Effect of Malaysian durian rind varieties on the yield and degree of esterification of the pectin extracted

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Abstract. Durian rind which composed more than half of the total weight of durian had produced massive waste to the environment, especially during the peak season. Durian rind was found to be a potential source of pectin. The yield and characteristic of pectin present at different varieties of durian rinds could differ. Thus, the objective of this study is to examine the yield and degree of esterification (DE) of pectin extracted from Musang King (D197), D24, Hajjah Hasmah (D168) and mixed varieties of durian rinds. The pectin was extracted from different varieties of durian rinds in pH 2 citric acid solution (solid to liquid 1:30) at 90 °C for 2 hours. Fourier-transform infrared spectroscopy (FTIR) was used to examine the chemical structure and DE of the pectin. The results showed that the pectin extracted from D168 has the least amount (5.6%) while the pectin yield of other varieties does not differ distinctly (7.1 – 7.73%). All the pectin isolated with DE range 60.79 – 76.71% is high methoxyl pectin with DE > 50%. This study shows that different varieties of durian rinds produce pectin with varying yield, gelling and structural properties which could provide a new alternative of pectin for various industries such as the pharmaceutical and food industries.

1. Introduction

Durian (Durio zibethinus) is a famous tropical fruit with a distinct flavour and unique aroma. It is mainly found in Southeast Asia, Thailand being the biggest durian producer, followed by Malaysia and other countries including Indonesia and the Philippines [1]. Durian fruit has either ovoid or ovoid oblong to almost round shape which weighs around 2 to 4.5 kg depending on their varieties [2]. In Malaysia, there are numbers of popular varieties of durian in Malaysia such as Musang King (D197), Hajjah Hasmah (D168) (also known as “Durian IOI”), D24 and Udang Merah (D175). The Department of Agriculture of Malaysia (DOA) had reported an increase in the production of durian fruits from 297,696 metric tons to 341,332 metric tons of durian which worth about RM 5 million from the year 2010 to 2018 (14.66% increment) (unpublished data). In 2020, the hit of coronavirus (COVID-19) pandemic worldwide had not hindered the demand for durian, especially in China. The export of durian fruits to China had resumed to almost 80% after the pandemic in China was under controlled and the demand is expected to increase during the peak season (July to August) [3]. Besides, amid the pandemic in Thailand, more than 100,000 tons of durian fruits had been shipped from Thailand to China [4]. Nevertheless, the large production of durian could lead to massive disposal of durian rinds to the environment, especially during the peak season [5]. These wastes are either burnt or buried in landfills, leading to several problems to the environment [6,7].
These wastes could be turned into assets with commercial importance, for example, particleboard as construction panel, low-cost absorbent of acid dye and the water-soluble polysaccharides extracted from durian rinds could bind tablet, moisturize the skin and form antibacterial dressing film [8–12]. The water-soluble polysaccharide obtained from durian rinds is mainly made up of pectin [13]. Pectin is composed of galacturonic acid backbones linked by α-1,4-bonds, which are esterified with methyl alcohol at carboxylic acid. The main chain is interspersed with L-rhamnose residues linked to neutral sugar side chains, mainly arabinans and arabinogalactan side chains [14]. Generally, pectin is categorized into two groups that are high methoxyl pectin (HMP) with a degree of esterification (DE) more than 50% and low methoxyl pectin (LMP) with DE less than 50%. HMP forms a gel at acidic condition (pH ≤ 3.5) and high sugar level whereas LMP forms a gel in the presence of calcium ions at a larger pH range (2.0 < pH < 6.0) [15,16]. Pectin plays significant roles in food, pharmaceutical, dental and cosmetic industries due to its gelling, stabilizing and emulsifying properties [17,18]. The demands for pectin had been increasing by 4 to 5% every year [19]. Previously, citrus peels and apple pomace are the main sources to produce commercial pectin [20]. Due to the high demand for pectin, many studies were done to seek new pectin sources such as passion fruit peel, jackfruit rind, papaya peel and dragon fruit peel, while durian rind could be an alternative source of pectin [21–24].

