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Optimisation of Microwave-Assisted Extraction of *Artocarpus integer* Peel Pectin with Choline Chloride based Deep Eutectic Solvent

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ABSTRACT

Extraction of pectin from natural resources using a biodegradable solvent is an alternative to wasteful and corrosive acidic extraction. Traditionally, attempts to extract pectin from biomass have resulted in low yields and a large consumption of corrosive solvents. Therefore, microwave-assisted extraction (MAE) with deep eutectic solvents (DES) of choline chloride and malonic acid (ChCl:MA) at a mole ratio of 1:2 was used in this study. The objective of this study was to extract pectin from Artocarpus integer peels (AIP) and to optimise the extraction conditions in terms of the percentage of DES, solid-to-liquid ratio of AIP to DES, microwave power, and extraction time. The results showed that the pectin yield increased as the percentage of DES, solid-to-liquid ratio, power, and time increased. The extraction of pectin up to 30.17 % with MAE was optimised at a 1:50 ratio, 50 W, 4 min, and 5 % DES. These results indicate that the proposed MAE-DES technique is excellent for extracting pectin with high yield from AIP.

Keywords: Microwave-Assisted Extraction; Deep Eutectic Solvents; Artocarpus Integer Peels; Pectin; Optimisation.

INTRODUCTION

Pectin, a complex polysaccharide, is highly functional and widely used in the food, pharmaceutical, and cosmetic industries [1]. Pectin is commercially extracted from various raw materials, including apple pomace and citrus peel, using acids at high temperatures [2]. Another potential source of pectin, AIP which is derived from the tropical fruit commonly known as cempedak, has sparked interest among researchers owing to its enormous waste, as it can be converted to valuable pectin with a high pectin content and high quality [3]. However, efficient extraction of pectin from these peels remains a challenge, necessitating the exploration of a novel extraction technique. The extraction of pectin from AIP involves several factors such as extraction techniques, solvent used, solid-to-liquid ratio, and extraction time. Conventionally, strong acids, such as nitric acid, sulfuric acid, and hydrochloric acid, have been used to extract pectin because of their high effectiveness and higher yields. However, there is a problem related to their nature, as they are corrosive and lead to environmental hazards. Organic acids, such as citric acid, are popular replacements for corrosive acids; however, the pectin yields obtained are low [4]. Therefore, ionic liquids (ILs) have been introduced as alternatives because of their promising properties such as low volatility, high thermal stability, high polarity, and tunability [5]. However, these common ILs are expensive and difficult to prepare because of the need for purification steps. Therefore, deep eutectic solvents (DES), which are third-generation IL, are preferable alternatives to pectin solvent extractors because they are easy to prepare, non-toxic, biodegradable, and have low melting points [6].

DES are a class of ILs formed by combining a hydrogen bond acceptor (HBA) and hydrogen bond donor (HBD) in a specific ratio. These solvents exhibit unique properties, such as low volatility, tunability, and eco-friendliness, making them attractive alternatives to conventional solvents in extraction processes [7,8]. DES have demonstrated exceptional solubilising capabilities for bioactive compounds, enhancing extraction efficiency, and offering opportunities for sustainable extraction practices [8]. Recently, new advanced methods such as enzyme-assisted extraction (EAE), ultrasonic-assisted extraction (UAE), and microwave-assisted extraction (MAE) have been introduced to enhance pectin extraction. EAE uses pectinases as enzymes to selectively break down the cell wall structure to release

pectin from plant materials. EAE offers advantages such as selectivity and improved pectin yields; however, the enzyme is very expensive, requires a longer extraction time, and is inefficient [9]. Later, UAE was used as a promising new technique for pectin extraction, as it requires less solvent and reduces the environmental impact in terms of energy and time. Even so, the use of this method is limited due to the potential of ultrasonic changes in the morphology of pectin, which leads to greater porosity and lower pectin quality. The MAE is a modern extraction technique that employs microwave irradiation to accelerate the extraction process. By utilising microwave energy, MAE promotes rapid heating, improves extraction kinetics, reduces extraction time, and enhances yield [10,11]. This technique has gained popularity for natural product extraction because of its efficiency, reduced solvent consumption, and energy-saving benefits [12].

