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BIODETERIOGENIC CAPACITY OF A MICROFUNGAL SPECIES ISOLATED FROM TEXTILE CULTURAL HERITAGE ITEMS ON CONTEMPORARY WOOL MATERIALS

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Microorganisms are omnipresent in the ambient and have the ability to colonize diverse natural habitats depending on the temperature, humidity, pH, salt concentration etc. Microfungi are equipped with a rich enzymatic system, being able to degrade various substrates, from inorganic to organic. If from the biotechnological point of view, the microfungal bio-degradative capacities bring benefits to human economy and are exploited and improved, in the Cultural Heritage field, the microfungi produce the biodeterioration process, which can leads to a complete degradation and loss of the heritage good. Textile Heritage items made of natural fibres are continuously exposed to the biological risk represented by the fungal contamination. This risk can manifest solely or can be synergic with other types of degradation such as physical and chemical. In our study, an accelerated *in vitro* biodeterioration test was performed using some microfungi strains isolated form The National Museum for Romanian Peasant (Bucharest) on a textile material made of 100% wool fibres. After the incubation period, the biodeterioration effect of the microfungal strains was assessed through macroscopical, microscopical, physical-mechanical testing, infrared spectrophotometry and electron microscopy. The results of the study showed that the wool textile material presented a high degradation, especially when the microfungi were cultivated in a rich glucose culture medium. The biodeterioration of the wool woven could be observed at all levels: macroscopically, microscopical and structural level.

Keywords: natural fibers**,** wool, Cultural Heritage, microfungi, *Penicillium*, biodeterioration, heritage textiles, ethnographic textiles, FTIR.

INTRODUCTION

The process of biodeterioration is currently seen as a risk factor regarding long time conservation of heritage collections[1]. Heritage Textiles made of natural fibers, such as wool, hemp, flax, cotton can be severely damage by microfungi, especially if they are kept in high humidity. The biodeterioration process can produce an aesthetic damage but also can induce structural modifications to the natural biopolymers[2]. Microorganisms can deposit on the surface of materials different metabolic products that modify the local pH, towards an acidic value, favorable for other types of deteriorations[3]. The colonization of the organic substrates from the Textiles Heritage collections by microorganisms and their further development are influenced by many factors, such as atmospheric humidity, the water content of the material, the chemical composition of the substrate, the degree of illumination[4-6]. Thus, the investigation of microorganisms inducing biodeterioration and of the biological, chemical and physical mechanisms that accompany this process can offer an important insight for the development of proper conservation strategies.

Among the natural fibers, wool has been widely used since early times either for clothing or house items, such as carpets.

In our study an *in vitro* biodeterioration process was reproduced using a fungal strain belonging to *Penicillium* genera. The strain was isolated from a carpet made of wool exposed at the National Museum for Romanian Peasant (Bucharest). The biodeterioration study was done on a modern woven made of 100% wool fibres. The assessment of the fungal biodeterioration by

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means of macroscopically, microscopically, physical-mechanical testing, infrared spectrophotometry (FTIR), electron microscopy and mathematical bivariate analysis (not presented here) revealed changes at morphological and structural levels of the wool fibers material.

MATERIALS AND METHODS

TEXTILE MATERIAL

For the biodeterioration *in vitro* testing it was chosen an woven made of 100% wool fibres. The physical-mechanical characteristics of the woven are presented in Table 1.

BIODETERIORATION TEST

The biodeterioration testing was done accordingly to SR EN 14119: 2004[7]. The textile specimens were exposed to a fungal cell density during a period of 28 days at 300C. Regulary at 3, 7, 14, 21 and 28 days macroscopical and microscopical observations were made in order to observe the growth of the fungal mycelium on the surface of woolen specimens. The intensity of growth was correlated with marks from 0 to 5, were 0 means no growth of the microfungi and 5 means 100% coverage of the specimen with fungal mycelium.

The fungal strain used in the textile biodeterioration study was *Penicillium crustosum* (Fig. 1), being isolated from a woolen carpet from the Heritage Texttile Colection of the National Museum for Romanian Peasant, from Bucharest, Romania. *P. crustosum* was inoculated on Czapek-Dox agar medium suplimented with glucose and on Czapek-Dox agar medium without glucose. The density of the inoculum was established at 10^6 CFU/ml using the hemacytometer method.

