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# Synthesis of silver nanoparticles using aqueous extracts of *Heterotheca inuloides* as reducing agent and natural fibers as templates: *Agave lechuguilla* and silk



Raúl.A. Morales-Luckie<sup>a,\*</sup>, Aldo Adrián Lopezfuentes-Ruiz<sup>a</sup>, Oscar F. Olea-Mejía<sup>a</sup>, Argueta-Figueroa Liliana<sup>a</sup>, Víctor Sanchez-Mendieta<sup>b</sup>, Witold Brostow<sup>c</sup>, Juan P. Hinestroza<sup>d</sup>

<sup>a</sup> Universidad Autónoma del Estado de México, Centro Conjunto de Investigación en Quínica Sustentable UAEM-UNAM, Carretera Toluca-Atlacomulco Km 14.5, San Cayetano, 50200 Toluca, Estado de México, Mexico

<sup>b</sup> Facultad de Química, Universidad Autónoma del Estado de México. Paseo Colón y Paseo Tollocan, Toluca, Estado de México 50120, Mexico

<sup>c</sup> Laboratory of Advanced Polymers & Optimized Materials (LAPOM), Department of Materials Science and Engineering, University of North Texas, 1150 Union Circle #305310, Denton, TX, United States

<sup>d</sup> Department of Fiber Science and Apparel Design, Cornell University, 242 Van Rensselaer Hall, Ithaca, New York 14853-4203, United States

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### ABSTRACT

Silver nanoparticles (Ag NPs) were synthesized using a one-pot green methodology with aqueous extract of *Heterotheca inuloides* as a reducing agent, and the support of natural fibers: *Agave lechuguilla* and silk. UV–Vis spectroscopy, X-Ray photoelectron spectroscopy XPS and transmission electron microscopy TEM were used to characterize the resulting bionanocomposite fibers. The average size of the Ag NPs was 16 nm and they exhibited low polydispersity. XPS studies revealed the presence of only metallic Ag in the nanoparticles embedded in *Agave. lechuguilla* fibers. Significant antibacterial activities against gram-negative *Escherichia coli* and gram-positive *Staphylococcus aureus* were determined. AgO as well as metallic Ag phases were detected when silk threads were used as a substrates hinting at the active role of substrate during the nucleation and growth of Ag NPs. These bionanocomposites have excellent mechanical properties in tension which in addition to the antibacterial properties indicate the potential use of these modified natural fibers in surgical and biomedical applications.

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# 1. Introduction

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Replacement of artificial fibers by natural fibers is encouraged for several reasons including a more responsible use of resources, the manufacturing of biodegradable products, and the potential reduction of production costs [1]. Industrial natural fibers are attractive due to their low environmental impact, renewability, biodegradability, relative low cost, lightness, carbon dioxide neutrality, acoustic and thermal insulation properties [2,3]. According to the 2020 Technology Road Map for Plant/Crop Based Renewable Resources, authored by the US Department of Energy (DOE), the application of plant derived renewable resources may increase to 10% as basic chemical building blocks by 2020, and by 50% by 2050 [4]. In fact, the insertion of natural fibers in the industrial, building and commercial markets has experienced a compounded growth rate of 13% over the last 10 years [5].

The use of biocomposites is expected to improve manufacturing and recycling of products with enhanced environmental compatibility [6].

\* Corresponding author.

*E-mail addresses*: rmoralesl@uaemex.mx, ramluckie@gmail.com (R.A. Morales-Luckie).

The biodegradability of biocomposites is due to the existence of microorganisms with specific enzyme systems capable of hydrolyzing carbohydrate polymers from the cell wall into digestible units.

It is well known that the properties of nanocomposite materials depend not only on the properties of their individual components but also on the morphological and interfacial characteristics between nanomaterials and substrates [7]. It has been demonstrated that natural cellulose fibers can be used as supports for nanoparticle synthesis using *in situ* approaches and electrostatic assembly methods [8]. However, most literature reports on natural fiber's surface modification with nanoparticles refer to cotton and few references are made to other natural fibers. The use of unconventional natural fibers as solid supports for metal nanoparticles synthesis is attractive as they exhibit heterogeneous and oxygen-rich surface structure [9]. These unique surface properties can result in diverse nanoparticle shapes, sizes and distributions that could confer, due to the shape- and size-dependent properties of the nanoparticles, varied macroscopic properties such as color, conductivity and improved mechanical resistance to the modified fibers [10].

