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Water-based extraction of pectin from flavedo and albedo of orange peels

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Abstract

The extraction of pectin from orange peels has been studied at different sample to solvent ratios, different pHs and with different extraction techniques using water. The highest pectin and total nitrogen was produced by a sample to solvent ratio of 1:12.5. The amount of pectin extracted reduces as the pH increased, while the extractability of crude protein is not affected so significantly. Previous workers have found similar results. Considerably more pectin was obtained by the Soxhlet method than by microwave extraction by a factor of two, with a longer extraction duration than the microwave extraction by a factor of 240, so microwave extraction showed a much higher extraction rate (per unit time) by a factor of 120. Pectin existed mainly in the albedo, but the flavedo still contained 27% of the amount of pectin in the total extract. The total pectin yield from the dried peel was 2.2%. The combination of hand-pressure and microwave on pectin yield from flavedo was 12% better than hand-pressure alone, which was also better than microwave extraction alone.

Keywords: Extraction; Wastes; Products; Oranges; Pectin; Citrus; Microwave; Soxhlet

1. Introduction

When processing fruit and vegetables, the capacity of waste management systems is challenged by the concomitant by-products, such as peels, skins, and pips. Current research has been classified into five areas of by-products use [1]: (1) bioremediation, included with anaerobic fermentations, methanogenesis and initial remediation; (2) land fertilizer and animal feed; (3) bio-transformations and bio-peroxidase catalysts; (4) biochemical reactions such as bio-fuels and bio-degradable plastics; (5) separation and recovery of components, such as extraction. Many by-products are currently reused to fertilize the land and soil, to feed animals as nutrients, and to generate energy. More valuable re-uses of by-products would be a significant benefit. For biomass by-products, there are still some useful contents that can be extracted from peels before using the peels as fertilizers, animal feeds or bio-fuels. In this work, extraction from orange peels is discussed and analysed.

Orange production is a significant agricultural product in Australia and competes with the production of mandarins. According to the Australia Bureau of Statistics [2], orange production has progressed substantially, and the manufacture of juice has increased to become a significant industry. There has been a dramatic increase in fresh orange juice sales in the US$ 825 million fruit juice industry in Australia. Orange juice now enjoys the largest share of juice production [2]. Orange production was 8.40% of the total agricultural production in NSW, Australia, from 2002 to 2003, and Navel oranges are widely planted. The peel represents about half of the fruit mass. The orange by-products (peels and tissues) are roughly four-ninths of the complete fruit [3]. With significant juice production, orange peels, as major wastes, have become a substantial burden to the environment. Hence it is necessary to find a feasible way to dispose of the peels to have a positive environmental impact or to turn them into useful products.

There are two types of fibres, insoluble fibre and soluble fibre. The insoluble fibre includes hemicellulose, cellulose and lignin and they were found in beans and fruits with edible seeds. The soluble fibre includes gums and pectin [4]. The benefit of insoluble fibre is that water binds to it, giving the fibre a bulking effect and improving the efficiency of the gut and colon, while the beneficial effects of soluble fibre include an increase in the growth rate of natural bacteria in the digestive tract, improving the process of digestion [5].

Pectin is a complex carbohydrate, which is a significant component of the primary cell wall of plants. Pectin is a polymer of α-galacturonic acid (at least 65%) with a variable number of methyl ester groups. Pectin from orange peels has wide

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applications. It has been used in food industry as a thickener, a texturiser, an emulsifier and stabilizer; it has been used as a gelling agent in jams and jellies; it has also been used as a fillers or stabilizer in confections, dairy products, fruit preparations, bakery fillings, icings and frostings; other applications include fat replacers in spreads, salad dressings, ice cream and emulsified meat products [6]. Behall and Reiser [7] have suggested that pectin, as a kind of dietary fibre, lowers blood cholesterol levels and low density lipoprotein cholesterol fractions without changing high density lipoprotein cholesterol or triglycerides, which are good for human health.

