SILVER NANOPARTICLES BASED ON CAFFEIC ACID APPLIED FOR TEXTILES PRESERVATION

Mihaela-Cristina LITE[1], Irina-Mariana SĂNDULACHE[2], Elena-Cornelia TĂNĂSESCU[3], Roxana CONSTANTINESCU[4], Camelia UNGUREANU[5], Nicoleta BADEA[6]

The present study investigates the potential application of green synthesized silver nanoparticles (AgNPs) for textile preservation. Thus, silver nanoparticles were produced, using caffeic acid as reducing agent. For synthesis optimization, the average particle size was evaluated by DLS technique and confirmed by SEM. The physical stability of the resulting dispersions was determined by zeta potential measurements. The dispersion with the optimal ratio CA:$AgNO_3$ (v:v) was applied on textile samples and its effect was further investigated, in terms of chromatic parameters and antimicrobial activity against bacteria (Salmonella typhimurium and Bacillus subtilis) and fungi (Aspergillus brasiliensis and Penicillium hirsutum) strains.

Keywords: Silver nanoparticles, caffeic acid, antimicrobial activity, textile preservation

1. Introduction

The degradation of textile objects is influenced by several factors: the presence of microorganisms, the availability of oxygen, temperature, humidity, light, chemical factors (pH, electrolytes, etc.) [1, 2]. The main method of preserving them involves maintaining an appropriate microclimate, with a temperature below 22°C and a relative humidity of 65% [3]. In addition to maintaining the microclimate, the literature describes several physical and chemical methods for

[1] PhD student, Department of General Chemistry, Faculty of Chemical Engineering and Biotechnologies, University POLITEHNICA of Bucharest, Romania, e-mail: lite.cristina@gmail.com
[2] Eng., INCDTP - National Research & Development Institute for Textiles and Leather, Bucharest, Romania, e-mail: irina.sandulache@incdtp.ro
[3] PhD student, INCDTP - National Research & Development Institute for Textiles and Leather, Bucharest, Romania, e-mail: cornelia.tanasescu@incdtp.ro
[4] Eng., Department of Biotechnologies and Environmental Protection, ICPI - Leather and Footwear Research Institute, Bucharest, Romania, e-mail: rodica.constantinescu@icpi.ro
[5] Assoc. Prof. PhD Eng., Department of General Chemistry, Faculty of Chemical Engineering and Biotechnologies, University POLITEHNICA of Bucharest, Romania, e-mail: camelia.ungureanu@upb.ro
[6] Prof. PhD Eng., Department of General Chemistry, Faculty of Chemical Engineering and Biotechnologies, University POLITEHNICA of Bucharest, Romania, e-mail: nicoleta.badea@upb.ro

disinfecting heritage textile objects, which aim to prevent their degradation due to microorganisms. Physical methods refer to dehydration, exposure to gamma radiation, and placing objects in a low-oxygen environment. Chemical methods involve the use of various chemical compounds, such as alcohols, phenols, azoles, quaternary ammonium salts, nanomaterials, etc. [4].

The research in the field of nanomaterials demonstrated that certain nanoparticles (NPs) exhibit UV-blocking, deacidification, and antimicrobial properties, making them potential candidates for conservation treatments [5-6].

Silver ions and silver-based compounds are very toxic to microorganisms. Silver is used for its antibacterial effect in a variety of applications, for example dental work, burn treatment and water treatment [7-10]. Metal nanoparticles are the most promising systems, as they show improved antibacterial properties due to their high surface-to-volume ratio. The interest of research in this direction is in continuous development due to the increasing microbial resistance against metal ions, antibiotics, and the development of resistant strains.

There are several methods for the synthesis of silver nanoparticles (AgNPs). They can be obtained using chemical, physical, photochemical, and biological methods. Each method has advantages and disadvantages, the main issues being the cost, its applicability, and the particle size. The synthesis of silver nanoparticles by green methods is of great interest due to its nontoxicity and low costs [11-13].

Caffeic acid (3,4-dihydroxycinnamic acid) appears as a yellow solid and is found in many plants, being an intermediate in lignin biosynthesis. It is found in moderate amounts in thyme, sage, mint, in sunflower seeds, in higher amounts in black tea, and in the yerba mate plant from South America [14, 15]. Caffeic acid presents potential anti-inflammatory, antioxidant, antihyperglycemic and antineoplastic activities. It prevents oxidative stress, both in vitro and in vivo, therefore preventing DNA damage induced by free radicals [16, 17].

