(REFEREED RESEARCH)

ASSESSMENT OF ANTIMICROBIAL ACTIVITY OF NATURAL LEATHERS TREATED WITH *Pseudevernia furfuracea* (L.) Zopf EXTRACTS

Pseudevernia furfuracea (L.) Zopf EKSTRAKTLARI İLE MUAMELE EDİLEN DOĞAL DERİLERİN ANTİMİKROBİYAL AKTİVİTESİNİN DEĞERLENDİRMESİ

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ABSTRACT

In the present study, antimicrobial activity of raw skin and chrome-tanned leather samples treated with acetone and chloroform extracts of Pseudevernia furfuracea (L.) Zopf were tested against Bacillus subtilis (ATTC 6633), Bacillus cereus (CCM 99), Staphylococcus aureus (ATCC 6538-P), Escherichia coli (ATCC 11228), Enterococcus faecalis (ATCC 29212), Klebsiella pneumoniae (CCM 2318), Pseudomonas aeruginosa (ATCC 27853), Aspergillus niger, Aspergillus fumigatus, Aspergillus candidus, Aspergillus flavus, Penicillium jensenii, Geotrichum candidum and Candida albicans by the disc diffusion method. The study showed that acetone and chloroform extracts of Pseudevernia furfuracea (L.) Zopf had antimicrobial activity on raw skin and chrome-tanned leather. The inhibition zones of both extracts for the bacteria and fungi tested were found to vary between 11.0 mm to 31.0 mm in raw skins, but between 11.2 mm and 29.0 mm in chrome-tanned leathers.

Key Words: Raw skin, Chrome-tanned leather, Pseudevernia furfuracea (L.) Zopf, Antimicrobial activity.

ÖZET

Bu çalışmada, Pseudevernia furfuracea (L.) Zopf'ın aseton ve kloroform ekstraktları ile muamele edilmiş ham deri ve krom tabaklanmış deri numunelerinin antimikrobiyal aktivitesi disk difüzyon yöntemiyle, Bacillus subtilis (ATTC 6633), Bacillus cereus (CCM 99), Staphylococcus aureus (ATCC 6538-P), Escherichia coli (ATCC 11228), Enterococcus faecalis (ATCC 29212), Klebsiella pneumoniae (CCM 2318), Pseudomonas aeruginosa (ATCC 27853), Aspergillus niger, Aspergillus fumigatus, Aspergillus candidus, Aspergillus flavus, Penicillium jensenii, Geotrichum candidum ve Candida albicans'a karşı test edilmiştir. Çalışma sonucunda, Pseudevernia furfuracea (L.) Zopf'un aseton ve kloroform ekstraktlarının antimikrobiyal aktivitesi, ham deri ve krom ile tabaklanmış derilerde gözlenmiştir. Her iki ekstraktın inhibisyon zonları ham derilerde, test edilen bakteri ve funguslar için 11.0 mm ile 31.0 mm arasında, kromlu derilerde ise 11.2 mm ile 29.0 mm arasında değiştiği bulunmuştur.

Anahtar Kelimeler: Ham deri, Krom tabaklanmış deri, Pseudevernia furfuracea (L.) Zopf, Antimikrobiyal aktivite.

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1. INTRODUCTION

Raw skin used in the production of final leather is a natural and specific product in terms of its fibre structure, chemical composition and physical properties. Skin, as an outer covering, performs many functions while the animal is alive; it protects the organism against heat, cold, mechanical impacts and microorganisms (1).

A great diversity of microorganisms from air, water, soil, faeces and external dirt are found on animal skin. Many microorganisms have a limited impact on skin when the animal is alive, but after slaughter and flaying, the microorganisms start to spread over the flesh side of the skin and proceed to grow quickly. (2-3).

Because the threat from microorganisms starts from the time of flaying, skins are commonly conserved with NaCl. However, in spite of the many precautions taken to prevent damage from microorganisms during the various processes performed in the tannery, the skins may still constitute a nutrient source for microorganisms (4). During leather production, the skins may have basic or acidic properties which leave them open to damage by bacteria or fungi respectively; they may be degraded by bacteria in the initial processes of leather production such as pre-soaking and main soaking while being damaged by fungi in the pickling, tanning and post-tanning processes (5). For this reason, they are also treated with bactericides and fungicides to prevent damage from these microorganisms.

