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Bioactive composites containing TEGDMA-functionalized calcium phosphate particles: Degree of conversion, fracture strength and ion release evaluation

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ABSTRACT

Objective. To evaluate the strength and ion release of experimental composites containing TEGDMA-functionalized calcium phosphate particles.

Methods. Seven composites containing equal parts (in mols) of BisGMA and TEGDMA and 60 vol% of fillers were manipulated. Filler phase was constituted by silanized barium glass and 0% (control), 10% or 20% (volume) of dicalcium phosphate dihydrate (DPCD) particles, either non-functionalized or functionalized with two different TEDGMA contents. DCPD particles were synthesized and characterized by X-ray diffraction (XRD), elemental analysis, surface area and dynamic light scattering. Composites were tested for degree of conversion (DC) by near-FTIR. Biaxial flexural strength (BFS) was determined after 24 h and 28 days in water. Calcium and phosphate release after 7 days was assessed using inductively coupled plasma optical emission spectrometry (ICP-OES). Data were analyzed by ANOVA/Tukey test (alpha:5%).

Results. XRD confirmed the crystalline structure corresponding to DCPD. Elemental analysis revealed particles with zero, 14% or 22% TEGDMA, with similar D_{50} (around 19 μm) and surface areas from 3.5 to 11.4 m^2/g. The presence of DCPD did not reduce DC. After 24 h, functionalization (both 14% and 22% TEGDMA) improved composite strength in comparison to non-functionalized DCPD, both at 10% and 20% levels. After 28 days, BFS of materials containing 10% functionalized DCPD were statistically similar to the control containing only barium glass. Among composites containing 10% DCPD, particle functionalization with 14% TEGDMA did not jeopardize ion release.

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Significance. At 10 vol%, the use of TEGDMA-functionalized CaP particles improved composite strength in relation to non-functionalized particles, while maintaining similar ion release levels.

1. Introduction

Over the years, the incorporation of calcium orthophosphates [1–4] and bioactive glass particles [5,6] in resin-based materials has been studied as a possible path to promote tooth remineralization and reduce the risk of secondary caries at the tooth/restoration interface [1,7–10]. These particles behave as ion sources, releasing calcium and phosphate to the surrounding medium and creating a supersaturated environment that favors mineral deposition on the tooth.

Unfortunately, the addition of CaP particles to unfilled resins significantly lowers their strength [11,12]. This impaired mechanical behavior is ascribed to the lack of a strong chemical bond between the bioactive particles and the resin phase, as well as the low cohesive strength of the CaP agglomerate itself [11–13]. Several approaches have been tested to increase the mechanical properties of bioactive composites. For example, an experimental composite containing hydroxyapatite particles functionalized with acrylic acid showed a significantly higher flexural strength in comparison with a control material containing unmodified CaP particles [14]. Silanization of CaP particles was also tested and, though composite strength increased in comparison to the use of non-silanized particles, ion release was negatively affected [15,16].

The association between reinforcing fillers and bioactive particles seems to offset some of the negative effect of the latter on composite mechanical properties. For example, when 40 wt% of tetracalcium phosphate (TTCP) was added to an unfilled resin, 20 wt% of silanated glass had to be added in order to recover the flexural strength of the unfilled resin [12]. In another study, the addition of 10 wt% reinforcing fillers to an experimental material containing 40 wt% amorphous calcium phosphate (ACP) also contributed to improve composite strength, with no adverse effect on ion release [17].

Recently, DCPD nanoparticles functionalized with triethylene glycol dimethacrylate (TEGDMA) were synthesized [18]. Among the different calcium orthophosphate phases, dicalcium phosphate dihydrate (DCPD, $CaHPO_4.2H_2O$) presents a relatively high solubility [19], which makes it more suitable as ion source than less soluble phases, such as hydroxyapatite (HA). Also, its refractive index (1.54–1.55) [20] is similar to barium glass (1.53) [21]. HA, for example, has a refractive index of 1.63–1.67, which compromises light transmission during photoactivation, as well as esthetics [14]. By adding the monomer to one of the reacting solutions (*i.e.*, prior to DCPD crystallization), it would attach to the Ca^{2+} at the nanoparticle surface at early stages of its growth, when reactivity is at its maximum. Also, the TEGDMA in the functionalizing layer would co-polymerize with monomers of the resin matrix, increasing the compatibility between the nanostructured agglomerates and the resin. Preliminary studies showed that the incorporation of TEGDMA-functionalized DCPD in a BisGMA:TEGDMA resin matrix increased biaxial flexural strength by 32% in comparison with a material containing non-functionalized particles.

