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Physicochemical and biological characterization of silica-coated alumina particles

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ABSTRACT

Objectives: A tribochemical silica-coating (TSC) method has been developed to improve the adhesion of dental resin composites to various substrates. The method utilizes airborne-particle abrasion using particles having a silica surface and an alumina core. The impact of the TSC method has been extensively studied but less attention has been paid to the characterization of the silica-modified alumina particles. Due to the role of silicate ions in cell biology, e.g. osteoblast function and bone mineralization, silica-modified alumina particles could also be potentially used as a biomaterial in scaffolds of tissue regeneration. Thus, we carried out detailed physicochemical characterization of the silica-modified alumina particles.

Methods: Silica-modified alumina particles (Rocatec, 3 M-ESPE) of an average particle size of 30 µm were studied for the phase composition, spectroscopic properties, surface morphology, dissolution, and the capability to modify the pH of an immersion solution. The control material was alumina without silica modification. Pre-osteoblastic MC3T3-E1 cells were used to assess cell viability in the presence of the particles. Cell viability was tested at 1, 3, 7 and 10 days of culture with various particle quantities. Multivariate ANOVA was used for statistical analyses.

Results: Minor quantities of silica enrichment was verified on the surface of alumina particles and the silica did not evenly cover the alumina surface. In the dissolution test, no change in the pH of the immersion solution was observed in the presence of the particles. Minor quantities of silicate ions were dissolved from the particles to the cell culture medium but no major differences were observed in the viability of pre-osteoblastic cells, whether the cells were cultured with silica-modified or plain alumina particles.

Significance: Characterization of silica-modified alumina particles demonstrated differences in the particle surface structure compared to control alumina. Dissolution of silica

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layer in Tris buffer or SBF solution varied from that of cell culture medium: minor quantities of dissolved Si were observed in cell culture test medium. The cell viability test did not shown significant differences between control alumina and its silica-modified counterpart.

1. Introduction

The development of minimally invasive dental treatments has given rise to demand for the improved adhesion of restorations to tooth substance. This in turn has led to a variety of physical and chemical conditioning methods of the substrates, which are used in so-called unibody restorations where the remaining tooth structure is restored using dental materials adhered to the tooth substrate [1]. Zirconia is an example of a substrate of indirectly made restoration, which requires special surface conditioning for adhesion to dental adhesives. It has relatively good physical properties but limited long-term adhesive properties to resin luting cements [2–5]. Many attempts have been made to improve the bonding of zirconia and other materials to resin composites. These include chemical modification of the substrate surface to contain more hydroxyl groups for better bonding and developing acidic primer systems tailored for specific dental prosthodontic systems [6,7]. Out of these, a tribochemical silica-coating (TSC) seems to be a good option to improve the bonding of several kinds of substrates to resin cements [8–10].

Applying TSC creates chemical bonds to the substrate material via kinetic energy. This is achieved by air-blown silica-modified alumina particles in optimized conditions, i.e. air pressure and working time [11]. In other words, TSC exposes the substrate to simultaneous chemical and mechanical effects. Consequently, the substrates surface is cleaned and obtains favorable wettability by increasing number of hydroxyl groups via silica entrapment on the surfaces [12,13]. At the end of the TSC process, the substrate is ready to be primed with silane coupling agents and bound to resin composites via a covalent polysiloxane network on the substrate [14].

Although TSC has been extensively studied [2,4,8,12, 15,16], less attention has been paid to detailed chemical characterization of the silica surface and the alumina core in the air-abrasion particle (Al_2O_3-SiO_2), used in the TSC. Chemical characterization and dissolution behavior of these particles are also of interest from the perspective of cell biological studies, where cell and tissue cultures are performed in the presence silica containing particles. Silicate ions have been reported to positively correlate with bone formation, metabolism, homeostasis, mineral density, and decreasing bone loss [17–19]. In addition, silica as sodium silicate nanoparticles has been reported to stimulate osteoblast differentiation [18,20] and inhibit osteoclast activity in cell culture studies [21]. The surfaces of certain orthopedic implants have even been treated with silica hydroxyapatite and tricalcium phosphate, although it is not known, whether the biological effects are due to silicate ions, other ions, or the materials' topography [19,22]. In addition, it has been observed that silica particles of different sizes can lead to different cell responses [19].

The aim of this study was to characterize silica-modified particles of Al_2O_3-SiO_2 used in TSC for their physicochemical properties, such as phase composition, spectroscopic properties, surface morphology, dissolution, and capability to modify the pH of the immersion solution. In addition, we studied the viability of preosteoblastic MC3T3-E1 cells in the presence of Al_2O_3-SiO_2 particles.

