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Extraction and characterisation of pomace pectin from gold kiwifruit (*Actinidia chinensis*)

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ABSTRACT

Gold kiwifruit pomace extracted using citric acid, water and enzyme (Celluclast 1.5L) were studied in terms of pectin yield, protein, ash, non-starch polysaccharide, galacturonic acid (GalA), neutral sugar composition, molar mass (M_w), viscosity and degree of branching. Water-extracted pectin was considered closest to its native form. Enzyme extracted pectin showed the highest yield (~4.5% w/w) as compared with the acid and water extraction methods (~3.6–3.8% w/w). Pectin obtained from different extraction methods showed different degree of branching. The M_w and root mean square (RMS) radius varied with the extraction methods with values of 8.4×10^5 g/mol and 92 nm, 8.5×10^5 g/mol and 102 nm, 6.7×10^5 g/mol and 52 nm for acid, water and enzymatic extraction methods, respectively. Similar trend was observed for pectin viscosity, with water-extracted pectin giving a slightly higher viscosity followed by acid and enzyme-extracted pectin. This study showed that gold kiwifruit pomace pectin has potential application in food products.

1. Introduction

Pectin is a structural polysaccharide found in the cell walls of growing plants. Pectin is regarded as a complex heteropolysaccharide consisting mainly of α-(1,4)-linked D-galacturonic acid backbone with different degrees of esterification (Mohnen, 2008). Pectin can be further classified based on their occurrences as three main structures namely, homogalacturonan (HG), rhamnogalacturonan type I (RG-I) and substituted galacturonan (Ridley, O'Neill, & Mohnen, 2000). The HG group has a backbone with a linear chain of α-(1,4)-linked D-galacturonosyl acid. The RG-I group has a backbone of α-(1,2)-linked L-rhamnosyl and α-(1,4)-linked D-galacturonosyl acid residues, with neutral sugars such as arabinose and galactose as the side chains. The substituted galacturonan group consists of α-(1,4)-linked D-galacturonosyl acid residues as the backbone such as rhamnogalacturonan II and xylogalacturonan (O'Neill, Albersheim, & Darvil, 1990).

Pectin is widely used as a thickener and stabiliser in food products such as jams and yoghurt drinks. Apple pomace and citrus peels are currently the two main sources of commercial pectin (May, 1990). They are usually obtained from the by-products of fruit juice manufacture. Pectin polymers from other plant sources have been isolated and studied. They include sugar beet pulp (Wang & Chang, 1994), peach (Pagan, Ibarz, Llorca, Pagan, & Barbosa-Canovas, 2001), mango (Ketsa, Chidtragool, Klein, & Lurie, 1999), banana peel (Emaga, Ronkart, Robert, Wathelet, & Paquot, 2008) and chicory roots (Panouille, Thibault, & Bonnin, 2006). The extraction and isolation of pectin can be carried out using different methods including chemical, physical or enzymatic treatments (Panouille et al., 2006). Chemical extraction method using dilute acid solution is commonly employed in pectin manufacture. However, the use of chemical method to extract pectin could give a negative connotation as consumers increasingly prefer chemical-free ingredients to be used in food products. Instead, the use of specific enzymes could be advantageous due to the low enzyme concentrations required and the low amount of waste generated (Yuliarti, Matia-Merino, Goh, Mawson, & Brennan, 2012).

New Zealand kiwifruit growers produced approximately 400,000 tonnes of kiwifruit from 13,000 hectares split between green (80%) and gold (20%) kiwifruit (MPI, 2013). Kiwifruit is either consumed as a fruit or processed into kiwifruit juice. In the processing of kiwifruit juice, a large proportion of the fruit goes into waste which includes the fruit peels, pomace and seeds. Kiwifruit

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waste has been reported by Wabnitz (2008) to be approximately 18% of the total kiwifruit crop. Gold kiwifruit pomace obtained after juice extraction constitutes about 40–50% of the weight of the fresh fruit (data not published). Despite the smaller percentage of gold kiwifruit crop based on the current data, the demand for gold kiwifruit is envisaged to grow as consumer demand increases.

The physico-chemical properties of pectin from the whole gold kiwifruit have recently been reported (Yuliarti et al., 2015). However, the physico-chemical properties of pectin solely from the pomace of gold kiwifruit have not been well-understood. The chemical composition of pomace pectin and its functional properties could differ from pectin extracted from whole kiwifruit. It is useful for growers and manufacturers to understand the potential functional benefits of pomace where the whole kiwifruit is utilised for juice processing. The data from this study could provide useful knowledge to maximise the utilisation of gold kiwifruit that will not only value-add but also reduce kiwifruit waste generated. It was therefore deemed necessary to characterise and compare the properties of pomace pectin with the pectin obtained from the whole kiwifruit. Knowledge of the physical functionality of pomace pectin could provide useful information for manufacturers to consider utilising the kiwifruit pomace as another source of pectin that is capable of providing certain unique functional properties. The objectives of this study were to examine the yield, composition, molar mass and the rheological properties of gold kiwifruit pomace pectin extracted using different extraction methods (acid, water and enzyme). Comparisons were made with pectin isolated from the whole kiwifruit (main-harvested fruit; MHF), as previously reported by Yuliarti et al. (2015).

