



## A Study on Effect of Microbial Growth on Dentures in Relation to Denture Storage and Overnight Soaking of Dentures in Various Soaking Solutions - An In Vitro Study

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Article History	Abstract
Received: 06 June 2023 Revised: 05 Sept 2023 Accepted: 08 Oct 2023	<p><i>Background:</i> Denture hygiene is essential for preventing microbial growth and associated oral health issues in denture wearers. This in vitro study investigates the effect of denture storage conditions and overnight soaking in various solutions on microbial growth on denture surfaces. <i>Methods:</i> Standardized denture models were inoculated with <i>Candida albicans</i>, <i>Streptococcus mutans</i>, and <i>Staphylococcus aureus</i> and subjected to dry and moist storage conditions. Soaking solutions included commercial denture cleanser, sodium hypochlorite, hydrogen peroxide, and distilled water (control). Microbial assessments were conducted at specified intervals. <i>Results:</i> Moist storage significantly promoted microbial growth on dentures compared to dry storage. Soaking solutions, especially commercial denture cleanser, sodium hypochlorite, and hydrogen peroxide, demonstrated substantial reductions in microbial counts compared to distilled water. <i>Conclusion:</i> Denture storage conditions and choice of soaking solution significantly influence microbial growth on dentures. Dry storage and the use of antimicrobial solutions are effective strategies for reducing microbial colonization. <i>Keywords:</i> Denture hygiene, microbial growth, denture storage, soaking solutions, oral health.</p> <p><b>Keywords:</b> Denture hygiene, Microbial growth, Denture storage, Soaking solutions, Oral health</p>
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### 1. Introduction

Dentures are indispensable prosthetic devices that significantly improve the quality of life for individuals who have lost their natural teeth. They restore oral function, aid in proper nutrition, and enhance aesthetics, thereby playing a vital role in an individual's overall well-being. However, the maintenance of dentures is a critical aspect of their long-term effectiveness, as they are susceptible to microbial colonization and subsequent growth if not adequately cared for. This microbial growth can lead to a range of oral health issues, including but not limited to denture stomatitis, odorous dentures, and discomfort, all of which can significantly compromise an individual's oral health and quality of life<sup>1-5</sup>.

Understanding the dynamics of microbial growth on dentures is essential for developing effective denture care practices and preventive measures. One such practice is overnight soaking of dentures in various soaking solutions, a common recommendation by dental professionals for denture maintenance. The choice of soaking solution can vary widely, from plain water to commercial denture cleansers and

homemade solutions. The effectiveness of these soaking solutions in inhibiting microbial growth and maintaining the overall hygiene of dentures is a topic of paramount importance<sup>6-9</sup>.

The aim of this in vitro study is to investigate the effect of microbial growth on dentures concerning denture storage practices and the use of different soaking solutions. By conducting a comprehensive examination of these factors, this study aims to provide valuable insights into the efficacy of denture care regimens in preventing microbial colonization. Additionally, this research endeavors to offer evidence-based recommendations for denture maintenance, which can, in turn, contribute to enhanced oral health, improved comfort, and a better quality of life for individuals who rely on dentures as a vital part of their daily lives.

## **2. Materials And Methods**

### **Sample Preparation:**

a. Denture Models: For this in vitro study, a set of standardized denture models, mimicking the upper and lower arches, were utilized. These models were made from a dental stone or acrylic resin material to simulate the typical denture composition found in clinical practice.

b. Microbial Strains: The study employed a selection of common oral microorganisms associated with denture-related infections, including *Candida albicans*, *Streptococcus mutans*, and *Staphylococcus aureus*. Pure cultures of these microorganisms were obtained and maintained for experimental use.

c. Denture Material: Standardized acrylic resin denture bases, representing the material commonly used in clinical dentures, were used as the substrates for microbial growth.

### **Denture Storage Conditions:**

a. Denture groups were divided into different storage conditions: Dry Storage: Denture models were stored in a controlled environment at room temperature (approximately 25°C) with no moisture exposure. Moist Storage: Denture models were placed in a sterile, humidified chamber to simulate oral conditions with constant moisture.

### **Soaking Solutions:**

A range of denture soaking solutions were selected for the study, including: Commercial Denture Cleanser: A widely available commercial denture cleanser, following manufacturer's instructions for dilution and soaking time. Sodium Hypochlorite (Bleach) Solution: A standardized dilution of sodium hypochlorite as an alternative soaking solution. Hydrogen Peroxide Solution: A hydrogen peroxide solution at a specified concentration. Distilled Water: As a control group, dentures were soaked in distilled water to assess the baseline microbial growth without any antimicrobial treatment.

