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Sulphates for skin preservation—A novel approach to reduce tannery effluent salinity hazards

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ABSTRACT

In tanneries microorganisms are able to find environment suitable for their growth. Raw hide of buffalo and other animals like goat that are economically important, are an ideal source of nutrients for bacterial and fungal growth. In the past, preservatives like sodium chloride provided effective protection to fresh hides however the ill effect of their excessive use was not evaluated. But recently concern over potential ecological hazards has become more deliberate and sodium chloride features lot of disadvantages in agriculture as most of the tannery effluent is flown in agricultural fields in India. After rigorous laboratory experimentation on moisture content, SEM of hide, pure sodium sulphate as well as sodium sulphate in addition with sodium chloride (i.e. 10% w/w and 20% w/w) proved as most preferable option for curing of buffalo hide which gives effective preservation. Pollution load studies put forward sodium sulphate as an effective curing agent for buffalo hide to apply at industrial scale also.

1. Introduction

Raw hides are the most valuable by product of meat industry and most of those hides are converted into leather. Skins of cow, goat, sheep and buffalo are found to be most useful and widely available hence utilized in leather making. This is evident that buffalos occupies the important place in Indian rural economy and are major source of agriculture-based products like milk and meat also. After death or slaughtering buffalo skins are preserved and supplied to the leather industries where they are processed into finished leather articles.

1.1. Raw hide deterioration, preservation and hazards of traditional preservatives

Autolysis of raw hides is a spontaneous process responsibly done by the microorganisms. Many practical experiences have indicated that this simplified assumption was not completely accurate. In the last few years the role of halophilic bacteria has become more realistically appraised in the hide curing and storage process [1]. However simple salt NaCl perfectly preserve raw hides at its best. Salt curing acts to preserve hides in double manner. Firstly it combines with moisture for removal of water from the hide and secondly it lowers the water activity of remaining moisture. Salt packs are historically the first method for hide preservation. In this process the first hide is placed on a bed of salt, is then covered with salt and the next hide is placed on top. This process is repeated until the stack is several feet high. It requires about a pound of salt per pound of hide to do a thorough job of curing. The process is much slower and takes as much as 30 days to complete [2,3]. Bacteria cannot live in the presence of certain chemical treatments. There are two classes of chemical treatments for bacteria, bacteristats that limit the growth of microorganisms at whatever stage they are in and bactericides that kill the organisms outright. Both these types of materials are, at best, useful for short-term preservation [4]. NaCl is bacteriostatic compound which have good binding compatibility with collagen fiber of buffalo skin hence dehydrates skin rapidly so the growth of microorganism is inhibited due to lack of appropriate moisture content available in raw hide. Beside these properties of NaCl, there are several disadvantages found in form of increase in salinity. The dry matter yield at maturity of plant tops decreased with increasing salinity of irrigation water [5]. An observation in greenhouse experiment irrigation with sea water undiluted or diluted with an equal volume of fresh water, reduced plant height, grain yield and straw yield of spring barley and oats [6]. It was reported that reduction in growth, yield components, grain yield and 1000 grain weight produced by irrigation with diluted sea water varied among 13 barley cultivars [7]. Hence to replace NaCl from traditional skin curing process, lot of scientific efforts have been attempted on laboratory scale and also on industrial level. Initial studies were concentrated on determination of efficiency of experimental alternatives. Treated

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hides/skins were monitored periodically for physical changes like smell and hair slip, which are indications for putrefaction [8] Further several other analytical parameters like moisture content, volatile nitrogen, bacterial colony count, hydro thermal characterization, physical strength and properties and pollution load analysis, were included for the study of the goodness of the curing agent.

Numerous compounds and combinations were scrutinized for experimentation to preserve skin efficiently and to decrease pollution load from final effluent. Boric acid [9], boric acid in combination with NaCl [10], potassium chloride [11], sodium meta-bisulphite (SMBS) with acetic acid [12], Salt-less preservation by the use of neem oil (1%) with alcohol could also preserve the skin but the leather quality is affected [13]. Short-term preservation of hide/skin with hypo at an offer level of 5% on hide weight could preserve the skin/hide for 10 days [14]. Silica gel was also found to be a good dehydratant comparatively to the NaCl [15]. Eventually abovementioned and other practiced hide curing methods were standardized in laboratory and not cost effective for Indian scenario of leather industry.

