Physicochemical and functional properties of pectin extracted from the edible portions of jackfruit at different stages of maturity

Nidhina K[1,2#], Billu Abraham[1,2], Cynthia Fontes-Candia[3], Antonio Martínez-Abad[3], Marta Martínez-Sanz[3], P Nisha[1,2,3*], Amparo López-Rubio[3*]

[1]CSIR – National Institute for Interdisciplinary Science and Technology (NIIST), Thiruvananthapuram, Kerala 695019, India.

[2]Academy of Scientific and Innovative Research (AcSIR), Ghaziabad - 201002, India.

[3]Food Safety and Preservation Department, IATA-CSIC, Avda. Agustin Escardino 7, 46980 Paterna, Valencia, Spain

ORCID iD of Corresponding Authors:

Amparo López-Rubio: https://orcid.org/0000-0001-6469-9402

P. Nisha: http://orcid.org/0000-0002-9292-2226

Declaration of interest: None

** Correspondence:*

Amparo López-Rubio

Ph: Tel.: +34 963900022; fax: +34 963636301

E-mail: amparo.lopez@iata.csic.es

P. Nisha

Ph: + 91-471-2515348

E-mail: pnisha@niist.res.in; bp.nisha@yahoo.com

Abstract

BACKGROUND

The physico chemical and functional properties of pectin (JFP) extracted from edible portions (including pericarp and seed) of raw jackfruit (underutilized tropical fruit), at four different maturity stages (referred to as stage-I, II, III, and IV), were characterized in terms of extraction yields, chemical composition, molecular weight, functional and antioxidant properties to evaluate its applicability foods.

RESULT

Yield of JFP increased from 9.7 to 21.5% with fruit maturity, accompanied by an increase in the galacturonic acid content (50.1, 57.1, 63.6 and 65.2%) for stages I-IV respectively. The molecular weight increased from 147 kDa in stage-I to 169 kDa in stage-III, but decreased to 114 kDa in stage-IV, probably due to cell wall degradation during maturation. JFP was of the high-methoxyl type and the degree of esterification (DE) increased from 65% to 87% with fruit maturity. The functional properties of JFP were similar or better than those reported for commercial apple pectin, thus highlighting its potential as food additive. Although the phenolics and flavonoids content of JFP decreased with fruit maturity, their antioxidant capacity increased, which may be correlated with the increased content of galacturonic acid upon fruit development. Gels prepared from JFP showed viscoelastic behavior. Depending on maturity stage in which they were obtained, different gelation behavior was seen.

CONCLUSION

The study confirmed the potential of pectin extracted from edible parts of jackfruit as a promising source of high-quality gelling pectin with antioxidant properties, for food applications.

52 Keywords: jackfruit; high-methoxyl pectin; galacturonic acid; degree of esterification;
53 antioxidant activity
54

1. Introduction

Pectin belongs to the family of structural polysaccharides found in the primary cell walls and middle lamellae of higher plants. These polysaccharides include homogalacturonan (HG), rhamnogalacturonan I (RG-I), rhamnogalacturonan II (RG-II), xylogalacturonan (XGA) and apiogalacturonan (AGA). This heterogeneous structure of pectin confers them with interesting properties for many different applications, especially it has been used for many years as a gelling agent (in jams, jellies, marmalades, preserves and bakery fillings), thickening agent (in sauces, ketchup, and flavored syrups), texturizing agent (in fruit-flavored milk desserts), stabilizer (yogurts and milk drinks) and emulsifying agent (cod liver oil, ice-cream) in the food industry.[1] Currently, the primary sources of commercial pectin are apple pomace and peel of citrus fruits such as oranges, lemons, and grapefruits. In order to lower the production costs and meet the growing demand, alternative cheaper sources are being sought. However, obtaining pectin with similar composition and functional properties as the commercially available pectin is a challenging task and, thus, alternative sources need to be explored.

Jackfruit (*Artocarpus heterophyllus*) is a tropical fruit indigenous to many parts of Southeast Asia. Despite its high nutritional value and numerous health benefits, jackfruit remains underutilized due to insufficient post-harvest processing facilities and loopholes in the supply chain.[2] Approximately 60% of the whole jackfruit is either discarded as waste or used as cattle feed thus considerably reducing its commercial value.[3] An early study indicated that jackfruit is a rich source of pectin. Different parts of the jackfruit such as seeds, kernel, and pericarp were reported to contain 21-25%, 23-28%, and 25-30% of pectin, respectively.[4] Recent studies have focused on the isolation of pectin from jackfruit seed and waste, including rind and peel.[5,6] However,

extraction of pectin from edible portions of the jackfruit (including the fleshy pericarp and seed) has not been attempted.

Tender jackfruits are known to evolve through four maturity stages (Fig. 1). Besides the various physicochemical changes, the fruit hardness significantly varies with progress in fruit maturity. It decreases from 8052 g in immature fruits to 840 g in fully ripe fruits. The reduction in fruit hardness has been attributed to changes in the pectin content of jackfruit through the different maturity stages.[7] Nevertheless, a comprehensive investigation on the relationship between fruit maturity and the physicochemical and functional properties of pectin is still lacking. Therefore, in the present study is designed on the hypothesis that the physico-chemical and functional properties of the pectin from jackfruit depends on the fruit maturity and therefore its application in foods determined by the maturity stage at which the pectin is extracted.

Thus, in the present study, jackfruits from different maturity stages were used for pectin extraction. The extraction yields, chemical composition, molecular weight and functional properties of the jackfruit pectin (JFP) were evaluated as a function of the fruit maturity. Specifically, amongst the functional properties studied, the rheological properties of extracted pectins were explored, given their relevance for their practical application as a texturizing agent.[8]

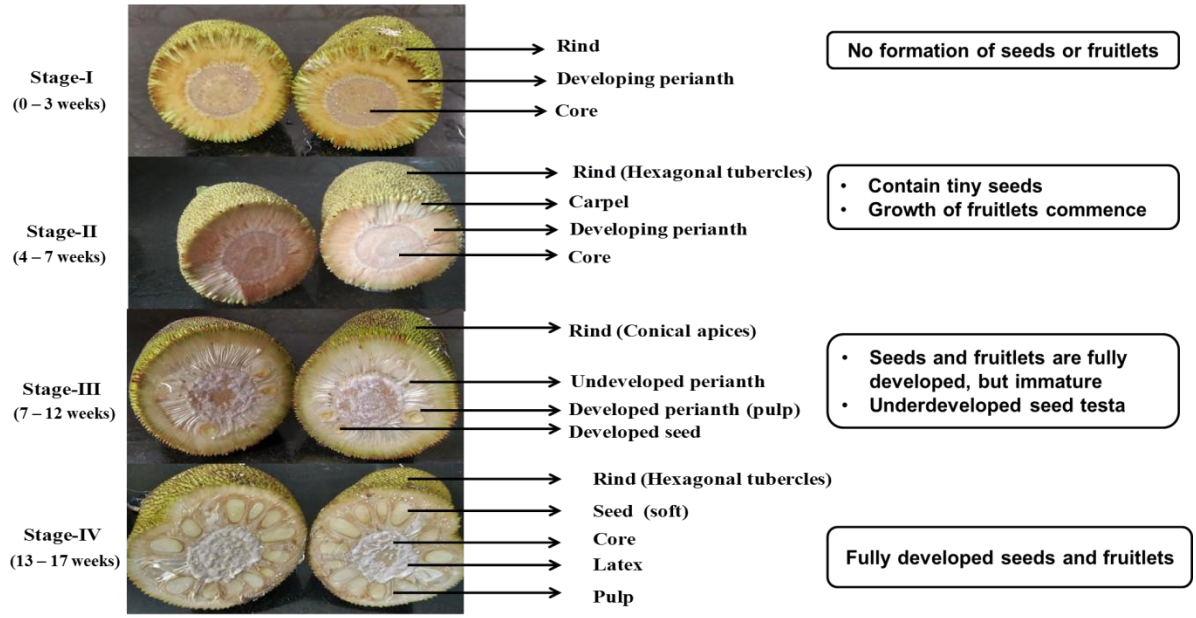

Figure 1. Different maturity stages of jackfruit.

2. Materials and Methods

2.1. Materials

Jackfruits at different maturity stages were collected from trees in the campus of CSIR-NIIST (Thiruvananthapuram, Kerala, India). Commercial pectin (CAS No 9000-69-5, DE 65-70%) was purchased from Himedia, India. Acetone, sodium chloride, sodium hydroxide, sodium carbonate, potassium acetate, methanol, trifluoroacetic acid (TFA), and Folin-Ciocalteau reagent were purchased from Sisco Research Laboratories (SRL) Pvt. Ltd. (Mumbai, India). Hydrochloric acid was acquired from Ranbaxy Fine Chemicals Limited (Mumbai, India). Orthophosphoric acid, aluminum chloride, gallic acid, quercetin, ascorbic acid, ABTS and DPPH were obtained from Merck India (Mumbai, India). All the reagents were of the analytical grade.

