EFFECT OF PROCESS PARAMETERS ON PECTIN EXTRACTION FROM KAGZI LIME PEEL

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ABSTRACT

The experiments were conducted at Department of Processing and Food Engineering, College of Agricultural Engineering & Technology, Junagadh Agricultural University, Junagadh. The standard operating condition was prepared to extracts the pectin from lemon peel. The results suggested that the temperature were in line with the pH, one of the factors for extraction of pectin yield and quality parameters like moisture content, ash content, methoxyl content, equivalent weight and yield. Different pH was used with temperature of 70^0C, 80^0C and 90^0C. The results suggested that wider variation was observed in yield and quality of pectin extracted at these temperature. At 70^0C, the pectin extracted from lemon peel at irrespective of pH. It was ranges between 12.60 to 13.60 % yield and methoxyl Content was recorded higher at 4.65% and lower at 4.28%. Similarly, at 80^0C, methoxyl content was recorded at 6.63% and equivalent weight of the pectin extracted was ranges between 724.64 to 793.65g. Thus, the moisture, ash and methoxyl content was increased with increased temperature and reverse trend was observed in equivalent weight of pectin content. The higher equivalent weight of pectin extracted at a pH of 1.6 (963.68g) at 70°C and the lower equivalent weight (332.62g) of pectin extracted at a pH of 2.9 and 90^0C. The result of SEM indicated that the surface of pectin powder is rough, and heterogeneous.

KEY WORDS: Ash, lemon, lime, pectin, pH, methoxyl, moisture content,

INTRODUCTION

The major lime/lemon producing districts in Gujarat are Ahmedabad, Kheda, Mehsana and Bhavnagar. The total area under citrus fruits during 2012-13 in Gujarat is 31.2 thousand ha with production of about 331.89 thousand and productivity of 10.6 tonne/ha (Anonymous, 2015). Despite of these, local market having problems of post harvest losses during handling, separation, storage and through biotic and abiotic stresses. The Peel of citrus fruits after the extraction of juice will not used by any manner. Therefore, pectin extraction may be one of the sources for value addition and Industrial application.

Pectin is used in jellybeans. In addition to this, pectin has many other uses in food and pharmaceutical industries. Pectin is also being recommended for use as fat replacer. On account of its ever-increasing use

and demand, pectin has become an indispensable ingredient in food industry. The citrus peel processing industry can get a complete makeover if due importance is given for separation of useful ingredient from citrus peel. Indigenous production of high grade pectin is one of economic significance, as it will not only reduce the imports, but also the desired food formulation can be made by using lesser quantities of it. Moreover, production of high-grade pectin has not been achieved successfully on commercial scale. The annual global market for pectin in 2009 was approximately 42,000 tonnes, and is continuously expanding at a rate of approximately 3% per annum. Out of these, approximately 70%, 25% and 5% of pectin are produced from citrus peel, apple peel and other fruit & vegetable residues, respectively (Erika *et al.*, 2009). Thus, a wide gap was exists between production and demand for pectin. Numerous researchers and scientists had worked on the separation of pectin from lemon peel and reporting their findings, but the citrus have larger variation in cultivars as well as composition. Therefore, efforts are being made to optimize the process and see the effect of process parameter like as temperature and pH in the extraction of pectin from Kagji lime waste peel with some modification in process.

Lemon peel and its composition

The peel is a by-product of lemon juice processing, with a high potential use. Two different tissues are found in what is colloquially called lemon peel, flavedo and albedo (Augusti, 2003). Flavedo is the peel's outer layer, whose colour varies from green to yellow. Albedo is the major component of lemon peel and is a spongy and cellulosic layer laid under flavedo (Figure 1). The thickness of the albedo fluctuates according to several variables like variety and degree of ripeness.

Chemical composition of Lemon peel

Composition	**Per 100g**
Protein	1.5 g
Fat (total Lipids)	0.3 g
Carbohydrate, Total (by Difference)	16.0 g
Moisture	81.6 g
Sugar, Total	4.17 g
Fibre, Total Dietary	10.6 g
Energy (kilojoules)	197 kJ
Calories (energy)	47 Cal

MATERILAS AND METHODS

Sample preparation and determination of moisture content

The fresh lemon peels was brought and washed thoroughly with tap water to remove the adhering pulp and any other foreign matter attached to the lemon peels. The sample was placed inside the hot air oven at 50 ± 2^0C till it gets constant weight; the sample was cooled in a desiccator and weighed (5 to 6 h). The difference in the initial and the final weights of the sample was taken as the water removed and expressed in per cent dry basis and wet basis (A.O.A.C., 1952).

