



Investigation of antifungal potency of capsaicin and tea tree oil against oral infection in pediatric patients

Zakeeyah Saliam Mohammed Qajjam ^{1*}, Rumadhanah Abraheem Mohammed ²
^{1,2} Department of Botany, Faculty of Science, University of Zawiya, Al-ajalat, Libya

دراسة فعالية الكابيسيسين وزيت شجرة الشاي كمضادات فطرية في عدوى الفم عند الأطفال

زكية سالم محمد قجام ^{1*}، رمضانة ابراهيم محمد النقا ²
^{2,1} قسم النبات، كلية العلوم، جامعة الزاوية، العجيلات، ليبيا

*Corresponding author: z.qajjam@zu.edu.ly

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Abstract

Oral fungal infections, primarily caused by *Candida* species, are prevalent in pediatric patients. The antifungal efficacy of capsaicin and tea tree oil appears promise; however, their efficiency against *Candida* species in oral infections has been largely unexamined. This study evaluated the antifungal efficacy of capsaicin and tea tree oil against *Candida albicans* and *Candida tropicalis* in pediatric patients with oral fungal infections. This is a cross-sectional, laboratory-based investigation conducted from March to June 2024. The study included children aged 2 weeks to 24 months diagnosed with oral *Candida* infections. Oral swabs were collected from the diseased areas, and *Candida* species were identified with Sabouraud Dextrose Agar, germ tube assays, and CHROMagar *Candida*. Antifungal susceptibility testing was conducted against capsaicin and tea tree oil using a twofold serial dilution in liquid media. This study included 30 children, predominantly male (55%), aged between 6 and 12 months. Of the identified *Candida* species, *Candida albicans* comprised 70%, while *Candida tropicalis* constituted 30%. Maternal attributes indicated that 40% of mothers possessed a high school education, whilst 60% participated in breastfeeding. Capsaicin and tea tree oil exhibited antifungal properties, with capsaicin demonstrating superior fungicidal efficacy, especially against *Candida albicans*. Conversely, tea tree oil shown activity, mostly in a fungistatic capacity, against *Candida tropicalis*. Capsaicin and tea tree oil have antifungal properties against *Candida albicans* and *Candida tropicalis*, with capsaicin proving to be more effective. These natural compounds may serve as supplementary therapy to conventional antifungals in pediatric oral infection cases.

Keywords: *Candida*, fungi, pediatric, antifungal, oral infection.

المخلص

تعد العدوى الفطرية الفموية، والتي تُسبب بشكل رئيسي بواسطة أنواع *Candida*، من المشكلات الشائعة لدى المرضى الأطفال. وقد أظهرت الفعالية المضادة للفطريات لكل من الكابيسيسين وزيت شجرة الشاي نتائج واعدة؛ إلا أن كفاءتهما ضد أنواع *Candida* في حالات العدوى الفموية ما تزال غير مدروسة بشكل كافٍ. هدفت هذه الدراسة إلى تقييم الفعالية المضادة للفطريات للكابيسيسين وزيت شجرة الشاي ضد *Candida albicans* و *Candida tropicalis* لدى الأطفال المصابين بعدوى فطرية فموية. أجريت هذه الدراسة المقطعية المعتمدة على المختبر خلال الفترة من مارس إلى يونيو 2024. شملت الدراسة أطفالاً تتراوح أعمارهم بين أسبوعين و24 شهراً تم تشخيصهم بعدوى فطرية فموية ناتجة عن *Candida*. جُمعت مسحات فموية من المناطق المصابة، وتم التعرف على أنواع *Candida* باستخدام وسط Sabouraud Dextrose Agar، واختبار أنبوب الإنبات (Germ tube test)، ووسط CHROMagar *Candida*. كما أُجري اختبار الحساسية لمضادات الفطريات للكابيسيسين وزيت شجرة الشاي باستخدام طريقة التخفيف التسلسلي الثنائي في وسط سائل. شملت الدراسة 30 طفلاً، غالبيتهم من الذكور (55%)، وتتراوح أعمارهم بين 6 و12 شهراً. ومن بين أنواع *Candida* المعزولة، شكّلت *Candida albicans* نسبة 70%، في حين بلغت نسبة *Candida tropicalis* نحو 30%. وأظهرت الخصائص المتعلقة بالأمهات أن 40% منهن يحملن شهادة الثانوية العامة، بينما كانت نسبة الرضاعة الطبيعية 60%. أظهر كل من الكابيسيسين وزيت شجرة الشاي خصائص مضادة للفطريات، حيث أبدى الكابيسيسين فعالية قاتلة للفطريات أعلى، لا سيما ضد *Candida albicans*. وعلى النقيض، أظهر زيت شجرة الشاي نشاطاً مضاداً للفطريات في الغالب بصيغة مثبّطة للنمو (fungistatic)، وخاصة ضد *Candida tropicalis*.

