Research Article

Bovine Serum Albumin and Chitosan Coated Silver Nanoparticles and Its Antimicrobial Activity against Oral and Nonoral Bacteria

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Antimicrobial agents have been developed for drug-resistance infections, which have been rapidly increasing; however, the control of involved microorganisms is still a challenge. In this work, SNP with bovine serum albumin (BSA) and chitosan (CS) coatings were prepared with an aqueous reduction method, characterized using dispersion light scattering, transmission electron microscopy, and thermal analysis. Antibacterial activity was tested on seven oral and nonoral bacteria by microdilution test and scanning electron microscopy. Six different sizes and shapes of coated SNP were prepared and used. Characterization revealed narrow size and good distribution of particles, spherical and pseudospherical shapes, and the presence of coatings on the SNP surfaces. All samples showed antimicrobial activity, although smaller sizes and CS samples had the best inhibition effects. The highest microbial resistance was shown by Gram-positive bacteria. Although coated SNP action depends on particular bacterium, BSA and CS coated SNP could be used for drug-resistance infections.

1. Introduction

Nowadays the increase in bacterial drug resistance is considered a serious health problem concern worldwide. Multiple antibiotic-resistant bacteria are rapidly developing resistance to clinically useful antimicrobials and making them ineffective, reaching alarming limits to antimicrobial therapeutics

[1]. *Staphylococcus aureus* (*S. aureus*) has been one of the most frequent drug-resistant bacteria in infections acquired from nosocomial institutions and community, including superficial skin and other soft tissue infections, toxic shock, pneumonia, endocarditis, and septicemia [2]. *Escherichia coli* (*E. coli*) is the most common pathogen causing serious intestinal and extraintestinal infections; it possesses several

virulence factors that are necessary for resistance, survival, and colonization of the bacteria, overcoming host defenses [3]. *Enterococcus faecalis* (*E. faecalis*) is also one of the most common bacteria found in clinical setting, responsible for approximately 80% of enterococcal infections; it has been associated with endodontic infections after root canal treatments showing multidrug resistance [4, 5]. Moreover, *Streptococcus mutans* (*S. mutans*) and *Streptococcus sobrinus* (*S. sobrinus*) have been considered as principal oral pathogens involved in the beginning and development of dental caries [6]; they have shown specific levels of bacterial resistance to antibiotics [7].

Several improved treatments for prevention and control of the microorganisms described above have been relevantly successful protocols in many countries of the world [3, 8, 9] although other alternative approaches of treatments should be investigated for microbial resistance. An option is to use silver nanoparticles (SNP) due to their great bactericidal effect in a wide range of microorganisms including drug-resistant bacteria [10, 11]. It has been reported that SNP could show toxic effects in human cells depending on their physical and chemical properties [12, 13]; therefore, several studies have designed new nanostructured materials using biocompatible components [14]. Different organic materials have been employed to modify nanoparticles properties through surface modification [15]. Two of them are bovine serum albumin (BSA) and chitosan (CS); the former is a protein with a wide range of physiological functions [16] and the latter is a biopolymer with very important biological properties [17]. Various studies have successfully designed new and novel nanomaterials using different methods for green synthesis [18, 19]; however, limited information of the antibacterial activity of SNP with BSA and CS coatings on drug-resistant microorganisms is available considering SNP as relevant antimicrobial agents for potential medical uses. In the present work, different sizes and shapes of coated SNP using BSA and CS compounds were prepared and characterized. Antimicrobial activity of the coated SNP was determined by minimal inhibitory concentrations (MIC) using *S. aureus*, *E. coli*, *E. faecalis*, *S. mutans*, *S. sobrinus*, *S. oralis*, and *S. salivarius* strains.

2. Materials and Methods

2.1. Materials and Reagents. Silver nitrate (AgNO_3 , CTR Scientific), bovine serum albumin (BSA, Sigma-Aldrich), chitosan (low molecular weight 85%, Sigma-Aldrich), gallic acid ($\text{C}_7\text{H}_6\text{O}_5$, Sigma-Aldrich), acetic acid ($\text{C}_2\text{H}_4\text{O}_2$, 99.7%, J. T. Baker), brain-heart infusion (BHI, DIFCO Laboratories, Detroit, MI, USA), *S. aureus* (ATCC, 29213), *E. coli* (ATCC, 25922), *E. faecalis* (ATCC, 29212), *S. mutans* (ATCC, 25175), *S. sobrinus* (ATCC, 27351), *S. oralis* (ATCC, 10557), and *S. salivarius* (NCTC, 8618) strains were purchased, used, and stored according to manufacturer's recommendations. All used reagents were of analytical grades. Deionized water (18 M) was prepared using a Milli-Q Biocel Purification System.

