Journal of Scientific & Industrial Research
Vol. 81, May 2022, pp. 455-461

Synthesis and Characterization of Edible Films from Garlic*(Allium sativum)* Husk Components

Anand Kumar[1], Gopesh Patel[1], Madhuresh Dwivedi [1]*, Salman Hashmi[2] and Rama Chandra Pradhan[1]

[1]Department of Food Process Engineering, National Institute of Technology, Rourkela 769 008, Odisha, India

[2]Centre of Food Technology, Institute of Professional Studies, University of Allahabad, Allahabad 211 002, UP, India

Received 18 January 2022; revised 31 March 2022; accepted 04 April 2022

Garlic husks are waste biomass and unutilized sources of essential compounds like lignin, pectin, cellulose, hemicellulose, lignocellulose and dietary fibre. Pectin was extracted from garlic husk using ammonium oxalate solution. Lignin is a complex biopolymer consisting of p-coumaryl alcohol, sinapyl alcohol, and coniferyl alcohol. Lignin was extracted from garlic husk using alkali pulping method; the sample was subjected to 10% (w/w) NaOH solution in a pressurized condition at high temperature. The extracted lignin was characterized using Fourier Transform Infrared Spectroscopy (FTIR). Transmittance ratios in FTIR Spectra of extracted lignin were similar to those of the commercial lignin. The edible film was prepared by solution casting method where the film was casted from the solution of pectin and lignin. The physical and chemical properties of the film like moisture content, color, solubility of the film, antimicrobial activity, water vapour transmission rate, pectin and lignin were characterized. The lowest water vapour permeability of the film is 17.48 ± 0.12 g/h m^2. Whiteness index and 'L' value of film decreased from film 1 to film 4. An inhibition zone was formed against the *Bacillus cereus* proving the antimicrobial activity of the lignin extracted from garlic husk.

Keywords: Antimicrobial activity, Biodegradable film, Lignin extraction, Packaging, Valorization

Introduction

Waste utilization is essential in every aspect to maintain the process sustainable towards achieving less environmental impact and ecological footprint and is a process of using the biomass for other processes directly or altering it into another form, where it is applicable in a particular field.[1] The composition of biomass determines its application. Food waste is generated throughout the supply chain, from the farm field to consumption in the household. With biomass from fruits and vegetables contains a significant amount of pectin, and a minor amount of lignocellulose materials; one example is garlic husk waste which is produced throughout the chain that includes the agricultural field, processing industries and household consumptions.[2,3]

Garlic (*Allium sativum*) is an indigenous vegetable plant of central Asia and northeast Iran. China is the leading producer of garlic in the world, while India holds the second position with 2.91 million tons per annuum.[4] One kg of garlic produces about 240 g of husk, which includes husk and straw.[5] Garlic husk is rich in pectin and lignin.[2,6] Mostly this husk is burnt into ashes or dumped in the soil.

Lignin is an aromatic biopolymer mainly present in plant cell walls which is packed tightly with cellulose and hemicellulose present in the plant.[1] It is used to produce biofuel, wastewater treatment, asphalt emulsification, dye-water reduction, controlled release of fertilizers and pesticides, drug delivery as antiviral, anticancer, antioxidants and antibacterial. It can also be used for pet and human food as a fibre source and can also be used in different kinds of packaging solutions.[7] The three monomers of lignin are namely p-coumaryl, coniferyl, and sinapyl alcohol.[8] It possesses antimicrobial and antioxidant activity due to its phenolic compounds. Coating solutions prepared with lignin extracted from sugarcane bagasse have shown antifungal activity.[9] It has an antiradical activity that inhibits lipid oxidation and also shows antimicrobial activity against the *Listeria monocytogenes, Staphylococcus aureus and Bacillus spp*.[10]

Pectin is present in the plant cell wall, responsible for tissue integrity and firmness.[11] It is used as a gelling agent, stabilizer, emulsifier and cation-binding agent. It is consumed over 20000 tons per year and its consumption is getting higher gradually every year. It is also present in the peels and pomace of utilized fruits.[12]

*Author for Correspondence
E-mail: madhureshd@gmail.com

Biodegradable packaging is one of the vital recommendations of all environmental-based organizations. The food packaging must be made sustainable by utilizing waste from the food industry, such as whey protein from milk and fruit and vegetable peels from F and V processing industries.[13] Replacing inorganic polymer packaging material with biopolymer is essential as it has significant limitations of recyclability and biodegradability.[14,15] Plastics have entered the human food chain. Edible packaging is one of the desired applications for extracted pectin and lignin from the biomass of garlic husk. The casting of edible film requires a polysaccharide or a protein as a significant compound to form a structure.[16]

Lignin from the garlic husk (Yamuna Safed (G-1)) has not been extracted yet. The pectin and lignin extracted from the garlic husk have not been utilized for the edible film casting. The objective of the present work is to use garlic husk for edible film preparation by extracting pectin and lignin and studying the properties of the prepared film.

