

The Effect of Three Different Disinfectant Materials on Polyether Impressions by Spray Method

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ABSTRACT

Background: Dentists, dental equipments and dental laboratories are exposed to different types of pathogenic microorganisms. The aim of this study is to investigate the effect of three different types of disinfectant agents: Sodium hypochlorite 0.525%, Epimax and Deconex, on polyether impressions after 5 and 10 minutes.

Materials and methods: In this *in vitro* experimental study, 66 circular samples of polyether impression material of 1 cm diameter and 2 mm thickness were contaminated with *Staphylococcus aureus* (ATCC29213) *Pseudomonas aeruginosa* (ATCC27853) and *Candida albicans* fungus (PTCC5027). Except for control samples, all of them were disinfected with sodium hypochlorite 0.525, Deconex and Epimax by way of spraying. Afterward, they kept in plastic bags with humid rolled cotton for 5 and 10 minutes. In order to isolate bacteria, the samples were immersed in 2% trypsin for 1 hour and then the solution was diluted with normal saline in portion of 1, 1/2, and 1/4. The trypsin suspensions were transferred to culture plates and the number of colonies was counted after 24 and 48 hours for bacteria and after 72 hours for fungus. For data analysis Mann-Whitney statistical test was used ($\alpha = 0.05$).

Results: Epimax and sodium hypochlorite thoroughly eliminated *Candida albicans* as time elapsed from 5 to 10 minutes; however, the other two microorganisms were not eliminated completely. Deconex was completely efficient for all microorganisms when immersion time was enhanced from 5 to 10 minutes. There was a significant difference between efficacy of Deconex-sodium hypochlorite and sodium hypochlorite-Epimax for *Pseudomonas aeruginosa* in 5-minute immersion (p-value:0/046) Moreover, Deconex and sodium hypochlorite were significantly different for *Staphylococcus aureus* (p-value:0/046) and *Pseudomonas aeruginosa* in 10-minute immersion (p < 0.05).

Conclusion: According to the results of this study, none of the three disinfection materials could eradicate three different kinds of microorganisms in 5 minutes, but in 10 minutes, Deconex could completely eradicate all microorganisms compared to other disinfection agents, which is a good indicator for high efficacy of this agent in disinfecting polyether impressions.

Keywords: Disinfectant agents, Impression materials, Polyether, Spray.

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INTRODUCTION

Dentists, dental materials and dental laboratories are exposed to different kinds of pathogenic microorganisms. The main sources of cross-infection between patients and dentists are the impression materials, impression trays and poured stone casts.¹

New researches have shown that 67% of materials sent to dental laboratories are infected by various microorganisms.² The most identified microorganisms are *Streptococcus species*, *Staphylococcus species*, *Escherichia coli species*, *Actinomyces species*, *Antitratrus species*, *Pseudomonas species*, *Enterobacter species*, *Klebsiella pneumoniae* and *Candida species*.³ Taking this into account, efforts should be made to eliminate most of these microorganisms and reduce the rate of infection transmission in dental laboratories. The International Dentistry Federation consequently insists on disinfecting all impressions taken from patients before sending them to laboratories.⁴ Also the American Dental Association (ADA) has advised all dental workers to disinfect patients' impression trays.⁵ In some studies, it has been declared that washing the impression materials with tap water only removes 40% of bacteria, even though some studies indicated that it has the capacity to reduce 90% of microorganisms.⁶ The most common chemical disinfectants which are used by dentists are alcohols, aldehydes, chlorine combination, phenols, biguanides, iodide combinations and ammonium.⁷ Based on the type of chemical disinfectant, there are two common methods to disinfect dental materials: (1) Immersion, (2) spraying.⁶ disinfecting by immersing in chemical materials has proved to cover all surfaces of impression in one time⁸ while spraying is not capable of disinfecting all surfaces effectively and also cannot cover all undercuts. But contrarily to soaking, it significantly reduces the amount of shrinkage and impression distortion.⁶ Some impression materials, such as alginate, which are common in dentistry,⁹ absorb water and distort by immersing in disinfectant solutions.¹⁰

In the study by Westerholm et al in 1992,¹¹ the efficacy of eight different disinfectant agents was assessed; among them Sporidicin and 0.525% sodium hypochlorite were able to eliminate 99.99% of *Staphylococcus aureus*.

