



Detection ability of *Candida species* isolated from oral kids with oral Candidiasis on production of phospholipase and proteinase in vitro

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Abstract

Background and objective: Over the past thirty years, there has been a growing amount of reports of cases of superficial and invasive mycoses have triggered by newly discovered species of *Candida*. Fungal infection is primarily caused by the generation of enzymes that hydrolyze. Protease and phospholipase secretion are two well-known virulence factors of *Candida* infections. Therefore, the aim of the study is to determine if *Candida* species isolated from oral children with oral Candidiasis can be detected by measuring their in vitro phospholipase and proteinase production. **Material and methods;** Specimen were taken from 150 kids who had oral candidiasis symptoms which sleeping in AL-Batool Teaching Hospital in Baqubah City. Kids age groups from months to > 2 years provided samples. Macro and microscopical methods were used for diagnosis oral Candidiasis. Chromogenic Agar *Candida* (CAC) was used to selectively develop the isolates with features characteristic of *Candida spp*. The synthesis of proteinase was confirmed in a medium comprising bovine serum albumin, whereas the phospholipase reaction was carried out in an egg yolk media. **Results;** Results of conducted study showed 100 (66.66%) of 150 kids were positive with oral candidiasis than kids without oral candidiasis 50 (33.34%) . Most infected kids were males (72%) with age groups 1-2 years (54%) with . *C. albicans* scored highest infection in oral kids (69%) followed by *C. glabrata* (16%), *C. krusei* (9%), and *C. tropicalis* (6%) . *C. albicans* scored highest percentage for phospholipase and proteinase production (92.75% and 85.50%), while *C. krusei* scored least percentage (55.55% and 22.22%) than another *Candida* species. Finally, these difference among gender, age groups, *Candida* species, and production of phospholipase and proteinase were significant ($p < 0.05$). **Conclusions;** There is a correlation between children's oral candidiasis incidence and compromised immune function. Male children in the 1-2 year age range were the most infected. Because *Candida albicans* is the most harmful kind of yeast, it had the highest frequency in oral children when compared to other kinds of *candida*. When it came to the generation of phospholipase and proteinase, *Candida albicans* received the greatest percentage, next to another species of *Candida*.

Keywords; oral Candidiasis, *C. albicans*, Oral infection, phospholipase and proteinase



قدرة الكشف عن أنواع الكانديدا المعزولة من الأطفال المصابين بداء المبيضات الفموي على إنتاج الفسفوليبيز والبروتيناز في المختبر

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المخلص

الخلفية والهدف: على مدى الثلاثين عامًا الماضية، كانت هناك كمية متزايدة من التقارير عن حالات الإصابة بالفطريات السطحية والغزوية التي تسببها أنواع الكانديدا المكتشفة حديثًا. تحدث العدوى الفطرية في المقام الأول بسبب تكوين الإنزيمات التي تتحلل. يعد إفراز البروتيناز والفسفوليبيز عاملين معروفين لضراوة عدوى الكانديدا. لذلك، فإن الهدف من الدراسة هو تحديد ما إذا كان من الممكن الكشف عن أنواع الكانديدا المعزولة من الأطفال المصابين بداء المبيضات الفموي عن طريق قياس إنتاج الفسفوليبيز والبروتيناز في المختبر. المواد والطرق: تم أخذ العينات من 150 طفلاً مصابين بأعراض داء المبيضات الفموي والذين يترقدون في مستشفى البتول التعليمي في مدينة بعقوبة. تم أخذ العينات من فئات عمرية من أشهر إلى < 2 سنة. تم استخدام الطرق المجهرية والعيانية لتشخيص داء المبيضات الفموي. تم استخدام أجار الكانديدا الكروموجيني (CAC) لتطوير العزلات بشكل انتقائي ذات السمات المميزة لأنواع الكانديدا. تم تأكيد تخليق البروتيناز في وسط يحتوي على ألومين مصل البقر، في حين تم إجراء تفاعل الفسفوليبيز في وسط صفار البيض. النتائج: أظهرت نتائج الدراسة التي أجريت أن 100 (66.66%) من 150 طفلاً كانوا إيجابيين مع داء المبيضات الفموي مقارنة بالأطفال غير المصابين بداء المبيضات الفموي 50 (33.34%). كان معظم الأطفال المصابين من الذكور (72%) في الفئة العمرية 1-2 سنة (54%). سجلت الكانديدا البيضاء أعلى نسبة إصابة في الأطفال الفمويين (69%) تليها الكانديدا غلابراتا (16%) و الكانديدا كروساي (9%) و الكانديدا تروبيكالييس (6%). سجلت الكانديدا البيضاء أعلى نسبة لإنتاج الفسفوليبيز والبروتيناز (92.75% و 85.50%)، بينما سجلت الكانديدا كروساي أقل نسبة (55.55% و 22.22%) من أنواع الكانديدا الأخرى. أخيرًا، كانت هذه الاختلافات بين الجنس والفئات العمرية وأنواع الكانديدا وإنتاج الفسفوليبيز والبروتيناز مهمة ($p < 0.05$). الاستنتاجات: هناك علاقة بين حدوث داء المبيضات الفموي لدى الأطفال وضعف وظيفة المناعة. كان الأطفال الذكور في الفئة العمرية 1-2 سنة هم الأكثر إصابة. ولأن الكانديدا البيضاء هي أكثر أنواع الخميرة ضررًا، فقد كان لها أعلى معدل انتشار في الأطفال الفمويين مقارنة بأنواع الكانديدا الأخرى. عندما يتعلق الأمر بتوليد الفسفوليبيز والبروتيناز، حصلت المبيضة البيضاء على النسبة الأكبر، بعد نوع آخر من المبيضات.

