



Vapourised hydrogen peroxide (VHP) and ethylene oxide (EtO) methods for disinfecting historical cotton textiles from the Auschwitz-Birkenau State Museum in Oświęcim, Poland

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ABSTRACT

The research aims were: to assess the microbiological contamination of historical cotton textiles from the Auschwitz-Birkenau State Museum in Oświęcim, Poland; to establish the antimicrobial effectiveness of vapourised hydrogen peroxide (VHP) and ethylene oxide (EtO); to determine concentration and time of VHP disinfection and to assess the impact of VHP and EtO on the new and historical cotton textiles properties. The microbiological contamination of historical cotton textiles were 3.1×10^4 – 4.6×10^4 CFU/100 cm² (bacteria) and 7.5×10^3 – 1.8×10^4 CFU/100 cm² (fungi). VHP was more effective against fungi and had similar biocidal effect to EtO. The optimal parameters of VHP disinfection were: concentration 300 ppm, time 20 min. VHP and EtO did not affect the fibre morphology, chemical composition or colour of the new and historical cotton fabrics. Both methods changed the strength parameters and cellulose polymerisation degree of new fabric. Increasing the time (30 min) or concentration (400 ppm) gave a similar biocidal effect but resulted in a decrease in strength parameters and cellulose polymerisation (new fabric) and colour change (historical fabric). VHP is an effective and safe method for historical textiles and can be applied in mass treatment.

1. Introduction

Textiles are one of the numerous types of objects in the collections of the Auschwitz-Birkenau State Museum in Oświęcim, Poland (A-BSM). The textile products are made from different types of fabric, with the predominance of natural fibres such as cotton and linen, some of them having extra wool and viscose fibres. The collection has over 2200 artefacts consisting of both clothes and other useable textile products. The first group in the collection includes prisoners' clothes (sweatshirts, coats, dresses, trousers, caps, functional prisoners' bands, camp number patches), civilian's clothes (coats, jackets, dresses, hats, underwear), SS uniforms and accessories (uniform badges, hats, flags) and children's wear (Fig. 1 a). Striped camp clothing is the largest group (approximately 370 objects) of clothing in the A-BSM collection. Prisoners arriving at KL Auschwitz-Birkenau were deprived of their clothes and received clothes made of a drill fabric (a strong, thick cotton fabric

made in a diagonal weave with a triple warp) in blue-grey stripes. The striped camp clothing, distinguished the prisoners from afar and prevented them from hiding in case of an escape. The museum's collection also includes a smaller group of infant's and children's civilian clothes (approximately 80 objects). This group includes sweaters, matinee jackets, dresses, underwear, gloves, hats and socks, which children most likely wore in the camp (Iwaszko, 2000).

The second group consists of tallisim and ataras (Jewish prayer shawls and ornamental belts), straw mattresses, furniture elements and selected objects made by prisoners, such as a felt Christmas tree, a commemorative shawl and a puppet (Fig. 1 b–c). Furthermore, in addition to the textile collection, selected fragments of some objects from set of objects in mass quantities, such as shoes, suitcases and prostheses, are made of textiles (Fig. 1 d–e). In the collections of the A-BSM, there are approximately 3800 suitcases made of various materials, including fabrics (cotton, linen, jute, hemp and artificial silk). Fabrics were used

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Fig. 1. Samples of textile objects from Auschwitz-Birkenau State Museum: a. prisoner's clothes; b. tallit; c. Christmas tree; d. suitcase; e. shoe (author: a-b. Michał Dziejewski; c-e. Adam de Sas Topolnicki).

in suitcases to cover the top of the suitcase and as a lining. Suitcases brought by prisoners to the camp were thoroughly inspected to steal hidden valuable items and money in favour of the Third Reich. Empty suitcases, together with footwear and leather goods, were collected in the camp warehouses and outdoor (Strzelecki, 2000).

Historical objects are susceptible to destruction by microorganisms that come from soil, water and air. These microorganisms may lead to biodeterioration, which is a multistage and complex phenomenon that causes undesirable changes in the physical properties of materials. As a result of microbiological decomposition, stains, deposits, structural weakness and colour changes may appear in historical textile objects. The progress of this process depends primarily on the chemical composition of the textile materials and storage conditions (relative humidity and temperature of the air) (Szostak-Kotowa, 2004). Stable microclimatic conditions that maintain the air temperature at approximately 20 °C and the relative air humidity (RH) below 60% should inhibit the biodeterioration process (Wolf, 2002). Literature examples indicate that the historical textiles most susceptible to biodeterioration are archaeological textiles in permanent contact with water and soil, such as mummies, textiles in graves, tombs, crypts, sunken ships and soldiers' uniforms (Gutarowska et al., 2016). Additionally, textiles made of cellulose and keratin-based fibres are more susceptible to biodeterioration than fibroin and sericin-based fibres (Pekhtasheva et al., 2012).

Microorganisms involved in the biodeterioration of historical textiles produce extracellular enzymes (cellulolytic and proteolytic), pigments and acids. Biodeterioration involves two processes: assimilation – when fibres are used as a nutrient source; and/or degradation – when fabrics are damaged due to the growth of microorganisms and secretion of metabolites. The degradation of cellulosic and proteinaceous archaeological textiles is mainly connected with filamentous fungi, while silk is primarily destroyed by bacteria (Pekhtasheva et al., 2012).

Suitcases and children's clothes from the A-BSM collections are

stored in storage warehouses in which the environmental conditions are monitored and at the exhibition. The biodeterioration process depends on the objects' storage location and on the type of material from which they are made. The suitcases from the exhibition show greater signs of biodeterioration than those stored in storage warehouses. Microorganisms from the environment and those introduced by visitors may be the reason for their more extensive deterioration. Due to the period from which the suitcases originated (the first half of the 20th century), most of the fabrics used to make suitcases are made from fibres of natural origin. Therefore, they are more susceptible to biodeterioration.

Disinfection of historical objects is a difficult process because it should be effective, not negatively affect the historical objects and also be safe for people and the environment. Many methods of chemical (e.g., alcohols, quaternary ammonium compounds, azoles, essential oils, nanometals and ethylene oxide) and physical (high and low temperature, pressure, modified atmospheres and irradiation) disinfection of historical textile objects are currently available. All of the mentioned disinfection methods have some disadvantages, such as changes in pH, colour, or structure and the occurrence of depolymerisation, hydrolysis, acidolysis and accelerated ageing, or they pose a threat to human health, while some of them cannot be used for mass treatment (Paulus, 2004; Sequeira et al., 2012; Gutarowska et al., 2016).

Thus, there is a need to find new, effective and safe methods for mass disinfection that are adjusted to the specificity of historical textiles and inhabiting microorganisms. Due to the need for historical objects (textiles, suitcases) from the A-BSM to be protected from the destruction caused by biodeterioration, new research has been started on the use of hydrogen peroxide in the gas form (vapourised hydrogen peroxide - VHP) to find an effective and safe technique for fabric disinfection.

The biocidal properties of hydrogen peroxide (H₂O₂) have been known for over 100 years and are widely used in the food, medical and

healthcare industries. The biocidal activity of H₂O₂ consists of the interaction of hydroxyl radicals with a cell, leading to its destruction. Liquid H₂O₂ requires a high concentration and a long period of interaction to obtain an antimicrobial effect. The vapourisation process provides a gas with a higher concentration of H₂O₂ peroxide than in the liquid from which it was vapourised (Hultman et al., 2007). In 1990, the Environmental Protection Agency (EPA 2011. List A: Antimicrobial Products Registered with the EPA as Sterilizers, online at https://www.epa.gov/sites/production/files/2016-12/documents/list_a_sterilizer.pdf) registered H₂O₂ in a gas form as a sterilising agent. In 1992, VHP products were introduced to the pharmaceutical and research market. VHP shows a broad spectrum of virucidal, bactericidal, fungicidal and sporicidal activities, as confirmed by several studies (Klapes and Vesley, 1990; Neighbor et al., 1994). However, this method has not been used in museums until now, and its impact on the disinfected historical material has not been established.

