ORIGINAL RESEARCH

Novel Ag nanocrystals based dental resin composites with enhanced mechanical and antibacterial properties

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Received 25 April 2013; accepted 1 November 2013
Available online 5 December 2013

KEYWORDS
Dental resin composite; Ag nanocrystals; Mechanical properties; Antibacterial property

Abstract The aim of this work was to investigate the effect of trace addition of oleic acid coated Ag nanocrystals (Ag NCs) on mechanical and antibacterial properties of dental resin composites. Composites (70 wt% of silica loading) with 25 ppm, 50 ppm, 75 ppm and 100 ppm (wt) Ag NCs were prepared and the composite without Ag NCs served as a control. The experimental results showed that the addition of Ag NCs significantly improved the strength and modulus of the resin composite without compromising the shade. For the composite with 50 ppm Ag NCs, flexural strength (140.3 MPa), modulus (13.2 GPa) and compressive strength (347.2 MPa) were increased by 4.1%, 22.2%, 13.3%, respectively, compared with the control. The antibacterial test demonstrated that trace Ag NCs provided the resin composites with an antibacterial effect. Such strong and antibacterial dental resin composites might be advantageous to prevent secondary caries and be potential for future clinical applications.

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1. Introduction

Resin composites, as most popular dental restorative materials, possess excellent properties such as: good esthetic properties, acceptable mechanical properties, fast restoration, etc. [1]. Long-lasting and high-effective performance of a restorative resin composite is always expected in clinical applications. It is widely believed that restoration failure is mainly related to the mechanical properties of composites and the secondary caries [2]. In the past decades, the development of hybrid filling technology and novel reinforcing-toughening fillers facilitated significant improvement of strength, modulus and wear-resistance of resin composites [3,4], reducing the restoration failure effectively. However, micro-chinks and micro leakage are still inevitable which provide places for absorption and proliferation of bacteria, leading to secondary caries subsequently [5,6]. Though increasing filler loading and
introducing novel low-shrinkage monomers decreased polymerization shrinkage [7,8], bacterial breeding was still not prevented effectively. Therefore, more attention was given to antibacterial dental resin composites.

The antibacterial effect of dental resin composite is often realized by altering the resin matrix and the filler, such as modification of the polymer, incorporation of the filler with an organic or inorganic antibacterial agent [9,10]. Organic antibacterial agents provide a rapid and strong antibacterial effect, but easy migration, poor heat resistance and toxicity might limit their applications in dental composites [11]. Inorganic antibacterial agents have better compatibility, long-lasting and wide broad-spectrum antibacterial properties, and Ag-based agent is one of the most commonly used inorganic antibacterial agents. For dental composites, usage of silver-zeolite, silver-apatite and silver-supported zirconium phosphate had been reported and good antibacterial effects were achieved [12–15]. Although higher amount of addition and higher specific surface area of an Ag-based agent improved the antibacterial properties, the subsequent poor dispersion in the composite and unsatisfied color stability would adversely influence the physical and mechanical properties of the resin composite.

On the basis of above considerations, organic-functionalized silver nanoparticles, possessing improved compatibility with organic components and high antibacterial performance due to a higher specific surface area, might be a superior antibacterial agent to realize better dispersion and combination in the composite and achieve long-lasting antibacterial performance. To date, such nano hybrid silver particles have rarely been applied into dental composite. In our previous work [16], oleic acid coated Ag NCs with novel properties was studied and herein, we employed the Ag NCs into dental resin composite. Owing to its higher specific surface area and excellent dispersion in organic solvent, small amounts of Ag NCs was supposed to realize better antibacterial function of the resin composite without compromising the shade. Moreover, Ag NCs with double bonds might enhance the filler packing density and interfacial interaction. Therefore, the effect of trace addition of Ag NCs on the mechanical and antibacterial properties of resin composite was investigated.

2. Experimental

2.1. Materials

Bisphenol A glycidyl methacrylate (Bis-GMA) and triethylene glycol dimethacrylate (TEGDMA) were obtained from Sigma-Aldrich. Camphorquinone (CQ), 4-dimethylamino-benzoate (EDMAB) and γ-methacryloyloxypropyltrimethoxy silane (γ-MPS) were purchased from J&k Scientific Ltd. Propylamine and cyclohexane were obtained from Sinopharm Chemical Reagent Co., Ltd. Spherical micro-silica (average size 1 μm) particles were obtained from Zhejiang Tongda Weipeng Electric Co., Ltd and nano-silica (Aerosil OX50, average size 40 nm) from Degussa. All materials were of analytical grade and used as received without further purification.