الكلمات المفتاحية: داء المبيضات الفموي، المبيضة البيضاء، العدوى الفموية، الفسفوليبيز والبروتيناز

Introduction

Oral candidiasis, a condition caused by *Candida albicans* in the dental cavity, was first reported by pediatrician Francois Veilleux in 1838. Gonçalo et al. (2024) state that immunological decreasing, the more general cause of the condition, can be attributed to a number of factors, including immunocompromising diseases like HIV/AIDS, long-term systemic steroid and prescription antibiotics, and extremes in age (infants and elderly).

Oral candidiasis may impact both immunocompetent and immunocompromised individuals, albeit it is more common in those who are immunocompromised. Nearly 90% of HIV-positive individuals experience oral candidiasis at some point throughout their disease (Taverne-Ghadwal et al., 2022). Oral candidiasis



can affect both sexes equally. Rarely does it occur within the first week of life; babies and neonates are often affected. It becomes more common during the fourth week of existence and less common in newborns older than six months of age, most likely because of the development of immunity from the host. In these people, immunosuppression manifests as hepatosplenomegaly, breakouts, diarrhea, and ongoing infections (Lass-Flörl et al., 2024).

Early childhood caries (ECC) patients have oral *Candida albicans*, which has also been shown to have cariogenic characteristics in animal studies of the illness. The only species of fungus known to attain an extensive biomass in the cavity of the mouth, *Candida*, has been shown during the past 20 years to flourish in reduced oral pH and ECC (Diaz and Dongari-Bagtzoglou, 2021). Numerous investigations conducted at different species levels have demonstrated a favorable correlation between the severity and frequency of ECC and the amount and quantity of *Candida albicans*, indicating that *C. albicans* may also be a significant etiological risk factor for ECC (Menon et al., 2022).

Clinical examinations, information collection about health conditions, and associated risk evaluations are often used to diagnose oral candidiasis. In certain cases, a biopsy is recommended in addition to empirical treatment. Cultures are frequently performed in cases when antifungal medication is ineffective (Hellier and Wrynn, 2023; Gómez-Gaviria et al., 2024).

Candida is often seen in the oral microbiota of immunocompetent individuals. In the oral cavity, 30–60% of adults and 45–65% of infants contain *candida* spp., according to Hellstein and Marek (2019). Oral candidiasis is most commonly caused by the species *Candida albicans*, that has been identified isolated from more than 80% of lesions. Macias-Paz et al. (2023) describe *Candida albicans* as a dysmorphic yeast that, based on the environment, can appear as either a yeast or a hyphal.

