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Surface pre-conditioning with bioactive glass air-abrasion can enhance enamel white spot lesion remineralization

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ABSTRACT

Objective. To evaluate the effect of pre-conditioning enamel white spot lesion (WSL) surfaces using bioactive glass (BAG) air-abrasion prior to remineralization therapy.

Methods. Ninety human enamel samples with artificial WSLs were assigned to three WSL surface pre-conditioning groups ($n = 30$): (a) air-abrasion with BAG-polyacrylic acid (PAA-BAG) powder, (b) acid-etching using 37% phosphoric acid gel (positive control) and (c) unconditioned (negative control). Each group was further divided into three subgroups according to the following remineralization therapy ($n = 10$): (I) BAG paste (36 wt.% BAG), (II) BAG slurry (100 wt.% BAG) and (III) de-ionized water (negative control). The average surface roughness and the lesion step height compared to intra-specimen sound enamel reference points were analyzed using non-contact profilometry. Optical changes within the lesion subsurface compared to baseline scans were assessed using optical coherence tomography (OCT). Knoop microhardness evaluated the WSLs' mechanical properties. Raman micro-spectroscopy measured the υ-$(CO_3)^{2-}/\upsilon_1$-$(PO_4)^{3-}$ ratio. Structural changes in the lesion were observed using confocal laser scanning microscopy (CLSM) and scanning electron microscopy–energy dispersive X-ray spectrometry (SEM–EDX). All comparisons were considered statistically significant if $p < 0.05$.

Results. PAA-BAG air-abrasion removed $5.1 \pm 0.6\,\mu$m from the lesion surface, increasing the WSL surface roughness. Pre-conditioning WSL surfaces with PAA-BAG air-abrasion reduced subsurface light scattering, increased the Knoop microhardness and the mineral content

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of the remineralized lesions ($p < 0.05$). SEM–EDX revealed mineral depositions covering the lesion surface. BAG slurry resulted in a superior remineralization outcome, when compared to BAG paste.

Significance. Pre-conditioning WSL surfaces with PAA-BAG air-abrasion modified the lesion surface physically and enhanced remineralization using BAG 45S5 therapy.

1. Introduction

The optimal goal of minimal invasive (MI) dental caries management is to prevent and "heal" the incipient lesion by inhibiting the demineralization process, preventing any further mineral loss and enhancing the remineralization repair process [1]. The enamel white spot lesion (WSL) results from the physical changes occurring in enamel due to the caries process before it has reached the enamel–dentin junction (EDJ) [2]. Different protocols have been described to remineralize enamel WSLs including bioactive glass (BAG) 45S5 [3–7]. BAG 45S5 is an inorganic amorphous, calcium, sodium phosphosilicate material which contains fivefold ratio of Ca/P [8]. It interacts with aqueous solutions such as saliva to form a hydroxycarbonate apatite (HCA) layer, attached chemically to the treated surfaces [9,10]. In vitro and in vivo studies demonstrated that BAG 45S5 allowed close contact of the living cells at its surface and did not contain leachables which produce inflammation [11]. It has been shown that BAG 45S5 mixture prepared to treat dentin hypersensitivity and incipient enamel caries is a biocompatible material concluded by assessing the viability and the morphological alternations of pulp cells [12].

The remineralization of enamel WSL is a complex physicochemical process where the remaining mineral crystals are less reactive, covered by salivary proteins and the limited diffusion of ions lessens the net mineral gain [13–15]. In order to promote the WSL remineralization process, an additional stage of pre-conditioning the lesion surface using phosphoric acid prior to the remineralization therapy has been described and shown to increase the remineralization of WSLs [16–18]. In the present study, air-abrasion with PAA-BAG powder was used to pre-condition the lesion surface as opposed to cutting it, in order to promote remineralization using different topical therapies including mixtures of BAG 45S5. Polyacrylic acid (PAA) altered the hydroxycarbonate apatite (HCA) induced by BAG 45S5, with smaller structures deposited on the surfaces of enamel WSLs, remineralized using PAA-BAG slurry and assessed using Raman micro-spectroscopy and SEM [7]. The (—COOH) functional group of PAA may bind the calcium and phosphate ions to form nano-precursors small enough to penetrate the carious lesion more effectively [19,20].

The objectives of this study were to assess the physical effects of this WSL surface pre-conditioning and to study the impact of this modification on overall lesion remineralization. The physical changes were assessed using non-contact white light confocal profilometry and optical coherence tomography (OCT). The mineral content at the lesion surface following the application of the remineralization therapies was evaluated using Raman micro-spectroscopy used in a StreamLine™ scanning technique to map the lesion surface measuring the υ-$(CO_3)^{2-}/\upsilon_1$-$(PO_4)^{3-}$ ratio of 2880 spectra per sample. The biomechanical properties of the WSLs were assessed using Knoop microhardness testing. Confocal laser scanning microscopy (CLSM) and scanning electron microscopy (SEM) coupled with energy dispersive X-ray spectrometry (EDX) were used to study the ultra-structural changes within remineralized artificial WSLs created using a bi-layer demineralization protocol. The two null hypotheses investigated in this study were that pre-conditioning the WSL using PAA-BAG air-abrasion had no effect on lesion surface characteristics and that this pre-conditioning had no effect on the following remineralization therapy using BAG 45S5.

2. Materials and methods

2.1. *Sample preparation*

Caries-free human molars were collected using ethics approval reviewed by the East Central London Research Ethics Committee (Reference 10/H0721/55), stored in refrigerated deionized water and used within a month from the extraction. One buccal enamel slab from each tooth was sectioned using a diamond wafering blade (XL 12205, Benetec Ltd., London, UK). The slab's initial surface integrity was inspected using a confocal tandem scanning microscope (TSM) (Noran Instruments, Middleton, WI, USA), with an 20× air objective in reflection scanning mode. Ninety samples were included in this study. The samples were included face down in acrylic resin using a hard-anodized aluminum and brass sample former (Syndicad Ingenieurbüro, München, Germany). The superficial enamel layer was removed to create more consistent, reproducible artificial enamel lesions [14,21], using a water-cooled rotating polishing machine (Meta-Serv 3000 Grinder-Polisher, Buehler, Lake Bluff, IL, USA) with a sequential standard polishing protocol started from 600- to 4000-grit silica carbide disks, followed by 3 min of ultrasonication to remove the smear layer. Dental wax was applied to protect part of the enamel leaving an exposed window of 3 mm × 1 mm in the central area of the exposed enamel slab. The specimens were submitted to a previously documented bi-layer demineralization protocol of 8% methylcellulose gel buffered with a layer of lactic acid solution (0.1 mol/L, pH 4.6) for 14 days at 37 °C, to create artificial WSLs with an average depth of 70–100 μm [22,23]. The sound enamel areas around the WSL were covered with polyvinyl chloride tape throughout the experimental procedures, and removed at the end of the remineralization therapies. The samples were assigned into nine experimental groups according to the