2.2. Silver Nanoparticles Preparation. All preparations started with a 0.01 M AgNO_3 solution were placed in a 250 mL

reaction vessel. Under magnetic stirring, 10 mL of deionized water containing gallic acid (0.1 g for first size sample and 0.5 g for second size sample) was added to 100 mL Ag^+ solution. Afterwards, solution's pH value was immediately adjusted (for first size sample the pH was raised to 11 with NaOH 1.0 M and for second size sample pH was raised to 10 with NH_4OH) [20]. Later, first and second size suspensions were agglomerated using nitric acid (63%) to reach a pH 1.5; all suspensions were filtrated using nitrocellulose filter (Millipore, 0.1 μm pore) in a vacuum filter flask and SNP suspensions on the filter were washed several times with deionized water to reach pH 8; finally, membranes with SNP solutions were redispersed and diluted to a metered volume. SNP with pseudospherical shape and big size were also synthesized. This preparation began with a 0.01 M AgNO_3 solution placed in a 250 mL reaction vessel; under magnetic stirring, gallic acid was added to 100 mL of Ag^+ solution and immediately was irradiated with UV light (254 nm, 15 W) during 30 min (pH was not modified), and it was heated during 30 min at 80°C [20]. Finally, solution was centrifuged at 9000 rpm for 10 minutes (Biofuge, USA) and washed three times to discard supernatants; particles were redispersed and diluted to a meter volume with deionized water.

2.2.1. Preparation of BSA Coated SNP. A solution containing 1% (wt/v) of BSA was made using deionized water. Three different sizes of SNP and 1% of BSA solutions were mixed in tubes of 50 mL according to the following proportions (SNP/BSA): 7 : 1 for first sample; 14 : 1 for second sample, and 7 : 1 for third sample, respectively. Each solution was mixed for 12 h on an electronic mixer at environmental temperature.

2.2.2. Preparation of CS Coated SNP. CS solutions with different concentrations of acetic acid were prepared by dissolving purified CS in 1%, 0.5%, and 0.25% acetic acid solutions to reach a concentration of 1% (wt/v) under magnetic stirring for 12 h until solutions were transparent. Once dissolved, each solution was placed in different proportions (SNP/CS). To obtain CS coated SNP in smaller size (first sample), a solution containing 0.5% of acetic acid dissolved with 1% of CS was mixed with first size of SNP in a proportion of 56 : 1 (SNP/CS); for second size of CS coated SNP, a solution containing 0.5% of acetic acid with 1% of CS was collocated with the second size of SNP in a proportion of 28 : 1 and finally, for bigger size of CS coated SNP, an acetic acid solution of 0.25% dissolved with 1% of CS was placed with third size of SNP in a proportion of 28 : 1, respectively. Each solution was mixed for 12 h on an electronic mixer at environmental temperature. Each procedure for BSA and CS samples was made in three different times.

2.3. Characterization. Ultraviolet-visible (UV-Vis) absorption spectroscopy analysis was used in nanoparticles samples dispersed in water using a UV-Vis spectrometer (CHEMUSB4-VIS-NIR, Ocean Optics) with an integrated lamp of visible light. Dynamic light scattering assay (DLS, Malvern Zetasizer Nano ZS, Instruments Worcestershire, United Kingdom) operating with a He-Ne laser at a wavelength of 633 nm and a detection angle of 90 degrees

TABLE 1: Physical and chemical properties of BSA and CS coated SNP.