2.2. Pectin extraction

Jackfruits from the four different maturity stages (stage I–IV) were pre-processed to remove the outer rind. The edible part of the fruit, including the pericarp and seed,

was cut into small pieces, blanched and dried in a Refrigeration Adsorption Dehumidified Drier (RADD) at 50 °C, until a constant weight was reached and powdered using a laboratory mill (Retsch Laboratory Mill, Germany). The jackfruit powder thus obtained was passed through a sieve of mesh size 50 mm and stored in an air-tight container until further processing. For the extraction of JFP, the protocol described by Ranganna, 2002 was followed after slight modifications.[9] (Supplementary material; Section 1.1)

2.3. Compositional analysis

2.3.1. Moisture content

Moisture content was determined by gravimetric analysis. [10] Pectin sample (1 g) was accurately weighed and heated to 50 °C in a hot air oven until a constant weight was attained. The analysis was performed in duplicate.

2.3.2. Protein

Crude protein content in JFP samples was calculated based on the determination of nitrogen content in the sample and then using a conversion factor to convert it to protein content. (Supplementary material; Section 1.2)

2.3.3. Ash

1 g of pectin was incinerated at 550°C in a muffle furnace, overnight. The ash thus obtained was weighed after cooling in a desiccator.[10]

2.3.4. Monosaccharide and galacturonic acid content

The JFP samples were hydrolyzed at 120 °C for 3 h using trifluoroacetic acid (TFA; 2M). Subsequently, the hydrolysates were centrifuged at 3000×g for 15 min and then vacuum-dried to remove the TFA. The dried residue was dissolved in deionized water and centrifuged at 3000×g for 15 min.[11] The aqueous solutions of pectin

hydrolysate (5 mg/mL), standard monosaccharides (0.25 – 2.5 mg/mL), and standard galacturonic acid (0.25 – 2.5 mg/mL) in deionized water were filtered through a 0.22 micron PTFE filter before injecting them into the HPLC device. (Supplementary material; Section 1.3)

2.3.5. Degree of esterification (DE)

The degree of esterification (DE) of JFP was calculated according to the titrimetric method described by Ranganna, 2002. [9]

2.4. Structural characterization

2.4.1. Fourier Transform Infrared (FTIR) Spectroscopy

IR spectra were recorded on a Bruker Alpha-E FTIR spectrometer (Bruker Optics Inc., Ettlingen, Germany), using attenuated total reflection (ATR). The spectra were obtained in a wavelength range between 600 and 4000 cm^{-1} by averaging 24 scans at a resolution of 4 cm^{-1}. [12] The degree of esterification was confirmed using FTIR data following the method Duan et al (2022)[13] with slight modifications.

2.4.2. Determination of molecular weight by Matrix-Assisted Laser Desorption/Ionization Time-of-Flight (MALDI TOF-TOF)

The mass spectra of JFP samples were acquired in the positive ion mode over a mass-to-charge ratio (m/z) range of 150–2000 Da using a Bruker – auto flex speed MALDI-TOF TOF MS/MS analyzer, by following the procedure of Bermúdez-Oria, Rodríguez-Gutiérrez, Fernández-Prior, Vioque & Fernández-Bolaños, 2019, with slight modifications.[14]

The instrument was operated at an accelerating voltage of 26.45 kV with an extra voltage of 13.399 kV. Each spectrum was generated by accumulating data from 1000 to 2000 laser shots. The matrix solution of jackfruit pectin was prepared in 10

mg/mL ACN:H_2O:TFA (50:47.5:2.5) (v/v/v) in the presence of sodium trifluoroacetate (based on Bermúdez-Oria, Rodríguez-Gutiérrez, Fernández-Prior, Vioque & Fernández-Bolaños, 2019, with slight modifications).[14]

Molecular weight (M_w) was directly estimated from the m/z ratio using the following equation.

$$M_w = n(y - 1) \quad (4)$$

In the above equation, n is the charge state of ion given by,

$$n = \frac{x-1}{y-x} \quad (5)$$

Where x is the lower m/z value for the $n+1$ ion state, and y is the higher m/z value for the $n+$ ion state for two ions in series for a polymer of molecular weight M_w[15-17]

2.5. Differential Scanning Calorimetry (DSC)

The thermal behavior of jackfruit pectins was evaluated in comparison with that of commercial pectin, by differential scanning calorimetry (DSC), using the methodology of Wang et al. (2016).[18] DSC measurements were carried out using a differential scanning calorimeter (DSC Q2000, TA Instruments, USA). 5 mg of pectin sample were directly weighed into an aluminum pan and duly sealed by pressure. An empty aluminum pan was used as a reference. The reference and sample were then subjected to a linear temperature increase from 30 °C to 250 °C at a constant heating rate of 10 °C/min, under an inert atmosphere maintained by a constant flow of nitrogen at 50 mL/min.

2.6. Functional properties of jackfruit pectin

2.6.1 Water holding capacity and oil holding capacity

185 Briefly, 0.1 g of jackfruit pectin sample was added to a previously weighed centrifuge
186 tube. Then, the sample was soaked in 10 mL of distilled water/sunflower oil and
187 incubated for 24 h at 37 °C. After incubation, the suspension was centrifuged at
188 14000 rpm for 10 min. The excess water/oil was decanted, and the tubes were
189 inverted for 1 h at room temperature. For the calculation of WHC, the residues after
190 hydration and decanting were dried at 105 °C until constant weight was obtained.
191 For the estimation of OHC, the tubes were weighed to determine the weight of
192 residue after oil absorption.[19] Then, the water and oil holding capacities were
193 calculated as follows:

$$Water\ holding\ capacity\ (g/g) = \frac{Weight\ of\ hydrated\ residue - Weight\ of\ dry\ residue}{Weight\ of\ dry\ residue} \quad (6)$$

$$Oil\ holding\ capacity\ (g/g) = \frac{Weight\ of\ residue\ after\ oil\ absorption - Weight\ of\ sample\ before\ oil\ absorption}{Weight\ of\ sample\ before\ oil\ absorption} \quad (7)$$

194

195 ***2.6.2 Swelling capacity***

196 0.2 g of sample were added to 10 mL of distilled water in a graduated tube and
197 incubated for 18 h.[20] The final volume of the swollen sample was measured, and the
198 swelling capacity was calculated as:

$$Swelling\ capacity\ (mL/g) = \frac{Volume\ of\ swollen\ sample}{Initial\ weight\ of\ sample} \quad (8)$$

199 ***2.6.3 Oscillatory shear rheology***

200 Solutions were prepared from 6% (w/v) aqueous dispersions of JFP samples. For
201 solution preparation, the pectin powder was dissolved in distilled water at 75°C under
202 constant stirring for 15 min. Subsequently, the prepared solution was transferred

onto the rheometer plate. Rheological tests were performed by small-amplitude oscillatory shear using a strain-controlled rheometer (MCR 102, Anton Paar Gmbh, Germany) equipped with a Peltier-thermostated surface (20 °C). A cone-plate geometry (40 mm diameter, 2° angle, and 53 μm gap) was used for the measurement of material properties.

Initially, amplitude sweep tests were done to ascertain the linear viscoelastic range (LVR) of the JFP gel samples. Then, the mechanical spectra of the gels prepared from stage-I and stage-II pectin samples were obtained by frequency sweeps from 0.01 to 10 Hz at 0.4% strain (set based on the LVR established from the amplitude sweep tests done previously; data not shown) and 20°C. The mechanical spectra of gels prepared from Stage-III and Stage-IV pectin samples were obtained by frequency sweep from 0.01 to 10 Hz at 0.7% strain (set based on the LVR established from the amplitude sweep tests; data not shown) and 20 °C.

The gel point (T_{gel}) of JFP samples was determined from temperature ramps. Freshly prepared hot pectin solutions were loaded onto the rheometer, which plate was pre-heated at 80°C. The pectin solutions were heated from 20 °C to 80 °C, at 0.2 °C/min. After that, a controlled cooling step from 80 to 20 °C was carried out at 1 °C/min and a fixed frequency of 1 Hz. The strain was set at 0.4% for stage-I and stage-II pectin gels and 0.7% for stage-III and stage-IV pectin gels (set based on the LVR).

2.7 Total phenolic content (TPC), total flavonoids content (TFC) and antioxidant activity

2.7.1 TPC and TFC

Folin-Ciocalteau method was used to obtain the total phenolic content (TPC) in the JFP samples, with gallic acid as standard. Different concentrations of the samples in methanol were prepared in a total volume of 750 µL. To this solution, 80 µL of Folin's reagent and 200 µL of 7.5% sodium carbonate solution were added. The solution was kept in the dark for 90 minutes, and then the absorbance was measured at 750 nm[21]. The total phenolic content of JFP was expressed as milligrams of gallic acid equivalent per gram of pectin (mg GAE/g pectin).

The total flavonoid content (TFC) was estimated by following the aluminum chloride colorimetric method, with quercetin as standard.[21] Different concentrations of the samples were mixed with 10 µL of 10% aluminum chloride, 10 µL of 1M potassium acetate, and 280 µL of distilled water. The mixture was incubated for 40 min at room temperature (27 ± 3 °C), and its absorbance was read at 415 nm against blank using a multimode reader (Synergy BioTek, BioTek Instruments, Inc., Mumbai, India). TFC was expressed as milligram Quercetin equivalent per gram of pectin (mg QE/g pectin).