Curing alone contributes 40–50% total dissolved solid (TDS) in final effluent so curing agent should follow the criteria of pollution control, i.e. less chloride (Cl^-) ion concentration and also in reach of flayers and slaughterhouses belongs basically to rural India because they use NaCl due to its economic feasibility and availability. Sodium sulphate was used as curing agent alone and in combinations due to having similar properties as like of NaCl but not has chloride in the formulation and proved better for goat skin preservation. In our previous work sodium sulphate was opted experimentally as initiative to find an alternative to sodium chloride, thus we optimized curing against sodium chloride and showed remarkable differences. Firstly, in reduction of the quantity used for curing, hence sodium sulphate referred as novel became cost comparative in term of Indian leather industries requirement. It was observed in our earlier work that the quantity of sodium chloride required for curing of goat skin was nearly half of weight of hide, but sodium sulphate showed same curing level only with the use of one-fifth the quantity of the hide weight [16].

Buffalo hides are somewhat different in physicochemical and structural properties from goat skin so the affectivity of sodium sulphate was also been tested against the numerous parameters for buffalo hide which found to be equally efficient as it was proved on goat skin.

2. Materials and methods

2.1. Materials

2.1.1. Buffalo hides/skins

Freshly flayered two buffalo skins 16 kg each of weight and area of approximately 20 ft^2 were purchased from local slaughterhouse near Indian Institute of Technology, Kanpur used in study.

2.1.2. Sodium sulphate

Commercial grade sodium sulphate white in color, were purchased from Neha Chemicals an authorized stockist of laboratory chemicals and Grasim Industries in Kanpur region.

2.1.3. Sodium chloride

Commercial NaCl purchased from outlet known for supply in slaughterhouses and hometown flayer community which have purity of 60–70% and grey in color.

2.2. Methods

Two freshly flayed buffalo hides were purchased from local slaughterhouse, and were treated with experimental salt and control salt. Experimental salt was prepared by mixing NaCl in two ratios, i.e. 10% and 20% of total weight used for curing with Na_2SO_4 hence the two ratios were prepared for experimentation 90:10 and 80:20 (Na_2SO_4:NaCl). One hide was cut into two equal halves and treated with the two ratios, i.e. 90:10 and 80:20 (Na_2SO_4:NaCl). Out of the four hide halves of about 8 kg each, three were treated with pure sodium sulphate and combination of sodium sulphate:sodium chloride separately, so three halves were treated with experimental salt [ratios of 90:10 and 80:20 (Na_2SO_4:NaCl) and pure sodium sulphate] and rest one half piece of hide was treated with control salt (sodium chloride). For sodium chloride treatment 4.5 kg of salt was used as tradition and for experimental salt [ratios of 90:10 and 80:20 (Na_2SO_4: NaCl) and pure sodium sulphate] about 900 g (rationally one-fifth of sodium chloride mediated curing quantity) was utilized. When lesser quantities of both experimental salt mixtures and controlled salt were used the hide samples started deteriorating very fast even at 30–35 °C (as an attempt for reducing pollution load, but it failed). The skin halves were folded and kept at ambient temperature of 30–35 °C. These were monitored periodically for putrification, hair slip and physical changes like odor that are indication of auto proteolysis. The experimental skin halves were kept for 21 days (3 weeks). Efficiency of curing with both experimental and control salt was systematically assessed by analysis of treated skin halves for change in moisture content, total extractable nitrogen content, bacterial colony count. Scanning electron micrographs are taken after treatment of experimental salt and control salt at different intensifying zooms, i.e. 250× and 1500×. Method for pollution analysis adopted for biochemical oxygen demand (BOD), chemical oxygen demand (COD), TDS, total suspended solid (TSS) and chloride ion content in effluent.