Classically, the process steps for the industrial production of pectin include extraction from the raw material, purification of the liquid extract and isolation of pectin from the liquid extract [8]. Microwave pretreatment of fresh orange peels can increase the capillary-porous characteristics and the water absorption capacity of the plant material. The high temperatures involved in microwave pretreatment can also inhibit the pectinesterase activity that can otherwise degrade the pectin before extraction in the oranges. These changes provide an opportunity for improving the yield of pectin and its molecular mass and gel strength [9].

Ethanol, hexane petroleum, HCl, methanol, chloroform and CO_2 are often used as solvents in the extraction process. Water, as a non-flammable, non-toxic, readily available and cheap solvent, is heavily used in the food industry. Pectic substances normally have little solubility in organic solvents, except for formamide, dimethylsulfoxide, dimethylformaide and warm glycerol. Bucher [10] carried out a comparison of extraction solvents for pectic substances in vegetables and fruits (apple, cabbage, pea, potato and spinach) by using EDTA, CDTA, ammonium oxalate/oxalatic acid, urea with sodium acetate, urea without sodium acetate, guanidine hydrochloride and 4-methylmorhpoline-4-oxide. It was found that the quantity of pectic substances extracted by EDTA at neutral pH was significantly influenced by temperature, by as little as a 2 °C shift. Much more galacturonan was extracted by EDTA, CDTA, and ammonium oxalate than by milder, hydrogen-bond breaking agents, such as urea and guanidine.

Campbell and Palmer [11] found that citrus fruits, such as oranges, grapefruit and lemons, all have more than 2% pectin. Two chemical methods have been used to quantify galacturonic acid in pectin, including both free and esterified forms: (1) reaction with cysteine and (2) reaction with carbazole [12]. The carbazole method, modified by Bitter and Muir [13], gave more sensitivity and stability in the colour reaction used to quantify pectin. This method is based on the principle that galacturonic acid is one of the hydrolysates of pectin, which can be condensed by carbazole ethanol.

Fishman et al. [14] successfully extracted pectin from orange albedo by microwave preheating under pressure at a maximum level of 50 ± 2 psi (345 ± 14 kPa) and a maximum temperature of 195 °C. According to them, the molar mass, size and intrinsic viscosity of pectin were improved by microwave preheating under pressure, because the process of microwave preheating improved the structure of the plant matrix, helping pectin to be released from this matrix. In Table 1, the results of different extraction methods are compared, from which it can be seen that sequential extractions can produce higher yield than single extractions alone.

While the use of water to extract pectin is not, in itself, novel, previous research has not usually studied the separate parts, flavedo and albedo, in the orange peels, which is part of the novelty of this work. The main experimental techniques used in this study have included different methods, such as microwave extraction, Soxhlet extraction and hand-pressing extraction, and comparing all these methods for the same source of samples is also novel. The concentration of pectin in the extracts has been measured to give the pectin yield of each method. Total nitrogen in the extracts has been tested to indicate the level of crude protein. By comparing the pectin yield, total nitrogen concentration and the relative amount of extraction of extracts, a characteristic description of the final products is given.

2. Materials and methods

2.1. Materials preparation

Navel orange, purchased in the local fruit market, was chosen as the raw material in these experiments. The peels were stripped with a paring knife. The flavedo, which is the outside skin of the orange peel, was removed by a fruit peeler. The flavedo and albedo were cut into small pieces using a Tiffiny Fruit proces-

Table 1
Comparison of work on pectin extraction

Extraction method	Extraction conditions	Raw material	Results	References
Steam injection heating	*P*: 188.7 kPa; *T*: 95–100 °C; HCl solution with pH 2.0 as solvent; sample to solvent ration: 1–25; sequential extraction three times with 3 min each	Albedo	Sequence 1: 6.9% pectin; sequence 2: 7.5% pectin; sequence 3: 2.5% pectin; total: 16.9% pectin; all on dried albedo base	Fishman et al. [20]
Microwave extraction	*T*: 147 °C; HCl solution with pH 2.0 as solvent; sample to solvent ration: 1–5 and 1–25; sequential extraction three times with 3 min each	Albedo	Sequence 1: 7.2% pectin; sequence 2: 2.8% pectin; sequence 3: 5.3% pectin; total: 15.3% pectin; all on dried albedo base	Fishman et al. [14]
Microwave pretreatment	HCl solution with pH 1.5 as solvent; microwave pre-treatment at 0.45, 0.63 and 0.9 kW for 5, 10 and 15 min; conventional heat to 80–82 °C with continuous stirring, and duration for 1 h	Orange peel	14–18% pectin on dried peel base; yield increased with increasing of microwave power	Kratchanova et al. [9]

