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Microwave-assisted Extraction of Pectin from Dalanghita (*Citrus reticulata Blanco*) Peels

Rnie Shayne G. Gaurana[1,2*], Sean Alec Lois B. Sunga[1], Bernice Lyrem Joy S. Lim[1], Amelia B. Hizon-Fradejas[1], Sheryl Lozel B. Arreola[1]

[1] *Institute of Chemistry, University of the Philippines Los Baños, Laguna, Philippines 4031*
[2] *Department of Chemistry, University of the Philippines Visayas, Miagao, Iloilo, Philippines 5023*

*Author to whom correspondence should be addressed; email: rggaurana@up.edu.ph

ABSTRACT

Pectin is one of the important high value-added products today due to its wide applications as a functional ingredient in food science, cosmetics, and pharmacy. Because of the rising demand, pectin research has become increasingly relevant. Commercial pectins are mainly obtained from citrus peels. Unlike oranges, pectin extraction from dalanghita is not well established. In this study, microwave-assisted extraction was employed for the isolation of pectin from dalanghita (*Citrus reticulata Blanco*) peels. Using a Box-Behnken response surface methodology design, the effects of three process parameters (pH, irradiation time, and solid/liquid ratio) on the % yield of pectin were observed. The optimum conditions for a maximum pectin yield of 35.43% were pH=1.5, irradiation time=120s, and S/L ratio=30. The extracted pectin was pale yellowish (wet) to brownish (dry) in color, with a 31.75% actual yield, 4.94% methoxyl content, 171.67% anhydrouronic acid (AUA) content, and a 16.37% degree of esterification (DE). Given this data, it was concluded that dalanghita peels are a viable source of pure, low-ester, food-grade pectin that can be used for low-sugar applications. The microwave-assisted process is also a fast and accessible way of extracting pure pectin.

Keywords: *pectin; optimization; microwave-assisted; green extraction; Citrus; dalanghita*

INTRODUCTION

Agricultural-based industries generate large amounts of by-products every year, often discarded as waste. Fruit processing alone produces a lot of by-products, such as peels and seeds, which account for more than 50% of the fresh fruit. These by-products, however, have been an important source of high-quality products like pectin (Torres-León et al., 2018). Pectin is a complex polysaccharide (carbohydrate) found in the cell walls of almost all higher plants, which varies considerably in composition and structure. It is widely used in the cosmetics, food, and pharmaceutical industries as a gelling, thickening, and stabilizing agent (Belkheiri et al., 2021). It is also a good source of dietary fiber, which aids digestion and lowers cholesterol and blood sugar

levels (Mishra et al., 2012). Consequently, pectin is recognized as a safe additive that can be consumed daily with no limits if manufactured properly (WHO, 2017).

Commercially available pectin is produced mainly from citrus peels due to its high extraction yield (20-30%) and good gelling properties (Rolin, 1993). Although several other pectin sources have been identified (e.g., pomace apple, papaya, guava, pineapple, calamansi), some do not have a good yield or quality (Normah and Hasnah, 2000). The conventional method of producing pectin is quite expensive and time-consuming. Recently, greener technologies such as microwave-assisted extraction (MAE), supercritical water extraction (SWE), and ultrasound-assisted extraction (UAE) have emerged as potential alternatives to the conventional method (Marić et al., 2018). Among these techniques, MAE has been regarded as a practical and effective approach for pectin extraction due to its simplicity, short processing time, high extraction rate, and large handling capacity with moderate cost (Maran et al., 2015). Several studies have shown that MAE increases the yield and quality of pectin by allowing better penetration of extracting solvent into the plant tissues through microwave exposure (Chan et al., 2017; Sandarani, 2017).

Moreover, pectin extraction is influenced by various process parameters such as pH, temperature, extraction time, solid/liquid ratio, and the type of solvent used (May, 1990). It is recommended to develop an optimization method to obtain the optimal conditions for maximum pectin yield. The Design of Experiments (DoE) is a systematic and efficient method that allows analysts to investigate the link between various input variables (aka factors) and key output variables (aka responses) (JMP, n.d.). One of the most widely used experimental designs for optimization is the response surface methodology (RSM). RSM is an effective mathematical and statistical technique that deals with complex processes involving two or more variables that influence the response of interest. Unlike other optimization tools, RSM requires fewer experimental trials to analyze the effects of all the variables and reveal the best combination (Kanmani et al., 2014; Pasandide et al., 2018; Aydar, 2018).

