

Extraction and characterization of pectin from grapefruit peels

Abstract

The present study was focused on the potential of red and white grapefruit peel as a source of pectin. Grapefruit peels were treated with 95% alcohol and the obtained alcohol-insoluble solids (AIS) were subjected to a sequential extraction with hot distilled water and hot 0.5% HCl. Comparative investigations were carried out with purified commercial citrus pectin. Chemical and physicochemical characterization of all pectin was accomplished. The yield of grapefruit peel pectin was 25% (dwb) It was found that pectin of the two types of grapefruit peels were similar in anhydrouronic acid content (AUA 60.95%). The grapefruit peel pectin for the red and white differed in their degree of esterification (DM 51.24-55.01%) and 50.848% for corresponding citrus pectin. Both grapefruit peel pectin were higher in their methoxyl content 8.875% for the red and 7.542% for the white, while corresponding citrus pectin was 6.798%. The acetyl content for the white grapefruit peel pectin was 1.634% and for the red was 0.55%, lower in comparison with the corresponding citrus pectin of 0.624. The grapefruit peel pectin contained L-arabinose, D-galactose, L-rhamnose and D-xylose. Gel-forming property of grapefruit peel pectin was of good quality similar to commercial pectin.

Keywords: grapefruit peel pectin, commercial citrus pectin, neutral sugars composition, degree of esterification, methoxyl content, acetylation, gel-forming property

Volume 2 Issue 1 - 2016

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Received: October 25, 2015 | **Published:** February 19, 2016

Introduction

Pectic substances in fruits were discovered by the French chemist Louis Nicolas Vauquelin in tamarind fruit. The term "pectin" was introduced by Henri Braconnot because of the gelling properties of these substances.^{1,2} Pectins are structural polysaccharides present within all dicotyledonous plant cell walls. The primary structural feature of pectin is a linear 1, 4 α linked D-Galacturonic acid chain with varying degrees of methylation, where they are responsible for different physiological processes.³ In the cell walls they serve as one of the main agents cementing the cellulose fibrils and may be linked covalently to other polymers. Intracellular pectins provide the channels for passage of nutrients and water.⁴ The main raw materials used to produce commercial pectin are apple pomace and citrus peel,⁵ sugar beet and sunflower heads.⁶ Commercial pectins are extracted at low pH and high temperature. Pectins are widely used as food additives (E440) with gelling and stabilizing properties in jams, jellies, marmalades, milks and confectionery products.⁷ Citrus is cultivated in native to tropical and subtropical regions. Nowadays, it is grown all over the world wherever there is sufficient rainfall and irrigation to sustain the trees. Total area of citrus in Sudan is estimated as 100000Fed (National Horticulture Administration, 2001) and considered as an important cash crop in the Sudan. In addition, it is one of the major sources of human diet due to its high nutritive value, especially vitamin C.⁸ However, the productivity of this area may not satisfy the ever-increasing demand for citrus products for local consumption and export. Therefore, the national strategy of citrus expansion is directed towards the large national schemes.⁹ The grapefruit (*Citrus paradisi*) is one of the most important citrus species in Sudan. Sudan ranked second, after South Africa, in production of

grapefruit in Africa, with a total production of 65000million metric tons (mmt) compared to 147000mmt in South Africa.¹⁰ Sudanese grapefruit is well known for its large size, excellent quality and good coloration. A productivity as high as 40tons per hectare is obtained.¹¹

The aim of this study was to investigate the physical and chemical properties of pectin extracted from grapefruit peels and compare these properties with that of commercial pectin.

Materials and methods

Materials

Mature grapefruit of two types (white and red) were purchased from local market. Commercial citrus pectin were granted from Saeed factory for food industry. All the chemicals used in the study were of analytical grade.