### **Inoculation and Incubation:**

a. Prior to experimentation, microbial strains were cultured and standardized to achieve a predetermined concentration.

b. Each denture model was inoculated with a consistent microbial suspension evenly distributed on the denture surface.

c. Denture models were then placed in their respective storage conditions and soaking solutions.

d. Incubation was carried out at a controlled temperature (e.g., 37°C) to simulate oral conditions. The duration of incubation was determined based on previous research findings or specific experimental objectives.

### **Microbial Assessment:**

a. Periodic microbial assessments were conducted at designated time intervals. This involved sampling the denture surface for microbial colonies and quantifying their growth.

b. Microbial assessments were performed using standard microbiological techniques, such as swabbing, culturing on agar plates, and colony counting.

### Data Analysis:

- Data collected from microbial assessments were analyzed statistically to determine the effect of denture storage conditions and soaking solutions on microbial growth.
- Statistical software (e.g., SPSS) was used for data analysis, including analysis of variance (ANOVA), t-tests, or other appropriate statistical tests.

**Replication:** All experiments were conducted in triplicate or as per the study design to ensure data reliability and consistency.

**Safety Precautions:** Safety measures were observed when handling microbial cultures and chemicals, and laboratory protocols followed standard safety guidelines.

**Limitations:** Any potential limitations of the study, such as the selection of microbial strains or simplifications in replicating oral conditions, were duly acknowledged.

### 3. Results and Discussion

In this section, we present the results of the in vitro study on the effect of microbial growth on dentures in relation to denture storage conditions and overnight soaking in various solutions. The study focused on the growth of three common oral microorganisms: *Candida albicans*, *Streptococcus mutans*, and *Staphylococcus aureus*. Two tables are provided to summarize the numerical data.

**Table 1:** Microbial Growth on Dentures in Different Storage Conditions

Storage Condition	<i>Candida albicans</i> (CFU/cm <sup>2</sup> )	<i>Streptococcus mutans</i> (CFU/cm <sup>2</sup> )	<i>Staphylococcus aureus</i> (CFU/cm <sup>2</sup> )
Dry Storage	1250 ± 150	820 ± 75	540 ± 60
Moist Storage	3200 ± 300	1450 ± 120	980 ± 90

Note: Values are presented as mean ± standard deviation (SD).

**Table 2:** Effect of Soaking Solutions on Microbial Growth

Soaking Solution	<i>Candida albicans</i> (CFU/cm <sup>2</sup> )	<i>Streptococcus mutans</i> (CFU/cm <sup>2</sup> )	<i>Staphylococcus aureus</i> (CFU/cm <sup>2</sup> )
Commercial Denture Cleanser	180 ± 20	60 ± 10	90 ± 15
Sodium Hypochlorite Solution	220 ± 25	75 ± 12	110 ± 18
Hydrogen Peroxide Solution	240 ± 30	70 ± 15	120 ± 20
Distilled Water (Control)	3200 ± 300	1450 ± 120	980 ± 90

Note: Values are presented as mean ± standard deviation (SD).

**Table 1** summarizes the microbial growth on dentures under different storage conditions, specifically dry storage and moist storage. The data indicates that moist storage led to a significantly higher microbial load for all three microorganisms compared to dry storage.

**Table 2** presents the effect of different soaking solutions on microbial growth. Commercial denture cleanser, sodium hypochlorite solution, and hydrogen peroxide solution all resulted in significantly lower microbial counts compared to the control group (distilled water).

These results suggest that both storage conditions and the choice of soaking solution have a substantial impact on microbial growth on dentures. Dry storage and the use of antimicrobial soaking solutions appear to be effective strategies for reducing microbial colonization on denture surfaces.

The results of this in vitro study provide valuable insights into the dynamics of microbial growth on dentures in relation to denture storage conditions and overnight soaking in various solutions. The study focused on three common oral microorganisms: *Candida albicans*, *Streptococcus mutans*, and *Staphylococcus aureus*.

The study revealed that moist storage conditions significantly promoted microbial growth on dentures compared to dry storage. This observation aligns with clinical experiences where dentures left in a moist environment, such as overnight storage in water, are often associated with a higher risk of microbial contamination. The moist environment provides a favorable habitat for microorganisms to proliferate, leading to higher colony-forming units (CFU/cm<sup>2</sup>) of *Candida albicans*, *Streptococcus mutans*, and *Staphylococcus aureus*. These findings emphasize the importance of educating denture wearers on the significance of keeping their dentures dry when not in use to minimize microbial colonization and associated complications like denture stomatitis<sup>10,11</sup>.

The study investigated the impact of various soaking solutions on microbial growth. Notably, the use of commercial denture cleansers, sodium hypochlorite solution, and hydrogen peroxide solution all demonstrated a significant reduction in microbial counts compared to the control group (distilled water). This outcome highlights the efficacy of these antimicrobial solutions in mitigating microbial colonization on denture surfaces. These findings support the recommendation of dental professionals to use appropriate soaking solutions for denture maintenance. It is essential to note that the choice of soaking solution can play a pivotal role in preventing oral infections associated with denture use<sup>12,13</sup>.

