**Efficacy of Sodium hypochlorite vs Sodium perborate as Denture Disinfectants**

**- In Vitro Study**

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**Abstract**

**Aim:** To find the efficacy of sodium hypochlorite and sodium perborate as denture disinfecting agents

**Materials and methods:** Autoclaved heat cure acrylic denture strips were immersed 25ml of sterilised artificial saliva and 0.5 McFarland standard Candida albicans suspension. It was incubated for 3 days at 37 Celsius, after that it was cleaned with mineral water and then immersed in 50ml of disinfectant for 6 hours. 2% sodium hypochloriteand 4% sodium perboratewere used as denture cleansing agents, saline and 0.2% chlorhexidine were taken as the negative and positive control respectively.After 6 hours, a swab was taken from the rough surface of the denture base sample and streaked on the SDA plate. It was incubated for 24 hours. Growth pattern of Candida albicans was observed. A broth culture was made to confirm the results obtained in the above study

**Background:** The prevalence of *Candida albicans* in the denture is significantly higher than that in mucosa.Sodium hypochlorite is a reducing agent & Sodium perborate is an oxidizing agent and helps in removing stains and kills the microbes on the denture. Denture disinfectant is used to control growth of microorganisms like *Candida albicans* preventing denture related stomatitis

**Results:** It is concluded that 2% sodium hypochlorite is found to be better denture disinfecting agent compared to 4% sodium perborate.

**Conclusion:** 2% sodium hypochlorite is found to be better denture disinfecting agent compared to 4% sodium perborate

**Keywords:** Denture disinfectants, denture stomatitis, Candida albicans, Sodium hypochlorite, Sodium perborate

**Introduction**

A complete denture is defined as a dental prosthesis, which replaces the entire dentition and associated structures of the maxilla and mandible.[1] A complete denture restores the aesthetic, phonetic and masticatory functions of the individual. A denture placed in the oral environment forms a biofilm on the surfaces of thedenture, which makes it susceptible for infections [2]. Patients who wear dentures present with a variety of symptoms and abnormal intra oral findings [3]. The advancing age of the denture wearer and the nature of the denture bearing mucosa appear to influence the nature of the problem. Superimposed infection with Candida organisms and traumatic lesions are the most commonly encountered abnormalities. Denture stomatitis had been reported in 11-67% of complete denture wearers [4]. Denture bio film is an important factor in the pathogenesis of denture stomatitis. Candida albicans found in the biofilm has been reported as an important agent for the installation and maintenance of denture stomatitis.[5] The prevalence of Candida albicans in the denture is significantly higher than that in mucosa. In healthy individuals it has a prevalence rate of 45-65% with a higher in children and young adults. In denture wearers the prevalence of candida increases to 60-100% due to the decreased flow of oxygen and saliva caused by the denture to the underlying tissue producing a local acidic and anaerobic micro-environment that favours yeast overgrowth. [6]

Candida species are yeasts and within the oral cavity. It is one of the main causative organisms of denture- induced stomatitis which is primarily due its ability to adhere and form biofilms on oral cavity tissues and denture surfaces as well as due to its resistance to anti-fungal agents [7] This biofilm grows extensively on acrylic resin denture material and its effective removal is a significant challenge by both chemical and mechanical methods. Dentures can be cleaned mechanically, chemically or through a combination of both these methods. Mechanical methods comprises of brushing and ultrasonic treatment through the use of ultrasonic cleansers. [8] The ultrasonic cleaning method is limited due to the lack of information and discouraging cost. Brushing is easier, inexpensive and an effective method when used methodically in removing denture biofilm. However, abrasive action could result in the wear of the denture base and relining materials [9]. Another disadvantage of the mechanical methods is among the physically challenged or geriatric denture wearers. Hence the efficient chemical denture cleansers could be an important alternative or adjunctive to mechanical cleansing. Chemical methods include soaking the dentures in commercial (peroxides, acids, mouth washes and enzymes) or household (hypochloride, sodium chloride vinegar) products [10]. These chemicals are easy to use and can easily reach undercuts of the denture base which are otherwise overlooked during denture cleaning. The acrylic resins surface roughness remains unchanged compared to the abrasion due to brushing and the surfaces are less susceptible tobio film accumulation [11].

