# Efficacy of Three Denture Cleansers on Candida-Related Denture Stomatitis

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#### الخلاصة

الاهداف: تحدف الدراسة الى تقييم كفاءة ثلاث مطهّراتِ لطقم الأسنان على النهاب الفمّ الناتج عن طقم الأسنان والمتعلق بفطرٍ المبيّضات بجساب قابليتها على تقليل الأعدادِ الحية مِنْ فطرٍ المبيّضات بَعد غمرٍ طقم الأسنان بالمطهرات ولأوقات مختلفة. المواد و طرائق العمل: اثنا عشر مريضا ممن لديهم النهاب الفم الناتج عن طقم الاسنان وزعوا الى اربع مجاميع بالاعتماد على انواع مطهر الطقم و هي ( بروتيفكس ، ٢% محلول الصوديوم هايوكلورايت ،المخلول المشبع لملح كلوريد الصوديوم ومجموعة القياس الماء ). احري الاحتماد اللمكتبري لتقييم كفاءة المطهرات باحتساب الأعدادِ الحية مِنْ فطرٍ المبيّضات بعد غمرٍ طقم الأسنان بالمطهرات ولأوقات مختلفة الأعدادِ الحية مِنْ فطر المبيّضات ( يوم قبل المعالجة وبعد ٤ ايوم و ٢٨ يوم من المعالجة ) وقد احري احتبار تحليل التباين واختبار دنكن لاحتساب الفرق المعنوي بين المجموعات عند مستوى الاحتمالية(٥%) النتائج : اظهرت النتائج ان هناك تاثير معنوي للمطهرات الثلاثة على تقليل الاعداد الحية من فطر المبيضات بعد معالجة التهاب الفم لمدة (١٤ و ٢٨) يوما بالمطهرات ولكن تاثير الماء كان غير معنويا وان مطهر صوديوم هايوكلورايت هو الاكثر كفاءة. الاستناجات الخهسرت النتائج انه لا يوجد فرقا معنويا بين كفاءة المطهرات الثلاثية وانحيا جميعا كفوءة في معالجة التهاب الفيم النتاتج عن طقم الاسينان.

### **ABSTRACT**

Aim: The aim of the study is to evaluate the efficacy of three dentures cleanser on *Candida* related denture stomatitis through assessing their ability to reduce the number of colony forming unit (CFU) of *Candida* on palate of the patient for different times. **Materials and methods:** Group of 12 patients who have denture stomatitis were divided into four groups according to the type of denture cleaners (Protefix, sodium hypochlorite solution 0.02%, saturated sodium chloride salt solution and control water). The microbiological examination was done to determine the efficacy of the cleansers that used by assessing the number of (CFU) at different times (day before treatment and after 14, 28days of treatment). The data were statistically analyzed, (ANOVA) followed by Duncan's multiple Range test to assess the significant difference between the groups at  $P \le 0.05$ . **Results:** The results showed that there was a significant effect of three cleansers on reducing number of (CFU) of *Candida* species in the treatment of denture stomatitis after 14 and 28 days of treatment, but it was not significant for control water group. Sodium hypochlorite showed the highest efficacy. **Conclusions:** Result showed that there was no significant difference in the efficacy between the three cleansers indicating that all cleansers are effective in the treatment of denture stomatitis.

Key words: Denture cleaners, Candida, Stomatitis

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# **INTRODUCTION**

Acrylic resin is the most commonly used material for denture bases construction. (1, 2) Denture stomatitis is one of the oral infections associated to the use of dentures. It is a chronic disease characterized by either localized or generalized inflammation or inflammatory papillary hyperplasia that may affect patients wearing complete or partial removable prostheses in one or both dental arches. (3,4)

Candida is a significant opportunistic pathogen of humans and is a major cause

of denture stomatitis, an inflammation of tissues underlying dentures. Adherence is critical to the pathology of *Candida*, serving as a first step of infection for many microorganisms. In healthy mouth saliva flows and scraping by the tongue limit the accumulation of microorganisms. Prostheses impair this flushing, facilitating establishment of a focal infection by retention of *Candida* close to the basal seat. (5,6) The disinfection of the prosthesis is recommended as prophylactic treatment and necessary for patients with high risk of

stomatitis. (7,8)

The aim of the study is to evaluate the efficacy of three denture cleansers on *Candida* related denture stomatitis through assessing their ability to reduce the number of colony forming unit (CFU) of *Candida* on palate of the patient with denture stomatitis for different times (day before treatment and after 14, 28days of treatment).

