# Bio-Intervention Phyto-Based Material For Raw Goatskin Preservation: A Cleaner-Sustainable Approach

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**Research Article**

**Keywords:** Phyto-based, Wastewater, Chloride, Preservation, Persicaria hydropiper, Total Dissolved Solids

**Posted Date:** August 9th, 2021

**DOI:** https://doi.org/10.21203/rs.3.rs-627961/v1



**Version of Record:** A version of this preprint was published at Environmental Science and Pollution Research on January 10th, 2022. See the published version at https://doi.org/10.1007/s11356-022-18544-0.

# Abstract

The regular practice of using sodium chloride to preserve raw animal skin triggers increasing salinity and total dissolved solids (TDS) in the surface and groundwater during rehydration soaking operations. The process disrupts the lives of animals, plants, and human beings. This paper is focused on the phyto-based short-term preservation of goatskin to reduce salinity in tannery soaking operations. The indigenous *Persicaria hydropiper* leaf was investigated to assess the preservation of animal skin to diminish salinity and TDS of tannery soaking wastewater. Methanol extracted leaf was characterized by GC-MS and FTIR for chemical composition analysis and affiliated functional groups. Fresh goatskins were preserved at the preliminary, laboratory, and pilot-scale scenarios to establish the best possible mixture, monitor the moisture and nitrogen content, shrinkage temperature, microorganism analysis, and pollution load at each level. The processed leathers derived from the preserved skins with an optimal mixture of 10% leaf paste with 8% salt and conventional 50% salt were tested for their physical strength. Finally, the modification in fiber structure due to the varieties of preserving chemicals was evaluated through a scanning electron microscope (SEM) and detected insignificant variation of leather fibers. The findings reported in this study can be applied to the industrial level and remove certain amounts of salinity and TDS from tannery soaking wastewater.


# Highlights

- Leaf paste of *Persicaria hydropiper* is used for eco-friendly goatskin preservation
- Significant reduction of chloride, TDS, BOD and COD in soaking wastewater
- Fiber structures of optimized experimental and control leathers are similar

# Introduction

The contribution of the leather industry to the economy of Bangladesh is huge. In mid-2017, the government of Bangladesh shifted the tannery-based cluster of Hazaribagh, Dhaka to the Savar tannery industrial park, Hemayetpur, also in Dhaka. In terms of export revenue, the leather industry holds the second position in Bangladesh's economy with export revenue of $1.08 billion (Hong, 2018). However, the quality of leather produced in Bangladesh is well known throughout the world and controls at least 10% of the global leather market. The essential material of the leather industry is the raw hides/skins which are produced from meat industries as by-products. Hides and skins are composed of protein-polymer matrix which is called collagen but the principal components of hides/skins are keratin, moisture, fat, protein, minerals, and carbohydrates (Unango et al. 2019). Hides and skins are associated with different organisms which remain in a dormant state in the living animal owing to the body's defense mechanism. After slaughtering, the natural defense reaction switches off and the dormant bacteria go into action within 5–6 hours (Shede et al. 2008). It has been observed that in one square centimeter of the skin's surface about one thousand bacteria are present. Gram-positive bacteria are the most common types of bacteria which are present on the skin's surface (Santos and Gutterres, 2007). Due to bacterial action, a

variety of defects have appeared like hair slip, looseness, holes, discoloration, and other weaknesses which are responsible for the deterioration in leather quality (Rangarajan et al. 2003).

To produce high quality leather, raw hides and skins have to go through a temporary ‘preservation’ or ‘curing’ period (Vinodhkumar et al. 2016). Hides and skins can be preserved in two strategies: firstly, stopping the bacterial action; or secondly, destroying all the bacteria. Based on these strategies, preservation methods can be categorized as chemical, biological, phytochemical, or physical in character (Balasubramanian et al. 2019). The traditional and most common preservation method is the use of 40–50% salt based on the pelt weight. Salt curing is the most widely employed method due to its cost-effectiveness but from the environmental point of view, it is damaging. The conventional preservation process creates more than 70% of TDS and 40% of chlorides in total tannery effluent (Peng et al. 2014). It is estimated that from one ton of wet salted skins 7–9 $m^3$ wastewater is generated in soaking operations. This wastewater contains huge amounts of organic and inorganic pollutants like COD, TDS, BOD, and $Cl^-$ which have become major problems for tanners (Islam et al. 2014).

Different biological, chemical and physical technologies can remove some pollutants to safer levels. On the other hand, some pollutants cannot be removed at all. Among them, TDS remains at a higher level even in treated effluent (Lofrano et al. 2013). The major source of TDS is the salt used in the preservation process. The discharged wastewater containing high TDS has an adverse effect on soil quality and seed germination (El-Sheikh et al. 2011). Again, the salt curing process cannot destroy or resist the activity of halophilic bacteria which is responsible for creating red heat defects in animal skins (Caglayan et al. 2015). Furthermore, 75% of the utilized salts end up as effluent in the soaking operation which creates serious environmental pollution. The released salts compromise the immobilization of heavy metals in soil and water bodies which further damage or kill flora and fauna (Kahlown and Azam, 2003).

It is now an obligatory issue to find alternative preservation methods that can replace salt curing. In recent times, researchers have attempted to develop different chemical, biological, physical, and phytochemical strategies. Chemical methods include different chemicals to replace salts. The most commonly used chemicals are sodium hexafluorosilicate (Virgilijus et al. 2017), sodium sulfate (Vankar and Dwivedi, 2009a), boric acid (Kanagaraj et al. 2005), silicate, and ozone (Sivakuma et al. 2010). The main disadvantage of chemical-based preservation is the strong possibility of producing secondary air and water pollutants that need to be treated. Physical preservation methods comprise electrical current (Birbir et al. 2015b), vacuum (Gudro et al. 2014), direct sunlight, and frost (Narayanan et al. 2014). Except for the sun-drying process, all the other physical methods of preservation are expensive. However, the sun-drying preservation technique requires more time, a warmer climate, and special arrangements (Wu et al. 2017). Recently, phyto-based preservation process has increased in popularity and this method is carried out by using different natural sources, for instance *Semecarpus Anacardium* (Iyappan et al. 2013), *Tamarindus Indica* (Tamil Selvi et al. 2015), *Moringa oleifera* (Hashem et al. 2018), de-oiled neem cake (Vedaraman et al. 2016), and *Aristolochia bracteolata L.* (Vaghasiya and Chanda, 2007).

Mohammad et al. (2016) found that 10% of *Rumex abyssinicus* root with 15% of NaCl can effectively preserve goatskin for 30 days and most importantly this process can reduce the chloride content and TDS in soaking liquor. Nur-A-Tomal et al. (2020) reported that the extracted *Aphanamixis polystachya* oil is an environmentally friendly preservative agent which could preserve goatskin for more than 30 days. The use of a natural source in the preservation process mainly relies on its antimicrobial activity and availability (Kanagaraj et al. 2014). *Persicaria hydropiper* is a member of the Polygonaceae family, it has a good reputation in medical disciplines and most commonly used in Western and Eastern herbal medicine (Ayaz et al. 2014). Al-Mahmud and Lina (2017) claimed that *Persicaria hydropiper* leaf paste has antioxidant, antibacterial, anti-inflammatory, antihyperglycemic, and cytotoxicity properties which indicate its suitability as a curing agent. The local word for *Persicaria hydropiper* is 'Biskatali' and it grow on the banks of rivers, lakes, canals, and near roadsides in south Asian countries (Mabberley, 2017).

This research paper applies *Persicaria hydropiper* leaf paste to preserve goatskins by replacing common salts. The capacity of *Persicaria hydropiper* as a curing agent was assessed by monitoring different factors: moisture content, hair slip, Total Kjeldahl Nitrogen (TKN), thermal stability, and bacterial count. During preservation period, the efficacy of *Persicaria hydropiper* was compared to traditional preservation. Preservation via *Persicaria hydropiper* leaf paste could significantly reduce pollutants derived soaking liquor and make the preservation process eco-friendly.

# Materials And Method

### Sample collection

For conducting the preservation process, fresh goatskins of 4-5 sq. ft. were collected from a local slaughterhouse in Khulna, Bangladesh. The collected goatskins were washed to clean away dirt, dung, blood and other impurities and hung for 30 min. *Persicaria hydropiper* which is otherwise known as 'water pepper leaf' was collected from Khulna, Bangladesh. The leaf was cleaned with water and turned into a paste in the laboratory using a mortar and pestle. The leaf paste was prepared immediately before the preservation process so that it could be used while it was fresh.

