

Investigation of the Effect of L-Glutamic Acid Content on the Biodegradation of Poly(Vinyl Alcohol) Membranes and Poly(Vinyl Alcohol) Foamed with Poly(Ethylene Glycol)

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Abstract

The study aims to evaluate the influence of L-glutamic acid content on the degradation of modified PVA materials and PEG-porous modified PVA materials, based on previous research, was investigated. To assess the biodegradation, measurements such as mass analysis were employed to evaluate the weight loss of the materials. In addition, SEM was utilized to analyze the morphological changes, and FT-IR spectroscopy was applied to monitor the variations in functional groups. The results showed that the PVA-c-Glu material was degraded by 24-32% after 150 days. For the PVA-c-Glu/PEG material, the mass loss ranged from approximately 25-46% after 150 days due to the addition of pores. Based on the SEM observations, significant surface changes were detected, which were attributed to microbial colonization forming clusters on the material surface. Moreover, the FT-IR results indicated that the signals corresponding to functional groups were attenuated after degradation.

Keywords: PVA, L-glutamic acid, porous polymer, biodegradation.

1. Introduction

With the rapid expansion of industry and increasing consumer demand, plastic pollution has become a serious global environmental challenge [1, 4]. Improperly managed plastic waste accumulates in oceans, rivers, and landfills, posing significant threats to ecosystems and human health [1, 3]. Furthermore, both the production and degradation of plastics release harmful substances, exacerbating air, soil, and water pollution. Addressing this issue requires the development of eco-friendly materials, improvements in recycling systems, and a reduction in single-use plastics. These efforts align with sustainable development goals and contribute to the advancement of a circular economy.

Poly(Vinyl Alcohol) (PVA) is a biodegradable, non-toxic, and environmentally friendly polymer known for its excellent water solubility and film-forming ability [5, 6]. Due to its natural degradability, PVA is considered a sustainable alternative to conventional synthetic polymers, helping to minimize long-term environmental impact. Notably, it is the only vinyl polymer that can undergo biodegradation through bacterial activity [7]. Additionally, PVA is biocompatible and can be processed using aqueous

methods, ensuring its safety for human use in various applications, including membrane technology [8, 9].

However, a major drawback of PVA is its linear polymer structure with a high density of hydrophilic hydroxyl (–OH) groups, and the PVA chains are straight, which easily slide over each other in aqueous environments, resulting in poor mechanical strength of PVA in aqueous environments [10, 11]. This characteristic makes PVA prone to dissolution, limits its flexibility, and significantly restricts its practical applications, especially in wet conditions. To overcome these limitations while preserving its biodegradability and environmental safety, researchers have focused on modifying PVA using natural and eco-friendly additives.

An effective approach is the incorporation of L-glutamic acid, a natural amino acid found in various plants [12] and meat [13], which has the ability to cross-link to enhance the mechanical stability of PVA due to the presence of two –COOH groups, which help cross-link with PVA chains through solid-phase esterification reaction [14]. In addition, the amine group (–NH₂) in L-glutamic acid also has the potential for heavy metal adsorption due to its strong coordination ability with metal ions [15], improving adsorption

efficiency compared to the hydroxyl (–OH) groups in PVA. Therefore, L-glutamic acid is a safe, naturally derived, and environmentally friendly crosslinking option.

The crosslinking process can reduce the free volume within the polymer structure, as PVA chains become more tightly packed, leading to decreased water permeability of the membrane [14, 16]. To overcome this limitation, Poly(Ethylene Glycol) (PEG) was incorporated to create a porous structure within the material. This modification increases the surface area while also enhancing the free volume within the polymer matrix. Moreover, PEG is a water-soluble polymer that can biodegrade under suitable environmental conditions [17], contributing to the overall eco-friendliness of the composite material.

This study aims to evaluate the effect of L-glutamic acid on the biodegradability of PVA membranes and PEG-foamed PVA in soil. To analyze changes after biodegradation, gravimetric analysis of weight loss, Scanning Electron Microscopy (SEM) and Fourier Transform Infrared Spectroscopy (FT-IR) will be used to examine mass change of materials, surface morphology, degradation characteristics, and chemical structural changes in the materials. The findings of this study will contribute to a deeper understanding of the role of eco-friendly modifying agents in optimizing biodegradable materials, promoting their sustainable applications in real-world scenarios.