Micro and nano silica particles were silanized respectively following a typical method [17] as shown in Fig. 1 to improve their compatibility and dispersion in the resin matrix. The silanized silica particles were characterized by fourier transform infrared spectroscopy (FTIR) and thermogravimetric analysis (TGA). Ag NCs was synthesized following the reported method [16] and characterized by transmission electron microscopy (TEM) and FTIR. According to the Ag NCs and silica loadings added in the resin composites, silanized silica (micro/nano, 55/15) [18] and Ag NCs at different mass ratios (e.g. silica loading 70 wt%, and addition of Ag NCs 100 ppm wt) were represented by silica/Ag NCs 7/0.001) were dispersed in cyclohexane by ultrasonic and then the solvent was removed to obtain functional inorganic mixtures, as illustrated in Fig.1.

2.2. Fabrication of resin composites

The resin composites containing 70 wt% of silica, 30 wt% of resin base (Bis-GMA/TEGDMA 49.5/49.5 wt%, CQ/EDMAB 0.2/0.8 wt%) and Ag NCs (0, 25, 50, 75, 100 ppm wt) were fabricated following a procedure as shown in our previous work [19,20]. The obtained pastes were placed in oven under vacuum at room temperature for 8 h to remove air bubbles and then maintained in a refrigerator (4 °C).

2.3. Characterization

2.3.1. Degree of conversion

Degree of conversion of the monomers in the resin composite was evaluated by FTIR spectroscopy with a universal method as reported [21]. Each thin specimen was light-cured with a LED curing unit (blue light, 470 nm, SLC-VIII B Hangzhou Sifang Medical Apparatus Co., Ltd., Zhejiang, China) for 120 s. The aliphatic C¼C peak at 1637 cm⁻¹ and aromatic carbon–carbon peak at 1608 cm⁻¹ before and after curing were used to calculate the conversion, according to the following equation:

\[
\text{Degree of conversion} \% = \left[ 1 - \frac{(A_{1637}/A_{1608})_{\text{polymer}}}{(A_{1637}/A_{1608})_{\text{monomer}}} \right] \times 100
\]

Fig. 1 Schematic of the preparation of resin composites containing Ag nanocrystals.
where \( \frac{A_{1637}}{A_{1608}} \text{polymer} \) means the ratio of the absorbance peaks at 1637 cm\(^{-1}\) and 1608 cm\(^{-1}\) of cured specimen, and \( \frac{A_{1637}}{A_{1608}} \text{monomer} \) is the ratio of the absorbance peaks at 1637 cm\(^{-1}\) and 1608 cm\(^{-1}\) of uncured specimen.

2.3.2. Mechanical properties and morphology of the fracture surfaces

The mechanical properties of the resin composites were measured by a universal testing machine (Instron 5900, USA). Specimen preparation and test procedure were based on the method described in ANSI/ADA Specification No. 27-2009 (ISO 4049). Six rectangular-shaped specimens (25 mm \( \times \) 2 mm \( \times \) 2 mm) were used for three-point bending test (span distance 20 mm, cross-head speed 0.75 mm/min) and six disc-shaped specimens (Φ 4 mm \( \times \) 6 mm) for compressive test (loading rate 1 mm/min). Each specimen in the mold was light-cured for 120 s over both sides. All specimens were polished using a sandpaper of grit number 1500# before mechanical testing. The fracture surfaces of the resin composites after three-point bending test were observed by FE-SEM.

2.3.3. Antibacterial properties

The antibacterial property of the cured composites was tested via both quantitative analysis in a liquid medium and qualitative analysis on a solid agar disc-diffusion plate.