2. Materials and methods

2.1. Materials

Silica-modified alumina (Rocatec Soft, 3 M Espe, Germany) (Al_2O_3-SiO_2) particles with a grain size of 30 µm were studied. Based on the product information, Rocatec is aluminium oxide (> 95 %) coated with silica (1–5 %) and containing a low amount of sodium oxide (< 0.5 %). Alumina particles (Duralum White F320, Washington Mills, USA) with a diameter of 28–31 µm and composition of Al_2O_3 99.75 %, Na_2O 0.25 %, SiO_2, 0.02 %, Fe_2O_3 0.02 % were used as control material for chemical and cell culture analysis.

2.2. Cell culture

A mouse cell line MC3T3-E1, subclone 4, (ATCC, CRL2593) was cultured in phenol-red free Minimum Essential Medium (α-MEM, Gibco, 41061) with 10 % fetal bovine serum (FBS) (Gibco, 10270–106) and 1 % penicillin at 37 °C in a humidified atmosphere containing 5 % CO_2. At sub-confluency, the cells were trypsinized and plated on 96-well plates for viability assays. All experiments were performed with cells < passage 20 and cell culture medium was changed every 3–4 days.

2.3. XRD analysis

X-ray powder diffraction (XRD) was used to characterize the phase composition of the particles. The particles were characterized with a Bruker D8 Discover instrument (Bruker) with Cu Kα radiation (λ = 1.54 Å). The samples were measured in the 2-theta range 5°– 80°, using an increment of 0.04° and data collection of 0.2 s per step.

2.4. ATR-FTIR analysis

Attenuated total reflectance Fourier transform infrared spectroscopy (ATR-FTIR) was used to analyze the spectroscopic properties of the silica-modified alumina and alumina using a PerkinElmer Spectrum (Version 10.4.2) spectrometer.

The ATR-FTIR instrument averaged from 16 scans collected for wavelengths from 650 cm^{-1} to 2 500 cm^{-1} at 4 cm^{-1} resolutions. Analysis was done with the CPU32 Main 00.09.9934 22–4-2011 software and a UATR crystal combination diamond/ZnSe at triplicate of bounces was used.

2.5. *pH change in vitro*

The impact of silica-modified alumina and alumina on the pH of a fresh Tris buffer was studied in a continuous flow-through reactor. The pH change was also measured for simulated body fluid (SBF) and Tris buffer in a static system. The pH of the (50 mM) Tris buffer (Tris, 2-amino-2-hydroxymethyl-propane-1,3-diol, Trizma base, Sigma-Aldrich, pKa 8.06) was adjusted to 7.3 with 1 M HCl (J-T. Baker). The SBF solution was of a following composition: NaCl, $NaHCO_3$, KCl, $K_2HPO_4 \cdot 3\ H_2O$, HCl, $MgCl_2 \cdot 6\ H_2O$, $CaCl_2 \cdot 2\ H_2O$, Na_2SO_4, Tris buffer (Table 1.).

In the dynamic system, the solution was fed continuously (0.2 ml/min) through a particle bed at 40 °C. The sample chambers were filled with silica-modified alumina (0.33 g) or alumina [23]. The pH was measured for solution samples (4 ml) collected at the time points of 20, 40, 60, 80, 100, 120, 240, 480, and 1440 min. In the static system, tubes with silica-modified alumina particles (~ 75 g) and 50 ml Tris buffer or SBF were placed in an incubator (Orbital incubator, SI500, Stuart) and rotated at 100 rpm at 40 °C [24]. The pH was measured for samples at the same time points as in the dynamic system.

In both systems the pH was measured with a Mettler Toledo Seven-Easy electrode. For the ion analysis, 1 ml of the solution was diluted with ultrapure water (1:10) and acidified with concentrated HNO_3. The ion concentrations in the solutions were measured with inductively coupled plasma optical emission spectrometry (ICP-OES, PerkinElmer Optima 5300 DV, Waltham, MA, USA). The released ions were examined for silicon (LOQ 0.04 ppm, 251.611 nm), sodium (LOQ 0.2 ppm, 589.592 nm), calcium (LOQ 0.7 ppm, 393.366 nm), phosphorus (LOQ 0.03 ppm, 213.617 nm) and aluminium (LOQ 0.01 ppm, 396.153 nm).

