

Investigating the Effects of Spraying Deconex on Disinfection of three Different Impression Materials

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KEY WORDS

Disinfection;
Impression Materials;
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ABSTRACT

Statement of Problem: Dentists and dental laboratory technicians' awareness of the dangers of cross-contaminations has made them seek the best ways to control and eradicate the main sources of infections.

Purpose: The aim of this study was to evaluate the effects of spraying Deconex on three different impression materials.

Materials and Method: In this in vitro experimental study, 30 circular samples of different impression materials, such as alginate, silicone and polyether impression materials were separately contaminated with *Staphylococcus aureus* (ATCC29213), *Pseudomonas aeruginosa* (ATCC27853), and *Candida albicans* fungus (PTCC5027). Except for the control samples, all the other samples were disinfected with Deconex through spraying. Then, they were kept in plastic bags which were stuffed with humid rolled cotton for 5 and 10 minutes. In order to isolate bacteria, the samples were immersed in 2% trypsin for one hour and then the solution was diluted with normal saline in proportion 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. Data analysis was done through running Mann-Whitney Test ($\alpha = 0.05$).

Results: There was a significant difference between the effect of Deconex on silicone and its effect on polyether for all the mentioned microorganisms after 5 minutes ($p < 0.05$). The highest percentage of bacterial growth prevention was recorded for silicone impression material. Deconex completely eradicated the three kinds of microorganisms after 5 and 10 minutes in the silicon group. Deconex was not capable to eliminate all three microorganisms in the polyether and the alginate groups; with the exception of *Pseudomonas aeruginosa* after 10 minutes.

Conclusion: The results of the present study indicated that Deconex has the highest capacity when it is used with silicone; it eradicates all microorganisms in both 5 and 10 minutes.

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Introduction

Dentists and dental laboratory technicians' awareness of the risk and probability of contaminations around them has forced them to look for the best way to control and eradicate the main sources of cross-contaminations. In the field of prosthodontics, disinfection should be done in all the procedures but it is especially significant in

two steps: 1) disinfection of impressions 2) disinfection of prostheses which are sent from the dental laboratory [1].

Impressions are in contact with oral tissues and saliva, thus they act as a reservoir for many hazardous microorganisms [2]. New researches have shown that 67% of materials which are sent to dental laboratories

are infected by various microorganisms [3]. The most frequently identified microorganisms are Streptococcus species, Staphylococcus species, Escherichia coli species, Actinomyces species, Antitratrus species, Pseudomonas species, Enterobacter species, Klebsiella pneumonia, and Candida species [4]. The most common chemical disinfectants which are used by dentists are Alcohols, Aldehydes, Chlorine combinations, Phenols, Biguanides, Iodide combinations and Ammonium [5]. Efficacy of disinfectants is highly dependent on the type of microorganism [6], disinfectant concentration, contact time, turbidity and the temperature [7]. As a result, in order to maximize the efficacy of these materials, it is necessary to recognize all these parameters [8].

Quaternary Ammonium Compounds (QACs) are widely used as disinfectants and can be a good alternative for other disinfectants. All QACs are cationic compounds that penetrate into the bacterial cell wall and lyse them [7]. American Dental Association (ADA) has announced that an acceptable disinfectant must eradicate the vegetative forms of hazardous pathogens, such as Mycobacterium-Tuberculosis (MT) in 30 minutes [9]. ADA claimed that Quaternary Ammonium Compounds (QACs) are not suitable to disinfect surfaces. Best et al. [10] surveyed the efficacy of QACs and declared that old-generation QACs are not effective enough in disinfecting both MT and environmental surfaces.

In 1990, new generations of QACs (Uniseptic Quick and Deconex Solarsept) were introduced and they were approved by many associations, such as AOAC (Association of Official Agriculture Chemists, USA) and BSI (British Standards Institute) [9]. Based on the laboratory test results, the recommended dosage of deconex is 0.25% and its spectrum of activity has been identified as: Bactericidal - fungicidal - tuberculocidal - effective against enveloped viruses including HBV and influenza virus as well as against rotavirus and FCV [11].

