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# Comparative Analysis of Different Chemical Methods for Removing Biofilm from Complete Dentures

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## Abstract

The aim of this study was to make a comparative evaluation of three chemical substances for cleaning complete dentures, as regards their efficacy of biofilm removal. The sample consisted of 20 maxillary complete dentures. The dentures were randomly divided into groups according to the chemical cleaning method to be used: Group 1 - water (control), Group 2 - sodium hypochlorite solution, Group 3 - sodium perborate (Corega Tabs®) and Group 4 - 2% chlorhexidine. The groups were evaluated in terms of the quantity of biofilm before and after application of the chemical cleaning method by applying a revealer, and later, analysed by the Denture Hygiene Index (DHI). The results showed that only sodium hypochlorite solution was effective for biofilm removal. There was statistically significant difference among the groups, pointing out greater efficacy of the method used in Group 2 in comparison with Groups 1 and 4. Group 3 did not differ statistically from any other group evaluated. The results allowed to conclude that sodium hypochlorite solution is the most efficient chemical agent for removing biofilm from complete dentures. However, when used alone, all the tested chemical cleaning methods were incapable of eliminating all the biofilm from denture surfaces.

Keywords: Complete denture; Hygiene; Biofilm; Disinfection

## Introduction

The increase in the population group formed by elderly individuals is a well described demographic phenomenon in Brazil and worldwide. It is estimated that 9% (18 million inhabitants) of the Brazilian population will be aged 65 years or over in the year 2020. This population requires better living conditions, and oral health is a contributory factor that favors their well-being [1].

It is certain that the majority of elderly persons today underwent curative dentistry, in which the main treatment therapy recommended was extraction, now involving a deficiency of the oral structures, reduced number of healthy teeth and a large number of edentulous individuals and complete denture wearers [2]. In Brazil, according to the Ministry of Health, 57.9% of the population over the age of 65 years wears dentures in the maxillary arch and 24.8% in the mandibular arch [2].

It is perceived that patients who become complete denture wearers continue with the same precarious cleaning habits, as well as their dentures [3]. The correct use of and care taken of dentures are of great importance to patients, not only for esthetic and functional reasons, but also for the health of the supporting tissues and adequate preservation of the denture itself [2]. It is known that daily and adequate denture and soft tissue cleaning is imperative, with the goal of maintaining a healthy oral mucosa. Rehabilitative treatment with complete dentures performed by dentists must include motivating and instructing the patient to promote effective biofilm control by means of cleaning methods [3].

Biofilm present on complete dentures must be removed, and this can be done by means of mechanical and chemical methods, or association of the two. The mechanical method consists of using a tooth brush and dentifrice or neutral soap. The chemical method is performed by immersing the denture in chemical products that have a solvent, detergent, fungicidal and/or bactericidal action. Whereas the chemical-mechanical method is based on a combination of brushing and dentifrice/neutral soap followed by immersing the denture in chemical solutions. Among the chemical agents, hypochlorite, alkaline peroxides, diluted acids, enzymes and chlorhexidine are outstanding [3].

Although there are a diversity of methods for cleaning complete dentures, there is still no consensus in the literature about which method is most effective for removing biofilm from acrylic resin denture base surfaces. Thus, the aim of this study was to make a comparative evaluation of three chemical substances for cleaning complete dentures: sodium hypochlorite solution, sodium perborate – Corega Tabs® (Block Drug Company, Inc. – USA) and 2% chlorhexidine, as regards the efficacy of biofilm removal from complete denture surfaces. The null hypothesis admitted is that the chemical cleaning methods proposed in this research are not effective in removing biofilm from complete dentures.

## **Material and Methods**

The sample consisted of 20 maxillary complete dentures from patients of an School of Dentistry. The clinical charts were consulted in order to call patients who were maxillary complete denture wearers to come for a follow-up consultation of the prosthetic treatment they had received in the Institution. These patients were informed of the nature, purpose and form of the work proposed in this research, and only after they signed the Term of Free and Informed Consent, they participated of the study, which was approved by the Research Ethics Committee of the Institution (098.2010.2).

Each denture was washed in running water for 5 seconds and then dried with jets of air for a further 10 seconds. A swab was used to apply a biofilm revealer (5% Erythrosine) on the internal surface of each complete denture and remained in contact with the acrylic base for 1 minute. After this, the complete dentures were again washed in running water for 5 seconds and dried with jets of air for 10 seconds for the purpose of removing the excess revealer [4].