2. Experiment

2.1. Materials

Poly(Vinyl Alcohol) (CAS: 9002-89-5; $M_w = 89000\text{--}98000$; $\geq 99\%$ hydrolyzed), and Poly(Ethylene Glycol) (CAS: 25322-68-3; $M_n = 6000$) were purchased from Sigma-Aldrich (USA), L-Glutamic acid (CAS: 1002910250) was purchased from Merck (Germany). Other chemicals used in this research were supplied by Shanghai (China) and Sigma-Aldrich (USA).

2.2. Preparation of the Materials

2.2.1. Synthesis of PVA-c-Glu

Solution A was prepared by dissolving PVA in distilled water under stirring (250 rpm) at 85 °C for 2 hours. Separately, Solution B was prepared by dissolving L-glutamic acid hydrochloride (L-Glu.HCl) in distilled water with stirring at room temperature for 1 hour. Solutions A and B were then combined and stirred (250 rpm, at room temperature, 2 hours) to form a homogeneous PVA/L-Glu.HCl mixture, which was cast into a Petri dish and dried at 50 °C to obtain a crude membrane. Crosslinking was performed by heating the dried membrane at 130 °C for 1 hour, facilitating ester bond formation between PVA's hydroxyl groups and the carboxyl groups of L-Glu. The content of L-Glu.HCl relative to PVA was varied from 10 to 50% (following

the PVA content) to investigate the effect of crosslinker concentration on the membrane properties (Fig. 1).

2.2.2. Synthesis of PVA-c-Glu/PEG

Solution A was prepared similarly to the experiment mentioned above. Solution C contained L-glutamic acid hydrochloride (L-Glu.HCl, 10-50 wt% relative to PVA) and PEG (fixed at 50% of PVA content, %w/w) dissolved in distilled water under room-temperature stirring for 1 hour. The two solutions were combined and stirred (250 rpm, at room temperature, 2 hours) to form a homogeneous PVA/PEG/L-Glu.HCl mixture, which was cast onto a silicone mold and dried at 50 °C to obtain a crude membrane. Crosslinking was achieved by heating the membrane at 130 °C for 1 hour. To remove the pore-forming PEG phase from the material, the crosslinked membrane was extracted in distilled water (room temperature, 48 hours) and dried at 50 °C, yielding the final porous PVA-c-Glu/PEG membrane (Fig. 2).

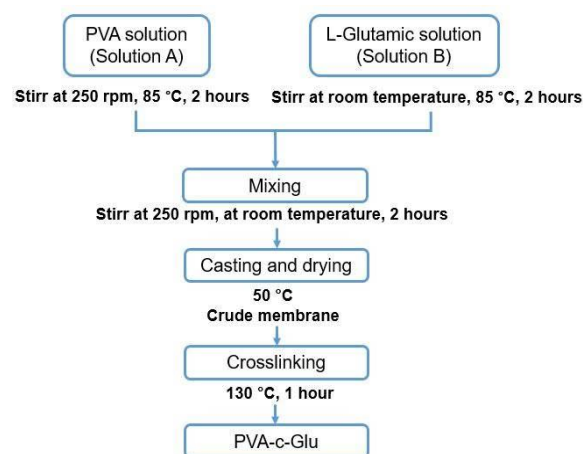


Fig. 1. Summary diagram of the synthesis process of PVA-c-x%Glu material ($x = 10; 20; 30; 40; 50$)

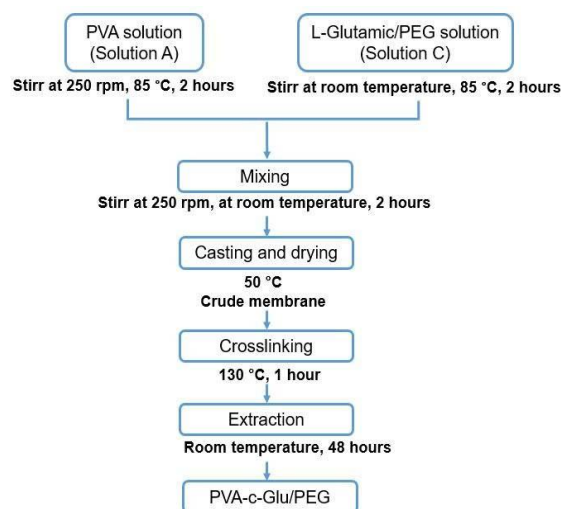


Fig. 2. Summary diagram of the synthesis process of PVA-c-x%Glu/PEG material ($x = 10; 20; 30; 40; 50$)

2.3. Methods

2.3.1. Biodegradable in soil

PVA-c-Glu and PVA-c-Glu/PEG films were prepared with varying contents of L-Glutamic acid (L-Glu) ranging from 10-50% by weight relative to PVA, while PEG was maintained at a constant concentration of 50% as a porogen. All samples were synthesized following a previously established procedure. Prior to the burial experiment, the films were cut into square pieces measuring $x \text{ cm} \times x \text{ cm}$ and their initial weight (m_0) was accurately determined using a high-precision analytical balance.