Physicochemical analysis

Grapefruit peels were analyzed for different attributes such as moisture, ash, protein, crude fiber titratable acidity, sugars, ascorbic acid, calcium, magnesium, alcohol insoluble solids (AIS) and total pectin. Dry matter was measured by calculating the weight difference after drying the samples following.¹² Ash was determined by the incineration of 2g samples placed in a muffle furnace (LMF4 from Carbolite, Bamford, Sheffield UK) maintained at 550°C for 5hours. Crude protein (% total nitrogenx6.25) was determined by Kjeldahl method,¹³ using 2g samples; Crude fat was determined crude fat was obtained by exhaustively extracting 2g of each sample in a Soxhlet apparatus using petroleum ether(boiling range 40-60°C) as the

extractant.¹⁴ Crude fiber is loss on ignition of dried residue remaining after digestion of sample with 1.25% (W/V) H₂SO₄ and 1.25% (W/V) NaOH solutions (W/V) under specific conditions.¹⁵

Titrateable acidity

Ten grams of material were homogenized in a blender and the volume made up to 250ml with distilled water and filtered. One hundred milliliters aliquots were taken and titrated with 0.1N NaOH to pH 8.0 using phenolphthalein as indicator.¹⁶ The results were expressed as percent citric acid as follows:

$$\text{Titrateable acidity \%} = \text{No. of ml of NaOH} \times C.F$$

C.F=Conversion factor which equal to 0.07 citric acid hydrous

Reducing sugars

Were determined by the method of Lane and Eynon. Where ten grams of sample were extracted with 200ml of ethanol (70%) for 6hours in Soxhlet, then evaporated to 100ml, clarified by adding lead acetate and filtered, sodium oxalate was added to remove lead acetate by filtration. The burette was filled with sugar solution prepared above, then titrated against Fehling solution using methylene blue indicator. The titration was competed by appearance of red brick colour.¹⁷

Total sugars

Total sugars were determined according to the method of Lane and Eynon. Ten milliliters of HCl (1:1) were added to 50mls sugar solution and left for 8 hours. The solution was neutralized by NaOH (40%), the volume was completed to 100ml and titrated against Fehling solution as mentioned above. Total sugars were calculated from the table¹⁷ as shown above.

Ascorbic acid

Ascorbic acid was determined by titrating the filtrate of samples against standard solution of 2,6-dichlorophenol-indophenol to faint pink colour end point persist for 15seconds. The result expressed as mg/100 gm ascorbic acid.¹⁷

$$\text{Mg / 100 gm ascorbic acid} = \frac{\text{titration figure} \times \text{dye strength} \times 100}{\text{Factor}}$$

Calcium and magnesium

One gram of the peel is ignited in a muffle furnace at 500°C overnight. The contents were dissolved into 5ml of 20% HCl, the solutions were warmed and filtered through acid washed filter paper into 50ml volumetric flask and made to volume with distilled water.¹⁸ From one milliliter of this solution, the amount of calcium and magnesium were determined spectrophotometrically at wave length of 422.7nm and 285.2nm respectively using Atomic Absorption Spectrophotometer (3110)

Alcohol Insoluble Solids (AIS)

Twenty grams from each type of grape fruit peels were weighed into a 600ml beaker then 300mls of 95% alcohol were added, stirred, brought to boiling, simmered for 30min and then filtered through Buchner fitted with filter paper which was dried previously in a bottomed dish for 2 hours at 100°C, covered with fit cover and

weighed. The residue then washed with 80% alcohol until washings are clear and colourless. The paper was then transferred to the previous dish and dried at 100c for 2hours.¹⁹

The final weight minus first weight was recorded as weight of alcohol insoluble solids and its percentage was then calculated as follows:

$$\text{AIS\%} = \frac{\text{final weight} - \text{first weight} \times 100}{\text{Weight of sample}}$$

Crude lignin

Duplicate samples (0.5g) of alcohol insoluble solids were weighed and 6ml of 72% sulphuric acid were added and the mixture left at room temperature for 2hours. One hundred and forty four milliliters of distilled water were added and the mixture refluxed for 4hours. The insoluble residue was filtered in a coach crucible, washed with hot water till freed from acid, dried and weighed as crude lignin. Finally, the residue was ignited and the weight of lignin was obtained by difference.¹⁹