Additional genera that are believed to be connected are *Candida glabrata*, *Candida tropicalis*, *Candida kruesi*, *Candida guilliermondii*, *Candida lusitanae*, *Candida parapsilosis*, *Candida pseudotropicalis*, and *Candida stellatoidea*; nevertheless, they are far less common (Hellstein and Marek, 2019). According to research, non-*albicans* *Candida* species infiltrate adults 80 years of age and older more frequently than they do young people (Ghojoghi et al., 2024).

During pregnancy and adolescence, both the mother and the child seem to be particularly susceptible to fungal infections since fungi are opportunistic microbes. According to Azevedo et al. (2023), vaginal *Candida* spp. colonization affects 30 to 40 percent of pregnant women. This colonization may be caused by higher levels of estrogen that promote yeast adhesion and penetration of the vaginal mucous membrane. This tendency seems to continue after delivery, as demonstrated by the finding of Khadija et al. (2019) that



postpartum females have enhanced virulence traits and are more susceptible to oral *Candida* spp. colonization. It is hypothesized that *Candida* spp. colonization occurs in the early hours after delivery in relation to the child. Moreover, a limited body of data corroborates the hypothesis that fungal isolates can be transferred from mother to child (Alkhars et al., 2024). Concurrently, infants who use diapers often get fungal infections, and oral candidiasis—which is usually diagnosed in neonates as thrush in the mouth—is a common ailment in children, particularly up to the sixth month of life (Ota et al., 2024).

Important stages in the development of superficial candidiasis include fungal adherence, settlement, and eventual penetration into the tissue. The synthesis of hydrolytic enzymes is a crucial process in each of these stages (Amini and Mohammadi, 2023). Because proteins and phospholipids are the main chemical components of the membrane of the host cell and should be demolished to be able to provide minerals for fungus development and expansion along with facilitate the fungal's distribution within the body of the host, two key categories of enzymes—proteases and phospholipases—have prominent and central roles in this regard (Aparna et al., 2023).

In Iraq/ Diyala province, researches were losing about in vitro production of phospholipase and proteinase by *Candida species* that isolated from oral kids with oral Candidiasis. Thus, the goal of the research is detection ability of *Candida* species isolated from oral kids with oral Candidiasis on production of phospholipase and proteinase in vitro

Materials and Methods

Data collection Sample Collection and Candida Isolation

Specimen were taken from 150 kids who had oral candidiasis symptoms which sleeping in AL-Batool Teaching Hospital in Baqubah City. Kids age groups from months to > 2 years provided samples. Swabs made of cotton were used to gather oral specimens, which were subsequently streaked on Sabouraud's dextrose agar (SDA) plates supplemented with 0.005 g/L chloramphenicol to inhibit the development of bacteria. For 48 hours, incubation was conducted at 30°C. Next, the isolates were inspected for size, shape, color, odor, and texture using both macro and microscopical methods (Khaleefa et al. 2020). The genus *Candida* is characterized through its oval or oblong form and bipolar budding, an asexual method of development. The newly formed *Candida* cells are cream-white in color on culture medium. Their cells have a diameter of three to six microns, and they occasionally form pseudohyphae (Samy and Aljaza, 2023; Mohammed et al., 2019).

Species Identification by Biochemical Tests



Chromogenic Agar Candida (CAC) was used to selectively develop the isolates with features characteristic of Candida spp. (Al-Ani et al. 2023). After growing for 24 hours on SDA, a piece of the clean colony is plotted on CAC. At 37°C, incubating lasts for 24 to 48 hours. The color of the communities helps identify the various species. As per Al-Ani et al. (2023) Candida albicans is green, Candida glabrata is pink to purple, Candida krusei is dark pink, and Candida tropicalis is blue.

Phospholipases production

The isolates' ability to generate phospholipases was examined using Price et al.'s (1982) egg yolk agar plate technique. Sabouraud dextrose agar with 2% egg yolk, 0.005M calcium chloride, and 1M sodium chloride served as the test media. There were three inoculations of each strain. Seven days after the the injection, the Petri plates were kept warm at 37°C, and the diameters of the colonies, precipitation area, and colony measurement was performed. The phospholipase activity zone (Pz) was measured and computed using the procedure outlined by Price et al. (1982). To find the average Pz, the Pz of three samples of each strain were measured. Pz between 0.9 and 1 (+), very low Pz group; 0.89 – 0.80 (+ +), low Pz group; 0.79 – 0.70 (+ + +) high Pz group; and 0.69 (+ + + +), very high Pz group were the four groups into which the Pz coefficients of the studied Candida strains were divided.

