



## Efficacy of commercial chlorhexidine gluconate mouth rinse on candida colonization in patients with complete dentures

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### ABSTRACT.

The aim of the present study was to evaluate the efficacy of application of chlorhexidine mouthwash on *Candida* colonization in patients with complete dentures. Sixty-eight patients with complete denture were included in this study and divided in two groups. One group of patients were instructed to rinse their mouth with chlorhexidine mouthwash once per day for one week. The second group of patients were instructed to use placebo mouthwash containing normal saline solution. Palatal swabs and smears were taken from each patient before and after chlorhexidine and saline application and specimens were examined mycologically. For each group, *Candida* colony forming units (CFU) were assessed using Sabouraud agar culture. The difference between *Candida* colonization before and after treatment and the differences between pre-treatment and post-treatment clinical findings were assessed using paired t test. Application of chlorhexidine mouthwash significantly reduced the CFU and improved the palatal inflammation ( $p < 0.05$ ). This reduction was found in all patients even in smokers and diabetic patients, as well as in the subjects without suitable denture health. We demonstrated the effectiveness of application of a commercial chlorhexidine gluconate mouthwash in the management of *Candida* colonization in patients with complete dentures specially in high-risk candidates.

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### Introduction

Complete dentures cause many changes in the oral cavity including bone resorption, morphologic changes in the alveolar ridge as well as oral mucosal reaction.(1,2) Oral candidiasis is one of the most common mucosal infections in the denture wearers which is caused by overgrowth of *Candida* species, particularly *Candida Albicans*.(3,4) The infection caused by these micro-organisms usually appears as denture stomatitis, which is found in 24-60% of denture wearers that might happen at anytime.(5) Failure of therapy in antifungal-resistant candidiasis is not uncommon.(6) Increased incidence of candidal infection in these patients necessitates effective prevention.(7) It seems that excellent oral hygiene and denture care are not sufficient for infection prevention. Therefore, application of appropriate chemical agents (such as Chlorhexidine gluconate) can minimize mucosal or denture related microbial colonization.(8) Chlorhexidine gluconate (CHX) is a broad spectrum antimicrobial agent with very few side-effects.(9) Its effective antifungal action has been demonstrated by many *in vivo* and *in vitro* studies.(8-10) It has been shown that different concentrations of CHX may be effective to reduce the risk of candidiasis through reduction of *Candida* colonization and adhesion to the oral epithelial cells.(10) Therefore, the present study was designed to evaluate the efficacy of early application of 0.12% CHX (the most common commercial concentration) on *Candida* colonization in patients with complete dentures.

### Methods and Materials

Sixty-eight patients with complete dentures were participated in this study. They were selected randomly from volunteer patients who referred to prosthodontic department of Shiraz University of Medical Sciences, School of Dentistry. Complete

medical and dental history were obtained from each participant and the patients who presented history of taking antibiotics, and steroids or using other mouthwashes within the past three months were excluded. All the experiments in this study were reviewed and approved by Shiraz Dental School ethical board. The patients were randomly divided into two groups: 1) CHX groups: who were instructed to use CHX mouthwash once per day for one week, where patients rinsed their mouth with 0.12% CHX mouthwash (Peridex, 3M, St Paul, MN) for 1 minute; and 2) control group (used normal saline (NS) solution (NaCl 9%). In both groups, before and after use of either of mouthwashes, the dorsal portion of tongue was swabbed with a sterile cotton tip applicator, with one firm movement in about 3-cm length to collect the samples. These samples streaked on to Sabouraud Dextrose Agar (SDA) containing chloramphenicol plates.<sup>11</sup> The second samplings were performed after one week of CHX application with the same method and transferred to the culture. Subsequently, well-formed colonies were counted and colony-forming unit (CFU) was evaluated for each sample and microbiological measurements were quantified. In addition, in order to score the clinical effects of application of CHX, the Budtz-Jorgensen, index was utilized. Palatal examination was categorized as either positive: no inflammation, improvement: decreased inflammation, or negative: no change in inflammation.(12,13) Clinical data including age, gender, history of diabetes mellitus, and smoking as well as patients' personal denture-cleansing method were recorded.

One-way analysis of variance (ANOVA) and paired t-test was used for pair wise comparisons using SPSS (Version 15) statistical analysis software. Statistical significance was assigned at  $p < 0.05$ .

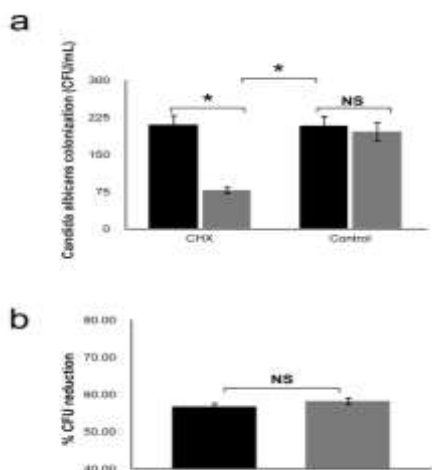
## Results

The study groups were included 33 males and 35 females from 61 to 80 years with mean of 73 years. According to denture hygiene program, 40% (n=28) were cleaning the denture with water, 20% (n=14) with toothpaste, 15% (n=10) with soap and 25% (n=16) were not cleaning their denture at all. Thirty five percent of the participants were smokers and 32% were diabetics (Table 1).