Materials and Methods

Raw Materials

About 10 kg of Garlic husk (skin) was collected from the market area of Rourkela, Odisha. Commercial lignin (alkali) of 500 g was purchased from Sigma Aldrich. Ammonium oxalate, sodium hydroxide, commercial pectin and guar gum were purchased from HiMedia Laboratories. Lignin was extracted from garlic husk by alkaline pulping method at high temperature with high pressure.[2] *Bacillus cereus* colonies are large, feathery, dull, gray, granular, spreading colonies and opaque with a rough matted surface and irregular perimeters.[17] Inoculum of *B. cereus* were prepared by transferring a frozen culture to nutrient agar media by spread plate method and incubated at 35°C for 24 hours. DMSO (dimethyl sulfoxide) was used for dissolving lignin for immersing filter paper in it.[18]

Pectin Extraction

Garlic husk was washed and dried at 103 ± 2°C for 24 h. Pectin extraction is based on the method[19] and also 30g garlic husk was subjected to hydrothermal pretreatment in 500 ml of solution and subsequently treated with the solution containing 0.5% (w/v) of ammonium oxalate dissolved in the distilled water. The extraction was carried out in a water bath at 95°C for 30 min. The extraction process was repeated 3–4 times to obtain higher pectic substances. After treatment, the solution was cooled down to room temperature. The extracts were separated from garlic residues by filtration. For pectin separation, the solution containing ethanol of 95% (v/v) with 0.1N HCl (hydrochloric acid) was added to the extract. Pectin is precipitated and dried for 8 h at 40°C.

Lignin Extraction

The residue obtained from the pectin extraction process was utilized again for the lignin extraction. The residue was dried at 80°C for 24 h before processing. The sample was subjected to hydrothermal pre-treatment in a solution containing 10% (w/w) sodium hydroxide with distilled water and a 500 ml conical flask used in the extraction process. The extraction process was carried out in the autoclave at 121°C and 15 psi for 30 minutes. After autoclave treatment, the solution was cooled down to room temperature. The dissolved lignin alkaline solution was separated from the solid particles by vacuum filtration. The filtered liquid part is known as black liquor and mainly consists of lignin. The pH of black liquor was brought below two using concentrated sulphuric acid. Precipitated acid-insoluble lignin was separated and washed using the centrifugation process at 5000 rpm for 20 minutes. The final solid lignin was dried in a hot air oven at 60°C for 8 hours.

Edible Film Preparation

Guar gum solution 1% (w/v) was prepared by dissolving 1 g of guar gum in 48 ml of distilled water. Pectin flakes of 10% (w/v) were dissolved to 40 ml of distilled water. The two dispersions were homogenized using a magnetic stirrer for 60 min (600 rpm). Lignin solution of 0.25% (w/v) was prepared by dissolving 0.25 g of lignin in 9.75 ml of 1 M sodium hydroxide solution. The lignin solution was added dropwise during homogenization. Glycerol was added to the mixture as a plasticizer at a concentration of 1% (w/v). The film-forming solution was subjected to ultrasonication treatment at the amplitude of 30 for a pulse time of 10 s. The mixture was used to prepare the film 2. The film was prepared by the solution casting method. The dried films were moved to the desiccator and removed from the casted Petri plate. The film was prepared at the four different lignin percentages. Film 1 has 0% (w/v) of lignin content, film 2 has 0.25% (w/v) of the lignin content, Film 3 has 0.5 % (w/v) of the lignin content, and film 4 has 1% (w/v) of lignin content.[20] The formulation

percentage is attached in Table 1. The prepared edible film with different concentrations is shown in Fig. 1.

