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Research Paper

Comparison of antibacterial activity and cytotoxicity of silver nanoparticles and silver-loaded montmorillonite and saponite

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ABSTRACT

Although silver nanoparticles are known for their antibacterial activity, little research has been carried out on what synthesis method provides the most effective particles. In this study, silver nanoparticles were synthesised via chemical reduction by using silver nitrate as the silver precursor, ascorbic acid as the reducing agent and sodium citrate as the stabilising agent. The solutions were adjusted to several pH values employing sodium hydroxide, citric acid or nitric acid. Dynamic light scattering and absorption spectra in the ultraviolet/visible region characterisation revealed that employing nitric acid to adjust the pH produced more varied and larger silver particle sizes. Then, silver nanoparticles were supported on montmorillonite and saponite through wet impregnation or ion exchange methods. Scanning electron microscopy, energy-dispersive X-ray spectroscopy and transmission electron microscopy characterisation confirmed that silver nanoparticles were successfully loaded onto the clay minerals. Next, the antibacterial activity of the samples was evaluated against *Escherichia coli* and *Staphylococcus aureus* by determining their minimum inhibitory concentrations and minimum bactericidal concentrations. The free silver nanoparticles did not show any antibacterial activity at 125 mg/L. In contrast, the silver-loaded samples obtained by wet impregnation and with a higher silver content displayed the strongest antibacterial effect. Finally, the cytotoxicity of the samples was determined in GM07492-A cell line by using an XTT colorimetric assay. The calculated IC_{50} values revealed that the supported silver nanoparticles were barely toxic. Thus, the silver-loaded clay minerals obtained here are promising antibacterial materials with a high-grade safety profile.

1. Introduction

The emergence and spread of antimicrobial resistance have recently been highlighted as one of the top 10 threats to global health (Micoli et al., 2021; Liguori et al., 2022). Infectious-resistant microbes are expected to kill 2.4 million people worldwide in the next 30 years, costing 3.5 billion US dollars in medical care (Hofer, 2019). In fact, some predictions stand that resistant bacterial infections will become the leading cause of death worldwide by 2050 (O'Neill, 2014). In this context, nanoantibiotics have been shown to be promising alternatives to traditional antibiotics to overcome this challenge (Mamun et al., 2021). And, within antibiotics, nanoparticles with antimicrobial activity are arousing increasing interest (Tran et al., 2016).

Nanoparticles are particles that have a diameter of less than 100 nm in at least one dimension (Garcés et al., 2021). Silver nanoparticles have become one of the most compelling nanomaterials due to their exponential number of applications (Akter et al., 2018). Among nanoparticles that present high bioactivity, silver nanoparticles are the most promising because of their broad-spectrum activity (Roy et al., 2017). Thus, they have been demonstrated to be efficient against bacteria, fungi and viruses (Tariq et al., 2022). For example, recent studies have shown that they can present antiviral activity against SARS-CoV-2 (Almanza-Reyes et al., 2021; Baselga et al., 2022).

These antimicrobial properties have led to silver nanoparticles being

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applied in different fields. For instance, they have been used in polymers in the food packaging industry, such as beverage bottles or storage containers (Duncan and Pillai, 2015). Examples of this are several products commercialised in the USA, such as Kinetic Go Green Basic Nanosilver Food Storage container, Oso Fresh Food Storage container and FresherLonger™ Plastic Storage bags (Echegoyen and Nerín, 2013; Azizi-Lalabadi et al., 2021).

They can also be used in the disinfection and purification of water. For example, Vilela et al. (Vilela et al., 2017) built microbots decorated with silver nanoparticles that were able to kill 80% of bacteria from a water sample in 15 min. There are also commercialised products in the USA like water filters (e.g., 989 Bacteriostatic Water Filter Media and NATURE2 G45-VC40) or algaecides (e.g., Algeadyn and Nu-Clo Silvercide) (Nowack et al., 2011). Likewise, silver nanoparticles have also been used as air disinfectants in air filters, creating both antibacterial (Xiao et al., 2021) and antiviral (Joe et al., 2016) materials.

Finally, where the antimicrobial properties of silver nanoparticles have stood out the most is in medical applications. For instance, they have been employed to speed up the healing of infected wounds (Liu et al., 2022) or treat burn injuries (Ying et al., 2019). They have also been utilised in surgical implants (Ziąbka et al., 2018), tissue engineering (Srivastava et al., 2019), and healthcare products such as AgTive® silver-nanoparticle-impregnated central venous catheters (Antonelli et al., 2012) and Acticoat™ wound dressings (Khundkar et al., 2010). Moreover, they have been shown to be effective against medical device-associated infections (Tran and Tran, 2021) and even dental biofilms (Takamiya et al., 2021).

There are multiple possible methods for the synthesis of silver nanoparticles. On the one hand, there is chemical reduction, where a precursor silver, usually a silver salt like $AgNO_3$, is reduced to elemental silver by reducing agents like ascorbic acid (Zhang et al., 2022) or borohydride (Khatoon et al., 2023). The drawback of this method is that most of the chemicals and solvents employed are toxic (Sharma et al., 2022). Another possibility is the photochemical method, in which the reduction is achieved through the aid of photo irradiation (Zahoor et al., 2021), like LEDs (Leong et al., 2022). Third, silver nanoparticles can be synthesised by physical methods (Vishwanath and Negi, 2021), such as evaporation-condensation, ball milling and laser ablation (Naganthran et al., 2022). Finally, there is the biological method. This recently appeared methodology consists of using plant parts to produce environmentally friendly materials (Garg et al., 2022).

This example of green chemistry allows the preparation of green-synthesised silver nanoparticles, which have been reported to present lower toxicity levels compared to chemically synthesised nanoparticles (Akter et al., 2018; Younas et al., 2022). Thus, there are numerous examples in the literature that employ vegetal compounds as reducing agents, like extracts of marine algae (Algotiml et al., 2022), eucalyptus leaves (Balčiūnaitienė et al., 2022), saffron (Khorasani et al., 2022), coconut shell fiber (Das et al., 2021), coffee waste (El-Desouky et al., 2022) or banana peels (Sengupta and Sarkar, 2022).

The physicochemical characteristics of nanoparticles favor their aggregation, which in turn limits their effectiveness (Shameli et al., 2011). To overcome this fact, several support strategies have been proposed, like cyclodextrins (Gill et al., 2021), surfactants (Pisárčik et al., 2021) or polysaccharides such as pectin (Shankar et al., 2016). However, these strategies can be difficult to find and manipulate (Giraldo et al., 2016) or be expensive, which complicates their application in low-resource settings, where they could benefit from their use.

Clay minerals have proven to be excellent carriers for metal nanoparticles in general, and silver nanoparticles in particular, due to their numerous advantages, like natural abundance, cost-effectiveness, non-toxic nature, chemical inertness, and high absorptive capacity (Roy et al., 2017). This allows obtaining a material with great bioactivity and prolonged duration since a slow release of silver species from the support is achieved (Giraldo et al., 2016; Roy et al., 2017). The low cost and toxicity and environmentally friendliness of clay minerals (Giraldo et al., 2016) have also led to their use in other fields, like catalysis (Cecilia and Jiménez-Gómez, 2021), as drilling fluids (Morariu et al., 2022) or as adsorbent materials for CO_2 capture (Tao et al., 2022) or contaminants removal (Gil et al., 2021), such as antibiotics (Hacıosmanoğlu et al., 2022) or heavy metals (Jiang et al., 2021) from wastewater.

Clay mineral-supported metal nanoparticles can be synthesised employing a wide variety of methods (Gil et al., 2000; Varadwaj and Parida, 2013). Firstly, it is necessary to introduce the metal precursor by chemical vapor deposition, deposition-precipitation, wetness impregnation, ion exchange and sol-gel, among other procedures. If the synthesis of the metal is required, additional steps such as hydrogen reduction under atmospheric pressure, polyol or borohydride methods or other chemical reduction routes are required.

In the case of silver nanoparticles, the most common methodologies to introduce them into the structure of clay minerals are wet impregnation and ion exchange (Sato et al., 1997; Holešová et al., 2013; Cao et al., 2014; Lavorgna et al., 2014; Tian et al., 2014; Jou and Malek, 2016). As the silver precursor is an aqueous solution of $AgNO_3$, a subsequent reduction is needed, with chemical reducing agents like sodium borohydride (Miyoshi et al., 2010; Praus et al., 2013; Nunes Pessanha et al., 2014), ascorbic acid (Liu et al., 2007), hydrazine (Papp et al., 2008; Bagchi et al., 2014), hydrothermal treatment (Wu et al., 2010), phenolic derivatives (Makwana et al., 2020), plant natural extracts (Sohrabnezhad et al., 2015) or even electrochemical procedures (Huang et al., 2012).

Nonetheless, although silver nanoparticles are appealing for their antimicrobial efficacy, their toxicity toward mammalian cells is causing increasing concern (Chang et al., 2021a, 2021b). For this reason, the potential risks against human health (Hakimov et al., 2022) are motivating more studies to focus on evaluating their cytotoxicity.

The main task of this study is the comparison of the antibacterial activity and cytotoxicity of silver nanoparticles prepared by chemical reduction and silver-loaded montmorillonite and saponite prepared by wet impregnation and ion exchange were compared. The objectives of this work are notably original for several reasons. On the one hand, there is limited evidence in the literature of the comparison of different synthesis methods since most works focus on analysing a specific methodology without comparing it with previous ones. On the other hand, saponite was employed as a carrier of silver nanoparticles. Again, in the literature, there are limited examples of the use of this clay mineral for the preparation of materials with antibacterial activity since other clay minerals such as montmorillonite or kaolinite are more often utilized (Liu et al., 2007; Miyoshi et al., 2010; Huang et al., 2012; Holešová et al., 2013; Praus et al., 2013; Cao et al., 2014; Lavorgna et al., 2014; Tian et al., 2014; Sohrabnezhad et al., 2015; Jou and Malek, 2016; Makwana et al., 2020). Finally, the cytotoxicity of the silver-loaded clay mineral samples was evaluated. Although it is true that there is an increasing number of studies that analyses the potential toxicity of silver nanoparticles, the evidence of this fact in silver-loaded clay minerals still remains extensively limited.

2. Experimental section

2.1. Materials and reagents

For the synthesis of silver nanoparticles, silver nitrate ($AgNO_3$, 99.0%, Panreac, Barcelona, Spain) was used as the silver precursor. Ascorbic acid ($C_6H_8O_6$, 99.0%, Panreac, Barcelona, Spain) and sodium citrate ($Na_3C_6H_5O_7{\cdot}2H_2O$, 99.0%, Probus, Badalona, Spain) were utilised as reducing and stabilising agents, respectively. Citric acid ($C_6H_8O_7$, 99+%, Alfa Aesar, Kandel, Germany), sodium hydroxide (NaOH, 98.0%, Panreac, Barcelona, Spain) and nitric acid (HNO_3, 65%, Panreac, Barcelona, Spain) were employed to adjust the pH of the solutions. A commercial colloidal silver solution with a concentration of 500 mg/L (Fairvital, Landgraaf, The Netherlands) was used as reference material. All these reagents were utilised as received without further

purification. Ultrapure water (conductivity < 1 μS/cm) was employed throughout all experiments to prepare the aqueous solutions. Montmorillonite (Tsukinuno) and synthetic saponite (Kunimine Industries Co., Ltd.), both purchased from The Clay Science Society of Japan, were used as solid supports for silver nanoparticles. Some of the characteristics of these materials are a specific surface area (S_{BET}) of 23 m^2/g and a Cationic Exchange Capacity (CEC) of 1.19 x 10^{-3} eq/g in the case of montmorillonite, and an S_{BET} = 216 m^2/g in the case of saponite. The CEC value for this last clay mineral was not specified by the supplier.

2.2. Synthesis of silver nanoparticles

For the preparation of silver nanoparticles, the procedure reported previously by Qin et al. (Qin et al., 2010) was followed. Like that, the synthesis was performed by reducing an $AgNO_3$ solution with ascorbic acid (see Eq. (1)) (Piñero et al., 2017). Sodium citrate was used as a stabilising agent, thus controlling the size of nanoparticles and ensuring a size as small as possible.

$$2Ag^+ + C_6H_8O_6 \rightarrow 2Ag^\circ + C_6H_6O_6 + 2H^+ \quad (1)$$

Briefly, an 8.0 mL aqueous solution containing ascorbic acid 6.0 x 10^{-4} mol/L and sodium citrate 3 x 10^{-3} mol/L was prepared, with a measured pH of 6.6 (from now on this solution will be **reference sample 0CN**). Then the solution was adjusted to pH values between 6 and 11 (**samples 1C-6C**) by adding NaOH 0.1 mol/L or citric acid 0.2 mol/L. After these values were reached, 0.08 mL of an $AgNO_3$ 0.1 mol/L aqueous solution was added under constant stirring at 700 rpm at room temperature. The reaction samples were observed to change from colorless to yellow and then brown (pH=11) or turbid blue (all other samples). After 15 min, no further change in color took place, indicating the reactions were completed (see Figure S1). As a modification of the reported procedure, a strong acid was also employed to adjust the pH, with the aim of obtaining lower pH values and thus completing the study. Like that, the solution was also adjusted to pH values between 3 and 6 (**samples 7N-10N**) by adding HNO_3 0.1 mol/L, keeping the resting methodology unchanged. This acid was specially selected to avoid the incorporation of new components in the samples that could interfere with the results. Just as citric acid and sodium citrate contribute the same citrate ion, HNO_3 and $AgNO_3$ provide the same NO_3^- ion. These samples were seen to change from turbid blue to turbid green, stopping the reactions after 15 min. The **reference sample 0CN** was used as a negative control without silver in the following experiments of this study. The specific reagents used in the synthesis of each sample are shown in Table 1.

2.3. Synthesis of silver nanoparticles supported on clay minerals

In this study, two lamellar clay minerals were used as solid supports: montmorillonite and synthetic saponite. Two methods were employed to fulfill the support: wet impregnation, which allows a fixed amount of metal to be introduced either in one or successive impregnations through physical incorporation (Giraldo et al., 2016), and ion exchange, which allows a quantity of metal to be introduced by substituting the cations present on the surface of the clay mineral. This second method depends on the ion exchange capacity of the clay mineral and achieves, a priori, a greater dispersion of silver on the surface and smaller particle size than the first method (Shameli et al., 2011; Giraldo et al., 2016; Roy et al., 2017).

For the support of silver nanoparticles by wet impregnation, the procedure formerly described by Giraldo et al. (Giraldo et al., 2016) was followed. In this way, a first impregnation was made by adding 2 mL $AgNO_3$ 0.1 mol/L to 5 g of montmorillonite. When the situation of incipient humidity was reached, the mixture was dried at 70°C for 16 h, obtaining **sample 11M**. This impregnation process was repeated a total of three times to prepare **sample 12M**, as well as a total of five times to synthesise **sample 13M**. The same methodology was followed, utilising saponite instead of montmorillonite, to get **samples 14S** (one impregnation), **15S** (three) and **16S** (five).

The support of silver nanoparticles by ion exchange was performed according to the procedure presented beforehand by Shameli et al. (Shameli et al., 2011). Like this, a solution was prepared by dissolving 0.5670 g $AgNO_3$ in 150 mL of ultrapure water, which represents an amount of silver equivalent to six times the cation exchange capacity of montmorillonite, a quantity proposed by Roy et al. (Roy et al., 2017). 5 g of montmorillonite was introduced into the solution and was left for 6 h at room temperature without stirring. Then the suspension was centrifuged at 7000 rpm for 30 min. The obtained precipitate was washed with ultrapure water several times until it became free of residual silver ions. After drying at 70°C for 16 h, **sample 17IM** was finally obtained. The same methodology was followed, utilising saponite instead of montmorillonite and employing the same $AgNO_3$ solution (because the CEC value of this clay mineral was not specified by the supplier), to get **sample 18IS.** Montmorillonite and saponite as received constituted **reference samples 0M and 0S**, which were used as negative controls without silver loading in the following experiments of this study. The specific reagents used in the synthesis of each sample are shown in Table 2.

2.4. Characterisation techniques

The absorption spectra in the ultraviolet/visible region (UV-Vis) of silver nanoparticles were collected over the range of 300-800 nm with a Jasco V-730 UV-Vis spectrophotometer. The size of the nanoparticles was investigated through a Dynamic Light Scattering (DLS) analysis carried out in a Malvern DLS/Zetasizer 3000.

The surface morphologies of silver-loaded clay minerals were studied through Scanning Electron Microscopy (SEM) and the elemental composition through Energy-Dispersive X-ray spectroscopy (EDX), both techniques on a Tescan Vega 3 Model EasyProbe. Samples were dispersed in the stub with carbon tape and then coated with a thin layer of gold by a Quorum SC7620 coating system.

Finally, the qualitative morphology of the supported silver

Table 1
Description of reagents used in the synthesis of samples of silver nanoparticles.

Sample	Reagent used to adjust pH	Final pH	Silver source	Silver concentration (mg/L)
1C	Citric acid 0.2 mol/L	6	0.08 mL $AgNO_3$ 0.1 mol/L	154
2C	NaOH 0.1 mol/L	7	0.08 mL $AgNO_3$ 0.1 mol/L	154
3C	NaOH 0.1 mol/L	8	0.08 mL $AgNO_3$ 0.1 mol/L	154
4C	NaOH 0.1 mol/L	9	0.08 mL $AgNO_3$ 0.1 mol/L	154
5C	NaOH 0.1 mol/L	10	0.08 mL $AgNO_3$ 0.1 mol/L	154
6C	NaOH 0.1 mol/L	11	0.08 mL $AgNO_3$ 0.1 mol/L	154
7N	HNO_3 0.1 mol/L	3	0.08 mL $AgNO_3$ 0.1 mol/L	154
8N	HNO_3 0.1 mol/L	4	0.08 mL $AgNO_3$ 0.1 mol/L	154
9N	HNO_3 0.1 mol/L	5	0.08 mL $AgNO_3$ 0.1 mol/L	154
10N	HNO_3 0.1 mol/L	6	0.08 mL $AgNO_3$ 0.1 mol/L	154
0CN	None	6.6	Reference sample without silver	0

Table 2
Description of reagents used in the synthesis of samples of supported silver nanoparticles.

Sample	Starting clay mineral	Support method	Silver source	Silver concentration (mass %)
11M	Montmorillonite	Wet impregnation	2 mL $AgNO_3$ 0.1 mol/L	0.43
12M	Montmorillonite	Wet impregnation	6 mL $AgNO_3$ 0.1 mol/L	1.29
13M	Montmorillonite	Wet impregnation	10 mL $AgNO_3$ 0.1 mol/L	2.15
14S	Saponite	Wet impregnation	2 mL $AgNO_3$ 0.1 mol/L	0.43
15S	Saponite	Wet impregnation	6 mL $AgNO_3$ 0.1 mol/L	1.29
16S	Saponite	Wet impregnation	10 mL $AgNO_3$ 0.1 mol/L	2.15
17IM	Montmorillonite	Ion exchange	0.5670[a] g $AgNO_3$ in 150 mL H_2O	6.72
18IS	Saponite	Ion exchange	0.5670[a] g $AgNO_3$ in 150 mL H_2O	6.72
0M	Montmorillonite	None	Reference sample without silver loading	0
0S	Saponite	None	Reference sample without silver loading	0

[a] This represents an amount of silver equivalent to six times the cation exchange capacity of montmorillonite, a quantity proposed by Roy et al. (Roy et al., 2017).

nanoparticles was observed through Transmission Electron Microscopy (TEM) on a Tecnai Spirit Twin TEM microscope operating at 120 kV. Samples were prepared by grinding the material into fine particles and then deposited on carbon-coated palladium films supported on 300 mesh-capped grids.

2.5. Evaluation of the antibacterial activity

To evaluate the *in vitro* antibacterial activity of the samples, their minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were determined according to Clinical and Laboratory Standards Institute (CLSI) guidelines. *Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 25923 were used for these antibacterial effect assays. As culture media, tryptic soy broth (TSB) was obtained from bioMérieux (Marcy l'Étoile, France), and tryptic soy agar (TSA) was prepared by adding 15 g of agar per litre of TSB, according to the supplier. Sterilisation of all glassware and materials was carried out in an autoclave at 121°C for 30 min before the experiments.

MICs were calculated by serial two-fold dilutions in TSB, performed in 96-well round bottom polystyrene plates (TPP® Merck KGaA, Darmstadt, Germany). Briefly, the samples with silver nanoparticles (**samples series C and N**) were adjusted to an initial concentration of 125 mg/L by diluting them in TSB, and the samples with supported silver nanoparticles (**series M, S and I**), to an initial concentration of 250 mg/L, in order to make all the samples comparable. Then, the serial two-fold dilutions were carried out in TSB, achieving concentrations between 125 and 0.12 mg/L (**series C and N**) and between 250 and 0.24 mg/L (**series M, S and I**), in a volume of 100 μL per well. Negative controls without samples and with **reference samples 0CN, 0M and 0S** were also included. As a positive control, a commercial colloidal silver solution 500 mg/L (Fairvital, Landgraaf, The Netherlands) (from now on, **reference sample Ag-Ref**) was used as received without further dilution, taking advantage of its activity in these strains proven in previous experiments of the authors of this study.

Both *E. coli* and *S. aureus* strains were grown aerobically in TSA for 12 h at 37°C. Then, these cultures were diluted in TSB and adjusted to a concentration of 1.0×10^6 CFU/mL. 100 μL was inoculated in every well, achieving a final concentration of 5.0×10^5 CFU/mL, and incubated aerobically for 24 h at 37°C without shaking. All tests were done in triplicate. MICs of samples were determined by the lowest concentration of the sample which completely inhibited the growth of bacteria indicated by a clear well after incubation.

Lastly, MBCs were determined by culturing 10 μL of the well with no apparent growth and the two following more concentrated wells in TSA plates. Incubations were performed aerobically for 24 h at 37°C. All tests were done in triplicate. MBCs of samples were determined by the lowest concentration of the sample which completely inhibited the growth of bacteria in these TSA plates.

2.6. Cytotoxicity assay

A cytotoxicity assay was performed to ensure the safety of the samples. **Samples 0M, 0S, Ag-Ref, 13M and 15S** were selected based on the results of the previous antibacterial activity studies. The cytotoxicity was evaluated in human fibroblasts (GM07492-A cell line) through an XTT colorimetric assay (Roche Diagnostics, São Paulo, Brazil), following the manufacturer's guidelines.