To date, there is limited published information about the extraction and characterization of pectin from durian rinds. The yield and characteristics of pectin could be affected by different varieties, tissues and plants’ stages of growth. There is no published data about the influence of varieties of durian rinds on the yield and characteristic of the pectin isolated, to the best of our knowledge. Hence, in the present study, the objective is to examine the yield and DE of the pectin extracted based on different durian’s varieties in Malaysia.

2. Material and Methods

2.1. Chemical and Methods

Citric acid monohydrate was purchased from Bendosen (Norway), whereas ethanol 95% was obtained from HmBG (Malaysia).

2.2 Durian rinds collection

2.2.1 Collection of rinds from durian variety. Musang King (D197), D24 and Hajjah Hasmah (D168) (also known as Durian IOI) were purchased from a local fruit store in Muar, Johor, Malaysia in mid-December 2019. Upon collection, durian varieties were verified by an officer from the Department of Agricultural of Malaysia (DOA). The durian fruits chosen were ranged from 1.5 to 2 kg depending on durian variety. The fruits were checked to ensure freshness and free from defects such as visible holes by worms or squirrels, cracking and soft or discoloured patches on the fruits. A fresh and mature durian can be identified through a combination of few indices, for example, hollow tapping sound of the fruit, fruit stem which is not broken or damaged nor shrivelled due to loss of water, firm and hard fruit stem, and flexible thorns when two spines are pressed to each other [25]. The durian fruits collected were cut open manually using a sharp knife. The arils were separated from durian rinds.

2.2.2 Collection of mixed varieties durian rind. The durian wastes rind were obtained from a local durian wholesaler in Muar, Johor, Malaysia in mid-December 2019. The durian rinds collected contains a mixture of various durian varieties and were collected on the same day of disposal. The durian rinds collected were free from defects, for example, wet and watery rind, rotten patches, the outgrowth of mould, visible holes by pests, and browning on the rind.

2.3 Drying of durian rinds

The durian rinds were washed with distilled water to remove dirt and the remaining durian flesh on the rinds. The washed rinds were left on the laboratory benchtop to air dry for 30 minutes. Then, the sharp
2.4 Extraction of pectin from durian rinds

Each variety of dried durian rind powder samples was extracted in citric acid solution adjusted to pH 2 (solid-liquid ratio: 1:30) at 90 °C for 2 hours in a shaking water bath (SWB-B, Biobase, China) [22]. The solutions were centrifuged at 4200 rpm (3234 × g) for 20 minutes (5804R, Eppendorf, Germany). The supernatants were collected and cooled to room temperature. The solutions were precipitated with 95% ethanol in a 1:2 ratio to allow pectin precipitation. The samples were left at room temperature of 25 °C for 1 hour to allow pectin flotation. Then, the polysaccharide containing pectin was separated by filtration and washed twice with 75% ethanol, once with 95% ethanol to remove mono- and disaccharide and unwanted colour. The resulting pectin was filtered and dried in a vacuum oven (VDL 23, Binder, Germany) at 55 °C for 16 hours, then grounded into powder form. The percentage of pectin yield was calculated (Equation 1) and expressed as yield (%).

\[
Pectin\ yield\ (\%) = \frac{\text{Weight (g)dried pectin}}{\text{Weight (g)dried durian rinds powder}} \times 100\%
\]