تستنتج الدراسة أن الكابيسين وزيت شجرة الشاي يمتلكان خصائص مضادة للفطريات ضد *Candida albicans* و *Candida tropicalis*، مع تفوق الكابيسين من حيث الفعالية. وقد تمثل هذه المركبات الطبيعية خيارًا مساعدًا للعلاج التقليدي بمضادات الفطريات في حالات العدوى الفطرية الفموية لدى الأطفال.

الكلمات المفتاحية: الكانديدا، الفطريات الفموية، الأطفال، مضادات الفطريات الطبيعية، عدوى الفم.

Introduction

Approximately 200 recognized *Candida* species are classified within the Saccharomycetes order of fungus. Of them, 20 can induce human illnesses [1]. The range of *Candida* infections extends from superficial mucosal conditions such as oral thrush and vulvovaginal candidiasis to severe systemic infections, particularly in hospitalized or immunocompromised individuals [2]. Multiple factors influence the virulence of *Candida* species, such as biofilm formation, the production of hydrolytic enzymes, and phenotypic flipping [3].

Oral fungal infections, particularly those induced by *Candida species*, represent a significant health concern in pediatric patients, notably in neonates and immunocompromised children [4]. Oral thrush manifests as white plaques on the tongue, inner cheeks, and palate, accompanied by discomfort and feeding challenges [5, 6].

Adhesion is a critical factor in the pathogenicity of *Candida spp.*, facilitating its colonization of the oral mucosa and subsequent infection. *Candida species*, particularly *C. albicans*, have adhesions and surface proteins that facilitate adherence to epithelial cells and extracellular matrix constituents [7, 8]. Subsequently, attached *Candida species* develop biofilms: structured assemblages of fungal cells encased in an extracellular matrix [9]. Biofilms safeguard against environmental pressures and antifungal drugs by sustaining infections [10].

Candida spp. synthesize hydrolytic enzymes, including proteases, lipases, and phospholipases, that decompose host tissues and facilitate invasion. The secreted aspartyl proteases are essential for compromising epithelial barriers and degrading host immunological elements, such as antibodies. Additionally, *Candida* has a phenotypic transition between yeast and hyphal forms, with the hyphal form demonstrating greater invasiveness. It also attempts to evade the host immune system by synthesizing candidalysin, a protein that damages epithelial cells and suppresses immunological responses. The fungus can alter the host immune response by disrupting cytokine signaling and evading detection by immune cells through the concealment of surface antigens with mannan-rich glycans [11, 12].

Antifungal resistance is a direct result of the pathogenic processes in *Candida spp.* Biofilms create both a physical and biochemical barrier to antifungal medications, diminishing penetration and causing sub-lethal exposure of fungal cells, which fosters the emergence of resistance [13, 14]. Active transport efflux pumps include those produced by the CDR and MDR genes, which aggressively expel antifungal drugs from cells. Alterations in the ergosterol production pathway, which is the target of most antifungal agents, result in reduced susceptibility [13, 15, 16].

The management of *Candida* infections typically requires antifungal drugs, including azoles such as fluconazole, polyenes like nystatin and amphotericin B, and echinocandins such as caspofungin. Such chemicals target fungal cell membranes or walls to impede growth and induce the demise of fungal cells [17, 18].

The pharmacokinetics of antifungal agents in pediatric populations varied markedly, necessitating meticulous dose modification to prevent toxicity or inadequate therapeutic levels [19]. The restricted availability of pediatric-friendly formulations, such as oral suspensions, hinders treatment adherence. The immature immune system in neonates and pediatric patients may extend recovery time and heighten vulnerability to repeated infections [20, 21].

The increasing resistance to synthetic antifungal agents and their severe side effects have generated interest in phytotherapeutic alternatives. Bioactive phytoconstituents from medicinal plants serve as a significant source of potent antifungal agents. In addition to providing novel mechanisms of action, these agents align with the contemporary movement towards sustainable and environmentally acceptable therapeutic approaches [22, 23, 24]. Natural compounds generally exhibit reduced toxicity and enhanced biocompatibility relative to manufactured pharmaceuticals. Their extensive efficacy and minimal likelihood of resistance emergence render them intriguing choices for antifungal treatment. The multifunctional properties of plant agents, including concurrent antifungal, anti-inflammatory, and immunomodulatory actions, enhance their therapeutic potential, particularly in vulnerable populations such as children [25, 26, 27].

Safety and tolerability are critical factors in antifungal treatment for pediatric populations. Capsaicin and tea tree oil have demonstrated minimal systemic antifungal efficacy at suitable dosages. Their natural nature and less risk of severe side effects render these products attractive as prospective therapies for fungal infections in children [28, 29, 30].

This study is to evaluate the effectiveness of capsaicin and tea tree oil as antifungal agents against *Candida species* implicated in juvenile oral candidiasis. The objective is to investigate their viability as secure and efficacious substitutes for traditional antifungals, tackling resistance challenges and side effects while advocating for plant-derived therapeutic alternatives.

Material and methods

Study design: The present study is a cross-sectional, laboratory-based investigation aimed to assess the antifungal potential of capsaicin and tea tree oil against *Candida species*, which cause oral infections in pediatric patients.

