



# Synthesis and evaluation of silver nanoparticles loaded with Gemini surfactants: Surface and antimicrobial activity



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## ABSTRACT

Silver nanoparticles loaded by three Gemini cationic surfactants having different spacer chain length were prepared and characterized using ultraviolet–visible (UV–vis) absorption spectroscopy and transmission electron microscope (TEM) analysis. TEM analysis showed the homogeneity and stability of the formed silver nanoparticles in the presence of the Gemini surfactants. The results showed increased activity of the silver nanoparticles in the presence of the Gemini cationic surfactants. Antimicrobial results of inhibition zone diameter and minimum inhibitory concentration were correlated to Gemini surfactants and their nanoparticles including surface activity and tendency toward adsorption at interfaces.

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## Introduction

Silver nanoparticles (AgNPs) are exhibiting very strong bactericidal activity against both gram-positive and gram-negative bacterial strains [1,2] and also can be considered as potential antifungal agents. The antibacterial inhibition of silver nanoparticles (AgNPs) as well as silver nanocomposites or silver nanoparticle-based materials has been intensively studied recently due to the growing bacterial resistance to common antibiotics [3–5]. Antibacterial activities of silver nanoparticles containing materials help in reducing hazardous infections by bacteria [6], and in preventing bacterial colonization of prostheses and catheters [7]. The already published studies on bactericidal activity have proved that AgNPs kill bacteria at such low concentrations (units of mg/L) [1], which does not reveal acute toxic effects on human cells [8,9]. The antimicrobial mechanism of cationic surfactants was described in recent study and the results showed their high efficiency as antimicrobial agents [10]. In this study, we prepared three cationic Gemini surfactants with different spacer chain length and their silver nanoparticles. The structures of silver nanoparticles were determined using UV–vis

absorption spectroscopy, and transmission electron microscope (TEM) analysis. The silver nanoparticles were evaluated as antibacterial agents against different types of bacteria.

## Experimental

### Synthesis of aminopyridine schiff base (SB)

A mixture of benzaldehyde (0.15 mol, 15.75 g), *p*-aminopyridine (0.15 mol, 14.1 g) and xylene (150 mL) was heated in a round flask under a continuous removal of water of the reaction [11]. The reaction was stopped after the removal of 2.7 mL of water. The obtained schiff base (SB) was viscous and brown in color, C<sub>12</sub>H<sub>10</sub>N<sub>2</sub>O, elemental analysis % (calculated/found): C: 77.6/77.1, H: 5.9/5.8, N: 16.5/16.3.

### Synthesis of cationic Gemini surfactants

A mixture of the prepared schiff base (SB) (0.05 mol, 8.51 g), 0.025 mol of dibromoalkanes namely: 1,2-dibromoethane, 1,6-dibromohexane and 1,12-dibromododecane (4.7 g, 6.1 g and 8.2 g, individually) and 100 mL of acetone was refluxed in a round flask for 12 h. Then the reaction mixture was cooled overnight, and the products were filtered and washed twice with diethyl ether and

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finally dried under vacuum [12–14]. The obtained products were designated as: SB-2, SB-6 and SB-12, Scheme 1.

#### Preparation of silver nanoparticles (AgNPs)

The silver nanoparticles colloidal solution was prepared using chemical reduction method. All solutions of reacting materials were prepared in bidistilled water. In a typical experiment 50 mL of  $\text{AgNO}_3$  solution ( $1 \times 10^{-3}$  M) was boiled, then 5 mL of 1% trisodium citrate was added drop wise under vigorous stirring until the color changed to pale yellow. Then, heating was stopped and the reaction was cooled to room temperature [15,16].

#### Preparation of silver nanoparticles loaded with Gemini surfactants

The silver nanoparticles solution (20 mL) was mixed with 5 mL solution of the Gemini surfactants (SB-2, SB-6 and SB-12) ( $1 \times 10^{-2}$  M). The mixture was stirred continuously for 24 h until the colour change. The resulting solution was used for UV–vis absorption spectroscopy and transmission electron microscope (TEM) analysis [17].

#### Measurements

Fourier transform infrared (FTIR) spectra were recorded at room temperature using AT<sup>TM</sup> Mattsonm Infinity series, Bench top 961 controlled by Win First<sup>TM</sup> V2.01 software. Proton nuclear magnetic resonance ( $^1\text{H}$  NMR) spectra were measured using Spect. Varian, GEMINI 200 ( $^1\text{H}$  200 MHz) in dimethyl sulfoxide ( $\text{DMSO}-d_6$ ) as a solvent and tetramethyl silane TMS as a reference.