Protease production

Protease generation was measured in accordance with Aoki et al. (1990). Plates with agar containing bovine albumin serum (BSA) made up the test condition. 60 mL of a solution of 0.04 g of MgSO₄.7H₂O, 0.5 g of K₂HPO₄, 1 g of NaCl, 0.2 g of yeast extract, 4 g of glucose, and 0.5 g of BSA (milk serum). The pH was adjusted to 4.0 and Fraction V (Sigma Chem Co., St. Louis, Mo., USA) was produced. After filtering the solution to sterilize it, 140 milliliters of melted agar were added. For seven days, plates containing this medium were incubated at 37°C. In accordance with the procedure outlined by Price et al. (1982), proteinase activity was quantified and computed as the ratio of colony diameter to colony plus inhibition zone. To get the average Pz values, the investigation was conducted three times for every strain. As previously noted, the Pz coefficients were divided into 4 groups.

Statistical analysis

SPSS (Statistical Package for Social Science) version 22 was utilized to analyze current data. Chi-square test was depended to measure significant variation among percentage at levels of significant $P \leq 0.05$.

Results

1. Distribution of kids depend on fungal infection

Outcomes of conducted research showed (66.66%) of kids have oral candidiasis than kids without oral candidiasis (33.34%) with significant different ($p<0.05$) (table 1).

Table 1; distribution of kids based on oral Candidiasis

Results	N	%	P value
Positive	100	66.66%	P<0.01**
Negative	50	33.34%	
Total	150	100%	

2. Distribution infected kids according to gender

Outcomes of our investigation showed the most infected kids were males (72%) than females (28%) with high significant differences ($p<0.05$) (table 2).

Table 2; distribution of kids with oral Candidiasis based on gender

Gender	N	%	'P value
Males	72	%72	p<0.01**
Females	28	%28	
Total	100	%100	

3. Distribution infected kids according to age groups

Results of current study mentioned the most infected kids were lie at age groups 1-2 years (54%) and least of them at >2 years (16%) with significant different ($p<0.05$) among age groups of infected kids (table 3).

Table 3; distribution of kids with oral Candidiasis depend on age groups

Age groups	N	%	P value
<1 year	30	30%	p<0.001***
1-2 years	54	54%	
>2 years	16	16%	
Total	100	100%	

4. Distribution infected kids according to *Candida species*

Results of our study showed the most kids were positive for *C. albicans* (69%) and least of them were positive for *C. glabrata* (16%), *C. krusei* (9%), and *C. tropicalis* (6%) with significant differences ($p<0.05$) among candida species (table 4).



Table 4; distribution of kids with oral Candidiasis based on candida species

Candida types	N	%	P value
<i>C. albicans</i>	69	%69	P<0.001***
<i>C. glabrata</i>	16	%16	
<i>C. krusei</i>	9	9%	
<i>C. tropicalis</i>	6	%6	
Total	100	100%	

5. Distribution Candida species according to phospholipase production

Results of current study showed the *C. albicans* scored highest production of phospholipase (92.75%), followed by *C. glabrata* (68.75%), *C. tropicalis* (66.66%), and then *C. krusei* (55.55%) with significant differences ($p<0.05$) among positivity of phospholipase production by Candida species (table 5).

Table 5; distribution of kids with oral Candidiasis based on phospholipase production

Candida types	NT	NP	%	P value
<i>C. albicans</i>	69	64	92.75%	P<0.001***
<i>C. glabrata</i>	16	11	68.75%	P<0.01**
<i>C. krusei</i>	9	5	55.55%	P>0.05
<i>C. tropicalis</i>	6	4	66.66%	P<0.05*
Total	100	84	84%	P<0.001***
P value	P<0.01**			

6. Distribution Candida species according to proteinase production

Results of our study showed the *C. albicans* scored highest production of phospholipase (85.50%), followed by *C. tropicalis* (50%), *C. glabrata* (31.25%), and then *C. krusei* (22.22%) with significant differences ($p<0.05$) among positivity of proteinase production by Candida species (table 6).