Total pectin

Duplicate samples (0.1gm each) of AIS were mixed with 300ml of 0.05 of the sodium salt of Ethylene Diamine Tetra Acetic Acid (EDTA), treated with 1N NaOH to reach pH of 11.5, allowed for 30min at room temperature and the pH was adjusted to 5.0 with 1N acetic acid. To this mixture, 0.1gm pectinase was added and stirred for about an hour, diluted to 500ml with distilled water, filtered through whatman No.1 first few filtrates were discarded. Two milliliters of the filtrate were diluted to 50ml from which two milliliters were taken for colorimetric determination in spectronic 20 the absorbance of total pectin was measured at wave length 520nm with known amount of galacturonic acid ranged from 0.5-3.5mg/2ml.²⁰

Preparation of Alcohol Insoluble Solids (AIS)

The peels were sliced into small cubes which were mixed thoroughly and immediately treated in 3 volumes of boiling 95% (v/v) ethanol for 10min and cooled to room temperature. Alcohol-insoluble solids (AIS) were continuously washed with 70% (v/v) ethanol to remove free sugars, pigments (especially phenolics and tannins), and other impurities as much as possible. The residue was then dried by solvent exchange (95% ethanol and acetone), placed in a fume hood for 5h for residual acetone evaporation and oven-dried overnight. Dried AIS was ground in a hammer mill (Model 912, Winona Attrition Mill Co, Winona, MN) to pass through a 60-mesh sieve and was kept under moisture-free conditions until use for further analysis i.e. yield of pectin, moisture, ash.²⁰

Isolation of pectin from grapefruit peel

The alcohol insoluble solids (AIS) obtained from grapefruit peel was weighed into a beaker and distilled water was added at 1:15 ratio. The mixture was stirred to dissolve the AIS powder in water. Then the pH of the mixture was measured and adjusted to pH 2 using 1.5N hydrochloric acid. Then the mixture was left for 30 minutes at room temperature then heated at 80°C for 1hour. The mixture was cooled to room temperature then centrifugated at 15000rpm for 10minutes. The precipitate was suspended in 600ml distilled water, acidified to pH 2 with conc. hydrochloric acid, heated at 80°C for 10minutes and centrifuged again. The liquid recovered was bulked with the first extract then filtered under vaccum. The clear filtrate was added to 96%

ethanol at 1:2 ratios. Then it was left to precipitate pectin for 12 hours. After precipitation of pectin in ethanol, the precipitate was filtered using cheese cloth and firstly washed with acidified 80% ethanol and left for 1 hour. Secondly, it was washed by 95% ethanol and finally washed by acetone and got the washed pectin. Then it was dried in vacuum oven at 60°C (5mm mercury), to obtain constant weight. Finally the dried pectin was ground to pass through mesh NO. 60 and kept for further analysis, i.e. moisture, ash, anhydrous acid content, Equivalent Weight, methoxyl content, degree of esterification, acetyl value, sugar composition and gelling characteristics. All these analysis were carried out for commercial pectin as well.

Analysis and characterization of pectin

Qualitative tests

- i. **Colour:** This was done by visual observation
- ii. **Solubility of dry pectin in cold and hot water:** (0.25%) of the pectin samples were separately placed in a conical flask with 10ml of 95% ethanol added followed by 50ml distilled water. The mixture was shaken vigorously to form a suspension which was then heated at 85-95°C for 15min.²¹
- iii. **Solubility of pectin solution in cold and hot alkali (NaOH):** To 1ml and 0.1N NaOH was added 5ml pectin solution and then heated at 85-90°C for 15min.²²
- iv. **pH determination:** The choice of the pH was made by preparing a buffer at pH 7.0 and the temperature adjusted to 28°C, the glass electrode standardized with standard buffer solution with the electrode rinsed with distilled water before inserting into the pectin solution and pH determined read off.