2.7.2. 1,1-diphenyl-2-picrylhydrazyl (DPPH) and ABTS radical scavenging activity

Approximately, 50 µL of 0.2 mM DPPH solution were added to 50 µL of pectin solutions at different concentrations. The sample was mixed well and allowed to stand for 30 min in an amber-colored 96 well plate. The absorbance of the reaction mixture was measured at 517 nm against a blank, using a multimode reader (Synergy BioTek, BioTek Instruments, Inc., Mumbai, India). Gallic acid was used as the standard.[21] Percentage inhibition of DPPH was calculated using the below equation.

$$\%\ Inhibition\ of\ DPPH = \frac{(Absorbance\ of\ control - Absorbance\ of\ sample)\ \text{x}\ 100}{Absorbance\ of\ control} \quad (9)$$

ABTS•+ radical cation (2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) was produced by reacting 7 mmol/L ABTS in water (ABTS stock solution) with 2.45 mmol/L solution of potassium persulfate, stored at room temperature for 12 h. The ABTS•+ solution was then diluted by phosphate buffer to an absorbance of 0.70 at wavelength 734 nm. Pectin solution (2 mL) with different concentration were added into 4 mL ABTS•+. Then the mixtures were placed in the dark at room temperature for 10 min, the absorbance of the mixtures were recorded at 734 nm. Ascorbic acid was used as a positive control [12] The ABTS•+ scavenging activity was calculated using the same formula as DPPH radical scavenging activity calculation.

2.8. Statistics

The means of all the parameters were examined for significance by one-way analysis of variance (ANOVA) at a confidence level of 95%, using the MS-Excel Data Analysis Toolpak add-in (Microsoft Excel, MS Office 2010, USA).

3. Results and discussion

3.1. Extraction yield of pectin

The extraction yields of pectin obtained from jackfruits at four different maturity levels are compiled in Table 1. An increase in pectin yield from 9.7 to 21.5% was observed with the progress in fruit maturity. The above range of pectin yield is comparable to that reported in other studies that involved pectin extraction from different parts of jackfruit using sodium hexametaphosphate: jackfruit waste: 22.5% [22], jackfruit rinds and cores: 14.81%.[23] The higher yield from sodium hexametaphosphate may be attributed to its ability for cell wall disruption, thus enhancing extraction rates. Pectins are physically bound to the middle lamella of plant cells through metallic cations,

specifically divalent cations. Being a sequestering agent, sodium hexametaphosphate can readily bind these cations and aid in releasing the pectins from cell walls.[24]

The increase in pectin yield with the maturity level was also reported in the case of banana pulp, wherein it increased from 0.37% at maturity stage-V to 0.66% in stage-VII. [25] The reason for increase in pectin yield may be the conversion of water-insoluble protopectin to water-soluble pectin during fruit maturation.[26] Yet another reason for the higher pectin yield could be that stage-I jackfruit did not contain seeds, as mentioned earlier. But, the stage-IV fruits had seeds which were reported to contain about 25% of pectin.[4]

3.2. Composition of jackfruit pectin

A comparison between the moisture content, protein content and ash content of Jackfruit pectin against those of pectin derived from other sources and their variation with fruit maturity are discussed in Section 1. of Supplementary Material.

Table 1. Yield and composition of jackfruit pectin

	Stage-I	**Stage-II**	**Stage-III**	**Stage-IV**
Yield (%)	9.7 ± 0.2^{a}	16.1 ± 0.2^{b}	16.5 ± 0.2^{b}	21.5 ± 0.5^{c}
Moisture (% w.b)	6.8 ± 0.1^{a}	7.0 ± 0.00^{a}	7.5 ± 0.1^{a}	9.9 ± 0.1^{b}
Protein (%)	4.4 ± 0.00^{a}	4.6 ± 0.00^{a}	4.6 ± 0.00^{a}	4.3 ± 0.00^{a}
Ash (%)	13.3 ± 0.1^{a}	8.0 ± 0.1^{b}	4.9 ± 0.03^{c}	4.4 ± 0.02^{c}

Values with different letters are significantly different ($P < 0.05$) from each other

3.2.1. Galacturonic acid and monosaccharide composition

Table 2 compiles the galacturonic acid (GalA) and monosaccharide composition of the pectins obtained from the various maturity stages. The GalA content of pectins obtained from stage I-IV jackfruits ranged from 50 to 65% (Table 2) and it increased

with fruit development. This may be ascribed to the degradation of water-insoluble pectic substances by polygalacturonase, which leads to the release of water-soluble galacturonic acids during fruit development.[27] Moreover, GalA is an important parameter that determines the suitability of pectin for different applications (ex. use in jams and jellies etc.). Its value should not be less than 65% for pectin to be used as food additives or for pharmaceutical applications.[28] Therefore, the purity and suitability for food applications was higher for pectin derived from stage-IV jackfruits. Apart from galacturonic acid, the major neutral sugars present in the jackfruit pectin were galactose, arabinose and rhamnose (cf. Table 2). Pectin is a heteropolysaccharide with α-(1-4)-linked linear homogalacturonic units alternated with two types of highly branched rhamnogalacturonan regions, RGI and RGII. RG-I region is substituted with side chains of arabinan, galactan, and arabinogalactan units whereas RG-II, that forms a minor component of pectin, includes (1→4) linked D-galactosyluronic residues, branched with eleven different monosaccharides.[1] The presence of residual glucose in pectin even after the acidic and alcoholic washing during pectin extraction process can be correlated with the presence of glucan linked to pectin.[29] Pectin is reported to be covalently bound to the other cell-wall polysaccharides such as xyloglucan, arabino glucan etc. [30] Basu et al., reported that glucans are responsible for the presence of glucose in pectin.[31] Therefore, the higher value of glucose in the present study may be correlated with the presence of glucan as reported by these authors.

The degree of esterification (DE) of pectins derived from jackfruits at different maturity levels is also included in Table 2, which ranged between 65% and 87%. The degree of esterification that was calculated using FTIR (Fig 2) followed similar trend of 67.78, 80.48, 83.61 and 86.10% respectively for JFP stage I-IV. Therefore,

irrespective of its maturity level, jackfruit pectin can be categorized as high-methoxyl pectin. Similarly, freeze-dried pectin prepared from jackfruit waste was also found to be of the high methoxyl type (DE = 63.9 ± 2.4%).[23]

Table 2. Monosaccharide composition of jackfruit pectin.

Pectin sample	Monosaccharide composition (%)				Galacturonic acid (%)*	DE (%)
	Glucose	Arabinose	Rhamnose	Galactose		
Stage-I JFP	0.59 ± 0.05^{a}	1.04 ± 0.00^{a}	1.06 ± 0.15^{a}	4.08 ± 0.09^{a}	50.05 ± 0.07^{a}	64.5 ± 0.49^{a}
Stage-II JFP	1.45 ± 0.09^{b}	1.03 ± 0.05^{a}	3.62 ± 0.15^{b}	8.17 ± 0.00^{b}	57.11 ± 0.13^{b}	76.7 ± 0.63^{b}
Stage-III JFP	7.04 ± 0.08^{c}	1.39 ± 0.00^{b}	2.57 ± 0.09^{c}	6.49 ± 0.05^{c}	63.56 ± 0.05^{c}	80.0 ± 0.82^{c}
Stage-IV JFP	9.33 ± 0.00^{d}	1.10 ± 0.07^{a}	2.68 ± 0.00^{c}	5.45 ± 0.00^{d}	65.21 ± 0.09^{d}	86.7 ± 0.56^{d}

*The fact that the sum does not amount to 100% is connected with the presence of unidentified substances.
The difference in values between the different maturity stages for all the parameters were statistically significant, with *P-value<0.05.*

3.3. Structure and molecular weight

FTIR analysis of JFP samples was carried out to confirm proper pectin extraction by identifying the characteristic spectral peaks from the pectin backbone and was compared with that of commercial pectin. Fig. 2 shows the FTIR spectra of jackfruit pectin and commercial pectin samples. As it can be seen, both JFP and commercial pectin exhibited the characteristic peaks of pectin. The broad peak at 3300 cm^{-1} corresponds to the O–H stretching vibrations, and the peak at 2900 cm^{-1} is related to the C–H stretching vibrations of CH, CH_2, and CH_3 groups. The peak at 1750 cm^{-1}, corresponding to esterified carboxyl groups (C=O stretching vibration of methyl-esterified carboxyl), is typical of high-methoxyl pectin.[32] As inferred from the FTIR spectra of JFP (Fig. 2), the presence of an intense spectral band at 1750 cm^{-1} confirms the titrimetry-determined JFP as high-methoxyl pectin. Moreover, the spectral range between 1200 and 800 cm^{-1} is deemed as the *'fingerprint'* region of polysaccharides[32], wherein the spectral region 1200–950 cm^{-1} depicts the highly

coupled modes of polysaccharide backbones [33] and the deformation of C-O stretch, the ring mode and the distortion of C-O-H, C-C-H, and O-C-H in the side groups.[34] The fingerprint region described above was present in the IR spectra of all the JFP samples. The band at 1540–1560 cm^{-1}, corresponding to the amide groups, indicate the presence of protein.