In another scientific research, Rueggeberg et al found that spraying disinfectant agents on the surface of alginate

cannot cause any dimensional changes in poured stone casts when compared with casts from water-rinsed controls. Disinfection by immersing method caused dimensional distortion in both anterior and posterior segments. Both spraying and immersion methods decreased surface details to the same extent. The antimicrobial effect of spraying was similar to the immersion method, while mere water rinsing did not show any significant disinfection effect.¹²

Ghahremanlo et al investigated the antimicrobial effect of 0.525%, sodium hypochlorite, Deconex and Sanosil. It was concluded that the use of 0.525% sodium hypochlorite sprayed on the surface of alginate, effectively disinfected 96.6% of the samples.⁹

As none of the methods and materials above has been accepted as a standard gold for disinfecting dental materials and impressions, finding an appropriate method seems rational. So the aim of this study is to investigate the disinfection effect of Deconex Solarcept solution, 0.525% sodium hypochlorite and Epimax on polyether impressions in 5 and 10 minutes.

MATERIALS AND METHODS

This randomized experimental study was carried out with the cooperation of dentistry faculty and the Department of Microbiology of the Medical School, aiming at evaluating the disinfection effect of 0.525% sodium hypochlorite (Chloran, Tehran, Iran), Deconex (Borer chemie, Switzerland) and Epimax (Emad, Isfahan, Iran) on the polyether (Impregum, 3M ESPE AG Co. St Paul, MN) impression material.

Sampling Methods

An appropriate mixture of water and powder of impression material was prepared in a sterile bowl with a sterile spatula according to the instructions of the manufacturer company.

Then the mixture was poured into a 5-cc sterile syringe; after some time for material setting, the impression material was cut-off and removed with a no. 10 surgical blade from the end part of the syringe in 2 mm thick slices. Eventually, 66 samples with 2 mm thickness were prepared. In order to ensure that samples were kept sterile during preparation, three samples were selected as negative controls (blank) and were incubated on TSB culture for 24 to 48 hours; after which the bacterial growth was examined. For each bacterial type, 21 samples were used. Sodium hypochlorite 0.525% was used to disinfect three of them for 5 minutes and three others for 10 minutes. Three samples were used to be disinfected with Deconex for 5 minutes and three others for 10 minutes, and three samples for disinfecting with Epimax for 5 minutes and three others for 10 minutes. Last,

three more samples were used as positive controls to check any microbial contaminations.

Preparation of Bacterial Suspension and Yeast

For many type of susceptibility testing, standard inoculums of bacteria must be used. The standard inoculums were prepared according to 0.5 McFarland (1.5×10^8 cfu/ml) by transferring 1 to 2 colonies of 18 to 24 hours cultures to TSB medium and incubate at 35°C until 0.5 McFarland turbidity was gained. For *Candida albicans* fungus, the sample was taken from 48 hours Dextrose agar cultures.

Contamination of Samples

To evaluate the disinfection effect of abovementioned three substances, samples were separately contaminated with microbial solutions of *Staphylococcus aureus* (ATCC29213) *Pseudomonas aeruginosa* (ATCC27853) and *Candida albicans* fungus (PTCC5027). The impressions were put in sterile test tubes separately with 1 cc of microbial suspension and then incubated at 35°C for 1 hour.

Disinfection of Samples and Microbiological Surveys

After contamination, all samples were rinsed with sterile distilled water for 30 seconds. In order to disinfect all samples, except controls, sodium hypochlorite, Deconex and Epimax were used on each sample, applying spraying method, in 10 puffs in 15 seconds. Then the samples were put into sterile plastic bags containing sterile cotton humidified with sterile distilled water for 5 and 10 minutes.