The increased importance of ecological and toxicological criteria in all industrial fields has greatly affected the leather industry also. With the development of new production techniques and technologies, a tendency towards more healthy production has occurred. The use of many toxic chemical substances has been abandoned. Various restrictions, or even prohibitions, have been introduced, especially on exported products containing antimicrobial agents which may damage human health and the environment (6-7).

Synthetic antimicrobial agents that have been used industrially have included quarternary ammonium salts, chlorinated phenols, silylquaternary compounds, tolysulfone compounds, polyvinyl pyridines, diamonium rings, n-halamin compounds, and photosynthesizing compounds (8). Many natural products with antimicrobial activity derived from plants, animals and microorganisms have low toxicity, and are biodegradable and biocompatible (9). However, some are toxic, poorly effective and expensive, which makes them unsuitable for applications in health, food, textiles, filters and leathers, and for the prevention of pollution (10).

Lichens have highly diversified areas of use, and have antimicrobial features. They have been used in the fields of medicine and pharmacy (11-13). Although many studies have been carried out on the antimicrobial effects of lichens, no research was found related to use of lichen extracts in leather production process and antimicrobial properties of leather treated with it

This study aimed to examine the antimicrobial activity of acetone and chloroform extracts of *Pseudevernia furfuracea* (L.) Zopf, a lichen commonly found on trees in Turkey on raw skin and chrome-tanned leather.

2. MATERIAL AND METHOD

2.1. Material

Seven species of bacteria and fungi were used as test microorganisms. These microorganisms were *Bacillus subtilis* (ATTC 6633), *Bacillus cereus* (CCM 99), *Staphylococcus aureus* (ATCC 6538-P), *Escherichia coli* (ATCC 11228), *Enterococcus faecalis* (ATCC 29212), *Klebsiella pneumoniae* (CCM 2318), *Pseudomonas aeruginosa* (ATCC 27853); *Aspergillus niger, Aspergillus fumigatus, Aspergillus candidus, Aspergillus flavus, Penicillium jensenii, Geotrichum candidum and Candida albicans*. All microorganisms were provided from Ege University, Science Faculty, Biology Department, Basic and Industrial Microbiology Section.

Four domestic wet-salted sheep skins without any bactericide or fungicide were used. All the analytical grade chemicals were provided by Merck (Germany).

2.1. Method

2.1.1. Processing of Skins

Raw skin discs 10 mm in diameter were first subjected to microbiological analysis. Pre-tanning and chrome-tanning processes of the skins were then carried out according to the prescription provided in Table 1.

Leather samples with a diameter of 10 mm were obtained from raw skin and chrome-tanned leather by cutting them carefully with a sterile stainless steel gauge manufactured from with an inner diameter of 10 mm.

2.1.2. Preparation of Lichen Extracts

Lichen samples collected from *Quercus* sp. trees in Bursa, Turkey were used in the study. The samples were dried at room temperature and then pulverized in a porcelain mortar. 20 g of each of those pulverized lichen samples was taken and extracted in 150 ml chloroform (Merck) at 62°C and in 150 ml acetone (Merck) at 52°C in a Soxhlet device (Electro-Mag) for 24 hours. The Chloroform-Lichen (C-LE) extract solution and the Acetone-Lichen (A-LE) extract solution were stored at +4°C for use in the microbiological analyses (14).

2.1.3. Preparation of Microorganism Cultures

The Disc Diffusion Method was used for the determination of antimicrobial activity (14-19).

Raw skin and chrome-tanned leather samples were put separately into two sterile Erlenmeyer flasks, one containing 200 ml C-LE and the other 200 ml A-LE. Sterile Erlenmeyers containing only 200 ml chloroform and 200 ml acetone were used as a negative control. As a positive control, Tetracycline and Nystatin antibiotics were used for bacteria and molds/yeasts respectively. These were absorbed by sterile antibiotic assay discs prepared with the same diameters as the skin discs (Tetracycline 10µg/disc and Nystatin 25µg/disc). The antimicrobial activity of chrome tanning material was not considered in chrome-tanned leather samples

Bacteria cultures were inoculated into a Nutrient Broth medium and incubated for 24 hours at 37°C, while mold and yeast cultures were inoculated into a Malt Broth medium and incubated for 48 hours and 5 days respectively at 27°C. Then 0.1 ml was taken from each test culture and inoculated into petri dishes containing Müller Hinton Agar using L-baget. The leather and paper samples were placed onto agar plates. All processes were carried out with aseptic techniques. The Petri dishes inoculated test bacteria were incubated for 24-48 hours at 37°C while fungi were incubated for 5-7 days at 27°C. The trials were repeated 3 times. Following the incubation, the zone diameters around the discs were measured and averaged.

