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Antimicrobial Potential of *Albizia lebbeck* Extracts for Sustainable Sheepskin Preservation and Leather Production

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Abstract

Raw skins are preserved before processing them into leather, to protect the skin protein from microbial attack. Preservation of hides or skins usually involves the application of common salt, resulting in environmental pollution. This is because the use of salt to preserve hides/skins could result in the generation of quantities of total dissolved solids regarded as one of the most difficult pollutants to manage. This study evaluates the antimicrobial potential of Albizia lebbeck extracts for sheepskin preservation and leather production. The biocide recipe was applied on the flesh side of the skin at different concentrations and preserved for a period of 30 days. The preserved skins were tanned and retanned, and the leather's properties were assessed. Ethyl acetate extracts demonstrated the highest antimicrobial activity, particularly against Staphylococcus spp. (37 mm zone of inhibition). Leather produced from the experimentally preserved sheepskin shows similar physical properties in comparison to the leather produced from conventionally preserved sheepskin. Moreover, the results suggested that Albizia lebbeck extracts are highly potential biocidal agents for preserving sheepskins at the concentration range of 1 - 5 mg/mL. Conclusively, leather preserved with A. lebbeck exhibited comparable physical properties to salt-cured leather, highlighting its potential as an eco-friendly alternative to conventional preservation methods.

Keywords: Albizia lebbeck, Biocide, Leather, Preservation, Sheepskins

INTRODUCTION

Traditional salt-based preservation methods for hides and skins contribute to environmental pollution through high salinity in tannery effluent. This study investigates the potential of *Albizia lebbeck* extracts as a sustainable alternative, evaluating their antimicrobial efficacy and impact on leather quality during processing. Pre-tanning, tanning, post-tanning, and hide/skin finishing are the steps involved in producing leather. Since the hide or skin is mainly composed of 25–30 % protein and 60–70 % water by weight, microorganisms typically impact it (Turzo *et al.*, 2023). Within 6 to 8 hours of an animal's death, hide or skin begins to denature if preservation is not applied. Bacteria may proliferate after 15 to 24 hours, leading to significant grain damage and holes in the hide or skin (Wu *et al.*, 2017). Although affordable and widely accessible, the traditional method of preserving hide or skin by utilizing 30 to 70 % of the salt (NaCl) based on sample weight is ecologically unfriendly. It causes total dissolved solute (TDS) in tannery effluent (Vedaraman *et al.*, 2016). It is crucial to replace this preservative, and researchers have studied physical, chemical, and biocidal treatments as alternate methods of hide and skin preservation (Turzo *et al.*, 2023). Plant sources used in the phyto-based preservation process, which has become more and more popular, include *Aristolochia bracteolata L.* (Uddin *et al.*, 2021), *Tamarindus indica* (Alagumuthu *et al.*, 2015), *Moringa oleifera* leaf paste (Hashem *et al.*, 2018) and *Ficus hispida* leaf paste (Hashem *et al.*, 2021). The amount of essential protein in raw skin determines the quality of the leather.

As a result, protecting skin protein from deterioration throughout the preservation process is crucial. Since the protein in the skin is very vulnerable to bacterial breakdown, stopping the microbial attack is crucial for skin preservation. The raw skin's intact protein content represents the leather's quality. Numerous microbiological flaws, including looseness, hair slippage, weakness, coloring, odor, holes, and fiber degradation of finished leather, are caused by microorganisms (Selvi *et al.*, 2020). Therefore, raw skin protein must be well preserved to create high-quality leather. The hair slip, hydrothermal stability, moisture content, and organoleptic characteristics of *Albizia lebbeck* biocide were measured to assess its effectiveness as an antibacterial agent. Throughout the preservation time, the effectiveness is evaluated by comparing it to traditional preservation methods. The search for environmentally friendly hides/skin preservatives necessitated the development of an organic curing agent with proven properties compared to the inorganic preservatives. Conventionally, salts are generally used to preserve hides/skins and are environmentally unfriendly to the ecosystem. With this new development, plant extracts in this report have proven good potency in the stabilization of the hides/skins. This research was limited to only one plant out of many that could be utilized. There is a need for more research to be conducted on screening plants that could have antimicrobial potential on animal skin preservation.

MATERIALS AND METHODS

Plant Samples Collection and Extract Preparation

We purchased the fresh *Albizia lebbeck* leaves from Samaru-Zaria in Kaduna State, Nigeria. Thereafter, the identity of the plant was carried out in the herbarium of the Department of Botany, Faculty of Life Sciences, Ahmadu Bello University, Zaria. It was done by comparing the plant with the original herbarium specimen with a voucher specimen number of 900247. After being thoroughly sorted, the plant leaves were cleaned with regular water and allowed to dry at room temperature. The dried material was ground into a fine powder and extracted using an analytical-grade solvent. Additionally, 500 g of the plant materials were extracted using the maceration method with 2,000 cm^3 of ethyl acetate, methanol, acetone, and water each. Afterward, the mixes were shaken vigorously and sporadically for 72 hours at room temperature. The extract was then obtained by filtering through Whatman filter paper No. 1. Using a drying cabinet, the filtrate extracts were dried at 37°C and stored in a cold location until they were needed.