This paper describes the synthesis of silver nanoparticles using caffeic acid as reducing agent. The efficiency of the reactions is first evaluated by UV-Vis absorption spectrometry, highlighting the formation of silver nanoparticles by the presence of the specific plasmon resonance (SPR) band in the UV-Vis spectrum. Subsequently, the average size and stability of the nanoparticles was evaluated by DLS technique (Dynamic Light Scattering) and zeta potential measurements. For a clearer picture of the size, shape, and distribution of silver nanoparticles, they were characterized using a scanning electron microscope (SEM) equipped with an X-ray detector for the elemental analysis of the components present in the obtained dispersions.

The AgNPs dispersion was used to treat textile samples (cotton and wool) and they were further characterized by measuring the chromatic parameters. The presence of AgNPs on the textile support was studied by scanning electron microscopy and X-ray dispersive spectroscopy. The antimicrobial effect of the AgNPs dispersion applied

on textile samples was tested against bacteria (*Salmonella typhimurium* and *Bacillus subtilis*) and fungi (*Aspergillus brasiliensis* and *Penicillium hirsutum*) strains.

2. Experimental

2. 1. Material and methods

The silver precursor used in the AgNPs synthesis was silver nitrate, purchased from Anal-R NORMAPUR. The textile samples were provided by the National Research and Development Institute for Textiles and Leather–INCDTP (Bucharest, Romania). The rest of the regents were purchased from Merck. The antimicrobial activity tests were conducted using *Salmonella typhimurium* ATCC 14028 (Gram negative) and *Bacillus subtilis* ATCC 6633 (Gram positive) bacteria strains and *Aspergillus brasiliensis* ISM 73/34 and *Penicillium hirsutum* ATCC 52323 fungi strains.

2. 2. Synthesis and characterization of silver nanoparticles

An aqueous solution of caffeic acid 1 mM was prepared, adjusting the pH to 11.80 with NaOH, a value at which its complete dissolution occurred. Sodium hydroxide was used to adjust the pH. A Crison GLP 21+ pH meter was used to monitor the pH variation of the reaction mixtures.

The synthesis of AgNPs was carried out using different ratios CA:$AgNO_3$, 1:1, 1:3, 1:5 and 1:9 (v:v). The formation of AgNPs was indicated by the dark color of the reaction mixtures. After two hours, they were centrifuged for 30 minutes at 5000 rpm and the supernatant was collected and characterized.

The formation of AgNPs was monitored by recording the UV-Vis absorption spectrum, using a Lambda 950 spectrophotometer from Perkin Elmer, and confirmed by the presence of the characteristic absorption band, the SPR (surface plasmon resonance) [18, 19]. Subsequently, the size and polydispersity of the AgNPs was evaluated by Dynamic Light Scattering (DLS) using a Zetasizer Nano ZS instrument (Malvern Instruments Ltd., Worcestershire, UK). The physical stability of the dispersions containing AgNPs was determined by measuring the zeta potential, using the same instrument.

The morphology of the AgNPs was evaluated by Scanning Electron Microscopy (SEM) using a FEI Quanta 200 microscope equipped with an Everhart-Thornley (ET) detector. By coupling an EDAX-AMETEK X-ray detector to the electron microscope, the nature of the AgNPs was confirmed, by performing X-ray dispersive spectroscopy (EDS).

2. 3. Evaluation of the effect of AgNPs on textile samples

The deposition of silver nanoparticles on the selected textile samples was performed by directly soaking the cotton and wool samples (10 cm × 10 cm) in the resulting dispersion from synthesis. After drying, the chromatic parameters were measured and expressed in the CIE L*a*b* system of colors, for the treated textile

samples and compared to the reference fabrics, using the Datacolor spectrophotometer (with a D65/10 lamp).

SEM characterization was conducted to evaluate the distribution of the AgNPs on the textile fibers and their nature was confirmed by EDS.

Prior to the antibacterial assessment, all samples (1 cm^2) were UV sterilized using the Vilber Lourmat equipment (5 minutes on each side). The samples were tested in triplicate. The bacterial strains were grown on Luria Bertani (L.B.) culture medium, according to Ansari et.al [20] at 37 $^{\circ}$C. The L.B. composition consists of peptone (10 g/L), yeast extract (5 g/L), and NaCl (5 g/L). Sterile culture medium (L.B., 10 mL) was inoculated with 1% bacterial suspension. The sterile samples were incubated for 18 hours in test tubes containing 0.5 mL culture medium inoculated with the tested microorganism. The optical densities were determined using the UV-VIS spectrophotometer Jenway Spectrophotometer instrument, after 18 hours of incubation in a Laboshake Gerhardt incubator. The inhibition rate was calculated with the following formula:

$$I\% = \frac{(B_{18} - B_0) - (C_{18} - C_0)}{(B_{18} - B_0)} \times 100 \quad (1)$$

where: I% represents the rate (degree) of inhibition of the pathogen by the antibacterial agent, B_{18} represents the optical density value at 600 nm of the bacterial suspension after 18 hours of incubation, B_0 represents the optical density value at 600 nm of the bacterial suspension before incubation, C_{18} represents the density value optical density at 600 nm of the bacterial suspension in the presence of the treated sample after 18 hours of incubation and C_0 represents the value of the optical density at 600 nm of the bacterial suspension in the presence of the treated sample before incubation.