2.2.1. Determination of moisture content

Skins were preserved with experimental salt and control salt, were unhaired without disturbing the moisture content and weighed. After each period of experimentation the moisture content of skin pieces was determined by using the procedure outlined in [17].

2.2.2. Determination of nitrogen content

Cured samples of known weight (5 g) were cut from the experimental and control-treated skins, mixed with distilled water in 1:10 w/v, and shaken well in a bottle for 3 h at 30–35 rpm. The liquid was then filtered through a filter paper, digested and the amount of nitrogen was determined using the kjeldahl method [17].

2.2.3. Determination of bacterial count

Bacterial colony count determines the number of bacterial colonies present in per gram of medium at different preservation duration after curing with experimental salt mixtures and control salt. 5 g preserved skin pieces were weighed and soaked in 50 ml sterile water; the skin extract was prepared by shaking it in the arbitral shaker at 200 rpm for 30 min. 1 ml of liquid in which skin pieces had been soaked was taken in 9 ml of sterile water and shaken well to get uniform suspension of the bacteria. A volume of 0.1 ml of the resulting diluted solution was taken in sterile petri plates, and molten nutrient agar at 40 °C was poured and shaken gently to obtain uniform distribution of the bacteria. The plates were incubated at 37 °C for 48 h and colony forming units were determined by serial dilution method [18].

2.2.4. SEM studies

After 3 week of storage period the preserved skin samples were subjected to processing at a tannery house. Processed leather samples were taken for SEM studies as follows. First miniature pieces from same area of leather sample have been taken for gold plating. After 15–20 min of gold plating, samples were subjected to scanning electron micrograph (SEM-EDAX). The micrographs for the grain surface and cross section were obtained by operating the SEM at an accelerating voltage of 20 kV with different magnification 250× and 1500×.

2.2.5. Pollution load generated in leather processing

The spent liquid from the primary unit operation was quantitatively collected and analyzed for pollution parameters such as BOD, COD, TDSs, TSSs and Cl^-s content using standard analytical procedures [19]. The results were expressed in terms of emission, in mg per kg of the raw material, as compared with conventional salt cured stock, expressed in terms of emission load per kg of the raw material.

3. Results and discussion

Evaluation of the experimental salt and control salt treatment outcomes instigated with analysis of moisture content. It helps in diagnosis of the presence and activity of microorganisms during preservation process. Due to microbial activity structural component breakdown take place that leads to the formation of amino compounds that are estimated as nitrogen content on different time scale, points towards rate of hydrolysis of structural protein component caused by microorganism. Quantity of microorganism on preserved skin further estimated by serial dilution method in terms of colony forming unit. Curing agents inhibits bacterial growth through the interaction with the hide structural component. These interactions change fibrous striation and various structural properties of hide so to find these alterations scanning electron micrographs are taken. Finally for the preservation of environmental safety, pollution load is analyzed which was generated by the experimental salt and control salt separately during preservation.

The buffalo hide, preserved with experimental salt ratios [Na_2SO_4:NaCl (90:10), Na_2SO_4:NaCl (80:20) and Na_2SO_4] and control salt (NaCl) were studied for various properties during the incubation period. Moisture content was to assess the curing competency. Successive loss in the water (moisture) in all four samples was observed till the end of the experimentation period. An apparent digression in the values of moisture content between the experimental salt ratio [Na_2SO_4:NaCl (90:10), Na_2SO_4:NaCl (80:20) and Na_2SO_4] and control salt (NaCl) has been shown in Table 1 and Fig. 1. For instance, controlled salt treated buffalo hide lost 28%, 13%, 4% and 5% moisture during four consequent intervals, i.e. 0–1st day, 1st–7th and so on, while a continuous and steady dehydration was also observed in case of experimental salt ratio [Na_2SO_4:NaCl (90:10), Na_2SO_4:NaCl (80:20) and Na_2SO_4]. Evidently dehydration rate creates hyper tonicity which was found to be greater in case

Table 1
Moisture content (in %)

