



## Beeswax-poly(vinyl alcohol) composite films for bread packaging

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### ARTICLE INFO

#### Keywords:

PVA  
Beeswax  
Thermal treatment  
Characterization  
Biodegradability  
Food packaging

### ABSTRACT

The study aimed to improve the barrier characteristics of polyvinyl alcohol (PVA) film by adding beeswax (BW) and glycerin and by heat treatment in order to become a potential suitable material for bread packaging. XRD, FTIR and XPS showed new cross-links between PVA and beeswax demonstrating the composite nature of the film. TGA-DTA showed that glycerin reduced the dehydration and that the beeswax improved their thermal stability. The best PVA-beeswax film formula has water vapor permeability (WVP) only 7.5 times higher than that of polyethylene (PE), while PVA is 29 times more permeable. The correlation coefficients, linear (MLR) and step-wise (SR) regression modelling showed a direct correlation between WVP, solubility in food simulants, swelling capacity and biodegradability. Principal Component Analysis (PCA) demonstrated that the analyzed films have different behavior from PE, and among them the most similar is that of the PVA with 2 g of beeswax and no glycerin sample. This film type also showed efficiency in bread packaging.

### 1. Introduction

Food packaging plays a crucial role in controlling the quality and safety of food products and therefore must have very good barrier properties against gases, light, odors and liquids and excellent mechanical resistance to tearing, bending and tensile strength (Mujtaba et al., 2022). These requirements are best met by plastics (Ong et al., 2022), i.e. polymers such as polyvinyl chloride (PVC), polyethylene terephthalate (PET), polypropylene (PP), PE and polyamide (PA) (Jain & Tiwari, 2015; Mujtaba et al., 2022). The disadvantages of their use are multiple and consist of non-degradability, persistence in the environment for a very long time, emission of greenhouse gases such as carbon dioxide and methane and difficulty of recycling (Shen et al., 2020).

There is a need to replace these materials with more environmentally friendly formulas in order to reduce the environmental impact. This is the reason why biopolymers are more and more intense studied as

materials in the packaging industry. Some of them (polycaprolactones, poly-ester-amides, polyglycols and polybutene derivatives with succinic and adipic acids) are obtained from non-renewable sources and others from renewable ones. The latter ones are classified into agrobiopolymers (carbohydrates, lipids, protein), chemically synthesized biopolymers (polylactic acid, polyvinyl alcohol) and biopolymers synthesized by microbial mechanism (curdlan, fucopol, gellan, pullulan, polyhydroxyalkanoates, polyhydroxybutyrates, xanthan). Resins, waxes, fatty acids, acylglycerols and vegetable oils are lipids and proteins that can have vegetable (gluten, soy and zein protein) or animal (casein, collagen, gelatin and whey protein) origin (Mujtaba et al., 2022). The use of biopolymers as packaging materials has advantages and disadvantages. They are biodegradable, non-toxic and biocompatible (Reichert et al., 2020) and moreover, they have low barrier properties and mechanical strength (Moeini et al., 2021).

PVA is a non-toxic, odorless and tasteless polymer with promising

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<https://doi.org/10.1016/j.fochx.2024.102053>

Received 29 October 2024; Received in revised form 22 November 2024; Accepted 28 November 2024

Available online 30 November 2024

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applicability in many industry sectors (Bellelli et al., 2018), prepared by hydrolysis of polyvinylacetate or other vinyl polymers with formiate or chloroacetate groups, when transesterification in basic catalysis with ethanol occurs (Hallensleben, 2000). PVA is water-soluble, biodegradable and has very good ability to form hydrophilic film (Ulaganathan et al., 2022). These properties recommend it as a food packaging material, but there is a need to improve its barrier properties, especially WVP. Thus, it can be modified with chitosan and anthocyanins (Pereira Jr. et al., 2015), cellulose nanocrystals (Ulaganathan et al., 2022; Van Nguyen & Lee, 2022), polyvinylpyrrolidone (Franca et al., 2022), starch, propolis, gliadin and anthocyanins (Mustafa et al., 2020; Phattarateera et al., 2023; Zheng et al., 2023), zeolites (Youssef et al., 2019) in order to overcome these challenges. PVA enrichment aimed to increase its antimicrobial activity (Mustafa et al., 2020; Pereira Jr. et al., 2015; Zheng et al., 2023) or to improve its barrier properties (Cazón et al., 2018; Fortunati et al., 2016; Jahana et al., 2016; Phattarateera et al., 2023; Ulaganathan et al., 2022).

However, the use of lipid-based compounds such as petroleum-based or organic waxes and vegetal oils as modifying agents is highly effective in reducing the WVP due to their significant hydrophobic properties. Beeswax is one of the most effective hydrophobic compound to reduce the water sensitivity of hydrophilic films (Liu et al., 2022). Beeswax is edible, has negligible toxicity and is approved for food use (E901) (Scientific Opinion 2007). Beeswax is a mixture of palmitate, palmitoleate and oleate esters with of long chain aliphatic alcohols (Liu et al., 2022).

Because the polyols are very efficient for plasticizing hydrophilic polymers and due to their role as crosslinking agents, glycerin has been widely included in polymeric films in order to increase the flexibility of the films (Cazón et al., 2018), but it has also a negative effect, namely it increases the film permeability to water vapors, because is a highly polar molecule with a strong attraction to water, promoting entrapment of large amount of water molecules inside the polymer network (Cazón et al., 2018). However, the positive role of glycerin, namely the increase of the film flexibility is more important to the film properties than the disadvantage given by the hydrophilicity. The literature data report about the preparation of the cellulose-glycerol-polyvinyl alcohol composite films for food packaging and shows that glycerol produced an UV protective effect, thus preventing the lipid oxidation (Cazón et al., 2018).

The literature data also report the significant role of the thermal curing in improving the barrier properties of the polymeric films. The heat treatment of PVA determines a many-fold gain in resistance to water absorbance compared to virgin PVA, and such improvement it was attributed to the elimination of water and to the alignment of polymeric chains, with a consequent loss of polar nature (Bellelli et al., 2018).

This study aims to develop a PVA-based composite material enriched with beeswax and heat treated that could replace polyethylene or other petroleum-originated plastics currently in use as food packaging material.

Bread was used as food which the packaging capacity of the film developed in the article was tested on, because it is a food that dries quickly becoming inappropriate for consumption, so the degree of permeability for water vapors of the packaging must be as low as possible.

## 2. Materials and methods

### 2.1. Materials

PVA was purchased from Merck, Germany. Beeswax was purchased from a local bee shop from Baia Mare, Romania. Glycerin was supplied by Chemical Company, Romania. Cocamidopropyl betaine extracted from coconut was used as surfactant and was supplied from Mayam, Romania. Acetic acid and ethanol were purchased from Chemical Company Romania. Polyethylene film used as a reference was purchased

from the Kaufland supermarket from Baia Mare. The bread was purchased from a local bakery in Baia Mare, Romania.

### 2.2. Beeswax-PVA composite films preparation

A series of 18 types of beeswax-PVA composite films was prepared that differ by beeswax content, glycerin and heat treatment (Table 1). The procedure is described in **Supplementary material – A.1.**

### 2.3. Beeswax-PVA composite films characterization

Morphology, elemental analysis, structure, differential thermal analyses (DTA) and thermogravimetry (TGA), physical-chemical properties, biodegradability, testing the most efficient material as bread packaging and statistical processing are detailed in **Supplementary material – A.2.**

## 3. Results and discussion

### 3.1. Morphology and elemental analysis of beeswax-PVA composite films

According to the macroscopic images, the virgin PVA films were transparent and colorless, they appears slightly elastic and have good bending resistance when they are stretched (Table S1 **from Supplementary material**). The same results regarding the PVA films were obtained by Jayakumar et al. (2019). The PVA film developed by them was flexible, transparent and could be easily and clearly peeled off from the Petri plate.

The PVA films turned yellow as the beeswax content increased, but their elasticity and resistance to bending were preserved. Up to a content of 0.8 g beeswax, heat-treated films do not differ from non-heat-treated ones in terms of color and appearance. Still at a content higher than 0.8 g beeswax, heat-treated films are more opaque and less elastic than the non-heat treated ones, but retain their resistance to bending (Table S1 **from Supplementary material**). PVA 2 G film is more transparent than PVA 2 no G, so the presence of glycerin reduces the opacity of the untreated film.

In the case of heat-treated films, if PVA 2 G tt is compared with PVA 2 no G tt it is observed that glycerin does not influence the appearance and color of the film. Jayakumar et al. (2019) also showed that the opacity of the PVA films increased in the presence of different additives like starch, ZnO nanoparticles and jamun extract.

Optical and SEM microscopy images show that PVA 0 is a homogeneous network and by heat treatment the network densifies (Table S1 **from Supplementary material**). The presence of beeswax is evidenced by the presence of yellowish areas homogeneously distributed in the PVA network and it is noticed that the yellow color intensity rises with the wax content.

