*Journal of Molecular Science*
*Volume 35 Issue 3, Year of Publication 2025, Page 587-593*
*DoI-10.004687/1000-9035.2025.080*

Journal of Molecular Science

Journal of Molecular Science
www.jmolecularsci.com ISSN:1000-9035

# Study of the Chemical and Degradation Behavior of a Sustainable Biopolymer Derived from Keratin – Lignin Blend.

**Olibha Xalxo[1], Kush Kumar Nayak[2*], Varaparasad Kolla[1*]**
[1]Amity Institute of Biotechnology, Amity University Chhattisgarh, Raipur.
[2] School of Studies in Biotechnology, Shaheed Mahendra Karma Vishwavidyalaya, Bastar.
Corresponding Author Email id: **nayak23bt@gmail.com**

***Article Information***
Received: 18-09-2025
Revised: 25-09-2025
Accepted: 02-10-2025
Published: 08-10-2025

**Keywords**
*keratin, sustainable, crosslinkers, hydrogen, biodegradation, crystallinity.*

## ABSTRACT

**The increasing demand for environmentally friendly alternatives to petroleum-based plastics has prompted research into biopolymers made from renewable resources. In this study, keratin from poultry feather waste and lignin from rice husks have been extracted and combined in optimized ratios to create environmentally sustainable composite mat. A 70:30 keratin-lignin mixture produced the optimum mat forming ability and structural stability. Four crosslinkers were tested for performance enhancement: formaldehyde, glutaraldehyde, acetic acid, and polyvinyl alcohol (PVA). Fourier-transform infrared spectroscopy (FTIR) demonstrated successful chemical interactions, particularly in formaldehyde and glutaraldehyde crosslinked systems via Schiff base and acetal linkages, whereas acetic acid caused partial esterification and PVA produced hydrogen-bonded networks. X-ray diffraction (XRD) revealed mostly amorphous structures, with aldehyde-based crosslinkers dramatically lowering crystalline and boosting flexibility. Among all systems, formaldehyde crosslinked mat demonstrated excellent casting characteristics and stability. Biodegradation experiments showed approximately 35% mass loss over 10 days, with some materials exceeding 50% degradation. These findings point to keratin-lignin biopolymer blends as interesting options for sustainable material development, as they combine biodegradability with variable structural performance via crosslinking techniques.**

## 1. INTRODUCTION: -

The shift in focus from petrochemical based materials to sustainable and renewable alternatives has been accelerated by the growing urgency of global environmental issues (Tabassum et al., 2023). To reduce environmental pollution without depleting natural resources, comprehensive green solutions had to be developed. The development of materials that are safer and of higher quality than the biological system materials already in use is prompted by the need to control future issues brought on by plastic waste and petroleum sources. By investigating environmentally friendly products, a lot of research is being done to create bio-based polymers that can replace petroleum derived and related substances (Chincholikar et al., 2023).

In this regard, lignin and keratin show promise as biopolymeric options. Widely obtained from poultry waste, particularly chicken feathers, keratin has a great deal of potential for value-adding in a circular bioeconomy, which would help cut down on pollution and resource depletion (Tabassum et al., 2023). The second most prevalent biopolymer, lignin, is derived from lignocellulosic biomass and is usually burned or thrown away as low-value fuel. Because of its highly crosslinked, aromatic nature, it offers a great chance to be incorporated into polymer networks to improve mechanical and thermal performance (Stanzione & La Scala, 2016). Despite these promising characteristics, isolated keratin-derived materials frequently have low molecular weight and poor mechanical capabilities, whilst lignin's structural variability and limited reactivity present problems for uniform integration. This emphasizes the necessity for novel crosslinking techniques to efficiently create keratin-lignin biopolymer networks with specific functionality (Chinomso Iroegbu & Ray, 2022; Stanzione & La Scala, 2016). Due to their reactivity, classic crosslinkers like aldehydes continue to be widely used. Formaldehyde effectively facilitates crosslinking between functional polymers with hydroxyl or amine groups, such as PVA and lignin, improving water resistance and thermal stability in composite films of alkaline lignin and polyvinyl alcohol. Formaldehyde's high reactivity allows for strong network development, increasing thermal breakdown temperatures and decreasing crystallinity when compared to clean PVA films(Su & Fang, 2014). Similarly, the capacity of glutaraldehyde, a dialdehyde with two formyl endings, to join primary amines to form imines and stiffen protein structures is well-known. Glutaraldehyde is a strong crosslinker for amine-rich proteins or polymers in the field of biomaterials, improving structural integrity through condensation between amino groups. Its strong crosslinking makes keratin networks more thermally stable and stiff, but usually at the expense of greater cytotoxicity, which is something that greener design needs to take into consideration (Grabska-Zielińska, 2024). It has a better ability to crosslink proteins by condensing with primary amines, creating imine linkages, and stiffening networks. Although its toxicity necessitates cautious use, it has been extensively utilized in biomaterials to improve stability, stiffness, and resistance to biodegradation. Glutaraldehyde crosslinked samples in keratin films showed distinct FTIR alterations and changed thermal degradation behaviors, indicating successful crosslinking, albeit occasionally with lower thermal stability when compared to other agents (Steitz et al., 2023; Thonpho et al., 2015).