Quantitative tests

Yield: The pectin yield was calculated by using the following equation:

$$\text{Pectin (g/100g)} = \frac{\text{weight (g) of dried pectin}}{\text{Weight (g) of dried alcohol insoluble powder taken for extraction}} \times 100$$

Moisture content: 1g of sample in previously dried and tared dish was weighed and placed in the oven for 2 hours at 130°C. The dish was removed and cooled in a desiccators and then weighed.

$$\text{Moisture \%} = \frac{W_1 - W_2 \times 100}{W_1 - W}$$

Where W is weight of petri dish (g), W_1 is weight of petri dish with sample (g) W_2 weight of petri dish with dried sample (g).

Ash content: The ash content was determined by weighing 1g of pectin in a tared crucible and then heated in a muffle furnace at 600°C for four hours. The residue was cooled in a desiccator and weighed to constant weight.

Determination of ash alkalinity: The ash was dissolved in 25ml of 0.1N HCl heated gently and then titrated with 0.1N NaOH using phenolphthalein indicator. Then ash alkalinity calculated as the number of milliliters of acid required neutralizing one gram ash.

Protein content: Nitrogen content was determined by Kjeldahl method.²³ Protein content was calculated by multiplying the nitrogen content by a factor of 6.25.

Acetyl content: The acetyl content of the pectin was determined.²⁴

$$\text{Acetyl content \%} = \frac{\text{net ml of NaOH} \times \text{normality of NaOH} \times 4.3}{\text{Weight of sample (g) in the aliquot}}$$

Where

netml of NaOH = totalml of ml of NaOH required to titrate the distillate - totalml of ml of NaOH required to titrate the distillate of blank run.

Equivalent weight: Equivalent weight of the pectin was determined by titrating a known weight of pectin against standardized 0.1N NaOH solution to faint pink endpoint.²⁵

The equivalent weight was calculated as follows:

$$\text{Equivalent Weight} = \frac{\text{Weight of sample (mg)}}{\text{meq. Of sodium hydroxide}}$$

where

meq. of sodium hydroxide = normality x titre value

This titre is known as initial titre (IR) or free acid titre.

Methoxyl content: The methoxyl content was determined by saponification of extracted pectin and titrating the liberated carboxyl group against standardized 0.1N NaOH solution using phenol red as indicator to faint pink end point.²⁵

$$\text{Methoxyl content} = \frac{\text{meq. of NaOH} \times 31 \times 100}{\text{Weight of sample}}$$

The degree of esterification and anhydrous acid: content were calculated as follows:

The degree of esterification was calculated from the observed value of methoxyl content and anhydrous acid content as per following expression given by Schult.²⁶

$$\text{Degree of estrification} = \frac{176 \times \text{methoxyl content} \times 100}{31 \times \text{anhydrous acid content}}$$

$$\text{Anhydrous acid content (AUA)} = \frac{176 \times 100}{Z}$$

Where

$$Z = \frac{\text{Weight of sample}}{\text{meq. of alkali for free acid} + \text{meq. of alkali for methoxyl}}$$

Sugar composition: The acid hydrolysis of the sample was carried out then thin Layer chromatography was used for the separation of sugars in the neutralized hydrolysate.²⁷

Determination of intrinsic viscosity and molecular weight:

Determination of Intrinsic Viscosity was performed at 25°C. The studied pectin was solubilized in distilled water the filtered. The obtained filtrate was used to prepare solutions with concentrations varying between 0.15, 0.1 and 0.05 g/100 ml. The relative viscosity was calculated. To get the intrinsic viscosity the ratio (η_{sp}/c) was plotted against c and extrapolated to zero to obtain the intrinsic viscosity