2.6. *Ion release in vitro*

Si release from silica-modified alumina into α-MEM was measured by a colorimetric method. Silica-modified alumina particles were immersed in α-MEM (without penicillin or FBS) in five different concentrations (0, 0.3125, 0.625, 1.25, 2.5, 5 mg/ml) and incubated at 37 °C, 5 % CO_2. The samples were collected at 1, 3, 7 and 10 days. The solution samples were centrifuged (10 G, 5 min) and supernatant was pipetted into new test tubes and stored in the fridge (4°C) until colorimetric analyses. Si concentrations were analyzed by the molybdenum blue method [25] using dilutions of a Si standard ($(NH_4)_2SiF_6$ in H_2O, Certipur) in five concentrations (0, 0.1, 0.3, 0.5, 0.7, 1 mg/L). Samples were diluted (1:4) with ultra-pure water. Samples and standards were mixed with antimony phosphomolybdate complex, reduced with ascorbic acid, and finally, the silicomolybdate complex was adjusted by mixing 1-amino-2-naphthol-4-sulphonic acid, sulphate and tartaric

Table 1 – The reagents of SBF.

Reagent	NaCl	$NaHCO_3$	KCL	$K_2HPO_4 \cdot 3H_2O$	1M HCl	$MgCl_2 \cdot 6H_2O$	$CaCl_2 \cdot 2H_2O$	Na_2SO_4	TRIS
Manufacturer	VWR Chemicals	J.T. Baker	Sigma Aldrich	Sigma Aldrich	Sigma Aldrich	Sigma Aldrich	VWR Chemicals	Sigma Aldrich	Sigma Aldrich (Trizma® base)
Amount of reagent per 2 l	16 g	0.70 g	0.45 g	0.46 g	70 ml	0.61 g	0.74 g	0.14 g	12.11 g

acid. Absorbances at 820 nm were measured by a UV-1601 spectrophotometer (Shimadzu, Australia).

2.7. Morphology of Silica-modified alumina

Silica-modified alumina particles were investigated with a scanning electron microscope equipped with an energy-dispersive X-ray analyser (SEM-EDX) for their surface structure before and after static dissolution tests in Tris-buffered and SBF solutions (4 h and 24 h). For SEM examination, the particles were washed with ethanol and cast in epoxy resin. The electron beam was accelerated with voltages of 2.7 kV and the SEM used was a Leo Gemini 1530 (Carl Zeiss, Oberkochen, Germany).

2.8. Cell viability assay

For assessing the viability of preosteoblasts in the presence of silica-modified alumina or alumina particles, MC3T3-E1 cells were seeded at 5 000 cells/well in 96-well plates. The total volume of each well was 200 µl. The cells were adhered for one day before the medium was changed to the medium containing different concentrations of silica-modified alumina or alumina particles. Particles were sterilized in an autoclave (121°C, 20 min) and materials were suspended in cell culture medium right before the viability experiment was started. Five different quantities, i.e. 0.3125, 0.625, 1.25, 2.5, 5 mg/ml of both materials were used, and a control of 0 mg/ml was included. Cell viability at 1, 3, 7 and 10 days was determined by a WST method (Cell Counting Kit-8, CK04, Dojindo, 1:10 dilution) by measuring the absorbance at 450 nm (Thermo Scientific, Multiscan FC with SkanIt software for microplate readers, UI version 4.1.0.43). The background absorbance without any cells (silica-modified alumina or alumina particles at each quantity in cell culture medium) was also measured and subtracted from sample values, when viability results were analyzed. The viability test was repeated three times.

2.9. Statistical analysis

Statistical analysis was accomplished by using JMP pro 16.2.0 (570548). Differences in the cell viability with different concentrations of silica-modified alumina or alumina were compared to the control group at each time point. Statistical significance was analysed by using the nonparametric multiple comparisons. P-values ˂ 0.05 were considered statistically significant. Statistically significant differences between silica-modified alumina and alumina on same concentrations and day were analysed by using Wilcoxon test were statistically significant P-value were used Bonferroni correction.

3. Results

3.1. Phase analysis

XRD diffractograms of the silica-modified alumina and the alumina particles are shown in Fig. 1. The major crystalline phase in both samples was equivalent to the standard data for the alpha-Al_2O_3 (corundum) phase (ICDD card 00–010-0173) with a minor contribution from the beta-Al_2O_3 phase (ICDD card 00–051–0769). Both samples also display a broad hump in the 10–20 2theta region, which indicates an amorphous phase. The XRD data verifies no crystalline SiO_2.

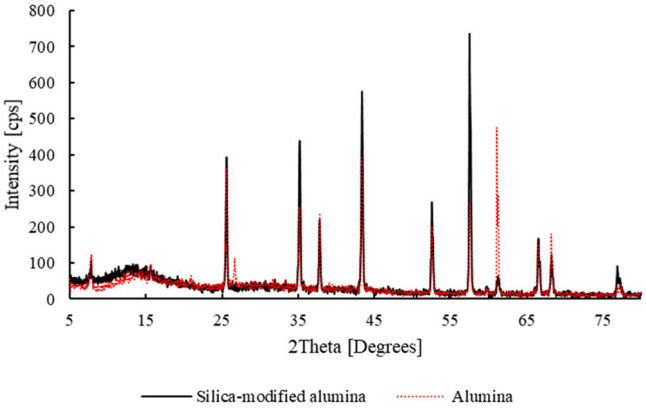


Fig. 1 – XRD patterns of the silica-modified alumina and alumina samples.

