



A COMPARATIVE EVALUATION OF THE EFFICACY OF PERACETIC ACID-BASED DISINFECTANT FOR DECONTAMINATION OF ACRYLIC RESINS- AN IN VITRO STUDY

Dental Science

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ABSTRACT

Aim: To assess the antimicrobial efficacy of peracetic acid and chlorhexidine for the decontamination of Acrylic resins.

Material and method: sixty specimens of chemically activated acrylic resin were fabricated and contaminated in vitro by inoculum of *Candida* sp. (ATCC 10231). Twenty specimens from each group were immersed in the following disinfecting agents: Peracetic acid, Chlorhexidine and Sodium perborate. The contaminated specimens were immersed in the disinfecting solution for 10 mins. After disinfection, suspensions were prepared of each group. Plating was performed on Sabraud's Dextrose agar culture medium and incubated at 37°C for 48 hrs. Bacterial growth was observed by analyzing the number of colony forming units (CFU) after incubation. Values of each group were statistically compared.

Results: The results showed that colony forming units were less on decontamination by Peracetic acid as compared to Chlorhexidine and Sodium perborate.

Conclusion: Immersion for atleast 10 min in 5% peracetic acid promoted high-level disinfection of the acrylic resins.

KEYWORDS

Peracetic acid, Acrylic resins, *Candida albicans*, disinfection

INTRODUCTION:

In pediatric dental practice, acrylic resins have been used in day to day basis in the form of removable prosthesis. Oral cavity of healthy individuals with or without teeth may be colonized by yeast and bacteria coexisting in a relationship of commensalism which may result in several infections including denture stomatitis, aspiration pneumonia, and lung and gastrointestinal infections.¹ Most commonly found microbes in oral cavity of children are streptococcus, staphylococcus, *Neisseria*, *Candida* etc. which may serve as a reservoir of infection.² *Candida albicans* is known to be the most causative factor of denture stomatitis.³ Of all the microbes, *Candida albicans* has high capacity to adhere to denture base resins and form structured biofilm. Other factors that may result in oral candidiasis are denture base fit, metabolic disorder, patient's age, mucosa conditions, epithelial changes, poor diet, appropriate denture hygiene, xerostomia and salivary flow.⁴

Correct prosthetic use and daily hygiene are important factors for good oral health, greater longevity of the prosthesis and health of supporting tissue. It was reported that daily hygiene has been essential to prevent oral mucosal inflammation and lesions and that the patients do not receive professional instructions on how to clean their denture.¹ MacCallum et al. (2000) reported that patients themselves choose a denture cleanser without informing and thus, it becomes difficult for dentists to recommend cleansing agents. They use homecare products which could cause harmful effects.⁵ Andruccioli et al. (2004) reported that the chemical methods were not routinely applied, either due to lack of information or knowledge about these methods, cost or lack of access, or nonavailability of these products in the market.⁶

Dentures can be cleaned mechanically, chemically, and a combination of them.² Ideal denture care products should remove inorganic/organic deposits and stains; have cost-effective, bactericidal, and fungicidal properties; and be easy to handle for the human health and harmless for the denture materials. The American Dental Association and Centers for Disease Control and Prevention guidelines 2009 recommend that dental prostheses should be disinfected before being sent to the laboratory and before delivery to the patient.³

For the disinfection of acrylic resin several disinfectants have been suggested. Peracetic acid (C₂H₄O₃) is a strong chemical disinfectant with a broad antimicrobial spectrum. Peracetic acid-based disinfectants have been used in food industry, as well as for

decontamination and sterilization of thermosensitive medical and hospital equipment and devices.⁷ Peracetic acid is a more powerful oxidant agent than chloride and chloride dioxide, causing the rupture of the cell membrane by means of protein denaturing. In this study, our aim is to evaluate the effectiveness of peracetic acid based disinfectants on acrylic resin surfaces.

MATERIALS AND METHOD:

Sixty heat cured acrylic resin cubes of polymethyl methacrylate resin (DPI company) of size 10×10×2mm according to American society for testing and material standard were made. These are prepared according to manufacturer's instructions, by mixing 3 parts of polymer to 1 part of monomer. After curing using short curing cycle they were polished with pumice and kept in water until use. *Candida albicans* (ATCC 10231) was included for antimicrobial activity test.

The following disinfectants were included in study are 5% Peracetic acid, 0.2% chlorhexidine gluconate and 3.8% sodium perborate. The specimens were distributed into three groups for each disinfectant. The specimens and the control group were contaminated with inoculum and incubated.

Disinfection of samples:

After contamination, all samples were rinsed with sterile distilled water for 30 sec. except the control group, remaining samples were immersed in 10 ml of each disinfectant to be tested for 10 minutes. After 10 minutes, again the disinfected specimens were immersed in sterile distilled water for 2 seconds to remove excess disinfectant solution.

Microbiological survey:

After excess disinfectant removal, the samples were transferred to 10 ml of normal saline solution and were agitated for 5 min to remove the adhered cells. From this suspension, 100 microliters were plated on Sabaroud's Dextrose Agar (SDA) media and were spread uniformly. After drying, the petri dishes were placed in incubator at 25°C for 48 hours. After incubation, the grown fungal colonies on SDA were counted before and after disinfection.

The data was submitted to statistical package for social sciences (SPSS) software and wilcoxon test was run to analyze the data.

RESULTS:

The number of Colony forming units obtained during initial cultures is

constant i.e., 1.5×10^8 cells/ml (P - 0.05). Confluent growth (100% growth) of CFU was observed. Among all the groups, the control group showed viable bacteria at all experimental times, which indicated the efficiency of method.