Briefly, fibroblasts were cultured in a 96-well plate at a concentration of 10^4 cells/well, in Dulbecco's Modified Eagle Medium (DMEM) with Nutrient Mixture F-10 Ham (Ham's F-10) (1:1, v/v) (Sigma-Aldrich, São Paulo, Brazil) supplemented with 10% fetal bovine serum (FBS) (Nutricell, Thermo Fisher Scientific, São Paulo, Brazil). After an incubation of 24 h at 37°C without shaking and with 5% of CO_2, the cell cultures were treated with different concentrations of **samples 0M, 0S, 13M and 15S** ranging from 312.5 to 2500 mg/L, and with concentrations of **reference sample Ag-Ref** that varied from 56.25 to 450 mg/L. After this treatment, the plate was incubated under the same conditions for 24 h. Then, the cells were washed with phosphate-buffered saline (PBS) and exposed to 50 μL of Ham's F-10 medium (Cultilab, São Paulo, Brazil) supplemented with 25 μL of XTT, and incubated again for 17 h under the same conditions. At the end of the indicated time, the cell viability was assessed by determining the absorbance of the wells in a microplate reader (Biochrom Asys UVM340/ MicroWin 2000) at 450 nm. Cell viability was expressed as a percentage of untreated cells, once the negative control without any sample was designated as 100%. Finally, the cytotoxic activity of the samples was assessed by calculating the IC_{50} value i.e., the concentration required to inhibit 50% of cell viability, which was determined by the linear regression model using GraphPad Prism 5.0 (La Jolla, California, USA). All tests were done in triplicate.

3. Results and discussion

3.1. Synthesis of silver nanoparticles

In this study, silver nanoparticles were synthesised by chemical reduction, using $AgNO_3$ as the silver precursor, ascorbic acid as the reducing agent and sodium citrate as the stabiliser agent, a procedure previously reported by Qin et al. (Qin et al., 2010). The critical step of this methodology was the addition of $AgNO_3$ to the solution with the other two reagents under stirring conditions. As soon as the silver source was added, the colour of the solutions began to change, indicating the initiation of the chemical reaction (Qin et al., 2010; Shameli et al., 2011). This colour change is a sign that the reduction of silver ions is taking place due to the reducing action of the ascorbic acid (Bhakya et al., 2016). Although in this case the colour change occurred immediately after the addition of the silver precursor, it can take much longer if other synthesis methods alternative to chemical reduction are used, such as biosynthesis. For example, Nithya Deva Krupa and Raghavan (Nithya Deva Krupa and Raghavan, 2014) observed that the colour

change began to occur after 24 h when reducing $AgNO_3$ with the aqueous fruit extract of *Aegle marmelos*, attributable to the slow action of the enzymes present in the plant root extracts compared to chemical reagents.

The literature also highlights the importance of this step in the prevention of the aggregation of nanoparticles. Thus, after the entire silver source has been added and the colour change has stopped, agitation should be discontinued to prevent aggregation (Mulfinger et al., 2007). The same happens if the salt is not added continuously and the reaction stops. For this reason, in this study the stirring conditions were brought to an end when observing that the colour no longer varied, which occurred 15 minutes after adding the silver source. This was in accordance with what was described by the authors of the procedure. It has been also described that aggregation may be suspected if the colour of the sample changes to dark gray (Mulfinger et al., 2007; Badiah et al., 2019). In this research, the colour of the samples remained unchanged for months, suggesting silver nanoparticle stability.

In their work, Qin et al. (Qin et al., 2010) demonstrated that the size of spherical silver nanoparticles was tunable by using ascorbic acid as the reducing agent, since the variation in pH varies the reactivity of ascorbic acid, thus being effective in mediating the reduction rate of the silver precursor. Like that, the reduction of $AgNO_3$ is promoted at elevated pH values due to risen activity of ascorbic acid, which means a decrease in the size of the nanoparticles with the increase of the pH of the reaction. The employment of pH variation to modify the reactivity of the reagents has also been described in the synthesis of gold nanoparticles (Ji et al., 2007), platinum nanoparticles (Sadalage et al., 2022) and copper nanocrystals (Lyu et al., 2022).

The absorbance spectra of the samples were studied using UV-Vis absorption spectroscopy. The measurements showed absorbance maxima ranging from 400 to 432 nm (see Fig. 1). In addition, the absorption peaks became narrower as the pH increased. These results agree with those described by Qin et al. (Qin et al., 2010). The authors found absorption peaks between 412 and 480 nm. Furthermore, numerous studies have shown that spherical silver nanoparticles exhibit a characteristic peak in the region of 400 to 420 nm (Song et al., 2009; Nakamura et al., 2011; Chhatre et al., 2012; Jiraroj et al., 2014; Lu et al., 2017; Singh et al., 2020). This typical maximum, also shown by the nanoparticles in this work, represents the dipole component of the plasmon resonance of the surface of small spherical silver particles (Evanoff and Chumanov, 2004). The narrowing of the spectra as the pH increases can be related to the decrease in particle size and the degree of anisotropy (Slistan-Grijalva et al., 2005; Qin et al., 2010).

Qin et al. (Qin et al., 2010) also describe that their product synthesised at pH=6 presented a shoulder band located at 390 nm, which indicated the anisotropic character of those particles. In this study, a shoulder band can also be observed at 652, 612 and 700 nm in the case of samples 1C (pH=6), 2C (pH=7) and 10N (pH=6), respectively. This demonstrates the concordance of the results with those of the literature.

Finally, it should be noted that the **samples of the C series**, prepared according to the methodology reported by Qin et al. (Qin et al., 2010), presented a spectra morphology and absorption peaks wavelengths almost identical to those described by the authors. This suggests that the results are comparable, despite the minimal changes in the synthesis procedure (a stirring speed of 700 rpm versus their conditions of 900 rpm for the same 15 min until the color of the reactions changed). The **samples of the N series**, whose preparation methodology has been proposed for the first time in this work, also presented similar morphology and wavelengths, which suggests that these nanoparticles are comparable to those of the C series. **Reference sample Ag-Ref** also exhibited alike characteristics. This implies that it can be considered a valid positive control for subsequent antibacterial activity studies.

Lastly, the characterisation of silver nanoparticles was completed with the measurement of their size through DLS technique. The results of this analysis are shown in Table 3. The **samples of the C series** contained nanoparticles with diameters ranging from 10.9 to 2.9 nm. It was also observed that as the pH of the samples increased, the size of the nanoparticles decreased. In their work, Qin et al. (Qin et al., 2010) obtained average sizes and standard deviations of 73 (±22%), 63 (±15%), 56 (±20%), 50 (±19%), 40 (±17%) and 31 (±19%) nm in the particles prepared at pH of 6, 7, 8, 9, 10 and 10.5, respectively. They used TEM characterisation to investigate these data. It is possible that they did not get diameters as small as those achieved in this study because their maximum pH reached was 10.5, instead of 11. The authors also argued, as mentioned before, that the decrease in nanoparticle size with

Table 3
Measurement of the diameters of silver nanoparticles through DLS technique.

Sample	pH	Diameter (nm)
1C	6	10.9 ± 0.53
2C	7	11 ± 0.57
3C	8	5.7 ± 2.30
4C	9	28 ± 4.95
5C	10	3.6 ± 3.23
6C	11	2.9 ± 0.65
7N	3	171.3 ± 15.93
8N	4	43.9 ± 0.76
9N	5	29 ± 3.63
10N	6	7.4 ± 3.58
Ag-Ref	-	4.8 ± 2.54

Results are presented as average diameter ± standard deviation of 10 independent measurements.

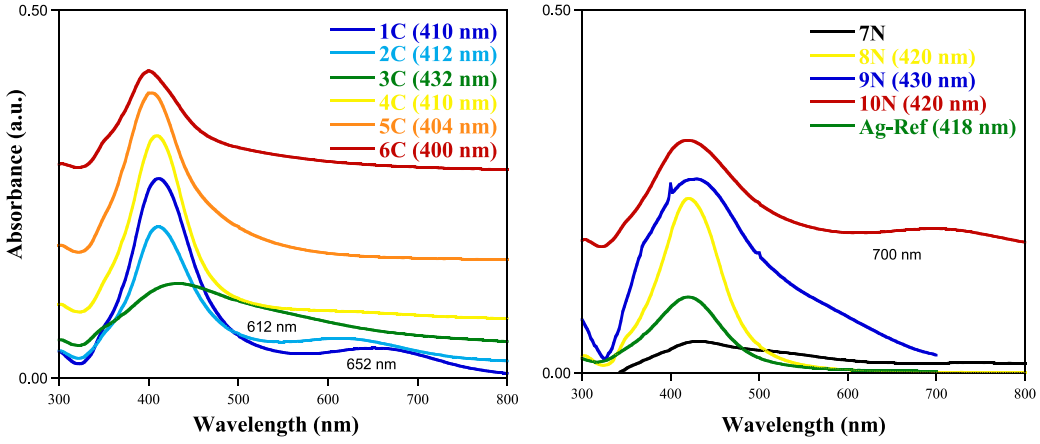

Fig. 1. UV-Vis absorption spectra of silver nanoparticles prepared via reduction of $AgNO_3$ using variable pH values. **Samples series C** (**left**), **N** and the **reference commercial product Ag-Ref** (**right**) are shown.

increasing pH occurred because the reduction of the silver precursor was promoted at elevating pH, alleging an enhancement of ascorbic acid activity.

Regarding **reference sample Ag-Ref**, a diameter of 4.8 ± 2.54 nm was found. Finally, **the samples of the N series**, whose synthesis procedure has been proposed for the first time in this work, contained nanoparticles with diameters ranging from 171.3 to 7.4 nm. Again, there was a trend of smaller sizes as the pH increased. Although sizes as small as those described in the samples of the C series were not achieved (perhaps because such high pH values were not reached), this series is more interesting than the one proposed by Qin et al. (Qin et al., 2010) since more varied and larger sizes were obtained. This suggests that the use of nitric acid instead of citric acid is more appealing if what is being sought is to synthesize nanoparticles with diameters as diverse as possible.

3.2. Synthesis of silver nanoparticles supported on clay minerals

In this study, silver nanoparticles supported on clay minerals were synthesised to overcome their potential aggregation. There are numerous works in the literature that employ different matrices to incorporate silver species, most of which use $AgNO_3$ as the silver source (Duncan and Pillai, 2015). For instance, inorganic polymers such as poly (vinyl alcohol) (PVA) (Kyrychenko et al., 2017) or polyethylene (Ibarra-Alonso et al., 2015) have been utilised as matrices. However, montmorillonite has been extensively employed as a carrier due to its unnumbered advantages, such as a large specific surface area, lack of toxicity, high natural availability, low cost and high intercalation and sportive capabilities (Liu et al., 2007; Miyoshi et al., 2010; Wu et al., 2010; Holešová et al., 2013; Praus et al., 2013; Cao et al., 2014; Lavorgna et al., 2014; Saha et al., 2014; Tian et al., 2014; Sohrabnezhad et al., 2015; Roy et al., 2017). In fact, montmorillonite has also been reported as a support for numerous species seeking for antibacterial activity, such as CuO nanocomposites (Nouri et al., 2018) and Cu (Yan et al., 2019), SnO_2 (Phukan et al., 2017), Ag_2CO_3 (Sohrabnezhad et al., 2015), Zn (Malachová et al., 2011) or Ag/TiO_2 (Wu et al., 2010) nanoparticles.

However, silver-montmorillonite nanohybrids have been reported to present better antibacterial properties than other metal species, due to the clay's high adsorptive capabilities for silver ions and nanoparticles (Roy et al., 2017). In addition, their cheap preparation (Giraldo et al., 2016) has made them a promising material with described applications in medicine as a wound curative (Subha et al., 2022), food packaging systems (Costa et al., 2012; Kuorwel et al., 2015) and paint products (Kaegi et al., 2010). For example, 20 mg of silver-montmorillonite nanoparticles were enough to prolong the shelf life of fresh fruit salad (Costa et al., 2011).

To synthesise these silver-clay hybrids, some metal cations of montmorillonite surface, like Mg^{2+}, Al^{3+} and $Fe^{2+/3+}$, are substituted for others with similar atomic size, without altering the SiO_4 groups of its octahedral sheet (Giraldo et al., 2016; Roy et al., 2017). Traditionally, the incorporated cations have come from surfactants, like polyurethane (Wang et al., 2012), but ionic liquids i.e., salt in the liquid state, are being increasingly employed since a more efficient substitution is achieved in less time (Takahashi et al., 2012; Giraldo et al., 2016). In this study an ionic liquid was utilised, as the clay minerals were impregnated with an $AgNO_3$ solution.

Once the Ag^+ ions are incorporated into the montmorillonite surface, it has been observed that the reduction of Ag^+ to Ag^0 and further coalescence of Ag^0 results in the formation of silver nanoparticles. Their immobilization on montmorillonite sheets prevents further agglomeration and allows a slow diffusion from the clay mineral, providing a prolonged antibacterial activity (Roy et al., 2017). To achieve this reduction, silver-loaded montmorillonite has been after-treated with reducing agents like glucose (Shabanzadeh et al., 2015; Sadeghianmaryan et al., 2021), ethylene glycol (Wang et al., 2018), quaternary ammonium nitrate (Zhang et al., 2018), N,N-dimethylformamide (Wang et al., 2018), sodium citrate (Bonga et al., 2016), hydrazine (Bagchi et al., 2014), sodium borohydride (Praus et al., 2013), ethanol (Wei et al., 2013), formaldehyde (Praus et al., 2010) or glycerol (Valásková et al., 2008). In addition, it has also been described how the use of electrochemical methods (Yuan and Golden, 2020), UV (Gabriel et al., 2017), microwave (Kesavan Pillai et al., 2013; Kheiralla et al., 2014) or γ (Kamyar et al., 2010) radiations can also promote this reduction. Even so, the formation of silver nanoparticles without the use of any reducing agents has also been reported, because montmorillonite itself not only prevents nanoparticle aggregation but also assists in the chemical reduction process of silver (Shameli et al., 2011). This is not unexpected, since silver element has a high reduction potential (+ 0.80 V), and silver nanoparticles may develop under the influence of heat and light (Roy et al., 2017). For this reason, no reducing agents nor further reducing processes were employed in this work other than the silver source and the solid support.

Although the support of silver nanoparticles in montmorillonite has been widely described, other clay minerals have also been employed as carriers, like kaolinite (Papp et al., 2008; Jou and Malek, 2016; Moosa, 2019; Bekissanova et al., 2022), halloysite (Hong et al., 2021; Shevtsova et al., 2022), sepiolite (Kahangi et al., 2020; Li et al., 2022), synthetic Laponite (Wu et al., 2018) or organovermiculite (Holešová et al., 2013). However, little has been reported about the antibacterial activity of silver nanoparticles supported on saponite, only the works of Sprynskyy et al. (Sprynskyy et al., 2019) and Sato et al. (Sato et al., 1997) to the best of the knowledge of the authors of this study. Saponite, although synthetic, serves as an important industrial material due to its low cost and easy production (Marchesi et al., 2020), finding applications in agriculture, construction, ceramics, catalysts and polymer materials (Carniato et al., 2020). Its high specific surface area, acidity, adsorption and thermal stability have been of great interest in these applications (Tao et al., 2016; Zhou et al., 2019). For this reason, in this work more research was done in this direction to compare which support, montmorillonite or saponite, results in a product with greater antibacterial activity.

In this study, silver nanoparticles were supported on montmorillonite and saponite employing two methods: wet impregnation and ion exchange. Both procedures allow the introduction of Ag^+ ions on the surface of the clay mineral substituting native ions, but their mechanisms of action are different. Wet impregnation consists of immersing the porous support in a solution, which enables fast and sufficient dispersion of the species of the solution into the porous structure of the support, if the solid support is porous (Krūmiņš et al., 2022), which montmorillonite is (Cecilia et al., 2013; Onwuka et al., 2021). Thus, wet impregnation allows more species to be loaded onto the carrier than other methods (Yuan et al., 2018; Tao et al., 2022), in addition to being a simpler, cheaper and faster procedure (Martín et al., 2016). However, the amount introduced into the clay mineral is theoretical, since in practice the exact amount that has been introduced is not known, although reaching the situation of incipient humidity allows controlling the amounts deposited (Jaber et al., 2014). The ion exchange method relies on the CEC value of the clay mineral, which describes its physical property of cation retention and diffusion processes of charged and uncharged molecules (Meier and Kahr, 1999). Therefore, this procedure requires knowing the CEC value, information that is not always available, and whose experimental determination is not easy and varies according to the method used. However, it achieves a greater dispersion of silver on the surface and smaller particle size than wet impregnation (Shameli et al., 2011; Giraldo et al., 2016; Roy et al., 2017).

It has been described how the initial concentration of $AgNO_3/Ag^+$ is the most important factor in determining the size of the silver nanoparticles that will form in the lamellar space of the clay minerals (Shameli et al., 2011; Giraldo et al., 2016). This is the reason why successive impregnations were carried out in this study to obtain **samples 11M-13M** and **14S-16S** since what was sought was to expose the clay

minerals to different concentrations of $AgNO_3$. Finally, the supported silver nanoparticles were characterised through SEM, EDX and TEM techniques.

The elemental composition of the samples was analysed through EDX technique. The mapping generated by this characterisation method allowed the observation of the silver content of the samples, which indicated successful incorporation of this species. Apart from the silver content, their aluminum and silicon content also stood out, which makes sense if the fact that both montmorillonite and saponite are layered aluminosilicates is considered. The characterisation also showed a regular distribution of the elements, which suggests that the samples were homogeneous and no dense particles of any kind were formed. For example, the elemental composition of **sample 13M** is shown in Fig. 2, where this description can be observed.

The surface morphology of the samples was investigated through SEM and TEM techniques. SEM studies revealed the typical morphology of these clay minerals, consisting of a "house of cards" morphology with layered flaky platelets superimposed on top of each other (Shameli et al., 2011; Roy et al., 2017). It has been described how the cause of such aggregated superstructure can be the hydration produced by Na^+ ions (Roy et al., 2017). The silver nanoparticles could also be observed, which suggests their adequate introduction in the structure of these materials, as shown in Fig. 3. The samples with montmorillonite and the samples with saponite showed practically indistinguishable morphology. In addition, the morphology of **reference samples 0M and 0S**, consisting of montmorillonite and saponite without silver loading, was also very similar to that of the samples, suggesting that the silver incorporation process did not imply a transgression of the morphology of the supports.

TEM studies showed small and spherical silver nanoparticles decorating the surface of the layers of the clay minerals, as seen in Fig. 4. The images allow observing darker zones, which correspond to the clay mineral platelets, and black circles on these darker zones, which are silver nanoparticles deposited on the platelets (Roy et al., 2017). In addition, a large number of silver nanoparticles were deposited at the outer surface of the clay minerals layers, which was also noticed by Shameli et al. (Shameli et al., 2011). Lastly, like in SEM micrographs, no significant morphological differences between **reference samples 0M and 0S** and the silver-loaded samples were appreciated.

As mentioned before, it has been described how different-sized silver nanoparticles can be synthesised in the lamellar space of clay minerals by varying the initial concentration of $AgNO_3/Ag^+$ (Shameli et al., 2011;

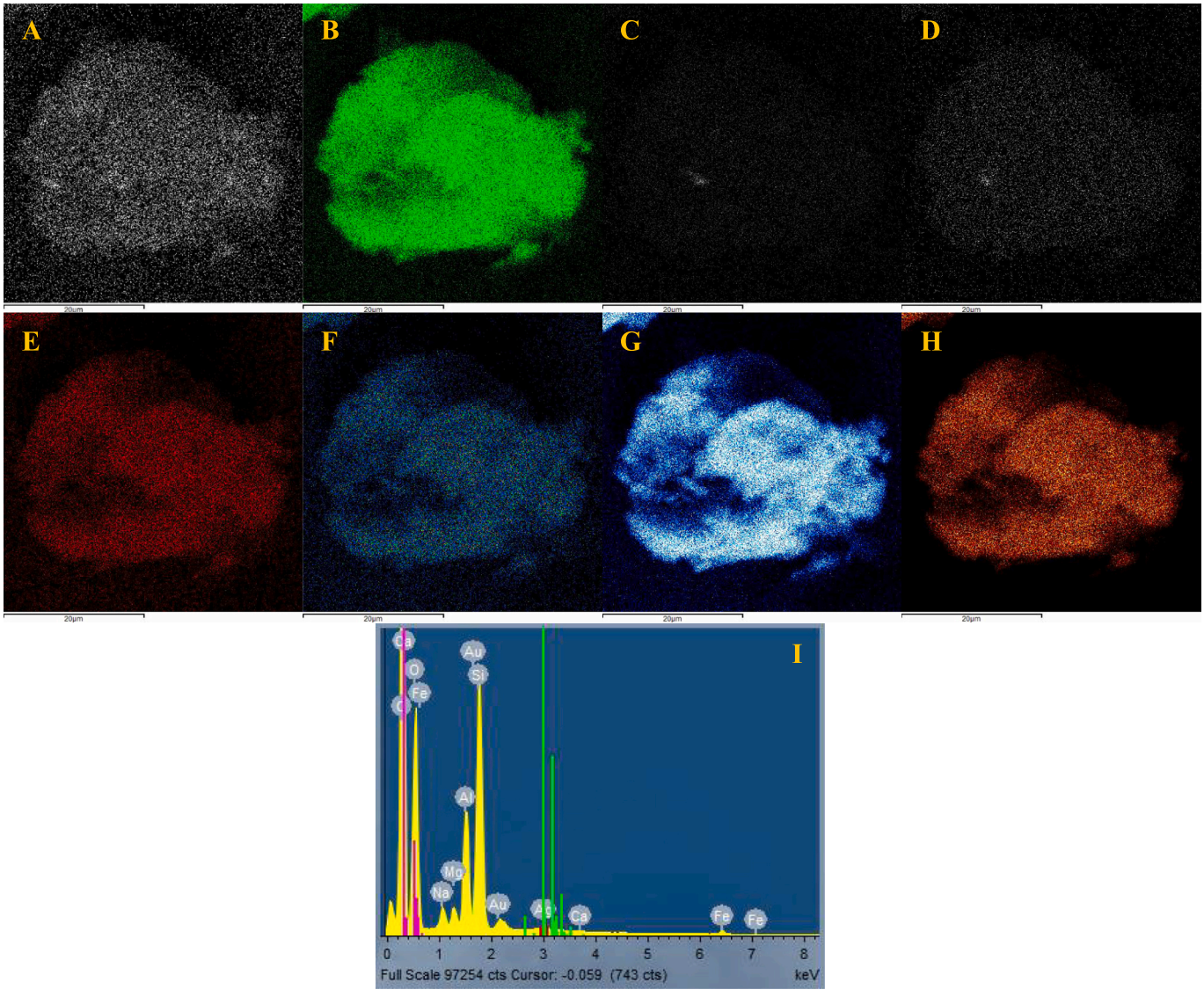

Fig. 2. Elemental composition of **sample 13M** analysed through EDX technique. Elemental mappings of silver (**A**), aluminum (**B**), calcium (**C**), iron (**D**), magnesium (**E**), sodium (**F**), oxygen (**G**), and silicon (**H**), in addition to the elemental analysis (**I**) are shown.