3.2. ATR-FTIR

ATR-FTIR spectra of the silica-modified alumina and alumina are shown in Fig. 2. The materials had wavenumber at 699 cm^{-1} (84 % transmittance) and 1095 cm^{-1} (97 % transmittance). No signs of silica on the alumina was found at wavenumber 650–2450 cm^{-1}. Typically signs of silica were found at wavenumbers 800 and 1 100 cm^{-1}.

3.3. pH change

The pH changes induced by the silica-modified alumina were measured in a static test in SBF and in continuous and static tests in Tris buffer (data not shown). In the SBF static dissolution test, the pH variation was minor, about 0.06 pH unit.

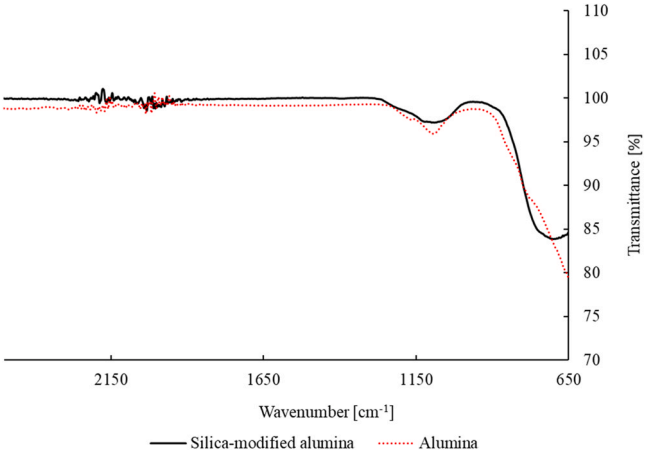


Fig. 2 – ATR-FTIR spectra of the silica-modified alumina and alumina samples.

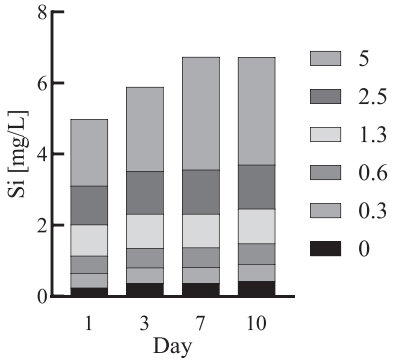


Fig. 3 – Si release to α-MEM with different particle concentrations at different immersion times.

In both the continuous and static systems with Tris buffer, the variation in pH was minor, only 0.04 and 0.07 pH units, respectively.

3.4. *Ion release*

In the continuous dissolution system, some Na, Ca, and P ions were released from the silica-modified alumina particles during the first two hours. No other ions were detected. In the static dissolution system into Tris-buffer, the ions were below the detection limits. In contrast, Si was released from the silica-modified alumina into the α-MEM at increasing particle concentration and with prolonged immersion (Fig. 3).

The Si released from the silica-modified alumina into α-MEM did not change during the dissolution period except for day 10, which showed different Si content during immersion time (Fig. 3).

3.5. *Surface morphology*

SEM images at different magnification of the silica-modified alumina particles are shown as-received and after 24 h of static dissolution in Tris buffer and SBF for magnifications of 1000 X, 5000 X and 25000 X, the surface areas indicated are 120×90 µm, 24×18 µm, 4×3.6 µm, respectively (Fig. 4). No changes of the particle morphology were visually seen in the SEM images before or after static dissolution tests regardless of dissolution time (0–24 h). SEM-EDXA of randomly selected silica-modified alumina particle areas gave silica concentrations between 0–42% indicating that the silica was not evenly covering the alumina surface (Fig. 5). The silica particles shown to be about 100 nm spherical nanoparticles above alumina (Fig. 4).

3.6. *Cell viability*

No differences were observed in the viability of pre-osteoblastic MC3T3-E1 cells between silica-modified alumina and alumina (Fig. 6). However, cell viability was significantly decreased ($P < 0.05$) in the presence of alumina at 5 mg/ml on day 10 ($P = 0.0066$) when compared to the control group on same day. Cells were also visually inspected under the light microscope during the 10-day culture period and no major effects on cell morphology were observed (data not shown).