The MALDI spectra of stage I-IV JFP are shown in Figs. S1 [a-d] (Supplementary information). The calculated molecular weights of stage I-IV pectin samples were 147 kDa, 158 kDa, 169 kDa, and 114 kDa, respectively. The average molecular weight of pectin ranges from 50-150 kDa [35] and it has been observed to vary with the growth stage of the fruits.[36,37] Rahman et al. (1995) [38]studied the microscopic and chemical changes that occur during the ripening of jackfruit and found that high molecular weight material was very less in immature fruits (10-20 g per kg of jackfruit perianth dry matter) and the value increased with maturity (40-60 g per kg of jackfruit perianth dry matter). Accordingly, in this study, the molecular weight of JFP increased with maturity from stage I-III and reduced in that isolated from stage-IV fruits. The decrease in pectin molecular weight in the final maturity stage may be attributed to the cell wall degradation, a phenomenon that is typical of the fruit maturation process. During the above phenomenon, pectin is depolymerized by the action of pectolytic enzymes such as polygalacturonase. In other words, protopectin, the insoluble, high molecular weight parent pectin is converted into soluble polyuronides.[39] This reduces the content of high molecular-weight uronic acid polymer and increases the amount of low molecular-weight polymer in matured fruits.[40,41] However, the increase in M_w of pectin from stages I-III may be accredited to the initial rise in protopectin before attaining the physiological maturity.[42]

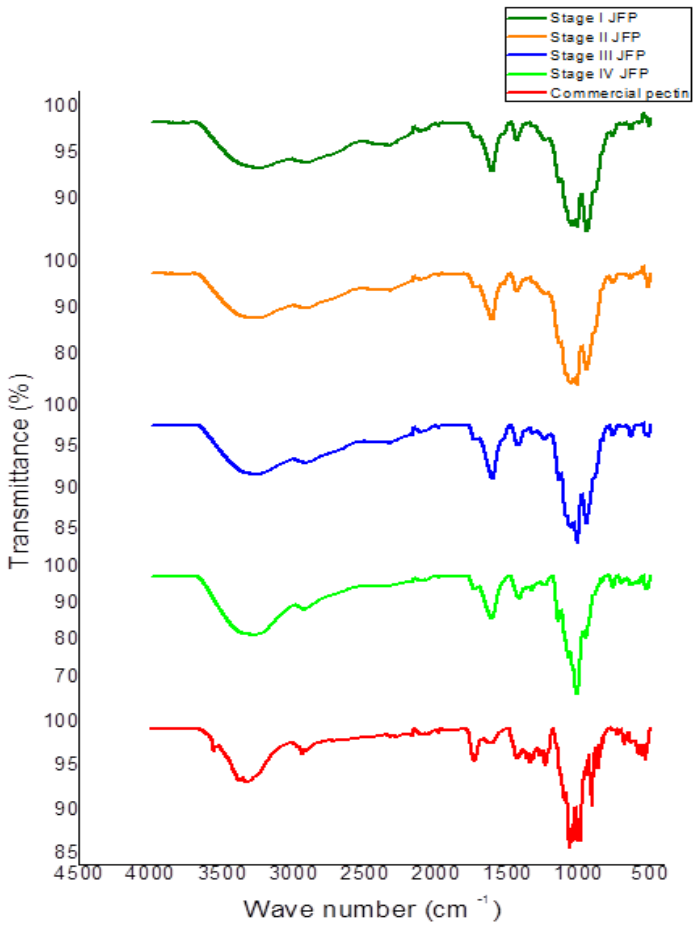

Figure 2. FTIR spectra of stage I-IV jackfruit pectin samples and of commercial pectin.

3.4. Thermal behavior

DSC was performed to investigate the thermal behavior of jackfruit pectins. The DSC thermograms of JFP and commercial pectin are shown in Fig. 3. From the DSC thermograms, it could be observed that all the JFP samples showed two endothermic peaks, the first one corresponding to the evaporation of adsorbed water and the second one which could be ascribed to the melting temperature of pectin (correlated to the loosening of intermolecular hydrogen bonds in crystalline pectin structures). In contrast, the commercial pectin (CP) showed three endothermic peaks, the two latter ones being much more defined, probably indicating a greater

purity degree. Table 3 compiles the thermal parameters, i.e. the melting temperature (T_m) and degradation temperature (T_d), respectively. The degradation temperature was determined from the small exothermic pyrolysis peak. T_m values ranged from 197 to 202°C for JFP samples while the two melting peaks from CP appeared at 184 and 204°C, respectively. This may be attributed to the presence of proteins or other unknown substances as reported earlier[43].

The melting peak from pectins has been normally ascribed to a conformational change from the more compact and partly crystalline state, in which the pectin chains are in the $^4C_1$ conformation, to the amorphous and more stretched $^1C_4$ conformation, which occurs because of the electrostatic repulsion of dissociated carboxyl groups when water is eliminated from the pectin structure. These endothermic peaks are usually observed just before the exothermic pectin degradation starts.[44] It is interesting to note that T_m values vary depending on the pectin source and extraction conditions, while the degradation temperature mainly depends on the pectin source.[45]

The T_m values for the stage-IV and stage-III jackfruit pectins and CP were significantly higher than those from the other pectins, which was indicative of their higher DE and galacturonic acid content that made the pectin molecules to tightly adsorb to water.[46] Stage-IV pectin showed the highest melting point (Table 3). Also, JFP exhibited higher melting point than pectins extracted from other sources. This indicates that the interactions via hydrogen bonds with water molecules of the galacturonan rings were stronger, which may have implications in the functional properties of the extracted pectins.[18,45]

The exothermic peak is caused by the degradation of pectin during the heating process.[47] The exothermic transition of all the JFP samples occurred at the same

temperature (230°C), being slightly lower than that from CP (T_d of 240 °C) (Table 3). Stability below 240 °C has been reported in citrus peel and apple pomace pectins.[45] Thermal stability of pectin is of relevance to its application as an additive in food products that can be processed at high temperature, such as cakes, bread and pastries.[48] Therefore, pectin with high thermal stability is more useful in the food industry. In the present study, the thermal analysis using DSC showed that JFP possessed comparable thermal stability as the commercial pectin, indicating its potential to replace CP in thermal food processing.[18]

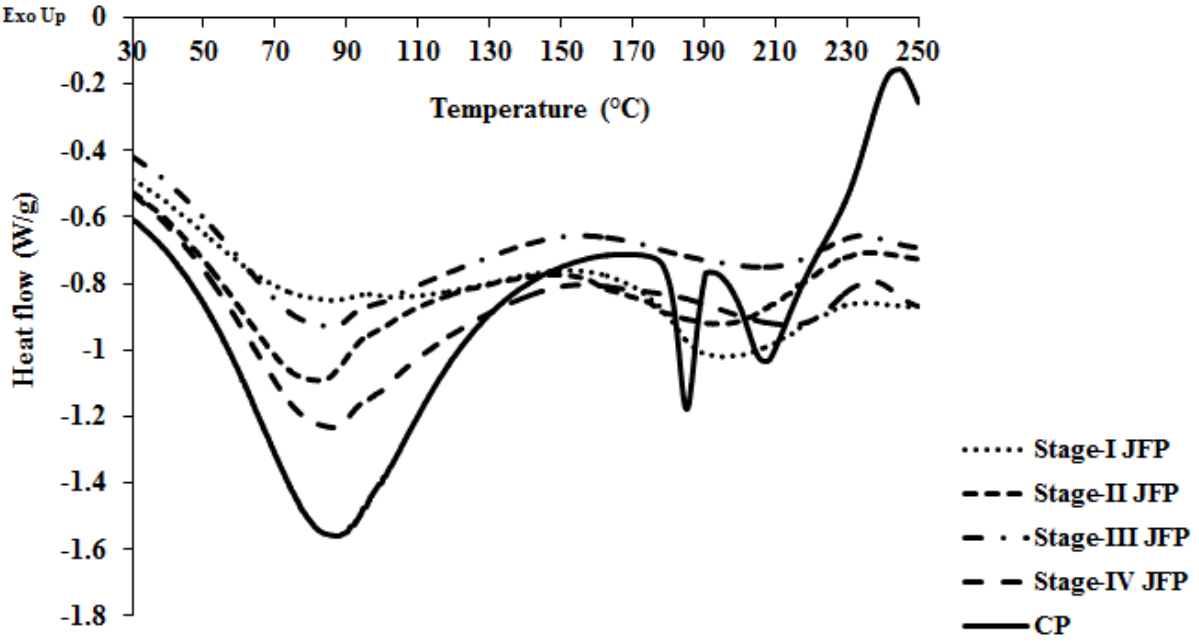

Figure 3. DSC thermograms of jackfruit pectins and commercial pectin.