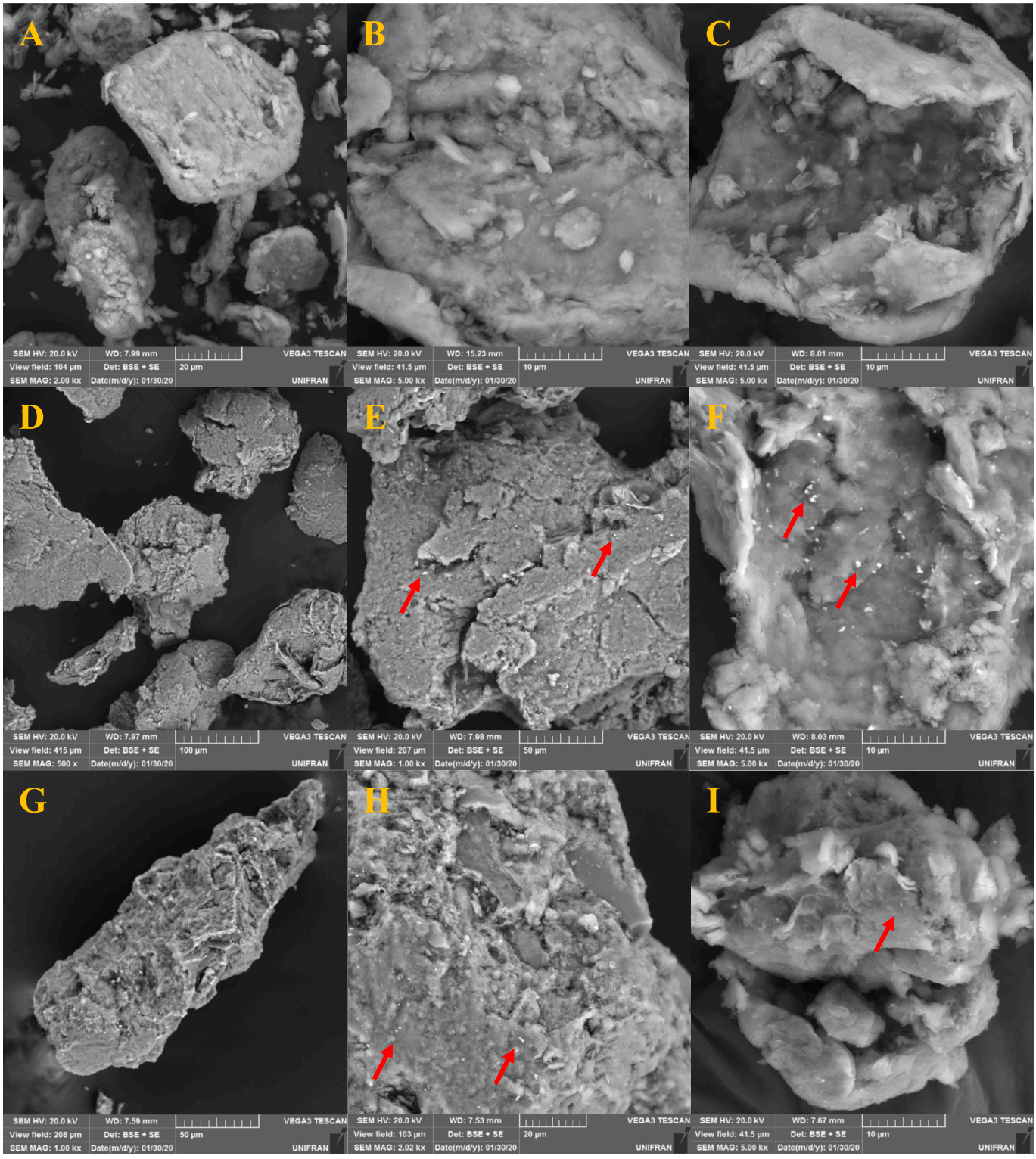

Fig. 3. SEM micrographs of **reference sample 0M** (**A-C**), **sample 13M** (**D-F**) and **sample 15S** (**G-I**). The typical montmorillonite morphology, consisting of a layered surface with some large flakes (Roy et al., 2017), and the supported silver nanoparticles (**red arrows**) can be observed. No significant morphological differences between montmorillonite without silver loading (**reference sample 0M**), the sample with montmorillonite (**sample 13M**) and the sample with saponite (**15S**) are appreciated.

Giraldo et al., 2016). However, there have been contradictions on this point. While some studies affirm that an increasing concentration of Ag^{+} ions allows larger particle sizes (Shameli et al., 2011) to be obtained, others state that larger particle size is obtained by decreasing the concentration of Ag^{+} ions (Roy et al., 2017). In this work it was not possible to determine the size of the supported silver nanoparticles, since TEM characterisation was used to qualitatively investigate the morphology of the samples and not to measure the particle size.

The conclusion that can be drawn from the results of the three characterisation techniques is that it was possible to verify the

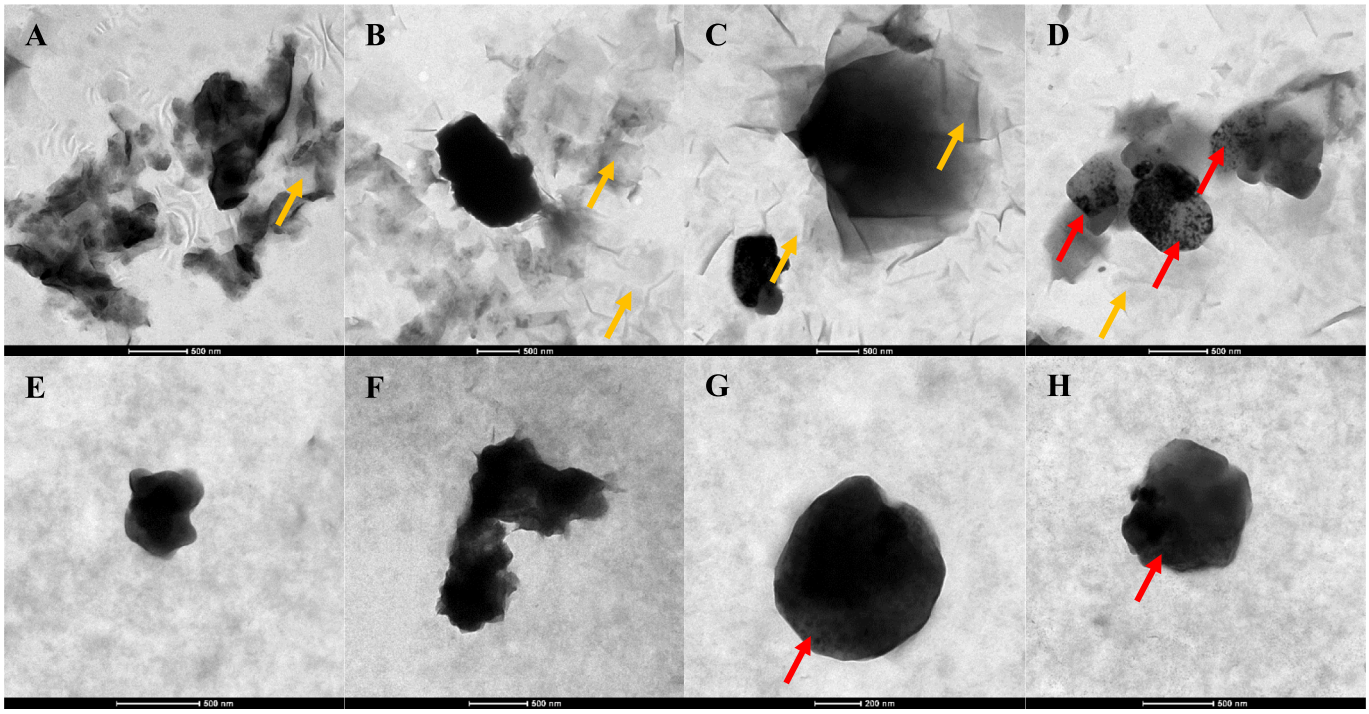

Fig. 4. TEM images of **reference sample 0M** (**A-B**), **sample 11M** (**C-D**), **reference sample 0S** (**E-F**) and **sample 14S** (**G-H**). Note the laminar structure of the clay minerals (**yellow arrows**). The darker zones correspond to the clay mineral platelets and the black circles on these darker zones are the silver nanoparticles deposited on the platelets (**red arrows** in **samples 11M and 14S**) (Roy et al., 2017). Note how a large number of silver nanoparticles are deposited at the outer surface of the clay minerals layers. No significant morphological differences between montmorillonite without silver loading (**reference sample 0M**), the sample with montmorillonite (**sample 11M**), saponite without silver loading (**reference sample 0S**) and the sample with saponite (**14S**) are appreciated.

incorporation of silver nanoparticles into clay minerals. Furthermore, no significant morphological differences were observed between **reference samples 0M and 0S** and the silver-loaded samples, suggesting that the silver incorporation process did not imply a transgression of the morphology of the supports.

3.3. Evaluation of the antibacterial activity

In this study, the antibacterial activity of silver nanoparticles and silver-loaded clay minerals was evaluated against Gram-negative *E. coli* and Gram-positive *S. aureus*. Most *E. coli* strains live harmlessly in the intestines, as this species is the predominant facultative anaerobe of the human colonic microbiota, and rarely causes disease in healthy individuals (Nataro and Kaper, 1998; Gomes et al., 2016). Nonetheless, there are pathogenic strains that present the ability to produce a broad spectrum of human diseases, like urinary tract infections, neonatal meningitis, enteritis, central nervous system disease, diarrheal illness and sepsis (Nataro and Kaper, 1998; Allocati et al., 2013; Gomes et al., 2016). In fact, diarrheagenic strains of *E. coli* are traditionally divided into dissimilar categories according to their pathogenic mechanism: enteropathogenic (EPEC), enterohemorrhagic (EHEC), enteroaggregative (EAEC), enterotoxigenic (ETEC) and enteroinvasive (EIEC) *E. coli* (Nataro and Kaper, 1998). Taken together, these organisms are probably the most common cause of severe pediatric diarrhea worldwide, especially in developing countries (Gomes et al., 2016; Ipshita et al., 2022) and a prominent cause of traveler's diarrhea (Hyesuk et al., 2022; Jones Jr. et al., 2022). *E. coli* is also an important cause of nosocomial infections (Alhazmi et al., 2022), and it is even possible to acquire the infection via the maternal-fetal pathway (Teweldemedhin et al., 2017). The treatment of *E. coli* infections is increasingly complicated because it is one of the most common multi-drug resistant bacteria (Abdi et al., 2022; Huang et al., 2022a), whose arousal is mainly due to the spread of mobile genetic elements, like plasmids (Allocati et al., 2013). The emergence of antimicrobial resistance has become a serious problem (Shahbazi et al., 2018, 2022), since multidrug-resistant bacteria cause 35000 deaths every year in the USA (Mahdi et al., 2022). It is estimated that the global annual mortality rate due to antibiotic resistance will exceed 10 million by 2050 (Opoku-Temeng et al., 2019; Mahdi et al., 2022).

On the other hand, *S. aureus* is part of the usual microbiota of the skin and nasopharynx (Foster, 2002), to the point that it is estimated that 25-40% of the human population will be colonised by this organism at some point (Lowy, 1998; Tong et al., 2015). However, it can also provoke a wide variety of infections like bacteremia, endocarditis, pneumonia and ocular, bone, skin, soft tissues and central nervous system infections (Foster, 2002; David and Daum, 2017; Ondusko and Nolt, 2018). Community-acquired (Muñoz et al., 2022) and nosocomial infections (Ghanbari et al., 2022) caused by *S. aureus* still pose a growing concern due to its high morbidity and mortality. In addition, this species is among the main ones presenting problems related to multidrug resistance (Rosa et al., 2022). In fact, already in the 1960s *S. aureus* strains resistant to methicillin were isolated, which is why these strains are still known today as "methicillin-resistant *S. aureus*" (MRSA), although methicillin is not used anymore (David and Daum, 2017). Likewise, *S. aureus* is the most common organism responsible for surgical site infections (Shetye et al., 2022), where it can also form biofilms on implants, catheters and other medical devices (Otto, 2008; Niraj et al., 2022; Pinto et al., 2022). Finally, *S. aureus* can also induce food poisoning by contaminating it with enterotoxins (Chopra et al., 2023).

E. coli and *S. aureus* have been chosen as target strains in this study because they embody the most representative species of the two main groups of bacteria based on their cell wall structure: Gram-negative and Gram-positive bacteria, respectively. Outside the bacterial cell membrane, Gram-positive bacteria present a thick layer of peptidoglycan of about 80 nm, reinforced with teichoic and teichuronic acids. On the other hand, in Gram-negative bacteria the plasma membrane is surrounded by the periplasmic space, followed by a thin peptidoglycan layer of approximately 8 nm and an additional outer membrane

enriched in lipopolysaccharide (LPS) 1-3 μm thick (Slavin et al., 2017). Outside these layers, both types may or may not present a capsule comprised of polysaccharides (Whitfield et al., 2020; Huang et al., 2022c). It has been suggested that this difference in wall structure may be responsible for the distinguished antibacterial activity of metal nanoparticles.

Regarding the antibacterial activity of silver nanoparticles, their exact mechanism of action is not completely understood. Several possibilities have been described in the literature, although it is not known which of them contributes the most to this effect. Firstly, silver nanoparticles may interfere with cell wall synthesis, denature ribosomes and impede DNA replication and other metabolic pathways (Yin et al., 2020). Silver nanoparticles may also release Ag^+ ions, which also participate in the antibacterial effect (Feng et al., 2000; Tran et al., 2016). Therefore, both Ag^+ and Ag^0 contribute to the antibacterial activity of silver nanoparticles (Morones et al., 2005).

One of the most described mechanisms of action is the disruption of the membrane and its power functions, like permeability and respiration (Murray et al., 1965; Feng et al., 2000). Silver nanoparticles are electrostatically attracted to negatively charged bacterial cell wall (Pan et al., 2013; Ivask et al., 2014). In the process, these particles accumulate in the bacterial membrane and periplasmic space, forming pits in the cell wall that increase its permeability and cause irreversible damage and bacterial death (Sondi and Salopek-Sondi, 2004). It has been suggested that the shape of the nanoparticles may influence this mechanism, since it seems that triangular ones have more effect than spherical or rod ones (Sukdeb et al., 2007). It has also been seen how Ag^+ can interact with phosphate, carboxyl and hydroxyl groups of lipoproteins of the bacterial cell wall (Giraldo et al., 2016), which can provoke plasmolysis (separation of cytoplasm from bacterial cell wall) and inhibition of bacterial cell wall synthesis (Song et al., 2006).

Another widely described mechanism of action is an increased production of reactive oxygen species (ROS) (Bragg and Rainnie, 1974; Ivask et al., 2014). ROS are highly reactive species of oxygen that are produced during basic cell metabolism when oxygen enters unwanted reduction states and transforms into free radicals, superoxides, and peroxides, rather than water (Slavin et al., 2017). Stress on the cell, like UV light, DNA damage or metal nanoparticles, can increase ROS production to a toxic level, causing important cell damage and even cell death by oxidising vital biomolecules (Madl et al., 2014). To counteract the action of ROS, cells have several antioxidant strategies, such as glutathione (GSH). GSH is a tripeptide that, when oxidised to glutathione disulfide (GSSG), can reduce ROS molecules and eliminate their harmful action (Ramalingam et al., 2016). In addition, the oxidative stress produced by ROS is quenched by the action of ubiquinone or coenzyme Q_{10}, a component of the electron transport chain that is essential for aerobic respiration (Ivask et al., 2012). However, the toxic level of ROS can surpass this antioxidant defense of the cell causing glutathione depletion. This produces damage to the cell membrane (Leung et al., 2014; Wang et al., 2014a), in addition to being able to produce lipid peroxidation, which inhibits bacterial growth (Kim et al., 2007; Jahnke et al., 2016). In addition to this lipid damage (Juan et al., 2021), ROS also induces DNA damage (Srinivas et al., 2019) and alterations in the structure and activity of proteins, as sulfur-containing amino acids like cysteine and methionine are highly susceptible to ROS action (Ezraty et al., 2017).

Increased ROS production has been also described as a mechanism of other metal nanoparticles, like ZnO (Padmavathy and Vijayaraghavan, 2008; Applerot et al., 2009), TiO_2 (Kumar et al., 2011), MgO (Leung et al., 2014) and Se (Geoffrion et al., 2020) nanoparticles. In this late case, Se presents a special effect in biofilm-forming bacteria (Tran and Webster, 2011; Shoeibi and Mashreghi, 2017), particularly if polymeric medical devices like polyvinyl chloride (PVC), polyurethane (PU) and silicone are coated with this species to avoid biofilm formation (Wang and Webster, 2012; Tran and Webster, 2013). Lastly, another example is the work of Habeeb Rahman et al. (Habeeb Rahman et al., 2018), where they were able to investigate through Field Emission Scanning Electron Microscopy (FE-SEM) how the increased production of ROS caused by ZnO:Fe nanoparticles was able to produce cell membrane rupture in *Salmonella typhimurium* cultures in just 45 min.

In addition to membrane disruption and increased ROS production, other antibacterial mechanisms of silver nanoparticles have also been described, like inhibition of proper ribosome function, enzyme inactivation or direct interaction with DNA, proteins and other metabolites (Shockman and Barren, 1983; Jiang et al., 2004; Raffi et al., 2007; Guzman et al., 2012; Slavin et al., 2017). Ag^+ also can produce protein denaturation, loss of DNA replication ability and inactivation of other cytoplasmic components (Feng et al., 2000; Kim, 2007; Gupta et al., 2008; Kumar et al., 2008). Although not yet known with certainty, it has been described that the most influential mechanism in Gram-negative bacteria may be membrane disruption and pits formation (Li et al., 2010; Mirzajani et al., 2011), while in Gram-positive bacteria it may be the destruction of the thick peptidoglycan layer (Mirzajani et al., 2011).

In this study, the antibacterial activity of silver nanoparticles and silver nanoparticles supported on clay minerals was evaluated against the reference strains *E. coli* ATCC 25922 and *S. aureus* ATCC 25923 by determining their MICs and MBCs according to CLSI guidelines. The results of these tests, which are shown in Table 4, were determined from three independent experiments, whose values obtained were identical. An MBC higher than 250 mg/L has been indicated in those situations where the MIC was 250 mg/L but when studying the MBC, growth of the bacteria was observed. No antibacterial activity was observed in **reference samples 0CN, 0M and 0S**, which were used as negative controls without silver loading. This suggests that the original solution of ascorbic acid and sodium citrate, montmorillonite and saponite do not contribute to the antibacterial activity of samples. Therefore, any observed effect must be attributed, in principle, to the incorporation of silver in the samples.

The first thing that stood out was that none of the samples of silver nanoparticles showed antibacterial activity at the maximum concentration of 125 mg/L. There may be various explanations for this. Firstly, it is possible that the specific synthesis procedure used in this study is not effective in preparing silver nanoparticles with antibacterial activity. In fact, Qin et al. (Qin et al., 2010) reported only the synthesis procedure but did not evaluate its antibacterial performance. This is a very useful finding because it implies that this methodology is not useful for synthesising silver nanoparticles with an antibacterial effect, contrary to other procedures reported in the literature (Ugwoke et al., 2020; Acharya et al., 2021; Bharti et al., 2021). In addition, those samples in which nitric acid was used to adjust the pH (**samples 7N-10N**) did not show any activity either. This is a valuable fact, because as the samples with citric acid did not present any effect, if the **samples 7N-10N** had

Table 4
MICs and MBCs for **reference sample Ag-Ref** and silver nanoparticles supported on clay minerals[a].

Sample	Silver concentration (mass %)	MIC (mg/L)		MBC (mg/L)	
		E. coli ATCC 25922	*S. aureus* ATCC 25923	*E. coli* ATCC 25922	*S. aureus* ATCC 25923
Ag-Ref	500[b]	31.25	125	125	> 250
11M	0.43	62.5	125	250	250
12M	1.29	31.25	62.5	31.25	250
13M	2.15	31.25	62.5	31.25	125
14S	0.43	125	125	125	250
15S	1.29	15.625	62.5	15.625	250
16S	2.15	31.25	62.5	31.25	62.5
17IM	6.72	250	250	250	> 250
18IS	6.72	250	250	250	250

[a] MICs and MBCs were calculated according to CLSI guidelines. These results were determined from 3 independent experiments, whose values obtained were identical.
[b] mg/L

done so, it would not have been possible to know whether to attribute it to the effect of the silver or to the low pH reached, since it has been described that acidic pH induces bacterial membrane disruption (Belfiore et al., 2007; Lou et al., 2011, 2012; Valle-González et al., 2018). The synergistic action of low pH and antimicrobials has also been reported. For instance, histidine-rich peptides (Mason et al., 2006; Kacprzyk et al., 2007), pyrazinamide (Zhang et al., 2003) or hydrogels with nanofiber networks (Wang et al., 2019) only produce membrane lysis under acidic pH conditions. Therefore, although as described in section 3.1 the use of nitric acid was more interesting since more varied and larger nanoparticle sizes were obtained, this did not translate into a higher antibacterial activity than the employment of citric acid.

Another possible explanation for the lack of activity of silver nanoparticles is that, despite the stabilising agent and the fact that the color of the samples remained unchanged for months and did not vary to dark gray, which may indicate aggregation (Mulfinger et al., 2007; Badiah et al., 2019), it is possible that eventually this agglomeration may have occurred. This could have reduced the interaction of the silver nanoparticles with the bacteria, thus averting the development of their effect. If so, it is likely that the aggregation caused a wide particle size distribution, and it has been described that high-performance antibacterial activity is only observed if silver nanoparticles have narrow particle size distribution (Girase et al., 2011).