3. RESULTS AND DISCUSSION

3.1. Effects of Lichen Solutions on Bacteria

The antimicrobial activity of lichen extracts on raw skin and chrome-tanned leather are provided in Table 2.

Table 1. Methodology of pre-tanning and chrome tanning process

Processes	Chemical Additivities (%)	Temperature (°C)	Time	Remarks
Pre-soaking	Water, 500%	20 °C	240 min	Rest, drain
Main soaking	Water, 500% Nonionic surface active agent, 0.5%	20 °C	18 hours	5'run/55'rest, drain
Unhairing: Painting solution contained 15°Bé Na_2S and 25°Bé $Ca(OH)_2$. After 4 hours the skin was unhaired.				
Liming	Water, 400% Na_2S, 2% $Ca(OH)_2$, 4%	20 °C	30 min run + 5'run/55'rest Total 24 hours	pH: 11-13, drain
Washing	Water, 300%	35 °C	10min	Drain
Deliming and Bating	Water, 300% $(NH_4)_2SO_4$, 1,5% Proteolitic enzyme, 1%	35 °C	30 min 60 min	pH = 8.2 – 8.5, drain
Washing	Water, 200%	20 °C	10 min	Drain
Degreasing	Water, 100% Degreasing agent, 5%	35 °C	90 min	Drain
Washing	Water, 100% NaCl, 2%	35 °C	30 min	Drain
Washing	Water, 100% NaCl, 2%	35 °C	30 min	Drain
Pickling	Water, 150% NaCl, 6% HCOOH (1:10 diluted), 1% H_2SO_4 (1:10 diluted), 0.8%	20 °C	15 min 30 min 90 min.	pH = 2.8-3.0, drain
Chrome Tanning	Water, 100% NaCl, %6 Basic chrome sulphate, 4% (% 33 Basicity) Fatliquor, 0.5% Basic chrome sulphate, 4% (% 33 Basicity) HCOONa (1:10 diluted), 1% $NaHCO_3$ (1:10 diluted), 0.5%	20 °C	30 min. 30 mim. 240 min. 30min. 60 min.	pH: 3.8-4.0, drain, 48 hours horse up
Drying and process mechanics				

Table 2. Antimicrobial activity of *Pseudevernia furfuracea* (L.) Zopf extracts

Microorganisms	Raw skin disc*				Chrome tanned leather disc*				Paper disc* (Positive control)	
	A-LE	Acetone**	C-LE	Choloroform**	A-LE	Acetone**	C-LE	Choloroform**	Tetracycline 30µg	Nystatin 100 µg
Bacteria										
B. cereus	18,0	10,0	15,0	10,0	11,2	10,0	13,5	10,0	25,0	-
B. subtilis	15,0	10,0	15,0	10,0	12,5	10,0	14,5	10,0	27,0	-
S. aureus	25,0	10,0	16,0	10,0	12,5	10,0	14,0	10,0	26,0	-
E. coli	17,0	10,0	18,0	10,0	12,0	10,0	13,0	10,0	28,0	-
E. faecalis	16,0	10,0	16,0	10,0	13,5	10,0	15,2	10,0	25,0	-
K. pneumoniae	14,0	10,0	16,0	10,0	11,8	10,0	13,0	10,0	30,0	-
P. aureginosa	20,0	10,0	30,0	10,0	12,75	10,0	15,0	10,0	34,0	-
Fungi										
A. .niger	20,0	10,0	17,0	10,0	17,0	10,0	15,5	10,0	-	22,0
A. fumigatus	17,0	10,0	18,0	10,0	25,0	10,0	21,0	10,0	-	24,0
A. candidus	20,0	10,0	21,0	10,0	21,8	10,0	18,0	10,0	-	22,0
A. flavus	20,0	10,0	16,0	10,0	22,0	10,0	19,0	10,0	-	23,0
P. jensenii	13,0	10,0	11,0	10,0	20,5	10,0	23,2	10,0	-	24,0
G. candidum	29,0	10,0	30,0	10,0	26,0	10,0	29,0	10,0	-	21,0
C. albicans	31,0	10,0	31,0	10,0	21,0	10,0	21,5	10,0	-	23,0

