Journal of Cultural Heritage 23 (2017) 138–147

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Original article

Microbial diversity of pre-Columbian archaeological textiles and the effect of silver nanoparticles misting disinfection

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ARTICLE INFO

Article history:
Received 4 April 2016
Accepted 25 July 2016
Available online 16 August 2016

Keywords:
Silver nanoparticles
Microorganisms
Disinfection
Archaeological textiles
SEM-EDS

ABSTRACT

Biodeterioration of archaeological materials contribute to significant economic losses and the destruction of invaluable pieces of cultural heritage. The study materials were 5 pre-Columbian fibres (1250–1450 A.D., Argentina). The microscopic analyses (SEM-EDS) showed that they were made of cotton, sisal and wool, as well as they were contaminated by mineral impurities and dust. So far, no research has been conducted on determining the effectiveness of disinfection with silver nanoparticles (AgNPs) misting of historical textiles. The studies showed that the reduction of microorganism number was between 30.8–99.9%, which depended on the qualitative microbial contamination and its amount. Different AgNPs sensitivity of microorganisms was noted, with the least susceptible being endospore-forming bacteria *Bacillus*, more easily inhibited were bacterial genus *Oceanobacillus*, *Kocuria*, *Paracoccus* and moulds *Cladosporium*, *Penicillium*. AgNPs misting does not adversely influence the pH and chemistry textiles. The presented in this paper disinfection method with AgNPs misting can be used for disinfection of archaeological textiles made of wool, cotton and sisal, as an alternative to the currently available methods.

1. Research aims

Biodeterioration of historical fabric causes the destruction of priceless cultural heritage. Detailed knowledge of these materials and present microbiota allow for ideal protection. Earlier studies showed that the use of silver nanoparticles (AgNPs) leads to a high protection of textiles against microbial development. AgNPs disinfection did not significantly influence the mechanical and optical parameters of textiles (cotton, flax, wool, silk), even those which were artificially aged. However, so far AgNPs has not been used for the disinfection of historical textiles. Hence, the aims of this paper were to identify archaeological pre-Columbian textiles (1250–1450 A.D.) based on microscopic analysis of their surface; the assessment of the diversity of microorganisms on pre-Columbian textiles; the AgNPs misting disinfection efficiency, the determination of the sensitivity of isolated microorganisms to AgNPs and whether AgNPs misting changes pH and chemistry of disinfected textiles.

2. Introduction

The problem of fabric biodeterioration is often discussed in literature. Biodeterioration not only causes economic losses but in the case of artefacts also an immeasurable loss of cultural heritage. Archaeological fibres, which usually undergo biodeterioration, include wool, cellulose fibres and silk [1,2]. The biodeterioration of textiles leads to a musty odour, change in pH, permanent staining, decrease in strength [3]. Depending on a fibre's origin, different kinds of microorganisms are responsible for their degradation. In animal origin textiles, the carbon and nitrogen source are proteins (e.g. keratin in wool, fibroin and sericin in silk) [4,5]. Wool is mostly degraded by keratinolytic fungi genus: *Acremonium*, *Alternaria*, *Aspergillus*, *Cephalothecium*, *Chaetomium*, *Chrysosporium*, *Dematium*, *Fusarium*, *Microsporum*, *Oospora*, *Penicillium*, *Rhizopus*, *Scolulariopsis*, *Stachybotrys*, *Thrichoderma*, *Trichophyton*, *Ulocladium*; as well as bacteria from genera *Alcaligenes*, *Bacillus*, *Proteus*, *Pseudomonas*, *Streptomyces* [5–8]. In the case of plant origin textiles (e.g. cotton, linen) cellulose, hemicellulose, lignin are a food source. Plant origin fabrics are colonized by cellulolytic microorganisms, such as fungi: *Alternaria*,

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http://dx.doi.org/10.1016/j.culher.2016.07.007
1296-2074/

Aspergillus, *Aureobasidium*, *Chaetomium*, *Cladosporium*, *Fusarium*, *Mnemoniella*, *Mucor*, *Myrothecium*, *Paeciliomyces*, *Penicillium*, *Rhizopus*, *Stachybotrys*, *Trichoderma*, *Trichotecium*, *Verticillium* and bacteria: *Arthrobacter*, *Bacillus*, *Cellulomonas*, *Cellvibrio*, *Clostridium*, *Cytophaga*, *Microbispora*, *Nocardia*, *Pseudomonas*, *Sporocytophaga Streptomyces* [2,7,9–11].

The protection of cultural heritage from the effects of biodeterioration should consist mainly of appropriate storage of objects. The microclimatic conditions for storage of antique textiles should be stable: temperature about 10 °C, relative air humidity below 50% [12,13]. Any light intensity is not safe for stored item. Recommended 50 lux is a compromise between deterioration and visibility [14,15]. However, if the material has visible signs of microbial growth, it should be immediately isolated from the remaining collection and the appropriate method of disinfection should be applied to protect the material against progressive biodeterioration and people from contact with harmful microorganisms. The choice of the method should be preceded by a detailed research of the material. It must include microscopic and chemical analyses, as well as microbiological procedures. The disinfection should be carried out in the case of the mass contamination of museum collections and prevent the destruction of valuable exhibits. In some cases, conservator may decide that any disinfection method will cause more harm than good, then the object should be kept in RH < 60% to slow down the deterioration. The range of disinfection methods is extensive, chemical methods involve the use of biocides (e.g. ethanol, quaternary ammonium salts, titanium dioxide), gases and vapours (e.g. ethylene oxide, essential oils), while physical factors include: gamma radiation, UV, low or high temperature [16–19]. Each of the methods has a different efficacy, from sterilization to biostatic effect. Most of them were tested for protection of culture heritage objects, but due to many limitations (change the material properties (colour, strength), low effectiveness against spores, harmfulness to humans and the environment) without great success. Hence, the search for a new effective method of disinfection of historical materials is a focus of scientific interest.

