Keratin Production by The Enzymatic Method and Its Reuse in The Leather Process

Aykut Sancakli [1], Fatih Arican [1], Aysegul Uzuner Demir [1], Carmen Gaidau [2], Mihaela Niculescu [2], Maria Stanca [2], Daniela Berechet [2], Cosmin Alexe [2], Stoica Tonea[3]*

[1] Kazlicesme R&D Center and Test Laboratories, Tuzla, Istanbul, Turkey

[2] Research & Development National Institute for Textiles and Leather-Division Leather and Footwear Research Institute, Ion Minulescu Str., Bucharest, 031215, Romania,

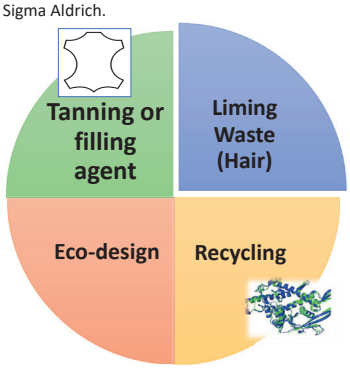
[3]SC Taro Comimpex Srl, Drumul Sarii Str, 60151, Bucharest, Romania,

Introduction

Unhairing and liming are the main steps carried out during leather production in the tannery, with impacting on effluent and leather quality. In this study, present work aims to show the possibility of recovery and recycling of saved hair from unhairing process based on keratinase enzyme which is a greener process. The separated hair was solubilized by chemical hydrolyses and characterized for dry substance, pH, total nitrogen, ash and molecular weight determination. Furthermore, different keratin/acrylic polymer ratios and reaction parameters were experimented with final product optimization, given us tanning, retanning, or filling agent for leather processing. The new products were characterized by Fourier Transform Infrared (FTIR) spectroscopy, scanning electron microscopy (SEM), differential scanning calorimeter (DSC), and thermal gravimetric analyses (TGA).

Material and Method

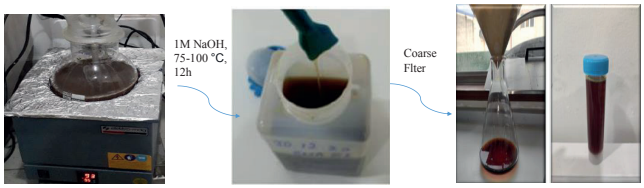
Bovine hair was supplied from Iskefe Leather tannery as a waste of the leather processing liming and used to obtain keratin. Acrylamide (AAm), keratinase enzyme, sodium hydroxide (NaOH), sodium dodecyl sulfate (SDS) and hydrogen peroxide(H2O2) were supplied from Sigma Aldrich.



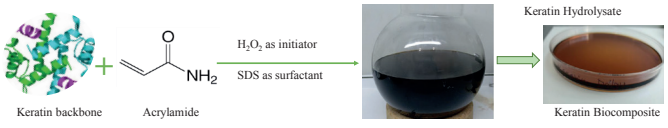
Keratin Extraction and Its Biocomposite Synthesis



Leather Liming Waste (Hair) obtained by Enzymatic Method (keratinase)



Keratin Hydrolysate obtained by Leather Liming Waste (Hair)



Keratin Backbone Grafted with Acrylamide Monomer

Result & Discussion

Table 1. RSM trial design applied for keratin extraction.

	Solution Amount (L)	% Raw Material Cons. (w/v)	Temperature (°C)	% Alkaline cons. (w/w)	Type of Alkaline	Reaction time (h)
1	1	5.5	75	36	NaOH	12
2	1	10	100	30	NaOH	12
3	1	10	75	40	NaOH	12
4	1	10	75	20	NaOH	12
5	1	50	100	16	NaOH	12
6	1	50	100	24	NaOH	12

Table 2. Spesifications for extracted keratin.

Characteristics	Keratin
Dry substance, % SR EN ISO 4684:2006	15,80
Ash, % SR EN ISO 4047:2002	14,56
Total nitrogen, % SR ISO 5397:1996	7,91
Protein substance, % SR ISO 5397:1996	47,97
pH, STAS 8619/3:1990	7,13
Aminic nitrogen, % Method ICPI	15,70
Cysteine, % SR 13206:13206:1994	0,45
Cystinic sulphur, % SR 13206:13206:1994	0,12
Molecular weight (Mn)	3644

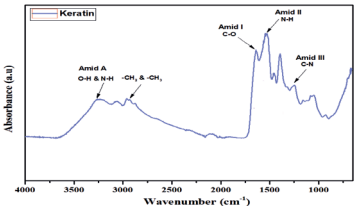


Figure 1. FT-IR spectrum for keratin.

Table 3. The synthesis of keratin biocomposite.

Volume (ml)	T/ (oC)	Protein (ml)	Monomer (g)	Initiator (ml)	Surfactant (gr)	Destile water (ml)	Time (h)
200	80	40	40	0,05	0,04	140	6
200	80	40	40	0,1	0,04	140	6
200	80	40	40	0,3	0,04	140	6
1000	80	200	200	0,3	0,4	700	6
1000	80	200	200	0,3	0,4	700	6
1000	80	200	200	0,3	0,4	700	6

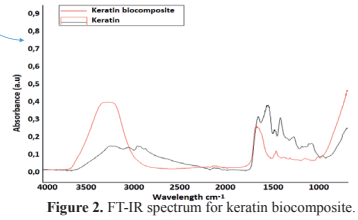




Figure 2. FT-IR spectrum for keratin biocomposite.

Table 4. Physical-mechanical characteristics of bovine leathers retanned with new keratin biocomposite (MZK)

Characteristics	Control	Keratin Biocomposite (MZK)
Elongation at cracking, %	14.40	**12.05**
Elongation at breaking, %	14.40	**12.05**
Cracking strength, N/mm²	14.05	**18.35**
Tensile strength, N/mm²	14.05	**18,35**
Tear force, N	24,85	**27.20**
Softness, Ø 25 mm	3.36	**3.08**

Conclusion

The main purpose of liming is to separate the hair from the hides, to release the interfibrilar soluble proteins, greasy matters and to transform the raw hide in pelt. This study obtained hair waste from the liming process with the enzymatic method, and the alkaline method extracted keratin hydrolysate from hair. Then, optimum process parameters were achieved for obtaining keratin biocomposite from the keratin hydrolysate. Also, all synthesized materials during study were characterized in detail. Finally, keratin biocomposite obtained from the leather process wastes was reused in the retanned process with other additives (MZK) to increase process efficiency.

As a result, the mechanical properties of the retanned leather produced with the new material were examined. The results of experiments performed in hide tanning and leather retanning processes showed the possibility to use a new material based on recovered hair keratin as circular technology.

References

[1] T. Luo, S. Hao, X. Chen, J. Wang, Q. Yang, Y. Wang, Y. Weng, H. Wei, J. Zhou, B. Wang, Development and assessment of keratein nanoparticles for use as a hemostatic agent, Mater. Sci. Eng. C-Mater. Biol. Appl. 63, 352–358, 2016.
[2] N. Eslahi, F. Dadashian, N.H. Nejad, Prep. Biochem. Biotechnol. 43, 624–648, 2013.
[3] X.-C. Yin, F.-Y. Li, Y.-F. He, Y. Wang, R.-M. Wang, Biomaterials Science 1, 258–536, 2013.
[4] S.-W. Ha, A.E. Tonelli, S.M. Hudson, Biomacromolecules 6, 1722–1731, 2005.

Acknowledgements

The research was carried out within a grant funded Scientific and Technological Research Council of Turkey, project number 9200010, E! 13359 KER_COL_CE.