The aims of the study were: 1) to assess the microbiological contamination of historical cotton textile objects (suitcases, clothes) from the Auschwitz-Birkenau State Museum in Oświęcim, Poland; 2) to establish the antimicrobial effectiveness of vapourised hydrogen peroxide (VHP) and ethylene oxide (EtO) against the microorganisms inhabiting the historical textile objects; 3) to determine the optimal concentration and time of VHP disinfection; and 4) to assess the impact of VHP and EtO on the morphological (SEM), structural (FTIR, degree of cellulose polymerisation), optical (changes in colour), and mechanical properties (breaking strength, elongation) of new and historical cotton textiles. The scope of the study included: A) isolation and identification of microorganisms from the historical textiles from the A-BSM; B) evaluation and comparison of VHP and EtO disinfection effectiveness (under different conditions of concentration and time) carried out under model conditions on the new cotton fabric inoculated with the microorganisms C) evaluation and comparison of the impact of VHP and EtO disinfection on structural, morphological, optical and mechanical properties of the new cotton fabric D) evaluation and comparison of VHP and EtO disinfection effect on structural (FTIR, degree of cellulose polymerisation), morphological (SEM) and optical changes (changes of colour) in the historical cotton fabric from the A-BSM.

2. Materials and methods

2.1. Historical textile objects

Historical suitcases (number of samples, N = 250) and children's clothes (N = 30) dated to the first half of the 20th century from the collections of A-BSM in Oświęcim were tested for microbial contamination and isolation of microorganism strains for the model study.

Moreover, historical cotton fabric from a historical replica of a suitcase from the A-BSM collection (Fig. 2) was tested to evaluate the impact of the disinfection methods (VHP and EtO) on the material properties. The replica of the suitcase came from the same period, i.e.,

the first half of the 20th century, and it was stored under the same conditions as the original objects. The suitcase has a wooden construction, a cladding made of vulcanised fibre (treated with zinc chloride and leached and pressed cellulose), and a lining inside with fabric, under which cardboard is placed. The VHP and EtO disinfection processes were performed on selected textile fragments (4 × 4 cm) from the suitcase replica.

2.2. New textile samples

In model studies, new cotton fabric samples (N = 130) (130 g/m², Andropol S.A., Poland) were used. The fabric was bleached with H₂O₂, did not contain any dyes and additives. Samples were 5 × 2 cm and sterilized before disinfection efficiency test (121 °C, 20 min).

2.3. Assessment of microbial contamination

In order to determine the microbial contamination on the surface of suitcases' textile elements and clothes, imprints method using contact plates was applied. To collect the sample, the surface of the media was pressed against the tested surface for 3–5 s. TSA (Tryptic Soy Agar, Merck, Germany) with nystatin and Sabouraud Agar (Merck, Germany) with chloramphenicol media were used to determine the total number of bacteria and fungi, respectively. The plates were incubated at temperature 30 ± 2 °C for 48 h (bacteria) and 28 ± 2 °C for 5 days (fungi). After incubation, the colonies were counted and the result was presented as Colony Forming Units (CFU) per 100 cm².

2.4. Identification of microorganisms

The identification of isolated bacteria was performed using molecular method. Bacterial strains were identified based on the nucleotide sequence of gene 16S rRNA according to the methodology presented by Krakova et al. (2017). Genomic DNAs of microorganisms were extracted using the described method, the obtained nucleotide sequences of 16S rRNA genes were analysed 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).

Fungi were identified based on macroscopic and microscopic observations on MEA (Malt Extract Agar, Merck, Germany) and CYA (Czapek Yeast Agar, Oxoid, UK) media using diagnostic keys (Samson et al., 1996; Pitt and Hocking, 2009).

2.5. Cultures of microorganisms in model studies

For tests of disinfection efficiency, microorganisms with the highest isolation frequency (> 15%) were used: *Alternaria alternata*, *Aspergillus flavus*, *Cladosporium cladosporioides*, *Epicoccum nigrum*, *Fusarium poae*, *Penicillium chrysogenum*, *Bacillus licheniformis*, *Lysinibacillus fusiformis*,



Fig. 2. Historical: a. suitcase replica from A-BSM; b. cotton fabric from suitcase replica (author: a. Margrit Borman; b. Adam de Sas Topolnicki).

Micrococcus luteus, *Psychrobacillus psychrodurans*, *Staphylococcus epidermidis*. Pure cultures of isolates were activated before experiments: fungi on MEA for 5–10 days at temperature $28 \pm 2^\circ\text{C}$ depending on the strain, and bacteria on TSA at temperature $30 \pm 2^\circ\text{C}$ for 2 days. Inoculum formed suspension of fungal conidia or bacterial cells in sterile saline (0.85% NaCl). The concentration of inoculum was established using Thoma cell counting chamber and plate methods and equalled for bacteria and fungi 10^8 CFU/ml and 10^7 conidia/ml, respectively. Except of pure strains inoculum, the mixtures of bacteria and fungi isolates were also prepared by mixing pure strains inoculum in equalled proportions. Standardized inoculum (0.2 ml) was transferred on sterile new cotton sample on M_0 medium ($\text{MgSO}_4 \times 7\text{H}_2\text{O}$ - 5 g; $(\text{NH}_4)_2\text{SO}_4$ - 3 g; KH_2PO_4 - 1 g; glucose - 20 g; agar - 15 g for 1000 ml H_2O) and incubated for 21 days in climatic chamber (temperature $28 \pm 2^\circ\text{C}$, relative humidity RH 80%). Experiment was performed in duplicate. Next, disinfection with VHP and EtO were performed and efficiency was evaluated.

2.6. Vapourised hydrogen peroxide (VHP) disinfection

Samples were treated with VHP in a model chamber designed for low-temperature treatment with oxidising chemicals in the gas phase and consisting of the following three modules: a 1 m^3 hermetic chamber, a central unit in which a VHP generator is located along with instrumentation, and an absorption column with a filter (Sójka-Ledakowicz et al., 2015). The model chamber allows disinfection with VHP concentrations of up to approximately 900–1000 ppm at a temperature of up to approximately $65 \pm 5^\circ\text{C}$. The processes were performed under a controlled concentration, temperature, relative humidity and pressure so that the actual concentration of VHP in the decontamination chamber did not exceed the condensation point. The process conditions were as follows: RH 80–85%; temperature 30°C ; VHP concentration/exposure time 300 ppm/20 min (designated VHP 1), 400 ppm/20 min (VHP 2) and 300 ppm/30 min (VHP 3).

2.7. Ethylene oxide (EtO) fumigation disinfection

Fumigation with EtO was performed in a fumigation chamber of type UDA 800 (TERPO S.A., Vacuum Techniques Plant in Koszalin) using a 1:9 mixture of EtO and CO_2 (Rotanox) for 8–12 h and a 10-day quarantine period.

2.8. Disinfection efficiency

The number of microorganisms before and after VHP or EtO disinfection was established to assess the disinfection efficiency according to AATCC Test Method 100-2012 using culture method on TSA at temperature $30 \pm 2^\circ\text{C}$ for 48 h (bacteria) and MEA at $28 \pm 2^\circ\text{C}$ for 5 days (fungi). The reduction in the microorganism number (total and particular species) due to disinfection was calculated using the equation:

$$R = (N_0 - N / N_0) \times 100\%$$

Where: N is the number of microorganisms in the sample after disinfection (CFU/100 cm^2); N_0 is the number of microorganisms in the sample before disinfection (CFU/100 cm^2).

