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REVIEW ARTICLE

Bio-preservation of raw hides/skins: A review on greener substitute to conventional salt curing

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Abstract

Raw hides/skins are considered to be the prime component for leather industry, which once flayed from animals, plummets to microbial attack. Their preservation combats putrefaction wherein curing using sodium chloride (NaCl) is by and large the most widely accepted method. However, there are few stumble blocks in using NaCl in terms of pollution load generated such as high total dissolved solids (TDS), total suspended solids (TSS), biological oxygen demand (BOD), chemical oxygen demand (COD) and chlorides (Cl^-). Additionally, this effluent when discharged affects the quality of the water, soil and plants causing huge ecological damage. To evade these problems, researches are being carried out to explore alternative preservation techniques which are either salt free or with reduced amount of salt. Different methods were proposed time and again which remained unfeasible due to associated drawbacks like high cost, health hazards and environmental concerns. Therefore, finding cheaper, eco-friendly and sustainable method for preservation has become the need of the hour for this industry. This review meticulously summarizes the changing trends in preservation techniques for past few decades with special emphasis on bio-based preservation. The diversity of the natural preservatives explored for the said purpose has been systematically reviewed. The enormous environmental benefits that can be obtained by adopting bio-based preservation and future avenues of research have been discussed.

Keywords Antimicrobial activity · Bio-preservation · Pollution parameters · Physical strength properties · Skin putrefaction

Introduction

The leather industry has been a key player in the global commerce market for millennia. Today it is unquestionably a major industry of huge economic importance on an international. In spite of the colossal benefits the leather industry brings, it is categorised under the 'red category' due to the damage it causes to the environment owing to high amounts of solid and liquid wastes generated during the processing. The leather making process predominantly consists of three stages viz. pre-tanning, tanning and post-tanning (Fig. 1), of which about 50–60% of the total pollution is generated only in the pre-tanning stage (Sivasubramanian et al. 2008). The tannery effluent possesses bazillions of environment polluting factors like TDS, TSS, BOD and COD (Jahan et al. 2014), wherein the common effluent treatment has been successful in removing the suspended solids and has appreciably lowered the BOD and COD values (Sabumon 2016). Nevertheless, there is no fool-proof technology available to treat the salt liquor, although, in tropical countries, the liquor is evaporated using solar evaporation ponds and salt is recovered manually from drying pans. However, this curing method is affected by climatic conditions, space availability, recovery and reuse of salts (Kanagaraj et al. 2020). Even though the prescribed Indian standard of TDS in released tannery effluent is 2100 ppm (Sivakumar et al. 2005), however, the actual value continues to range from 7000 to 10,000 ppm or even higher (Reda 2016). The conventional preservation process generates more than 70% of TDS and 40% of chlorides of total tannery effluent (Peng et al. 2014; Hashem et al. 2022). On processing 1 tonne of

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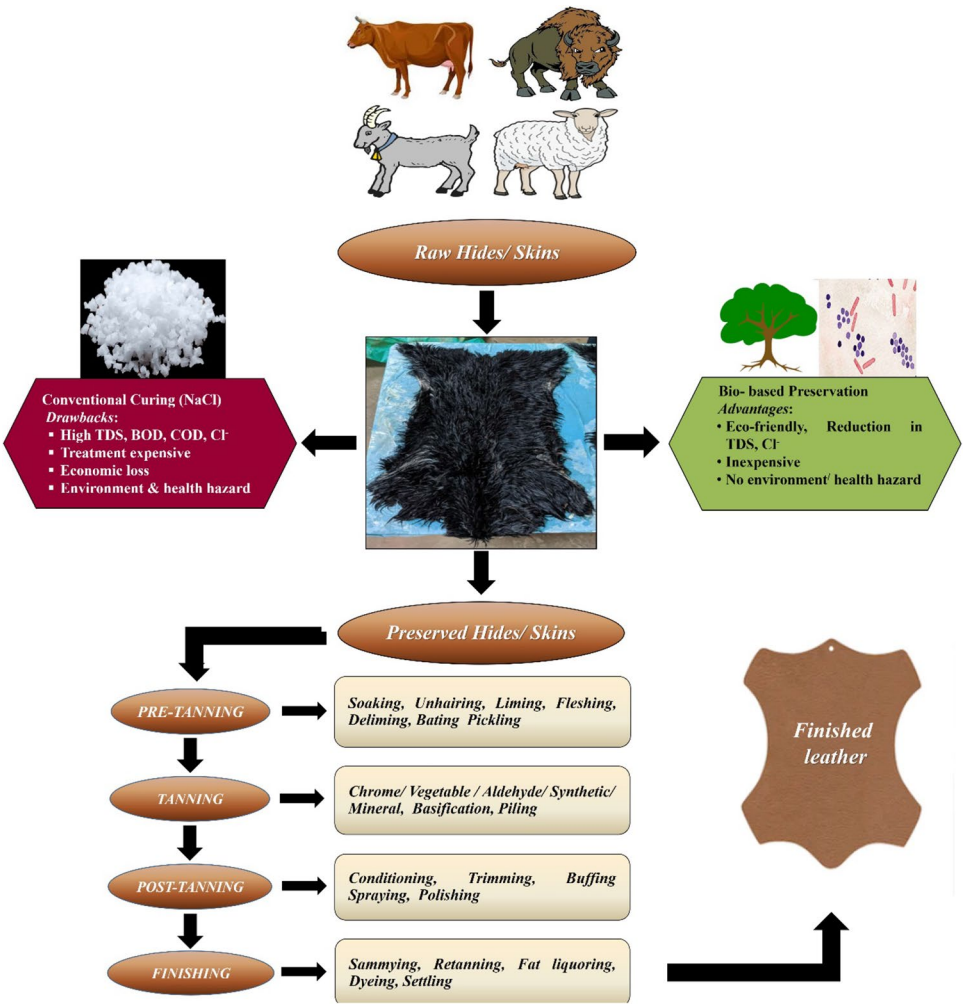

Fig. 1 Overview on various stages of leather making

hides/skins, around 40,000 L of effluent is generated contributing ~ 350–450 kg salt as TDS (Preethi et al. 2006). Despite such enormous pollution, preservation of raw hides/skins is inevitable, since its major constituents are moisture (65%), protein (33%), minerals (0.5%) and fatty substances (2–6%), which makes them highly susceptible to bacterial attack (Vijayalakshmi et al. 2009). Once the hides/skins are flayed from the animal in the slaughterhouse, they are normally taken to curing premises before their delivery to the tanneries for processing into leather. Therefore, the primary step of leather making involves preservation also known as 'curing' of hides/skins. Un-cured hides/skins harbour vast range of microorganism, derived from manure, air, soil, water and extraneous filth. To add to this, the high protein and lipid present in them provide a suitable substrate for prolific growth of microorganisms (Orlita 2004). Production of proteolytic and collagenolytic substance from these organisms will cause the disintegration of hides/skins eventually leading to poor quality leather thereby de-valuating it (Kayalvizhi et al. 2008). Hence, curing becomes unavoidable unless and until the hides/skins are going to be processed immediately into leather, in which case they may be stored at lower temperatures for a short time. Since most of the bacteria exhibit optimum growth temperature between 15 and 37 °C, better preservation of hides/skins could be achieved at temperature range between 10 and 18 °C (Kanagaraj and Chandra Babu 2002).

The use of NaCl imparts certain attributes which makes it the method of choice for preservation. It dehydrates the hides/skins creating a totally unfavourable environment for the growth of the bacteria. It also exhibits bacteriostatic effect and prevents their growth by plasmolysis (Elias et al. 2020). The major reason for using NaCl is also due to its low-tech process and cost feasibility (Kanagaraj and Chandra Babu 2002). As we know, before processing the cured hides/skins into leather, they need to be soaked in water which imparts re-hydration but also leads to the release of huge amount of NaCl in the soak liquor. This NaCl accounts for the high TDS and chloride levels rendering the ground water saline, which eventually affects the quality of soil and plants (Nur-A-Tomal et al. 2021). There is a limited scope to reuse the salt recovered in salt pans, as it is contaminated

with halophilic bacteria and organic matter (Alagamuthu et al. 2015).

Need for preservation: an insight into probable damage caused to un-cured hides/skins by putrefying microbes

Raw hides/skins contain majorly protein and moisture—the bacteria utilise them for their growth and proliferation, a process known as 'putrefaction'. Lots of research has been carried out on the types of bacteria causing putrefaction, wherein facultative aerobes and anaerobes have been reported to be majorly involved. A predominant role of Gram-positive bacteria in skin putrefaction has been reported (Mohamed et al. 2016), wherein among a total of 414 bacterial strains isolated from cattle hide and sheep skins, 379 were found to be Gram-positive (91.6%). The most primarily involved were species of *Staphylococcus* (47%), *Micrococcus* (21%) and *Corynebacterium* (19%). This was further confirmed in a study wherein species of *Staphylococcus*, *Bacillus* and *Micrococcus* were observed to be largely involved in skin putrefaction (Oruko et al. 2019). However, anaerobic bacteria were considered to be more perilous as they degraded collagen and converted into amino acids, thereby affecting the quality of leather (Kuttalam et al. 2020). If the animal care is improper, the flayed hide/skin may contain ~ 1–2 billion/cm^2 microbes which are retained even after the animal is dead (Pekhtasheva et al. 2012). The microbes are also contributed from sources such as dung, storage area etc., as secondary contaminants. Hence, the window between hide/skin flaying and its preservation remains highly prone to bacterial attack (Shede et al. 2008).

Once the animal is slaughtered and the oxygen delivery and defence mechanism ceases, degradation is accelerated by proteolytic enzymes produced by bacteria, eventually involving the breakdown of proteins followed by carbohydrate and fats (Enquahone et al. 2020). This results in an altered chemical composition leading to structural damage. Temperature above 18 °C facilitates the proteolytic action leading to rapid tissue putrefaction (Colak et al. 2006). Initially, there is no visible change in hide/skin as the aerobic putrefaction starts from the surface and penetrates deep into the layers gradually (Marzo 1995). The keratinous cells present in the upper epidermis were found to lose their integrity and further led to the destruction of the epidermis (Pekhtasheva et al. 2012). The grain region of the hide/skin was eventually damaged, when curing process was delayed for some time (David and Bailey 1996). After this, the subcutaneous region is affected, which is susceptible to microbes due to the presence of loosely arranged fats. The reticular region remains uncontaminated for some time as it contains densely arranged complex collagen (Thiele 1980). Eventually, the contamination spreads to the inter-fascicular region degrading the collagen and elastin fibres resulting in the delamination of the dermis layer causing complete destruction of skin (Pekhtasheva et al. 2012). The rods were found to penetrate the collagen fibres whereas the cocci penetrated into the hair bags. Collagenase enzyme produced by the bacteria disrupts the intact collagen structure (Sarker et al. 2020). The cocci present in the hair bulb produces proteolytic enzymes which leads to tissue damage eventually causing hair slip and is considered as one of the main corroborations of skin putrefaction, which directly infers the preservation technique to be poor (FAO 1995). Histological staining of the skin structure revealed absence of cellular structure and thin epidermis. The follicle structure of the hair was seen to be destroyed and the dermis found to be detached from the epidermal layer. The examination of sub-cutis revealed the presence of bacilli and cocci, thus confirming the association of microbes with skin putrefaction (Mohammed et al. 2016).

