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Extraction and Characterization of Pectin from Apple (Malus Pumila. Cv Amri) Peel Waste

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ABSTRACT

Apple peel, the major waste in preserves manufacturing, contained 1.21% pectin. Peel waste could be stabilized with 600 ppm sulphur dioxide and 1% citric acid. Pectin extraction was better in citric acid than hydrochloric acid. The pectin powder was prepared by triple extraction with citric acid solution (1%) clarification through sedimentation, concentration (24°B), precipitation using ethyl alcohol, vacuum drying and grinding. Physico-chemical properties of pectin powder revealed moisture (10.0%), total ash (1.4%), equivalent weight (652.48), methoxyl content (3.7%), anhydrouronic acid (62.82%), degree of esterification (33.44%), acetyl value (0.68), and jelly grade (80).

Key Words: Apple peel; Pectin; Waste; Gelly grade; Characteristics.

INTRODUCTION

Fruits processing generates solid waste as high as 50% of raw material. Generally the wastes consist of peel, core, pomace, unripe fruits, cull fruits, mechanically damaged fruits etc. Pectin is one of the important products recovered primarily

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from citrus and apple wastes. The average price of the high and low methoxyl pectin is about \$10 and 12 kg⁻¹ respectively. Pectin is used not only in food processing in jam, jellies, marmalades, ketchups, sauces, juices, concentrate, syrups, and yoghurt etc., but also in pharmaceutical preparation such as medical formulations for stabilizing the suspensions. It has an important role in cosmetic industry.^[11] Its tremendous potential has drawn the attention of food scientists for several decades. Mandarin orange peel and pomace,^[2] guavas,^[3,4] jack fruit,^[3,5,6] papaya,^[3,5,6] Assam lemon,^[7] mango peel,^[8] and apple pomace^[9,10] have been studied for their pectin.

In India apples are produced in plenty especially in Himachal Pradesh, Jammu and Kashmir, and Uttranchal.^[9] Other than fresh consumption, the majority of the produce is utilized in manufacturing apple juice concentrate while apple preserve constitutes the next important products. Preserve processing units use *Amri* variety that is medium in size, compact, green in color and capable of withstanding processing conditions. Peel is the major waste since entire fruit is used for preserve making after peeling. At present peel is either fed to animals or thrown into garbage. This study was designed to explore the possibility of utilizing the peel waste for the production of dry pectin.

MATERIALS

Apple Peel

Fresh apple peel was collected from a preserve manufacturing unit located at Amritsar during the month of November 2002.

Pectin

Commercial pectin was procured from CDH Lab reagent, Mumbai.

METHODS

Proximate Analysis^[11]

The fresh peel was analyzed for moisture, ash, acidity, crude fibre, and total sugars following standard techniques.

Pectic Substances^[11]

Peel (50 g) was extracted with HCl (400 mL, 0.05 M) for 2 h at 80–90°C. The mixture was then cooled, volume was made to 500 mL and filtered (Whatman No. 4). Filtrate (100 mL) was neutralized and then NaOH (10 mL, 1 M) was added with



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constant stirring. It was allowed to stand overnight. Acetic acid (50 mL, 1 M) was added, allowed to stand for 5 min, calcium chloride solution (25 mL, 0.5 M) was added, allowed to stand for 1 h, boiled for 1-2 min and filtered through previously prepared filter paper (filter paper wet in hot water, dried in an oven at 102°C for 2 h, cooled in a desiccator and weighed in a covered dish). The precipitates were washed with hot water until free from chloride (tested using 1% silver nitrate). Filter paper containing calcium pectate was dried over night at 100°C, cooled in desiccator and weighed.

Preservation of Peel

Chemical preservatives and citric acid were used for extending the storage life of peel. Three combinations were tried.

- Potassium meta-bisulphite (0.1032% or 600 ppm SO₂) and citric acid (1%).
- Sodium benzoate (0.0413% or 350 ppm benzoic acid) and citric acid (1%).
- Citric acid (1%).

The peel was incubated at 28° C and observed for any off odor or mould growth.

Extraction of Pectin^[12]

The apple peel was extracted three times using extractants (0.02 M HCl, 0.05 M HCl, and 1% citric acid). The clarified extracts were tested for pectin content as calcium pectate and best extractant was selected based on pectin yield.

Pectin Powder

Peel was washed and extracted with citric acid solution (1%) (Fig. 1). In the first extraction equal quantity, in second and third extractions half quantity, of extractant was added and boiled for 25 min. The combined extract was strained through muslin cloth, cooled, mixed with potassium meta-bisulphite, and kept overnight for clarification. The supernatant was concentrated to $24^{\circ}B$ (Abbe's Refractometer), precipitated with alcohol, vacuum dried, and powdered.