### Chemicals and reagents

Commercial grade sodium chloride, pre-tanning, and tanning materials were procured from a local tannery at Khulna, Bangladesh. Laboratory-grade reagents were purchased from the largest scientific store in Khulna. The physicochemical analysis was carried out with laboratory-grade reagents and leather processing was continued by commercial-grade chemicals.

### Characterization of leaf paste

### GC-MS

For GC-MS analysis, fresh *Persicaria hydropiper* leaf was dried for 48 h at 70°C in an oven and turned into a fine powder. The methanol extract was prepared following the Soxhlet extraction method with 20 g leaf

powder. After extraction, the extracted material was cooled and then dried at 50°C to evaporate the solvent. The extracts were stored at 4°C in a dark place and utilized for GC-MS study. About 2 µl of methanol extracted sample was placed in a GC-MS (GCMS-QP2010, Ultra) device where helium flowed at a 1 mL/min rate as carrier gas. The temperature was increased from 40°C to 250°C maintaining a 10°C/min rate of increase. The temperature of the injector and detector was controlled at 250°C and 280°C, respectively.

### FTIR analysis

Fourier transforms infrared (FTIR) spectroscopy identifies the functional groups present in a molecule. The methanol-extracted *Persicaria hydropiper* leaf was investigated for residual functional groups employing an FTIR spectrometer (Perkin Elmer, USA). The recorded FTIR spectra were within the 4000 to 400 $cm^{-1}$ wavelength range for the adsorbent placed on KBr discs.

### Working Procedure

### Preliminary experiment

For the preliminary experiment, a freshly flayed goatskin was divided into four pieces of 25 cm × 25 cm (Figure 1) and weighed separately. The skin pieces were applied with 10% leaf paste and/or varying amounts of salt (0%, 4%, 8%, and 12%) based on weight. A preliminary experiment was carried out for two weeks (14 days) and assessed to find the organoleptic properties, signs of putrefaction, and microbial growth.

### Laboratory experiment

For laboratory testing, another fresh goatskin was cut around the backbone to split it into two parts. One section was examined for commercial preservation purposes and this became the ‘control sample’, while the other was tested with the best preserving materials obtained from preliminary data and termed the ‘experimental sample’. Figure 2 depicts the control and experimental samples as being preserved by 50% salt and 10% plant paste with 8 % salt, respectively, for one month (30 days). During preservation, the moisture, total nitrogen, bacterial load, and hydrothermal stability were continuously monitored at fresh, $1^{st}$, $4^{th}$, $7^{th}$, $14^{th}$, $21^{st}$, and $30^{th}$ day.

### Pilot experiment

For small-scale pilot production, five fresh goat skins were gathered from local shambles in Khulna and cleaned with water. After draining the water, the skins were cured with 10% plant paste and 8% salt mixture based on their weight and preserved for 30 days. During this period, the moisture and hydrothermal stability were monitored. The leather prepared from the plant-cured skins was assessed for its physical strength properties.

### Process monitoring

### Dehydration rate analysis

The moisture's changing dehydration rate due to the application of a curing agent was assessed by following the Bureau of Indian Standards (BIS, 1971). In this method, three separately weighed skins ($W_1$) were dried in an oven at 105°C±5°C and weighed and dried again until the weight change was <0.1 mg. The dried samples were cooled using a desiccator and finally weighed ($W_2$). The loss of moisture, its percentage, and standard deviation were calculated from the mean values of $W_1$ and $W_2$.

### Total Kjeldahl Nitrogen (TKN) assessment

The skin extract after the preservation period was prepared from 3 g of skin in 30 mL distilled water after agitating it at 200 rpm for half an hour. The extract was filtered and nitrogen was measured by the Kjeldahl method of APHA (2012) after the filtrate was properly digested. This exercise was carried out in triplicate and the mean value and standard deviation were calculated.

### Shrinkage temperature assessment

Shrinkage temperature denotes the hydrothermal stability of the hide and skin. It was tested by the SATRA shrinkage tester (STD 114, UK) according to the standard method ISO 3380:2015 (ISO, 2015).

### Microorganism analysis

The growth of microorganisms, especially bacteria in the experimental and control sample was analyzed from the skin extracts as collected for nitrogen determination. The extract was serially diluted with 1 mL extract into 9 mL distilled water three times for a 0.001 dilution. A molten nutrient agar solution was prepared from 3.9 g agar powder in 100 mL of distilled water. From the diluted extract, 0.1 mL sample was poured onto a pre-sterilized Petri plate and into it, the molten agar was added at 40°C. The mixture was shaken clockwise and counter-clockwise for the proper distribution of bacteria. The plates were retained in the incubation chamber for 24 h at 37°C. The bacterial colony was counted with a colony counter (Cruickshank, 1965).

### Leather processing

The experimental and control preserved goatskin samples were processed to produce shoe-upper leather following a commercial recipe after 30 days. The leathers were then tested for their physical properties.

### Assessment of leathers

To assess the physical strength of the experimental and control leathers, they were firstly conditioned and treated at 20±2°C and relative humidity of 65±2% for 48 hours. After conditioning, the leather samples were tested according to IUP/2 (1958). The physical strength properties, namely tensile strength, elongation, and ball bursting strength were determined following standard procedure ISO 3376 (2011) and ISO 3379 (2015).

### Pollution load estimation

To estimate the pollution load of preserved samples, physicochemical parameters of the soaking wastewater during rehydration of skins were evaluated through chloride ($Cl^-$), total dissolved solids (TDS), biochemical oxygen demand (BOD), and chemical oxygen demand (COD). For this part of the analysis, standard protocols of the APHA (2012) were employed. This helped to estimate the efficiency of the proposed preservation strategy to reduce pollution during soaking operations.

### Fiber structure investigation

The impact of the preservation process on the fibres of the leathers was scrutinized through scanning electron microscope (SEM) (JEOL JSM-7600F, USA). The leathers placed on conducting carbon tape were photographed at 20 kV with a 250X magnification.

## Results And Discussion

## GC-MS analysis

Table 1 summarizes the major chemical components obtained by GC-MS of the *Persicaria hydropiper* leaf paste: Phytol (55.327%), 9,12-Octadecadienoic acid (8.669%), Phthalate (diethyl) (7.623%), Di-n-octyl phthalate (5.182%), Limonene (4.789%), n-Hexadecanoic acid (4.094%) and 9-Octadecenamide (3.877%). The presence of phytol is supreme with a maximum concentration of 55.327% which is found to have both antimicrobial and antioxidant properties (Costa et al. 2012). Agoramoorthy et al. (2007) observed that both 9,12-Octadecadienoic acid and n-Hexadecanoic acid creates unfavorable conditions for the growth of Gram-negative bacteria. Although Phthalate (diethyl) and Di-n-octyl phthalate are considered carcinogenic pollutants, these compounds did demonstrate antifungal and antibacterial activity against *Aspergillus flavus, Bacillus subtilis*, *Escherichia coli*, and *Staphylococcus aureus* (Ortiz and Sansinenea, 2018). Limonene is found to have antimicrobial properties and is used as a preservative (Hąc-Wydro et al. 2017). Other chemical compounds like Iso E Super (1.108%), Octanal, 2-phenylmethylene (0.972%), 9,9-Dimethoxybicyclo nona-2,4-dione (0.628%), Phthalic acid, butyl undecyl ester (1.1916%), and Diisooctyl phthalate (1.027%) are also present in *Persicaria hydropiper* leaf paste. Due to having these phytochemicals in the *Persicaria hydropiper* leaf, it could be used to preserve raw animal skin.