SNP sample (nm)	Diameter DLS (nm)	Diameter TEM (nm)	Shape	Zeta potential (mV)
10.7*	10.7	—	—	-22.7 ± 8.2
38.3*	38.3	—	—	-15.4 ± 12.9
110.8*	110.8	—	—	-24.0 ± 5.6
BSA 22.9	22.9 ± 8.5	11.0 ± 3.2	Spherical	-21.2 ± 7.7
BSA 32.7	32.7 ± 8.3	19.4 ± 5.2	Spherical	-20.9 ± 5.8
BSA 113.3	113.3 ± 39.3	94.4 ± 17.6	Pseudospherical	-19.1 ± 5.4
CS 29.7	29.7 ± 12.1	11.9 ± 2.7	Spherical	17.3 ± 5.4
CS 41.0	41.0 ± 6.8	27.6 ± 4.1	Spherical	36.5 ± 5.6
CS 116.6	116.6 ± 4.6	96.0 ± 13.5	Pseudospherical	22.0 ± 6.8

Diameters and zeta potential are expressed in average and standard deviation; * noncoated samples were prepared one time only; DLS: Dynamic Light Scattering; TEM: Transmission Electron Microscopy. —: not applicable.

for 60 seconds at 25°C for each sample was performed to confirm size and zeta potential. Transmission Electron Microscopy (TEM, JEOL JEM-1230, Tokyo, Japan) at an accelerating voltage of 100 kV determined shape of particle. Thermogravimetric analysis (TGA) and differential scanning calorimetry (DSC) analysis were used in dehydrated coated and noncoated SNP samples using a DSC calorimeter (Brand Waters Model DSC 500) with a constant heating rate of 20°C/min at room temperature to 1000°C under nitrogen atmosphere in a range of temperature from 20°C to 510°C.

2.4. Antibacterial Assay. Seven bacterial strains of *S. aureus*, *E. coli*, *E. faecalis*, *S. mutans*, *S. sobrinus*, *S. oralis*, and *S. salivarius* were tested. Previously to the antimicrobial test, the identity of all microorganisms was confirmed by polymerase chain reaction (PCR). The antibacterial test used in this study was previously reported [20] and some modifications were made. Bacterial strains were cultured in BHI by 18 h at 37°C before the test. MIC were determined by incubating each microorganism in 96-well microdilution plates; 200 µL of each SNP dispersion was placed in the first column and it was diluted 1:1 with BHI medium (containing 2% of sucrose for oral bacteria) inoculated with each microorganism strain at 6×10^5 CFU/mL; finally, plates were incubated at 37°C for 24 h. After that, the last well that presented turbidity was considered as minimum inhibitory concentration (MIC). To confirm MIC results for oral bacteria (*E. faecalis*, *S. mutans*, *S. sobrinus*, *S. oralis*, and *S. salivarius*), wells were washed with phosphate buffer solution (PBS) and fixed with 0.1% glutaraldehyde for 5 min; and then wells were washed with PBS once and stained using 0.5% crystal violet confirming antimicrobial activity in the last well that presented coloration. All antibacterial tests were made in triplicate.

2.5. Scanning Electron Microscopy. *S. mutans* cells were observed using scanning electron microscopy (SEM) after their exposition to SNP. Wells with SNP samples at MIC for smaller coated (BSA 22.9 nm and CS 29.7 nm) and noncoated (10.7 nm) SNP and negative control well were prepared using 0.1% glutaraldehyde for 5 minutes, washed with PBS (pH 7.4), and immersed in solutions with increasing ethanol concentrations (50, 60, 70, 90, 95, and 100%) during 20 minutes at 4°C. Finally, they were air dried and sputter coated

FIGURE 1: UV-Vis spectra of BSA coated SNP.

with a gold layer. Micrographs were taken with a SEM (JEOL JSM-6510) at an accelerating voltage of 20 kV.

3. Results and Discussion

3.1. UV-Vis and DLS Analysis. UV-Vis spectra of coated SNP are shown in Figure 1. SNP samples showed the characteristic surface plasmon of Ag nanometric (430 nm for larger SNP and 425 nm for smaller sample); however, the presence of biomolecules can promote larger bands from 600 to 800 nm. The presence of the characteristic surface plasmon confirms good stability and dispersion of SNP samples. DLS results of coated SNP are shown in Table 1. BSA and CS coated SNP samples showed narrow size distributions and the average sizes obtained were slightly bigger in BSA and CS coated SNP samples than those without coatings but there were no representative changes in their original shapes (Figure 2). Zeta potential values showed low negative values in BSA coated SNP compared to noncoated samples increasing

FIGURE 2: TEM micrographs and DLS analysis of BSA and CS coated SNP. (a) BSA 22.9 nm; (b) BSA 32.7 nm; (c) BSA 113.3 nm; (d) CS 29.7 nm; (e) CS 41.0 nm; (f) CS 116.6 nm.