The antifungal activity was assessed by Kirby-Bauer disk diffusion method [21, 22] which determines the degree of inhibition on the solid medium by measuring the diameter of the inhibition zone of the antifungal agent. The culture medium (PDA, Potato-Dextrose-Agar) was autoclaved and then poured into Petri dishes. After its solidification, the Petri dish was seeded with microorganism suspension to be tested. Seeding over the entire surface of the plate was performed using a sterile Drigalski spatula. The sterile samples were placed on the center of the plate. All samples (1 cm^2) were UV sterilized using the Vilber Lourmat equipment (5 minutes on each side) and the tests were performed in triplicate.

The evolution/involution of the pathogen culture was followed at 72-96 hours by comparison with the negative control plate (untreated sample) and the inhibition zone (I.Z.) was measured in millimeters (mm), using the formula:

$$IZ = \frac{D - d}{2} \quad (2)$$

where D is the total diameter of the sample and the inhibition zone (mm), and d is diameter of the sample (mm).

3. Results and discussion

3. 1. Physico-chemical, morphological, and biological characterization of silver nanoparticles

The synthesis of silver nanoparticles using different ratios of caffeic acid and $AgNO_3$ was spectrally monitored. The overlapped absorption spectra are presented in Fig. 1. The absorption spectrum of caffeic acid exhibits an absorption maximum at the wavelength of 267 nm, specific to the deprotonated form. According to specialized literature, for pH values between 7 and 10.5, the absorption spectrum of caffeic acid shows two absorption maxima, at 290 nm and 320 nm, which decreases in intensity as the pH decreases [23]. At pH=11, this doublet is replaced with a single band at $\lambda \sim 270$ nm. The intensity of this peak decreases during the formation of AgNPs, indicating the role of caffeic acid in the reduction of Ag ions.

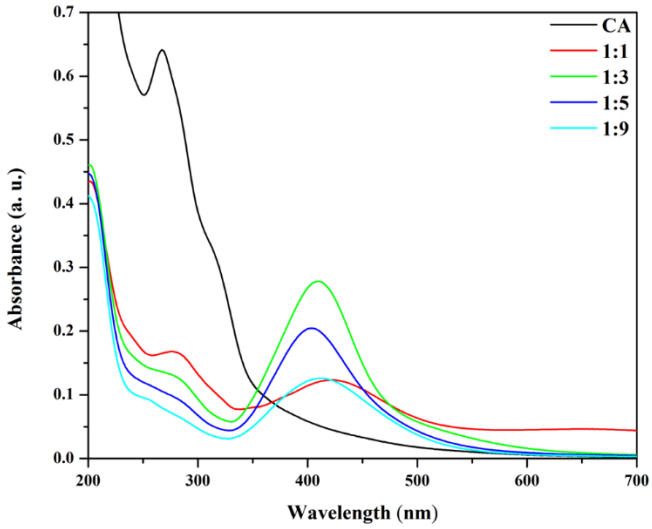


Fig. 1. Overlapped UV-Vis absorption spectra for AgNPs dispersions resulting from the caffeic acid reaction in different CA: $AgNO_3$ ratios.

The presence of the absorption bands in the UV-Vis spectra at wavelengths in the range 400-420 nm, specific to the surface plasmon resonance (SPR) of silver nanoparticles, demonstrates the formation of silver nanoparticles following the reaction of silver nitrate with caffeic acid.

The intensity, shape, and shift of the absorption bands are indicators of the size, distribution, and stability of the AgNPs [24]. In the case of the 1:1 and 1:9 ratios, the SPR band is wide (absorption maxima at 422 nm and 412 nm, respectively), indicating the formation of large silver nanoparticles or their agglomeration, compared to the 1:3 and 1:5 ratios, which present absorption

maxima at 410 nm and 404 nm. The highest band intensity and sharpness was recorded when the ration CA: $AgNO_3$ was 1:3.