2.5 Spectroscopic analysis and determination of degree of esterification (DE)

FT-IR spectra of the pectin were collected using a Perkin Elmer Spectrum Two (USA) with wavenumber from 450 – 4000 cm\(^{-1}\) in attenuated total reflection (ATR) mode at a resolution of 4 cm\(^{-1}\). The DE of the pectin was calculated using the equation as shown in Equation 2 [26]. The peak area for the esterified carboxyl peak between 1730 cm\(^{-1}\) – 1720 cm\(^{-1}\) (limits of bands: 1830 – 1695 cm\(^{-1}\)) and non-esterified carboxyl peak between 1630 cm\(^{-1}\) – 1600 cm\(^{-1}\) (limits of bands: 1695 – 1570 cm\(^{-1}\)) were identified using instrument software (Spectrum, Version 10.4, UK)) [27].

\[
DE\ (%) = \frac{\text{Area of esterified carboxyl group}}{\text{Area of esterified carboxyl group + area of non-esterified carboxyl group}} \times 100\%
\]

2.6 Experimental design and statistical analysis

The extraction of pectin from each durian rind varieties, the FTIR analysis and the DE of each pectin were done and obtained in triplicate. ANOVA was performed followed by Tukey’s test with significant difference considered at values of p < 0.05, using a statistical software (Minitab, Version 18.1, USA).

3. Results and Discussion

3.1 Yield of pectin according to durian variety

The yield of pectin extracted from Musang King (D197), D24, Hajjah Hasmah (D168) and mixed varieties durian range from 5.60 to 7.73% as shown in Table 1. Among all varieties, pectin extracted from Hajjah Hasmah (D168) has the least yield (5.60%), which is significantly lower than other varieties. On the other hand, pectin yield produced from rinds of Musang King (D197), D24 and mixed varieties showed no significant difference among each other (refer to Figure 1). The amount, chemical structure and chemical composition of pectin could differ relying on plant species and varieties due to genetic factors (e.g., gene expression) or development factors (e.g., climate) of the plants [28–30]. This is in agreement with the findings of Canteri-Schemin et al. 2005 which had reported a statistical difference of pectin yield among different varieties of apple pomace [31]. Besides, a significant difference in the amount of pectin isolated from six genotypes of okra was also found [32]. Generally,
the yield of pectin obtained in this study is lesser than the pectin isolated from other sources, for example, apple pomace, citrus, passion fruit, dragon fruit, and jackfruit peel [21,24,33–35].

Table 1. Yield and DE of pectin extracted from different durian rind varieties

| Durian Varieties       | Yield (%)   | DE (%)     |
|------------------------|-------------|------------|
| **D24**                | 7.37 ± 0.45\textsuperscript{a} | 67.18 ± 1.70\textsuperscript{b} |
| **Hajjah Hasmah**      | 5.60 ± 0.20\textsuperscript{b} | 76.71 ± 3.57\textsuperscript{a} |
| *(D168)*               |             |            |
| **Musang King**        | 7.73 ± 0.23\textsuperscript{a} | 65.01 ± 3.26\textsuperscript{b} |
| *(D197)*               |             |            |
| **Mixed Varieties**    | 7.10 ± 0.40\textsuperscript{a} | 60.79 ± 2.09\textsuperscript{c} |

\textsuperscript{a, b, c} Different small case superscript letters in the same column indicate a significant difference (p < 0.05), with the same letters, show an insignificant difference (p > 0.05)

Figure 1. Yield of pectin extracted from different type of durian varieties

3.2 Degree of esterification of pectin according to durian variety

DE of pectin indicates “the percentage of esterified galacturonic acids over the total galacturonic acids residues” which determines the gelling properties and setting time of pectin. As shown in Table 1, the pectin extracted from all varieties of durian rinds (DE range from 60.79 to 76.71%) can be classified as high methoxyl pectin (HMP) with DE exceeds 50%. Wai et al. 2010 reported that most of the durian rind pectin extracted had DE more than 50% [16]. The durian rind pectin has a similar range of DE as compared to banana peel, mango peel, pumpkin peel and jackfruit peel [36], [22,37,38].