Table 6; distribution of kids with oral Candidiasis based on proteinase production

Candida types	NT	NP	%	P value
<i>C. albicans</i>	69	59	85.50%	P<0.001***
<i>C. glabrata</i>	16	5	31.25%	
<i>C. krusei</i>	9	2	22.22%	P<0.001***

<i>C. tropicalis</i>	6	3	50%	p>0.05
Total	100	71	71%	P<0.001***
P value	P<0.01**			

Discussion

In addition to visual examination of removable white plaque or erythema tissue in the cavity, microscopic examination of a sample of the oral mucosa revealing diagnostic findings is frequently used to make a determination of oral candidiasis. Additionally, oral candidiasis can be diagnosed using microbial and medical tests (Qadir et al., 2023). Using a magnifying glass, oral candidiasis was discovered in 72 (65%) of the 110 oral swabs that were used in the prior study. A previous study indicated that the overall incidence of Candida strains that are part of the oral microbes was 35%, despite the fact that their geographical location varies (Kim and Ahn, 2021). These findings above had been 35% lower than those of the current research, wh ich found that 100 oral swabs (66.66%) included Candida.

However, in a previous study, it was predominantly observed in adults 60 years of ages and older, both male and female, and there was no discernible correlation between the incidence of oral candidiasis and the presence of a dry mouth, alcohol or tobacco use, dentures wear, or hormonal problems (Qiu et al., 2023). These characteristics are often indicators of risk for oral candidiasis development.

Keyvanfar et al. (2024) state that while oral candidiasis is rare in the first week following delivery, it can occur in infants and young children. It is more common around the fourth week of existence and less common in newborns older than six months of age, most likely because to the development of host immunity. Such results are not consistent with the present data, which show a considerable prevalence of oral candidiasis in neonates older than six months. There is a correlation between the higher incidence of oral candidiasis in infants and babies and their weakened immune systems (Keyvanfar et al., 2024).

Similar to the present data, Cook et al.'s (2023) findings showed that oral candidiasis was more prevalent in women (55%) than in males (45%). According to Chuey Chuan et al. (2023), women (61%) had oral candidiasis at a higher rate than males (39%). These results did not align with current studies, which indicated that males had a higher prevalence of Candida albican (72%) compared to females (18%). Study discrepancies are influenced by the number of samples and the host's immunological status.

The investigators found a substantial association between breastfeeding with bottles and a higher risk of respiratory diseases as well as GIT infections, despite the fact that nursing women's breastbones had a considerably higher volume and



more diversity of Candidal species. They also found that only a small percentage of breastfed babies liked this type of yeast expanding into within their mouths, which lends credence to the idea that protective factors like breast milk provide protection against the *Candida* genus settling in the oral cavity of breastfed only babies (Bao et al., 2024).

Previous studies have demonstrated that the child may inherit strains of *Candida albicans* and *Candida parapsilosis* via their mother. In addition, the researchers revealed the first-ever vertical transmission of a *C. albicans* strain resistant to fungicides. This new pathogen induced oral candidiasis (Azevedo et al., 2023).

Recent studies show that oral *Candida albicans* uptake in early infants is significantly influenced by mother transmission. By adding screening for mom fungus oral bearers and implementing oral health education programs during the prenatal period, it would be able to prevent fungal transmission in the early stages of infancy (Alkhars et al., 2024).

Up to 40% of individuals in good health have naturally occurring, or entering, *Candida* in their dental cavities. Immunosuppressed individuals have been found to have a greater incidence of *Candida* species in their dental cavities when compared to healthy individuals. discovered that up to 80% of patients' salivary glands had *Candida* spp. and that oral yeasts were detected in the saliva of 55% of those admitted to hospitals with advanced malignancies (Rajendra Santosh et al., 2021).

