EDITORA VERDE
Revista Verde de Agroecologia e Desenvolvimento Sustentável
Green Journal of Agroecology and Sustainable Development
GVAA GRUPO VERDE DE AGROECOLOGIA E ABELHAS

The influence of dispersive medium on efficient and eco-friendly synthesis of silver nanoparticles for application in antimicrobial fabric

Meio dispersivo na síntese eficiente de nanopartículas de prata para aplicação em tecidos antimicrobianos

Bianca Pizzorno Backx[1]; *Thais Delazare*[2]

[1]Professora e Doutora na Universidade Federal do Rio de Janeiro, Campus Duque de Caxias, Rio de Janeiro, 2679-2098, biapizzorno@xerem.ufrj.br; [2]Professora Doutora na Universidade Federal do Rio de Janeiro, Instituto de Química, Rio de Janeiro 3938-3700, thdelazare@iq.ufrj.br

A R T I G O
Recebido: 30/11/2018
Aprovado: 05/03/2019

Key words:
Nanostructures
Plant extract
Green synthesis
Melissa Officinalis Leaves
Smart textiles

A B S T R A C T

Silver nanoparticles have been increasingly used in industrial applications for a long time. Chemical methods of synthesizing silver nanoparticles can be toxic and expensive. Therefore, many studies have emerged to create ecologically correct synthetic routes that provide good dispersion and stability in the extractive medium. Plant species are investigated for the discovery of a new dispersive medium for silver nanoparticles (AgNPs) synthesized by the green route. AgNPs are disperse in plant extract, by plants available in Brazil. In this work, *Melissa Officinalis* leaves were used as a dispersive medium of nanoparticles. The characterization techniques used became possible to observe the superficial modifications with the presence of well-dispersed nanostructures, which are fundamental for the production of intelligent textiles with antimicrobial action. Thus, it was possible by means of each synthesis route to evaluate the dispersion and morphology of AgNPs in different dispersive media of *Melissa Officinalis*, concluding that it is very efficient for the stabilization of AgNPs.

Palavras-chave:
Nanoestruturas
Extrato vegetal
Síntese verde
Folhas de *Melissa Officinalis*
Têxteis inteligentes

R E S U M O

As nanopartículas de prata têm sido cada vez mais utilizadas em aplicações industriais há muito tempo. Os métodos químicos de síntese de nanopartículas de prata podem ser tóxicos e caros. Portanto, muitos estudos têm surgido para criar rotas de síntese ecologicamente corretas que forneçam boa dispersão e estabilidade no meio extrativo. Espécies vegetais são investigadas para a descoberta de um novo meio dispersivo para nanopartículas de prata (AgNPs) sintetizadas por via verde. As AgNPs estão dispersas em extrato vegetal, por plantas disponíveis no Brasil. Neste trabalho foram utilizadas folhas de *Melissa Officinalis* como meio dispersivo das nanopartículas. Através das técnicas de caracterização utilizadas foi possível observar as modificações superficiais com a presença de nanoestruturas bem dispersas, que são fundamentais para a confecção de tecidos inteligentes com ação antimicrobiana. Assim foi possível por meio de cada rota de síntese avaliar a dispersão e morfologia das AgNPs em diferentes meios dispersivos de *Melissa Officinalis*, concluindo-se que o mesmo é muito eficiente para estabilização das AgNPs.

INTRODUCTION

In the last decade, the study of nanometer-scale particles has grown exponentially around the world. Due to the vast field of applications of nanoparticles (NPs) new properties that are not found in micro and macro scale, can be observed at the nanoscale, such as temperature tolerance, color variety, changes in chemical reactivity and electric conductivity.

Supramolecular chemistry as well as quantum physics, take us to understand peculiar characteristics of nature, like the different forms of interactions of matter. That could bring out an extensive discussion about other ways to observe the world. Interaction forces is an essential field of science because studies based on this theory can be effective due to a precise justification of the different properties of matter.

In contemporary science, the already studied at the level of knowledge science is highly addressed to the fundamental relations of matter, its behavior in the face of adversities over time and how the nanostructure of that analyzed system is perceived according to these parameters in a different medium. Those points bring us out the concepts such as polymorphism, hysteresis, and supramolecular complexes. The understanding of the all-supramolecular interactions between nanostructure and the dispersive medium become an important field in nanoscience (BACKX AND ANTUNES FILHO, 2018).