The experiment was conducted in standard garden soil under strictly controlled conditions: the relative humidity of the soil was maintained at 50-70 %, the soil was watered twice a day to maintain moisture, and the temperature was kept stable at $25 \pm 2 \text{ }^\circ\text{C}$ (the samples were performed simultaneously at room temperature) [18, 19]. The samples were buried at a depth of 10 cm with a spacing of 5 cm between each sample to ensure uniform decomposition conditions. Burial times were set at 0, 30, 60, 90, and 150 days.

After each predetermined burial period, the samples were retrieved and processed according to a standardized procedure. First, the samples were gently rinsed with distilled water to remove any adhered soil. They were then dried at $50 \text{ }^\circ\text{C}$ until reaching a constant weight. The remaining weight of each sample (m_1) was accurately measured using an analytical balance with a precision of $\pm 0.1 \text{ mg}$. The biodegradation percentage was calculated using the formula:

$$\text{Biodegradation (\%)} = \frac{m_0 - m_1}{m_0} \times 100 \quad (1)$$

where m_0 is the initial weight before burial and m_1 is the weight after burial and post-treatment. This method enables a precise quantitative assessment of the material's degradability under natural soil conditions.

2.3.2. Scanning electron microscopes

SEM was employed to investigate the surface morphology and microstructure of the films. SEM provides high-resolution images by scanning the sample surface with a focused electron beam, enabling detailed analysis of surface roughness, particle size, and structural features without destroying the sample.

The sample after biodegradation was washed to remove soil on the surface of the material with distilled water, and dried at $30 \text{ }^\circ\text{C}$. SEM observations were performed using a Thermo Fisher Scientific instrument (USA) at an accelerating voltage of 15 kV.

2.3.3. Fourier transform infrared spectroscopy

Fourier Transform Infrared Spectroscopy (FT-IR)

was employed to analyze the chemical composition and molecular structure of the materials. FT-IR identified functional groups by measuring the absorption of infrared radiation, providing information on chemical bonds and structural changes. The technique offers rapid, non-destructive analysis with high sensitivity. FT-IR measurements were performed using an IRAffinity-1S spectrometer (Shimadzu Corporation, Japan) in the range of $400\text{-}4000 \text{ cm}^{-1}$ with Attenuated Total Reflectance (ATR) mode.

3. Result and Discussion

3.1. Effect of L-Glutamic Acid Content in PVA-c-Glu Membranes on Biodegradation in Soil

The biodegradability of PVA crosslinked with varying percentages of glutamic acid (PVA-c- $x\%$ Glu) was evaluated via soil burial at ambient temperature (approx. $30 \text{ }^\circ\text{C}$) over a 150 day period, without enzymatic intervention, as shown in Fig. 3 and Fig. 4.

Gravimetric analysis revealed a progressive mass loss, ranging from 11-18% after 30 days to 24-32% after 150 days. Notably, the rate of degradation diminished significantly between 60 and 150 days, with mass loss plateauing within the 12-22% range during the 30-60 day interval. Concomitantly, visual inspection indicated a gradual color fading, indicating a change in material properties. These findings demonstrate the biodegradability of the PVA-c-Glu material under environmental conditions, albeit with a decelerating degradation rate over extended exposure.

The observed biodegradation was attributed to alterations in the material's structure influenced by varying glutamic acid content. The inherent water solubility of PVA facilitates initial dissolution and interaction with environmental enzymes, promoting microbial access [20]. According to the published study by R Ray *et al*, the biodegradability of the original PVA film in soil at ambient temperature ($27.5 \text{ }^\circ\text{C}$) was 10 to 25% after 3 and 6 days [18]. Therefore, it can be seen that the presence of L-glutamic acid reduces the biodegradability of the PVA film, increasing the L-glutamic acid content correlates with a reduction in biodegradability, possibly due to the enhanced mechanical strength and modified hydrophilicity, due to increased crosslinking between PVA and L-glutamic acid, which hinders the degradation rate. However, despite this inhibitory effect at higher concentrations, the presence of L-Glutamic acid, a natural amino acid, may also contribute to the increased biodegradability of the PVA-c-Glu material by providing accessible degradation sites for microorganisms and enzymes, thus counteracting the increased degradation resistance.