2.9. Optical analysis of the textiles

The colour changes of the textiles after the disinfection were determined spectrophotometrically according to European Standard EN ISO 105-J01:1999. Colorimetric values of the cotton fabric specimens were measured on V-670 UV-Vis-NIR spectrophotometer (JASCO, USA), using the CIEL*a*b* colour system, created by Commission Internationale de l'Eclairage. The colour coordinates representing lightness, red-green axis (a^*) and yellow-blue axis (b^*) were recorded. An average

of four colour readings was taken for each sample. The differences between disinfected and undisinfected cotton fabrics samples are expressed as ΔL^* , Δa^* , Δb^* . The total colour changes after disinfection processes were calculated using the equation (Abdel-Kareem, 2005; Mokrzycki and Tatol, 2011):

$$\Delta E^* = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$$

2.10. Structural analysis of the textiles

To determine changes in the molecular structure of the textiles after disinfection, the samples were examined by Fourier transform infrared (FTIR) spectroscopy. Analysis of new cotton fabric was performed using a Nicolet 6700 FTIR spectrophotometer (Thermo Scientific, USA), and a Nicolet 8700 FTIR spectrometer (Thermo Scientific, USA) was employed for analysis of the historical cotton fabric; both of the instruments were equipped with an attenuated total reflectance (ATR) accessory, and spectra were collected in the range of $400\text{--}4000\text{ cm}^{-1}$ with a resolving power of 4 cm^{-1} . The test results were analysed with the software application OMNIC 3.2. (Thermo Scientific, USA).

To determine the degree of cellulose polymerisation before and after the disinfection processes, gel permeation chromatography (SEC-MALLS-RI analyses) was performed. Samples were comminuted and derivatised using phenyl isocyanate (Fluka, GC) to obtain cellulose tricarbonyl (CTC) (Łojewski et al., 2010). A minimum of 90 mg of sample was dried for 30 min at 105°C , treated with 1 cm^3 of water-free pyridine and 0.1 cm^3 of phenyl isocyanate (PIC) and conditioned at 80°C for 72 h. To terminate the substitution reaction, 0.1 ml of methyl alcohol was added, and the solutions were cooled. Samples were dissolved in tetrahydrofuran (THF, HPLC grade) and filtered prior to each SEC analysis. The average molecular mass was determined using a Waters chromatographic system, which consists of an isocratic pump 1515, an autosampler 717+, a column oven, a MALLS detector (Dawn Heleos, Wyatt Technology, working at 658 nm) and a differential refractive index detector (Optilab T-rEX, Wyatt Technology, working at 658 nm) acting as a concentration sensitive detector (Pawcenis et al., 2015). The degree of cellulose polymerisation of the samples was calculated from the equation:

$$DP = M_w / M_{\text{mono}}$$

Where M_w is the average molecular mass and M_{mono} is the molecular mass of the glucose molecule substituted in the cellulose chain (510 g/mol).

2.11. Mechanical analysis of the textiles

Mechanical tests of textiles were performed to estimate the changes in the strength due to the disinfection processes. The strength parameters (breaking strength and elongation at break) of textiles conditioned for 24 h at a temperature of 20°C and RH 65%, were determined using a INSTRON Model 5944 braking testing machine (Instron, USA), according to EN ISO 13934-1:2013. The strength tests were performed in 4 repetitions.

2.12. Morphological analysis of the textiles

The surface morphology of cotton fibres in the textile samples before and after disinfection was investigated with scanning electron microscopy (SEM). The samples of fabrics were deposited onto an aluminum specimen mount with a carbon tape and sputter-coated with gold before examination using a JFC-1200 evaporator (JEOL, Japan) in an argon atmosphere (argon pressure - 8 Pa) for 30 s at an amperage of 15 mA. For imaging at magnification $5000 \times$ Nova Nanosem 230 FEI scanning electron microscope (Thermo Scientific, USA) was employed for analysis of new cotton fabric and Quanta 3D FEI scanning electron

microscope (Thermo Scientific, USA) for historical cotton fabric. The tests were performed under high vacuum using accelerating voltage of 5 kV.

2.13. Statistical analysis

The arithmetic mean and standard deviation for microorganism numbers, mechanical parameters and degree of cellulose polymerisation of textiles were calculated. Differences between the results obtained for control samples and samples after disinfection processes were analysed using one-way Analysis of Variance (ANOVA) at significance level $p < 0.05$. All data were analysed using the statistical software STATISTICA 6.0 (Statsoft, USA).

3. Results and discussion

3.1. Microbiological contamination of the Auschwitz-Birkenau State Museum (A-BSM) historical cotton textile objects

The microbiological contamination of the A-BSM historical cotton textile objects ranged from 10^1 to 10^6 CFU/100 cm². The average concentration of bacteria was 4.6×10^4 CFU/100 cm² (suitcases) and 3.1×10^4 CFU/100 cm² (clothes). Fungi contaminated the surfaces by 7.5×10^3 CFU/100 cm² (suitcases) and 1.8×10^4 CFU/100 cm² (clothes) on average (Table S1). The suitcases were more contaminated by bacteria than by fungi. Most of the tested suitcases (64.3%) showed microbial contamination at the level of 10^4 – 10^5 CFU/100 cm². The clothes were contaminated to a similar extent by bacteria and fungi. Most of the tested clothes (66.6%) showed low microbial contamination at the level of 10^0 – 10^3 CFU/100 cm² (Table S1).

Based on the 16S rRNA gene nucleotide sequences, 13 species of bacteria were detected from the A-BSM historical textile objects: *Bacillus* (6 species), *Micrococcus* (1), *Psychrobacillus* (1), *Paenibacillus* (1), *Enterobacter* (1), *Lysinibacillus* (1), *Pseudomonas* (1), and *Staphylococcus* (1). The degree of similarity in the nucleotide sequence of the 16S rRNA gene was over 99% for all analysed strains (Table 1). Additionally, 11 species of filamentous fungi belonging to the following genera were identified: *Aspergillus* (2), *Penicillium* (1), *Rhizopus* (1), *Cladosporium* (1), *Fusarium* (1), *Alternaria* (1), *Epicoccum* (1), *Chaetomium* (1), *Trichoderma* (1), and one unidentified *Mycelium sterillum*; furthermore, one yeast species, *Stephanoascus ciferrii*, (1) was identified (Table 1).

Historical textile objects from A-BSM were inhabited by bacteria mainly belonging to the genera *Bacillus*, while the most frequently isolated filamentous fungi included the genera *Aspergillus*, *Penicillium* and *Cladosporium*. Many species detected in the present study were previously found to inhabit the interior surfaces (walls, floors, door frames, bunk bed, etc.) of the wooden and brick barracks at the A-BSM (Koziróg et al., 2013; Rajkowska et al., 2014). Furthermore, most of these species were also isolated from different museum textile objects. The occurrence of *Chaetomium* sp., *Alternaria* sp., *Trichoderma* sp., *Aspergillus* sp. and *Penicillium* sp. on various textile fabrics stored in the Egyptian Museum, Wawel Royal Cathedral in Krakow (Poland), La Plata Museum (Argentina) and religious institutions in Slovenia (churches and cloisters) was described, and the authors showed the destructive potential of the isolated strains (Abdel-Kareem, 2010; Pangallo et al., 2013; Kavkler et al., 2015; Lech, 2017; Pietrzak et al., 2017). It is noteworthy that all of the aforementioned fungi colonising the cotton samples may degrade cellulose fibres, which highlights their biodegradation potential (Szostak-Kotowa, 2004; Pekhtasheva et al., 2012). Moreover, in contrast to the relatively well-documented fungal deterioration, the genera of bacteria isolated from historical textiles have been reported only in a few studies (Seves et al., 1998; Szostak-Kotowa, 2004). Different bacterial strains mainly cause staining of the fabrics. Stains appear due to the action of exopigments, which are secreted by cells and diffuse into the fabric (Gutarowska et al., 2016).

Table 1

Results of identification of bacteria and fungi isolated from the surfaces of suitcases and clothes.