In the study done by Ezoddini et al., it was concluded that the use of deconex on radiographic equipment is more effective than the use of Micro10 and Alprocid [12].

Previous studies on new generations of QACs were conducted on environmental surfaces. Considering the capacity and the high efficacy of Deconex, the aim

of this study was to determine the efficacy of this disinfectant on different impression materials.

Materials and Method

This randomized experimental study was carried out with the cooperation of the School of Dentistry and Department of Microbiology of Isfahan University of Medical Sciences. This study was planned to evaluate the disinfection effects of Deconex (Borer Chemie, Switzerland) on alginate (Zhermak, Roma, Italy), polyether (Impregum, 3M ESPE AG Co. St. Paul, MN) and condensational silicon (Spidex, Coltene AG, Altstatten, Switzerland) impression materials.

Three impression materials were separately prepared according to their manufacturers' instructions.

The prepared alginate and polyether were drawn into a 5-CC sterile syringe (Atlas, Tehran, Iran) after a pause for material setting based on the manufacturers' recommendations for each material. Alginate and polyether impression materials were then cut off and removed with a No. 10 surgical blade (Ambala, Haryan, India) from the end part of the syringe in 2-mm thick slices. With regard to silicon, the heavy body impression material (putty) was mixed with the catalyst according to the manufacturer's instructions. The mixture was drawn into a syringe, one centimeter in diameter, and samples with 1.5 millimeter thickness were accurately measured by a digital caliper. Then the light body impression material (Wash) and the catalyst were mixed on a paper pad with a sterile spatula (Santam, Tehran, Iran) and were transferred into the upper 0.5 millimeter of the syringe.

30 samples were provided from each type of impression materials. Three of the samples were considered as negative controls to assure that samples were not contaminated and then they were incubated in TSB culture for 24 to 48 hours; the time after which the bacterial growth was examined. Another 27 samples were divided into three groups for each impression material and based on different types of microorganisms (Staphylococcus aureus, Pseudomonas aeruginosa, and Candida albicans). In each group, three samples were disinfected with Deconex for 5 minutes, three for 10 minutes and the remaining three were regarded as positive controls.

Preparation of bacterial suspension and yeast

For any types of susceptibility testing, a standard inoculum of bacteria must be employed. The standard inoculums were prepared according to 0.5 McFarland (a reference to adjust the turbidity of bacterial suspensions so that the number of bacteria will be within a given range) (1.5×10^8 cfu/ml) by transferring 1-2 colonies of 18-24 hour cultures to Tryptic Soy Broth (TSB) culture and incubating them at 35°C until the turbidity; the cloudiness or haziness of a fluid caused by individual particles (suspended solids) which are generally invisible to the naked eye, of media was equal to 0.5 McFarland. For *Candida albicans* fungus, the sample was taken from 48 hour Sabouraud and Dextrose Agar cultures.

Contamination of Samples

Samples were separately polluted with microbial solutions of *Staphylococcus aureus* (ATCC2-9213), *Pseudomonas aeruginosa* (ATCC27853) and, *Candida albicans* fungus (PTCC5027). The impressions were separately put in sterile test tubes with 1 cc of microbial suspension and then they were incubated at 35°C for one hour.

Disinfection of Samples and microbiological Surveys

After contamination, all the samples were rinsed with sterile distilled water for 30 seconds. In order to disinfect all the samples, except the controls, Deconex was sprayed over samples with 10 puffs in 15 seconds. Then the samples were put into sterile plastic bags stuffed with sterile cotton, humidified with sterile distilled water to form a moisturized environment for 5 and 10 minutes.