C =concentration

η_{sp} =relative viscosity

The average molecular weight was determined from intrinsic viscosity data according to the equation:-

$$N = 1.4 \times 10^{-6} M^{1.34}$$

Where

N = intrinsic viscosity

M =molecular weight

Preparation of jellies: One gram of the sample is added to 22 gram of sucrose and stirred well with 30 ml of distilled water. About 5 ml of 3% citric acid is added in a beaker and boiled for 15 minutes.²⁸

Results and discussion

Physicochemical analysis

The main components of grapefruit peels used for preparation of AIS were listed in Table 1. The moisture content of grapefruit peel sample was (75.25%, 75.37%) for red and white respectively. The grapefruit peels had low ash (1.5, 1.6%), protein (1.05, 1.15%) and crude fiber content (1.73, 1.82%) for both type compared to banana peels which gave (12.8%, 4.2% and 37.3%) for three attributes.²⁹ The ash contents of orange peel contains 2.61%.³⁰ The results of ash content obtained for grapefruit peel were lower than the value of other researchers^{31,32} who reported that grapefruit peel comprises 3.55%, 3.7% ash respectively. Similarly, Annon et al.³³ stated that orange byproduct powder consists of 3.43% ash. In comparison with the peels of other fruits, the protein and fat content of the grapefruit peel were lower than those of lemon peel (7 and 2.5 g/100 g dry matter, respectively) and sweet orange peel (9.1 and 2.6 g/100 g dry matter, respectively). Also lower than that grapefruit peel with 6.8%, 6.65% crude protein reported by other researchers.^{31,32} The lemon peel was reported to contain crude fiber (15.18%) and protein (9.42%) and crude fat (4.98%).³⁴ Whereas for grapefruit peel it was reported to contain 4.9% crude fat and that orange peel consists of 4.53% crude fat.^{32,30} respectively. The variation in all these results could be attributed due type of soil, varieties, season, maturity and environmental changes.

Our results revealed lower titrable acidity of 0.22% for red type and 0.16% for white type which differs from the value of 0.74% as citric acid obtained by other researchers.³⁵ This could be referred to the presences of a comparatively small amount of organic acids and salts in the peel of grapefruit.³⁶ The total sugars obtained was (19.78, 18.89) while reducing sugar amounted to (10.4, 10.2) for white and red type respectively this result in contrast with other who reported low value of 6.35% as reducing sugars.³⁵ The amounts of soluble carbohydrate of the peels of citrus fruit correlate with the stage of fruit

maturity. The ascorbic acid content of two types of grapefruit peels was 150 mg/100 gm and 158 mg/100 gm respectively, this higher than the value 142 mg/100 for grapefruit from Florida³⁶ and similar to the values obtained for orange peels and lemon peels.³⁷ The calcium content was 0.71 mg/100 gm in red type and 0.69 mg/100 gm in white type this result agrees well with the range of 0.55-0.96 mg/100 gm calcium reported earlier.³⁸ The magnesium content was 0.05 mg/100 gm for both types which lower than the range of 0.18-0.28 mg/100 gm magnesium reported.³⁸ The alcohol insoluble solids was 9.5% and 10.5% for red and white respectively (on fresh weight basis) which amounted to 38.0% and 42.0% (on dry weight basis). This value is higher than the value (33.69%) estimated by other researchers.³⁹

Table 1 Components of grapefruit peels

Parameters (%)	Grapefruit peels type	
	Peels of white type	Peels of red type
Moisture	75.25	75.37
Ash	1.5	1.6
Protein	1.05	1.15
Oil	0.2	0.4
Fiber	1.73	1.82
Alcohol Insoluble Solids (AIS)	10.5	9.5
Titrable acidity	0.16	0.22
Ascorbic acid	0.158	0.15
Reducing sugars	10.4	10.2
Total sugars	19.78	18.89
Calcium mg/100g	0.69	0.71
Magnesium mg/100g	0.17	0.17

Each value is a mean of duplicate determinations.