Acetic acid has two complementary functions: it can cause acid triggered degradation in lignin-based thermoplastics and acts as a moderate acid catalyst in specific crosslinking reactions. In moderate acetic acid solutions (acetic acid/water 1:1), for instance, lignin containing films have been shown to dissolve in a matter of hours, indicating both rapid degradability and the possibility of controlled processing pathways (Chinomso Iroegbu & Ray, 2022). Acetic acid makes a variety of contributions. Being a mild acid, it improves the solubility of biomass components in ionic liquids and catalyzes regulated hydrolysis or dissolution in lignocellulosic systems, opening more benign processing avenues. In lignin-specific applications, acetic acid is used in microwave-assisted selective acetylation, which enhances solubility and antioxidant capacity while maintaining phenolic activities by selectively acetylating aliphatic hydroxyls in kraft lignin (de Oliveira et al., 2020; Trzcinski & Stuckey, 2015). Blending polyvinyl alcohol (PVA) with keratin or lignin makes it a synergistic companion. PVA improves mechanical qualities and processability when heat-treated, keratin-PVA mix fibers have more waterproof stability and tenacity than wool, most likely because of crosslinking through hydrogen bonding or disulfide production (Katoh et al., 2003). In the meantime, crosslinked films with remarkable water resistance, and thermal stability are produced by combining alkaline lignin with PVA in the presence of formaldehyde. (Su & Fang, 2014).

Biomaterials made from blends of two or three components are widely researched. Two main techniques for creating these blends are mixing aqueous solutions in solvents and combining in the molten state. Fourier transform infrared spectroscopy (FTIR) is commonly used to examine the interactions and miscibility of these blends (Nuutinen et al., 2022). Crosslinking is often used to improve the mechanical, barrier, and stability properties of lignin and keratin-based materials (Grabska-Zielí Nska et al., 2021) . The establishment of chemical links between polymer chains results in a more robust and interconnected network structure. This study looks at the usage of different crosslinking agents, such as formaldehyde, glutaraldehyde, acetic acid, and polyvinyl alcohol. Formaldehyde and glutaraldehyde are popular crosslinkers recognized for their efficiency in biopolymer modification (Kawamura et al., 2021). Polyvinyl alcohol, a water-soluble synthetic polymer, is also investigated for its capacity to form films and interact with biopolymers, potentially increasing the mechanical and barrier properties of the resulting bio composites (Nasalapure et al., 2021). Thus, PVA functions as a reactive scaffold as well as a structural enhancer (Su & Fang, 2014).

The current study attempts to generate long lasting keratin-lignin composite matte by carefully testing several crosslinking procedures. Four crosslinkers (formaldehyde, glutaraldehyde, acetic acid, and polyvinyl alcohol) were used to improve the blends' structural stability, processability, and performance. The capacity to form films, chemical interactions, and structural ordering were evaluated using FTIR and XRD techniques, as well as biodegradability tests under soil burial circumstances. The study's methodology aims to determine the most effective crosslinking strategy for producing eco-friendly, biodegradable films with potential uses in sustainable packaging, agriculture, and related industries.

## 2. MATERIALS AND METHODOLOGY:

### 2.1 Materials required:

Fresh chicken feathers were collected from a local butcher shop in Raipur, Chhattisgarh, and used as keratin sources. Banana leaves were collected from the Amity University campus in Chhattisgarh for use as lignin biomass. All reagents used in this study were analytical grade. HiMedia (India) supplied urea, sodium dodecyl sulphate (SDS), ethanol, and glutaraldehyde, while Merck (India) provided β-mercaptoethanol, sodium hydroxide (NaOH), sulphuric acid ($H_2SO_4$), dioxane, acetic acid, and formaldehyde solution. Polyvinyl alcohol (PVA) was purchased from HiMedia (India). The experimental methods were carried out with deionized water manufactured in-house at Amity University Chhattisgarh laboratory. All compounds were utilized as obtained, without further purification.