Duration of preservation	NaCl	Na_2SO_4:NaCl (90:10)	Na_2SO_4:NaCl (80:20)	Na_2SO_4
0 h	75 ± 2	75 ± 2	75 ± 2	75 ± 2
1st day	47 ± 2	55 ± 2	51 ± 2	58 ± 2
7th day	34 ± 2	45 ± 2	42 ± 2	49 ± 2
15th day	30 ± 2	37 ± 2	32 ± 2	42 ± 2
21st day	25 ± 2	29 ± 2	28 ± 2	35 ± 2

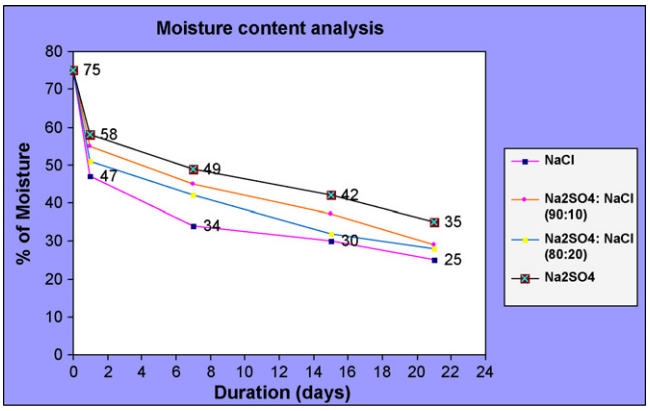

Fig. 1. Moisture content analysis of buffalo skin on different time scale when treated with experimental and control salt separately.

of control salt (NaCl) treatment till the end of day 21. On the other hand, experimental salt ratios [Na_2SO_4:NaCl (90:10), Na_2SO_4:NaCl (80:20) and Na_2SO_4], despite of a higher level of moisture content, was able to preserve the skin.

The amount of extractable nitrogen is an index of microbial attack and putrification of proteins of skin by which amino group containing components are obtained that leads to generation of bad odor and hair slip during preservation. Fig. 2 shows the total extractable nitrogen values during curing experiment of the skin using experimental salt [Na_2SO_4:NaCl (90:10), Na_2SO_4:NaCl (80:20) and Na_2SO_4] and control salt (NaCl). According to the results, control salt treated skin showed increase in total nitrogen content by twofold from initial value while with the experimental system there is slight increment because initially total nitrogen content was low as compared to the control salt treated skin values, i.e. 2.40, 2.84 and 2.71 for [Na_2SO_4:NaCl (90:10), Na_2SO_4:NaCl (80:20) and Na_2SO_4] and 3.32 for control salt (NaCl), respectively. The difference in the total extractable nitrogen between experimental and control salt during experimentation phase showed the extraordinary inhibitory potential of experimental salt in curing which is also due to the mutual effect of the antimicrobial property of experimental salt and the simultaneous decrease in the moisture content that is likely to preserve the buffalo skin efficiently (Table 2).

The curing efficacy of agents depends mainly on the growth inhibitory properties for proteolytic bacterial species on the skin protein. The skin degradation can be directly analyzed by the presence of proteolytic bacteria present on the skin under preservation during the incubation period. Table 3 shows the bacterial population of the fresh buffalo skin and preserved skin at different

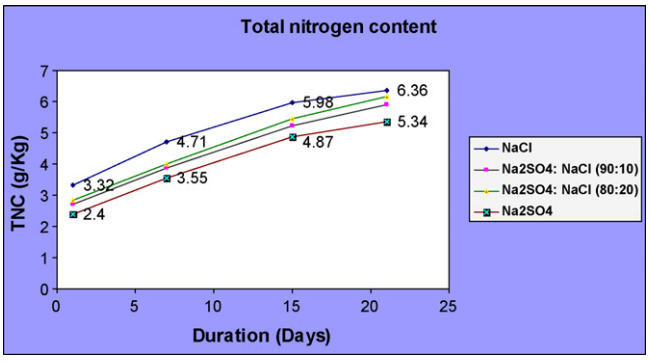

Fig. 2. Total nitrogen content in buffalo skin after the treatment of experimental and control salt.

