Advanced Materials Research *Online: 2014-02-27*
ISSN: 1662-8985, Vol. 898, pp 292-295
doi:10.4028/www.scientific.net/AMR.898.292

Type IV dental stone incorporated with antimicrobial agents and its physical properties

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Keywords: dental stone, antimicrobial, setting time, strength

Abstract. Contamination of dental casts can occur via direct contact with impression materials that are contaminated by patients' fluids. Thus, the development of dental stone with antimicrobial activity to reduce cross-contamination between patients and laboratory personnel is needed. This study aims to evaluate the influence of incorporation of disinfectants into type IV dental stone on its antimicrobial activities and physical properties such as setting time and dry compressive strength. Type IV dental stone incorporated with 4 types of disinfectants; Diamond Rock D (3-iodo-2-propynylbutylcarbamate), Diamond Rock B (zeolite), Diamond Rock Z (thiabendazole) and Diamond Rock T (2-benzimidazole carbamic acid), were tested in this study compared with the control, Diamond Rock O (type IV dental stone without disinfectant). Microorganisms tested were *Staphylococcus aureus* ATCC 6538, *Pseudomonas aeruginosa* ATCC 27853 and *Candida albicans* ATCC 1023. One hundred μL of each microbial suspension (10^8 CFU/mL) was dropped on hydrocolloid impression (Jeltrate; Caulk/Dentsply, Milford, DE) and left dry. All types of stone mixes were prepared and poured into the impressions and allowed to set for 60 min. Then the stone samples were removed and the microbial contact surfaces were imprinted on Brain Heart Infusion agar plates. After incubation, colonies appeared on agar were gram-stained and counted. The setting time and dry compressive strength were tested in accordance with International Standard (ISO) 6873: 1998 (E). Dental stone incorporated with 4 types of disinfectants showed antimicrobial activity against all tested microorganisms with the percentage of microbial reduction ranging from 83% to 100%. Among all types of dental stone, no significant difference in setting time was observed. In dry compressive strength testing, the disinfectant-containing dental stone had significant higher strength than that without the disinfectant. The newly developed type IV dental stone incorporated with disinfectants had antimicrobial effects against all tested microorganisms. The physical properties of the modified dental stones were within the ISO standards. However, further investigation on other properties such as dimensional stability, detail reproduction and clinical usage are still needed.

Introduction

In dentistry, infection control is becoming increasingly more important since dentists and their supporting staffs are constantly exposed to variety of microorganisms found in patients' mouth. Center for Disease Control has developed recommendations to prevent the spread of these contaminants in dental offices such as the wearing of gloves, masks and eye protectors. In prosthodontics, dental impressions have been considered to be potential sources of microbial contamination from patients' blood and saliva to dental personnel and prostheses fabricated on stone casts. Pathogenic microorganisms recovered from these impression materials and stone casts include *Staphylococcus aureus, Pseudomonas aeruginosa* and *Candida albicans* [1].

The disinfection of impressions is one of protocols developed to break this chain of cross-contamination. This is usually carried out through spraying the impression or immersing it in a disinfectant solution [2]. In these procedures, effective disinfection requires a long period of contact between the disinfectant and the impression material. This may become a problem since some impression materials, with their hydrophilic nature, can have change in dimensional accuracy [3]. Therefore, the incorporation of disinfectant in dental stone used in prosthesis fabrication has been regarded as a promising alternative. The aim of the present study was to evaluate the influence of incorporation of various disinfectants into type IV dental stone on its antimicrobial activity. In addition, dry compressive strength and setting time were also measured.

Materials and Methods

Dental stone

Type IV dental stone (calcium sulfate hemihydrate; Noritake SCG Plaster Co., Ltd, Thailand) incorporated with 4 types of disinfectants; Diamond Rock D (3-iodo-2-propynylbutylcarbamate), Diamond Rock B (zeolite), Diamond Rock Z (thiabendazole) and Diamond Rock T (2-benzimidazole carbamic acid), were tested in this study compared with the control, Diamond Rock O (type IV dental stone without disinfectant) which is well-accepted and commonly dental stone used in dental offices.

Antimicrobial testing

Three types of microorganisms (*Staphylococcus aureus* ATCC 6538, *Pseudomonas aeruginosa* ATCC 27853 and *Candida albicans* ATCC 10231) were used. They were grown on Brain Heart Infusion (BHI) agar (BBL, USA). Colonies were subcultured in BHI broth and incubated at 37°C for 24 h. Microbial suspensions were prepared at the concentration of 10^8 CFU/mL using 0.5 MacFarland Standard. One hundred µL of each microbial suspension was dropped on irreversible hydrocolloid impression (Jeltrate; Caulk/Dentsply, Milford, DE) and left dry for 10 min. All types of stone mix were prepared and poured onto the impressions and allowed to set for 60 min. Then the stone mass samples were removed and the microbial contact surfaces were imprinted on BHI agar plates. The plates were incubated at 37°C for 24-48 h. Colonies appeared on the agar were gram-stained and counted. Dental stone without disinfectant (Diamond Rock O) was used as the control. All experiments were done triplicate in 3 separate occasions.

Physical properties testing

The testing of setting time and dry compressive strength was carried out in accordance with International Standard (ISO) 6873: 1998 (E) [4] which states that conditions for testing dental stone products should be conducted at 23 ± 2 °C and 50 ± 10 % relative humidity.

For setting time determination, Vicat needle apparatus was used. A metallic needle connected to a vertically mobile rod under a total weight of 300 g was placed at a distance of 0.5 mm from the stone surface and suddenly set free 1 min prior to the stone mass lost its surface shine. Then, sequential penetration of the needle was carried out every 15 sec in different areas until the needle could not penetrate the stone any longer. The total time spent from the start of the mixing to the time when the needle first failed to penetrate the stone mass to a depth of at least 2 mm was defined as setting time.