sor (Model: Mini Food Processor No MC9, Tiffiny) and were sealed in glass bottles. The diced peel particles looked granular. To get a representative sample, the flavedo and albedo from four oranges were well mixed within each (flavedo and albedo) sample, separately. During the experiment, the pH and concentration of samples were measured at different times. The duration of the extraction and its temperature were also controlled.

2.2. Experimental procedure

The experiments were conducted under different conditions and with different extraction methods. The flow chart, shown in Fig. 1, outlines the whole experimental procedure from material preparation to chemical analysis. Different methods, including microwave heating extraction and Soxhlet extraction, were studied in this experiment for the same source of oranges. Water, which is a low health impact, low cost, easily accessible agent, was chosen as the extraction solvent.

2.3. Microwave extraction

Microwave heating was performed in an oven made by Milestone Microwave Lab Stations. The chopped orange peel albedo or flavedo was washed with deionized water three times. The weighed albedo or flavedo sample and 50 ml of deionized water were placed in TFM (Teflon) vessels of volume 100 ml, which were surrounded by protection shields sealed with overpressure release valves. About four drops of 1 M HCl were added to adjust the pH to 2.00 ± 0.05 in most of the experiments. This pH was chosen for the microwave extraction based on previous research results, which indicate that orange pectin with a high degree of esterification generally results from acid extraction processes [15]. After an initial heating-up period of 5 min, the temperature was maintained at 150 °C for 15 min. An ATC-400-FO temperature probe was inserted into the assembled reference segment to measure and control the internal temperature by applying different microwave power levels. The products, after extraction, were cooled down for about 1 h to reach room temperature. The cold products were filtered with a piece of 80-mesh cloth.

2.4. Soxhlet extraction

To compare with the microwave extraction, a Soxhlet extraction was performed on a mass of orange albedo. In this experiment, the conical flask was filled with 200 ml deionized water as a solvent. The solvent was heated by a heater (Heidolph, MR30001) to the boiling point. Sixteen gram of albedo tissue was placed in a Whatman single thickness cellulose thimble (33 mm external diameter × 80 mm external length). The Soxhlet extraction was assumed to be complete when the solution in the Soxhlet extraction chamber was the same orange colour as the one in the flask. The extraction was allowed to proceed for 6 h at the boiling point of water. After 6 h, the product and the extraction residue were cooled down before doing further chemical analysis.

For both types of extraction, the total solids content of albedo and flavedo (on a wet basis) was measured after heating samples to a constant weight in a drying oven at 100 °C for 48 h. The solid contents of flavedo or albedo per unit mass of dried orange peel were calculated based on the following equation:

$$\text{Solid content of flavedo (albedo)(\% of dried peel)} = \frac{\text{Weight of flavedo(albedo)} \times \text{Total solids of flavedo(albedo)(\%)}}{\text{Weight of flavedo} \times \text{Total solids of flavedo} + \text{Weight of albedo} \times \text{Total solids of albedo}} \quad (1)$$

2.5. Hand-pressure extraction

In the hand-pressure procedure (a type of pressure extraction method), an external pressure that is applied to the orange