An additional possible explanation could be the low concentration of silver in the samples, which was 125 mg/L. However, **reference sample Ag-Ref** (commercial colloidal silver solution) showed antibacterial activity with a MIC against *E. coli* of 31.25 mg/L. As this is lower than 125 mg/L, it does not seem that the lack of effect can be attributed to a concentration problem. In fact, even lower concentrations have been reported to present antibacterial activity, showing MICs of 15.6 mg/L against *E. coli* and 9.4 mg/L against *S. aureus* (Platania et al., 2022).

Regarding the antibacterial performance of silver-loaded clay minerals, all samples showed effect. This suggests that the incorporation of the nanoparticles in a support allows their controlled release, which prevents their agglomeration. This enables these samples to have more effect than silver nanoparticles (Roy et al., 2017). This was also seen in this work, as the MICs and MBCs of the samples were generally lower than those of the **reference sample Ag-Ref**. It has even been reported that the effect of supported silver nanoparticles is greater even when the silver content is at least 10 times less compared to solutions of silver nanoparticles (Roy et al., 2017).

It stood out that the samples synthesised via ion exchange (**samples 17IM and 18IS**) showed less activity than those prepared through wet impregnation (**samples 11M to 16S**). This fact may be because in these samples there were free particles between the clay layers due to the synthesis process, which makes silver nanoparticles more available to interact with bacterial components. This did not occur in ion exchange samples, where there were only particles on the slides themselves. This could also justify a much slower release, which could also explain the lower activity of these samples.

Within the samples synthesised through wet impregnation, although the trend was not entirely clear, it was observed that the higher the silver content of the sample, the greater its antibacterial effect, both in the samples with montmorillonite (**samples 11M to 13M**) and with saponite (**samples 14S to 16S**), which is according to what is described in the literature. This trend may be due to two reasons. On the one hand, the more silver there is in the support, the more it can be released and therefore the greater the effect (Roy et al., 2017). On the other hand, the higher the silver loading in the clay mineral, the smaller the size of the released nanoparticles (Roy et al., 2017; Huma et al., 2018). The smaller the particle, the larger the surface available for interaction with bacterial components, which gives it a stronger antibacterial performance (de Lacerda Coriolano et al., 2021; Mehata, 2021). In addition, lesser particle sizes lead to increased particle instability, which provokes the generation of more Ag^{+} ions that also participate in the bactericidal effect (Roy et al., 2017). Therefore, the concentration of $AgNO_3$ loaded on the clay minerals directly affected their antibacterial activity.

Finally, no noteworthy differences were observed between the activity developed by the samples with montmorillonite and the samples with saponite. This supports that saponite could be more commonly used as a support, not only because of what was discussed in section 3.2 on the correct incorporation of silver nanoparticles, but also because of the antibacterial activity similar to that of montmorillonite demonstrated in these MIC and MBC studies.

Likewise, it was observed that most of the samples presented a higher activity against *E. coli*, with greater MICs and MBCs than against *S. aureus*. It has been widely described that Gram-positive bacteria are more resistant to the mechanisms of action of metal nanoparticles (Slavin et al., 2017). The reason for this phenomenon is believed to be the differing cell walls. Although it has been determined that the cell wall of *E. coli* is more negative than that of *S. aureus* (Sonohara et al., 1995), which could attract silver nanoparticles with more intensity, this fact may not influence so much because both Gram-positive and Gram-negative bacteria have a negatively charged cell wall (Varghese and Balachandran, 2021). The reason for this increased resistance could be that Gram-positive bacteria present a much thicker peptidoglycan layer than Gram-negative bacteria (80 nm vs 8 nm), in addition to being reinforced with teichoic and teichuronic acids (Slavin et al., 2017). This makes the thick peptidoglycan layer a protective coating, without which the membrane disruption produced by metal nanoparticles is more injurious for Gram-negative bacteria.

In conclusion, the incorporation of silver nanoparticles into montmorillonite and saponite resulted in products that exhibited potent antibacterial activity. These agents are attractive because the synthesis processes are advantageously low-priced and uncomplicated, so they can be carried out even in the most underprivileged environments. In addition, they have an environment-friendly nature because the reagents employed in the preparation are not toxic nor dangerous, as some of those used in the synthesis of solutions of silver nanoparticle solutions could be. Therefore, these materials could be used as effective antibacterial agents, making the applicable to biomedical systems (Persano et al., 2021), food packaging (Azizi-Lalabadi et al., 2021) or even food containers (Diana-Carmen et al., 2022) in circumstances with few resources. Further research in this direction is needed, both to validate the effect of the materials on other bacterial, fungal and viral species, and to ensure that the material maintains its antibacterial activity over time.

3.4. Cytotoxicity assay

To ensure the safety of the samples, their cytotoxicity was evaluated in human fibroblasts (GM07492-A cell line) through an XTT colorimetric assay. In the same way that silver nanoparticles can be harmful to prokaryotic cells, they can also damage eukaryotic cells. It has been described how the strong oxidative activity of silver nanoparticles induces cytotoxicity, genotoxicity, immunological responses and even cell death in biological systems (Składanowski et al., 2016; Wypij et al., 2020).

Recent studies have focused on examining what characteristics determine the cytotoxicity of silver nanoparticles. Firstly, this activity depends on the size. The smaller the size, the greater the toxicity (Devanesan and Alsalhi, 2021), because a higher production of ROS (Nguyen et al., 2021), lactate dehydrogenase (LDH) (Sriram et al., 2012) or inflammation (Wang et al., 2014b) is induced, among other mechanisms. This inflammatory response is mediated by increased production of inflammatory mediators like tumor necrosis factor (TNF-α) and interleukins 6 and 8 (Zorraquín-Peña et al., 2020).

Their cytotoxicity also depends on the dose, since the higher the dose, the greater the toxicity (Iram et al., 2021), and on the aggregation, since more aggregated particles produce less toxicity (Lankoff et al., 2012). It also depends on the shape, as it can influence the cellular uptake mechanism, which in turn modulates cytotoxicity (Akter et al., 2018). For example, spherical nanoparticles appear to be more toxic

than other forms (Lee and Park, 2020). Likewise, when a coating is used to prevent aggregation, it also provides protection against cytotoxicity (Barbalinardo et al., 2021). This occurs because it maintains the surface chemistry of nanoparticles by increasing stability (Akter et al., 2018). Although the effect depends on the coating material, citrate (Malysheva et al., 2021) or PVP (Ilić et al., 2021) have been shown to reduce cytotoxicity more than other substances.

Silver nanoparticles can penetrate eukaryotic cells through different uptake mechanisms, such as diffusion, phagocytosis, endocytosis, the membrane flip-flop mechanism or even direct penetration via ion channels (Haase et al., 2011; Murugan et al., 2015). Once inside the eukaryotic cell, their main cytotoxic effect is apoptosis-mediated cell death (Mohamed et al., 2021), which can occur through multiple mechanisms, some of which are common to those already described in prokaryotic cells. Firstly, silver nanoparticles can produce an increased generation of ROS (Hailan et al., 2022), which induces oxidative stress that finally leads to apoptosis (Salama et al., 2022). The reduction of GSH levels through the inhibition of GSH synthesising enzyme (Aragoneses-Cazorla et al., 2022; El-Samad et al., 2022) and the depletion of the activity of other antioxidant enzymes (Amjad et al., 2022) also contribute to oxidative stress. The overproduction of ROS also induces apoptosis via p53, AKT or MAPK signaling pathways (Li et al., 2021; Chang et al., 2021b; Mohd Faheem et al., 2022), as well as through upregulation of pro-apoptotic Bax protein (Acharya et al., 2022).

Silver nanoparticles also can include an increase in LDH activity and leakage (Akter et al., 2021; Barbasz et al., 2022), epigenetic dysregulation through DNA methylation, which can cause reprogramming of gene expression (Mytych et al., 2017), arrest of the cell cycle in G_1 phase (Sun et al., 2021), membranolytic action (Alves et al., 2022) and loss of cytoskeleton proteins like β-tubulin and filamentous actin (Akter et al., 2018). They can also disrupt mitochondrial function (Jabeen et al., 2021) since they increase mitochondrial membrane permeability (Kaplan et al., 2022) and decrease mitochondrial membrane potential (Badirzadeh et al., 2022). This can induce the release of cytochrome c into the cytosol, which leads to apoptosis via caspase activation (Li et al., 1997; Plackal Adimuriyil George et al., 2018; Tang et al., 2019).

Finally, it has also been described that Ag^+ ions released from nanoparticles may be the initial factor for toxicity induction (Abdellatif et al., 2021), in addition to empowering the global toxicity of nanoparticles (Youssef et al., 2021). There are also studies that suggest that Ag^+ ions are responsible for the overall toxicity of nanoparticles and not the nanoparticles themselves (Xiu et al., 2012). In conclusion, silver nanoparticles reduce cell viability (Salesa et al., 2021).

Other metal nanoparticles also induce cytotoxicity by similar mechanisms. For example, TiO_2 nanoparticles cause increased production of ROS (Vigneshwaran et al., 2021; Pedrino et al., 2022). This mechanism also occurs in gold nanoparticles (Lorenzo-Anota et al., 2021).

In addition, several works have studied in vivo the effects of silver nanoparticles in mice and rats. For instance, they have been shown to produce neurotoxicity in brain regions like the caudate nucleus, frontal cortex and hippocampus (Rahman et al., 2009; Dziendzikowska et al., 2022). This happens because they can pass through the blood-brain barrier (BBB) by transcytosis of capillary endothelial cells (Gallardo-Toledo et al., 2021) or crossing the tight junctions of the BBB (Akter et al., 2018). They can also cause increased permeability of the BBB (Choudhary et al., 2022), astrocyte swelling (Opris et al., 2022), neuronal degeneration (Elblehi et al., 2022), synaptic disturbance (Chang et al., 2022) and depletion of dopamine (Attia et al., 2021). Silver also accumulates in other tissues like the cerebellum, muscle, spleen, duodenum and myocardial muscle (Pelkonen et al., 2003). It can also cause malformations and morphological deformities during embryo development in zebrafish (Cunningham et al., 2021). However, little is known about the impact on human physiology (González-Vega et al., 2022).

Although the cytotoxicity mechanisms of silver nanoparticles are still not entirely clear, there are many studies in this regard. However, there is a limited amount of work that focuses on the toxicity of silver-loaded clay minerals. It is generally accepted that they present lower cytotoxicity than nanoparticles with the same silver content (Long et al., 2022), perhaps because the existence of support reduces toxicity. The limited number of recent investigations that evaluated the cytotoxicity of silver-loaded montmorillonite found that there was hardly any disruption of cell viability or changes in cell morphology (Zhu et al., 2018; Ge et al., 2019; Huang et al., 2022b). Furthermore, this material was also not neurotoxic in mice (Daniel et al., 2010). However, to the best of the knowledge of the authors of this study, there are no studies that evaluate the cytotoxicity of silver-loaded saponite.

In this study, the cytotoxicity of selected samples was evaluated in human fibroblasts (GM07492-A cell line) through an XTT colorimetric assay. The chosen samples were **reference sample Ag-Ref**, and **samples 13M and 15S**, as they showed promising results in the antibacterial activity studies. Likewise, **reference samples 0M and 0S** were also included in this assay to determine the toxicity of montmorillonite and saponite without silver loading. The results of the cytotoxicity assay are shown in Table 5.

The XTT assay is a colorimetric method first described by Scudiero et al. in 1988 (Scudiero et al., 1988). It is a fast, simple, accurate and highly sensitive way to determine cytotoxicity (Karatop et al., 2022). It consists of evaluating how cells respond to different substances by assessing cell proliferation, quantification and viability. After incubation of the cell line, the substances to be tested are added. After further incubation, XTT is also added. Metabolically active cells initiate a redox reaction that consists in reducing the tetrazolium salt XTT to orange-colored formazan, which is soluble in water and thus the color intensity can be measured with a spectrophotometer (Karatop et al., 2022). As the intensity of the color detected is proportional to the number of metabolically active cells (Riss et al., 2004), cell viability and values like the IC_{50} can be assessed after designating the negative control without testing substances as the 100%.

Reference sample Ag-Ref presented an IC_{50} of 35.3 ± 6.8 μg/mL. It has been described in the literature how silver nanoparticles presented similar IC_{50} values, such as 16.3 μg/mL in human macrophages (RAW 264.7 cell line) (Wypij et al., 2021), 27.98 μg/mL in cervical cancer cells (HeLa cell line) (Wei et al., 2020), 40 μg/mL in breast cancer cells (MCF-7 cell line) (Jiao et al., 2014), or 80 μg/mL in lung adenocarcinoma cells (A549 cell line) (Majeed et al., 2016). Other obtained higher toxicity levels, with values of 147.175 μg/mL in MCF-7 cell line (Das et al., 2022). In this work, the cytotoxicity of the samples of silver nanoparticles was not evaluated due to their lack of antibacterial activity.

Reference samples 0M and 0S presented IC_{50} values of 1446 ± 162.9 and 687.7 ± 17.9 μg/mL, respectively. This suggests that the cytotoxicity of montmorillonite and saponite is scarce. As expected, after loading silver, the IC_{50} values decreased to 308.6 ± 7.4 and 553.9 ± 36.8 μg/mL for **samples 13M and 15S**, respectively. These values are highly promising because they suggest that silver-loaded clay minerals have an exceptional safety profile. These IC_{50} values may be lower than those of **reference sample Ag-Ref** because, as it has been previously

Table 5
IC_{50} values obtained against human fibroblasts (GM07492-A cell line) through an XTT colorimetric assay after 24 h of treatment with the samples.

Sample	Silver concentration (mass %)	IC_{50} (μg/mL)
Ag-Ref	500[a]	35.3 ± 6.8
0M	-	1446 ± 162.9
13M	2.15	308.6 ± 7.4
0S	-	687.7 ± 17.9
15S	1.29	553.9 ± 36.8

IC_{50} represents the sample concentration that inhibits 50% of cell viability. Results are presented as mean ± standard deviation of 3 independent experiments.
[a] mg/L

discussed, coating reduces cytotoxicity. No IC_{50} values could be found in the literature for comparison. This is because, to the best of the knowledge of the authors of this study, the cytotoxicity of silver-loaded montmorillonite has been evaluated through alternative methods and there is no prior estimation of the cytotoxicity of silver-loaded saponite. Therefore, this study presents valuable results since it represents the first time that IC_{50} values of silver-loaded montmorillonite and saponite have been calculated. The promising obtained results suggest that these materials present a high-grade safety profile.

Therefore, these materials have the potential to be employed safely in biological systems. Extensive research in this direction is needed to further evaluate the safety of silver nanoparticles and silver-loaded clay minerals.

4. Summary and conclusions

In summary, in this study the antibacterial activity and cytotoxicity of silver nanoparticles and silver nanoparticles supported on clay minerals were compared. Silver nanoparticles were synthesised by chemical reduction using $AgNO_3$ as the silver precursor, ascorbic acid as the reducing agent and sodium citrate as the stabiliser agent. Silver nanoparticles were supported on montmorillonite and saponite via wet impregnation or ion exchange methods. The antibacterial activity of the samples was evaluated against *E. coli* ATCC 25922 and *S. aureus* ATCC 25923 by determining their MICs and MBCs according to CLSI guidelines. Finally, the cytotoxicity of the samples was determined in GM07492-A cell line employing an XTT colorimetric assay.

The results found that the use of nitric acid instead of citric acid to adjust the pH of the silver nanoparticles during the synthesis process was more interesting since more varied and larger nanoparticle sizes were obtained. Also, as the pH of the samples increased, the nanoparticle size decreased.

The characterization techniques employed verified the successful incorporation of silver nanoparticles into clay minerals. No significant morphological differences were observed between montmorillonite and saponite and before and after silver incorporation, which suggests that both clay minerals provide similar support and that the silver incorporation process does not imply a transgression nor modification of their morphology.

The silver nanoparticles synthesised did not present any antibacterial activity. Therefore, the synthesis procedure used is not useful for this purpose, whether citric acid or nitric acid is used to adjust the pH during the synthesis process. In contrast, silver-loaded clay minerals did show antibacterial activity, which was greater in those samples synthesised by wet impregnation and with a higher silver content. The effect was greater against *E. coli*. The effect was similar whether montmorillonite or saponite was used as the support.

Finally, silver-loaded montmorillonite and saponite were less toxic than reference sample Ag-Ref and presented IC_{50} values suggestive of a high-grade safety profile. This was the first time, to the best of the knowledge of the authors and from the revised literature, that the IC_{50} value was calculated in silver-loaded montmorillonite and saponite.

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CRediT authorship contribution statement

Adrián Gil-Korilis: Project administration, Supervision, Conceptualization, Methodology, Resources, Investigation, Data curation, Validation, Writing – original draft, Writing – review & editing, Visualization. **Mihail Cojocaru:** Conceptualization, Investigation, Data curation, Writing – review & editing, Visualization. **Melibea Berzosa:** Methodology, Investigation, Writing – review & editing. **Carlos Gamazo:** Supervision, Methodology, Resources, Writing – review & editing. **Natália J. Andrade:** Methodology, Resources, Investigation, Writing – review & editing. **Katia J. Ciuffi:** Supervision, Resources, Writing – review & editing.

Declaration of Competing Interest

The authors declare no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request from the corresponding author.

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Appendix A. Supplementary data

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References

Abdellatif, A.A.H., Alsharidah, M., Al Rugaie, O., Tawfeek, H., Sameh, N., 2021. Silver nanoparticle-coated ethyl cellulose inhibits tumor necrosis factor-α of breast cancer cells. Drug Des. Devel. Ther. 15, 2035–2046. https://doi.org/10.2147/DDDT.S310760.

Abdi, H., Hassan, M., Abdulaziz, H., Omer, N., 2022. Antiboitic resistance of uropathogenic Eshcherichia coli in pateints of Hargeisa Group Hospital, Hargeisa, Somaliand. Adv. Microbiol. 12, 333–342. https://doi.org/10.4236/aim.2022.125024.

Acharya, D., Pandey, P., Mohanta, B., 2021. A comparative study on the antibacterial activity of different shaped silver nanoparticles. Chem. Pap. 75, 4907–4915. https://doi.org/10.1007/s11696-021-01722-8.

Acharya, D., Satapathy, S., Yadav, K.K., Somu, P., Mishra, G., 2022. Systemic evaluation of mechanism of cytotoxicity in human colon cancer HCT-116 cells of silver nanoparticles synthesized using marine algae Ulva lactuca extract. J. Inorg. Organomet. Polym. Mater. 32, 596–605. https://doi.org/10.1007/s10904-021-02133-8.

Akter, M., Sikder, Md.T., Rahman, Md.M., Ullah, A.K.M.A., Hossain, K.F.B., Banik, S., Hosokawa, T., Saito, T., Kurasaki, M., 2018. A systematic review on silver nanoparticles-induced cytotoxicity: Physicochemical properties and perspectives. J. Adv. Res. 9, 1–16. https://doi.org/10.1016/j.jare.2017.10.008.

Akter, M., Atique Ullah, A.K.M., Banik, S., Sikder, Md.T., Hosokawa, T., Saito, T., Kurasaki, M., 2021. Green synthesized silver nanoparticles-mediated cytotoxic effect in colorectal cancer cells: NF-κB signal induced apoptosis through autophagy. Biol. Trace Elem. Res. 199, 3272–3286. https://doi.org/10.1007/s12011-020-02463-7.

Algotiml, R., Gab-Alla, A., Seoudi, R., Abulreesh, H.H., El-Readi, M.Z., Elbanna, K., 2022. Anticancer and antimicrobial activity of biosynthesized Red Sea marine algal silver nanoparticles. Sci. Rep. 12, 2421. https://doi.org/10.1038/s41598-022-06412-3.

Alhazmi, A., Abuallut, I., Alwadani, I., Haddad, M., Ageeli, B., Majrabi, H., Muslihi, I., AlAli, L., Homadi, H., Madkhli, E., Shami, J., 2022. Neonatal healthcare-associated conjunctivitis: A descriptive study from Saudi Arabia. Medicina (B Aires) 58, 1448. https://doi.org/10.3390/medicina58101448.

Allocati, N., Masulli, M., Alexeyev, M.F., Di Ilio, C., 2013. Escherichia coli in Europe: An overview. Int. J. Environ. Res. Public Health 10, 6235–6254. https://doi.org/10.3390/ijerph10126235.

Almanza-Reyes, H., Moreno, S., Plascencia-López, I., Alvarado-Vera, M., Patrón-Romero, L., Borrego, B., Reyes-Escamilla, A., Valencia-Manzo, D., Brun, A., Pestryakov, A., Bogdanchikova, N., 2021. Evaluation of silver nanoparticles for the prevention of SARS-CoV-2 infection in health workers: In vitro and in vivo. PLoS One 16, e0256401. https://doi.org/10.1371/journal.pone.0256401.

Alves, M.F., Paschoal, A.C.C., Klimeck, T.D.F., Kuligovski, C., Marcon, B.H., de Aguiar, A. M., Murray, P.G., 2022. Biological synthesis of low cytotoxicity silver nanoparticles (AgNPs) by the fungus Chaetomium thermophilum-Sustainable nanotechnology. J. Fungi 8, 605. https://doi.org/10.3390/jof8060605.

Amjad, T., Afsheen, S., Iqbal, T., 2022. Nanocidal effect of rice husk–based silver nanoparticles on antioxidant enzymes of aphid. Biol. Trace Elem. Res. 200, 4855–4864. https://doi.org/10.1007/s12011-021-03067-5.

Antonelli, M., De Pascale, G., Ranieri, V.M., Pelaia, P., Tufano, R., Piazza, O., Zangrillo, A., Ferrario, A., De Gaetano, A., Guaglianone, E., Donelli, G., 2012. Comparison of triple-lumen central venous catheters impregnated with silver nanoparticles (AgTive®) vs conventional catheters in intensive care unit patients. J. Hosp. Infect. 82, 101–107. https://doi.org/10.1016/j.jhin.2012.07.010.

Applerot, G., Lipovsky, A., Dror, R., Perkas, N., Nitzan, Y., Lubart, R., Gedanken, A., 2009. Enhanced antibacterial activity of nanocrystalline ZnO due to increased ROS-mediated cell injury. Adv. Funct. Mater. 19, 842–852. https://doi.org/10.1002/adfm.200801081.

