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Characterization of passion fruit seed fibres—a potential fibre source

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Abstract

Raw passion fruit (*Passiflora edulis*) seed is rich in crude lipid (24.5 g/100 g) and insoluble dietary fibre (64.1 g/100 g). After defatting, the insoluble fibre-rich fractions (FRFs), including insoluble dietary fibre, alcohol-insoluble solids and water-insoluble solids (84.9–93.3 g/100 g) became the predominant component in the (defatted) seed, and were mainly composed of cellulose, pectic substances and hemicellulose. These insoluble FRFs had water- and oil-holding capacities comparable with those of cellulose, while their bulk densities and cation-exchange capacities were significantly (P < 0.05) higher than those of cellulose. All FRFs exhibited significant (P < 0.05) effects in absorbing glucose and retarding amylase activity, and might help control postprandial serum glucose. These results underline the value of consumption of these FRFs as fibre sources or low calorie bulk ingredients in food applications. Further investigations on the in-vivo hypoglycemic effect and other physiological properties of these FRFs, using animal feeding experiments, are underway.

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1. Introduction

Sufficient consumption of dietary fibre could reduce the risk of civilization diseases, such as cardiovascular disease, colon cancer and obesity (Marlett, 2001; Slavin, 2001). Some dietary fibres have been reported to decrease the digestion and absorption of carbohydrate and postprandial serum glucose levels (Flourie, 1992; Ou, Kwok, Li, & Fu, 2001). Accordingly, dietary fibrerich products and diets have gained popularity as food ingredients for health benefits, and have encouraged food scientists to search for new fibre sources and to develop high-fibre products.

Both the composition and physicochemical properties of dietary fibres might explain their functionalities in foods. All this information could be further extended to the understanding of the physiological effects of dietary fibres (Gordon, 1989). Thus, a study of the chemical and physical properties of dietary fibre is important for exploiting the fibre as an ingredient in low-calorie foods.

Passion fruit (Passiflora edulis), which is native to Brazil, is a popular tropical fruit throughout the world. The soft, orange pulp of this fruit is full of tiny albuminous seeds (up to 25% of the fresh pulp by weight), and all of these are edible (Davidson, 1999). The passion fruit is usually used for juice production in Taiwan and elsewhere in Asia, and works best as a flavouring in many delicacies. In the juice industry, the passion fruit produces many thousand tons of seeds as agricultural byproducts during juice extraction. These seeds, containing large amounts of fibre and oil, are generally discarded after being crushed. In recent years, many studies have aimed to investigate dietary fibres from the byproducts and pomace of apple, citrus fruits, grape skin and seed, guava, mango and pineapple with a view to explore their potential applications and physiological activities (Baker, 1997; Gourgue, Champ, Lozano, & Delort-Laval, 1992; Igartuburu, Pando, Rodriguez-Luis, & Gil-Serrano, 1998; Jiménez-Escrig, Rincón, Pulido, & Saura-Calixto, 2001; Larrauri, Ruperez, Bravo, & Saura-Calixto, 1996; Larrauri, Ruperez, & Calixto, 1997; Leontowicz et al., 2001; Valiente, Arrigoni, Esteban, & Amado, 1995). However, information about the composition and functionality of dietary fibres prepared from the passion fruit pomace is scarce.

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The objective of this study was to evaluate and compare the composition, physicochemical properties and in-vitro hypoglycemic effect of different fibre-rich fractions (FRFs) prepared from the seeds of passion fruits (hybrid, Tai-Nong-1) indigenous to Taiwan. This study might allow an improved use of this resource, which is currently underexploited. Since the way of processing this agricultural byproduct might determine the functionality of food fibres, differences among the dietary fibres, alcohol-insoluble solids and water-insoluble solids, as well as the potential applications of these FRFs as fibre sources, are considered in this study.

2. Materials and methods

2.1. Seed samples

After juice extraction, the seed samples of passion fruit (hybrid, Tai-Nong-1) were collected from the farmers' association of Chiayi county in Taiwan. The seed sample was cleaned and finely ground to 0.5 mm size for analyses. The seed powder was defatted with petroleum ether (1:4 w/v), four times, and the content of residual lipid in the defatted sample was below 0.01 g/100 g. The defatted sample was kept in a desiccator until used.

2.2. Chemical analyses

Protein was calculated by multiplying the nitrogen content with a factor of 6.25. Total ash was determined by the method 4.1.10 according to AOAC (1995). Crude lipid was determined by using petroleum ether with a Soxhlet apparatus. Moisture was estimated by drying the sample to a constant weight at 105 °C.