Table 1
Chemical composition of methanol extracted Persicaria hydropiper leaves paste

| No. | Name of the compound | Concentration (%) | Molecular formula |
|---|---|---|---|
| 1 | Limonene | 4.789 | $C_{10}H_{16}$ |
| 2 | Phthalate (diethyl) | 7.623 | $C_{12}H_{14}O_4$ |
| 3 | Iso E Super | 1.108 | $C_{16}H_{26}O$ |
| 4 | Octanal, 2-phenylmethylene | 0.972 | $C_{15}H_{20}O$ |
| 5 | n-Hexadecanoic acid | 4.094 | $C_{16}H_{32}O_2$ |
| 6 | Phytol | 55.327 | $C_{20}H_{40}O$ |
| 7 | 9,12-Octadecadienoic acid | 8.669 | $C_{18}H_{32}O_2$ |
| 8 | 9,9-Dimethoxybicyclo nona-2,4-dione | 0.628 | $C_{11}H_{16}O_4$ |
| 9 | Phthalic acid, butyl undecyl ester | 1.916 | $C_{23}H_{36}O_4$ |
| 10 | 9-Octadecenamide | 3.877 | $C_{18}H_{35}NO$ |
| 11 | Diisooctyl phthalate | 1.027 | $C_{24}H_{38}O_4$ |
| 12 | Di-n-octyl phthalate | 5.182 | $C_{24}H_{38}O_4$ |

# FTIR analysis

The methanol-extracted *Persicaria hydropiper* leaf showed several peaks at a different wavelength ($cm^{-1}$) as illustrated in Fig. 3. The peaks indicate the presence of functional groups from different compounds. Stretching and bending of O-H functional group were observed at 2918, 3356, and 1382 $cm^{-1}$ wavelengths, respectively, strongly suggesting there are weak and strong intermolecular bonds of an alcohol group. A peak at 844, 1045 $cm^{-1}$ interacts strongly with the C-Cl group from halo compound and CO-O-CO stretching of anhydride. The presence of C-O stretching was also observed at 1134, 1292 $cm^{-1}$ and these numbers reveal strong aliphatic ether, aromatic ester groups. Stretching of S = O group at 1045, 1382 $cm^{-1}$ introduces the sulfoxide and sulfone group. C-N, C = C stretching of aromatic amine and conjugated alkene at 1292 and 1620 $cm^{-1}$ represent strong and medium interaction, respectively. The functional group N-H bending and N-H stretching was found at 1620, and 2918, 3356 $cm^{-1}$ which indicating the amine, an amine salt, aliphatic primary amine groups. N-H stretching was also reported by Balasubramanian (2019) during the preservation of skin using sodium polyacrylate. C-H bending and C-H stretching at 1826, 2918 $cm^{-1}$ demonstrate weak aromatic compound and medium alkane compound.

# Optimization of preserving agent

Different percentages of *Persicaria hydropiper* leaf paste with or without common salt was applied (Table 2) to find the ideal amount of leaf paste that could preserve goat skins. The efficacy of leaf paste was assessed by both visual observation and with physically hand feel. Visual observation reveals that *Persicaria hydropiper* leaf paste could preserve goatskin in any condition without odor creation and no hair slip (Fig. 2). However, physical feeling was different according to the leaf paste percentage. Preservation of goatskin with only 10% plant paste makes the skin hard which could create difficulties in further processing. It should be noted that 10% plant paste with 12% salt produces very soft and flexible leather after 14 days which could enhance the damaging possibilities if it is processed in a drum. It seems that preservation with 10% plant paste along with 8% salt followed a similar trend to conventional salt curing and after 14 days of preservation the skins became flexible. It means they could be easily processed in a drum and there will be no deterioration (Fig. 2). Hence, 10% plant paste + 8% salt was chosen as the optimized combination for experimental purposes.

Table 2
Leaf paste optimized for the preservation method (14 days)

| No. | % of curing agents | Hair slip | Odor | Physical feel |
|---|---|---|---|---|
| 01 | 10% plant paste | No | No | Hard |
| 02 | 10% plant paste + 4% salt | No | No | Medium hard |
| 03 | 10% plant paste + 8% salt | No | No | Flexible |
| 04 | 10% plant paste + 12% salt | No | No | Soft and flexible |

# Bacterial load

Bacterial count reveals the number of bacteria present in the preserved skins which helps in determining the efficacy of the new preservation method. The identified bacterial colonies that exist in the control and experimental skins during different time intervals of the short preservation period are presented in Fig. 4. In the early days of preservation, the bacterial load rose for both experimental and control samples. A possible explanation for this is that there might be certain types of bacteria present which can tolerate both the leaf paste and common salts for a certain period of time. By this time salt and leaf paste tolerant bacteria increased the number of bacteria in the colony through binary fission (Masoodi et al. 2021). Although the bacterial count rose for both methods up to the 7th day, results were comparable and in some cases experimental samples confirmed better results. From the 14th day, the bacterial count gradually declined which indicates that salt and leaf paste tolerant bacteria had gone astray. At the end of the 30th day of the preservation period the bacteria load was higher in the control samples compared to the experimental samples. This means that *Persicaria hydropiper* leaf paste showed better results

when compared to conventional preservation methods. This phenomenon was observed due to the presence of an antimicrobial agent in the *Persicaria hydropiper* leaf paste (Prota et al. 2014).

# TKN content

The value of the TKN content is serving as an indicator of the breakdown of the polypeptide chain of the collagen matrix. Due to bacterial degradation, the polypeptide chain of the goatskin is the breakdown by which components with ammonia and the amino group increase, and this generates a bad odor and promotes hair slip defects in the preserved skin (Vankar and Dwivedi, 2009b). Figure 5 depicts the extractable nitrogen content during preservation of the experimental and control skins. The figure shows that the value of the TKN for both the experimental and control samples increased during the initial 4 days of preservation. From the 5 day to 14 day the extractable nitrogen content value fell drastically which indicates that preservation efficiency was significantly increased for both samples. On the 21st day for the experimental and control samples, extractable nitrogen content was 3.12 g/kg and 2.87 g/kg, respectively. From the 21st day to the last day of preservation (30th day) the values remained almost unchanged for both samples. Throughout the preservation period the experimental sample showed slightly higher nitrogen value compared to the control sample, but there was no sign of hair slip and odor. Consequently, it can be stated that using 10% *Persicaria hydropiper* leaf paste with 8% salt goat skin makes preservation for 30 days possible.

# Moisture content

The percentage of moisture content is considered one of the major parameters when assessing the preservation efficiency. Moisture content in the raw skin is a major parameter for assessment of the curing competency. Raw skin contains 65–70% moisture which is a favorable state for bacterial survival. One of the main aims is to reduce the moisture content to a significant level. The moisture content (%) of the experimental and control goatskins during time intervals of preservation is illustrated in Fig. 6. It is observed from Fig. 6 that for both samples in the first 7 days the moisture content fell remarkably. On 14th, 21$^{st,}$ and 30th days the moisture content remained static for both samples. For the experimental samples, moisture content decreased from 62.13–44.7%, which indicates that *Persicaria hydropiper* leaf paste has dehydrating properties. Sivabalan and Jayanthi (2009) have used *Cassia fistula* for the preservation of skins and observed a similar outcome. At the end of the 30 days' preservation, moisture content in both samples was 44.7% (experimental) and 42.4% (control), which are far below the critical moisture content (50%) (Kanagaraj et al. 2014). Despite larger moisture content in the experimental samples compared to the control sample, there was no symptom of putrefaction. This reveals the effectiveness of the preservation methods.

# Hydrothermal stability

The shrinkage temperature value acts as an index to determine the hydrothermal stability of the skins/hides. Shrinkage temperature indicates the structural stability of the collagen matrix which fluctuates due to the disintegration of stable linkages (Babu et al. 2012, Kannan et al. 2009). Natural plant extracts can alter the structural properties of collagen matrix negatively or positively which depends on the kinds of reaction involved (Vijayalakshmi et al. 2009). Shrinkage temperature value decreases if the hides/skins undergo detritions and increases if the plant extracts can give some tanning properties. The shrinkage temperature value during 30 days' preservation of the skin using experimental curing agent (10% plant paste + 8% salt) and conventional curing agent (50% NaCl) is depicted in Fig. 7. It was observed in this figure that throughout the preservation period, no crucial change in the shrinkage temperature of both samples was evident. For experimental and control samples, the shrinkage temperature values rose slightly from 65.41°C to 65.43°C and 65.25°C to 65.37°C, respectively, after 30 days' preservation. This slight increase in the shrinkage temperature value suggests that *Persicaria hydropiper* leaf paste positively alters the hydrothermal stability of the collagen matrix and can be used to preserve the skin protein.

# Estimation of pollution load

Both the experimental and control preserved goatskin samples were subjected to a soaking operation. The main purpose here was to restore the moisture content in the goatskins which was lost during the preservation period. Apart from water different detergents, enzymes, and fat remover are used as chemicals in this operation (Sawalha et al. 2019). The pollution loads emitted during the soaking operation for the experimental and control samples are listed in Table 3. The soaking wastewater of the control sample contains a high level of chloride, TDS, BOD and COD compared to the experimental sample. The main pollution loads of the tannery wastewater are chloride content and TDS which diminished to 53.56% and 53.13%, respectively, because the common salt was replaced by 10% *Persicaria hydropiper* leaf paste with 8% salt. It is observed that due to high $Cl^-$ and TDS wastewater, treatment cost increases and it is estimated that for every 4 kg of wastewater the approximate extra expense in the treatment process is US$1 (Balasubramanian et al. 2019). Other pollution parameters like BOD and COD revealed a significant reduction to 50.92% and 47.10%, respectively, in the experimental soaking wastewater when compared to the control. The results demonstrate that the developed *Persicaria hydropiper* leaf paste-based preservation method not only retains the goatskin but is also environmentally friendly.