Table 3. Thermal properties of jackfruit pectins determined by DSC

	T_m (°C)	T_d (°C)
Stage-I JFP	196.7	230.1
Stage-II JFP	196.7	230.2
Stage-III JFP	202.4	230.1
Stage-IV JFP	202.4	230.1

Commercial pectin	184; 204	240.6

3.5. Functional properties of jackfruit pectin

3.5.1 Water holding capacity (WHC)

Water-holding capacity (WHC) of the JFP samples was in the range of 7-12 g water/g pectin (cf. Table 4). The increase in WHC of JFP samples from stage I-IV can be attributed to the increase in their uronic acid content with fruit maturity (Table 2).[49] However, there were no significant differences between the values of WHC between stage-III and stage-IV JFP samples. The WHC values of JFP samples were significantly higher compared to that of dragon fruit pectin (5.5 g water/g) and apple pectin (5.45 g water/g).[50] However, it was lower than that of citrus fruit pectins (37.8 - 43.7 g water/g pectin) [51] and sunflower pectin (57 g water/g organic material).[52]

3.5.2 Oil holding capacity (OHC) and swelling capacity (SC)

The oil-holding capacity (OHC) and swelling capacity (SC) of pectin exhibited a similar trend as that observed for the WHC (cf. Table 4). The stage-III JFP sample showed the maximum OHC and SC. Even the minimum OHC of stage-I JFP (5.1 g oil/g sample) observed in this study was higher than that of apple pectin (2.96 g oil/g sample), citrus pectin (0.73 g oil/g sample), and dragon fruit peel pectin (1.24 g oil/g sample).[50] The oil sorption capacity of pectin is normally related to its DE and the subsequent increase in its hydrophobicity.[53] A combination of high WHC and OHC is an indication of good emulsifying properties and ability to promote the solubilization or dispersion of two immiscible liquids.[54] Similarly, even the lowest swelling capacity of stage-I JFP (11.3 mL/g) was higher than that of apple pectin (8.08 mL/g), citrus

pectin (5.98 mL/g) and dragon fruit peel pectin (6.97 mL/g).[55] These findings suggest the potential of JFP for its application as a food additive.

Table 4. Functional properties of jackfruit pectin.

Functional property	Stage-I	Stage-II	Stage-III	Stage-IV
Water holding capacity (g water/g pectin)	7.2 ± 0.2[a]	11.1 ± 1.4[b]	12.7 ± 1.04[c]	12.21 ± 1.3[c]
Oil holding capacity (g oil/g pectin)	5.1 ± 0.2[a]	6.4 ± 0.1[b]	8.1 ± 0.1[c]	7.0 ± 0.1[b]
Swelling capacity (mL/g)	11.3 ± 1.1[a]	14.0 ± 0.7[b]	21.8 ± 1.1[c]	15.0 ± 0.7[d]
$G'_{20°C}$ (Pa)	723.7[a]	726.9[b]	943.5[c]	737.2[d]
Gel point (°C)	65.9[a]	59.8[b]	50.8[c]	46.6[d]

The difference in values between the different maturity stages for all the parameters were statistically significant, with *P-value<0.0.* Values with different letters are significantly different (P < 0.05) from each other

3.5.3 Rheological properties

Another functional attribute that was studied from the extracted pectins was their gelling ability, being the gelation mechanism governed by their DE. To investigate the rheological behavior of the jackfruit extracts, gels were prepared and subjected to frequency sweeps at 20 °C. Representative curves for the pectin gels obtained from differently matured jackfruits are shown in Figures 4[a-d]. The log-log representation of the storage modulus (G') and the loss modulus (G'') as a function of frequency shows a gel-like behavior, with $G' > G''$. Except for the G' of stage-I JFP (Fig. 4[a]) that showed a frequency-independent behavior, both the moduli (G' and G'') of all the other pectin gels showed significant frequency dependence (Figs. 4[a-d]). Thus, it seems that JFP from stage-I was able to form better-structured networks.

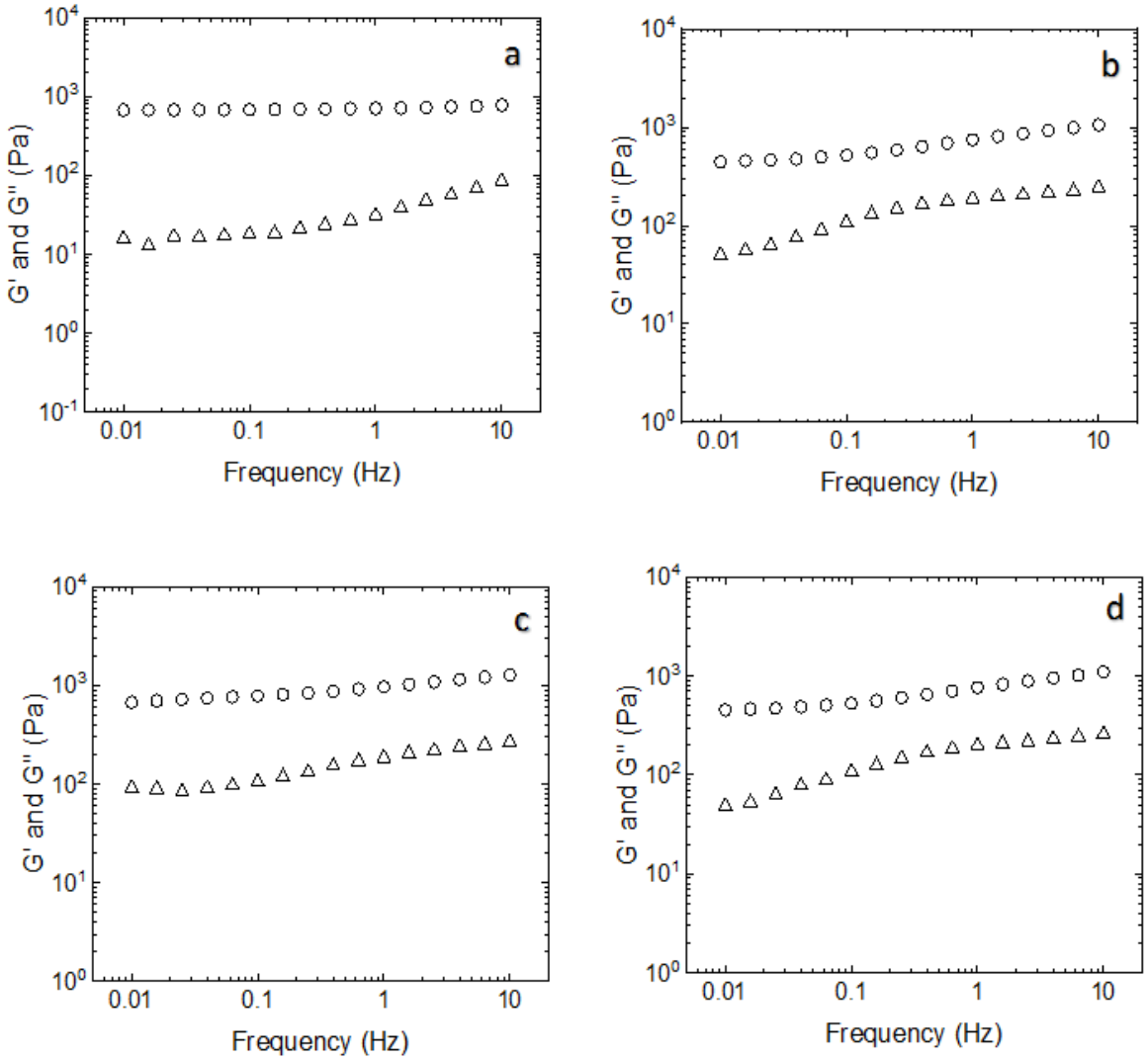

Figure 4. *G'* (circles) and *G"* (triangles) as function of frequency for the gels from aqueous solutions of jackfruit pectin obtained at different maturity stages: (a) stage-I; (b) stage-II; (c) stage-III;(d) stage-IV.

Fig. 5 [a-d] shows the characteristic plots of G' and G'' versus temperature, obtained from the small amplitude oscillatory measurements at constant frequency during controlled cooling. The temperature dependence of both moduli for all the gel samples is evident from the temperature sweep curves. The gel point (T_{gel}) was estimated from the crossover point between G' and G'' and the obtained values are listed in Table 4. As observed, the T_{gel} values significantly decreased from ca. 66 °C

464 to 47 °C with the maturity stage of jackfruit. T_{gel} bears a significant correlation with
465 the degree of methylation (DE) of the pectin.[55] Accordingly, high-methoxyl pectin has
466 been sub-classified into fast gelation pectin (T_{gel}: 75-85 °C); medium gelation pectin
467 (T_{gel}: 55-75 °C) and slow gelation pectin (T_{gel}: 45-60 °C).[51] Thus, pectin derived from
468 stage-I and stage-II jackfruits can be classified as medium gelation type, while that
469 extracted from stage-III and stage-IV as slow gelation type. Consequently, JFP may
470 find applications in jams and jellies and those gel-based products packed in large
471 containers, for which the formation of homogeneous gels by avoiding premature
472 gelation is critical, which would otherwise hinder the filling of packages.[57] After
473 equilibration at 20 °C, the samples were heated to 75 °C. Fig. 5 shows a slightly
474 decrease in the G' and G'' values when increasing the temperature; nevertheless, no
475 melting transition was observed. This confirms the non-reversible behavior of pectin
476 gels, as previously reported. [58]

477 The G' values for the gels at 20°C, gathered in Table 4, indicate that the pectin
478 extracted from stage I, II, and IV jackfruits formed gels with a very similar elastic
479 behavior. In contrast, the pectin extracted from stage-III jackfruit formed slightly
480 firmer gels. This can be related to the higher molecular weight of the latter, enabling
481 the formation of a higher number of cross-linking points between the pectin chains.
482 The temperature sweep test revealed that the JFP gels changed from a
483 predominantly liquid-like structure ($G' < G''$) to a typically gel-like structure ($G' > G''$)
484 at the gel point.

