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Microbiological and Chemical Analysis of Bamboo Textile Materials and Leathers Modified with Bamboo Extract at the Tanning Stage

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Abstract

In this paper, microbiological and chemical analysis were carried out in the case of bamboo textile materials and leathers modified by bamboo extract at the tanning stage. Microbiological resistance was examined for some fungi, i.e.: Aspergillus niger, Trichophyton mentagrophytes and Candida albicans and some bacteria strains: Escherichia coli, Salmonella enteritidis and Pseudomonas aeruginosa. In parallel, a safety analysis was conducted through the determination of heavy metals, certain aromatic amines and dimethyl fumarate. The main goal of the above-mentioned research was a comprehensive examination of materials which will be used as footwear components i.e.: linings, uppers and insoles. These issues are very important from the footwear manufacture point of view because of the opportunity to find new solutions in the field of hygienic and healthy materials which can be applied as footwear elements. The anti-microbial and anti-fungal resistance of materials are features important for the reduction of the probability of dermatosis. For this reason, they should be taken into account when the improvement of hygienic properties is pursued.

Key words: *bamboo fibres, extract, leather, microbiological.*

Introduction

Textile or leather materials used for footwear construction require the monitoring of microbiological conditions from the first stage of their lifecycle, that is, the raw material acquisition and selection. The optimal quality control process should include not only the final products, as occurs for textiles, but also focus on the number of proceeding processes connected with finishing aspects. For example, in the case of leather, the tanning of raw hides endows the material with fungi or microorganism resistance; but full antimicrobial protection is not guaranteed. This aspect is very important especially when the specific temperature and humidity conditions inside a shoe volume will be taken into account.

In accordance with the growing awareness of consumers, more and more new materials with improved functionalities are put on the market. Literature sources give a wide spectrum of methods used for enriching footwear materials – both leathers and textiles. One of the methods includes the use of the following additives: bioactive silver ions, aloe extract [1], fibres from coconut shells [2], natural minerals, and essential oil encapsulation [3]. Another type of material modification is shown by the authors in [6-8], where leather was protected against UV-A and UV-B aging by using optical brightened agents.

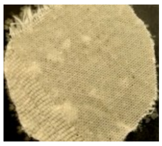
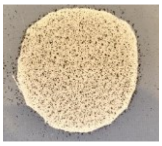
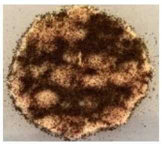
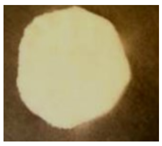
All activities aimed at the development of footwear material functionality are translated into the improvement of some physico – mechanical and hygienic parameters of materials. This is very important from the user's point of view because the changes are observable in a wide range of parameters, such as water vapour permeability, water vapour absorption, and material behaviour in contact with water. These aspects have an impact on thermal and humidity conditions inside a shoe during usage.

Another important aspect of footwear component modifications is the antimicrobial activity of materials. The microbiological purity of a shoe interior is one of the factors which decide the reduction of the probability of the occurrence of dermatosis, ulcers or other infections. Under real conditions of use, footwear materials are a barrier against moisture and temperature flow. Thus, when water condenses on the skin and lining materials, the temperature rises, and the specific conditions can affect the foot's microbiom in the closed space around it [4]. Literature sources show that the relative humidity inside a shoe volume at a level greater than 90%, causes a rapid increase in sweat – degrading bacteria [9]. On the other hand, the microbiological contamination of shoe materials may cause the degradation and disintegration of the footwear construction. Therefore, the correct selection of

Table 1. Basic characteristics of textile bamboo materials.

Sample name	Type of textile product	Mass per square metre, g/m^2	Thickness, mm	Percentage of fibres, %
M1	woven fabric	170	0.40	100% bamboo
M2	woven fabric	500	1.74	100% bamboo
M3	woven fabric	300	0.61	95% bamboo, 5% polyester
M4	woven fabric	170	0.35	50% bamboo, 50% flax
M5	knitted fabric	290	1.39	85% bamboo, 15% polyester
M6	knitted fabric	220	0.38	95% bamboo, 5% elastane

Table 2. Anti-fungal activity of textile materials M1 – M6 (5 – denotes the intensive growth, where fungi covers the whole sample area; 1 – denotes small growth).