The antimicrobial properties of silver have been known for centuries. Due to the wide range of properties including antimicrobial it is widespread and used in e.g. medicine, chemistry, electronics, papermaking, in everyday products such as underwear, air filters, coatings in refrigerators, washing machines [20–22].

Previous studies have shown that silver nanoparticles in the form of a mist may be used for disinfection of historical materials [23]. We have carried out model studies which allowed for the optimization of the process in order to achieve the highest antimicrobial efficiency [24–26]. The disinfection process is carried out in a closed chamber (safe for both the operator and environment) (Fig. 1). The detailed description of process parameters was published by Gutarowska et al. [23]. The obtained results were promising and material studies (also artificial ageing) showed that this process does not substantially affect the strength (elongation at break, tensile and tear indices, breaking strength) and colour parameters (CIELab and whiteness) for textiles (wool, silk, cotton, flax), papers (5 different types), wood (pine, oak, beech) and leather [23,26]. An additional advantage of this method is the protection for the future (applied AgNPs remain on the surface), as compared to the short-term methods [26]. The best antimicrobial results were obtained for textiles (linen and silk) and groundwood paper (mean reduction equalled to 69–92%) and it was higher for moulds than bacteria [23]. The disinfection of historical objects (19th–20th century) resulted in satisfactory reduction of 99.9% on wood and paper maps and lower on painting canvas and parchment: 68–80% [25]. In addition, there was no threat to the employee when working with the disinfection chamber. The level of AgNPs in the air was 0.22 mg/m^3 and decreased over two-fold after chamber airing [26]. The noted values are lower than any limitations for nanoparticles suggested by the National Institute for Occupational Safety and Health (NIOSH) [27,28]. In view of the previously obtained results, in the presented paper AgNPs misting was applied for the first time to archaeological textile objects – the fragments of the pre-Columbian (1250–1450 A.D.) fabrics La Plata Museum in Argentina.

Fig. 1. Disinfection chamber for sliver nanoparticle misting.

Table 1
Characteristics of tested fibre samples.

No.	Type		Origin
1	Archaeological textile	Cotton	1250–1450 AD Puna Argentina, Santa Rosa de Tastil (Argentina)
2		Sisal	
3		Wool	
4		Wool	
5		Wool	
6	Animal hair	*Lama pacos*	Municipal Zoological Garden in Lodz (Poland)
7		*Lama guanicoe*	
8		*Vicugna vicugna*	

3. Materials and methods

3.1. Textile samples

The pre-Columbian archaeological textiles (5 samples, Nos. 1–5) were deposited in Deposit 25 at La Plata Museum (Argentina) (Table 1, Fig. 2). According to Museum data, all textiles were made of wool (analysis showed that two are made of cotton and sisal). For morphological analysis of archaeological fibres, as a comparison, the animal hair of three camelids was tested: *Lama pacos* (alpaca), *Lama guanicoe* (guanaco) and *Vicugna vicugna* (vicugna) (3 samples, Nos. 6–8) (obtained from the Municipal Zoological Garden in Lodz, Poland) (Table 1).

Fig. 2. Macroscopic images of selected archaeological textiles. A – sample 1 (cotton); B – sample 2 (sisal); C – sample 4 (wool).

3.2. Microscopic analysis

The scanning electron microscopy with energy-dispersive X-ray spectroscopy (SEM-EDS) was performed to chemically and morphologically analyze the surface of archaeological textiles and animal hair. The analysis was done with Nova NanoSEM 230, SE Detector, HV: 15 kV, Low Vacuum (FEI, USA) with an X-ray microanalyser EDS (EDAX, USA). The quantitative chemical analysis of the studied materials was carried out with the use of ViP Quant software with ZAF and low vacuum correction procedures. The samples were prepared by fixing the fabric to a holder by use of conducting carbon adhesive tape.

The morphological comparison of archaeological fibres with the hair of camelids was also performed using an optical microscope (transverse and longitudinal images).

3.3. Microbial analysis of textiles

Microbial numbers on all samples were determined using serial dilutions in sterile saline (0.85% NaCl) and culturing on microbiological media: MEA (Malt Extract Agar, Merck, Germany) with chloramphenicol for fungi, and TSA (Tryptic Soy Agar, Merck Germany) with nystatin for bacteria. Samples were incubated using the following conditions: bacteria 24 h, 28 ± 2 °C and fungi 72 h, 28 ± 2 °C. After incubation, the microbial colonies were counted and expressed as microorganism number (CFU/g). All samples were done in triplicate.

The identification of isolated microorganisms was done by molecular methods. Bacterial strains were identified based on the nucleotide sequence of gene 16S rRNA according to the methodology presented by Kręgiel et al. [29]. Identification of moulds was performed based on the ITS1/2 of the rDNA region [30]. Genomic DNAs of microorganisms were extracted using the described method [29,31]. The resulting nucleotide sequences of 16S rRNA genes and ITS1/2 region for the studied microorganisms were analyzed and compared with the sequences published in the National Center for Biotechnology Information (NCBI) database, using the BLASTN 2.2.32+ program and Vector NTI Express software (Thermo Fisher Scientific, USA) [32]. The sequences obtained for microorganisms were deposited in the NCBI GenBank database. The pure cultures of microorganisms were deposited in the Pure Culture Collection at the Institute of Fermentation Technology and Microbiology at Lodz University of Technology (ŁOCK – Łódzki Ośrodek Czystych Kultur) (Table 3).