Table 2 summarized our findings of the AIS taken from grapefruit peels which contained (7.05%, 7.17%) moisture, (3.37%, 3.275%) ash, (0.083%, 0.062%) protein, (0.089%, 0.079%) lignin, for red and white type respectively. As shown in Table 2, the calcium content of two types of AIS was 1.04 and 1.83 mg/100 g respectively and the magnesium content were 0.19 and 0.17 mg/100 g respectively. The total pectin of the two types was 2.4, and 2.6% (on fresh weight basis) which amounted to 25.0% and 25.6% (on dry weight basis). Citrus peels contain 20-40% pectic substances calculated on dry weight basis.⁴⁰ The total pectin obtained in this study (25.6, 25.0%) is higher than the range of 15-17% reported by for Indian citrus peels,⁴¹ and lower than the range (30.7%) for Spanish grape fruit peels.⁴² The differences in pectin obtained may be attributed to varietal differences and/or stage of maturity of the fruits.

From Table 3, the characteristic color of pectin obtained from the three samples were all brownish according to IPPA,⁴³ pectin are usually light in color, factors such as surface contamination, environmental factors, types of fruits used and human error might have contributed to the discrepancy in color, this could be due to the amount of acetone used for precipitation and purification during the experiment not been enough.⁴⁴ In cold alkali (NaOH), the pectin suspension obtained from the fruits gave a yellow gelatinous color which turned white when

heated at 85-90°C for 15min, however, it is stated that pectins are unstable under alkaline solution⁴⁵ which corresponded with what was obtained from this research. The pH determination for the pectins were 4.1 for red type, 4.0 for white type and 3.6 for citrus pectin, aqueous solution of pectins are slightly acidic

Table 2 Analysis of alcohol insoluble solids (AIS) grams/100grams (on dry weight basis)

Parameters (%)	Alcohol soluble solids(AIS) of grapefruit peels	
	AIS of white type	AIS of red type
Moisture	7.170	7.050
Ash	3.275	3.370
Protein	0.062	0.083
Lignin	0.079	0.089
Calcium mg/100g	1.830	1.040
Magnesium mg/100g	0.170	0.190
Total pectin	2.60	2.40

Each value is a mean of duplicate determinations.

Table 3 Qualitative test for grapefruit peels compared to commercial citrus pectin

Parameter	Pectic substances		
	Red grapefruit	White grapefruit	Commercial
Color	Brownish	Brownish	Brownish
Solubility in cold water	Insoluble, inibibes, swells after vigorous shaking, form suspension	Insoluble, inibibes, swells after vigorous shaking, form suspension	Dissolved slightly and form suspension after vigorous shaking
Solubility at 85-90°C for 15min	The mixture dissolves	The mixture dissolves	The mixture dissolves
Solubility of pectin suspension in cold alkali	The pectin suspension forms yellow precipitated	The pectin suspension forms yellow precipitated	The pectin suspension forms yellow precipitated
Solubility of pectin suspension in hot alkali	The pectin suspension dissolve and turned milky	The pectin suspension dissolve and turned milky	The pectin suspension dissolve and turned milky

Yield of pectin

Red and white grapefruit peel yielded 25.26-25% pectin on dry basis respectively. This yield was higher than the yields of 15-17% for Indian citrus peel⁴¹ and the other three varieties of grapefruit peel from Florida with yields of 16-19%.³⁶ The results also supported by other researchers who concluded the amount of pectin extracted from grapefruit peel was 21.1% also Khan et al.⁴⁶ were obtained maximum yield of 22.55%.⁴⁷ White and red grapefruit varieties may be considered to be potential good source of pectin.