Table 3
Reduction of pollution load during soaking operation

| Parameters | Experimental | Control | Unit | Reduction (%) |
|---|---|---|---|---|
| $Cl^-$ | 8965 ± 23 | 19303 ± 156 | mg/L | 53.56 |
| TDS | 21293 ± 61 | 45431 ± 72 | mg/L | 53.13 |
| BOD | 586 ± 31 | 1194 ± 23 | mg/L | 50.92 |
| COD | 2987 ± 32 | 5647 ± 59 | mg/L | 47.10 |

# Physical properties of processed leather

The type of preservation methods applied in the preservation process has a profound effect on the physical properties of the final leather product. The physical quality of the leather is assessed by deciding how it is used in the final product's preparation. The value of the leather mainly depends on the physical characteristics of the end product (Kim et al. 2018). The physical properties of the processed upper crust leather of the experimental and control samples are reported in Table 4. Judging by the results, it was observed that physical properties viz. tensile strength, elongation at break, and grain crack load of experimental skins preserved with 10% *Persicaria hydropiper* leaf paste along with 8% salt, showed better physical properties compared to the control samples. Tensile strength of the finished leather is considered a prime feature and it plays a significant role in how well manufactured products function (Sudha et al. 2009). The tensile strength of the experimental and control finished leather samples were 218.7 kg/$cm^2$ and 210.6 kg/$cm^2$, respectively. Though experimentally processed leather indicates lower grain crack distension value compared to the control sample, it fulfils the requirements of the standard. The physical properties of the produced leather demonstrate that *Persicaria hydropiper* leaf paste has a positive impact on the mechanical and structural properties of skin protein.

Table 4
Physical properties of the processed experimental and control leather

| Parameters | Experimental | Control | Required (Kanagaraj et al. 2001) |
|---|---|---|---|
| Tensile strength (kg/$cm^2$) | 218.7 | 210.6 | 200 |
| Elongation at break (%) | 44.03 | 43.5 | 40–65 |
| Bursting strength: | 7.8 | 8.1 | 7 |
| Distension, grain crack (mm) Load, grain crack (kg) | 25.3 | 24.8 | 20 |

# Physical properties of processed leather

Fiber arrangement plays an important role when assessing the quality of the final leather since the physical strength of final leather depends on fiber orientation (Covington 2011). The fiber composition of the final leather produced from the experimental and control samples are assessed through SEM images which are depicted in Fig. 8. These images showed that the fiber structure of the experimental sample is very similar to the control sample, suggesting that *Persicaria hydropiper* leaf paste played no part in disordering the fiber orientation. However, any destruction of the fiber structure was not observed in the experimental sample, meaning that the recommended curing agent could be used in the preservation process.

# Pilot trial

# Moisture content of pilot-scale trial samples

Figure 9 illustrates the moisture content (%) during the 30-day preservation period of the pilot trial samples (curing agent: 10% *Persicaria hydropiper* leaf paste + 8% salt). The percentage of moisture content for all five pilot experiments showed a similar kind of situation. Up to the 7th day, moisture content was marginally reduced, while from the 14th day to 30th day the moisture content virtually remained in a steady-state condition in all five experiments. During this 30-day preservation period no damage to the skins like bad odor, or hair slip was observed.

# Hydrothermal stability of pilot trial samples

Figure 10 presents the hydrothermal stability during the 30 days preservation period of the pilot-scale trial samples (curing agent: 10% *Persicaria hydropiper* leaf paste + 8% salt). Throughout the preservation period, no substantial change in the shrinkage temperature among the pilot-scale trial samples occurred. For instance, shrinkage temperatures of the fresh samples were 65.75°C, 66.32°C, 66.35°C, 65.98°C, and 65.65°C. These fell slightly to 64.68°C, 64.94°C, 64.98°C, 64.88°C, and 64.79°C, respectively, after 30 days of preservation for the pilot trial samples P1, P2, P3, P4, and P5.

# Physical properties of pilot trial samples

Table 5 summarizes the physical properties of the final leather processed from the pilot trial samples. All the physical characteristics of the five trial samples fulfil the standard requirements. The tensile strength value ranged from 207 kg/cm$^2$ to 219 kg/cm$^2$ which is above the standard value. Tensile strength, elongation at break, and grain crack load were also in the standard range for all five pilot trial samples.

Table 5
Physical properties of the processed pilot-scale experimental leather

| Parameters | P1 | P2 | P3 | P4 | P5 | Kanagaraj et al. 2001 |
|---|---|---|---|---|---|---|
| Tensile strength (kg/cm$^2$) | 209 | 213 | 219 | 207 | 215 | 200 |
| Elongation at break (%) | 45 | 43 | 46 | 44 | 42 | 40–65 |
| Bursting strength: | | | | | | |
| Distension at grain crack (mm) | 7.7 | 8.1 | 7.8 | 7.5 | 7.2 | 7 |

# Conclusion

*Persicaria hydropiper* plants contain several antimicrobial components which are analyzed through GC-MS analysis, and to what extent they can preserve goatskin for up to 30 days. The efficacy of *Persicaria hydropiper* leaf paste as a curing agent was assessed by investigating the moisture content, bacterial count, and shrinkage temperature of the preserved skins. Results disclosed that *Persicaria hydropiper* leaf paste-aided preserved skins showed indistinguishable or even better properties compared to traditional salt-preserved skins. The physical-organoleptic characteristics of the processed final leather which was preserved with *Persicaria hydropiper* leaf paste also meet the standard prerequisite. The SEM images confirmed that the fiber structure of the skins did not degrade when *Persicaria hydropiper* leaf paste was added. The newly devised preservation method is an eco-friendly approach as it reduced the $Cl^-$, TDS, BOD, and COD by 53.56%, 53.13%, 50.92%, and 47.10%, respectively, from the soaking wastewater compared to the traditional method. Subsequently, *Persicaria hydropiper* leaf paste appears to be a sustainable and convincing alternative curing agent that can replace pollution-creating salts.

# Declarations


### Acknowledgements

This work was supported by the Advanced Studies & Research (CASR), Khulna University of Engineering & Technology (KUET), Khulna, Bangladesh


### Conflict of interest

The authors declare no conflict of interest.

### Ethical Approval

The authors declare that the submitted manuscript is original. Authors also acknowledge that the current research has been conducted ethically and the final shape of the research has been agreed by all authors. Authors declared that this manuscript does not involve researching about humans or animals.

### Consent to Participate

The authors consent to participate in this research study.

### Consent to Publish

The authors consent to publish the current research in ESPR journal.

### Authors Contributions

Md. Abul Hashem: visualization/conceptualization, investigation, methodology, and writing-review. Md. Anik Hasan: investigation, methodology, supervision, data managing-organizing. Md. Abdul Momen: sampling and data collection. Sofia Payel: writing-original draft, and editing, Mehedi Hasan: writing-review Md. Zillur Rahaman Shaikh: review and editing. All authors read and approved the final manuscript.

### Availability of data and materials

The datasets generated and analyzed during the current study are not publicly available but are available from the corresponding author on reasonable request.