Cook et al., (2023) mentioned the *C. albicans* showed large prevalence in oral neonates (35%), followed by *C. glabrata* (5%), and *C. tropicalis* (2%). These outcomes were nearly to present findings. Additionally, Cook et al., (2023) mentioned the most *Candida* strains were resistance to fluconazole. Another study showed *C. albicans* showed high prevalence in oral infants (67%), followed by *C. tropicalis* (30%) and *Candida parapsilbsis* (15%) and *Candida krusei* (17%) (Neamah, 2024). Another study In 52 children, *Candida* was isolated, and the identified species were *C. albicans* (87.8%), *C. glabrata* (11.5%), *C. krusei* (5.8%) and *C. parapsilosis* (1.9%) (Pinto-Almazán et al., 2022). Malnutrition; age fluctuations (young children and elderly), metabolic diseases, immunocompromising illnesses concurrent infections, chemotherapy, transplantation of organs, long-term treatment with steroids, therapy with antibiotics, and hypofunction of the salivary glands are indicators of risk for the pathological the colonization of *Candida* species (Alves et al., 2023).

Proteinase and phospholipase are produced by 100% and 87% of candida strains from oral kids, respectively, according to Ilkhanizadeh-Qomi et al. (2020). These results are in line with our own research, which revealed that 71% and 84% of candida strains from oral kids generate these exoenzymes. The investigators found that *C. albicans* strains are more effective secretors of



aspartyl proteinase enzymes than non-albicans strains due to the considerable variations in aspartyl proteinase enzyme synthesis between the two strains. It has been shown that *C. albicans*'s phospholipase and proteinase activities are crucial factors for virulence, and that other *Candida* species may be less virulent as a result of their absence or lower activity (Ilkhanizadeh-Qomi et al., 2020). Contrary to the majority of research that demonstrate *C. albicans*, the authors observe greater levels of proteinase and phospholipase activity within *C. tropicalis*. The results above conflict with a recent study that found that *Candida albicans* had stronger proteinase and phospholipase activity than *Candida tropicalis*. Extracellular enzyme release is undoubtedly a possible source of virulence, although the host's constitution greatly influences both the amount of these enzymes' production and their effects (Aparna et al., 2023). According to Chaula et al. (2024), the enzyme assay revealed that *Candida albicans* produces the greatest quantity of extracellular enzymes for hydrolysis, which is why it is considered the most dangerous species when compared to non-albicans.

Conclusions

There is a correlation between children's oral candidiasis incidence and compromised immune function. Male children in the 1-2 year age range were the most infected. Because *Candida albicans* is the most harmful kind of yeast, it had the highest frequency in oral children when compared to other kinds of *Candida*. When it came to the generation of phospholipase and proteinase, *Candida albicans* received the greatest percentage, next to another species of *Candida*.

Conflict of interest:

The authors declare that no conflict of interests.

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References

- Al-Ani, D. K. J., Musa, F. H., & Buniya, H. K. (2023). Isolation and identification of *Candida albicans* from children patient with candidiasis from Ramadi city, Iraq. *HIV Nursing*, 23(2), 441-449.
- Alkhars, N., Al Jallad, N., Wu, T. T., & Xiao, J. (2024). Multilocus sequence typing of *Candida albicans* oral isolates reveals high genetic relatedness of mother-child dyads in early life. *Plos one*, 19(1), e0290938.
- Alves, A. M. C. V., Lopes, B. O., Leite, A. C. R. D. M., Cruz, G. S., Brito, É. H. S. D., Lima, L. F. D., ... & Rodrigues, C. F. (2023). Characterization



of Oral Candida spp. Biofilms in Children and Adults Carriers from Eastern Europe and South America. *Antibiotics*, 12(5), 797.