The study was conducted in AL-Jamil hospital in Al-Jmail city, Libya and continued for 4 months from the beginning of march 2024 till the end of June 2024.

Inclusion and exclusion criteria: The inclusion criteria are children aged 2 weeks to 24 months exhibiting evident manifestations of oral fungal infection, such as white lesions on the tongue, inner buccal mucosa, or palatal region. Consent was obtained from the parents or guardians for the involvement of the child, but children without a history of any chronic systemic disorders that may cause immunity, such as cancer or HIV-positive status, did not be excluded from this experiment.

The exclusion criteria encompassed children with documented allergies to chili peppers, tea tree oil, or analogous substances, as well as those who have undergone antifungal therapy within the two weeks preceding the experiment. Other oral infections, excluding those caused by *Candida*, such as bacterial infections, was also omitted to ensure the trial remains concentrated on *Candida* oral infections.

Data collection: Demographic data about the child and his mum was collected in an organized questionnaire from the patients attending AL-Jmail hospital. The data needed include age of both child and mother, residence, gender, type of feeding and mother's education level.

Samples collection: Each participant's oral cavity was swabbed for infections in areas such as the tongue and inner cheeks. Samples were obtained using sterile swabs and promptly transported to the laboratory for further processing.

Isolation and Identification of *Candida* Species: Oral swabs were collected and inoculated into Sabouraud Dextrose Agar plates, which were supplemented with chloramphenicol antibiotic to suppress bacteria growth. The plates will next be incubated at 30-37°C for 24–48 hours. The colonies on the SDA plates were examined for characteristic *Candida* traits such as creamy, smooth, and elevated colonies [31].

Germ tube test: One colony of *Candida* was placed into a sterile test tube containing 1 mL of broth medium. The tube was then incubated at 37°C for 2 to 3 hours, allowing germ tubes to form. After incubation, a wet mount was created by applying a little amount of growth medium on a clean glass slide and covering it with a coverslip. Microscopy (40× magnification) was used to examine the sample [32].

CHROMagar *Candida* for Differentiation of *Candida* Species: The *Candida* sample was streaked directly onto a CHROMagar plate using a sterile inoculation loop. The plate was then placed in an incubator at 37°C for 24–48 hours, allowing *Candida species* to grow. During this incubation, colonies changed colors due to chromogenic reactions. Following incubation, the colony morphology and color were observed [33].

Antifungal Testing: 30 mg of capsaicin powder was dissolved in 3 ml of absolute ethanol (99.9%), and 3 ml of tea tree oil was dissolved in 3 ml of ethanol. The suspension was mixed well using a vortex mixer to ensure that all the powder dissolved. Further, the stock solution was sterilized in a water bath for 10 minutes at 60°-63°C. Twofold serial dilution of capsaicin and tea tree oil from 20% to 80% was done in sterile tubes. A suspension of *Candida species* was prepared in sterile saline to match a 0.5 McFarland standard (~1-5 x 10⁶ CFU/mL) [34, 35].

For accurate antifungal testing, control tubes were constructed. To test antifungal effectiveness, the positive control tube has fluconazole. To demonstrate unrestricted growth, the negative control tube contained only *Candida* inoculum without antifungals. A solvent control tube lacking the active component was utilized to test fungal growth. To promote fungal development, these tubes were incubated at 37°C with shaking for 24–48 hours. After incubation, the optical density of each tube was measured at 475 nm using a spectrophotometer to assess fungal growth. The MIC was defined as the lowest concentration of capsaicin and tea tree oil exhibiting a minimum of 90% suppression of fungal growth relative to the negative control. To ascertain the MFC, 100 µL were extracted from tubes exhibiting MIC and higher concentrations, and subsequently plated on SDA to assess regrowth. The samples were incubated at 37°C for 48 hours. The MFC was established as the minimal concentration at which no fungal regrowth was detectable, indicating fungicidal efficacy.

Results and discussion:

Sociodemographic characteristics distribution among pediatric patients:

The sociodemographic data presented outlines essential aspects of the pediatric patients in the study as shown in table 1. The predominant age groups of the patients are younger, with 36.7% aged 1-6 months and 33.3% aged 7-12 months. Regarding gender, 60% of the patients are male and 40% are female, which may indicate sample variability or disparities in healthcare-seeking habits rather than an authentic gender-based tendency. A majority of patients (63.3%) live in rural regions, whilst 36.7% inhabit metropolitan areas. Breastfeeding is the primary feeding method, observed in 80% of instances, whilst bottle feeding constitutes 20%.

Table 1: Distribution of Sociodemographic of participants.