Silver nanoparticles (Ag NPs) have been extensively studied because of their use in antimicrobial gels [11], preservation of fruits and vegetables [12], healing materials [13], and antiseptic sprays [14]. There are several methods of synthesize nanoparticles including laser ablation [15], chemical reduction [16], mechanical synthesis, and biological methods. In recent years, the bio-reduction of noble metal ions with plant extracts to obtain nanoparticles has been gained importance [15, 17–19]. The use of bioreductors does not require expensive equipment or chemical reactants while simple methods follow the principles of green chemistry- which refers to the design of products and processes friendly to the environment [20].

*Heterotheca inuloides* is a medicinal plant that had been used for centuries to heal skin wounds and bruises, as it exhibits anti-inflammatory properties. Several studies have shown that these properties are given by its main compounds: cadalen-15-oic acid, 3,7-dihydroxy-3isocadalen-4-one, and dicadalenol, which found in the aerial parts of *Heterotheca inuloides* [21].

When a wound is sutured there is a risk of infection [22] which not only delays the healing process but also can cause bigger damages to the wound. When suture threads are used, patients are required to take external antibiotic drugs to avoid infection; however, many of these drugs show undesirable secondary effects [23]. A suture thread that could reduce the risk of infection would also minimize the need to take external antibiotic drugs.

In this work, *Heterotheca inuloides*, also known as *Mexican arnica*, is used for the first time as a bio-reducing agent of silver ions to generate Ag NPs on two natural fibers: *Agave Lechuguilla* and silk. Structural and morphological characterizations, mechanical properties, and their capability to inhibit bacteria growth, were determined for both natural fibers.

# 2. Materials and methods

#### 2.1. Materials

All reagents used were analytical grade. AgNO<sub>3</sub> was purchased from Sigma-Aldrich. *Heterotheca inuloides (Mexican arnica)* was purchased from Anahuac Mexican teas, 99.90% of purity. *Agave lechuguilla* fibers were obtained directly from agave producers in the State of Mexico. Commercial suture threads of silk with regular tapered point, black, braided and non-absorbable were obtained from Atramat, Inc. (Mexico).

### 2.2. Agave lechuguilla fiber preparation

Fibers were boiled in deionized water for 5 min to remove impurities. Because the fibers do not present a uniform diameter along their length, they were cut to eliminate the heart base and the tapering towards the tip to achieve a uniform diameter (400  $\mu$ m approximately) along 40 cm of length. Additionally, some of these fibers were treated with methylene blue (MB) to soften them and improve their handling for suture applications.

#### 2.3. Synthesis of silver nanoparticles on Agave lechuguilla and silk

1 g of *Heterotheca inuloides* dry leaves was poured into 100 ml of deionized water. The mixture was boiled for 20 min, vacuum filtered and let to cool at room temperature.  $1 \times 10^{-2}$ ,  $5 \times 10^{-3}$ ,  $1 \times 10^{-3}$ ,  $1 \times 10^{-4}$  M AgNO<sub>3</sub> solutions were prepared. *Agave lechuguilla* fibers were submerged inside these solutions, at 1, 10, 30 and 60 min and, after the set immersion time, the fibers vacuum filtered. The fibers impregnated with Ag<sup>1+</sup> ions were soaked in solutions of *Mexican arnica* at room temperature during different times: 1, 10, 30 and 60 min. Fibers were removed and dried at room temperature.

#### 2.4. Characterization of the silver nanoparticles

UV–Vis spectroscopy was performed in a Cary 5000 UV–Vis Spectrophotometer using a quartz cell and the wavelength range was from 300 to 600 nm. Scanning electron microscopy (SEM) and Energy dispersive spectroscopy (EDS) analysis were done in a JSM-6510-LV microscope (JEOL) at 20 kV of acceleration and using secondary electrons.