The antimicrobial properties of silver it has long been known. With increasing surface area, AgNPs present an even more efficient antimicrobial action. The metallic NPs, more specifically, the silver nanoparticles (AgNPs) are fundamentally silver ions (Ag^+) reduced in aqueous solutions, forming Ag° atoms (BACKX et al., 2018). Different scientific results by observing the nanoscale phenomena of different forms become favorable to introduce new approaching in the science. That discovery and knowledge of science in its essence leads use to a better understanding of the action of the nanostructures in antimicrobial action.

Silver nanoparticles are beginning to be considered viable alternatives to antibiotics and may have a high potential for solving the problem of bacterial resistance (RAI et al., 2008). Studies of antibacterial activities by AgNPs were collected and reported by FRANCI et al. (2015) on 13 species of bacteria and biofilms, with emphasis on methicillin-resistant *Staphylococcus aureus* (MRSA), *Pseudomonas aeruginosa* with multidrug resistance, *Escherichia coli* and *Streptococcus pyogenes* resistant to erythromycin (LARA et al., 2010).

The use of plant extract may be advantageous compared to other methods, since the reaction rate for the synthesis is very high, allows easy production in large scale and does not use microorganisms, which require asepsis and maintenance costs (KEAT et al., 2015).

In green chemistry, it is known that vegetables produce several antioxidant molecules, which could act in the production of nanoparticles. Biomolecules such as amino acids, proteins, polysaccharides, in addition to secondary metabolites such as flavonoids, tannic acid, and terpenoids, can act both reducing the metal precursor, as well as stabilizing the nanoparticles, preventing their agglomeration.

Plants can act as antioxidants because of their various components (MITTAL et al., 2013). These include amino acids, flavonoids, phenolic compounds, terpenoids, citric acid, functional groups (alcohols, aldehydes, amines), heterocyclic compounds, dehydrogenases, intracellular CO2, membrane proteins, NADP reductases, peptides, polysaccharides, tannic acid (Akhtar et al., 2013). Some of these compounds can also stabilize NPs, covering the surface of the particles, limiting their growth (BACKX AND SANTANA, 2018). These can be found in several plants used as phytotherapics, and *Melissa Officinalis* leaves present high levels of these compounds (MIMICA-DUKIC et al., 2004).

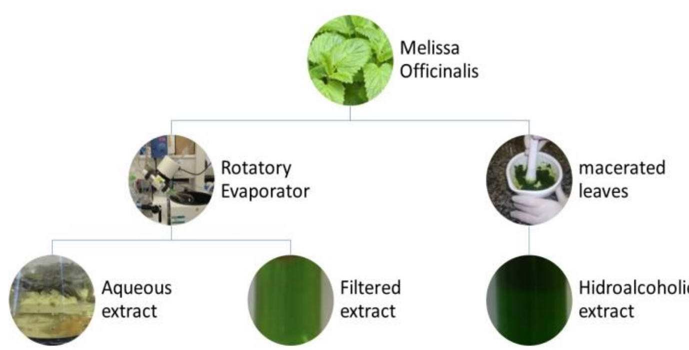
Supramolecular parameters can solely be to understand interactions of the fabric surfaces with AgNPs and the dispersive medium. This important approach is also useful to understand nanoscience sourcing modern science different forms of comprehending the nanoworld based on evolved interaction force to producing brand new materials, like smart fabrics, that has an antimicrobial activity, for example.

The main objective of this work is to evaluate the dispersive potential of AgNPs in *Melissa Officinalis* medium as the main stabilizing agent of these nanostructures to stimulate future studies concerning the antimicrobial potential of the plant extract conjugated to the AgNPs, ratifying the prominent influence of efficiency of dispersive medium to AgNps envolving supramolecular chemistry and green chemistry.