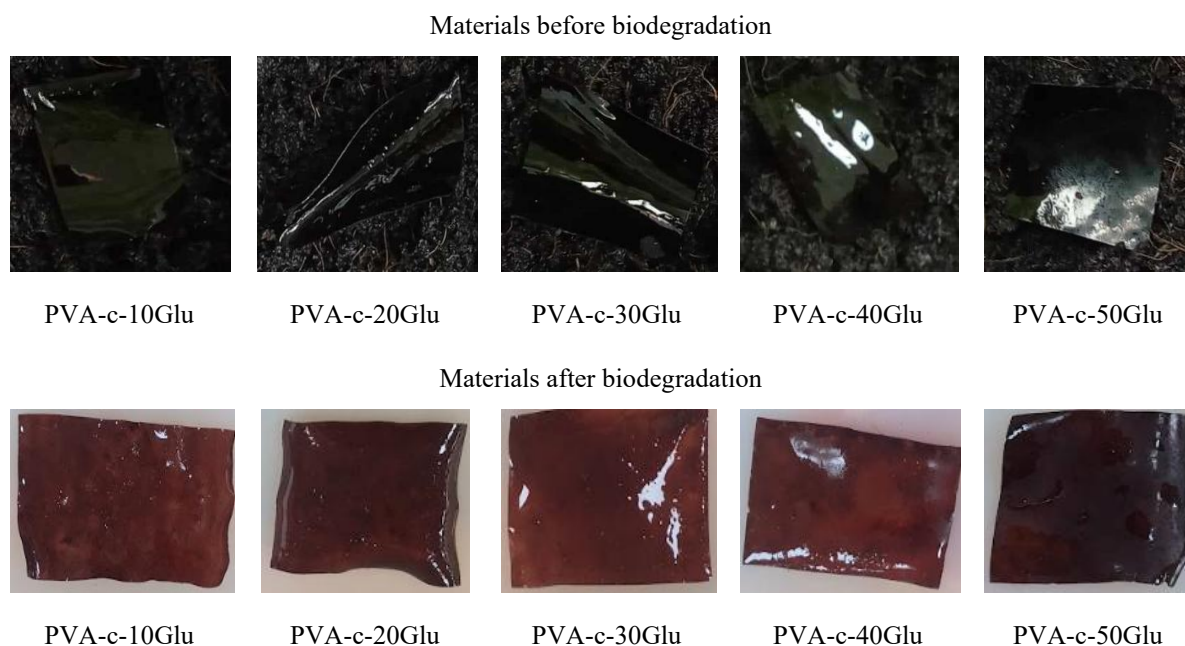


Fig. 3. Photograph of biodegradation of PVA-c-x%Glu Membrane ($x = 10; 20; 30; 40; 50$)

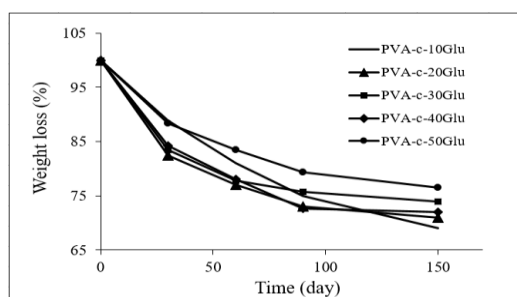


Fig. 4. Mass loss graph of PVA-c-x%Glu material during biodegradation in natural soil

3.2. Effect of L-Glutamic Acid Content on Soil Biodegradation in PVA-c-Glu/PEG Membranes

The biodegradability of PVA-c-x%Glu/PEG materials ($x = 10, 20, 30, 40, 50$) was assessed through soil burial over 150 days at ambient temperature (circa 30 °C), mirroring the methodology used for varying glutamic acid content but with the incorporation of PEG, as shown in Fig. 5 and Fig. 6.