The particle size (Z_{av}) and the polydispersity index (PdI) of the samples were estimated by DLS measurements (Fig. 2a). AgNPs present a mean diameter between 89 and 767 nm and a bimodal size distribution. A general tendency to increase the average particle size and the polydispersity index is observed at high concentrations of $AgNO_3$ and caffeic acid. This behavior indicates that the higher availability of silver ions leads to a faster growth of the silver nanoparticles, which might aggregate into clusters, while higher concentrations of caffeic acid might work as a steric barrier inhibiting the long-distance growing process. Moreover, while for the ratios 1:3 and 1:5 the average particle size is maintained in the nanoscale, the size of the silver particles produced using 1:1 and 1:9 ratios exceed the nanoscale (particle sizes above 100 nm). The DLS values correlate with the UV-Vis spectroscopy results. For the ratios with the lowest AgNPs average particle sizes (1:3 and 1:5) presented sharp, well defined SPR bands in the UV-Vis spectrum, while in the case of 1:1 and 1:9 ratios, the wide SPR bands confirms silver particles sizes, which are situated in the microscale [25].

The physical stability of silver nanoparticles was evaluated by measurement to zeta potential (ξ), the values of this parameter illustrated in Fig. 2b.

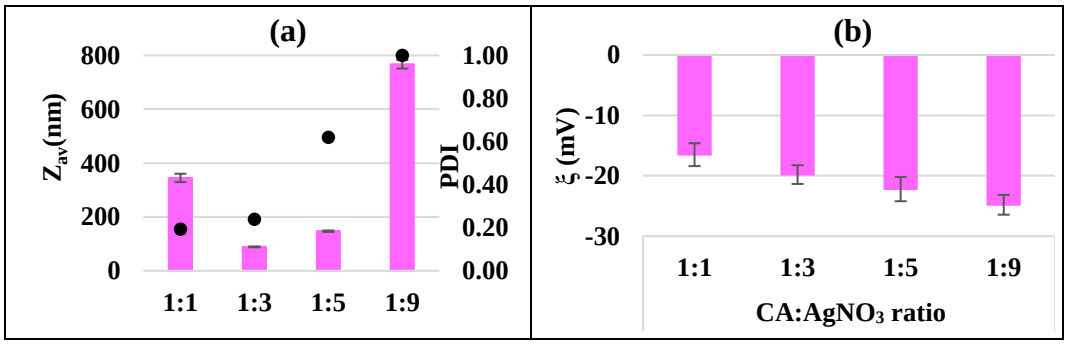


Fig. 2 Average size, polydispersity index (PdI) and Zeta potential of the AgNPs produced for different CA: $AgNO_3$ ratios

The physical stability of the AgNPs dispersions follows an increasing trend with the increase of the silver ions concentration.

The AgNPs dispersion synthesized using the ratio 1:3 (CA: $AgNO_3$) was selected for further experiments, due to its low average size (88.87 ± 2.53 nm), with a polydisperse index of 0.240, and a moderate physical stability (ξ = -19.80 ± 1.55).

Morphological characterization of AgNPs. The SEM images and EDS spectra are presented in Fig. 3.

SEM results confirm the DLS values obtained for AgNPs synthesized using the ratio 1:3 CA: $AgNO_3$. Moreover, it is observed that the small particles, with

diameters in the range 57.6 – 106.2 nm, aggregate into clusters of different shapes. The EDS spectrum confirmed the nature of the AgNPs.

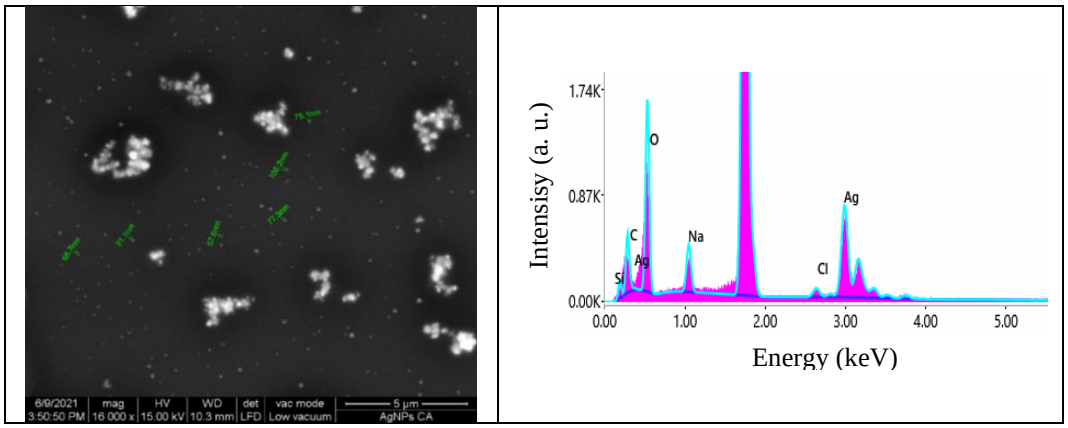


Fig.3. SEM images and EDS spectrum of the AgNPs synthesized using the ratio CA:$AgNO_3$ of 1:3

3. 2. Characterization of the textile samples treated with the AgNPs dispersion

The effect of AgNPs on textile materials was determined by visual evaluation, by measuring the chromatic parameters and by testing the antimicrobial activity.

