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Characterisation and physiochemical properties of mango peel pectin extracted by conventional and phase control microwave-assisted extractions

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Abstract

Natural pectins from fruit peel have applications in the food industry as food stabilisers, sources of dietary fiber and as bio-waste. Here, we investigated the physicochemical properties of mango-peel pectin, after using conventional extraction and phase control microwave-assisted extraction (PCMAE). Due to its effectiveness as a bio-waste, a high peel-to-fruit weight ratio and fiber content, we used the Sam-pee mango cultivar from Northern Thailand for this study. The highest pectin yield found was 10.45% (w/w) after using PCMAE at 500W. The characterisation of extracted pectin revealed that the equivalent weight was higher when pectin was extracted using PCMAE, while the methoxyl content was lower and the degree of esterification remained constant (~55%). The PCMAE at 500W provided an emulsion activity pattern similar to that of the conventional extraction method. Phytochemical properties of the pectin however, measured by total phenolic content and antioxidant scavenging activities, were not significantly different ($p < 0.05$) between PCMAE and conventional methods. At a higher microwave radiation during the PCMAE (900W), the levels of antioxidants measured in the mango-peel pectins declined.

Introduction

Pectin is a polysaccharide primarily found in plant cell walls, of which the chemical structure consists of a homopolymer of partially methyl-esterified(1→4)-d-galacturonic acid. Extracted pectin of plant origin is commercially used as a food stabilizer, thickener and emulsifier, as well as a gelling agent in both the food and the pharmaceutical industries (Joye and Luzio, 2000). Pectin is also considered to be an important dietary fiber, in that consuming food that contains this non-digestible component is claimed to reduce the risk of developing chronic and degenerative diseases, such as atherosclerotic cardiovascular diseases, insulin resistance and type II diabetes, along with certain types of cancer (Goñi and Hervert-Hernández, 2011). Even though the demand for pectin in the global food industry is continuously increasing, pectin production is limited by plant source availability, particularly from the fruit wastes generated during food processing, such as apple pomace and citrus rind (Thakur *et al*., 1997). The price of pectin is mainly based on its quality and type, which are mostly ranked according to physicochemical characteristics, including the degree of esterification, the level of galacturonic acid, neutral molecular weight and other important biological functionalities (Jiang *et al.*, 2012). However, such parameters can be largely influenced by the extraction conditions applied to fruit peel in order to obtain pectin of commercial quality (Fishman *et al*., 1984; Joye and Luzio, 2000; O'Donoghue and Somerfield, 2008; Jiang *et al.,* 2012).

Conventionally, pectin is extracted with a diluted HCl solution coupled with heating, a process that consumes large amounts of time and energy (May, 1990; Bagherian *et al.*, 2011). Moreover, thermal processing leads to the degradation of pectin, thus impairing its biological functionality. Other methods of pectin extraction are currently under investigation,

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some of which are giving acceptable outcomes. Among these, microwave-assisted extraction (MAE) has been suggested to be a promising technique, as it increases in the yield and the quality of extracted pectin (Fishman *et al*., 1999; Fishman *et al.*, 2006; Bagherian *et al*., 2011). In conventional extraction methods, the "on-off" operating of the microwave, can lead to overheating of the raw material, which ultimately results in low product quality. To overcome this problem, the MAE unit used in this study included a phase controller (PCMAE), which regulates the electrical power input into the magnetron, thus generating continuous and adjustable microwave power, and enabling the operator to control both the heating rate and the extraction temperature simultaneously (Sommano *et al.,* 2015).