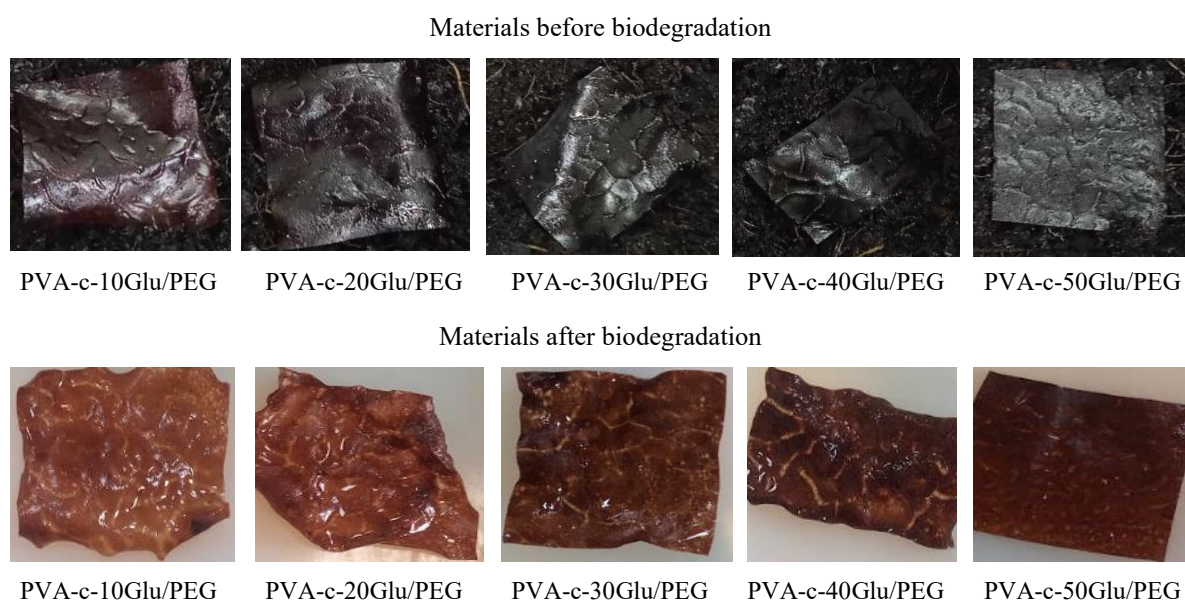


Fig. 5. Photograph of biodegradation of PVA-c-x%Glu/PEG membrane ($x = 10; 20; 30; 40; 50$)

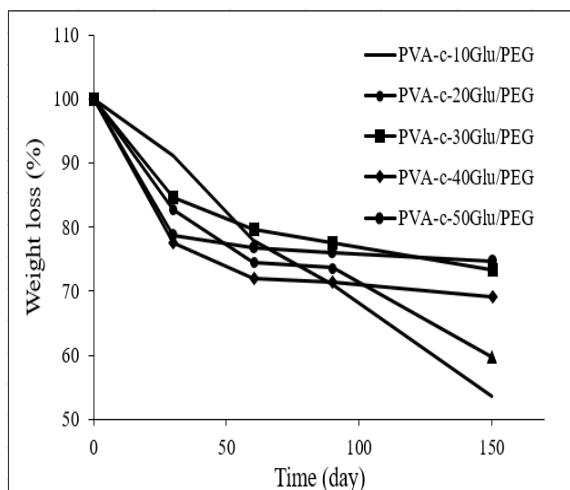


Fig. 6. Mass loss graph of PVA-c-x%Glu/PEG material during biodegradation in natural soil

Gravimetric analysis revealed a mass loss ranging from 9-22% after 30 days, progressing to 25-46% after 150 days. A minimal change in mass (11-28% loss) was observed between 30 and 60 days, indicating a deceleration in the degradation rate. Visual observation showed a gradual color fade compared to the undegraded material, consistent with material decomposition.

This enhanced biodegradation observed in PEG-containing materials was attributed to alterations in their structure and morphology, characterized by the formation of numerous surface pores. This increased porosity significantly expands the material's surface area exposed to the degrading soil environment. Consequently, natural microorganisms and enzymes gain more facile access to the polymer matrix, accelerating the degradation process. Furthermore, the increased surface area facilitates greater water penetration into the material, establishing a more conducive environment for microbial proliferation and subsequent decomposition.

