Spectroscopic Characterization of Structural and Functional Properties of Dentin Adhesive with Buffering Capability

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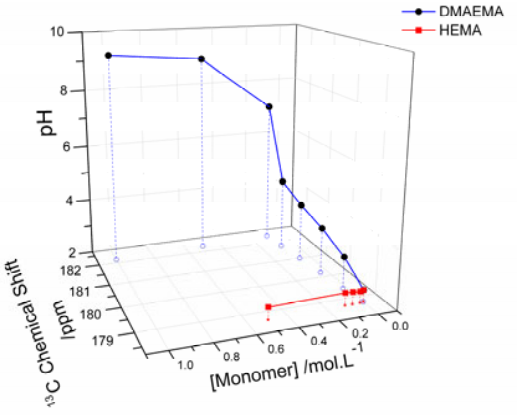
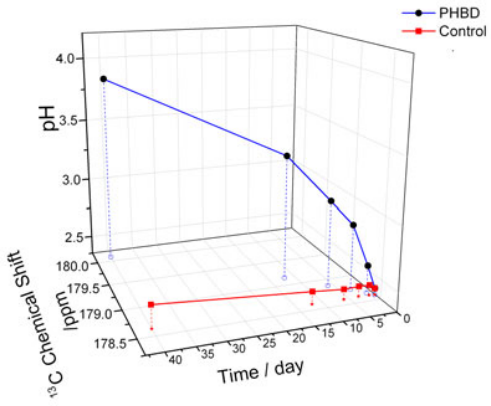
Statement of Purpose: Adhesion of *S. mutans* to surfaces in the mouth creates an environment that supports the subsequent attachment and growth of other bacterial species, ultimately forming a micro-ecosystem known as a biofilm [1]. In addition to its role as a "pioneer" organism in biofilm formation, *S. mutans,* produces lactic acid which damages the tooth surface by demineralization. Dental plaque biofilm cannot be eliminated[2] but the pathogenic impact of the biofilm at the tooth/composite interface could be reduced by engineering novel dentin adhesives that neutralize the micro-environment to prevent damage by demineralization. The objective of this research is to characterize, through spectroscopic techniques including ^{1}H/ ^{13}C NMR and FTIR, the neutralizing capacity of basic functional monomers and polymers containing buffering moieties.

Methods: 2-(dimethylamino) ethyl methacrylate (DMAEMA, 98%), 2-hydroxyethylmethacrylate (HEMA, 99%), 2,2-bis[4-(2-hydroxy-3-methacryloxypropoxy) phenyl]-propane (BisGMA), L(+)-lactic acid (LA, 98%), and deuterium oxide (99.8 atom % D) were purchased from Sigma Chemical Co., St. Louis, MO, USA. Experimental adhesives (PHBD) containing HEMA, BisGMA and DMAEMA 25/55/20 (w/w) were polymerized with visible light and compared to control adhesives [HEMA/BisGMA, 45/55 w/w]. The 0.1M solution of LA in D_2O was mixed with different molar concentrations of DMAEMA or HEMA. The samples were cast on the diamond crystal top-plate of an attenuated total reflectance accessory using a Perkin-Elmer Spectrum 400 FTIR. LA is the primary compound produced during acidification of the oral cavity by microbes and thus, the assays focused on determining behavior in LA. The addition of basic monomeric units or hydrated polymers that contain buffering moieties alters the pH of the sample. The degree of change was tracked and the buffering capacity quantified using NMR-based assay. NMR spectra of the solution were obtained using a Bruker Avance DRX 500 MHz NMR spectrometer.

Results: After interaction with lactic acid, the appearance of the FTIR characteristic peaks of COO^- asymmetric stretching vibration at 1587 cm^{-1} indicates the formation of amino-LA complex. The NMR chemical shift is extremely sensitive to small changes that most other methods cannot detect, making it an excellent probe for monitoring perturbations to the nucleus of interest. Here, the chemical shift of the carbonyl ^{13}C in LA has been correlated with pH and monitored as a function of increasing concentration of monomer or incubation time with polymer (Figure 1). HEMA or control adhesive does not alter the pH of the 0.1 M LA solution, whereas increasing amounts of DMAEMA which contains a basic amine or longer incubation of experimental adhesive PHBD shift the pH of the acidic LA solution making it more neutral.



(A)



(B)

Figure 1. Three-dimensional plot of pH vs ^{13}C NMR chemical shift (ppm) of the carbonyl group (C=O) from lactic acid (LA) in solutions (A) titrated with increasing concentrations of HEMA or DMAEMA monomers; (B) incubated for 40 days with experimental adhesive (PHBD) or control adhesive.

Conclusions: Inclusion of HEMA, the monomer currently used in the methacrylate dentin adhesive, has no impact on the pH of the solution and cannot buffer or neutralize lactic acid. In comparison, the results show that neutralization can be achieved by basic functional compounds such as DMAEMA and that the buffering capacity can be measured in a simple spectrometry experiment using lactic acid as a probe.

References:

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Acknowledgement: Supported by grant R01DE14392 and 3R01DE014392-08S109 (Spencer), from the NIDCR/NIH, Bethesda, MD 20892.