Overview on reviews published on the alternative techniques proposed for the preservation of raw hides/skins

In order to overcome the demerits of conventional salt preservation, research is being carried out globally and many optional methods have been proposed time and again. A bird's eye view on these methods has been well summarised in the reviews below and their major advantages/drawbacks have been depicted in Table 1.

A review published way back in 2002 discussed about the alternative preservation techniques researched about two decades back (Kanagaraj and Chandra Babu 2002). Physical methods like sun drying (Roddy and Hermosa 1943), controlled drying (Waters et al. 1997), cooled air or use of ice (Hausam 1939; 1951), freezing (Haines 1981), irradiation (Bailey 1997; Ross 1997), preservation using chemicals like benzaalkonium chloride (Cordon et al. 1963), potassium chloride (Bailey 1995; Bailey and Gosselin 1996), liricure (Russell et al. 1997), boric acid (Barret 1985), silica gel (Kanagaraj et al. 2000; 2001), sodium sulphite alone (Hopkins et al. 1973) or in combination with acetic acid (Bailey and Hopkins 1977) and other chemicals (Money 1974) were discussed. Nevertheless, the authors opined that a combination of eco-friendly chemicals and biocidal curing system could render good preservation efficiency. A technique for preservation of hides using low levels of SO_2 gas was also undertaken (Hopkins 1983). Much later in 2017, another review discussed about the status and development trends on cleaner preservation methods used in curing and soaking processes

Table 1 An overview on various methods proposed for the preservation of raw hides/skins

No	Method	Mechanism/advantage	Drawback	Reference
A. Physical preservation methods				
1	Sun drying	Cheap and eco-friendly	Produce inferior quality leathers, applicable only for countries with warm dry climate	Roddy and Hermosa 1943
2	Controlled drying	Control on moisture content is possible, spoilage due to putrefaction was virtually eliminated	Requires high running cost and installation price	Waters et al. 1997
3	Cooled air treatment or addition of ice	Automated handling, no increase in weight, low investment and running cost	Needs equipment installation, addition of ice increases the weight, applicable only for short duration	Hausam 1939; 1951
4	Refrigeration/freezing	Hides can be safely held at 0 °C for 3 weeks without any additional treatment, whereas at − 10 °C the hides could be preserved for 3 months	High cost & energy inputs required, failure of machinery or interruption in power supply could lead to putrefaction, ice crystals formed during freezing causes rupture of skin	Haines 1981
5	Irradiation	Minimizes energy expenditure and reduces the need for toxic chemicals	Need for expensive equipment's, chances of reduction in tensile strength, need for protection of workers	Bailey 1997; Ross 1997
6	Chilling	A greener method to address the TDS problem	Chances of fibre damage is high, installation of mobile chillers may not be cost-effective for small-scale leather industries	Chandrababu et al. 2012a; 2012b
7	Vacuum	It can store raw hides/skins up to 21 days. It preserves the protein matrix and also arrests the microbial attack	Higher initial cost, loss of preservation once vacuum is opened	Gudro et al. 2014
8	Gamma irradiation	Ensures preservation of hides/skins at higher level, leading to production of quality leather, reduction in 40% of TDS value and 78% of chloride concentration	Higher cost, handling hide after irradiation to avoid recontamination can be troublesome, implantation of instrument in all tannery units is not possible	Bailey 1999; Bailey et al. 2001; Gaidau et al. 2021
9	Electric current	Kills almost all the halophilic microbes within 15 min at 0.5A of electric current, high efficiency and ease of application	Higher initial cost, not practical for the long-term preservation of hides and skins	Birbir et al. 2008
B. Less sodium chloride methods				
1	Antiseptic + brine solution	Reduced the usage of salt by 50%	Higher cost, additionally toxicity of the chemicals might create environmental problem	Sarker et al. 2020
2	NaCl + Benzalkonium chloride (BAC)	No adverse effect on preserved skin	The bi-products produced by using BAC may have environmental impacts	Cordon et al. 1963
3	NaCl + EDTA + chlorinated phenol + sawdust (pine)	EDTA inhibits bacterial growth and proliferation by inactivating enzyme active sites, reduction in NaCl by 20%	EDTA may cause difficulties in precipitating chromium in effluent treatment and also possess health hazards	Russell et al. 1997

Table 1 (continued)

No	Method	Mechanism/advantage	Drawback	Reference
4	NaCl + boric acid (BA)	Inhibition due to the bactericidal or bacteriostatic effect of BA, does not pose any serious health issue, is stable to heat, light, air and is not volatile	It comes under effluent restriction in Australia for curing process	Barret 1985, Kanagaraj et al. 2005a
5	NaCl + silica gel	Silica acts as dehydrating agent thus inhibiting bacterial growth. Reduction in TDS by 70–75%	May cause some health issues	Kanagaraj et al. 2000; 2001
6	NaCl + sodium meta-bisulphite (SMBS)	The antiseptic property of SMBS and the gradual decrease in the moisture content preserves the skin	Inhalation of SMBS during curing process may cause asthma like allergy, has harmful health effects	Kanagaraj et al. 2005a
7	NaCl + Polyethylene glycol (PEG) + crude glycerol	The PEG polymer acts as dehydrating agent and glycerol acts as humectant making the skin softer, no degradation in collagen fibres of crust leather	The PEG polymer is non-biodegradable	Aldema-Ramos et al. 2015
8	NaCl + Superabsorbent	The superabsorbent acts as a dehydrating agent and allows the salt to penetrate into the collagen structure via inter-fibrillar elements	The polymer is non-biodegradable, which again poses environmental threat	Brosse and Sabatier 2005
C. Sodium chloride–free methods				
1	Potassium chloride	Physical and chemical properties similar to NaCl, prevention of red heat, potassium is a macro nutrient to the plants	Higher cost of KCl, decreased solubility of KCl with drop in temperature	Bailey 1995; Bailey and Gosselin 1996
2	Sodium sulphite and acetic acid	Using acid-sulphites combination on flesh side of hides	Acetic acid has harmful environmental and health effects. Not eco-friendly, results in higher TDS	Hopkins et al. 1973; Bailey and Hopkins 1977
3	Zinc chlorite or calcium hypochlorite	Preservation mechanism exerted by zinc chloride/calcium hypochloride is similar to NaCl	High chloride, TDS release which might pose environmental threat	Money 1974
4	Use of SO_2 gas	Hides and skins are frozen using SO_2 gas, which limits bacterial growth	SO_2 emission can be harmful to human health. Release of SO_2 can damage trees and plants and also causes acid rain	Hopkins 1983
5	Sodium sulphate	50% reduction in weight of NaCl used, preserves the skin for longer duration at 80:20 ratio of Sodium sulphate to NaCl	Health hazards due to SO_2 gas production	Vankar et al. 2006; Vankar and Dwivedi 2009, 2009a
6	Silicate	Significant reduction of TDS and nearly full elimination of salt in soaking liquors, application of silicate-containing soaking liquors for crop irrigation lead to improved plant growth and yield	May pose some health issue	Munz 2007
7	Calcium hydroxide + Polyethylene glycol (PEG)	Preservation is carried out using a water-soluble polymer of ethylene dioxide in combination of a sparingly soluble alkali	Non-natural product and the non-biodegradability property could pose threat to the environment	Rao et al. 2009; Subramanian et al. 2014

Table 1 (continued)

No	Method	Mechanism/advantage	Drawback	Reference
8	Peracetic acid (PAA)	Strong oxidising agent to preserve skins, 11-fold reduction in chloride value	Process pose health hazards due to organic solvents, PAA is considered as weak carcinogen	Valeika et al. 2013; 2016
9	Ozone	Strong oxidising agent which can be used as sterilant for short-term preservation of skins	Ozone is considered as toxic	Sivakumar et al. 2010
10	Super critical carbon dioxide	Reduction in chloride content by 90%	Implantation of reactor units for small-scale industries is difficult	Endlweber 1991; Gopinath et al. 2020
11	Emulsion of IPBC, TCMTB, polyoxyethylene triglyceride, glycol ether, polyalkylene, xanthan gum and dipropylene glycol	Combination of IPBC & TCMTB acts as stable compound and provides strong resistance against fungal contamination, prevents grain damage caused due to fungi	It does not possess antibacterial activity	Bonjour et al. 1999; 2000
12	Phenolic and Azole compounds (3,5-dimethyl-4-chlorophenol + methyl benzimidazolyl-carbamate (MBC))	Prevents the damage to hides by providing resistance against microbial infection by suppressing their activity, resulting in longer shell life	Development of azole resistance to fungi should be considered	Rother et al. 2000