**Table S1 from Supplementary material** also includes SEM images of four types from the prepared PVA-based films. SEM image of sample PVA 0 tt (not included) was similar with that of PVA 0 and those of the other types of PVA-based films (not included) were similar with those of PVA 2 G tt, PVA 2 no G and PVA 2 no G tt. The SEM images show that the beeswax is both impregnated and coated on the PVA network, leaving PVA meshes and that the beeswax distribution on the surface is homogeneous. Comparing the SEM images of PVA 2 no G and PVA 2 no G tt samples, it is noticed that the wax layer on the surface is reduced by heat treatment, behavior explained by its interpenetration into the PVA network. Jenno et al. (2022) developed beeswax modified with pine-needle extract and its morphological structure showed a yellowish, flaky and layered appearance. Similar layered morphological patterns of the beeswax were observed also by Espolov et al. (2014) and Huang et al. (2020).

Carbon is in the highest content in the beeswax-PVA based films and oxygen is also present but its content is approximately half of that of carbon. A small amount of sodium and chloride also appears in the PVA-

**Table 1**  
Preparation conditions for the investigated beeswax-PVA composite films.

Sample code	PVA (g)	Surfactant cocamidopropyl betaine (10 g/L) (mL)	Beeswax (g)	Glycerin (1 g)	Thermal treatment at 125 °C, 30 min	Thickness (mm)
PVA 0	1.25	12.5	–	Yes	No	0.12 ± 0.0057
PVA 0 tt	1.25	12.5	–	Yes	Yes	0.12 ± 0.0051
PVA 0.4 no G	1.25	12.5	0.4	No	No	0.22 ± 0.0025
PVA 0.4 no G tt	1.25	12.5	0.4	No	Yes	0.22 ± 0.0012
PVA 0.4 G	1.25	12.5	0.4	Yes	No	0.12 ± 0.0034
PVA 0.4 G tt	1.25	12.5	0.4	Yes	Yes	0.12 ± 0.0042
PVA 0.6 G	1.25	12.5	0.6	Yes	No	0.24 ± 0.0052
PVA 0.6 G tt	1.25	12.5	0.6	Yes	Yes	0.12 ± 0.0042
PVA 0.8 G	1.25	12.5	0.8	Yes	No	0.24 ± 0.0024
PVA 0.8 G tt	1.25	12.5	0.8	Yes	Yes	0.24 ± 0.0053
PVA 1 G	1.25	12.5	1	Yes	No	0.16 ± 0.0024
PVA 1 G tt	1.25	12.5	1	Yes	Yes	0.16 ± 0.0062
PVA 1 no G	1.25	12.5	1	No	No	0.12 ± 0.0012
PVA 1 no G tt	1.25	12.5	1	No	Yes	0.16 ± 0.0024
PVA 2 G	1.25	12.5	2	Yes	No	0.15 ± 0.0034
PVA 2 G tt	1.25	12.5	2	Yes	Yes	0.05 ± 0.0027
PVA 2 no G	1.25	12.5	2	No	No	0.15 ± 0.0031
PVA 2 no G tt	1.25	12.5	2	No	Yes	0.05 ± 0.0032

based films probably as impurity from the surfactant solution. EDXS and XPS analysis showed reduced amounts of oxygen, calcium, magnesium and silicon in addition to carbon which is higher than 98 % in PE, probably from the production process (*Table S2 from Supplementary material*).

### 3.2. Structure of the beeswax-PVA composite films

The XRD patterns of the beeswax-PVA films are presented in *Fig. S1 from Supplementary Material*. X-ray diffraction is a useful tool to establish the crystal lattice arrangements of beeswax-PVA composite films. PVA exhibited the major crystalline peak at Bragg angles  $2\theta = 19.72^\circ$  in agreement with the literature values (Feng et al., 2024). Two strong and sharp XRD peaks at Bragg angles  $2\theta = 22.24^\circ$  and  $24.24^\circ$  are observed as a result of the orthorhombic structure presence of hydrocarbon/monoester fractions of beeswax (Liu et al., 2022). The peak intensity at Bragg  $2\theta = 19.72^\circ$  corresponding to PVA is higher in the samples with beeswax than in unchanged PVA, which shows the interconnection of the PVA network with that of beeswax and the formation of a crystalline composite structure.

By comparing the XRD pattern of PVA 2 no G with that of PVA 2 no G tt, it can be noticed that the intensity of the signals increases by thermal treatment showing an enhanced crystallization degree of the material. The same behavior was observed also in the case of PVA 2G and PVA 2 G tt. The PE film exhibits also a crystalline nature. The diffraction peaks of PE are observed at Bragg angles  $2\theta = 19.08^\circ$  and  $23.92^\circ$  corresponding to the Miller indices of PE (120) and (112) atomic plane, respectively. However, the third peak at  $28.66^\circ$  is ascribed to the second order of the former peak (240) (Telfah et al., 2022).

*Fig. 1* illustrates the FTIR spectra of the beeswax-PVA composite films. The FTIR spectra of PVA 0 and PVA 0 tt contain signals at  $3329\text{ cm}^{-1}$  corresponding to the vibration of OH groups, at 2931, 2863, 1339, 1240 and  $851\text{ cm}^{-1}$  due to the vibration of methylene groups, at  $1650\text{ cm}^{-1}$  assigned to the C=O group vibration, at  $1425\text{ cm}^{-1}$  attributed to vibration of simple C—H bonds and in the region  $1102\text{--}919\text{ cm}^{-1}$  due to vibration of C—O saturated bonds (Franca et al., 2022). There are no differences in PVA 0 and PVA 0 tt spectra, as a result of heat treatment, which shows that by treatment at  $125\text{ }^\circ\text{C}$  for 30 min, the PVA network does not degrade. The same signals appear in the FTIR spectra of PVA 2 no G and PVA 2 no G tt, but at a lower intensity than in neat PVA. This is explained by a lower vibration of PVA-specific bonds due to their hardening as a result of the connection with molecules of esters, hydrocarbons, alcohols and fatty acids in the composition of beeswax (Tulloch, 1980). By comparing the FTIR spectra of PVA 2 no G and PVA 2 no G tt with that of beeswax it can be observed commonly peaks at  $1735\text{ cm}^{-1}$  assigned to the vibration of C—O bonds, in the region of

$1174\text{--}1416\text{ cm}^{-1}$  and at  $719\text{ cm}^{-1}$  due to the vibration of the C=O bond in carboxyl groups from free fatty acids (Zuber et al., 2021, Josef-Leenose-Helen et al. 2022). However, in the FTIR spectra of PVA 2 no G and PVA 2 no G tt respectively, new peaks appear, at 1632, 1093 and  $837\text{ cm}^{-1}$ . These are absent in the FTIR spectra of neat PVA and of beeswax and are assigned to the vibration of new bonds generated by esterification between the carboxyl groups from beeswax's fatty acids and the PVA's hydroxyl groups and by etherification between the hydroxyl groups of PVA and of polyols from beeswax (Tulloch, 1980). This shows the composite structure formation and agrees with the XRD results. The difference between the FTIR spectrum of PVA 2 no G and that of PVA 2 no G tt occurs only at  $3329\text{ cm}^{-1}$  and consists of a weaker signal in the heat treated sample, explained by the elimination of water as a result of heat treatment. As in the case of PVA 0, FTIR analysis proved that the structure of PVA 2 no G has not been degraded by heat treatment, but has only compacted.

The XPS curves are observed in *Figs. S2a) and S2b) from Supplementary material*. C1s can be divided into four peaks, C—C inter-connected ( $283\text{--}283.5\text{ eV}$ ), C—H/C—H ( $284.6\text{ eV}$ ), C—O—H/C—O—C ( $286.2\text{--}286.5\text{ eV}$ ) and C=O/O—C=O ( $288.1\text{--}288.6\text{ eV}$ ). The binding energy (BE) of C1s increased by wax adding and by thermal treatment (*Fig. S2a) from Supplementary material*), disclosing that the density and distribution of electron cloud around carbon changed due to the intermolecular interaction (Xu et al., 2023). The signal from 283 to  $283.5\text{ eV}$  in the XPS spectra of waxed PVA samples, which is absent in PVA 0, due to the presence of C—C bonds between the PVA lattice and beeswax components. The share of the bond decreases by heat treatment, but of these, the largest share occurs in PVA 2 no G tt (*Fig. S2b) from Supplementary material*). This sample shows the best water vapor barrier properties (*Table 2*). The presence of beeswax is proven by the higher share of C—C and C—H bonds from  $284.6\text{ eV}$  in wax samples compared to PVA 0 (*Fig. S2b-A) from Supplementary material*). In addition, O1s can be divided into three peaks, C=O ( $531.2\text{--}532\text{ eV}$ ), C—O—C ( $531.6\text{--}532.5\text{ eV}$ ) and C—O—H ( $532.8\text{--}533.3\text{ eV}$ ) (*Fig. S2b-B) from Supplementary material*). The binding energy of O1s increased after adding the beeswax and by thermal treatment (*Fig. S2a) from Supplementary material*), including the increase of C—O—H/C—O—C binding energy together with the reduction of the C=O binding energy (*Fig. S2b) from Supplementary material*). The heat treatment decreases the share of C=O bonds ( $531.3\text{ eV}$ ) and of C—O—H bonds ( $533\text{ eV}$ ) and increases that of C—O—C bonds ( $288.1\text{--}288.6\text{ eV}$ ). This spectral behavior indicates that C=O and C—O—H bonds were consumed and C—O—C bonds were generated by interconnection of O—H groups from PVA and carboxyl groups from long chain acids from beeswax. This also explains that the impregnation of PVA with beeswax improves the hydrophobicity of PVA composite films by increasing the hydrophobic waxes