N°	Species	Compared sequence	S [%]	Accession number
Bacteria				
1	<i>Bacillus atrophaeus</i>	<i>Bacillus atrophaeus</i> Y41	99	KF641819.1
2	<i>Bacillus cereus</i>	<i>Bacillus cereus</i> ML267	99	KC692161.1
3	<i>Bacillus licheniformis</i>	<i>Bacillus licheniformis</i> WAS3-5	99	JF496512.1
4	<i>Bacillus megaterium</i>	<i>Bacillus megaterium</i> GC61	99	KF158230.1
5	<i>Bacillus simplex</i>	<i>Bacillus simplex</i> IHB B 7066	99	KJ721216.1
6	<i>Bacillus subtilis</i>	<i>Bacillus subtilis</i> Van3	99	JX049584.1
7	<i>Enterobacter</i> sp.	<i>Enterobacter</i> sp. LB37	99	JQ692868.1
8	<i>Lysinibacillus fusiformis</i>	<i>Lysinibacillus fusiformis</i> KNUC423	99	JK071512.1
9	<i>Micrococcus luteus</i>	<i>Micrococcus luteus</i> 3A	99	KF993658.1
10	<i>Paenibacillus provencensis</i>	<i>Paenibacillus provencensis</i> 4401170	99	NR_044179.1
11	<i>Pseudomonas stutzeri</i>	<i>Pseudomonas stutzeri</i> TH-31	99	KF783212.1
12	<i>Psychrobacillus psychrodurans</i>	<i>Psychrobacillus psychrodurans</i> fwzy181	99	KF208476.1
13	<i>Staphylococcus cohnii</i>	<i>Staphylococcus cohnii</i> HNS003	99	JN128237.1
Fungi				
1	<i>Alternaria alternata</i>			
2	<i>Aspergillus flavus</i>			
3	<i>Aspergillus niger</i>			
4	<i>Chaetomium globosum</i>			
5	<i>Cladosporium cladosporioides</i>			
6	<i>Epicoccum nigrum</i>			
7	<i>Fusarium poae</i>			
8	<i>Penicillium chrysogenum</i>			
9	<i>Rhizopus nigricans</i>			
10	<i>Trichoderma viride</i>			
11	<i>Stephanoascus ciferrii</i>			
12	<i>mycelium sterillum</i>			

S – similarity; Accession number – GenBank, NCBI 16S rRNA gene sequence comparison.

Arthrobacter sp., *Bacillus* sp., *Corynebacterium* sp., *Pseudomonas* sp., and *Streptomyces* sp. are listed among the pigment-producing bacteria (Joshi et al., 2003; Pietrzak et al., 2016a). Studies by Seves et al. (1998), documented the ability of *Pseudomonas cepacia* to form a biofilm and hydrolyse fibroin, which may cause irreversible damage to silk artefacts of historical interest. Furthermore, some of the bacteria isolated from the A-BSM textiles surface may pose a danger to human health: *Enterobacter* sp. (group 2 of health risk according to Directive 2000/54/EC (the European Parliament and the Council of 18 September 2000 on the protection of workers from risks related to exposure to biological agents at work (seventh individual directive within the meaning of Article 16(1) of Directive 89/391/EEC) and regulation of the Polish Minister of Health of April 22, 2005) and *Bacillus subtilis* (group 2 according to the regulation of the Minister of Health of 22 April 2005). Among the fungi, some species known for their allergenic or toxic properties, such as *A. alternata*, *A. flavus*, *A. niger*, and *C. cladosporioides*, were isolated (Flannigan et al., 2001; Chapman, 2006).

3.2. VHP and EtO disinfection efficiencies

Disinfection using the VHP and EtO methods resulted in a significant reduction in the number of microorganisms (Table 2). A minimum 3 log reduction in the number of microorganisms is considered a good disinfection effect (EN 1650:2008; EN 1276:2009). In the case of VHP-1, VHP-2 and VHP-3, we observed for particular fungal strains that the number was 3.5–8.3 log reduced, and for bacteria, the reduction ranged between 1.5 and 8 log (Table 2). Longer times or higher VHP concentrations (VHP 2 and VHP 3) increased the

Table 2
Number of microorganisms on cotton fabric before (control) and after VHP and EtO disinfection.

Microorganism	Number of microorganisms [CFU/100 cm ²]				
	Control	VHP 1	VHP 2	VHP 3	EtO
<i>A. alternata</i>	$1.5 \times 10^8 \pm 9.5 \times 10^7$	$5.0 \times 10^4 \pm 2.4 \times 10^4^*$	$0 \pm 0^*$	$0 \pm 0^*$	$1.5 \times 10^3 \pm 3.4 \times 10^2^*$
<i>A. flavus</i>	$2.1 \times 10^8 \pm 2.4 \times 10^8$	$9.0 \times 10^1 \pm 1.1 \times 10^1^*$	$0 \pm 0^*\#$	$0 \pm 0^*\#$	$0 \pm 0^*$
<i>C. cladosporioides</i>	$6.0 \times 10^7 \pm 3.2 \times 10^7$	$0 \pm 0^*$	$0 \pm 0^*$	$0 \pm 0^*$	$2.0 \times 10^1 \pm 2.0 \times 10^1^*$
<i>E. nigrum</i>	$8.0 \times 10^7 \pm 5.5 \times 10^7$	$3.0 \times 10^1 \pm 4.0 \times 10^1^*$	$0 \pm 0^*\#$	$0 \pm 0^*\#$	$0 \pm 0^*$
<i>F. poae</i>	$4.0 \times 10^7 \pm 2.0 \times 10^7$	$0 \pm 0^*$	$0 \pm 0^*$	$0 \pm 0^*$	$0 \pm 0^*$
<i>P. chrysogenum</i>	$7.5 \times 10^7 \pm 6.1 \times 10^7$	$0 \pm 0^*$	$0 \pm 0^*$	$0 \pm 0^*$	$0 \pm 0^*$
Mix	$8.6 \times 10^7 \pm 3.5 \times 10^7$	$9.4 \times 10^6 \pm 7.2 \times 10^6$	$1.4 \times 10^6 \pm 8.6 \times 10^5$	$1.2 \times 10^6 \pm 9.0 \times 10^5$	$2.0 \times 10^3 \pm 8.2 \times 10^2^*$
<i>B. licheniformis</i>	$8.5 \times 10^6 \pm 4.0 \times 10^6$	$6.5 \times 10^3 \pm 7.6 \times 10^3^*$	$1.1 \times 10^3 \pm 1.3 \times 10^3^*$	$1.0 \times 10^3 \pm 9.6 \times 10^2^*$	$0 \pm 0^*$
<i>L. fusiformis</i>	$1.7 \times 10^6 \pm 9.2 \times 10^5$	$1.0 \times 10^4 \pm 8.5 \times 10^3^*$	$1.0 \times 10^2 \pm 1.5 \times 10^2^*\#$	$3.0 \times 10^2 \pm 1.5 \times 10^2^*\#$	$1.0 \times 10^1 \pm 1.0 \times 10^1^*$
<i>M. luteus</i>	$6.0 \times 10^5 \pm 1.8 \times 10^5$	$1.7 \times 10^4 \pm 1.5 \times 10^4$	$3.0 \times 10^3 \pm 2.8 \times 10^3^*$	$3.2 \times 10^3 \pm 7.5 \times 10^2^*$	$0 \pm 0^*$
<i>P. psychrodurans</i>	$1.1 \times 10^8 \pm 9.8 \times 10^7$	$2.3 \times 10^5 \pm 5.8 \times 10^4^*$	$0 \pm 0^*\#$	$5.0 \times 10^2 \pm 1.8 \times 10^2^*\#$	$2.7 \times 10^4 \pm 1.7 \times 10^4^*$
<i>S. epidermidis</i>	$1.8 \times 10^7 \pm 2.2 \times 10^7$	$3.0 \times 10^4 \pm 1.5 \times 10^4^*$	$1.3 \times 10^3 \pm 8.5 \times 10^2^*$	$2.3 \times 10^3 \pm 1.7 \times 10^3^*$	$0 \pm 0^*$
Mix	$9.6 \times 10^5 \pm 1.5 \times 10^5$	$1.2 \times 10^4 \pm 7.9 \times 10^3$	$1.1 \times 10^3 \pm 4.3 \times 10^2^*$	$1.7 \times 10^3 \pm 9.2 \times 10^2^*$	$0 \pm 0^*$

Mix – mixture of fungi or bacteria; VHP 1: 300 ppm, 20 min; VHP 2: 400 ppm, 20 min; VHP 3: 300 ppm, 30 min; * significantly different to control sample before disinfection; # statistical difference between VHP 1 and VHP 2/VHP 3; ANOVA with significance level $p < 0.05$.

effectiveness of the process at 7.6–8.3 log for certain fungal strains and 2.3–8.0 log for bacteria depending on the strain and parameters used. Analysis of the results showed that all applied disinfection conditions reduced the number of fungi and bacteria a statistically significantly amount compared to the control sample (Table 2). There were also statistically significant differences between the VHP 1 and VHP 2 conditions and between the VHP 1 and VHP 3 conditions for several species of fungi and bacteria. No significant differences were observed between VHP variants in the case of mixed fungal and bacterial cultures, so increasing the time or the VHP concentration gave a similar biocidal effect.