The samples were coated with a thin film of gold (approx 20 nm) using a Denton Vacumm DESK IV sputtering equipment. Transmission electron microscopy (TEM) was carried on in a JEOL-2100 Microscope (JEOL) at 200 kV of acceleration in the bright field mode. In order to prepare the samples for TEM, the specimens were sonicated during 4 h to detach the nanoparticles from the fibers; Then a drop of the suspension was carefully placed on a carbon coated TEM grid. Thermogravimetric analysis were done on a SDT Q600 from TA Instruments in a nitrogen atmosphere at a heating rate of 10 °C min<sup>-1</sup> from 25 to 600 °C.

TGA and DSC analysis for pyrolysis and combustion were performed at 10 °C/min heating rate. Infrared spectra of the relevant materials have been obtained using a Bruker FTIR spectrometer (IFS 113v) for wavelengths between 2 and 200  $\mu$ m (5000–500 cm<sup>-1</sup>).

XPS wide and narrow spectra were acquired using a JEOL JPS-9200, equipped with a Mg X-ray source (hv = 1253.6 eV) at 200 W over an area of 1 mm<sup>2</sup>. The spectra was analyzed using the specsurf<sup>TM</sup> software and the spectra were corrected by means of the carbon signal (C1s) at 284.5 eV. The Shirley method was used for background subtraction, whereas curve fitting was done with the Gauss-Lorentz method.

#### 2.5. Antibacterial activity

The antimicrobial activity was assessed using procedures from the Clinical and Laboratory Standards Institute [22]. For culturing *Staphylococcus aureus* and *Escherichia coli*, mannitol salt agar [23] and eosin methylene blue agar [24], were used respectively. Antimicrobial activity of the synthesized Ag NPs on *Agave lechuguilla* and silk was tested against human pathogenic bacteria *Staphylococcus aureus* and *Escherichia coli* by determining the halo of inhibition following the agar diffusion test using Muller-Hinton agar.

Agar plates were prepared and inoculated with 200  $\mu$ l of bacterial culture. The culture was adjusted with sterile saline to achieve a turbidity equivalent to a 0.5 McFarland standard. *Agave lechugilla* and silk fibers with and without Ag nanoparticles were firmly placed on the agar plates. Tests were performed three times for each strain. The inoculated petri dishes were incubated at 37 °C for 24 h, and then, the antibacterial halos were observed over the agar plates.

#### 2.6. Mechanical properties

10 fiber samples of *Agave lechuguilla* and *silk* were selected for mechanical testing at an initial deformation speed of 50 mm/s using an universal testing machine MTS Qtest/5. For statistical analysis, the descriptive analysis including mean and standard deviation was performed. For significant differences between the groups, Kruskal-Wallis test and Mann-Whitney U tests were used. Correlations between elastic modulus and stress at break were determined using Spearman correlation analysis. Statistical analysis was performed using SPSS Version 19 (SPSS, Inc., Chicago, IL, USA). *P*-values of  $\leq 0.05$  were considered to indicate statistical significance.

#### 3. Results

In order to find the best conditions for obtaining Ag nanoparticles well dispersed throughout the surface of the fibers, without agglomeration and in a sufficient amount to deliver antibacterial properties, the effect of each of the the synthesis parameters was judiciously explored. It was found that the optimum silver ions impregnation time was 30 min. At shorter impregnation times, silver was not detected on the surface of the fibers, while, at longer times big agglomreations of AgNP were found.

Regarding  $AgNO_3$  concentration, the optimum concentration was found to be  $10\times10^{-3}$  M. A higher concentration would yield big

micrometric aggregates, while smaller concentrations would lead to a scarce amount of surface silver particles on the surface of the fibers. It was also found that 10 min of reduction time was enough to reduce all the silver ions and that longer times did not result in higher amounts of Ag on the fiber's surface. Finally, the most suitable bioreductor concentration was 1 mg/100 ml. In order to verify the effectiveness of *Heterotheca inuloides* as a bio-reducing agent, the plant infusion was mixed with AgNO<sub>3</sub> in solution for 10 min at ambient conditions. The solution's color changed from light yellow to light brown as the reduction of silver ions to nanoparticles took place. The color change was confirmed by UV–Vis spectroscopy as shown in Fig. 1.