Aragoneses-Cazorla, G., Buendia-Nacarino, M.P., Mena, M.L., Luque-Garcia, J.L., 2022. A multi-omics approach to evaluate the toxicity mechanisms associated with silver nanoparticles exposure. Nanomaterials 12, 1762. https://doi.org/10.3390/nano12101762.

Attia, A., Ramadan, H., ElMazoudy, R., Abdelnaser, A., 2021. Disruption of brain conductivity and permittivity and neurotransmitters induced by citrate-coated silver nanoparticles in male rats. Environ. Sci. Pollut. Res. 28, 38332–38347. https://doi.org/10.1007/s11356-021-13397-5.

Azizi-Lalabadi, M., Garavand, F., Jafari, S.M., 2021. Incorporation of silver nanoparticles into active antimicrobial nanocomposites: Release behavior, analyzing techniques, applications and safety issues. Adv. Colloid Interf. Sci. 293, 102440 https://doi.org/10.1016/j.cis.2021.102440.

Badiah, H.I., Seedeh, F., Supriyanto, G., Zaidan, A.H., 2019. Synthesis of silver nanoparticles and the development in analysis method. In: IOP Conference Series: Earth and Environmental Science. Institute of Physics Publishing. https://doi.org/10.1088/1755-1315/217/1/012005.

Badirzadeh, A., Alipour, M., Najm, M., Vosoogh, A., Vosoogh, M., Samadian, H., Hashemi, A.S., Farsangi, Z.J., Amini, S.M., 2022. Potential therapeutic effects of curcumin coated silver nanoparticle in the treatment of cutaneous leishmaniasis due to Leishmania major in-vitro and in a murine model. J. Drug. Deliv. Sci. Technol. 74, 103576 https://doi.org/10.1016/j.jddst.2022.103576.

Bagchi, B., Thakur, P., Kool, A., Das, S., Nandy, P., 2014. In situ synthesis of environmentally benign montmorillonite supported composites of Au/Ag nanoparticles and their catalytic activity in the reduction of p-nitrophenol. RSC Adv. 4, 61114. https://doi.org/10.1039/C4RA11108G.

Balčiūnaitienė, A., Liaudanskas, M., Puzerytė, V., Viškelis, J., Janulis, V., Viškelis, P., Griškonis, E., Jankauskaitė, V., 2022. Eucalyptus globulus and Salvia officinalis extracts mediated green synthesis of silver nanoparticles and their application as an antioxidant and antimicrobial agent. Plants 11, 1085. https://doi.org/10.3390/plants11081085.

Barbalinardo, M., Bertacchini, J., Bergamini, L., Magarò, M.S., Ortolani, L., Sanson, A., Palumbo, C., Cavallini, M., Gentili, D., 2021. Surface properties modulate protein corona formation and determine cellular uptake and cytotoxicity of silver nanoparticles. Nanoscale 13, 14119–14129. https://doi.org/10.1039/D0NR08259G.

Barbasz, A., Czyżowska, A., Piergies, N., Oćwieja, M., 2022. Design cytotoxicity: The effect of silver nanoparticles stabilized by selected antioxidants on melanoma cells. J. Appl. Toxicol. 42, 570–587. https://doi.org/10.1002/jat.4240.

Baselga, M., Uranga-Murillo, I., de Miguel, D., Arias, M., Sebastián, V., Pardo, J., Arruebo, M., 2022. Silver nanoparticles-polyethyleneimine-based coatings with antiviral activity against SARS-CoV-2: A new method to functionalize filtration Media. Materials 15, 4742. https://doi.org/10.3390/ma15144742.

Bekissanova, Z., Railean, V., Brzozowska, W., Wojtczak, I., Ospanova, A., Buszewski, B., Sprynskyy, M., 2022. Synthesis, characterization of silver/kaolinite nanocomposite and studying its antibacterial activity. Colloids Surf. B: Biointerfaces 220, 112908. https://doi.org/10.1016/j.colsurfb.2022.112908.

Belfiore, C., Castellano, P., Vignolo, G., 2007. Reduction of Escherichia coli population following treatment with bacteriocins from lactic acid bacteria and chelators. Food Microbiol. 24, 223–229. https://doi.org/10.1016/j.fm.2006.05.006.

Bhakya, S., Muthukrishnan, S., Sukumaran, M., Muthukumar, M., 2016. Biogenic synthesis of silver nanoparticles and their antioxidant and antibacterial activity. Appl. Nanosci. 6, 755–766. https://doi.org/10.1007/s13204-015-0473-z.

Bharti, S., Mukherji, Soumyo, Mukherji, Suparna, 2021. Enhanced antibacterial activity of decahedral silver nanoparticles. J. Nanopart. Res. 23, 36. https://doi.org/10.1007/s11051-020-05106-z.

Bonga, D.L.S., Pinto Ma, M.F.B., Tayad, M.F.T., 2016. Synthesis and characterization of silver nanoparticles anchored on montmorillonite via chemical reduction. Nano Hybrids Compos. 11, 30–37. https://doi.org/10.4028/www.scientific.net/NHC.11.30.

Bragg, P.D., Rainnie, D.J., 1974. The effect of silver ions on the respiratory chain of Escherichia coli. Can. J. Microbiol. 20, 883–889. https://doi.org/10.1139/m74-135.

Cao, G.F., Sun, Y., Chen, J.G., Song, L.P., Jiang, J.Q., Liu, Z.T., Liu, Z.W., 2014. Sutures modified by silver-loaded montmorillonite with antibacterial properties. Appl. Clay Sci. 93–94, 102–106. https://doi.org/10.1016/j.clay.2014.03.007.

Carniato, F., Gatti, G., Bisio, C., 2020. An overview of the recent synthesis and functionalization methods of saponite clay. New J. Chem. 44, 9969–9980. https://doi.org/10.1039/D0NJ00253D.

Cecilia, J.A., Jiménez-Gómez, C.P., 2021. Catalytic Applications of Clay Minerals and Hydrotalcites. Catalysts 11. https://doi.org/10.3390/catal11010068.

Cecilia, J.A., García-Sancho, C., Franco, F., 2013. Montmorillonite based porous clay heterostructures: Influence of Zr in the structure and acidic properties. Microporous Mesoporous Mater. 176, 95–102. https://doi.org/10.1016/j.micromeso.2013.03.037.

Chang, C.-H., Lee, Y.-H., Liao, Z.-H., Chen, M.H.-C., Peng, F.-C., Lin, J.-J., 2021a. Composition of nanoclay supported silver nanoparticles in furtherance of mitigating cytotoxicity and genotoxicity. PLoS One 16, e0247531. https://doi.org/10.1371/journal.pone.0247531.

Chang, X., Wang, X., Li, J., Shang, M., Niu, S., Zhang, W., Li, Y., Sun, Z., Gan, J., Li, W., Tang, M., Xue, Y., 2021b. Silver nanoparticles induced cytotoxicity in HT22 cells through autophagy and apoptosis via PI3K/AKT/mTOR signaling pathway. Ecotoxicol. Environ. Saf. 208, 111696 https://doi.org/10.1016/j.ecoenv.2020.111696.

Chang, X., Niu, S., Shang, M., Li, J., Zhang, W., Sun, Z., Li, Y., Wu, T., Zhang, T., Tang, M., Xue, Y., 2022. Silver nanoparticles induced hippocampal neuronal damage involved in mitophagy, mitochondrial biogenesis and synaptic degeneration. Food Chem. Toxicol. 166, 113227 https://doi.org/10.1016/j.fct.2022.113227.

Chhatre, A., Solasa, P., Sakle, S., Thaokar, R., Mehra, A., 2012. Color and surface plasmon effects in nanoparticle systems: Case of silver nanoparticles prepared by microemulsion route. Colloids Surf. A Physicochem. Eng. Asp. 404, 83–92. https://doi.org/10.1016/j.colsurfa.2012.04.016.

Chopra, A., Swami, A., Sharma, R., Devi, N., Mittal, S., Sharma, R.K., Wangoo, N., 2023. Femtomolar detection of staphylococcal enterotoxin 'B' using a fluorescent quantum dot based hybrid Apta-immunosensor. Spectrochim. Acta A Mol. Biomol. Spectrosc. 287, 122036 https://doi.org/10.1016/j.saa.2022.122036.

Choudhary, A., Singh, S., Ravichandiran, V., 2022. Toxicity, preparation methods and applications of silver nanoparticles: an update. Toxicol. Mech. Methods 32, 650–661. https://doi.org/10.1080/15376516.2022.2064257.

Costa, C., Conte, A., Buonocore, G.G., Del Nobile, M.A., 2011. Antimicrobial silver-montmorillonite nanoparticles to prolong the shelf life of fresh fruit salad. Int. J. Food Microbiol. 148, 164–167. https://doi.org/10.1016/j.ijfoodmicro.2011.05.018.

Costa, C., Conte, A., Buonocore, G.G., Lavorgna, M., Del Nobile, M.A., 2012. Calcium-alginate coating loaded with silver-montmorillonite nanoparticles to prolong the shelf-life of fresh-cut carrots. Food Res. Int. 48, 164–169. https://doi.org/10.1016/j.foodres.2012.03.001.

Cunningham, B., Engstrom, A.M., Harper, B.J., Harper, S.L., Mackiewicz, M.R., 2021. Silver nanoparticles stable to oxidation and silver ion release show size-dependent toxicity in vivo. Nanomaterials 11, 1516. https://doi.org/10.3390/nano11061516.

Daniel, S.C.G.K., Tharmaraj, V., Sironmani, T.A., Pitchumani, K., 2010. Toxicity and immunological activity of silver nanoparticles. Appl. Clay Sci. 48, 547–551. https://doi.org/10.1016/j.clay.2010.03.001.

Das, G., Shin, H.-S., Kumar, A., Vishnuprasad, C.N., Patra, J.K., 2021. Photo-mediated optimized synthesis of silver nanoparticles using the extracts of outer shell fibre of Cocos nucifera L. fruit and detection of its antioxidant, cytotoxicity and antibacterial potential. Saudi J. Biol. Sci. 28, 980–987. https://doi.org/10.1016/j.sjbs.2020.11.022.

Das, P., Dutta, T., Manna, S., Loganathan, S., Basak, P., 2022. Facile green synthesis of non-genotoxic, non-hemolytic organometallic silver nanoparticles using extract of crushed, wasted, and spent Humulus lupulus (hops): Characterization, anti-bacterial, and anti-cancer studies. Environ. Res. 204, 111962 https://doi.org/10.1016/j.envres.2021.111962.

David, M.Z., Daum, R.S., 2017. Treatment of Staphylococcus aureus infections. In: Bagnoli, F., Rappuoli, R., Grandi, G. (Eds.), Staphylococcus Aureus: Microbiology, Pathology, Immunology, Therapy and Prophylaxis. Springer International Publishing, Cham, pp. 325–383. https://doi.org/10.1007/82_2017_42.

de Lacerda Coriolano, D., de Souza, J.B., Bueno, E.V., de Medeiros, S.M.F.R., dos Cavalcanti, S., I.D.L., Cavalcanti, I.M.F, 2021. Antibacterial and antibiofilm potential of silver nanoparticles against antibiotic-sensitive and multidrug-resistant Pseudomonas aeruginosa strains. Braz. J. Microbiol. 52, 267–278. https://doi.org/10.1007/s42770-020-00406-x.

Devanesan, S., Alsalhi, M.S., 2021. Green synthesis of silver nanoparticles using the flower extract of abelmoschus esculentus for cytotoxicity and antimicrobial studies. Int. J. Nanomedicine 16, 3343–3356. https://doi.org/10.2147/IJN.S307676.

Diana-Carmen, M., Dumitra, R., Ana-Maria, G., Ana-Maria, R., Andrei, C.V., Valentin, Z., Ileana-Denisa, N., 2022. Silver nanoparticles incorporated on natural clay as an inhibitor against the new ISO SS bacteria isolated from Sewage Sludge, involved in malachite green dye oxidation. Molecules 27. https://doi.org/10.3390/molecules27185791.

Duncan, T.V., Pillai, K., 2015. Release of engineered nanomaterials from polymer nanocomposites: diffusion, dissolution, and desorption. ACS Appl. Mater. Interfaces 7, 2–19. https://doi.org/10.1021/am5062745.

Dziendzikowska, K., Wilczak, J., Grodzicki, W., Gromadzka-Ostrowska, J., Węsierska, M., Kruszewski, M., 2022. Coating-dependent neurotoxicity of silver nanoparticles-An in vivo study on hippocampal oxidative stress and neurosteroids. Int. J. Mol. Sci. 23, 1365. https://doi.org/10.3390/ijms23031365.

Echegoyen, Y., Nerín, C., 2013. Nanoparticle release from nano-silver antimicrobial food containers. Food Chem. Toxicol. 62, 16–22. https://doi.org/10.1016/j.fct.2013.08.014.

Elblehi, S.S., Abd El-Maksoud, E.M., Aldhahrani, A., Alotaibi, S.S., Ghamry, H.I., Elgendy, S.A., Soliman, M.M., Shukry, M., 2022. Quercetin abrogates oxidative neurotoxicity induced by silver nanoparticles in Wistar rats. Life 12, 578. https://doi.org/10.3390/life12040578.

El-Desouky, N., Shoueir, K., El-Mehasseb, I., El-Kemary, M., 2022. Synthesis of silver nanoparticles using bio valorization coffee waste extract: photocatalytic flow-rate performance, antibacterial activity, and electrochemical investigation. Biomass Convers Biorefin. 1–15 https://doi.org/10.1007/s13399-021-02256-5.

El-Samad, L.M., Bakr, N.R., El-Ashram, S., Radwan, E.H., Abdul Aziz, K.K., Hussein, H.K., El Wakil, A., Hassan, M.A., 2022. Silver nanoparticles instigate physiological, genotoxicity, and ultrastructural anomalies in midgut tissues of beetles. Chem. Biol. Interact. 367, 110166 https://doi.org/10.1016/j.cbi.2022.110166.

Evanoff, D.D., Chumanov, G., 2004. Size-controlled synthesis of nanoparticles. 1. "Silver-only" aqueous suspensions via hydrogen reduction. J. Phys. Chem. B 108, 13948–13956. https://doi.org/10.1021/jp047565s.

Ezraty, B., Gennaris, A., Barras, F., Collet, J.-F., 2017. Oxidative stress, protein damage and repair in bacteria. Nat. Rev. Microbiol. 15, 385–396. https://doi.org/10.1038/nrmicro.2017.26.

Feng, Q.L., Wu, J., Chen, G.Q., Cui, F.Z., Kim, T.N., Kim, J.O., 2000. A mechanistic study of the antibacterial effect of silver ions on Escherichia coli and Staphylococcus aureus. J. Biomed. Mater. Res. 52, 662–668. https://doi.org/10.1002/1097-4636(20001215)52:4<662::AID-JBM10>3.0.CO;2-3.

Foster, T., 2002. Staphylococcus aureus. In: Sussman, M. (Ed.), Molecular Medical Microbiology. Academic Press, London, pp. 839–888. https://doi.org/10.1016/B978-012677530-3/50258-0.

Gabriel, J.S., Gonzaga, V.A.M., Poli, A.L., Schmitt, C.C., 2017. Photochemical synthesis of silver nanoparticles on chitosans/montmorillonite nanocomposite films and antibacterial activity. Carbohydr. Polym. 171, 202–210. https://doi.org/10.1016/j.carbpol.2017.05.021.

Gallardo-Toledo, E., Velasco-Aguirre, C., Kogan, M.J., 2021. Inorganic nanoparticles and their strategies to enhance brain drug delivery. In: Morales, J.O., Gaillard, P.J. (Eds.), Nanomedicines for Brain Drug Delivery. Springer US, New York, NY, pp. 149–172. https://doi.org/10.1007/978-1-0716-0838-8_6.

Garcés, M., Cáceres, L., Chiappetta, D., Magnani, N., Evelson, P., 2021. Current understanding of nanoparticle toxicity mechanisms and interactions with biological systems. New J. Chem. 45, 14328–14344. https://doi.org/10.1039/D1NJ01415C.

Garg, Rajni, Rani, P., Garg, Rishav, Khan, M.A., Khan, N.A., Khan, A.H., Américo-Pinheiro, J.H.P., 2022. Biomedical and catalytic applications of agri-based biosynthesized silver nanoparticles. Environ. Pollut. 310, 119830 https://doi.org/10.1016/j.envpol.2022.119830.

Ge, M., Li, J., Song, S., Meng, N., Zhou, N., 2019. Development and antibacterial performance of silver nanoparticles-lecithin modified montmorillonite nanoparticle hybrid. Appl. Clay Sci. 183, 105334 https://doi.org/10.1016/j.clay.2019.105334.

Geoffrion, L.D., Hesabizadeh, T., Medina-Cruz, D., Kusper, M., Taylor, P., Vernet-Crua, A., Chen, J., Ajo, A., Webster, T.J., Guisbiers, G., 2020. Naked selenium nanoparticles for antibacterial and anticancer treatments. ACS Omega 5, 2660–2669. https://doi.org/10.1021/acsomega.9b03172.

Ghanbari, F., Nourmoradi, H., Nazari, A., Sadeghifard, N., Pakzad, I., Soroush, S., Ashrafi, B., Taherikalani, M., 2022. Molecular epidemiology and recycling of Staphylococcus aureus resistant to methicillin among the staff, patients, and surfaces in university Hospital in West Iran, Ilam. Infect. Disord. Drug Target. 23, 42–51. https://doi.org/10.2174/1871526522666220928145550.

Gil, A., Gandía, L.M., Vicente, M.A., 2000. Recent advances in the synthesis and catalytic applications of pillared clays. Catal. Rev. 42, 145–212. https://doi.org/10.1081/CR-100100261.

Gil, A., Santamaría, L., Korili, S.A., Vicente, M.A., Barbosa, L.V., de Souza, S.D., Marçal, L., de Faria, E.H., Ciuffi, K.J., 2021. A review of organic-inorganic hybrid clay based adsorbents for contaminants removal: Synthesis, perspectives and applications. J. Environ. Chem. Eng. 9, 105808 https://doi.org/10.1016/j.jece.2021.105808.

Gill, A.A.S., Singh, S., Nate, Z., Pawar, C., Chauhan, R., Thapliyal, N.B., Karpoormath, R., Patel, R., 2021. One-pot synthesis of β-cyclodextrin modified silver nanoparticles for highly sensitive detection of ciprofloxacin. J. Pharm. Biomed. Anal. 203, 114219 https://doi.org/10.1016/j.jpba.2021.114219.

Giraldo, L.F., Camilo, P., Kyu, T., 2016. Incorporation of silver in montmorillonite-type phyllosilicates as potential antibacterial material. Curr. Opin. Chem. Eng. 11, 7–13. https://doi.org/10.1016/J.COCHE.2015.11.003.

Girase, B., Depan, D., Shah, J.S., Xu, W., Misra, R.D.K., 2011. Silver–clay nanohybrid structure for effective and diffusion-controlled antimicrobial activity. Mater. Sci. Eng. C 31, 1759–1766. https://doi.org/10.1016/j.msec.2011.08.007.

Gomes, T.A.T., Elias, W.P., Scaletsky, I.C.A., Guth, B.E.C., Rodrigues, J.F., Piazza, R.M.F., Ferreira, L.C.S., Martinez, M.B., 2016. Diarrheagenic Escherichia coli. Braz. J. Microbiol. 47, 3–30. https://doi.org/10.1016/j.bjm.2016.10.015.

González-Vega, J.G., García-Ramos, J.C., Chavez-Santoscoy, R.A., Castillo-Quiñones, J. E., Arellano-Garcia, M.E., Toledano-Magaña, Y., 2022. Lung models to evaluate silver nanoparticles' toxicity and their impact on human health. Nanomaterials 12. https://doi.org/10.3390/nano12132316.

Gupta, P., Bajpai, M., Bajpai, S.K., 2008. Investigation of antibacterial properties of silver nanoparticle-ioaded poly (acrylamide-co-itaconic acid)-grafted cotton fabric. J. Cotton Sci. 12, 280–286.

Guzman, M., Dille, J., Godet, S., 2012. Synthesis and antibacterial activity of silver nanoparticles against gram-positive and gram-negative bacteria. Nanomedicine 8, 37–45. https://doi.org/10.1016/j.nano.2011.05.007.

Haase, A., Tentschert, J., Jungnickel, H., Graf, P., Mantion, A., Draude, F., Plendl, J., Goetz, M.E., Galla, S., Mašić, A., Thuenemann, A.F., Taubert, A., Arlinghaus, H.F., Luch, A., 2011. Toxicity of silver nanoparticles in human macrophages: uptake, intracellular distribution and cellular responses. J. Phys. Conf. Ser. 304, 012030 https://doi.org/10.1088/1742-6596/304/1/012030.

Habeeb Rahman, A.P., Misra, A.J., Das, S., Das, B., Jayabalan, R., Suar, M., Mishra, A., Tamhankar, A.J., Lundborg, C.S., Tripathy, S.K., 2018. Mechanistic insight into the disinfection of Salmonella sp. by sun-light assisted sonophotocatalysis using doped ZnO nanoparticles. Chem. Eng. J. 336, 476–488. https://doi.org/10.1016/J.CEJ.2017.12.053.

Hacıosmanoğlu, G.G., Mejías, C., Martín, J., Santos, J.L., Aparicio, I., Alonso, E., 2022. Antibiotic adsorption by natural and modified clay minerals as designer adsorbents for wastewater treatment: A comprehensive review. J. Environ. Manag. 317, 115397 https://doi.org/10.1016/j.jenvman.2022.115397.

Hailan, W.A., Al-Anazi, K.M., Farah, M.A., Ali, M.A., Al-Kawmani, A.A., Abou-Tarboush, F.M., 2022. Reactive oxygen species-mediated cytotoxicity in liver carcinoma cells induced by silver nanoparticles biosynthesized using Schinu molle extract. Nanomaterials 12, 161. https://doi.org/10.3390/nano12010161.

Hakimov, S., Kylychbekov, S., Harness, B., Neupane, S., Hurley, J., Brooks, A., Banga, S., Er, A.O., 2022. Evaluation of silver nanoparticles attached to methylene blue as an antimicrobial agent and its cytotoxicity. Photodiagn. Photodyn. Ther. 39, 102904 https://doi.org/10.1016/j.pdpdt.2022.102904.

Hofer, U., 2019. The cost of antimicrobial resistance. Nat. Rev. Microbiol. 17, 3. https://doi.org/10.1038/s41579-018-0125-x.