$$\text{Moisture content (\%)} = \frac{\text{Weight of residue}}{\text{Weight of the sample}} \times 100$$

Ash content

The pectin extracted at temperatures of 70^0C, 80^0C and 90^0C, a sample of 0.5 g was taken in silica crucibles from each temperature for the estimation of ash content. The crucible was kept in the electrical muffle furnace for 5 h at 600^0C temperature. The crucible with completely ignited pectin sample was removed after completion of process then it was cooled to room temperature in desiccators and reweighed. The difference between initial weight of empty crucible and final weight of crucible with ash was taken as weight of pectin ash. The percent ash content was calculated as per A.O.A.C., 1952, the formula given below:

$$\text{Ash Content, \%} = \frac{\text{Weight of ash}}{\text{Weight of pectin}} \times 100$$

Equivalent weight

The pectin sample of 0.5 g and 5 ml ethanol were taken into the 250 ml conical flask. 1 g of sodium chloride to sharpen the end point, 100 ml of demonized distilled water and 5 drops of phenol red indicator was added to pectin ethanol solution. The solution was shake thoroughly till all the pectin substances was dissolved and no lumps were retained on the side of the flask. The solution was titrated slowly with 0.1N NaOH until the colour of the indicator changed; the colour change should persist at least for 30 sec. The equivalent weight of pectin was estimated and calculated according to Zhao-Mei *et al*., 2010.

$$\text{Equivalent weight, g} = \frac{\text{Weight of sample} \times 1000}{\text{ml of alkali} \times \text{Normality of alkali}}$$

Methoxyl content

Methoxyl content was determined by saponification of the pectin and titration of the librated carboxyl group (COOH) with standard NaOH as per the procedure described by Chakraborty and Ray (2011). The neutral solution was titrated for equivalent weight; containing 0.5 g pectin. In the solution 25 ml of 0.25N NaOH was added and was thoroughly shacked. The solution was allowed to stand for 30 min. at room temperature. After 30 min. 25 ml of 0.25N HCl was added to the solution. The solution was shacked thoroughly and titrated by 0.1N NaOH until the colour of the solution changes. Methoxyl content was calculated according to following formula given by Zhao-Mei *et al*., 2010.

$$\text{Methoxyl Content, \%} = \frac{\left(\text{Normality of Alkali} * \text{ml of alkali} * 3.1\right)}{\text{Weight of sample}}$$

Scanning electron microscopy of pectin

Scanning electron microscopy blocks of processed dry pectin powder were viewed in longitudinal and observed in a scanning electron microscope (Zeiss model) at 5 kV. The dried powder size of 20, 30, 100 micrometer of samples dehydrated at critical point. The dry powder were then mounted on surface upwards, onto aluminium stubs using silver conducting paint and coated put in a sputter coater. Representative specimens were examined with a Zeiss scanning electron microscope and photographed.

Flow chart

The extracting conditions and flowchart for extraction of pectin as depicted in Figure 2. The extraction of pectin was done at 70^0C, 80^0C and 90^0C for 1 h with constant starring from dried lemon, the pH of mixture was measured with the help of pH meter. The pH of mixture was in the ranges from 1.5 peels. After extraction the extract and residues were separated by using centrifuge at 8000 rpm for 10 min. Pectin was precipitated by treating extract with equal volumes of 95 percent acidified acetone. The precipitated pectin was separated by simple filtration using muslin cloth. Separated pectin was again washed with two volumes of 95% ethanol. Washed, separated pectin was spread on thin polyethylene paper or petri-plates and dried in hot air oven at 45^0C till it gets constant weight. After drying the dried pectin was grinded into 85 mesh size powders by using power driven grinder. The dried pectin powder was filled in airtight polyethylene bag and kept in deep freezer to avoid moisture absorption and microbial activities.