### 2.2 Biomass Pre-treatment:

Chicken feathers were collected from a local butcher store and cleaned to eliminate any unwanted dirt before being thoroughly washed with deionized water. To further purify, the chopped feathers were soaked in 1% (v/v) ethanol overnight, then drained, rinsed with deionized water, and air dried. The feathers were air dried at ambient temperatures (about 25 °C) before they were chopped into smaller pieces. Portions of approximately 5 g were prepared to achieve a 5% (w/v) loading when dispersed in 250 ml of solution for subsequent experiments.

Rice husks were collected from a nearby rice mill, thoroughly washed with water to remove dirt and air dried to room temperature (about 25 °C). The dried husks were manually separated, diced into smaller bits, then milled with a grinder. The resultant material was sieved using a 350 µm mesh to ensure a consistent particle size distribution. The sieved rice husk powder was then sealed in sterile, airtight polyethylene pouches and stored in a dry environment, following which it was immersed overnight in an aqueous sodium hydroxide (NaOH) solution of about 250ml solution.

### 2.3 Extraction protocol of keratin & lignin:

To extract keratin, pre-treated chicken feather pieces were carefully cleaned with distilled water to remove any remaining pollutants before drying in a hot air oven. To solubilize the dried feather material, it was mixed with 250 ml of an extraction solution containing 1.2% (w/v) urea, 0.3% (v/v) β-mercaptoethanol, and 0.35% (w/v) sodium dodecyl sulphate. The mixture was constantly stirred on a magnetic stirrer and incubated for 3-4 days to total feather decomposition. This procedure produced a semi-solid keratin extract (Mattiello et al., 2023).

To extract lignin, the suspension was incubated on a heated magnetic stirrer for 2 hours at 40-60 °C to promote partial delignification and rupture of the lignocellulosic matrix. The alkaline slurry was then allowed to stand for 48 hours. Following incubation, the mixture was filtered to separate the solid fraction from the alkali-soluble liquid. Approximately 0.5 mL of 5% (v/v) sulfuric acid solution was added to the filtrate to adjust the pH to around 1.5. The crude lignin fraction was washed twice with a solution made of 1 g NaOH in 100 ml deionized water and a 96:4 (v/v) mixture of dioxane and water. The final lignin precipitate was extensively rinsed with deionized water until neutral pH was obtained, and then oven-dried at 40-50°C for storage(Nazimudheen et al., 2020).

### 2.4 Preparation of Keratin-Lignin Blend (70:30 Ratio):

A semi-aqueous keratin solution was produced and kept at a suitable concentration. Separately, lignin powder was dissolved in distilled water (1:10 w/v) to form a homogenous solution. Various concentrations were examined, as stated in table 1. The two solutions were mixed in a 70:30 ratio (lignin: keratin, v/v), yielding 70 ml of keratin and 30ml of lignin solution, respectively. To ensure equal dispersion, the liquid was homogenized using a magnetic stirrer at 30°C.

### 2.5 Crosslinking Treatment:

The blended solution was separated into parts and treated with various crosslinking agents to determine their structural stabilizing effects. Four crosslinkers were tested: glutaraldehyde, acetaldehyde, acetic acid, and polyvinyl alcohol (PVA) (Su et al., 2014.). Each crosslinker was added at varied quantities (0.2-0.4 ml) in table 2 with continuous stirring.
(Don et al., 2006; Dou et al., 2015)

### 2.6 Film Casting and Drying:

The crosslinked mixes were cast into sterile Petri plates, resulting in thin polymer mats. The cast solutions were then dried in a hot air oven at 60 °C overnight, resulting in stable keratin-lignin bio composite suitable for further applications.

### 2.7 Biodegradation testing mass-loss method:

Three mat specimens (S1, S2, and S3) of size 2X2cm were incubated in soil to determine their biodegradation rates. The initial mass of each specimen was properly measured using an analytical balance on the first day of the experiment. After a 10-day soil burial period, the specimens were carefully recovered and gently washed to remove any soil particles that had adhered to. The samples final masses were recorded. The percentage mass loss was computed using the difference between the initial and end weights by incorporating in the given formula (Chowdhury et al., 2022). Triplicate measurements were taken, and the findings are shown as mean ± standard deviation (SD)(Vasek, 2011). Table 3 summarizes the original and final weights percentage.

Percentage mass loss: $Biodegradability\ (\%) = W1 - W2/W1 \times 100$ (equation 1)

## 3. RESULTS AND DISCUSSION:

### 3.1 Extracted keratin and lignin and its blended solution:

After extraction, the raw materials' initial dry weight was reduced by approximately 5.0% (w/w) for keratin and 26.0% (w/w) for lignin. Figure 1 shows the physical features of the two extracted biomaterials. The lignin appeared as a granular, light-brown particle mass, whereas the keratin sample was fibrous and light white in color. The keratin-lignin blends were prepared in various proportions Table 1, with the 70:30 ratio demonstrating the best balance of biomaterial forming ability and structural stability. As a result, this optimized composition was chosen for further mat biopolymer development, with several crosslinking agents like (formaldehyde, glutaraldehyde, acetic acid, and polyvinyl alcohol) introduced to test their effect on material qualities.