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# Figures

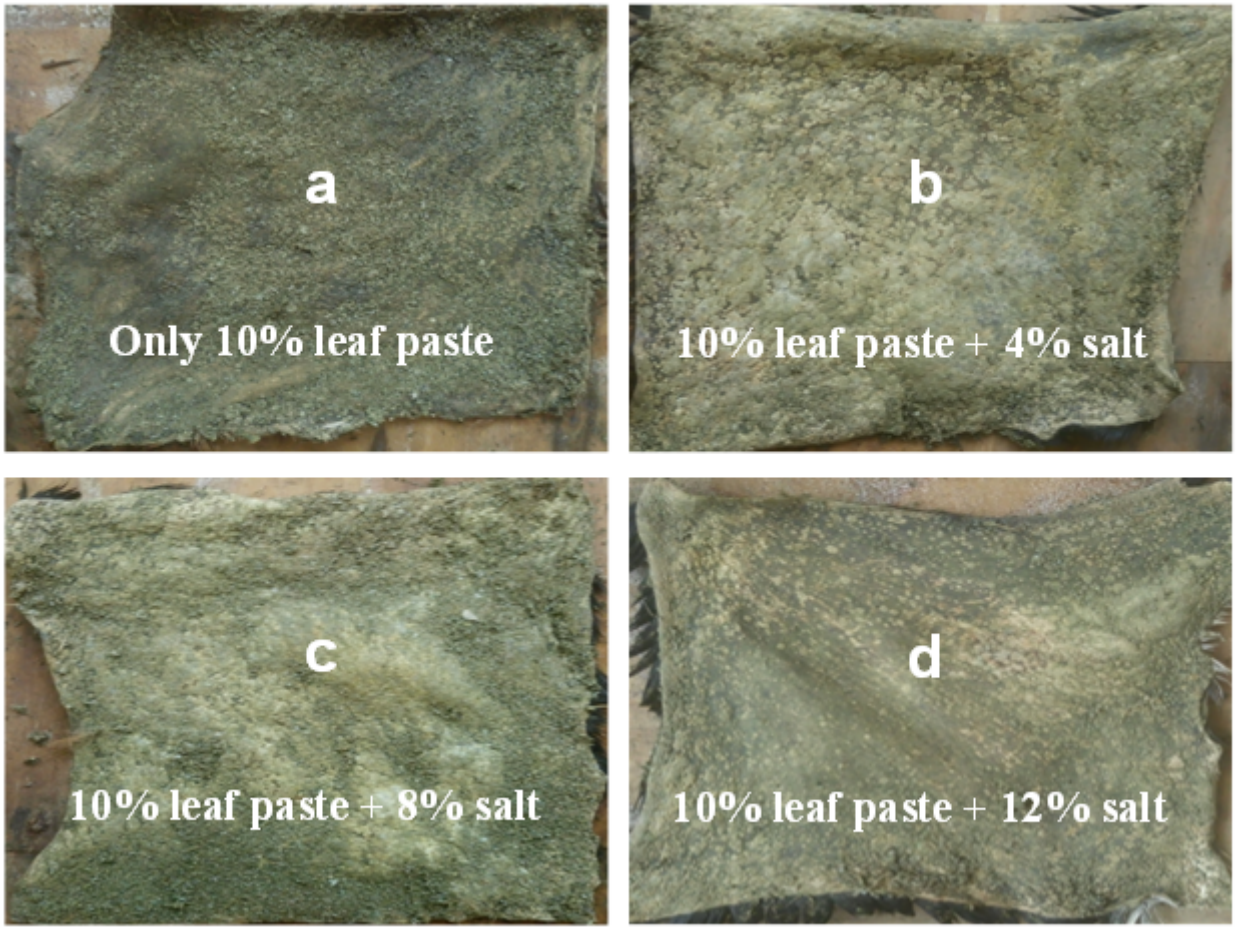



**Figure 1**

Preliminary experiments: a) leaf paste 10% b) leaf paste 10%+ salt 4% c) leaf paste 10%+ salt 8% and d) leaf paste 10% + salt 12%


**Figure 2**

Preservation with the present experimental (leaf paste 10% + salt 8%) and conventional (50% salt) methods