In the qualitative analysis, the well-distributed Escherichia coli suspension (10 \( \mu \)L) was first spread onto the agar plate (diameter 10 cm) and the disc samples (15 mm \( \times \) 1 mm) of the cured composite were pasted onto the agar plate. All the samples were sterilized for 3 h by UV radiation before they were pasted. The bacteria were then incubated at 37 °C for 12 h and the inhibition zone for each sample on the plate was visually inspected. In the quantitative analysis, Escherichia coli, Staphylococcus aureus, and Lactobacillus were added into a 5 mL liquid medium with an OD value of 0.1 to 0.2 at 625 nm for each test tube, then disc sample (8 mm \( \times \) 1 mm, \( n = 3 \)) of the cured composite was added into each test tube. For comparison, the tube without the cured composite was used as a control. All the treated bacterial samples were then incubated at 37 °C with gentle shaking (100 rpm), and the absorbance at 625 nm was monitored using the Lambda 25 UV–vis spectrophotometer after 12 h of incubation.

The percentage of bacterial inhibition was calculated by the following equation:

\[
\text{Inhibitory rate (\%)} = \left( \frac{\text{OD}_{\text{cont}} - \text{OD}_{\text{expe}}}{\text{OD}_{\text{cont}}} \right) \times 100
\]  

where \( \text{OD}_{\text{expe}} \) (experimental group) and \( \text{OD}_{\text{cont}} \) (control group) are the average ODs of the experimental group and the control group, respectively.

3. Results and discussion

3.1. Analysis of silanized silica and Ag NCs

The FTIR spectra of the untreated silica and the silanized silica shown in Fig. 2(a) illustrate the characteristic peaks of \( \gamma \)-MPS at 1720 cm\(^{-1}\) (C = O) and 1637 cm\(^{-1}\) (C = C). Fig. 2(b) shows the TGA curves of the untreated silica and the silanized silica. The graft ratio was calculated to be 2.9 wt% for the micro-silica and 3.8 wt% for nano-silica, respectively. Both FTIR and TGA results confirmed that the \( \gamma \)-MPS was covalently attached on the surface of silica particles successfully.

The morphology of synthetic Ag NCs was spherical with an average of 6 nm in diameter as shown in Fig. 3 [16]. For the obtained Ag NCs in the FTIR analysis (Fig. 4), the characteristic peaks at 2922 cm\(^{-1}\) and 2841 cm\(^{-1}\) were C–H stretching of CH\(_2\) groups, and peak at 3006 cm\(^{-1}\) was the C = C stretching of oleic acid. N–H stretching around 3180–3500 cm\(^{-1}\) and at 1631 cm\(^{-1}\) and asymmetric –COO\(^-\) stretching of the acid-base complex (–\(\text{COO}^-\text{NH}_3^+\)) at 1534 cm\(^{-1}\) were attributed to the reaction of oleic acid with the alkylamine, which proved the successful synthesis of oleic acid coated Ag NCs [16]. With C=C bonds and the organic layer on the surface, Ag NCs would be inclined to serve as cross-linking points during photo-polymerization and enhance the interfacial compatibility and combination.

![Fig. 2](image)

**Fig. 2** FTIR (a) and TGA (b) analysis of silanized silica: curve 1, untreated SiO\(_2\); curve 2, silanized micro SiO\(_2\); curve 3, silanized nano SiO\(_2\).
3.2. Degree of conversion and mechanical properties of the resin composites

The degree of conversion was influenced by factors such as monomer activity, light strength, curing time, characteristics of the filler, shade of the composite, etc. [22]. In this study, the addition of Ag NCs decreased the mean conversion values of the resin composites from 61.6% to 55.8% (Fig. 5(a)). With a higher amount of dark purple Ag NCs, the shade of uncured composite paste turned to light yellow and the cured composite changed to light brown (Fig. 5(b)). The dark Ag NCs and the shade of the composite might influence the transmission efficiency of blue-light [23,24] and have adverse effect on the photo-polymerization of monomers, leading to more unreacted monomers and relatively incomplete polymer network consequently.