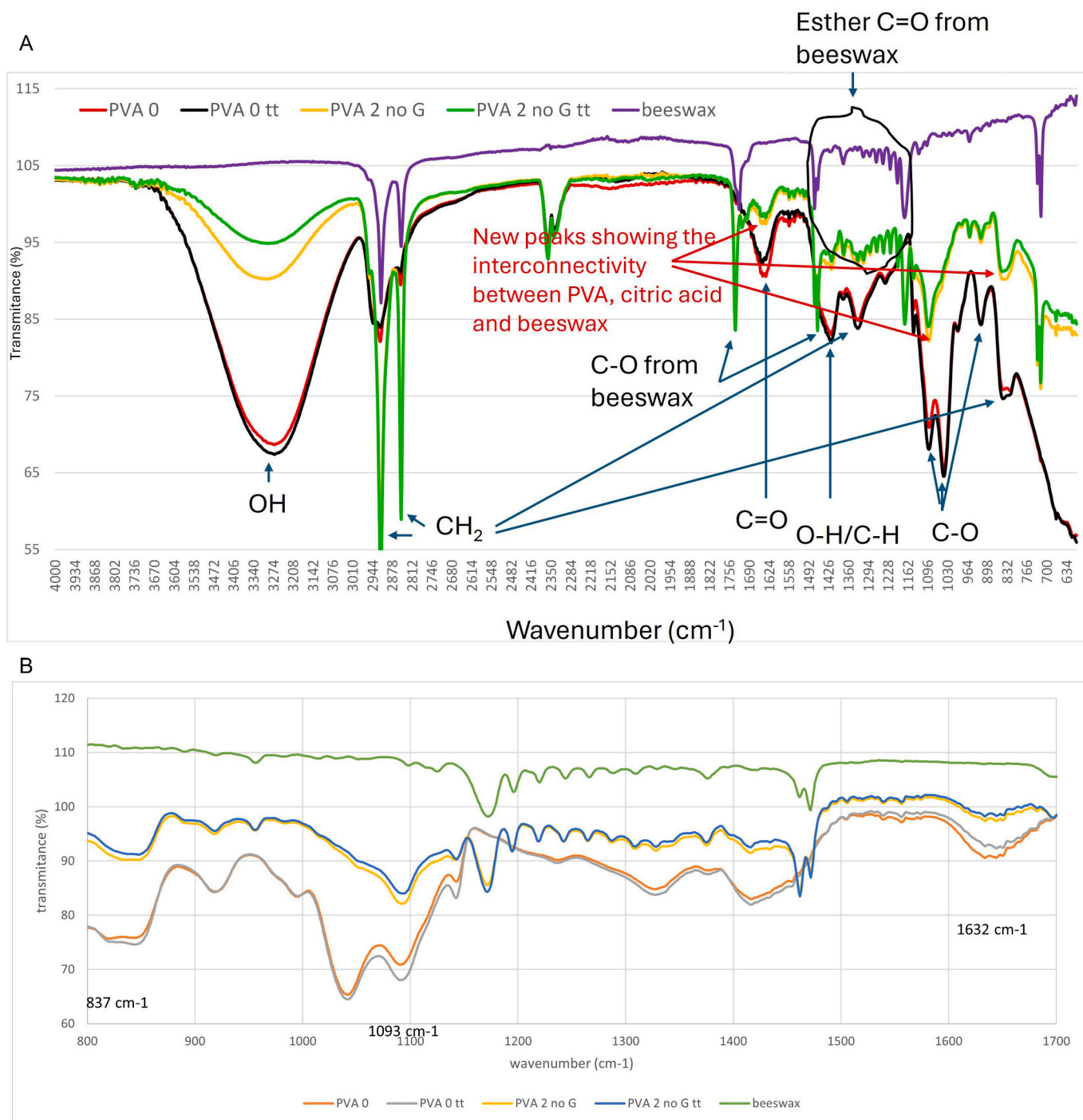


Fig. 1. a) FTIR of the beeswax-PVA composite films; b) Zoom on the region 1700–800  $\text{cm}^{-1}$ .

inside and decreasing the hydrophilic groups in PVA, which synergistically act physically and chemically to increase the hydrophobicity.

### 3.3. Differential thermal analyses (DTA) and Thermogravimetry (TGA) of the beeswax-PVA composite films

Thermal curves resulting from simultaneous DTA/TGA measurements for all samples are shown in **Fig. S3 from Supplementary material**. From the differential thermal and thermogravimetric curves obtained on the beeswax-PVA composite films (**Figs. S3a) and S3b) from Supplementary material**), were determined the water loss due to hydrophilic nature of PVA, and thermal decomposition of the samples

during their heating up to 800 °C. Both samples PVA 0 and PVA 2 G tt, show a small mass loss of 3 and 4 % up to 140 °C. After this temperature, PVA 0 (**Fig. S3a) from Supplementary material**) shows two associated endothermic events in the melting TG curve. After 220 °C, the sample decomposes in 4 steps with a total mass loss of nearly 92 %. The TGA curve for the PVA 2 G tt sample (**Fig. S3b) from Supplementary material**) does not clearly highlight melting, although two endothermic signals are observed in the DTA curve, with the maximum temperature at appropriate values to those of PVA 0; this may be due to the simultaneous production of melting and decomposition, which occurs in 5 stages, resulting in a total mass loss of 96.7 %. The thermal profiles of the PVA 2 no G tt and PVA 2 no G (**Figs. S3c) and S3d) from Supplementary**