Table 2
Total extractable nitrogen content (g/kg) of skin

Duration of preservation	NaCl	Na_2SO_4:NaCl (90:10)	Na_2SO_4:NaCl (80:20)	Na_2SO_4
1st day	3.32 ± 0.10	2.71 ± 0.10	2.84 ± 0.10	2.40 ± 0.10
7th day	4.71 ± 0.10	3.86 ± 0.10	3.99 ± 0.10	3.55 ± 0.10
15th day	5.98 ± 0.10	5.23 ± 0.10	5.46 ± 0.10	4.87 ± 0.10
21st day	6.36 ± 0.10	5.89 ± 0.10	6.17 ± 0.10	5.34 ± 0.10

Table 3
Bacterial count against time scale (no. of colonies per gram of medium)

Duration of preservation	NaCl	Na_2SO_4:NaCl (90:10)	Na_2SO_4:NaCl (80:20)	Na_2SO_4
0 h	2×10^3	2×10^3	2×10^3	2×10^3
1st day	3×10^{10}	2×10^9	2×10^9	2×10^8
7th day	2×10^8	2×10^7	3×10^7	2×10^7
15th day	3×10^7	3×10^5	3×10^5	3×10^5
21st day	4×10^6	2×10^5	3×10^5	2×10^5

intervals. The experimental salt showed relatively lower bacterial count in comparison to the control salt treated buffalo skins in per gram of growth medium. This clearly demonstrates the antimicrobial property of experimental salt. The microbial growth data reveals that the experimental salt starts preservation hides not only at initial stage but also at later stages on applied concentrations and in the given experimental conditions.

3.1. SEM analysis

SEM pictures were taken after treatment of skin for both experimental and control salt separately. Essentially, to analyze effect of salt treatment on skin, common regions of all skin were chosen for scanning. Obtained micrographs were based on the magnification of conjoint bundles of constituting fibers of skin which have been taken on two intensified zooms 250× and 1500×. Every micrograph has been concentrated to put emphasis on the bundling of fibers because as the bundles will be properly gyrated on each other, there efficiency of absorption and physical properties will enhance evenly and leather quality will be better, thus SEM pictures have given an insight to the effect caused by the experimental salt and control salt treatments. As it can be seen that NaCl-treated skin (Fig. 3) micrographs showed dispersed bundles, lack of proper striation and fabrication while experimental salt treated skin either from Na_2SO_4:NaCl (90:10) (Fig. 4), Na_2SO_4:NaCl (80:20) (Fig. 5) or from pure Na_2SO_4 (Fig. 6)) showed proper convention in fabrication (shown in 250× zoom at left which when magnified on 1500× in right micrograph shown by arrow). Scanning electron micrographs of hide cured with experimental composition exhibit

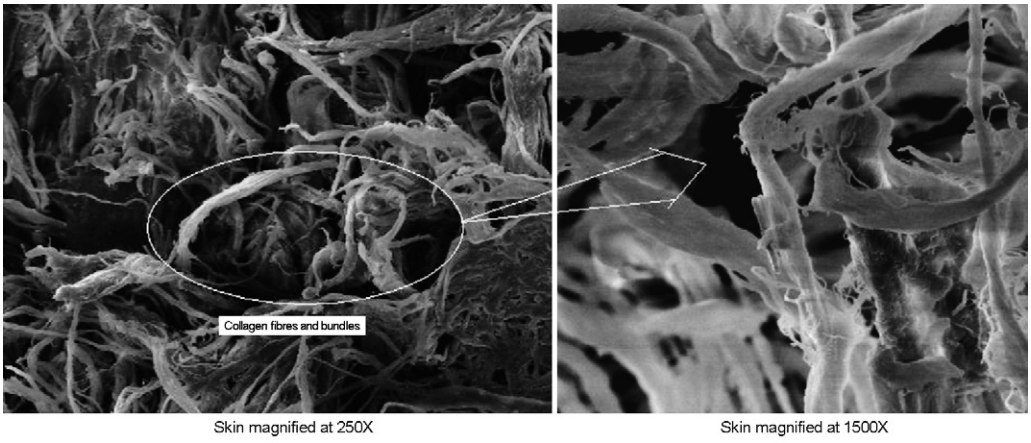

Fig. 3. NaCl-treated skin micrographs.