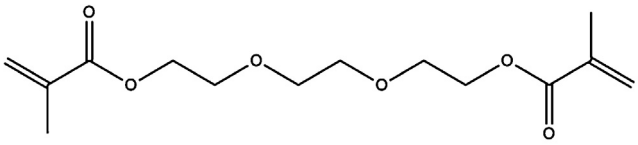

Fig. 1 – Structural formula of triethylene glycol dimethacrylate (TEGDMA) used as functionalizing agent.

This investigation is a follow-up from a previous study where the substitution of silanized barium glass with proprietary, non-functionalized DCPD nano-structured agglomerates in experimental composites resulted in up to 35% reduction in fracture strength [13]. In the present investigation, TEGDMA-functionalized DCPD particles were tested partially replacing the reinforcing fillers. The ultimate goal was to determine if the use of functionalized DCPD particles would improve composite strength (in comparison to the material containing non-functionalized particles) without jeopardizing ion release. Additionally, the effect of the amount of TEGDMA in the functionalizing layer was also verified. The null hypotheses were: (1) replacing 10 vol% or 20 vol% of silanized glass fillers by DCPD particles does not affect composite degree of conversion and strength (prior and after aging in water for 28 days), regardless of the DCPD content or the presence of a TEGDMA functionalizing layer, (2) ion release is not affected by DCPD content or by functionalization of the DCPD particles.

2. Methods

2.1. *Synthesis and characterization of dicalcium phosphate dihydrate (DCPD) nanoparticles*

DCPD ($CaHPO_4.2H_2O$) nanoparticles were synthesized through the reaction between ammonium phosphate $(NH_4)_2HPO_4$, and calcium nitrate $Ca(NO_3)_2.4H_2O$ (both from Sigma–Aldrich Co., St. Louis, MO, USA) by a sol–gel process [18]. Equimolar solutions (0.078 mol/L) were prepared with deionized water at room temperature and mixed with a magnetic stirrer. The ammonium phosphate solution received 6.86 g (0.024 mol) of triethylene glycol dimethacrylate (Fig. 1, TEGDMA, Mw = 286 g/mol, Sigma–Aldrich). 400 mL of the calcium nitrate solution was added to the same volume of the ammonium phosphate/TEGDMA solution using a peristaltic pump (9 mL/min). The resulting mixture was kept under stir-

ring for 30 min, followed by 30 min of decantation. Then, the supernatant was removed, and the gel was washed by adding deionized water and centrifuging it (1085 × *g*, Sorvall RC6 Plus, Thermo Fisher Scientific, Asheville, NC, USA). After centrifugation, the TEGDMA-rich supernatant was removed and a new washing cycle initiated. TEGDMA content on the functionalizing layer was modulated by varying the number of washing cycles. The first two batches were washed two and seven times, respectively, while in the last synthesis the gel was washed four times using water plus one cycle using a TEGDMA/water mixture. The pastes were freeze-dried and a white powder was obtained.

Nanoparticles were characterized according to different methods. In order to identify the calcium orthophosphate phase, X-ray diffraction (XRD) measurements were taken using nickel CuKα radiation at 40 kV and 30 mA (MultiFlex, Rigaku Corp., Tokyo, Japan). The equipment geometry was $\theta/2\theta$. Readings were continuous from 10° to 60° at 0.05° intervals, 10 s per interval. Elemental analysis (2400 CHN Elemental Analyzer, PerkinElmer, Waltham, MA, USA) was used to determine TEGDMA content, based on the percentage of carbon detected in the sample. Approximately 1 mg of each powder was heated to 925 °C under pure oxygen. Resultant gases (CO_2, H_2O and N_2) were homogenized and quantified with a resolution of 0.3%.