The antimicrobial effect obtained in the model studies of VHP and EtO disinfection is higher than that of other disinfection methods described in the literature, such as misting with silver nanoparticles (AgNPs, R = 31.6–99.9%) (Gutarowska et al., 2012; Pietrzak et al., 2017), low-temperature plasma LTP (R = 84.0–99.9%) (Szulc et al., 2017), and treatment with cinnamon essential oil (CEO) (R = 78.3–99.9%) (Matusiak et al., 2017). The conducted studies also showed that the effectiveness of VHP disinfection is higher than the disinfection of archival documents using the AgNP, LTP and thyme essential oil (TEO) techniques (Pietrzak et al., 2016b).

The effectiveness of VHP disinfection is comparable to that of fumigation with EtO. VHP disinfection resulted in a similar biocidal effect as EtO disinfection on fungi and a lower effect on bacteria. Previous studies on the EtO disinfection effectiveness also showed a high fungicidal efficacy against fungal strains on paper (Hanus et al., 1997; Michaelsen et al., 2013; Karbowska-Berent, 2014). However, it has been determined that ethylene glycol, generated during disinfection with EtO, is highly hygroscopic, which increases the moisture content in the material. Such conditions are conducive to the germination of fungal spores, which makes the material more susceptible to microbial growth (Valentin, 1986).

Satisfactory results for the reduction in the number of microorganisms have been reported for VHP and EtO disinfection: all filamentous fungi species and 4 out of 5 bacterial species were reduced to the level R = 99% (Table 3). The VHP disinfection method was more effective against fungi (R = 99.97–100%) than bacteria (R = 97.17–100%). The group of microorganisms (R = 99.99%) that are very sensitive to the VHP disinfection method include all the following filamentous fungi and bacteria: *P. psychrodurans*, *B. licheniformis*, *L. fusiformis*, and *S. epidermidis*. Only *M. luteus* belongs to medium-sensitivity microorganisms (R = 97.17%) of VHP disinfection (Table 3). The VHP disinfection method proved to be less effective against mixed cultures of fungi and bacteria (R = 89.07–99.88%) than monocultures.

The majority of previous studies on the effectiveness of VHP

Table 3
Reduction of microbial number after VHP and EtO disinfection.

Microorganism	Reduction [%]			
	VHP 1	VHP 2	VHP 3	EtO
<i>A. alternata</i>	99.97	100	100	99.99
<i>A. flavus</i>	99.99	100	100	100
<i>C. cladosporioides</i>	100	100	100	99.99
<i>E. nigrum</i>	99.99	100	100	100
<i>F. poae</i>	100	100	100	100
<i>P. chrysogenum</i>	100	100	100	100
Mix	89.07	98.37	98.57	100
<i>B. licheniformis</i>	99.92	99.98	99.99	100
<i>L. fusiformis</i>	99.42	99.99	99.99	99.99
<i>M. luteus</i>	97.17	99.50	99.80	100
<i>P. psychrodurans</i>	99.89	100	99.99	99.98
<i>S. epidermidis</i>	99.83	99.99	99.99	100
Mix	98.74	99.88	99.82	100

Mix – mixture of fungi or bacteria; VHP 1: 300 ppm, 20 min; VHP 2: 400 ppm, 20 min; VHP 3: 300 ppm, 30 min.

disinfection focused on pathogenic microorganisms, for which the very high effectiveness of this method has been demonstrated (R = 100%) (Rickloff and Oreliski, 1989; Neighbor et al., 1994). Earlier studies showed that moulds from the species *Aspergillus niger* and *Trichophyton mentagrophytes* are more susceptible to VHP than the bacteria *Staphylococcus aureus* and *Mycobacterium terrae* (Meszaros et al., 2005). This is consistent with the results for microbes isolated from historical textile materials obtained in this work.

Comparing the obtained results, it was found that the VHP 2 and VHP 3 disinfection conditions are suitable for obtaining satisfactory antimicrobial effects. However, the disinfection parameters should be determined after studying the disinfection effect on the material properties.

3.3. Influences of VHP and EtO on the textile properties

The results of the optical analysis show that disinfection with VHP and EtO caused slight colour changes in the new and historical cotton fabrics. The lightness of the new undisinfected (undyed) cotton fabric ($L^* = 97.83$) in the historical cotton fabric is much higher than the lightness of the contaminated fragments (background) ($L^* = 66.4–67.6$) (Table 4). The tested disinfection processes changed the lightness of undyed samples of the new cotton fabric as well as both the background and coloured stripes of the historical fabric. However,

Table 4
The colour changes of new and historical cotton fabric after VHP and EtO disinfection.

Tested section of textile	Sample		Optical properties							
			Colour shades			Colour differences				
			L*	a*	b*	ΔE^*	ΔL^*	Δa^*	Δb^*	
New cotton fabric										
background	untreated		97.83	-0.17	1.76	-	-	-	-	
	EtO		96.94	-0.11	1.64	0.91	-0.89	0.06	-0.12	
	VHP 1		97.26	-0.26	1.33	0.74	-0.57	-0.09	-0.43	
	VHP 2		97.36	-0.20	1.26	0.69	-0.47	-0.03	-0.50	
	VHP 3		98.52	-0.36	2.13	0.80	0.69	-0.19	0.37	
Historical cotton fabric										
background	EtO	before	67.41	4.14	18.33	-	-	-	-	
		after	67.70	4.16	18.16	0.34	0.29	0.03	-0.17	
	VHP1	before	67.62	4.00	17.61	-	-	-	-	
		after	67.45	3.42	16.50	1.26	-0.17	-0.58	-1.11	
	VHP2	before	66.42	3.85	17.29	-	-	-	-	
		after	66.45	3.25	16.11	1.32	0.03	-0.61	-1.18	
	VHP3	before	66.71	4.20	18.08	-	-	-	-	
		after	65.96	3.90	17.00	1.35	-0.75	-0.29	-1.09	
	black stripe	EtO	before	41.15	3.00	0.28	-	-	-	-
			after	40.65	2.99	0.14	0.52	-0.50	0.00	-0.14
		VHP1	before	41.54	3.51	-0.58	-	-	-	-
			after	42.01	3.67	-0.73	0.52	0.47	0.16	-0.15
VHP2		before	40.01	3.17	-0.36	-	-	-	-	
		after	40.55	3.28	-0.19	0.57	0.54	0.11	0.17	
VHP3		before	39.75	2.90	0.15	-	-	-	-	
		after	40.95	3.25	0.24	1.25	1.19	0.35	0.09	
violet stripe		EtO	before	46.16	7.28	-1.45	-	-	-	-
			after	47.11	7.28	-1.43	0.95	0.95	0.00	0.02
		VHP1	before	47.69	7.68	-2.36	-	-	-	-
			after	47.35	7.62	-2.81	0.56	-0.34	-0.06	-0.44
	VHP2	before	46.92	7.89	-1.83	-	-	-	-	
		after	48.07	7.90	-2.01	1.16	1.15	0.01	-0.18	
	VHP3	before	47.76	6.94	-0.04	-	-	-	-	
		after	48.00	6.62	-0.29	0.47	0.24	-0.32	-0.25	
	brown stripe	EtO	before	64.78	6.49	19.28	-	-	-	-
			after	63.93	6.78	19.64	0.98	-0.86	0.29	0.36
		VHP1	before	63.02	7.46	19.48	-	-	-	-
			after	63.93	7.35	19.10	1.00	0.91	-0.11	-0.38
VHP2		before	61.92	7.05	19.35	-	-	-	-	
		after	63.12	7.26	18.79	1.33	1.20	0.21	-0.55	
VHP3		before	52.44	8.64	18.35	-	-	-	-	
		after	52.71	9.49	18.66	0.95	0.27	0.87	0.31	

- not applied; EtO – ethylene oxide fumigation; VHP 1: 300 ppm, 20 min; VHP 2: 400 ppm, 20 min; VHP 3: 300 ppm, 30 min.

the changes in L^* are not greater than 1.2.