MATERIAL AND METHODS

The *Melissa Officinalis* leaves extraction were made in two ways (Figure 1). In the first one, the rotatory evaporator was used to extraction on aqueous solution. Leaves of the *Melissa Officinalis* were collected one day before use. The leaves were rinsed with Milli - Q water. The leaves were cut using scissors to get a smaller size. 5 g of the leaves pieces were carried in a 250 mL beaker with 200 mL Milli-Q and heated in rotatory evaporator. After 5 minutes of boiling, the extract was filtered using the filter with 3μm of porosity Brand Nalgon. For hydroalcoholic extract, 5 g of the *Mellisa Officinalis* leaves were macerated and added 40 mL of ethyl alcohol solution 40% (v/v). The heating occurred in microwaves for 1 min. The hydroalcoholic extract was filtered and then stored into an amber bottle to minimize the light influence and the evaporation of the ethanol.

Figure 1: Schematic of preparation of *Melissa Officinalis* leaves extraction to a dispersive medium of AgNPs.



Source: Authors (2019).

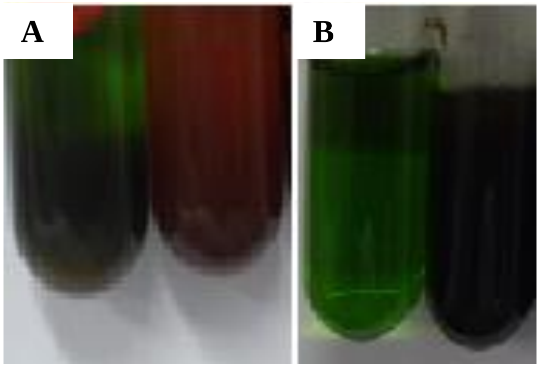
Synthesis of silver nanoparticles by green chemistry route

The synthesis of AgNPs was fulfilled by protocol adjusted to that studied by RAVEENDRAN et al. (2006), 200 μl of 0.1 M AgNO3 interacted with 500 μL of 0.1 M glucose under mechanical agitation. After that, 10.0 ml of starch solution was inserted into this solution and heated for 10 min to boiling in a heating plate. The reaction is represented below, in which, the aqueous medium is represented by (aq), solid and liquid physical state was represented by (s) and (l), respectively.

$$2AgNO_3(aq) + C_6H_{12}O_6(aq) + H_2O\ (l) \rightarrow 2Ag°(s) + 2HNO_3(aq) + C_6H_{12}O_7(aq)$$

1 mL of aqueous or hydroalcoholic of *Mellissa Officinalis* leaves were spiked of 50 µL AgNPs (Figure 2A and 2B), then stored at an amber bottle.

Figure 2: A) Aqueous extract (on the left) and aqueous extract with AgNPs spike (on the right). B) Hydroalcoholic extract (on the left) and hydroalcoholic extract with AgNPs spike (on the right).



It can be possible to observe that the final color of both solutions was modified to brown color, probably due to the formation of the AgNPs.

Ultraviolet–visible spectroscopy (UV-Vis)

The spectrophotometer, Thermo Evolution 60S, was used in the 300-1100 nm range using Milli-Q as white. The 600 µL of aqueous and hydroalcoholic extract with AgNPs were analyzed.

Nanoparticle Tracking Analysis

Nanosight is an equipment able to measure the size and the dispersion of nanoparticles and analyze the Brownian motion of the nanostructures. This equipment allows rapid and accurate analysis of the size distribution and concentration of all types of nanoparticles from 10 nm in diameter.

Characterization by Scanning Electron Microscope (SEM) and energy-dispersive spectroscopy (EDS)