PHYSCAL-MECHANICAL TESTING

After the biodeterioration test, the wool fibers specimens were sterilised in autoclave and treated with 70% ethanol solution, cleaned in tap water, and dried at room temperature. The specimens togheter with a blank, not biodeteriorated specimen, were tested for the maximum breaking force and ellongation at break according to SR EN ISO 13934-1:2002 standard[8]. Before testing, all specimens were kept in standard temperature and humidity according to SR EN 139:2005[9].

Table 1

Physical-mechanical characteristics of the woolen material

Characteristic (unit)		Value
Mass (g/m^2)		72
Density (threads /10 cm)	*warp*	180
	weft	170
Breaking force (N)	*warp*	132.8
	weft	120.9
Elongation at break (%)	*warp*	11.00
	weft	15.60
Thickness (mm)		0.32
Air permeability (l/m^2/s)		3454
Water vapors permemability (%)		37.7

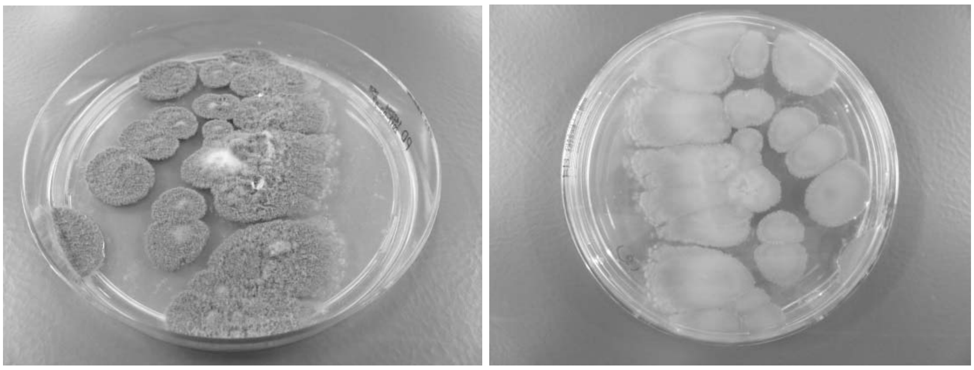

Figure 1. *Penicillium crustosum* strain used for the *in vitro* biodeterioration.

FTIR ANALYSIS

The FTIR investigation was done with *FTS Excalibur 3000* (*Digilab*, SUA), in transmitance mode (T%), and the scans were made in the range of 4000 cm^{-1} to 400 cm^{-1} wave number.

MACROSCOPICAL AND MICROSCOPICAL OBSERVATIONS

The surface modifications of the wool fibers were analysed by SEM- *Scanning electron microscopy*, using the *Quanta 200* (*FEI*, Netherlands) and optical microscopy using *Stereo Discovery.V8* (*Carl Zeiss*, Germany) and *AxioImager A2* (*Carl Zeiss*, Germany) microscopes.

RESULTS AND DISCUSSIONS

The *in vitro* biodeterioration test was realised using a species of the *Penicillium genera*, *P. crustosum* which was previously isolated from an ethnographic heritage textile[10]. Cultivated on a carbon rich culture medium and a carbon poor culture medium *P. crustosum* showed two growth intensity patters of wool biodeterioration. On the carbon rich medium the fungal strain developped quickly starting even from the 3rd day of incubation. Thus, until the end of the testing period the percentage of coverage of the wool specimen was of 100% (Tabel 1).

Without the carbon source the mycelium was poorly developed, presenting mainly hiphae without reproductive structures, thought the percentage of coverage was over 75% of the wool specimen surface.

On the rich carbon source medium, *P. crustosum* formed colonies with a dense mycelium and with specific morphological characteristics such as the color and the texture, even from the 3rd day of incubation period (Fig. 2).

The *in vitro* biodeterioration proces determined by the *P. crustosum* strain was intense for the wool material in the presence of a carbon source. The analysis of the physical-mechanical parameters revealed the loss of breaking force of over 85% and a loss of elongation to break of over 60% comparing with the non-biodeteriorated specimen (Fig. 3 and Fig. 4).

The most important structure of the wool fibre is the cortex. The characteristics of the cortex cells give specific physical-mechanical and chemical properties to wool. The main chemical component of the wool is cheratine which is a biopolymer made of different amino acids with varying composition depending on the structural level of the fibre. In wool fibres, the most prevalent aminoacids are cysteine, arginine and serine[11].

The FTIR spectrofotometry investigations of the biodeteriorated wool specimen showed a clear modification of the transmitance bands in the region of 3444 cm^{-1} and 739 cm^{-1} (Fig. 5 and Fig. 6). The 3444 cm^{-1} region corresponds with valence vibration ν(O-H)[12] and the region of 739 cm^{-1} corresponds to deformation vibration δ(C-H) from hydrocarbons like alkanes, alkenes, aromatic hydrocarbons.