Particle surface area was determined after nitrogen adsorption isotherms were obtained using a volumetric adsorption analyzer (Quantachrome Instruments, Boynton Beach, FL, USA). In order to maintain the organic coating around the particles, samples were purged at 50 °C for 14 h. Surface area was determined according to the Brunauer, Emmet and Teller (BET) method (NOVAWin, Quantachrome Instruments, Boynton Beach, FL, USA). Size distribution of the nanoparticle agglomerates was estimated using a laser diffraction particle analyzer (Mastersizer 2000 Ver. 5.54, Malvern Instruments Ltd., Worcestershire, UK).

2.2. Composite formulation

Seven composites were prepared, with the organic phase containing 1:1 in mols of bisphenol-A glycidyl dimethacrylate (BisGMA) and TEGDMA, plus 0.5 wt% of camphorquinone and ethyl-4-dimethylamino benzoate (EDMAB, all components from Sigma–Aldrich) as photoinitiators. All composites had a total filler of 60 vol%. Filler phase was constituted by 40, 50 or 60 vol% of silanated barium glass and 20, 10 or 0 vol% of DCPD particles. Three different DPCD particles with different TEGDMA contents (obtained as previously described) were used. Composites were mechanically mixed under vacuum (Speedmixer DAC 150.1 FVZ-K, FlackTek Inc., Landrum, SC, USA) and kept under refrigeration until 2 h before use.

2.3. Degree of conversion

Degree of conversion (n = 3) was determined by Fourier-transformed near-infrared spectroscopy (near-FTIR) [22]. Unpolymerized composite was placed in a silicon mold (7 mm diameter and 1 mm thickness) pressed between two microscopy glass slides. The spectrum of the unpolymerized material was obtained between 4000 and 10,000 cm^{-1} by the co-addition of 32 scans with 4 cm^{-1} resolution (Vertex 70; Bruker Optik GmbH, Ettlingen, Germany). The composite was then photoactivated through the glass slide (Radii Cal, SDI, Bayswater, Australia), receiving 24 J/cm^2 (1200 mW/cm^2 for 20 s) and the whole set was dry-stored at 37 °C for 24 h. Then, a new spectrum was obtained and the degree of conversion was calculated as the ratio between the areas of the absorption band located at 6165 cm^{-1} (corresponding to the absorption of the =C—H_2 group) of the polymerized and the unpolymerized material.

2.4. Biaxial flexural test

Disk-shaped specimens (12 mm × 1 mm, n = 20) were built using a stainless steel split mold. Half of the specimens were stored in water at 37 °C for 24 h and the other half for 28 days. Photoactivation was performed by irradiating each quadrant for 10 s. After storage, the specimens were placed on a "piston on three balls" device positioned on a universal testing machine (Kratos KE series, Cotia, Sao Paulo, Brazil) at a cross-speed of 0.5 mm/min. The discs were positioned with the irradiated surface facing the flat piston and loaded until fracture. Biaxial flexural strength (BFS), in MPa, was calculated according to the following equations:

$$BFS = \frac{-0.2387P(X - Y)}{b^2}$$

$$X = (1 - \nu)\ln\left(\frac{r_2}{r_3}\right)^2 + \left[\left(\frac{1 - \nu}{2}\right)\right]\left(\frac{r_2}{r_3}\right)^2$$

$$Y = (1 - \nu)\left[1 - \ln\left(\frac{r_1}{r_2}\right)^2\right] + (1 - \nu)\left(\frac{r_1}{r_3}\right)^2$$

where P is the fracture load (in Newtons); b is the specimen thickness (in mm); ν is the Poisson's ratio; r_1 is the radius of the supporting spheres circumference (5 mm); r_2 is the radius of the loading piston (0.6 mm): r_3 is the specimen radius (in mm). The Poisson's ratio was set for 0.30 for all composites. Fractured surfaces were gold-sputtered and observed under scanning electron microscopy (LEO 430, Electron Microscopy Ltd., Cambridge, UK).

2.5. Ion release

Disk-shaped specimens (5 mm × 1 mm, n = 3) were dry-stored for 24 h at 37 °C and then immersed individually for 7 days in 5 mL of NaCl solution (133 mmol/L) buffered to pH = 7 with 50 mmol/L HEPES. The concentrations of calcium and phosphate ions released in the solution were determined by inductively coupled plasma-optical emission spectrometry (ICP-OES, 700, Agilent Technologies, Santa Clara, CA, USA). Prior to analysis, the specimen was removed from the vial and the solution was filtered (pore size: 3 μm) before being acidified with 5 mL of 10% HNO_3 (pH = 0.2). ICP-OES uses photon detection (emitted by the elements of interest when decaying from an excited state reached after collision with ions from an argon plasma) to quantify element concentration in the solution. Calcium and phosphorous readings were obtained simultaneously, based on the emission wavelengths of 184 nm and 177 nm, respectively.