of leather industry (Wu et al. 2017). In addition to the methods discussed above, the authors have considered in detail about the usage of less NaCl in combination with EDTA (Russell et al. 1997), silica gel (Kanagaraj et al. 2000; 2001), sodium meta-bisulphite (Kanagaraj et al. 2005a), boric acid (Barret 1985; Kanagaraj et al. 2005b) and 1% of sodium hexafluorosilicate (Valeika et al. 2017). On the other hand, the NaCl-free methods included the use of inorganic chemicals viz., potassium chloride (Bailey 1995; Bailey and Gosselin 1996), sodium sulphate (Vankar et al. 2006; Vankar and Dwivedi 2009, 2009a) and silicate (Munz 2007), synthetic preservatives viz., polyethylene glycol (Rao et al. 2009; Subramanian et al. 2014; Aldema-Ramos et al. 2015), various acids (Valeika et al. 2013; 2016) and other chemical antiseptic materials like merpin TKE, Nercolan GLO and Vantocil CL (Kanagaraj and Chandra Babu, 2002). Some of the authors also touched upon the use of natural materials as preservatives and other physical methods like chilling (Chandrababu et al. 2012a; 2012b), vacuum (Gudro et al. 2014), irradiation (Bailey 1999; Bailey et al. 2001) and electric current (Birbir et al. 2008). These are the methods where both proprietary products and standard chemicals were used in place of salt to preserve the raw hides/skins. Most of these research revolved around the usage of less NaCl in combination with other chemicals as well as sodium chloride-free methods involving the use of organic and inorganic preservatives like silicate, ozone (Sivakumar et al. 2010), salts of different metals viz., zinc, sodium, potassium, nickel and mercury. However, these are yet again problematic from environmental and health perspective, since majority of these come under the hazardous chemical category and are classified as toxic, harmful or irritant, corrosive and dangerous for the environment. Additionally, the expense involved in their usage is quite high due to their bulk requirement that too in pure form. Another disadvantage is the difficulty in the handling of the chemicals by the personnel involved in the curing process. Synthetic preservatives were also investigated for their antibacterial effect and used in preservation or soaking process. Nevertheless, these are either economically inconceivable or has other environmental impacts (Shede et al. 2017). A recent review highlighted the limitations and drawbacks associated with various salt free/less salt methods for preservation of hides/skins (Sivakumar et al. 2019). The authors concluded that the use of natural eco-benign materials with antimicrobial property could be a viable option for the short-term preservation of hides/skins as novel approach for sustainable development. The use of solvent (Buechler et al. 1987), CO_2 gas (Endlweber 1991), aqueous fungicidal emulsion of 2-(thiocyanomethylthio) benzothiazole and 3-iodo-2 propynyl-n-butylcarbamate (Bonjour et al. 1999; 2000), mixture of phenolic compound

and azole/morpholine compound (Rother et al. 2000), superabsorbent polymers (Brosse and Sabatier 2005) and stepwise drying of skins under vacuum in the temperature range of 30–45 °C (Rai et al. 2009) have been highlighted. Some recent research involving the use of alkali and water-soluble polymer of ethylene oxide (Sundar and Muralidharan 2019), supercritical CO_2 (Gopinath et al. 2020) and gamma irradiation (Gaidau et al. 2021), are also available. Yet another recently published review discussed about the trends and advancements in sustainable leather processing and future directions and challenges (Kanagaraj et al. 2020). Most of these research where different chemicals were used have efficiently preserved the skins. However, due to their chemical nature, the environmental parameters needs be taken into consideration since they are either hazardous themselves or practically not adaptable or are expensive. Under such a scenario, researchers are now changing their focus from chemical preservation to bio-based preservation.

Changing trends of research from chemical to bio-based preservation

In the past two decades, natural, bio-based preservation has emerged as prospective alternative to conventional NaCl, as they are eco-friendly and non-harmful. Research has been focused worldwide on developing preservation methods using various bio-based products. Since quite a number of researches have been published on other preservation techniques, as discussed in the previous section, the current review has been written with an emphasis on summarising the developments in bio-based preservation techniques during the past two decades. As shown in

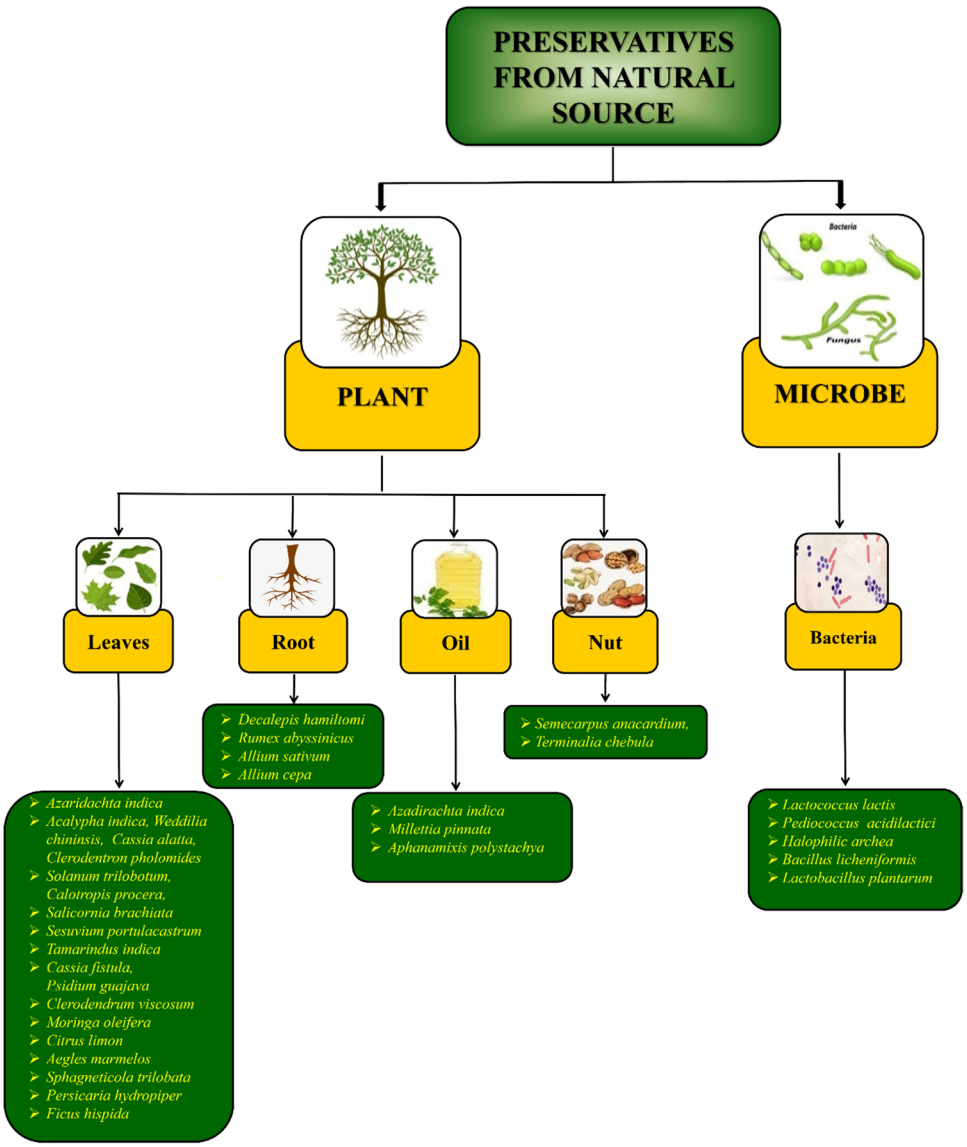

Fig. 2 Bio-based formulations exploited for preservation of raw hides/skins

Fig. 2, the phyto-based formulations involve the use of different plant parts like paste/powder/extracts of leaves, root extracts and essential oils. On the other hand, the use of antibiotics produced by bacteria has also been envisaged as a potential substitute to conventional curing in a few studies. These researches have been discussed in details in the following section:

Phyto-based preservation

It is a well-known fact that the phytochemicals are non-nutritive plant chemicals that have protective properties. Plants produce these chemicals to protect itself, but research demonstrates that many phytochemicals can protect humans against diseases. The various plants discussed in the subsequent section used for the preservation and the

Table 2 Plants used for preservation and their phyto-constituents anticipated being responsible for imparting antimicrobial activity

Plant	Phyto-constituents detected	Optimum Concentration	Reference
Curing			
Acalypha indica	Eodin, loliolide, acid, gallic acid, rutin, nicotinic, b-sitosterol	15% plant formulation	Vijayalakshmi et al. 2009
Weddilia chininsis, Solanum trilobotum, Cassia alatta, Calotropis procera, Clerodentron pholomides	–	5 mL plant extract + 3% salt	Sivabalan and Jayanthi 2009
Sesuvium portulacastrum	trans-4-hydroxyprolinebetaine, proline and 3,5,4′-trihydroxy6,7-dimethoxy-flavone 3-glucoside	10% leaf powder (dry weight)	Kanth et al. 2009
Salicornia brachiata	2-hydroxy-6,7-methylenedioxyisoflovane, -2-hydroxy- 6,7-methylene dioxy isoflavanone, 2,7-dihydroxy 6-methoxy isoflovane	10% leaf (dry weight)	Kannan et al. 2010
Semecarpus anacardium	Phenolics, flavonoids, terpenes, saponins, glycosides, alkaloids	1% nut extract oil + 10% acetone	Kuttalam et al. 2013
Azadirachta indica	Nimbin, nimbidin, azadirachtin B, salannin	De-oiled neem cake	Vedaraman et al. 2016
Rumex abyssinicus	Anthraquinones, flavonoids, triterpenoids	5% root powder + 15% salt	Mohammed et al. 2016
Cassia fistula and *Psidium guajava*	7-Bioflavonoids, 9-epiafzelechin, procyanidin β-2 epiafzelechin, fistulic acid, rhein glucoside, rhein, sennoside A & B, flavonoid, proanthocyanidin	15% leaf paste + 15% salt	Vinodhkumar et al. 2016
Moringa oleifera	–	10% leaf paste + 10% salt	Hashem et al. 2018
Clerodendrum viscosum	Clerodin (α-sterol), tannin, oleic, stearic acid, lignoceric acids, glucuronide, gallic acid, phenolics, alkaloids, flavonoid, saponin, anthraquionone	10% leaf paste + 10% salt	Minhaz Uddin et al. 2019
Citrus limon	Cyclopropanecarboxylic acid, 1,2-benzenediol, 3-[[[3,5-dichlorophenyl] imino] methyl], pentadecanoic acid	10% leaf paste + 8% salt	Alagamuthu et al. 2020
Aphanamixis polystachya	–	15% essential oil	Nur-A-Tomal et al. 2021
Dried neem leaf powder (DNL)	Nimbolide, dehydro salannol, desacetyl nimbinene	15% DNL powder + 15% salt	Velappan et al. 2022
Sphagneticola trilobata	–	20% of leaf paste	Hashem et al. 2021
Persicaria hydropiper	Phytol, 9,12-octadecadienoic acid, phthalate (diethyl), di-n-octyl phthalate, limonene, n-hexadecanoic acid, 9-octadecenamide	10% plant paste + 8% salt	Hashem et al. 2021a
Allium cepa and *Allium sativum*	Flavonoid, alkaloid, glycosides, saponin, steroids, phenols, terpenoids	6% peel powder + 10% salt	Alagamuthu et al. 2022

phytochemicals responsible for imparting antimicrobial activities is depicted in Table 2.