Tabel 2

The marks reflecting the percentage of coverage of the textile materials with mycelium of *P. crustosum*

Culture medium	Marks according to the percentage of coverage of the textile surface				
	3 days	**7 days**	**14 days**	**21 days**	**28 days**
Medium with glucose	3–4	4	4–5	4–5	4–5
Medium without glucose	2–3	2-3	3	3	4

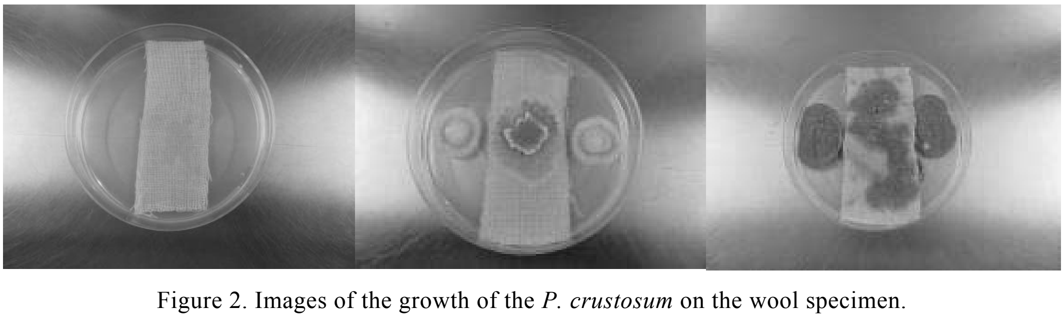

Figure 2. Images of the growth of the *P. crustosum* on the wool specimen.
(*a*) Initial image; (*b*) day 3; (*c*) day 28. Medium with glucose.

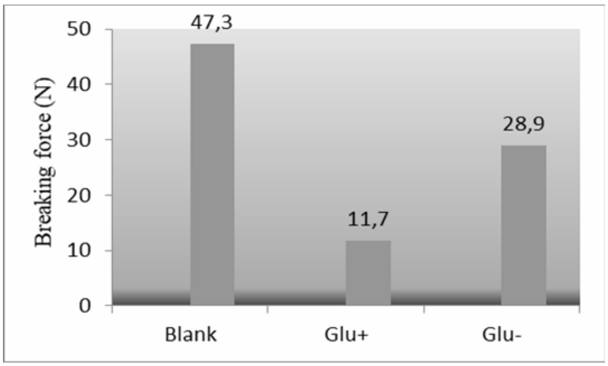

Figure 3. The breaking force (N) for the wool specimens: the blank nonexposed and the biodeteriorated wool specimens cultivated on medium with glucose (Glu+) and without glucose (Glu-).

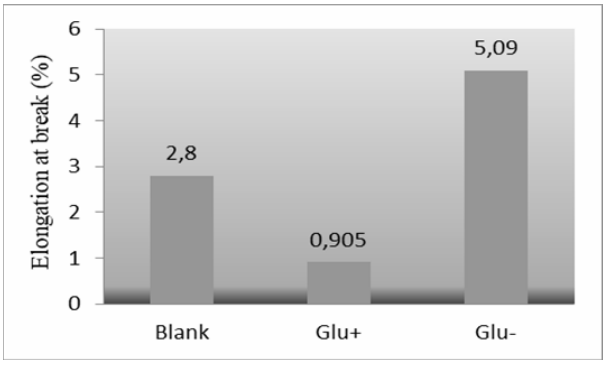

Figure 4. The elongation at break (%) for the wool specimens: the blank nonexposed and the biodeteriorated wool specimens cultivated on medium with glucose (Glu+) and without glucose (Glu-).