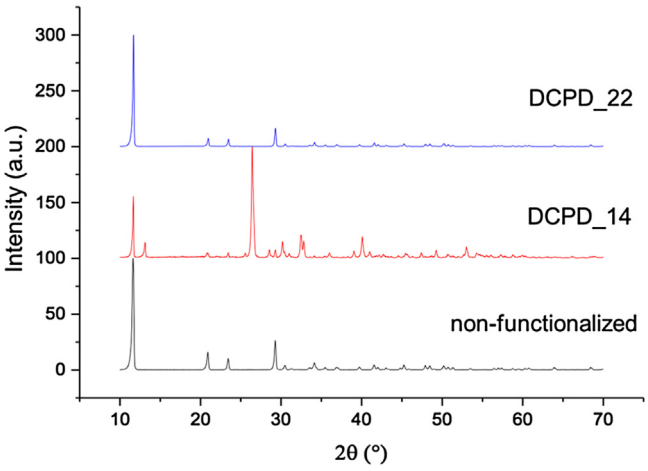

Fig. 2 – Difractograms of the calcium phosphate nanoparticles (Top: DCPD functionalized with 22% TEGDMA, middle: 14% TEGDMA, bottom: non-functionalized DCPD).

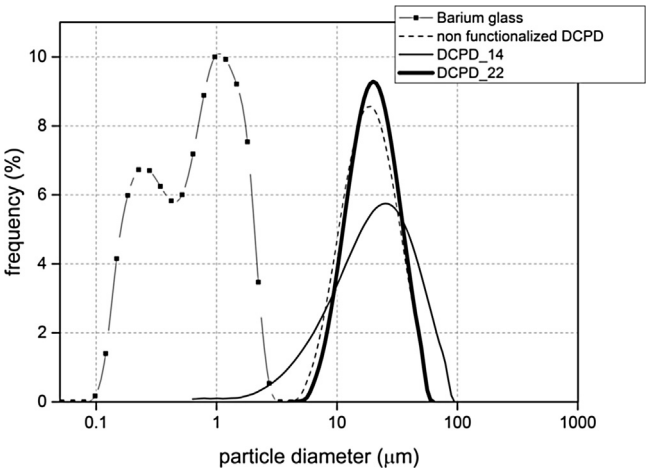

Fig. 3 – Filler size distributions for barium glass and functionalized DCPD particles.

2.6. *Statistical analysis*

Data were subjected to either one-way ANOVA (degree of conversion) or two-way ANOVA (biaxial flexural strength and ion release). For the mechanical testing, the main factors were DCPD content (seven levels, *i.e.*, six combinations between two DCPD levels and three TEGDMA contents, plus the control material without DCPD) and storage time (24 h or 28 days), while for ion release the main factors were DCPD content (10% or 20%) and particle TEGDMA content. In all cases, Tukey test was used for pair-wise comparisons. The global significance level (α) was set at 0.05.

3. Results

3.1. *Particles characterization*

Difractograms (Fig. 2) identified the crystalline structure of DCPD for the three synthesized calcium phosphate powders (International Center for Diffraction Data/Joint Committee on Powder Diffraction Standards, ICDD/JCPDS, PDF 09-0077). Additionally, the powder obtained in the synthesis that used two washing cycles also revealed the presence of DCPA (dicalcium phosphate anhydrous, ICDD/JCPDS PDF 090080).

Elemental analysis revealed only traces of carbon in the powder obtained after seven washing cycles and 8% of carbon in the powder subjected to two washing cycles, which corresponds to a TEGDMA content of 14%. The powder resulting from the synthesis where the last of four washing cycles used a TEGDMA/water mixture revealed a carbon content of 13%, corresponding to 22% of TEGDMA. Therefore, DCPD particles with 0% TEGDMA will be referred to as "non-functionalized particles", while those with 8% and 13% carbon will be referred to as "DCPD_14" and "DCPD_22", respectively. According to the BET method, the surface areas of the particles were 11 m^2/g (non-functionalized), 5 m^2/g (DCPD_14), and 4 m^2/g (DCPD_22).

