

Converting of animal-derived protein waste into new biomaterials in a circular economy concept



M. Râpă *, C. Gaidău **, Liliana Mititelu-Tartau ***, Andrei Constantin Berbecaru *, Ecaterina Matei *, Cristian Predescu *

*Faculty of Material Science and Engineering, University POLITEHNICA of Bucharest, Bucharest, Romania
(E-mail: maria.rapa@upb.ro; Andrei.Berbecaru@upb.ro; Ecaterina.matei@upb.ro; Cristian.predescu@upb.ro)

**The National Research & Development Institute for Textiles and Leather-Division, Bucharest, Romania
(E-mail: carmen_gaidau@hotmail.com)

***Pharmacology, Clinical Pharmacology and Algesioly Department, Faculty of Medicine "Grigore T. Popa", University of Medicine and Pharmacy, Iasi, Romania
(E-mail: ltartau@mail.umfiasi.ro)

Introduction

A significant amount of proteins results from the processing of mammal skins (cattle, sheep, etc.) and sheep's wool, which can be exploited due to their bioactive potential for the regeneration of damaged human tissues. For example, these proteins can be deposited in the form of nanofibers on a cotton support through the electrospinning process.

The use of animal-derived proteins in the electrospinning process for the fabrication of wound dressings compared to synthetic polymers, is an environmentally friendly approach. This is because non-toxic solvents are used in preparing solutions, and these proteins possess antimicrobial and biocompatibility properties.



Figure 1: Mono- and coaxial electrospinning equipment used to obtain assembled PLA/PEO/Keratin-PVP/Collagen nanofibers

Table 1. Compositions and optimal parameters for obtaining of electrospun biomaterials

Composition	Electrospinning
PLA/PEO	Mono
PLA/PEO/KH	1 st layer, Coaxial
PVP/CH	2 nd layer, Coaxial
PLA/PEO/KH - PVP/CH	Assembled structure
PLA/PEO/PVP	Mono

In this paper, we developed dual-layered wound dressings using biodegradable matrices, namely poly(lactic acid) (PLA), poly(ethylene oxide) (PEO), and poly(vinyl pyrrolidone) (PVP), loaded with hydrolysed keratin (KH) and bovine collagen glue (CH). The dressings were fabricated using both mono- and coaxial electrospinning technology. (Figure 1).

Results & Discussion

Table 2. Physico-chemical characteristics of concentrated collagen hydrolysate and keratin hydrolysate

Characteristics, U.M.	Value ± Standard Deviation	
	Collagen hydrolysate (CH)	Keratin hydrolysate (KH)
Dry matter, %	60.40 ± 0.42	9.02 ± 0.05
Ash, %	6.24 ± 0.27	13.73 ± 0.25
Total nitrogen, %	14.67 ± 0.66	14.40 ± 0.57
Protein, %	82.43 ± 2.66	80.84 ± 1.40
pH, pH units	8.54 ± 0.10	11.84 ± 0.09
Aminic nitrogen, %	1.43 ± 0.06	1.34 ± 0.06
Electrical conductivity, µs/cm	870 ± 0.1	13700 ± 20