Sample name	*Aspergillus niger*		*Trichophyton mentagrophytes*	
	Intensity of growth (unaided eye view)		Intensity of growth (unaided eye view)	
Control sample: cotton woven fabric	5		5	
M1	5		5	
M2	5		1	
M3	5		5	
M4	5		5	
M5	5		5	
M6	5		5	

footwear and construction materials is a guarantee of users; safety [5].

The main goal of this paper was the analysis of microbiological aspects of bamboo textile materials and leathers modified by bamboo extract. The research was conducted in order to make a holistic quality evaluation of these materials from the footwear construction perspective. Use of the above-mentioned materials, based on natural components, was focused on the improvement of the hygienic safety and microbiological purity of the shoe interior, which is a very important factor in the proper development of children's feet [10, 11]. The intention of the authors was the implementation of knowledge about bamboo fibres and bamboo extract properties [12-15] in a new field of footwear production.

The in-depth assessment which was undertaken in the project entitled: "Use of bamboo extract and fibres in the elements of leather, textile and combined leather and textile children's footwear" completed with the development of a footwear prototype. The new shoes were characterised by better properties – especially in hygienic aspects. The final product was completely safe for the health and environment, which was confirmed by the analysis of the hazardous substance (i. e. heavy metals) content.

The research conducted in this paper matches current trends by focussing on improving the safety of goods for humans and the environment. The microbiological aspect, which was underlined in this research, completes a holistic view of the possibility of the application of bamboo textiles and leathers modified by bamboo extract for children's footwear.

Materials and method

Materials

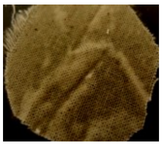
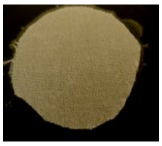
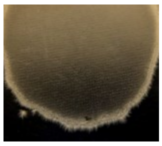
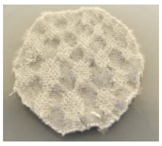
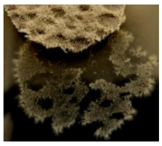
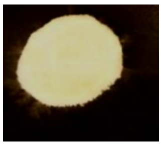
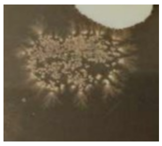
These studies were carried out on the following group of textiles: four woven and two knitted fabrics based on bamboo fibres. The thickness of these materials was in the range of 0.35 to 1.74 mm, and the mass per square metre was between 170 and 500 g/m^2. The bamboo fibre content in each material was greater than 50%. The basic characteristics of the textiles used are listed in ***Table 1***. For leather materials, modifications of semi-finished bovine lining leathers (with thickness 1.1-1.2 mm) and pig upper leathers (with thickness 3.9-4.1 mm) were conducted. Bamboo extract was used as a modification agent, consisting of about 70% silica recovered from stem and leaf. The percentage of bamboo extract additive in relation to the leather mass was equal to 1, 2 and 5%.

Methodology of research

Methodology of microbiological analyses

The antifungal resistance of the textiles was evaluated with the use of the method described in Standard PN-EN 14119:2005 'Testing of textiles – Evaluation of the action of microfungi – visual method A1'. Samples with a diameter of 40 mm, were treated with a spore sus-

Table 3. *Resistance of textile samples to Candida albicans.*

Sample name	Qualitative assessment of growth kinetics on agar substrate			Quality evaluation of material
Control sample: cotton woven fabric	Growth on the sample was invisible to the unaided eye and under a microscope. Growth visible around the edges and under the sample.			Lack of fungistatic effect
M1	Growth on the sample was invisible to the unaided eye and under a microscope. Growth visible around the edges and under the sample.			Lack of fungistatic effect
M2	Growth on the sample was invisible to the unaided eye and under a microscope. Growth visible around the edges and under the sample.			Lack of fungistatic effect
M3	Growth on the sample was invisible to the unaided eye and under a microscope. Growth visible around the edges and under the sample.			Lack of fungistatic effect
M4	Growth on the sample was invisible to the unaided eye and under a microscope. Growth visible around the edges and under the sample.			Lack of fungistatic effect
M5	Growth on the sample was invisible to the unaided eye and under a microscope. Growth visible around the edges and under the sample.			Lack of fungistatic effect
M6	Growth on the sample was invisible to the unaided eye and under a microscope. Growth visible around the edges and under the sample.			Lack of fungistatic effect