Fig. 2 shows SEM images of Ag NPs, synthesized with *Heterotheca inuloides* as bio-reducing agent. Fig. 2(a) shows a pristine silk thread and in Fig. 2 (b) Ag nanoparticles of spherical in shape are observed distributed over the fiber's surface. Fig. 2 (c) and (d) shows a SEM micrograph on *Agave lechuguilla fibers* similar in diameter to those of silk. Fig. 2 (e) shows the elemental mapping of *Agave lechuguilla* fiber.

Ag NPs were synthesized in both types of fibers using NaBH<sub>4</sub>, a wellknown reducer, for control purposes. In Fig. 3 (a) and (b) silk fibers show particle agglomerates. In Fig. 3 (c) and (d) *lechuguilla agave* fibers also show agglomerates on the surface of the fibers. In Fig. 3 (e) the elemental mapping confirms the presence of agglomerates.

Fig. 4(a) and (b) are TEM images showing that the shape of the Ag NPs tends to be spherical. (c) HRTEM shows an interplanares distance of 0.232 nm which correspond to plane (111) of Ag NP.

Fig. 5 shows TGA and DSC plots for pyrolysis and combustion at 10 °C/min heating rate. As seen in Fig. 5 (a) and (b), the DSC curve nearly matches the first weight of loss in the TGA curve. Note that the first weight loss in the TGA curve is around 100 °C corresponding to the loss of water. While in the TGA analysis the differences between the silk fibers with and without Ag NPs are also minimal, since the temperature for its pyrolysis occurs 314 °C and 312 °C, respectively as is shown in Fig. 5(a) and (b); however in 5 (b) a deeper peak is observed, which could correspond to the presence of metal. In the same manner, this endothermic and deeper peak is observed in the Fig. 5 (d) compared to (c). In regard to the TGA analysis, the differences between the *Agave lechuguilla* fibers with and without Ag NPs are minimal, since the

temperature for its pyrolysis occurs at 279.39 °C and 284.08 °C, respectively as is shown in Fig. 5(c) and (d).

Functional groups of silk and *Agave lechuguilla* fibers appear unaltered before and after the reduction of Ag nanoparticles - as shown in IR-spectra, see Supplementary material.

EDS studies are not enough to assure that only Ag metallic nanoparticles are present. XPS was used to elucidate the oxidation state of silver on the fibers' surface as shown in Fig. 6.

For the Agave lechuguilla specimens the AgO signal was not observed and only the metallic corresponding peaks are shown. By deconvoluting the Ag peak in the high resolution XPS spectrum of the Ag NPs synthesized over silk, it was found that silver oxide (AgO) was present with an abundance of about 14% compared to that of the Ag metal peak.

Two types of bacteria were tested in this study: gram-positive *Staphylococcus aureus* and gram-negative *Escherichia coli*. Functional composites of *Agave lechuguilla* and silk fibers bearing Ag nanoparticles were found to inhibit the growth of both types of bacteria, as shown in Fig. 7; the halos around the fibers are approximately three times the width of the fibers.

In the Fig. 8, the silk fiber with nanoparticles(b) shows a reduction of the elastic modulus of 25% respect to the silk fiber without nanoparticles(a). However, the *Agave lechuguilla* fibers both with Ag nanoparticles(d) and (e) show only a decrease of 5% and 9% in the elastic modulus. Also, Fig. 8 shows a reduction of the stress at break when Ag nanoparticles are adhered to the fibers. In silk fibers, this reduction is not substantial (only 2%) but for pure *Agave lechuguilla* fibers(c) the reduction was 53%. Statistical analysis shows that fiber (a) has significant higher stress at break (Z = -3.141, n = 10 per group, p = 0.002) and higher elastic modulus (Z = -3.780, n = 10 per group,  $p \le 0.0001$ ) than the fiber (c); as expected, pure silk has itself better mechanical properties than pure lechuguilla fiber.

