Effect of sodium bicarbonate on *Candida albicans* adherence to thermally activated acrylic resin

**Abstract:** The purpose of this study was to evaluate the effect of 5% sodium bicarbonate on the adherence of *Candida albicans* to thermally activated acrylic resin. Fifty 4 mm² specimens of acrylic resin were obtained using a metallic matrix. The specimens received chemical polishing, were sterilized and then immersed in Sabouraud broth, inoculated with *Candida albicans* standardized suspension. After 24 hours of incubation at 37°C, the specimens were divided into four groups according to the substance used for disinfection (5% sodium bicarbonate, 0.12% digluconate chlorhexidine, vinegar and Corega Tabs). A control group was included, in which distilled water was used. The adhered microorganisms were dispersed, diluted and plated onto culture media to determine the number of colony-forming units (cfu/mL). The results were analyzed through the Mann-Whitney statistical test at the 5% level of significance. Only 0.12% digluconate chlorhexidine and 5% sodium bicarbonate presented a statistically significant difference (p = 0.0010 and p = 0.0156, respectively) compared to the control group, decreasing the number of cfu/mL. However, when the different disinfecting solutions were compared with each other, only 0.12% digluconate chlorhexidine presented a statistically significant difference in the reduction of cfu/mL. It was concluded that although 0.12% digluconate chlorhexidine was more effective in the reduction of *Candida albicans* adherence values to thermally activated acrylic resin, 5% sodium bicarbonate also proved to be a viable alternative.

**Descriptors:** Sodium bicarbonate; Acrylic resins; *Candida albicans*; Cell adhesion.

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Introduction

In spite of all advances in dentistry, complete dentures are still essential for oral rehabilitation of edentulous dental arches. However, this type of prosthesis, which is basically confectioned with thermally activated acrylic resin, constitutes a favorable environment for the colonization and proliferation of Candida genus yeasts, since these microorganisms have the ability to strongly adhere to polymethylmethacrylate, which constitutes the acrylic resin.\(^1,2\)

This phenomenon, in association with factors such as trauma, diet, poor hygiene or predisposing systemic conditions (xerostomia, hormonal alterations and immunodepression caused by diabetes mellitus or HIV infection), may lead complete denture users to develop a condition known as denture stomatitis.\(^3-7\) This condition is clinically characterized by a discrete focal inflammation on the palate, which can evolve to an intense erythema in all area covered by the denture and, in some cases, to papillary hyperplasia.\(^6\)

According to Daniluk et al.\(^5\) (2006), Candida genus yeasts can be isolated from the human oral cavity in 64.9% of patients with denture stomatitis, being Candida albicans the most prevalent species (78.4%). In fact, studies have shown that regardless of the presence of clinical symptoms, Candida albicans is more prevalent in complete denture users than in the general population.\(^2,5,8\) This condition makes complete denture users more susceptible to the development of other forms of candidosis when submitted to conditions that might lead to a disequilibrium in the oral microbiota, such as usage of broad-spectrum antibiotics, anti-histamines, chemo- or radiotherapy. The presence of conditions that cause reduction in salivary flow, including diabetes mellitus, drug usage, poor nutrition and advanced age, for instance, may also cause oral microbiota disequilibrium.\(^9-12\)

However, simple attitudes such as the removal of the denture during the night and its correct hygiene with the daily usage of disinfecting solutions are mentioned in the literature as effective in the prevention and treatment of denture stomatitis.\(^4,13-15\) Thus, many studies have been performed with the objective of evaluating the efficiency of different disinfecting solutions in the control of Candida genus yeasts, searching for a disinfecting solution that would present excellent antimicrobial activity without harming the patient’s health or causing alterations in the physical-structural characteristics of dentures.\(^16-20\)

Within this context, the objective of the present study was to evaluate the effect of sodium bicarbonate in the adherence of Candida albicans to thermally activated acrylic resin, comparing it to other disinfecting solutions already mentioned and tested in the literature.

Material and Methods

Fifty 4 mm\(^2\) standardized specimens were obtained using thermally activated acrylic resin (Artigos Odontológicos Clássico Ltda., São Paulo, SP, Brazil) and an aluminum matrix. Chemical polishing of the specimens was performed using a Termotron PQ-9000 chemical polisher (Termotron do Brasil Ltda., Piracicaba, SP, Brazil). The specimens were kept in glass recipients containing physiological serum (0.85% NaCl), sterilized in autoclave (121°C/15 min) and submitted to adherence testing.\(^21\)

Candida albicans ATCC 1880 was plated on Sabouraud's dextrose agar and incubated at 37°C for 24 hours. After this period, a suspension containing \(10^6\) viable cells per milliliter was prepared in sterile saline solution (0.85% NaCl) with the aid of a UV-1203 Shimadzu spectrophotometer (Shimadzu Corp., Kyoto, Japan), adopting the wavelength of 530 nm and optical density of 0.284.

Adherence testing was performed in an aseptic environment, in a laminar air flow chamber using 24-wells plates for cell culture. For this purpose, 1.5 mL of Sabouraud broth (Difco Labs., Detroit, MI, USA), one specimen and 0.1 mL of Candida albicans standardized suspension were added to each well. The plates were sealed and incubated at 37°C for 24 hours.