For compressive strength testing, the stone was mixed and poured into mold (20 ± 0.2 mm in diameter and 40 ± 0.4 mm in length) to fabricate dental specimen. The dental specimens were stored in conditions mentioned above. At 60 ± 5 min after the start of mixing, specimens were stressed in the compressive mode until fractured using Instron testing machine (Instron Corp. Canton, USA) with a cross-head speed of 5 ± 2 kN/min. The maximum stress of each specimen (S) expressed in megapascal (MPa) using the recorded maximum force (F) was calculated according to the following formula:

$$S = F/314. \tag{1}$$

Kruskal-Wallis test was performed to find differences between the independent variables and specific comparisons was determined by Turkey test. The significance threshold was set at $p < 0.05$.

Results

Dental stone incorporated with disinfectants (Diamond Rock D, 3-iodo-2-propynylbutylcarbamate; Diamond Rock B, zeolite; Diamond Rock Z, thiabendazole and Diamond Rock T, 2-benzimidazole carbamic acid) showed antimicrobial activity against all microorganisms tested with the percentage of microbial reduction ranging from 83% to 100% compared to control (Diamond Rock D, dental stone without disinfectant) (Fig 1). No significant difference in microbial reduction was found among the groups of dental stone incorporated with disinfectants.

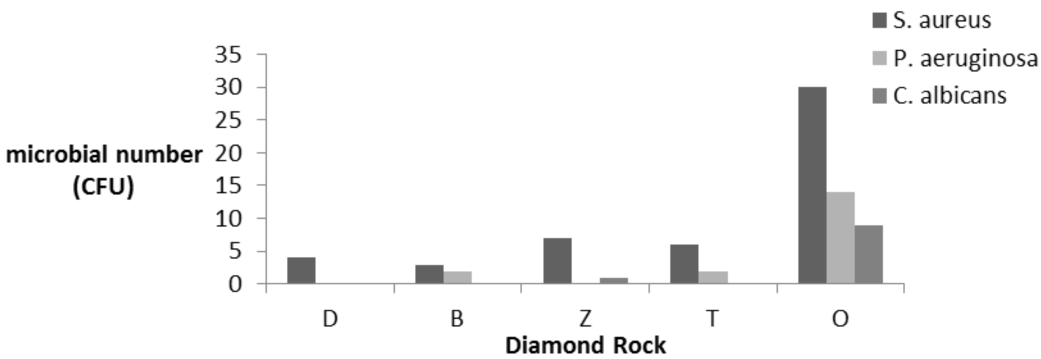

Fig. 1 Microorganisms detected from each type of disinfectant-containing dental stone

Regarding setting time and dry compressive strength, data are displayed in Table 1. Among all types of dental stone, no significant difference in setting time was observed. Dental stones incorporated with disinfectants seems to have increased setting time compared to that without the disinfectant. Diamond Rock D showed longest setting time. In dry compressive strength testing, the disinfectant-containing dental stone (Diamond Rock D, Diamond Rock B, Diamond Rock Z and Diamond Rock T) had significant higher strength than Diamond Rock O (control). Diamond Rock D exhibited the highest compressive strength at 41.9 MPa followed by Diamond Rock T at 40.6 MPa.

Table 1 Mean of physical properties of the dental stones

Dental stone	Setting time [min]	Compressive strength [MPa]
Diamond Rock D	7.75	41.9
Diamond Rock B	7.25	39.4
Diamond Rock Z	7.50	39.6
Diamond Rock T	7.50	40.6
Diamond Rock O	7.00	36.5

Discussion

Several pathogenic microorganisms have been detected from the surfaces of dental stone casts made from impressions and these casts may be a source of cross-infection transferred to dental personnel or other patients [1,5]. Therefore, recommendation has been set up to prevent this contamination by disinfection of impression materials before fabrication or incorporation of antimicrobial agents into dental stone. In a similar study, many types of disinfectants were added at the time of mixing dental stone powder with water such as iodophors, phenol, sodium hypochlorite, glutaraldehyde or chlorhexidine digluconate and it was demonstrated that these modified dental stones had varying activities against tested microorganisms [6]. In our study, all disinfectants incorporated with stone powder (3-iodo-2-propynylbutylcarbamate, zeolite, thiabendazole and 2-benzimidazole carbamic acid) showed effective antimicrobial action against both the tested bacteria and yeast. Even though the antimicrobial effect may occur due to many factors inherent in dental stone itself, such as heat of hydration of the setting stone, desiccation of the cast or by the effect of salt (calcium sulfate) on the microbes [7], significant microbial reduction was observed compared to dental stone without disinfectant. Considering antimicrobial effect against all tested microbes, *S. aureus* exhibited more resistance to the disinfectants. The emergence of antimicrobial or drugs resistance of *S. aureus* has been reported due to the efflux pumps which extrude the agents, hence decreasing intracellular concentration and increasing its survival [8]. Moreover, other cell wall and cell membrane modifications have also been documented [9].

In term of physical properties, stone-containing disinfectants had increased setting time compared with control without disinfectant. However, this retardation in the setting time is of minimal clinical significance and was still within the limit of ISO requirement which establishes setting time for type IV dental stone to be between 6 to 8 min. It has been proposed that some chemical agents such as low-solubility salts can delay the setting time of the stone [10]. Moreover, stone-containing disinfectants exhibited higher compressive strength. This could be an advantage of these newly developed stone materials even though the principle behind has still not been completely elucidated.

Acknowledgement

This study was supported by grants from Mahidol University Faculty of Dentistry and Siam Cement Group, Thailand.

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