**Table 2**  
Physical-chemical parameters of beeswax-PVA composite films.

Sample *	WVP x 10 <sup>10</sup> (g/s m Pa)**	Solubility (%) in simulant **			Tensile strength (N/mm <sup>3</sup> ) **	Bending resistance (no) **	Opacity (a.u./ mm) **	Swelling capacity (%)**	Biodegradability (%)**	
		A***	B ***	D1***					30 days	60 days
PVA 0	5.31 ± 0.23 <sup>gh</sup>	73.6 ± 2.51 <sup>a</sup>	75.7 ± 0.86 <sup>a</sup>	52.8 ± 2.42 <sup>a</sup>	0.57 ± 0.02 <sup>c</sup>	216 ± 2.99 <sup>b</sup>	0.74 ± 0.04 <sup>l</sup>	10,000 ± 109.36 <sup>a</sup>	50.3 ± 0.01 <sup>cde</sup>	100 ± 3.01 <sup>a</sup>
PVA 0 tt	5.76 ± 0.2 <sup>fg</sup>	42.3 ± 2.06 <sup>d</sup>	41.4 ± 1.81 <sup>e</sup>	40.1 ± 0.31 <sup>de</sup>	0.92 ± 0.01 <sup>a</sup>	215 ± 8.93 <sup>b</sup>	0.92 ± 0.01 <sup>i</sup>	1010 ± 21.9 <sup>b</sup>	41.9 ± 0.34 <sup>f</sup>	100 ± 3.01 <sup>a</sup>
PVA 0.4 no G	8.74 ± 0.16 <sup>b</sup>	55.6 ± 2.3 <sup>c</sup>	67.5 ± 3.34 <sup>b</sup>	35.4 ± 1.32 <sup>f</sup>	0.48 ± 0.01 <sup>d</sup>	202 ± 2.91 <sup>b</sup>	1.12 ± 0 <sup>j</sup>	950 ± 10.59 <sup>bc</sup>	52.4 ± 1 <sup>bcd</sup>	100 ± 3.01 <sup>a</sup>
PVA 0.4 no G tt	9.08 ± 0.02 <sup>b</sup>	31.8 ± 1.06 <sup>g</sup>	32.4 ± 1.52 <sup>gh</sup>	29.5 ± 0.19 <sup>g</sup>	0.75 ± 0.02 <sup>b</sup>	204 ± 6.12 <sup>b</sup>	3.52 ± 0.17 <sup>g</sup>	804 ± 6.5 <sup>efg</sup>	43.5 ± 0.42 <sup>f</sup>	100 ± 3.01 <sup>a</sup>
PVA 0.4 G	5.66 ± 0.21 <sup>fg</sup>	66.4 ± 2.22 <sup>b</sup>	65.2 ± 0.84 <sup>b</sup>	46.4 ± 1.59 <sup>b</sup>	0.3 ± 0 <sup>i</sup>	210 ± 2.46 <sup>b</sup>	0.81 ± 0.01 <sup>j</sup>	920 ± 41.1 <sup>bc</sup>	49.8 ± 0.4 <sup>cde</sup>	100 ± 3.01 <sup>a</sup>
PVA 0,4 G tt	6.04 ± 0.06 <sup>ef</sup>	35.8 ± 1.3 <sup>efg</sup>	39.2 ± 0.45 <sup>ef</sup>	42.1 ± 1.91 <sup>cd</sup>	0.43 ± 0.01 <sup>e</sup>	211 ± 5.49 <sup>b</sup>	1.75 ± 0.05 <sup>i</sup>	789 ± 6.02 <sup>efg</sup>	40.9 ± 0.34 <sup>fg</sup>	100 ± 3.01 <sup>a</sup>
PVA 0,6 G	10.3 ± 0.32 <sup>a</sup>	55.7 ± 0.84 <sup>c</sup>	53.2 ± 0.6 <sup>d</sup>	44.1 ± 0.51 <sup>bc</sup>	0.37 ± 0 <sup>gh</sup>	210 ± 2.46 <sup>b</sup>	1.15 ± 0.03 <sup>j</sup>	912 ± 29.38 <sup>cd</sup>	51.5 ± 0.81 <sup>cd</sup>	100 ± 3.01 <sup>a</sup>
PVA 0,6 G tt	4.93 ± 0.06 <sup>h</sup>	36.1 ± 1.12 <sup>efg</sup>	34.1 ± 0.32 <sup>gh</sup>	41.6 ± 2.04 <sup>cd</sup>	0.5 ± 0.01 <sup>d</sup>	205 ± 6.84 <sup>b</sup>	2.16 ± 0 <sup>i</sup>	711 ± 15.38 <sup>gh</sup>	42.7 ± 0.98 <sup>f</sup>	100 ± 3.01 <sup>a</sup>
PVA 0,8 G	10.4 ± 0.32 <sup>a</sup>	56.6 ± 1.26 <sup>c</sup>	49.3 ± 1.14 <sup>d</sup>	36.4 ± 1.6 <sup>ef</sup>	0.39 ± .01 <sup>fg</sup>	201 ± 3.39 <sup>b</sup>	2.89 ± 0.09 <sup>b</sup>	875 ± 5.05 <sup>cde</sup>	52 ± 0.32 <sup>cd</sup>	100 ± 3.01 <sup>a</sup>
PVA 0,8 G tt	10 ± 0.14 <sup>a</sup>	36.2 ± 0.34 <sup>ef</sup>	32.4 ± 1.52 <sup>gh</sup>	38.3 ± 1.52 <sup>def</sup>	0.6 ± 0.01 <sup>c</sup>	204 ± 6.12 <sup>b</sup>	3.56 ± 0.14 <sup>g</sup>	642 ± 29.55 <sup>hi</sup>	41.3 ± 0.71 <sup>f</sup>	100 ± 3.01 <sup>a</sup>
PVA 1 G	7.2 ± 0.1 <sup>c</sup>	41.8 ± 2.04 <sup>d</sup>	38.6 ± 1.62 <sup>ef</sup>	25.4 ± 1.14 <sup>h</sup>	0.34 ± 0.01 <sup>h</sup>	202 ± 2.91 <sup>b</sup>	3.75 ± 0.03 <sup>g</sup>	821 ± 11.46 <sup>def</sup>	52.5 ± 0.12 <sup>bc</sup>	100 ± 3.01 <sup>a</sup>
PVA 1 G tt	7.2 ± 0.1 <sup>c</sup>	32.7 ± 0.74 <sup>fg</sup>	31.8 ± 0.55 <sup>gh</sup>	39.4 ± 1.77 <sup>de</sup>	0.3 ± 0 <sup>i</sup>	203 ± 9.53 <sup>b</sup>	5.48 ± 0 <sup>e</sup>	554 ± 25.08 <sup>ij</sup>	47.2 ± 0.58 <sup>e</sup>	100 ± 3.01 <sup>a</sup>
PVA 1 no G	6.3 ± 0.05 <sup>e</sup>	42.5 ± 1.59 <sup>d</sup>	58.7 ± 2.25 <sup>c</sup>	22.1 ± 0.68 <sup>h</sup>	0.42 ± 0.01 <sup>ef</sup>	209 ± 3.92 <sup>b</sup>	4.25 ± 0.21 <sup>f</sup>	742 ± 1.48 <sup>fg</sup>	55.7 ± 2.05 <sup>b</sup>	100 ± 3.01 <sup>a</sup>
PVA 1 no G tt	8.74 ± 0.16 <sup>b</sup>	24.5 ± 0.39 <sup>h</sup>	24.6 ± 0.78 <sup>i</sup>	15.4 ± 0.01 <sup>ij</sup>	0.42 ± 0.01 <sup>ef</sup>	204 ± 6.12 <sup>b</sup>	10.98 ± 0.16 <sup>b</sup>	521 ± 21.31 <sup>jk</sup>	49.1 ± 1.55 <sup>de</sup>	100 ± 3.01 <sup>a</sup>
PVA 2 G	6.9 ± 0.23 <sup>cd</sup>	37.2 ± 1.18 <sup>e</sup>	35.5 ± 0.12 <sup>fg</sup>	23.4 ± 0.43 <sup>h</sup>	0.21 ± 0 <sup>j</sup>	202 ± 2.91 <sup>b</sup>	5.44 ± 0.07 <sup>e</sup>	524 ± 3.6 <sup>jk</sup>	67 ± 2.25 <sup>a</sup>	100 ± 3.01 <sup>a</sup>
PVA 2 G tt	1.7 ± 0.05 <sup>i</sup>	26.5 ± 1.09 <sup>h</sup>	30.1 ± 1.38 <sup>h</sup>	18.1 ± 0.46 <sup>i</sup>	0.15 ± 0.01 <sup>k</sup>	201 ± 3.39 <sup>b</sup>	7.92 ± 0.22 <sup>c</sup>	433 ± 12.49 <sup>kl</sup>	48.1 ± 1.55 <sup>e</sup>	100 ± 3.01 <sup>a</sup>
PVA 2 no G	6.5 ± 0.14 <sup>de</sup>	39.8 ± 0.51 <sup>de</sup>	52.3 ± 1.75 <sup>d</sup>	14.1 ± 0.6 <sup>j</sup>	0.36 ± 0 <sup>gh</sup>	204 ± 6.12 <sup>b</sup>	7 ± 0.3 <sup>d</sup>	498 ± 21.21 <sup>jk</sup>	52.9 ± 1.95 <sup>bc</sup>	100 ± 3.01 <sup>a</sup>
PVA 2 no G tt	1.5 ± 0.03 <sup>i</sup>	15.1 ± 0.18 <sup>i</sup>	15.2 ± 0.43 <sup>j</sup>	7.2 ± 0.35 <sup>k</sup>	0.27 ± 0 <sup>i</sup>	202 ± 2.91 <sup>b</sup>	14.4 ± 0.34 <sup>a</sup>	465 ± 12.55 <sup>kl</sup>	37.9 ± 0.7 <sup>g</sup>	100 ± 3.01 <sup>a</sup>
PE	0.2 ± 0.01 <sup>j</sup>	0 ± 0 <sup>j</sup>	0 ± 0 <sup>k</sup>	6.2 ± 0.08 <sup>k</sup>	0.5 ± 0.01 <sup>d</sup>	555 ± 13.24 <sup>a</sup>	2.15 ± 0.09 <sup>i</sup>	381 ± 7.99 <sup>j</sup>	2.8 ± 0.04 <sup>h</sup>	3.7 ± 0.16 <sup>b</sup>

\* Sample coding is given in Table S1 from Supplemental material.

\*\* Same letters means statistical nonsignificant difference according to Anova One Way, Tukey model.

\*\*\* Simulant A – ethanol solution 10 %, simulant B - acetic acid 3 %, simulant D1- ethanol 50 %.

**material**), look similar: the events in the DTA curve occur at values close to the temperature. The mass loss due to the dehydration of the samples occurs up to 140 °C, and is very little of 1 %. Melting takes place between 140 and 220 °C, as in the samples discussed above, and the decomposition occurs in 4 stages, with mass losses in the TG curve showing 96 % for PVA 2 no G tt and 93 % for PVA 2 no G.