Sodium perborate is a white, odour-less compound with the chemical formula Na2B2O4(OH)4[12]. It is an oxidizing agent with antimicrobial activity and their chemical effects can remove stains by releasing oxygen [13].Sodium perborate is widely used in dentistry as a bleaching agent for treating discolored teeth [14]. Sodium hypochlorite is a reducing agent with antibacterial and anti fungal activity [15]. Sodium hypochlorite does not affect the surface roughness but the chances of deteriorating the denture base material by corrosion and bleaching is possible [16].Sodium hypochlorite is commonly used as a root canal irrigant in dentistry [17]. Chlorhexidine acts against a wide range of microbes like Gram positive, Gram negative bacteria, dermatophytes, lipolytic viruses, fungi, yeasts. Based on the concentration it can be bacteriostatic or bactericidal [18].

There are many known denture disinfectants such as EDTA, Chlorhexidine, Sodium hypochlorite, Sodium perborate, Povidone iodine, Hydrogen peroxide, etc. In this study we have chosen to study the effectiveness of the very commonly available agents, namely Sodium hypochlorite and Sodium perborate [19, 20].

**Materials and Methods**

The effect of disinfectant was tested by two methods. One was by contamination of denture bases with candida suspension and the second method was by testing the effect of the standardised concentration of disinfectant in a broth.

**Sample fabrication**

A total of 40 heat-polymerized acrylic denture strips were obtained from a wax pattern with a standardised dimension of 5x1 cm. The wax pattern was invested with dental stone (type III gypsum) in a metallic flask. After the setting of dental stone, de-waxing is done by immersing the flask in a water bath at a temperature of 70-80C for about 10 minutes [21]. Heat-polymerized acrylic resin was mixed according to the manufacturer’s recommendation and packed into the mould at the dough stage. The metal flask was then closed and subjected to a short curing cycle at 740C for 2 hours followed by a terminal boiling at 1000C for 1 hour [22]. On completion of curing cycle, the flask was allowed to completely cool before opening and the denture sample was obtained. The denture strips of 5x1cm dimension were checked for any imperfections. The cameo surface of the strips was sandpapered and polished [22]. On completion of processing, the strips were packed and autoclaved.

**Contamination of specimen**

40 heat cured denture acrylic denture strips were selected and sterilized by autoclaving at 15lbs for 30 minutes. These denture strips were immersed in sterilizeduricol containers containing 25ml of sterilized artificial saliva. A Candida albicans suspension was made to the turbidity matching 0.5 Mcfarland standard by immersing for 30 minutes. 100µl of suspension is added to the artificial saliva and well shaken to ensure a good mix. The denture strips in the above suspension was incubated for 3 days at 37after which it was taken out, and cleaned with mineral water and then immersed in 50ml of disinfectant and kept for 6 hours. A subculture was made on Brain Heart Infusion agar and incubated for 24 hours.

**Preparation of disinfectants**

Commercially available oxidising agent 2% sodium hypochloriteand 4% sodium perboratewere used as denture cleansing agents in this study. Saline was taken as the negative control and 0.2% chlorhexidine containing commercially available mouthwash was taken as the positive control. After incubation for 48 hours, the denture samples were washed in drinking water and placed in a sterile container containing denture cleansing agent. 10 denture samples were placed in each denture cleansing agent (sodium hypochloriteand sodium perborate). The denture samples were left in the denture cleansing agent for 6 hours.

**Culture preparation**

After 6 hours, a swab was taken from the rough surface of the denture base sample and streaked on the SDA plate. This process was repeated for all the denture base samples. The SDA plates were incubated for 24 hours. After 24 hours, the growth pattern of Candida albicans was observed.