Morphology and Structure

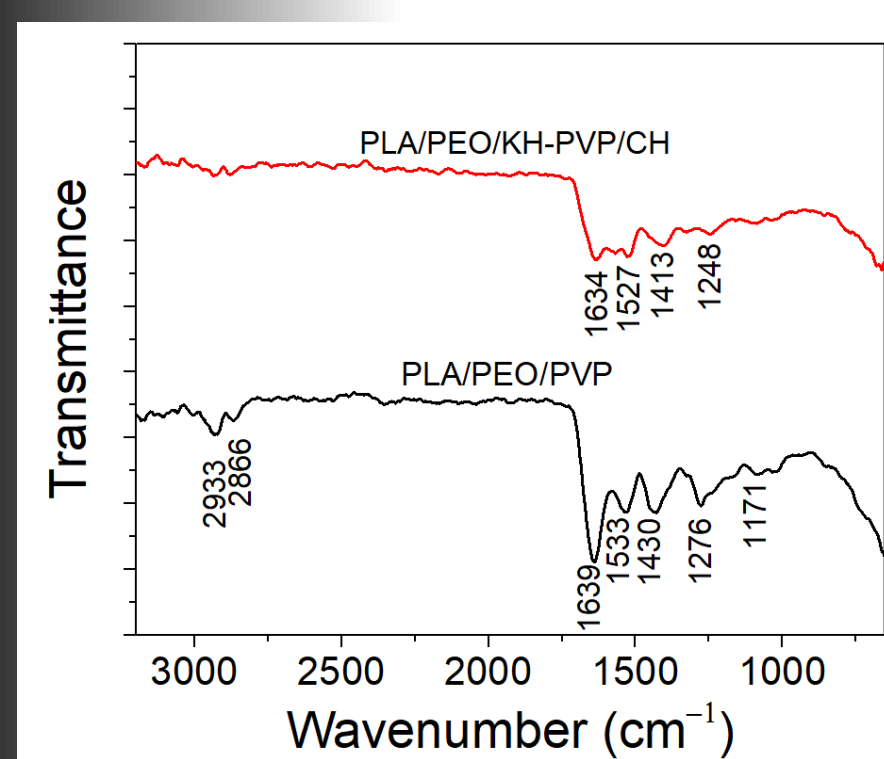