Particle size distributions are shown in Fig. 3. The median particle sizes (D_{50}) of the nanoparticles agglomerates were quite similar among the different powders (non-functionalized: 17 μm, DCPD_14: 20 μm and DCPD_22: 19 μm). Barium glass filler showed a D_{50} of 0.5 μm.

3.2. *Degree of conversion*

Degree of conversion (DC) results are shown in Table 1. The replacement of barium glass by non-functionalized or DCPD_22 particles did not significantly affect DC, regardless of the DCPD content. However, the use of DCPD_14 resulted in higher DC compared to the composites containing only silanized glass or non-functionalized DCPD particles ($p < 0.001$).

3.3. *Biaxial flexural test*

BFS results are shown in Table 1. After 24 h storage, the replacement of barium glass by calcium phosphate particles resulted in significant reductions in fracture strength ($p < 0.05$). More severe reductions were observed for composites containing non-functionalized nanoparticles (10% DCPD: 41%, 20% DCPD: 71%) than those containing DCPD_14 (10% DCPD: 28%, 20% DCPD: 36%) and DCPD_22 (10% DCPD: 17%, 20% DCPD: 37%). Among composites containing 10% DCPD, strength increased with the particle TEGDMA content. For materials containing 20% DCPD, particle functionalization increased strength; however, no difference was observed between DCPD_14 and DCPD_22. Strength values of composites containing 10% and 20% DCPD_14 were not statistically different.

After 28 days in water, all composites showed reductions in strength ranging between 21% and 39%. Among composites containing 10% DCPD, no statistically significant difference was found between DCPD_14 and DCPD_22. Noteworthy, their strength values were statistically similar to that of the control containing only barium glass. For the composites containing 20% DCPD, particle functionalization did not lead to improved strength after 28 days. Finally, no difference was observed between composites containing 10% or 20% of non-functionalized particles.

Table 1 – Degree of conversion and biaxial flexural strength results (average and standard deviations) of experimental composites as a function of DCPD content and amount of TEGDMA in the functionalizing layer, and storage period (strength only). Similar letters in a given test indicate lack of statistically significant differences (ANOVA/Tukey test, $p > 0.05$).

DCPD content (vol%)	TEGDMA in the particle (wt%)	Degree of conversion (%)	Biaxial flexural strength (MPa)	
			24 h	28 days
0 (control)		78.1 (0.1) ^{C}	161.4 (12.7) ^{A}	97.9 (10.5) ^{DE}
10	0	77.7 (0.6) ^{C}	95.4 (14.4) ^{DE}	65.0 (6.1) ^{GH}
	14	83.2 (1.3) ^{B}	116.8 (10.3) ^{C}	86.9 (11.5) ^{EF}
	22	79.6 (0.5) ^{BC}	133.6 (15.0) ^{B}	96.7 (9.3) ^{DE}
20	0	78.3 (0.7) ^{BC}	76.2 (9.9) ^{FG}	60.5 (4.6) ^{H}
	14	88.7 (4.6) ^{A}	103.5 ± 9.5^{CD}	65.8 ± 13.4^{GH}
	22	81.7 (0.2) ^{BC}	101.2 ± 11.8^{DE}	69.2 ± 8.1^{GH}