Trypsin protease, which is able to isolate the microbes from contaminated environments, was used. The time and concentration for the effective use of trypsin is 60 minutes and 2% respectively. This time and concentration are based on the maximum microorganisms which can be isolated from the samples. After washing the samples with sterile distilled water for 30 seconds, they were put in 2% trypsin solution for 60 minutes. The suspensions of ½ and ¼ trypsin solution were then prepared. Using 100 microliter samplers, these samples were transferred to Muller Hinton agar for the *Pseudomonas aeruginosa* and *Staphylococcus aureus*. Saborow Dextrose agar (SDA) medium was selected for *Candida albicans* fungus. Using a Pasteur pipet bent with heat at 90 degrees, the samples were spread on cultures. After 24 and 48 hours incubation, the grown bacterial colonies on cultures were counted. The grown fungus colonies of *Candida albicans* on SDA were counted after 72 hours. SPSS software was used for data analysis; edition 11.5 and statistical Mann-Whitney testing was used.

RESULTS

The difference between Deconex-sodium hypochlorite and sodium hypochlorite-Epimax regarding their capability in eliminating *Pseudomonas aeruginosa* after 5 minutes was significant ($p = 0.046$). Moreover, Deconex and sodium hypochlorite represented a significant difference in eliminating *Staphylococcus aureus* ($p = 0.046$) and *Pseudomonas aeruginosa* after 10 minutes ($p = 0.05$) (Tables 1 and 2).

Deconex, Epimax and sodium hypochlorite were able to thoroughly eliminate *Candida albicans* as time elapsed from 5 to 10 minutes; however, other two microorganisms were not eliminated completely. Deconex was completely efficient in eradicating all microorganisms when the time, samples were kept in plastic bag was enhanced from 5 to 10 minutes (Table 3). However, the efficacy of all types of disinfectant agents was increased as time elapsed.

Table 1: Comparison of disinfectant and control agents in 5 minutes and 1 dilution

Bacteria	<i>Candida albicans</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
Disinfectants	p-value	p-value	p-value
Deconex-control	0/046	0/046	0/046
Hypochlorite sodium control	0/043	0/046	0/050
Epimax-control	0/043	0/050	0/046
Deconex-hypochlorite sodium	0/072	0/814	0/046
Deconex-Epimax	0/346	0/637	0/197
Hypochlorite sodium-Epimax	0/099	0/637	0/046

DISCUSSION

Dentists practicing dentistry encounter potentially harmful microorganisms. Patients are the most common source of microorganisms.¹³ Studies indicate that the surface of impressions taken out of the mouth is contaminated with bacteria.¹⁴⁻¹⁷ As impressions and occlusal records cannot be sterilized by heat, chemical disinfection is still the most common practicable method to eradicate microorganisms.¹⁸⁻²⁰ So far there is no global way to disinfect impression materials.²¹ The American Dental Association (ADA) recommends to soak impressions in disinfectant solutions for less than 30 minute.²² Muller Bolla et al found that in European Schools of Dentistry, the soaking method is applied for 63% of alginate impressions and in 73% of silicon impressions. The approximate time of disinfection is 10.3 ± 6.3 minutes. Half of the dentistry schools in the study did not disinfect alginate impressions.¹⁹

But, Hiroshi Egusa et al in 2008 showed that alginate impressions from patients' mouths contain hazardous microorganisms and organisms like streptococci, *Staphylococcus aureus*, methicillin resistant *Staphylococcus*, *Candida*, *Pseudomonas aeruginosa* with rate of 100, 55.6, 25.9, 25.9 and 5.6% respectively.²¹ These are opportunist pathogens that spread and transfer through the oral cavity.²¹ *Candida* causes common opportunistic infections known as oral candidiasis found in patients with immune deficiency.²² *Pseudomonas aeruginosa* is an infectious agent that exists in hospital appliances and instruments.²¹ However, studies show that among the common population, the spreading rate of *S. aureus* to the nasopharynx is only 10%.¹³ This is

Table 2: Comparison of disinfectant and control agents in 10 minutes and 1 dilution