Film Characterization

Moisture content and solubility of the film

Each film was cut into 2 cm^2 each, and its initial weight was recorded before the film was heated in a hot air oven set to 105 ± 2°C. The dried sample's final weight was measured after 24 hours. The moisture content of the sample is calculated using the following equation.[21]

$$Moisture\ content = \frac{Initial\ weight - Final\ weight}{Initial\ weight} \times 100 \quad \ldots (1)$$

The completely dried film was used for determining the water solubility. The film was immersed in 50 ml of distilled water filled in a beaker. The beaker was placed in the incubator for maintaining a constant environment under shaking conditions (50 rpm). The suspension was filtered after 24 hours using filter paper (Whatman #1, GE Healthcare Life Sciences) to recover the residue. The residue was dried in a hot air oven at 105 ± 2°C for 24 hours. The dried residue was kept in a desiccator and weighed. The following equation is used to determine the water solubility of the film.[22]

$$Water\ solubility\ (\%) = \frac{WDF - WDR}{WDF} \times 100 \quad \ldots (2)$$

where, WDF is the weight of the dried film and WDR is the weight of the dried residue.

Optical Properties

The colour values of the prepared edible films were expressed as L*, a*, and b*. The colour values were calculated using Matlab software. The scale 'L' represents (lightness to darkness) while chromaticity parameters 'a' represents (red-green) and 'b' represents (yellow-blue). The total colour change (ΔE), whiteness index and yellowness index of the films were calculated using the following equations[23]:

$$\Delta E = \sqrt{(L^* - L)^2 + (a^* - a)^2 + (b^* - b)^2}\ . \quad \ldots (3)$$

$$WI = 100 - [(100 - L^*)^2 + a^{*2} + b^{*2}]^{0.5} \quad \ldots (4)$$

$$YI = 142.86b \times L^{-1} \quad \ldots (5)$$

Fourier Transform Infrared Spectroscopy

Lignin and pectin extracted from the garlic husk were confirmed and characterized by FTIR spectroscopy. The commercial pectin and lignin are considered as a standard and compared for similarities in peak value. The edible film was characterized through FTIR spectroscopy. Functional groups of lignin, pectin present in the edible film were analyzed at 4000 – 400 cm^{-1} wave number at 4 cm^{-1} resolution.[24]

Water Vapour Transmission Rate (WVTR)

Films were wrapped on the 10ml glass bottles filled with water. Sealed glass bottles were placed at constant RH (50%) and temperature (25°C). The weight of the glass bottles was measured periodically for each hour. The graph is plotted according to the change in weight per unit hour. The slope of the graph divided by the area of the film-covered bottle's mouth determines the water vapour permeability.[20]

Antimicrobial Activity of Garlic Extract (Lignin) in Film

The inoculum of *Bacillus cereus* was prepared by transferring a frozen culture to the nutrient broth and

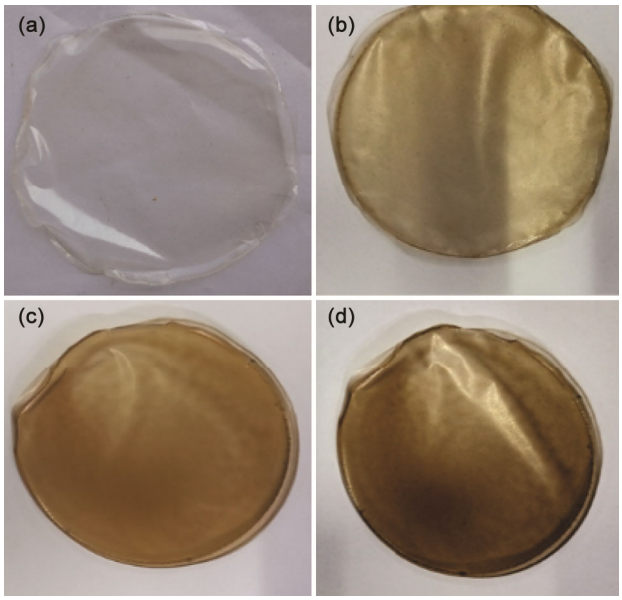

Fig. 1 — Edible film with: (a) only pectin, (b) 0.25% lignin, (c) 0.50% lignin, (d) 1% of lignin

Table 1 — Formulation table of prepared edible film

Film types	Pectin solution (g)	Guar gum solution (g)	Lignin % (w/w)	Glycerol (g)
Film 1	49	50	0	1
Film 2	49	50	0.25%	1
Film 3	49	50	0.50%	1
Film 4	49	50	1.00 %	1

incubated at 35°C for 24 hours. Autoclaved nutrient agar was poured into plates and solidified. Organisms were spread over the surface by a sterile L rod. The filter paper was soaked in the lignin solution; the lignin solution was prepared by dissolving 0.1 g of lignin in 1 ml of DMSO (dimethyl sulfoxide) solution. The filter paper was placed in the culture added Petri plate and incubated at 35°C for 24 hours. A zone of inhibition was formed around the extract; it was used for [1,25] determining the activity of the extracted lignin compound.

