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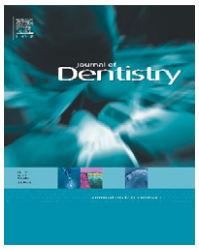

Influence of human dentine on the antibacterial activity of self-etching adhesive systems against cariogenic bacteria

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ABSTRACT

Objectives: The incorporation of antibacterial agents into adhesive systems has been proposed to eliminate residual bacteria from dentine. This study used the agar diffusion method to evaluate the antibacterial activity of Clearfil Protect Bond (CPB), Clearfil SE Bond (CSEB), Clearfil Tri-S Bond (C3SB) and Xeno-III (XIII) self-etching adhesive systems, with or without light-activation, against cariogenic bacteria, and to assess the influence of human dentine on the antibacterial activity of these materials.

Methods: An aliquot of 10 μl per material (and individual components) were pipetted onto paper and dentine discs distributed in Petri dishes containing bacterial culture in BHI agar. Positive control was 0.2% chlorhexidine digluconate (CHX).

Results: After incubation, the adhesive components of CPB and CSEB, liquid A of XIII and C3SB did not present antibacterial activity when applied to paper discs. The non-light-activated CPB primer + adhesive promoted the greatest inhibition of *Streptococcus mutans* ($p < 0.05$), whereas with light-activation, there was no significant difference between primer + adhesive and primer alone. For *Lactobacillus acidophilus*, CPB primer presented the greatest antibacterial activity in both light-activation conditions ($p < 0.05$). Regarding the dentine discs, only CHX promoted an inhibitory effect, though less intense than on paper discs ($p < 0.05$). CHX presented greater antibacterial activity against *S. mutans* than against *L. acidophilus* ($p < 0.05$).

Conclusions: Light-activation significantly reduced the antibacterial activity of the self-etching adhesive systems; MDPB incorporation contributed to the effect of adhesive systems against cariogenic bacteria; the components eluted from the adhesive systems were not capable to diffuse through 400 μm-thick dentine disc to exert their antibacterial activity against cariogenic bacteria.

1. Introduction

Although the etiological factors of caries disease and the methods for its prevention have been widely investigated, secondary caries is still the main cause for replacement of restorations.[1,2] During removal of carious tissue, bacteria invariably remain entrapped in the dentinal substrate because neither the clinical parameters of dentine hardness and colour

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nor the caries-detector dyes are able to ensure complete elimination of microorganisms.[3,4] As the contemporary dentine adhesive systems have not proved completely effective in eliminating microleakage at tooth/restoration interface, the incorporation of antibacterial agents into these materials has been proposed in an attempt to prevent caries recurrence.[5–15]

Among the antibacterial agents incorporated into adhesive materials, the resin monomer 12-methacryloyloxydodecyl-pyridinium bromide, known as MDPB, stands out.[8,9,13] The main advantage of MDPB is its capacity to copolymerize with other resin monomers being immobilised within the polymer matrix, which confers safety and prolonged antibacterial action to this agent, as it does not leach to the medium. This characteristic also ensures a good survival rate for the restoration, as MDPB, unlike soluble antibacterial agents, is not deleterious to the physical and mechanical proprieties of the materials to which it is incorporated.[8,9,16]

The incorporation of MDPB into self-etching adhesive systems has been advocated as extremely advantageous because, in the clinical practice, these materials are directly applied to the dentine with some level of contamination and, unlike the two-step or three-step total-etching adhesive systems, they do not have a separate acid etching step in their protocol, which could reduce significantly the number of residual microorganisms.[14,17,18] In spite of the acidic nature of self-etching adhesive systems, this characteristic does not seem sufficient to completely eliminate residual bacteria from dentine because this tissue acts as a solid buffering medium to acidic monomers.[15,19] Therefore, for the self-etching adhesive systems to inhibit the microorganisms that remain lodged inside the dentinal tubules, they should present antibacterial activity and capacity to diffuse to some extent through the dentin.[19]

However, there is little published information on the influence of dentine on the antibacterial properties of self-etching adhesive systems.[13,19,20] Therefore, the aim of this study was to evaluate the antibacterial activity of self-etching adhesive systems, with or without light-activation, against cariogenic bacteria, and to assess the influence of the interposition of 400 μm-thick human dentine discs on the antibacterial activity of these materials using the agar diffusion method.

2. Materials and methods

The specifications, main components and manufacturers of the materials used in this study are presented in Table 1. The antibacterial activity of the materials was evaluated against the following bacterial strains: *Streptococcus mutans* (UA-159) and *Lactobacillus acidophilus* (ATCC #IAL-523).

2.1. *Agar diffusion method: application on paper discs*

CPB, CSEB, C3SB and XIII self-etching adhesive systems, as well as their individual components, were submitted to the agar diffusion method. The positive and negative controls were, respectively, 0.2% chlorhexidine digluconate (CHX) and paper discs not impregnated with any material (PD).

Table 1 – Specifications, main components and manufacturers of the tested materials

Material (manufacturer)	Type	Composition	pH	Batch#
Clearfill Protect Bond (CPB) (Kuraray Medical Inc. Okayama, Japan)	Two-step self-etching adhesive system	Primer: HEMA, MDP, hydrophilic dimethacrylate, MDPB, water. Adhesive: HEMA, MDP, hydrophobic dimethacrylate, N,N-diethanol-*p*-toluidine, CQ, silanized colloidal silica, BisGMA, sodium fluoride	Primar: 1.9. Adhesive: 2.8	41137
Clearfil SE Bond (CSEB), (Kuraray Medical Inc., Okayama, Japan)	Two-step self-etching adhesive system	Primer: HEMA, MDP, hydrophilic dimethacrylate, N,N-diethanol-*p*-toluidine, CQ, water. Adhesive: HEMA, MDP, hydrophobic dimethacrylate, N,N-diethanol-*p*-toluidine, CQ, silanized colloidal silica, BisGMA	Primar: 1.9. Adhesive: 2.8	51308
Clearfil Tri-S Bond (C3SB) (Kuraray Medical Inc. Okayama, Japan)	One-step self-etching adhesive system	HEMA, BisGMA, MDP, hydrophobic dimethacrylate, silanized colloidal silica, CQ, ethanol, water	2.4	61113
Xeno-III (XIII) (Dentsply, Konstanz, Germany)	One-step self-etching adhesive system	Liquid A: HEMA, water, ethanol, THB, amorphous silica. Liquid B: Piro-EMA, PEM-F, UDMA, THB, CQ, Ethyl 4-dimethylaminobenzoate	1.0 (mixed)	0504002710
0.2% chlorhexidine digluconate (CHX) (University Pharmacy, UNESP, Araraquara, SP, Brazil)	Antibacterial solution	0.2% chlorhexidine digluconate aqueous solution	5.9	–

BisGMA: bisphenol A glycidyl dimethacrylate (BisGMA); THB: toluene hydroxybutyrate; CQ: D,1-camphorquinone; HEMA: 2-hydroxyethyl methacrylate; MDP: 10-methacryloxydecyl dihydrogen phosphate; MDPB: 12-methacryloyloxydodecyl pyridinium bromide; PEM-F: mono fluoro phosphazene modified methacrylate; Piro-EMA: phosphoric acid modified methacrylacte; UDMA: urethane dimethacrylate.