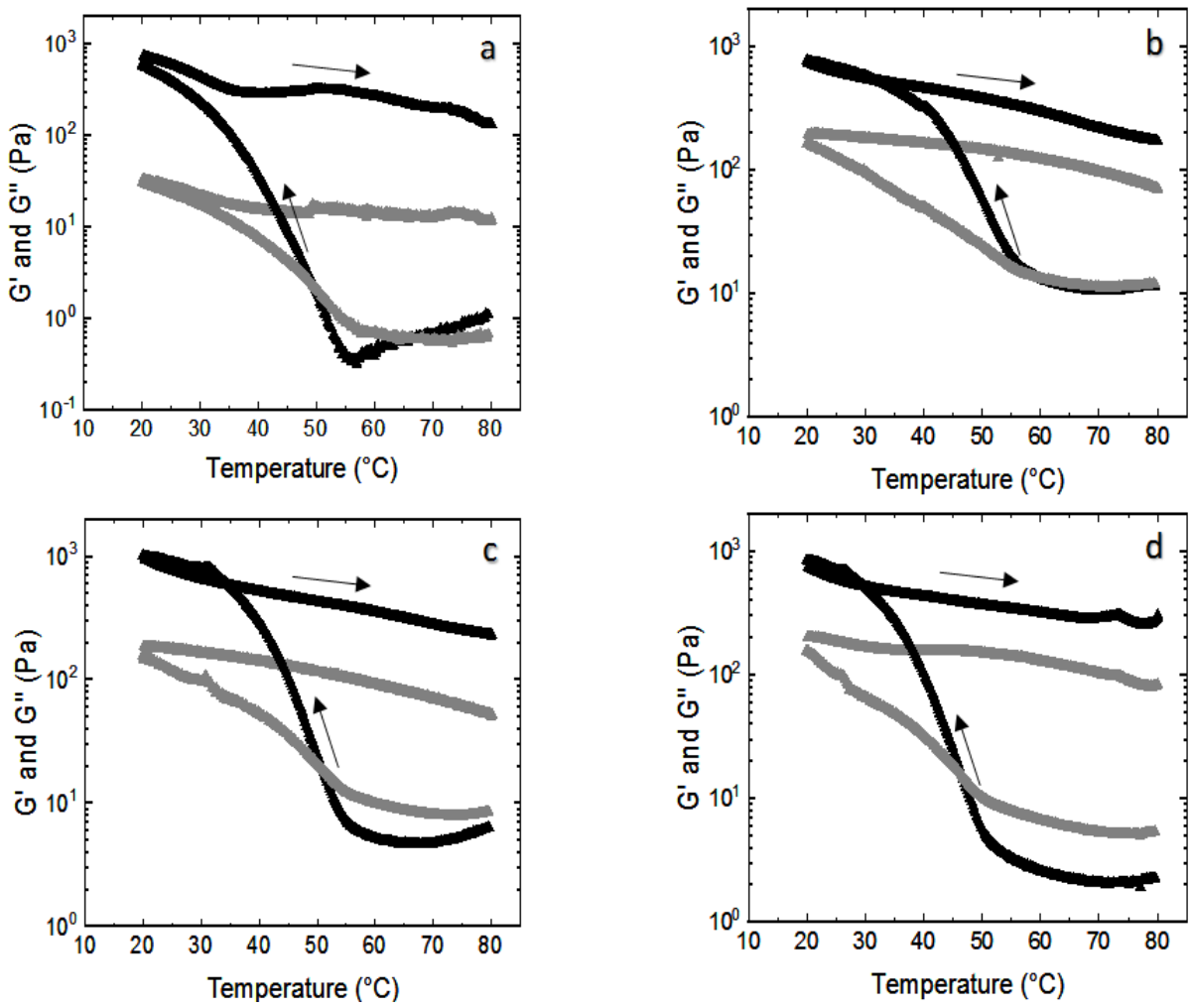

Figure 5. Temperature dependence of *G'* (black markers) and *G"* (grey markers) moduli of JFP gels during cooling and heating ramps of (a) stage-I JFP; (b) stage-II JFP; (c) stage-III JFP; (d) stage-IV JFP. Arrows indicate the direction of the temperature ramps.

3.5.4. Total phenolic and flavonoid content

The total phenolic content (TPC) and total flavonoid content (TFC) of JFP samples ranged between 21-62 mg GAE/g pectin and 30-47 mg QE/g pectin, respectively (Table 5). The TPC values of JFP were higher than those of pectin derived from eggplant calyx pectin (15.59 mg GAE/g pectin),[11] pistachio green hull pectin (18.18 mg GAE/g pectin),[59] pumpkin pectin (1.04 mg GAE/g pectin), citrus pectin (1.04 mg GAE/g pectin) and apple pectin (2.41 mg GAE/g pectin).[60] Similarly, the reported

497 TFC values of JFP were higher than the values found in pectin from *Cola milleni* peel
498 (10.17 mg QE/g pectin), *Theobroma cacao* (cocoa) (14.07 mg QE/g pectin) and
499 *Irvingia gabonensis* (wild mango) (5.94 mg QE/g pectin),[61] thus making this pectin of
500 special interest as a food additive, because phenolic compounds and their related
501 antioxidant capacity could provide benefits in terms of enhanced food preservation.
502 Extraction of pectin from the whole jackfruit rather than seed or peel may be the
503 reason behind the higher TPC and TFC of JFP than the values obtained in pectin
504 derived from other sources. An earlier study has reported that the fresh seeds and
505 flesh of jackfruit possess significant gallic acid equivalent phenolic content, which
506 contributes to approximately 70% of the total antioxidant activity.[62] Phenolic and
507 flavonoid contents in jackfruit pectin samples were found to decrease with maturity.

508 **3.5.5. Antioxidant activity**

509 Pectin is reported to exhibit antioxidant activity and the antioxidant potential of the
510 extracted pectin was assessed in terms of DPPH and ABTS radical scavenging
511 activity.

512 DPPH radical scavenging activity of jackfruit pectin increased with progress in fruit
513 maturity (Table 5) and was higher than the commercial pectin. ABTS radical
514 scavenging activity followed the order stage IV> stage III> stage II>CP> stage I. As
515 can be seen, the antioxidant activities did not follow the same trend as that detected
516 between total phenolic content and fruit maturity. Typically, in plant materials, the
517 radical scavenging activity increases with the increase in the concentration of
518 phenolic compounds.[63, 64] However, the unusual behavior observed in jackfruit pectin
519 may be due to the presence of antioxidant constituents other than phenolics and
520 flavonoids that are involved in inhibiting the radicals. It is reported that galacturonic
521 acid demonstrate promising antioxidant activities.[65, 66] It was suggested that the

carboxyl group of the galacturonic acid might act as a hydrogen donating and electron transfer agent facilitating radical scavenging potential. [67] Therefore, the increase in galacturonic acid during maturation may be correlated with the increased antioxidant activity. Gallic acid (IC_{50} -1.4 µg/µL), apple pectin (IC_{50} -2.75 mg/mL) and citrus pectin (IC_{50} -2.68 mg/mL) have been reported to have better DPPH activity than the JFPs obtained in the present study. However, when compared to okra (IC_{50} -8.9 mg/mL) and acid extracted apple pectin (IC_{50} -5.24 mg/mL), a better antioxidant activity of JFPs was observed. When considering the antioxidant activity measured through the ABTS radical scavenging assay, it was observed that JFP-IV outperformed both citrus and okra pectin(IC_{50} -3.33 mg/mL and 7.1 mg/mL, respectively)[68- 70], although it was much lower than that of the standard ascorbic acid employed (IC_{50} - 14.14 µg/mL).

Table 5. Total phenolic content, total flavonoid content and antioxidant activities of jackfruit pectin at different stages of maturity

Parameter	Stage-I	Stage-II	Stage-III	Stage-IV	Commercial pectin
TPC (mg GAE/g pectin)	62.4 ± 0.9[a]	40.2 ± 1.0[b]	30.3 ± 0.4[c]	21.5 ± 0.2[d]	30.7±0.4[c]
TFC (mg QE/g pectin)	46.6 ± 0.4[a]	41.9 ± 0.8[b]	42.4 ± 0.7[b]	30.4 ± 0.4[c]	19.91±0.7[d]
DPPH radical scavenging activity IC_{50} (mg/mL)	23.29±0.07[a]	12.2±0.05[b]	4.88±0.05[c]	3.62±0.02[d]	9.01±0.11[e]
ABTS radical scavenging activity IC_{50} (mg/mL)	12.75±0.04[a]	9.5±0.05[b]	4.8±0.1[c]	3.1±0.04[d]	2.45±0.08[e]

The difference in values between the different maturity stages for all the parameters were statistically significant, with *P-value<0.05s*

Values with different letters are significantly different (P < 0.05) from each other

Conclusions

This study was aimed at determining the influence of physiological maturity of jackfruits on the physicochemical and functional properties of pectin derived from their edible portions. The findings of this study confirm the potential of the edible

parts of jackfruit (including the pericarp and seed) as an alternative source of high quality pectin. Greater pectin extraction yields were obtained from the more mature jackfruits, with a greater GalA content and DE. Irrespective of the maturity level, the pectin obtained from jackfruit was of the high-methoxyl type, which was inherently capable of forming strong viscoelastic gels with medium-to-slow gel setting behavior. Furthermore, pectin from jackfruit demonstrated substantial antioxidant activity, water holding capacity, oil holding capacity, and swelling capacity, all of which increased with the progress in fruit maturity. Pectin derived from stage-I and II are found to be medium gelation type whereas that from stage-III and IV are slow gelation type with non-reversible behavior which can found application in jams and jellies and as a thickening agent. Thus, the results suggest that edible parts of the jackfruit at all maturity levels are a potential source of gelling pectin for food applications and hence can be utilized to obtain a value-added product with demonstrated functionalities. The scope for future research exists for elucidating the biological activity of jackfruit pectin and its role as a food additive. Further studies are needed to understand the distribution of pectic and other non-cellulosic polysaccharides and the possible changes in the structure of pectic polysaccharides during fruit development.