#### Ultraviolet–visible (UV–vis) absorption spectroscopy

UV–vis absorption spectroscopy measurements of the Gemini surfactants, AgNPs solution and the nanostructure of the Gemini surfactants were carried out using Jenway 6505 UV/vis spectrometer (UK) in water as a solvent.

#### Transmission electron microscope (TEM) analysis

For the TEM measurements, a drop of a solution containing the particles was deposited on a copper grid covered with amorphous carbon. After allowing the film to stand for 2 min, the extra solution was removed by means of blotting paper and the grid allowed drying before the measurement. Transmission electron

microscopy (TEM) studies were performed using a JEOL JEM-1230 electron microscope operating at an accelerating voltage of 200 kV attached to a CCD camera.

#### Surface tension measurements

Surface and interfacial tension measurements were performed using a Krüss K6 tensiometer by the platinum ring detachment method ( $\pm 0.5$  mN/m), Germany. Freshly prepared aqueous solutions of Gemini surfactants (SB-2, SB-6, and SB-12) with a concentration range of 0.01 to 0.00001 M at 25 °C. The solutions were poured into a clean Teflon cup with a mean diameter of 28 mm (Teflon cup was used to prevent the adhesion of the surfactant to the glass cup walls). The solutions were left for 2 h to allow the stabilization and complete adsorption of surfactant molecules at the solution surface, and then the apparent surface tension values were the average of 3 readings [18]. The platinum ring was removed, washed with dilute hydrochloric acid solution followed by bidistilled water.

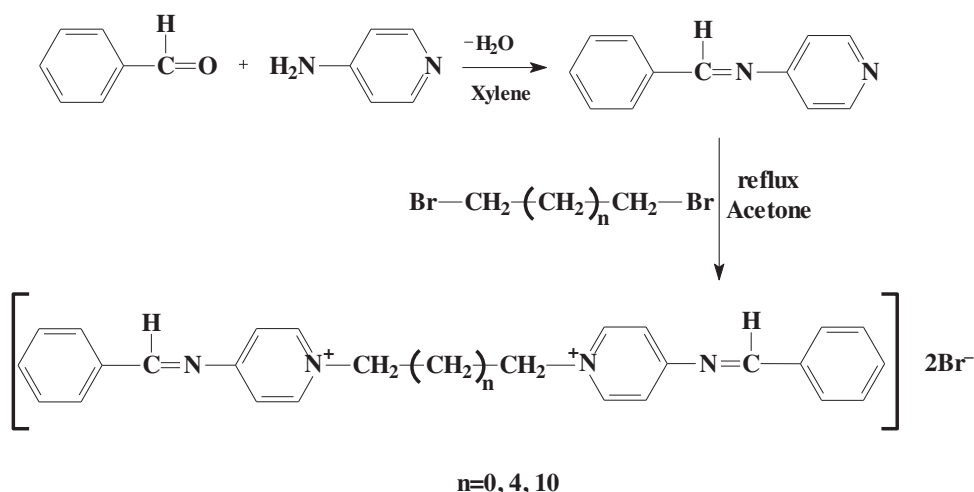
#### Antimicrobial activity measurements

##### Microorganisms

The biocidal activity of the synthesized surfactants was tested against different bacterial strains as follows: *Staphylococcus aureus*, *Escherichia coli*, *P. aeruginosa*, and *Bacillus subtilis*. The selected species cause dangerous diseases for the humans and animals.

##### Measurements of resistance and susceptibility

For preparation of discs and inoculation, 1.0 mL of inocula were added to 50 mL of agar media (40 °C) and mixed. The agar was poured into 120 mm petri dishes and allowed to cool to room temperature. Wells (6 mm in diameter) were cut in the agar plates using proper sterile tubes and filled up to the surface of agar with 0.1 mL of the synthesized biocides dispersed in  $\text{H}_2\text{O}$  (1, 2, 5 mg/mL DMF). The plates were left on a leveled surface, incubated for 24 h at 30 °C and then the diameters of the inhibition zones were read. The inhibition zone formed by these compounds against the particular test bacterial strain determined qualitatively the antibacterial activities of the synthetic compounds. The mean value obtained for three individual replicates was used to calculate the zone of growth inhibition of each sample. The antimicrobial activity was calculated as a mean of three replicates. The tested compounds were completely compatible with the medium of agar and no turbidity was observed during the mixing process [19,20].



Scheme 1. Synthesis of the Gemini cationic surfactants.

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