3.4. Silver nanoparticles misting disinfection

Colloidal solution of silver nanoparticles (concentration 90 ppm; ϕ = 10–80; pH = 7) (Mennica Polska S.A., Poland) was applied in the chamber (1.73 m^3) under specific conditions: temperature T = 25 °C, RH = 90%, air flow 1 m/s (Fig. 1). The chamber and disinfection procedure are patent protected [33]. The silver nanoparticles misting was described in detail by Gutarowska et al. [23].

3.5. Disinfection effectiveness

The number of microorganisms before and after disinfection was established to assess the disinfection efficiency. The reduction in the microorganism number (total and particular species) due to AgNPs treatment was calculated using Eq. (1).

$$R = \frac{N_0 - N}{N_0} \times 100\% \quad (1)$$

where N is the number of microorganisms in the sample with silver nanoparticles (CFU/g); N_0 is the number of microorganisms in the sample without silver nanoparticles (CFU/g).

A one-way ANOVA was performed to assess statistical significance between the number of microorganisms before and after AgNPs misting.

Table 2
Microscopic and chemical analyses of archaeological samples.

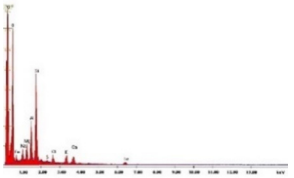
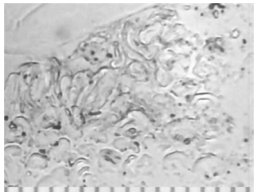
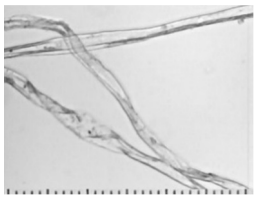
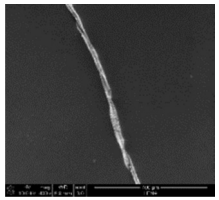
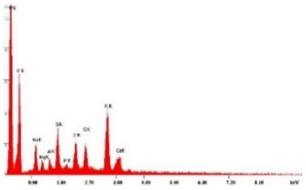
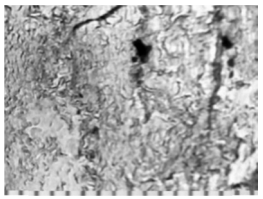
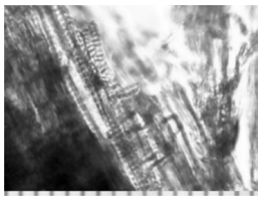
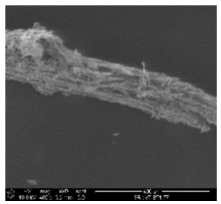
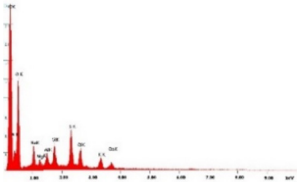
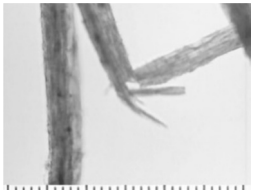
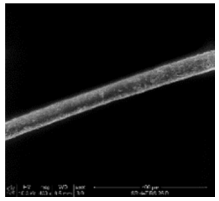
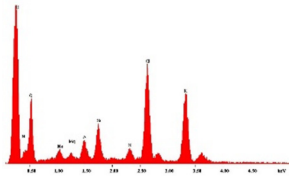
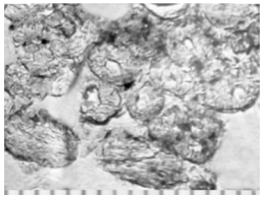
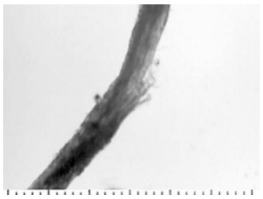
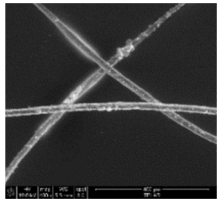
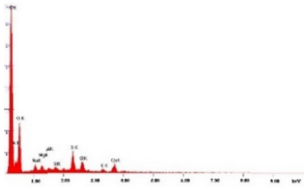
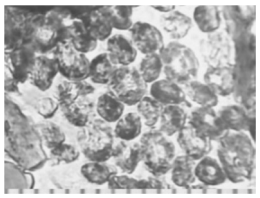
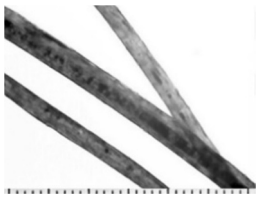
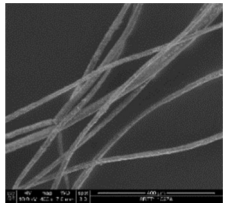
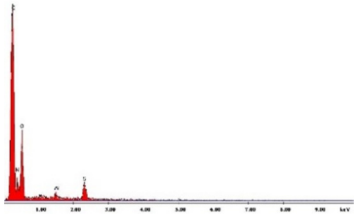
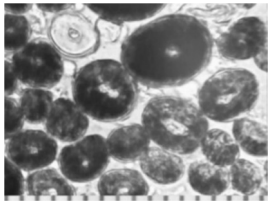
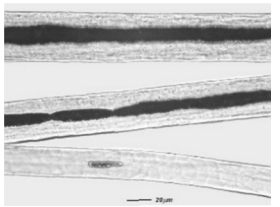
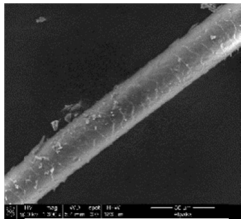
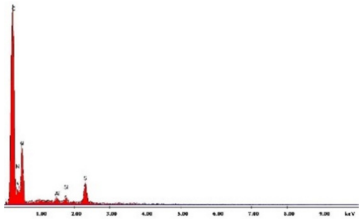
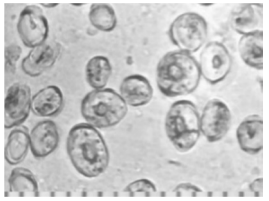
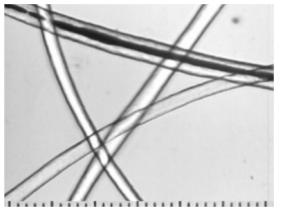
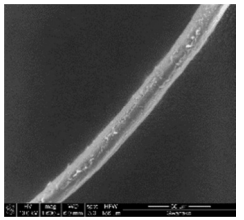
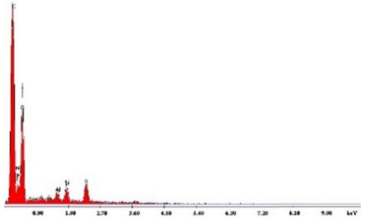
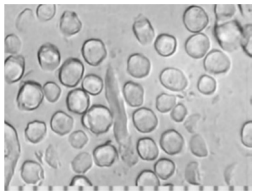
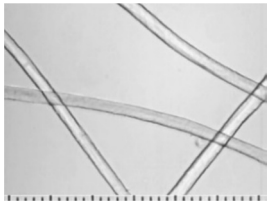
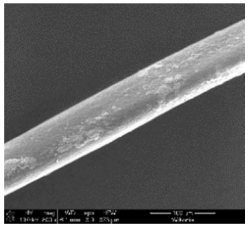
Textile	Chemical analysis (SEM-EDS)		OM image		SEM image
	Element content (%)	Spectrum	Transverse	Longitudinal	
1	C: 40.81; O: 39.29; Si: 8.16; Al: 3.30; Fe: 1.82; Ca: 1.46; Mg: 1.46; K: 1.41; Na: 1.31; Cl: 0.88; S: 0.11				
2	C: 46.19; O: 30.02; K: 9.24; Si: 2.88; Na: 2.34; Cl: 2.78; Ca: 2.70; S: 2.17; Mg: 0.70; Al: 0.65; P: 0.32				
3	C: 45.09; O: 29.03; N: 8.45; Cl: 3.96; Na: 3.43; S: 3.07; Si: 2.08; K: 1.78; Al: 1.09; Ca: 0.90; Fe: 0.74; Mg: 0.38				
4	C: 49.18; O: 20.44; K: 9.86; Cl: 9.23; N: 5.95; Si: 2.28; Al: 1.19; S: 0.82; Na: 0.75; Mg: 0.31				
5	C: 55.01; O: 26.36; N: 9.62; S: 2.39; Ca: 2.15; Cl: 1.76; Na: 0.89; K: 0.63; Mg: 0.63; Si: 0.38; Al: 0.17				