3.3. Morphology of PVA-c-Glu Materials Altered by L-Glutamic Acid Content after Biodegradation

The PVA-c-x%Glu material, after undergoing 150 days of degradation in soil, was observed by SEM and is presented in Fig. 7. The surface of the PVA-c-Glu material prior to biodegradation was characterized as smooth and flat. However, subsequent to biodegradation, nodular protrusions were observed across the material's surface, occurring in clusters and distributed throughout, indicating the activity of indigenous soil microorganisms [21, 22]. The figure revealed that microbial activity was most pronounced in the material with a 50% L-glutamic acid content, a phenomenon attributed to L-glutamic acid being an amino acid that facilitates the proliferation of microorganisms.

However, strong microbial activity was observed in the material containing 50% L-glutamic acid, but the mass loss after biodegradation was lower than that of the other L-glutamic acid contents. This could be attributed to the fact that the polymer network structure of the PVA material, after being cross-linked with L-glutamic acid, became denser and more durable. As a result, the polymer chains were no longer as loose as those in materials with lower L-glutamic acid content, leading to reduced biodegradability.

3.4. Morphology of PVA-c-Glu/PEG Materials Altered by L-Glutamic Acid Content after Biodegradation

The biodegradation of the PVA-c-x%Glu/PEG materials in soil after 150 days is presented in Fig. 8. It was observed that the addition of PEG resulted in the creation of pores on the material's surface, thereby facilitating easier accessibility of bacteria to the material. Consequently, bacteria were even able to readily penetrate the internal structure of the material, leading to a more rapid biodegradation of the material compared to PVA-c-x%Glu. This acceleration occurred because the degradation process in PVA-c-x%Glu was required to proceed from the surface into the bulk volume of the membrane [22, 23].

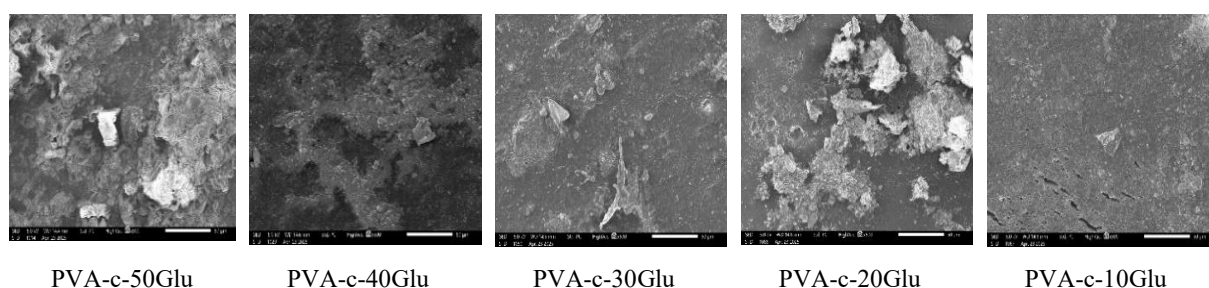
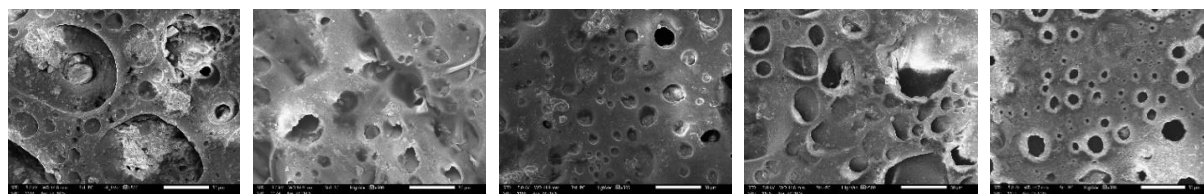


Fig. 7. SEM images of PVA-c-x%Glu ($x = 10, 20, 30, 40, 50$) material after 150 days of degradation (white bar indicates 50 μm)



PVA-c-50Glu/PEG PVA-c-40Glu/PEG PVA-c-30Glu/PEG PVA-c-20Glu/PEG PVA-c-10Glu/PEG

Fig. 8. SEM images of PVA-c-x%Glu/PEG ($x = 10, 20, 30, 40, 50$) material after 150 days of degradation (white bar indicates 50 μm)