FIGURE 3: Core-shell structure of coated SNP by TEM. (a) BSA 113.3 nm; (b) CS 116.6 nm.

negative charge slightly when its size is larger; moreover, positive charges in CS samples compared to negative values in noncoated samples were also obtained. We can assume that BSA and CS components, due to their high affinity to bind to SNP surface attaching thiols and amino groups might be modifying electrical charges on the surface of SNP producing differences on surface charge of outer region of SNP [21, 22].

3.2. TEM Analysis. TEM micrographs of BSA and CS coated SNP are shown in Figure 2. We can realize that smaller BSA and CS coated SNP samples show uniform spherical shapes while bigger nanoparticles in both BSA and CS coated SNP are pseudospherical particles with irregular hexagonal shapes. Coated SNP exhibit acceptable dispersion and good size distribution, although some SNP agglomerates can be seen in BSA coated SNP of 19.4 nm (Figure 2(b)) due to strong interactions between Ag^+ and BSA forming conglomerations of many BSA molecules [23]. A small shell in BSA and CS coated SNP samples (BSA 113.3 nm and CS 116.6 nm samples) was also observed around the pseudospherical SNP (Figure 3); BSA coatings of 5.93 nm and 2.52 nm of CS were clearly observed showing thickness similarities with others studies [24, 25].

3.3. Thermal Analysis. TGA and DSC results are shown in Table 2. Weight loss differences were found between coated (BSA 3.24–13.07% and CS 2.43–32.68%) and noncoated (1.5–2.18%) SNP samples. TGA curves showed that coated SNP samples have different mass profile and considerable amount of BSA and CS compounds than noncoated SNP. Smaller BSA and CS coated SNP displayed a higher percentage of coating weight than larger nanoparticles; this may be easily explained due to the fact that smaller nanoparticles have a high reactive area to bind to thiol, amino, and hydroxyl groups of BSA and CS molecules, respectively [21, 26]. DSC results are shown in general two peaks; the first weight loss appeared from 18 to 110°C for coated and noncoated SNP

indicating water loss (18 to 110°C) and the second peak, which corresponds to decomposition of BSA and CS components, started from 200–250°C up to 500°C and 250 up to 450°C, respectively. TGA-DSC results suggest the presence of BSA and CS coatings on SNP surfaces due to the different mass profile obtained from BSA and CS coated SNP compared with noncoated SNP samples. Probably, BSA as plasma protein showed a strong affinity for SNP in order that SNP surfaces and Ag^+ ions could bind with proteins and amino acids present in BSA attaching with thiol groups providing steric stabilization due to the bulky protein molecules [21, 27, 28]. CS coated SNP can assume that CS macromolecules are attached with SNP using electrostatic interactions with the presence of a large number of free amino ($-\text{NH}_2$) and hydroxyl ($-\text{OH}$) groups in CS chains forming bonds with SNP surfaces; the high affinity of CS from β (1–4) D-glucosamine units to bind to Ag surfaces has been demonstrated by previous studies [24, 26, 29].

3.4. Antibacterial Assay. MIC results from BSA and CS coated SNP on Gram-negative bacterium of *E. coli* and Gram-positive bacterium of *S. aureus*, *E. faecalis*, *S. mutans*, *S. sobrinus*, *S. oralis*, and *S. salivarius* are shown in Table 3. Coated and noncoated SNP samples showed different levels of antibacterial activity on different strains, although differences according to type of coatings, size of particle, and type of bacterium strain were also associated. For example, the antimicrobial effect of the two smaller nanoparticles was better in coated SNP than noncoated samples; moreover, the bigger size of BSA coated SNP sample showed higher growth inhibition than the other similar sizes of CS coated and noncoated SNP samples, respectively. The smallest coated and noncoated SNP samples obtained higher antimicrobial activity than bigger samples and the coating of CS sample showed the best inhibition activity. Furthermore, it can be observed in Figure 3 that poor bacterial adherence and low survival activities in bacterial strains (*S. mutans*) were

TABLE 2: TGA and DSC analysis of BSA and CS coated SNP.