Trypsin protease, which has the capacity to isolate the microbes from contaminated environments, was used. The implementation time and effecting concentration of Trypsin is 60 minutes and 2%, respectively. Within this time and concentration, the maximum number of microorganisms can be isolated from the samples. This enzyme separates the microorganisms which are adhered to impression materials through destroying adherent proteins. After washing the samples with sterile distilled water for 30 seconds, they were put in 2% Trypsin solution for 60 minutes. The suspensions of 1/2 and 1/4 Trypsin solution

were then prepared. Using 100 micro liter samplers, these samples were transferred to Muller Hinton Agar for the bacteria *Pseudomonas aeruginosa* and *Staphylococcus aureus*. Sabouraud Dextrose Agar medium was selected for *Candida albicans* fungus. Using a Pasteur pipet bent with heat of 90°C, the samples were spread on the cultures. After 24 and 48 hours of incubation, the grown bacterial colonies on the culture were counted. The grown fungus colonies of *Candida albicans* on SDA were counted after 72 hours of incubation.

Then the data were submitted to SPSS software (version 11.5) and 1 Mann Whitney test was run to analyze the data ($\alpha = 0.05$).

Results

As it can be seen from tables 1 and 2, there is a significant difference between the effect of Deconex on silicon and its effect on polyether for all the mentioned microorganism during 5 minutes ($p < 0.05$). The difference was meaningful only in 10 minutes and for *Candida albicans*. With regard to the efficacy of Deconex, a Significant difference was observed between alginate and silicone in 5 and 10 minutes and for the entire mentioned microorganism ($p < 0.05$) However, this difference was not significant for *Candida albicans* in 5 minutes ($p > 0.05$). The effect of Deconex was significantly different between alginate and polyether for *Staphylococcus aureus* in 5 minutes and for *Pseudomonas aeruginosa* in 10 minutes ($p < 0.05$).

As it is shown in table 3, the highest percentage of bacterial growth prevention, has been recorded for silicon impression material. Deconex entirely eradicated all three kinds of microorganisms after 5 and 10 minutes in the silicon group. Deconex was not capable of eliminating all these three microorganisms in the polyether and alginate groups with an exception to the *Pseudomonas aeruginosa* in 10 minutes when there are numerous bacterial colonies that are not countable, the number of colonies can be calculated by diluting the origin solution to 1/2 and 1/4 proportions. Since the real number of colonies was countable in the original solution (dilution#1) and all the results from other dilutions were similar to the aforementioned results, the number of colonies in the other two dilutions have not been reported.

Table 1 The comparative evaluation of deconex effects on different impression materials for three types of microorganism after 5 minutes

Impression material	Bacterial type	Candida albicans	Staphylococcus aureus	Pseudomonas aeruginosa
		P.value	P.value	P.value
Alginate group- Control group		0.050	0.046	0.043
Silicon group- Control Group		0.037	0.037	0.037
Polyether Group- Control Group		0.050	0.046	0.046
Alginate Group- Silicone Group		0.121	0.034	0.034
Alginate Group- Polyether Group		0.658	0.043	0.099
Silicon Group- Polyether Group		0.037	0.034	0.034

Discussion

During their dental practice, dentists and dental staff are exposed to a large number of microorganisms which are potentially harmful. It has been suggested that the main source of contaminations is the patients' saliva [13]. According to Miller's study, a saliva droplet contains more than 50,000 bacteria [14-15]. Unfortunately, these pathogens can be easily spread thorough impressions sent to the laboratories. As impressions and occlusal records cannot be sterilized by heat, chemical disinfection is still the common practical method to eradicate microorganisms [16-18]. So far, there has been no global way to disinfect impression materials [2]. The American Dental Association (ADA) recommends that impression materials be immersed in disinfectant solutions for less than 30 minutes [19]. Because this area of research is of great importance, the aim of this study was to evaluate the effects of spraying Deconex on three different impression materials.

