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Long-term stability of dental adhesive incorporated by boron nitride nanotubes

Felipe Weidenbach Degrazia^{a,b}, Vicente Castelo Branco Leitune^a,
Fernanda Visioli^c, Susana Maria Werner Samuel^a,
Fabrício Mezzomo Collares^{a,*}

^a Dental Materials Laboratory, School of Dentistry, Federal University of Rio Grande do Sul, Rua Ramiro Barcelos, 2492, Rio Branco, 90035-003, Porto Alegre, RS, Brazil

^b School of Dentistry, University of Federal District — UDF, SHCS Q 704/904 — Asa Sul, 70390-045, Brasília, Brazil

^c School of Dentistry, Federal University of Rio Grande do Sul, Rua Ramiro Barcelos, 2492, Rio Branco, 90035-003, Porto Alegre, Brazil

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ABSTRACT

Objective. The aim of this study was to evaluate physicochemical properties, long-term microtensile bond strength and cytotoxicity of methacrylate-based adhesive containing boron nitride nanotubes (BNNTs) as fillers.

Methods. A dental adhesive was formulated using BisGMA/HEMA, 66/33 wt% (control). Inorganic BNNT fillers were incorporated into the adhesive at different concentrations (0.05, 0.075, 0.1 and 0.15 wt%). Analyses of degree of conversion (DC), polymerization rate [R_p (s^{-1})], contact angle (CA) on dentin, after 24 h and 6 months microtensile bond strength (μ TBS-24 h and 6 months) were assessed. Cytotoxicity was performed through viability of fibroblast cells (%) by sulforhodamine B (SRB) colorimetry.

Results. DC and max. polymerization rate increased ($p < 0.05$) after incorporating 0.075 and 0.1 wt% BNNT. The contact angle on dentin increased ($p < 0.05$) after incorporating 0.15 wt% BNNT. The μ TBS-24 h showed no changes ($p > 0.05$) after incorporating up to 0.15 wt% BNNT comparing to control. After 6 months, μ TBS decreased ($p < 0.05$) for control and 0.15 wt% BNNT and BNNT groups up to 0.15 wt% showed higher μ TBS than control ($p < 0.05$). No difference of fibroblast growth was found among adhesives ($p > 0.05$) and up to 19% of cell viability was found comparing 0.05 wt% BNNT to positive control group (100%).

Significance. Incorporating boron nitride nanotubes up to 0.1 wt% into dental adhesive increased the long-term stability to dentin without decreasing viability of fibroblast cell growth. Thus, the use of BNNTs as filler may decrease failure rate of current dental adhesives.

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* Corresponding author.

E-mail addresses: fdegrazia@hotmail.com (F.W. Degrazia), vicente.leitune@ufrgs.br (V.C.B. Leitune), fernanda.visioli@ufrgs.br (F. Visioli), susana.samuel@ufrgs.br (S.M.W. Samuel), fabricao.collares@ufrgs.br (F.M. Collares).
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1. Introduction

Adhesive dentistry has advanced significantly over the past ten years, although premature reductions in bond strength, interface and marginal degradation, and biocompatibility of adhesives still pose challenge to the field [1]. These issues may occur due to the presence of non-entangled exposed collagen fibrils in the partially or completely demineralized dentin, after the etching step from acids or acidic resin monomers [2]. The incomplete monomer infiltration into the collagen matrix leads to the presence of mineral-depleted, resin-sparse, water-rich fibrils along the bonded interface challenging the interfacial integrity between resin and dentin [3]. Since the discovery of the hybrid layer, efforts have been focused on displace water within the extra- and intrafibrillar compartments of demineralized collagen matrix [4,5]. Hydrophilic monomers used to promote adhesive diffusion into demineralized dentin leads to higher water uptake and thus decrease its mechanical properties over time [6,7].

A recent review of innovative applications in dentistry [8] claimed that fortifying adhesives with nanomaterials that possess biological merits and enhancing their mechanical and physical properties help to attain and maintain a durable adhesive joint with enhanced longevity. Therefore, novel functionalizing nanofillers to improve current dental adhesives have been evaluated by several studies [9–12]. One-dimensional nanotubes have been investigated [13,14] and offer an excellent choice to reinforce dental composites with bioactive properties. Among such, boron nitride nanotubes (BNNTs) have been recently used as nanofillers of a dental adhesive [15] showing bioactivity and improved physicochemical properties. The mechanical properties of their nanocomposites is enhanced by various functional groups such as amines and nitriles [16,17] causing improvement on their dispersion in polymer matrices that facilitates better polymer-nanomaterial interaction and allows efficient load transfer from the polymer to the nanomaterial [18]. Besides, their hydrophobic character could act as a shield against water remained on demineralized collagen matrix avoiding hydrolytic softening of the polymer matrix [15] thus improving the longevity of the adhesive bonding to dentin [1,10].