The undyed new and historical cotton samples disinfected with VHP and EtO showed a slight colour shift in the direction of the blue shade ($\Delta b^* < 0$). Only in the case of the new fabric sample disinfected by VHP 3 was an increase in the yellowness ($\Delta b^* > 0$) observed. A similar increase in the yellowness of the undyed cotton fabric was found after disinfection with AgNPs and with treatment by CEO vapour (Gutarowska et al., 2014; Matusiak et al., 2017).

In the case of new cotton fabric, the total colour change (ΔE^*) following the disinfection processes was less than 1.0, while for the historical fabric, the value of ΔE^* did not exceed 1.35. There is no clear correlation between the VHP concentration and the total colour change (ΔE^*) of either fabric disinfected by this method.

Disinfection by LTP (process gas oxygen) had a significantly larger effect on the colour of the cotton fabric – the parameter (ΔE^*) reached a value of 2.9 (Szulc et al., 2017).

VHP and EtO disinfection tests on samples of historical paper of various compositions carried out by Karbowska-Berent (2014) showed colour changes of $\Delta E^* = 0.75$ – 6.24 for VHP and $\Delta E^* = 0.19$ – 0.51 for EtO, although for most of the samples the change was below $\Delta E = 2.0$. No change in the lightness of the 4 paper types after EtO disinfection

was also shown by Hanus et al. (1997). In the current studies on historical textiles, it was also shown that the colour change after VHP disinfection (300 ppm 20 min) was smaller than in the studies of archival documents disinfected by three techniques: AgNPs, LTP, TEO (Pietrzak et al., 2016b).

Infrared absorption spectra (ATR-FTIR) of the untreated and disinfected new cotton fibres samples are presented in Fig. S1 in supplementary materials, while Fig. S2 shows the spectra of samples of the historical cotton fibres. A comparison of the infrared spectra shows that no significant changes occur in the spectral profiles of the undisinfected and disinfected cotton samples. There are no new bands associated with new functional groups generated in the molecular structure of cellulose during the disinfection processes using VHP and EtO. However, very slight differences in the spectra of the disinfected fabric samples, both new and historical, were observed compared to the spectra of the relevant control samples.

In Fig. S1, it can be observed that the intensity of the broad band centred at 3600 cm^{-1} , which is assigned to the hydroxyl group (OH) stretching vibrations, decreased slightly as a result of the disinfection performed using the VHP methods. The peaks in that spectral region are indicative of inter- and intramolecular hydrogen bonds between

Table 5

The breaking strength, elongation at break and degree of cellulose polymerisation of new cotton fabric after tested disinfection processes.

Sample	Degree of cellulose polymerisation	Mechanical properties	
		Elongation [%]	Breaking strength [N]
control	5526 ± 58	5.15 ± 0.06	156.00 ± 1.38
EtO	5934 ± 99*	4.85 ± 0.13*	153.15 ± 1.91
VHP 1	3889 ± 5*	4.84 ± 0.09*	150.45 ± 1.52*
VHP 2	3537 ± 97*\$	4.80 ± 0.08*	149.23 ± 1.75*
VHP 3	3385 ± 57*\$	4.63 ± 0.13*\$	147.50 ± 1.52*\$

Mean ± standard deviation; EtO – ethylene oxide fumigation; VHP 1: 300 ppm, 20 min; VHP 2: 400 ppm, 20 min; VHP 3: 300 ppm, 30 min; * significantly different to control sample before disinfection; \$ statistical difference between VHP 1 and VHP 2/VHP 3, ANOVA with significance level $p < 0.05$.

adjacent cellulose macromolecules (El-Gaoudy et al., 2011). The slight decrease in absorption indicated that in the disinfected cotton fibres, some hydrogen bonds were broken. Therefore, for the disinfected samples of the new cotton fabric, a deterioration of the mechanical strength and a decrease in the cellulose polymerisation degree were observed (Table 5).

In the case of cotton fibres, the region between 1750 and 1600 cm^{-1} is the most convenient for assessing cellulose degradation by oxidation (Abdel-Kareem, 2005). Cellulose oxidation products (aldehydes and carboxylic acids) resulting from the degradation of glycosidic linkages in cellulose macromolecules give rise to a prominent band in that spectral region. In Fig. S1, however, no spectral changes are observed between 1750 and 1600 cm^{-1} , which means that the tested disinfection processes did not oxidise cotton cellulose or form aldehyde and carboxyl groups. The same results of the FTIR spectral comparison within the range 1750–1600 cm^{-1} were also obtained for the cotton fabric before and after disinfection by LTP (Szulc et al., 2017). Similarly, in Fig. S2, in this spectral range, there are no differences between the absorption spectra of the control and historical cotton samples disinfected with VHP. Thus, hydrolysis of cotton cellulose seems to be the main mechanism of degradation caused by the tested disinfection methods.

In general, one can see that the intensity of the band at 1750–1600 cm^{-1} for the historical cotton fibres (Fig. S2) is much higher than for the new cotton fibres (Fig. S1). This is proof of the higher concentration of cellulose oxidation products in the historical cotton fibres compared to in the new fibres.

The two bands at 1032 cm^{-1} and 1060 cm^{-1} are assigned to C-O bridge stretching, while the band at 1111 cm^{-1} corresponded to C-O-C pyranose ring skeletal vibrations (Li et al., 2010). In the spectra of the historical fabric disinfected with VHP, a slight increase in all the above-mentioned absorption bands can be observed (Fig. S2). It is believed that this unexpected result (one could expect a reduction in the intensity of the absorption bands corresponding to the elements of the cellulose molecular structure) is caused by the destruction of non-cellulosic impurities (present on the historical fabric surface) during the disinfection process. As a result, the cotton fibres in this fabric became more exposed to the IR radiation beam when performing FTIR analysis, and the discussed absorption bands were strengthened.

The analysis of the obtained FTIR results indicates that disinfection of cotton fabric by the VHP method causes virtually no changes in the structure of cotton cellulose, so it does not affect the susceptibility of the material to biodeterioration. The results of the FTIR analyses of the cotton fabrics after disinfection with CEO vapour (Matusiak et al., 2017) as well as after disinfection by the LTP (Szulc et al., 2017) indicate that these processes cause only very small changes in the cellulose molecular structure.

The degree of cellulose polymerisation (Tables 5 and 6) in the new cotton fabric changed after VHP disinfection by different variants and

Table 6

The degree of cellulose polymerisation of historical cotton fabric after tested disinfection processes.

Sample	Degree of cellulose polymerisation
control	558 ± 17
EtO	586 ± 11*
VHP 1	533 ± 13*

EtO – ethylene oxide fumigation; VHP 1: 300 ppm, 20 min; * significantly different to control sample before disinfection.

EtO. In the case of VHP disinfection, the degree of cellulose polymerisation of the samples was reduced by 30–39%, and VHP 1 resulted in the smallest degree of cellulose polymerisation reduction. Both VHP and EtO disinfection of the new cotton fabric samples resulted in reductions in the breaking strength and elongation by 2–6% and 4–10%, respectively. A similar decrease in tensile strength (almost by 10%) was observed for cotton fabric after AgNPs misting (Gutarowska et al., 2014). Among all the tested disinfection variants, VHP 3 influenced the degree of polymerisation, elongation and breaking strength the most. The determined changes in the mechanical parameters of the new cotton fabric caused by the tested disinfection processes should be considered very small. More extensive deterioration of the tensile strength of cotton fabrics (by approximately 10%) was found after their disinfection by the CEO vapour (Matusiak et al., 2017). It should be noted that a decrease in the degree of cellulose polymerisation is reflected by the tensile strength properties only for cotton fibres with a significant degree of fibre damage. The tensile strength of the cotton fibres remained almost unchanged even though the average degree of cellulose polymerisation was reduced to approximately 1000 (Bilkova, 2012). In the case of the historical material, which is characterised by a much lower degree of cellulose polymerisation in the control sample, the reduction in the degree of cellulose polymerisation caused by disinfection with VHP 1 was 4.5%. The degree of cellulose polymerisation in both the new and historical materials after disinfection with EtO, in contrast to VHP, was higher than that of the control. LTP treatment had a different effect on the mechanical strength of the cotton fabrics than the other methods of disinfection. Studies have shown (Szulc et al., 2017) that the breaking force of cotton fabrics treated with LTP increases by approximately 15%. This increase in the strength of the fabric is due to a change in the microtopography of the cotton fibre surface, which increases their kinetic coefficient of friction.