Sample (nm)	First weight loss (%)	Second weight loss (%)	Residues (%)	Total weight loss (%)	Coating weight (%)
10.7	0.02 (18–110)	1.41 (150–500)	98.5	1.5	—
38.3	0.0 (20–110)	1.7 (150–500)	98.24	1.76	—
110.8	0.17 (20–100)	1.84 (200–500)	97.82	2.18	—
BSA 22.9	0.38 (21–100)	3.4 (200–500)	96.37	3.63	2.13
BSA 32.7	1.32 (21–110)	11.44 (200–500)	86.93	13.07	11.31
BSA 113.3	0.33 (21–100)	2.74 (210–500)	96.76	3.24	1.06
CS 29.7	30.45 (23.5–100)	2.81 (200–500)	67.32	32.68	31.18
CS 41.0	0.16 (20–100)	2.39 (150–500)	97.57	2.43	0.67
CS 116.6	0.0 (20–100)	3.6 (175–500)	95.9	4.1	1.92

First and second weight loss columns describe in parentheses the temperature range for each sample. —: not applicable.

TABLE 3: Minimal inhibitory concentrations (MIC) of BSA and CS coated SNP.

SNP samples (nm)	<i>S. aureus</i>	<i>E. coli</i>	<i>E. faecalis</i>	<i>S. mutans</i>	<i>S. sobrinus</i>	<i>S. oralis</i>	<i>S. salivarius</i>	Total/SNP*
10.7	535.0 ± 0.0	535.0 ± 0.0	535.0 ± 0.0	222.9 ± 77.2	535.0 ± 0.0	535.0 ± 0.0	44.6 ± 19.3	420.4 ± 202
38.3	1070.0 ± 0.0	1070.0 ± 0.0	1070.0 ± 0.0	535.0 ± 0.0	1070.0 ± 0.0	1070.0 ± 0.0	178.3 ± 77.2	866.2 ± 363
110.8	1070.0 ± 0.0	1070.0 ± 0.0	1070.0 ± 0.0	1070.0 ± 0.0	1070.0 ± 0.0	1070.0 ± 0.0	178.3 ± 490.8	942.6 ± 337
BSA 22.9	469.2 ± 0.0	469.2 ± 0.0	469.2 ± 0.0	234.6 ± 0.0	469.2 ± 0.0	391.0 ± 135	39.1 ± 16.9	363.1 ± 167
BSA 32.7	998.9 ± 0.0	998.9 ± 0.0	998.9 ± 0.0	249.7 ± 0.0	998.9 ± 0.0	832.4 ± 288	41.6 ± 18.0	731.3 ± 409
BSA 113.3	938.5 ± 0.0	938.5 ± 0.0	938.5 ± 0.0	469.2 ± 0.0	938.5 ± 0.0	938.5 ± 0.0	625.6 ± 271	826.8 ± 196
CS 29.7	525.6 ± 0.0	87.6 ± 37.9	525.6 ± 0.0	262.8 ± 0.0	262.8 ± 0.0	262.8 ± 0.0	262.8 ± 0.0	312.9 ± 158
CS 41.0	1033.1 ± 0.0	516.5 ± 0.0	1033.1 ± 0.0	688.7 ± 298	1033.1 ± 0.0	1033.1 ± 0.0	344.3 ± 149.1	811.7 ± 293
CS 116.6	1033.1 ± 0.0	1033.1 ± 0.0	1033.1 ± 0.0	1033.1 ± 0.0	1033.1 ± 0.0	1033.1 ± 0.0	688.7 ± 298.3	983.9 ± 130
Total/MO [†]	852 ± 260	746 ± 353	852 ± 260	529 ± 336	823 ± 311	796 ± 316	267 ± 245	—

All values are measured in microgram per milliliter ($\mu\text{g/mL}$), average and standard deviation. Average values of MIC were obtained in triplicate. * Total average per SNP sample. [†] Total average of MIC per microorganism. —: not applicable.

observed in smallest SNP samples compared to larger SNP in which CS samples showed slightly higher antiadherence characteristics in the most bacterial strains than BSA samples (Figure 4). The results can suggest that the antimicrobial activity of coated SNP might depend on the type of coating on the SNP surface producing a synergic mechanism that it improves the bacterial growth inhibition and the ability of SNP to bind to cell membranes and to generate antimicrobial effect; however, the large surface area of small SNP might also be involved [26, 30, 31]. The most resistant bacteria to SNP were presented by *S. aureus* and *E. faecalis*, followed by *S. sobrinus*, *S. oralis*, *E. coli*, *S. mutans*, and *S. salivarius* strains. The main Gram-positive (*S. aureus* and *E. faecalis*) and Gram-negative (*E. coli*) bacteria, which are associated with multidrug-resistance infections, showed differences on their MIC results. *S. aureus* and *E. faecalis*, as Gram-positive bacteria had similar higher MIC values ($852 \pm 260 \mu\text{g/mL}$) than Gram-negative bacteria of *E. coli* ($746 \pm 353 \mu\text{g/mL}$). The Gram-positive bacteria revealed the highest microbial resistance to SNP although *E. coli* bacteria can offer more antimicrobial inhibitory resistance than *S. aureus* strain to similar conditions [32]. Our findings agree with the majority of previous reports indicating an antimicrobial effect against several types of bacterial strains (including oral and nonoral bacteria), improving their antimicrobial activity when BSA and/or CS coatings are present [24, 26, 32, 33].