In the case of the PE sample (**Fig. S3e**) from **Supplementary material**), no dehydration of the sample is observed, and the endothermic signal from ~73 °C is not identified as in the other PVA samples, demonstrating that the presence of glycerin reduces the degree of dehydration, a finding consistent with the results of **Cazón et al. (2018)**. Between 140 and 250 °C, a broad signal is identified with a maximum of ~222 °C, associated in all samples with the melting of the substance. After ~260 °C, the sample starts to lose mass due to the evaporation process of glycerol - the endothermic peak with the maximum at 289 °C, recording in the TGA curve a loss of 3.7 %.

The peak at 301 °C in the DTA profile of the PVA 0 sample, at 347 °C in that of PVA 2 G tt, at 342 °C of the PVA 2 no G tt sample profile, 337 °C of PVA 2 no G sample and at 395 °C of PVA 2 G sample is due to the PVA dehydroxylation (**Pereira Jr. et al., 2015**) and beeswax decomposition (**Namdar et al., 2009**) and demonstrates that modification of PVA with beeswax and the thermal treatment improved the thermal stability and the PVA's dehydroxylation process was slowed down. The presence of glycerin delayed dehydroxylation, which occurred at a higher

temperature in comparison with the samples without glycerin. The exothermic peak at 449 °C from DTA profile of PVA 0, at 461 °C in PVA 2 G tt, at 464 °C and 478 °C in PVA 2 no G tt and at 415 °C and 475 °C in PVA 2 no G is due to oxidation of organic material in air without forming a residue (**Pereira Jr. et al., 2015**).

### 3.4. Physical-chemical characterization of the beeswax-PVA composite films

Enhanced WVP is essential for packaging materials to protect the packaged foodstuffs (**Nguyen & Lee, 2022**). According to **Table 2**, WVP raised by adding beeswax from 0.4 g to 0.8 g and decreased in samples containing 1 g and 2 g beeswax, respectively, probably due to inadequate homogenization of the film-forming solution. The results are in agreement with the findings of **Farahnour et al. (2024)** who prepared mycelium-based composites containing coconut oil and beeswax and revealed that the highest tested composition of beeswax (80 g beeswax with 20 g coconut oil) leads to the lowest water absorption ability, due to the beeswax's water-repellent properties.

By comparing the non-heat-treated films, WVP is higher in the case of glycerin-containing films than for those without glycerin, due to the weak polar character of glycerin, but in the case of heat-treated glycerin including samples, the behavior is reversed, explained by the glycerin degradation and the formation of much denser hydrophobic networks.

The glycerin presence does not influence the WVP of the samples containing the maximum amount of beeswax (2 g) (samples PVA 2 G, PVA 2 G tt, PVA 2 no G and PVA 2 no G tt), explained by the fact that the ratio between beeswax and glycerin is high and the hydrophobicity is predominantly. [Cazón et al. \(2018\)](#) prepared PVA-based films modified with cellulose nanocrystals and glycerin and they have proved that the water adsorption increases with the glycerol content. Their conclusion can be confirmed in our study only for low amounts of beeswax (lower than 2 g). The heat treatment decreases WVP only to PVA 2, because condensation reactions (esterification, etherification) occur and reorganize and compact the film network. In the case of samples with maximum beeswax content, both heat-treated and not heat-treated, the presence of glycerin does not influence WVP, explained by the fact that the hydrophobic phase is predominant and imprints this character to the entire film. By modifying PVA with beeswax and thermal treatment at 125 °C for 30 min, the WVP was reduced by 74 %. Sample PVA 2 no G tt film has WVP only 7.5 times higher than PE (considered nonpermeable material to water vapors), while WVP of unchanged PVA is almost 29 times larger than PE. [Bellelli et al. \(2018\)](#) showed that water adsorption capacity of PVA-based films modified with citric acid and malic acid as surfactants decreases with the curing temperature as a result of water removal, alignment and crosslinking of the polymeric chains. [Mustafa et al. \(2020\)](#) who developed PVA-based films modified with starch, propolis and rosemmary antocyanins showed that cross-linker, polymers, plasticizers and intermolecular interactions mainly influence this property. The WVP decreases while propolis extract rises, attributed to hydrogen bonding between polyphenols from propolis extract and the PVA-starch matrix. [Fortunati et al. \(2016\)](#) obtained PVA films modified with hydroxytyrosol methyl (HTyr-MC) carbonate and showed that their water absorption capacity decrease as the content of HTyr-MC rises, explained by the fact that HTyr-MC acts as nucleation agent for the PVA matrix and influences the diffusion processes of the water through the polymer. [Nguyen and Lee \(2022\)](#) showed that the incorporation of cellulose nanocrystals into the PVA matrix significantly enhanced the water vapor barrier properties, by 30.3 %, due to the crosslinking that limited the transport of water vapor through the film. [Yu et al. \(2018\)](#) observed that the adding 0.6 % silica reduced by at least 0.1 times the WVP of PVA/chitosan biodegradable films, preventing moisture transfer. [Zheng et al. \(2023\)](#) showed that the WVP of PVA decreased with the increased replacement of octenyl succinic anhydride (OSA)/starch/gliadin, because OSA enhanced the hydrophobicity of the polymers.

The barrier to grease is one of the most important property of the film when is used to pack certain food products. No sign of migration was observed with castor oil, demonstrating the high resistance of these materials to grease.

The solubility in all three simulants decreases with increasing the beeswax mass, due to its hydrophobic character and by applying heat treatment, due to compaction of the film network ([Table 2](#)). These results are in agreement with those obtained in [Table 2](#), namely, solubility in food simulants is directly influenced by WVP. The presence of glycerin increases the solubility in simulants of non-heat-treated films with low wax content, because the composition of the hydrophilic and hydrophobic phases is homogeneous. In contrast, with increasing the hydrophobic phase, solubility decreases. The presence of glycerin causes a significant increase in the solubility of heat-treated films regardless of the beeswax content, explained by the remaining of non-degraded hydrophilic portions that would give polar character to the film. In the case of PVA 2 no G tt sample showing the lowest WVP value, solubility in simulant A (ethanol solution 10 %) and simulant B (3 % acetic acid solution) is higher than in simulant D1 (ethanol solution 50 %), so the film would be more suitable for packaging fatty foods than aqueous and acidic ones. This formula is best suited for storing dry food. [Phattarateera et al. \(2023\)](#) also proved the water-soluble character of films based on PVA modified with starch. The solubility was much intense when hydrolyzed starch was used due to its lower molecular weight. [Haghighi et al. \(2020\)](#) showed that the influence of the ethyl lauroyl

arginate (ELA) incorporation on the moisture content of PVA-chitosan films was not significant ( $p < 0.05$ ), but increased their water solubility as a result of the hydrophilic nature of PVA-chitosan film and low oil-water equilibrium partition coefficient of ELA. [Fortunati et al. \(2016\)](#) showed that after 10 days in ethanol 10 %, significant differences in overall migration ( $P < 0.05$ ) between the PVA matrix and PVA/Hydroxytyrosol methyl carbonate (HTyr-MC) films, namely the migration level in the modified PVA film was lower than that in virgin PVA.

Tensile strength ([Table 2](#)) decreases by adding beeswax to films with glycerin regardless of whether heat treatment is applied or not. This would be explained by the severe agglomeration of the beeswax microcrystals on the PVA's network and by the fact that beeswax is soft and has weak tensile strength. The weakly resistant character of the beeswax is also imprinted on the film. The same behavior was observed by [Van Nguyen and Lee \(2022\)](#) in the case of PVA-based films modified with titanium dioxide and cellulose crystals. The mechanical resistance of the films increased as the TiO<sub>2</sub> content raised at 1 wt% explained by the change in the crystallinity of the composite and by formation of the H bonds and decreased with TiO<sub>2</sub> content higher than 10 wt% due to the agglomeration of titania particles. The presence of glycerin has a detrimental effect, as it reduces the mechanical resistance of both untreated and heat-treated films. Heat treatment increases the mechanical resistance of films without glycerin and containing little beeswax (up to 0.4 g) and those with glycerin and up to 0.8 g beeswax. This is in agreement with the findings of [Bellelli et al. \(2018\)](#) and [Yoon \(2014\)](#) who showed that the tensile strength of PVA-starch films improved by 65–295 % through heat curing. Moreover, the correlation coefficient between WVP and bending resistance is negative and significant in a percentage higher than 95 % ([Table 3](#)), showing that WVP varies inversely proportional with the bending resistance. This conclusion makes the films good candidates for food packaging.

The opacity is also included in [Table 3](#) and the correlation coefficient between WVP and opacity are calculated in [Table 3](#). For the materials designed for food packaging, it is better to develop a preventive effect on photo degradation of food during storage. The UV and visible light can initiate food oxidation which results in food spoilage. Opacity ([Table 2](#)) increases with wax content, regardless of the presence of glycerin. Glycerin-free films are more opaque than glycerin-free films. Heat treatment increases the opacity of the films. The correlation coefficient between opacity and WVP is negative and statistical significant, meaning that WVP decreases, induced by increase in the beeswax content, thermal treatment and glycerin, generated also a rise of the samples opacity. [Bellelli et al. \(2018\)](#) showed that thermal curing did not affect the color of pure PVOH film samples, but that of the films incorporating organic acids, probably due to a slight thermal degradation. They discussed that the yellowness of PVOH films should be considered as a side-effect of acid incorporation and heat curing. [Jayakumar et al. \(2019\)](#) proved the decrease of the films transmittance by addition of starch, ZnO nanoparticles, nutmeg oil and jamun extract.

