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Extraction and Characterization of Pectin from Orange Peels

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Abstract

The sweet oranges (Citrus sinensis (L.)) are a very commonly growing tree fruit in the world. The present work explores the possibility of separation of essential oils and pectin from the orange peels. It is found from the experimental observations that the peel source, for extraction of pectin, when taken after extracting orange oil through simple distillation gives higher yield than leaching residue [1]. This work uses Soxhlet extraction for the extraction of oil and the pectin is extracted by acid extraction method from the remaining peels. Strong acids are corrosive and may be a potential threat to health. Moreover, the liquid waste generated from the industrial processes might lead to burden the environment and a high cost might incur for treating the strong acidic waste [2]. Varying pH solutions of citric acid are used for the pectin extraction. The effect of pH of the acid solutions and mesh size of the orange peels on pectin yield and composition was studied in a citric acid extraction process. The pectin yield and degree of esterification (DE) of the extracted pectin ranged from 7.3 to 52.90% and 5.1 to 71.0% respectively. It was found that extraction pH was the most important parameter influencing yield. These results demonstrate the successful extraction of essential oil and pectin, providing potential benefits for industrial extraction of pectin from an economic and environmental point of view.

Keywords: Orange peels, pH, pectin, Soxhlet extraction, degree of esterification, yield.

INTRODUCTION:

Citrus fruits are at the top not only in total production, but also in economic value. One of them; oranges, specifically, the oranges (Citrus sinensis (L.)) are a very commonly growing tree fruit in the world. Oranges are widely cultivated in tropical and subtropical climates, which is peeled and eaten whole, or processed to extract orange juice and also for the fragrance. These essential oils (fragrance) are a mixture of volatile compounds as terpenes and oxygenated derivatives such as aldehydes (citral), alcohols and esters [3]. They have a great commercial importance and various applications in many food industry products because of their aroma and functional properties (antifungal, antimicrobial, etc.) which make them excellent additives. The present work explores the possibility of separation of essential oils and pectin from the orange peels.

Pectin is a natural, biocompatible, biodegradable and renewable polysaccharide characterized as an emulsifier, gelling agent, glazing agent, stabilizer, and/or thickener in commercial applications; all of which are in fact subsets of the term ―rheology modifier [2-4]. Pectin gels are formed when the molecule chains are cross-linked, forming a three-dimensional network where water and co-solutes are retained [5].

Consumption of pectin has been shown to reduce blood cholesterol levels. In the large intestine and colon, microorganisms degrade pectin and liberate short-chain fatty acids that have positive influence on health [6].

The Joint FAO/WHO Expert Committee on Food Additives (JECFA) has recommended pectin as a safe additive with no limit on acceptable daily intake [7]. Pectin is quite stable under the acidic condition of the stomach, although a slight de-esterification can occur. Without the fermentation process, pectin would pass almost unchanged through the digestive system [8]. Increasing consumer awareness of a healthy lifestyle and the emerging trend to produce functional food has made pectin popular. It has been reported that pectin has numerous positive influences on health including improving colonic health, lowering of cholesterol and serum glucose levels, reducing cancer propensity, and stimulating the immune response [9]. The degree of methyl-esterification (also known as degree of methylation) (DM) of GalA units is used to classify pectin. DM is a percentage which expresses the molar ratio of methyl-esters present to GalA units (includes both free GalA and substituted GalA) [10]. It is the major parameter affecting gelling, influencing surface tension and emulsion formation.

The DM percentage above 50% is classified as high methyl ester (HM) pectin while those less than 50% is known as low methyl ester (LM) pectin [11]. The lower the DM, the faster pectin is hydrolysed, probably due to a lower amount of methyl-esterified target groups. This in turn influences gel strength, leading to the formation of a weaker gel [12]. However, at controlled or reduced temperatures, there is a higher possibility that LMP will be obtained without extensive main-chain breakdown. Enzyme de-esterification has becoming increasingly popular for obtaining LMP in an efficient and environmentally sustainable manner [13, 14].

Rapid-setting HMP is usually obtained after a short extraction time at temperatures close to boiling [15]. This is due to short extraction times with high temperature reduces de-esterification. Conversely, long extraction times with low temperature favour slow-setting HMP or even LMP. Therefore, it is important to select suitable extraction conditions to obtain pectin with the desired properties [16, 17].