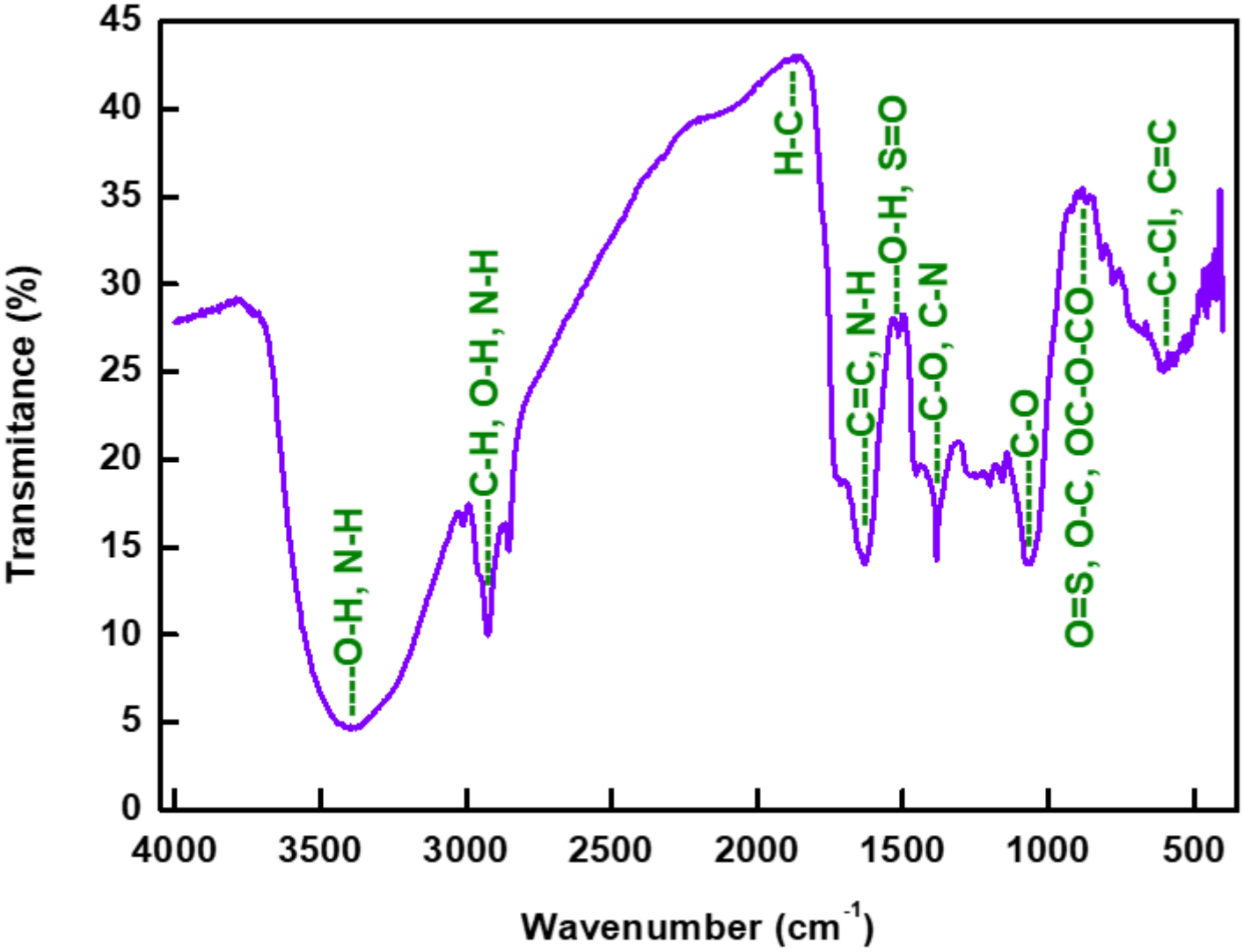



**Figure 3**

FT-IR spectrum of methanol extract of Persicaria hydropiper leaf

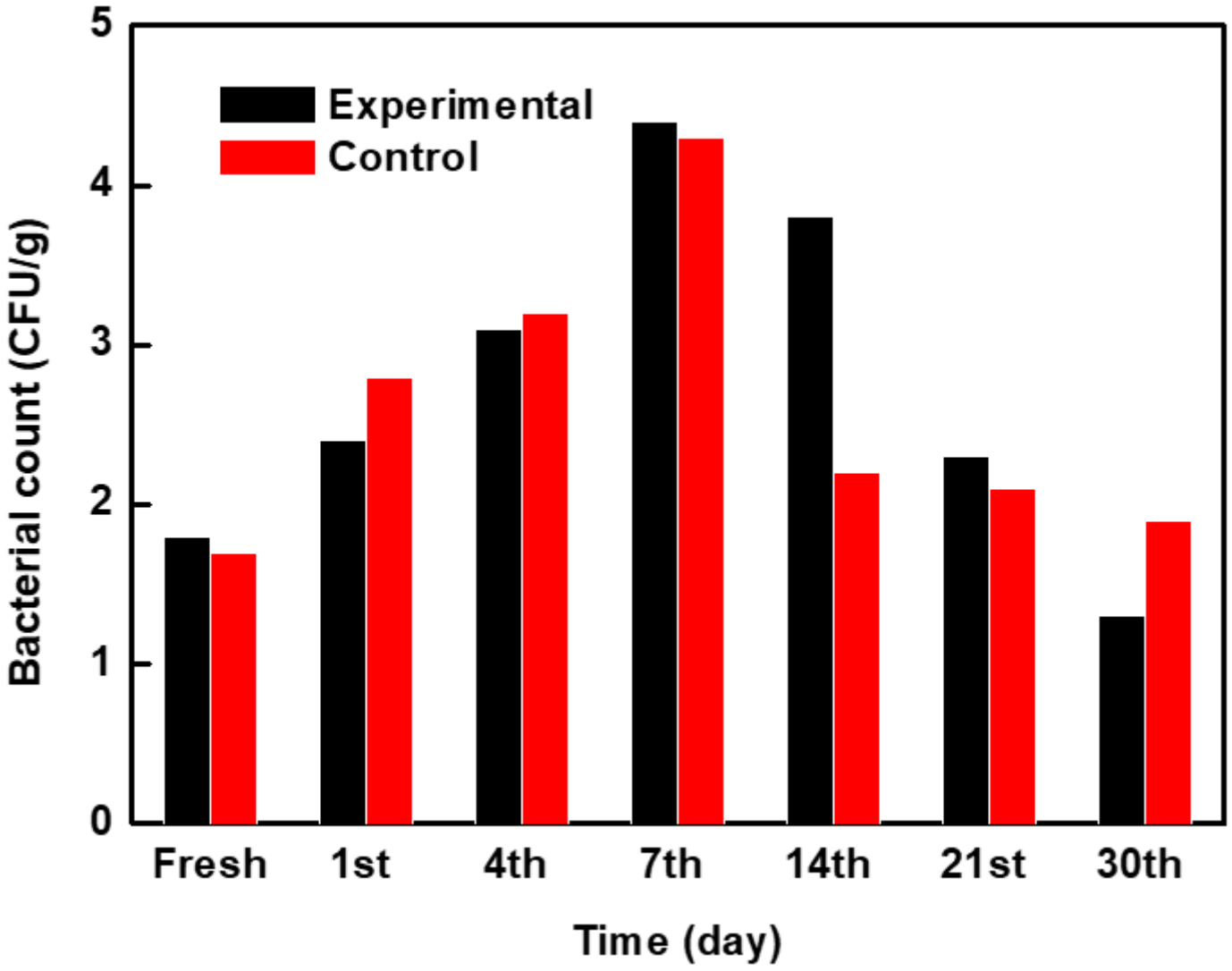



**Figure 4**

Bacterial loads in the Persicaria hydropiper leaf paste with lower (10% leaf paste + 8% salt) and experimental (50% salt) of the preserved goatskins. The y-axis value is expressed with×106 (CFU/g).

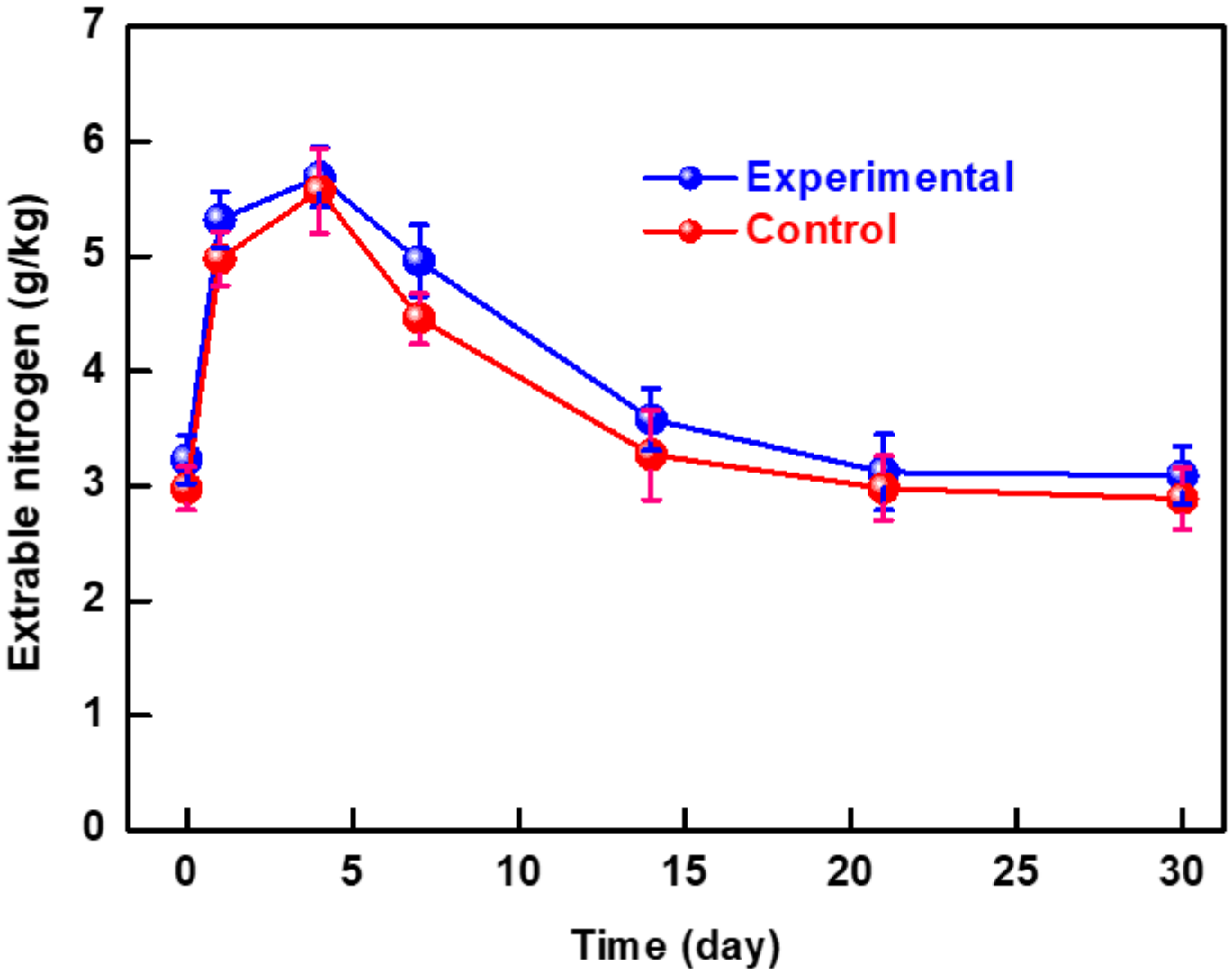



**Figure 5**

Extractable nitrogen content in control (50% salt) and experimental (10% leaf paste + 8% salt) of the preserved goat skins