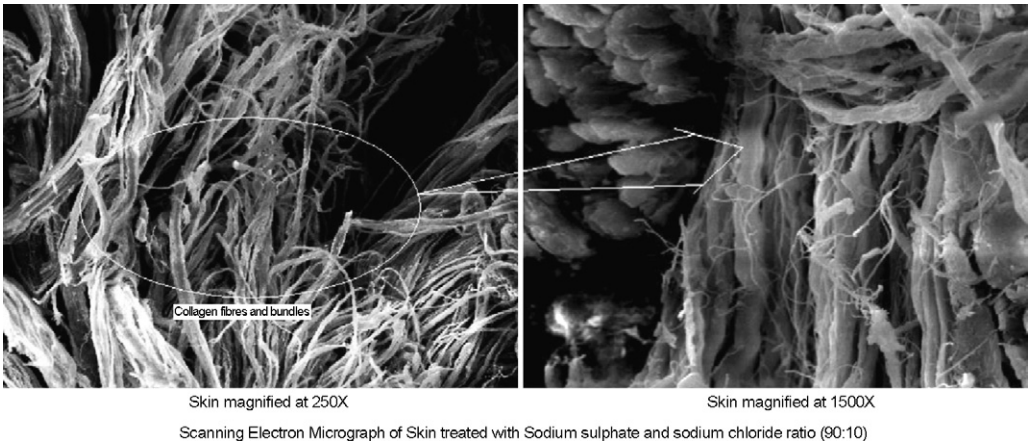

Fig. 4. Micrographs of skin treated with sodium sulphate and sodium chloride (90:10).

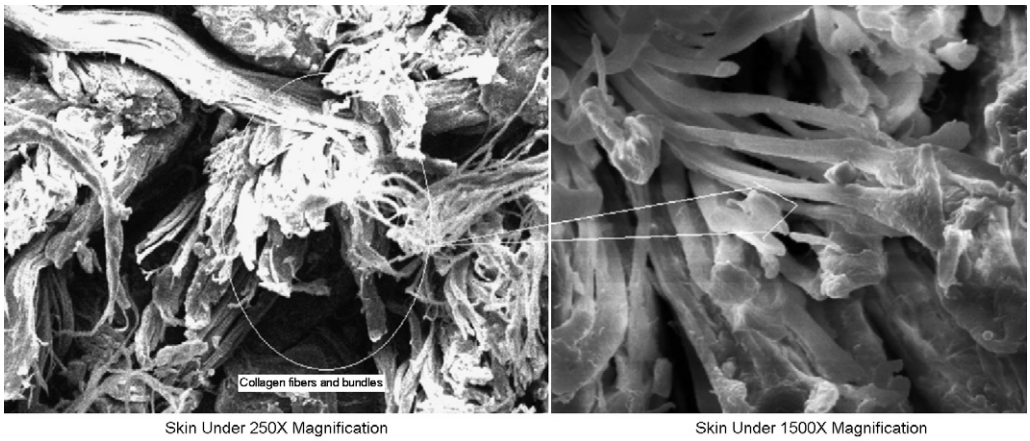

Fig. 5. Micrographs of skin treated with sodium sulphate and sodium chloride (80:10).