Plant leaves paste/powder

Azardirachta indica

Preethi et al. (2006) investigated on the efficacy of herbal-based formulation prepared using powder of *A. indica* (common name: Neem) leaves with isopropyl alcohol (or cucumber extract) to form a paste for the preservation of raw goat skins. The formulation was applied on the flesh side of goat skins and skin preserved with salt (40%) served as control. These skins were piled and incubated in a place with 40–75% humidity at 26–32 °C and monitored periodically. After ~15 days, the skins were processed into leathers after scrapping off the adhering material. No sign of putrefaction was found for all the tested skins in the laboratory as well as in field experiments. Pollution parameters such as total solids, TDS, COD and chloride in the soaking effluent exhibited a dramatic reduction in the experimental batch due to the absence of NaCl in curing. Further, the effluent devoid of NaCl could be easily recycled after suitable treatment thus saving considerable water and achieving nearly zero solid waste discharge. Additional advantage would be to convert the scrapped off material into organic compost with high nutritive value.

Yet another study reported the possibility of using the leaves extract from the same plant for the short-term preservation of goat skin (Ahmed et al. 2015). The authors have proposed various chemicals in combination with salt. Nevertheless, we discuss about the trials with only Neem leaves paste in combination with 10% KCl, wherein 40% NaCl served as control. Results revealed no discolouration of the skin, hair slip or bad odour until 30 days period. This span of time was considered to be sufficient enough for the handling and transfer of the preserved goat skins. The used *A. indica* leaves paste with KCl could be collected from the preserved goat skins before soaking and could be used as manure.

In a recent study, an eco-friendly approach using dried Neem leaves (DNL) as a phyto-preservative to reduce the environmental pollutants was carried out (Velappan et al. 2022). The antimicrobial activity of the DNL was tested against *Bacillus* sp., *E. coli* and *Salmonella* sp., which were previously isolated from putrefied goat skin. The acetone extract was found to show higher activity with zones of 8 mm, 10 mm and 16.9 mm against previously mentioned bacteria, respectively. The phytochemical compound nimbolide present in the Neem leaves was reported to impart the antimicrobial activity. Higher concentration of DNL viz., 15%, 20% and 25% in combination with 15% salt was found capable to preserve skins for 15 days without any putrefaction. The phytochemicals viz., dehydro salannol, desacetyl nimbinene and nimbolide were found to be the key components responsible for preventing bacterial growth. Furthermore, the hydrothermal and physical properties of DNL preserved leather were almost similar as control leather. The scanning electron microscope (SEM) results revealed uniformity in grain structure and compact fibres were observed in the dermal layer of both control and experimental leathers which indicates no structural damage in grain surface and tissue fibre. Hence, this study once again illustrated the efficacy of Neem leaves with less salt as an eco-friendly and viable preservation method for skins.

Acalypha indica

Another study on short-term preservation of hides/skins using fresh *A. indica* leaves paste (15%) in combination with 10% and 20% KCl and monitoring the variations in microbial population dynamics and various biochemical parameters was reported (Vijayalakshmi et al. 2009). Goat skin and cow hide samples (5 × 5 cm) were incubated at different temperatures for a period of 2 weeks and relative humidity of 85%. Additionally, isolation and screening of microbes for their protease, collagenase and keratinase activity from the raw hide/skin was undertaken. The rate of collagen degradation was monitored by hydroxyproline (HP) release and the non-collagen protein degradation using tyrosine release. The collagen degradation progressed slowly with a maximum HP release of 4.7 mg on the 10^{th} day, which was attributed to the production of hydrolytic enzymes by the skin putrefying bacteria, whereas the non-collagenous protein degradation reached a maximum of 0.88 mg on 8^{th} day followed by a reduction pattern. This decrease may be due to the utilisation of free amino acids by bacteria. This could also be correlated to an increase in bacterial population from 2×10^7 to 4×10^9. The rate of skin degradation was directly proportional to the increase in temperature, which might be due to activation of extracellular bacterial enzymes. Almost all the strains isolated from putrefied goat skin exhibited keratinolytic, collagenolytic or proteolytic activity and were predominantly Gram-positive rods or cocci. The putrefied cow hides yielded proteolytic strains viz., *Klebsiella*, *Proteus vulgaris*, *Stromatococcus*, *B. fusiformis*, *B. subtilis*, *B. sphaericus*, *Micrococcus halobilus*, *M. nishinomiyaensis* and *Moraxella nonliquefaciens*. Thus, this method using *A. indica* leaves with KCl effectively preserved the goat skins for a period of ~ 70 days and was found to be viable, cost-effective and eco-friendly.

Weddilia chininsis, Cassia alatta, Clerodentron pholomides, Solanum trilobotum and Calotropis procera

The possibility of using extract of medicinal plants in combination with less amount of salt for preservation was

attempted (Sivabalan and Jayanthi 2009). Different bacteria and fungi isolated from skin sample were further identified. Subsequently, twelve medicinally important plants were selected for their antimicrobial activity and evaluated against the isolated skin putrefying bacteria and fungi. All the extracts exhibited no antifungal activity; however, good antibacterial activity was observed. Chloroform extract exhibited maximum antibacterial activity against *Staphylococcus aureus*, *S. epidermidis* and *Pseudomonas aeruginosa*, exhibiting a zone of inhibition of 40, 20 and 22 mm respectively. Based on the results, among the twelve, only five plants viz. *W. chininsis*, *S. trilobotum*, *C. alatta*, *C. procera* and *C. pholomides* were selected for further studies.

For preservation experiment, extracts of leaves of these five plants was prepared, filtered and sterilised for 15 min. Freshly flayed goat skins were obtained and the control skin was applied with 30% salt. For less-salt preservation approach, 5 mL plant extract combined with 3% salt was directly applied on skin and stored up to 15 days. Among the five plant extracts assayed for preservation, *C. procera* was found to be more effective in bringing the moisture content of the skin to about 29.6% followed by *C. alatta* 29.07%, *W. chininisis* 28.82% and *C. pholmoides* 27.78%. It was also observed that *S. trilobotum*, *C. alatta* and *C. procera* showed no putrefaction, odour or hair loosening. For all the trials, the skins were found to be well preserved which could be attributed to the antimicrobial property of the herbal extracts against the microorganisms.

Salicornia brachiata

S. brachiata is a halophyte belonging to the Chenopodiaceae family and is distributed worldwide. Investigation on the possibility of using *S. brachiata* grown in tannery wastewater treated lands to preserve sheep skins was undertaken (Kannan et al. 2009; 2010). It is reported that around 364 µmol/g of Na^+ and 310 µmol/g of Cl^- is present in the plant; since *S. brachiata* when grown under alkaline condition absorbs all the salts and hoards it in its vacuoles. For the present study, various concentrations of *S. brachiata* powder 5, 10, 15, 20 and 40% (on the weight of the skins) were applied on the flesh side of the right halves of sheep skins. The corresponding left halves were treated with 40% NaCl. The skins were piled and stored at 40 °C and periodically monitored. The skin preserved using *S. brachiata* at 10% dry weight basis resulted in effective preservation. The extractable nitrogen content was found to be 3.42 mg/g and the bacterial count was also low as compared to the control sample which might be attributed to the synergistic action of salt and essential oils present in the plant. The phytochemicals present were also believed to exert some toxic effect against microbes by disrupting the cell membrane integrity. An increase in shrinkage temperature by 6 °C as compared to the conventionally preserved skin was also noted. The polyphenols present in the plant were believed to cause slight stabilisation of collagen coupled with decrease in moisture content thus leading to an increase in shrinkage temperature. No notable difference in the colour intensity value (ΔC) and the darkness value (ΔL) among the experimental and control leathers ruled out the possibility of the constituents imparting colour to the experimental leather. Additionally, this preservation system also added some extra attributes to the leather viz. slightly higher softness value as compared to control crust, no major difference in organoleptic properties and no structural modification in the collagen network. Hence, the proposed method for preservation of sheep skins using *S. brachiata* was considered as a cleaner approach to eliminate the environmental constraints caused by conventional salt curing system.

Sesuvium portulacastrum

S. portulacastrum, yet another perennial halophyte, was investigated for the possibility of curing of raw goat skins (Kanth et al. 2009). This plant is known for its antifungal, antibacterial and antioxidant property due to the presence of wide range of essential oils (Filipowicz et al. 2003). The dry weight of the plant leaf contains 18% Na^+ and 15% Cl^- (Joshi 1981). The major phyto-constituents were reported to be proline, trans-4hydroxyprolinebetaine,3,5,4′-trihydroxy6,7-dimethoxyflavone 3-glucoside. The compounds were found to play a crucial role in osmo-regulation. Skins preserved using 10% of phyto preserve showed no putrefaction, hair slip or odour at the end of 48 h of preservation. The moisture content of phyto preserve was found to be 28% at the end of 30 days. The bacterial count for salt preserved sample on days 1, 14 and 30 was 9×10^{10}/g, 5×10^{7}/g and 4×10^{6}/g respectively. However, for skins cured with 10% phyto preserve on the same days showed much reduced bacterial count. A marginal difference in shrinkage temperature of control (66 °C) and experimental samples (68 °C) indicates no structural modifications of the skin matrix. The organoleptic and physical properties of the processed leather were almost similar to salt-cured skin. Moreover, preservation by this method does not pose any safety problem or health hazard to the environment, which makes it a highly approachable preservation technique.

Tamarindus indica

T. indica is an economically important tree yet less utilised and less known. It is famous as 'crop for the future' the extract of which has promising antibacterial, fungicidal, antiviral, anti-nematodal, antidiabetic and antioxidant properties. With an attempt to reduce the pollution load, preservation using *T. indica* was explored (Alagamuthu

et al. 2015). Methanolic extract of *T. indica* leaves powder was prepared and used for minimal inhibitory concentration (MIC) determination against different collagenolytic bacteria viz., *Escherichia coli, Bacillus* sp., *Klebsiella* sp., *Pseudomonas* sp., *Proteus* sp., *Streptococcus epidermis*, *S. aureus* and *Salmonella* sp. The inhibition for all the above was 100% except *S. aureus* and *Bacillus* sp., which was 94.62% and 98.80% respectively. The presence of various groups like amine, alcohol, carboxylic acid etc., in the extract as revealed by Fourier Transform Infrared (FTIR) spectroscopy were supposed to be salient for preservation. The amino and carboxylic acid groups were reported to form weak hydrogen bonds (reversible) with skin collagen.

For preservation, various concentrations of extract and salt were applied based on the green weight of skins. Sensory evaluation like hair slip, smell and putrefaction was carried out until 21 days of preservation, which showed satisfactory results. The overall results showed that better preservation was achieved when 10–15% salt with 15% *T. indica* leaves extract was used. Additionally, the skins treated with *T. indica* showed equivalent or comparable strength property like control sample, thus making it evident that *T. indica* did not have any disagreeable impact on the collagen matrix. Based on the results, it was inferred that *T. indica* leaves extract has an enormous potential to emerge as viable alternative to conventional salt-based preservation.