Therefore, the combination of MAE and DES serves as a new promising pectin extraction technique because it reduces the amount of organic and toxic solvents, is easy to operate, and has low energy usage compared with hot acid extraction, which can degrade pectin and reduce its quality. This method involves the formulation of a DES tailored for pectin extraction from AIP, and the application of microwave irradiation to enhance the process. Thus, the objective of this study was to prepare DES and optimise the pectin extraction process based on factors such as the solid-to-liquid ratio of AIP powder to DES, microwave power, extraction time, and percentage of DES to achieve the maximum pectin yield. The main results of this study demonstrated the efficacy of DES and MAE in extracting pectin from AIP. The optimised extraction conditions resulted in higher pectin yields compared with conventional methods, highlighting the potential of DES and MAE as sustainable and efficient extraction techniques. These findings will contribute to the development of an eco-friendly extraction technique, utilisation of agricultural waste, and advancement of green chemistry practices.

METHODOLOGY

Preparation of deep eutectic solvents

The DES preparation was adopted and modified from the work of Shafie *et al.* [13]. The DES was composed of a 1:2 mixture of a hydrogen bond acceptor (choline chloride) and hydrogen bond donor (malonic acid) (ChCl:MA). The mixture was stirred (80 °C) until a colourless liquid was formed, which remained after 24 h at room temperature. To avoid moisture absorption, DES was stored in a desiccator.

Optimisation pectin extraction from AIP

The conditions for pectin extraction were optimised based on the solid-to-liquid ratio (1:10-1:50 g/mL), microwave power (90-50 W), extraction time (1-5 min), and DES (1-5 %v/v). In each experiment, one component was varied while the others remained constant to obtain the percentage yield of *Artocarpus integer* Peels Pectin (AIPP). The extraction procedure was modified based on the method described by Shafie *et al.* [13]. AIP (15 g) was lyophilised in a freeze dryer and sliced into small pieces. Prior to extraction, the lyophilised materials were crushed, meshed into powder, and stored (4 °C). The peel powder was suspended in DES and extracted in a microwave under the conditions listed in Table 1. The slurries were then centrifuged at 4500 rpm for 10 min and filtered through a muslin cloth. The filtrates were then precipitated using ethanol for 2 h (volume of (98 %) (v/v) ethanol = four times diluted DES) at room temperature (25 °C). Before analysis, the precipitates were filtered and lyophilised to calculate the yield on a dry basis. Eq. (1) was used to determine the percentage yield of *Artocarpus integer* Peels Pectin (AIPP).

$$\text{Yield of AIPP} = (a/b) \times 100\% \quad (1)$$

where ‘a’ represents the quantity of extracted pectin in grams, and ‘b’ is the original amount of crushed AIP powder (15 g). All experiments were conducted in triplicate to obtain the mean value, and the optimised conditions were determined by selecting the highest yield from each variable.

FINDINGS AND DISCUSSION

Prior to extraction, the moisture content of the AIP sample was determined to ensure that it was less than 12 % to limit the water content before being placed in a microwave [14]. For every parameter varied, the other parameters were held constant, as listed in Table 1.

Table 1: Parameters that held constant for every parameter varied

Parameters varied	Parameters constant
Solid-liquid ratio (1:10-1:50 g/mL)	3 %DES, 70 W, 3 min
Microwave power (90-50 W)	3 %DES, 1:30 g/mL, 3 min
Extraction time (1-5 min)	3 %DES, 70 W, 1:30 g/mL
Percentage of DES (1-5 %v/v)	1:30 g/mL, 70 W, 3 min

Effect of solid-to-liquid ratio

The amount of AIP powder required to dissolve the diluted DES was also determined. As shown in Figure 1, the solid-to-liquid ratio increased from 1:10 to 1:50 g/mL, and the yield increased from 0.74 % to 44.33 %.