2.3. Separation of dietary fibre (DF)

The separations and determination of total, insoluble and soluble DF in the defatted seed sample were done according to the method 991.43 (AOAC, 1995) using the fibre assay kit (Megazyme K-TDFR, Wicklow, Ireland). The DF contents were corrected for residual protein, ash, and blank.

2.4. Separation of alcohol-insoluble solids (AIS)

According to the method of Thomas, Crepeau, Rumpunen, and Thibault (2000) with slight modifications, defatted seed sample was homogenized in 85% (v/v) boiling alcohol (seed:alcohol 1:30 w/v), using the Osterizer (Sunbeam-Oster, IL, USA) at the "Hi" speed for 1 min. The suspension was further boiled for another 40 min and the sample was extracted twice. AIS was filtered, washed with 70% ethanol, and dried by solvent exchange and air at 30 °C.

2.5. Separation of water-insoluble solids (WIS)

Based on the method of Massiot and Renard (1997) with slight modifications, WIS was prepared by homogenizing the defatted seed sample in cold distilled water (seed:water 1:10 w/v), using the Osterizer (Sunbeam-Oster, IL, USA) at the "Hi" speed for 1 min. After filtration, the WIS collected was washed with cold distilled water, and dried by solvent exchange and air at $30 \,^{\circ}$ C.

2.6. Starch measurement

Following the dietary fibre separation, as described in Section 2.3, the amount of digestible starch was calculated from the difference between the glucose contents in the mixture with and without (control) enzymatic digestion.

2.7. Analysis of sugar composition

According to the methods of Englyst, Quigley, and Hudson (1994), the neutral sugars of the FRFs were determined using allose as an internal standard. The FRFs were hydrolyzed by 12 M H₂SO₄ at 35 °C for 60 min and then boiled in 2 M H₂SO₄ for a further 60 min. The residual insoluble matter in the hydrolysate was quantified as Klason lignin, gravimetrically. The released monosaccharides in the hydrolysate were quantified as alditol acetates by gas chromatography (Hitachi G-5000, Tokyo, Japan, fitted with a flame ionization detector). GC operating conditions were as follows: column: Quadrex 007-225 (15 m×0.53 mm i.d.); oven temperature: programmed from 100 °C isothermal for 3 min, to 220 °C at a rate of 4 °C/min; injector temperature: 270 °C; detector temperature: 270 °C; carrier gas: nitrogen; flow rates: 2.1 ml/min (nitrogen), 500 ml/min (air).

The cellulose content was calculated from the difference between the amounts of total glucose and non-cellulosic glucose which was determined by boiling the FRFs with 2 M H₂SO₄ only. Uronic acid was determined colorimetrically (45.4.11; AOAC, 1995) using Dgalacturonic acid monohydrate as reference. The pectin content was estimated by the amount of uronic acid in terms of polysaccharide residues.

2.8. Physicochemical properties

According to the methods of Chau, Cheung, and Wong (1997) with slight modifications, the oil-holding capacity (g/g) and water-holding capacity (ml/g) were measured by mixing the insoluble FRFs with vegetable oil (1:5 w/v) for 30 min and with distilled water (1:10 w/v) for 24 h, respectively. The density of vegetable oil was 0.88 g/ml. Moreover, the bulk density (g/ml) and cation-exchange capacity (meq/kg) of the insoluble FRFs were determined by the methods of Chau (1998).

2.9. Determination of glucose-adsorption capacity and amylase inhibitory effect

According to the method of Ou et al. (2001) with slight modifications, the glucose-adsorption capacity (mmol glucose/g fibre) of the FRF was estimated by stirring 1 g of FRF in 100 ml of glucose solution (100 mmol/l), followed by determining the final glucose content in the mixture. The amylase inhibitory effect (%) of the FRF was determined by the method of Ou et al. (2001) with slight modifications. FRFs (1 g) and 4 mg of alpha amylase (Cat. No. 100447, ICN, USA) were mixed in 40 ml potato starch solution (4% w/v) for 30 min, followed by measuring the final glucose content in the mixture. The amylase inhibitory effect (%) was defined as the percent decrease in the rate of glucose production (μ mol/h) over the control (without fibre).

2.10. Statistical analysis

Data collected from this study were analyzed by oneway analysis of variance and the Duncan test (Ott, 1988). Evaluations were based on the P < 0.05 significance level.