Characterization of Pectin Powder

Equivalent Weight^[11]

Pectin (0.5 g), ethanol (5 mL), sodium chloride (1.0 g), carbon dioxide free distilled water (100 mL), and six drops of phenol red indicator were dissolved and



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PEEL \downarrow WASHING \downarrow EXTRACTION WITH 1% CITRIC ACID SOLUTION [Peel:Extract Ratio; 1st 1:1, 2nd and 3rd 1:0.5] \downarrow FILTRATION OF EXTRACT \downarrow STORING THE EXTRACT WITH 600 PPM OF SO₂ \downarrow DECANTATION \downarrow CONCENTRATION OF EXTRACT (24°B) \downarrow PRECIPITATION OF PECTIN WITH ALCOHOL \downarrow WASHING WITH ABSOLUTE ALCOHOL \downarrow DRYING IN VACCUM OVEN (38-42°C, 10 mm Hg) \downarrow **GRINDING OF FLAKES** \downarrow

PECTIN POWDER

Figure 1. Flow diagram of extraction of pectin from apple peel.

titrated against standard 0.1 M NaOH until the color of indicator changed (pH 7.5) to pink and persisted for at least 30 s.

Equivalent weight = $\frac{\text{Wt of sample (g)} \times 1000}{\text{Vol of alkali (mL)} \times \text{Normality of alkali}}$

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Methoxyl Content^[11]

To the neutral solution titrated for equivalent weight containing 0.5 g of pectic substance, added 25 mL of 0.25 M NaOH, shaken thoroughly, and allowed to stand for 30 min at room temperature in a stoppered flask. HCl (25 mL, 0.25 M) was added and titrated with 0.1 M NaOH to the same end point as before.

Methoxyl content (%) =
$$\frac{\text{Vol of alkali (mL)} \times \text{Normality of alkali} \times 31 \times 100}{\text{Wt of sample (g)} \times 1000}$$

Alkalinity of Ash^[11]

The sample was ashed and HCl (0.1 M, 25 mL) was added. The mixture was heated to boil, cooled, and titrated against standard 0.1 M NaOH using phenolphthalein as indicator. Blank titration with HCl (0.1 M, 25 mL) was also carried out.

Alkalinity (%) =
$$\frac{(Blank - Titre) \times Normality of alkali \times 60 \times 100}{Wt of ash (g) \times 1000}$$

Anhydrouronic Acid^[11]

The alkali milli-equivalents from equivalent weight, methoxyl content, and alkalinity of ash were taken to calculate anhydrouronic acid (AUA) content.

 $=\frac{176 \text{ (m.e. for free acid } + \text{ m.e. for saponification } + \text{ m.e. for titrable ash}) \times 100}{\text{Wt of sample (mg)}}$

m.e. = milli-equivalents of alkali

Degree of Esterification^[9]

The degree of esterification was calculated from methoxyl and anhydrouronic acid content using the following expression.

Degree of esterification $=\frac{176 \times \text{Methoxyl content (\%)} \times 100}{31 \times \text{Anhydrouronic acid (\%)}}$

Acetyl Value^[11]

Pectin (0.5 g) and NaOH (25 mL, 0.1 M) were stirred until the pectin dissolved and allowed to stand overnight. The contents were diluted to 250 mL with water and



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an aliquot (20 mL) was placed into the distillation apparatus. Magnesium sulphatesulphuric acid solution (20 mL), magnesium sulphate (100 g) + sulphuric acid (1.5 g)diluted to 180 mL) was also transferred to distillation apparatus, distilled, and about 100 mL of distillate was collected. The distillate was titrated with NaOH (0.05 M)using phenol red indicator. A blank distillation with magnesium sulphate-sulphuric acid solution (20 mL) was carried out and distillate was titrated.

Acetyl value (%) = $\frac{\text{Vol of alkali (mL)} \times \text{Normality of alkali} \times 4.3}{\text{Wt of sample in aliquot (g)}}$

Jelly Grade^[11]

Jelly grade was measured following test jelly method under standard conditions (TSS 68°B, pH 3.2). The sugar, citric acid, and water were kept constant while pectin content was varied. The quality of the gel was judged and jelly grade was calculated for the well set and firm gel.