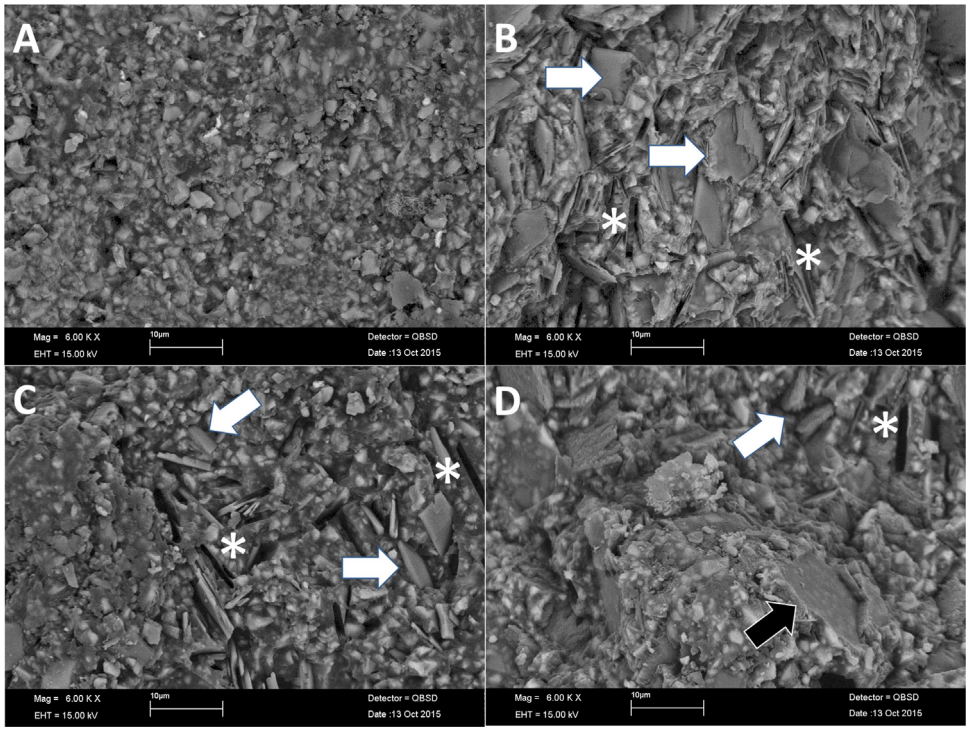

Fig. 4 – Backscattered SEM images of composite fractured surfaces. White arrows indicate the DCPD agglomerates, asterisk indicate empty spaces where the agglomerates were displaced upon fracture and the black arrow shows the imprint left by an agglomerate on the resin matrix. A: Control, 28 days; B: 20% non-functionalized DCPD, 28 days; C: 20% DCPD_14, 28 days; D: 20% DCPD_22, 28 days.

SEM images of fractured surfaces after 28 days storage in water are shown in Fig. 4. The control composite (Fig. 4A) revealed exposed particles suggesting that fractured occurred at the matrix/particle interface. The composite containing non-functionalized DCPD (Fig. 4B) showed several plate-like DCPD agglomerates protruding from the matrix, as well as slit-shaped spaces suggesting that agglomerates detached from the matrix upon fracture. Composites containing functionalized DCPD (Figs. 4C and D) presented a similar aspect, except for the smoother agglomerate edges, in comparison to the uneven, rugged edges displayed by the non-functionalized DCPD. No differences were observed between specimens fractured after 24 h and 28 days.

3.4. *Calcium and phosphate release*

Calcium release values are presented in Fig. 5A. The interaction between DCPD percentage and TEGDMA content was not statistically significant ($p = 0.275$), though both factors influenced calcium release ($p < 0.001$). Composites containing 20% DCPD released 23% more Ca^{2+} then those with 10% DCPD (20%: 0.242 ± 0.051 mmol/L; 10%: 0.186 ± 0.031 mmol/L). Overall, regardless of the DCPD content, particle functionalization reduced Ca^{2+} release between 22% and 32%, while TEGDMA content in the particle did not significantly affect Ca^{2+} release (0% TEGDMA: 0.261 ± 0.046 mml/L, DCPD_14: 0.203 ± 0.033 mmol/L, DCPD_22: 0.178 ± 0.030 mmol/L). How-