This work aims to extract and characterize pectin from orange peels using citric acid at various pH.

MATERIALS AND METHODS:

Chemicals

Citric Acid- 99.5% pure, Sodium hydroxide- 0.09-0.11 N (Concentration), Phenolphthalein- 0.1% w/v, Ethanol-99.9% pure, Hydrochloric Acid - 0.1±0.0005 N, Petroleum ether- 99.5% pure.

This work is divided into two parts: (a) Separation of essential oil (b) Extraction of pectin

Separation of essential oil

Orange essential oil is present in small ductless gland contained in the peel of the orange fruits. The main constituent of orange peel essential oil is d-limonene (present to the extent of at least 90 %), which is the only hydrocarbon present [18, 19]. Distillation is the most widely used method to produce essential oils. In this work the Soxhlet Extraction method has been adopted ensuring no alteration in the properties of the components and greater yield with appreciable solvent recovery.

Extraction of pectin

Extraction is the most important process in the pectin production. Pectin extraction in a hot diluted strong mineral acid solution is the most commonly used method [19]. Strong acids are corrosive and may be a potential threat to health. Moreover, the liquid waste generated from the industrial processes lead to burden the environment and a high cost might incur for treating the strong acidic waste [20, 21].

This work takes into account different variations starting with the size of the crushed orange peels followed by the time of extraction and the pH values of the acidic solution used for the extraction of pectin.

The orange peels are sun dried till their moisture content is negligible. They are then crushed and 16, 32 and 60 mesh screens are used to separate the powdered peels accordingly. 80 gms of powdered peel of 16 mesh size is taken and fed to the Soxhlet apparatus with 1000mL of petroleum ether (taken as solvent). The setup is maintained at 40^0C (B.P. of Petroleum ether) for 6 hours, 9 hours and 12 hours respectively. The oil collects at the bottom in the solvent which can be separated by simple distillation.

Further, the powdered peels are collected separately after the solvent extraction is complete.

Citric acid solutions of pH 1.0, 1.5, 2.0 and 2.5 are prepared. 5gms of powdered peels collected are heated with the pH solutions prepared for 30 minutes (optimum time) at temperature 65^0C with continuous stirring. After cooling the solution, it was filtered with muslin cloth. The filtrate was added to double amount of ethanol and allowed to precipitate. The jelly-like precipitate formed is nothing but pectin which was subsequently washed with ethanol two times. Pectin was then dried in a hot air oven at 40^0C for 20 minutes. The process was repeated for 32 and 60 mesh size *Fig. 1*.

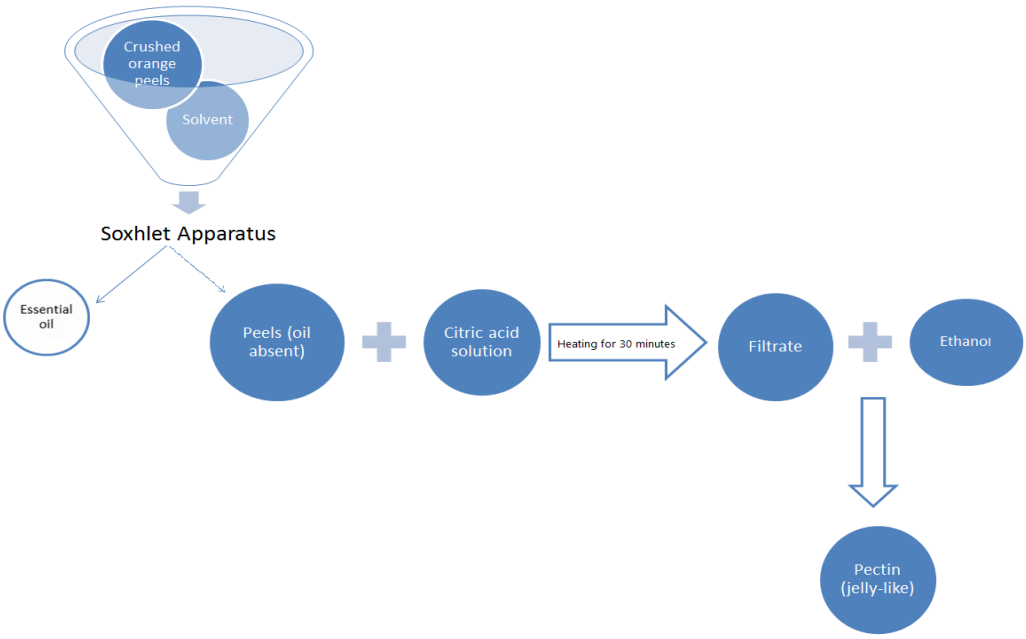


Fig 1: Flow sheet for the acid extraction of pectin

Now, the pectin yield can be calculated as:

$$\text{Yield}(\%) = \frac{\text{Amount of dry pectin obtained(g)}}{\text{Amount of orange powder taken(g)}} * 100$$