* :Diameter of all discs is 10 mm; **: Negative Control ; - : None detected

When the results were examined (Table 2), it was observed that raw skins treated with A-LE showed the greatest antimicrobial activity against *S. aureus* (25 mm) and showed the lowest activity against *K. pneumonia* (14 mm). Raw skins treated with C-LE were found to have the highest antimicrobial activity against *P. aeruginosa* (30 mm), while showing the lowest antimicrobial activity against *B. cereus* and *B. subtilis* (15 mm). In our findings, it was also revealed that raw skins treated with C-LE had lesser antibacterial effect on the *Bacillus* genus.

Chrome-tanned leathers treated with C-LE showed a greater antimicrobial activity than those treated with A-LE. Chrome-tanned leathers treated with A-LE had the greatest effect on *E. faecalis* (13.5 mm) while showing the lowest effect on *B. cereus* (11.2 mm). The antimicrobial effect of chrome-tanned leathers treated with C-LE was also found to be the highest on *E. faecalis* (15.2 mm) while its lowest effect was on *E. coli* and *K. pneumonia* (13 mm).

It was also determined in the present study that the antibacterial effect of A-LE and C-LE on raw skin was higher than on chrome treated leather.

Saenz et al. (2006) tested the antimicrobial activity of acetone extracts of some lichens (*A. radiosa, C. convoluta, C. firma, D. scruposus, D. repanda, L. muralis, P. mammosa, R. canariensis, R. subfarinacea, R. fuciformis* and *X. calcicola*) against *B. cereus* (ATCC 14579), *B. megaterium* (ATCC 33085), *S. aureus* (ATCC 25923), *E. coli* (CCM 180), *K. pneumoniae* (ATCC 13883) and *P. aeruginosa* (ATCC 27853). The researchers showed that the acetone extracts of none of the lichens had any antimicrobial effect on *E. coli* and *P. aeruginosa* while the acetone extract of *X. calcicola* had an antimicrobial effect against *K. pneumoniae* (19).

In our results, acetone and chloroform extract of *P. furfuracea* (L.) Zopf were effective against *P. aureginosa, E. coli* and *K. pneumoniae*. C-LE provided a larger inhibition zone value for *P. aureginosa, E. coli* and *K. pneumoniae* than did A-LE.

When Gücin et al. (1997) examined the antimicrobial effect of ethyl acetate, chloroform, acetone and ethanol extracts of *P. furfuracea* (L.) Zopf on *E. coli* ATCC 11230, *E. aerogenes* CCM 2531, *Staphylococcus epidermidis* ATCC 12228, *S. aureus* 6538P, *B. subtilis* La2114, *B. cereus*, *Mycobacteriıım smegmatis* CCM2067, *C. albicans* and *Candida utilis* La991 (20), they found that the extracts had no antimicrobial effect against *E. coli* ATCC 11230 and *E. aerogenes* CCM 2531. The antimicrobial effect of chloroform extract of *P. furfuracea* (L.) Zopf was the greatest on *S. epidermidis* ATCC 12228 (11.0 mm) and least on *Mycobacterium smegmatis* CCM2067. They also showed that acetone extract had the lowest effect on *C. albicans* (8.0 mm)(20).

In another study, it was found that acetone and chloroform extracts of *P. furfuraceae var. furfuracae* had no antimicrobial effect on *C. albicans*, *E. coli*, *K. pneumoniae* and *P. aeruginosa* (21).

