of ventral cervical lesion showing reduction in erythema and regrowth of hair.



Figure 13: Before treatment: Multifocal lesions characterized by alopecia and erythematous patches.



Figure 14: After treatment: Recovery showing reduction in erythema and restoration of hair coat.

4. Discussion

The present study revealed that mycotic dermatitis accounted for 21.02% of dermatological cases in dogs, indicating that fungal skin diseases remain an important clinical problem in veterinary canine dermatology. The occurrence observed in the present investigation closely corresponds with earlier reports documenting incidences ranging from approximately 17-20% in canine dermatological disorders (Chittawar and Rao, 1982; Bisui, 2005; Kalita *et al.*, 2013). Environmental factors such as warm and humid climatic conditions prevailing in tropical regions may favour fungal growth and transmission, thereby increasing the occurrence of mycotic dermatitis.

Among the fungal isolates identified in the present study, *Microsporum* spp. was the predominant organism followed by *Trichophyton* spp., whereas *Malassezia* spp., *Candida* spp., *Aspergillus* spp., and *Penicillium* spp. were isolated less frequently. These findings are in close agreement with previous studies reporting *Microsporum canis* as the major dermatophyte associated with canine dermatophytosis (Cafarchia *et al.*, 2004; Copetti *et al.*, 2006). The predominance of *Microsporum* spp. may be attributed to its highly contagious nature, ability to persist in the environment, and effective transmission through infected hairs, fomites, and asymptomatic carrier animals. *Trichophyton* spp. constituted the second major isolate

in the present study, supporting earlier reports indicating significant involvement of *Trichophyton* species in canine fungal dermatitis. The isolation of non-dermatophyte fungi such as *Aspergillus* spp., *Penicillium* spp., *Candida* spp., and *Malassezia* spp. observed during the present study further supports previous findings indicating that opportunistic fungi and yeasts may act either as primary pathogens or secondary invaders in compromised skin conditions (Sturzu *et al.*, 2002; Joshi, 2019). The comparatively lower prevalence of *Malassezia* spp. in the present study compared to earlier reports may be attributed to geographical variation, climatic differences, host factors, and differences in sampling populations.

Direct microscopic examination using KOH preparation and lactophenol cotton blue staining revealed fungal elements in the majority of cases, whereas fungal culture yielded a higher positivity rate. Similar observations have been reported previously, emphasizing that fungal culture remains the gold standard for definitive diagnosis of dermatophytosis (Kumar *et al.*, 2002; Shalaby *et al.*, 2016). The combined use of direct microscopy and culture in the present study improved diagnostic accuracy and facilitated species identification. The successful isolation of fungal organisms on sabouraud's dextrose agar and dermatophyte test medium further corroborates earlier findings recommending combined cultural and microscopic examination for reliable diagnosis of canine dermatomycosis (Kumar *et al.*, 2002; David *et al.*, 2004). Clinically, affected dogs in the present study exhibited pruritus, alopecia, erythema, scaling, crusting, hyperpigmentation, kerion formation, and lichenification. These findings are in accordance with previous reports describing alopecia, scaling, erythema, crust formation, and pruritus as characteristic manifestations of dermatophytosis (Beale, 2000; Foil, 1998). Generalized lesions were more common than localized lesions in the present investigation, suggesting widespread infection and chronicity in many affected animals. Kerion lesions observed in certain dogs may be associated with exaggerated inflammatory reactions commonly produced by dermatophyte infections.

Haematological evaluation during the present study revealed significant elevation in total leucocyte count and neutrophil percentage in infected dogs prior to treatment, indicating an inflammatory and infectious responses associated with active fungal infection. Similar findings of leukocytosis and neutrophilia have been reported previously (Scott *et al.*, 2001; Sindha *et al.*, 2015). However, haemoglobin, packed cell volume, total erythrocyte count, and platelet count remained within normal physiological limits and did not vary significantly, indicating minimal alterations in erythrocytic parameters in uncomplicated dermatophytosis.