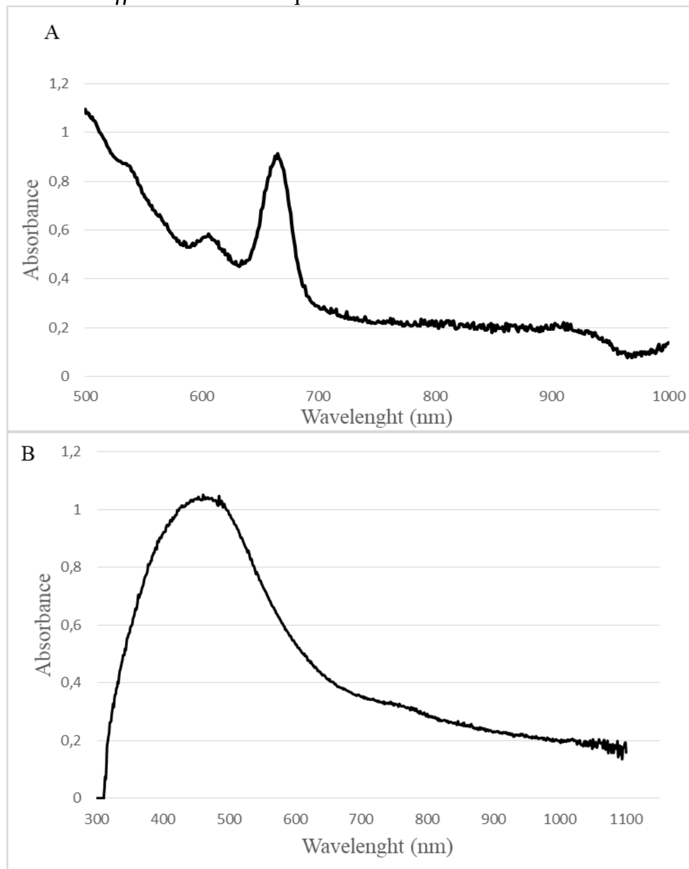
The microscopic characterization was fulfilled by VEGA 3 LMU MEV with a thermo EDX attachment. A piece of carbon adhesive tape (1cm x 1cm) was placed on top of each metal base; 250 µL of each sample was added on the surface of the carbon tape. The solution was evaporated by casting the samples were prepared to characterize the nanostructures obtained from green routes.

RESULTS AND DISCUSSION

For the hydroalcoholic extract, it was not possible to observe the colloidal peak of the AgNPs, located between 380 and 480 nm. Probably, there was a displacement of this peak (Figure 3A) it was possible to observe the peak located in the region around 660 nm. This may have occurred by the agglomeration or complexation of silver, reducing the activities of the nanoparticles, probably due to hydroalcoholic dispersive medium.

The position and peak intensities provide information about the nature of the phenolic compound and its substitution pattern. Probably the compounds extracted in alcohol influenced the dispersion and morphology of AgNPs.

Figure 3: UV-Vis spectrum of the AgNPs. A) AgNPs in *Melissa Officialis* leaves hidroalcoholic extract B) AgNPS in *Melissa Officialis* leaves aqueous extract



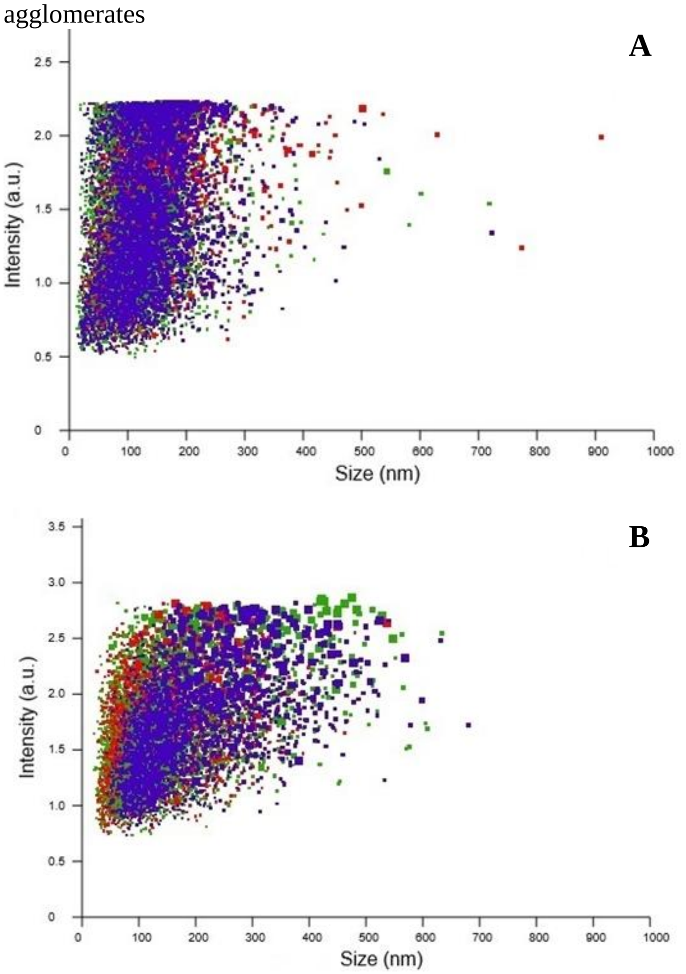
The solvent to be used in the preparation of the extract also has great relevance due to the influence of these solvents on the extraction of the leaves components. Different syntheses of AgNPs have been already tested with the variation in the solvents used for the preparation of the extracts (chloroform, hexane, butanol, ethyl acetate and water) and it was possible to observe that the aqueous fraction presented the most satisfactory results, preventing the extraction of the majority of lipids (KUMAR et al., 2014). It is possible to observe the characteristic colloidal peak can be revealed (Figure 3B) in the AgNPs dispersion in an aqueous extract. This result ratifies the efficient method of synthesis and dispersion of AgNPs in the aqueous extract of *Melissa Officinalis* leaves.