Jelly grade = $\frac{\text{Wt of sugar (g)}}{\text{Wt of pectin (g)}}$

RESULTS AND DISCUSSION

Proximate Composition of Apple Peel

Apple peel contained 75% moisture, 21.4% fibre, 0.28% acidity (malic acid), 9.27% total sugar (as invert), 1.60% ash, and 1.21% pectic substance (Table 1). Data are scanty regarding composition of apple peel. However, proximate composition of apple pomace was reported previously as 76.74% moisture, 16.16% crude fibre, 2.39% acidity (malic acid), 17.35% total sugar (as invert), 1.65% ash, and 16.95% pectin.^[10] Our results revealed that ash content of peel was similar to, but fibre content was higher than, that of pomace.

Table 1. Proximate composition of apple peel (n=3).

Parameters	Observed value (%)
Moisture content	75.00 ± 0.3288
Crude fibre	21.40 ± 0.8073
Total sugar	9.20 ± 0.0577
Total ash	1.60 ± 0.0208
Pectic substances	1.21 ± 0.1127
Acidity as malic acid	0.28 ± 0.009



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Preservation of Peel

Chemical preservatives along with organic acid were used for extending the storage life of perishable fresh peel. Results showed that the peel preserved with 1% citric acid and 600 ppm SO₂ was stable for about 120 days at 28°C. Another treatment of 1% citric acid and 350 ppm benzoic acid preserved the peel for 60 days at the same temperature. In third treatment only citric acid (1%) was used but it failed to preserve the peel. The study established that apple peel could be preserved with potassium meta-bisulphite and citric acid. The actual storage life would be longer since ambient temperature was lower than incubated temperature thus giving enough time to process the peel for pectin extraction. This method is very cost effective as compared to drying.

Extraction of Pectin

The first treatment using HCl (0.02 M) extracted 0.1896 g calcium pectate in the first, 0.1128 g in the second, and 0.61 g in the third extraction from 100 g of fresh peel (Table 2). Second treatment using HCl (0.05 M) extracted 0.2788, 0.1296, and 0.1822 g calcium pectate contents in the first, second, and third extractions from 100 g fresh apple peel respectively. The third treatment using citric acid (1%) recovered 0.4688, 0.1532, and 0.1456 g calcium pectate in the first, second, and third extraction from 100 g fresh peel respectively. The cumulative pectin showed that 0.02 M HCl, 0.05 M HCl, and 1% citric acid from 100 g of fresh peel extracted 0.4552, 0.5956, and 0.7856 g of calcium pectate. The total pectin in fresh peel was 1.21 g per 100 g thus the present treatments were able to extract majority of the pectin from fresh peel. The data has also been given for dry peel basis considering its moisture content i.e., 75%.

Total pectin extracted from peel revealed that citric acid was more effective than HCl. Moreover it is also used in many foods such as jellies, jams, and marmalades. Thus citric acid was selected for further study. Reports are available on the extraction of pectin using sulphurous acid,^[13] sulphuric acid,^[14] HCl,^[15] nitric acid,^[16,17] oxalic acid,^[18] ammonium oxalate,^[19] sodium hexametaphosphate,^[20] and tartaric acid.^[21]

Table 2. Extraction of pectin from apple peel using different acids (n = 3).

	Amount of	Extracted cal	cium pectate (g/10	0 g fresh peel)
Extraction	extractant used (mL/100 g fresh peel)	0.02 M HCl	0.05 M HCl	1% CA
First extract	100	0.1896 (0.7584)	0.2788 (1.1152)	0.4688 (1.8752)
Second extract	50	0.1128 (0.4512)	0.1296 (0.5184)	0.1632 (0.6528)
Third extract	50	0.1528 (0.612)	0.1822 (0.7288)	0.1456 (0.5824)
Cumulative	200	0.4552 (1.8208)	0.5956 (2.23824)	0.7856 (3.1424)

Values indicated in the parenthesis refer to extracted calcium pectate g per 100 g dry peel.



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Characterization of Pectin Powder

The characterization of powdered pectin obtained from apple waste was carried out for various parameters to evaluate its suitability in food systems. The commercial sample was also analyzed simultaneously for comparison.

Moisture Content

The moisture content of pectin powder was 10.0% and that of commercial sample 7.0% (Table 3). Jain et al.^[9] reported moisture content of pectin prepared from pomace of golden delicious apples to be 8.80%. The moisture content of our pectin was slightly higher whereas that of commercial pectin was less. However Dang^[22] reported the value of moisture content of pectin of papaya as 8.58%, Assam lemon as 7.08%, and galgal as 10.3%. Thus the values of moisture content reported in literature are very near to our results.