Based on the findings of the study done by Egusa et al. in 2008, impressions from patients' mouths contain hazardous microorganisms like Streptococci, Staphylococcus aureus, Methicillin resistant Staphylococcus, Candida, Pseudo-moans aeruginosa with the percentage rate of 100%, 55.6%, 25%, 9%, 5.6 % respectively [2]. The main reason for the selection of Candida albicans, Pseudomonas aeruginosa, and Staphylococcus aureus in the current study was that these microorganisms are common opportunists which can be easily spread through the population [2, 13, 19].

Deconex is a new generation QACs, which in the current study, effectively disinfected silicon in both 5 and 10 minutes but it was not effective in disinfecting alginate and polyether. Ghahramanloo et al. claimed that this agent could eradicate 70.4 % of microorganisms [20]. Maybe the main reason for the difference observed in Ghahramanloo et al study and the present study, with regard to Deconex effectiveness, is that they used only irreversible hydrochloride which is hydrophilic; and tends to be dissolved in water and seems to be able to attract water out of the air. This was in contrast with the hydrophobic characteristic; the physical property of a molecule that is repelled from a mass of water, such as silicon. This was observed in Ghahramanloo et al study, which produced more porosity and in turn caused deep penetration of microorganism into the impression material. Also by being hydrophilic, there is a great tendency for the impression material to attract and absorb the disinfectant agents. These differences explain limitations in capacity of a disinfectant agent in eradicating microorganisms.

In the study by Hoseini et al. [9] it was concluded that Pseudomonas aeruginosa and Staphylococcus aureus exist on contaminated handpieces which is in accordance with the results of our study.

Ghahremanloo et al. [20] and yilmaz et al. [21] both observed the higher efficiency of sodium hypochlorite, compared to Deconex, in disinfecting alginate impressions which were contaminated with the same microorganism as in the present study [22]. As the

Table 2 The comparative evaluation of deconex effect on different impression materials for three types of microorganism after 10 minutes

Impression material	Bacterial type	Candida albicans	Staphylococcus aureus	Pseudomonas aeruginosa
		P value	P value	P value
Alginate Group. Control Group		0.046	0.050	0.043
Silicon Group- Control Group		0.050	0.037	0.037
Polyether Group- Control Group		0.037	0.050	0.037
Alginate Group- Silicone Group		0.046	0.037	0.034
Alginate Group- Polyether Group		0.317	0.077	0.034
Silicon Group- Polyether Group		0.037	0.121	1.000

Table 3 Bacterial growth Prevention percentage by deconex after 5 and 10 minutes

Impression material dilution	Time (min)	Candida albicans (%)	Staphylococcus aureus	pseudomonas aeruginosa
		1	1	1
Silicone	5	100	100	100
	10	100	100	100
polyether	5	21.90	50.94	80.98
	10	100	98.82	100
Alginate	5	91.40	95.39	99.27
	10	99.21	96.83	100

findings of the current study indicate, Deconex can completely eradicate microorganisms just in the silicone group; therefore, the researchers should be cautious about using Deconex with alginate and polyether.

One of the disadvantages of this research is that it was an in-vitro experimental study which is obviously different from clinical and in-vivo studies. Usually, impression materials remain for 3 to 5 minutes in the patient's mouth, while in the present study it took 60 minutes in order to attach all the bacterial types to the samples because 60 minutes is an effective time for bacterial adherence. The pressure which is exerted while taking an impression and saliva can also alter the bacterial adherence capability.

Conclusion

Based on the findings of this study, there was a complete eradication of all microorganisms from the silicon impression by the use of Deconex disinfectant material after 5 minutes. It was also observed that after 10 minutes, the number of all microorganisms was significantly decreased on the silicon surface. It can be concluded that Deconex is mostly effective in silicon surface since all the microorganisms were eradicated from silicon surface in both 5 and 10 minutes. The present study revealed that *Pseudomonas aeruginosa* is the most susceptible microorganism to Deconex

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