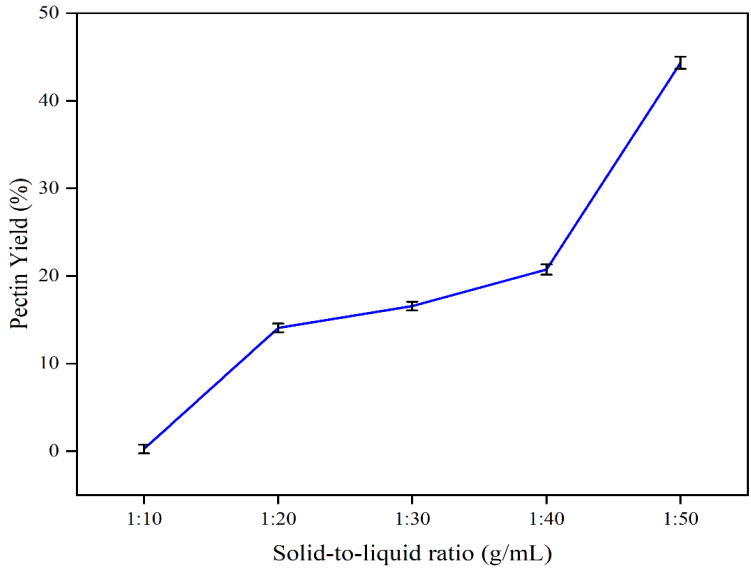

Figure 1: Effect of solid to liquid ratio in extracting AIPP with MAE

A higher solid-to-liquid ratio provides more contact between the solid matrix and solvent [15]. Thus, the dissolution of pectin in the DES and pectin yields were enhanced. According to Maran [16], the extraction solvent collects soluble pectin through mass transfer from the solid matrix to the liquid solvent. Thus, increasing the volume of the solvent led to a higher yield of pectin due to the larger volume of solvent, as it provides more space for the extraction process. According to Fick's law of diffusion, an increased solid-to-liquid ratio creates a concentration difference between the interior plant cells and the exterior solvent which drives the diffusion of solutes from areas of higher to lower concentrations [17]. During this process, it was observed that at a 1:10 g/mL ratio, the solvent was insufficient to extract the powder, resulting in a more concentrated extract. Thus, transferring the sample to a microwave was difficult, resulting in a low percentage and pectin quality. As the pectin extraction process proceeded, the pectin concentration in the extraction solvent gradually increased. Adetunji *et al.* [18] stated that this increase caused the solvent to become more viscous. Shafie *et al.* [13] reported a solid-to-liquid ratio of 1:30 g/mL for pectin extraction using ChCl-citric acid, with an optimum pectin yield of 24.68 %. However, the maximum pectin yield increased to 96 % with a solid-to-liquid ratio of 1:40 g/mL when extracted using ChCl:MA. This implies that the optimal solid-to-liquid ratio varied owing to the different effectiveness of the DES. The optimal solid-to-liquid ratio was from to 1:30-1:50 g/mL [19]. Therefore, selection of an appropriate solid-to-liquid ratio is critical for efficient extraction. In this study, 1:50 g/mL was selected as the optimised solid-to-liquid ratio because it yielded a higher percentage of pectin than other solid-to-liquid ratios.

Effect of microwave power

Microwave power is an important parameter in the pectin extraction process because it affects the efficiency and quality of pectin yield. As shown in Figure 2, the microwave power increased from 50 to 90 W and the pectin yield decreased from 17.26 % to 9.77 %, similar to the trends reported by Lefsih *et al.* [20] and Thirugnanasambandham *et al.* [21].The highest yield was observed at 50 W, whereas the lowest yield was obtained at 90 W. Based on these experiments, 50 W was chosen as the optimum microwave power to extract pectin from AIP. Xu *et al.* [22] reported that very high microwave power led to the degradation of pectin. In MAE, temperature control is

achieved by regulating the incident microwave power, which determines the amount of energy supplied to the matrix.