After this period of incubation, the specimens were washed with 1 mL of sterile distilled water and randomly divided into five groups (\(n = 10\)), according to the disinfecting solution tested: control group (distilled water); group B (5% sodium bicarbonate - Labsynth Produtos para Laboratórios Ltda., Di-
adema, SP, Brazil); group V (white vinegar - Castelo Alimentos, Jundiaí, SP, Brazil); group C (Corega Tabs - GlaxoSmithKline Brasil Ltda., Rio de Janeiro, RJ, Brazil); and group P (0.12% digluconate chlorhexidine - Periogard - Colgate Palmolive Ind. e Com. Ltda., São Paulo, SP, Brazil). The specimens were immersed for 10 minutes in 10 mL of the corresponding solution, according to the group.

After immersion, the specimens were then removed, washed with 1 mL of sterile distilled water and transferred to tubes containing 1 mL of sterile physiological solution (0.85% NaCl) and glass pearls. The tubes were agitated in a Vortex stirrer for 60 seconds and the adhered cells were dispersed. This initial suspension was diluted 10, 100 and 1,000 times in physiological solution, and 0.1 mL of each suspension was plated in duplicate on Sabouraud dextrose agar (Difco Labs., Detroit, MI, USA). After 48 hours of incubation at 37°C, the number of colony-forming units per specimen (cfu/specimen) was determined.

The data was statistically analyzed through the Mann-Whitney test, at the 5% level of significance, comparing groups two by two.

Results

The mean value of cfu/mL ± standard deviation in the control group was $7.50 \times 10^3 \pm 5.69 \times 10^3$. Meanwhile, groups B, V, C and P presented, respectively, the following results: $2.55 \times 10^3 \pm 1.80 \times 10^3$, $6.04 \times 10^3 \pm 7.96 \times 10^3$, $6.01 \times 10^3 \pm 6.55 \times 10^3$, and $1.05 \times 10^3 \pm 1.58 \times 10^3$. Group P presented the highest reduction in comparison to the control group (86.00%), followed by groups B (66.00%), C (19.90%) and V (19.50%). (Graph 1)

In fact, only groups P and B showed significant statistical difference when compared to the control group ($p = 0.0010$ and $p = 0.0156$, respectively). However, when the experimental groups were compared with each other, only group P presented significant statistical difference ($P \times B, p = 0.0211$; $P \times V, p = 0.0233$; $P \times C, p = 0.0046$).

Discussion

Chlorhexidine’s efficiency in the treatment of denture stomatitis is recognized since 1970. Budtz-Jørgensen and Løe, in 1972, showed that disinfection of complete dentures with 2% chlorhexidine for 15 minutes, twice a day, was capable of reducing the inflammation caused by stomatitis and also completely eliminated the presence of Candida hyphae. Lal et al. (1992) demonstrated similar results after the use of 0.12% chlorhexidine-based rinsing solutions.

Several studies have shown that the disinfection of complete dentures with either 2% or 4% chlorhexidine, as well as the use of either 0.12% or 0.2% chlorhexidine-based rinsing solutions is effective in the reduction of the total number of cfu/mL of Candida albicans adhered to the surface of acrylic resin and, consequently, effective in the prevention and treatment of denture stomatitis, especially when both methods are associated. In fact, the results presented in the present study corroborated the findings in the literature, since, among the disinfecting solutions, 0.12% digluconate chlorhexidine presented the best results, reducing in 86% the number of microorganisms adhered to the surface of the specimens.

However, although chlorhexidine presents excellent antimicrobial properties such as low-concentration efficiency, substantivity, minimal perception by the gastrointestinal tract, capacity to reduce biofilm formation and disorganize pre-formed biofilm, it also presents some side effects, including mild discomfort or burning sensation when chlorhexidine comes in contact with the oral mucosa, in addition to epithelium exfoliation, teeth staining, enhance-
ment of supragingival calculus and color alteration of artificial teeth and denture basis.22,23,25,28

Thus, there is a need to research alternative disinfecting solutions, aiming at minimizing the side effects of disinfecting solutions to the physical-chemical properties of dentures and to the patient’s health. Sodium bicarbonate has been used for disinfection of dentures and orthodontic appliances, but there is no corroboration in the literature to the efficiency of this substance regarding its influence on adherence. Thus, one of the study groups of the present study included sodium bicarbonate, and, based on the results, it has been shown to be a viable alternative, at least considering its anti-Candida adherence effect, since it significantly reduced the adherence of Candida albicans to the surface of the specimens.

In fact, many studies have shown that sodium bicarbonate at high concentrations has an antimicrobial effect over several microorganisms isolated from the oral cavity, including Candida albicans.29,30 However, none of these studies evaluated the effect of sodium bicarbonate on the adherence of these microorganisms. According to Newbrun29 (1997), sodium bicarbonate presents low abrasion, low cost, relative safety if accidentally ingested and compatibility with the fluoride present in most dentifrices, which is an important advantage over chlorhexidine. This compatibility associated to its efficiency against Streptococcus mutans30 makes sodium bicarbonate an excellent option for complete or partial denture users, who still present remaining teeth in the arches.

Regarding white vinegar and Corega Tabs, both were inefficient in the reduction of the number of Candida albicans colonies adhered to the specimens under the conditions in the present study. In addition, the lack of studies regarding these solutions in the literature contraindicates their use, especially by patients who have already presented signs of stomatitis and, therefore, need a more aggressive approach. However, new studies should be performed, using other concentrations and other immersion times, before a definitive conclusion can be reached.

Conclusion

Based on the results obtained in the present study, it was concluded that, among the disinfecting solutions tested, 0.12% digluconate chlorhexidine was the most effective in the reduction of Candida albicans adherence to thermally activated acrylic resin. However, 5% sodium bicarbonate also proved to be a viable alternative.

References