Following treatment, significant reductions in total leucocyte count and neutrophil percentage were observed in both therapeutic groups, indicating resolution of infection and a reduction in the inflammatory response. The non-significant changes in lymphocyte and platelet counts suggest that the infection predominantly elicited neutrophilic inflammatory reactions rather than generalized hematological disturbances. Biochemical analysis revealed that serum ALT, AST, and ALP values remained within normal physiological ranges throughout the study, indicating absence of hepatic involvement and safety of the therapeutic agents used. Total protein levels increased significantly following treatment, possibly due to improvement in nutritional status, immune response, and recovery from chronic inflammatory conditions. *In vitro* antifungal evaluation demonstrated that ketoconazole exhibited greater antifungal efficacy than *C. tora*

ointment against all isolated fungal species. The superior efficacy of ketoconazole observed in the present study is consistent with earlier reports documenting its strong antidermatophytic activity against *Microsporum* and *Trichophyton* species (Kumar *et al.*, 2002; Shalaby *et al.*, 2016). Ketoconazole acts by inhibiting lanosterol 14- α -demethylase, thereby interfering with ergosterol synthesis and compromising fungal cell membrane integrity.

The formulated *C. tora* ointment also demonstrated appreciable antifungal activity, although somewhat lower than that of ketoconazole. The antifungal effect observed may be attributed to anthraquinone compounds such as emodin, chrysophanol, physcion, rhein, and aloe-emodin present in *C. tora* seeds and leaves (Kim *et al.*, 2004; Charoenchai and Chankana, 2018). Additionally, protease inhibitory activity of *C. tora* may reduce fungal virulence and tissue invasion, thereby contributing to therapeutic efficacy (Tripathi *et al.*, 2011). Therapeutically, dogs treated with topical ketoconazole exhibited more rapid complete clinical recovery than those treated with *C. tora* ointment. Complete clinical recovery was observed in 70% of ketoconazole-treated dogs compared to 50% recovery in the *C. tora* group. These findings agree with earlier studies reporting high therapeutic efficacy of ketoconazole in canine dermatophytosis (Kalita *et al.*, 2013). Rapid reduction in pruritus, erythema, crusts, and scaling following ketoconazole therapy may be attributed to its potent fungicidal activity and ability to effectively penetrate keratinized tissues. The comparatively slower recovery observed in dogs treated with *C. tora* ointment might be due to lower potency, reduced tissue penetration, or variability in phytochemical concentration within herbal preparations. Nevertheless, the herbal ointment demonstrated gradual clinical improvement with reduction in scaling, crusting, erythema, and pruritus, indicating its potential as an alternative or supportive therapy for management of canine mycotic dermatitis (Srivastava *et al.*, 2017; Shadab *et al.*, 2019).

Overall, the findings of the present study confirm that dermatophytes, particularly *Microsporum* spp. and *Trichophyton* spp., are major etiological agents of canine mycotic dermatitis. Combined microscopic and cultural examination remains essential for accurate diagnosis, while ketoconazole continues to provide superior therapeutic efficacy. The formulated *C. tora* ointment demonstrated promising antifungal potential and may serve as a cost-effective semi-herbal alternative for supportive management of fungal dermatitis in dogs.

5. Conclusion

C. tora ointment demonstrated moderate antifungal efficacy with a 50% recovery rate in canine mycotic dermatitis. The formulation was observed to be safe and effective in alleviating clinical signs. It may serve as an economical alternative or adjunct to conventional antifungal therapy. Further studies focusing on formulation enhancement and combination therapy are recommended.

Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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Citation

M. B. Bukke, V. V. Karande, B. C. Ghumare, U. M. Tumlam, A. K. Barate, A. S. Khairmode and P. D. Pokharkar (2026). Evaluation of antifungal potential of *Cassia tora* L. ointment in canine mycotic dermatitis. *Ann. Phytomed.*, **15**(1):736-744, <http://dx.doi.org/10.54085/ap.2026.15.1.66>.