Application and Quality Evaluation of Sheepskins

Sheepskins that had been flayed (within one hour of flaying) were utilized for preservation, quality assessment, and curing using varying amounts of preserving agents. The study's percentages were determined using the skin's green weight, shown in Table 1. The half skins were stored for 30 days after being treated with 30% common salt (on the side of the new skin) as a positive control. After being preserved at varying concentrations of 30% *Albizia lebbeck* biocides, each quarter of the skin was stored in a laboratory setting for 30 days. The quality of preservation was evaluated organoleptically by monitoring the mucosal surface of the skin at room temperature and noting any physical changes, such as hair slippage, smell, and look daily, which are indicators of putrefaction (Kamaruzzaman *et al.*, 2024).

Table 1: Methods for Preserving the Materials to Check the Antimicrobial Activity

Experiment	Codes	Percent (w/v) 30% of preservation materials used	Sample weight (g)
Preservation with *Albizia lebbeck*	Sample 1	5mg/mL of *Albizia lebbeck*	261.25
	Sample 2	1mg/mL of *Albizia lebbeck*	226.14
	Sample 3	Control (NaCl)	670.00

Leather processing and evaluation of physical properties of leather

Following thirty (30) days of treatment, a standard tanning technique was used to turn the experimental and control sheepskin samples into finished leather. It was established how strong the experimental and control leathers were physically. Specimens were cut out using the procedures (ISO2418 2017). According to IULTCS guidelines, analyses were conducted for tensile strength and elongation percentage at break (IUP6, 2023), load at grain crack (IUP8, 2016), moisture content, tear strength, and hardness.

Statistical Analysis

The statistical tools used were descriptive and ANOVA. A one-way analysis of variance was used

because the data were divided into groups (from three above) according to only one factor.

This was carried out using Microsoft Office Excel 2019 version on the properties of the leather produced. From the menu bar, data was selected, and afterward, a drop box appeared, and data analysis was then selected. Subsequently, the input range was selected as well as the output. Finally, the "OK tab" was clicked, and the results that contained the p-value appeared on the output range.

RESULTS AND DISCUSSION

Table 2: Anti-microbial Activity of *Albizia lebbeck* extract on Indigenous Organisms from sheepskins

Plant	Extracts	Microbes	Inhibition (mm) Zone at Different Concentrations (mg/mL)				Positive control (mm)	*p* value
			10	5.0	2.5	1.25		
Albizia sp	ME	*Bacillus* spp	16	0	0	0	45	<0.0001
	EA		20	9	0	0		
	AC		25	0	0	0		
	AQ		0	0	0	0		
	ME	*Staphylococcus aureus*	22	0	0	0	39	<0.001
	EA		25	0	0	0		
	AC		20	0	0	0		
	AQ		10	0	0	0		
	ME	*Candida tropicalis*	25	16	11	6	55	0.0003
	EA		32	15	10	5		
	AC		22	15	12	7		
	AQ		7	0	7	0		
	ME	*Staphylococcus* spp	23	17	12	11	52	0.0026
	EA		37	18	15	13		
	AC		30	15	11	10		
	AQ		11	0	0	0		
	ME	*Corynerbacterium spp*	25	14	9	7	37	0.0182
	EA		18	12	10	9		
	AC		10	9	10	7		
	AQ		0	0	0	0		
	ME	*Micrococuss* spp	25	9	9	7	38	0.0016
	EA		31	13	10	9		
	AC		21	10	10	7		
	AQ		9	0	0	0		

ME: Methanol Extract, AC: Acetone Extract, EA: Ethyl Acetate Extract and AQ: Aqueous

Antimicrobial activity assay

The microorganisms isolated from the specimen were susceptible to the *Albizia lebbeck* extract at a concentration of 10 mg/mL, according to the results of the antimicrobial assay (Table 2). These findings showed that the putrefying bacterial isolates on the sheepskins were inhibited in their growth by the extracts of *Albizia lebbeck*. With a 37 mm inhibition zone, the most notable inhibitory performance was seen against *Staphylococcus species*. Furthermore, the results demonstrated that the plant extracts' potency rose with concentration. Aqueous extract showed no discernible antibacterial action, while ethyl acetate extracts had the strongest antibacterial activity among the four extracts examined, followed by acetone and methanolic extracts. The antibacterial activity of the ethyl acetate, acetone, and methanolic extracts ranged from 5 to 37, 7 to 30, and 6 to 25 mg/millimeters, respectively. In the current study, *Staphylococcus species* were the most vulnerable, followed by *Candida tropicalis*, whereas *Coryner bacterium* species were the most resistant to all the extracts examined. The p-values of the isolates were found to be less than 0.05 at a 95 % confidence level. This is an indication that the inhibitory properties of the plant are statistically significant.