3. Results and discussion

During the passion fruit juice extraction process, seed was separated from pulp by centrifugation, yielding a large quantity of seed. In this study, the contents of seed and pulp in the fresh passion fruit were about 11.1 ± 0.35 and 88.9 ± 0.35 g/100 g, respectively.

The proximate composition of the raw and defatted passion fruit seeds is shown in Table 1. The raw seed

was rich in total dietary fibre (TDF) (64.8 g/100 g) and crude lipid (24.5 g/100 g), and possessed small amounts of crude protein (8.25 g/100 g), ash (1.34 g/100 g) and carbohydrate (1.11 g/100 g). Moreover, the seed was almost free of digestible starch (< 0.01 g/100 g). After lipid removal, the TDF content was greatly increased up to 85.9 g/100 g defatted seed. In the agricultural byproducts of apple, citrus fruits, corn, oat, wheat and some other fruits and greens, TDF contents ranged from 10.2 to 87.9 g/100 g (Gorinstein et al., 2001; Grigelmo-Miguel & Martin-Belloso, 1999a, 1999b; Thomas et al., 2000). Insoluble dietary fibre (IDF) (84.9 g/100 g defatted seed) was the predominant fibre fraction (98.8% of TDF). In the pomace and agricultural byproducts of many other fruits and vegetables, insoluble fibres were also reported to be the major fibre fraction (Gorinstein et al., 2001; Grigelmo-Miguel et al., 1999a, 1999b; Thomas et al., 2000). Since the consumption of insoluble fibres is beneficial to intestinal peristalsis by increasing fecal bulk and decreasing transit time (Schneeman, 1987), the defatted passion fruit seed could be a good source of insoluble fibre with desirable physiological effects.

Table 2 presents the contents of AIS and WIS in the raw and defatted passion fruit seed. Both of these FRFs were determined on a weight basis and were not corrected for protein and ash. For the raw seed, the contents of AIS and WIS were 70.5 and 67.2 g/100 g, respectively. The slightly higher level of AIS relative to WIS was due to the alcohol precipitating substances such as protein and some inorganic substances (Ting, 1970). In some other fruit byproducts, such as mango, quince and citrus wastes, AIS contents were reported to be 30.7–49.7, 27.8–37.5 and 45.0–75.0 g/100 g, respectively (Gourgue et al., 1992; Thomas et al., 2000; Ting, 1970). After lipid removal, the amounts of AIS and WIS in the defatted seed were largely increased to 89.0 and 93.3 g/100 g, respectively, hence the contents of inso-

Table	1

Chemical	composition	of the raw	and defatted	passion	fruit seed

Composition	g/100 g raw seed, dry weight	g/100 g defatted seed, dry weight		
Moisture ^a	6.60 ± 0.28	_		
Crude protein ^b	8.25 ± 0.58	10.8 ± 0.75		
Crude lipid ^a	24.5 ± 1.58	-		
Total dietary fibre (TDF) ^{b,c}	64.8 ± 0.05	85.9 ± 0.07		
Insoluble dietary fibre (IDF) ^{b,c}	64.1 ± 0.02	84.9 ± 0.03		
Soluble dietary fibre (SDF) ^{b,c}	0.73 ± 0.07	0.97 ± 0.09		
Ash ^a	1.34 ± 0.08	1.77 ± 0.11		
Carbohydrate ^d	1.11	1.53		

^a Means±S.D. of triplicates.

^b Means±S.D. of duplicates.

^c The fibre contents have been corrected for protein and ash. The IDF and SDF contents in the defatted seed without correction were 93.8 ± 0.26 and 1.64 ± 0.18 g/100 g defatted seed, respectively.

^d The carbohydrate was defined as the residue, excluding protein, lipid, TDF, and ash, and was calculated by difference (=100-protein-lipid-TDF-ash). The digestible starch content was <0.01 g/100 g seed.

luble FRFs were high enough to be a potential fibre source.