The degree of esterification, DE is defined as the ratio of esterified galacturonic acid groups to the galacturonic acid groups present [22]. 0.2 g of dried pectin sample was moistened with ethanol and dissolved in 20 ml distilled water. Three drops of phenolphthalein were added into the sample. The sample was titrated with 0.1 N sodium hydroxide. The result was recorded as the initial titration volume once some pink colour appeared. The number of free carboxy group was calculated from the volume of 0.1 N sodium hydroxide solution spent for initial titration. 10 ml of 0.1 N sodium hydroxide was added to neutralize polygalacturonic acid. The sample was

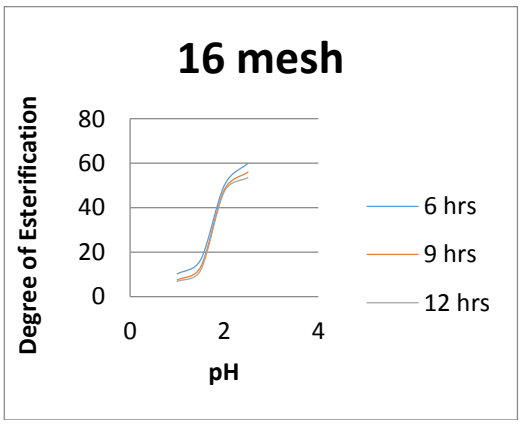
plugged with a stopper and shaken vigorously, then allowed to stand at room temperature for 2 hours to de-esterify pectin. 10 ml of 0.1 N hydrochloric acid was added to neutralize sodium hydroxide and the sample was shaken until its pink colour disappeared. Three drops of phenolphthalein were added into the sample and the sample was further titrated with 0.1 N sodium hydroxide. The volume of titration was recorded as final titration volume once some pink colour appeared. The number of the esterified carboxy group was calculated from the volume of 0.1 N sodium hydroxide solution spent for final titration [23]. The degree of esterification can be calculated as:

$$DE(\%) = \frac{Final\ titration\ volume(ml)}{Intital\ titration\ volume(ml) + Final\ titration\ volume(ml)} * 100$$

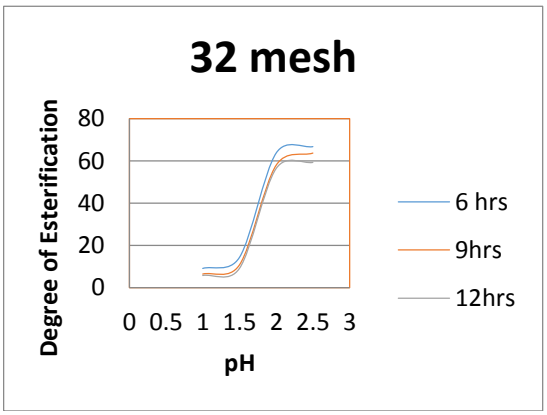
RESULTS AND DISCUSSIONS:

Degree of Esterification

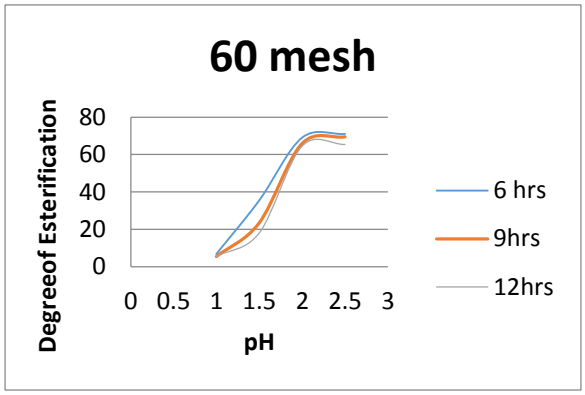
The influence of pH on Degree of Esterification was studied in a citric acid extraction process. As seen from the *Fig 2 (a, b, c),* at low pH (<2.0) Low Methoxyl(LM) pectin is formed whereas at pH>2.0, High Methoxyl (HM) pectin is formed. LM Pectin will form a thermo- irreversible gel, which means that it will stay gelled even when heated to temperatures that would normally melt it [24]. High Methoxyl pectin is the form of pectin traditionally used for canning applications. It requires high amounts of sugar to gel and is very sensitive to acidity [25]. Low Methoxyl pectin has been used in the food industry to create low-sugar jams because it does not require high sugar levels to gel and has become popular for pastries and molecular recipes designed not to be as sweet [26]. Low Methoxyl pectin is used as a gelling agent, thickening agent and stabilizer. Low Methoxyl pectin can also be used as a fat substitute in baked goods and to stabilize acidic protein drinks such as drinking yogurt [27].



(a)



(b)



(c)

Fig 2: Variation of Degree of esterification w.r.t pH at 6, 9 and 12 hours of Soxhlet run time with orange peels of mesh size (a) 16, (b) 32, (c) 60

Yield

pH is considered as one of the more crucial parameters affecting the amount and properties of extracted pectin. *Fig. 3* shows that pectin yield decreased with increasing pH, highest being 52.90% at pH 1 and 60-mesh size. The presence of high concentration of hydrogen ions in the solvent has stimulated the hydrolysis of protopectin [28- 30]. At low pH, as the hydrogen ion concentration of the solution is increased, ionization of the carboxylate groups is repressed, i.e., the highly hydrated carboxylate group is converted into hydrated carboxylic acid groups [31, 32]. The loss of carboxylate groups is able to reduce the repulsion of the polysaccharide molecules which promotes the gelation properties of pectin giving more precipitated pectin at lower pH [33]. This observation agreed with previous work who extracted pectin from apple pomace and sugar beet pulp where the yield increased with increasing acid strength. Further, increase in the yield of pectin is also noted as the size of the powdered peels decreased [11, 34]. This happens as the surface area available for mass transfer increases with the decreasing size thus increasing the yield [35].

CONCLUSIONS:

The present work revealed that the sweet orange peels are good source of orange oil and pectin and does have the potential to become important raw material for food processing industries. The maximum pectin yield is 52.90% clearly represented by the *Fig. 3.* The degree of esterification varies from 5.1 to 71.0% according to pH and mesh sizes as shown in fig. 2(a, b, c). The process in which essential oil is first extracted followed by acid extraction of pectin is most suitable for industrial production. These results demonstrate the successful extraction of essential oil and pectin, providing potential benefits for industrial extraction of pectin from an economic and environmental point of view.

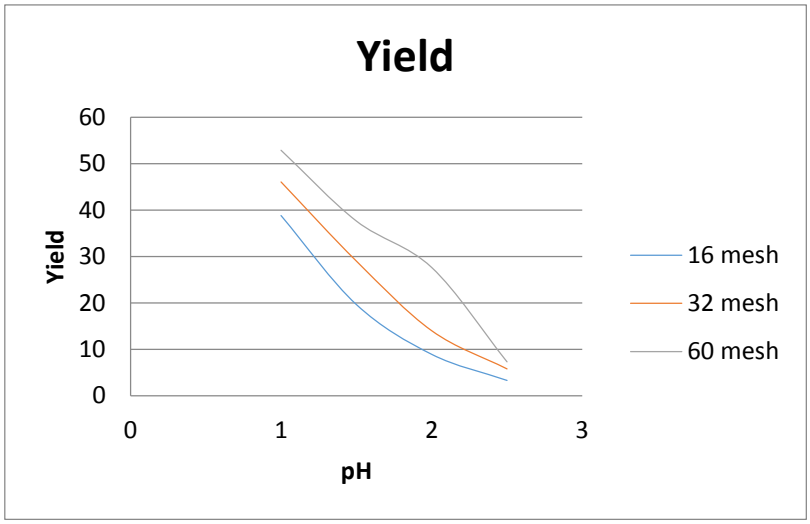


Fig 3: Variation of yield of pectin extracted from 16, 32 and 60 mesh size peels w.r.t pH.

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