Table 3: 30 Days Degradation Evaluation of the Preserved Skins at Different Concentrations of the Curing Agents

Percentage of curing agent (30%)	Skin degradation evaluation		
	Hair loosening	Odour	Putrefaction
5mg/ml of *Albizia lebbeck*	Hair slip absent	No odour	Absent
1mg/ml of *Albizia lebbeck*	Hair slip. No	Normal	Insignificant
Control (NaCl)	No hair slip	Odourless	No

Skin Degradation Evaluation

There is no putrefaction, odor, or hair slip (hair pull-out) in the conserved skins treated with the experimental *Albizia lebbeck* biocides and the salted skin (Table 3). This suggests that the Albizia lebbeck biocides mixture can effectively preserve the skin.

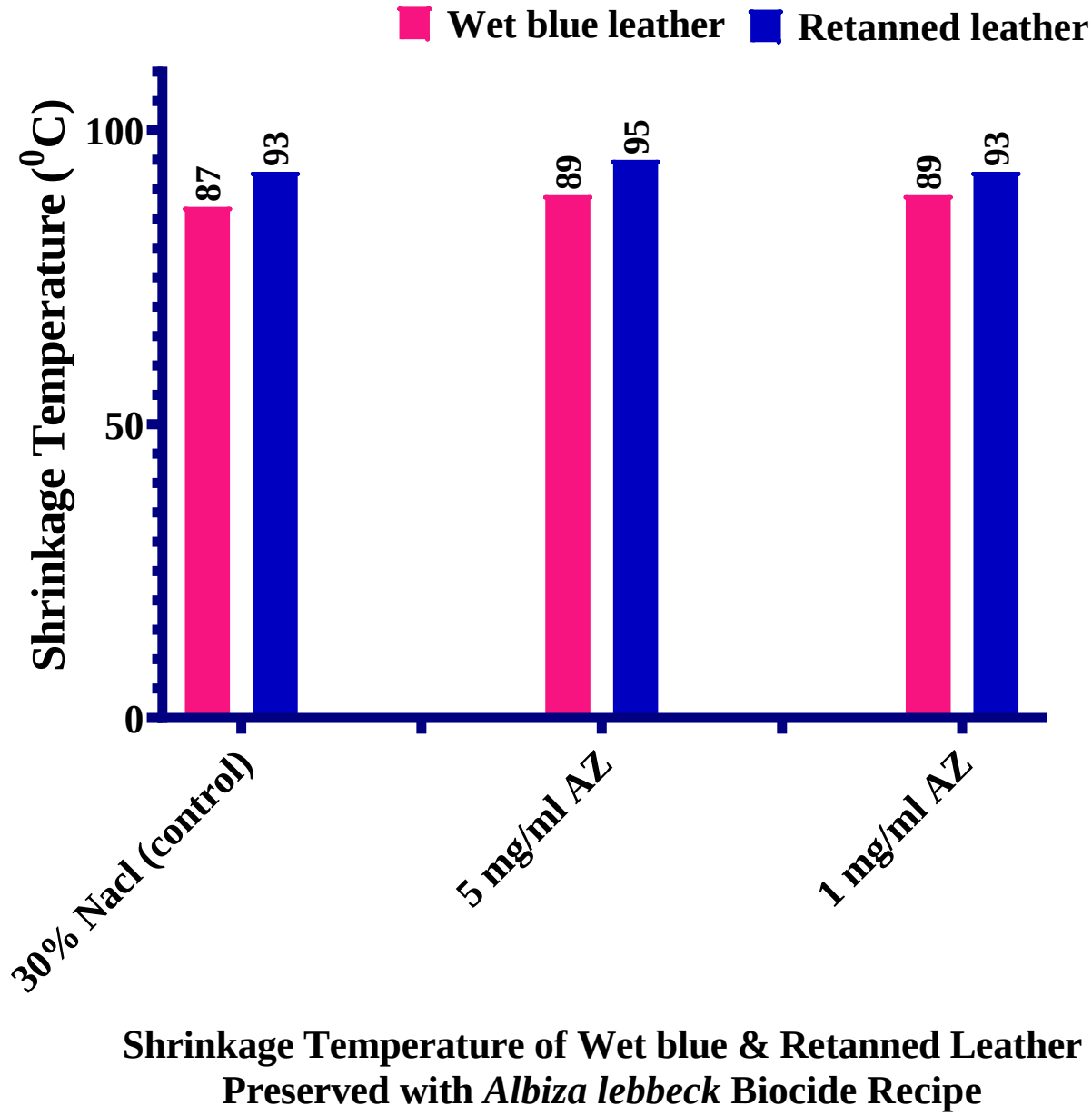

Figure 1: Shrinkage temperature of wet-blue leather of experimentally treated 30% of *Albizia lebbeck* and control (salted sheepskins). AZ = *Albizia lebbeck*, NaCl = Sodium chloride