Chromatic parameters measurements. Table 1 includes the values of the measured chromatic parameters L* a* b*, as well as the value of the color difference, ΔE*, compared to the untreated reference textile material.

Table 1

Chromatic parameters for cotton and wool samples treated with AgNPs dispersion.

Textile fabric	Sample	L*	a*	b*	ΔL*	Δa*	Δb*	ΔE*
Cotton	**Reference**	93.58	-0.01	3.19	–	–	–	–
	AgNPs	85.64	0.29	3.61	-7.94	0.30	0.42	**7.96**
Wool	**Reference**	85.32	-0.04	12.07	–	–	–	–
	AgNPs	83.16	-0.08	11.08	-2.16	-0.04	-0.99	**2.37**

The measurements of the chromatic parameters suggest that the nature of the textile fibers influences the appearance of the textile material following the application of the AgNPs dispersion treatment. Specifically, wool is less affected compared to cotton, having a color difference ΔE* of 2.37 compared to the reference wool sample, as opposed to cotton showing a ΔE* of 7.96 compared to the reference cotton sample. The parameter L* (lightness) is the most affected in both situations, but while in the case of cotton it decreases by almost 8 units, the decrease recorded for wool is only 2 units.

Morphological evaluation and nature confirmation of the AgNPs deposited on textile. Fig. 4 illustrates the morphology of the textile fibers before and after depositing AgNPs. The recorded EDS spectra are presented in Fig. 5.

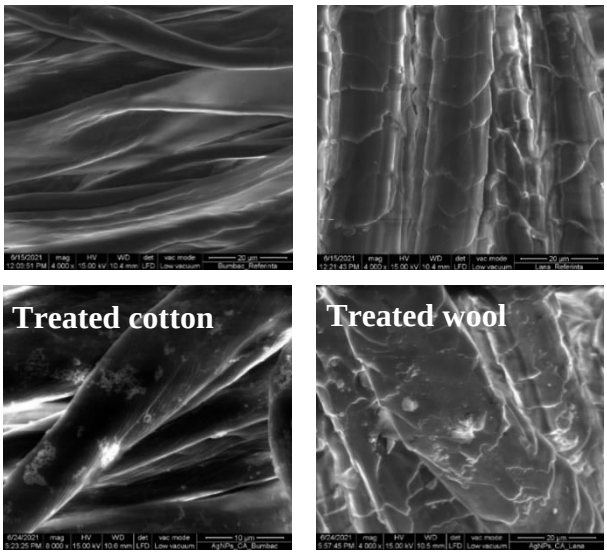


Fig. 4. SEM images obtained for untreated cotton and wool textile samples and for AgNPs synthesized using the ratio CA:$AgNO_3$ of 1:3 deposited on cotton and wool textile samples.