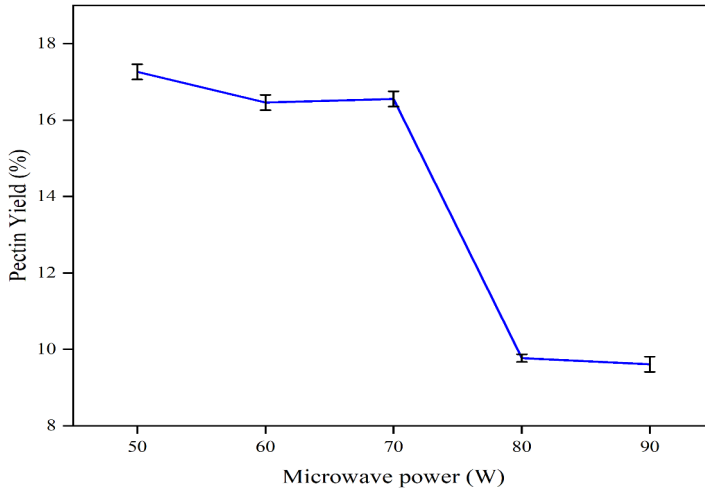

Figure 2: Effect of microwave power in extracting AIPP with MAE

This energy is converted into heat within the dielectric material. Thus, MAE delivers microwave energy directly to the sample, where the electromagnetic field interacts with pectin molecules, resulting in the conversion of electromagnetic energy into thermal energy [23]. Thus, increasing the microwave power increases the temperature, which significantly increases the pectin yield. This is primarily attributed to the faster and easier transfer of pectic polysaccharides from the AIP matrix. When the temperature is increased, it softens the plant tissues, which in turn improves the rate of diffusion [24]. Therefore, it is essential to select a solvent with a high extracting power that exhibits strong interactions with the matrix and the analyte being extracted.

In this study, ChCl:MA was used to extract AIPP (moisture content: ±0.05 %). The dielectric constant of a solvent typically influences its degree of microwave absorption. Choline chloride-based DES have been reported to possess higher dielectric constants in the range of 10-20 than common solvents such as hexane, toluene, and diethyl ether [25]. Water has a high dielectric constant, implying a low dissipation factor. This implies that the system absorbs more microwave energy than can dissipate, resulting in a

phenomenon known as " superheating" when water is present in the matrix [26]. Thus, selecting the optimal power range is crucial to avoid adverse effects on the pectin quality.

Effect of extraction time

The extraction time plays a critical role in pectin extraction, as it determines the duration of contact between the solid matrix of the AIP powder and the DES.

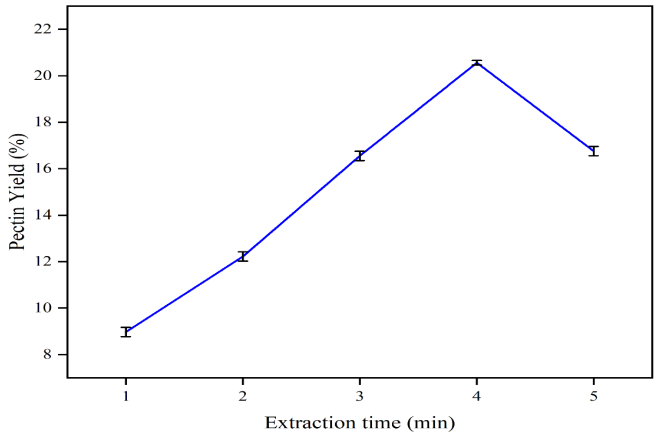

Figure 3: Effect of extraction time in extracting AIPP with MAE

As illustrated in Figure 3, the yield obtained was directly proportional to the extraction time up to 4 min, from 8.97 % to 20.57 %, and decreased slightly to 16.76 % at 5 min. Based on these findings, the optimal extraction time was determined to be 4 min. The decrease in yield at 5 min was due to pectin degradation caused by prolonged exposure, which provided more reaction time opportunities, as reported by Chua *et al.* [15] and Liew *et al.* [27]. Extending the duration provided sufficient time for DES to penetrate the AIP matrix and promote the hydrolysis of protopectin. This enhanced the mass transfer of soluble pectin from the matrix to the DES, resulting in a higher pectin yield [16, 28,29]. Mada *et al.* [30] reported that a sufficient extraction time allows for greater solubilisation and diffusion of pectin from the solid matrix into the extraction solvent. Several studies have investigated the effect of the extraction time on pectin extraction using MAE. Khamsucharit *et al.* [2] reported that pectin yield from apple

pomace increased up to 19.32 % at 20 min, while Fakayode and Abobi [31] found that pectin yield from orange peel increased with extraction time up to 15 min (22.2 %). Thus, the optimal extraction time balances extraction efficiency and pectin yield. An extraction time that is too short may result in incomplete extraction, while excessively long extraction times may lead to degradation or loss of pectin molecules.

Effect of percentage of DES

In this study, the DES was diluted in 100 mL of deionised water from 1 % to 5 %. The yield of pectin obtained differed owing to the polarity of the DES in solubilising pectin from the AIP powder solid matrix. The results showed that, as the percentage of DES increased, the yield increased, as illustrated in Figure 4.