Bacteria	<i>Candida albicans</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
Disinfectant	p-value	p-value	p-value
Deconex-control	0/034	0/05	0/037
Hypochlorite sodium-control	0/034	0/046	0/050
Epimax-control	0/034	0/046	0/046
Deconex-hypochlorite sodium	1/000	0/046	0/037
Deconex-Epimax	1/000	0/046	0/034
Hypochlorite sodium-Epimax	1/000	1/000	0/105

Table 3: Bacterial growth prevention percentage by different disinfectant agents in 5 and 10 minutes and 1 dilution

Bacteria	<i>Candida albicans</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>	
Disinfectant	Suspension time (min)	1	1	
Hypochlorite sodium 0.525%	5	897.82%	94.11%	95%
	10	100%	96.07%	96.42%
Epimax	5	92.38%	93.72%	95.95%
	10	100%	96.07%	97.61%
Deconex	5	90.21%	94.50%	98.80%
	10	100%	98.82%	100%

a good reason that in the present survey, *Staphylococcus aureus*, *Candida albicans* and *Pseudomonas aeruginosa* were selected to assess the disinfection ability of disinfectant agents.

By the year 1991, washing the impression materials with running water was the common way to remove microorganisms.²¹ This method could reduce about 90% of bacteria.²³ Running water can wash up saliva, blood and debris. But recent studies indicate that such methods cannot eliminate microorganisms from impression materials completely. Therefore washing the impression materials with running water, without disinfection, is not sufficient.²¹

In the present study, 0.525% hypochlorite sodium was used. This agent is also used in housework. This disinfectant agent could efficiently prevent microorganism's growth and disinfect the impression materials.

Westerholm,¹¹ Reuggeberg et al¹² also showed that spraying sodium hypochlorite can effectively disinfect the impression materials. The Westerholm et al showed that sodium hypochlorite could absolutely (99.99%) prevent the growth of *S. aureus*.¹¹ In Ghahremanloo et al study, spraying sodium hypochlorite could disinfect samples effectively (96.6%) in 10 minutes.⁹

Also in this study, 0.525% hypochlorite sodium spray effectively eradicated three types of microorganism and showed its highest potential against *Candida albicans* after 10 minutes (100% eradication). The results of the present study were in agreement with all mentioned studies, and this indicates that despite different kinds of impression materials, the efficacy of this disinfectant agent is almost equal in eradicating different kinds of microorganism.

Deconex is an alcohol based disinfectant agent, which in our study could impressively eradicate microorganisms. The efficacy of this agent enhanced as time elapsed, as it was not capable in eradicating all microorganisms in 5 minutes but it completely eradicated two kinds of microorganisms (*Candida albicans* and *Pseudomonas aeruginosa*) effectively. In the study by Ghahremanloo et al this agent could eradicate only 70.4% of samples.⁹ The main reason of this difference is probably the use of more resistant type of bacteria.

In this survey, for the first time the antimicrobial features of Epimax on impression materials were investigated. The effect of this agent was also satisfactory and could compete with other common disinfectant agents, such as Deconex and hypochlorite sodium in eradicating infectious microorganisms. Like other disinfectant agents in this study, the efficacy of this agent increased as time elapsed and this agent showed its highest disinfectant capability against *Candida albicans* in 10 minutes as it completely eradicated it.

However, it should be emphasized that the results of the present study are not comparable with the results of other studies, because of the different types of impression materials and different application methods of disinfectant agents in various studies.

One of the disadvantages of the present research is that it was an *in vitro* experimental study, which is different from clinical and situations. Usually, impression materials remain 3 to 5 minutes in patients' mouth, while in our study it took 60 minutes to attach all bacterial types to the samples. Also, pressure which is applied during impression procedure and saliva could alter bacterial adherence capacity. This survey investigated the effect of three common disinfectant agents on two types of bacteria and one fungus. As so many dentists are concerned about viruses, such as HIV and HBV, further studies should be conducted to find an effective way to eradicate these kinds of pathogens.

CONCLUSION

According to the results of this study, none of the three disinfectant materials could completely eradicate three different kinds of microorganisms in 5 minutes, but in 10 minutes, Deconex could completely eradicate all microorganisms compared to other disinfection agents, which is a good indicator for its high capacity in disinfecting polyether impressions.

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