Statistical Analysis

All the parameters have been reported as mean ± standard deviation. One way ANOVA and Tukey's HSD (honestly significant difference) multiple comparison test were performed in order to evaluate the statistical differences between the treatments using SPSS 17.0 for Windows (SPSS Inc., Chicago, IL, USA), $p < 0.05$ considered as the significance level.

Results and Discussion

Thickness, Moisture Content and Water Solubility

The moisture content of the Film 1 was higher as pectin had absorbed more moisture towards itself. Film 1 contained pectin and guar gum. Film 2, 3, and 4 had less moisture than control film that had lignin compound. The film's moisture content depends on the compounds present in the film. The addition of lignin-free hydrophilic groups would have formed bonding with lignin molecules via hydrogen and hydrophobic interaction, reducing the adhesion of water molecules.[26] It might be the reason for the low moisture content on the lignin added film. The solubility of the film also depends on the compounds present in the film. The lignin is soluble in alkaline solutions and organic solvents.[27] The water solubility of acacia lignin film was lower than the alginate film. The solubility of the film decreased due to the presence of lignin content in the film. The solubility of film 1 was higher compared to other films. Film 4 had lower solubility due to higher lignin content. Pectin, guar gum and glycerol were soluble in water. There was a significant difference between film 1 and film 4 for water solubility. The physical properties of the film are shown in Table 2.

Optical Properties

The colour values of the edible film are presented in Table 3. The lightness value (L) decreased from Film 1 (45.12 ± 1.01) to Film 4 (17.38 ± 4.29)) when lignin content increased. Similarly, the total colour value (ΔE) increased when compared to the standard white plate, and the whiteness index decreased with an increase in the lignin content of the film. The yellowness index increased for higher lignin content in the film. The lignin content is the main factor that affected the film's colour value. Zadeh *et al*.[28] studied the colour value of lignin film, where the film became darker with an increased lignin concentration. Sabiha-Hanim and Siti-Norsafurah[29] concluded that the hemicellulose film's colour was darker than other films because of the high lignin content.

FTIR Characterization

Lignin and pectin extracted from the garlic husk were analyzed through FTIR Spectroscopy. The lignin characteristic peaks, around 3500 – 3100 cm^{-1} represent associated -OH stretching vibrations. The band indicates the presence of alcoholic and phenolic hydroxyl groups involved in hydrogen bonds. This showed the presence of sinapyl and coniferyl alcohol groups in the compound, thus confirming the presence of lignin.[30] The peaks around the 1600 to 1700 cm^{-1} exhibited the presence of carbonyl and carboxyl groups. The peaks present around 1500 cm^{-1} represents the benzene ring, and 1314 cm^{-1} stand for syringyl ring.[31] Deformation vibrations of C-H Bonds are generally present between 988 – 960 cm.[1,32]

Table 2 — Physical properties of the film

Film type	Moisture content (%)	Solubility (%)	Water vapour permeability (g /h m^2)
Film 1	56.30 ± 1.58	93.82 ± 0.48	18.45 ± 0.12
Film 2	44.46 ± 0.82	59.90 ± 1.38	18.28 ± 0.21
Film 3	49.03 ± 1.87	43.73 ± 1.28	17.48 ± 0.12
Film 4	51.02 ± 1.87	35.66 ± 1.46	30.55 ± 0.12

Table 3 — Colour values of the edible film

Film types	L	a	b	ΔE	Whiteness index	Yellowness index
Film 1	45.12 ± 1.01	1.22 ± 0.02	−3.52 ± 0.07	46.30 ± 1.00	44.99 ± 1.00	−11.15 ± 0.01
Film 2	39.63 ± 1.44	1.14 ± 0.38	17.75 ± 0.75	55.10 ± 1.11	37.05 ± 1.19	63.98 ± 0.62
Film 3	28.47 ± 1.63	7.33 ± 0.15	21.96 ± 0.30	67.38 ± 1.54	24.81 ± 1.57	110.55 ± 6.29
Film 4	17.38 ± 4.29	8.00 ± 0.79	17.64 ± 1.60	76.77 ± 3.77	15.10 ± 3.87	149.61 ± 18.61

Vibrations of C–O bonds present in primary and secondary alcoholic groups created bands at 1100, 1072 and 1033 cm^{-1}. C-H bonds benzene ring are present around the 900 – 700 cm^{-1}. The FTIR spectra of lignin are plotted in Fig. 2.