As expected from the above analysis, the differences across all five groups for elastic modulus are significant (Kruskal-Wallis test, H = 44.321, df = 6, n = 10 per group,  $p \le 0.0001$ ). On the other hand, Mann-Whitney U tests confirm that the fiber (a) has a significantly higher elastic modulus than the fiber (b) (Z = -3.781, n = 10 per group,  $p \le 0.0001$ ). As well, Mann-Whitney U tests confirm that the fiber (c) has a significantly higher elastic modulus than the fiber (d) the fiber (d) has a significantly higher elastic modulus than the fiber (d) the fiber (d) has a significantly higher elastic modulus than the fiber (d) has a significantly higher elastic modulus than the fiber (d) has a significantly higher elastic modulus than the fiber (d)



Fig. 1. (a) UV-Vis spectra of Ag NPs obtained with *Heterotheca inuloides* as bio-reducing agent. Images of vials contain the bio-reducing agent infusion (b) without Ag NPs and (c) containing the bio-reducing agent infusion with Ag NPs.



Fig. 2. (a) and (b) SEM images of silk fiber with Ag NPs reduced with *Heterotheca inuloides* at different magnifications. (c) and (d) SEM images of *Agave lechuguilla* fiber with Ag NPs reduced with *Heterotheca inuloides* at different magnifications. (e) Element mapping of *Agave lechuguilla* fiber coated with Ag NPs.

(Z = -3.480, n = 10 per group, p = 0.001). However, Mann-Whitney U tests confirm that fiber (d) has no significant difference in the elastic modulus compared with fiber (e) (Z = -1.364, n = 10 per group, p = 0.173), meaning that the pretreatment with MB has not improved the elastic modulus of lechuguilla fiber with Ag NPs.

As it was expected after performing the first analysis, the differences across all five groups for stress at break are significant (Kruskal-Wallis test, H = 43.915, df = 4, n = 10 per group, p = 0.026). On the other hand, Mann-Whitney U tests confirm that the fiber (a) has a significantly higher stress at break than the fiber (b) (Z = -2.309, n = 10 per group, p = 0.021). As well, Mann-Whitney U tests confirm that the fiber (d) (Z = -3.782, n = 10 per group,  $p \le 0.0001$ ). However, Mann-Whitney U tests confirm that the fiber (d) (Z = -3.782, n = 10 per group,  $p \le 0.0001$ ). However, Mann-Whitney U tests confirm that the fiber (e) has a significantly higher stress at break than the fiber (d) (Z = -3.782, n = 10 per group,  $p \le 0.0001$ ).

break than the fiber (d) (Z = -3.781-, n = 10 per group,  $p \le 0.0001$ ), meaning that the pretreatment with MB is improved the stress at break of lechuguilla fiber with Ag NPs.

Besides, it was found correlation between the elastic modulus and stress at break by Spearman correlation analysis (Spearman's rank correlation coefficient = 0.870, p = 0.01). That result is in accordance with the congruent expected data from the both mechanical tests.

# 4. Discussion

Aramwit et al. [25] performed a study to generate silver nanoparticles using a similar green synthesis approach, obtaining antibacterial properties. By contrast, in this present study, the synthesis on the fiber itself was carried out. Moreover, its main finding was to investigate



Fig. 3. (a) and (b) SEM images of silk fiber with Ag NPs reduced with NaBH<sub>4</sub> at different magnifications. (c) and (d) SEM images of Agave lechuguilla fiber with Ag NPs reduced with NaBH<sub>4</sub> at different magnifications. (e) Element mapping of Agave lechuguilla fiber with Ag NPs.



Fig. 4. (a) and (b) TEM images of Ag NPs, at different magnifications. (c)HRTEM of Ag NPs, and (d) Size histogram of Ag NPs.

the antibacterial and others properties of natural fibers using different green technology. We used for the first time *Heterotheca inuloides* as reducing agent.