2. Materials and methods

2.1. Materials

The main-harvested gold kiwifruit (MHF) obtained from Zespri International Ltd (Hastings, New Zealand) was the raw material used in this study. The fruit was harvested approximately 20 weeks after pollination, with a firmness of ~32 Newton (N). The pomace of MHF used in this study was obtained by squashing whole kiwifruit in a fruit juicer (Avanti, Model 2000 Juicer, FED Australia and New Zealand, ~0.5 mm mesh). As the juice contained a mixture of very coarse fruit pulp fractions, skin and seeds, centrifugation (3300*g*, 20 min at 4 °C) was carried out to isolate the insoluble fraction from the juice. The fruit pulp from the juicer was hydraulically pressed to separate the remaining juice. The pellet, which was considered to be part of the pomace, was pooled and mixed with the fruit pulp that was retained in the juicer. The skin and seeds of the fruit were included in the pomace preparation. A flow diagram for the preparation of the pomace is shown in Fig. 1.

2.2. Pectin extraction

The pomace recovered from MHF was subjected to three different extraction methods using acid, water or enzyme respectively.

2.2.1. Acid extraction

Gold kiwifruit pomace (200 g) was mixed with 600 mL of 1.0% (w/v) citric acid (CA) solution of pH 2.20 ± 0.01 (1:3 w/v pomace to acid solution ratio) in a container and the mixture (pH 3.1 ± 0.05) was heated in a 50 °C water bath for 60 min under continuous stirring. The container was covered with aluminium foil to reduce moisture loss due to evaporation. Purified water obtained by reverse osmosis (RO water) was used throughout the study. After extraction, the mixture was cooled to approximately 20 °C in a bath of crushed ice and then centrifuged at 3300*g* for 20 min at 4 °C. The supernatant was filtered through four layers of cheese cloths to remove the pulp and seeds that were not separated during centrifugation. Warm water at 50 °C was added to the pellet, in a 1:1 (w/v) ratio, and the mixture was stirred for 30 min (to solubilise the remaining soluble polysaccharides) and centrifuged again as described above. All supernatants and filtrates were combined and precipitated with ethanol as commonly carried out in pectin production (May, 1990).

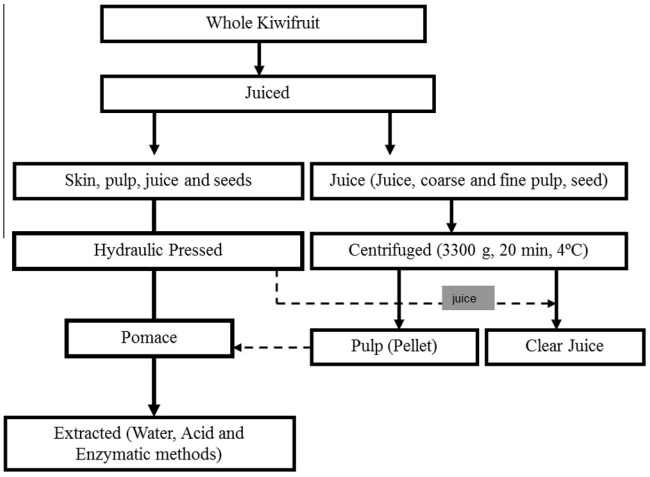

Fig. 1. Schematic flow chart of the preparation of gold kiwifruit pomace.

The polysaccharide in the supernatant was precipitated by ethanol to give a final concentration of 80% (v/v). This concentration was chosen because low molecular weight saccharides (e.g. monosaccharides and oligosaccharides) would remain soluble but polysaccharides would be precipitated out (Englyst, Quigley, Hudson, & Cummings, 1992). The mixture of ethanol and extract was stirred (10 min, ±25 °C) to obtain a uniform mixture and was then kept at 4 °C for 4 h to allow the polysaccharides to precipitate.

To separate the polysaccharide precipitate from the solvent, the mixture was centrifuged (3300*g*, 10 min, 4 °C). The pellet was washed twice with 95% ethanol (1:1, w/v) and then centrifuged again as before. The pellet was vacuum dried (Eyela, Vacuum Oven, Voc-300 SD, Science Technique Ltd, New Zealand) at 58 ± 3 °C, 65 cm Hg, for approximately 7–10 h until a constant weight was achieved.

For further purification of pectin, the vacuum-dried sample was dispersed in RO water (1.0% w/w) and stirred overnight (~15 h) in a 4 °C chiller. The dispersion was then centrifuged at 30,000*g* for 60 min (4 °C) to separate out the remaining insoluble particles. The soluble pectin in the supernatant was recovered by ethanol precipitation (80%), centrifuged, ethanol washed and vacuum oven dried as described earlier. The pectin yield was calculated using the equation below (Ptichkina, Markina, & Rumyantseva, 2008) and was denoted as the *acid extracted pomace pectin*:

$$D = 100\left(\frac{m_{\text{pectin}}}{m_{\text{kp}}}\right)$$

where D is the percentage yield of purified pectin (%), m_{pectin} is the mass of recovered pectin (g) and m_{kp} is the mass of kiwifruit pomace (g) used in the extraction.

2.2.2. Water extraction

The water extraction method was similar to the acid extraction method except that RO water was used instead of the citric acid solution. The extraction was carried out in a 25 °C water bath for 30 min with a pomace to water ratio of 1:3 (w/v) (*i.e.* 200 g of pomace mixed with 600 mL of RO water). The pH value of the mixture was 3.70 ± 0.05. Recovery of the soluble fraction from the