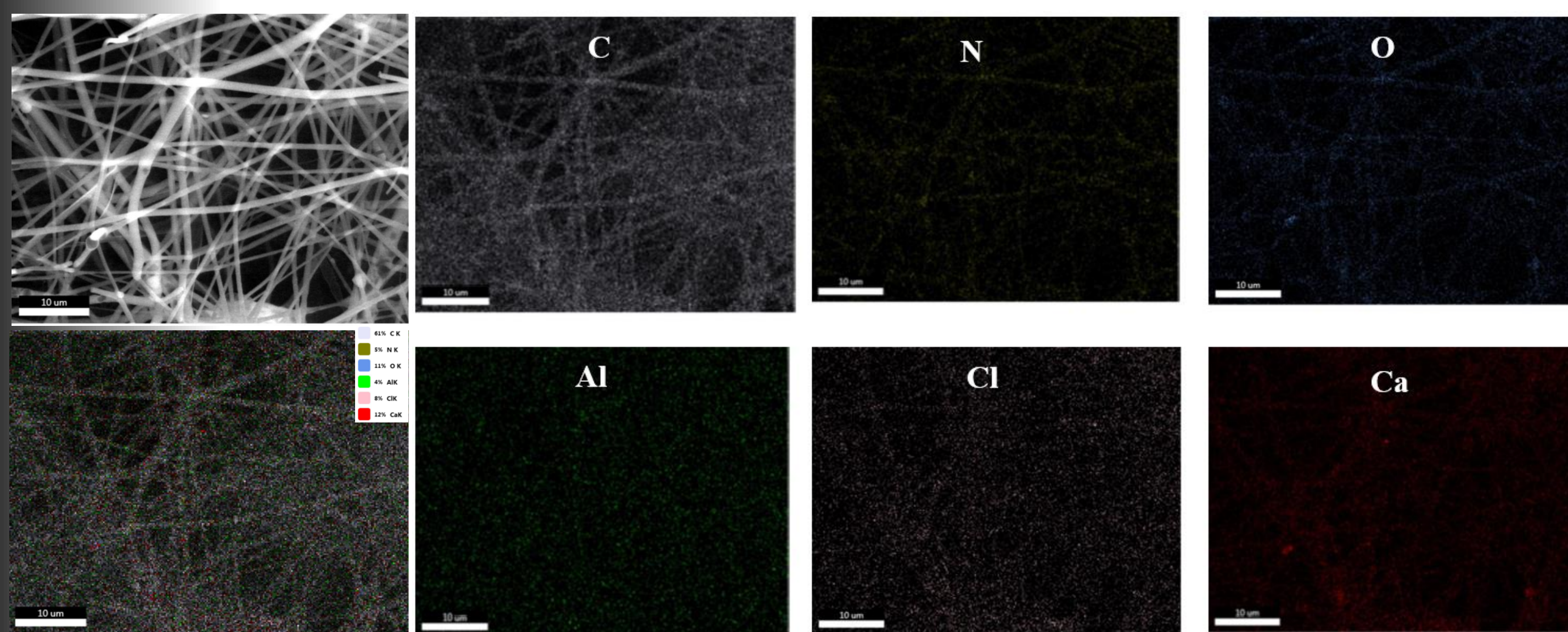
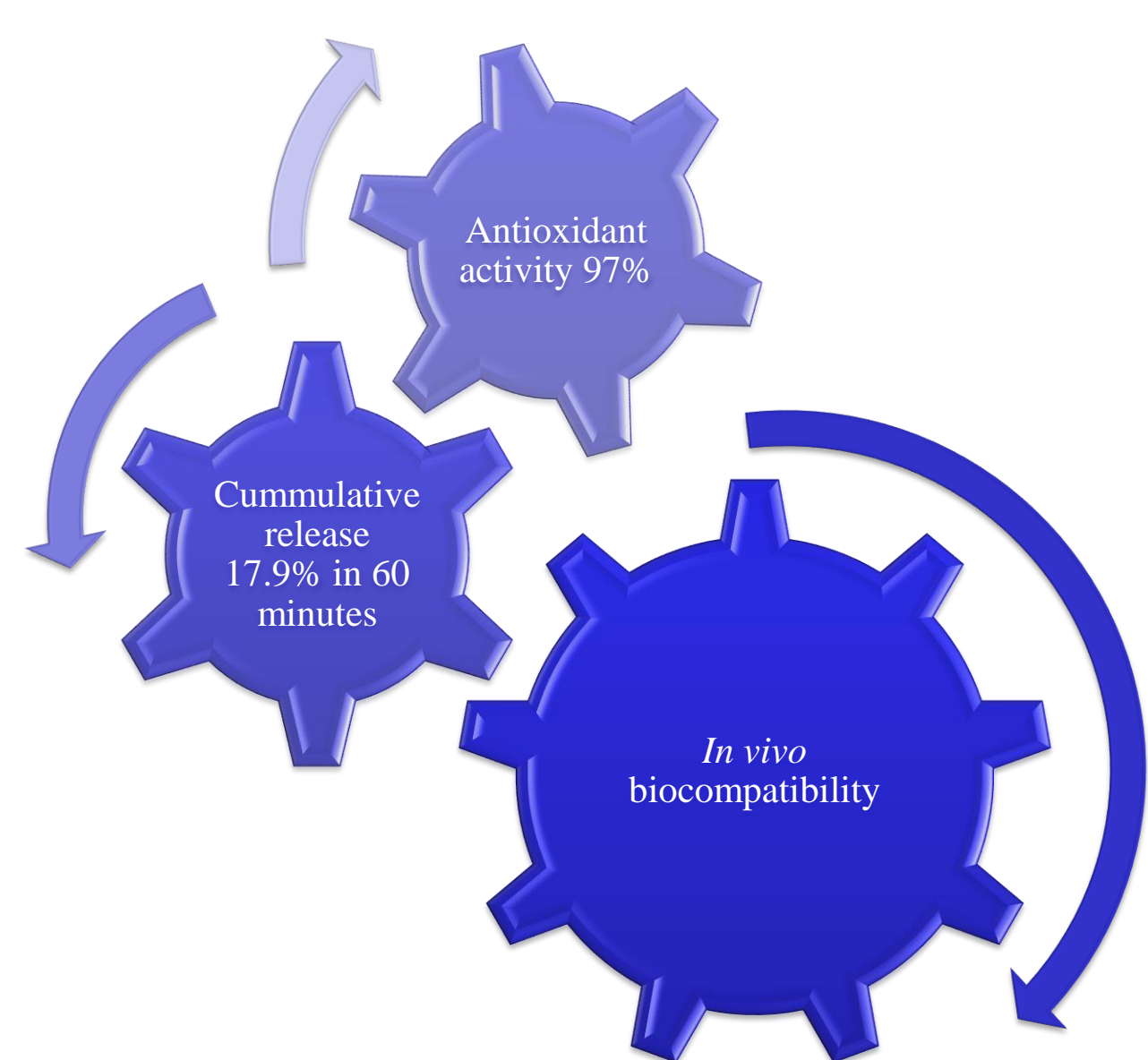