**Broth culture**

The disinfectant material is taken in a standardized concentration in 5 curettes of 1ml each, the candida suspension was made with turbidity matching 0.5 McFarland standardsand it is taken. 10µl of the suspension is added to disinfectants taken in cuvette. It was allowed to react for 6 hours at room temperature. After the 6 hour period 10µl of this preparation was transferred to saboraud’s dextrose agar and incubated for 12 hours at 37. The test was done along with positive and negative control.

**Experimental and control groups**

Four groups each containing 10 contaminated specimenswere assigned to various disinfectants.

Group 1: Chlorhexidine 0.2% (Positive control)

Group 2: Saline (Negative control)

Group 3: Sodium hypochlorite2%

Group 4: Sodium perborate4%

**Results**

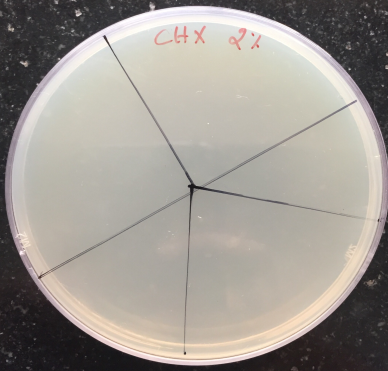
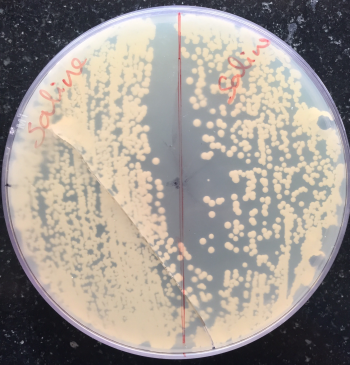
75% of the denture bases did not showany growth of Candida and and 25% of the denture bases developed Candida growth. Out of which, 90% negative results and 10% positive Candida growth was seen on use of 2% Sodium hypochlorite, whereas 40% of the positive results and 60% of negative results on using Sodium perborate. Finally, 100% of the denture bases showed no growth of Candida on using chlorhexidine. This study was designed to evaluate the change in the proportion of positive and negative growth among patient’s usage of chemical agents. When tested against the two controls, it was found that Sodium hypochlorite is better compared to Sodium perborate.

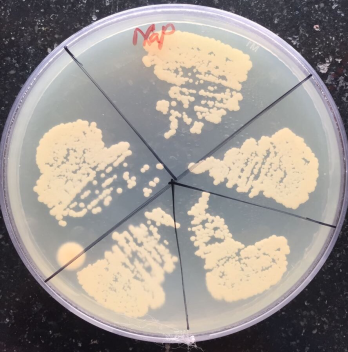
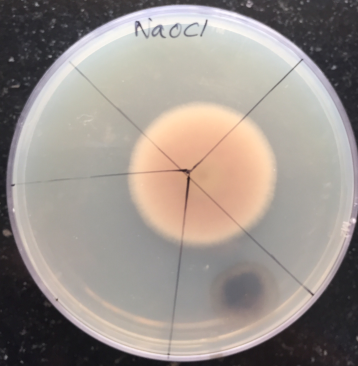
|  |  |  |
| --- | --- | --- |
| Agent | Positive | Negative |
| 2%Sodium hypochlorite | 1 | 9 |
| 4%Sodium perborate | 4 | 6 |
| Saline | 10 | 0 |
| Chlorhexidine | 0 | 10 |

**Table 1: The table below denotes the results of effectiveness of disinfecting agents against denture strips contaminated with Candida suspension.**

**Figure 1: Graphical representation of the results**

**Figure 2 shows Chlorhexidine 0.2% Figure 3 shows Saline**





**Figure 4 shows 2% Sodium hypochlorite Figure 5 shows 4% Sodium perborate**

**Discussion**

Sodium perborate is a white, odourless, economical and versatile water soluble oxidizing agent which releases hydrogen peroxide on reacting with water [23]. Hydrogen peroxide is a broad spectrum antimicrobial agent effective against dormant forms of known high resistance microbes such as bacterial spores, protozoal cysts and infectious proteins such as prions [24]. It degrades the microbes by chemical oxidation of the cellular contents [25].