Cassia fistula and *Psidium guajava*

Vinodhkumar and co-workers attempted to reduce/replace NaCl used for preservation by substituting with plant extracts of *C. fistula* and *P. guajava* (Vinodhkumar et al. 2016). These plants are already reported to exhibit brilliant antimicrobial properties. About 44 compounds have been identified through Gas Chromatography–Mass Spectrometry (GC/MS) analysis from *C. fistula* (Tzakou et al. 2007). The phytochemicals present in its leaves were found to be anthraquinones like fistulic acid, rhein glucoside, rhein sennoside A & B, 3-O-B-D-glucopyranoside, chrysophanol, physcion, 7-bioflavonoids, 9-epiafzelechin and two triflavonoids with procyanidin B-2 and epiafzelechin (Luximon-Ramma et al. 2002; Gupta et al. 2008). On the other hand, *P. guajava* have also demonstrated properties like antioxidant, antimicrobial, antiallergic, antidiabetic, antirheumatic and antigenotoxic by numerous pharmacological studies (Gutiérrez et al. 2008).

In this study, leaves extracts of *C. fistula* and *P. guajava* were evaluated for their antimicrobial activity against collagenolytic bacteria by agar well diffusion assay. Good activity was exhibited by ethanolic extracts of *C. fistula* and *P. guajava* against *Bacillus* and *Klebsiella* sp. For preservation experiments, these plant leaves extract with different combinations of salt were applied on skin for 21 days. The release of HP was found to be low in all the samples (below 500 µg/g of skin) on the 21st day, indicative of a good curing efficacy. Nevertheless, the presence of soluble protein in soak liquor indicated the degradation of skin proteins. *C. fistula*–cured skins showed tremendous decrease in soluble protein content by the 21st day of preservation, which was better than *P. guava*–cured skins.

The physical strength properties of the experimental skins showed comparable results with the control. Except for the neck portions of the leathers, tensile strength and tear load values of the experimental leathers were appropriate for upper leather. However, grain cracking property was observed to be comparable in all experimental leathers to the standards advised for upper leathers. Organoleptic evaluation proved that *C. fistula* and *P. guajava* leaves can be recommended for skin preservation. Especially 15% leaves paste each of *C. fistula* and *P. guajava* in combination with equal amount of salt (based on green weight) proved to be very effective for preservation.

Clerodendrum viscosum

C. viscosum is widely distributed in tropical and subtropical regions of the world particularly in India. Extracts from different parts of the plant have been widely used in Ayurveda and Unani for treating different illness (Nandi and Lyndem, 2016). Additionally, *C. viscosum* has also exhibited considerable inhibition against many skin putrefying bacteria e.g. *E. coli*, *S. aureus*, *Pseudomonous aureogenosa*, *Salmonella typhi* and *B. subtilis* (Oly 2011). Hashem et al. (2017) have reported a method for preservation of goat skin using *C. viscosum* leaves-based formulations in combination with salt. The goat skins preserved using only leaves paste and 10% leaf paste with 5% salt was hard and difficult to process in drum/paddle. Hence, experiments were carried out using 10% leaf paste + 10% NaCl. The moisture content for goat skins preserved using 10% leaf paste in combination with 10% and 15% salt was found to be 41.4% and 50.3% respectively. It was seen that the moisture content was increased when NaCl concentration was increased from 5% to 10% due to salt's hygroscopic nature. Until day 16, the bacterial count was higher in experimental skin than control skin but no hair slip, putrefaction or odour was observed. Although at the end of 30 days, bacterial count lowered in the experimental sample due to its biocidal effect. SEM analysis for all the processed leathers revealed no deterioration. The authors opined that application of 10% each of leaves paste and NaCl could preserve the goat skin for a period of 30 days and the physical properties of the produced leather fulfilled the requirements of shoe upper.

Preservation of goat skins using the same plant has also been explored by another group (Minhaz Uddin et al. 2019). Skins preserved using only leaves paste showed bacterial growth on the surface at the end of 30 days. However, the

plant-based treatment lowered the bacterial count by inhibiting bacterial actions like enzyme secretion and cellular metabolism until the 15th day of preservation. Although the bacterial count was high, there was no sign of putrefaction for leaves powder and leaves paste preserved skins. However, for skins treated without salt, especially leaves powder was found to be very hard and difficult to process. The more the salt, the more the moisture content was retained in the skin. This was due to salt's ability to absorb moisture from the environment. After 30 days, the moisture content lowered to 20% rendering unfavourable condition for proteolytic microorganisms and the bacterial count was found to be 4×10^6 CFU/mL. Goat skins treated using leaves paste exhibited shrinkage temperature between 60 and 70 ℃ indicating that no denaturation had occurred. Nevertheless, where treatment was given using leaves powder, a shrinkage temperature above 70 ℃ was observed which might be attributed to some tanning action in these skins. *C. viscosum* leaves paste with or without salt showed appreciable results by lowering the pollution load in soak liquor. Hence, this treatment method was reported to provide sufficient antimicrobial effect even without salt in tough and humid conditions (Minhaz Uddin et al. 2022).

Moringa oleifera

In another work by Hashem et al. (2018), an alternative curing system using *M. oleifera* leaves paste with less salt was applied on the flesh side of the goat skin for preservation and observed for 28 days. The effectiveness of preservation was monitored regularly and compared to conventional salt curing. It was observed that all the combinations were found to preserve the skin. However, the skins preserved using only leaves paste and leaves paste with less salt were hard and hence difficult to process in paddle or drum. Eventually, skins preserved with 10% leaf paste + 10% salt were found to be optimum for preservation, and therefore pilot-scale studies were carried out using this combination. Shrinkage temperature is considered to be a significant property, as it relies on the hydrothermal stability of the collagen and is indirectly dependent on the structural orientation of skin proteins. In the present study, the shrinkage temperature of the preserved skins met the standard value as for control. In terms of pollution load, phyto-preserved skins using 10% leaf paste + 10% salt reduced all the parameters viz., BOD, COD, TDS and chlorides. The results of pilot-scale study showed that the moisture content was almost constant from 14–28 days and did not show any degradation, odour or hair slip. The crust leather from both the experimental and control goat skins subjected to SEM analysis revealed no degradation in fibre structure and the texture and quality of the skin was intact. Hence, this low salt preservation method using *M. oleifera* could be used as a viable option for goat skin preservation.

Citrus limon

The rich antibacterial, antioxidant and antifungal property of *C. limon* (common name: Lemon) leaves as a substitute for preservation of goat skin was investigated (Alagamuthu et al. 2020). Various extracts (water, methanol and chloroform) of Lemon leaves were screened for phytochemicals, and revealed the presence of terpenoids, steroids, sterols, saponins, quinones, phenols, flavonoids, phlobatannins, cardiac glycosides, tannins and starch. The GC–MS analysis of methanol extract confirmed the presence of 1,2-benzenediol,3-[[[3,5-dichlorophenyl] imino] methyl], cyclopropanecarboxylic acid and pentadecanoic acid, which are reported to exert negative effect on Gram-negative bacteria (Agoramoorthy et al. 2007). Additionally Also, heptadecanoic acid as a formidable antifungal and antibacterial agent has also been reported (Al-Shammari et al. 2012). The presence of these antimicrobial compounds in Lemon leaves strengthens the idea of using the extract for the preservation of raw hides/skins. The methanol extract of Lemon leaves showed excellent antioxidant nature at 200 µg/mL concentration. The effectiveness of this extract as a potential preservative was further confirmed by the antibacterial effect of Lemon leaves against collagenolytic bacteria. HP release was found to be more in conventionally preserved skins than the experimental skins. The bacterial count was found higher initially, which then reduced gradually after 8 days of preservation in experimental skins. Estimation of pollution load revealed 75% decrease in TDS value and substantial reduction in other parameters like TS, BOD and COD. The skins treated using Lemon leaves showed comparable tensile strength, grain cracking property and elongation break with respect to salt-cured skins. Hence, preservation using Lemon leaves addresses the limitations caused due to conventional preservation and may turn up as a game changing technique. Additionally, the usage of Lemon leaves as preservative imparts good odour during leather processing.

Aegles marmelos

Preservation using ethanolic extract of *A. marmelos* which is totally devoid of salt has also been attempted (Kuttalam et al. 2020). Optimisation studies showed that 4% ethanolic extract was found to preserve skins for a period of 20 days whereas 5% was found to be effective for 30 days as evidenced by no hair slip or odour. The moisture content of the experimental skins did not show much difference on day one (5.7%), where as in the case of control skins, there was a drastic reduction from 74% to 65%. Although after 7 days, it was almost similar for both the experimental (45%)

and control (40%) skin samples. Higher osmotic pressure bestowed by salt is found to be the driving force for causing the water to migrate outside the skin. Reduction in moisture content was found to be directly proportional to the bacterial population. On day seven, the bacterial count was reduced to 6×10^8 CFU/mL in *A. marmelos*–treated skin and 6×10^{10} CFU/mL in salt-treated skins. The main advantage of adapting this method of preservation was the complete elimination of salt and the quality of leather processed from phyto-preserved skins was equally satisfactory as the leather processed from salt preserved skins.

Sphagneticola trilobata

S. trilobata plant, locally known as 'bhringraj', is grown as an ornamental herb that grows rapidly into the surroundings. In this study, *S. trilobata* leaves paste was applied to preserve the goat skin without any NaCl (Hashem et al. 2021). The leaves paste was offered at 10, 15, 20, 25 and 30%, based on skin weight. The comparison and assessment of the experimental proposed solution with the conventional wet salting method revealed no significant difference in case of moisture content, hydrothermal stability and bacterial count. The extractable nitrogen was also consistent with moisture content. On the 14th day, both moisture content and nitrogen content reached an equilibrium point. On the 28th days, the extractable nitrogen content for the experimental and control sample was 1.7 and 1.9 g/kg, respectively. The physical properties of the experimental leather fulfilled the requirement of shoe upper and its SEM image exhibited properly arranged bundle arrays as in control sample which helped absorb the dye properly during further processing of the skin, giving a well lustrous grain to leather. Moreover, no deterioration was observed in the fibre structure of the goat skin. This 'green' preservation method reduced the Cl^-, TDS, BOD and COD in soaking operation. Thus, the recommended preservation method could be a sustainable option to preserve goat skin, which would enormously reduce the pollution load, especially in soaking operations.