Holešová, S., Samlíková, M., Pazdziora, E., Valášková, M., 2013. Antibacterial activity of organomontmorillonites and organovermiculites prepared using chlorhexidine diacetate. Appl. Clay Sci. 83–84, 17–23. https://doi.org/10.1016/j.clay.2013.07.013.

Hong, J., Wu, T., Wu, H., Zeng, B., Zeng, S., Chen, T., Wang, X., Lu, Z., Yuan, C., Balaji, K., Petri, D.F.S., Dai, L., 2021. Nanohybrid silver nanoparticles@halloysite nanotubes coated with polyphosphazene for effectively enhancing the fire safety of epoxy resin. Chem. Eng. J. 407, 127087 https://doi.org/10.1016/j.cej.2020.127087.

Huang, R.-H., Chao, W.-K., Yu, R.-S., Huang, R.-T., Hsueh, K.-L., Shieu, F.-S., 2012. Facile synthesis of silver nanoparticles by electrochemical method in the presence of sodium montmorillonite. J. Electrochem. Soc. 159, E122. https://doi.org/10.1149/2.037206jes.

Huang, C.-H., Lee, C.-H., Chang, C., 2022a. Spontaneous bacterial peritonitis in decompensated liver cirrhosis-A literature review. Livers 2, 214–232. https://doi.org/10.3390/livers2030018.

Huang, Xinrong, Ge, M., Wang, H., Liang, H., Meng, N., Zhou, N., 2022b. Functional modification of polydimethylsiloxane nanocomposite with silver nanoparticles-based montmorillonite for antibacterial applications. Colloids Surf. A Physicochem. Eng. Asp. 642, 128666 https://doi.org/10.1016/j.colsurfa.2022.128666.

Huang, Xueting, Li, X., An, H., Wang, J., Ding, M., Wang, L., Li, L., Ji, Q., Qu, F., Wang, H., Xu, Y., Lu, X., He, Y., Zhang, J.-R., 2022c. Capsule type defines the capability of Klebsiella pneumoniae in evading Kupffer cell capture in the liver. PLoS Pathog. 18, e1010693. https://doi.org/10.1371/journal.ppat.1010693.

Huma, Z., Gupta, A., Javed, I., Das, R., Hussain, S.Z., Mumtaz, S., Hussain, I., Rotello, V. M., 2018. Cationic silver nanoclusters as potent antimicrobials against multidrug-resistant bacteria. ACS Omega 3, 16721–16727. https://doi.org/10.1021/acsomega.8b02438.

Hyesuk, S., Qiangde, D., Ipshita, U., Weiping, Z., 2022. Evaluation of multivalent enterotoxigenic Escherichia coli vaccine candidate MecVax antigen dose-dependent effect in a murine model. Appl. Environ. Microbiol. 88 https://doi.org/10.1128/aem.00959-22 e00959-22.

Ibarra-Alonso, M.C., Sánchez-Valdes, S., Ramírez-Vargas, E., Fernandez-Tavizón, S., Romero-Garcia, J., Ledezma-Perez, A.S., Ramos de Valle, L.F., Rodriguez-Fernandez, O.S., Espinoza-Martinez, A.B., Martinez-Colunga, J.G., Cabrera-Álvarez, E.N., 2015. Preparation and characterization of Polyethylene/Clay/Silver nanocomposites using functionalized polyethylenes as an adhesion promoter. J. Adhes. Sci. Technol. 29, 1911–1923. https://doi.org/10.1080/01694243.2015.1045247.

Ilić, K., Hartl, S., Galić, E., Tetyczka, C., Pem, B., Barbir, R., Milić, M., Vinković Vrček, I., Roblegg, E., Pavičić, I., 2021. Interaction of differently coated silver nanoparticles with skin and oral mucosal cells. J. Pharm. Sci. 110, 2250–2261. https://doi.org/10.1016/j.xphs.2021.01.030.

Ipshita, U., Lauder, K., Siqi, L., Galen, P., Weiping, Z., 2022. Intramuscularly administered enterotoxigenic Escherichia coli (ETEC) vaccine candidate MecVax prevented H10407 intestinal colonization in an adult rabbit colonization model. Microbiol. Spectr. 10 https://doi.org/10.1128/spectrum.01473-22 e01473-22.

Iram, F., Yasmeen, A., Massey, S., Iqbal, M.S., Asim, S., Irshad, M., Zahid, H., Khan, A.Y., Kazimi, S.G.T., 2021. Synthesis of gold and silver nanoparticles by use of arabinoglucan from Lallemantia royleana. Int. J. Biol. Macromol. 191, 1137–1150. https://doi.org/10.1016/j.ijbiomac.2021.09.096.

Ivask, A., Suarez, E., Patel, T., Boren, D., Ji, Z., Holden, P., Telesca, D., Damoiseaux, R., Bradley, K.A., Godwin, H., 2012. Genome-wide bacterial toxicity screening uncovers the mechanisms of toxicity of a cationic polystyrene nanomaterial. Environ. Sci. Technol. 46, 2398–2405. https://doi.org/10.1021/es203087m.

Ivask, A., ElBadawy, A., Kaweeteerawat, C., Boren, D., Fischer, H., Ji, Z., Chang, C.H., Liu, R., Tolaymat, T., Telesca, D., Zink, J.I., Cohen, Y., Holden, P.A., Godwin, H.A., 2014. Toxicity mechanisms in Escherichia coli vary for silver nanoparticles and differ from ionic silver. ACS Nano 8, 374–386. https://doi.org/10.1021/nn4044047.

Jabeen, S., Qureshi, R., Munazir, M., Maqsood, M., Munir, M., Shah, S.S.H., Rahim, B.Z., 2021. Application of green synthesized silver nanoparticles in cancer treatment—a critical review. Mater. Res. Express 8, 092001. https://doi.org/10.1088/2053-1591/ac1de3.

Jaber, M., Georgelin, T., Bazzi, H., Costa-Torro, F., Lambert, J.-F., Bolbach, G., Clodic, G., 2014. Selectivities in adsorption and peptidic condensation in the (arginine and glutamic acid)/montmorillonite clay system. J. Phys. Chem. C 118, 25447–25455. https://doi.org/10.1021/jp507335e.

Jahnke, J.P., Cornejo, J.A., Sumner, J.J., Schuler, A.J., Atanassov, P., Ista, L.K., 2016. Conjugated gold nanoparticles as a tool for probing the bacterial cell envelope: The case of Shewanella oneidensis MR-1. Biointerphases 11, 011003. https://doi.org/10.1116/1.4939244.

Ji, X., Song, X., Li, J., Bai, Y., Yang, W., Peng, X., 2007. Size control of gold nanocrystals in citrate reduction: the third role of citrate. J. Am. Chem. Soc. 129, 13939–13948. https://doi.org/10.1021/ja074447k.

Jiang, H., Manolache, S., Wong, A.C.L., Denes, F.S., 2004. Plasma-enhanced deposition of silver nanoparticles onto polymer and metal surfaces for the generation of

antimicrobial characteristics. J. Appl. Polym. Sci. 93, 1411–1422. https://doi.org/10.1002/app.20561.
Jiang, J., Ayaz, T., Jiang, Z., Lei, M., 2021. Green remediation of heavy metal polluted water and soil using clay minerals: A review. IOP Conf. Ser. Earth Environ. Sci. 632, 052079 https://doi.org/10.1088/1755-1315/632/5/052079.
Jiao, Z.-H., Li, M., Feng, Y.-X., Shi, J.-C., Zhang, J., Shao, B., 2014. Hormesis effects of silver nanoparticles at non-cytotoxic doses to human hepatoma cells. PLoS One 9, e102564. https://doi.org/10.1371/journal.pone.0102564.
Jiraroj, D., Tungasmita, S., Tungasmita, D.N., 2014. Silver ions and silver nanoparticles in zeolite A composites for antibacterial activity. Powder Technol. 264, 418–422. https://doi.org/10.1016/j.powtec.2014.05.049.
Joe, Y.H., Park, D.H., Hwang, J., 2016. Evaluation of Ag nanoparticle coated air filter against aerosolized virus: Anti-viral efficiency with dust loading. J. Hazard. Mater. 301, 547–553. https://doi.org/10.1016/j.jhazmat.2015.09.017.
Jones Jr., R.M., Seo, H., Zhang, W., Sack, D.A., 2022. A multi-epitope fusion antigen candidate vaccine for Enterotoxigenic Escherichia coli is protective against strain B7A colonization in a rabbit model. PLoS Negl. Trop. Dis. 16, e0010177.
Jou, S.K., Malek, N.A.N.N., 2016. Characterization and antibacterial activity of chlorhexidine loaded silver-kaolinite. Appl. Clay Sci. 127–128, 1–9. https://doi.org/10.1016/j.clay.2016.04.001.
Juan, C., Perez de la Lastra, J., Plou, F., Perez, E., Reinbothe, S., 2021. The Chemistry of Reactive Oxygen Species (ROS) Revisited: Outlining Their Role in Biological Macromolecules (DNA, Lipids and Proteins) and Induced Pathologies. Int. J. Mol. Sci. 22, 4642. https://doi.org/10.3390/ijms22094642.
Kacprzyk, L., Rydengård, V., Mörgelin, M., Davoudi, M., Pasupuleti, M., Malmsten, M., Schmidtchen, A., 2007. Antimicrobial activity of histidine-rich peptides is dependent on acidic conditions. Biochim. Biophys. Acta 1768, 2667–2680. https://doi.org/10.1016/j.bbamem.2007.06.020.
Kaegi, R., Sinnet, B., Zuleeg, S., Hagendorfer, H., Mueller, E., Vonbank, R., Boller, M., Burkhardt, M., 2010. Release of silver nanoparticles from outdoor facades. Environ. Pollut. 158, 2900–2905. https://doi.org/10.1016/j.envpol.2010.06.009.
Kahangi, F.G., Mehrdad, M., Heravi, M.M., Sadjadi, S., 2020. Bio-assisted synthesized Ag (0) nanoparticles stabilized on hybrid of sepiolite and chitin: efficient catalytic system for xanthene synthesis. Sci. Rep. 10, 15285. https://doi.org/10.1038/s41598-020-71866-2.
Kamyar, S., Ahmad, M., Yunus, W., Ibrahim, N., Gharayebi, Y., Sedaghat, S., 2010. Synthesis of silver/montmorillonite nanocomposite using γ-irradiation. Int. J. Nanomedicine 5, 1067–1077. https://doi.org/10.2147/IJN.S15033.
Kaplan, Ö., Gökşen Tosun, N., İmamoğlu, R., Türkekul, İ., Gökçe, İ., Özgür, A., 2022. Biosynthesis and characterization of silver nanoparticles from Tricholoma ustale and Agaricus arvensis extracts and investigation of their antimicrobial, cytotoxic, and apoptotic potentials. J. Drug. Deliv. Sci. Technol. 69, 103178 https://doi.org/10.1016/j.jddst.2022.103178.
Karatop, E.U., Cimenci, C.E., Aksu, A.M., 2022. Colorimetric cytotoxicity assays. In: Anil, S., Mansour, M.A. (Eds.), Cytotoxicity. IntechOpen, Rijeka. https://doi.org/10.5772/intechopen.105772. Ch. 1.
Kesavan Pillai, S., Sinha Ray, S., Scriba, M., Bandyopadhyay, J., Roux-van der Merwe, M. P., Badenhorst, J., 2013. Microwave assisted green synthesis and characterization of silver/montmorillonite heterostructures with improved antimicrobial properties. Appl. Clay Sci. 83–84, 315–321. https://doi.org/10.1016/j.clay.2013.08.014.
Khatoon, U.T., Velidandi, A., Nageswara Rao, G.V.S., 2023. Sodium borohydride mediated synthesis of nano-sized silver particles: Their characterization, antimicrobial and cytotoxicity studies. Mater. Chem. Phys. 294, 126997 https://doi.org/10.1016/j.matchemphys.2022.126997.
Kheiralla, Z.M.H., Rushdy, A.A., Betiha, M.A., Yakob, N.A.N., 2014. High-performance antibacterial of montmorillonite decorated with silver nanoparticles using microwave-assisted method. J. Nanopart. Res. 16, 2560. https://doi.org/10.1007/s11051-014-2560-6.
Khorasani, S., Ghandehari Yazdi, A.P., Saadatfar, A., Kamali Rousta, L., Nejatian, M., Abarian, M., Jafari, S.M., 2022. Valorization of saffron tepals for the green synthesis of silver nanoparticles and evaluation of their efficiency against foodborne pathogens. Waste and Biomass Valor. 13, 4417–4430. https://doi.org/10.1007/s12649-022-01791-0.
Khundkar, R., Malic, C., Burge, T., 2010. Use of Acticoat™ dressings in burns: What is the evidence? Burns 36, 751–758. https://doi.org/10.1016/j.burns.2009.04.008.
Kim, J.S., 2007. Antibacterial activity of Ag+ ion-containing silver nanoparticles prepared using the alcohol reduction method. J. Ind. Eng. Chem. 13, 718–722.
Kim, J.S., Kuk, E., Yu, K.N., Kim, J.-H., Park, S.J., Lee, H.J., Kim, S.H., Park, Y.K., Park, Y. H., Hwang, C.-Y., Kim, Y.-K., Lee, Y.-S., Jeong, D.H., Cho, M.-H., 2007. Antimicrobial effects of silver nanoparticles. Nanomed. 3, 95–101. https://doi.org/10.1016/j.nano.2006.12.001.
Krūmiņš, J., Kļaviņš, M., Ozola-davidāne, R., Ansone-bērtiņa, L., 2022. The Prospects of Clay Minerals from the Baltic States for Industrial-Scale Carbon Capture: A Review. Minerals. https://doi.org/10.3390/min12030349.
Kumar, A., Vemula, P.K., Ajayan, P.M., John, G., 2008. Silver-nanoparticle-embedded antimicrobial paints based on vegetable oil. Nat. Mater. 7, 236–241. https://doi.org/10.1038/nmat2099.
Kumar, A., Pandey, A.K., Singh, S.S., Shanker, R., Dhawan, A., 2011. Engineered ZnO and TiO2 nanoparticles induce oxidative stress and DNA damage leading to reduced viability of Escherichia coli. Free Radic. Biol. Med. 51, 1872–1881. https://doi.org/10.1016/j.freeradbiomed.2011.08.025.
Kuorwel, K.K., Cran, M.J., Orbell, J.D., Buddhadasa, S., Bigger, S.W., 2015. Review of mechanical properties, migration, and potential applications in active food packaging systems containing nanoclays and nanosilver. Compr. Rev. Food Sci. Food Saf. 14, 411–430. https://doi.org/10.1111/1541-4337.12139.
Kyrychenko, A., Pasko, D.A., Kalugin, O.N., 2017. Poly(vinyl alcohol) as a water protecting agent for silver nanoparticles: the role of polymer size and structure. Phys. Chem. Chem. Phys. 19, 8742–8756. https://doi.org/10.1039/C6CP05562A.
Lankoff, A., Sandberg, W.J., Wegierek-Ciuk, A., Lisowska, H., Refsnes, M., Sartowska, B., Schwarze, P.E., Meczynska-Wielgosz, S., Wojewodzka, M., Kruszewski, M., 2012. The effect of agglomeration state of silver and titanium dioxide nanoparticles on cellular response of HepG2, A549 and THP-1 cells. Toxicol. Lett. 208, 197–213. https://doi.org/10.1016/j.toxlet.2011.11.006.
Lavorgna, M., Attianese, I., Buonocore, G.G., Conte, A., Del Nobile, M.A., Tescione, F., Amendola, E., 2014. MMT-supported Ag nanoparticles for chitosan nanocomposites: Structural properties and antibacterial activity. Carbohydr. Polym. 102, 385–392. https://doi.org/10.1016/j.carbpol.2013.11.026.
Lee, Y.J., Park, Y., 2020. Green Synthetic Nanoarchitectonics of Gold and Silver Nanoparticles Prepared Using Quercetin and Their Cytotoxicity and Catalytic Applications. J. Nanosci. Nanotechnol. 20, 2781–2790. https://doi.org/10.1166/jnn.2020.17453.
Leong, Q.L., Cheng, H.-Y., Huang, C.-C., Tsai, J.-J., Lee, C.-W., Chen, J.-C., Lee, C., Huang, C.-L., 2022. Synthesis of silver nanoparticles with long-term storability for SERS applications using aqueous extracts of rice bran: A rapid and green photochemical approach. J. Mol. Struct. 1254, 132338 https://doi.org/10.1016/j.molstruc.2022.132338.
Leung, Y.H., Ng, A.M.C., Xu, X., Shen, Z., Gethings, L.A., Wong, M.T., Chan, C.M.N., Guo, M.Y., Ng, Y.H., Djurišić, A.B., Lee, P.K.H., Chan, W.K., Yu, L.H., Phillips, D.L., Ma, A.P.Y., Leung, F.C.C., 2014. Mechanisms of antibacterial activity of MgO: Non-ROS mediated toxicity of MgO nanoparticles towards Escherichia coli. Small 10, 1171–1183. https://doi.org/10.1002/smll.201302434.
Li, P., Nijhawan, D., Budihardjo, I., Srinivasula, S.M., Ahmad, M., Alnemri, E.S., Wang, X., 1997. Cytochrome c and dATP-dependent formation of Apaf-1/caspase-9 complex initiates an apoptotic protease cascade. Cell 91, 479–489. https://doi.org/10.1016/S0092-8674(00)80434-1.
Li, W.-R., Xie, X.-B., Shi, Q.-S., Zeng, H.-Y., OU-Yang, Y.-S., Chen, Y.-B., 2010. Antibacterial activity and mechanism of silver nanoparticles on Escherichia coli. Appl. Microbiol. Biotechnol. 85, 1115–1122. https://doi.org/10.1007/s00253-009-2159-5.
Li, L., Bi, Z., Hu, Y., Sun, L., Song, Y., Chen, S., Mo, F., Yang, J., Wei, Y., Wei, X., 2021. Silver nanoparticles and silver ions cause inflammatory response through induction of cell necrosis and the release of mitochondria in vivo and in vitro. Cell Biol. Toxicol. 37, 177–191. https://doi.org/10.1007/s10565-020-09526-4.
Li, D., Liu, P., Hao, F., Lv, Y., Xiong, W., Yan, C., Wu, Y., Luo, H., 2022. Preparation and application of silver/chitosan-sepiolite materials with antimicrobial activities and low cytotoxicity. Int. J. Biol. Macromol. 210, 337–349. https://doi.org/10.1016/j.ijbiomac.2022.05.015.
Liguori, K., Keenum, I., Davis, B.C., Calarco, J., Milligan, E., Harwood, V.J., Pruden, A., 2022. Antimicrobial resistance monitoring of water environments: A framework for standardized methods and quality control. Environ. Sci. Technol. 56, 9149–9160. https://doi.org/10.1021/acs.est.1c08918.
Liu, J., Li, X., Zuo, S., Yu, Y., 2007. Preparation and photocatalytic activity of silver and TiO2 nanoparticles/montmorillonite composites. Appl. Clay Sci. 37, 275–280. https://doi.org/10.1016/j.clay.2007.01.008.
Liu, Q., Zhang, Y., Huang, J., Xu, Z., Li, X., Yang, J., Huang, H., Tang, S., Chai, Y., Lin, J., Yang, C., Liu, J., Lin, S., 2022. Mesoporous silica-coated silver nanoparticles as ciprofloxacin/siRNA carriers for accelerated infected wound healing. J Nanobiotechnol. 20, 386. https://doi.org/10.1186/s12951-022-01600-9.
Long, Y., Hu, S., Lei, P., Li, Y., 2022. Preparation of green silver nanoparticles with high antibacterial ability using N-maleoyl chitosan and montmorillonite. Mater. Lett. 316, 132044 https://doi.org/10.1016/j.matlet.2022.132044.
Lorenzo-Anota, H.Y., Zarate-Triviño, D.G., Uribe-Echeverría, J.A., Ávila-Ávila, A., Rangel-López, J.R., Martínez-Torres, A.C., Rodríguez-Padilla, C., 2021. Chitosan-coated gold nanoparticles induce low cytotoxicity and low ROS production in primary leucocytes, independent of their proliferative status. Pharmaceutics 13, 942. https://doi.org/10.3390/pharmaceutics13070942.
Lou, Z., Wang, H., Zhu, S., Ma, C., Wang, Z., 2011. Antibacterial activity and mechanism of action of chlorogenic acid. J. Food Sci. 76, M398–M403. https://doi.org/10.1111/j.1750-3841.2011.02213.x.
Lou, Z., Wang, H., Rao, S., Sun, J., Ma, C., Li, J., 2012. p-Coumaric acid kills bacteria through dual damage mechanisms. Food Control 25, 550–554. https://doi.org/10.1016/j.foodcont.2011.11.022.
Lowy, F.D., 1998. Staphylococcus aureus infections. N. Engl. J. Med. 339, 520–532. https://doi.org/10.1056/NEJM199808203390806.
Lu, Q., Wang, H., Liu, Y., Hou, Y., Li, H., Zhang, Y., 2017. Graphitic carbon nitride nanodots: As reductant for the synthesis of silver nanoparticles and its biothiols biosensing application. Biosens. Bioelectron. 89, 411–416. https://doi.org/10.1016/j.bios.2016.05.064.
Lyu, Z., Shang, Y., Xia, Y., 2022. Shape-controlled synthesis of copper nanocrystals for plasmonic, biomedical, and electrocatalytic applications. Acc. Mater. Res. https://doi.org/10.1021/accountsmr.2c00134.
Madl, A.K., Plummer, L.E., Carosino, C., Pinkerton, K.E., 2014. Nanoparticles, lung injury, and the role of oxidant stress. Annu. Rev. Physiol. 76, 447–465. https://doi.org/10.1146/annurev-physiol-030212-183735.
Mahdi, R.A., Bahrami, Y., Kakaei, E., 2022. Identification and antibacterial evaluation of endophytic actinobacteria from Luffa cylindrica. Sci. Rep. 12, 18236. https://doi.org/10.1038/s41598-022-23073-4.
Majeed, S., Abdullah, M.S., Bin Dash, G.K., Ansari, M.T., Nanda, A., 2016. Biochemical synthesis of silver nanoprticles using filamentous fungi Penicillium decumbens (MTCC-2494) and its efficacy against A-549 lung cancer cell line. Chin. J. Nat. Med. 14, 615–620. https://doi.org/10.1016/S1875-5364(16)30072-3.

Makwana, D., Castaño, J., Somani, R.S., Bajaj, H.C., 2020. Characterization of Agar-CMC/Ag-MMT nanocomposite and evaluation of antibacterial and mechanical properties for packaging applications. Arab. J. Chem. 13, 3092–3099. https://doi.org/10.1016/j.arabjc.2018.08.017.