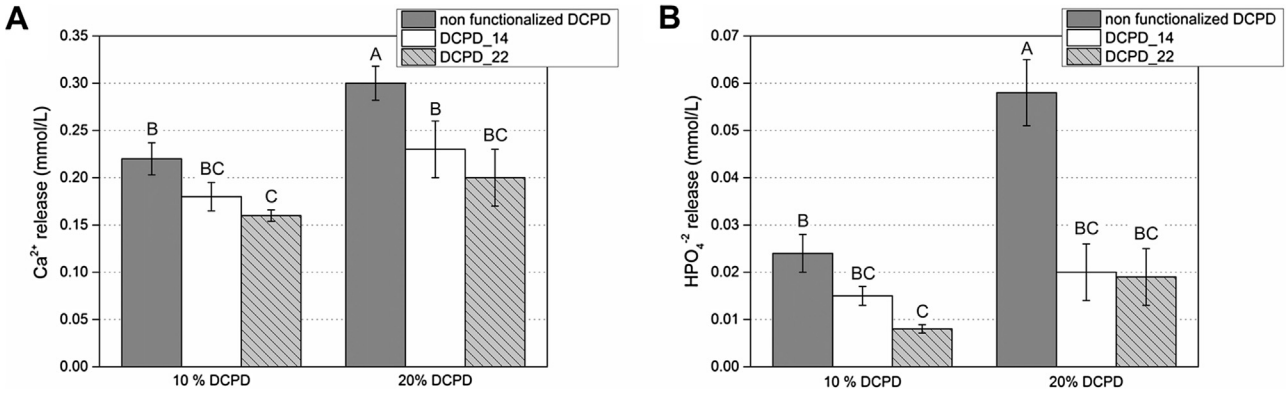

Fig. 5 – Ion release (in mmol/L) as a function of DCPD percentage and TEGDMA content (A: calcium, B: hydrogen phosphate). Same letters indicate lack of statistically significant differences ($p > 0.05$). Please, notice the difference in scale of y-axes between both graphics.

ever, at 10% DCPD, the difference between the material containing non-functionalized particles and the composite with DCPD_14 was not statistically significant.

Phosphate release after 7 days is shown in Fig. 5B. The interaction between bioactive filler level and TEGDMA content was statistically significant ($p < 0.001$). Only for composites with 10% DCPD, there was no difference in HPO_4^{2-} release between the materials containing non-functionalized and DCPD_14 particles. Regardless of the DCPD level, TEGDMA content in the particle did not significantly affect HPO_4^{2-} release.

4. Discussion

Materials containing CaP particles have demonstrated potential for remineralization of enamel and dentin *in vitro* [1,5,23,24]. However, since CaP particles are not cohesively strong nor have a chemical interaction with the resin matrix, they show a deleterious effect on the composite strength [13]. Functionalization did not contribute to reduce nanoparticles agglomeration. Therefore, in practical terms, they behave as "micro" particles or "nano-particles agglomerates", with plate-like morphology. Surface area values are low because the functionalizing layer was not removed prior to analysis, and TEGDMA fills the pores and surface irregularities of the CaP core. Non-functionalized and both functionalized DCPD particles presented agglomerates with similar median (D_{50}) sizes and particle size distribution. Since mechanical properties are influenced by the size of the agglomerates [25,26], this finding, though unforeseen, is useful to highlight the influence of particle functionalization on the strength results.

Composites containing DCPD_14 presented statistically higher DC compared to those containing non-functionalized particles. Increasing TEGDMA content in resin mixtures was shown to increase DC due to its small size and flexible structure [27,28]. Therefore, it could be hypothesized that TEGDMA-rich domains around functionalized DCPD particles were responsible for the increase in DC. Interestingly, this increase was slightly lower in composites containing DCPD_22. It can only be speculated that the effect of a higher mobility of the reactive species in TEGDMA-rich areas was offset by an increase in polymerization reaction termination through segmental diffusion, also associated with high TEGDMA fractions [29].

Composites containing DCPD presented significantly lower flexural strength after 24 h than the control. In agreement with previous investigations [12,13], increasing DCPD content from 10% to 20% also reduced strength, except for DCPD_14 where the reduction was only numerical. The lower strength observed with the addition of calcium orthophosphates particles occurs due to different reasons. First, while silanized glass particles increase composite fracture strength [30–32], calcium orthophosphate particles are nano-particles agglomerates with much lower cohesive strength than glass and, as such, they are much less effective as toughening agents. Second, due to the lack of a coupling interphase between DCPD and the organic matrix, these particles behave as large inclusions, increasing the risk of crack initiation at low stress levels [11,12,33–35].