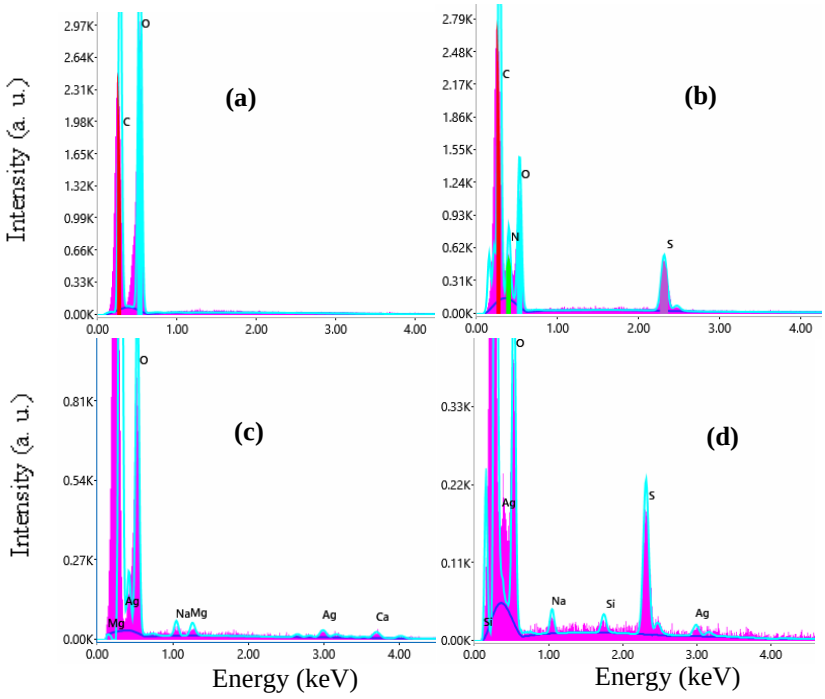


Fig. 5. EDS spectra of untreated cotton (a) and wool (b) and with deposited AgNPs cotton (c) and wool (d).

The SEM images reveal that the deposition of silver nanoparticles was achieved uniformly as aggregates, both on the cotton and wool fibers. Moreover,

the treatment with AgNPs depression obtained using caffeic acid as a reducing agent does not alter the integrity of the textile fibers. The EDS spectra confirmed the presence of silver on textile fibers.

Evaluation of the antimicrobial activity. The wool sample was selected for antimicrobial evaluation because it was the least affected by the treatment in terms of color change, and Fig. 6 shows the antimicrobial effect of AgNPs on these samples.

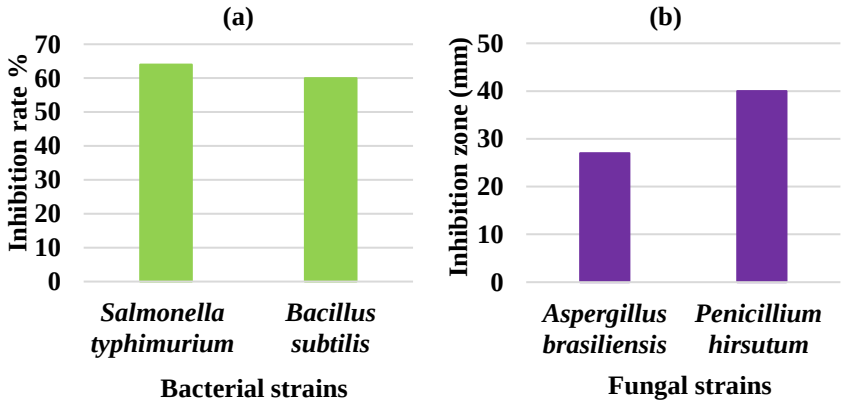


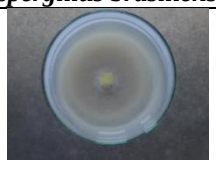
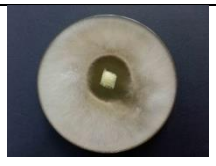
Fig. 6. Antimicrobial activity of the AgNPs dispersion, ratio 1:3 (CA:$AgNO_3$), applied on wool samples for (a) bacterial strains and (b) fungal strains.

The difference between the two values of the inhibition rate for the two types of bacteria tested (64 % for *Salmonella typhimurium* and 60% for *Bacillus subtilis*) is attributed to the structure of the cell wall. *Salmonella typhimurium* is a Gram-negative bacterium, unlike *Bacillus subtilis* which is a Gram-positive bacterium, with a more resistant cell wall, due to the basic unit - peptidoglycan.

The samples tested on *Aspergillus brasiliensis* and *Penicillium hirsutum* fungal strains had an inhibitory effect of 27 mm and 40 mm, respectively. Representative images of this effect are illustrated in Table 2.

Table 2

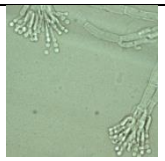
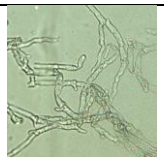
Images of the inhibition zone resulted when inoculating untreated and AgNPs treated wool samples with fungal strains.

	Aspergillus brasiliensis	***Penicillium hirsutum***
Untreated wool		
Treated wool		

The antifungal effect was evaluated in terms of reproductive inhibition, by investigating the colonies using the Epifluorescent FLUO 3 light microscope at 200X magnification. Representative images are shown in table 3.

Table 3

Microscopic appearance of the reproductive formations for the tested fungal strains.

	Aspergillus brasiliensis	***Penicillium hirsutum***
Untreated wool		
Treated wool		

In the case of *Aspergillus brasiliensis*, the control sample shows numerous chained conidiospores in contrast to the microscopic preparation of the treated wool sample, where hyphae with "naked" vesicles are observed and a reduced number of conidiospores. Microscopic observations showed that there are morphological alterations of hyphae that appear degraded with large vesicles. Similarly, in the case of the *Penicillium hirsutum* fungus the fungal colonies are much smaller and with a partial loss of spore formation for the treated wool samples, unlike the control sample where a pronounced sporulation is observed.