- Amini, N., & Mohammadi, R. (2023). Phospholipase activity of Candida species isolated from diabetic patients. *Advanced Biomedical Research*, 12(1), 19.
- Aoki, S., Ito-Kuwa, S., Nakamura, Y., & Masuhara, T. (1990). Comparative pathogenicity of a wild-type strain and respiratory mutants of Candida albicans in mice. *Zentralblatt für Bakteriologie*, 273(3), 332-343.
- Aparna, T., Raphy, H. M., & KS HK, I. S. (2023). Phospholipase, proteinase, esterase and haemolytic activity of Candida species isolated from oral cavity and its antifungal susceptibility pattern. *International Journal of Research in Medical Sciences*, 11(7), 2476.
- Aparna, T., Raphy, H. M., & KS HK, I. S. (2023). Phospholipase, proteinase, esterase and haemolytic activity of Candida species isolated from oral cavity and its antifungal susceptibility pattern. *International Journal of Research in Medical Sciences*, 11(7), 2476.
- Azevedo, M. J., Araujo, R., Campos, J., Campos, C., Ferreira, A. F., Falcão-Pires, I., ... & Sampaio-Maia, B. (2023). Vertical transmission and antifungal susceptibility profile of yeast isolates from the oral cavity, gut, and breastmilk of mother–child pairs in early life. *International journal of molecular sciences*, 24(2), 1449.
- Bao, L., Du, K., Gao, J., Jiang, R., Li, B., & Liu, T. (2024). An analysis of the risk factors for invasive fungal infections in preterm infants and a discussion of prevention strategies. *Technology and Health Care*, 32(1), 361-367.
- Chaula, Y., Swilla, J. D., Lyatuu, F., & Namkinga, L. A. (2024). Assessment of Proteinase and Phospholipases Enzymes Isolated from Pathogenic Candida species from Women Attending Antenatal Clinic at Mbeya Zonal Referral Hospital, Southern Highland Regions of Tanzania. *Journal of Biology and Life Science*, 15(1), 93-104.
- Chuey Chuan, T. A. N., Daniel, L. I. M., Nurin Qistina, M. O. H. D., Noralia Auni ELIAS, A. S., & AZLI, Y. C. G. (2023). Clinicopathological correlation of oral candidiasis–Our experience in a tertiary centre over two decades. *Malays J Pathol*, 45(2), 237-246.
- Cook, A., Ferreras-Antolin, L., Adhisivam, B., Ballot, D., Berkley, J. A., Bernaschi, P., ... & Sharland, M. (2023). Neonatal invasive candidiasis in low- and middle-income countries: data from the NeoOBS study. *Medical Mycology*, 61(3), myad010.
- Diaz, P. I., & Dongari-Bagtzoglou, A. (2021). Critically appraising the significance of the oral mycobiome. *Journal of Dental Research*, 100(2), 133-140.
- Ghojoghi, A., Khodavaissy, S., Mahmoudabadi, A. Z., Nazar, E., & Fatahinia, M. (2024). Exploring the diversity of uncommon oral yeast species



and associated risk factors among substance abusers in southwestern Iran. *Scientific Reports*, 14(1), 1906.

- Gómez-Gaviria, M., García-Carnero, L. C., Baruch-Martínez, D. A., & Mora-Montes, H. M. (2024). The Emerging Pathogen *Candida metapsilosis*: Biological Aspects, Virulence Factors, Diagnosis, and Treatment. *Infection and Drug Resistance*, 171-185.
- Gonçalo, R. I. C., De Souto Medeiros, M. R., Carlan, L. M., De Farias Morais, H. G., Gonzaga, A. K. G., De Oliveira, P. T., & Da Silveira, É. J. D. (2024). Oral Candidiasis In Head And Neck Cancer Patients: A Systematic Review. *Oral Surgery, Oral Medicine, Oral Pathology And Oral Radiology*, 137(6), E316.
- Hellier, S. D., & Wrynn, A. F. (2023). Beyond fluconazole: A review of vulvovaginal candidiasis diagnosis and treatment. *The Nurse Practitioner*, 48(9), 33-39.
- Hellstein, J. W., & Marek, C. L. (2019). Candidiasis: red and white manifestations in the oral cavity. *Head and neck Pathology*, 13, 25-32.
- Ilkhanizadeh-Qomi, M., Nejatbakhsh, S., Jahanshiri, Z., & Razzaghi-Abyaneh, M. (2020). Aspartyl proteinase and phospholipase activities of *Candida albicans* isolated from oropharyngeal candidiasis in head and neck cancer patients. *Jundishapur Journal of Microbiology*, 13(9).
- Keyvanfar, A., Najafiarab, H., Talebian, N., Tafti, M. F., Adeli, G., Ghasemi, Z., & Tehrani, S. (2024). Drug-resistant oral candidiasis in patients with HIV infection: a systematic review and meta-analysis. *BMC Infectious Diseases*, 24(1), 546.
- Khadija, B., Abbasi, A., Khan, S., Nadeem, M., Badshah, L., & Faryal, R. (2019). Isolation of pathogenic *Candida* species from oral cavity of postpartum females, and its association with obstetric and dental problems. *Microbial pathogenesis*, 131, 40-46.
- Khaleefa, R. H., Ahmed, L. T., & Hameed, D. S. (2020). Oral and Dermal Candidiasis Among Infants in Al-Battol Teaching Hospital. *Diyala Journal of Medicine*, 19(2), 147-156.
- Kim, J. H., & Ahn, J. M. (2021). Clinical characteristics of patients with oral candidiasis. *Journal of Oral Medicine and Pain*, 46(2), 33-40.
- Lass-Flörl, C., Kanj, S. S., Govender, N. P., Thompson III, G. R., Ostrosky-Zeichner, L., & Govrins, M. A. (2024). Invasive candidiasis. *Nature Reviews Disease Primers*, 10(1), 20.
- Macias-Paz, I. U., Pérez-Hernández, S., Tavera-Tapia, A., Luna-Arias, J. P., Guerra-Cárdenas, J. E., & Reyna-Beltrán, E. (2023). *Candida albicans* the main opportunistic pathogenic fungus in humans. *Revista Argentina de Microbiología*, 55(2), 189-198.