In Uv–vis spectroscopy, after 1 h of reduction, there is an incipient reduction and the plasmon peak is not observed. After 2 h of reduction, the beginning of the nanoparticle formation is noted as a wide Plasmon peak with low absorbance is noted. At 4 h of reduction, the plasmon peak is clearly observed at 451 nm indicating the formation of Ag NPs. At 6 h of reduction, a sharper peak is noted as well as a red shift of a few nanometers indicating an increase in size of the nanoparticles [17].

The SEM images of silk fiber and *Agave lechuguilla* fiber and their element mapping of the latter evidently show the almost homogeneous distribution of nanoparticles in both fibers. In Fig. 2 (d) a larger amount of nanoparticles on the surface of the fibers is observed and these nanoparticles exhibit a more homogenous size than those shown in Fig. 2 (b). We attribute this fact to the structure of the cellulosic fiber, which has many ester and hydroxyl groups, and those chemical groups work as attractors of positive ions of silver.

Based on these direct observations (Fig. 3), it is possible to affirm that there is an increased formation of nanoparticles when  $NaBH_4$  is used as reducer but unfortunately large agglomerates are present on the surfaces of both fibers.

According to the composition of *Heterotheca inuloides* [21], the main molecules responsible for the bio-reduction are those with phenolic



Fig. 5. TGA and DSC curves of (a)silk fiber (b)silk fiber/Ag NPs (c)Agave lechiguilla fiber (d) Agave lechiguilla fiber/Ag NPs.



Fig. 6. (a) High resolution XPS of Ag 3d<sub>5/2</sub>of Ag NPs on Agave lechuguilla fiber. (b) High resolution XPS spectrum of Ag 3d<sub>5/2</sub> of Ag NPs on silk suture thread.

groups, which act similarly than other plant's bio-reduction systems [26]. These molecules are also responsible for the stabilization of Ag NPs leading to narrow size dispersion and non-agglomerated particles, as confirmed by TEM. As far as our best knowledge, before us no reports have been found on Heterotheca inuloides a reducing agent for nanoparticles. Until now, it has not been clearly identified the compound or compounds with reducing activity of Heterotheca inuloides. However, Haraguchi et al. [27], concluded in their study that the sesquiterpenoids, 7-hydroxy-3,4-dihydrocadalin and 7-hydroxycadalin, and flavonoids, cathechine, quercetin, kaempferol and their glycosides, isolated from Heterotheca inuloides showed potent antioxidative activity. This gave to us the prospect to use this plant as a reducing agent; we hypothesized that such active components on Heterotheca inuloides can generate silver nanoparticles since it is well-known that the antioxidants from plants can be reducing agents [28]. In addition, it has been recognized by Delgado, et al. [22], that cathechine and quercetin are the main compounds in Heterotheca inuloides. Coballase-Urrutia, et al. [29], reported the potent concentration-dependent antioxidant effects against reactive oxygen species produced *in vitro*, and they suggest that action mechanism of the polyphenols due to scavenging capabilities associated with their hydroxyl groups and their chelation capacity transition metal. Soobrattee, et al. [30], concluded that structural characteristics of flavonoids such as quercetin are important for their activities as free radical scavengers, their chemical structure including two diphenolic groups each in the A and C rings.

Besides, the mean particle size was 16 nm, which is important to maintain antibacterial properties [31]. The size, size distribution and shape of Ag NPs synthesized on *Agave lechuguilla* and on silk were similar.

TGA is finding a great utility in investigations of the pyrolysis and combustion behavior of materials. Among materials, cellulose is extensively studied. One of the utilities of this analytical procedure is detecting small experimental deviations which may be introduced by amounts of impurities in the sample [32], due to this, the data collected in this study is valuable since there is a limited data about its behavior when cellulose is doped with metal nanoparticles. In the TGA for



**Fig. 7.** (a) *Agave lechuguilla* fiber with Ag NPs, in agar Muller-Hinton and *S. aureus* (gram +). (b) *Agave lechuguilla* fiber with Ag NPs in Eosin methylene blue agar with *E. coli* (gram -). (c) Silk suture thread with Ag NPs, in Eosin methylene blue agar with *S. aureus* (gram +). (d). Silk suture thread with Ag NPs, in Eosin methylene blue agar with *E. coli* (gram -).