Acknowledgements

Author, P. Nisha thanks the Council of Scientific and Industrial Research (CSIR), India for awarding the Raman Research Fellowship. Author, Nidhina, K. acknowledges the University Grants Commission (UGC) for awarding Research Fellowship.

Conflict of interest

The authors claim no conflict of interest

References

1. Padma Ishwarya, Sandhya, P Nisha. Advances & Prospects in the Food Applications of Pectin Hydrogels. *Critical Reviews in Food Science and Nutrition*, **62**(16):4393-417(2021) DOI: 10.1080/10408398.2021.1875394

2. Vazhacharickal PJ, Sajeshkumar NK, Mathew JJ, Kuriakose AC, Abraham B, Mathew RJ, Albin AN, Thomson D, Thomas RS, Varghese N and Jose S. Chemistry and medicinal properties of jackfruit (Artocarpus heterophyllus): a review on current status of knowledge. *International Journal of Innovative Research and Review,* **3**:83-95 (2015).

3. Subburamu K, Singaravelu M, Nazar A and Irulappan I. A study on the utilization of jack fruit waste. *Bioresour Technol,* **40**:85–6 (1992).

4. Krishnamurti CR and Giri KV. Preparation, purification and composition of pectins from Indian fruits and vegetables. *InProceedings of the Indian Academy of Sciences-Section B,* Springer India, **29**:155-167 (1949).

5. Koh PC, Leong CM and Noranizan MA. Microwave-assisted extraction of pectin from jackfruit rinds using different power levels. *Int Food Res J,* **21**:2091–7 (2014).

6. Li WJ, Fan ZG, Wu YY, Jiang ZG and Shi RC. Eco-friendly extraction and physicochemical properties of pectin from jackfruit peel waste with subcritical water. *J Sci Food Agric,* **99**:5283–92 (2019).

7. Ranasinghe RASN and Marapana RAUJ. Effect of Maturity Stage on Physicochemical Properties of Jackfruit (Artocarpus heterophyllus Lam .)

591 Flesh. *World J Dairy Food Sci,* **14**:17–25 (2019).

592 8. Strom A, Ribelles P, Lundin L, Norton I, Morris ER and Williams MAK.
593 Influence of Pectin Fine Structure on the Mechanical Properties of Calcium -
594 Pectin and Acid - Pectin Gels. *Biomacromolecules,* **8**:2668-74 (2007).

595 9. Ranganna S. Pectin, in *handbook of analysis and quality control for fruit and*
596 *vegetable products.* Tata McGraw-Hill Publishing Company Ltd, USA, pp. 31–
597 47 (2002).

598 10. AOAC. Official methods of analysis, (16th edn). AOAC International, Arlington,
599 VA, USA (1995).

600 11. Kazemi M, Khodaiyan F, and Hosseini SS. Utilization of food processing
601 wastes of eggplant as a high potential pectin source and characterization of
602 extracted pectin. *Food Chem*, **294**:339–346 (2019).

603 12. Xu SY, Liu JP, Huang X, Du LP, Shi FL, Dong R, Huang XT, Zheng K, Liu Y,
604 Cheong KL. Ultrasonic-microwave assisted extraction, characterization and
605 biological activity of pectin from jackfruit peel. *LWT*, **90**:577-82 (2018).

606 13 Duan H, Wang X, Azarakhsh N, Wang C, Li M, Fu G, Huang X. Optimization of
607 calcium pectinate gel production from high methoxyl pectin. J*ournal of the*
608 *Science of Food and Agriculture*, **102**(2):757-63 (2022).

609 14. Bermúdez-oria A, Rodríguez-gutiérrez G, Fernández-prior Á, Vioque B, and
610 Fernández-bolaños J. Strawberry dietary fiber functionalized with phenolic
611 antioxidants from olives . Interactions between polysaccharides and phenolic
612 compounds. *Food Chem Elsevier,* **280**:310–320 (2019).

15. Chapman JR, Gallagher RT, Barton EC, Curtis JM and Derrick PJ. Advantages of High-resolution and High-mass Range Magnetic-sector Mass Spectrometry for Electrospray Ionization. *Org MASS Spectrom,* **27**:195–203 (2000).

16. Covey TR, Bonner RF, Shushan BI, Henion J and Boyd RK. The determination of protein, oligonucleotide and peptide molecular weights by ion-spray mass spectrometry. *Rapid Communications in Mass Spectrometry,* **2**:249-56 (1988).

17. Mann M, Meng CK and Fenn JB. Interpreting mass spectra of multiply charged ions. *Analytical Chemistry,* **61**:1702-8 (1989).

18. Wang J, Hu S, Nie S, Yu Q and Xie M. Reviews on Mechanisms of in Vitro Antioxidant Activity of Polysaccharides. *Oxid Med Cell Longev,* **2016** (2016).

19. Suzuki T, Ohsugi Y, Yoshie Y, Shirai T and Hirano T. Dietary fiber content, water-holding capacity and binding capacity of seaweeds. *Fisheries science,* **62**:454-61(1996).

20. Gómez-Ordóñez E, Jiménez-Escrig A, Rupérez P. Dietary fibre and physicochemical properties of several edible seaweeds from the northwestern Spanish coast. *Food Research International*, **43**(9):2289-94 (2010). http://dx.doi.org/10.1016/j.foodres.2010.08.005

21 Arun KB, Persia F, Aswathy PS, Chandran J, Sajeev MS, Jayamurthy P and Nisha P. Plantain peel-a potential source of antioxidant dietary fibre for developing functional cookies. *Journal of Food Science and Technology,* **52**:6355-64 (2015).

635 22 Begum R, Aziz MG, Uddin MB and Yusof YA. Characterization of Jackfruit
636 (Artocarpus heterophyllus) Waste Pectin as Influenced by Various Extraction
637 Conditions. *Ital Oral Surg*, **2**:244–51 (2014)
638 http://dx.doi.org/10.1016/j.aaspro.2014.11.035

639 23. Leong CM, Noranizan MA, Kharidah M, Choo WS. Physicochemical properties
640 of pectin extracted from jackfruit and chempedak fruit rinds using various
641 acids. *International Food Research Journal*, **23**(3):973-978 (2016).

642 24. Yeoh S, Zhang S, Shi J and Langrish TAG. A comparison of different
643 techniques for water-based extraction of pectin from orange peels. *Chem Eng*
644 *Commun,* **195**:511–20 (2008).

645 25. Tapre AR, Jain RK. Study of advanced maturity stages of banana.
646 *International Journal of Advanced Engineering Research and Studies*,
647 **1**(3):272-4 (2012).

648 26. Inari T, Yamauchi R, Kato K and Takeuchi T. Changes in pectic
649 polysaccharides during the ripening of cherry tomato fruits. *Food Science and*
650 *Technology Research,* **8**:55-8 (2002).

651 27. Reeve RM. Histological and histochemical changes in developing and ripening
652 peaches. II. The cell walls and pectins. *American Journal of Botany,* **46**:241-8
653 (1959).

654 28. May CD. Industrial pectins: Sources, production and applications.
655 *Carbohydrate Polymers,* **12**:79–99 (1990).

656 29. Petkowicz CL, Vriesmann LC, Williams PA. Pectins from food waste:

657 Extraction, .characterization and properties of watermelon rind pectin. *Food*
658 *Hydrocolloids*, **65**:57-67 (2017).

659 30. Legentil A, Guichard I, Piffaut B, Haluk JP. Characterization of strawberry
660 pectin extracted by chemical means. *LWT-Food Science and Technology*,
661 **28**(6):569-76 9(1995).

662 31. Basu NG, Ghosal PK, Thakur S. Some structural features of an arabinoglucan
663 from the fruits of Cordia dichotoma Forst. *Carbohydrate research,* **146**(2):350-
664 1 (1986).

665 32. Naik M, Rawson A, Rangarajan JM. Radio frequency-assisted extraction of
666 pectin from jackfruit (Artocarpus heterophyllus) peel and its characterization.
667 *Journal of Food Process Engineering*, **43**(6):e13389 (2020).

668 33. Engelsen SB and Nørgaard L. Comparative vibrational spectroscopy for
669 determination of quality parameters in amidated pectins as evaluated by
670 chemometrics. *Carbohydr Polym,* **30**:9–24 (1996).

671 34. Chylińska M, Szymańska-Chargot M and Zdunek A. FT-IR and FT-Raman
672 characterization of non-cellulosic polysaccharides fractions isolated from plant
673 cell wall. *Carbohydr Polym,* **154**:48–54 (2016).