Physical and chemical properties of pectin (Table 4)

The moisture content: The moisture content of red and white grapefruit peel pectin were (8.96-7.88%) respectively, higher than that of commercial pectin (1.86%). A value of 8.42% for Sudanese grapefruit peel pectin was reported without mentioning the type of grapefruit investigated.⁴⁸ Literature data on moisture content of pectin extracted from different citrus peel lies in the range of 6.4-10%.^{2,49} Pectin should have low moisture content as possible for safe storage and to inhibit the growth of micro-organism that can affect the pectin quality due to the production of pectinase enzymes.⁵⁰

Table 4 Properties of grapefruit peels pectic substances compared to commercial citrus pectin

Parameter	Pectic substances		
	Red grapefruit	White grapefruit	Commercial
Acetyl content%	0.455	1.634	0.624
Methoxyl content%	8.875	7.542	6.798
Degree of esterification	55.01	51.24	50.848
Anhyrouronic acid%	60.95	60.95	68.90
Moisture	8.960	7.880	1.859
Ash	1.800	2.000	0.852
Intrinsic viscosity dl/g	1.500	1.500	3.800
Molecular weight	31620	31620	50380

Results were presented as a mean of two replicates.

The ash content: The ash content was ranged from 1.8%-2% for red and white grapefruit peel pectin respectively, while citrus pectin had low ash content (0.852%). The value reported was 1% for grapefruit which is lower than those of this study.⁴⁸ Low ash content is good for gel formation. The maximum limit for ash content for good quality gel criteria is 10%.⁵¹ The ash content increases as the pectin yield decrease.⁵² Therefore, with respect to this, the pectin isolated agrees with the statement since red grapefruit peel yield of pectin is slightly higher than that of white grapefruit peel yield of pectin.

The protein content: The protein content obtained was 0.0399% and 0.0405% for red and white pectin substances respectively while citrus pectin gave 0.129% commercial pectin usually have protein below 2%, since it prepared from sources of low protein content.

The methoxyl content is an important factor in controlling the setting time of pectin and ability of pectin to form gel.⁵³ The methoxyl content of red and white grapefruit peel pectin were (8.875-7.542%) respectively, higher than that of commercial pectin (6.987%). This

was also higher than the methoxyl reported as, 4.11% for lime, 3.9% for orange and 4.2% for grapefruit by El Mubarak,⁴⁸ Alexander et al.⁴¹ reported 8.62% for lime, 7.6% for orange, 7.73% for sweet orange and 7.4% for grapefruit. The results for methoxyl content is in contrast with findings of⁵² for dragon fruit pectin which showed lower methoxyl content and degree of esterification as compared to commercial apple pectin. Thus, methoxyl content of pectin can vary with the source of raw material used, the method used for extraction in addition to the method used for determination of methoxyl content. Based on the methoxyl content values in this study indicated that grapefruit peel pectin was categorized as high methoxyl pectin (HM) pectin (Methoxyl content >7%).

Degree of esterification: Degree of esterification was higher in red and white grapefruit peel pectin (55.01- 51.24%) respectively, than that of commercial pectin (50.848%) in comparison with 63.2% for lime, 56.1% for orange, 57% for sweet orange and 57.1% for grapefruit.⁴¹ Pectin is divided into two major groups: high-ester pectin, with degree of esterification higher than 50%, and low-ester pectin, with degree of esterification lower than 50%.⁵⁴ A higher degree of esterification causes more rapid setting of gel. Based on their classification, both grapefruit peel pectin are classified as high ester pectin (DE >50%).

Acetyl value: Acetyl value was higher in white grapefruit peel pectin (1.634%) compared to red grapefruit peel pectin and commercial pectin (0.55%-0.624%) respectively. Result of other researchers indicated that the gelling capacity of pectin decreased with increase in the degree of acetylation and samples containing 3.5%-4.0% acetyl gives weak gels while gelling power restored at levels around 2.4% acetyl.^{26,55} Based on acetyl value in this study indicates that both varieties of grapefruits peel pectin have good gelling power. The red grapefruit peel pectin has higher gelling power compared to white grapefruit peel pectin and commercial pectin.