**Figure 1: The extracted keratin and lignin image and their prepared blend image**

### 3.2 FT-IR spectrophotometry reveals the molecular interaction of keratin-lignin blends.

The FTIR spectra of lignin (L), keratin (K), and their crosslinked blends with formaldehyde (F), glutaraldehyde (G), acetic acid (A), and polyvinyl alcohol (P) show distinctive functional group changes, indicating successful network formation Figure 2. Lignin exhibited a large O-H stretching band (3600-3200 cm⁻¹), aromatic skeletal vibrations at 1600 and 1510 cm⁻¹, and C-O stretching in guaiacyl/syringyl units at 1265-1030 cm⁻¹, indicating its polyphenolic structure(Gea et al., 2020). Keratin's peptide backbone was confirmed by the presence of prominent amide I (~1650 cm⁻¹) and amide II (~1540 cm⁻¹) bands, amide III (~1230 cm⁻¹), and a broad N–H/O–H band (~3300 cm⁻¹) (Mattiello et al., 2023). Formaldehyde (F) crosslinking resulted in increased C–O–C bands (1100–1050 cm⁻¹) and new or enhanced absorption at 1640–1655 cm⁻¹ (C=N stretching), which overlapped with amide I and was suggestive of methylene bridges between lignin phenolic hydroxyls and keratin amines via Schiff-base and acetal formation (Hoffman et al., 2015). Due to the dual imine and acetal linkages characteristic of GA in aqueous solutions, glutaraldehyde (G) crosslinked samples showed comparable C=N characteristics, including a broadening of the O–H/N–H band and greater C–O–C vibrations (~1100 cm⁻¹) (Migneault & Waldron, 2004).

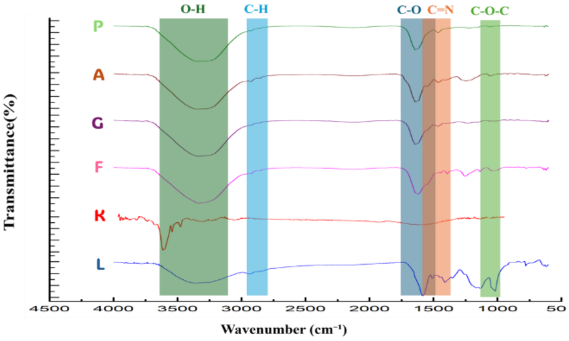

**Figure 2: FTIR analysis of blend graph L- Lignin, K- Keratin, F- Formaldehyde, G- Glutaraldehyde, A- acetic acid, P- Poly vinyl alcohol**

Due to OH consumption and changed H-bonding, acetylation of lignin/keratin side-chain hydroxyls decreases or broadens the O–H stretch (~3300 cm⁻¹) and introduces ester carbonyls, which produce a new or intensified band at ~1730–1740 cm⁻¹. It is typical to see increased intensity at ~1225–1260 cm⁻¹ (C–O of esters/aryl–O–C). These modifications show partial acetylation, which decreases lignin polarity and permits ester bonds to keratin side chains, improving K–L compatibility (Bennani et al., 2023; Md Salim et al., 2021; Schwanninger et al., 2011). Through acetal bridges between PVA-OH and aldehyde carbons, PVA can chemically crosslink in the presence of dialdehydes (F or G) and blend physically (H-bonding). This broadens/shifts the O–H band ~3200–3450 cm⁻¹ and increases ~1090–1141 cm⁻¹ (C–O/C–O–C). A 1730 cm⁻¹ shoulder may result from residual acetate groups in commercial PVA. With F/G present, further acetal/C=N contributions arise between 1100 and ~1640–1660 cm⁻¹. In K–L–PVA systems, expect enhanced ~1090–1141 cm⁻¹ and broader 3300 cm⁻¹ from substantial H-bond networks involving PVA-OH, lignin-OH, and keratin C=O/N–H (Rahmani et al., 2024).

### 3.3 X-Ray Diffraction (XRD) analysis investigation:

Figure 3 shows the XRD profiles of lignin (L), keratin (K), and their crosslinked blends with formaldehyde, glutaraldehyde, acetic acid, and polyvinyl alcohol. The diffraction pattern of lignin revealed a broad, diffuse halo, confirming its amorphous character, which is explained by the uneven organization of its aromatic phenylpropanoid units and the lack of long-range order. Keratin showed faint and broad peaks around 2θ = 10-20°, indicating the existence of organized α-helix and β-sheet domains in the protein backbone. These findings suggest that both pure lignin and keratin have primarily amorphous structures, which are consistent with prior studies on biomass-derived biopolymers.