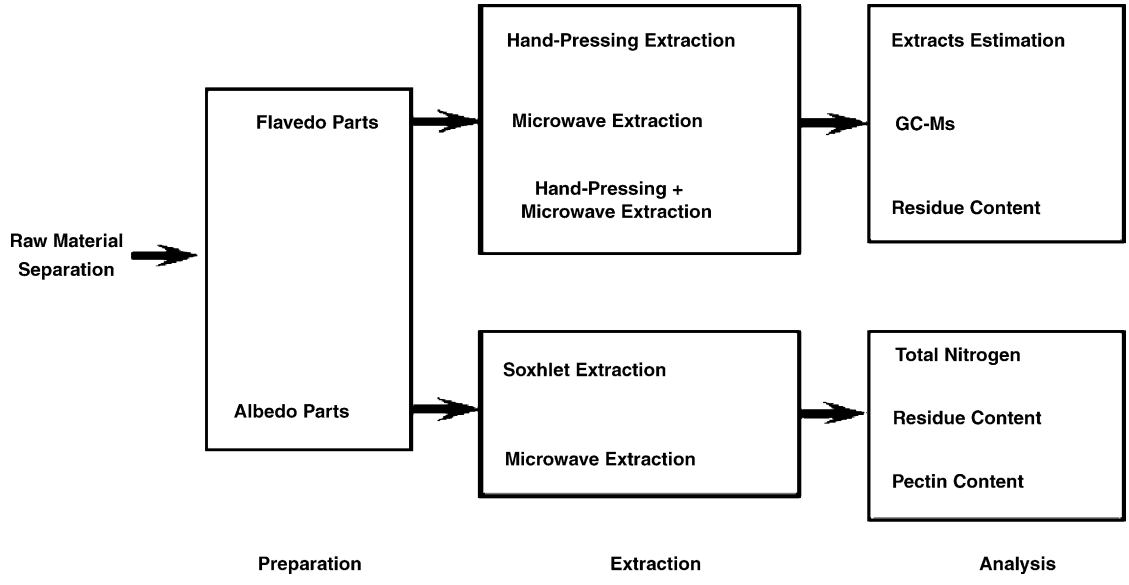

Fig. 1. Overall experimental procedure.

peels causes rupture of the cell walls, and subsequently pectin is liberated. Orange peel oil is also extracted in conjunction with the pectin. This type of extraction is usually favourable to the extraction of peel oil but not to the extraction of pectin.

1. A measured weight of chopped flavedo was washed with deionized water three times;
2. the washed flavedo was wrapped with a piece of 80-mesh cloth;
3. the cloth-wrapped flavedo was pressed by hand and leached with deionized water;
4. the solvent volume was controlled to a particular volume so that the pectin analysis and calculation on the same base;
5. the residue was collected for further analysis.

The pressure procedure for generating pectin from peel was carried out either along, as a separate method as its own, or combined with microwave extraction as a pretreatment in series.

The particle sizes of 10 samples of chopped flavedo and albedo were separately measured with a Vernier Caliper having a resolution of ±0.01 mm. The particle size in this paper means the length (the longest of three dimensions) of the granular peel particles.

The amount of protein, as a significant nutrient in food production, was assessed on the basis of the amount of total nitrogen in the extraction products. The amount of total nitrogen was determined by Merck Method 140. This method is based on the total nitrogen round-cell test using the Merck spectroquant analysis system with a Merck SQ 118 Photometer and a Merck Thermoreactor TR 300. According to Kennedy [16], the amount of crude protein can be calculated by multiplying the total nitrogen content by 6.25.

The amount of pectin was determined by IFJU Method 26. The principle of the method is that pectin is a polymer of α-galacturonic acid, which can be condensated by carbazole ethanol [12]. The pectin was coagulated with ethanol (95%) in a constant temperature water bath (Model: Thermoline BTC-9090) at 85 °C for 10 min. After discarding the supernatant, the pectin was washed with ethanol repeatedly until there was no sugar left by reacting the pectin with 5% naphthol–ethanol. If the colour of the mixture was purple, it indicated that there was sugar present. After coagulation and purification, the pectin from the products was collected in a 100 ml volumetric flask with 5 ml 1 M NaOH. One milliliter of the pectin mixture was flocculated with 0.5 ml 0.1% carbazole–ethanol indicator. The mixture had a white colour. After adding 6 ml 1 M H_2SO_4, the mixture was placed in a 85 °C water bath for 5 min. The concentration of pectin was measured by using a 1 cm cell in the spectrophotometer (Model: Cary 50 Spectrophotometer, Varian) at $\lambda = 525$ nm, immediately after a 15 min cooling period. To quantify the concentration of pectin in the extracts with the carbazole spectrophotometer method, in the preparation stage, pure pectin powder (Sigma product code P9135) from citrus fruit was used to calibrate a standard absorption curve for pectin. The absorbance measurements were repeated three times and then averaged. The relative amount of extraction in this experiment was calculated by dividing the change in weight between the dried raw sample and the dried residue by the weight of the dried raw sample. The weight of dried residue was measured after heating samples to a constant weight in a drying oven at 100 °C for 48 h.