The microscopic images (Figs. S3 and S4) of the surface of the cotton fibres from new and historical fabrics disinfected with the tested methods did not show any significant differences in comparison with cotton fibres from the corresponding fabrics before disinfection. All the fibres from the new cotton fabric, both untreated (control) and disinfected, showed the typical morphology of the cotton fibre surface. This indicates that the above-mentioned disinfection methods, under the conditions of the performed tests, caused no marked changes in the cotton fibre morphology. It is also shown that the exposure of cotton fabric to CEO vapour does not lead to visible changes in the surface state of the cotton fibres (Matusiak et al., 2017). It should be noted that the difference in mechanical strength of approximately 4–6% between untreated new cotton fabric samples and samples after treatment with VHP (Table 5) is not reflected by changes in the morphology of the fibre surface. Fig. S4 shows SEM images of the historical cotton fibre surface in the fabrics before and after disinfection. These images reveal a very high degree of contamination of the fibre surface in both the untreated and treated fabric samples. There are numerous impurities of different natures (organic, mineral) and origins as well as microorganisms and their spores on the surface. This contamination makes it impossible to observe the microtopography and morphology of individual cotton fibres in the historical fabric samples.

The main advantages of VHP disinfection are the cost and possibility

to perform it in any room, in contrast to disinfection with EtO, which requires the transport of objects outside the storage warehouse or the museum area. The cost of VHP disinfection is around € 4 per m³ depending on the total cubic capacity of the disinfected rooms. The cost of disinfection with EtO is approximately € 25 per metre of the current file (approximately 0.06 m³) or basket with a capacity of 0.144 m³. Moreover, VHP enables the quick and complex disinfection of enclosed rooms, including objects, surfaces and air, while EtO disinfects only the single objects located in the chamber. The main advantage of VHP over EtO is its good safety profile and environmental friendliness. VHP does not pose a threat to the environment because it disintegrates rapidly into oxygen and water vapour. In contrast, the viability of disinfection with EtO is doubtful due to its confirmed carcinogenic and mutagenic properties. After VHP disinfection, the room with the objects is immediately available. However, after disinfection with EtO, a quarantine of several days is required. VHP may be considered safe for historical cotton textiles and the environment.

4. Conclusions

The microbiological contamination of historical cotton textile objects (suitcases, clothes) from the Auschwitz-Birkenau State Museum was 3.1×10^4 – 4.6×10^4 CFU/100 cm² (bacteria) and 7.5×10^3 – 1.8×10^4 CFU/100 cm² (fungi). Most of the tested suitcases and clothes showed low or medium levels of microbial contamination. The VHP disinfection method was more effective against fungi than bacteria. The VHP disinfection method proved to be less effective against mixed cultures of fungi and bacteria than monocultures. VHP disinfection resulted in a biocidal effect similar to that of EtO disinfection for fungi and a lower effect for bacteria. The following optimal parameters of effective VHP disinfection of historical cotton textiles were established: concentration, 300 ppm, and time, 20 min.

Disinfection with VHP did not change the colour of the historical cotton fabric, the chemical composition, the morphology, or the degree of cellulose polymerisation to an extent deemed unacceptable by conservators. It is an effective disinfection method that can be applied in mass. However, the final decision on the application of VHP in the disinfection of historical cotton textiles should be made by conservators after analysis of the objects. Moreover, disinfection methods should be always selected individually for specific historical objects, as they have different effects on various materials.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.ibiod.2018.05.016>.

References

- AATCC Test Method 100-2012. Antibacterial finishes on textile materials: assessment of Abdel-Kareem, O., 2005. The long-term effect of selected conservation materials used in the treatment of museum artefacts on some properties of textiles. *Polym. Degrad. Stabil.* 87, 121–130.
- Abdel-Kareem, O., 2010. Fungal deterioration of historical textiles and approaches for their control in Egypt. *e-Preservation Sci.* 7, 40–47.
- Bilkova, L., 2012. Application of infrared spectroscopy and thermal analysis to the examination of the degradation of cotton fibers. *Polym. Degrad. Stabil.* 97, 35–39.
- Chapman, M.D., 2006. Challenges associated with indoor moulds: health effects, immune response and exposure assessment. *Med. Mycol.* 44, 529–532.
- El-Gaoudy, H., Kourkoumelis, N., Varella, E., Kovala-Demertzi, D., 2011. The effect of thermal aging and color pigments on the Egyptian linen properties evaluated by physicochemical methods. *Appl. Phys. A* 105, 497–507.
- EN 1276, 2009. Chemical Disinfectants and Antiseptics. Quantitative Suspension Test for the Evaluation of Bactericidal Activity of Chemical Disinfectants and Antiseptics Used