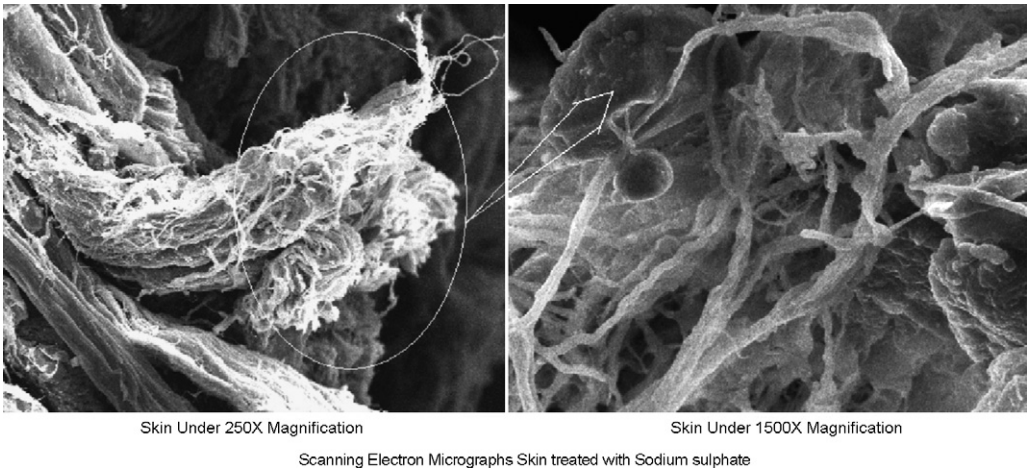

Fig. 6. Micrographs of skin treated with sodium sulphate.

properly arranged bundle arrays, i.e. non-over lapping and wrapped separately, which absorb the dye properly in the further processing of the hide; giving a well lustrous grain to leather.

SEM micrographs clearly show the formation of striations in the cases of sodium sulphate cured samples, however the exact role of SO^{-4} ion in the formation of bundles/striations is not known.

Table 4 and Fig. 7 represent the extent of pollution load contributed by experimental salt and control salt during the soaking operation. The results clearly show that there is substantial reduction in the values of the BOD, COD and TSS but there is huge reduction in values of TDS and Cl^- compared to the control salt mediated preservation. As mentioned earlier, the amount of sodium sulphate utilized in curing was nearly one-fifth less then that of sodium chloride, thus it lead to obvious reduction in TDS/Cl^- [16].

Table 4
Shows pollution load analysis in (g/kg)

Parameters	NaCl	Na_2SO_4:NaCl (90:10)	Na_2SO_4:NaCl (80:20)	Na_2SO_4
BOD	11 ± 1	8 ± 1	9 ± 1	7 ± 1
COD	32 ± 1	18 ± 1	19 ± 1	16 ± 1
TDS	255 ± 1	156 ± 1	160 ± 1	145 ± 1
Cl^-	219 ± 1	41 ± 1	46 ± 1	29 ± 1
TSS	26 ± 1	12 ± 1	15 ± 1	11 ± 1

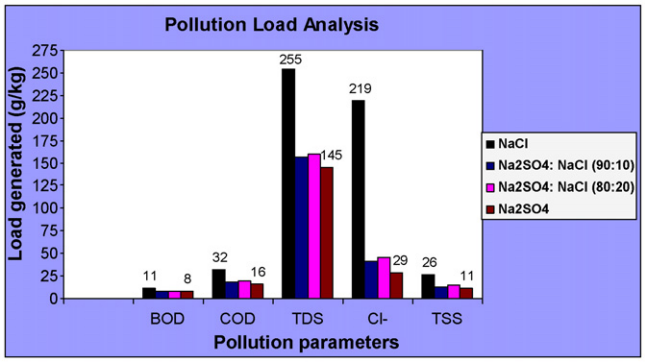

Fig. 7. Pollution load analysis with experimental salt ratio and control salt.

4. Conclusion

Study on sodium sulphate mediated buffalo hide preservation was carried with five perspective experiments. Every experiment enlightened the effect of sodium sulphate on buffalo hide preservation comparatively to the sodium chloride.

(1) The experiments related to the preservation efficiency like moisture content study, total nitrogen generated during exper-

imentation, sodium sulphate and its additive ratios showed better efficacy on buffalo hide as proved in previous work [16] on other types of hides like goat.

(2) Further scanning electron micrographs proved uttered utility and compatibility of sodium sulphate as curing agent because it enhance bundling and striation of fibers which is supposed as necessary requirement for tannage acceptance. Since initial aspect of this experimentation was to develop a method and novel curing agent for buffalo hide that would be able to reduce chloride mediated salinity up to 10-folds and TDS up to 2–3-folds which was achieved by using sulphate in place of chloride in less quantity it is able to preserve at the same. Since the quantity of sodium sulphate used per hide was 1/5 times as compared to sodium chloride, thus the TDS also reduces significantly in the former case.