4. Conclusions

The work provided results regarding the applicability of green synthesized AgNPs for textile preservation. Caffeic acid was used as reducing agent of the silver precursor, in order to obtain stable nanoparticles, with an average size of 88.87 ± 2.53 nm and a polydisperse index of 0.240, determined by DLS technique. SEM characterization confirmed the size results and provided a clear picture of the distribution of the particles and the manner they aggregate into clusters of different shapes. The dispersion with the optimal ratio (1:3), CA: $AgNO_3$ (v: v), was selected to be applied on textile samples (cotton and wool). The chromatic parameters measurements revealed that wool samples are less affected by the AgNPs treatment, with a color change of $\Delta E^* = 2.37$, compared to cotton treated samples ($\Delta E^* = 7.96$). The treatment applied on wool samples exhibited inhibition rates of 64% and 60% against Gram-negative bacterium (*Salmonella typhimurium*) and Gram-positive bacterium (*Bacillus subtilis*), respectively. The inhibition growth of the fungal strains was demonstrated by measuring the inhibition zone on inoculated Petri dishes

containing untreated and AgNPs treated samples. The values of the inhibition zone were 27 mm for *Aspergillus brasiliensis* and 40 mm in the case of *Penicillium hirsutum*. Microscopic observation showed that the fungal colonies are much smaller and with a partial loss of spore formation for the AgNPs treated wool samples, in contrast to the control sample where a pronounced sporulation is observed.

Acknowledgment

This work was supported by the Ministry of Research and Innovation within Program 1–Development of the national RD system, Subprogram 1.2–Institutional Performance–RDI excellence funding projects, contract no. 4PFE/2021, with contribution from the European Social Fund from the Sectoral Operational Programme Human Capital 2014-2020, through the Financial Agreement with the title "Training of PhD students and post-doctoral researchers in order to acquire applied research skills–SMART", contract no. 13530/16.06.2022–SMIS code: 153734.

R E F E R E N C E S

[1]. M. Zee, J. H. Stoutjesdijk, P. A. A. W. Heijden, D. Wit, "Structure biodegradation relationships of polymeric materials. 1. Effect of degree of oxidation on biodegradability of carbohydrate polymers", in Journal of Polymers and the Environment, **3**, 1995, pp. 235–242.

[2]. E. Pekhtasheva, A. Neverov, S. Kubica, G. Zaikov, "Biodegradation and biodeterioration of some natural polymers", in Chemistry and Chemical Technology, **6**, 2012, pp. 263–280.

[3]. Romanian government decision no. 1546/2003 for the approval of the conservation and restoration norms of classified movable cultural assets.

[4]. B. Gutarowska, K. Pietrzak, W. Machnowski, J. M. Milczarek, "Historical textiles – a review of microbial deterioration analysis and disinfection methods", in Textile Research Journal, **87**, 2017, 2388–2406.

[5]. M.C. Lite, R.R. Constantinescu, E.C. Tănăsescu, A. Kuncser, C. Romaniţan, I. Lăcătuşu, N. Badea, "Design of green silver nanoparticles based on Primula Officinalis extract for textile preservation", in Materials, **15**, 2022, pp. 7695-7715.

[6]. M. C. Lite, R. R. Constantinescu, E. C. Tănăsescu, O. G. Iordache, N. Badea, "Textile artefacts conservation using nanomaterials – Review", in Industria Textila, **73**, 2022, pp. 607-613.

[7]. J. S. Kim, E. Kuk, K. N. Yu, J. H. Kim, S. J. Park, H. J. Lee, S. H. Kim, Y. K. Park, Y. H. Park, C. Y. Hwang, Y. K. Kim, Y. S. Lee, D. H. Jeong, M. H. Cho, "Antimicrobial effects of silver nanoparticles", in Nanomedicine: Nanotechnology, Biology and Medicine, **3**, 2007, pp. 95–10.

[8]. M. Rai, A.Yadav, A. Gade, "Silver nanoparticles as a new generation of antimicrobials", in Biotechnology Advances, **27**, 2009, pp. 76–83.

[9]. M. E. Barbinta-Patrascu, N. Badea, C. Ungureanu, D. Besliu, S. Antohe, "Bioactive phyto-nanosilver particles "green" synthesized from clary sage, burdock, southernwood and asparagus", in Romanian Reports in Physics, **72**, 2020, pp. 1-13.