- in Food, Industrial, Domestic and Institutional Areas. Test Method and Requirements (Phase 2, Step 1).
- EN 1650, 2008. Chemical Disinfectants and Antiseptics. Quantitative Suspension Test for the Evaluation of Fungicidal or Yeast-like Activity of Chemical Disinfectants and Antiseptics Used in Food, Industrial, Domestic and Institutional Areas. Test Method and Requirements (Phase 2, Step 1).
- EN ISO 105–J01, 1999. Textiles. Tests for Colour Fastness. General Principles for Measurement of Surface Colour.
- EN ISO 13934–1, 2013. Textiles. Tensile Properties of Fabrics. Determination of Maximum Force and Elongation at Maximum Force Using the Strip Method.
- Flannigan, B., Samson, R.A., Miller, J.D., 2001. Microorganisms in Home and Indoor Work Environments: Diversity, Health Impacts, Investigation and Control. Taylor & Francis, London and New.
- Gutarowska, B., Rembisz, D., Zduniak, K., Skóra, J., Szykowska, M., Gliścińska, E., Koziróg, A., 2012. Optimization and application of the misting method with silver nanoparticles for disinfection of the historical objects. *Int. Biodeterior. Biodegrad.* 75, 167–175.
- Gutarowska, B., Pietrzak, K., Machnowski, W., Danielewicz, D., Szykowska, M., Konca, P., Surma-Ślusarska, B., 2014. Application of silver nanoparticles for disinfection of materials to protect historical objects. *Curr. Nanosci.* 10, 277–286.
- Gutarowska, B., Pietrzak, K., Machnowski, W., Milczarek, J.M., 2016. Historical textiles – a review of microbial deterioration analysis and disinfection methods. *Textil. Res. J.* 87, 2388–2406.
- Hanus, J., Minarikova, J., Durovic, M., Bacilkova, N., 1997. Influence of ethylene oxide sterilization on some properties of different types of paper. In: ARSAG, La Conservation: une science en evolution. Bilan et Perspectives, Actes des Troisiemes Journées Internationales d'Etudes de L'Arsag, (Paris).
- Hultman, C., Hill, A., McDonnell, G., 2007. The physical chemistry of decontamination with gaseous hydrogen peroxide. *Pharmaceut. Eng. Mag.* 27, 22–32.
- Iwaszko, T., 2000. The housing, clothing and feeding of the prisoners. In: Długoborski, W., Piper, F. (Eds.), *Auschwitz, 1940-1945 Central Issues in the History of the Camp, T2-the Prisoners, Their Life and Work*. Auschwitz-Birkenau State Museum, Oświęcim.
- Joshi, V.K., Attri, D., Bala, A., Bhushan, S., 2003. Microbial pigments. *Indian J. Biotechnol.* 2, 362–369.
- Karbowska-Berent, J., 2014. Dezynfekcja Chemiczna Zabytków Na Podłożu Papierowym - Skuteczność i Zagrożenia. Wydawnictwo Naukowe Uniwersytetu Mikołaja Kopernika, Toruń, in polish.
- Kavkler, K., Gunde-Cimerman, N., Zalar, P., Demsar, A., 2015. Fungal contamination of textile objects preserved in Slovene museums and religious institutions. *Int. Biodeterior. Biodegrad.* 97, 51–59.
- Klapes, N.A., Vesley, D., 1990. Vapour-phase hydrogen peroxide as a surface decontaminant and sterilant. *Appl. Environ. Microbiol.* 56, 503–506.
- Koziróg, A., Otlewska, A., Piotrowska, M., Rajkowska, K., Nowicka-Krawczyk, P., Hachułka, M., Wolski, G.J., Gutarowska, B., Kunicka-Styczyńska, A., Libudzisz, Z., Żakowska, Z., Żydzik-Białek, A., 2013. Colonizing organisms as a biodegradation factor on historical wood materials at the former concentration camp of Auschwitz II-Birkenau. *Int. Biodeterior. Biodegrad.* 86, 171–178.
- Krakova, L., Soltys, K., Otlewska, A., Pietrzak, K., Purkrova, S., Savicka, A., Puskarova, A., Buckova, M., Szemes, T., Budis, J., Demnerova, K., Gutarowska, B., Pangallo, D., 2017. Comparison of methods for identification of microbial communities in book collections: culture-dependent (sequencing and MALDI-TOF MS) and culture-independent (Illumina MiSeq). *Int. Biodeterior. Biodegrad.* <https://doi.org/10.1016/j.ibiod.2017.02.015> (in press).
- Lech, T., 2017. The impact of high-density polyethylene materials on microbiological purity in the process of storing and preserving textiles. *Textil. Res. J.* 87, 2076–2088.
- Li, L., Frey, M., Browning, K., 2010. Biodegradability study on cotton and polyester fabrics. *J. Eng. Fibers Fabr.* 5, 42–52.
- Matusiak, K., Machnowski, W., Wrzosek, H., Polak, J., Rajkowska, K., Śmigielski, K., Kunicka-Styczyńska, A., Gutarowska, B., 2017. Application of *Cinnamomum zeylanicum* essential oil in vapour phase for heritage textiles disinfection. *Int. Biodeterior. Biodegrad.* 2017. <https://doi.org/10.1016/j.ibiod.2017.02.011>.
- Meszáros, J.E., Antloga, K., Just, C., Plesnicher, C., McDonnell, G., 2005. Area fumigation with hydrogen peroxide vapor. *Appl. Biosaf.* 10, 91–100.
- Michaelsen, A., Pinzari, F., Barbabietola, N., Piñar, G., 2013. Monitoring the effects of different conservation treatments on paper-infecting fungi. *Int. Biodeterior. Biodegrad.* 84, 333–341.
- Mokrzycki, W.S., Tatol, M., 2011. Color difference ΔE: a survey. *Mach. Graph. Vis.* 20, 383–411.
- Neighbor, N.K., Newbury, L.A., Bayyari, G.R., Beasley, J.N., McNew, R.W., 1994. The effect of microaerosolized hydrogen peroxide on bacterial and viral poultry pathogens. *Poultry Sci.* 73, 1511–1516.
- Pangallo, D., Krakova, L., Chovanova, K., Buckova, M., Puskarova, A., Simonovicova, A., 2013. Disclosing a crypt: microbial diversity and degradation activity of the microflora isolated from funeral clothes of Cardinal Peter Pazmany. *Microbiol. Res.* 168, 289–299.
- Paulus, W., 2004. *Directory of Microbicides for the Protection of Materials – a Handbook*. Kluwer Academic Publishers, Dordrecht.
- Pawcenis, D., Jacob, L.T., Łojewski, T., Milczarek, J.M., Łojewska, J., 2015. Towards determination of absolute molar mass of cellulose polymer by size exclusion chromatography with multiple angle laser light scattering detection. *J. Chromatogr. A* 1409, 53–59.
- Pekhtasheva, E., Neverov, A., Kubica, S., Zaikov, G., 2012. Biodegradation and biodegradation of some natural polymers. *Chem. Chem. Technol.* 6, 263–280.
- Pietrzak, K., Otlewska, A., Puchalski, M., Gutarowska, B., Guiamet, P., 2016a. Antimicrobial properties of silver nanoparticles against biofilm formation by *Pseudomonas aeruginosa* on archaeological textiles. *Appl. Environ. Biotechnol.* 1, 1–9.

- Pietrzak, K., Otlewska, A., Danielewicz, D., Dybka, K., Pangallo, D., Kraková, L., Puskárová, A., Bucková, M., Scholtz, V., Durovic, M., Surma-Ślusarska, B., Demnerová, K., Gutarowska, B., 2016b. Disinfection of archival documents using thyme essential oil, silver nanoparticles misting and low temperature plasma. *J. Cult. Herit.* 24, 69–77.
- Pietrzak, K., Puchalski, M., Otlewska, A., Wrzosek, H., Guiamet, P., Piotrowska, M., Gutarowska, B., 2017. Microbial diversity of pre-Columbian archaeological textiles and the effect of silver nanoparticles misting disinfection. *J. Cult. Herit.* 23, 138–147.
- Pitt, J.I., Hocking, A.D., 2009. *Fungi and Food Spoilage*. Springer, LLC.
- Rajkowska, K., Otlewska, A., Koziróg, A., Piotrowska, M., Nowicka-Krawczyk, P., Hachułka, M., Wolski, G.J., Kunicka-Styczyńska, A., Gutarowska, B., Żydzik-Białek, A., 2014. Assessment of biological colonization of historic buildings in the former Auschwitz II-Birkenau concentration camp. *Ann. Microbiol.* 64, 799–808.
- Rickloff, J.R., Oreliski, P.A., 1989. Resistance of various micro-organisms to vaporized hydrogen peroxide in prototype tabletop sterilizer. In: 89th Annual Meeting of the ASM, (New Orleans).
- Samson, R.A., Hoekstra, E.S., Frisvad, J.C., 1996. *Introduction to Foodborne Fungi*. Centraalbureau voor Schimmennutturees, Baarn.
- Sequeira, S., Cabrita, E.J., Macedo, M.F., 2012. Antifungals on paper conservation: an overview. *Int. Biodeterior. Biodegrad.* 74, 67–86.
- Seves, A., Romanò, M., Maifreni, T., Sora, S., Ciferri, O., 1998. The microbial degradation of silk: a laboratory investigation. *Int. Biodeterior. Biodegrad.* 42, 203–211.
- Sójka-Ledakowicz, J., Walawska, A., Filipowska, B., Lewartowska, J., Olczyk, J., Kiwała, M., 2015. New eco-friendly method of cellulosic product bleaching with simultaneous disinfection. *Fibres Text. East. Eur.* 3, 115–119.
- Strzelecki, A., 2000. Plundering the victims' property. In: Długoborski, W., Piper, F. (Eds.), *Auschwitz, 1940-1945 Central Issues in the History of the Camp, T2-the Prisoners, Their Life and Work*. Auschwitz-Birkenau State Museum, Oświęcim.
- Szostak-Kotowa, J., 2004. Biodeterioration of textiles. *Int. Biodeterior. Biodegrad.* 53, 165–170.
- Szulc, J., Urbaniak-Domagala, W., Machnowski, W., Wrzosek, H., Łacka, K., Gutarowska, B., 2017. **Low temperature plasma for textiles disinfection**. *Int. Biodeterior. Biodegrad.* <http://doi.org/10.1016/j.ibiod.2017.01.021> (in press).
- Valentin, N., 1986. Biodeterioration of library materials disinfection methods and new alternatives. *J. Pap. Conserv.* 10, 40–45.
- Wolf, S.J., 2002. Appendix K. in: *NPS Museum Handbook. Part I. National Park Service*, Washington.
- Łojewski, T., Zięba, K., Łojewska, J., 2010. Size exclusion chromatography and viscometry in paper degradation studies. New Mark-Houwink coefficients for cellulose in cupri-ethylenediamine. *J. Chromatogr. A* 1217, 6462–6468.