The diffraction profiles changed significantly after blending and crosslinking. The keratin-lignin blend crosslinked with formaldehyde (F) showed a larger and less pronounced peak than the native polymers. This drop in peak sharpness reflects the breakdown of molecular order caused by formaldehyde-induced methylol couplings, which limit polymer chain alignment and lower crystallinity. Similarly, the glutaraldehyde crosslinked blend (G) showed a broad halo with a little shift in location, demonstrating that covalent crosslinking between aldehyde and hydroxyl/amino groups reduces crystalline packing. Both F and G blends illustrate the efficacy of aldehyde crosslinkers in generating an amorphous, network-like structure.

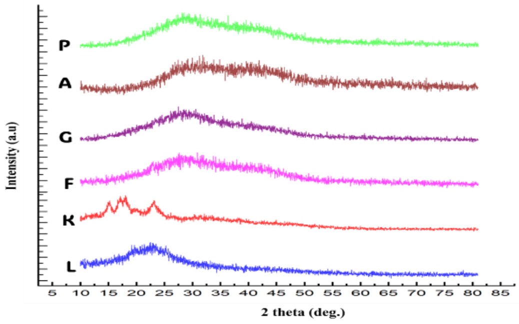

**Figure 3: X-Ray Diffraction XRD analysis of blend graph L- Lignin, K- Keratin, F- Formaldehyde, G- Glutaraldehyde, A- acetic acid, P- Poly vinyl alcohol**

The acetic acid-treated blend (A) had a diffraction profile like the native polymers, but with a small broadening of the distinctive halo. This shows that acetic acid primarily functions as a plasticizing and solubilizing medium, boosting hydrogen bonding interactions while avoiding real covalent crosslinks. In contrast, the polyvinyl alcohol blend (P) preserved a more significant diffraction peak about 2θ = 19-20°, corresponding to the semi-crystalline portions of PVA. However, the peak intensity was significantly reduced, indicating that the addition of keratin and lignin disturbed the crystalline domains of PVA and increased the amorphous fraction of the composite. Overall, the XRD data show that keratin and lignin are naturally amorphous, and the addition of crosslinking agents further lowers crystalline characteristics by limiting molecular ordering. The PVA composite retained partial crystallinity due to PVA's natural semi-crystalline structure, but the formaldehyde and glutaraldehyde systems suppressed crystalline domains the most. These results demonstrate that chemical crosslinking increases structural disorder and aids in the production of more amorphous, flexible, and stable keratin-lignin biomaterials. (Donato & Mija, 2020; Li et al., 2019.; Su et al., 2013)

### 3.3 Prepared keratin and lignin blend matte:

The prepared keratin and lignin blend of 70:30 ratio undergoes mat biopolymer formation where four crosslinkers were tested glutaraldehyde, acetaldehyde, acetic acid, and polyvinyl alcohol figure 4, table 2 in which the formaldehyde-based mat showed the better formation.

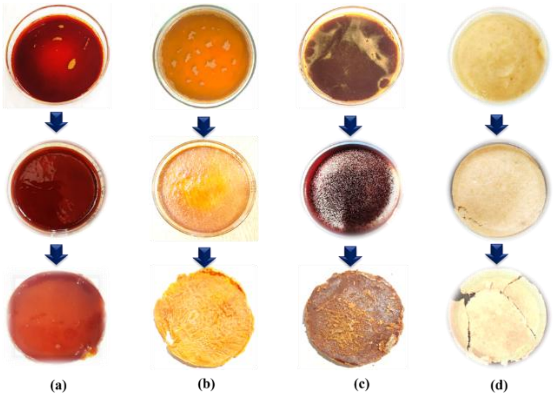

***Figure 4 The keratin -lignin blend based prepared mat,* (a)Formaldehyde, (b)Glutaraldehyde, (c)Acetic acid, (d)Poly vinyl alcohol**

**Table 1: Comparative analysis of keratin-lignin mix ratios for matte forming ability and stability.**

| Keratin: lignin ratio | Film formation | Structural stability | Observations |
|---|---|---|---|
| **90:10** | Film forms but brittle | Low | High Keratin leads to fragile, crack-prone mat |
| **80:20** | Moderate | Improved | Slight increase in flexibility, but poor homogeneity |
| **70:30** | **Good film formation** | **High** | **Good balance between flexibility, stability, and miscibility** |
| **60:40** | Poor | Low | Excess lignin increases brittleness, reduces cohesion |
| **50:50** | Very poor | Very low | Phase separation observed, unstable mat |