Figure 2: ATR-FT-IR for PLA/PEO/PVP and PLA/PEO/KH-PVP/CH



In vivo biocompatibility tests

- **Leukocyte formula:** neutrophil polymorphonuclear (PMN), lymphocytes (Ly), eosinophils (E), monocytes (M), and basophils (B)
- **Liver enzymes:** The serum values of glutamic-oxaloacetic transaminase (TGO), glutamic-pyruvic transaminase (TGP), and lactate dehydrogenase (LDH)

Table 3. Percentage values of leukocyte formula elements measured in animals that received nanofibers

	Period of time	Leukocyte formula (%)				
		PMN	Ly	E	M	B
Witness	24 h	29.4±7.7	63.9±18.3	0.1±0.01	6.1±1.1	0.2±0.05
	7 d	28.6±8.9	64.8±19.5	0.2±0.05	6.2±1.1	0.2±0.05
PLA/PEO	24 h	28.5±8.5	65.0±17.5	0.2±0.05	6.1±1.3	0.2±0.05
	7 d	28.7±9.1	64.6±18.7	0.2±0.01	6.3±1.3	0.2±0.1
PLA/PEO/KH	24 h	29.2±8.3	64.1±18.9	0.1±0.01	6.4±0.5	0.2±0.1
	7 d	28.9±9.3	64.5±18.3	0.1±0.01	6.3±1.1	0.2±0.05
PLA/PEO/PVP	24 h	29.3±7.9	64.3±17.9	0.1±0.05	6.1±1.1	0.2±0.1
	7 d	29.1±8.7	64.4±19.1	0.1±0.01	6.2±1.3	0.2±0.1
PVP/CH	24 h	28.6±8.5	64.8±18.5	0.2±0.01	6.2±1.1	0.2±0.1
	7 d	27.8±8.9	64.5±18.7	0.1±0.05	6.3±1.3	0.2±0.05
PLA/PEO/KH - PVP/CH	24 h	28.6±8.7	65.6±18.9	0.1±0.01	6.3±1.1	0.2±0.05
	7 d	28.8±9.1	64.4±17.7	0.2±0.05	6.4±0.5	0.2±0.1

Table 4. Changes in serum levels of TGP, TGO and LDH in animals that received nanofibers

	Period of time	TGP (U/ml)	TGO (U/ml)	LDH (U/l)
Witness	24 h	39.6±10.3	158.6±31.5	328.28±64.33
	7 d	40.2±10.5	160.4±30.7	333.56±70.67
PLA/PEO	24 h	40.5±10.9	161.7±32.9	332.34±71.13
	7 d	40.7±10.7	163.2±33.7	335.83±66.83
PLA/PEO/KH	24 h	39.4±9.7	159.8±35.3	330.45±69.45
	7 d	40.3±10.5	161.3±32.7	334.32±58.83
PLA/PEO/PVP	24 h	40.6±11.3	158.5±33.5	331.27±72.13
	7 d	39.8±9.9	162.7±31.7	335.19±71.33
PVP/CH	24 h	39.5±10.7	160.6±34.1	332.53±67.67
	7 d	39.8±10.3	163.8±33.5	336.13±69.27
PLA/PEO/KH - PVP/CH	24 h	39.7±10.5	160.4±35.3	330.67±71.33
	7 d	40.5±11.1	161.3±30.9	333.21±70.67

Conclusions

All studies demonstrated that, in our laboratory conditions, the use of nanofibers containing bovine glue and keratin hydrolysate, incorporated in biodegradable polymers, did not produce significant hematological and biochemical and did not significantly influence some specific stress parameters oxidative, compared to the use of patches with a textile support.

The use of collagen and keratin hydrolysates extracted from animal by-products and processing represents a sustainable and circular approach for added value new biomaterials and waste valorization.

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