With a global production that reached nearly 40 million tons in 2011, mangoes (*Mangifera indica* L.) are among the most commercially grown tropical fruits (FAOSTAT, 2013). Besides for personal consumption, mangoes are mainly produced for industrial food processing purposes and account up to 62% of the total exports of fruits and vegetables in India, the world's largest mango producer (Berardini *et al*., 2005). The pulp of the mango is mostly used as a raw material for juices, nectars and concentrates, and makes up to 33–85% of the fresh fruit, while the proportions of peel and kernel vary between 7–24% and 9–40%, respectively (Wu, Chen and Fang, 1993). Larrauri *et al.* (1996) estimated that during the course of industrial mango processing, the waste or the by-product (peel and kernel), adds up to 35–60% of the total weight of the fruit. The Sam-pee mango cultivar used in this study is indigenous to the Northern part of Thailand. Due to its brilliant golden-orange colour, wonderful aroma and flavorful taste, it is extensively used in the preparation of juices and fruit wine rather than for fresh consumption (Srisamatthakarn *et al*., 2003). Maneepun and Yunchalad (2004) and Monck and Pearce (2007) estimated that approximately 9% of the overall mango production (ca. 300,000 tons in 2014) is used towards food processing in Thailand, the Sam-pee and Kaew cultivars being generally preferred over other cultivars.

To the best of our knowledge, little is known about the effects of microwave processing on the qualitative and quantitative characteristics of pectin extracted from mango peel. In this study, we put into practice a novel microwave extraction technology known as PCMAE to investigate the yield, the physiochemical properties and the quality of pectin extracted from Sam-pee mango peel. We then compare our findings to those of conventional extraction methods.

Material and Methods

Raw material

Sam-pee mangoes were harvested at a commercial ripening stage at the Maejo University orchard in the Sansai district of Chiang Mai. The length, width, and thickness were measured at the maximum points of the fruit (n = 10). The ratio of peel weight to total fruit weight was determined by averaging the measures of 10 ripe fruits. The moisture, ash, and fiber contents of the fruit peels were determined by previously approved procedures (AOAC 2000; Jiang *et al*., 2012) and the peel color (CIE L^*, a^*, b^* system) was assessed using a Minolta CR300 chroma meter with illuminant D65 and observer 2° (Konica Minolta®, Tokyo, Japan). The peels were stripped with a peeling knife and then dried in a hot-air oven at 60°C. The dried peel was ground to a fine powder in a high-speed food processor, and passed through a sieve, resulting in a final mass of particles smaller than 0.6 mm in diameter (Bagherian *et al*., 2011; Maran *et al.*, 2015).

Extractions of mango peel pectin

Conventional method

The methods of Koubala *et al.* (2008) and Jiang *et al.* (2012) were followed. Twenty grams of peel powder were mixed with 600 mL of diluted acidic solution (distilled H_2O adjusted to pH 1.5 with 2 M HCl). The slurry mixture was then stirred in a water bath at 85°C for 1.5hr and filtered through a 200-mesh filter cloth. The filtrates were centrifuged at 4800rpm for 20 mins to eliminate any remaining coarse particles. After precipitating the filtrates with two volumes of 95% ethanol and keeping for 1hr prior to filtration, the wet pectins were successively washed three times with 65%, 85%, and 100% ethanol, respectively. The obtained pectins were first dried overnight at 50°C, and then ground to powder, prior to analysis. The yield (%) of pectin was calculated from the following equation (Maran *et al*., 2015)

$$\text{Yield}\ (\%) = \left(\frac{M_0}{M}\right) \times 100 \qquad (1)$$

Where; M_0 (g) = the weight of dried pectin
M (g) = the weight of dried mango peel powder

Microwave-assisted methods

The pectin from Sam-pee mango peel was extracted using a phase-power control microwave extraction system (Sommano *et al.*, 2015). After

treating the samples with the acidic solution, they were held in the microwave oven and irradiated at a frequency of 2450 MHZ at 500 W (PCMAE 500) and 900 W (PCMAE 900) of power for 3 mins and 1 mins, respectively, according to the conditions described by Wang *et al*. (2007) and Bagherian *et al.* (2011). This process was then followed by the precipitation, washing and drying methods mentioned above. The yield (%) of pectin was also calculated accordingly.