674 35. Mudgil D. The Interaction Between Insoluble and Soluble Fiber. In: Dietary
675 Fiber for the Prevention of Cardiovascular Disease: Fiber's Interaction
676 between Gut Micoflora, Sugar Metabolism, Weight Control and Cardiovascular
677 Health [Internet]. Elsevier Inc,. pp.35–59 (2017). Available from:
678 http://dx.doi.org/10.1016/B978-0-12-805130-6/00003-3

679 36. Sundarraj AA, Vasudevan RT, Sriramulu G. Optimized extraction and
680 characterization of pectin from jackfruit (Artocarpus integer) wastes using
681 response surface methodology. *International Journal of Biological*
682 *Macromolecules*, **106**:698-703 (2018).

683 37. Tsuchida Y, Yakushiji H, Oe T, Negoro K, Gato N, Kotani T, et al. Differences
684 in Cell-wall Polysaccharide Degradation during Softening Process in Two
685 Cultivars of Japanese Apricot Fruits. J*ournal of the Japanese Society for*
686 *Horticultural Science,* **83**:81–9 (2014).

687 38. Rahman AM, Huq E, Mian AJ and Chesson A. Microscopic and chemical
688 changes occurring during the ripening of two forms of jackfruit (Artocarpus
689 heterophyllus L.). *Food Chemistry,* **52**:405-10 (1995).

690 39. John MA and Dey PM. Postharvest changes in fruit cell wall. *Adv Food Res,*
691 **30**:139–93 (1986).

692 40. Paniagua C, Posé S, Morris VJ, Kirby AR, Quesada MA and Mercado JA. Fruit
693 softening and pectin disassembly: An overview of nanostructural pectin
694 modifications assessed by atomic force microscopy. *Ann Bot,* **114**:1375–83
695 (2014).

696 41. Tsuchida Y, Onishi S, Gato N, Naka Y, Oe T and Jomura N. Effect of maturity
697 and after-ripening on the formation of gel in the syrup made from Japanese
698 apricot 'Suiko' fruits. *Sci Hortic* (Amsterdam), **247**:101–6 (2019).
699 https://doi.org/10.1016/j.scienta.2018.12.005

700 42. Tandon DK and Kalra SK. Pectin changes during the development of mango
701 fruit cv Dashehari. *J Hortic Sci,* **59**:283–6 (1984).

43. Wang X, Lü X. Characterization of pectic polysaccharides extracted from apple pomace by hot-compressed water. *Carbohydrate polymers*, **102**:174-84 (2014).

44. Li WJ, Fan ZG, Wu YY, Jiang ZG, Shi RC. Eco-friendly extraction and physicochemical properties of pectin from jackfruit peel waste with subcritical water. *Journal of the Science of Food and Agriculture*, **99**(12):5283-92 (2019).

45. Wang X, Chen Q and Lü X. Food Hydrocolloids Pectin extracted from apple pomace and citrus peel by subcritical water. *Food Hydrocoll*, **38**:129–37 (2014). http://dx.doi.org/10.1016/j.foodhyd.2013.12.003

46. Iijima M, Nakamura K, Hatakeyama T and Hatakeyama H. Phase transition of pectin with sorbed water. *Carbohydrate Polymers,* **41**:101-6 (2000).

47. Godeck R, Kunzek H and Kabbert R. Thermal analysis of plant cell wall materials depending on the chemical structure and pre-treatment prior to drying. *European food research and technology,* **213**:395-404 (2001).

48. Combo AMM, Aguedo M, Quiévy N, Danthine S, Goffin D, Jacquet N, et al. Characterization of sugar beet pectic-derived oligosaccharides obtained by enzymatic hydrolysis. *Int J Biol Macromol,***52**:148–56 (2013). http://dx.doi.org/10.1016/j.ijbiomac.2012.09.006

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

50. Izalin MN, Kharidah M, Jamilah B and Noranizan MA. Functional properties of

724 pectin from dragon fruit (Hylocereus polyrhizus) peel and its sensory attributes.
725 *J. Trop. Agric. and Fd. Sc,* **44**:95-101 (2016).

726 51. Boulos NN, Greenfield H and Wills RB. Water holding capacity of selected
727 soluble and insoluble dietary fibre. *International Journal of Food Properties*,
728 **3**:217-31 (2000).

729 52. Miyamoto A and Chang KC. Extraction and physicochemical characterization
730 of pectin from sunflower head residues. *Journal of Food Science,* **57**:1439-43
731 (1992).

732 53. Rubio-Senent F, Rodríguez-Gutiérrez G, Lama-Muñoz A and Fernández-
733 Bolaños J. Pectin extracted from thermally treated olive oil by-products:
734 Characterization, physico-chemical properties, invitro bile acid andglucose
735 binding. *Food Hydrocoll,* **43**:311–21 (2015).

736 54. de Moura FA, Macagnan FT, Dos Santos LR, Bizzani M, de Oliveira Petkowicz
737 CL and Da Silva LP. Characterization and physicochemical properties of
738 pectins extracted from agroindustrial by-products. *Journal of food science and*
739 *technology,* **54**:3111-7 (2017).

740 55 Begum R, Aziz MG, Yusof YA, Saifullah M, Uddin MB. Evaluation of gelation
741 properties of jackfruit (Artocarpus heterophyllus) waste pectin. C*arbohydrate*
742 *Polymer 4Technologies and Applications*, **2**:100160(2021).

743 56. Soler MP. Processamento industrial. Industrialização de geléias, Instituto de
744 Tecnologia de Alimentos, vn **8**:1-20 (1991).

745 57. Rosenthal A and Torrezan R. Preservation and processing of subtropical fruits.
746 In: Yahia, E. M. (Ed.), Postharvest biology and technology of tropical and
747 subtropical fruits, Volume 1: Fundamental issues, Woodhead Publishing
748 Limited, PA, USA, pp. 419-484 (2011).

749 58. Chan SY, Choo WS, Young DJ and Loh XJ. Pectin as a rheology modifier:
750 Origin, structure, commercial production and rheology. *Carbohydrate polymers*
751 **161**:118-39 (2017).

752 59 Kazemi M, Khodaiyan F, Hosseini SS and Hojjati M. Pistachio green hull
753 pectin: optimization of microwave-assisted extraction and evaluation of its
754 physicochemical, structural and functional properties. *Food Chem,***271**:663-672
755 (2018). https://doi.org/10.1016/j.foodchem.2018.07.212

756 60. Torkova AA, Lisitskaya KV, Filimonov IS, Glazunova OA, Kachalova GS,
757 Golubev VN and Fedorova TV. Physicochemical and functional properties of
758 Cucurbita maxima pumpkin pectin and commercial citrus and apple pectins: A
759 comparative evaluation. *PloS one* **13**:e0204261 (2018).

760 61. Oloye MT, Lajide L 2, Adetuyi AO, Ayoade GW and Arogundade OL.
761 Phytochemical Constituents of Pectin Extracted from Peels of Cola milleni,
762 Theobroma cacao and Irvingia gabonensis [Internet]. *futa.edu.ng.* (2013).
763 Available from:
764 https://www.futa.edu.ng/journal/papers/paper_7_1549623186.pdf

765 62. Soong YY and Barlow PJ. Antioxidant activity and phenolic content of selected
766 fruit seeds. *Food Chem,* **88**:411–7 (2004).

767 63. Alothman M, Bhat R and Karim AA. Antioxidant capacity and phenolic content

of selected tropical fruits from Malaysia, extracted with different solvents. *Food chemistry,* **115**:785-8 (2009).

64. González-montelongo R, Lobo MG, González M. Antioxidant activity in banana peel extracts : Testing extraction conditions and related bioactive compounds. *Food Chem,* **119**:1030–9 (2010). http://dx.doi.org/10.1016/j.foodchem.2009.08.012

65. Bayar N, Bouallegue T, Achour M, Kriaa M, Bougatef A, Kammoun R. Ultrasonic extraction of pectin from Opuntia ficus indica cladodes after mucilage removal: Optimization of experimental conditions and evaluation of chemical and functional properties. *Food Chemistry*, **235**:275-82 (2017).

66. Kumar M, Potkule J, Tomar M, Punia S, Singh S, Patil S, Singh S, Ilakiya T, Kaur C, Kennedy JF. Jackfruit seed slimy sheath, a novel source of pectin: Studies on antioxidant activity, functional group, and structural morphology. *Carbohydrate Polymer Technologies and Applications*, **2**:100054 (2021).

67. Zhang H, Li J, Xia J, Lin S. Antioxidant activity and physicochemical properties of an acidic polysaccharide from Morinda officinalis. *International Journal of Biological Macromolecules*, **58**:7-12 (2013).

68. Wang MM, Wang F, Li G, Tang MT, Wang C, Zhou QQ, Zhou T, Gu Q. Antioxidant and hypolipidemic activities of pectin isolated from citrus canning processing water. *LWT,* **159**:113203 (2022).

69. Wikiera A, Grabacka M, Byczyński Ł, Stodolak B, Mika M. Enzymatically extracted apple pectin possesses antioxidant and antitumor activity. *Molecules*, **26**(5):1434 (2021).

70 Xiong B, Zhang W, Wu Z, Liu R, Yang C, Hui A, Huang X, Xian Z. Preparation, characterization, antioxidant and anti-inflammatory activities of acid-soluble

pectin from okra (Abelmoschus esculentus L.). *International Journal of Biological Macromolecules,* **181**:824-34 (2021).