		No.	%
Age per month	1-6	11	36.7%
	7-12	10	33.3%
	13-18	4	13.3%
	19-24	5	16.7%
Gender	male	18	60%
	female	12	40%
residence	Rural	19	63.3%
	Urban	11	36.7%
feeding	Breast	24	80%
	Bottle	6	20%

Mother’s characteristics:

The predominant age group of mothers is 26-35 years, with 53.4%, followed by 43.3% who are under 25 years, and a mere 3.3% who are above 35 years. Concerning education, 46.6% of moms possess a secondary education, 36.7% have attained only a primary education, and 16.7% hold a university degree. Moreover, 50% of moms reliant on breastfeeding reported getting breast mastitis, while 30% did not encounter this condition, as table 2 indicates.

Table 2: distribution of Mothers’ characteristics.

		No.	%
Age of mother	<25	13	43.3%
	26-35	16	53.4%
	>35	1	3.3%
Mother's education level	Primary	11	36.7%
	Secondary	14	46.6%
	university	5	16.7%
Breast mastitis of mother	Yes	15	50%
	No	9	30%

Prevalence of candida albicans and candida tropicalis in oral cavity of participants:

Candida albicans was detected in 63.3% of cases, establishing it as the predominant pathogen where, *Candida tropicalis* was observed in 16.7% of patients. Notably, 20% of subjects demonstrated co-infection with both *Candida albicans* and *Candida tropicalis*, as figure 1 illustrates.

Prevalence of candida spp.

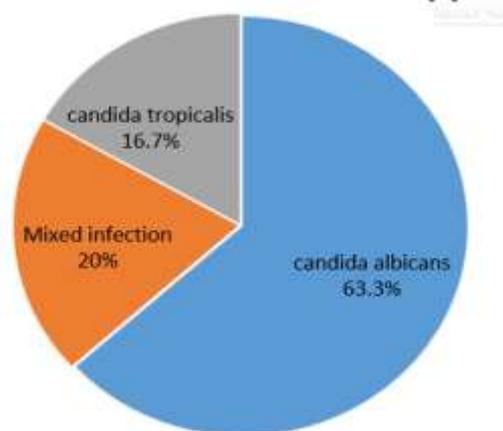


Figure 1: prevalence of *Candida* spp. among children.

Isolation and biochemical identification of *Candida* spp.:

The swift and characteristic germ tube production within 3 hours provides robust evidence for the identification of *C. albicans*, as this trait is a hallmark of the species with significant diagnostic accuracy. The inconsistent or delayed production of germ tubes, as observed in the 6-hour results, indicates the presence of *C. tropicalis* as observed in figure 2 .

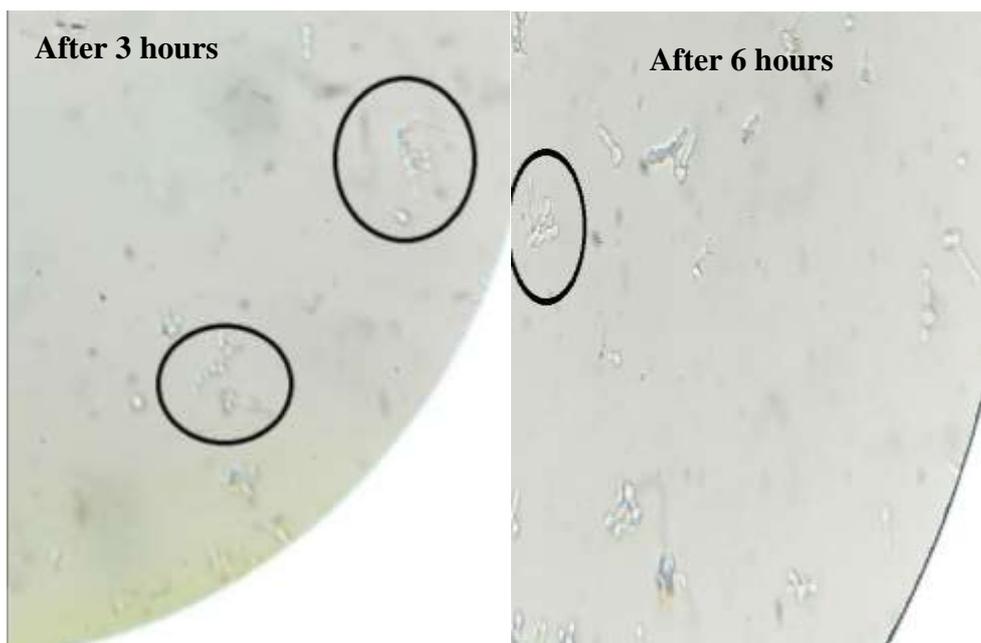


Figure 2: Germ tube of candida spp. under microscope.

Figure 3, depicting the CHROMagar test, effectively demonstrates the utility of this medium in differentiating *Candida species* based on colony color and form. CHROMagar *Candida albicans* typically forms green colonies, facilitating its swift identification. Conversely, *Candida tropicalis* forms colonies that are blue, providing a distinct contrast with other species.



Figure 3: differentiation between *Candida spp.* according to change in colours of colonies on CHROM agar media.