Characterisation of pectins

Methoxyl content (Mox), anhydrouronic acid content (AUA) and degree of esterification (DE). The methods suggested in Ranganna (1986) and Pinheiro *et al.* (2008) were followed. Dried pectin (0.2 g) was stirred in CO_2-free distilled water (20 mL) until fully dissolved. One gram of NaCl was added to the solution, prior to titrating with 0.1 N NaOH in the presence of phenolphthalein. The volume was recorded as the initial titre (V1). Then, a 0.1 N NaOH solution (10 mL) was added to a neutralized polygalacturonic acid sample after the determination of the free carboxy groups. The solution was mixed thoroughly until the color of the solution became purple. A few drops of the indicator (0.25 N HCl) were added, and the mixture was titrated with 0.1N NaOH until the color turned from yellow to pink. This time, the volume was recorded as V2. The Mox, AUA and DE were then calculated using the following equations:

$$\text{Mox} = \frac{(N)(V2)(E)}{1{,}000\ (S)} \quad (2)$$

$$\text{AUA} = \frac{17.6\ (V1+V2)}{S} \quad (3)$$

$$\text{DE} = \frac{V2 \times 100}{V1+V2} \quad (4)$$

Where; S = mass of dried pectin (g)
N = 0.1 N (NaOH)
V1 = Vol of NaOH used (V1)
V2 = Vol of NaOH used (V2)
E = equivalent weight of methoxyl = 31

FT-IR spectrometry

FT-IR analysis was performed using an infrared spectrometer (Nicolet 6700, USA) equipped with MCT Detector (Mercury cadmium telluride). Each sample was scanned by placing the sample side down on the ATR diamond crystal and applying the pressure tower. The spectrum was recorded in the transparent mode from 900 to 4000cm^{-1}, with a resolution of 4.0cm^{-1}. Each IR spectrum was corrected for optical effects with the ATR correction algorithm (OMNIC software).

Physiochemical properties of pectin

Scanning electron microscope (SEM)

Pectin powder was mounted onto a specimen stub with a double-sided tape and sputter coated with gold (Jiang *et al.*, 2012). In comparison to commercial citrus pectin, the samples were morphologically viewed using SEM (JELO JSM-5910, Japan) with an accelerating voltage of 10 kV. The images were zoomed at magnifications of × 100 and × 500.

Emulsion activity

The emulsion activity was measured according to the method discussed in Jiang *et al.* (2012) and Lv *et al*. (2013). Pectin solutions were prepared from citrate buffer (0.1 M, pH 5.0), 0.5% (w/v) pectin, and 0.02% NaN_3. Series of oil-in-water (O/W) emulsions of 5–40% (oil volume/total volume) soybean oil to pectin solutions were prepared, which were then homogenized using a high-speed homogeniser (Omni International, USA) for 1 mins at 15,000 rpm. The oil in the water emulsion (50 µL) was mixed with 5 mL of 0.1% (w/v) sodium docecylsulphate (SDS). The absorbance was measured, thereafter, at 500 nm with a UV spectrophotometer (Biochrom, UK), and 0.1% SDS solution was used as the blank. The emulsion activity was calculated as the turbidity of the solution, using the following equation:

$$T = \frac{2.303 \times A \times D}{L} \quad (5)$$

Where; T = turbidity (1/cm)
A = absorbance at 500 nM
D = dilution factor
L = path length of cuvette (1 cm)

Extraction of bound phenolic compounds

The phenolic compound extraction of the bound phenolic compound was carried out according to the modified method suggested by Tian *et al*. (2004) and Ajila and Rao (2013). The above-mentioned extracted pectin (2 g) was hydrolyzed twice with 10 mL of 1 M sodium hydroxide containing 0.5% sodium borohydride under a nitrogen atmosphere. The hydrolyzate (viz., 20 mL) was acidified to pH 1 with 4M HCl, which was solvent extracted with the same amount of ethyl acetate. The extraction was done thrice and the solvent layers (top) were combined, and then evaporated to dryness under N_2 steam. Prior to accessing phenolic acids and antioxidant activities, the dried fraction was redissolved in 200 µL of 15% methanol (and used as the methanolic extract).

Total phenolic content

One hundred microliters of the methanol extract and gallic acid standards, or a 95% (v/v) methanol blank (100 µL) were added to 1 mL cuvettes containing 200 µL of 10% (v/v) Folin–Ciocalteu reagent (Singthong *et al.*, 1999). After 8 mins of incubation at room temperature, 800µL of 700mM Na_2CO_3 solution was added to avoid the oxidation of the phenolic compounds. The mixture was blended thoroughly and left to stand at room temperature for 2 hr. The absorbance of the solutions was measured at 765 nm, and the total phenolic content was calculated against gallic acid standards, considering that gallic acid is a major phenolic acid in both raw and ripe mango peels, as discussed in other studies (Ajila *et al.*, 2010; Ajila and Rao, 2013), and expressed as g GAE (gallic acid equivalent) per gram pectin using a 50 mg/L to 500 mg/L gallic acid standard curve.