Notwithstanding, functionalization of DCPD particles significantly reduced the detrimental effect resulting from the replacement the silanized glass by non-functionalized particles. Compared to the control (without DCPD), the difference in strength observed with a 10% replacement decreased from 41% (non-functionalized) to 17% (with DCPD_22). For a 20% replacement, the difference in strength with the addition of DCPD was reduced from 71% to 36% (with DCPD_14). The superior performance of composites containing functionalized DCPD can be explained by the incorporation of the TEGDMA monomers bonded to the DCPD core into the polymer network. Only for composites containing DCPD_14 (both at 10% and 20% levels), the gain in strength could also be attributed to their higher DC; however, the fact that composites with DCPD_22 did not present higher DC than those with non-functionalized particles highlights the importance of the functionalizing layer. It is interesting to point out that the amount of TEGDMA around the particle was not relevant for the strength of composites containing 20% DCPD. The higher DC reached by the composite with DCPD_14 associated with the low reinforcing filler content could explain the similarity in strength with the DCPD_22 composite.

After 28 days of storage in water, all materials showed statistically significant reductions in strength. Composites containing 10% DCPD presented similar strength reductions, between 26% and 32%. The lower strengths verified after prolonged water immersion can be attributed to hydrolytic degradation of the matrix, as well as partial dissolution of the CaP particles [36]. For composites with 20% DCPD, however, a more severe strength reduction after water immersion was observed with the use of functionalized DCPD (between 32% and 36%), in comparison to the material containing non-functionalized DCPD (21% reduction). A possible explanation for this finding involves the higher TEGDMA content in composites with a higher fraction of functionalized nanoparticles leading to the formation of a more heterogeneous network, which facilitates water sorption [37]. That, associated with TEGDMA relatively high hydrophilicity [38] would lead to increased composite degradation. Also noteworthy was the fact that composites containing 10% of functionalized DCPD presented strength values statistically similar to the control material after 28-day storage. Albeit encouraging, this finding must be taken carefully, as the control composite presented an unexpectedly high reduction in strength, when compared with a previous study testing the same formulation (24 h: 157 MPa; 28 days: 127 MPa) [13].

Clinically, a sustained ion release is desirable for a long-term caries inhibition effect. Therefore, it becomes important to know the kinetics of ion release over time. The available literature shows different release profiles, depending on the particle characteristics and content, pH of the immersion medium and hydrophilicity of the resin matrix [17,39]. In the present study, however, measurements were restricted to seven days, as the main focus was to verify if functionalization could impair ion release. In fact, ion release was influenced by DCPD level and functionalization. The higher ion release verified with composites containing a higher DCPD fraction agrees with previous studies [40,41]. Overall, the use of functionalized DCPD particles reduced ion release, suggesting that the interaction by chemisorption between TEGDMA and calcium in the DCPD structure [18] represents an extra hindrance for ions to be released from the DCPD crystalline structure. Notwithstanding, the composite containing 10 vol% of DCPD_14 released calcium and phosphate in levels similar to the composite containing non-functionalized particles and concentrations similar to those from previous reports (Ca^{2+}: 0.18 mmol/L; HPO_4^{2-}: 0.015 mmol/L) [13,42]. Ion release levels reported in *in vitro* remineralization studies (Ca^{2+}: 0.3–0.5 mmol/L; HPO_4^{2-}: 0.1–0.3 mmol/L) [7,24] are in the same range of those obtained with the composites containing 20 vol% of non-functionalized DCPD, but higher than those obtained with 10 vol% DCPD. Therefore, it is important to verify in future investigations if the concentrations released from materials containing functionalized DCPD particles are capable of promoting mineral recovery in initial caries lesions.

Silanization of calcium phosphate particles has demonstrated to negatively affect ion release because of the hydrophobic nature of silane coating [16,25]. On the other hand, the similar calcium release among non-functionalized DCPD and DCPD_14 could be explained by TEGDMA hydrophilic character [16,37,38]. HPO_4^{2-} release followed the same patterns displayed by Ca^{2+}. Lower hydrogen phosphate release values are in agreement with previous studies [12,39], which can be attributed to the phosphate tetrahedral structure, which is not easily removed from the crystalline lattice [13].

In conclusion, the use of TEGDMA-functionalized DCPD particles significantly improved the strength of ion-releasing composites, in relation to the use of non-functionalized DCPD. Among the tested materials, the composite containing 10% DCPD_14 showed the best compromise between fracture strength and ion release, including strength statistically similar to the control composite after 28 days in water. Further research is needed to evaluate the remineralization potential of composites containing TEGDMA-functionalized DCPD particles, as well as to optimize parameters such as filler size distribution and TEGDMA level in the functionalizing layer in order to improve mechanical and biological properties.

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