

improvement of microbial resistance of resin-based dental sealant by sulfobetaine methacrylate incorporation

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Abstract

Prevention of dental caries is a key research area, and improvement of the pit and fissure sealants used for caries prevention has been of particular interest. This report describes results of incorporating a zwitterion, sulfobetaine methacrylate (SB), into photo-polymerized resin-based sealants to enhance resistance to cariogenic bacteria and protein adhesion. Varying amounts (1.5–5 wt%) of SB were incorporated into a resin-based sealant, and the flexural strength, wettability, depth of cure, protein adhesion, bacterial viability, and cell cytotoxicity of the resultant sealants were evaluated. The flexural strength decreased with the increasing SB content, but this decrease was statistically significant only for sealants containing ≥ 3 wt% SB. Incorporating a zwitterion led to a significant reduction in the water contact angle and protein adhesion. The colony-forming unit count showed a significant reduction in the bacterial viability of *S. mutans*, which was confirmed with microscopic imaging. Moreover, cell cytotoxicity analysis of SB-modified sealants using an L929 fibroblast showed a cytotoxicity comparable to that of an unmodified control, suggesting no adverse effects on the cellular metabolism upon SB introduction. Hence, we conclude that the addition of 1.5–3 wt% SB can significantly enhance the inherent ability of sealants to resist *S. mutans* adhesion and prevent dental caries.

Keywords: resin based sealant, zwitterion; oral bacteria; bacterial adhesion; protein adsorption

Introduction

Dental caries is a chronic oral ailment that has caused a worldwide oral health burden [1]. It is caused by an interplay of specific acidogenic bacteria, such as *Streptococcus mutans*, which are present in dental plaque surrounding external tooth surfaces. The interaction of such microbes with the tooth structure in the presence of carbohydrates leads to the initiation of irreversible damage in the form of dental caries [2]. Dental caries impacts people of all ages, and thus, it demands effective preventive measures. Given the multifactorial nature of dental caries, tooth morphology with deep pits and fissures is a risk factor because it presents potential sites for caries formation.

Various materials such as toothpaste, mouth rinse and fluoride varnish have been used to prevent dental caries. Since the pits and fissures of the occlusal surface have complicated and irregular structures, it is difficult to prevent dental caries using these materials [1]. To address the risk of caries formation due to pits and fissures, the adoption of preventive measures within the first few years of tooth eruption is commonly advised [2,3]. The preferred preventative measure is the use of dental resin-based sealants, which are designed to develop a physical barrier that prevents biofilm growth by blocking nutrition. Sealants are applied for the management of both the initial occlusal and proximal surface lesions. Thus researchers have explored several modification.

Materials and Methods**Incorporation of SB into Sealant**

Sulfobetaine methacrylate was used in the study. SB powder was mixed into a sealant at 1.5%, 3%, and 5% by weight. An unmodified sealant without the incorporation of SB was used as a control. From the results of preliminary experiments, it was confirmed that the content of SB at 5 wt% or more could dramatically decrease the mechanical properties of the base material, and based on this, the composition of the test group was confirmed in this study. Four sealant compositions were tested, as listed. In each experiment, at least five specimens were tested for each group.

Mechanical Properties

Mechanical properties were measured according to ISO 4049 [11]. For each group in Table 1, the bar-shaped samples (25 mm \times 25 mm \times 2 mm) were prepared. Samples were stored in distilled water at 37 ± 1 °C for 24 h after fabrication. A computer-controlled universal testing machine (Model 3366; Instron®, Norwood, MA, USA) was used to fracture the specimens in a three-point flexure. The flexural strength (σ) was calculated as $\sigma = 3Fl/(2bh^2)$

where F is the maximum load on the specimen (in N), l is the distance between the two supports (20 mm), b is the width (in mm) of the specimen measured before the test, and h is the thickness (in mm) of the specimen measured immediately before the test.

Wettability

Wettability was determined in accordance with previous studies, using a video contact angle goniometer (SmartDrop; Femtobiomed Inc., Gyeonggi-do, Korea) [11] with distilled water as the reference liquid. Four groups of samples were fabricated in a mold with a diameter of 15 mm and a thickness of 1 mm. A total of 2 μ L of distilled water was placed on the sample surface, and the contact angle with the sample surface was measured after 10 s

Results and Discussion

In this study, samples of a resin-based sealant containing varying amounts of SB, a zwitterionic additive, were evaluated and compared to an unmodified sealant. The results indicate that the null hypotheses can be rejected due to the observation of significant differences in the mechanical properties and bacterial resistance between the control and the SB-containing samples.

The approach of incorporating SB to maintain a balance of negative and positive charges and reduce the adhesion of serous protein and biomolecules has been extensively used in developing surface coatings for biomedical devices and implants [18]. However, reports have stated that the mode of action of SB in dental materials is different than that in other applications due to the complex interactions inherent to a dynamic oral environment [10]. The complexity of the oral

environment stems from its continuous interaction with the external environment, which results in multi-biome habitat formations. This predisposes the oral environment to pathologies like dental caries. Furthermore, *S. mutans* has been widely identified as one of the leading causative microorganisms for dental decay. Thus, the focus of the present research was to supplement the physical barrier mechanism of

resin sealant with anti-biofouling properties. In the current study, resin-based sealants containing varying concentrations of SB were tested to evaluate their mechanical properties and the effect of SB incorporation on protein and bacterial adhesion.

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