Antioxidant scavenging activities

2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical assay. The DPPH free radical microplate reading assay was followed according to the discussion given in Molan *et al.* (2009) and Duan *et al.* (2011). The methanol extract (25 µL) was incubated with 250 µL of 0.2 mM DPPH in 95% ethanol in a 96-well microplate and in the dark for 30 mins. Thereafter, the absorbance of each sample was measured at 517 nm using a microplate reader. The blank contained methanol instead of DPPH solution.

Trolox equivalent antioxidant capacity assay

This assay was carried out as discussed in Sommano *et al.* (2013). Briefly, a 1:0.035 molar ratio of ABTS radical solution ($ABTS^{\bullet+}$) was prepared by mixing ABTS (7 mmol/L) with 2.45 mmol/L of potassium persulfate ($K_2S_2O_8$). The solution was left to stand overnight, in the dark and at room temperature, to allow the completion of the reaction and a stable absorbance to take place. To prepare the working solution, the mixture was then diluted with a known amount of sodium phosphate buffer (PBS, pH 7.4) to obtain the absorbance as 0.40 at 734 nm. An adequate dilution (10 µL) of the sample was added to 200 µL of the diluted $ABTS^{\bullet+}$ solution in each well of the microplate, and the absorbance reading was taken immediately within 1 mins after the initial mixing, for 6 mins, at 734 nm.

In both methods, the dose-response curves were derived from seven concentration levels (triplicate readings) of trolox (50–500 mg/L). The values were expressed as Trolox equivalent (TE) per gram pectin (Dorta *et al*., 2013).

Statistical analysis

Unless otherwise stated, all analyses were conducted in true triplicates (i.e. from 3 extracted samples). The results are expressed as mean ± standard error (SE) of these three measurements. The differences between the mean values were analyzed using an Analysis of Variance (ANOVA) test in Minitab 15 software (Minitab, USA) and a significant difference was set at $p < 0.05$

Results and Discussion

Physiology and chemical composition of the Sam-pee mango cultivar

Sam-pee mango is considered to be a small mango cultivar (length = 9.4 cm, width = 5.6 cm, thickness = 4.8cm) in comparison to other cultivars such as Nam Dok Mai (length = 13.9 cm, width = 6.9 cm, thickness = 5.9 cm) and Chok Anan (length = 11.3 cm, width = 6.9 cm, and thickness = 6.0 cm). However, the ratio of the peel weight to the total fruit weight of this cultivar was significantly higher (7.6%), compared to the ratios of Nam Dok Mai (5.3%) and Chok Anan (6.01%) (Sommano *et al*., 2014). Because they generate more reusable biomass, Sam-pee mangoes are often preferred over the other cultivars as a raw material for pectin extraction in food processing. Physicochemical properties of Sam-pee mangoes, such as ash and fiber contents, which are accepted estimates of total pectin yield, were significantly higher than those of Nam Dok Mai and Chok Anan (Table 1). Further differences in physiology and chemical compositions between Sam-pee and the two other mango cultivars are reported in Table 1.

Characterisation of pectin extracted from Sam-pee mango

The biochemical characterisation of Sam-pee mango pectin is reported in Table 2. The average yield from the Sam-pee pectin extraction carried out by conventional heating was approximately 8.8%, which is relatively low when compared to the quantity of pectin extracted from the peels of the Améliorée mango cultivar (10%) and of lime (19.8%) (Koubala *et al.,* 2008). Heating with different microwave field intensities of 900 W and 500 W however yielded different relative amounts of pectin, 8.6% and 10.45%, respectively. The PCMAE 500 W method thus showed a remarkable increase in pectin yield in comparison to the conventional method. The total heating time was 90 (900 W) and 30 (500 W) times faster when the extraction was performed under microwave heating, which is in accordance to work by Bagherian *et al.* (2011). Microwave heating is

Table 1. Physiological and physicochemical characteristics of Sam-pee mango fruit and peel as compared to other ripe type cultivars.