Quantitative analysis of the biofilm present was performed visually by means of the standardized photographic technique, at an angle of 90°, using a digital camera (NIKON-P60, NIKON, Japan). The images were transferred to a computer and analyzed in the CorelDraw 2008 program. A standardized division of nine parts (Figure 1) was superimposed on each image, and the presence of biofilm was quantified using the method proposed by Schubert and Schubert [5] for classifying the cleaning level of complete dentures. Each part of the division traced on the image was evaluated by a blinded researcher, according to the following codes and criteria:

- 0 Absence of plaque;
- 1 small spots of plaque;
- 2 less than half covered with plaque;
- 3 more than half covered with plaque;
- 4 the entire area covered with plaque.

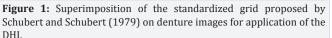
The scores, which were attributed to each segment by a trained and calibrated researcher, were added together and the sum divided by the total number of segments (nine).

Two photographic records were made of the internal surface of each denture after revealing biofilm: 1) before and 2) after application of a chemical complete denture cleaning method.

The dentures were divided into 4 groups, according to the chemical substance used:

• Group I - Control: immersion in water for 15 minutes.





- Group 2 sodium hypochlorite solution: Immersion in a solution consisting of 15ml of 2.25% sodium hypochlorite and 200ml water, for 15 minutes.
- Group 3 Sodium Perborate (Corega Tabs Block Drug Company, Inc. - USA): immersion in 100ml warm water containing one Corega Tabs tablet, for 5 minutes, according to the manufacturer's instructions.
- Group 4 2% Chlorhexidine (Villevie Joinville Santa Catarina Brazil): Immersion in 100ml of the substance for 15 minutes.

The chemical agent used for cleaning each denture was determined by draw, a process that guaranteed randomized research.

After the chemical cleaning process, the dentures were cleaned with a tooth brush and neutral soap for complete removal of biofilm revealer, before being returned to the patients.

The data collected in this research were processed and analyzed by Statistical Software SPSS (Statistical Package for Social Sciences) version 18.0, at a level of significance of 5%.

#### Results

The nominal values of biofilm quantified were tabulated in spreadsheets and analyzed by means of descriptive statistics using the SPSS program. The normality of distribution was verified by the Shapiro-Wilk test and homocedacity by Levene's Test. The biofilm values quantified before and after were compared in each experimental group by the paired *t*-test.

Data on the difference between biofilm quantified before and after, named "effect", were compared among the 4 experimental groups by the Analysis of Variance and Tukey's post hoc test.

The results showed that only the agent used in Group 2 was effective for biofilm removal. The agents in the other groups tested were not effective in removing biofilm from complete denture surfaces Table 1.

There was statistically significant difference among the groups, pointing out the greater efficacy of the agent used in Group 2 (Figures 2C and D) in comparison with the Group 1

(Figures 2A and B) and 4 (Figures 2G and H). Group 3 (Figures 2E and F) did not differ statistically from any other group evaluated.

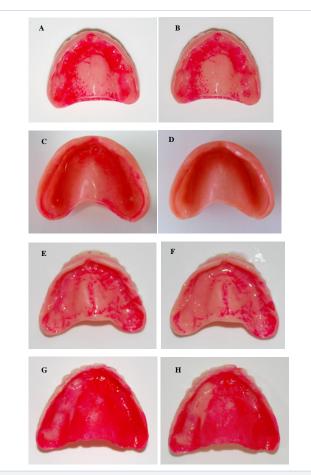
When used alone, none of the tested chemical cleaning methods were capable of eliminating all the biofilm from denture surfaces.

Table 1: Statistical analysis of different chemical cleaning methods.

GROUP	BIOFILM EFORE	BIOFILM AFTER	CLEANING EFFECT (BIOFILM BEFORE – AFTER)
1	1.88 (±0.48)	1.5 (±0.62)	0.37 (±0.31) B
2	2.79 (±1.16)	0.95 (±0.9)*	1.84 (±0.96) A
3	2.79 (±1.04)	1.81 (±1.32)	0.98 (±0.78) AB
4	2.17 (±0.29)	1.81 (±0.21)	0.35 (±0.37) B

\* Indicates significant difference between before x after within the same group.

Different letters indicate significant difference between the groups as regards cleaning effect.



**Figure 2:** Photographs of dentures before and after application of different cleaning methods, respectively: A and B) Immersion in water; C and D) Immersion in sodium hypochlorite solution; E and F) Immersion in sodium perborate (Corega Tabs®); G and H) Immersion in 2% chlorhexidine.

## Discussion

One of the measurements of effectiveness of a complete denture cleaning agent is its ability to remove biofilm. Nikawa et al. [6] drew attention to the need for this effectiveness to be evaluated by means of clinical and laboratory tests, as the *in vitro* results do not necessarily agree with those of *in vivo* tests. As regards complete denture cleaning agents, clinical studies prevail over laboratory studies.

In clinical experiments, biofilm levels can be evaluated on the denture as a whole; that is, without distinction between one surface and the other. However, in the majority of these, this evaluation is performed on the internal surface of the maxillary complete denture [7].