The DE of pectin isolated from mixed varieties of durian rinds is the lowest (60.79%), which is significantly lower than the pectin extracted from other varieties. In contrast, the pectin produced by Hajjah Hasmah (D168) shows the highest value of DE (76.71%), which is distinctly higher as compared to the other pectin. Among them, pectin extracted from D24 and Musang King (D197) do not differ
significantly from one another (refer Figure 2). These findings support that the variation in chemical structures of the cell wall polysaccharides is related to the genetic factors of plants [29].

HMP can be further classified as rapid set (DE% > 70%), medium rapid set (65% < DE < 70%) and slow set (58% < DE < 65%) [39,40]. In this study, pectin of Hajjah Hasmah (D168) (DE 76.71%) durian rind can be classified as rapid set pectin whereas pectin of Musang King (D197) (DE 65.01%) and D24 (DE 67.18) are medium rapid set pectin. On the other hand, pectin of mixed varieties durian rind (DE 60.79%) is slow set pectin. HMP forms gel at acidic condition (pH 2.0 – 3.5) with high soluble solids content [15]. Typically, HMP form gel at about pH 3.0 – 3.1 while slow set pectin needs a little lower pH than rapid set pectin to yield gels with a similar strength [41].

![Figure 2. DE of pectin extracted from different type of durian rind varieties](image)

3.3 Spectroscopic analysis

Figure 3 shows the FT-IR spectra of pectin extracted from the different variety of durian rinds. In the first region (3500 cm$^{-1}$ – 1800 cm$^{-1}$), there is a broad area of absorption between 3600 cm$^{-1}$ – 3200 cm$^{-1}$ which represents the O-H stretching absorption due to intra- and intermolecular hydrogen bonding of the galacturonic acid polymer in pectin samples. Another band found in this region is between 3000 cm$^{-1}$ – 2800 cm$^{-1}$ corresponds to C-H absorption which includes CH, CH$_2$ and CH$_3$ stretching and bending vibrations [42]. In the second region between 1800 cm$^{-1}$ – 1500 cm$^{-1}$, there are two peaks which are represented by C=O stretching vibration of methyl esterified groups (COOCH$_3$) and COOH (1730 cm$^{-1}$ – 1720 cm$^{-1}$) and the ionized carboxylate group (COO$^-$) found in the region (1630 cm$^{-1}$ – 1600 cm$^{-1}$) [32,43]. The ratio percentage of the area of these two absorption bands defines the degree of esterification of the pectin samples as shown in Equation 2. The third region lesser than 1500 cm$^{-1}$ indicates the ‘fingerprint region’ which are distinct and specific for each polysaccharide [27]. The band at about 1230 cm$^{-1}$ shows -CH$_3$CO stretching [44]. The strong absorptions between 1100 cm$^{-1}$ – 950 cm$^{-1}$ portray the ether linkage (C-O-C) and hydroxyl group in the pyranose ring [45]. The FT-IR spectra in Figure 3 show that the pectin from the different variety of durian rinds vary from each other in region II and III. This confirms that the pectin isolated from the different variety of durian rinds could differ from each other.
Figure 3. FT-IR spectra of pectin from different variety of durian rinds: (a) Hajjah Hasmah (D168), (b) D24, (c) Musang King (D197) and (d) Mixed Varieties

4. Conclusion
The pectin extracted from Musang King (D197), D24, Hajjah Hasmah (D168) and mixed varieties of durian rind has yield range from 5.60 to 7.73%. Hajjah Hasmah has the least yield of pectin (5.60%) which significantly lower than other varieties, whereas the pectin yield of the remaining varieties (7.10 to 7.73%) do not differ significantly. All the pectin obtained in this study can be classified as HMP with DE more than 50%. The result obtained from this study can be taken into consideration for the sample preparation of pectin extraction from durian rinds. In future research, studies on comparison among different methods of pectin extraction and more comprehensive physicochemical and functional characterization of pectin could build a good foundation for the application of pectin obtained in various industries including food and pharmaceutical industries.

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