	Sam-pee	Nam Dok Mai	Chok Anan
Physiological characteristics			
Length of fruit (max, cm)	9.36 ± 0.40^{c}	13.88 ± 0.11^{a}	11.29 ± 0.59^{b}
Width of fruit (max, cm)	5.57 ± 0.20^{b}	6.91 ± 0.25^{a}	6.92 ± 0.47^{a}
Thickness of fruit (max, cm)	4.76 ± 0.21^{b}	5.90 ± 0.13^{a}	6.01 ± 0.34^{a}
Peel colour (L*, a*, b*)	47.43 ± 3.51^{b}	50.90 ± 4.34^{b}	55.06 ± 3.24^{a}
	3.28 ± 4.41^{b}	4.82 ± 2.35^{b}	6.41 ± 2.22^{a}
	20.36 ± 3.79^{a}	16.59 ± 3.09^{b}	22.42 ± 2.70^{a}
Peel to total fruit weight ratio (%)	7.58 ± 0.91^{a}	5.31 ± 0.38^{c}	6.01 ± 0.75^{b}
Physicochemical characteristics			
Moisture (%)	42.63 ± 5.44^{a}	37.09 ± 0.32^{a}	33.24 ± 2.35^{b}
Ash (%)	5.19 ± 0.11^{a}	3.86 ± 0.39^{c}	4.37 ± 0.01^{b}
Fibre content (%)	16.72 ± 0.35^{a}	11.48 ± 0.54^{b}	8.64 ± 0.31^{c}

Values are means of five replications ± standard error.
Values within a row followed by a different lower case letter are significantly different ($p < 0.05$).

Table 2. Biochemical characterisation of pectin extracted from Sam-pee mango peel using conventional and PCMAE extraction methods.

Extraction methods	Conventional	PCMAE 500	PCMAE 900
Time (min)	90	3	1
Temp (°C)	85	75	83
Characterisation			
Pectin yield (%)	8.77 ± 1.33^{b}	10.45 ± 0.40^{a}	8.63 ± 0.60^{b}
Mox (%)	6.60 ± 0.88^{a}	1.21 ± 0.12^{b}	1.08 ± 0.26^{b}
AUA	63.43 ± 14.13^{a}	11.08 ± 1.57^{b}	13.36 ± 1.61^{b}
DE (%)	56.49 ± 5.91^{a}	52.33 ± 6.26^{b}	54.04 ± 5.28^{a}
Methanolic extract of bound dietary fiber			
Antioxidant scavenging activities			
DPPH (mg TE / g)	690.99 ± 1.33^{a}	691.25 ± 0.69^{a}	588.45 ± 0.75^{b}
TEAC (mg TE / g)	793.09 ± 12.23^{a}	801.02 ± 15.23^{a}	620.59 ± 13.13^{b}
Total Phenolic Content (mg GAE / g)	161.97 ± 6.15^{a}	138.40 ± 5.99^{b}	105.67 ± 5.05^{c}

Values are means of three replications ± standard error.
Values within a row followed by a different lower case letter are significantly different ($p < 0.05$).
PCMAE 500 = phase control microwave-assisted extraction at 500 W
PCMAE 900 = phase control microwave-assisted extraction at 900 W

indeed more efficient than other extraction methods due to the intense formation of vapour in polar substances generated by the electromagnetic field (Kratchanova *et al*., 2004). Heat vapour modifies the cell wall matrix and leads to the severing of parenchymal cells, which rapidly and extensively opens the skin tissues, thus increasing the interaction between the extracting agent and the plant material during the extraction process (Wang *et al*., 2007). Our findings not only show that pectin yield with a PCMAE method can be equal or higher than with conventional extraction techniques (Table 2), but also that a higher pectin yield can be obtained at a lower extraction temperature, i.e. at 75°C (PCMAE 500) vs. 85°C (conventional method) and 83°C (PCMAE 900).