4. Discussion

Materials containing silica or having silica-rich surfaces are used in dentistry because of several beneficial properties, including easily obtained adhesion to the hydroxyl group covered silica surface by using silane coupling agents [8,26]. In other contexts, such as in bone repair, filling materials or in vitro cell culture studies, silica as a source for silicate ions has been shown to influence osteoblast proliferation, differentiation and mineralization. However, the effects of silicate ions on osteogenic cells are not completely understood [19,27], although there are studies showing positive effects of zeolite and silicon-substituted calcium phosphate derived silicate ions to bone health [19]. On the other hand, when silicon-substituted calcium phosphates have been studied, the specific effects of silicate ions have been critically discussed because of lack of evidence of the resorption rates and release of silicate ions [28]. Thus, there is a clear need for studies on the dissolution behavior and effects of silica-containing minerals on cell biological parameters.

The ultrastructure of the silica-modified alumina was recently examined in another study, and a 50 nm thick silica layer was reported to cover the alumina core [14]. That study also showed small, silica agglomerates on the alumina surface in the SEM images. SEM-EDX analysis confirmed agglomerations to be silica. Detailed analysis of whether the silica was amorphous or crystallized was not performed. We also found agglomerates of randomly located silica in the present study but our analysis did not confirm that silica was amorphous and potentially prone to dissolution. From the perspective of the used chemical analysis methods and materials, we can conclude that the analyzed surfaces were not optimal because two materials overlapped. SEM examination of the silica-modified alumina particles after immersion in a continuous Tris buffer flow or static Tris buffer and SBF up to 24 h did not reveal any significant signs of dissolution. The ion concentration analyses of the Tris buffer and SBF solutions confirmed the SEM observations. Furthermore, the pH of the dissolution solutions did not change during the in vitro tests either. Interestingly, we however observed that the silicate ions dissolved into the cell culture medium (without FBS) of the cell culture study. The ion concentrations increased with particle amount and prolonged immersion. It should however be noted that the three different systems we used to study the ion dissolutions are not directly comparable. In the dynamic system, the experiment was performed in Tris buffer and in the static system in Tris buffer and SBF. The third system, where the particles were immersed in α-MEM, was also static but is still not directly comparable with SBF due to different solution ion compositions and static system methods between SBF and α-MEM.

Sodium metasilicate has previously been shown to affect the viability, differentiation, and mineralization of pre-osteoblastic cells [17]. Interestingly, in our study silica-modified alumina did not affect the viability of pre-osteoblastic cells, although our silica-modified alumina dissolution test gave