Sodium hypochlorite is a reducing agent with antimicrobial activity [19]. It is an organic, fat solvent which degrades fatty acids and convert them into salts, alcohol and neutralises amino acids forming water and salt [26].The high pH of sodium hypochlorite interrupts the cytoplasmic membrane integrity with biosynthetic alterations in cellular metabolism, irreversible enzymatic inhibition, phospholipid degradation [27].It also reduces the adhesion of *Candida albicans* to the epithelial cells of oral cavity [19].

From the above results, 2% Sodium hypochlorite is found to be more effective compared to 4% Sodium perborate. These resultsare similar to a study done by Cem et al., where 2% Sodium hypochlorite was found to be better compared to other chemical disinfecting agents [19]. Nikawa and coworkers have concluded a similar result that chemical agents are easier to use and it is more efficient in reducingthe formation of biofilm [15]. Denture wearers maintain the denture hygiene if denture cleansing products are feasible and easily available. The commercially available denture cleansers may not be feasible to the patient and this may lead them to neglect denture hygiene. The two disinfectants used in the study fulfill these criteria as they are cost effective, easily available and they have the ability to effectively disinfect a denture. For best results, mechanical cleansing of the denture using toothbrush or nailbrush should be done prior to chemical disinfection [28].

**Conclusion**

From the above results it is concluded that 2% sodium hypochlorite is found to be better denture disinfecting agent compared to 4% sodium perborate.

**Reference**

[1] Jain AR, Dhanraj M. A Clinical Review of Spacer Design for Conventional Complete Denture. Biology and Medicine. 2016 Jan 1;8(5):1.

[2]Mahtani AA, Nittla PP, Muralidharan, Dhanraj, Comparison on the Efficacy of Chemical and Natural agents on Disinfection of Denture Bases –In Vitro Study, International Journal of Pharmaceutical Sciences Review and Research, 2017July 1; 45(1): 164-171.

[3]Jeganathan S, Lin CC. Denture stomatitis—a review of the aetiology, diagnosis and management. Australian dental journal. 1992 Apr 1;37(2):107-14.

[4] Theilade E, Budtz-Jørgensen E, Theilade J. Predominant cultivable microflora of plaque on removable dentures in patients with healthy oral mucosa. Archives of Oral Biology. 1983 Jan 1;28(8):675-80.

[5] Dorey JL, Blasberg B, MacEntee MI, Conklin RJ. Oral mucosal disorders in denture wearers. Journal of Prosthetic Dentistry. 1985 Feb 1;53(2):210-3.

[6] Mangram AJ, Horan TC, Pearson ML, Silver LC, Jarvis WR. Guideline for prevention of surgical site infection, 1999. American journal of infection control. 1999 Apr 1;27(2):97-134.

[7] Silva MM, Vergani CE, Giampaolo ET, Neppelenbroek KH, Spolidorio DM, Machado AL. Effectiveness of microwave irradiation on the disinfection of complete dentures. International Journal of Prosthodontics. 2006 May 1;19(3).

[8] Sanita PV, Vergani CE, Giampaolo ET, Pavarina AC, Machado AL. Growth of Candida species on complete dentures: effect of microwave disinfection. Mycoses. 2009 Mar 1;52(2):154-60.

[9] Lal K, Santarpia III RP, Pollock JJ, Renner RP. Assessment of antimicrobial treatment of denture stomatitis using an in vivo replica model system: therapeutic efficacy of an oral rinse. The Journal of prosthetic dentistry. 1992 Jan 1;67(1):72-7.