Ash Content

The ash content of test pectin powder was 1.44% and that of commercial pectin was 1.164%. The ash content of pectin extracted from golden delicious pomace was 1.84%,^[10] from mandarin pomace was 0.45%^[2] and from mandarin peel was 0.50%. Dang^[22] reported that ash content of pectin was 4.79% in papaya, 0.70% in Assam lemon and 1.72% in galgal. The ash content of test and commercial pectin was lower than those of apple pomace but within the range of other sources.

Equivalent Weight

The equivalent weight of pectin extracted from apple peel was found to be 652.5 and that of commercial pectin was found to be 1030.9 (Table 3). The equivalent weight of pectins from golden delicious pomace and golden delicious partially ripe apples

Table 3. Physico-chemical characteristics of apple peel pectin (n = 3).

Parameters	Apple peel pectin	Commercial pectin
Moisture (%)	10.03 ± 0.020	7.02 ± 0.21
Equivalent weight	652.48 ± 1.48	1030.92 ± 0.69
Methoxyl content (%)	3.7 ± 0.02	5.3 ± 0.07
Anhydrouronic acid (%)	62.82 ± 0.12	70.50 ± 0.29
Acetyl value (%)	0.68 ± 0.04	0.50 ± 0.02
Degree of esterification (%)	33.44 ± 0.04	42.68 ± 0.19
Total ash (%)	1.44 ± 0.31	1.16 ± 0.01





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were 1384 and 1027.7 respectively.^[23] The equivalent weight of the pectin extracted from apple peel was lower than those for the commercial sample and reported values.

Methoxyl Content

Methoxyl content of pectin extracted was 3.7% and that of commercial sample was 5.3%. It has been reported that the methoxyl content from golden delicious pomace and golden delicious partially ripe apple as 8.73 and 7.4% respectively.^[9,23] The methoxyl content of pectin extracted from unblanched, washed, and dried apple peel and pomace was 8.0%^[7] and of blanched one was 8.74%. Srirangarajan and Shrikhande^[8] have reported the methoxyl content of pectin extracted from mango peel to be 8.25%. The methoxyl content of test and commercial pectin was lower than the reported values.

Anhydrouronic Acid Content

Anhydrouronic acid content of pectin is an important parameter which indicates the suitability of pectin for its use in jams, jellies etc. The anhydrouronic acid content of test pectin was 62.8% and that of commercial sample was 70.5%. It has been reported that the anhydrouronic acid content of pectin extracted from golden delicious pomace and golden delicious partially ripe apple was 74.1% and 73.9%, respectively.^[9,23] Anhydrouronic acid content of pectin extracted from peel was slightly low but that of commercial sample was in agreement with the reported values. Pectin obtained from mango peel was reported to contain 61.1% anhydrouronic acid.^[9]

Acetyl Value

The acetyl values of pectin prepared from apple peel and commercial were 0.62 and 0.50% respectively. It has been reported that the acetyl value of pectin extracted from golden delicious pomace and golden delicious partially ripe apple were 0.21 and 0.46% respectively.^[9,23] Thus the values obtained in current study were higher than the previously reported values.

Degree of Esterification

The degree of esterification of pectin extracted from peel was found to be 33.44% and that of commercial sample was 42.68%. The degree of esterification of pectin extracted from golden delicious pomace and golden delicious partially ripe apple was 72.40 and 56.9% respectively^[9,23] whereas that of mango peel was 76%.^[8] The degree of esterification for apple peel pectin was less than reported values.



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Jelly Grade

Jelly grade is the most important parameter, which indicates the number of parts of sugar required for one part of pectin to produce jelly of desirable consistency under standard conditions (TS 68%, pH 3.2). The jelly grade of extracted pectin from peel was 80 and that of commercial sample was 152. It has been reported that the jelly grade of pectin extracted from golden delicious pomace and golden delicious partially ripe apple were 160 and 170, respectively.^[9,23] Dang^[22] reported that jelly grade was 227 for papaya, 227 for Assam lemon, and 350 for galgal. Srirangarajan and Shrikhande^[8] reported the jelly grade of pectin obtained from mango peel was 200.

The low temperature storage of fresh raw apple pomace showed a decrease in methoxyl content and anhydrouronic acid content due to the activities of pectin esterase and polygalacturanase.^[12,24] The liquid pectin extract preserved with 359–700 ppm SO₂ and kept at 20–25°C indicated decreased methoxyl content and jelly grade.^[12,25] Thus low grade of pectin in current study might be due to enzymatic action on pectin during collection and transport of the fresh peel from processing unit to laboratory and the use of SO₂ for the preservation of pectin extract.

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