Table 2 (*Continued*)

Textile	Chemical analysis (SEM-EDS)		OM image		SEM image
	Element content (%)	Spectrum	Transverse	Longitudinal	
6	C: 49.62; O: 31.47; N: 15.09; S: 3.18; Al: 0.64				
7	C: 52.98; O: 25.71; N: 14.79; S: 4.41; Si: 1.27; Al: 0.84				
8	C: 43.06; O: 36.21; N: 13.48; S: 3.06; Si: 2.99; Al: 1.21				

SEM-EDS – scanning electron microscopy with energy-dispersive X-ray spectroscopy; OM – optical microscope.

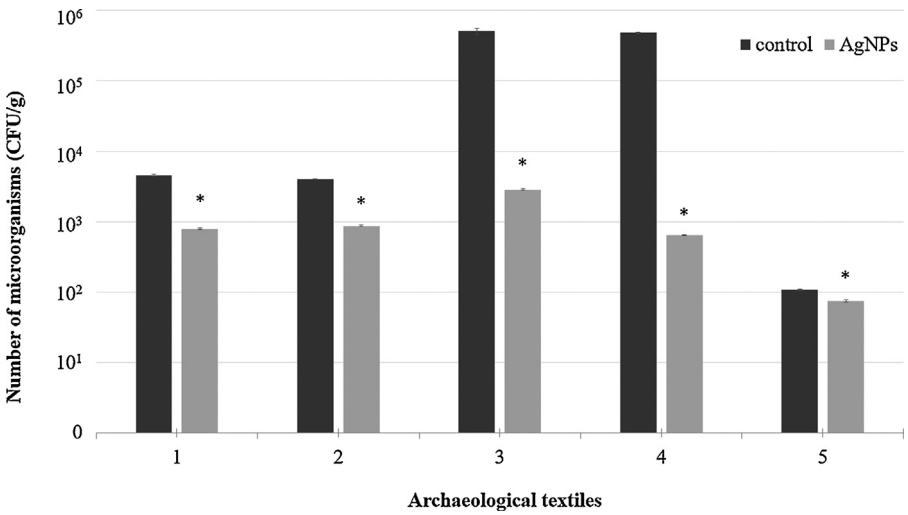


Fig. 3. The number of microorganisms on archaeological textiles before (control) and after AgNPs misting. *Significantly different to the sample without AgNPs, ANOVA with significance level $P < 0.05$.