**Table 1. Baseline data of the patients participated in this study.**

	Number
Gender	
Male	33 (59%)
Female	23 (41%)
Smoking	
Smoker	20 (35%)
Non-smoker	36 (65%)
Systemic status	
Diabetic	18 (32%)
Non- Diabetic	38 (68%)

The mean CFUs, before and after using CHX mouthwash are illustrated in Figure 1. After using CHX mouthwash, the mean *C. albicans* CFU counts showed a significant decrease ( $P<0.001$ ). However, for the control group no statistically significant difference was observed before and after application of the salt solution (Fig 1a). Furthermore, there was no significant difference in the reduction of CFU between smokers and non-smokers ( $P=0.65$ ) as well as diabetic and non-diabetic patients ( $P=0.55$ ). Application of CHX mouthwash did not show different effects on CFU reduction between patients with various denture-cleansing methods ( $P=0.7$ ), and also between males and females (Fig 1b).



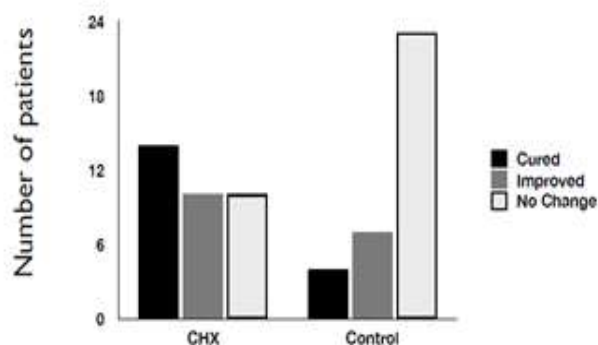
**Figure 1. (a) Candida albicans colonization (CFU/mL) before and after the treatment in the CHX and control groups. (b) Percentage of reduction of CFUs between males and females**

Results of clinical palatal examination after 7 days of application of CHX are exhibited in Figure 2 demonstrating significant reduction in palatal inflammation after application of CHX. Clinical examination revealed that more than 71% of the CHX group responded to CHX application positively (60% with positive results and 40% of the patients showed improvement). A statistically significant decrease ( $p=0.04$ ) in palatal inflammation was seen. However, no significant change or improvement was observed in the control group ( $P>0.05$ ).

## Discussion

Candida fungi exist in the oral cavity of about 60% of healthy adults without signs and symptoms of candidiasis. In various systemic or local conditions, such as in patients with complete

dentures or diabetes mellitus, this organism can proliferate and produce oral candidiasis with various presentations.



**Figure 2. Results of clinical palatal inflammation scores from the CHX and control groups**

Furthermore, growth of *Candida* species has been reported in 66% of denture wearers, especially in diabetic patients.(4) In the present study, we evaluated the efficacy of early application of 0.12% CHX mouthwash on colonization of *Candida* species in the patient with complete denture. We found that rinsing the mouth for 3 minutes; can significantly reduce the CFU. This finding is compatible with Vianna *et al* study, that showed 37% reduction in CFU after 1 minute of mouth washing.(14)Other studies have reported the antifungal activity of CHX on acrylic resin dentures, as well as improvement of *Candida*-associated mucositis beneath the denture bearing areas.(6,15)In addition, several studies have demonstrated sub-therapeutic dose of CHX can exhibit germ-tube formation of *Candida* species.(14, 15) Early studies have reported that 30% of CHX retains in the mouth for 24 hours after rinsing for 1 minute, although it is removed almost in the first few hours.(17) Efficacy of an early application of CHX mouthwash and spray has been shown to be effective in lowering the growth rate and reduction of culture scores of oral flora in ventilated patients.(18)We found the same efficacy of CHX in diabetic and non-diabetic patients who are extremely susceptible to thrush. Antifungal activity of CHX, but not complete elimination of *Candida*, has been shown in some systemic conditions such as diabetic patients, neutropenic children and bone-marrow transplant receivers. (19-21) Sneha *et al.* have shown an increased *Candida* growth in diabetic patients with complete dentures.(6) Reduction of *Candida* colonization also, has been revealed in hypo-salivation as well as in HIV-infected patients with compromised immune response.(22, 23) In addition to systemic conditions and wearing complete denture, some local factors such as smoking may increase the risk of *Candida*-associated mucositis.(24, 25)

In the current study, it was found that smoker patients derived benefits from mouth washing with 0.12% CHX. Also, efficacy of CHX in CFU reduction was similar in both genders. Cleansing denture plays an important role in prevention of infections in the oral cavity. In the present study, CHX had antifungal influence in patients with various methods of denture cleansing, even in patients who didn't clean their dentures at all. Some reports have shown that CHX mouthwash can be used as a denture disinfectant solution. 2% CHX has shown antifungal effects through reduction of *Candida* adhesion to denture base and even has been a disinfectant solution for fluconazole-resistant *Candida* species.(6) Different concentrations of CHX have been surveyed *in vivo* and *in vitro* to determine different antifungal functions. In addition to measurement of culture scores, reduction of *Candida* adhesion to epithelial cells as well as to the denture base, have been also reported. It has been shown that CHX mouth washing in a long duration (at least 4 weeks) increased the saliva

flow. Also, Kadir *et al.* have demonstrated that sub-therapeutic CHX can modulate phospholipase activity of *Candida albicans*. (26) These functions of CHX make it a promising disinfectant and preventive antifungal agent in susceptible patients.

### Conclusion

In the current study, we confirmed the effectiveness of application of commercial chlorhexidine gluconate mouthwash in the management of candida colonization and improving the clinical symptoms in patients with complete dentures. This mouthwash can be used as a preventive agent in the edentulous, complete denture users. Reduction of potentially pathogenic flora leads to obtain better oral intervention and control of potential infections of oral cavity.

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