Persicaria hydropiper

The *P. hydropiper* plant leaves were studied for the short-term preservation of animal skins (Hashem et al. 2022). The methanol extract of this plant leaves subjected to GC–MS analysis revealed the presence of antimicrobial compounds viz., phytol, which has antioxidant and antimicrobial properties, followed by 9,12-octadecadienoic acid and n-hexadecanoic acid which presents unfavourable condition for Gram-negative bacteria (Agoramoorthy et al. 2007). The di-n-octyl phthalate and phthalate (diethyl) demonstrated antibacterial and antifungal activity against *Aspergillus flavus*, *S. aureus*, *B. subtilis* and *E. coli* (Ortiz and Sansinenea 2018). The phytochemicals present in the leaves render them suitable for raw hides/skins preservation.

The concentration of leaves paste was optimised by both physical hands feel and visual observation of the skins. The skins preserved using 10% leaf paste was found very hard to process, on the other hand skins preserved using 10% leaf paste with 12% salt was found very flexible and soft which escalates the possibility of getting damaged in drum during leather processing. 10% plant leaf paste + 8% salt was taken as optimised concentration as the skins preserved using this combination was found flexible and exhibited similar trend as the control. The efficacy of the preservation was accessed using bacterial count, shrinkage temperature, moisture content etc. The skins were preserved successfully up to 30 days and the leather processed showed physical-organoleptic characteristics which met the standard. The fibre structure obtained by SEM image for the experimental skins was found almost same as the control. Like other plant preservatives this has successfully reduced the TDS and chloride levels, hence could be used as a convincing alternative preservativeon agent.

Ficus hispida

F. hispida plant is widely distributed throughout tropical and subtropical regions of India, China and Sri Lanka and is known to possess excellent antioxidant and antimicrobial activity. The leaves paste of this plant with a minimal amount of common salt was applied on the flesh side of raw goat skin. Efficacy of the method was evaluated by scrutinising thermal stability, hair slip, odour, moisture percentage, TKN and bacterial count in comparison with the conventional wet salting preservation method for 28 days. Analysis of wastewater indicated that the novel plant-based method decreased the Cl^-, TDS, TSS, BOD and COD by 51.02%, 41.6%, 37.1%, 2.9% and 14.6%, respectively (Hashem et al. 2021a).

Plant root

Decalepis hamiltomi

D. hamiltonii is a climbing shrub with aromatic tuberous roots distributed in Southern parts of Peninsular India. Its tuberous roots are widely used as a health drink and are well known for its medicinal properties (Venkatachalam et al. 1981). The very first attempt was made to preserve the raw skins and hides using *D. hamiltonii* way back in 1981. The boiled root extract when applied on flesh side could preserve the hides and skins for 3–5 days, whereas the soaked hides/skins in the decoction were found to remain in good condition for at least 2 weeks. Further, they conclude that in order to make it more effective, aryl alcohol has to be mixed with

inorganic salts and bases which could preserve the hides and skins for a month.

Rumex abyssinicus

Due to the persisting issues associated with conventional preservation method, a method with less salt using the herbal extract of *R. abyssinicus* (common name: Mekmeko) root has been proposed (Mohammed et al. 2016). The roots were shade dried, powdered and used for preservation, the efficacy of which was assessed at different intervals until 30 days. At the end of 30 days, the moisture content was seen to be lowered to 38–41% for experimental skins and 28–30% for salt-cured skins. The HP release of Mekmeko-based less-salt preservation was comparatively lower than corresponding conventional preservation, which were 23.86 mg/g and 33.98 mg/g respectively. Around 7–9 m^3/tonnes of effluent were generated during soaking process. The reduced pollution load and salinity in tannery effluent demonstrated that Mekmeko powder in combination with less salt could be used as good alternative for cleaner leather production. In Ethiopia, around 200 tonnes of wet-salted skins and hides are reported to be processed per day (Calabro 2012). The potential reduction in terms of chlorides and TDS for Memeko-based less salt preservation was calculated to be 13.45 and 17.5 tonnes respectively. Also, the proposed method did not require any additional facility or sophisticated instruments. The Mekmeko plant is available in abundant, and hence the introduction of this new preservation method on an industrial scale could be ventured without any additional cost.

Allium sativum and *Allium cepa*

Recently, in a novel study, the peel of *A. sativum* (garlic) and *A. cepa* (onion) were used for the preservation of raw goat skins. Garlic and onion production occupy a leading position worldwide due to their wide usage in the food industry. Nevertheless, during food processing, the outer scales and roots of garlic and onion bulb are removed, which poses a serious problem especially when it represents loss of valuable source of nutrients and phytochemicals. The extract of garlic peel and onion peel along with reduced salt was found to successfully preserve freshly flayed goat skins for 14 days. The moisture content was found to reduce from 38 to 35%, eventually reducing the bacterial load, which was further validated by very low release of HP for all experimental samples. The pollution load was reduced appreciably especially TDS at the level of 63–83% in the experimental samples. This work offers a dual advantage: (i) peels of garlic and onion which are available in plenty and discarded as waste can be effectively utilised and (ii) excellent antimicrobial property of these peels can be exploited for preservation of raw goat skin (Alagamuthu et al. 2022).

Plant nut extract

Semecarpus anacardium

A different short-term method to preserve hides/skins using *S. anacardium* nut extract was proposed (Kuttalam et al. 2013). The prime focus of the study was to lower the TDS content in tannery effluent by exploiting the dehydration property of acetone and antimicrobial property of *S. anacardium* nut extract. The significant compounds found to be present in *S. anacardium* were bhilwanols, phenolic compounds, sterols, glycosides (Rao et al. 1973), ursuhenol, anacardoside, semecarpetin and gullaflavonone (Murthy 1986).

Around 1% of *S. anacardium* nut extract was found to be sufficient to preserve skins for more than 30 days. A combination of 1% extract along with 10% acetone resulted in no hair slip or odour indicating no sign of putrefaction. However, the volatile nitrogen content was more for 2% essential oil (2.75 g/kg) than 1% (2.20 g/kg). The moisture content was seen to decrease after 7 days (46%) and 53% after 30 days for extract-treated skin sample. Whereas at the same time, the moisture content lowered to 20% and 33% respectively for salt-treated skins. The bacterial count reduced from 6×10^{10} CFU/mL on day 1 to 3×10^{10} CFU/mL on day 30 in case of conventionally cured skin, whereas it lowered from 4×10^{10} CFU/mL (Day 1) to 2×10^{6} CFU/mL (Day 30) in extract-treated skins.

After 30 days of preservation, collagen was extracted and isolated from skins preserved using both the techniques inferring that preservation using plant extract did not degrade or affect the collagen, as there was no significant difference in the banding pattern of both the treatment (Preethi et al. 2006). The phenolic compounds were said to be responsible for the antibacterial activity, which exerts toxic effect on microorganisms by inhibiting the enzymes essential for the growth and metabolism of the microbe. There was no noticeable change in shrinkage temperature of nut extract-cured skins and salt-cured skins, which indicates no detrimental effect on skin matrix by the newly suggested preservation method. Appreciable reduction in pollution load was observed in the soak liquor of *S. anacardium* nut extract-treated skins. This method paves way as eco-friendly substitute to the harmful pollution-causing chemicals used for skins/hides preservation in leather industry.

Terminalia chebula Retz.

A phyto-preservation approach using *T. chebula Retz.* (common name: Myrobalan) nut powder has also been proposed

(Sivakumar et al. 2016). Antimicrobial activity was carried out for 10% salt and Myrobalan nut powder separately using disc and well diffusion assay. It was observed that even low concentration of Myrobalan nut powder produced zone of clearance inhibiting all microorganisms including halophiles. However, in the case of antimicrobial activity of salt, growth of halophiles was observed around the well or salt impregnated discs. The highest zone of clearance was produced when 5% of Myrobalan nut powder was used in combination with 5% salt. The combination of Myrobalan nut powder and salt acted in synergy resulting in better preservation for more than 45 days. The skins preserved using 10% of the mixture did not show any hair slip or other putrefaction effects. The nut extract was found to have various effects on microorganisms like cell wall lysis, cytoplasmic membrane protein damage, cell content leakage, cytoplasm coagulation, cellular process inhibition and energy impairment (Burt 2004; Benli et al. 2008). The leather processed from the Myrobalan mixture preserved skin exhibited comparable result as control leather.

Plant oil

Plant oils are used for various applications like biologically active additives, dietary supplements, drugs, in food industry, aromatherapy and cosmetics (Dadalioğlu and Evrendilek 2004; Edris 2007). The components of these oils are phenol derivatives, oxygen analogues and low molecular weight mono-sesquiterpene hydrocarbons (Adminis et al. 2002). The biochemical processes are reported to be effortlessly terminated as the essential oil penetrates the bacterial cell wall which is facilitated by their molecular size. Depending upon the composition of essential oil, the biological activity may vary. Essential oils that hold replaced phenol groups (eugenol, thymol, carvacrol and guaiacol) manifest strong antioxidant and antimicrobial properties (Lee and Shibamoto 2002; Misharina and Samusenko 2008).

Azadirachta indica

The microbial deterioration of green hides, the key component for leather industry, has sought the attention of many researchers, as the deterioration leads to low quality products, therefore economic loss. To address this problem, studies on efficacy of *A. indica* oil to control green hide deterioration were carried out (Khan et al. 2005). The microflora associated with different skin regions like shoulder, neck, belly, butt and hind shank was estimated. Around nine hides were obtained from slaughtered animal of ~4 years old. Three trials were carried out with different concentrations of Neem oil and NaCl. At the end of 10 days, bacterial count was estimated, and on parallel, the preserved hides were subjected to visual examinations like spot, sliminess, hair slip and colour change. All the trails brought down the bacterial count effectively; however, the trial 2 which included the usage of 10% Neem oil + 20% NaCl exhibited maximum anti-coliform property. The mean value of TVC for this trail was found to be 6.96 ± 0.30, which was the lowest. However, none of the formulated treatment had any effect on Staphylococcal contamination, indicating the non-susceptibility nature against the curing agent. The highest number of Staphylococcal counts was found to prevail in the butt and neck regions (log 7.63 and log 7.49). The efficacy of the treatment was further determined by monitoring the hides at three different time periods viz., 10, 20 and 30 days. A combination of 40% Neem oil + 10% NaCl was found to preserve the skins successfully for 10 days. However, after 20/30 days, negligible amount of hair slip and sliminess was observed in the neck and butt region. Hides preserved using 10% Neem oil + 20% NaCl did not show any colour change or sliminess until 10 days. However, after 30 days, sliminess was fairly noticed throughout neck and butt region. The treatment involving 10% Neem oil + 10% NaCl exhibited noticeable putrefactive symptoms like foul odour and hair slip in the butt region on 30 days of storage time. The strong antibacterial activity of Neem oil was found due to the presence of a compound called Margosic acid. The results have proved the potential of Neem oil with salt as an effective curing agent against the skin contaminating microorganisms. However, it is found to be less effective against halophilic microbes. The problem could be overcome when it is used in combination with a strong antiseptic agent against halophiles.