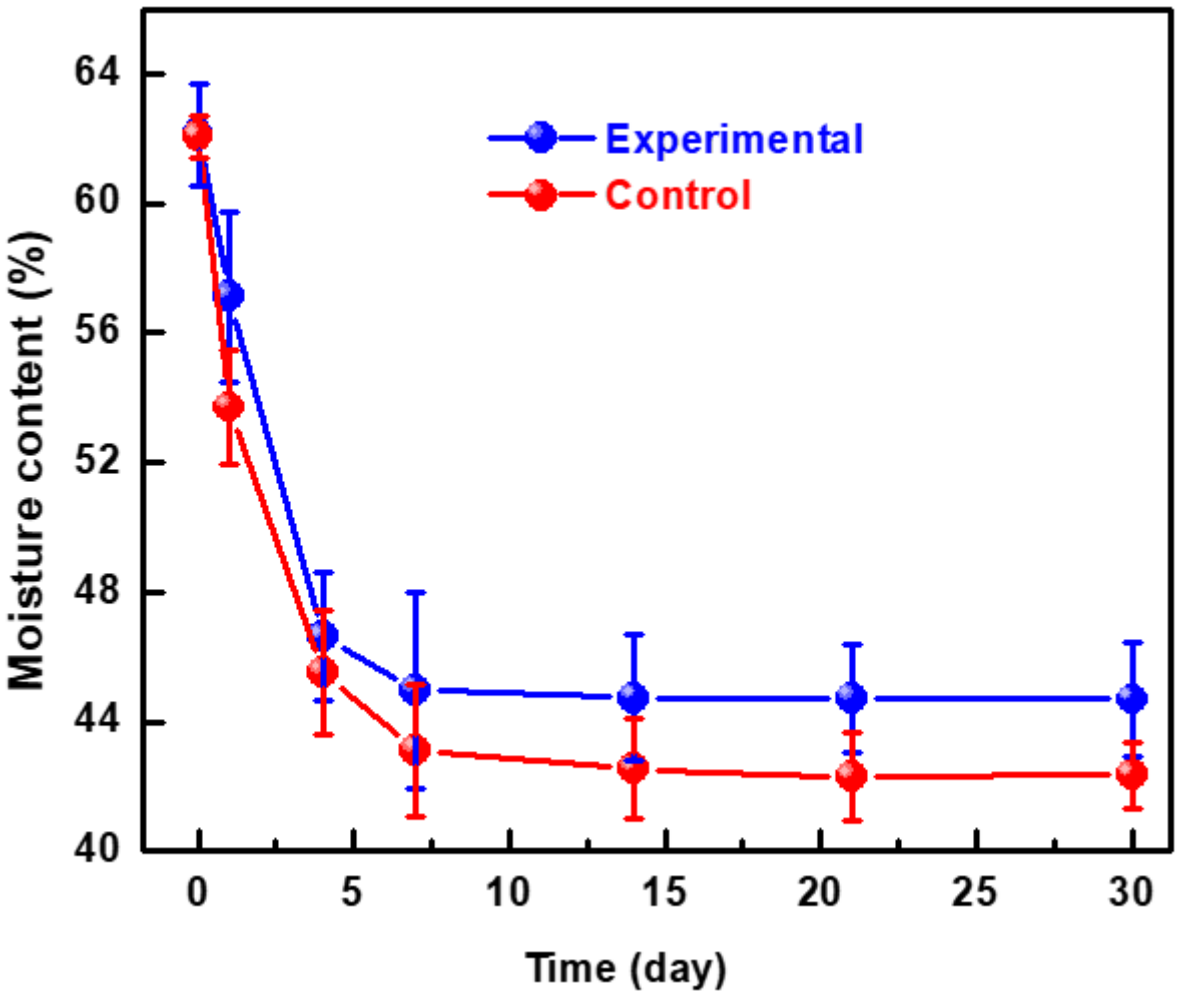



**Figure 6**

Moisture content in control (50% salt) and experimental (10% leaf paste + 8% salt) of the preserved goat skins

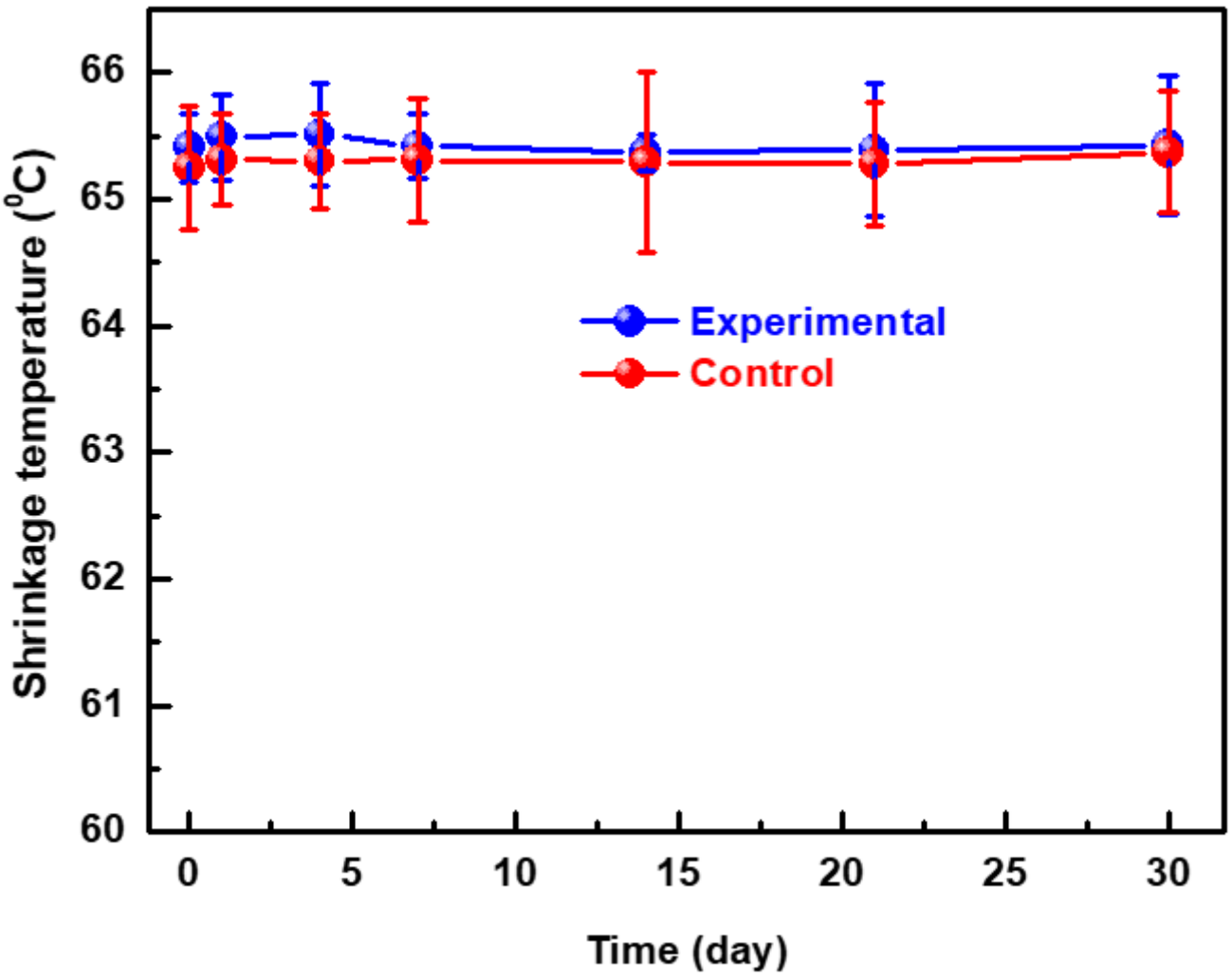
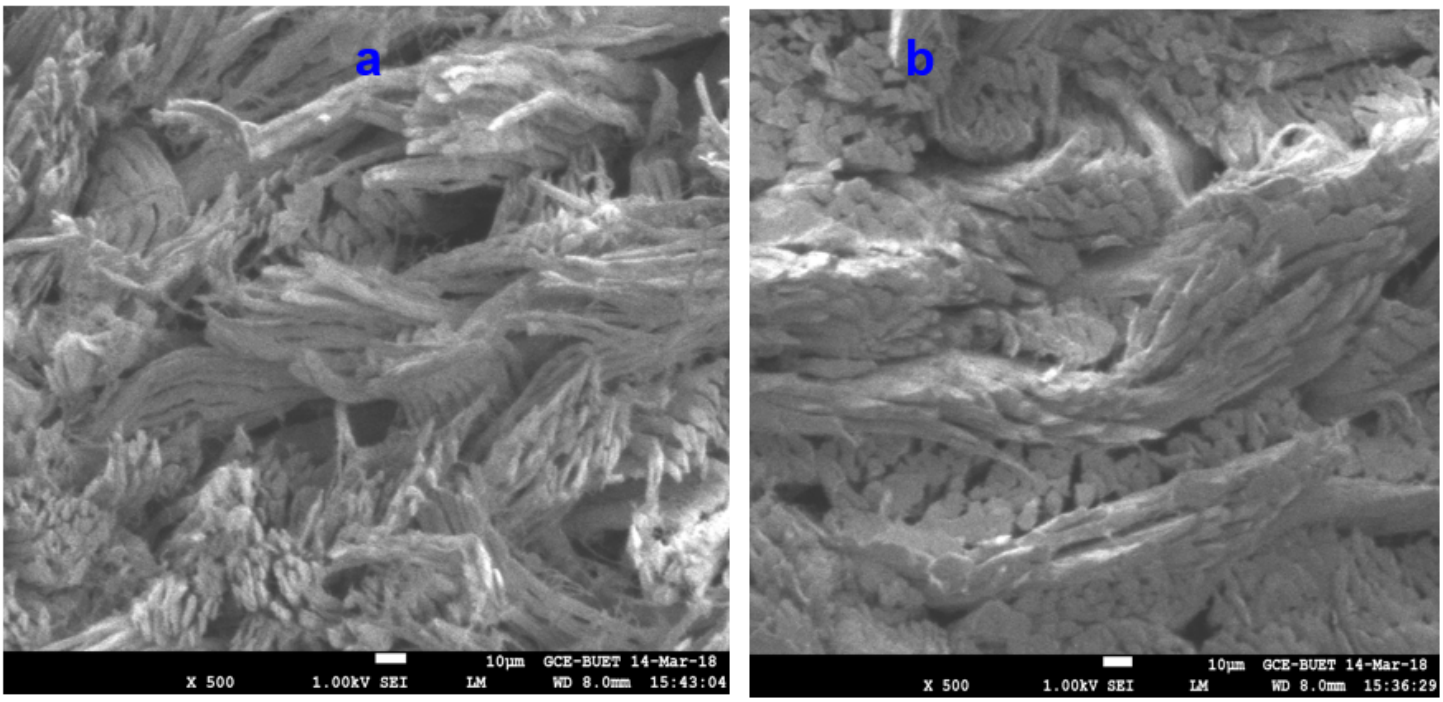