3. Results and discussion

3.1. Solids content and particle size

The solid contents of the flavedo (53%) and the albedo (47%) were similar, which meant that the moisture content was almost the same throughout the orange peels. It was found that the total solid contents in both parts of the orange peel (flavedo and albedo), on a wet basis, were around 80%. The moisture contents were, accordingly, about 20%. The average particle sizes of chopped flavedo and albedo were both found to be 2.0 mm and the standard deviations were both 0.1 mm. The food processor appears to have handled the flavedo and albedo similarly. Generally, a larger specific surface area of the extraction sample will result in quicker extractions. However, a larger specific surface area of the peel requires more time and energy to shred the peel, which may be a practical trade-off in industrial situations. According to Mira and Blasco [17], the intraparticle diffusion resistance has a significant effect on the extraction rate for most extraction methods, when the particle size is around 5–10 mm. Hence these samples are probably sufficiently fine for mass-transfer effects not to be rate limiting.

3.2. Effect of the ratio of sample to solvent

To determine the most appropriate sample to solvent ratio to extract pectin, albedo to water ratios of 1:12.5, 1:25 and 1:50 were tested, while other conditions, such as the time for the experiment, the temperature, the pH and the experimental procedure, were kept the same, as shown in Table 2. Fig. 2 shows a decreasing trend of yield for crude protein and pectin in the range of ratios from 1:12.5 to 1:50.

The albedo to water ratio of 1:12.5 showed the highest yield of pectin and total nitrogen by a factor of two. Compared with the yields for the 1:25 and 1:50 ratios, the ratio of 1:12.5 appears to be the best among the three. Fishman et al. [14] used ratios of

Table 2
Effect of the sample to solvent ratio on the pectin yields

Sample to solvent ratio	1:12.5	1:25	1:50
Colour after extraction	Dark	Dark	Light
Total nitrogen (%)	0.4	0.1	Undetected
Crude protein (% of dried albedo)	2.3	0.5	Undetected
Crude protein (% of dried peel)	4.9	1.1	Undetected
Concentration of pectin (mg/l)	518	209	Undetected
Pectin yield (% of dried albedo)	0.8	0.5	Undetected
Pectin yield (% of dried peel)	1.6	1.1	Undetected
Extraction rate (min^{-1})	0.11	0.07	0

All solutions were adjusted to a pH of 2.00 ± 0.05. Microwave extraction was used for 15 min at 150 °C. All samples gave bitter aromas after extraction. The dry weight basis was determined by drying samples at 100 °C for 48 h.

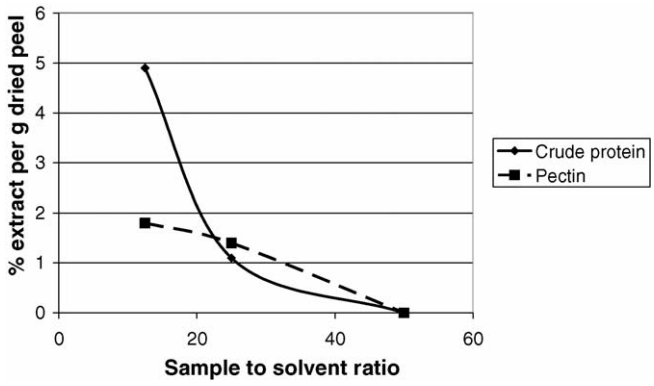

Fig. 2. The effect of different sample to solvent ratios on the crude protein and pectin levels from microwave extraction.