3.6. Textile properties

3.6.1. Fourier transform infrared spectroscopy (FT-IR) attenuated total reflectance (ATR) spectroscopy

The samples were subjected to image analysis by the Fourier transform infrared spectroscopy (FT-IR) Thermo Scientific Nicolet 6700 using the attenuated total reflectance (ATR) for the rapid analysis of solids, equipped with a diamond crystal in the range of 400–4000 cm^{-1}, with a resolving power of 4 cm^{-1} and a scan rate of 0.6329 cm/s.

Results were analyzed using the program OMNIC 3.2. (Thermo Scientific).

3.6.2. pH measurements

The pH values of textiles aqueous extracts were measured according to ISO 3071:2005 [34].

4. Results and discussion

The fragments of archaeological fabrics provided by the Museum of La Plata (Argentina) had visual biodeterioration signs (Fig. 2). They were brittle and the majority of them had disintegrated into separate fibres. Most tested fibres were broken (Table 2). According to the museum's data, all of them were made of wool, however, the microscopic analyses of cross-sections and

Table 3
Microorganisms isolated from archaeological textiles.

No.	Microorganisms	ŁOCK number	NCBI GenBank accession number	Closely related species (GenBank accession number)	Similarity (%)	Archaeological textile (isolation percentage %)
1.	*Bacillus atrophaeus*	1013	KT728844	*Bacillus atrophaeus* strain M14 (HQ699515)	99.9	5: 9.59
2.	*Bacillus cereus* I	1002	KT728833	*Bacillus cereus* strain LH8 (KC248215)	99.9	1: 1.51; 2: 2.09; 4: 0.55
3.	*Bacillus cereus* II	1003	KT728834	*Bacillus cereus* strain LH8 (KC248215)	99.9	1: 0.15
4.	*Bacillus licheniformis* I	1014	KT728845	*Bacillus licheniformis* strain CCMMB 935 (KF879276)	99.9	5: 6.85
5.	*Bacillus licheniformis* II	1015	KT728846	*Bacillus licheniformis* strain CCMMB 907 (KF879261)	99.9	5: 31.51
6.	*Bacillus pumilus*	1008	KT728839	*Bacillus pumilus* strain NF6 (KM613155)	100	1: 0.15
7.	*Bacillus tequilensis*	1010	KT728841	*Bacillus tequilensis* strain Y10 (KF641803)	100	3: 0.03
8.	*Kocuria rosea*	1001	KT728832	*Kocuria rosea* strain YJ-ST6 (KF876863)	100	1: 61.28; 2: 33.51; 3: 56.21; 4: 14.89
9.	*Micrococcus luteus*	1009	KT728840	*Micrococcus luteus* strain VSG-5 (JQ272845)	99.7	1: 1.56; 2: 3.65; 3: 0.03; 4: 0.03
10.	*Oceanobacillus picturae*	1012	KT728843	*Oceanobacillus picturae* strain R-5321 (NR028952)	99.4	1: 0.30; 2: 12.21; 3: 0.03; 4: 1.67
11.	*Paracoccus yeei*	1000	KT728831	*Paracoccus yeei* strain H13 (AY014178)	99.9	1: 22.52; 2: 30.43; 3: 43.05; 4: 38.61
12.	*Pseudomonas luteola*	1011	KT728842	*Pseudomonas luteola* strain NBRC 103146 (NR114215)	99.7	1: 0.15; 4: 0.02
13.	*Staphylococcus epidermidis*	1005	KT728836	*Staphylococcus epidermidis* strain BBN3N-03d (FJ357607)	100	1: 5.29; 2: 2.61; 3: 0.28; 4: 44.12
14.	*Staphylococcus pasteuri* I	1006	KT728837	*Staphylococcus pasteuri* strain CSB10 (KM203879)	99.9	1: 6.05; 2: 12.89; 3: 0.25; 4: 0.03
15.	*Staphylococcus pasteuri* II	1007	KT728838	*Staphylococcus pasteuri* strain CSB10 (KM203879)	100	3: 0.13
16.	*Aspergillus niger*	1115	KU925905	*Aspergillus niger* strain BPb1 (KP940588)	99.9	1: 1.03; 2: 2.61; 3: 0.01; 4: 0.09
17.	*Cladosporium macrocarpum*	1116	KU925906	*Cladosporium macrocarpum* strain CBS299.67 (EF679372)	99.9	3: 0.01
18.	*Penicillium chrysogenum*	1117	KU925907	*Penicillium chrysogenum* strain ATCC10106 (HQ026745)	100	5: 52.05

ŁOCK – Pure Culture Collection at Institute of Fermentation Technology and Microbiology at Lodz University of Technology; NCBI GenBank – National Center for Biotechnology Information GenBank database; 1–5 – number of archaeological textile.

longitudinal views showed that two of them (samples 1 and 2) are of plant origin (Table 2). Sample 1 was identified as cotton, while sample 2 was sisal. Wool fibres were identified as from alpaca (Table 2). Andean people used as a primary materials bast fibres (bromeliads, cotton and agaves) and wool from American camelids. Camelids llama (*Lama glama*), alpaca (*L. pacos*), vicugna (*V. vicugna*), guanaco (*Lama hunchus* or *Lama guanicoe*), which are very common in South America, and as a consequence, most Argentinian fabrics are made of llama and alpaca hair (domestic animals), and less from vicuna and guanaco (wild animals) [35].