Antifungal activity of tea tree oil and capsaicin against isolated *Candida spp.*:

Tea tree oil demonstrated varying minimum inhibitory concentrations (MIC) and minimum fungicidal concentrations (MFC), ranging from 20% to 80%, with effects shifting between fungistatic and fungicidal. This variability may reflect differences in fungal susceptibility across isolates of *Candida albicans*, with fungicidal activity being predominant at lower concentrations (e.g., MIC/MFC of 20%). Conversely, capsaicin displayed more consistent MIC and MFC values, typically at higher concentrations (80% in most cases), and was largely fungicidal, although occasional fungistatic effects were observed.

Table 3: antifungal activity, MIC and MFC of tea tree oil and capsaicin and tea tree oil against *Candida albicans*.

	Tea tree oil			Capsasin		
	MIC	MFC	Effect	MIC	MFC	Effect
1	60%	60%	fungicidal	80%	80%	fungicidal
2	40%	40%	fungicidal	80%	80%	fungicidal
3	40%	60%	fungistatic	80%	80%	fungicidal
4	20%	20%	fungicidal	80%	80%	fungicidal
5	40%	60%	fungistatic	80%	80%	fungicidal
6	40%	40%	fungicidal	80%	80%	fungicidal
7	40%	60%	fungistatic	20%	40%	fungistatic
8	40%	60%	fungistatic	80%	80%	fungicidal
9	20%	20%	fungicidal	20%	60%	fungistatic
10	80%	80%	fungicidal	80%	80%	fungicidal
11	40%	40%	fungicidal	80%	80%	fungicidal
12	20%	80%	fungistatic	80%	80%	fungicidal
13	20%	60%	fungistatic	80%	80%	fungicidal
14	80%	80%	fungicidal	80%	60%	fungistatic
15	20%	60%	fungistatic	60%	60%	fungicidal
16	80%	80%	fungicidal	60%	60%	fungicidal
17	20%	40%	fungistatic	60%	60%	fungicidal
18	20%	20%	fungicidal	60%	80%	fungicidal
19	80%	80%	fungicidal	40%	60%	fungistatic

The MIC results for capsaicin and tea tree oil against *Candida albicans* reveal unique antifungal activity patterns between the two substances. Capsaicin demonstrated its minimum inhibitory concentration mostly at elevated levels, with 63.15% of isolates necessitating 80% for inhibition, signifying restricted effectiveness at lower doses. Only 10.53% and 5.26% of isolates exhibited susceptibility at 20% and 40%, respectively, whereas 21.06% demonstrated susceptibility at 60%.

Conversely, tea tree oil exhibited a more advantageous MIC distribution, with the majority of isolates (36.84%) inhibited at concentrations between 20% and 40%. This suggests that tea tree oil effectively inhibits most isolates at lower doses. A lesser percentage (5.26%) demonstrated vulnerability at 60%, whereas 21.06% necessitated 80%, indicating that tea tree oil possesses superior potency and a wider spectrum of activity in comparison to capsaicin.

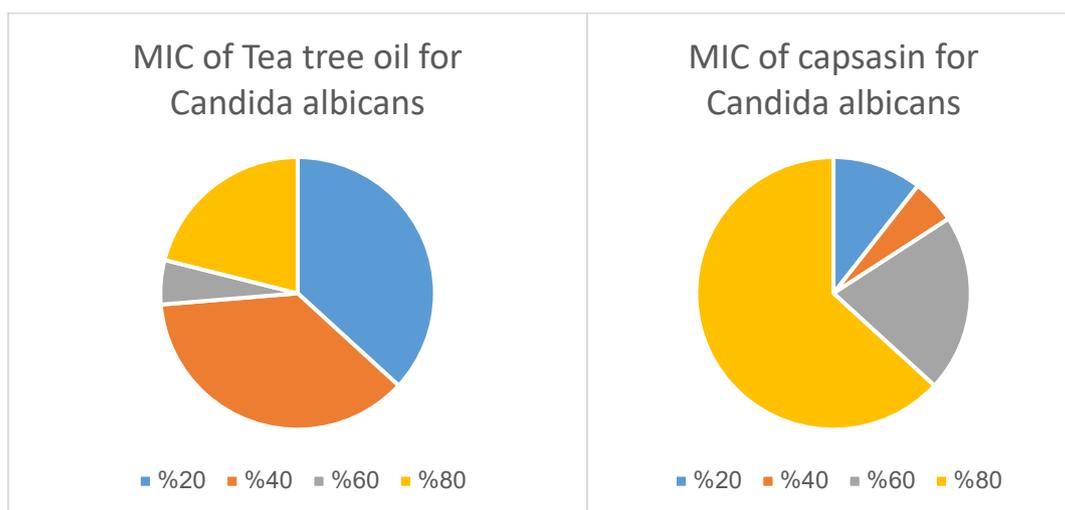


Figure 4: MIC of tea tree oil and capsaicin for *Candida albicans*.