[10]. M. E. Barbinta-Patrascu, C. Ungureanu, N. Badea, M. Bacalum, A. Lazea-Stoyanova, I. Zgura, C. Negrila, M. Enculescu, C. Burnei, "Novel ecogenic plasmonic biohybrids as multifunctional bioactive coatings", in Coatings, **10**, 2020, pp. 659-681.

[11]. Q. H. Tran, V. Q. Nguyen, A. Le, " Silver nanoparticles : synthesis , properties , toxicology, applications and perspectives", in Advances in Natural Sciences: Nanoscience and Nanotechnology, 2013, **4**, pp. 1-20.

[12]. M. E. Barbinta-Patrascu, N. Badea, C. Ungureanu, M. Constantin, C. Pirvu, I. Rau, "Silver-based biohybrids "green" synthesized from *Chelidonium majus L.*" in Optical Materials, **56**, 2016, pp. 94–99.

[13]. M. E. Barbinta-Patrascu, C. Nichita, N. Badea, C. Ungureanu, M. Bacalum, I. Zgura, L. Iosif, S. Antohe, "Biophysical aspects of bio-nanosilver generated from urtica dioica leaves and vitis vinifera fruits' extracts", in Romanian Reports in Physics, **73**, 2021, pp. 1-14.

[14]. M. Bojić, V. Simon Haas, D. Šarić, Ž. Maleš, "Determination of Flavonoids, Phenolic Acids, and Xanthines in Mate Tea (Ilex paraguariensis St.-Hil.)", in Journal of Analytical Methods in Chemistry, **2013**, 2013, pp. 1–6.

[15]. K. A. S. Berté, M. R. Beux, P. K. W. D. S. Spada, M. Salvador, R. Hoffmann-Ribani, "Chemical Composition and Antioxidant Activity of Yerba-Mate (Ilex paraguariensis A.St.-Hil., Aquifoliaceae) Extract as Obtained by Spray Drying", in Journal of Agricultural and Food Chemistry, 2011, **59**, pp. 5523–5527.

[16]. L. M. C. Coc, I. Lacatusu, N. Badea, M. E. Barbinta-Patrascu, A. Meghea, "Effective lipid nanocarriers based on linseed oil for delivery of natural polyphenolic active", in Journal of Nanomaterials, 2021, 8853941, pp. 1-9.

[17]. I. Gulcin, "Antioxidant activity of caffeic acid (3,4-dihydroxycinnamic acid)", in Toxicology, **217**, 2006, pp. 213–220.

[18]. M. E. Barbinta-Patrascu, Y. Gorshkova, C. Ungureanu, N. Badea, G. Bokuchava, A. Lazea-Stoyanova, M. Bacalum, A. Zhigunov, S. Petrovic, "Characterization and antitumoral activity of biohybrids based on turmeric and silver/silver chloride nanoparticles", in Materials, **14**, 2021, pp. 4726-4751.

[19]. S. L. Smitha, K. M. Nissamudeen, D. Philip, K. G. Gopchandran, "Studies on surface plasmon resonance and photoluminescence of silver nanoparticles", in Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy, **71**, 2008, pp.186–190.

[20]. M. A. Ansari, H. M. Khan, A. A. Khan, A. Malik, A. Sultan, M. Shahid, F. Shujatullah, A. Azam, "Evaluation of antibacterial activity of silver nanoparticles against MSSA and MRSA on isolates from skin infections", in Biology and Medicine, **3**, 2011, pp. 141-146.

[21]. B. Bonev, J. Hooper, J. Parisot, "Principles of assessing bacterial susceptibility to antibiotics using the agar diffusion method", in Journal of Antimicrobial Chemotherapy, **61**, 2008, pp. 1295–1301.

[22]. A. Mohanty, "Physiochemical and antimicrobial study of polyherbal", in Pharmacie globale, **4**, 2010, pp. 1-3.

[23]. A. Belay, "Self-association, sodium ion complexation and optical transition probabilities of caffeic acid determined spectrophotometrically", in Journal of Biological Physics and Chemistry, **12**, 2012, pp. 23–28.

[24]. M. Moldoveanu, I. Rau, M. Fleancu, M. Mihaly, "Size effect at nanometric scale based on spectral measurements on water in oil microemulsions", in UPB Scientific Bulletin, Series B: Chemistry and Materials Science, **73**, 2011, pp. 41-54.

[25]. D. Paramelle, A. Sadovoy, S. Gorelik, P. Free, J. Hobley, D. G. Fernig, "A rapid method to estimate the concentration of citrate capped silver nanoparticles from UV-visible light spectra", Analyst, **139**, 2014, pp. 4855–4861.