## MATERIALS AND METHODS

Patients selection and Data Collection

The patients enrolled in the current study have complete denture with conventional heat cured acrylic resin denture base material for more than 2 years, attending the Department of Prosthetic Dentistry, Mosul University. All the subjects underwent a routine dental checkup. A total of patients, 50 patients were chosen (who do not have the exclusion criteria). The group consisted of 30 males and 20 females with age vary between (43years – 75 years). The exclusion criteria were: (1) Subjects taking antifungal agents or antiseptic mouthwashes.(2) Subjects taking medication known to predispose them to oral candidiosis, such as antibiotics or steroid therapy. (3) Subjects with a medical history of any disease or medical condition that predisposed them to oral candidiosis or promoted subjects oral carriage of Candida species. (9)

#### Clinical Examination

The oral mucosa covered by the maxillary complete denture was examined to determine and diagnose denture stomatitis. Denture stomatitis (denture sore mouth) is a form of erythematous *candidiasis* in which the diffuse redness of the mucosa corresponds to the area covered by the denture, it most diverts on the palate and may occur in acute or chronic form. It is characterized by pin-point hyperemia, diffuse erythema or granular inflammation of the denture-bearing areas. (10,11)

## Microbiological Examination

A cotton swab was taken from the posterior mid-palatal part of the palatal mucosa of the patients. (9,12) Then the swab placed in a screw capped bottle containing (4.5ml) of nutrient broth (Hemadia laboratorie ) as a transparent medium. The broth was diluted up to 10<sup>-4</sup> by serial dilution , and 0.1 ml of diluted broth was spread over two of Sabouraud's dextrose agar Petri dish (Oxoid/England) and incubated for (48 hrs at 37 °C) in incubator (Fisher Scientific / Russia) and counted as (CFUs/ml) for the viable *Candida*. (13,14)

Identification of *Candida* was performed by the following diagnostic laboratory tests: (Culture characteristics): On Sabouraud' Dextrose Agar medium within (24-48) hrs at 37°C, *Candida* species produce soft creamy colored colonies with a yeast odor.<sup>(13, 14)</sup> (Figure 1).



Figure (1):Colony Morphology of Candida

The smears obtained from the culture of patient's specimens were examined by light microscopic (Lomo/Micmed/Russia) using crystal violet staining; the *Candida* 

species took gram positive stain. Microscopically it appeared as spherical to oval budding cells (3-6 m $\mu$ ); the yeast or the blastospore form  $^{(15)}$  (Figure 2) .

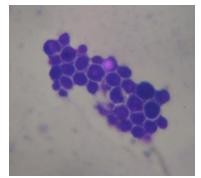


Figure (2): Candida under light microscope

Efficacy of Three Types of Denture Cleansers on Candida Related Denture Stomatitis

Group of 12 patients from 50 patients that chose of denture stomatitis were determined in the clinical examination. Patients were randomly divided into four groups according to the type of denture cleaners. Each group of three patients; first their denture soaked group, Protefix/Germany (its pH is 7.49) which prepared according to the manufacturer's instruction. Second group soaked in sodihypochlorite solution (Clorox/Saudi Arabia). (16,17) Third group soaked in saturated salt solution 40gm/ml Sodium chloride (without iodine) /Iraq. (18,19) Fourth group as control group dentures were soaked in water. Dentures were soaked in cleaning solutions overnight on each day of the study periods (8 hours). The microbiological examination

was done by the same way to determines the efficacy of the cleanser that used according to (the number of colony forming unit at each time). The efficacy of denture cleanser agents is assessed in reducing the number of microorganisms on palate of the patients at three different times (day before treatment and after 14, 28 days of treatment).

The data were statistically analyzed including; Descriptive statistic to show the mean and standard deviation. Analysis of variance (ANOVA) followed by Duncan's multiple Range test to assess the significant difference between the groups at  $P \le 0.05$ .

## **RESULTS**

In this study, the mean number of CFUs of microorganisms on the palatal mucosa for the different cleansers for three different times are shown in Tables (1).

Table (1): The mean CFUs of *Candida* on the Palatal mucosa for patients used different cleanser before treatment and after 14 and after 28 days of treatment.