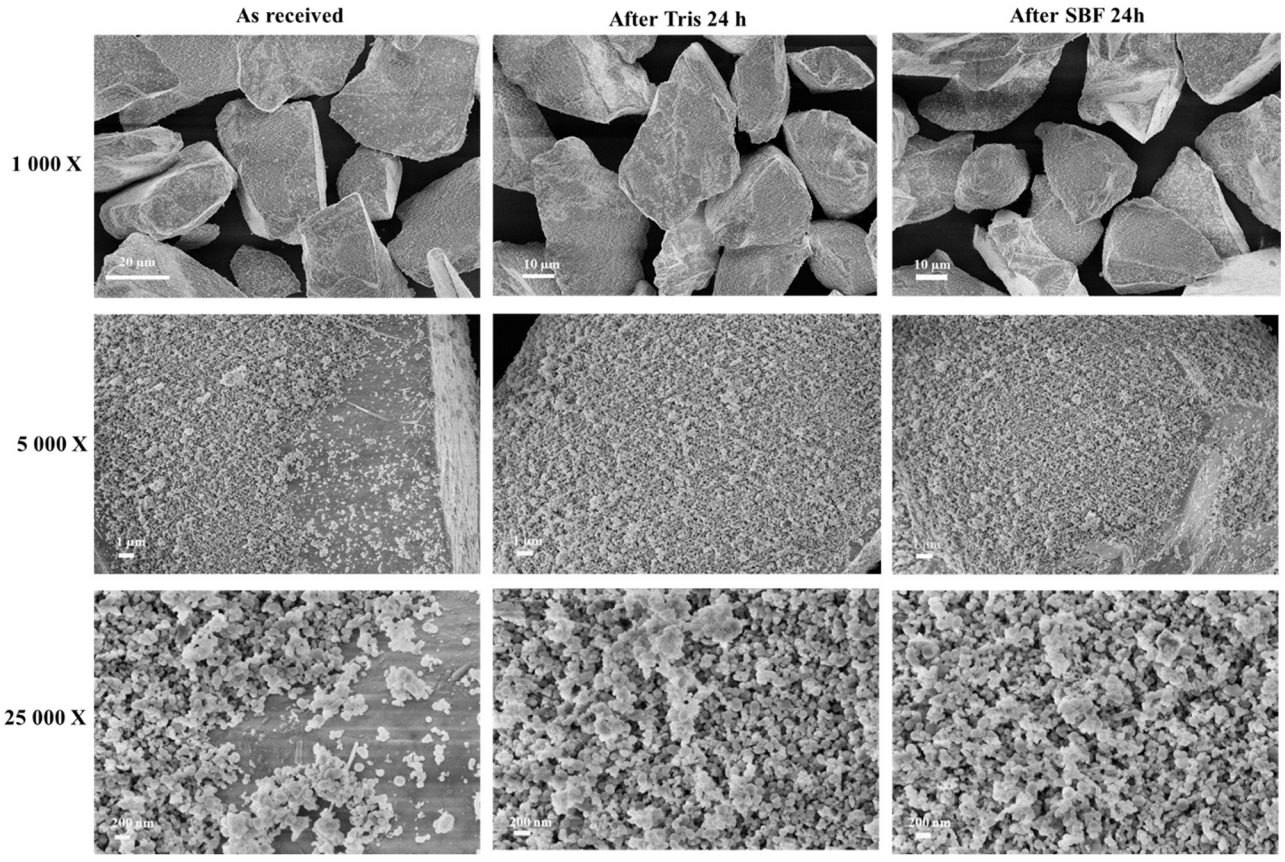
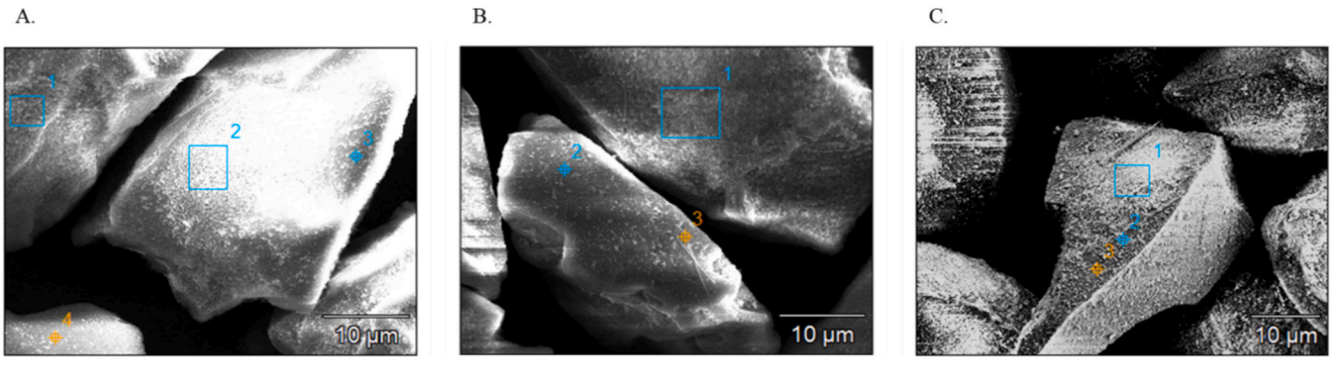


Fig. 4 – SEM images of the silica-modified alumina before ("As received") static dissolution test in the Tris-buffer solution and after the static dissolution test in Tris-buffer in SBF ("After Tris / SBF 24 h").



Surface	Amounts of compound [%]		
	O	Al	Si
1	42.59	44.92	12.5
2	49.17	39.19	11.64
3	50.87	49.13	
4	51.05	48.95	

Surface	Amounts of compound [%]		
	O	Al	Si
1	49.54	35.1	15.36
2	53.36	46.38	0.26
3	51.88	48.12	

Surface	Amounts of compound [%]			
	O	Na	Al	Si
1	53.29	1.25	3.16	42.29
2	51.6		48.4	
3	51.64		48.36	

Fig. 5 – Areas of SEM-EDX analysis of the silica-modified alumina surface for detecting presence of silica on the surface.

approximately the same silica concentration (0.5–2.5 mg/L) as was used in the previous study [17]. However, the release of silica from silica-modified alumina in α-MEM probably differed from the dissolution in cell culture conditions, where α-MEM contained FBS and cells were metabolically active. In addition, it should also be noted that previous studies have