albedo to solvent of 1:5 and 1:25 under very similar conditions to ours (see the comparison of work on pectin extraction summarized in Table 1). They found that, when the albedo:solvent ratio was 1:25 (w/v), 20.2% of the pectin in the albedo was recovered after 3 min of heating, while at an albedo:solvent ratio of 1:5 (5:25, w/v), 19.6% of the pectin was recovered after 6 min of heating. The yield of pectin from the albedo (fresh weight basis) at a ratio of albedo to solvent of 1:5 and 3 min of microwave heating was 3.30%, compared with the literature value of 3.36% for fresh orange peels heated by conventional methods [9]. Combining the ratios of 1:5 and 1:25 published by Fishman et al. [14] and ours of 1:12.5, 1:25 and 1:50, this suggests that an albedo to solvent ratio of 1:12.5 gives the highest pectin yield.

For a sample to solvent ratio of 1:12.5, the total amount of nitrogen from the microwave extracts was 0.4% per gram of dried albedo, or 2.3% of crude protein per gram of dried albedo, or 4.9% of crude protein per gram of dried peel, which was close to the result of 6% from Kar and Arslan [18]. This ratio of 1:12.5 gave the best result of the three ratios studied here for both protein and pectin yield, and literature results also support the suggestion that this ratio is best.

3.3. Effect of pH

To determine an appropriate pH for maximum pectin extraction, a pH of 2 and the initial pH of the sample were chosen, while the other conditions were kept the same. Table 3 gives the conditions in this experiment, showing that the amount of extracted pectin reduced as the pH was increased, while the amount of crude protein was similar. Traditionally, orange pectin is extracted in heated water at 85 °C from citrus peels at a pH range of 1–3 for 30 min. In the previous study by Fishman et al. [14], pHs from 1 to 2 were usually chosen, with HCl used to acidify the samples. According to BeMiller [19], the highly hydrated carboxylate groups are repressed at the larger hydrogen ion concentrations corresponding to lower pHs. These carboxylate groups are converted into slightly hydrated carboxylic acid groups. This loss of charge can cause the polysaccharide molecules to become less repulsive, which can help pectin to form gel, giving more precipitated pectin at lower pHs.

Table 3
Effect of the pH on the pectin yields

	pH adjusted	No pH adjustment
Sample to solvent ratio	8.1736 g:100 ml H_2O	8.0345 g:100 ml H_2O
pH	2.00 ± 0.05	4.97 ± 0.05
Total nitrogen (% of dried albedo)	0.4	0.3
Crude protein (% of dried albedo)	2.3	2.2
Crude protein (% of dried peel)	4.9	4.7
Concentration of pectin in solvent (mg/l)	518	342
Pectin yield (% of dried albedo)	0.8	0.5
Pectin yield (% of dried peel)	1.6	1.1

Experiments were carried out for the same duration of 15 min, the same sample to solvent ratio of 1:12.5 and the same extraction temperature of 150 °C on albedo. Both samples gave a dark colour and a bitter aroma. The dry weight basis was determined by drying the samples at 100 °C for 48 h.

3.4. Comparison of pectin yield between microwave extraction and Soxhlet extraction

The same ratio of sample to solvent (water) was chosen in the experiment as before. Detailed conditions and results for albedo extraction using microwaves and by the Soxhlet apparatus are listed in Table 4. Microwave extraction gave a higher rate amount of extraction and a higher amount of total nitrogen by a factor of four than the Soxhlet method. The long duration of extraction in the Soxhlet method may lead to volatilization of nitrogen from the albedo sample, and the higher temperature in the microwave oven may contribute to more nitrogen dissolving in the water. However, with a longer extraction duration, twice as much pectin in total was obtained in the Soxhlet extraction as in the microwave one, suggesting that microwave extraction also has some limitations compared with the Soxhlet method.

Table 4
Comparison of the pectin yields from microwave and Soxhlet extractions

	Microwave extraction	Soxhlet extraction
Sample to solvent ratio	8.1736 g:100 ml H_2O	16.0175 g:200 ml H_2O
pH	2.00 ± 0.05	4.50 ± 0.05
Duration	15 min	6 h (9 cycles)
Temperature (°C)	150	100
Colour	Dark	Light
Aroma	Bitter	Sweet
Total nitrogen (% of dried albedo)	0.4	0.1
Crude protein (% of dried albedo)	2.3	0.8
Crude protein (% of dried peel)	4.9	1.7
Concentration of pectin in solvent (mg/l)	518	1128
Pectin yield (% of dried albedo)	0.8	1.7
Pectin yield (% of dried peel)	1.6	3.6

The dry weight basis was determined by drying the samples at 100 °C for 48 h.