3.5. Confirmation of the Structure of PVA-c-Glu and PVA-c-Glu/PEG Materials after Biodegradation

The FT-IR spectra of PVA-c-x%Glu and PVA-c-x%Glu/PEG materials before and after biodegradation were presented in Fig. 9 and Fig. 10. Characteristic absorption bands were observed, including the $-\text{OH}$ stretching vibration in the range of $3000\text{--}3500\text{ cm}^{-1}$, $-\text{CH}_2$ stretching between $2800\text{--}3000\text{ cm}^{-1}$, $-\text{C}=\text{O}$ stretching around $1600\text{--}1700\text{ cm}^{-1}$, and $-\text{NH}_2$ bending in the $1500\text{--}1600\text{ cm}^{-1}$ region. From the FT-IR results, shifts in the $-\text{C}=\text{O}$ and $-\text{NH}_2$ bands were detected in Fig. 9, and the disappearance of the $-\text{C}=\text{O}$ signal was observed in Fig. 10.

After degradation in soil, it was observed that the FT-IR spectra of all materials exhibited lower peak intensities compared to the original films. This was attributed to significant surface degradation of the materials, leading to a considerable reduction in functional groups [21]. In addition, the shift and gradual disappearance of the $-\text{C}=\text{O}$ band were attributed to the fact that during the biodegradation process, the $-\text{C}=\text{O}$ group was one of the most vulnerable and easily degraded functional groups [21].

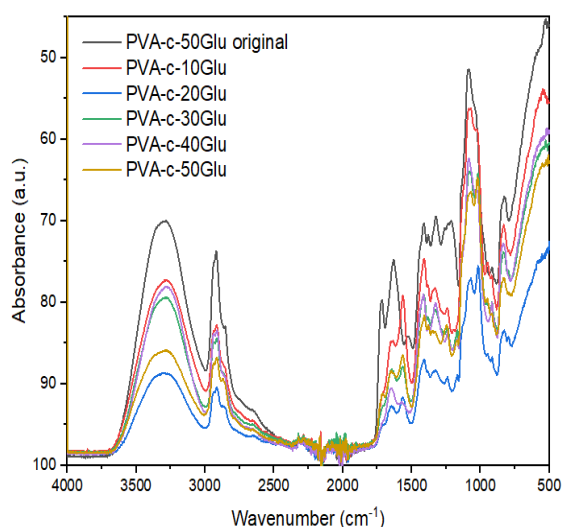


Fig. 9. FT-IR spectra of the original PVA-c-50Glu material and PVA-c-x%Glu material after biodegradation

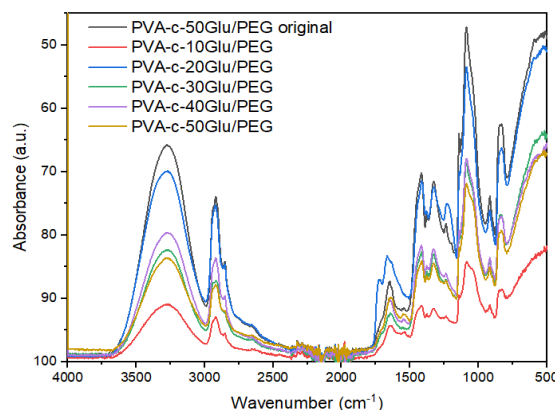


Fig. 10. FT-IR spectra of the PVA-c-50Glu/PEG original material and PVA-c-x%Glu/PEG material after biodegradation

4. Conclusion

In this study, the variation of L-glutamic acid content and the introduction of porosity by PEG were found to significantly affect the biodegradation of the materials. According to the results, the PVA-c-Glu material exhibited a mass loss of approximately 24-32% after 150 days, while the original PVA degraded up to 25% after only 6 days. Thereby, the use of L-glutamic acid as a cross-linking agent for the material reduced the biodegradability of the material due to the more tightly bound structure of the material. For the PVA-c-Glu/PEG material, a loss of 25-46% was observed after 150 days. Therefore, the presence of PEG-induced pores enhanced the biodegradability of the material. The fastest biodegradation was observed at a 10% L-glutamic acid content, and incorporating PEG at 50% of the PVA weight further improved the degradation rate of the modified PVA. However, at 50%Glu content, the most vigorous activity of natural soil microorganisms was observed.

The increase in L-glutamic acid content was found to promote the growth of microorganisms on the material surface, as clearly observed through SEM images, which helped in the rapid decomposition of the material thanks to the natural microorganisms in the soil. In addition, the porosity of the material due to PEG increased the accessibility of microorganisms

into the inner structure of the materials, thereby improving the biodegradation efficiency of the PVA-c-Glu material.

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