As shown in Fig. 6, mechanical properties of the resin composites were improved by the addition of Ag NCs. In the case of the composite with 50 ppm (wt) Ag NCs, the flexural strength (140.3 MPa), modulus (13.2 GPa) and compressive strength (347.2 MPa) were increased by 4.1%, 22.2%, 13.3%, respectively, compared with the composite without Ag NCs. Mechanical properties of dental resin composite containing Ag nanoparticles or silver-support agent have been investigated previously [25–27], however, the flexural strength (64–118 MPa) and compressive strength (94–264 MPa) were undesirable, which might possibly be resulted from the method of composite fabrication, dispersion quality of filler and untibacterial agent, and the interaction among components in the composite. The Ag NCs (5–10 nm) used in this study were modified by oleic acids with C=C double bonds. By solution mixing method as described in 2.1., Ag NCs could disperse well in the composite and have a good compatibility with resin matrix. On one hand, the C=C double bonds provided more cross-linking points and they might form covalent bonds with the matrix to enhance a multiphase combination. On the other hand, as small volume filler, Ag NCs could fill voids of the composite to improve the packing density. As a result, the mechanical properties of the composite with Ag NCs were improved. However, when more Ag NCs were added, the decrease of conversion might affect the formation of the matrix
network and decrease the strength and the modulus consequently. And this agreed well with the results as previously reported [25,26].

The morphologies of the fracture surface of the resin composites after three-point bending test were observed by FE-SEM. The images showed that the fracture surface of the composite without Ag NCs (Fig. 7(a)) was relatively flat and smooth. By comparison, the fracture surface of the composites with 50 ppm wt (Fig. 7(b)) and 100 ppm wt (Fig. 7(c)) exhibited relatively more and larger fracture steps, which might prove the enhanced multiphase interaction. Rougher fracture surfaces with a higher surface area, would consume more energy when new surface was created and facilitate stress propagation and deflection, therefore, improving the strength and modulus [28,29].

3.3. Antibacterial effect of the resin composite

As shown in Fig. 8, the inhibition zone of the resin composites with Ag NCs was observed and the zone of the composite with 100 ppm Ag NCs was relatively larger and clear, implying that the Ag NCs provided the resin composite with an antibacterial effect. Meanwhile, further quantitative analysis confirmed the inference, which was that the Ag NCs in the composite inhibited bacteria growth effectively and higher addition of Ag NCs enhanced the antibacterial performance as shown in Table 1. Since the oleic acid coated Ag NCs had better compatibility and combination with the resin matrix as analyzed above, Ag NCs served as immobilized antibacterial molecules to a greater extent [9]. The antibacterial effect might be not due to the release of silver ion but mostly to the activation of oxygen based on the catalytic action of silver [26], so the effect was not able to reach the farther area around the composite, but the accumulation of activated oxygen would reduce the bacteria on the surface. And that may be the reason why the Ag NCs filled composite studied possessed relatively lower inhibition rate than Ag-releasing filled composites. However, long-lasting antibacterial effect of the composite might be advantageous in clinical application and would be researched in latter work.

4. Conclusions

(1) The addition of Ag NCs into the dental resin composite enhanced the mechanical properties. The composite with 50 ppm (wt) Ag NCs possessed improved flexural strength (140.3 MPa), modulus (13.2 GPa) and compressive strength (347.2 MPa), which were increased by 4.1%, 22.2% and 13.3%, respectively, compared with the composite without Ag NCs. As more Ag NCs were added, the composite was discolored and the mechanical properties were inclined to decrease.

(2) The addition of Ag NCs provided the resin composite with an antibacterial effect. And superior performance could be achieved as more Ag NCs were added. Long-lasting antibacterial dental resin composite without compromising the mechanical properties could be fabricated and might be applied in future clinical applications.

Acknowledgements

This work was financially supported by the National High-Tech Research and Development Program of China (2012AA030309),

Table 1 Antibacterial performance of the cured resin composite with different amounts of Ag NCs by a liquid tube method.

<table>
<thead>
<tr>
<th>Bacterial</th>
<th>Composite without Ag NCs (%)</th>
<th>Composite with 50 ppm Ag NCs (%)</th>
<th>Composite with 100 ppm Ag NCs (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Staphylococcus aureus</td>
<td>1.1</td>
<td>51.2</td>
<td>64.6</td>
</tr>
<tr>
<td>Lactobacillus</td>
<td>0.8</td>
<td>57.9</td>
<td>66.5</td>
</tr>
<tr>
<td><em>Escherichia coli</em></td>
<td>2.1</td>
<td>54.8</td>
<td>69.1</td>
</tr>
</tbody>
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Program for Changjiang Scholars and Innovative Research Team in University (IRT1221), the National Natural Science Foundation for Distinguished Young Scholar of China (No. 50925312), the Shanghai Nano Special Projects (0952nm05600) as well as Doctoral Innovation Foundation of University (CUSF-DH-D-2013005).

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