The total monomeric sugar contents released from the IDF, AIS and WIS of the defatted seed were 43.1, 42.5 and 39.7% by weight, respectively (Table 3). The total sugar contents, among these FRFs, were comparable to each other. In addition to sugars, dietary fibres might also contain protein, ash, polyphenols and lignin, to different extents (Thomas et al., 2000). Table 3 shows that the amounts of impurities (protein and ash) in the insoluble FRFs were about 9.43-16.2 g/100 g FRF, while the other non-sugar components (e.g. Klason lignin) ranged from 41.3 to 47.5 g/100 g FRF. Among the sugars of the insoluble FRFs, the predominant sugars were glucose (50.1–53.7%) and xylose (17.7–22.4%), followed by arabinose (10.7–14.8%), uronic acid (7.05– 9.07%), rhamnose (1.48-4.94%) and galactose (2.67-3.27%). The high proportion of cellulosic glucose (43.1– 52.4%) revealed that cellulose was the major polysaccharide in the insoluble FRFs. The sugars, including arabinose, rhamnose and uronic acid, constituted up to 21.2–25.8% of the total sugars, suggesting the presence of arabinose-rich pectic substances. The xylose-containing polymer in the insoluble FRFs could possibly be a xylose-rich hemicellulose, comprising xylose, arabinose, galactose, glucose and mnannose (Igartuburu et al., 1998; Schneeman, 1986). A hemicellulose rich in xylose was reported to he the major polysaccharide in grape seed DF (Igartuburu et al., 1998). Table 3 shows a great similarity in the polysaccharide profiles among the three insoluble FRFs. In the SDF (Table 3), arabinose, rhamnose and uronic acids accounted for 53.0% of the total sugar content, implying that the arabinose-rich pectic substances were the major polysaccharides in the SDF.

Table 4 shows the bulk densities, water- and oilholding capacities (WHCs and OHCs) and cationexchange capacities among the three insoluble FRFs, compared with cellulose, the most-used food fibre. It was found that the bulk densities of these FRFs were comparable to each other and significantly (P < 0.05) higher than that of cellulose. The WHCs among these FRFs (2.37–3.20 ml/g) were comparable but lower than the WHC of cellulose (3.81 ml/g). The comparable WHCs could be explained by the similarities in the FRFs compositions (Table 3), number and nature of the water-binding sites, and structure (Gordon, 1989; Robertson & Eastwood, 1981). As compared to cellulose, the lower WHCs of the FRFs might be partly attributed to their higher densities as well as the fewer water-binding components, due to the presence of impurities and lignin (Chau & Cheung, 1999; Dufour-Lescoat, Le Coz, Andrieux, & Szylit, 1995). Table 4 reveals that the FRFs and cellulose have comparable OHCs (2.07-3.72 g/g), which were relatively greater than those of some orange byproduct fibres (0.9-1.3 g)g) (Grigelmo-Miguel & Martin-Belloso, 1999a). In general, the physicochemical properties, such as bulk density, OHC and WHC among different fibres were correlated with their particular chemical and physical structures as well as the preparation methods (Thibault and Ralet, 2001). In Table 4, the cation-exchange capa-

Table 2

Table 3

The contents of AIS and WIS prepared from the raw and defatted passion fruit s	eed
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Fibre-rich fractions ^a	g/100 g raw seed, dry weight	g/100 g defatted seed, dry weight		
Alcohol-insoluble solids (AIS) ^{b,c}	70.5±0.18	93.3±0.24		
Water-insoluble solids (WIS) ^{b,c}	67.2 ± 0.16	89.0 ± 0.22		

^a The fibre-rich fractions were determined on a weight basis and were not corrected for protein and ash.

 $^{\rm b}$ Means \pm S.D. of triplicates.

^c The digestible starch contents, in both the AIS and WIS, were <0.01 g/100 g FRF.

Monosaccharide composition ^a of the fibre-rich fractions ^b prepared from the defatted passion fruit seed											
Fibre-rich fractions	Protein	Ash	Rhamnose	Fucose	Arabinose	Xylose	Mannose	Galactose	NC-Glc ^c	C-Glc ^d	Uronic acid
IDF	9.00x	0.43x	1.48x	tr ^e	4.63x	9.67x	0.15x	1.15x	0.40x	22.6x	3.04x
AIS	14.6y	1.60y	0.63x	tr	6.29x	7.52y	0.24x	1.39x	0.55x	22.3x	3.61x
WIS	14.0y	0.63z	1.96x	tr	4.68x	8.30xy	0.10x	1.17x	2.80xy	17.1y	3.60x
SDF	41.2z	-	1.13x	tr	5.55x	1.75z	3.76y	1.03x	4.15y	-	5.39y

Values in the same column with different letters (x-z) are significantly different (Duncan, P < 0.05).

^a Expressed as g/100 g FRF.

^b The fibre-rich fractions were determined on a weight basis and were not corrected for protein and ash. The non-sugar components including Klason lignin in IDF, AIS and WIS were 47.5, 41.3 and 45.7 g/100 g FRF, respectively.

^c NC-Glc: non-cellulosic glucose.

^d C-Glc: cellulosic glucose.