Therefore, the purpose of this paper was to evaluate the long-term stability on dentin, polymerization kinetic and contact angle of a methacrylate-based adhesive incorporated by BNNTs and their cytotoxicity to dental fibroblast cells. The null hypothesis tested is that the incorporation of BNNT into a dental adhesive will not influence its chemical properties, bonding and cytotoxicity.

2. Material and methods

2.1. Adhesive formulation and incorporation of BNNT

The experimental adhesives were formulated by mixing 66.6 wt% bisphenol A glycerolate dimethacrylate (BisGMA) and 33.3 wt% of 2-hydroxyethyl methacrylate (HEMA) from Sigma-Aldrich (St. Louis, MO, USA) as described in a previous study

[12]. BNNTs (BNNT, LCC, Newport News, VA, USA) synthesized via a pressurized vapor/condenser method [19] were first immersed in a 65% HNO₃ bath for 3 h and into pure H₂O₂ for 48 h [20]. BNNTs were then used in four concentrations (0.05, 0.075, 0.1 and 0.15 wt%) and firstly incorporated into BisGMA, hand mixed and dispersed via ultrasonication for 1 h, and then HEMA was mixed. A control group without BNNT was used. As a photoinitiator system, camphorquinone and ethyl 4-dimethylaminobenzoate at 1 mol% each were added to all groups.

2.2. Polymerization kinetic

The degree of conversion (DC) and polymerization rate [Rp.(s⁻¹)] of each experimental adhesives were evaluated using Fourier transform infrared (FTIR) spectrometer Vertex 70 (Bruker Optics, Ettlingen, Germany) equipped with Attenuated Total Reflectance (ATR) device. A support was coupled to fix the light source and standardize the distance between the tip and the top of the 2 mm-height sample. Analysis was performed at a controlled room temperature of 23 ± 1 °C and 60 ± 1% relative humidity during sample (3 μL) polymerization, which was directly dispensed onto the diamond crystal and light activated for 50 s (n = 3) by a light-emitting diode (RadiiCal, SDI, Bayswater, VIC, Australia).

DC and Rp.(s⁻¹) were calculated for the experimental adhesives as described in a previous study [21] based on the intensity of the C=C stretching vibrations (peak height) as a function of time at 1635 cm⁻¹ and using the symmetric ring stretching at 1608 cm⁻¹ as an internal standard from the polymerized and non-polymerized samples. The IR-Solution software was used to set the standard parameters and to check the scanning in Happ-Genzel apodization function mode in a range of 4000–800 cm⁻¹, resolution of 4 cm⁻¹, and scanning time of 50 s. This setup allowed the acquisition of each single scan, every 0.49 s during adhesive photo-activation. The data was plotted in SigmaPlot 12.0 (Systat Software Inc, San Jose, CA, USA) and a sigmoidal curve fitting method was applied using linear regression. DC (%) × time (s) and [Rp.(s⁻¹)] × time (s) were plotted for each amount of BNNT.

2.3. Contact angle

Twenty-five bovine teeth were embedded in acrylic resin and their superficial dentin was exposed and ground flat (600-grit silicon-carbide paper) under running water for 1 min. A dentin area of 5 mm diameter was etched with 37% phosphoric acid for 15 s, rinsed with water, kept moist and primed actively for 20 s (Primer Scotch Bond MultiPurpose; 3 M ESPE), and the solvent evaporated for 5 s. Analysis was performed using an optical tensiometer (Theta, Biolin Scientific, Stockholm, Sweden) to measure the static CA (θ) among dentin and adhesive [22]. The drop out size, drop rate, displacement rate, and speed dispersion of adhesive were: 3.0 μL, 2.0 μL/s, 20.0 μL/s, and 50 mm/min, respectively. The test period was performed over 20 s and the mean contact angle (θ) between the droplet and the solid surface was registered after 10 s, following previous study [15].

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