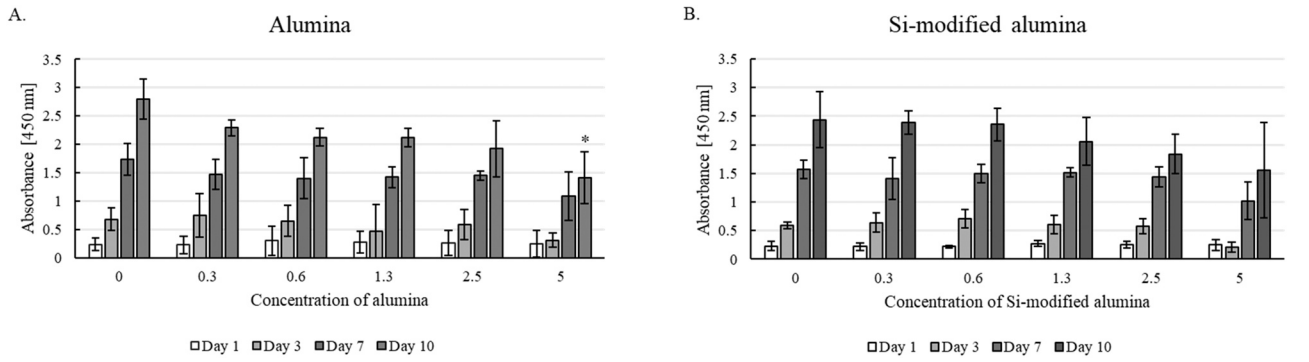


Fig. 6 – MC3T3-E1 cells viability as WST-methods (450 nm) with different concentrations of silica-modified alumina and alumina reference particles. Data are presented as an average of the repeated experiments (n = 3) with SD, *p < 0.01 vs. control group 0 mg/ml at respective time point.

been performed with aqueous sodium silicate solution, while we did the cell cultures with a biomaterial, i.e. silica-modified alumina.

In this study, no differences in cell viability were observed, whether the cells were grown with alumina or with silica-modified alumina particles. However, the silica could be the reason for decreased cell viability on day 10 in 5 mg/ml of alumina but not in silica-modified alumina.

Interestingly, in a previous study, silica nanoparticles with the same size (50 nm) as silica in our silica-modified alumina was shown to stimulate osteoblast differentiation and mineralization and to inhibit osteoclastogenesis. These silica nanoparticles did not affect MC3T3-E1 proliferation, which is in line with our result [20]. Silicon doping of alumina tubes was shown to stimulate cellular activity at the bone-tube interface in vivo but led to impaired osteogenic maturation within the tubes at 0.5 mol. % of silica, while osteogenesis in tubes was enhanced at 5 mol. % [29]. Taken together, this suggests that even though silica-modified alumina does not stimulate cell proliferation and viability in vitro, it might have positive effects on osteogenesis and mineralization in vivo. This warrants further investigation on the possibilities of silica-modified alumina as a material in bone research.

5. Conclusion

Characterization of silica-modified alumina particles demonstrated differences in the particle surface structure due to the presence of minor quantities of unevenly spread silica. Dissolution of the silica layer was not found to occur in Tris buffer or SBF solutions, but minor quantities of dissolved silicate ions were detected in α-MEM. However, the cell viability test did not demonstrate any differences between control alumina and its silica-modified counterpart.

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REFERENCES

[1] Vallittu PK, Durgesh BH, AlKheraif AAA, Hjerppe J. From body-on-frame to unibody constructions and designs mimicking biological structures – an overview. Eur J Oral Sci 2018;126:95–101.
[2] Galvão Ribeiro BR, Galvão Rabelo Caldas MR, Almeida AA, Fonseca RG, Adabo GL. Effect of surface treatments on repair with composite resin of a partially monoclinic phase transformed yttrium-stabilized tetragonal zirconia. J Prosthet Dent 2018;119(2):286–91.
[3] Gross C, Bergfeldt T, Fretwurst T, Rothweiler R, Nelson K, Stricker A. Elemental analysis of commercial zirconia dental implants - is “metal-free” devoid of metals? J Mech Behav Biomed Mater 2020:107.
[4] Hjerppe J, Närhi TO, Vallittu PK, Lassila LVJ. Surface roughness and the flexural and bend strength of zirconia after different surface treatments. J Prosthet Dent 2016;116(4):577–83.
[5] Heikkinen TT, Matinlinna JP, Vallittu PK, Lassila LVJ. Send orders for reprints to reprints@benthamscience [Internet]. Open Dent J 2013 (Available from: www.biomaterials.utu.fi).
[6] Thammajaruk P, Inokoshi M, Chong S, Guazzato M. Bonding of composite cements to zirconia: a systematic review and meta-analysis of in vitro studies. J Mech Behav Biomed Mater Elsevier Ltd 2018;80:258–68.
[7] Ruyter EI, Vajeeston N, Knarvang T, Kvam K. A novel etching technique for surface treatment of zirconia ceramics to improve adhesion of resin-based luting cements. Acta Biomater Odontol Scand 2017;3(1):36–46.
[8] Khan AA, Mohamed BA, Mirza EH, Syed J, Divakar DD, Vallittu PK. Surface wettability and nano roughness at different grit blasting operational pressures and their effects on resin cement to zirconia adhesion. Dent Mater J 2019;38(3):388–95.
[9] Matinlinna JP, Lassila LV, Özcan M, Yli-Urpo A, Vallittu PK. An introduction to silanes and their clinical applications in dentistry. Int J Prosthodont 2004;17(2):155–64.
[10] Özcan M, Vallittu PK. Effect of surface conditioning methods on the bond strength of luting cement to ceramics. Dent Mater 2003;19(8):725–31.
[11] Heikkinen TT, Lassila LVJ, Matinlinna JP, Vallittu PK. Effect of operating air pressure on tribochemical silica-coating. Acta Odontol Scand 2007;65(4):241–8.
[12] Martins SB, Abi-Rached F de O, Adabo GL, Baldissara P, Fonseca RG. Influence of particle and air-abrasion moment