Equivalent weight: The equivalent weight of extracted pectin obtained was 624 and 749 for red and white pectin respectively which were considerably higher than that of citrus pectin (620) and lower than apple pomace pectin (833.33 to 1666.30) grapefruit peel pectin was 953.⁵⁶ The cocoa husk pectin was reported in other studies to be (510.68 to 645.190,⁵⁷ which is lower than that of grapefruit peel pectin reported in this study. The lower equivalent weight could be due to higher partial degradation of pectin, thus increased or decreased of the equivalent weight might be dependent upon the amount of free acid.⁵⁷

The anhydrouronic acid content (AUA): The anhydrouronic acid content value of the red and white grapefruit peel pectin was 60.95% for both of them and 68.9% for the commercial pectin, higher than that reported by other researcher with a value of 32.94% for grapefruit.⁴⁸ The AUA indicates the purity of the extracted pectin and its value should not be less than 65%.⁵⁸ The low value of AUA means that the extracted pectin might not be sufficiently pure with the possible presence of protein, starch and sugars in the precipitated pectin.⁵¹

Composition of neutral sugars in pectin: Composition of neutral sugars in pectin Determination of neutral sugars associated with grapefruit peels pectin by the use of Thin Layer Chromatography techniques, will be helpful to signify its side chain. Individual neutral sugars are summarized in Table 5. The main neutral sugars were arabinose, galactose, rhamnose and xylose. The two grapefruit types of pectin sample studied were similar in their sugar composition. Pectin from citrus peels is characterized by the presences of arabinose, galactose and rhamnose.⁴¹ Neutral sugars such as arabinose, galactose, galacturonic acid and rhamnose are the structural components of

pectin from fruit peel, which have free hydroxyl group (-OH) that can be methylated to methoxyl groups (OCH₃) and the methoxyl content of pectin can vary with the source of raw material used for the extraction of pectin.⁵⁹

Table 5 Sugar composition of grapefruit peel pectin

Sample	Arabinose	Galactose	Rhamnose	Xylose
Pectin of Red grapefruit peel	+	+	+	+
Pectin of white grapefruit peel	+	+	+	+

+ indicates presence of the sugar.

Intrinsic Viscosity and Molecular Weight: The pectins obtained from two types of grapefruit peels exhibited lower viscosity (1.5dl/g) and hence lower molecular weight (31620) compared to citrus pectin which gave (3.8dl/g) viscosity and molecular weight (50380), similar to values of pectin extracted from gold kiwifruit.⁶⁰

Gelling characteristics: Gelling characteristics of pectin is determined by its chemical characteristics. The behavioural properties of the pectin correlate it with its applicability in food formulations. If the gel sets in 10-25min, the pectin is considered as 'rapid setting' and if the time required for the setting of gel is more than 25min, it is 'slow setting'. Jellies prepared using both types of grapefruit peel pectin set within the 10-25min indicating them to be rapid set pectin. Results showed that pectin from grapefruit peels can form gels similar to that formed with commercial pectin. An interesting observation was that grapefruit pectin gel was not very transparent like citrus jelly due to the colour

Conclusion

This study emphasized on pectin extraction and characterizations from two types of Sudanese grapefruit peel. The result of 25% pectin content obtained was promising, as compared with 20-40% total citrus peel pectin reported in literature. The extracted pectin owing to its high methoxyl content >7% can be used as stabilizer/thickening agent in various food. Both grapefruit peel pectin are classified as high ester pectin (DE >50%) indicating more rapid setting of gel. Jellies prepared using both types of grapefruit peel pectin were classified as rapid setting demonstrating that they might be used as a functional food ingredient domestically. In accordance with the national strategy of citrus expansion in Sudan and implementation of citrus juice concentrates industry, the waste or the byproduct such as the grapefruit peel, as a way of remediating to ecological problems and adding value to them could be used as a potential source of pectin. Further investigations need to be directed at the gelling properties of this pectin by means of rheological studies.

Acknowledgements

The author expresses his gratitude to the Ministry of Higher Education and Scientific Research of Sudan who sponsored this research. Thanks are extended to the staff members and technicians of the Department of Food Research Industries for their support.

Conflict of interest

The author declares no conflict of interest.

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