**Figure 7**

Shrinkage temperature in control (50 % salt) and experimental (10% leaf paste + 8% salt) of the preserved skins




## Figure 8

SEM photographs of the prepared crust leathers a) control (50% salt) and b) experimental (10% leaf paste + 8% salt) of the preserved skins

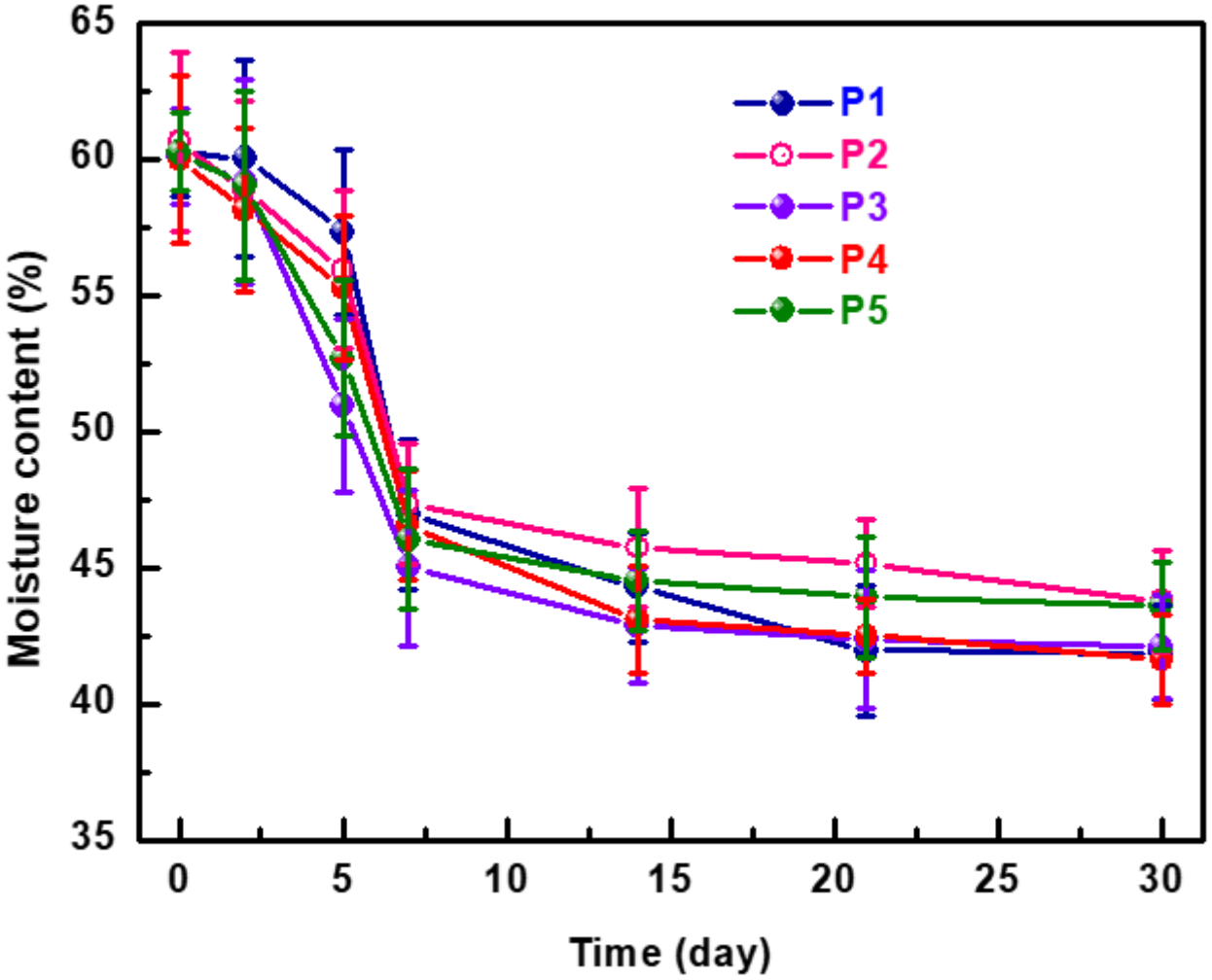



Figure 9

Moisture content of the pilot scale experiment with the preserving agents: 10% leaf paste + 8% salt

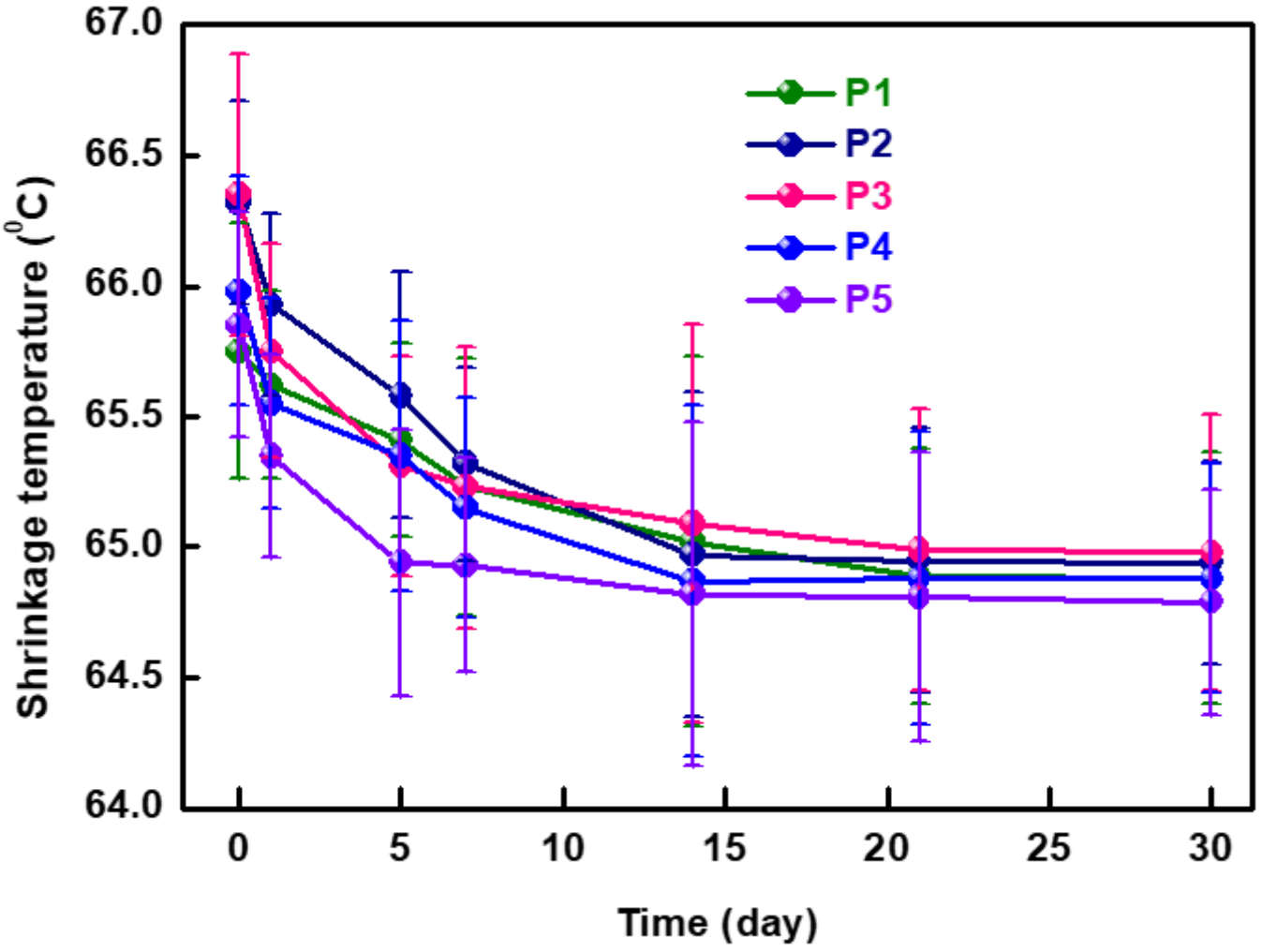



Figure 10

Shrinkage temperature of the pilot scale experiment with the preserving agents: 10% leaf paste + 8% salt