**Table 2: The effects of various crosslinking agents and concentrations on the structural and functional properties of keratin-lignin (70:30) blend films.**

| Crosslinker | Concentrations | Expected Outcome in 70:30 Blend |
|---|---|---|
| **Glutaraldehyde** | 0.2ml | Improved rigidity, reduced water solubility, more amorphous structure. |
| **Acetaldehyde** | 0.2ml | Moderate crosslinking, balanced flexibility and stability. |
| **Acetic Acid** | 0.3ml | Increased flexibility, but less structural consolidation. |
| **PVA** | 0.4ml | Improved mat integrity and compatibility, with diminished crystallinity. |

### 3.4 Quantitative findings of biodegradability test:

Mass-loss measurements revealed uneven degradation among the three specimens. S1 dropped 31.6% of its initial mass (0.38 → 0.26 g), S2 lost 23.1% (0.26 → 0.20 g), and S3 lost 50.0% (0.28 → 0.14 g). The average mass loss for the three specimens was 34.9 ± 13.8% (SD, n = 3). In terms of mass remaining (Day 10 / Day 0 × 100), samples retained 68.4% (S1), 76.9% (S2), and 50.0% (S3) of their starting mass. These data reveal figure 5, figure 6, table 3 varying susceptibility to biodegradation throughout the replicates, with S3 demonstrating the fastest breakdown under the test conditions.

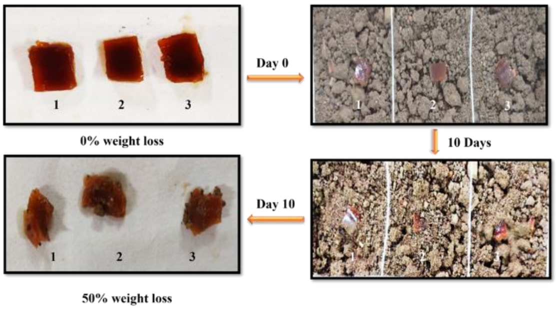

**Figure 5: The biodegradation of keratin lignin blended mat crosslinked with formaldehyde shows 50% degradation over 10days.**

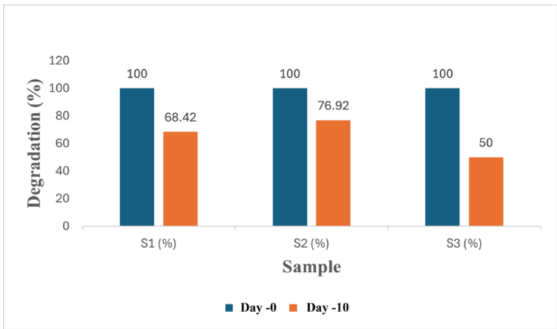

**Figure 6: The graph given of mat biodegradability shows the series of S1, S2, S3 from day -0 to day- 10.**

**Table 3: Biodegradability testing indicating the initial weight and final weight percentage of sample 1,2, and 3.**

| Day | S1(%) | S2(%) | S3(%) |
|---|---|---|---|
| 0 | 100.00 | 100.00 | 100.00 |
| 10 | 68.42 | 76.92 | 50.00 |

## 4. CONCLUSION:

This research illustrates the successful extraction, mixing, and crosslinking of keratin and lignin into functional biodegradable films. The optimized 70:30 keratin-lignin ratio improved film integrity and crosslinking dramatically increased network formation. FTIR and XRD measurements confirmed that formaldehyde produced the most homogeneous and stable mat of the agents tested. Biodegradability tests demonstrated effective decomposition under soil burial circumstances, demonstrating the material's eco-friendliness. Keratin-lignin composites crosslinked using green techniques may find use in sustainable packaging, agricultural mulching films, and biomedical scaffolds. Future research should focus on replacing harmful aldehyde crosslinkers with safer bio-based alternatives, increasing scalability, and customizing characteristics for a wide range of industrial applications.

### Author Contributions:

Olibha Xalxo: execution and conducting of all relevant experiments and the article writing.

Kush Kumar Nayak: identification of research gaps and designing of methodology and supervision of all experiments.

Varaprasad Kolla: Supervision of all work and guidance in article writing and preparation.

## CONFLICTS OF INTEREST:

There are no conflicts to declare.

### Data Availability:

The data supporting this article is available in the Supporting Information section.