The displacement of silver nanoparticle SPR band was studied since 1989 (HENGLEIN, 1989). They accounted for observing this fact-based in a decrease of electron dispersion on the nanostructure surface upon chemic absorptions of Ag^{+} on the surface and the SPR band transition, in the visible region and the interband transitions electron of Ag in 4d to 5sp, generally occur in the region of 420 nm, become unavailable. The typical peak of silver nanoparticles in the colloidal state should be revealed between 380 and 480 nm (RAVEENDRAN et al., 2006 and PAL et al 2007). The SPR transition depends on the supramolecular interactions of the dispersion medium.

Through the results obtained it was verified that the peak of plasmon resonance, typical of the silver in the colloidal state, was obtained efficiently in the solution of the aqueous extract.

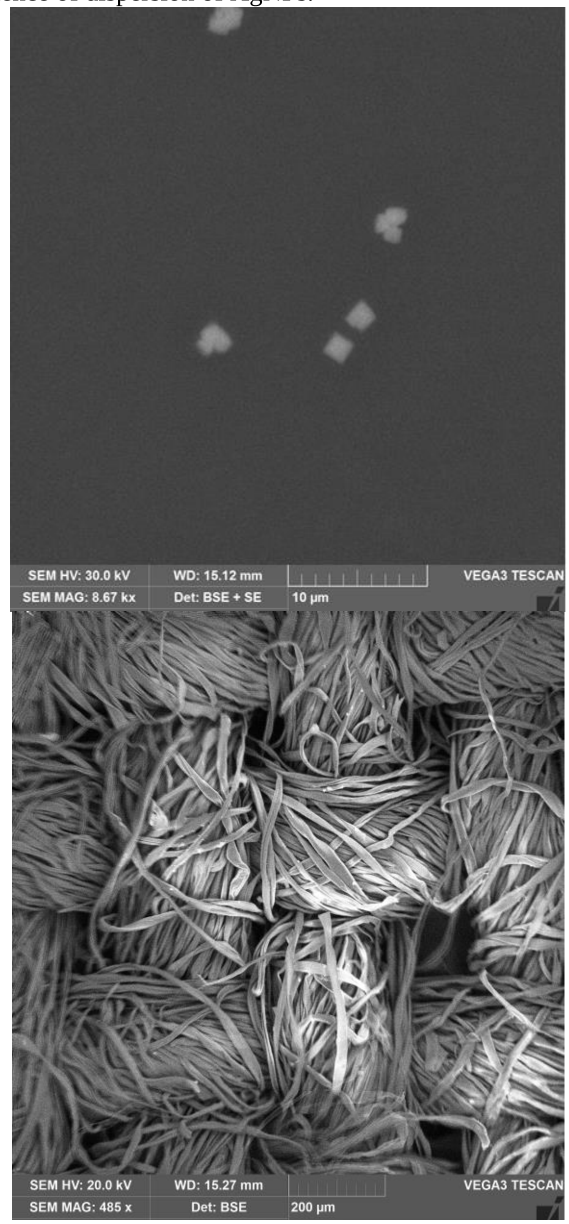
Silver nanoparticles synthesized by the green route were characterized by Nanosight equipment. The graph, in Figure 4 a, shows the aqueous extract of *Mellissa Officinalis* leaves with a spike of AgNPs, in which, most of the nanoparticles synthesized in the procedure are less than 100 nm. In Figure 4 b, it can be possible to observe that hydroalcoholic extract of *Mellissa Officinalis* leaves is not an efficient method of extraction when compared with aqueous extract, because, it was observed that the size of nanoparticles is bigger than observed on Figure 4 A, perhaps by the formation of possible agglomerates.