Malachová, K., Praus, P., Rybková, Z., Kozák, O., 2011. Antibacterial and antifungal activities of silver, copper and zinc montmorillonites. Appl. Clay Sci. 53, 642–645. https://doi.org/10.1016/j.clay.2011.05.016.

Malysheva, A., Ivask, A., Doolette, C.L., Voelcker, N.H., Lombi, E., 2021. Cellular binding, uptake and biotransformation of silver nanoparticles in human T lymphocytes. Nat. Nanotechnol. 16, 926–932. https://doi.org/10.1038/s41565-021-00914-3.

Mamun, M.M., Sorinolu, A.J., Munir, M., Vejerano, E.P., 2021. Nanoantibiotics: Functions and properties at the nanoscale to combat antibiotic resistance. Front. Chem. 9, 687660 https://doi.org/10.3389/fchem.2021.687660.

Marchesi, S., Carniato, F., Guidotti, M., Botta, M., Marchese, L., Bisio, C., 2020. Synthetic saponite clays as promising solids for lanthanide ion recovery. New J. Chem. 44, 10033–10041. https://doi.org/10.1039/C9NJ05983K.

Martín, C.F., Sweatman, M.B., Brandani, S., Fan, X., 2016. Wet impregnation of a commercial low cost silica using DETA for a fast post-combustion CO2 capture process. Appl. Energy 183, 1705–1721. https://doi.org/10.1016/j.apenergy.2016.09.081.

Mason, J., Gasnier, C., Kichler, A., Prévost, G., Aunis, D., Metz-Boutigue, M.-H., Bechinger, B., 2006. Enhanced membrane disruption and antibiotic action against pathogenic bacteria by designed histidine-rich peptides at acidic pH. Antimicrob. Agents Chemother. 50, 3305–3311. https://doi.org/10.1128/AAC.00490-06.

Mehata, M.S., 2021. Green route synthesis of silver nanoparticles using plants/ginger extracts with enhanced surface plasmon resonance and degradation of textile dye. Mater. Sci. Eng. B 273, 115418. https://doi.org/10.1016/j.mseb.2021.115418.

Meier, L.P., Kahr, G., 1999. Determination of the Cation Exchange Capacity (CEC) of clay minerals using the complexes of copper(II) ion with triethylenetetramine and tetraethylenepentamine. Clay Clay Miner. 47, 386–388. https://doi.org/10.1346/CCMN.1999.0470315.

Micoli, F., Bagnoli, F., Rappuoli, R., Serruto, D., 2021. The role of vaccines in combatting antimicrobial resistance. Nat. Rev. Microbiol. 19, 287–302. https://doi.org/10.1038/s41579-020-00506-3.

Mirzajani, F., Ghassempour, A., Aliahmadi, A., Esmaeili, M.A., 2011. Antibacterial effect of silver nanoparticles on Staphylococcus aureus. Res. Microbiol. 162, 542–549. https://doi.org/10.1016/j.resmic.2011.04.009.

Miyoshi, H., Ohno, H., Sakai, K., Okamura, N., Kourai, H., 2010. Characterization and photochemical and antibacterial properties of highly stable silver nanoparticles prepared on montmorillonite clay in n-hexanol. J. Colloid Interface Sci. 345, 433–441. https://doi.org/10.1016/j.jcis.2010.01.034.

Mohamed, E.M., Kattaia, A.A.A., Abdul-Maksoud, R.S., Abd El-Baset, S.A., 2021. Cellular, molecular and biochemical impacts of silver nanoparticles on rat cerebellar cortex. Cells 10, 7. https://doi.org/10.3390/cells10010007.

Mohd Faheem, M., Bhagat, M., Sharma, P., Anand, R., 2022. Induction of p53 mediated mitochondrial apoptosis and cell cycle arrest in human breast cancer cells by plant mediated synthesis of silver nanoparticles from Bergenia ligulata (Whole plant). Int. J. Pharm. 619, 121710 https://doi.org/10.1016/j.ijpharm.2022.121710.

Moosa, Salmah, 2019. Synthesis of Silver Nanoparticles in Kaolinite and Their Antibacterial Behaviour. Int. J. Eng. Res. Technol. 8, 65. https://doi.org/10.17577/IJERTV8IS080065.

Morariu, S., Teodorescu, M., Bercea, M., 2022. Rheological investigation of polymer/clay dispersions as potential drilling fluids. J. Pet. Sci. Eng. 210, 110015 https://doi.org/10.1016/j.petrol.2021.110015.

Morones, J.R., Elechiguerra, J.L., Camacho, A., Holt, K., Kouri, J.B., Ramírez, J.T., Yacaman, M.J., 2005. The bactericidal effect of silver nanoparticles. Nanotechnology 16, 2346. https://doi.org/10.1088/0957-4484/16/10/059.

Mulfinger, L., Solomon, S.D., Bahadory, M., Jeyarajasingam, A.V., Rutkowsky, S.A., Boritz, C., 2007. Synthesis and study of silver nanoparticles. J. Chem. Educ. 84, 322. https://doi.org/10.1021/ed084p322.

Muñoz, L.N., Jaramillo, V., Gantiva-Diaz, M., Cifuentes, J., Muñoz-Camargo, C., Cruz, J. C., González Barrios, A.F., 2022. Formulation of a novel antibacterial topical treatment based on Magnetite-Buforin-II-silver nanobioconjugates. Front. Bioeng. Biotechnol. 10, 1003004. https://doi.org/10.3389/fbioe.2022.1003004.

Murray, R.G.E., Steed, P., Elson, H.E., 1965. The location of the mucopeptide in sections of the cell wall of Escherichia coli and other Gram-negative bacteria. Can. J. Microbiol. 11, 547–560. https://doi.org/10.1139/m65-072.

Murugan, K., Choonara, Y.E., Kumar, P., Bijukumar, D., du Toit, L.C., Pillay, V., 2015. Parameters and characteristics governing cellular internalization and trans-barrier trafficking of nanostructures. Int. J. Nanomedicine 10, 2191–2206. https://doi.org/10.2147/IJN.S75615.

Mytych, J., Zebrowski, J., Lewinska, A., Wnuk, M., 2017. Prolonged effects of silver nanoparticles on p53/p21 pathway-mediated proliferation, DNA damage response, and methylation parameters in HT22 hippocampal neuronal cells. Mol. Neurobiol. 54, 1285–1300. https://doi.org/10.1007/s12035-016-9688-6.

Naganthran, A., Verasoundarapandian, G., Khalid, F.E., Masarudin, M.J., Zulkharnain, A., Nawawi, N.M., Karim, M., Che Abdullah, C.A., Ahmad, S.A., 2022. Synthesis, characterization and biomedical application of silver nanoparticles. Materials 15, 427. https://doi.org/10.3390/ma15020427.

Nakamura, T., Magara, H., Herbani, Y., Sato, S., 2011. Fabrication of silver nanoparticles by highly intense laser irradiation of aqueous solution. Appl. Phys. A 104, 1021–1024. https://doi.org/10.1007/s00339-011-6499-5.

Nataro, J., Kaper, J., 1998. Diarrheagenic Escherichia coli. Clin. Microbiol. Rev. 11, 142–201. https://doi.org/10.1128/CMR.11.1.142.

Nguyen, D.D., Lue, S.J., Lai, J.-Y., 2021. Tailoring therapeutic properties of silver nanoparticles for effective bacterial keratitis treatment. Colloids Surf. B: Biointerfaces 205, 111856. https://doi.org/10.1016/j.colsurfb.2021.111856.

Niraj, G., Thapa, G.T., Khushi, P., Paul, S., 2022. The influence of patterned surface features on the accumulation of bovine synovial fluid-Induced aggregates of Staphylococcus aureus. Appl. Environ. Microbiol. 0, e01217–e01222. https://doi.org/10.1128/aem.01217-22.

Nithya Deva Krupa, A., Raghavan, V., 2014. Biosynthesis of silver nanoparticles using Aegle marmelos (Bael) fruit extract and its application to prevent adhesion of bacteria: A strategy to control microfouling. Bioinorg. Chem. Appl. 2014, 949538 https://doi.org/10.1155/2014/949538.

Nouri, A., Yaraki, M.T., Ghorbanpour, M., Agarwal, S., Gupta, V.K., 2018. Enhanced antibacterial effect of chitosan film using montmorillonite/CuO nanocomposite. Int. J. Biol. Macromol. 109, 1219–1231. https://doi.org/10.1016/j.ijbiomac.2017.11.119.

Nowack, B., Krug, H.F., Height, M., 2011. 120 years of nanosilver history: Implications for policy makers. Environ. Sci. Technol. 45, 1177–1183. https://doi.org/10.1021/es103316q.

Nunes Pessanha, N.F., Fausta Kawase, K.Y., Luiz Vieira Coelho, G., 2014. Preparation and characterization of silver/organo-clay nanocomposites. Chem. Mater. Eng. 2, 173–178. https://doi.org/10.13189/cme.2014.020802.

O'Neill, J., 2014. Antimicrobial Resistance: Tackling a Crisis for the Health and Wealth of Nations, Review on Antimicrobial Resistance. Wellcome Trust & HM Government London.

Ondusko, D.S., Nolt, D., 2018. Staphylococcus aureus. Pediatr. Rev. 39, 287–298. https://doi.org/10.1542/pir.2017-0224.

Onwuka, K., Achilike, K., Eze, K., Enenwa, N., 2021. Montmorillonite clay enhanced TiO2 nanoparticle for photocatalytic degradation of organic pollutants: Mini review. Int. J. Pharma. Sci. 1, 1–2021. https://doi.org/10.51626/ijps.2021.01.00004.

Opoku-Temeng, C., Kobayashi, S.D., DeLeo, F.R., 2019. Klebsiella pneumoniae capsule polysaccharide as a target for therapeutics and vaccines. Comput. Struct. Biotechnol. J. 17, 1360–1366. https://doi.org/10.1016/j.csbj.2019.09.011.

Opris, R.V., Toma, V., Baciu, A.M., Moldovan, R., Dume, B., Berghian-Sevastre, A., Moldovan, B., Clichici, S., David, L., Filip, G.A., Florea, A., 2022. Neurobehavioral and ultrastructural changes induced by phytosynthesized silver-nanoparticle toxicity in an in vivo rat model. Nanomaterials 12, 58. https://doi.org/10.3390/nano12010058.

Otto, M., 2008. Staphylococcal biofilms. In: Romeo, T. (Ed.), Bacterial Biofilms. Springer, Berlin Heidelberg, Berlin, Heidelberg, pp. 207–228. https://doi.org/10.1007/978-3-540-75418-3_10.

Padmavathy, N., Vijayaraghavan, R., 2008. Enhanced bioactivity of ZnO nanoparticles—an antimicrobial study. Sci. Technol. Adv. Mater. 9, 035004 https://doi.org/10.1088/1468-6996/9/3/035004.

Pan, X., Wang, Y., Chen, Z., Pan, D., Cheng, Y., Liu, Z., Lin, Z., Guan, X., 2013. Investigation of antibacterial activity and related mechanism of a series of nano-Mg (OH)2. ACS Appl. Mater. Interfaces 5, 1137–1142. https://doi.org/10.1021/am302910q.

Papp, S., Patakfalvi, R., Dékány, I., 2008. Metal nanoparticle formation on layer silicate lamellae. Colloid Polym. Sci. 286, 3–14. https://doi.org/10.1007/s00396-007-1728-3.

Pedrino, M., Brassolatti, P., Maragno Fattori, A.C., Bianchi, J., de Almeida Rodolpho, J. M., de Godoy, K.F., Assis, M., Longo, E., Zambone, Nogueira, Pinto Rossi, K., Speglich, C., de Freitas Anibal, F., 2022. Analysis of cytotoxicity and genotoxicity in a short-term dependent manner induced by a new titanium dioxide nanoparticle in murine fibroblast cells. Toxicol. Mech. Methods 32, 213–223. https://doi.org/10.1080/15376516.2021.1994075.

Pelkonen, K.H.O., Heinonen-Tanski, H., Hänninen, O.O.P., 2003. Accumulation of silver from drinking water into cerebellum and musculus soleus in mice. Toxicology 186, 151–157. https://doi.org/10.1016/S0300-483X(02)00743-6.

Persano, F., Gigli, G., Leporatti, S., 2021. Halloysite-based nanosystems for biomedical applications. Clay Clay Miner. 69, 501–521. https://doi.org/10.1007/s42860-021-00135-8.

Phukan, A., Bhattacharjee, R.P., Dutta, D.K., 2017. Stabilization of SnO2 nanoparticles into the nanopores of modified Montmorillonite and their antibacterial activity. Adv. Powder Technol. 28, 139–145. https://doi.org/10.1016/j.apt.2016.09.005.

Piñero, S., Camero, S., Blanco, S., 2017. Silver nanoparticles: Influence of the temperature synthesis on the particles'morphology. In: Journal of Physics: Conference Series. Institute of Physics Publishing. https://doi.org/10.1088/1742-6596/786/1/012020.

Pinto, R.M., Seabra, C.L., De Jonge, M., Martins, M.C.L., Van Dijck, P., Reis, S., Nunes, C., 2022. Antibiofilm combinatory strategy: Moxifloxacin-loaded nanosystems and encapsulated N-acetyl-L-cysteine. Pharmaceutics 14, 2294. https://doi.org/10.3390/pharmaceutics14112294.

Pisárčik, M., Lukáč, M., Jampílek, J., Bilka, F., Bilková, A., Pašková, Ľ., Devínsky, F., Horáková, R., Březina, M., Opravil, T., 2021. Silver nanoparticles stabilized with phosphorus-containing heterocyclic surfactants: Synthesis, physico-chemical properties, and biological activity determination. Nanomaterials 11, 1883. https://doi.org/10.3390/nano11081883.

Plackal Adimuriyil George, B., Kumar, N., Abrahamse, H., Ray, S.S., 2018. Apoptotic efficacy of multifaceted biosynthesized silver nanoparticles on human adenocarcinoma cells. Sci. Rep. 8, 14368. https://doi.org/10.1038/s41598-018-32480-5.

Platania, V., Kaldeli-Kerou, A., Karamanidou, T., Kouki, M., Tsouknidas, A., Chatzinikolaidou, M., 2022. Antibacterial effect of colloidal suspensions varying in silver nanoparticles and ions concentrations. Nanomaterials 12. https://doi.org/10.3390/nano12010031.

Praus, P., Turicová, M., Machovič, V., Študentová, S., Klementová, M., 2010. Characterization of silver nanoparticles deposited on montmorillonite. Appl. Clay Sci. 49, 341–345. https://doi.org/10.1016/j.clay.2010.06.009.

Praus, P., Turicová, M., Karlíková, M., Kvítek, L., Dvorský, R., 2013. Nanocomposite of montmorillonite and silver nanoparticles: Characterization and application in catalytic reduction of 4-nitrophenol. Mater. Chem. Phys. 140, 493–498. https://doi.org/10.1016/j.matchemphys.2013.03.059.

Qin, Y., Ji, X., Jing, J., Liu, H., Wu, H., Yang, W., 2010. Size control over spherical silver nanoparticles by ascorbic acid reduction. Colloids Surf. A Physicochem. Eng. Asp. 372, 172–176. https://doi.org/10.1016/J.COLSURFA.2010.10.013.

Raffi, M., Hussain, F., Bhatti, T., Akhter, J., Hameed, A., Hassan, M., 2007. Antibacterial characterization of silver nanoparticles against E. Coli ATCC-15224. J. Mater. Sci. Technol. 24, 192–196.

Rahman, M.F., Wang, J., Patterson, T.A., Saini, U.T., Robinson, B.L., Newport, G.D., Murdock, R.C., Schlager, J.J., Hussain, S.M., Ali, S.F., 2009. Expression of genes related to oxidative stress in the mouse brain after exposure to silver-25 nanoparticles. Toxicol. Lett. 187, 15–21. https://doi.org/10.1016/j.toxlet.2009.01.020.

Ramalingam, B., Parandhaman, T., Das, S.K., 2016. Antibacterial effects of biosynthesized silver nanoparticles on surface ultrastructure and nanomechanical properties of Gram-negative bacteria viz. Escherichia coli and Pseudomonas aeruginosa. ACS Appl. Mater. Interfaces 8, 4963–4976. https://doi.org/10.1021/acsami.6b00161.

Riss, T.L., Moravec, R.A., Niles, A.L., Duellman, S., Benink, H.A., Worzella, T.J., Minor, L., 2004. Cell Viability Assays, in: Assay Guidance Manual. Eli Lilly & Company and the National Center for Advancing Translational Sciences, Bethesda (MD).

Rosa, D.S., Oliveira, S.A.S., Souza, R.F.S., Neto, A.A.S., Cunha, B., Fernandes, L.S., Soares, A.W.C., Teixeira, I.F.S., Aquino, S.K.R., Gouveia, S.R., Almeida, G.V., Naue, J.R.G.S., C.R., Costa, M.M, 2022. Characterization of multidrug-resistant Staphylococcus aureus isolates and comparison of methods of susceptibility to vancomycin. Curr. Microbiol. 79, 374. https://doi.org/10.1007/s00284-022-03070-6.

Roy, A., Butola, B.S., Joshi, M., 2017. Synthesis, characterization and antibacterial properties of novel nano-silver loaded acid activated montmorillonite. Appl. Clay Sci. 146, 278–285. https://doi.org/10.1016/J.CLAY.2017.05.043.

Sadalage, P.S., Dar, M.A., Bhor, R.D., Bhalerao, B.M., Kamble, P.N., Paiva-Santos, A.C., Nimbalkar, M.S., Sonawane, K.D., Pai, K., Patil, P.S., Pawar, K.D., 2022. Optimization of biogenic synthesis of biocompatible platinum nanoparticles with catalytic, enzyme mimetic and antioxidant activities. Food Biosci. 50, 102024 https://doi.org/10.1016/j.fbio.2022.102024.

Sadeghianmaryan, A., Zarbaf, D., Montazer, M., MahmoudiRad, M., 2021. Green synthesis of organo-montmorillonite/silver nanocomposites on dyed cotton with vat dyes to achieve biocompatible antibacterial properties on fashionable clothing. J. Nat. Fibers 1–16. https://doi.org/10.1080/15440478.2021.1993494.

Saha, K., Butola, B.S., Joshi, M., 2014. Synthesis and characterization of chlorhexidine acetate drug–montmorillonite intercalates for antibacterial applications. Appl. Clay Sci. 101, 477–483. https://doi.org/10.1016/j.clay.2014.09.010.

Salama, B., Alzahrani, K.J., Alghamdi, K.S., Al-Amer, O., Hassan, K.E., Elhefny, M.A., Albarakati, A.J.A., Alharthi, F., Althagafi, H.A., Al Sberi, H., Amin, H.K., Lokman, M. S., Alsharif, K.F., Albrakati, A., Abdel Moneim, A.E., Kassab, R.B., Fathalla, A.S., 2022. Silver nanoparticles enhance oxidative stress, inflammation, and apoptosis in liver and kidney tissues: Potential protective role of thymoquinone. Biol. Trace Elem. Res. https://doi.org/10.1007/s12011-022-03399-w.

Salesa, B., Assis, M., Andrés, J., Serrano-Aroca, Á., 2021. Carbon nanofibers versus silver nanoparticles: Time-dependent cytotoxicity, proliferation, and gene expression. Biomedicines 9, 1155. https://doi.org/10.3390/biomedicines9091155.

Sato, K., Fujimotoa, T., Kanaia, S., Kintaichib, Y., Inabab, M., Hanedab, M., Hamada, H., 1997. Catalytic performance of silver ion-exchanged saponite for the selective reduction of nitrogen monoxide in the presence of excess oxygen. Appl. Catal. B 13, 27–33. https://doi.org/10.1016/S0926-3373(96)00091-4.

Scudiero, D.A., Shoemaker, R.H., Paull, K.D., Monks, A., Tierney, S., Nofziger, T.H., Currens, M.J., Seniff, D., Boyd, M.R., 1988. Evaluation of a soluble tetrazolium/formazan assay for cell growth and drug sensitivity in culture using human and other tumor cell lines. Cancer Res. 48, 4827–4833.

Sengupta, A., Sarkar, A., 2022. Synthesis and characterization of nanoparticles from neem leaves and banana peels: a green prospect for dye degradation in wastewater. Ecotoxicology 31, 537–548. https://doi.org/10.1007/s10646-021-02414-5.

Shabanzadeh, P., Yusof, R., Shameli, K., 2015. Artificial neural network for modeling the size of silver nanoparticles prepared in montmorillonite/starch bionanocomposites. J. Ind. Eng. Chem. 24, 42–50. https://doi.org/10.1016/j.jiec.2014.09.007.

Shahbazi, S., Asadi Karam, M.R., Habibi, M., Talebi, A., Bouzari, S., 2018. Distribution of extended-spectrum β-lactam, quinolone and carbapenem resistance genes, and genetic diversity among uropathogenic Escherichia coli isolates in Tehran. Iran. J. Glob. Antimicrob. Resist. 14, 118–125. https://doi.org/10.1016/j.jgar.2018.03.006.

Shahbazi, S., Sabzi, S., Noori Goodarzi, N., Fereshteh, S., Bolourchi, N., Mirzaie, B., Badmasti, F., 2022. Identification of novel putative immunogenic targets and construction of a multi-epitope vaccine against multidrug-resistant Corynebacterium jeikeium using reverse vaccinology approach. Microb. Pathog. 164, 105425 https://doi.org/10.1016/j.micpath.2022.105425.

Shameli, K., Ahmad, M., Bin Zargar, M., Yunus, W.M.Z.W., Rustaiyan, A., Ibrahim, N.A., 2011. Synthesis of silver nanoparticles in montmorillonite and their antibacterial behavior. Int. J. Nanomedicine 6, 581–590. https://doi.org/10.2147/ijn.s17112.

Shankar, S., Tanomrod, N., Rawdkuen, S., Rhim, J.-W., 2016. Preparation of pectin/silver nanoparticles composite films with UV-light barrier and properties. Int. J. Biol. Macromol. 92, 842–849. https://doi.org/10.1016/j.ijbiomac.2016.07.107.

Sharma, D., Gulati, S.S., Sharma, N., Chaudhary, A., 2022. Sustainable synthesis of silver nanoparticles using various biological sources and waste materials: a review. Emergent Mater. 5, 1649–1678. https://doi.org/10.1007/s42247-021-00292-5.

