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Isolation and identification of candida species from oral of diabetic patients

Abstract

Diabetes increases the risk of fungal infections in the mouth and in other parts of the body. Candidiasis may occur if there is a decrease in the flow of saliva, which allows the fungi to grow. The present study aimed to determine the prevalence of Candida spp and identify the potential risk factors among diabetic and non-diabetic groups. This cross-sectional study was conducted from November 2021 to May 2022. During this period, 130 oral swabs were collected, the following tests were done: direct microscopic examination, gram stain and cultured on sabaroud dextrose agar with chloramphenicol, complimentary tests such as germ tube test, sugar assimilation test, growing on chromogenic media, were carried out to differentiate between the Candida species. Out of 130 oral swab cultured, there were 102 (78.4%) yielded Candida species. 80 (89%) were diabetics, the results showed strong statistical relationship between diabetics and Candida infections p=0.001. The frequencies of isolated Candida species including *C. albicans* were 41(40.2%), while *C. Glabrata* 23 (22.5%) *C. tropicalis* 26 (25.5%), and *C. krusei* 12 (11.8 %) The results of this study revealed a higher prevalence of *Candida* infections in patients with diabetes, so the study recommends continuous follow-up and the necessary tests in response to any symptoms, and recommends health education and attention to clean food and drink. Furthermore, the study underscores the necessity for further research on the correlation between Candida infections and diabetic complications.

Keywords: Candida species, diabètes, Candidiasis, Ibb, Yemen

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Enas F Al-Awadhi,[1] Hussein Al-Towiti,[2] Albaraa Al-Qadri,[2] Sadam Alhashdi,[2] Najmaldeen Alkhawlani,[2] Abdullaziz Alhamoodi[2]

[1]Department of Biology, Faculty of Science, Sana'a University, Yemen

[2]Laboratory Department, Faculty of Medical Sciences, National University, Yemen

Correspondence: Enas F Al-Awadhi, Microbiology Branch, Department of Biology, Faculty of Science, Sana'a University, Sana'a, Yemen, Tel 00967777626667, Email enasfadh@yahoo.com

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Introduction

Candida is a polymorphic fungus, oval, Gram positive, budding yeast cell that produces pseudo hyphae both in culture and in tissues and exudates[1] *Candida albicans* is considered the most common fungal infection of the human oral cavity. Nonetheless, candidiasis caused by non-albicans Candida species, such as *C. tropicalis, C. parapsilosis, C. krusei, C. glabrata, and C. dubliniensis*, are also becoming common among certain groups of patients.[2,3]

Diabetes mellitus is a metabolic disorder that arises due to a reduction, either partial or total, in the production of insulin by the pancreas. This reduction can be caused either by an autoimmune response that destroys the beta cells responsible for insulin synthesis, or by inadequate insulin production. As a consequence, the level of sugar in the blood increases, leading to a hyperglycemia. This condition can be particularly harmful to individuals with diabetes, especially those who have had the disease for a long time, as it can cause various changes in the body.[4]

Diabetes mellitus is a known risk factor for candidiasis,[5] a fungal infection caused by Candida species. The increased blood sugar levels in diabetic patients provide a favorable environment for the growth of yeast due to the higher number of receptors available to Candida.[6] Additionally; hyperglycemia can lead to a decrease in the defensive capability of polymophonucear neutrophils and T lymphocytes, further promoting the growth of Candida.[7]

Fungal pathogens and infections are an increasing global public health concern. People most at risk are those with underlying health problems or a weakened immune system, such as chronic lung disease, cancer, and diabetes mellitus.[8]

The purpose of this study was to determine the prevalence of Candida spp infections in diabetic and non-diabetic groups, as well as identify potential risk factors for such infections.

Materials and methods

This particular cross-sectional study was conducted between November 2021 and May 2022. During this time frame, 130 oral swabs were collected from each patient who visited certain hospitals and clinics located in Ibb city, Yemen.