Figure 4: Nanosight graphic (Size x intensity). A) Nanoparticles around 100 nm dispersed on the aqueous extracts of the plant. B) Hydroalcohol extract with AgNPs, possible agglomerates



SEM micrographs revealed that the hydroalcoholic extract presented AgNPs tetrahedral structures in using a carbonaceous substrate (Figure 5 A). To observe the dispersion of the AgNPs in the textile, 500 µL of de AgNPs synthesized by hydroalcoholic extract was spiked on the substrate of 100% cotton fabric. Figure 5 b shows a low efficiency of dispersion of AgNPs in hydroalcoholic extract on the fibers of the tissue.

Figure 5: A) AgNPs in hydroalcoholic extract B) low efficience of dispersion of AgNPs.



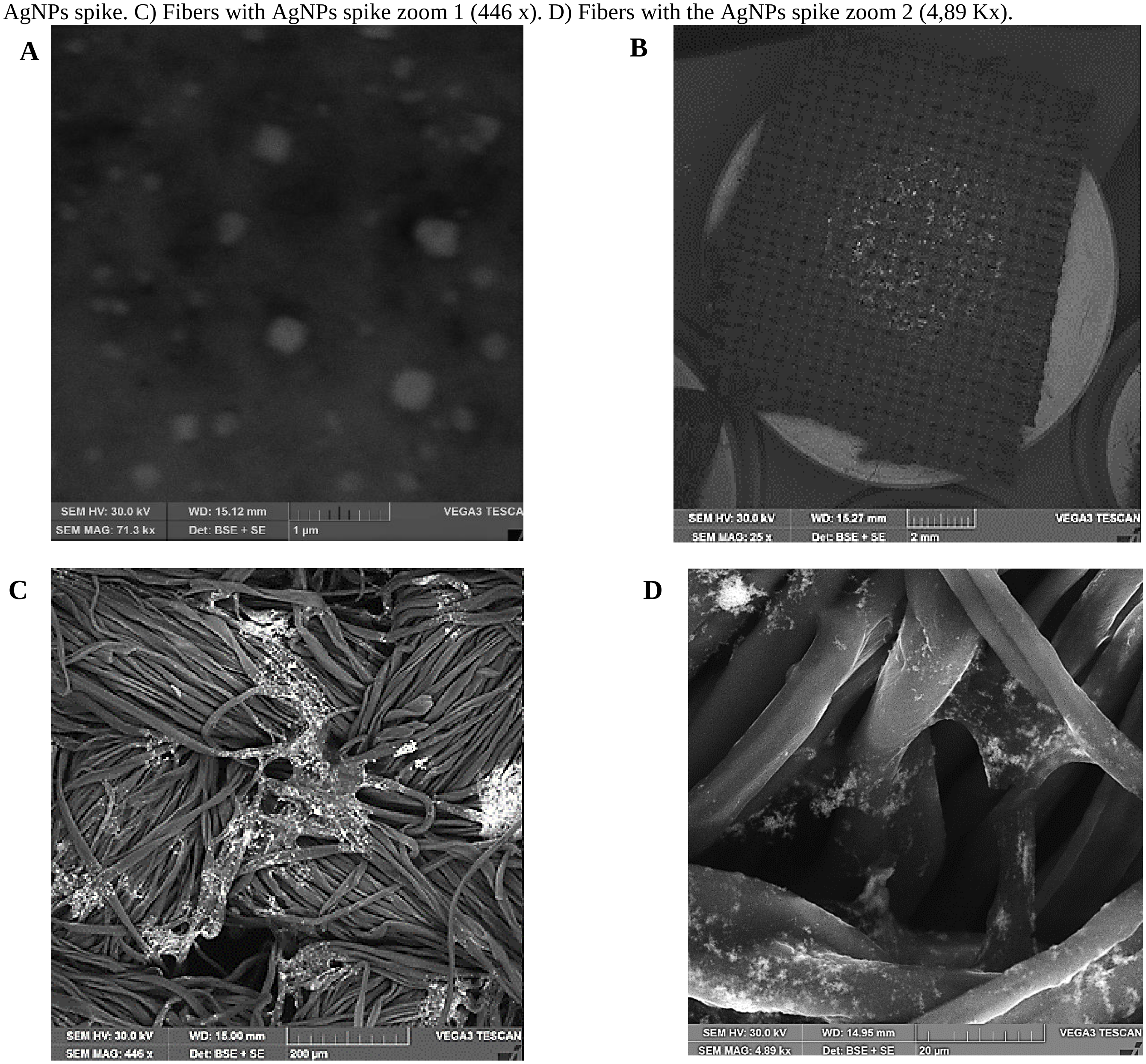
A

B

SEM analysis was fulfilled to confirm that the best dispersion of AgNPs occurred in aqueous extract. Initially, a micrograph of the AgNPs solution was made without the action of the dispersive aqueous extract medium comprising a carbonaceous substrate (Figure 6 A).

To verify the efficiency of the nanoparticles in the tissue, the posterior micrographs were made on the substrate of 100% cotton fabric. In Figure 6 B, 1cm x 1cm of 100% cotton fabric fibers was treated with 500 µL of the solution of AgNPs synthesized by starch and starch. It can be shown the impregnated fibers of the textiles with AgNPs. In Figure 6 C, the 446 x zoom of a region with them. Figure 6 D shows SEM micrographs of the tissue fibers with impregnation with AgNPs in a good dispersion thereof in *Melissa Officinalis* aqueous leaves extract, the 4,89 Kx zoom. From the EDX spectrum, it is evident that AgNPs were successfully synthesized, as can be shown in Figure 7.