- Menon, L. U., Scoffield, J. A., Jackson, J. G., & Zhang, P. (2022). *Candida albicans* and early childhood caries. *Frontiers in Dental Medicine*, 3, 849274.
- Neamah, R. A. (2024). Isolation and identification of some types of yeast *Candida* spp and study of their sensitivity to some antifungals. *Iraqi Journal of Humanitarian, Social and Scientific Research*, 4(12A-1).
- Ota, Y., Ito, T., Sashida, M., Hori, E., Kimijima, M., Narisawa, N., ... & Shimizu, T. (2024). Association between *Candida albicans* and childhood dental caries in Japanese children. *Pediatric Dental Journal*, 34(1), 1-7.
- Pinto-Almazán, R., Frías-De-León, M. G., Fuentes-Venado, C. E., Arenas, R., González-Gutiérrez, L., Chávez-Gutiérrez, E., ... & Martínez-Herrera, E. (2022). Frequency of *Candida* spp. in the oral cavity of asymptomatic preschool Mexican children and its association with nutritional status. *Children*, 9(10), 1510.
- Price, M. F., Wilkinson, I. D., & Gentry, L. O. (1982). Plate method for detection of phospholipase activity in *Candida albicans*. *Sabouraudia: Journal of Medical and Veterinary Mycology*, 20(1), 7-14.
- Qadir, M. I., Bashir, H., & Ahmad, M. H. (2023). Human Oropharyngeal Candidiasis: From Etiology to Current Treatment. *Critical Reviews™ in Immunology*, 43(3).
- Qiu, J., Roza, M. P., Colli, K. G., Dalben, Y. R., Maifrede, S. B., Valiatti, T. B., ... & Gonçalves, S. S. (2023). *Candida*-associated denture stomatitis: clinical, epidemiological, and microbiological features. *Brazilian Journal of Microbiology*, 54(2), 841-848.
- Rajendra Santosh, A. B., Muddana, K., & Bakki, S. R. (2021). Fungal infections of oral cavity: diagnosis, management, and association with COVID-19. *SN comprehensive clinical medicine*, 3(6), 1373-1384.
- Samy, R., & Aljaza, D. (2023). Morphology and molecular identification of *Candida* Species isolated from oral pediatric. , 10(1).
- Sharma, M., & Chakrabarti, A. (2023). Candidiasis and other emerging yeasts. *Current Fungal Infection Reports*, 17(1), 15-24.
- Taverne-Ghadwal, L., Kuhns, M., Buhl, T., Schulze, M. H., Mbaitolum, W. J., Kersch, L., ... & Groß, U. (2022). Epidemiology and prevalence of oral candidiasis in HIV patients from chad in the post-HAART era. *Frontiers in Microbiology*, 13, 844069.