Results show how important the identification of fibres stored in museums is. Each kind of textile requires different storage conditions and treatment. The chemical composition of wool makes it more resistant than cotton to stretching, tearing, environmental factors, including enzymatic degradation than cotton. Inappropriate treatment of wrongly identified textiles may cause damage.

The chemical analysis (SEM-EDS) of archaeological fibres proved identification of the previous fibre's origin. The element content of samples 3–5 included nitrogen (3: 4.45%; 4: 5.95%; 5: 9.62%), one of the main components of proteins (Table 2). The high concentration of carbon (sample 1: 40.81%; 2: 46.19%; 3: 45.09%; 4: 49.18%; 5: 55.01%) proves that archaeological fibres are a good carbon source for microorganisms [36]. Moreover, SEM-EDS showed that fibres were mineralized. Mineralization contributes to textile preservation since corrosive products can be toxic to microorganisms causing microbial growth inhibition [37]. Many chlorite crystals (Na: 0.75–3.43%; K: 0.63–9.86%; Cl: 0.88–9.23%) were detected as well as sand grains (Si: 0.38–8.16%).

Wool is mostly composed from keratin (up to 75%), which includes carbon (50%), oxygen (22%), nitrogen (17%), hydrogen (7%) and sulphur (4%). Similar values were obtained for animal hair (Table 2). Keratin, dirt, suint, grease, and water are present in wool in different concentrations. The high amount of nitrogen is a reason for the rapid biodegradation of wool [38,39]. Cotton fibres, which consist mostly of cellulose (83–92%), is composed of carbon (45%), oxygen (49%), and hydrogen (6%). The rest of the constituents are proteins, ash, pectin, oil, fat, wax, sugars [40,41].

The microbiological analysis showed that archaeological textiles were highly contaminated (1.1×10^2 to 5.0×10^5 CFU/g) (Fig. 3, control – before disinfection). The highest microbial number was detected in samples 3 and 4 (4.8×10^5 to 5.0×10^5 CFU/g). The lowest contamination was noted for wool 5 (1.1×10^2 CFU/g), which was also the least biodeteriorated.

The wool gathered from sheep can be more highly contaminated than those presented in this study by bacteria (1.0×10^3 to 4.0×10^8 CFU/g) and fungi (3.0×10^2 to 7.0×10^3 CFU/g) [42]. Raw cotton is highly contaminated: bacteria 5.0×10^5 to 6.9×10^9 CFU/g and fungi 2.0×10^2 to 1.8×10^7 CFU/g [43]. The archaeological textiles may be highly contaminated due to their contact with soil, e.g. canvas: 1.8×10^6 CFU/cm^2 – fungi [25] or fleece (sheep wool): 2.0×10^9 CFU/g – bacteria; 7.0×10^3 CFU/g – fungi [44]. Studies carried out in this work have not confirmed such a high concentration of microorganisms on historical textiles.

From archaeological textiles, 15 species of bacteria (8 bacilli, 6 cocci, 1 rod) and 3 species of mould were isolated (Table 3). The most biodiversified were sample 1 (cotton) with 11 species isolated, which is consistent with the literature, since cotton is contaminated by a more diverse group of microorganisms when compared to the wool [7]. The least common microorganism species were isolated from sample 5 (4 isolated species), which corresponds to the low contamination and degradation of the fibres. The most microbially contaminated samples 3 and 4 were also very biodiversified (9–10 isolated species). Seven species were detected in four of five samples, while nine species were only present in one sample. *Kocuria rosea* (15–61%) and *Paracoccus yeei* (23–43%) were detected with the highest isolation frequency. *Micrococcus luteus* and *Aspergillus niger* were detected in four out of five samples, however their isolation frequency was very low (0.01–3.65%). On the other hand, *Penicillium chrysogenum* was isolated from textile sample five, but with a 52% isolation frequency.

The studies proved the occurrence of typical microbial species infected archaeological textiles made of cotton (*Aspergillus* sp., *Cladosporium* sp., *Penicillium* sp., *Bacillus* sp., *Pseudomonas* sp.) and wool (*Aspergillus* sp., *Bacillus* sp., *Pseudomonas* sp.) [5,7,8,10]. Cotton fibres are among all textile types, the most susceptible to biodeterioration [2].

Some identified microorganisms were also isolated in previous studies from archaeological materials: *K. rosea* was isolated with