Shetye, S., Kamat, M., Singh, N., 2022. Incidence of surgical site infection in patients undergoing clean, clean contaminated cases with respect to antibiotic prophylaxis: a prospective observational study. Int. Surg. J. 9, 1835. https://doi.org/10.18203/2349-2902.isj20222938.

Shevtsova, T., Cavallaro, G., Lazzara, G., Milioto, S., Donchak, V., Harhay, K., Korolko, S., Budkowski, A., Stetsyshyn, Y., 2022. Temperature-responsive hybrid nanomaterials based on modified halloysite nanotubes uploaded with silver nanoparticles. Colloids Surf. A Physicochem. Eng. Asp. 641, 128525 https://doi.org/10.1016/j.colsurfa.2022.128525.

Shockman, G.D., Barren, J.F., 1983. Structure, function, and assembly of cell walls of Gram-positive bacteria. Annu. Rev. Microbiol. 37, 501–527. https://doi.org/10.1146/annurev.mi.37.100183.002441.

Shoeibi, S., Mashreghi, M., 2017. Biosynthesis of selenium nanoparticles using Enterococcus faecalis and evaluation of their antibacterial activities. J. Trace Elem. Med. Biol. 39, 135–139. https://doi.org/10.1016/j.jtemb.2016.09.003.

Singh, A., Gaud, B., Jaybhaye, S., 2020. Optimization of synthesis parameters of silver nanoparticles and its antimicrobial activity. Mater. Sci. Energ. Technol 3, 232–236. https://doi.org/10.1016/j.mset.2019.08.004.

Składanowski, M., Golinska, P., Rudnicka, K., Dahm, H., Rai, M., 2016. Evaluation of cytotoxicity, immune compatibility and antibacterial activity of biogenic silver nanoparticles. Med. Microbiol. Immunol. 205, 603–613. https://doi.org/10.1007/s00430-016-0477-7.

Slavin, Y.N., Asnis, J., Häfeli, U.O., Bach, H., 2017. Metal nanoparticles: understanding the mechanisms behind antibacterial activity. J Nanobiotechnol. 15, 65. https://doi.org/10.1186/s12951-017-0308-z.

Slistan-Grijalva, A., Herrera-Urbina, R., Rivas-Silva, J.F., Ávalos-Borja, M., Castillón-Barraza, F.F., Posada-Amarillas, A., 2005. Classical theoretical characterization of the surface plasmon absorption band for silver spherical nanoparticles suspended in water and ethylene glycol. Phys. E Low Dimens Syst. Nanostruct. 27, 104–112. https://doi.org/10.1016/j.physe.2004.10.014.

Sohrabnezhad, Sh., Pourahmad, A., Mehdipour Moghaddam, M.J., Sadeghi, A., 2015. Study of antibacterial activity of Ag and Ag2CO3 nanoparticles stabilized over montmorillonite. Spectrochim. Acta A Mol. Biomol. Spectrosc. 136, 1728–1733. https://doi.org/10.1016/j.saa.2014.10.074.

Sondi, I., Salopek-Sondi, B., 2004. Silver nanoparticles as antimicrobial agent: a case study on E. coli as a model for Gram-negative bacteria. J. Colloid Interface Sci. 275, 177–182. https://doi.org/10.1016/j.jcis.2004.02.012.

Song, H.Y., Ko, K.K., Oh, L.H., Lee, B.T., 2006. Fabrication of silver nanoparticles and their antimicrobial mechanisms. Eur. Cell Mater. 11, 58.

Song, K.C., Lee, S.M., Park, T.S., Lee, B.S., 2009. Preparation of colloidal silver nanoparticles by chemical reduction method. Korean J. Chem. Eng. 26, 153–155. https://doi.org/10.1007/s11814-009-0024-y.

Sonohara, R., Muramatsu, N., Ohshima, H., Kondo, T., 1995. Difference in surface properties between Escherichia coli and Staphylococcus aureus as revealed by electrophoretic mobility measurements. Biophys. Chem. 55, 273–277. https://doi.org/10.1016/0301-4622(95)00004-H.

Sprynskyy, M., Sokol, H., Rafińska, K., Brzozowska, W., Railean-Plugaru, V., Pomastowski, P., Buszewski, B., 2019. Preparation of AgNPs/saponite nanocomposites without reduction agents and study of its antibacterial activity. Colloids Surf. B: Biointerfaces 180, 457–465. https://doi.org/10.1016/j.colsurfb.2019.04.066.

Srinivas, U.S., Tan, B.W.Q., Vellayappan, B.A., Jeyasekharan, A.D., 2019. ROS and the DNA damage response in cancer. Redox Biol. 25, 101084 https://doi.org/10.1016/j.redox.2018.101084.

Sriram, M.I., Kalishwaralal, K., Barathmanikanth, S., Gurunathani, S., 2012. Size-based cytotoxicity of silver nanoparticles in bovine retinal endothelial cells. Nanosci. Meth. 1, 56–77. https://doi.org/10.1080/17458080.2010.547878.

Srivastava, C.M., Purwar, R., Gupta, A.P., 2019. Enhanced potential of biomimetic, silver nanoparticles functionalized Antheraea mylitta (tasar) silk fibroin nanofibrous mats for skin tissue engineering. Int. J. Biol. Macromol. 130, 437–453. https://doi.org/10.1016/j.ijbiomac.2018.12.255.

Subha, V., Ranu, A., Shankar, A., Kirubanandan, S., Satheeshkumar, E., Suresh, S., Pugazhendhi, A., Ilangovan, R., 2022. Functionalization of spray coated cellulose nanofiber sheet with montmorillonite (MMT) and silver nanoparticles (AgNPs) to biomedical nanocomposite as wound regeneration scaffold. Prog. Org. Coat. 166, 106782 https://doi.org/10.1016/j.porgcoat.2022.106782.

Sukdeb, P., Kyung, T.Y., Myong, S.J., 2007. Does the antibacterial activity of Silver nanoparticles depend on the shape of the nanoparticle? A study of the Gram-negative bacterium Escherichia coli. Appl. Environ. Microbiol. 73, 1712–1720. https://doi.org/10.1128/AEM.02218-06.

Sun, J., Wan, J., Zhai, X., Wang, J., Liu, Z., Tian, H., Xin, L., 2021. Silver nanoparticles: Correlating particle size and ionic Ag release with cytotoxicity, genotoxicity, and inflammatory responses in human cell lines. Toxicol. Ind. Health 37, 198–209. https://doi.org/10.1177/0748233721996561.

Takahashi, C., Shirai, T., Fuji, M., 2012. Study on intercalation of ionic liquid into montmorillonite and its property evaluation. Mater. Chem. Phys. 135, 681–686. https://doi.org/10.1016/j.matchemphys.2012.05.044.

Takamiya, A.S., Monteiro, D.R., Gorup, L.F., Silva, E.A., de Camargo, E.R., Gomes-Filho, J.E., de Oliveira, S.H.P., Barbosa, D.B., 2021. Biocompatible silver nanoparticles incorporated in acrylic resin for dental application inhibit Candida albicans biofilm. Mater. Sci. Eng. C 118, 111341. https://doi.org/10.1016/j.msec.2020.111341.

Tang, J., Lu, X., Chen, B., Cai, E., Liu, W., Jiang, J., Chen, F., Shan, X., Zhang, H., 2019. Mechanisms of silver nanoparticles-induced cytotoxicity and apoptosis in rat tracheal epithelial cells. J. Toxicol. Sci. 44, 155–165. https://doi.org/10.2131/jts.44.155.

Tao, Q., Fang, Y., Li, T., Zhang, D., Chen, M., Ji, S., He, H., Komarneni, S., Zhang, H., Dong, Y., Noh, Y.D., 2016. Silylation of saponite with 3-aminopropyltriethoxysilane. Appl. Clay Sci. 132–133, 133–139. https://doi.org/10.1016/j.clay.2016.05.026.

Tao, H., Qian, X., Zhou, Y., Cheng, H., 2022. Research progress of clay minerals in carbon dioxide capture. Renew. Sust. Energ. Rev. 164, 112536 https://doi.org/10.1016/j.rser.2022.112536.

Tariq, M., Mohammad, K.N., Ahmed, B., Siddiqui, M.A., Lee, J., 2022. Biological synthesis of silver nanoparticles and prospects in plant disease management. Molecules 27, 4754. https://doi.org/10.3390/molecules27154754.

Teweldemedhin, M., Gebreyesus, H., Atsbaha, A.H., Asgedom, S.W., Saravanan, M., 2017. Bacterial profile of ocular infections: a systematic review. BMC Ophthalmol. 17, 212. https://doi.org/10.1186/s12886-017-0612-2.

Tian, L., Oulian, L., Zhiyuan, L., Liuimei, H., Xiaosheng, W., 2014. Preparation and characterization of silver loaded montmorillonite modified with sulfur amino acid. Appl. Surf. Sci. 305, 386–395. https://doi.org/10.1016/j.apsusc.2014.03.098.

Tong, S., Davis, J., Eichenberger, E., Holland, T., Fowler, V., 2015. Staphylococcus aureus infections: Epidemiology, pathophysiology, clinical manifestations, and management. Clin. Microbiol. Rev. 28, 603–661. https://doi.org/10.1128/CMR.00134-14.

Tran, H.A., Tran, P.A., 2021. In situ coatings of silver nanoparticles for biofilm treatment in implant-retention surgeries: Antimicrobial activities in monoculture and coculture. ACS Appl. Mater. Interfaces 13, 41435–41444. https://doi.org/10.1021/acsami.1c08239.

Tran, P., Webster, T., 2011. Selenium nanoparticles inhibit Staphylococcus aureus growth. Int. J. Nanomedicine 6, 1553–1558. https://doi.org/10.2147/IJN.S21729.

Tran, P.A., Webster, T.J., 2013. Antimicrobial selenium nanoparticle coatings on polymeric medical devices. Nanotechnology 24, 155101. https://doi.org/10.1088/0957-4484/24/15/155101.

Tran, P.A., O'Brien-Simpson, N., Reynolds, E.C., Pantarat, N., Biswas, D.P., O'Connor, A. J., 2016. Low cytotoxic trace element selenium nanoparticles and their differential antimicrobial properties against S. aureus and E. coli. Nanotechnology 27, 045101. https://doi.org/10.1088/0957-4484/27/4/045101.

Ugwoke, E., Aisida, S.O., Mirbahar, A.A., Arshad, M., Ahmad, I., Zhao, T., Ezema, F.I., 2020. Concentration induced properties of silver nanoparticles and their antibacterial study. Surf. Interf. 18, 100419 https://doi.org/10.1016/j.surfin.2019.100419.

Valásková, M., Simha Martynková, G., Lesková, J., Capkova, P., Klemm, V., Rafaja, D., 2008. Silver nanoparticles/montmorillonite composites prepared using nitrating reagent at water and glycerol. J. Nanosci. Nanotechnol. 8, 3050–3058. https://doi.org/10.1166/jnn.2008.088.

Valle-González, E.R., Jackman, J.A., Yoon, B.K., Park, S., Sut, T.N., Cho, N.-J., 2018. Characterizing how acidic pH conditions affect the membrane-disruptive activities of lauric acid and glycerol monolaurate. Langmuir 34, 13745–13753. https://doi.org/10.1021/acs.langmuir.8b02536.

Varadwaj, G.B.B., Parida, K.M., 2013. Montmorillonite supported metal nanoparticles: an update on syntheses and applications. RSC Adv. 3, 13583–13593. https://doi.org/10.1039/C3RA40520F.

Varghese, M., Balachandran, M., 2021. Antibacterial efficiency of carbon dots against Gram-positive and Gram-negative bacteria: A review. J. Environ. Chem. Eng. 9, 106821 https://doi.org/10.1016/j.jece.2021.106821.

Vigneshwaran, R., Ezhilarasan, D., Rajeshkumar, S., 2021. Inorganic titanium dioxide nanoparticles induces cytotoxicity in colon cancer cells. Inorg. Chem. Commun. 133, 108920 https://doi.org/10.1016/j.inoche.2021.108920.

Vilela, D., Stanton, M.M., Parmar, J., Sánchez, S., 2017. Microbots decorated with silver nanoparticles kill bacteria in aqueous media. ACS Appl. Mater. Interfaces 9, 22093–22100. https://doi.org/10.1021/acsami.7b03006.

Vishwanath, R., Negi, B., 2021. Conventional and green methods of synthesis of silver nanoparticles and their antimicrobial properties. Curr. Res. Green. Sustain. Chem. 4, 100205 https://doi.org/10.1016/j.crgsc.2021.100205.

Wang, Q., Webster, T., 2012. Nanostructured selenium for preventing biofilm formation on medical devices. J. Biomed. Mater. Res. A 100, 3205–3210. https://doi.org/10.1002/jbm.a.34262.

Wang, M.-C., Lin, J.-J., Tseng, H.-J., Hsu, S., 2012. Characterization, antimicrobial activities, and biocompatibility of organically modified clays and their nanocomposites with polyurethane. ACS Appl. Mater. Interfaces 4, 338–350. https://doi.org/10.1021/am2014103.

Wang, L., He, H., Yu, Y., Sun, L., Liu, S., Zhang, C., He, L., 2014a. Morphology-dependent bactericidal activities of Ag/CeO2 catalysts against Escherichia coli. J. Inorg. Biochem. 135, 45–53. https://doi.org/10.1016/j.jinorgbio.2014.02.016.

Wang, X., Ji, Z., Chang, C.H., Zhang, H., Wang, M., Liao, Y.-P., Lin, S., Meng, H., Li, R., Sun, B., Van Winkle, L., Pinkerton, K.E., Zink, J.I., Xia, T., Nel, A.E., 2014b. Use of coated silver nanoparticles to understand the relationship of particle dissolution and bioavailability to cell and lung toxicological potential. Small 10, 385–398. https://doi.org/10.1002/smll.201301597.

Wang, H., Xie, H., Wang, S., Gao, Z., Li, C., Hu, G.-H., Xiong, C., 2018. Enhanced dielectric property and energy storage density of PVDF-HFP based dielectric composites by incorporation of silver nanoparticles-decorated exfoliated montmorillonite nanoplatelets. Compos Part A Appl. Sci. Manuf. 108, 62–68. https://doi.org/10.1016/j.compositesa.2018.02.020.

Wang, J., Chen, X.-Y., Zhao, Y., Yang, Y., Wang, W., Wu, C., Yang, B., Zhang, Z., Zhang, L., Liu, Y., Du, X., Li, W., Qiu, L., Jiang, P., Mou, X.-Z., Li, Y.-Q., 2019. pH-switchable antimicrobial nanofiber networks of hydrogel eradicate biofilm and rescue stalled healing in chronic wounds. ACS Nano 13, 11686–11697. https://doi.org/10.1021/acsnano.9b05608.

Wei, J.-C., Yen, Y.-T., Wang, Y.-T., Hsu, S., Lin, J.-J., 2013. Enhancing silver nanoparticle and antimicrobial efficacy by the exfoliated clay nanoplatelets. RSC Adv. 3, 7392. https://doi.org/10.1039/c3ra23476b.

Wei, S., Wang, Y., Tang, Z., Hu, J., Su, R., Lin, J., Zhou, T., Guo, H., Wang, N., Xu, R., 2020. A size-controlled green synthesis of silver nanoparticles by using the berry extract of Sea Buckthorn and their biological activities. New J. Chem. 44, 9304–9312. https://doi.org/10.1039/D0NJ01335H.

Whitfield, C., Wear, S.S., Sande, C., 2020. Assembly of bacterial capsular polysaccharides and exopolysaccharides. Annu. Rev. Microbiol. 74, 521–543. https://doi.org/10.1146/annurev-micro-011420-075607.

Wu, T.-S., Wang, K.-X., Li, G.-D., Sun, S.-Y., Sun, J., Chen, J.-S., 2010. Montmorillonite-supported Ag/TiO2 nanoparticles: An efficient visible-light bacteria photodegradation material. ACS Appl. Mater. Interfaces 2, 544–550. https://doi.org/10.1021/am900743d.

Wu, Z., Huang, X., Li, Y.-C., Xiao, H., Wang, X., 2018. Novel chitosan films with laponite immobilized Ag nanoparticles for active food packaging. Carbohydr. Polym. 199, 210–218. https://doi.org/10.1016/j.carbpol.2018.07.030.

Wypij, M., Jędrzejewski, T., Ostrowski, M., Trzcińska, J., Rai, M., Golińska, P., 2020. Biogenic silver nanoparticles: Assessment of their cytotoxicity, genotoxicity and study of capping proteins. Molecules 25, 3022. https://doi.org/10.3390/molecules25133022.

Wypij, M., Jędrzejewski, T., Trzcińska-Wencel, J., Ostrowski, M., Rai, M., Golińska, P., 2021. Green synthesized silver nanoparticles: Antibacterial and anticancer activities, biocompatibility, and analyses of surface-attached proteins. Front. Microbiol. 12, 632505 https://doi.org/10.3389/fmicb.2021.632505.

Xiao, Y., Wang, Y., Zhu, W., Yao, J., Sun, C., Militky, J., Venkataraman, M., Zhu, G., 2021. Development of tree-like nanofibrous air filter with durable antibacterial property. Sep. Purif. Technol. 259, 118135 https://doi.org/10.1016/j.seppur.2020.118135.

Xiu, Z., Zhang, Q., Puppala, H.L., Colvin, V.L., Alvarez, P.J.J., 2012. Negligible particle-specific antibacterial activity of silver nanoparticles. Nano Lett. 12, 4271–4275. https://doi.org/10.1021/nl301934w.

Yan, Y., Li, C., Wu, H., Du, J., Feng, J., Zhang, J., Huang, L., Tan, S., Shi, Q., 2019. Montmorillonite-modified reduced graphene oxide stabilizes copper nanoparticles and enhances bacterial adsorption and antibacterial activity. ACS Appl. Bio. Mater. 2, 1842–1849. https://doi.org/10.1021/acsabm.8b00695.

Yin, I.X., Zhang, J., Zhao, I.S., Mei, M.L., Li, Q., Chu, C.H., 2020. The antibacterial mechanism of silver nanoparticles and its application in dentistry. Int. J. Nanomedicine 15, 2555–2562. https://doi.org/10.2147/IJN.S246764.

Ying, W., Tan, J., Chen, C., Sun, T., Wang, S., Zhang, M., 2019. Biofabrication of silver nanoparticles and its application for development of wound dressing system in nursing care for burn injuries in children. J. Drug. Deliv. Sci. Technol. 54, 101236 https://doi.org/10.1016/j.jddst.2019.101236.

Younas, W., Khan, F.U., Zaman, M., Lin, D., Zuberi, A., Wang, Y., 2022. Toxicity of synthesized silver nanoparticles in a widespread fish: A comparison between green and chemical. Sci. Total Environ. 845, 157366 https://doi.org/10.1016/j.scitotenv.2022.157366.

Youssef, M.M., El-Mansy, M.N., El-Borady, O.M., Hegazy, E.M., 2021. Impact of biosynthesized silver nanoparticles cytotoxicity on dental pulp of albino rats (histological and immunohistochemical study). J. Oral Biol. Craniofac. Res. 11, 386–392. https://doi.org/10.1016/j.jobcr.2021.04.002.

Yuan, Q., Golden, T.D., 2020. A novel method for synthesis of clay/polymer stabilized silver nanoparticles. Surf. Interf. 20, 100620 https://doi.org/10.1016/j.surfin.2020.100620.

Yuan, M., Gao, G., Hu, X., Luo, X., Huang, Y., Jin, B., Liang, Z., 2018. Premodified sepiolite functionalized with triethylenetetramine as an effective and inexpensive adsorbent for CO2 capture. Ind. Eng. Chem. Res. 57, 6189–6200. https://doi.org/10.1021/acs.iecr.8b00348.

Zahoor, M., Nazir, N., Iftikhar, M., Naz, S., Zekker, I., Burlakovs, J., Uddin, F., Kamran, A. W., Kallistova, A., Pimenov, N., Ali Khan, F., 2021. A review on silver nanoparticles: classification, various methods of synthesis, and their potential roles in biomedical applications and water treatment. Water (Basel) 13, 2216. https://doi.org/10.3390/w13162216.

Zhang, Y., Wade, M.M., Scorpio, A., Zhang, H., Sun, Z., 2003. Mode of action of pyrazinamide: disruption of Mycobacterium tuberculosis membrane transport and energetics by pyrazinoic acid. J. Antimicrob. Chemother. 52, 790–795. https://doi.org/10.1093/jac/dkg446.

Zhang, L., Chen, J., Yu, W., Zhao, Q., Liu, J., 2018. Antimicrobial nanocomposites prepared from montmorillonite/Ag+/quaternary ammonium nitrate. J. Nanomater. 2018, 6190251. https://doi.org/10.1155/2018/6190251.

Zhang, J., Ahmadi, M., Fargas, G., Perinka, N., Reguera, J., Lanceros-Méndez, S., Llanes, L., Jiménez-Piqué, E., 2022. Silver nanoparticles for conductive inks: From synthesis and ink formulation to their use in printing technologies. Metals (Basel) 12, 234. https://doi.org/10.3390/met12020234.

Zhou, C.H., Zhou, Q., Wu, Q.Q., Petit, S., Jiang, X.C., Xia, S.T., Li, C.S., Yu, W.H., 2019. Modification, hybridization and applications of saponite: An overview. Appl. Clay Sci. 168, 136–154. https://doi.org/10.1016/j.clay.2018.11.002.

Zhu, W., Li, J., Lei, J., Li, Y., Chen, T., Duan, T., Yao, W., Zhou, J., Yu, Y., Liu, Y., 2018. Silver nanoparticles incorporated konjac glucomannan-montmorillonite nacre-like

composite films for antibacterial applications. Carbohydr. Polym. 197, 253–259. https://doi.org/10.1016/j.carbpol.2018.06.005.
Ziąbka, M., Menaszek, E., Tarasiuk, J., Wroński, S., 2018. Biocompatible nanocomposite implant with silver nanoparticles for otology—In vivo evaluation. Nanomaterials 8, 764. https://doi.org/10.3390/nano8100764.
Zorraquín-Peña, I., Cueva, C., de Llano, D., Bartolomé, B., Moreno-Arribas, M.V., 2020. Glutathione-stabilized silver nanoparticles: Antibacterial activity against periodontal bacteria, and cytotoxicity and inflammatory response in oral cells. Biomedicines 8, 375. https://doi.org/10.3390/biomedicines8100375.