Pectin FTIR spectra exhibited characteristic peaks around 3200 – 3600 cm^{-1} assigned for OH bonds. The bands around the 1700–1600 cm^{-1} confirmed the presence of COO- antisymmetric stretching vibration of polygalacturonic acid.[33] The peaks present around 1100 – 1000 cm^{-1} assigned to C-O ring. The peaks present around 950 – 960 cm^{-1} denoted C-O bending vibration. The higher intensity, around 1630 cm^{-1} exhibited the presence of carboxylic groups. The presence of pectin was confirmed by C = O bond and carboxylic groups.[34] The FTIR spectra of extracted pectin are plotted in Fig. 3.

FTIR Spectra of edible film represented both the lignin and pectin IR (Infrared Spectroscopy) peaks. Bands confirmed lignin presence has occurred around 1028 cm^{-1}. The peak shifted from 1032 cm^{-1} to 1028 cm^{-1} was due to the bond created between the lignin and guar gum. The peaks assigned to the OH group were reduced due to hydrogen bonding between lignin and pectin molecules. The peaks present around the 3500 – 3200 cm^{-1} assigned to the alcoholic groups present in the lignin compounds such as coniferyl and sinapyl alcohol. The pectin peak 1648 cm^{-1} occurred due to COO- vibrations thus, confirming the presence of galacturonic acid. The wavelength assigned for functional groups and structures present in the film containing lignin, pectin and guar gum are represented in Table 4. The FTIR spectra of the edible film prepared are shown in Fig. 4.

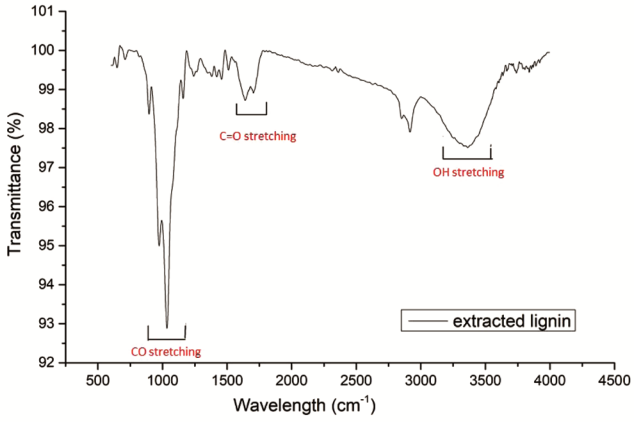

Fig. 2 — Extracted lignin analyzed by FTIR Spectroscopy

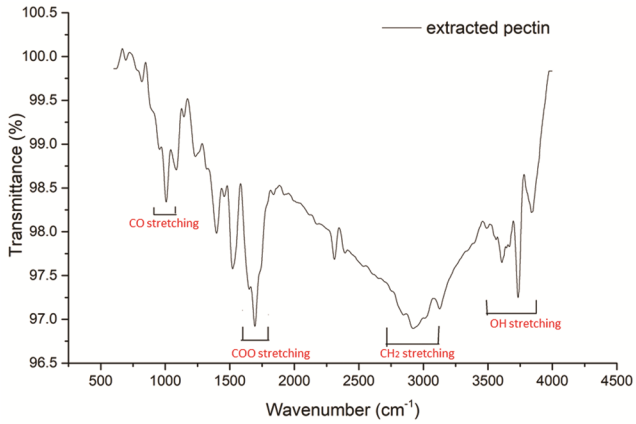

Fig. 3 — Extracted pectin FTIR Spectroscopy

Water Vapour Permeability (WVP)