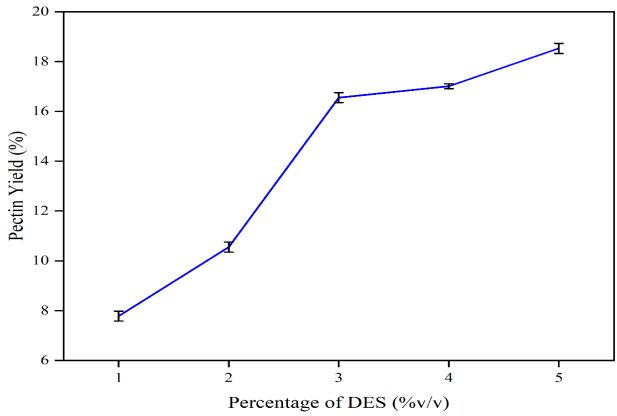

Figure 4: Effect of percentage of DES in extracting AIPP with MAE

The highest pectin yield was obtained with 5 % DES (18.53 %), whereas the lowest was obtained with 1 % DES (7.78 %). A higher percentage of DES enhanced pectin solubility. This leads to increased swelling of the AIP solid matrix, facilitating greater contact between DES and pectin. The optimum percentage of DES was determined to be 5 % because it produced the highest pectin yield among the others. Naturally, the prepared DES is quite viscous; therefore, another 95 % of water within the DES enables better extraction efficiency because the DES can easily move towards the sample matrix. This is because the presence of water in the DES

promotes an increase in the charge density at the micelle interface, leading to the formation of less elongated globular micelles and enhanced interactions between the DES and pectin molecules [32,33]. Therefore, the electrostatic attraction between the positively charged micelles and negatively charged pectin facilitates the extraction process with a better pectin yield. Duan *et al.* [34] mentioned that a higher percentage of DES facilitates faster mass transfer by enhancing diffusion during extraction. In contrast, excessive DES concentrations lead to phase separation, precipitation, and increased viscosity, impeding the pectin extraction efficiency [35]. Thus, optimisation of the DES percentage is crucial for maximising pectin yield.

Extraction optimisation

Based on the optimisation process, the optimal conditions were 1:50 g/mL, 50 W, 4 min, and 5 % ChCl: MA-based DES with an average pectin yield of 30.17 %. Few studies have utilised DES with MAE to extract pectin from AIP. Therefore, this study was compared with the study conducted by Shafie *et al.* [13]. They applied response surface methodology to optimise pectin extraction conditions based on molar ratios of DES of 1:1, 80°C, 2.5 h, and 3.74 % DES of ChCl:citric acid monohydrate, with a percentage yield of 14.14 %. This proved that MAE improved the pectin yield with a shorter extraction time. The high yield obtained in this study was related to the acidic conditions of malonic acid, which is the hydrogen bond donor in the prepared DES. This study was approved by Elgharbawy *et al.* [36], who reported that a ChCl-based carboxylic acid DES is the best choice for pectin extraction. In comparison with the study by Luo *et al.* [37], who extracted pectin using acetic acid, the obtained yield was 18.21 %. Notably, the pectin yield obtained using DES was as good as that obtained using the conventional method. Furthermore, the method used a small amount of DES and a shorter extraction time, indicating that it was more effective than other methods. There are no studies in the literature on pectin extracted from AIP using DES. Therefore, the obtained yield was compared with that reported by Islam *et al*. [38], who extracted pectin from the peel of *Artocarpus heterophyllus*, the closest relative of AIP, with yields of 29.40 % using hydrochloric acid. Thus, this study demonstrated that AIP could act as a novel potential source of pectin.

CONCLUSION

The MAE-DES combination demonstrated an effective extraction method for obtaining pectin from AIP. The effects of solid-to-liquid ratio, microwave power, and DES percentage were directly proportional to the obtained yield. Even so, the extraction time showed an increase in pectin yield up to 4 min and a decrease at 5 min. Thus, the optimal conditions were 1:50 g/mL, 4 min, 50 W, and 5 % DES, yielding a 30.17 % yield. This study is significant because it employs green principles to reduce the solvent consumption and waste production. This method also increased the pectin yield efficiency. Further research should focus on optimising the MAE-DES method to cover all possible factors such as pH and temperature. The use of response surface methodology is also recommended to predict pectin yield for comparison with experimental results.

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