The sample size for this study consisted of 130 individuals, including both diabetic and non-diabetic patients who were all over the age of 40. The diabetic patients had a known history of diabetes mellitus lasting approximately five years and presented with a history of oral ulcers lasting for at least 30 days.

In this study, several tests were conducted to identify the presence of Candida spp and differentiate between species. These tests included direct microscopic examination, Gram stain, and culture on Sabouraud dextrose agar with chloramphenicol. Complimentary tests such as the germ tube test, sugar assimilation test, and growing on chromogenic media were also performed to help distinguish between different Candida species. In addition to these tests, patient information was collected using a questionnaire. This likely included demographic information such as age, gender, and medical history, as well as information on risk factors for Candida spp infection such as smoking, alcohol consumption, and antibiotic use. Collecting this information can help researchers identify potential risk factors for Candida spp infection and assess their relevance to the studied population.

The statistical analysis for this study was conducted using the SPSS Chi-square test. A P value of less than 0.05 was considered statistically significant. Additionally, odds ratios and 95% confidence intervals (CI) were calculated to assess the strength of the association between the studied variables and the prevalence of Candida spp infections.

Results

The results of the present study indicate that the prevalence of Candida spp infection was significantly higher in diabetic patients compared to non-diabetic patients. Specifically, out of the 90 diabetic patients included in the study, 80 (89%) were found to have Candida spp infection. In contrast, only 22 (55%) of the non-diabetic patients were found to have Candida spp infection. The difference between these two groups was statistically significant with a P value of less than 0.001. The odds ratio calculated for this comparison was 2.196, suggesting that diabetic patients are 2.196 times more likely to have Candida spp infection compared to non-diabetic patients (Table 1).

Figure 1 shows the percentage of different species of Candida. Out of the 102 diabetic patients and non-diabetics, the percent of the *C. albicans* was 41 (40.2%) and *C. glabrata* was 23(22.5%) while *C. tropicalis* and *C. krusi* were 26 (25.5%) and 12(11.8 %) respectively.

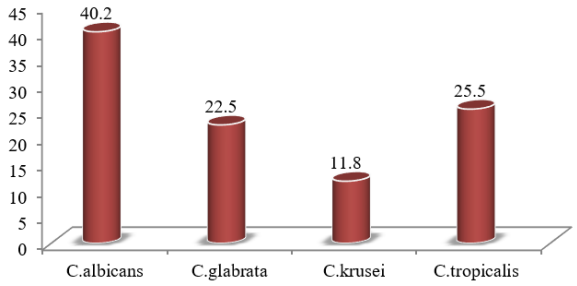

Figure 1 Percentage of isolated different species of Candida in diabetics and non-diabetics.

According to the results presented in Table 2 the prevalence of *Candida spp* infection in diabetic and non-diabetic patients based on ages was studied by using *Chi* square test, the results showed there was no significant difference in percentage of positive cases for Candida infection among different age groups in diabetic and non-diabetic patients (P>0.05).

We also studied the prevalence of *Candida spp* in study subjects in regard to gender, education level and smoking by using *Chi* square test. The results showed that there was no significant difference in prevalence of Candida infection based on gender, education level and smoking among diabetic and non-diabetic patients where the percent of positive cases for Candida were not significant, (P>0.05) as show in the Table 2.

In our study there were 25 diabetic patients that suffered from oral ulcer. The prevalence of *Candida spp* in diabetic patients with oral ulcer was studied by *Chi* square test the results found that the prevalence of Candida spp in diabetic patients with oral ulcer was 100% while in diabetic patients without oral ulcer was 71.4%. This difference was statistically significant (P≤0.05).

The prevalence of Candida spp in respect to hypertension and obesity in our study was studied by *Chi* square test; out of the 80 diabetic patients' positive for Candida, the prevalence of Candida spp among hypertensive diabetic patients was significantly higher (97.1%) than non-hypertensive diabetic patients (69.1%) with statistical significant difference (P<0.05).