The film's water vapour permeability was based on the quality of the dispersion of solute content in the film.[35] The lower water vapour permeability of film is necessary to prevent the mass transfer from the food. The permeability had reduced up to approximately 0.5% of lignin content. Still, there was no significant difference between films 1, 2 and 3 (Table 5). The water vapour permeability increased rapidly after 0.5% of the lignin.[28] There was a significant difference between film 3 and film 4 due to variation in the lignin content of the film. The optimum amount

Table 4 — FTIR peaks for functional groups of the compounds

	Wave number (cm^{-1})		
Characteristic functional groups	Lignin	Pectin	Guar gum
O-H stretching	3500 – 3100	3600–3200	3447.61
C-H stretching	988 – 960	3000–2800	—
C-O stretching	1100 – 1000	950 –960	—
C–H bond	—	—	1350-1300
COO^-	—	1648	—

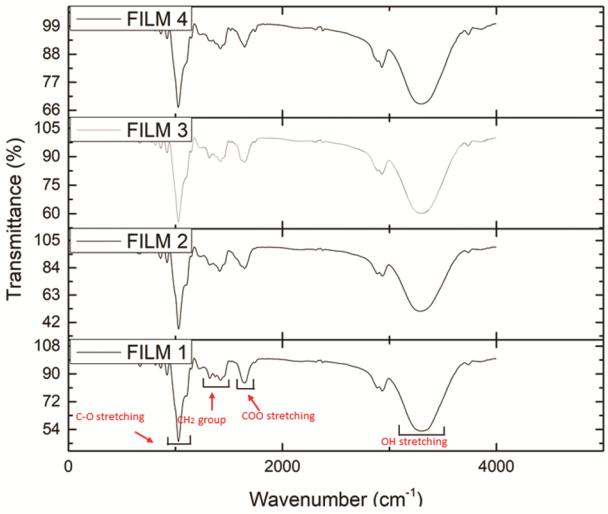

Fig. 4 — FTIR spectroscopy of the edible film

of lignin in the film required for better water vapour permeability might be below 1% of lignin content.

Antimicrobial Activity of the Lignin

Lignin inhibits the growth of *Bacillus cereus*. The percentage of Lignin content in the filter paper influences the zone of inhibition diameter. Lignin inhibits the growth of gram-positive bacteria (*E. coli* and *B. subtilis*), and the Gram-negative, (*Salmonella enterica* and *S. aureus*).[36] as represented in Fig. 5. The inhibitory activity of extracted lignin is lower than that of commercial lignin. Lignin antibacterial activity is due to the presence of phenolic compounds present in it. The presence of a double bond in the Cα = Cβ position of the side chain and a methyl group in the γ position is responsible for its inhibitory activity.[10]

Scanning Electron Micrograph

SEM images of the untreated sample and extracted lignin have shown differences in their surface morphology Fig. 6. The changes observed in raw and treated samples indicate the removal of substances like cellulose, hemicellulose, and other derivatives. The final compound is homogenous, and that is lignin.

Table 5 —Water vapour permeability of the film

Film type	Water vapour permeability (g/h m^2)
Film 1	18.4536 ± 0.0125
Film 2	18.2855 ± 0.0216
Film 3	17.4805 ± 0.0125
Film 4	30.5555 ± 0.0125

Fig. 5 — Activity of lignin on the *Bacillus cereus*

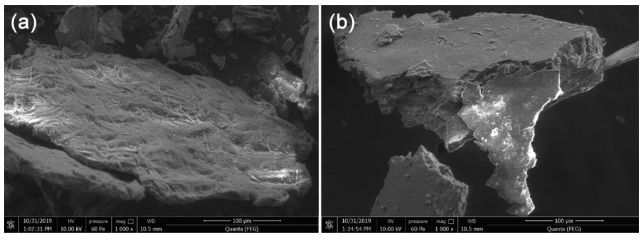

Fig. 6 — (a) SEM image of the pre-treated compound. (b) SEM image of extracted lignin