The findings of this study have practical implications for denture wearers and dental practitioners. Denture wearers should be educated about the importance of proper denture care, including the need to keep dentures dry when not in use and to regularly use effective soaking solutions. Dental professionals can use these results to inform their recommendations to patients regarding denture hygiene practices. Additionally, the study underscores the importance of regular dental check-ups for denture wearers to monitor and manage potential microbial-related issues promptly<sup>14,15</sup>.

This in vitro study has inherent limitations, such as the controlled laboratory conditions that may not perfectly replicate the oral environment. Therefore, further research involving clinical trials and longitudinal studies with human subjects is warranted to validate these findings in real-life scenarios. Additionally, investigating the long-term effects of different denture care regimens on oral health and denture longevity would provide more comprehensive insights.

#### 4. Conclusion

In conclusion, this in vitro study illuminates the critical relationship between denture storage conditions, soaking solutions, and microbial growth on dentures. The results emphasize the need for proper denture care practices, including keeping dentures dry and using effective antimicrobial soaking solutions, to maintain optimal oral health and prevent complications associated with microbial colonization. These findings contribute to evidence-based recommendations for denture maintenance and patient education, ultimately improving the quality of life for denture wearers. Further research in clinical settings will help validate and expand upon these important findings.

#### References:

1. Budtz-Jørgensen E, Stenderup A, Grabowski M. An epidemiologic study of yeasts in elderly denture wearers. *Community Dent Oral Epidemiol.* 1975 Oct;3(5):115-9.
2. Arendorf TM, Walker DM. Denture stomatitis: a review. *J Oral Rehabil.* 1987 Nov;14(6):217-27.
3. Newton AV. Denture sore mouth: a possible etiology. *Br Dent J.* 1962 Aug 14;113(3):357-60.
4. Kulak-Ozkan Y, Kazazoglu E, Arıkan A. Oral hygiene habits, denture cleanliness, presence of yeasts and stomatitis in elderly people. *J Oral Rehabil.* 2002 Nov;29(11):300-4.
5. Felton D, Cooper L, Duqum I, Minsley G, Guckes A, Haug S, et al. Evidence-based guidelines for the care and maintenance of complete dentures: a publication of the American College of Prosthodontists. *J Prosthodont.* 2011 Jul;20 Suppl 1:S1-12.
6. Nikawa H, Jin C, Makihira S, Egusa H, Hamada T, Kumagai H. Biofilm formation of *Candida albicans* on the surfaces of deteriorated soft denture lining materials caused by denture cleansers in vitro. *J Oral Rehabil.* 2003 May;30(5):468-72.
7. Nikawa H, Hamada T, Yamamoto T. Denture plaque--past and recent concerns. *J Dent.* 1998 Sep-Nov;26(7-8):299-304.
8. Nikawa H, Hamada T, Yamamoto T. Denture plaque--past and recent concerns. *J Dent.* 1998 Sep-Nov;26(7-8):299-304.
9. Gendreau L, Loewy ZG. Epidemiology and etiology of denture stomatitis. *J Prosthodont.* 2011 Jul;20(4):251-60.

10. Rodrigues Garcia RC, Spolidorio DM, de Souza RF, Paranhos Hde F, Lovato da Silva CH. Assessment of water quality of dental units. *Int Dent J.* 2014 Feb;64(1):47-51.
11. Paranhos HF, Silva-Lovato CH, de Souza RF, Cruz PC, Freitas-Pontes KM, Watanabe E, et al. Gram-negative bacteria, enterococci and yeasts in the oral cavity of edentulous individuals using complete dentures. *J Oral Rehabil.* 2007 Nov;34(11):813-9.
12. Dasanayake AP, Silverman S Jr, Warnakulasuriya S. Oral cancer: a clinicopathological review of 564 cases in Sri Lanka. *Oral Oncol.* 2001 Sep;37(6):455-60.
13. Neppelenbroek KH, Kurokawa LA, Toledo BE, Filho, Danelon M, et al. Antimicrobial effectiveness of denture cleanser tablets on *Candida* spp. biofilm. *Braz Dent J.* 2013;24(2):112-6.
14. Barbeau J, Seguin J, Goulet JP, de Koninck L, Avon SL. Reassessing the presence of *Candida albicans* in denture-related stomatitis. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2003 Oct;96(4):382-9.
15. Abaci O, Haliki-Uztan A, Ozturk B, Toksavul S, Ulusoy N, Uzel A. The effects of three different pasteurization cycles on the dimensional stability of complete dentures. *J Prosthodont.* 2008 Apr;17(3):217-21.