pension on a substrate poor in nutrients (agar substrate without a carbon source). The influence of the test fungi on the sample examined was evaluated as the growth rate on the samples, which was an average score from four repetitions. In this way, the resistance to *Aspergillus niger* was evaluated. For the other strains: *Trichophyton mentagrophytes* and *Candida albicans*, the methodology was modified by the application of mineral salts solution of pH 5.6 with the addition of peptone and glucose. This procedure allowed to prevent cellulose decomposition.

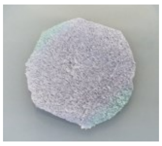
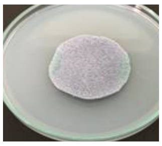
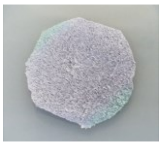
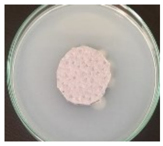
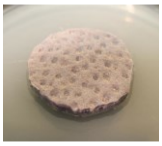
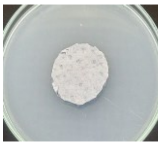
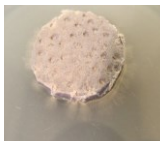
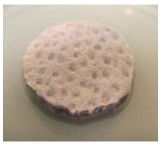
The antifungal properties of the leathers modified by bamboo extract were obtained with the use of the same standard as for textiles. The microbial activity was measured against the following strains: *Aspergillus niger*, *Trichophyton mentagrophytes* & *Candida albicans*. For the bovine lining materials, the antibacterial activity against *Escherichia coli, Salmonella enteritidis & Pseudomonas aeruginosa* was also examined. Samples of 20 x 50 mm size modified by bamboo extract were tested against the above-mentioned bacteria with the use of AATCC Test Method 147:2011 'Antibacterial Activity Assessment of Textile Materials: Parallel Streak Method'.

The test microorganisms in the suspension were cultured on the middle section of the solidified agar substrate. With the use of an inoculation loop with a diameter of 4 mm, 5 parallel scratches at 10 mm distances were made. The leather samples were put across the culture line in such a way as to cover the middle part of each line. After the incubation, the bacteria growth was rated. The bacteriostatic effect was visible as a zero growth rate under the sample and along the sample edges. In order to rate the growth inhibition, visual examination was carried out.

Methodology of chemical analyses

The determination of heavy metals (Sb, As, Pb, Cd, Cr, Co, Cu, Ni, Hg) in the textile materials examined was conducted with the use of Standard PN-EN ISO 17072-1: 2011: Leather – chemical determination of metal content – Part 1: Extractable metals. This standard specifies a method using extraction with an acid artificial – perspiration solution and subsequent determination with the use of hydride generation atomic absorption spectroscopy (HGAAS) and flame atomic absorption spectroscopy (FAAS). Moreover, hazardous substances were determined for the materials examined,

Table 4. *Anti-fungal activity of bovine and pig leathers modified with the use of 5%, 2% and 1% bamboo extract solution (5 – denotes intensive growth, where fungi covers the whole sample area; 1 – denotes small growth, which is not visible to the human eye, but visible at a microscopic scale at a magnitude of 60x; 0 – denotes the lack of invisible growth at a microscopic scale).*

Sample name		***Aspergillus niger***		***Trichophyton mentagrophytes***		***Candida albicans***	
		Growth intensity		**Growth intensity**		**Growth intensity**	
Growth medium		5		5		5	
Bovine leather	5%	0		0		0	
	2%	0		0		0	
	1%	1		0		0	
Pig leather	5%	0		0		0	
	2%	0		0		0	
	1%	0		0		0	

i.e. dimethyl fumarate, using the gas chromatography technique with a mass detector (GC/MS), aromatic amines – using liquid chromatography with a diode detector (HPLC/DAD), and formaldehyde according to PN EN ISO 14184-1:2011 Standard: Textiles – Determination of formaldehyde- part 1: free and hydrolysed formaldehyde (water extraction method) using of spectrophotometric method UV-VIS.