## REFRENCES:

1. Bennani, G., Brassard, P., Ndao, A., Konan, D., Leroux, É., Godbout, S., & Adjallé, K. (2023). *Acetic Acid Production From Lignocellulosic Biomass Pyrolysis and Recovery via Membrane Processes*. https://doi.org/10.20944/preprints202309.0238.v1
2. Chincholikar, P., Singh, K. R. B., Natarajan, A., Kerry, R. G., Singh, J., Malviya, J., & Singh, R. P. (2023). Green nanobiopolymers for ecological applications: a step towards a sustainable environment. In *RSC Advances* (Vol. 13, Issue 18, pp. 12411–12429). Royal Society of Chemistry. https://doi.org/10.1039/d2ra07707h
3. Chinomso Iroegbu, A. O., & Ray, S. S. (2022). Lignin and Keratin-Based Materials in Transient Devices and Disposables: Recent Advances Toward Materials and Environmental Sustainability. *ACS Omega*, *7*(13), 10854–10863. https://doi.org/10.1021/ACSOMEGA.1C07372
4. Chowdhury, M. A., Hossain, N., Noman, T. I., Hasan, A., Shafiul, A., & Mohammod Abul, K. (2022). Biodegradable, physical and microbial analysis of tamarind seed starch infused eco-friendly bioplastics by different percentage of Arjuna powder. *Results in Engineering*, *13*. https://doi.org/10.1016/j.rineng.2022.100387
5. de Oliveira, D. R., Avelino, F., Mazzetto, S. E., & Lomonaco, D. (2020). Microwave-assisted selective acetylation of Kraft lignin: Acetic acid as a sustainable reactant for lignin valorization. *International Journal of*

*Biological Macromolecules*, *164*, 1536–1544. https://doi.org/10.1016/j.ijbiomac.2020.07.216