In another study, the preservation of raw goat skins using de-oiled Neem cake was attempted (Vedaraman et al. 2016). Goat skin is normally preferred for the study, since the structure and grain pattern of goat skin allows detection of even minute changes during the course of preservation. Agar well diffusion assay for antimicrobial studies was done against isolated skin putrefying bacteria and fungi. Almost 100% inhibition was exhibited by methanolic extract against all the bacterial isolates tested. During the preservation period of 21 days, skins were regularly monitored for microbial growth. At the end of this period, the bacterial count was 50×10^3 CFU/mL for skins preserved using 8% de-oiled Neem cake + 15% salt, whereas it was around 66×10^3 CFU/mL for conventionally preserved skin. High-Pressure Liquid Chromatography (HPLC) was used to analyse the bioactive compounds present in the de-oiled Neem cake, and clearly showed the presence of compounds like Azadirachtin B, Nimbin and Salannin. After preservation period, the skins were processed into chrome-tanned leathers and their physical strength properties were tested in comparison with conventionally preserved skins. A considerable reduction in pollution load was achieved in soak liquor of de-oiled Neem cake-preserved skin. The Neem cake is a by-product

of Neem oil industry and is available in large quantities in tropical countries. The ease of application in combination with low salt and antimicrobial properties has envisaged its usage in large scale preservation of raw hides/skins in leather industry.

Millettia pinnata oil cake

M. pinnata (common name-Karanja) is a species of tree in the pea family, Fabaceae, native to eastern and tropical Asia, Australia and Pacific islands. An eco-friendly method using Karanja de-oiled cake without salt or with lower quantities of salt was studied for its antibacterial activity, MIC and minimum bactericidal concentration (Velappan et al. 2020). The reverse phase HPLC analysis showed the presence of Karanjin (0.2%) and Pongamol (0.02%) which were anticipated to be responsible for the antibacterial activity against skin deteriorating bacteria. The preservation trials with Karanja de-oiled cake (15%) along with less salt (15%) showed no hair slip, no putrefaction and reduced bacterial load by ~35% as compared to control skins at 25–35 °C when preserved for a period of 21 days. The preserved skins on re-hydration showed that the soak liquor of experimental skins had a considerable reduction in TDS compared to conventional soak liquor. On processing these skins into leather, the experimental skins were found to possess comparable strength properties (tensile, tear strength) and organoleptic properties as compared to control. This study concluded that conventional salt-based preservation of skins can be replaced by an eco-friendly preservation method using Karanja de-oiled cake along with less salt.

Aphanamixis polystachya

Nur-A-Tomal and co-workers attempted the preservation of goat skin using seed oil of *A. polystachya* plant (Nur-A-Tomal et al. 2021). The *A. polystachya* is a medicinal plant and the oil extracted from its seeds possess antimicrobial activity (Khare 2007). Optimum oil concentration for preservation was found by monitoring the most tangible parameters like hair slip and odour emission. At the end of 30 days, no putrefaction odour or hair slip was found at 15% oil concentration, which indicates no bacterial attack. Thus 15% oil concentration was found to be optimum to preserve goat skins. The total Kjeldahl nitrogen (TKN) content helps to analyse the putrefaction level by evaluating the amount of nitrogenous compound release. The bacteria degrade the intact skin proteins and leads to generation of low molecular weight nitrogen compounds. The TKN value was found to be more (~6 mg/g) for salt preserved skins than oil preserved skins. The oil used for preservation was believed to stabilise the polypeptide link present in the skin (Chandrababu et al. 2012).

At the end of 30 days, the pollution load was reduced drastically as compared to conventional preservation method. The physical strength properties of leather met the standard requirement of elongation at break, distension and load at grain crack and tensile strength. SEM analysis of leather preserved using 15% oil showed finer results compared to salt preservation. There was no destruction in the fibre structure, which further confirmed the effectiveness of oil to be an alternative curing agent. The results imply that the oil induced goat skin preservation proved to be a cleaner alternative preservation method and could be commercially implemented as the oil is inexpensive.

The insight into bio-preservation gives an enormous prospect to plant-based substances with antimicrobial activities. As seen in the sections above, the plant products were reported to preserve the hides/skins but with the aid of chemicals. Hence, different arrays of plant substances with much more potential are needed to be explored. Plenty of plant substances are known to have the bactericidal as well as the bacteriostatic activities. Nevertheless, the basis for plant selection should be their prevalence, abundance, ease of propagation, inexpensive cultivation methods and broad-spectrum antibacterial activities.

Microbes-based preservation

Antibiotics

Antibiotics as preserving agents offer certain advantages over the methods discussed above. These include their biodegradability, efficacy at low concentrations and their relative ease of production by bacterial fermentation. An approach to inhibit the collagenolytic test bacterium *Vibrio alginolyticus* that could degrade the fibrous matrix from hide was used way back in 1990 (Berwick et al. 1990). Microbiological bioassays were performed to determine the antibacterial activity of selected β-lactams, tetracyclines and aminoglycosides against *V. alginolyticus*. Consequently, antibiotics with high activity were selected for their potential application in short-term preservation of green hides. Assay results indicated that the tetracycline type antibiotics were most effective at 1% w/v concentrations. Hence, the interaction of tetracycline HCl with hide powder was investigated, to determine any inactivation of the antibiotic due to drug/collagen binding. Results showed minimal interaction of the antibiotics confirmed by shrinkage temperature measurements of green hide samples treated with antibiotic. Subjective (hair slip, development of mal odour) and objective (determination of extractable nitrogen ratio and volatile nitrogen ratio) measurements of antibiotic-cured skin on laboratory level was performed. Maximum total extractable nitrogen ratio and total volatile nitrogen ratio assessed the

deterioration level of antibiotic-cured green hide. Based on a maximum extractable nitrogen ratio, the hides could be stored for a period of 10 days. These authors have further reported their research for preservation using antibiotics like auromycin and terramycin (Berwick et al. 1996).

In yet another study, a wide range of antibiotics were tested to identify their effectiveness in short-term hide preservation (Stockman et al. 2007). In the initial screening, 14 antibiotic preparations were tested, along with five other chemical preservatives and a control. Raw hide pieces were dipped in solutions of the various preservative treatments, allowed to drain, and incubated at 30 °C. The hide pieces were evaluated for hair slip and odour after 4, 24, 48 and 72 h. Results revealed that combinations of certain widely used clinical antibiotics provide excellent efficacy for the short-term hide preservation and as a result, doxycycline has found its way into the hide preservation market in South America. In spite of their high cost, low-dosage requirements provide a 48-h preservation at a competitive application cost. Nevertheless, the authors opined that as long as specific antibiotics provide demonstrated benefit to humans against pathogens, their use for hide preservation or bacterial control cannot be promoted.

Bio-preservation is a technique for extending the shelf life of food by using natural or controlled microbiota or antimicrobials. The organisms of interest for this purpose are lactic acid bacteria (LAB) and their metabolites. They are capable to exhibit antimicrobial properties and helpful in imparting unique flavour and texture to the food products (Singh 2018). The LAB, generally considered as 'food-grade' organisms, show promise for selection and implementation as protective cultures. Some LAB's exhibit potent antimicrobial activities in the form of small, heat-stable, peptides (Nithya et al. 2012), which are ribosomally synthesised. They are generally effective in inhibiting the growth of similar or closely related bacterial strains (Silva et al. 2018). These are already used in the food industry for the long-term preservation of food materials, since they seem to be effective in inhibiting the growth of food spoilage microbes (Bungenstock et al. 2020).

Nisin and Pediocin AcH from *Lactococcus lactis* and *Pediococcus acidilactici*

In 1995, Hanlin and his co-workers proposed an ecologically acceptable and effective means of preserving cattle hides using Nisin and Pediocin from *L. lactis and P. acidalactici* respectively (Hanlin et al. 1995). A total of 58 isolates were obtained from 23 hides, out of which only 12 were selected for further studies; nevertheless, all 58 isolates were tested for their sensitivity to a mixture of nisin and pediocin AcH by plate assay. Further, the effectiveness of this mixture in reducing cell viability of the 12 isolates over 16 h was determined. It was observed that the cocci grew in their presence, but after 14 h had less OD than the respective control. The sensitive cells were reported to be killed; however, the resistant ones survived and grew. It was concluded that the mixture of nisin and pediocin used was not able to prevent growth of all the isolates over a period of time even at 25 °C. Finally, the results indicated that cattle hides could harbour different types of bacteria initially and the predominant types could change during storage of 2 days at 25 °C prior to curing. Here, the mixture along with 0.25% SDS controlled growth especially of the Gram-positive bacteria and also reduced the growth of Gram-negative rods initially, which later resumed.

Halocin from halophilic archea

Natural proteinaceous antimicrobials such as halocins produced by halophilic archaea may be an effective and pollution free alternative to inhibit the proteolytic halophilic archaea in brine solutions. For this reason, Birbir and coworkers conducted a microbial survey of the salt and brine samples collected from different salt sources in Turkey (Birbir et al. 2004; Birbir and Eryilmaz 2007).

Fifty-six strains of extremely halophilic bacteria were isolated from different salt sources like Tuz Lake, Kayacik saltern, Kaldirim saltern and Turkoy salt mine, since these are the main sources from which the crude salt used for hide/skin preservation was extracted. Twelve of the 18 strains isolated from Kaldirim saltern, 12 of 19 strains from Tuz Lake, 8 out of 12 from Turkoy salt mine and 5 of 7 Kayacik salterns strain were found to exhibit gelatinolytic activity. Many among these strains were found capable of producing halocin, notable was 89% of the isolated strains from Kaldirim saltern produced halocin. Further, the lipolytic activity of these isolated strains and the halocin activity against these isolates by the non-lipolytic halocin producing strains was also conducted (Birbir and Eryilmaz 2007). The results revealed halocin to be effective against most of the isolates. Based on the results, it was found that the halocin produced was active against the lipase and gelatinase-producing *Archaea*. Due to this, the authors recommended the usage of these strains which are neither proteolytic nor lipolytic in nature to prevent the haloarchaeal damage in hides/skins. Combination of halocin from different archaea could be employed to completely inhibit all the strains residing the hide.