The swelling degree is influenced by the cross-linked density of polymeric network including polymers, additives, cross-linkers, plasticizers and other compounds that can generate intermolecular linkage. The swelling capacity of untreated films was very high within 48 h ([Table 2](#)), while the SC of the heat-treated films was less pronounced. The results showed that the swelling capacity decreases with WVP ([Table 3](#)) and with wax content as a result of the hydrophobic character of the beeswax components and is lower in glycerin-free films compared to those containing glycerin. The results are in agreement with the findings of [Pereira Jr. et al. \(2015\)](#) and are explained by the crosslinking that induced two processes: 1) reduction in chain's mobility, thus hindering the water's molecules access; 2) formation of hydrogen bonds between the PVA and beeswax's hydroxyl groups and 3) hydroxyl groups of beeswax's components may cause a steric effect restricting the interaction of water in the polymer. [Musetti et al. \(2014\)](#) showed that the PVA-based film with poly-ethylene-glycol (PEG) displayed lower water content and degree of swelling if compared to neat PEG, because

**Table 3**

Correlation coefficients corresponding to the physical-chemical and biochemical characteristics of the beeswax-PVA composite films (p1-WVP, p2, p3 and p4-solubility in sim A, B and D1, respectively, p5-tensile strength, p6-bending resistance, p7-opacity, p8-swelling capacity, p9 and p10-biodegradability after 30 h and 60 h, respectively; \* means difference statistical significant >95 %; \*\* means difference statistical significant >99 % determined by applying the Student test).

	p1	p2	p3	p4	p5	p6	p7	p8	p9	p10
p1		0,5161*	0,4360	0,4669*	0,2144	-0,5217*	-0,2767	-0,0441	0,5506*	0,5191*
p2			0,9466**	0,7778**	0,0801	-0,5253*	-0,5606*	0,5375*	0,6213**	0,5511*
p3				0,6444**	0,0588	-0,5077*	-0,5021*	0,5090*	0,6329**	0,5335*
p4					0,3181	-0,3934	-0,7214**	0,4552	0,3006	0,4258
p5						0,1139	-0,4525	0,2175	-0,2987	-0,0847
p6							-0,1657	-0,0582	-0,8493**	-0,9983**
p7								0,0537	0,0537	0,1352
p8									0,1055	0,0911
p9										0,8455**
p10										

PVA chains are held together more tightly by interchain bridges formed by PEG, additionally to the low hydrophilicity of PEG. Mustafa et al. (2020) obtained PVA-based films modified with starch and propolis extract with high rigidity and less swelling degree in comparison with neat PVA.

3.5. Biodegradability of the beeswax-PVA composite films

The biodegradability results are included in Table 2 and the correlation coefficient between WVP and biodegradability is included in Table 3. After 30 days of preservation in soil, the biodegradability of PVA-based films was ~50 % higher than PE, considering that the biodegradability of PE was 2.8 %. The biodegradability of the PE film after 60 days was 3.7 %, while the PVA samples were completely destroyed. In the case of the non-heat-treated and heat-treated films with glycerin, the biodegradability increases with beeswax mass and in the case of those without glycerin, there was a slight decrease by adding hydrophobic agent. Biodegradability decreases with heat treatment. All PVA-based films were dissolved after 60 days of preservation in the soil. WVP is directly correlated with biodegradability according to the results

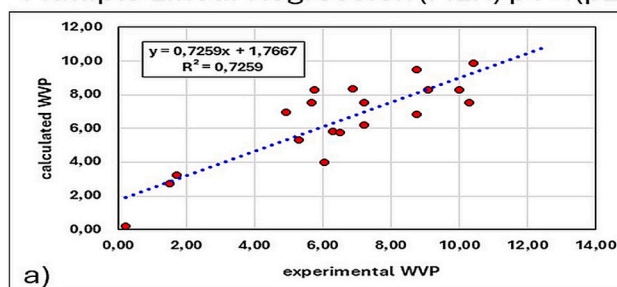
from Table 3, namely the WVP increase induced a variation in the same direction of the biodegradability.

Yu et al. (2018) showed that PVA films modified with chitosan can degrade quickly, namely they can disappear quickly themselves in soil due to the dissolution. Meanwhile, the presence of SiO<sub>2</sub> reduced the dissolution degree of PVA film by 24 % percent compared to the same film without SiO<sub>2</sub>. Zheng et al., 2023 tested the soil burial degradation of composite plastics based on PVA modified with starch and gliadin and consisted that many cavities and holes appeared after 56 days. The composite plastic's spots increased with the starch/gliadin ratio.

Moreover, the results in Table 2 were subjected to multiple linear and antegrade regression analysis (Fig. 2). The obtained results are correlated with those obtained in Table 2, i.e. with the decrease in WVP decreases the tensile strength and biodegradability, explained by formation of much denser hydrophobic network assigned by the presence of beeswax and the compaction generated during the thermal treatment.

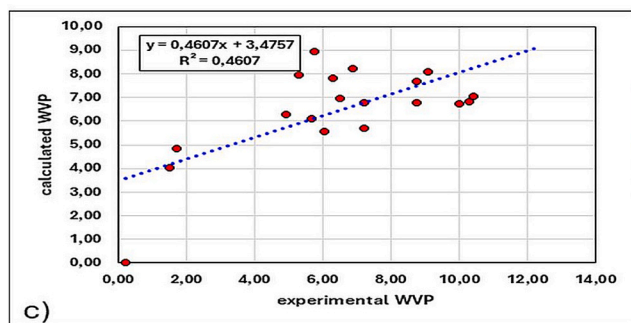
The PCA analysis (Fig. 3) highlights that the variability of the WVP data is majorly influenced by 6 components that explain 99.3037 % of the data variability. The first component explains 49.7967 % of the data variability, the second explains 26.8183 %, the third explains 9.7297 %.

Multiple Linear Regression (MLR) p1=f(p2-p10)



b)	beta	standard deviation error of beta	B	standard deviation error of B	t(9)	p-level	
intercept			146,6017	98,7845	1,4841	0,1719	ns
p2	0,4652	0,8447	0,0781	0,1418	0,5508	0,5952	ns
p3	0,0549	0,7323	0,0086	0,1149	0,0750	0,9418	ns
p4	0,9615	0,7088	0,2034	0,1499	1,3566	0,2079	ns
p5	0,7346	0,2871	11,6527	4,5539	2,5588	0,0307	p<0.05
p6	-7,4941	4,8346	-0,2721	0,1755	-1,5501	0,1555	ns
p7	0,7543	0,6514	0,5956	0,5144	1,1580	0,2767	ns
p8	-0,4687	0,2921	-0,0006	0,0004	-1,6048	0,1430	ns
p9	0,9576	0,5374	0,2240	0,1257	1,7820	0,1084	ns
p10	-8,4643	4,9836	-1,1162	0,6572	-1,6984	0,1237	ns

Stepwise Regression (SR)



d)	beta	standard deviation error of beta	B	standard deviation error of B	t(9)	p-level	
intercept			-3,7366	2,8340	-1,3185	0,2059	ns
p9	0,6748	0,1924	0,1578	0,0450	3,5081	0,0029	p<0.05
p5	0,4160	0,1924	6,5983	3,0513	2,1624	0,0461	p<0.05

Fig. 2. a) and c) Calculated WVP versus experimental WVP obtained by MLR and SR processing, respectively; b) and d) levels of significance between p1 and p2-p10 for MLR and SR model, respectively; (p1-WVP, p2, p3 and p4-solubility in sim A, B and D1, respectively, p5-tensile strength, p6-bending resistance, p7-opacity, p8-swelling capacity, p9 and p10-biodegradability after 30 h and 60 h, respectively; ns-statistical non-significant).