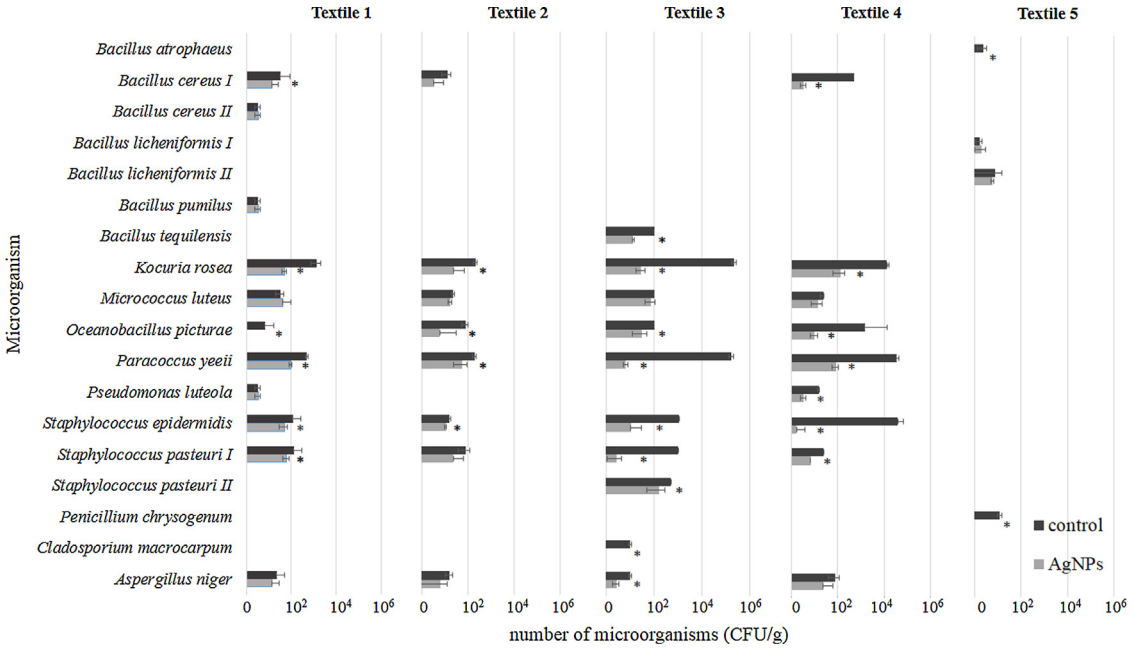


Fig. 4. The sensitivity of microorganisms isolated from archaeological textiles to silver nanoparticles. *Significantly different to the sample without AgNPs, ANOVA with significance level $P < 0.05$.

a high isolation frequency on surfaces (e.g. textiles) in the Central Museum of Textiles, Poland [45], as well as *A. niger* [25]. *K. rosea* and *P. yeei* were also isolated on historical objects (including paintings) by Pangallo et al. [46]. *M. luteus* is known from literature for colonizing textiles [47], while *Oceanobacillus picturae* was firstly isolated from canvas paintings [48]. *Bacillus atrophaeus*, *B. cereus*, *M. luteus*, *Aspergillus* sp. and *Penicillium* sp. were also isolated from different historical objects, such as bricks, wood, paintings, masks (linen, papyrus) [49–52].

The silver nanoparticles misting disinfection reduced, in a statistically significant way, the microbial number by 30.8–99.9%. The highest reduction was noted for samples 3 and 4 (99.4–99.9%), which was the most highly contaminated. Microorganism numbers were reduced by almost 3 log units, which is a very significant achievement. The lowest effectiveness was obtained for sample 5, which was the least contaminated, only 31%. It means that the higher the contamination, the better the expected results are (Fig. 3).

Previously conducted model studies showed that silver nanoparticles misting can significantly reduce the microorganism number on textiles. The reduction of moulds were 91.8% and 31.6–93.4% on wool and cotton, respectively. Bacteria were reduced by 40.6–49.6% and 32.6–99.9% on wool and cotton, respectively [23,25]. It was also proven that the higher reduction after the disinfecting with silver nanoparticles mist was obtained with higher initial microbial contamination [25]. Moreover, the previous studies showed that this disinfection method does not result in a significant deterioration of the mechanical (below 9%) and optical (below 6%) parameters of textile materials and does not lead to their accelerated ageing [23].

Four microorganism species were eliminated from particular samples: *B. atrophaeus* (sample 5), *O. picturae* (1), *P. chrysogenum* (5) and *Cladosporium macrocarpum* (3) (Fig. 4). There were some microorganisms that exhibit different AgNPs sensitivities depending on the isolation sample: *Pseudomonas luteola* (sample 1: 0%; 2: 77.8%) and *Staphylococcus epidermidis* (samples 1–2: 34.0–57.1%; 3–4: 99–100%). Most of the microorganisms from archaeological textiles were sensitive to AgNPs action. Those insensitive to silver nanoparticles were mostly bacilli: *Bacillus cereus* I, *B. licheniformis* I and II, *B. pumilus* and cocci *M. luteus*. These results are compatible with the authors' previous studies [24].

FT-IR ATR analyses were performed to establish whether AgNPs misting have the influence on chemical composition of disinfected textiles. Spectra analyses of control and AgNPs misted samples showed that presented in this paper disinfection process did not have adverse effect on chemistry of textile samples (exemplary