Hydrothermal Stability Properties

The breakdown of the collagen matrix was indicative of sheepskin spoiling. When assessing the skin's overall quality, one important consideration is the raw hides' or skins' hydro-thermal stability (Tsigab *et al.*, 2020). The collagen matrix's shrinkage temperature, which varies in response to the disintegration of fixed connections, illustrates its structural stability (Zhang *et al.*, 2020). The shrinkage temperature values kept dropping as the skins degrade due to bacterial activity (Hashem *et al.*, 2022). Figure 1 shows the temperature increase that took place during the 30-day skin preservation period using the commercial salt (30 % NaCl) and the curing agent for the experiment (30 % of 5 mg/mL *Albizia lebbeck* and 30 % of 1 mg/mL *Albizia lebbeck*). The experimentally preserved wet blue leather shrinkage temperatures were 89 ℃, 89 ℃, and 87 ℃ for 30 % of 5 mg/mL *Albizia lebbeck*, 30 % of 1 mg/mL *Albizia lebbeck*, and control samples, respectively. 30% of 5 mg/mL *Albizia lebbeck*, 30% of 1 mg/mL *Albizia lebbeck*, and control samples of 95 ℃, 93 ℃, and 93 ℃, respectively, were the shrinkage temperatures of the retanned leather (Plate 1 and 2). This could be in accordance with the report of Uddin *et al.* (2021), who reported that the shrinkage temperatures of *Clerodendrum viscosum* leaf pastes treated goatskins show no significant changes between experimental and conventional salt curing techniques. As there are very few changes in the shrinkage temperature during the curing period for the experimental skins, this may be an excellent indication of the hydrothermal stability of the preserved skins. The minimal variation in the experimental leather shrinkage temperature may be a valuable indicator of the hydrothermal resistance of the preserved skins. Because of the curing agent's power of preservation, bacterial attacks do not compromise the integrity of the collagen protein matrix in sheepskin.

Table 4: Effect of technological processes on retanned leather properties of *Albizia spp* biocide recipe and salt-preserved goatskins

Index	30% of 5mg/mL recipe (Mean±SD)	30% of 1mg/mL recipe (Mean±SD)	30% NaCl (Mean±SD)
Tensile strength [N/mm^2]	17.50±0.01	16.30±0.14	22.40±0.01
Elongation at break (%)	112.9±0.11	49.70±0.14	59.60±0.01
Hardness	66.00±0.07	73.70±0.14	73.70±0.14
Moisture content (%)	50.19±0.01	47.26±0.01	49.28±0.02
Resistance to compression	2.59±0.01	1.64±0.01	1.90±0.01
Lastometer	46.00±0.07	26.46±0.01	50.58±0.06
Water vapour permeability	0.102±0.001	0.031±0.21	0.025±0.00
Thickness (mm)	2.14±0.01	1.93±0.01	1.79±0.02
Apparent density	0.36±0.01	0.36±0.01	0.64±0.02

Physical Characterization of Processed Leather

Leathers' physical characteristics, which are impacted by preservation methods, are essential in assessing their value. The physical properties of the chrome retanned leather that had undergone experimental treatment were evaluated using tests for tensile strength, grain crack, percentage of elongation, tear strength, etc. (Table 4). Tensile strengths were 17.50±0.01, 16.30±0.14, and 22.40±0.01 kg/cm^2 for the finished leather with a 30 % concentration of 5 mg/mL of the recipe and 1 mg/mL of the recipe, as well as standard samples. According to Gendaszewska *et al.* (2024), the moisture content of the chrome retanned leather was 50.19±0.01 % (30 % of 5mg/mL of recipe), 47.26±0.01 % (30 % of 1mg/mL of recipe), and 49.28±0.02 % (30 %' NaCl, control) are in cognizance with their report. These results may be related to the high bonding quality and fiber strength, which enhances the overall strength of the leather. The current technique of sheepskin preservation with *Albizia* biocide can serve as a worthwhile alternative to traditional salt-curing approaches.

Plate 1: Tanned Leather Preserved with Biocide Recipe

Plate 2: Tanned Leather Preserved with Salt

CONCLUSION

This study highlights the efficacy of *Albizia lebbeck* extracts as eco-friendly biocides for sheepskin preservation, achieving antimicrobial activity comparable to conventional salt methods. Future studies should focus on scaling production, evaluating long-term environmental benefits, and assessing in vivo safety for broader applications.

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