 $^{\rm e}\,$ tr, trace amount ($<\!0.01$).

Table 4	
Physicochemical properties of the insoluble fibre-rich fractions ^a	prepared from the defatted passion fruit seed relative to cellulose

Fibre samples	Bulk density (g/ml)	Water-holding capacity (ml/g)	Oil-holding capacity (g/g)	Cation-exchange capacity (meq/kg)
Cellulose ^b	0.38x	3.81x	2.76xy	22.7x
IDF	0.68y	2.37y	3.72x	50.3y
AIS	0.71y	3.20z	3.52x	35.3z
WIS	0.68y	2.77yz	2.07у	42.5yz

Values in the same column with different letters (x–z) are significantly different (Duncan, P < 0.05).

^a The fibre-rich fractions were determined on a weight basis and were not corrected for protein and ash.

^b Alphacel-Nonnutritive fibre, ICN Nutritional Biochemicals, Cleveland, USA.

Table 5

Glucose-adsorption capacity and amylase inhibitory effect (%) of the insoluble fibre-rich fractions^a prepared from the defatted passion fruit seed relative to cellulose

Fibre samples	Glucose-adsorption capacity ^b (mmol glucose/g fibre)	Amylase inhibitory effect ^c (%)		
Cellulose ^d	8.74w	7.29w		
IDF	9.62x	27.9x		
AIS	10.4y	21.8y		
WIS	10.2z	26.7z		

Values in the same column with different letters (w–z) are significantly different (Duncan, P < 0.05).

^a The fibre-rich fractions were determined on a weight basis and were not corrected for protein and ash.

^b The concentration of the glucose solution used in the experiment was 100 mnmol/l.

^c The amylase inhibitory effect (%) was defined as the percent decrease in the glucose production rate (μ mol/h) over the control (without fibre; 121 μ mol/h).

^d Alphacel-Nonnutritive fibre, ICN Nutritional Biochemicals, Cleveland, USA.

cities of the FRFs (35.3–50.3 meq/kg) were significantly (P < 0.05) greater than that of the cellulose (22.7 meq/kg). As the cation-exchange capacity is related to the uronic acid content of a fibre (Gordon, 1989; Thibault et at., 2001), the stronger ion binding capacity of the FRFs relative to cellulose might be attributed to the presence of uronic acids (3.04–3.61 g/100 g FRF) (Table 3). As described by Furda (1990), fibres of higher cation-exchange capacity could entrap, destabilize and disintegrate the lipid emulsion, consequently decreasing the diffusion and absorption of lipids and cholesterol in the small intestine.

The glucose-adsorption capacity and amylase inhibitory effect (%) of the insoluble FRFs relative to cellulose are shown in Table 5. The glucose-adsorption capacities of the FRFs (9.62-10.4 mmol glucose/g fibre) were significantly (P < 0.05) higher than that of cellulose (8.74) mmol glucose/g fibre). The stronger ability of the FRFs to bind glucose might be beneficial to lower the concentration of available glucose in the small intestine (Ou et al., 2001). The insoluble FRFs were also found to exhibit a significantly (P < 0.05) stronger effect in reducing the α amylase activity (21.8–27.9%) than cellulose (7.29%). This in-vitro study indicated that the FRFs could retard the glucose production rate (µmol/h) more efficiently than cellulose. Findings from Gourgue et al. (1992) and Ou et al. (2001) demonstrated that dietary fibres could retard α -amylase activity by capsulating starch and

enzyme, and even inhibiting the enzyme. All these results imply that the apparent effects, on glucose-adsorption and amylase inhibition, of the FRFs might create a concerted function in decreasing the absorption rate of glucose and the concentration of postprandial serum glucose.

4. Conclusions

This study revealed that the edible passion fruit seed was rich in insoluble FRFs (IDF, AIS and WIS) which were mainly composed of cellulose, pectic substances and hemicellulose. These FRFs had water- and oilholding capacities comparable with those of cellulose, while their bulk densities and cation-exchange capacities were significantly (P < 0.05) higher than those of cellulose. All insoluble FRFs showed significant (P < 0.05) effects in absorbing glucose and retarding amylase activity, it is speculated that these FRFs might have potential benefit for controlling postprandial serum glucose, and potential applications as low calorie bulk ingredients for fibre enrichment and dietetic snacks. Since this fibre-rich passion fruit seed is available in large quantity as a byproduct of juice production, it could be exploited as a good source of food fibre. Further investigations of the physiological functions of these insoluble FRFs, using animal feeding experiments, are underway.

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