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Abstract

Introduction: In order to keep their dentures clean and properly stored, patients generally soak them in water at night. Candida albicans is a commensal yeast fungus that colonizes dentures, and in some conditions, it becomes an opportunistic pathogen and causes fungal infections known as candidiasis. **Objective:** This pilot study aimed to evaluate the effect of distilled water on Candida albicans colonizing dentures. **Materials and methods:** Twenty patients (9 men, 11 women; age range 40-75 years) with complete maxillary dentures infected by Candida albicans were included in this study. The dentures of these patients were soaked in distilled water for 4 days (8 hours at night). Swab samples from the dentures were collected before and after distilled water use and examined mycologically. **Results:** The Candida albicans colony counts increased after soaking the dentures in distilled water for 8 hours for 4 days. **Conclusion:** Patients should be dissuaded from soaking their dentures overnight in distilled water as the result is a significant increase in fungal colonization.

The Effect of Dentures' Overnight Soaking in Distilled Water: A Mycological Pilot Study

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ABSTRACT

Introduction: In order to keep their dentures clean and properly stored, patients generally soak them in water at night. *Candida albicans* is a commensal yeast fungus that colonizes dentures, and in some conditions, it becomes an opportunistic pathogen and causes fungal infections known as candidiasis. **Objective**: This pilot study aimed to evaluate the effect of distilled water on *Candida albicans* colonizing dentures. **Materials and methods:** Twenty patients (9 men, 11 women; age range 40-75 years) with complete maxillary dentures infected by *Candida albicans* were included in this study. The dentures of these patients were soaked in distilled water for 4 days (8 hours at night). Swab samples from the dentures were collected before and after distilled water use and examined mycologically. **Results:** The *Candida albicans* colony counts increased after soaking the dentures in distilled water for 8 hours for 4 days. **Conclusion:** Patients should be dissuaded from soaking their dentures overnight in distilled water as the result is a significant increase in fungal colonization.

Keywords: *Candida albicans*, denture, distilled water.

INTRODUCTION

Candida albicans is a commensal yeast fungus. In some conditions, it becomes an opportunistic pathogen and causes fungal infections known as candidiasis [1]. Among these conditions enabling this transformation is denture wear [2]. Gendreau and Loewy report that 15 to 70% of denture wearers have denture stomatitis, a form of oral candidiasis, and that the oral hygiene-related risk factors for this condition are significantly connected with morbidly increased colonization of *Candida albicans* found in biofilm highly adherent to the base material of dentures [3].

In this respect, daily cleaning of dentures is an important factor in the elimination of biofilm formation. Several modalities of denture-cleaning techniques have been suggested, such as mechanical brushing, microwave sterilization, and denture immersion in effervescent tablets and antiseptics [4, 5].

In a previous study investigating the hygiene habits of acrylic denture wearers in a sample of the Lebanese population, we found that 15.9% of patients soak their dentures in water (tap water or distilled water) during the night because, for them, chemicals from the cleanser may irritate their soft tissues [6].

To our knowledge, the effect of the distilled water on *Candida albicans* colonizing dentures has not been investigated yet. The aim of this pilot study was to evaluate the effect of distilled water on *Candida albicans* biofilm on dentures.

MATERIALS AND METHODS

This pilot study was conducted in accordance with the Helsinki agreement for research. All patients were informed in advance about the experimental procedure and their consent was obtained. Complete maxillary edentulous denture-wearing patients were assessed for clinical evidence of Newton's type II denture stomatitis (erythema involving part or all of the mucosa, which is covered by the denture and usually caused by *Candida albicans*) [7].

Twenty patients aged between 40 and 75 years who had been wearing their dentures for more than one year were included in this study. In order to standardize, they were asked not to clean their dentures during the experimental procedure.

Candida albicans colony counts were performed on the first day (D1) from the fitting side of the denture of each patient, and the dentures were soaked in distilled water during the night for 8 hours (from 10 pm to 6 am) for 4 consecutive nights.

A second swab collection destined for a new *Candida albicans* colony count was taken on day 4 (D4).

One investigator carried out microbiological procedures. The Becton-Dickinson (New Jersey, USA) Microbiology System, BBL CultureSwab, was used. These systems are sterile devices for collecting and transporting microbiological specimens (Amies, Stuart, and agar gel).