The majority of tea tree oil isolates (60%) exhibited fungistatic activity at a concentration of 60%, with the minimum fungicidal concentration (MFC) at 80%. This suggests that tea tree oil efficiently suppresses the proliferation of *Candida tropicalis* at moderate doses. In certain isolates (20%), tea tree oil exhibited fungicidal activity at a 20% minimum inhibitory concentration (MIC).

Conversely, capsaicin demonstrated a far stronger fungicidal impact, with MIC values reaching as low as 20% for the majority of isolates, indicating its fungicidal properties. The minimum fungicidal concentration (MFC) for capsaicin was also modest (20% for certain isolates), corroborating its ability to not only inhibit but also eradicate *Candida tropicalis* at comparatively low doses.

Table 4: antifungal activity, MIC and MFC of tea tree oil and capsaicin and tea tree oil against *Candida tropicalis*.

	Tea tree oil			Capsasin		
	MIC	MFC	Effect	MIC	MFC	Effect
1.	60%	80%	fungistatic	20%	20%	fungicidal
2.	40%	60%	fungistatic	20%	20%	fungicidal
3.	20%	20%	fungicidal	60%	80%	fungistatic
4.	20%	20%	fungicidal	60%	80%	fungistatic
5.	60%	80%	fungistatic	60%	60%	fungicidal

The findings indicate that *Candida tropicalis* exhibits a notable resistance to capsaicin. No inhibition was detected at 40% or 80%, and only 60% of isolates exhibited inhibition at 60%, with 40% inhibited at 40%. Conversely, tea tree oil demonstrated a more reliable antifungal efficacy. At 20% and 40% concentrations, 40% and 20% of the isolates exhibited inhibition, respectively, with a more pronounced inhibition observed at 60% (40%). No inhibition was observed at 80%. *C. tropicalis* has heightened vulnerability to tea tree oil compared to capsaicin.

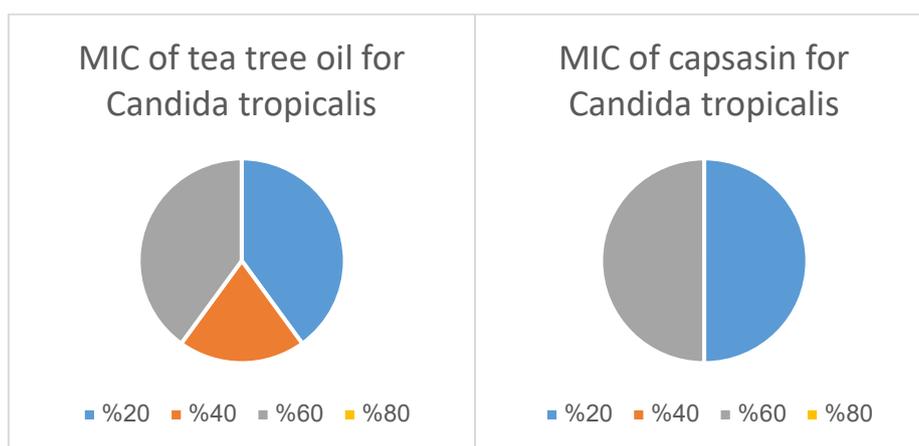


Figure 5: MIC of tea tree oil and capsasin for *Candida tropicalis*.

Effect of tea tree oil and capsasin on both *Candida albicans* and *Candida tropicalis*:

The antifungal properties of tea tree oil and capsasin against *Candida albicans* and *Candida tropicalis* exhibit notable disparities in effectiveness as presented in table 5. Tea tree oil shown a primarily fungicidal impact against *Candida albicans*, resulting in the death of 57.89% of isolates, while 42.11% exhibited fungistatic activity, indicating growth inhibition without cell death. Conversely, capsasin demonstrated more potency, resulting in the killing of 78.94% of isolates, while merely 21.06% shown fungistatic properties. This suggests that capsasin is especially potent against *C. albicans*, with most isolates eliminated at reduced doses.

In the case of *Candida tropicalis*, tea tree oil shown a higher propensity for fungistatic action, inhibiting the growth of 60% of isolates without causing death, while only 40% showed fungicidal effect. This indicates that tea tree oil is less efficacious against *C. tropicalis* compared to *C. albicans*. Conversely, capsasin exhibited a more potent antifungal action against *C. tropicalis*, resulting in having fungicidal effect against 60% of *Candida tropicalis* isolates and 40% displaying fungistatic activity.

Table 5: Antifungal activity of Tea tree oil and capsaicin against *Candida albicans* and *Candida tropicalis*.

		<i>Candida albicans</i>		<i>Candida tropicalis</i>	
		No.	%	No.	%
Tea tree oil	fungicidal	11	57.89%	2	40%
	fungistatic	8	42.11%	3	60%
capsaicin	fungicidal	15	78.94%	3	60%
	fungistatic	4	21.06%	2	40%

Discussion:

Oral candidiasis is one of the most common fungal infections among pediatric patients, mostly caused by *Candida albicans* and *Candida tropicalis*. The conventional therapy for fungal infections is effective but encounters some drawbacks, such as resistance and adverse effects, thus encouraging the finding of other therapies. Among natural molecules, capsaicin and tea tree oil, for its antimicrobial properties, represent candidates of interest for their antifungal action. The study explores the prevalence of *Candida* in young children and the antifungal properties of capsaicin and tea tree oil. The results are expected to contribute to the possible application of natural agents in treating oral fungal infections of pediatric patients.