3.2. Effects of Lichen Solutions on Fungi

In Table 2, it is seen that raw skin and chrome-tanned leather treated with both A-LE and C-LE have an antifungal effect. It was found that raw skin treated with A-LE showed the greatest antifungal effect on *C. albicans* (31 mm) and treated with C-LE exhibited the lowest effect on *P. jensenii* (11 mm). Chrome-tanned leathers treated with A-LE and C-LE showed the greatest antifungal effect against *G. candidum* (26 mm and 29 mm respectively) while showing the lowest effect against *A. niger* (17 mm and 15.5 mm respectively).

In the present study, it was found that A-LE and C-LE have an antifungal effect on *Aspergillus* species, which commonly appear on chrome-tanned leather (22). It was determined that chrome-tanned leathers treated with A-LE had a greater antifungal impact on *A. niger, A. fumigatus, A. candidus* and *A. flavus* compared to those treated with C-LE.

According to the findings of research carried out by Osmanoğlu et al. (1991), acetone and chloroform extracts of *P. furfuraceae var. furfuracae* had no antimicrobial effect against *C. albicans* (21). Similarly, it was determined that acetone extract of *P. furfuracea (L.)* Zopf had the lowest antifungal activity on *C. albicans* (8.0 mm) (20). In our research, however, the raw skin and chrome-tanned leathers treated with A-LE and C-LE showed antifungal activity on all test fungi used.

According to results obtained from other studies the lichens *Pseudevernia furfuracea* and *Pseudevernia furfuracea var. furfuracea* have little or no antibacterial and antifungal effect on selected bacteria and fungi. However, the results of present study provide a new finding, namely that *Pseudevernia furfuracea* (L.) Zopf has antibacterial and also antifungal activity on test bacteria and fungi in raw skin and chrome-tanned leathers treated with A-LE and C-LE. Thus, unlike the results of other studies in which the antimicrobial effects of lichens were examined, the results of our study indicated that *P. furfuracea (L.)* Zopf had antibacterial and antifungal effects on samples treated with A-LE and C-LE.

4. CONCLUSION

This study has demonstrated the antimicrobial activities of A-LE and C-LE applied to raw skin and chrome-tanned leather. It has been found that *P. furfuracea* (L.) Zopf has an antimicrobial effect on both raw skin and chrome-tanned leather. With regard to its antibacterial and antifungal effect on raw skin, C-LE had the greatest effect against *P. aeruginosa* (30mm), and *C. albicans* (31mm) and A-LE was most effective against *S. aureus* (25mm) and *C. albicans* (31mm).

The results showed that *P. furfuracea* (L.) Zopf, which is commonly found in Turkey, can be used in the leather industry as an alternative eco-friendly antimicrobial agent. Thus, it will be possible to obtain a better-quality final leather which does not contain harmfull compounds. Additionally, with use of lichens instead of traditional chemical substances as antimicrobial agents in the leather industry, it will be possible to minimize environmental pollution, and protect human health and the ecological balance.