In this study, the effectiveness products was evaluated on the internal surface of maxillary complete dentures, as it consists of an area where there is great biofilm accumulation, and this is of great clinical importance as regards the pathologies found in complete denture wearers, with the objective being to evaluate the efficacy of different chemical agents for removing biofilm from complete dentures. This study focused on using some of the products available on the market and reported on in the literature as being helpful in denture cleaning.

Surveys have confirmed that the mechanical method of hygiene is still the most popular among complete denture wearers [6-8]. Immersion in effervescent alkaline peroxide solutions is also widely used (9). Jagger and Harrison (10) in a survey of 120 complete denture and partial removable denture wearers, demonstrated that approximately 35% used a chemical cleanser in the form of effervescent tables as part of the daily hygiene routine.

In Brazil, a small portion of the population uses this method as a cleaning aid. In spite of being controversial, this may be explained by the lack of access to materials of a chemical nature, as they are sold to a large extent, or even due to the cost factor. A cause that could also be pointed out is the lack of knowledge of patients themselves about this method as a coadjuvant for complete denture cleaning [9, 10].

The agreement of this study with those of Keng and Lim [11] and Nicholson, Stark and Scott [12] shows that studies in which comparisons were made with control groups using water, chemical cleansers presented limited effectiveness.

Differently from the results obtained by Sheen and Harrison [13] and Paranhos et al. [14], who, in standardized clinical experiments found effectiveness of immersion in peroxides and silicone polymers respectively, in this study it was demonstrated that the method of immersion in alkaline peroxide was not effective for the removal of biofilm from dental prosthetic appliances, even when using a period of immersion of 5 minutes, according to the manufacturer's instructions.

It was found that sodium hypochlorite solution, in terms of effectiveness for removing biofilm from complete dentures, was not shown to be very efficient and presented no result of inefficiency, the same results as those obtained in the study of

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Catão et al. [3] in which around 100% of biofilm was removed in approximately 37% of the sample and around 75% of biofilm present in 30% of the complete dentures.

Normally 0.12% chlorhexidine gluconate is used for prevention of dental biofilm formation and antimicrobial action against Candida albicans, whereas 2% chlorhexidine is used for removing biofilm from surfaces. In the study of Semenoff et al [15], 2% chlorhexidine was more effective in inhibition microbial growth than 0.12% chlorhexidine. In the study of Segundo et al. [16], in which the two products were tested to verify cleaning, the results showed no statistical difference between them, and had similar antimicrobial action. The same result was observed in this research and by Catão et al. [3], in which 2% chlorhexidine was considered inefficient, since the regions previously revealed remained colored and unaltered in the majority of the sample. However, the effectiveness of 2% chlorhexidine in a study against the microbial activity of Candida albicans was the best in comparison with substances such as 1% sodium hypochlorite [17].

When analyzing the factor effectiveness, an item that must be considered is the immersion time used. Previous studies have indicated that short periods of immersion in chemical cleansers are not effective [18,19], and greater effectiveness could be obtained if a prolonged immersion period were used [20]. Although denture wearers are reluctant to remove their appliances, even during the period of sleep, it would be important to conduct a clinical study with the use of a prolonged period of immersion to verify the possibility of increasing the effectiveness of these immersion products. The use of additional daily immersions (three times a day) may be an alternative, however the cost factor must be analyzed [21].

Although evaluation of the efficiency of chemical complete denture cleansers is recognized by means of the methodology used in this study, it is necessary for this analysis to be complemented by other tests, such as evaluation of the action of chemical agents against specific pathogenic microorganisms in a similar clinical situation. It is also necessary to use a larger, and consequently, more representative sample of the studied population, with the purpose of seeking more significant and more scientifically reliable results.

It is suggested that future studies should be conducted to evaluate the cleaning capacity of different chemicals used at present, in different concentrations and exposure times, so that the dentist may have better scientific understanding and be able to provide complete denture wearing patients with guidance.

#### Conclusions

Analysis of the results found in this research permitted one to conclude that:

- The sodium hypochlorite solution tested is effective for removing biofilm from complete dentures, denying the null hypothesis of this research;
- 2% chlorhexidine is not efficient for removing biofilm from complete dentures, confirming the null hypothesis;

- Sodium perborate (Corega Tabs) is not efficient for removing biofilm from complete dentures, confirming the null hypothesis;
- The sodium hypochlorite solution tested is more efficient for removing biofilm from complete dentures than 2% chlorhexidine;
- When used alone, the tested chemical cleaning methods were incapable of eliminating all the biofilm from denture surfaces.

In view of the above findings, the authors of this research concluded that the immersion of complete dentures in the sodium hypochlorite solution tested for 10 minutes must be the chemical method of choice for indication to patients as an aid to mechanical cleaning of their complete dentures.

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