Figure 6: SEM micrographs. A) AgNPs without dispersive medium. B) fibers of the fabric with leaves aqueous extract with AgNPs spike. C) Fibers with AgNPs spike zoom 1 (446 x). D) Fibers with the AgNPs spike zoom 2 (4,89 Kx).

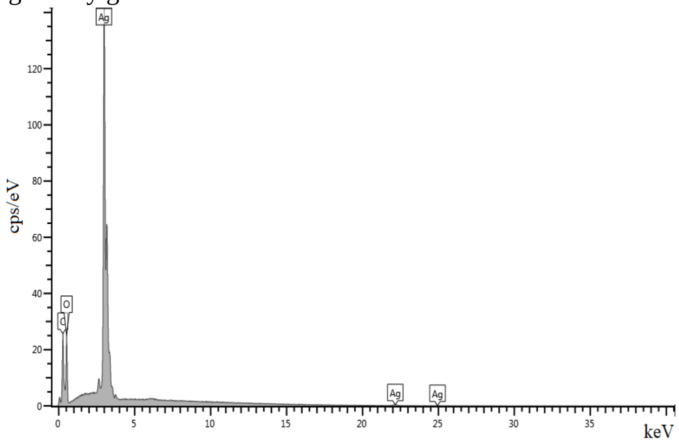


The SEM micrographs demonstrated that the AgNPs were synthesized by green synthesis (Figure 7) which was confirmed by EDX spectrum. It is evident that AgNPs were successfully synthesized, through the high intensity of silver peak, and it is possible to observe that the aqueous extract of *Melissa Officinalis* leaves can be used as an efficient dispersive medium for the AgNPs synthesized.

The leaves aqueous extract of *Melissa Officinalis* is efficient as AgNPs dispersive medium. The supramolecular interactions indicated that the aqueous dispersive medium is more efficient compared to the hydroalcoholic medium.

The characteristic shift of the SPR band in hydroalcoholic solution compare to silver nanoparticles in aqueous extract supports the formation of a stable silver nanoparticle. The shape of SPR band indicates the homogeneous distribution of AgNPs on the aqueous dispersive medium of *Melissa Officinalis* leaves. The typical peak, around 420 nm of the AgNPs was obtained by UV-Vis analysis. SEM micrograph, EDX spectrum, and Nanosight analysis allowed concluding that the aqueous extract of *Melissa Officinalis* leaves studied is a powerful dispersive medium for impregnation of AgNPs in the fabric fibers was efficient.