Results and discussion

Results of microbiological analyses for textile materials

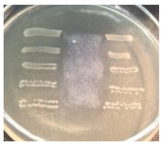
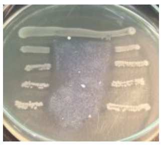
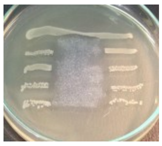
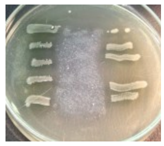
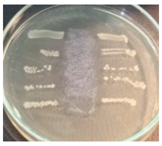
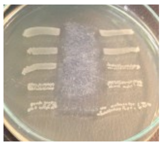
Qualitative analysis of the fungistatic and bacteriostatic activity of textile materials with a bamboo content was conducted by comparison with other commonly used textiles, for example cotton (***Tables 2*** and ***3***).

Due to the specific growth of *Candida albicans*, the results are given in ***Table 3***.

A lot of papers describe key factors which decide the biodeterioration affinity of fabrics [16, 17]. Among the most important factors are thread thickness, material thickness, the number and distribution of pores, and surface curvature. Another element is the bulk density of fibres because this parameter determines the fabric surface shape and area of the potential growth of microorganisms. All of the woven fabrics used have a plain weave with a high density of threads. Moreover, the fabrics used have a similar pattern, where between the weft and warp, the surface of abrasion was enlarged. Both of these parameters gave an opportunity for the uninhibited growth of fungi and microorganisms. Correction of the textile surface in order to reduce surface abrasion can provide a fungistatic and bacteriostatic effect.

In case of the knitted fabrics, the high number of rows and columns created favourable conditions for microorganism growth. The surface area of the textiles was increased because the columns were thin and rows – short. One of the ways to protect materials against biodeterioration without loss of hygienic and physico – mechanical properties is hydrophobic finishing or specific treatment in the finishing processes [18]. It should be noted,

Table 5. *Anti-bacterial effects of bovine leather samples modified with the use of 5%, 2% and 1% bamboo extract solution.*

Sample name		*Escherichia coli*		*Salmonella enteritidis*		*Pseudomonas aeruginosa*	
		Growth inhibition zone, mm		Growth inhibition zone, mm		Growth inhibition zone, mm	
Control sample without addition		0		0		0	
Bovine leather	5%	1-3		2-4		0-5	
	2%	2-4		2-4		2-4	
	1%	1-6		1-4		2-4	

that biodeterioration processes are more intensive for natural fibres because of the high proportion of cellulose subjected to enzymatic hydrolysis [19, 20]. Another element which is important from the microbiological point of view is the hydrophilicity level of materials, which is related with hygroscopic properties and porosity. Hydrophilic fibres swell faster, causing an increase in susceptibility to deterioration [21].

It should be noted that the materials based on bamboo fibres examined in this work do not differ significantly from another from a microbiological perspective. The materials are safe for health and the environment, which is very important due to their potential purpose. The materials will be used as footwear linings or upper elements, and thus the risk of microorganism growth is reduced by the stable temperature and humidity conditions inside the shoe volume. This fact is a consequence of the excellent hygienic properties mentioned in previous papers done within the framework of this project [10, 11].

Results of microbiological analyses for leather materials

Analysis of the fungi resistance of leather modified by bamboo extract showed a fungistatic effect (***Table 4***). After incubation the growth of spoilage mould *A. niger* in the form of white mycelium was observed. Other moulds were also observed: *T. mentagrophytes* and *C. albicans*. The fungistatic effect was found for bovine and pig leather samples modified by bamboo extract against the following microorganisms: *A. niger*, *T. mentagrophytes* and *C. albicans*. Also, the bacteriostatic effect for the modified leathers was observed for *E. coli*, *S. enteritidis* and *P. aeruginosa* (***Table 5***). The inhibition area was observed under and around the edges of the samples depending on the percentage addition of bamboo extract. The diversity of modified leather surfaces with the addition of 5%, 2% and 1% of the bamboo extract was examined by SEM/EDS analysis made at Nicolaus Copernicus University in Toruń, Poland. In order to determine the incorporation rate of the bamboo extract, the content of silica in the modified samples was measured.