Fig. 8. Mechanical properties of the fibers with and without Ag NPs. (a) Pure silk. (b) Silk with Ag NPs. (c) Pure Agave lechuguilla. (d) Agave lechuguilla with Ag NPs. (e) Treated MB Agave lechuguilla with Ag NPs.

cellulose the pyrolysis occurs at 344 °C [32], the results in the present study are similar in the silk fibers without Ag NPs, considering the silk fibers are mainly constituted of cellulose. Note that minimal changes in the temperatures for pyrolysis are indicated the thermal stability of both silk and *Agave lechuguilla* fibers. Therefore, it is possible to assume that both fibers, doped with Ag NPs using this *in situ* method are not affected in their thermal stability.

From the XPS spectrum is not possible to know if there are metal oxide particles as well as metallic Ag particles or there are metallic particles covered with a thin layer of oxide, or both situations co-exist. It is possible that the oxide phase appeared due to the presence of certain proteins that can lead to a partial oxidation of the Ag when supported on silk fibers [33]. This behavior is not observed in *Agave lechuguilla* fiber, as it is mostly composed of nitrogen-free cellulose [34].

It is also observed that the inhibitory halo in silk is smaller than the halo exhibited for *Agave lechuguilla*. This lower antibacterial activity of the silk fibers can be due to the presence of AgO. The general antibacterial mechanism of silver is by cell wall breakdown and inhibition of intracellular enzyme activity [19]. In solution, the antibacterial behavior of Ag nanoparticles arises from the direct contact bacteria-nanoparticle; however, when nanoparticles are attached to a natural fiber, the inhibitory halo appears because of the releasing of Ag ions on the agar medium [35].

Since these fibers could be potentially used as suture threads, it is important to study their mechanical properties before and after the Ag nanoparticles are impregnated. In regards to *Agave lechuguilla* fibers, several factors can influence their mechanical properties as these fibers are boiled to remove lignin and prolonged boiling may lead to degradation of chains of cellulose and hemicellulose of the fibers. It is suggested in present study that the *Agave lechuguilla* fiber treatment with methylene blue (MB), described before, could be a useful method for improvement of the natural *Agave lechuguilla* fiber composites while retaining their desirable mechanical properties. Only *Agave lechuguilla* fibers were treated with MB. MB is used as redox indicator because is capable of releasing oxygen from other molecules, especially on their surfaces. It is hypothesized that the *Agave lechuguilla* fiber treated with MB (in aqueous solution) should be negatively charged. This could generate an attraction between Ag<sup>+1</sup> ions and the fiber.

In the present study, the elastic modulus for all specimens decreases when nanoparticles are incorporated and the incorporation of Ag NPs affects more drastically silk fibers than the *Agave lechuguilla* ones. Silk fibers have a protein structure which is more affected by the presence of water while the *Agave lechuguilla* fibers are mostly composed of cellulose chains. Another important mechanical property for these materials is the stress at break, which is the maximum load that the thread can bear before rupture. In silk fibers, this reduction is really low but in *Agave lechuguilla* fibers the reduction was significant. Due to this, further studies need to be conducted in order to determine their clinical relevance.

#### 5. Conclusions

To summarize, *Heterotheca inuloides* was successfully used to synthesize Ag NPs on *Agave lechuguilla* and silk fibers. This bioreduction method proved to be simple, cost-effective and environmentally friendly and capable of obtaining silver nanostructures with low polydispersity with an average size of 20 nm. Another important finding in this study is that Ag nanoparticles obtained on *Agave lechuguilla* fibers are AgO free, which could be important for potential applications of these materials as antimicrobials towards gram(+) and gram(-) bacteria. In addition, the mechanical properties of *Agave lechuguilla* thread bearing Ag NPs are improved in comparison to those of commercial silk suture threads, making these natural fibers a good candidate to be used in surgical procedures.

Supplementary data to this article can be found online at http://dx. doi.org/10.1016/j.msec.2016.06.066.

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