on Y-TZP surface characterization and bond strength. J Prosthodont 2019;28(1):e271–8.
[13] Thammajaruk P, Buranadham S, Thanatvarakorn O, Guazzato M. Influence of nano-structured alumina coating on composite-zirconia bonding and its characterization. J Adhes Dent 2018;20(3):233–42.
[14] Nagaoka N, Yoshihara K, Tamada Y, Yoshida Y, Meerbeek B van. Ultrastructure and bonding properties of tribochemical silica-coated zirconia. Dent Mater J 2019;38(1):107–13.
[15] Abi-Rached FO, Martins SB, Campos JA, Fonseca RG. Evaluation of roughness, wettability, and morphology of an yttria-stabilized tetragonal zirconia polycrystal ceramic after different airborne-particle abrasion protocols. J Prosthet Dent 2014;112(6):1385–91.
[16] Özcan M, Melo RM, Souza ROA, Machado JPB, Felipe Valandro L, Botttino MA. Effect of air-particle abrasion protocols on the biaxial flexural strength, surface characteristics and phase transformation of zirconia after cyclic loading. J Mech Behav Biomed Mater 2013;20:19–28.
[17] Kim EJ, Bu SY, Sung MK, Choi MK. Effects of silicon on osteoblast activity and bone mineralization of MC3T3-E1 cells. Biol Trace Elem Res 2013;152(1):105–12.
[18] Uribe P, Johansson A, Jugdaohsingh R, Powell JJ, Magnusson C, Davila M, et al. Soluble silica stimulates osteogenic differentiation and gap junction communication in human dental follicle cells. Sci Rep 2020;10(1).
[19] Zhou X, Zhang N, Mankoci S, Sahai N. Silicates in orthopedics and bone tissue engineering materials. J Biomed Mater Res - Part A John Wiley Sons Inc 2017;105:2090–102.
[20] Beck GR, Ha SW, Camalier CE, Yamaguchi M, Li Y, Lee JK, et al. Bioactive silica-based nanoparticles stimulate bone-forming osteoblasts, suppress bone-resorbing osteoclasts, and enhance bone mineral density in vivo. Nanomed: Nanotechnol Biol Med 2012;8(6):793–803.
[21] Mladenović Ž, Johansson A, Willman B, Shahabi K, Björn E, Ransjö M. Soluble silica inhibits osteoclast formation and bone resorption in vitro. Acta Biomater 2014;10(1):406–18.
[22] Bohner M, Miron RJ. A proposed mechanism for material-induced heterotopic ossification. Materials Today vol. 22. Elsevier B.V; 2019. p. 132–41.
[23] Fagerlund S, Ek P, Hupa L, Hupa M. Dissolution kinetics of a bioactive glass by continuous measurement. J Am Ceram Soc 2012;95(10):3130–7.
[24] Maçon ALB, Kim TB, Valliant EM, Goetschius K, Brow RK, Day DE, et al. A unified in vitro evaluation for apatite-forming ability of bioactive glasses and their variants. J Mater Sci: Mater Med 2015;26(2):1–10.
[25] Ballo AM, Kokkari AK, Meretoja V v, Lassila LL, Vallittu PK, Narhi TO. Osteoblast proliferation and maturation on bioactive fiber-reinforced composite surface. J Mater Sci: Mater Med 2008;19(10):3169–77.
[26] Matinlinna J.P., Lassila L. Article in The International journal of prosthodontics [Internet]. 2004. Available from: ⟨https://www.researchgate.net/publication/8585276⟩.
[27] Wang W, Yeung KWK. Bone grafts and biomaterials substitutes for bone defect repair: a review. Bioact Mater KeAi Commun Co 2017;2:224–47.
[28] Bohner M. Silicon-substituted calcium phosphates - a critical view. Biomaterials 2009;30(32):6403–6.
[29] Pabbruwe MB, Standard OC, Sorrell CC, Howlett CR. Effect of silicon doping on bone formation within alumina porous domains. J Biomed Mater Res - Part A 2004;71(2):250–7.