Methoxyl (Mox) content is an important factor in indicating the setting time of pectins, their sensitivity to polyvalent cations and their beneficial properties in the preparation of low solid gels, films, and fibers (Shaha *et al.,* 2013). Mox content of pectin extracted from plant resources generally varies from 0.2% to 12%, depending on the source of the raw material and on the mode of extraction. The Mox content obtained by using the conventional method falls within this range (6.6%). The PCMAE method seemed to have a greater impact on Mox (1.08%, and 1.21%, for PCMAE 900 and 500 respectively). Also, the DE content of Sam-pee mango was slightly lower with the PCMAE method. According to these results, PCMAE seems to privilege the extraction of "low methoxyl pectin" (LMP) from the peels of Sam-pee mango fruit, which is believed to be an indicator of good quality pectin for food processing (Aina *et al.* 2012). Indeed, LMP can form gels with lower amounts of sugar or even without sugar in divalent cations, and is thus preferable for low sugar diet food (Shaha *et al.*, 2013). The lower DE we found could also be attributed to a lesser deesterifying action of the hydrochloric acid upon pectin solubilisation (Yapo, 2009; Kumar and Chauhan, 2010; Jiang *et a*l., 2012).

A high AUA content suggests an impurity of the extracted pectin (> 65%), which is mainly caused by the presence of sugar during alcohol precipitation (Food Chemicals Codex 1996; Shaha *et al.*, 2013). The AUA content of the pectin extracted with PCMAE was very low (~13%) compared to the conventionally extracted sample (~63 %). However, since the same alcohol precipitation method was followed in both extracting methods, the large difference in AUA may not be due to the presence of sugar moiety alone. Ismali *et al.* (2012) suggested that fruit proteins could also contribute to the impurity of the pectin that alcohol precipitation alone would not be able to eliminate. Following this argument, PCMAE may also facilitate the dissolvability of proteins present in the plant extract. Further investigations on protein impurity as well as into an appropriate precipitation

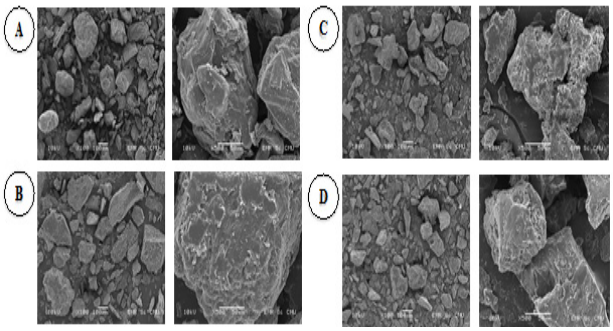

Figure 1. The SEM images of commercial pectin (citrus) (A), pectin conventionally extracted from Sam-pee mango (B), pectin obtained using microwave-assisted extraction from Sam-pee mango at 500 W (C), and pectin obtained using microwave-assisted extraction from Sam-pee mango at 900 W (D). The images were viewed at × 100 (left) and × 500 (right).

technique are required.

To identify the major functional groups of Sam-pee mango pectin, an analysis of the FT-IR spectra was performed. Figure 2 shows the FT-IR region ranging from 900 to 4000 cm^{-1} of conventionally extracted pectin and PCMAE extracted pectin from Sam-pee mango. The FT-IR spectra of the Sam-pee mango extracted with different modes of extractions exhibited similarities in their transmittance (%T) patterns. A distinct peak at around 3400cm^{-1} was likely due to the stretching of the hydroxyl groups, whereas a band at around 3000cm^{-1} was attributed to the C–H stretching of the CH_2 groups (Jiang *et al.,* 2012). The strong absorption observed at 1730–1760 cm^{-1}, characteristic of esterified pectins, arising from the ester carbonyl stretching band, and peaks at 1600–1630 cm^{-1} and 1400–1450 cm^{-1} were due to the anti-symmetric and symmetric stretching frequencies of the ionic carboxyl groups (Jiang *et al.*, 2012; Posé *et al.*, 2012). The absorptions at ~1750 cm^{-1} and ~1600 cm^{-1} were also used to estimate the DE value of pectin. The rationale for this is that the absorbance intensity of the ester carbonyl groups has a positive relation with DE, whereas the correlation with the intensity of the ionic carboxyl stretching is negative (Singthong *et al*., 2004). The intensities of the peaks at ~1750 cm^{-1} and ~1600cm^{-1} were similar, suggesting that the different extraction methods had a lesser influence on the DE; the calculated DE results were also in line with this result (Table 2). However, the intensity of the absorbance between 900 cm^{-1} and 1300 cm^{-1} increased in the PCMAE extracts, suggesting variability in their sugar compositions (Posé *et al.,* 2012). These absorption patterns are collectively known as the finger print region of C–O–C stretching, OH bending, and CH_3 deformation (Gan *et al.*, 2010; Jiang *et al*., 2012).