Table 1 The prevalence of Candida infection in diabetics and non-diabetics groups

Variables		Diabetic	Non diabetic	Total No. (%)	*OR*	95% Confidence intervals	*P* value
		No. (%)	No. (%)				
Candida	Positive	80 (89)	22 (55)	102(78.4)	2.196	1.322– 3.647	<0.001
	Negative	10 (11)	18 (45)	28(21.6)			
	Total	90 (100)	40 (100)	130(100)			

OR (95% CI odd ratio, 95 confidence interval, and p < 0.05 significant

Table 2 Mean percentages of Candida load in the oral cavity and baseline characteristics of the diabetic patients compared to non-diabetics

Variables		Positive Candida			X^2	P value
		Diabetic	Non diabetic	Total		
		No. (%)	No. (%)	No. (%)		
Age group	40-50	29 (69.1)	13 (30.9)	42 (41.1)	2.140	0.141
	51-60	33 (86.9)	5 (13.1)	38 (37.2)		
	> 60	18 (81.8)	4 (18.1)	22 (21.5)		
	Total	80 (78.4)	22 (21.6)	102 (100)		
Gender	Males	51 (79.6)	13 (20.3)	64 (62.7)	0.160	0.689
	Females	29 (76.3)	9 (23.6)	38 (37.3)		
	Total	80 (78.4)	22 (21.6)	102 (100)		
Education level	Illiterate	33 (82.5)	7 (17.5)	40 (39.2)	0.875	0.646
	Primary	25 (73.5)	9 (26.4)	34 (33.3)		
	Secondary	22 (78.5)	6 (18.1)	28 (27.4)		
	Total	80 (78.4)	22 (21.6)	102 (100)		
Smoking	Yes	20 (68.9)	9 (31.1)	29 (28.4)	2.146	0.143
	No	60 (82.2)	13 (17.8)	73 (71.6)		
	Total	80 (78.4)	22 (21.6)	102 (100)		
Oral ulcer	Yes	25 (100)	0 (0)	25 (24.5)	9.107	0.003
	No	55 (71.4)	22 (28.6)	77 (75.5)		
	Total	80 (78.4)	22 (21.6)	102 (100)		

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Table 2 Continued...

Variables		Positive Candida			X^2	P value
		Diabetic	Non diabetic	Total		
		No. (%)	No. (%)	No. (%)		
Obesity	Yes	42 (95.5)	2 (4.5)	44 (43.1)	13.255	<0.001
	No	38 (65.5)	20 (34.5)	68 (56.9)		
	Total	80 (78.4)	22 (21.6)	102 (100)		
Hypertension	Yes	33 (97.1)	1 (2.9)	34 (33.3)	10.461	0.001
	No	47 (69.1)	21 (30.9)	68 (66.7)		
	Total	80 (78.4)	22 (21.6)	102 (100)		

Also, the results showed that the prevalence of Candida spp in obese patients was (95.5%) which was higher than non-obese patients (65.5%) with statistical significant difference (P<0.05).

Discussion

The present study was a cross sectional study that included 130 patients, 90 diabetic and 40 non diabetic as a control. Out of the 90 diabetic patients 57 (63.3%) were males and 33 (36.7%) were females while 24 (60%) from the control were males and 16 (40%) were females. The results showed that out of the 90 diabetic patients 80 (89%) had Candida infection whereas only 22 (55%) from the non-diabetic patients showed Candida infection. This difference was with statistical significant difference P<0.001 and odds ratio =2.196. Out of the 102 diabetic patients and non-diabetics, the percent of the *C. albicans* was 41 (40.2%) and *C. glabrata* was 23 (22.5%) while *C. tropicalis* and *C. krusi* were 26 (25.5%) and 12 (11.8 %) respectively.

Premkumar et al.,[9] conducted a study and reported that *C. albicans* was the most commonly identified species. However, they also observed the presence of *C. dubliniensis*, *C. tropicalis*, and *C. parapsilosis.*

According to Mohammadi et al.,[10] Candida spp. was detected in the oral cavity of diabetic patients, with *Candida albicans* being the most prevalent species (43.1%). In comparison, non-diabetic controls had a lower prevalence of Candida spp. (27%).