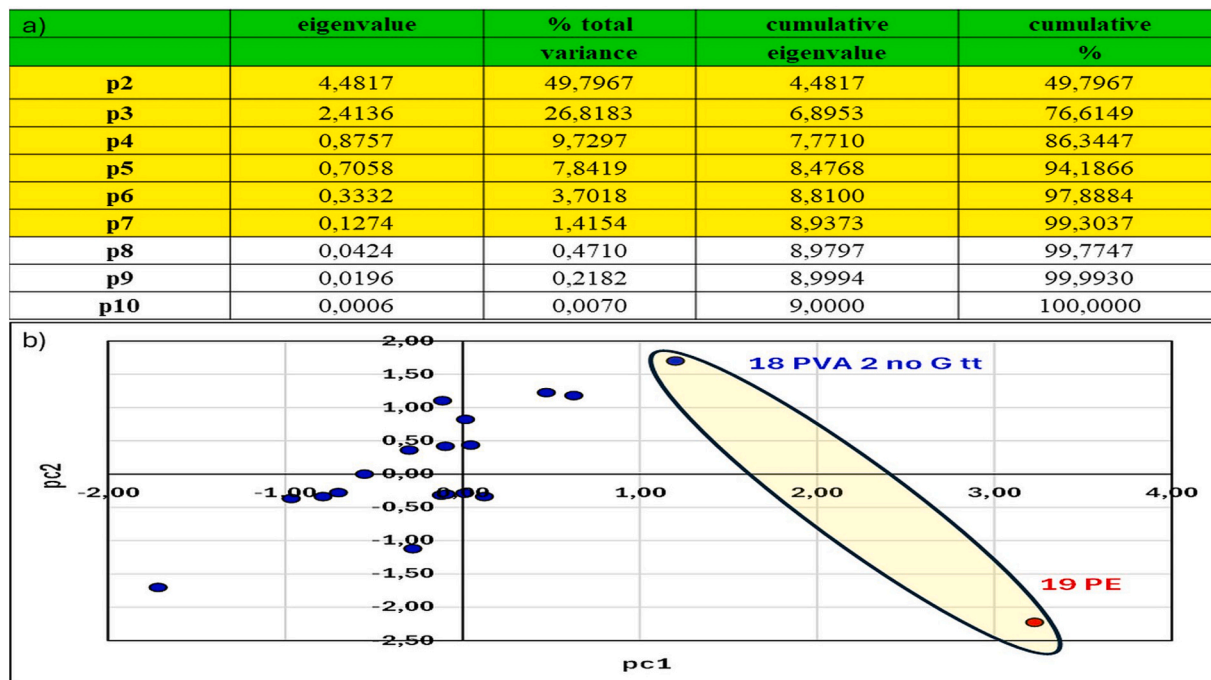


Fig. 3. a) Principal component analysis (PCA) on the physical-chemical parameters of beeswax-PVA composite films (p1-WVP, p2, p3 and p4-solubility in sim A, B and D1, respectively, p5-tensile strength, p6-bending resistance, p7-opacity, p8-swelling capacity, p9 and p10-biodegradability after 30 h and 60 h, respectively); b) Modelling dispersion of beeswax-PVA composite films as a function of pc1 and pc2 (pc1, pc2 – values of the first two principal components obtained by applying the PCA model).

The characteristics of PVA 2 no G tt film are the closest to PE and the most differed from those of the beeswax-PVA films.

### 3.6. Testing the beeswax-PVA composite films as bread food package

Because the best results for packaging analysis have been obtained for PVA 2 no G tt, this material will be subsequently used as packaging for bread rolls.

The inner portion of a bakery product has about 90 % moisture, therefore it tends to dry quickly and harden, so undesirable changes during bread storage include moisture and freshness loss and aging. The crust has low humidity and tends to soak in conditions of too high humidity and dry excessively at too low humidity. Aging of the bakery product occurs within 3–4 days after its manufacture. This is an inherent property of the type of flour, baking procedure and storage conditions. Water migrates from starch to protein during drying, thus the bread is becoming dry and lost its texture. Thus, the packaging material must have good moisture barrier properties to increase shelf life (Han, 2014).

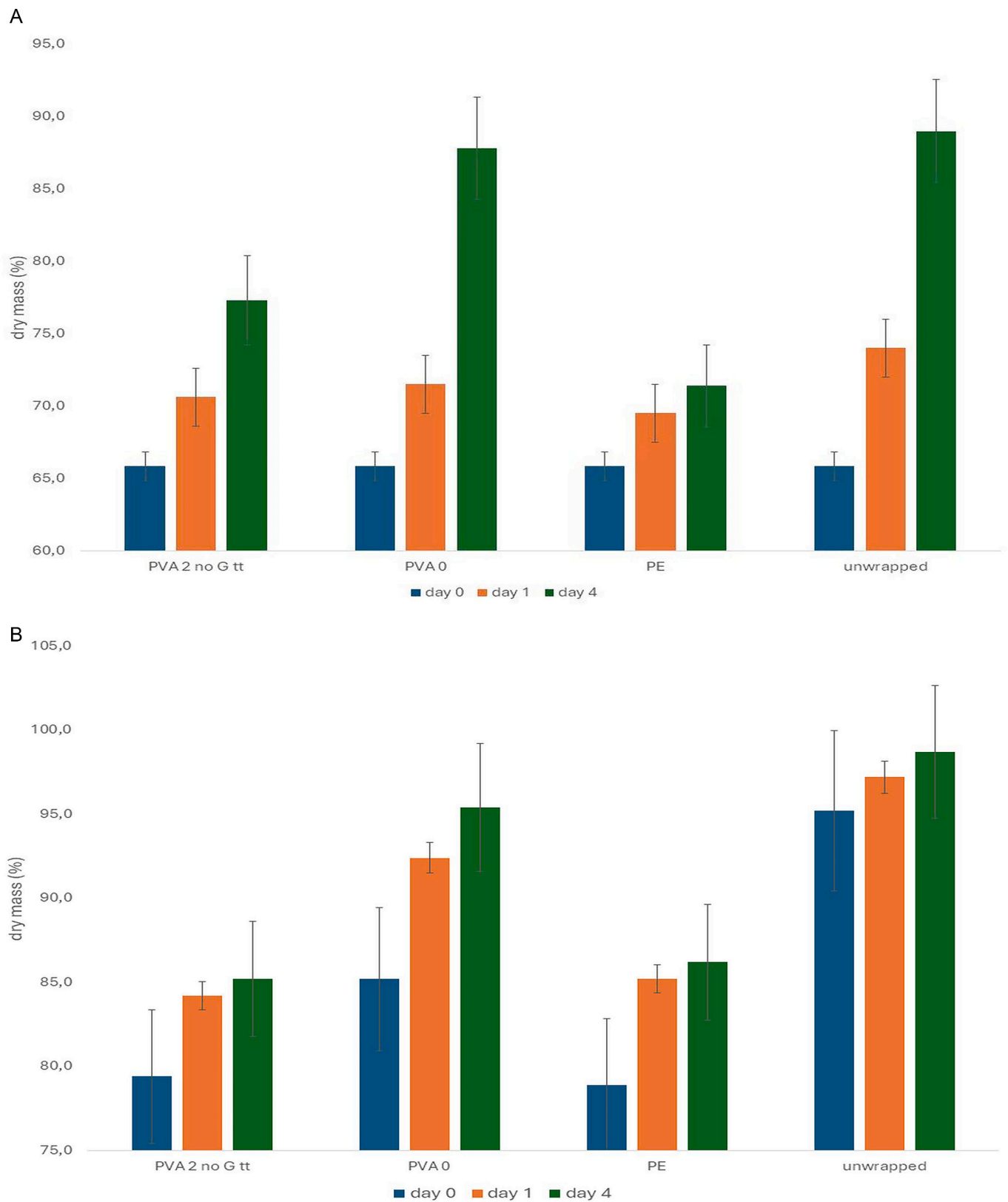
PVA 2 no G tt has been tested as a bread packaging material, as it has the lowest vapor retention capacity, PVA 0 and PE were used as references. At the beginning of the experiment, the bread appears fresh, well grown, unflattened, with pleasant golden color, uniform, slightly yellow and without lumps or traces of impurities. The crust is slightly browned and has no cracks or foreign odor and the shape is specific to the assortment. The core is well loosened, without clumps of flour, structurally connected to the shell, elastic and contains fine pores. After a day of packing in PVA 0, the bread bun became harder and stiffer, the core was very dry and the shell crumbly. After 4 days in PVA 0, the bread was much drier, which means that air has penetrated the packaging. The color began to be whitish and the peel to be very easy to crush (Table S3 from Supplementary material). The bread packed in PVA 2 no G tt, after a day, shows pleasant appearance, had relatively soft shell and the core was soft. After 4 days the bun presented harder peel, the core was more crumbly, displayed whitish color and the core was not very dry. The bread packed in PE, after a day of storage, got a pleasant appearance, a

little crumbly and a soft core. After 4 days, the appearance begins to change, the shell became hard and whitish due to drying and the core began to be hard. The unpacked bread, after few hours, began to have a stiffer shell, the core began to dry, was slightly crumbly but with a pleasant appearance. After 4 days, however, it was very hard both outside and inside, which means that it was dried out entirely. The appearance was still pleasant, whitish, but not edible due to its hardness.

The dry matter of the bread crumb increases during storage in all packaging materials tested (Fig. 4a). The most pronounced increase was registered in the case of the unpacked sample (by 23 % compared to day 0), followed by the sample packed in PVA 0 (by 22 % compared to day 0). The smallest increase was registered for samples packaged in PE (by 5,5 % compared to day 0) and PVA 2 no G tt (by 11 % compared to day 0). Thus, after 4 days of storage at 20 °C, the dry matter of crumb stored in PVA 2 no G tt was only 8 % higher than that of packaged bread in PE, while the dry mass of bread in PVA 0 was 23 % higher than that of PE. The same trend was obtained by analyzing the variation of the bread crust (Fig. 4b).