As a conclusion of the microbiological analyses of the textile and leather materials, it should be mentioned that in this paper the authors conducted research within a group of specific textiles primarily selected on the basis of the hygienic and mechanical properties, which are determinants of healthy footwear. But it is important to remember that there is an another set of variables which determine the biodeterioration properties of textiles connected with the fabric composition i.e. thread thickness, fabric thickness, linear density, yarn density, weight per unit area, and surface smoothness and roughness – both frictional and geometrical, among others [22, 23]. These parameters determine the dimension of the area that may be treated with microorganisms. When the area grows – for example as a result of textile surface roughness – the probability of extensive biodeterioration also increases. On the contrary, for leathers – or more precisely – for semifinished wet-blue products, particles of bamboo extract remain stuck to the leather surface, being a barrier against microbes. However, the full qualitative and quantitative effect of bamboo extract's antimicrobial activity should be comprehensively studied under various conditions.

Results of chemical analyses

The results of chemical analyses show that heavy metal content in all of the textile materials is below the limits specified in Oeko Tex Standard 100 [24]. In samples M1-M6 the lack of amine aromates (4-aminodiphenyl, benzidine, 4-chloro-o-toluidine, 2-naphthylamine, o-anisidine, o-aminoazotoluene, p-chloroaniline, 2,4 diaminoanisole, 4-aminoazobenzene, p-cresidine, 4,4 diaminodiphenylmethane, 3,3 dichlorobenzidine, 3,3 dimehoxybenzidine, 2,4-toluenediamine, 3,3 dimethtylobenzidene, 4,4 – oxydianiline, 2-amino-4-nitrotoluene, 3,3-dimethylo-4,4-diaminodiphenylmethane, 4,4' – methylene bis (2-chloroaniline), 4,4'-thiodioaniline, 2,4,5- trimethylaniline, o-toluidine) was observed. The range of measurement was below 30.0 ppm. Moreover, in the textiles examined, the formaldehyde and dimethyl fumarane (DFU) were also below the limits of quantification, equal to 16 mg/kg and 0.1 mg/kg, respectively. Thus, it can be concluded that all of the textile materials chosen are safe for human health and the environment.

Summary

The Łukasiewicz Research Network – Institute of Leather Industry has long been active in creating product and technology innovations addressed to the leather or textile branches. A lot of works are focused on the improvement of material functionality and a reduction in the environmental burden connected with the storage and treatment of leather materials within the framework of the circular economy [25-33]. It is commonly known that leather is still the most optimal raw material for footwear as it has many advantages, especially in hygienic and rheological properties, which are important from a comfort and healthiness point of view. Leather's structure allows to provide very good water vapour permeability as well as the sorption and desorption of moisture, which are some of the most important factors in the qualitative evaluation of footwear. On the other hand, the mechanical properties related to wearing comfort and stability in various environmental conditions are also (very) important.

With a view to deterioration processes, which happen under the influence of sweat and contaminations, the constructive search for new solutions is very important and desirable. All innovations in the field of textile and leather footwear material engineering should be aimed at those elements which decide the optimal temperature and humidity conditions as well as microbiological purity inside a shoe volume during the product's whole lifecycle. By using antiseptic agents based on natural components, like the above-mentioned bamboo extract, it is possible to create an alternative solution to the standard tannery processes, which consume a lot of chemical substances. Regarding textile fabrics with natural bamboo fibres, antimicrobial activity can be obtained with specific finishing processes without the loss of hygienic and physico – mechanical properties.

The research conducted in this and previous works [10, 11] were the basis for developing a children's footwear prototype where knitted fabric M6 and bovine leather with a 1% addition of bamboo extract were used as linings and filling of the shoe interior. Three types of footwear were created: leather, leather – textile and textile shoes. All prototypes received the Health Foot trademark, which is a hallmark of the very good quality of a final product dedicated for a special target group of users i. e. children under the age of 15. This certificate makes sure that healthy development and conditions are provided in shoe interiors.

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