Figure 7: Energy-dispersive spectroscopy spectrum of the AgNPs by green rote.



CONCLUSION

The *Melissa Officinalis* leaves are as a feasible dispersing aqueous medium for the AgNPs stabilization synthesized by green route with starch and glucose. This result is very optimistic because AgNPs has very important microbicidal properties. This action allied to a green dispersive medium, without toxic waste for the environment, brings the hope of associating the advances of nanomaterials with the environment. Future studies will continue to investigate other parameters related to the final product, an intelligent antimicrobial fabric.

REFERENCES

AKHTAR, B. S.; PANWAR, J.; YUN, Y.S. Biogenic synthesis of metallic nanoparticles by plant extracts. ACS sustainable Chemistry and Engineering, v. 1, p. 591-602, 2013.

BACKX, B. P.; ANTUNES FILHO, S. Green Dispersive Systems and the Formation of Micro- and Nanostructured Multiphase in Leaves Extract from *Psidium guajava L*. SciFed Nanotec Res Let 2:2, 2018.

BACKX, B. P.; PEDROSA, B. R.; DELAZARE, T.; DAMASCENO, F.; SANTOS, O. A .L. Green Synthesis of Silver Nanoparticles: A Study of the Dispersive Efficiency and Antimicrobial Potential of the Extracts of *Plinia Cauliflora* for Application in Smart Textiles Materials for Healthcare. J Nanomater Mol Nanotechnol, v. 7, n.1, p., 2018.

BACKX, B. P.; SANTANA, J. C. S. Green Synthesis of Polymer Blend impregnated with silver nanoparticles in *Euterpe Oleracea* dispersive medium. International Journal of Green and Herbal Chemistry, Sec. A; v.7, No.2, p. 424-429, 2018.

FRANCI, G.; FALANGA, A.; GALDIERO S.; PALOMBA, L. Silver nanoparticles as potential antibacterial agentes Molecules. Molecules, 20(5), p. 8856-8874, 2015.

HENGLEIN, A. Small-particle research: physicochemical properties of extremely small colloidal metal and semiconductor particles. Chemical Reviews, v. 89, n. 8, p. 1861–1873, 1989.

KEAT, C. L.; AZIZ, A.; EID, A.; ELMARZUGI, N. Biosynthesis of nanoparticles and silver nanoparticles. Bioresources and Bioprocessing, v.2, n. 1, p. 47, 2015.

KUMAR P. P. N.; PAMMI, S. V. N.; KOLLU, P.; SATYANARAYANA, K. V. V.; SHAMEEM, U. Green synthesis and characterization of silver nanoparticles using *Boerhaavia diffusa* plant extract and their antibacterial activity. Industrial Crops and Products, v. 52, p. 562-566, 2014.

LARA, H. H.; AYALA N.; TURRENT, L.; PADILLA, C.R. Mode of antiviral action of silver nanoparticles against HIV-1, Journal of Nanobiotechnology, 8:1, 2010.

MIMICA-DUKIC, N.; BOZIN, B.; SOKOVIC, M.; SIMIN, N. Antimicrobial and antioxidant activities of *Mentha officinalis* L. (Lamiaceae) essential oil. J. Agric. Food Chem., 52(9), p. 2485-2489, 2004.

MITTAL, A. K.; CHISTI, Y.; BANERJEE, U. C. Synthesis of metallic nanoparticles using plant extracts. Biotechnology advances, v. 31, n. 2, p. 346-356, 2013.

PAL, S.; TAK, Y. K.; SONG, J. M. 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, v.73, p. 1712-1720, 2007.

RAI, M.; YADAV, A.; GADE, A. Silver nanoparticles as a new generation of antimicrobials. Biotechnology advances, p.76-82, 2008.

RAVEENDRAN, P.; FU, J.; WALLEN, S. L. A simple and green method for the synthesis of Au, Ag, and Au–Ag alloy nanoparticles. Green Chemistry, v. 8, p. 34-38, 2006.