Conclusions

The utilization of by-products sincerely presents alleviation to agro-food processing industries concerning their infrastructure and economic views as waste storage, and processing are their important issues. Garlic husk is an unutilized waste produced commonly from processing industries and cooking kitchens. This waste had enormous potential for utilizing in the field of food packaging. The water solubility of the film was decreased for obtaining a higher amount of lignin. Lignin content in the film influenced the film's water solubility. Water vapour permeability is affected negatively by a higher percentage of lignin content. Lignin was added as an additive to the film-forming solution for its antimicrobial properties. The Colour value of the film was significantly affected by the lignin content in the film. The whiteness index decreased from film 1 (44.99) to film 4 (15.10). The yellowness index increased from film 1 (–11.15) to film 4 (149.61). It exhibited promising reinforcement abilities to improve mechanical and water vapor barrier properties of the films, due to their high lignin. The remaining residue can be further utilized for ethanol production. The antioxidant activity of lignin in the film can be investigated. However, this methodology, is not designed to be scaled up to industrial settings, such as extrusion, which needs heat-resistant biopolymer ingredients to generate ecologically stable packaging films. The incorporation of plant by-products into bio plastics may hasten the biodegradability of these composite films. As a result, more research is needed to understand the efficacy of the new plant-by-products in real-world food packaging applications.

References

1 Santhosh R, Nath D & Sarkar P, Novel food packaging materials including plant-based byproducts: A review, *Trends Food Sci Technol*, **118** (2021) 471–489.

2 Reddy J P & Rhim J W, Extraction and characterization of cellulose microfibers from agricultural wastes of onion and garlic, *J Nat Fibers*, **15** (2018) 465–473.

3 Senthil C & Lee C W, Biomass-derived biochar materials as sustainable energy sources for electrochemical energy storage devices, *Renew Sustain Energy Rev*, **137** (2021) 110464.

4 Kıraç H, Dalda Şekerci A, Coşkun Ö F & Gülşen O, Morphological and molecular characterization of garlic

(Allium sativum L.) genotypes sampled from Turkey, *Genet Resour Crop Evol*, (2022) doi:10.1007/s10722-022-01343–4.

5 Qureshi A A, Din, Z Z, Abuirmeileh N, Burger W C, Ahmad Y & Elson C E, Suppression of avian hepatic lipid metabolism by solvent extracts of garlic: Impact on serum lipids, *J Nutr*, **113** (1983) 1746–1755.

6 Abdel-Fattah A F & Edrees M, A study on the composition of garlic skins and the structural features of the isolated pectic acid, *J Sci Food Agric*, **23** (1972) 871–877.

7 Wahba S M R, Darwish A S, Shehata I H & Abd Elhalem S S, Sugarcane bagasse lignin, and silica gel and magneto-silica as drug vehicles for development of innocuous methotrexate drug against rheumatoid arthritis disease in albino rats, *Mater Sci Eng C*, **48** (2015) 599–610.

8 Vanholme R, Demedts B, Morreel K, Ralph J & Boerjan W, Lignin biosynthesis and structure, *Plant Physiol*, **153** (2010) 895–905.

9 Jonglertjunya W, Juntong T, Pakkang N, Srimarut N & Sakdaronnarong C, Properties of lignin extracted from sugarcane bagasse and its efficacy in maintaining postharvest quality of limes during storage, *LWT - Food Sci Technol*, **57** (2014) 116–125.

10 Espinoza-Acosta J L, Torres-Chávez P I, Ramírez-Wong B, López-Saiz C M & Montaño-Leyva B, Antioxidant, antimicrobial, and antimutagenic properties of technical lignins and their applications, *Bio Resources*, **11** (2016) 5452–5481.

11 Jolie R P, Duvetter T, Van Loey A M & Hendrickx M E, Pectin methylesterase and its proteinaceous inhibitor: A review, *Carbohydr Res*, **345** (2010) 2583–2595.

12 Ptichkina N M, Markina O A & Rumyantseva G N, Pectin extraction from pumpkin with the aid of microbial enzymes, *Food Hydrocoll*, **22** (2008) 192–195.

13 Rifna E J, Misra N N & Dwivedi M, Recent advances in extraction technologies for recovery of bioactive compounds derived from fruit and vegetable waste peels: A review, *Crit Rev Food Sci Nutr*, (2021) 1–34.

14 Rifna E J & Dwivedi M, Optimization and validation of microwave–vacuum drying process variables for recovery of quality attribute and phytochemical properties in pomegranate peels (Punica granatum L. cv. Kabul), *J Food Meas Charact*, **15** (2021) 4446–4464.

15 Murakonda S & Dwivedi M, Powders from Fruit Waste, in *Food Powders Properties and Characterization* (Springer) 2021, 155–168

16 Rifna E J & Dwivedi M, The microbiological safety of food powders, in *Food powders properties and characterization* (Springer, 2021), 169–193

17 Patra J K & Baek K H, Novel green synthesis of gold nanoparticles using Citrullus lanatus rind and investigation of proteasome inhibitory activity, antibacterial, and antioxidant potential, *Int J Nanomedicine*, **10** (2015) 7253–7264.