Cleansers	Period	No.	Mean	<u>+</u> SD
Saturated Salt Solution	Before treatment	3	500.0	173.20
	14 Days	3	16.67	5.77
	28 Days	3	0.00	0.000
Sodium Hypochlorite	Before treatment	3	416.6	175.59
0.02%	14 Days	3	10.0	0.000
	28 Days	3	0.00	0.000
Protefix	Before treatment	3	333.3	57.73
	14 Days	3	123.3	92.91
	28 Days	3	3.3	5.77
Control	Before treatment	3	400.0	132.28
Water	14 Days	3	346.6	100.16
	28 Days	3	363.3	125.03

No. Number of samples

The result showed that sodium hypochlorite 0.02% has the highest efficacy in reduce number of CFUs after 14 and 28 days of treatment, while water group has the least effect. One way analysis of vari-

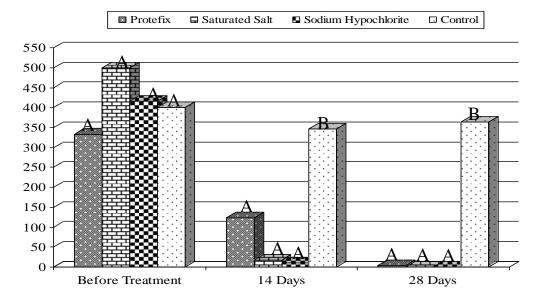
ance (Table 2) shows that there is a significant difference on the CFUs of *Candida* between all cleansers after 14 and 28 days of treatment .

Table (2): Analysis of variance (ANOVA) among different cleansers for the mean of CFUs of *Candida* taken from palatal mucosa of the patients

Contains taken from patients interest to patients						
Before treatment	Sum of Squares	D.F.	Mean Square	F–value	<i>p</i> –value	
<b>Between Groups</b>	42291.667	3	14097.22	00.690	0.853	
Within Groups	163333.33	8	20416.66			
Total	205625.00	11				
14 Days after treat-	Sum of Squares	D.F.	Mean Square	F-value	*p-value	
ment						
<b>Between Groups</b>	222291.66	3	74097.22	15.850	0.001	
Within Groups	37400.00	8	4675.00			
Total	259691.66	11				
28 Days after treat-	Sum of Squares	D.F.	Mean Square	F-value	*p-value	
ment						
<b>Between Groups</b>	295233.33	3	98411.11	25.126	0.000	
Within Groups	31333.33	8	3916.66			
Total	326566.66	11	_			

<sup>\*</sup>P-value: Means are highly significant different at  $P \le 0.01$  **DF:** Degree of freedom

The Duncan's Multiple Range Test for the effect of cleansers on the *Candidal* CFUs (taken from the Palate of the patients) after 14 and after 28 days of treatment (Figure 3).



Means with the same litter have no statistically significant difference at  $P \le 0.05$  Figure (3): Duncan's Multiple Range Test (DMRT) for the effect of different cleansers on the *Candidal* CFUs (taken from the palate of the patients) after 14, 28 days of treatment.

The result revealed that there is no significant difference between the effect of three denture cleaners on CFUs by *Candida* after 14 and 28 days of treatment, but there is a significant difference be-

tween the three cleaners and that of control group (water). The three agents that were used by the patients in this study are significant in reducing the No. of CFUs of *Candida* species. The application of

Protefix (commercial denture cleanser) modulates the candidal adherence to denture acrylic and affect the colonization rate of *Candida* on the surfaces of dentures and palatal mucosa, this agrees with Nalbant *et al.* <sup>(9)</sup>

Sodium hypochlorite is one of the earliest and most widely used disinfectant. It can be a bactericide and fungicide, because it acts directly on the organic matrix of the plaque, resulting in the dissolution of the polymer's structure, probably because of oxidation of the protean component or significantly reducing the adhesion of most Candida sp. to the oral epithelial cells<sup>(17)</sup>. These characteristics allow the hypochlorite to reduce Candida sp. adhesive ability, but it does not work as an anti invasive barrier, as it is not able to prevent the production of proteinases by the Candida sp. These findings show that sodium hypochlorite acts as an anti-fungi agent if used as a solution to submerge dentures in cases of denture stomatitis. (20)

Sodium chloride is commonly called as salt, common salt, table salt. It is an ionic compound having the structural formula NaCl. The most important property of sodium chloride is its inflammability. Many microorganisms cannot live in excess salty environment, water is drawn out of the cells by the process of osmosis. For this reason, salt is used for disinfecting. (21,22)

The results showed that there is a significant difference between saturated salt solution and distilled water as Protefix and Sodium hypochlorite 0.02%, therefore, it is considered as effective method in disinfection of denture contaminated with *Candida* species, this agrees with Al-Sumaidae.<sup>(19)</sup>

This study showed that disinfection of the denture is a successful method of treatment, and the denture is the source of the infection and no other treatment necessary, and this agrees with Webb *et al.*<sup>(16)</sup>

# **CONCLUSIONS**

The results revealed that the three cleansers were have significant effect in reducing the No. of CFUs of *Candida* species. Also, the result showed that there is no significant difference in treatment efficacy between the three cleansers indicat-

ing that all cleansers were effective in the treatment of denture stomatitis.

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