Table 5
Comparison of the pectin yields from albedo and flavedo extractions

	Albedo extraction	Flavedo extraction
Concentration of pectin in solvent (mg/l)	518	213
Pectin yield (% of dried peel)	1.6	0.6

Experiments were carried out at the same sample to solvent ratio (1:12.5), the same pH (2.00 ± 0.05) and the same temperature (150 °C) for the same duration (15 min). Both samples gave a dark colour and a bitter aroma. The dry weight basis was determined by drying the samples at 100 °C for 48 h.

3.5. Comparison of flavedo and albedo extraction

To determine the pectin content in the flavedo and albedo, microwave extractions of samples of each part were undertaken, with the same experimental conditions as before. The results are shown in Table 5, where it can be seen that pectin exists mainly in the albedo. The percentage of pectin in the albedo was two times higher than that in the flavedo. However, the flavedo part still held a significant amount of pectin, and the extract of pectin from flavedo was still 27% of the total extract (0.6/2.2). The total pectin of percentage in the dried peel was 2.2%, which is slightly lower than the result of 2.36% by Campbell and Palmer [11]. The pectin yield of 2.36% was the highest value recorded before 1997, while recent research on pectin by Fishman et al. [14,20], reported pectin extractions of 15.3% (2000) and 16.9% (2003), respectively. This higher yield was obtained by using three sequential microwave extractions rather than single extractions.

3.6. Comparison of hand-pressure extraction, microwave extraction and combined hand-pressure extraction and microwave extraction

Detailed conditions and results for the three processes of hand-pressure extraction, microwave extraction and a combined method with hand-pressure extraction and microwave extraction are given in Table 6. From the pectin testing of the flavedo extracts, combined hand-pressure and microwave extraction led to higher relative amounts of extraction. The pectin content was around two times more than that of the albedo for hand-pressure and microwave extraction. Hand-pressure and microwave extraction led to the highest amounts of pectin among the three extraction methods, and this combined technique was 12% better than hand-pressure alone. This finding suggests that microwaves help to break down the cell structure, improving the subsequent extraction of pectin, confirming the findings and explanation of Fishman et al. [14] and Kratchanova et al. [9].

Table 6
Effect of extraction methods on the pectin yields

	Hand-pressure extraction	Microwave extraction	Hand-pressure + microwave extraction
Sample to solvent ratio	4.0680 g:30 ml H_2O	4.0319 g:30 ml H_2O	4.0671 g:30 ml H_2O
Duration (min)	2	10	12
Temperature (°C)	Room temperature	80	80
Colour	Light	Dark	Medium
Aroma	Strong	Mild	Medium
Concentration of pectin (mg/l)	394	191	444
Pectin yield (% of dried peel)	0.7	0.3	0.8

The dry weight basis was determined by drying the samples at 100 °C for 48 h.

4. Conclusions

A ratio of sample to solvent of 1:12.5 extracted the most pectin and total nitrogen from the albedo. The amount of extracted pectin reduced as the pH was increased, while the extractability of crude protein was similar. These results are similar to those of previous workers. With a longer extraction duration than the microwave extraction by a factor of 240, although twice as much pectin was obtained by the Soxhlet method, the microwave method had a much better productivity, by a factor of 120. According to the results from microwave extractions of flavedo and albedo, it was shown that pectin existed mainly in the albedo, but the extract from flavedo was still 27% of the total extract. The total pectin yield in the dried peel was 2.2%, which is slightly lower than the result of 2.36% by Campbell and Palmer [11]. Combined hand-pressure and microwave treatment extracted 12% more pectin from the flavedo than using hand-pressure alone, among the three methods of hand-pressure extraction, microwave extraction and combined hand-pressure and microwave extraction.

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