18 Alexander M M & Sulebele G A, Pectic substances in onion and garlic skins, *J Sci Food Agric*, **24** (1973) 611–615.

19 Vilas Dhumal C, Pal K & Sarkar P, Synthesis, characterization, and antimicrobial efficacy of composite films from guar gum/sago starch/whey protein isolate loaded with carvacrol, citral and carvacrol-citral mixture, *J Mater Sci Mater Med*, **30** (2019) 1–14.

20 Giteru S G, Coorey R, Bertolatti D, Watkin E, Johnson S & Fang Z, Physicochemical and antimicrobial properties of citral and quercetin incorporated kafirin-based bioactive films, *Food Chem*, **168** (2015) 341–347.

21 Kavoosi G, Dadfar S M M & Purfard A M, Mechanical, physical, antioxidant, and antimicrobial properties of gelatin films incorporated with thymol for potential use as nano wound dressing, *J Food Sci*, **78** (2013) E244–E250.

22 Mehdizadeh T, Tajik H, Razavi Rohani S M & Oromiehie A R, Antibacterial, antioxidant and optical properties of edible starch-chitosan composite film containing Thymus kotschyanus essential oil., in *Veterinary research forum : an International Quarterly Journal* vol. 3 (Faculty of Veterinary Medicine, Urmia University, Urmia, Iran) 2012, 167–73

23 Vanholme R, Morreel K, Ralph J & Boerjan W, Lignin engineering, *Curr Opin Plant Biol*, **11** (2008) 278–285.

24 Alzagameem A, Klein, S E, Bergs M, Do X T, Korte I, Dohlen S, Hüwe C, Kreyenschmidt J. Kamm B, Larkins M & Schulze M, Antimicrobial activity of lignin and lignin-derived cellulose and chitosan composites against selected pathogenic and spoilage microorganisms, *Polymers (Basel)*, **11** (2019) 670.

25 Aadil K R, Prajapati D & Jha H, Improvement of physcio-chemical and functional properties of alginate film by Acacia lignin, *Food Packag Shelf Life*, **10** (2016) 25–33.

26 Sameni J, Krigstin S & Sain M, Solubility of lignin and acetylated lignin in organic solvents, *Bio Resources*, **12** (2017) 1548–1565.

27 Zadeh E M, O'Keefe S F & Kim Y T, Utilization of lignin in biopolymeric packaging films, *ACS Omega*, **3** (2018) 7388–7398.

28 Sabiha-Hanim S & Siti-Norsafurah A M, Physical properties of hemicellulose films from sugarcane bagasse, *Procedia Eng*, **42** (2012) 1390–1395.

29 Derkacheva O & Sukhov D, Investigation of lignins by FTIR spectroscopy, in *Macromolecular Symposia* vol. 265 (Wiley Online Library) 2008, 61–68

30 Lu Y, Lu Y-C, Hu H-Q, Xie F-J, Wei X-Y, Fan X, Structural characterization of lignin and its degradation products with spectroscopic methods, *J Spectrosc*, **2017** (2017).

31 Hergert H L, Infrared spectra of lignin and related compounds. II. conifer lignin and model compounds1,2, *J Org Chem*, **25** (1960) 405–413.

32 Ognyanov M, Georgiev Y, Petkova N, Ivanov I,Vasileva I & Kratchanova M, Isolation and characterization of pectic polysaccharide fraction from in vitro suspension culture of *Fumaria officinalis* L., *Int J Polym Sci*, **2018** (2018).

33 Sato M de F, Rigoni D C, Canteri M H G, Petkowicz C L de O, Nogueira A & Wosiacki G, Caracterização química e instrumental de pectinas isoladas de bagaço de 11 cultivares de maçã, *Acta Sci - Agron*, **33** (2011) 383–389.

34 Park H J & Chinnan M S, Gas and water vapor barrier properties of edible films from protein and cellulosic materials, *J Food Eng*, **25** (1995) 497–507.

35 Lobo F C M, Franco A R, Fernandes E M & Reis R L, An overview of the antimicrobial properties of lignocellulosic materials, *Molecules*, **26** (2021) 1749.