The current investigation identified the greatest prevalence of oral candidiasis in children between 1 and 6 months of age. This finding corresponds with a 2023 study that indicated a notable prevalence of oral candidiasis in children aged 0 to 2 years, implying a comparable trend in the susceptibility of younger demographics to oral fungal infections [36]. A 2022 study indicated that children aged 1 to 5 years were the most adversely affected by oral candidiasis, reinforcing the idea that younger children are at heightened risk [37].

A contradictory results were reported in a study from Mexico, indicating that the prevalence of oral candidiasis was greater among older children, notably those aged 6 to 12 years [38].

The present investigation revealed that most instances were prevalent among younger age groups, with 36.7% of infections in children aged 1 to 6 months and 33.3% in those aged 7 to 12 months. This age-related pattern underscores the heightened vulnerability of newborns under one year, potentially due to immature immune systems, inadequate oral hygiene practices, or feeding strategies like breastfeeding or pacifier use. Infants in this age group may have little exposure to antifungal therapies or prophylactic measures, rendering them more susceptible to oral candidiasis.

In the context of gender, in contrast to the findings of the present study, a 2023 investigation indicated a greater prevalence of oral candidiasis in females, comprising 53.7% of cases [36]. These discrepancies may be ascribed to gender-related hormonal variances that could influence immunological responses, potentially leading to a somewhat diminished defense against fungal infections in male babies. Moreover, behavioral and environmental factors, like variations in feeding patterns, hygiene protocols, or the frequency of pacifier using, may further account for the heightened vulnerability shown in males.

An investigation in 2020 revealed that urban individuals exhibit superior oral health conditions compared to their rural counterparts [39]. The elevated incidence in rural pediatrics may be ascribed to multiple reasons, likely resulting from insufficient health services. Moreover, hygiene procedures in these regions are often inadequate, with the frequent use of unsterilized feeding bottles or pacifiers acting as a reservoir for *Candida* species. Maternal health is a contributing factor; inadequate prenatal care in rural regions may neglect maternal infections, heightening neonates' vulnerability to infections.

The type of feeding significantly influences the incidence of oral candidiasis, as yeast may be transmitted through nursing. A 2020 study indicated an increased prevalence of *Candida* spp. colonization on breast nipples, which can directly lead to mouth candidiasis in newborns [40]. The elevated occurrence may suggest suboptimal feeding habits, including faulty latch techniques or insufficient breast cleaning, fostering an environment favorable to *Candida* development. Maternal factors, including undiagnosed or untreated nipple thrush, can substantially facilitate the transmission of *Candida* to the newborn.

Regarding the education level of mother, a recent investigation in 2024 showed the absence of significant variations were noted for mothers' educational levels, which included middle school, high school, an associate degree (usually two years), a bachelor's degree (generally four years), and postgraduate studies [41].

Aligning with the current study findings regarding prevalence of *candida* spp., A study in Brazil indicated that *Candida albicans* was the predominant species, identified in 70.7% of cases, followed by *C. glabrata* at 6.1% and *C. krusei* at 3% [42]. A meta-analysis indicated that *C. albicans* is the predominant *Candida* species, including 50.25% of all isolates, followed by *C. parapsilosis* at 21.40% and *C. tropicalis* at 9.45%. Other species, including *C. glabrata*, *C. lusitaniae*, *C. krusei*, *C. famata*, and *C. pseudotropicalis*, were identified at far lower prevalences, varying from 0.25% to 2.73% [43].

The prevalence of *C. albicans* and *C. tropicalis* corresponds with their common occurrence in the normal oral microbiota of infants. These organisms, while integral to a healthy microbiome, can opportunistically proliferate and result in oral candidiasis. This fungal infection is especially prevalent in newborns and young children, likely

attributable to their underdeveloped immune systems, feeding habits, or environmental factors that promote fungal proliferation [44].

The germ tube is utilized for the identification of *Candida* species, distinguishing between *albicans* and non-*albicans* based on the presence or absence of germ tubes [45]. The production of germ tubes by *C. tropicalis* may be delayed, partial, or nonexistent; hence, an isolate that forms a germ tube after 6 hours may be *Candida tropicalis* [46].

Chrome agar serves as an effective instrument for the identification of *Candida spp.* in clinical microbiology laboratories, facilitating precise and swift detection of these organisms [47]. Utilizing CHROMagar enhances specificity in identification by allowing differentiation of species based on colony coloration. The colonies of *C. albicans* are light green, *C. glabrata* are mauve, *C. tropicalis* are dark metallic blue, and *C. krusei* are fuzzy pink [48].