Our findings are consistent with those of Al-Attas and Amro,[11] who reported a higher frequency of Candida species isolation in diabetic individuals compared to non-diabetic individuals. They also found that the prevalence of *C. albicans* among diabetic patients was 68.9%, while it was 40.0% among healthy individuals.[11]

According to Mohammadi et al.,[10] diabetic patients are more susceptible to oral candidiasis due to several factors, including high levels of salivary glucose, reduced saliva secretion, impaired chemotaxis, and a defect in phagocytosis caused by a deficiency in polymorphonuclear leukocytes.[10]

The prevalence of *Candida spp* in diabetic and non-diabetic groups based on ages was studied by using *Chi* square test, the results showed there was no significant association in percentage of positive cases for Candida infection among different age groups in diabetic and non-diabetic patients (P>0.05). Sopian IL et al,[12] who showed no relationship between diabetes showing that there was no significant association between infection and age group.[1]

However, two previous studies (Natale & Rajagopalan,[13] Nguyen, Nguyen, & Tran,[14] reported that age is a risk factor for oral candidiasis in diabetic patients[3,4]. In contrast, our study found no significant association between Candida spp. and smoking (P = 0.143). These results are consistent with those of Mersil and Lailiqonita,[15] who found no statistically significant in the number of oral Candida between smokers and non-smokers (P = 0.820) (P>0.05).[14]

In our study 25 diabetic patients suffered from oral ulcer. The prevalence of *Candida spp* in diabetic patients with oral ulcer was studied by *Chi* square test. Our findings revealed that the prevalence of Candida spp in diabetic patients with oral ulcer was 100% whereas it was 71.4%. In diabetic patients without oral ulcer these findings suggest that the presence of oral ulcers may be associated with a higher likelihood of Candida infection in diabetic patients. However, further research is needed to confirm these results and explore potential underlying mechanisms.

The results of our study showed a statistically significant difference (P≤0.05) in the prevalence of Candida spp among hypertensive diabetic patients compared to non-hypertensive diabetic patients. Specifically, the prevalence of Candida spp was significantly higher among hypertensive diabetic patients (97.1%) compared to non-hypertensive diabetic patients (69.1%).

We also investigated the prevalence of Candida spp based on obesity status using the Chi square test. Our findings revealed that the prevalence of Candida spp was significantly higher (P<0.05) in obese patients (95.5%) compared to non-obese patients (65.5%).

The results found that hypertension and obesity considered as risk factors for Candida infections where that prevalence of Candida infection was significantly higher in hypertensive and obese patients compared to non- hypertensive, non -obese patients (P<0.05).

Conclusion

The overall prevalence of Candida spp. in our study population, including both diabetic and non-diabetic individuals, was found to be 78.4%. We observed a higher prevalence of Candida spp. in diabetic patients (89%) compared to non-diabetic patients (55%). Our study also found a strong and statistically significant association between Candida infection and diabetes (P=<0.001). Furthermore, our results revealed a significant association between Candida infection and oral ulcer, hypertension, and obesity, with a P-value of <0.001 for each factor.

Our findings showed that there was no statistically significant association between Candida infection and age, gender, education levels, smoking, or chewing Qat, with a P-value of >0.01 for each factor. However, *C. albicans* was the most frequently isolated Candida species, with a prevalence of 40.2%, while *C. krusei* was less common.

Based on the study results, we recommend that individuals at risk of diabetes take measures to prevent the spread of Candida through good personal, food, and drinking hygiene. Additionally, frequent monitoring of Candida infections should be conducted as a risk factor for diabetes. The study recommends the detection of oral Candida through confirmed diagnoses, such as biochemical tests. Future studies should focus on genotyping. Additionally, educating the general population about Candida infections and implementing hygiene programs are also recommended.

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Conflicts of interest

The authors declare no conflict of interest.

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