[10] Senna PM, Sotto‐Maior BS, Silva WJ, Del Bel Cury AA. Adding denture cleanser to microwave disinfection regimen to reduce the irradiation time and the exposure of dentures to high temperatures. Gerodontology. 2013 Mar 1;30(1):26-31.

[11] Senna PM, da Silva WJ, Del Bel Cury AA. Denture disinfection by microwave energy: influence of Candida albicans biofilm. Gerodontology. 2012 Jun 1;29(2).

[12] Brotherton BJ. Boron: Inorganic Chemistry. Encyclopedia of Inorganic Chemistry. 1994;1:372.

[13] Oliveira Paranhos HF, Silva‐Lovato CH, De Souza RF, Cruz PC, Freitas‐Pontes D, Karina M, Watanabe E, Ito IY. Effect of three methods for cleaning dentures on biofilms formed in vitro on acrylic resin. Journal of Prosthodontics. 2009 Jul 1;18(5):427-31.

[14] Jiménez-Rubio A, Segura JJ. The effect of the bleaching agent sodium perborate on macrophage adhesion in vitro: implications in external cervical root resorption. Journal of endodontics. 1998 Apr 1;24(4):229-32.

[15] Nikawa H, Hamada T, Yamashiro H, Kumagai H. A review of in vitro and in vivo methods to evaluate the efficacy of denture cleansers. International Journal of Prosthodontics. 1999 Mar 1;12(2).

[16] Harrison A, Jagger DC. An in vitro investigation of the abrasive qualities of a selection of denture-cleaning pastes on poly (methyl methacrylate) denture base material. Primary dental care: journal of the Faculty of General Dental Practitioners (UK). 1997 Jan;4(1):21-4.

[17] Hülsmann M, Hahn W. Complications during root canal irrigation–literature review and case reports. International Endodontic Journal. 2000 May 1;33(3):186-93.

[18] Balagopal S, Arjunkumar R. Chlorhexidine: the gold standard antiplaque agent. J Pharm Sci Res. 2013;5(12):270-4.

[19] Sahin C, Ayyildiz S, Ergin A, Uzun G. Effect of chemical denture cleansers on microorganisms over heat-polymerized acrylic resin. Afr J Dent. 2013;1(2):6-9.

[20] Shay K. Denture hygiene: a review and update. The journal of contemporary dental practice. 2000 Dec 25;1(2):1-8.

[21] Taylor PR. An illustrated history of lost wax casting. InProceedings of the 17th Annual BICTA Conference, Washington, DC 1983 Sep.

[22] Yau WF, Cheng YY, Clark RK, Chow TW. Pressure and temperature changes in heat-cured acrylic resin during processing. Dental Materials. 2002 Dec 1;18(8):622-9.

[23] McKillop A, Sanderson WR. Sodium perborate and sodium percarbonate: cheap, safe and versatile oxidising agents for organic synthesis. Tetrahedron. 1995 May 29;51(22):6145-66.

[24] Russell AD. Principles of antimicrobial activity. Disinfection, sterilization and preservation. 1991:29-58.

[25] Finnegan M, Linley E, Denyer SP, McDonnell G, Simons C, Maillard JY. Mode of action of hydrogen peroxide and other oxidizing agents: differences between liquid and gas forms. Journal of Antimicrobial Chemotherapy. 2010 Aug 16;65(10):2108-15.

[26] Spanó JC, Barbin EL, Santos TC, Guimarães LF, Pécora JD. Solvent action of sodium hypochlorite on bovine pulp and physico-chemical properties of resulting liquid. Braz Dent J. 2001 Jul;12(3):154-7.

[27] Estrela C, Estrela CR, Barbin EL, Spanó JC, Marchesan MA, Pécora JD. Mechanism of action of sodium hypochlorite. Brazilian dental journal. 2002;13(2):113-7.

[28] Jagger DC, Harrison A. Denture cleansing--the best approach. British dental journal. 1995 Jun 10;178(11):413.