Swabs were cultured in Sabouraud's dextrose agar (40 g/l dextrose, 10 g/l peptone, and 20 g/l agar) and containing chloramphenicol 0.5 g/l and actidione 0.5 g/l. A Candida count was carried out after 48 hours of incubation at 37°C in aerobic conditions. *Candida albicans* was differentiated from the other species by their production of filaments in 0.5 ml of animal serum.

The primary outcome measure was the analysis of *Candida albicans* colony count expressed in CFU/ml (colony-forming unit) collected from the denture surface at Day 1 (D1) and after the 4 nights of immersion at Day 4 (D4).

RESULTS

After immersion in distilled water for 8 hours for 4 consecutive nights, *Candida albicans* colony counts were even higher than before distilled water use in the majority of the patients' dentures.

Table 1 summarizes the details of the study (ages of the patients and *Candida albicans* colony counts-CFU/ml from the infected oral mucosa and the fitting side of the dentures before and after the dentures' immersion in distilled water).

Patient	**Name and Birth Date**	**D1-Denture** Before Immersion **(CFU/ml)**	**Immersion in distilled water**	**D4-Denture** After Immersion **(CFU/ml)**
1	J.M (M:72 years)	$>10^6$		$>10^6$
2	J.A (M: 67 years)	800		15500
3	O.M (F:71 years)	$>10^6$		$>10^6$
4	J.N (F:71 years)	$>10^6$		$>10^6$

5	E.A (F:57 years)	$>10^6$		$>10^6$
6	R.A (F:48 years)	6000		6200
7	N.H (F:75 years)	1100		1750
8	M.M (M:69 years)	1300		9800
9	K.M (M:55 years)	1500		1200
10	S.R (M:53 years)	400		900
11	R.H (M:57 years)	13500		10500
12	S.K (M:45 years)	400		850
13	Y.M (F:74 years)	12000		8500
14	N.F (F:63 years)	120		180
15	J.R (F:61 years)	40		40
16	G.K (F:64 years)	$>10^6$		$>10^6$
17	M.J (M:44 years)	40		40
18	M.K (F:75 years)	13000		13300
19	T.K (F:40 years)	142		155
20	S.S (M:57 years)	4000		2450

Table 1: Patients' age and counts of CFU/ml from the fitting side of the dentures at D1 and after the denture's immersion in distilled water at D4

DISCUSSION

The Candida genus is amongst the top fungi, and *Candida albicans* is the major fungal pathogen of humans [8]. In the mouth, the most common fungal infection is candidiasis, caused by *Candida albicans* colonizing the oral cavity of 40 to 60% of healthy people [9]. Dentures, reduced saliva, and poor oral hygiene predispose to candidiasis by producing an anaerobic environment with low oxygen and low pH conducive to *Candida albicans* outgrowth and biofilm

formation [10]. A study by Budtz-Jørgensen et al. (1996) detected DS in 72% of denture wearers in an elderly population living in a geriatric institution [11].

It has been widely agreed that appropriate routine cleaning of dentures prevents them from being infected by *Candida albicans* [6, 10, 12]. Kulak-Ozkan et al. (2002) evaluated clinically and mycologically 70 complete denture wearers and concluded that there is a statistically significant relationship between yeast presence and denture cleanliness [13]. It is well accepted that denture immersion, especially with chemical solutions, has some advantages over mechanical cleaning, such as effective disinfection and ease of use [10, 14].

Furthermore, patients wearing complete dentures are generally advised to soak them in clean water at room temperature overnight for two reasons: a) to keep them clean and properly stored; b) to prevent them drying out; and consequently, to prevent an increase in surface roughness.

In their study comparing the retention of *Candida albicans* on smooth and rough acrylic resin, Verran and Maryan concluded that microfissures and cracks within the material and an increased surface roughness promote *Candida albicans* colonization [15].

The present study investigated the effect of distilled water on *Candida albicans* colonizing dentures. Our results showed the preservation of high numbers of *Candida albicans* colony counts.

Our findings agree with Stafford et al., who found that *Candida albicans* colonization on the maxillary denture base is significantly reduced when dentures are left in the air to dry for 8 h, whilst the density increases if stored in water for a similar period [16].

To our knowledge, no other studies have been conducted on the same topic.

CONCLUSION

Distilled water presents negative effect on *Candida albicans* biofilm colonizing dentures after soaking them for eight hours on four consecutive nights. Patients should be dissuaded from soaking their dentures overnight in distilled water as the result is a significant increase in fungal colonization.

Conflict of interest statement: The author declares that there is no conflict of interest.

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