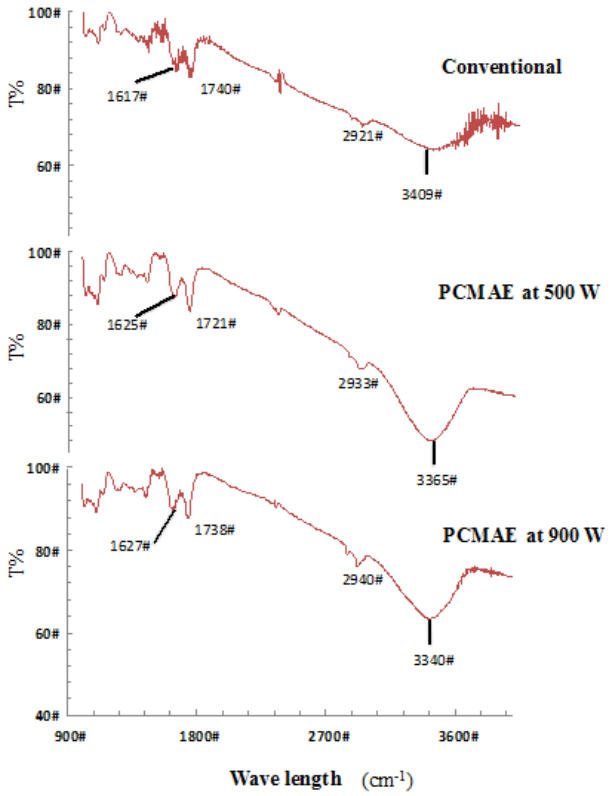

Figure 2. The FT-IR spectra of pectins extracted using conventional method, phase control microwave-assisted extraction (PCMAE) at 500 W, and PCMAE at 900 W, from 900 to 4000 cm^{-1} (x axis). T% is the percentage of transmittance (y axis).

Physiochemical properties of extracted pectin from Sam-pee mango

The SEM images in Figure 1 show the distinct surface structures of commercial citrus pectin (Figure 1A), conventionally extracted pectin from Sam-pee mango (Figure 1B), pectin extracted by MAE at 500 W (Figure 1C), and pectin extracted by MAE at 900 W (Figure 1D). The pectin particles of commercial citrus pectin and of Sam-pee mango pectin extracted by the conventional method displayed different shapes, Sam-pee mango pectin exhibiting pellets to lumpish particles, which differed greatly from the shape of the commercial citrus pectin, although the particles of both pectin types were of relatively smooth surfaces. However, the pectin particles extracted using the PCMAE 500 and PCMAE 900 methods were flakier in shape and with more porous surfaces (Figures 1 C and D compared to B). Therefore, our pectin morphology analysis indicates that the pectin formation was greatly influenced both by the extraction method applied and the source of the raw material. These results are in agreement with Jiang *et al.* (2012); our findings also suggest that different extractant methods can produce different pectin morphologies. Pectin particles with more

porous structures usually have a better solubility, than particles with the rigid structure and lower porosity, thereby increasing solution viscosity (Tamnak *et al*., 2016).