(3) Finally pollution load analysis proved sodium sulphate least hazardous in terms of salinity compared to the traditional sodium chloride curing agent.

(4) Thus we can conclude from the abovementioned benefits that sodium sulphate can easily replace sodium chloride as it is fortunately a commonly available chemical and can be used as an smart alternative in hide preservation technology.

References

[1] W.E. Kallenberger, Halophilic bacteria in brine curing, J. Am. Leather Chem. Assoc. 79 (1984) 104–114.
[2] R.G.H. Elliott, Methods for the treatment and preservation of raw hides and skins. A review, J. Soc. Leather Technol. Chem. 67 (1983) 31–34.
[3] J.W. Mitchell, Prevention of bacterial damage on brine cured and fresh cattle hides, J. Am. Leather Chem. Assoc. 82 (1987) 372–382.
[4] Lakshmi Muthusubramanian, R.B. Mitra, A cleaner production method for the synthesis of bronopol—a bactericide that is useful in leather making, J. Clean. Prod. 14 (2006) 536–538.
[5] M.S. Heakal, A.S. Modaihsh, A.S. Mashhady, A.I. Metwally, Combined effects of leaching fraction salinity and potassium content of waters on growth and water use efficiency of wheat and barley, Plant Soil 21 (1990) 177–184.
[6] Z. Koszanski, S. Karczmarczyk, Use of saline water for irrigation of spring barley and oats., Zeszyty-Naukowi-Akademii-Rolniczej-w-zczecinie, - Rolnictwo. 36 (1985) 95–105.
[7] E.R.R. Iyengar, J. Chikara, P.M. Sutaria, Relative salinity tolerance of barley varieties under semi-arid climate, Trans. Indian Sot. Desert Technol. Univ. Center Desert Stud. 9 (1984) 27–33.
[8] M. Sivaparvathi, S.C. Nandy, Evaluation of preservatives for skin preservation, J. Am. Leather Chem. Assoc. 69 (1974) 349–362.
[9] I.R. Hughes, Temporary preservation of hides using boric acid, J. Soc. Leather Trade Chem. 58 (1974) 100–103.
[10] J. Kanagaraj, J.V. Sundar, C. Muralidharan, S. Sadulla, Alternatives to sodium chloride in prevention of skin protein degradation—a case study, J. Clean. Prod. 13 (2005) 825–831.
[11] D.G. Bailey, J.A. Gosselin, The preservation of animal hides and skins with potassium chloride, J. Am. Leather Chem. Assoc. 91 (1996) 317–333.
[12] J. Kanagaraj, T.P. Sastry, C. Rose, Effective preservation of raw goat skins for the reduction of total dissolved solids, J. Clean. Prod. 13 (2005) 959–964.
[13] P.S. Venkatachalam, S. Sadulla, B. Duraisamy, V.S. Krishnamurthi, Short-term preservation of hide with neem oil, J. Soc. Leather Technol. Chem. Assoc. 61 (1977) 24.
[14] P.S. Venkatachalam, S. Sadulla, B. Duraisamy, Preliminary curing trials without the use of common salt, Leather Sci. 28 (1981) 151.
[15] J. Kanagaraj, N.K. Chandra babu, S. Sadulla, R.V. Suseela, G. Visalakshi, K.N. Chandra, Cleaner techniques for the preservation of raw goat skins, J. Clean. Prod. 9 (2001) 261–268.
[16] P.S. Vankar, A. Dwivedi, R. Saraswat, Sodium sulphate as a curing agent to reduce saline chloride ions in the tannery effluent at Kanpur: a preliminary study on techno-economic feasibility, Desalination 201 (2006) 14–22.
[17] Bureau of Indian Standards, Chemical Testing of Leather, 1971, p. 2–80.
[18] R. Cruickshank, Determination of bacterial count method, Med. Microbiol. (1965) 768–769.
[19] A.D. Eaton, L.S. Clesceri, A.E. Greenberg, Standard Methods of the Examination of Water and Wastewater, 19th ed., APHA, Washington, DC, 1995.