The biological activity of TTO is attributed to its principal constituents, namely methyl eugenol and methyl cinnamate, which exhibit antimicrobial and antifungal properties. TTO has demonstrated effectiveness against MDR microbial strains, with MIC values of 0.5% and 2% (v/v) [49].

TTO exerts its antifungal effect by altering fungal cell membranes, resulting in a compromise of their structural integrity and functionality. This method is thought to involve the lipophilic characteristics of TTO, facilitating contact with and eventual breakdown of the fungal cell membrane. Although the precise mechanism of action is not well elucidated, research indicates TTO's effectiveness against various fungal strains, including *Candida albicans*, *Candida glabrata*, *Aspergillus*, and *Trichophyton species* [50].

TTO exhibit fungistatic properties at lower doses and fungicidal effects at elevated ones. The validation of robust activity against *C. albicans* cells via MIC and MFC assays encompasses in vitro regulation of biofilm growth [51].

The rising application against bacterial and fungal illnesses emphasizes its versatility and effectiveness. The results on this specific characteristic of TTO further reinforce its status as a natural and efficacious agent against fungal infections, even those induced by drug-resistant strains.

An investigation evaluated the antifungal and antiparasitic properties of capsaicin. The researchers aimed to determine the efficacy of crude extract and capsaicin in inhibiting biofilm formation, eliminating biofilms, and reducing hemolysin secretion by *Candida* species. The results demonstrated that the extracts inhibited biofilm formation and successfully eliminated the biofilm produced by *C. tropicalis*. Sub-inhibitory doses of the extracts markedly impeded the hemolytic activity of *C. glabrata* and *C. tropicalis* [52].

Capsaicin, in addition to its many medicinal and physiological applications—such as pain alleviation, cancer prevention, and cardioprotection—has recently garnered increased attention for its antibacterial, antifungal, and antiviral characteristics [53]. Research on the antifungal properties of capsaicin against *Candida albicans*, a leading cause of oral candidiasis, shows promise [54]. Capsaicin, at a MIC value of 25 µg/ml, had inhibitory actions against *C. albicans* [55]. Furthermore, *Candida* species have greater sensitivity to capsaicin than certain bacterial strains [56]. The minimum inhibitory concentration (MIC) range of capsaicin against 30 oral isolates of *Candida albicans* was determined to be between 12.5 and 50 µg/mL. Capsaicin demonstrated significant efficacy in eradicating the mature biofilm of *C. albicans* at a minimum inhibitory concentration [29].

The disparity in the efficacy of tea tree oil against both species of *Candida* can be attributed to the composition of tea tree oil, particularly its active component, terpinen-4-ol, which has previously demonstrated antifungal properties by disrupting fungal membranes and inhibiting ergosterol synthesis. Nonetheless, *C. tropicalis* may possess structural or metabolic adaptations that enable it to withstand such effects, resulting in a fungistatic outcome.

Capsaicin presumably exerts its fungicidal effect by compromising fungal membrane integrity through the breakdown of lipid components, resulting in cell lysis. The fungistatic impact against *C. albicans* was 21.06%, whereas it was 40% against *C. tropicalis*, indicating a highly lethal action against *C. albicans* and a balanced antifungal profile against *C. tropicalis*, resulting in both growth inhibition and lethality. The disparity in susceptibility between *C. albicans* and *C. tropicalis* to both drugs is significant. *C. albicans* exhibited heightened sensitivity to both treatments, tea tree oil and capsaicin, demonstrating significant fungicidal action. This may pertain to variations in cell wall composition or ergosterol levels, which make *C. albicans* more vulnerable to membrane-disruptive chemicals. Conversely, *C. tropicalis* exhibited enhanced fungistatic responses, signifying increased resistance to membrane disruption or adaptation to the environmental stress induced by these agents.

Conclusion

The study revealed that younger age groups were prevalent, with a higher prevalence of males and rural residents. The majority of moms were aged between 26 and 35 years, with a considerable number possessing secondary education. The majority of lactating mothers experienced breast mastitis. The incidence of infection caused by *Candida albicans* is markedly greater than that of *Candida tropicalis*, and the co-infection rate among patients is exceedingly high. Tea tree oil and capsaicin shown differing antifungal efficacy against the two yeast types. Tea

tree oil primarily exhibited fungicidal effects against *C. albicans*, while demonstrating a greater tendency for fungistatic action against *C. tropicalis*. Capsaicin exhibited predominant fungicidal properties, particularly against *C. albicans*, for which it shown greater potency. Tea tree oil demonstrated efficacy at lower doses against *C. albicans*; however, capsaicin exhibited consistent and more robust antifungal action, particularly against *C. albicans*, indicating its potential as a more effective antifungal agent at reduced concentrations. The data indicate the varying susceptibilities of the two *Candida* species to both drugs, with capsaicin demonstrating greater overall efficacy. Additional research is necessary to comprehend the prospective application of these chemicals as therapeutic agents for fungal infections.

Compliance with ethical standards

Disclosure of conflict of interest

The author(s) declare that they have no conflict of interest.

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