Besides their ability to increase the viscosity of the aqueous phase, pectins can also decrease the tendency of oil globules in the oil-water phase to move and coalesce; moreover, they can absorb surface oil to form a coating around the dispersed oil particles and can stabilise emulsions due to electrostatic repulsion (Zouambia *et al.,* 2009; Jiang *et al*., 2012). Figure 3 illustrates the turbidity of pectin emulsions, with higher turbidity indicating a greater emulsion activity (Jiang *et al.*, 2012). At 5% oil concentration, pectin extracted conventionally or using the PCMAE 500 method both demonstrated a very high emulsion activity. As oil concentration increased, emulsion activity equally showed a flat increase, with a peak of activity at 30% oil concentration. This increase in turbidity is seemingly due to the impedance of the flow water molecules through the oil mixture, which becomes stronger at higher oil concentrations (Sharma *et al*., 1997). However, the turbidity of the pectin extracted using the PCMAE 900 method was highest at the 25% oil concentration. The overall pattern suggested that Sam-pee mango pectin extracted with the conventional method had a higher emulsion activity at lower ranges of oil concentration (10-15%), while the pectin extracted using microwave assisted techniques exhibited higher activity at higher oil concentrations (20-40%).

Table 2 shows the bound phenolic content and the antioxidant activities of pectin extracted conventionally, compared to using the PCMAE 500 and PCMAE 900 methods. The phenolic content level in mango peels is approximately 20-100 mg GAE/g, depending largely on the stage of fruit ripening, on mango cultivars, as well as on the mode of extraction (Ajila *et al*., 2010; Kim *et al.*, 2010; Dorta, Lobo and González, 2013). Our results suggest that the methanolic extract of pectin exhibited a relatively high phenolic content (100-160 mg GAE/g). Pectin extracted conventionally had the highest phenolic content in our experiments, followed by pectin extracted using the PCMAE 500 and PCMAE 900 methods, respectively. The antioxidant scavenging activities exhibited by the DPPH and ABTS assays are also illustrated in Table 2. The pectin extracted conventionally exhibited higher levels of antioxidant activity, i.e. 600-800 mg TE/g, against both the DPPH• and $ABTS^{\bullet+}$ radicals, as compared to the levels of antioxidant activity obtained from mango peel of Keitt cultivar (150-200 mg TE/g) (Dorta *et al.,* 2012). The antioxidant activities resulting from the PCMAE 500 method, were in the same ranges as from the conventional method; however, at higher levels of microwave power (and for a shorter period of time), antioxidant activities of mango peel pectins significantly decreased.

Chen and Gong (2007) and Dorta *et al.* (2013) explained this phenomenon by stating that the faster heating induced by the PCMAE method results in a lower risk of thermal degradation, decomposition, or oxidation of the phytochemicals found in mango peel, in comparison to conventional extraction. The PCMAE methods also required considerably less extraction time, which is a non-negligible factor in the food processing industry. Li *et al.* (2012) previously concluded that the dissolubility of phenolic compounds can reach equilibrium after a short time and that they are thus not readily affected by a lengthening of the extraction time. Our findings were in agreement with these observations, in that 1 min of heating at a higher microwave power (900W) seemed to have an overall negative effect on the levels of plant phytochemicals, suggesting that a PCMAE method at lower microwave power (i.e. 500 W) with a slightly longer heating time (3 min) would be more beneficial in conserving the nutritional properties of mango peel pectin.

Conclusion

Among new natural sources of extractable pectin, mango peel has a promising potential as a raw material for the food industry, due to its worldwide production capacity and to the tremendous amounts of mango peel produced every year as unused bio-waste. The PCMAE techniques that we tested in this study have been successfully assessed as complementary methods for the extraction of mango pectin. Indeed, we obtained a significantly higher pectin yield with the PCMAE 500 technique and at a 30-fold lower extraction time than with the conventional method. The PCMAE 900 technique that we tested proved to be as efficient as the conventional extraction method in terms of yield, however with a 90-fold gain in extraction time. The characterisation of the extracted pectin suggested that equivalent weights were much higher, but that the methoxyl contents were lower in the PCMAE extracted pectin than in the pectin extracted with the conventional method. Most likely due to the reduction of extraction time and to the direct heating of the raw material, phytochemicals was generally well preserved. Our results however suggest that higher levels of microwave power (900W) during PCMAE may significantly reduce the intensity of phenolic and antioxidant scavenging

activities. Overall, our comparison of conventional and PCMAE techniques provides a solid basis for promoting the use of PCMAE in food processing methods in the future and offers a simpler and more time-efficient approach to extract pectin from fruit peel, which would be otherwise considered as bio-waste.

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