Bacteriocin from *Bacillus licheniformis*

In 2010, a patent described about the production and optimisation of a bacteriocin from *B. licheniformis*, isolated from the sediments of slaughter house (Kayalvizhi et al. 2010) which was used for the preservation of raw hides/skins.

The genus *Bacillus* is widely used in many industries for various applications (Paik et al. 1997). The produced bacteriocin was ~1.3–1.5 kDa, and was found to possess strong bactericidal activity of 8800–11,500 U against both Gram-positive and Gram-negative bacteria. Additionally, it was found to exert potential antifungal property. It was found to be active in broad pH range of 3.0–10.0, was thermotolerant (30–100 °C) up to 60 min and was stable in organic solvents at 30–37 °C. The antibacterial activity of the bacteriocin was tested against an indicator strain *Kurthia gibsonnii* GCS6 and determined by the presence of clear zone, expressed in arbitrary units (AU). Powdered bacteriocin at various concentrations (5–20%) was applied to the flesh side of goat skin. Simultaneously, a combination of spray-dried bacteriocin powder and sodium sulphite (both at 5 and 10% concentrations of each) was also applied. The skins were piled up from flesh-to-flesh side and stored at 38–40 °C. The efficacy of preservation was monitored by assessing parameters like moisture content, bacterial count, total extractable nitrogen, shrinkage temperature and other physical properties of the skin sample. Based on the results, it was seen that combination of 10% sodium sulphite and 10% spray-dried bacteriocin was effective for preserving goat skins.

Bacteriocin from *Lactobacillus plantarum*

The use of bacteriocin from *L. plantarum* as an alternative bio-preservative for hide/skin was attempted (Kanagaraj et al. 2014). This strain exhibited maximum bactericidal activity of 470 AU/mL; therefore, it was used for further studies. The bacteriocin produced from *L. plantarum* showed potential activity against the test isolates viz. *Pseudomonas aeruginosa* (200 AU/mL) and *Bacillus putrefaciens* (340 AU/mL). It was observed that when goat skins were treated using 10% bacteriocin, the bacterial count was reduced to $1.0 \pm 0.1 \times 10^1$ CFU/mL. However, there was no growth of microorganism on skin sample treated using 15% and 20% bacteriocin. Skins treated using 15% bacteriocin was found to preserve effectively for 7 days at room temperature. The SEM analysis on microbial cells treated with bacteriocin confirmed the cell wall lysis of skin putrefying microbes. Simultaneously, SEM images of the leather processed from bacteriocin treated skins and control skins revealed no noticeable difference in fibre orientation or grain damage. The experimental leather showed good strength, softness and dyeing property as compared to the control leather. The waste liquor produced also met the standard discharge norms and was safe for human and environment.

Antibiotics as preservation agents offer specific advantages; however, if the equipment's and facilities required for their production are not owned by the tanners, then their use as preservatives may increase the cost of leather manufacturing. Additionally, the possibility that the small-scale tanners would not necessarily afford the setup and running cost of fermentation facilities has to be considered. In such cases, the dependency on the commercially available antibiotics increases.

Points to ponder and way forward

The primary aim of this review was to discuss the enormous work done towards the bio-based preservation of raw hides/skins in order to overcome the problems caused due to conventional salt preservation. As seen from the section above, the bio products are either plant or microbial based, and there are several advantages of using these bio-based preservatives. Most of the plant parts used for preservation are reported to have excellent antimicrobial activity attributed to the presence of phyto-constituents, especially against proteolytic bacteria which are found prominent in skin spoilage. These phyto-constituents are reported not to alter the collagen structure of the skin and produced leather with equivalent strength and quality as the conventionally produced leather. Additional traits like higher softness and stabilisation of collagen as evident from increase in shrinkage temperature are also worth mentioning. Enormous reduction in pollution load especially the TDS, and chlorides, achieved by using bio-based preservatives (Table 3), in turn helps to cut the treatment cost of liquors which in other case would be highly cost intensive and tedious. Few of the plant material are reported to impart fragrance and some of them could be scrapped off from the skin surface and used as manure after the preservation period. From another view point, the phyto-preservation does not require any skill or sophisticated instruments, thus, once again confirming their candidature as potential preservatives. Nevertheless, there are certain research avenues that can be explored like, use of already dried or withered leaves which may save time and energy associated with drying process (unless they are sun dried), also the change in phytochemical composition associated, if any, needs to be looked upon. The availability of the plant globally, and seasonal dependency also needs to be worked out. A recent review has discussed about the development of new preservation method using the tannins from the plant ash or bark powder of *Hagenia abyssinica* as an alternate curing process since the plant extract is already traditionally used for leather softening (Unango et al. 2019). Research towards exploration of new plants/its oils with good antimicrobial properties can be focused upon, however, a few short comings like instability and durability of oils should be taken into consideration. On the other hand, the use of antibiotics for preservation has also been discussed. Several advantages behind their use viz., easy production in bulk amount, no restriction with reference to global availability or seasonal dependency, safe, non-toxic to human health as

Table 3 A comparison on pollution generated as a result of conventional curing versus bio-based preservation

Plant	Pollution parameters										Reference
	Units	**BOD**		**COD**		**TDS**		**TSS**		**Chlorides**		
		Control	**Exp**	**Control**	**Exp**	**Control**	**Exp**	**Control**	**Exp**	**Control**	**Exp**	
Tamarindus indica	kg/tonne	8.8 ± 1.2	6.5 ± 0.5	26.1 ± 0.6	21.3 ± 0.8	270.6 ± 6.8	85.1 ± 1.5	295.0 ± 8.0	30.5 ± 1.0	190.5 ± 4.0	46.3 ± 1.4	Alagamuthu et al. 2015
Rumex abyssinicus	kg/tonne	11.5 ± 1.0	9.8 ± 0.8	24.3 ± 1.0	23.1 ± 1.0	181.2 ± 2.0	81.6 ± 1.4	–	–	109.7 ± 3.6	32.8 ± 0.8	Mohammed et al. 2016
De-oiled neem cake	mg/L	8.7 ± 1.2	7.6 ± 0.9	28.7 ± 0.8	26.1 ± 1.1	50.01 ± 0.81	14.5 ± 0.2	280.0 ± 9.0	127.5 ± 1.5	200.4 ± 0.8	97.8 ± 1.5	Vedaraman et al. 2016
Cassia fistula* & *Psidium guajava	mg/L	–	–	–	–	–	21	–	22	–	288	Vinodhkumar et al. 2016
Clerodendrum viscosum	mg/L	1260 ± 36	1360 ± 11	5250 ± 63	5644 ± 23	42,259 ± 153	21,215 ± 45	–	–	18,223 ± 173	9980 ± 14	Hashem et al. 2017
Moringa oleifera	mg/L	1257 ± 37	673 ± 23	5250 ± 63	2753 ± 27	44,350 ± 48	27,040 ± 53	–	–	18,311 ± 173	9847 ± 13	Hashem et al. 2018
Clerodendrum viscosum	mg/L	–	–	–	–	38,355	9980	–	–	20,846	359	Minhaz Uddin et al. 2019
Aegle marmelos	mg/L	13	7	28	9	270	3	12	24	–	–	Kuttalam et al. 2020
Citrus limon	mg/L	7.9 ± 0.9	7.6 ± 0.6	28.3 ± 1.7	28.8 ± 0.8	290.6 ± 0.8	99.7 ± 0.5	298.5 ± 0.7	35.0 ± 0.5	–	–	Alagamuthu et al. 2020
Dried neem leaf powder	mg/L	4300 ± 15	6105 ± 11	7800 ± 10	8971 ± 11	3350 ± 10	445 ± 12	3660 ± 19	4020 ± 1	40 ± 5	11.5 ± 0.9	Velappan et al. 2022
Sphagneticola trilobata	mg/L	1240 ± 0.01	122 ± 0.03	4480 ± 0.06	650 ± 0.5	4115 ± 0.5	291 ± 0.3	–	–	24,942.3 ± 0.02	488.9 ± 0.03	Hashem et al. 2021
Persicaria hydropiper	mg/L	1194 ± 23	586 ± 31	5647 ± 59	2987 ± 32	45,431 ± 72	21,293 ± 61	–	–	19,303 ± 156	8965 ± 23	Hashem et al. 2021a
Aphanamixis polystachya	g/L	1.3 ± 0.02	1.4 ± 0.03	5.3 ± 0.6	0.7 ± 0.04	42.3 ± 0.5	7.5 ± 0.1	–	–	18.2 ± 0.2	0.3 ± 0.01	Nur-A-Tomal et al. 2021
Allium cepa* and *Allium sativum	g/kg	8.9 ± 0.4	7.2 ± 1 6.8 ± 0.5	30.2 ± 0.5	26.3 ± 0.8 26.1 ± 0.4	272.6 ± 4.8	72.4 ± 1.2 73 ± 1.5	81.0 ± 4.2	22 ± 2 26 ± 1	185.2 + 0.6	46.23 ± 0.2 46.48 ± 0.7	Alagamuthu et al. 2022

well as environment emerge them as potential alternatives. Customised bacteriocin production targeted towards their application on specific skin putrefying bacteria can also be researched. However, certain constrains behind their usage needs to be addressed viz., lowering the production cost, stability in narrow temperature range which might require the tanners to make necessary arrangements for their storage at low temperatures involving capital investment, occurrence of unused antibiotics if introduced in natural habitats may create health problems, emergency of antimicrobial resistance by microorganisms constantly exposed to them etc. However, these can be monitored by using the right choice of antibiotics at apt dosage.

Conclusion

The quest for eco-friendly and economic bio-preservation, which has advantages over other preservation methods, has intensified in leather industry. An in-depth insight into the articles discussed in this review suggest that the curing efficiency of the bio-preservative is mainly dependent on the antimicrobial nature of the compound which inhibits the bacterial growth through various mode of action, thereby preventing the putrefaction of hides/skins. More importantly, no compromise in the quality of leather made from skins preserved using bio-preservatives makes it a competitive alternative to salt curing. Parallelly, the potential of the lesser explored peptides with respect to preservation of hides/skins cannot be denied mainly due to its non-hazardous nature to human or environment. Replacement of synthetic media by natural–cheaper nutrient sources for its production can cut down the cost enormously making it economically viable. Furthermore, the major advantage of using them is they can be combined based upon the predominance/occurrence of skin putrefying bacteria. To conclude, utilisation of natural products in preservation of hides & skins averting the environmental pollution caused by conventional preservation in leather industry unfurls to the evolution of a cleaner curing technology.

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