The hardness of the bread crust increases over time in all packages (Fig. 4c). The most intense increase was recorded for unpacked bread and bread packaged in PVA 0 and the lowest for bread packaged in PE and PVA 2 no G tt. After 6 days of investigation, the crust hardness of PVA 2 no G tt was only 1.11 times higher than that retained in PE, while that corresponding to PVA 0 was 4.81 times higher than in PE. So, by modifying PVA with beeswax, a packaging material that delayed the drying of the bread crust by 4.33 times was realized. This is an encouraging result for continuing to optimize the preparation conditions for the PVA-based films, Liu et al. (2017) showed that the PVA film modified with starch, antocyanins and limonene exhibits good color indication and antimicrobial activity in pasteurized milk, due to their intensive role in reducing the lactic acids bacteria growth at the initial spoilage stage for pasteurized milk. The intelligent PVA-based film developed by them showed good capability for both alerting and inhibiting food spoilage. Mustafa et al. (2020) also tested the preservative role of PVA-based films modified with propolis and rosemary





**Fig. 4.** Parameters of the bread wrapped in the investigated packages: crumb dry mass a), crust dry mass b) and crust hardness c).

antocyanins during pasteurized milk storage. The investigated active films showed significant changes by changing color on the pasteurized milk in comparison with the neat PVA. [Nguyen and Lee \(2022\)](#) tested the

preservative role of PVA modified with nanocellulose and  $\text{TiO}_2$  during the storage of fresh garlic and they have proven that the active films effectively prevented weight loss and spoilage by external influences,

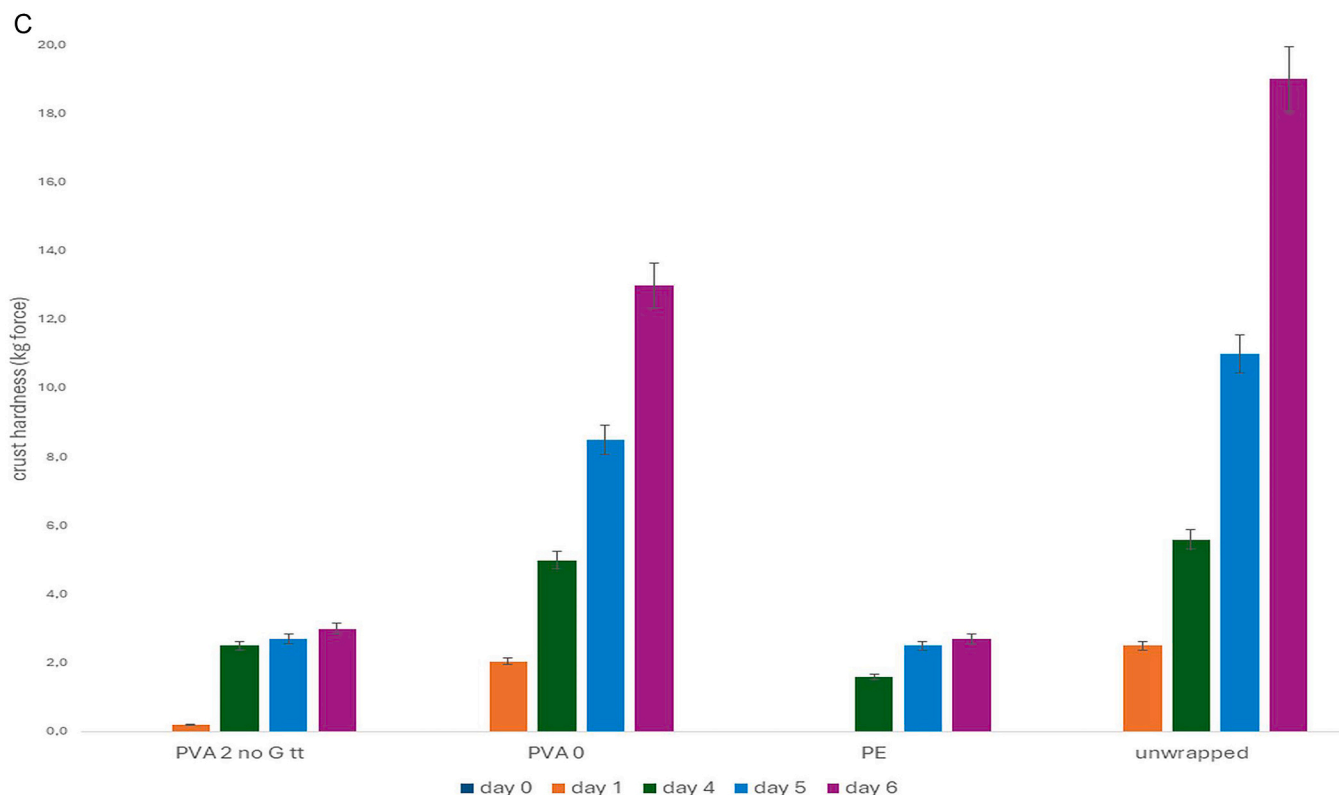


Fig. 4. (continued).

indicating their potential for food-packaging applications. Joseph-Leenose-Helen et al. (2022) explored the use of beeswax and pineapple for the enhancement of the self-life and nutraceutical potential of the Himalayan cheese, kradi. The coated cheese samples displayed retention of water activity. They showed that the designed coatings containing beeswax have great potential to be used in the cheese-making industry.

#### 4. Conclusions

The study aimed to improve the barrier properties of polyvinyl alcohol by adding beeswax and/or heat treatment. SEM microscopy revealed that the beeswax is coated on PVA's surface as well as in bulk PVA in the nontreated films and it penetrated the PVA matrix for the thermal treated ones. FTIR investigations showed that the film was not degraded during the thermal treatment at 125 °C for 30 min, but compacted and new connections were formed between the PVA and beeswax functional groups. Moreover, according to XRD, FTIR and XPS, new connections between the PVA and beeswax networks have been generated, demonstrating the composite structure of the films. TGA-DTA showed that glycerin reduced the dehydration and the presence of beeswax improved the thermal stability of the films. By modifying PVA with beeswax and thermal treatment at 125 °C for 30 min, the WVP was reduced by 74 %. Sample PVA 2 no G tt film has WVP only 7.5 times higher than PE (considered almost impermeable material to water vapor), while WVP of virgin PVA is almost 29 times larger than PE. In the case of PVA 2 no G tt sample showing the lowest WVP value, solubility in simulant A (distilled water) and simulant B (3 % acetic acid solution) is higher than in simulant D (ethanol solution 65 %), so the film would be more suitable for packaging fatty foods than aqueous and acidic ones. This formula is best suited for storing dry food. The correlation coefficients, linear and stepwise regression modelling showed that WVP varies reverse proportional with solubility in food simulants, swelling capacity and with biodegradability. Principal Component

Analysis model (PCA) proved that from all prepared beeswax-PVA samples, PVA 2 no G tt is the most similar with PE. The bread packaging analyses showed that the packaging efficiency of PVA 2 no G tt was only 8 % lower than PE, while that of virgin PVA was 23 % lower. So, modifying PVA with beeswax was able to preserve the bread softness by ~15 % in comparison with neat PVA.

#### CRedit authorship contribution statement

**Anca Peter:** Writing – original draft, Methodology, Data curation, Conceptualization. **Lucica Pop:** Visualization, Methodology. **Leonard Mihaly Cozmuta:** Writing – review & editing, Visualization, Supervision, Formal analysis, Data curation. **Camelia Nicula:** Writing – review & editing, Visualization, Investigation, Conceptualization. **Anca Mihaly Cozmuta:** Writing – review & editing, Supervision, Conceptualization. **Goran Drazic:** Writing – review & editing, Supervision, Methodology. **Klara Magyari:** Writing – review & editing, Visualization, Supervision, Methodology. **Marieta Muresan-Pop:** Writing – review & editing, Visualization, Methodology. **Milica Todea:** Writing – review & editing, Visualization, Supervision, Methodology. **Lucian Baia:** Supervision.

#### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

#### Data availability

Data will be made available on request.

#### Acknowledgements

The analyses were conducted at Research Center for Quality Control

and Environmental and Product Safety, Department of Chemistry and Biology, Technical University of Cluj-Napoca Romania, at the Electron Microscopy Laboratory at the National Institute of Chemistry, Ljubljana, Slovenia, and at Nanostructured Materials and Bio-Nano-Interface Center, Interdisciplinary Research Institute on Bio-Nano-Sciences, Babes-Bolyai University, Cluj-Napoca Romania.

## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.fochx.2024.102053>.

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