FIGURE 4: Antimicrobial activity of coated SNP against *S. mutans* strain (ATCC, 25175). Purple wells show bacterial growth.

It is possible that a combined antimicrobial action between SNP and coatings can be present. In BSA coated SNP, an antimicrobial mechanism of electrostatic interactions between the nanoparticles and the negatively charged cell surface [34], as well as the control in the interactions of nanoparticles with the cell by the protein on the nanoparticle surface, could be synergistically related [35]. Conversely, CS is

FIGURE 5: SEM micrographs of *S. mutans* bacteria treated with SNP. (a) Noncoated SNP (10.7 nm); (b) BSA-SNP (22.9 nm); (c) CS-SNP (29.7 nm); (d) control (*S. mutans*).

considered unable to enter into the cell and their antibacterial effect would be only on the cell surface using electrostatic contacts that could bind and disrupt the cytoplasmic membrane of bacteria leading to damage of membrane functions [36]. Also, CS could help to alter the dynamic balance of Ag-O bond at the same time as CS interacted with bacterial membrane in order to result in free radicals, Ag^+ ions, and/or small SNP that may easily penetrate inside cells [26] inhibiting respiration process, activation of proteins, replication of DNA [37], and the possibility of the induction of a bacterial apoptosis-like response [38] leading to the cell death.

3.5. SEM Analysis. SEM micrographs of *S. mutans* cells treated with small coated (BSA 22.9 nm and 29.7 nm) and noncoated (10.7 nm) SNP at the minimum inhibitory concentration are shown in Figure 5. Micrographs showed inhibition zones and alterations of the biofilm structure in coated (BSA 22.9 nm and 29.7 nm) and noncoated (10.7 nm) SNP than control group. Poor growing of bacteria cells on wall surfaces was observed in the coated (Figures 5(b) and 5(c)) and noncoated (Figure 5(a)) SNP finding isolated cells and significant alterations of the bacteria cell bindings. It can also be seen that noncoated SNP (Figure 5(a)) modified much more the morphological structure of the biofilm and the shape of cells than those SNP with BSA and CS coatings (Figures 5(b) and 5(c)). It is very probable that both coated and

noncoated SNP can promote alterations of biofilm structure and cellular binding due to the limited production of free glucosyltransferases (cellular binding's principal enzyme) to produce glucans, extracellular polysaccharides, and cellular breathing from bacteria metabolism [20] leading to death; moreover, other specific types of bacteria cell membranes could be involved according to their metabolisms. BSA and CS coated SNP were less aggressive on *S. mutans* cells than noncoated SNP; this may indicate that coated SNP could have a low toxicological potential in cells due to limited interactions between SNP surfaces and cell membranes; therefore, other *in vitro* and *in vivo* studies are necessary to understand coated SNP's action mechanism.

4. Conclusions

SNP with different sizes, shapes, and BSA and CS coatings were successfully prepared and characterized. The coated SNP showed good antibacterial activity against seven oral and nonoral bacteria strains. The antimicrobial effectiveness of the coated SNP was associated with the size of the SNP; the presence and type of coating on the particle surface, principally CS component, produced significant antimicrobial activity thus suggesting a combination of various antimicrobial mechanisms that may depend on specific properties of SNP and individual characteristics of microorganisms. Bio-functionalization of SNP using organic components might

be promising candidates for several biomedical applications. According to our knowledge, this is the first study where BSA and CS coated SNP were prepared with different sizes and shapes and their antimicrobial effects on seven oral and nonoral microorganisms were tested. Certainly, further investigations are necessary for a better understanding of coated SNP and their potential use for prevention and/or control of diseases considered as public health problems.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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