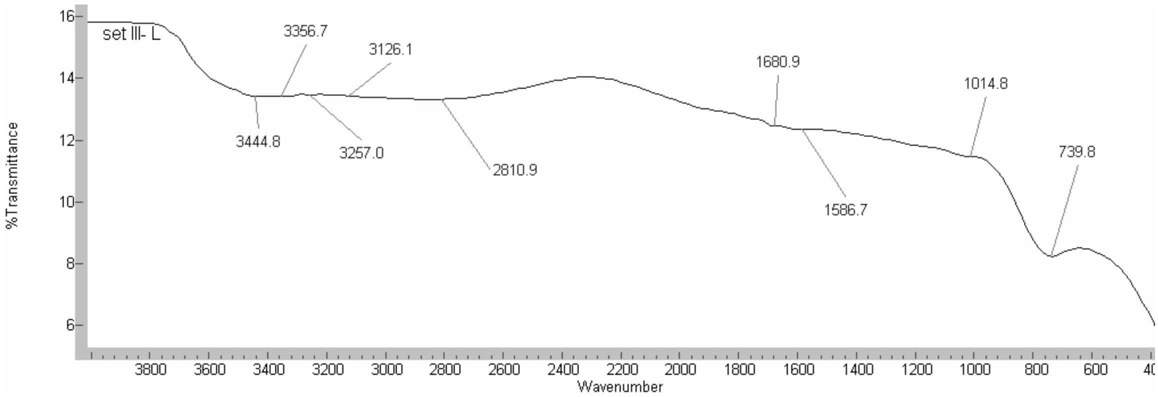

Figure 5. The FTIR spectrum of the wool specimen non-biodeteriorated.

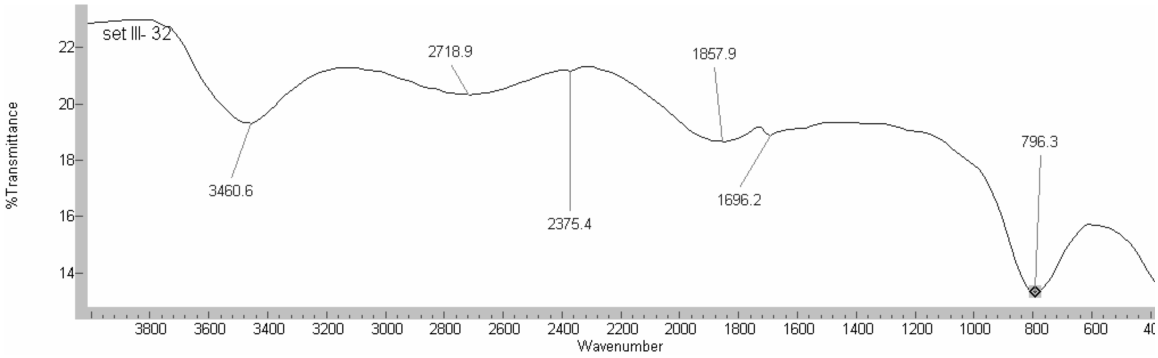

Figure 6. The FTIR spectrum of the wool specimen biodeteriorated by *P.crustosum*.

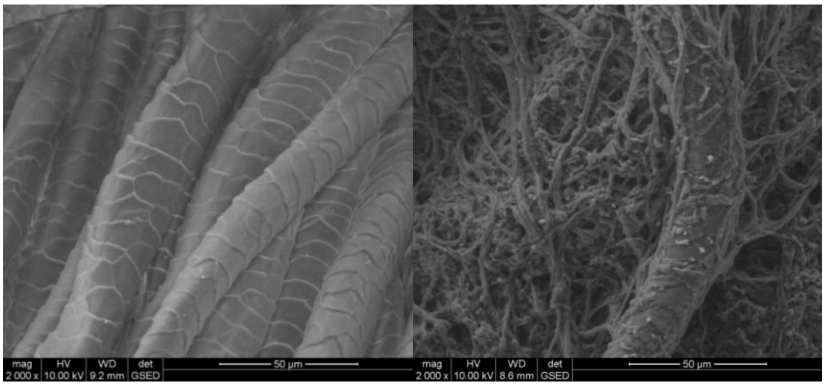

Figure 7a,b. Micrographs of the non-biodeteriorated wool specimen (left) and biodeteriorated wool specimen (right).

The surface of the biodeteriorated wool fibres were clearly degraded, with the culticle cells deformed and covered with organic material (Fig. 7). The microscopical degradation of the wool fibres accompany the macroscopical process of deterioration. Thus, some samples of tested wool specimen presented yarns and fiber breakage at the end of the *in vitro* biodeterioration process.

CONCLUSIONS

The process of wool fibres biodeterioration was reproduced at lab scale using a fungal strain, *Penicillium crustosum*, which was isolated from a museum environment. At the end of *in vitro* exposure, the textile material made of 100% wool fibres presented morphological and structural changes, which were more intensive when the fungal strain was cultivated on a carbon rich culture medium.

The study shows the importance of microfungi in the biodeterioration of natural fibres, especially the ones that are part of textile heritage goods. The complete understanding of the bio-chemical, chemical and physical mechanisms of the fungal induced textile biodeterioration can offer an optimum methodological approach for the preventive and curative conservation of the Textiles Heritage Collections.

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