RESULTS AND DISCUSSION

Individual effect on quality of pectin at 70^0C temperature

Statistically analyzed results showed variation among the different temperature in all the treatments as shown in Table 1. At 70^0C the pectin extracted from lemon peel at irrespective of pH. It was ranges between 12.60 to 13.60 % yield. In case of moisture content, it was ranges from 46 to 50 %. The ash content of the pectin at 70 ^{0}C, ranged between 7.45 to 7.84%. Similarly, methoxyl content was recorded higher at 4.65% and lower at 4.28%. Equivalent weight of the pectin extracted was ranges between 909.9 to 1020.41g. Estimated parameter at 70^0C for pectin, the co-efficient of variance for moisture content, Ash content, methoxyl content, equivalent weight and yield were 4.17, 2.55, 4.17, 5.78 and 3.82 respectively, while Standard error of mean was 1.15, 0.11, 0.11, 32.15 and 0.29 for the respective parameters. Similar variation were observed by Erika *et al*. (2009) during pectin acid extraction from passion fruit peel using three different acids (citric, hydrochloric or nitric) at different temperatures (40–90 ^{0}C), pH (1.2–2.6) and extraction times (10–90 min), with and without skins using a 24 factorial design. Temperature, pH and extraction time had highly significant effects on the pectin yield.

Individual effect on quality of pectin at 80^0C temperature

Individual effects on quality of pectin extracted at 80^0C temperature are depicted in Table 2. Statistically results showed variation among the different temperature in all the treatments. At 80^0C the pectin extracted from lemon peel, irrespective of pH. It was ranges between 11.67 to 12.07% yield. Moisture content was ranged from 70 to 76 %. Ash content

of the pectin at 80^0C, it was in between 8.16 to 9.39%. Similarly, in case of methoxyl content, higher was recorded at 6.63% and lower at 6.32 %. Equivalent weight of the pectin extracted was ranges between 724.64 to 793.65g. Similar findings were observed by (Kar and Arslan, 1999; Masmoudi *et al*., 2010b and Masmoudi *et al.*, 2012).

Individual effect on quality of pectin at 90^0C temperature

Individual effects on quality of pectin extracted at 90^0C temperature are depicted in Table 3. Statistically results showed variation among the different temperature in all the treatments. At 90^0C the pectin extracted from lemon peel at irrespective of pH. It was ranges between 10.33 to 12.00 % yield. Moisture content was ranged from 70 to 80 %. Ash content of the pectin at 90^0C, it was in between 9.22 to 9.80%. Similarly, in case of methoxyl content, higher was recorded at 8.00% and lower at 8.37 %. These results are in agreement with Zhao-Mei *et al*. (2010), who studied the extraction technology by optimized single factor experiment and orthogonal experiment. They assessed the influence of pH, time, temperature, and liquid-solid ratio on the extraction of low methoxyl pectin. Their results showed that optimum extraction condition for liquid-solid ratio (ml/g) of 21:1 and pH 2.0 at 80 ^{O}C for 70 minutes. Equivalent weight of the pectin extracted was ranges with 328.95 to 335.57g. Estimated parameter at 90^0C for pectin, the co-efficient of variance for moisture content, Ash content, methoxyl content, equivalent weight and yield were 6.68, 3.10, 2.27, 1.01 and 7.47 respectively. Standard error of mean were 2.91, 0.17, 0.11, 1.94 and 0.48 for the respective parameters.

Scanning electron microscopy of Lemon pectin

The Scanning Electron microscopy images are the first direct evidence of the interactions between other molecules in structure with high-methoxyl pectin (HMP). Using SEM, a new type of microstructure formed in pectin of lemon was characterized. It composed of continuous, porous network at the length scale of 20, 30, 100 micro meter as depicted in Figure 2. SEM are appears to offer unique possibilities to characterize the structural elements of pectin over a wide range of length scales 300x and 509x, the result of SEM indicated that the surface of (chitosan-poly (vinyl alcohol)-pectin) pectin film is rough and heterogeneous. The results were compared with the different pH and temperature extracted pectin by scanning electron microscopy. Masmoudi *et al.* (2010a) studied the microstructure and the rheological properties of lemon-pectin mixtures and compared to those of pure lemon (high methoxyl: HM) and date (low methoxyl: LM) pectins. They confirmed results by scanning electron microscopy, which revealed in homogeneous gels where dense aggregated network and loose, open network areas were present. Lam *et al.* (2009) also studied on image analysis through SEM and indicated that at pH 7.5 and 3.5 pectin showed a bimodal distribution of sizes with an average [d (0.5)] of about 0.05 micro meters, but at pH 3.8 the proteins formed larger aggregates than at high pH.

CONCLUSION

From the results, it can be concluded that wider variation was observed in yield and quality of pectin extracted at different temperature. The moisture, ash and methoxyl content was increased with increased temperature and reverse trend was

observed in equivalent weight of pectin content. The result of SEM indicated that the surface of pectin powder is rough, and heterogeneous.

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