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REFERENCES

1. Işık, N. O. and Karavana, H. A., 2012, "Determination of Some Physical Characteristics of Artificially Aged Chrome Tanned Garment Leather", *Tekstil ve Konfeksiyon*, Vol: 22, pp: 64-69.
2. Birbir, M., 1997, "Investigation of Salted-cured France and Russian Hides for Halophilic Bacteria", *Journal of Turkish Microbiology Society*, Vol: 27, pp: 68-73.
3. Orlita, A., 2004, "Microbial Biodeterioration of Leather and Its Control: a Review", *International Biodeterioration & Biodegradation*, Vol: 53, pp: 157-163.
4. Bitlisli, B. O., Karavana, H. A., Başaran, B., Sarı, Ö., Yasa, İ. and Birbir, M., 2004, "The Effect of Conservation Defects on The Suede Quality of Doubleface", *Journal of the American Leather Chemists Association*, Vol:12, pp: 494-501.
5. Durmuş, D., 2007, "Tabaklama ve Sonrası Yaş İşlemlerde Mikrobiyal Yükün Tespiti Üzerine Bir Araştırma", *Yüksek Lisans Tezi*, Çanakkale Onsekiz Mart Üniversitesi Fen Bilimleri Enstitüsü, pp:1-71.
6. Karavana, H. A., Başaran, B., Aslan, A., Bitlisli B.O. and Gülümser, G., 2011, "Heavy Metal Contents of Bootee Leathers Tanned With Different Process Recipes", *Tekstil ve Konfeksiyon*, Vol:21, pp: 305-310.
7. Aslan, A. and Gülümser, G., 2011, "Evaluation of Chrome-Tanned Leather Solid Wastes As Lubricating Filler Agents", *Tekstil ve Konfeksiyon*, Vol:21, pp: 405-409.
8. Williams, J. F., Halo Source, V. and Cho, U., 2005, "Antimicrobial Functions for Synthetic Fibers: Recent developments", *AATCC Review,* Vol:5, pp: 17-21.
9. Afşar, A., Gülümser, G., Aslan, A. and Ocak, B., 2010, "A Study on Usability of Collagen Hydrolysate Along with Oxazolidine in Leather Processing", *Tekstil ve Konfeksiyon*, Vol:20, pp: 37-40.
10. Khalili B., Bardana, E. J. Jr. and Yunginger, J. W., 2003, "Psyllium-Associated Anaphylaxis and Death: a Case Report and Review of The Literature" *Annals of Allergy, Asthma and Immunology*, Vol: 91, pp: 579-84.
11. Nash III, T.H., 1996, "Inroduction", *Lichen Biology*, Cambrigde University Press, United Kingdom, pp: 1-8.
12. Brodo, I. M., Sharnoff, S. D. and Sharnoff, S., 2001, " About the Lichens", *Lichens of North America*, Yale University Pres, New Haven and London, pp: 3-8.
13. Aslan, A., 2006, "Likenler, Kullanım Alanları ve Palandöken Likenleri", *106Y110 Nolu Proje*, TÜBİTAK, Ankara, pp: 107-121.
14. Dülger, B., Gücin, F. and Aslan, A., 1998, "Cetraria islandica (L.) Ach. Likeninin Antimikrobiyal Aktivitesi", *"Turkish Journal of Biology"*, Vol:22, pp: 111-118.
15. Tay, T., Özdemir Türk, A., Yılmaz, M., Türk, H., and Kıvanç, M., 2004, "Evaluation of the Antimicrobial Activity of the Acetone Extract of the Lichen Ramalina farinacea and its (+)-Usnic Acid, Norstictic Acid, and Protocetraric Acid Constituents", Verlag der Zeitschrift für Naturforschung, Vol: 59, pp: 384-388.
16. Balaji, P. and Hariharan, G. N., 2007, "In vitro Antimicrobial Activity of Parmotrema raesorediosum Thallus Extracts", Research Journal of Botany, Vol: 1, pp: 54-59.
17. Rankovi´c, B., Miši´c, M., and Sukdolak, S., 2007, Evaluation of Antimicrobial Activity of the Lichens Lasallia pustulata, Parmelia sulcata, Umbilicaria crustulosa and Umbilicaria cylindrica, Microbiology, 76(6):723-727p.
18. Rankovi´c, B., Miši´c, M., and Sukdolak, S., 2009, "Antimicrobial Activity of Extracts of The Lichens Cladonia Furcata, Parmelia Caperata, Parmelia Pertusa, Hypogymnia Physodes and Umbilicaria Polyphylla", Biologia, Vol: 64, pp:53-58.
19. Saenz, M.T., Garcia, M.D. and Rowe, J.G, 2006, "Antimicrobial Activity and Phytochemical Studies of Some Lichens From South Of Spain", Fitoterapia, Vol: 77, pp:56-159.
20. Gücin, F., Dülger, B., and Aslan, A., 1997, "Pseudevernia Furfuracea (L.) Zopf Likeninin Antimikrobiyal Aktivitesi", Ekoloji Çevre Dergisi , Vol: 7, pp: 22-24.
21. Osmanağaoğlu, Ö., Yıldız, A. and Saçılık, S. C., 1991, "Türkiye'deki Farklı Bölgelerden İzole Edilen Likenlerin Antimikrobiyal Aktiviteleri", Türk Mikrobiyoloji Cemiyeti Dergisi, Vol:30, pp: 17-19.
22. Meriçli Yapıcı, B. and Karaboz, İ., 1997, "The Effect of Two Anti-Fungal Compounds on The Growth of Molds That Frequently Appear on Tanned Leather". Journal of the American Leather Chemists Association, Vol:92, pp:38-45.