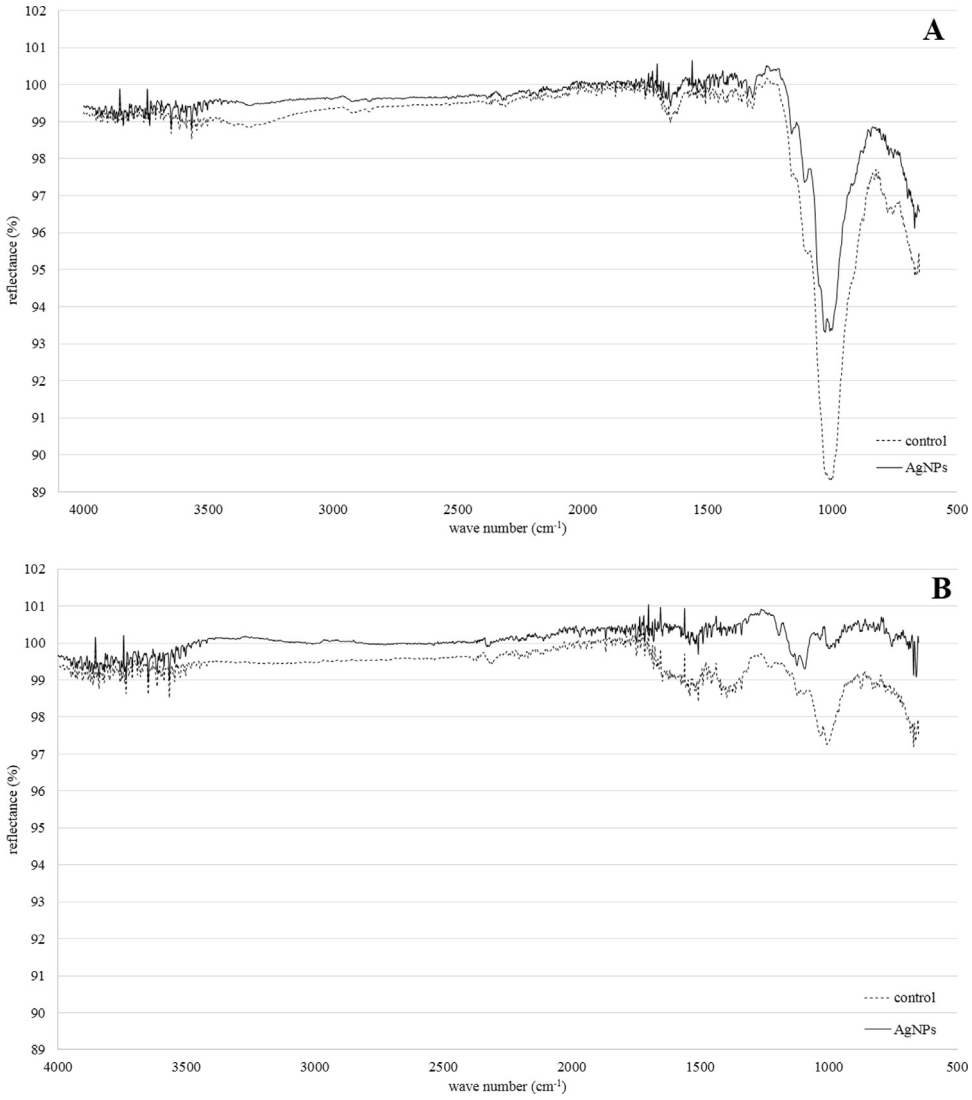


Fig. 5. FT-IR ATR spectra. A – sample 1 (cotton); B – sample 2 (sisal).

Table 4
The pH values for aqueous extract of textiles.

Textile	Control	AgNPs
1	7.74 ± 0.02	7.76 ± 0.01
4	7.21 ± 0.01	7.20 ± 0.01

results, Fig. 5). Moreover, AgNPs colloid (pH = 7) did not change pH of aqueous extracts of analyzed textile samples (exemplary results, Table 4).

The chamber has the capacity of 1.73 m^3. It contains two perforated shelf, which allow to disinfect at one time around 36–42 books (A4 size). It is possible to disinfect one big item, which fits through the chamber door. The process has been optimized for 8 cycles (8 h) with one silver nanoparticles colloid (90 ppm) [25]. During one process, the amount of used nanosilver colloid is about 500 ml, so during this procedure ca. 45 mg of nanosilver is used (5.6 mg/cycle). After disinfection process, the object can be transport to storage unit directly. No waiting period is needed. The price of AgNPs misting process include: price of silver nanoparticles preparation, utilities (water, electricity), maintenance, which gives about 50$ per process.

5. Conclusions

The crucial part of studying archaeological analysis is the fibre identification and determination of chemical and microbiological contamination. Microscopic analyses (SEM-EDS) of pre-Columbian fibres (1250–1450 AD, Argentina, $N = 5$) showed that two samples were made of cotton and sisal (classified in the museum documents as wool), the remainder were composed of wool. It is also important to check the presence of mineral impurities and dust. They should be removed before disinfection, increasing the effectiveness of disinfection. Detailed fibre analysis indicates the use of proper maintenance of the fabric, its storage and selection of disinfection methods. The disinfection with silver nanoparticles misting was effective for pre-Columbian fabrics. The reduction of microbial number was 30.8–99.9%, depending on the microbial species present on the material and the amount of initial microbial contamination. However, different sensitivity of microorganisms on AgNPs disinfection was noted, with the least susceptible being endospore-forming bacteria *Bacillus*, which are known for their high resistance to disinfection. More easily inhibited were bacteria *Oceanobacillus*, *Kocuria*, *Paracoccus* and moulds *Cladosporium*, *Penicillium*. There were no visual changes after disinfection of the material, also pH and chemistry of disinfected textile samples did not change. All results confirm earlier findings that AgNPs misting does not adversely affect the material parameters. Disinfection method with AgNPs misting can be used for the disinfection of archaeological textiles made of wool, cotton and sisal as an alternative to currently used methods.

Funding

This work was supported by the National University of La Plata [UNLP 11N713] and CONICET [PIP 0200].

Acknowledgements

Authors are grateful to Ana Igareta for allowing microbial sampling at Deposit 25, Archaeological Division, La Plata Museum (Argentina), Tomasz Jóźwik and Krzysztof Rochmiński from Municipal Zoological Garden for providing animal fur and Jan Czyżyk from Instal Warszawa S.A. (Poland) for the possibility to use the AgNPs misting chamber, Dr Waldemar Machnowski for scientific advices.

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