Table 1: results for antimicrobial effectiveness

Disinfectant solution	n	Initial culture (Mean)	After disinfection (Mean)	P-value
5% peracetic acid	20	1.5×10^8 cells/ml	0.13×10^8 cells/ml	0.016
0.2% chlorhexidine	20	1.5×10^8 cells/ml	0.40×10^8 cells/ml	0.025
3.8% sodium Perborate	20	1.5×10^8 cells/ml	0.66×10^8 cells/ml	0.045

Specimens disinfected with 5% Peracetic acid showed statistically significant difference (P - 0.016) among final counts of *Candida albicans*, after disinfection which was found to be 0.13×10^8 cells/ml. specimens disinfected with 0.2% chlorhexidine gluconate showed statistically significant difference (P - 0.025) among final counts which was found to be 0.40×10^8 cells/ml and remaining specimens disinfected with 3.8% sodium perborate showed no statistically significant difference (P - 0.045) among the final counts which was found to be 0.66×10^8 cells/ml. 5% Peracetic acid and 0.2% chlorhexidine gluconate showed similar effectiveness among the tested disinfectants against *Candida albicans*. 3.8% sodium perborate showed least effect against *Candida albicans* compared with initial growth or cultures, all of the immersion solutions were found to reduce the growth of *Candida albicans*.

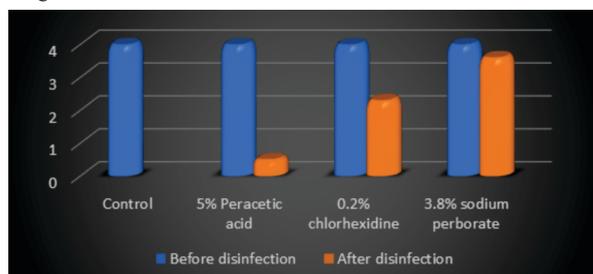


Fig 1: graph determining significant difference between the groups

DISCUSSION:

Oral environment temperature and acquired pellicle formed over dentures promote *Candida* adhesion to resin materials, indicating need of an adequate plaque control for maintaining oral health. Dantas AP et al (1997)⁸ showed that development of yeast on acrylic resins happens in three distinct stages. The initial stage lasts upto 11 hours from colonization when some microcolonies begin to be formed. Intermediate stage from 12-30 hours after colonization when some extracellular material begins to accumulate over colonies.

Maturation stage from 38-72 hours after colonization when *Candida albicans* colonies become totally involved by extracellular matrix forming a biofilm. They also concluded that antifungal resistance increases during biofilm development, as extracellular matrix acts as barrier to action of antifungal agents.

In the literature, Budtz – Jorgensen and Bertram, et al (1970) found that *Candida albicans* were found to be most prevalent of all candidal species both in healthy and diseased oral cavity. Arendof and walker et al (1987) reported *Candida albicans* to be most pathogenic and capable of adhering to epithelial cells and acrylic resin surfaces.

In this study we have determined the efficacy of three commonly used chemical agents: Sodium Perborate, Chlorhexidine and Peracetic acid. When sodium perborate is dissolved in water, they become alkaline hydrogen peroxide, which decomposes when it comes into contact with certain substances and releases small oxygen bubbles with the mechanical action of detaching the biofilm from the denture surface. The oxidant agents help to remove stains and have some antibacterial action. This type of solution can be used alone or in combination with a mechanical method.¹⁹

Sodium hypochlorite has the capacity to dissolve mucins and other organic substances in the biofilm matrix, inhibiting its formation and microorganism recolonization, yet promoting the degradation of the acrylic resin, depending on its concentration and immersion time. 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. 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.^{1,2,9,10}

Chlorhexidine has a broad spectrum of activity against a variety of organisms, including *C. albicans*. It has been pointed out that the resistance of biofilms originates typically from the recalcitrance of a small subpopulation. Chlorhexidine is a cationic agent, which is adsorbed by oral surfaces and negatively charged microbes, interfering with osmotic equilibrium, the formation of acquired films and the microbial adsorption of oral surfaces. Therefore, it has the capacity to reduce biofilm formation and inhibit the synthesis of insoluble polysaccharides in the microbial matrix of the dental biofilm. However, continued use may cause staining and alterations of color in the acrylic.^{1,2,10,13}

A.C.R.D. Salvia et al. (2013) evaluated the antimicrobial effectiveness of 1% sodium hypochlorite, 2% chlorhexidine digluconate and 50% vinegar and suggested that 50% vinegar was as effective as 1% sodium hypochlorite and 2% chlorhexidine digluconate against *C. albicans*, *E. coli* and *S. mutans* for the disinfection of heat-polymerized acrylic resin.¹¹

An in-vitro and in-vivo study reported by A.L.C. Chassot et al. (2006) revealed immersion for at least 5 min in a 0.2% peracetic acid-based disinfectant promoted high-level disinfection of heat polymerized, chemically activated and microwave-polymerized acrylic resins contaminated with either human saliva or *Bacillus subtilis* or *Bacillus stearothermophilus*.¹²

The results of present study demonstrated that peracetic acid showed best antimicrobial effectiveness against tested microorganisms. These data are in accordance with previous studies analyzing disinfection with this solution.

Unlike most chemical disinfectants, peracetic acid-based disinfectants are not inactivated in the presence of organic matter. Furthermore, peracetic acid does not leave residues and does not produce harmful byproducts because its mechanism of action involves release of free oxygen and hydroxyl radicals decomposing in oxygen, water and acetic acid and it acts rapidly against all microorganisms even at low concentrations.^{12,14}

CONCLUSION:

Immersion for at least 10 min in 5% peracetic acid-based disinfectant promoted high level disinfection of chemically activated acrylic resins contaminated with *Candida* sp.

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