6. Don, T. M., King, C. F., Chiu, W. Y., & Peng, C. A. (2006). Preparation and characterization of chitosan-g-poly (vinyl alcohol)/poly (vinyl alcohol) blends used for the evaluation of blood-contacting compatibility. *Carbohydrate Polymers*, *63*(3), 331–339. https://doi.org/10.1016/j.carbpol.2005.08.023
7. Donato, R. K., & Mija, A. (2020). Keratin associations with synthetic, biosynthetic and natural polymers: An extensive review. In *Polymers* (Vol. 12, Issue 1). MDPI AG. https://doi.org/10.3390/polym12010032
8. Dou, Y., Zhang, B., He, M., Yin, G., Cui, Y., & Savina, I. N. (2015). Keratin/polyvinyl alcohol blend films cross-linked by dialdehyde starch and their potential application for drug release. *Polymers*, *7*(3), 580–591. https://doi.org/10.3390/polym7030580
9. Gea, S., Siregar, A. H., Zaidar, E., Harahap, M., Indrawan, D. P., & Perangin-Angin, Y. A. (2020). Isolation and characterisation of cellulose nanofibre and lignin from oil palm empty fruit bunches. *Materials*, *13*(10). https://doi.org/10.3390/ma13102290
10. Grabska-Zielí Nska, S., Sionkowska, A., Olewnik-Kruszkowska, E., Reczy´nska, K., Reczy´nska, R., & Zbieta Pamuła, E. (2021). Is Dialdehyde Chitosan a Good Substance to Modify Physicochemical Properties of Biopolymeric Materials? *International Journal of Molecular Sciences Article J. Mol. Sci*. https://doi.org/10.3390/ijms
11. Grabska-Zielińska, S. (2024). Cross-Linking Agents in Three-Component Materials Dedicated to Biomedical Applications: A Review. In *Polymers* (Vol. 16, Issue 18). Multidisciplinary Digital Publishing Institute (MDPI). https://doi.org/10.3390/polym16182679
12. Hoffman, E. A., Frey, B. L., Smith, L. M., & Auble, D. T. (2015). Formaldehyde crosslinking: A tool for the study of chromatin complexes. In *Journal of Biological Chemistry* (Vol. 290, Issue 44, pp. 26404–26411). American Society for Biochemistry and Molecular Biology Inc. https://doi.org/10.1074/jbc.R115.651679
13. Katoh, K., Shibayama, M., Tanabe, T., & Yamauchi, K. (2003). *Preparation and Properties of Keratin-Poly (vinyl alcohol) Blend Fiber*.
14. Kawamura, T., Yunoki, S., Ohyabu, Y., Uraoka, T., & Muramatsu, K. (2021). Crosslinking efficacy and cytotoxicity of genipin and its activated form prepared by warming it in a phosphate buffer: A comparative study. *Materials*, *14*(21). https://doi.org/10.3390/ma14216600
15. Li, W., Chen, Y., & Zhang, S. (n.d.). *Hydrolysis of Abandoned Bovine Hair by Pulping Spent Liquor and Preparation of Degradable Keratin-based Sprayable Mulch Film*.
16. Mattiello, S., Guzzini, A., Del Giudice, A., Santulli, C., Antonini, M., Lupidi, G., & Gunnella, R. (2023). Physico-Chemical Characterization of Keratin from Wool and Chicken Feathers Extracted Using Refined Chemical Methods. *Polymers*, *15*(1). https://doi.org/10.3390/polym15010181
17. Md Salim, R., Asik, J., & Sarjadi, M. S. (2021). Chemical functional groups of extractives, cellulose and lignin extracted from native Leucaena leucocephala bark. *Wood Science and Technology*, *55*(2), 295–313. https://doi.org/10.1007/s00226-020-01258-2
18. Migneault, I., & Waldron, K. C. (2004). *Glutaraldehyde: Behavior in aqueous solution, reaction with proteins, and application to enzyme crosslinking*. https://doi.org/10.2144/3705A0790
19. Nasalapure, A. V., Chalannavar, R. K., Kasai, D. R., Reddy, K. R., & Raghu, A. V. (2021). Novel polymeric hydrogel composites: Synthesis, physicochemical, mechanical and biocompatible properties. *Nano Express*, *2*(3). https://doi.org/10.1088/2632-959X/ac11bf
20. Nazimudheen, G., Sekhar, N. C., Sunny, A., Kallingal, A., & Hasanath, B. (2020). ScienceDirect Physiochemical characterization and thermal kinetics of lignin recovered from sustainable agrowaste for bioenergy applications. *International Journal of Hydrogen Energy*, *xxxx*. https://doi.org/10.1016/j.ijhydene.2020.03.172
21. Nuutinen, E. M., Valle-Delgado, J. J., Kellock, M., Farooq, M., & Österberg, M. (2022). Affinity of Keratin Peptides for Cellulose and Lignin: A Fundamental Study toward Advanced Bio-Based Materials. *Langmuir*, *38*(32), 9917–9927. https://doi.org/10.1021/acs.langmuir.2c01140
22. Rahmani, A., Abdulkhani, A., Ashori, A., & Hosseinzadeh, J. (2024). Development of high-performance biocomposites through lignin modification and fiber reinforcement. *Scientific Reports*, *14*(1). https://doi.org/10.1038/s41598-024-80256-x
23. Schwanninger, M., Stefke, B., & Hinterstoisser, B. (2011). Qualitative assessment of acetylated wood with infrared spectroscopic methods. *Journal of Near Infrared Spectroscopy*, *19*(5), 349–357. https://doi.org/10.1255/jnirs.942
24. Stanzione, J., & La Scala, J. (2016). Sustainable polymers and polymer science: Dedicated to the life and work of Richard P. Wool. In *Journal of Applied Polymer Science* (Vol. 133, Issue 45). John Wiley and Sons Inc. https://doi.org/10.1002/app.44212
25. Steitz, M., Zouhair, S., Khan, M. B., Breitenstein-Attach, A., Fritsch, K., Tuladhar, S. R., Wulsten, D., Wolkers, W. F., Sun, X., Hao, Y., Emeis, J., Lange, H. E., Berger, F., & Schmitt, B. (2023). A Glutaraldehyde-Free Crosslinking Method for the Treatment of Collagen-Based Biomaterials for Clinical Application. *Bioengineering*, *10*(11). https://doi.org/10.3390/bioengineering10111247
26. Su, L., & Fang, G. (2014.). *Characterization of Cross-linked Alkaline Lignin/Poly (Vinyl Alcohol) Film with a Formaldehyde Cross-linker*.
27. Su, L., Xing, Z., Wang, D., Xu, G., Ren, S., & Fang, G. (n.d.). *Mechanical Properties Research and Structural Characterization of Alkali Lignin / Poly (vinyl alcohol) Reaction Films*.
28. Tabassum, Z., Mohan, A., Mamidi, N., Khosla, A., Kumar, A., Solanki, P. R., Malik, T., & Girdhar, M. (2023). Recent trends in nanocomposite packaging films utilising waste generated biopolymers: Industrial symbiosis and its implication in sustainability. In *IET Nanobiotechnology* (Vol. 17, Issue 3, pp. 127–153). John Wiley and Sons Inc. https://doi.org/10.1049/nbt2.12122
29. Thonpho, A., Srihanam, P., & Songeon, W. (n.d.). *Effect of cross-linked agents on keratin films property*. https://www.researchgate.net/publication/299174350
30. Trzcinski, A. P., & Stuckey, D. C. (2015). Contribution of acetic acid to the hydrolysis of lignocellulosic biomass under abiotic conditions. *Bioresource Technology*, *185*, 441–444. https://doi.org/10.1016/j.biortech.2015.03.016
31. Vasek, Vladimir. (2011). *Recent researches in energy environment entrepreneurship innovation: International conference on energy, environment, entrepreneurship, innovation (ICEEEI '11)*. WSEAS Press.