According to statistics, the global pectin market is expected to grow at a compound annual growth rate (CAGR) of around 6.5% in 2021-2028 due to increasing consumer demand (Market Research, 2021). Currently, the Philippines imports all its pectin requirements from other countries due to a lack of large-scale pectin production facilities. Government data showed that the country imported around 93,150 kg of pectin in 2011, with a total customs value of US $ 52,383,487 or P2.2 billion (Caliguiran, 2014). Because of this, efforts have been made to address the gap. Two leading technologies have successfully produced local pectin that meets United States Pharmacopeia (USP) standards for pharmaceutical grade pectin. One is obtained from calamansi peels (10-14% recovery) but on a laboratory scale only (Torres et al., 2011); the other is derived from carabao mango peels (21.65% recovery) and is commercially viable (Gragasin et al., 2012). These initiatives established the idea that local pectin production could be a promising venture, given the country has a rich collection of fruits, many of which are underutilized.

Citrus reticulata Blanco (dalanghita) is one of the economically important fruits that is locally grown in the Philippines but is underutilized (Coronel 2010). Dalanghita is a citrus fruit smaller than an orange and has smooth, oblong leaves. Its fruits are high in Vitamin C, usually green, but can turn yellow, greenish-yellow, or orange over time. It is usually eaten as a fruit, and the peels are discarded as waste. Up to 50% of the fruit's weight can be attributed to its peel (Philippine Medicinal Plant, 2021). Considering that it belongs to the *Citrus* genus, dalanghita could be a potential source of commercial pectin. Unlike oranges, there have been no comprehensive studies on pectin extraction from dalanghita peels to date. The main objective of this study is to design a microwave-assisted extraction (MAE) procedure for pectin from dalanghita peels. Instead of using conventional mineral acids (which caused environmental and economic concerns), citric acid, a milder organic acid, was used as the acidifying agent. Studies showed that using citric acid with microwave heating produced high-quality pectin. This combination is less pectin-degrading and more effective at altering the physicochemical properties of pectin (Kurita, 2008; Li et al.,

2019; Belkheiri et al., 2021). Using the designed procedure, the effects of different process variables (pH, irradiation time, and solid/liquid ratio) on the response variable (pectin yield) were examined. The optimized process parameters for maximum extraction yield were obtained using the Box-Behnken response surface methodology (RSM) design. The isolated pectin was then characterized in terms of its color, pH, solubility in selected solvents, and chemical composition.

METHODS

Materials and Equipment. The materials and chemicals used in this study were provided by the Analytical and Environmental Chemistry Division, Institute of Chemistry, University of the Philippines, Los Baños. The chemicals used were purchased from J.T. Baker. All chemicals are analytical grade unless otherwise stated. All measurements were done in triplicates.

Sample Collection and Preparation. Fresh dalanghita fruit (10 kg) was procured from the local market in Brgy. Batong Malake, Los Baños, Laguna, Philippines. Fruit peels were manually separated from the pulp, washed, and then air-dried for seven days. The air-dried peels were oven-dried for four hours at 40°C before homogenization using a Wiley mill. The dry, homogenized peel sample (1.80 kg) was placed in resealable bags and then stored in the refrigerator before extraction.

Microwave-assisted Extraction. Pectin extraction was performed using a household microwave oven (3D The Smart Wave). About 1.0g of dalanghita peel powder was placed in a 250 mL beaker and added with acidified water of varying pH (1.0, 1.5, 2.5) for different solid/liquid ratios (10, 20, and 30 mL/g). Citric acid was used as an acidifying agent, and the pH of the solvent was adjusted with 1 M HNO_3. The mixture was placed in the middle of the microwave plate and exposed to different irradiation times (60, 120, and 180 s) at maximum microwave power (800W). After exposure to the microwave, the mixture was allowed to cool down to room temperature before being subjected to filtration using Whatman No. 1 filter paper. The insoluble residue was discarded, and the supernatant containing the pectin was collected and precipitated with an equal volume of ethanol. The recovered pectin was washed with 96% ethanol three times to remove impurities. The pectin was then filtered again, air-dried, oven-dried, and weighed. The pectin yield was calculated using the equation below.

$$\text{Pectin yield} = \frac{Mass\ of\ Dried\ Recovered\ Pectin}{Mass\ of\ Dried\ Dalanghita\ Peels}\ x\ 100\%$$

Design of Experiment and Process Optimization. The process parameters (pH, irradiation time, and solid/liquid (S/L) ratio) for the extraction of pectin from dalanghita peels were optimized using the response surface methodology (RSM). A 3-factorial design using the Box-Behnken model was applied to create a matrix design that would give the optimal conditions to get the maximum pectin yield. The range for the pH, irradiation time, and S/L ratio was originally set from 1.0 to 2.0, 60 s to 180 s, and 10 to 30, respectively. There were fifteen plot points in the Box-Behnken design, and each plot point was done in triplicates. These treatments were closely monitored and accurately measured during experimentation. From the actual data in the experiment, different response surface plots and contour plots were obtained. The model generated a total of 60 treatments. Some of the data points, which were considered outliers, were removed from the matrix design to achieve good linearity. A Pareto plot was also generated to observe the significance of each explanatory variable on the response. All statistical analyses were processed through JMP 11 Statistical Software (JMP, NC, USA). A prediction expression was generated from the experimental values to obtain the optimum conditions for maximum yield. The expression is as follows:

$$Pectin\ Yield = 8.936 - 1.760\left(\frac{X_1-1.5}{0.5}\right) - 6.568\left(\frac{X_2-120}{60}\right) + 16.076\left(\frac{X_3-20}{10}\right) + \left(\left(\frac{X_1-1.5}{0.5}\right)\left(\left(\frac{X_2-120}{60}\right) * -4.699\right)\right) + \left(\left(\frac{X_1-1.5}{0.5}\right)\left(\left(\frac{X_3-20}{10}\right) * 1.576\right)\right) + \left(\left(\frac{X_1-1.5}{0.5}\right)\left(\left(\frac{X_1-1.5}{0.5}\right) * 5.482\right)\right) + \left(\left(\frac{X_2-120}{60}\right)\left(\left(\frac{X_2-120}{60}\right) * 2.258\right)\right) + \left(\left(\frac{X_3-20}{10}\right) * \left(\left(\frac{X_3-20}{10}\right) * 10.419\right)\right) \quad (1)$$

where X_1 is pH, X_2 is irradiation time, and X_3 is S/L ratio.

The predictive ability of the model was assessed using linear regression analysis.

Characterization of Extracted Pectin. The structure of pectin from dried dalanghita peels was characterized using the IRSpirit Fourier Transform Infrared Spectrometer (Shimadzu, Philippines). The equivalent weight, methoxyl content, anhydrouronic acid content, and degree of esterification of the isolated pectin were determined through titration according to the method by Ismail et al. (2012) with some modifications. Its pH, color, and solubility in 0.1N HCl, 0.1N NaOH, ethanol, room temperature distilled H_2O, cold H_2O, hot H_2O, and DMSO were also observed.

Equivalent Weight. Equivalent weight was determined by weighing 0.5 g of the pectin sample and 1.0 g of sodium chloride in a 250 ml Erlenmeyer flask. Then, 5 mL of ethanol and 100 mL of carbon dioxide-free distilled water were added to the flask and shaken until the pectin was dissolved completely. About six drops of phenol red indicator were added and titrated with 0.1 N standardized NaOH until the color of the indicator changed to pink (pH 7.5) and persisted for at least 30 seconds. The neutralized solution was set aside for methoxyl content determination. The equivalent weight was calculated using the formula below:

$$\text{Equivalent weight} = \frac{weight\ of\ sample\ (g)\ x\ 1000}{volume\ of\ alkali\ (mL) x\ normality\ of\ NaOH}$$

Methoxyl content. About 25 mL of 0.25 N NaOH was added to the neutralized solution titrated for the equivalent weight, shaken thoroughly, and allowed to stand for 30 min at room temperature in a stoppered flask. Then, about 25 ml of 0.25 N HCl was added and titrated with 0.1 N standardized NaOH to the same endpoint as before. The equivalent weight was calculated using the formula below:

$$\%\text{MeO} = \frac{meq\ of\ alkali\ x\ 31\ x\ 100}{weight\ of\ sample\ (mg)}$$

where 31 is the molecular weight of the methoxyl group

Anhydrouronic acid content (AUA). The values of the equivalent weight and the methoxyl content were taken to calculate the anhydrouronic acid content using the formula below:

$$\%\text{AUA} = \frac{176\ x\ (meq\ of\ alkali\ for\ free\ acid + meq\ of\ alkali\ for\ methoyl)\ x\ 100}{weight\ of\ sample\ (mg)}$$

where 176 is the molecular weight of AUA

Degree of esterification (DE). The values of the methoxyl content and anhydrouronic acid content were taken to calculate the degree of esterification using the formula below:

$$\text{DE} = \frac{176\ x\ \%MeO\ x\ 100}{31\ x\ \%AUA}$$

RESULTS AND DISCUSSION

Optimization of Microwave-assisted Extraction of Pectin from Dalanghita Peels. Response Surface Methodology (RSM) was employed to optimize the process parameters for microwave-assisted extraction of pectin from dalanghita peels. Box-Behnken design is widely used in RSM optimization studies because it only requires three factors to run the experiment. It basically suggests which data points to choose from a three-level factorial combination, allowing for efficient estimation of the first order and second order coefficients of the mathematical model. The generated mathematical models are then used to create 3D surface plots that illustrate the optimal conditions (Kanmani et al., 2014; Hidayanti et al., 2016; Pasandide et al., 2018).

In this study, three independent variables (pH, irradiation time, and S/L ratio) and one response variable (% yield) were used in the Design of Experiment (DoE). The points used and their corresponding responses are summarized in Table 1. The predicted and actual values of %pectin yield were also included in the table. Then, the parameter estimates in Table 2 were used as plot points to generate the contour and response surface plots (Figure 1).

Table 1. Box-Behnken Experimental Design Matrix with Observed Pectin Yield.

Pattern	pH	Irradiation Time, s	S/L Ratio	% Yield, Actual	s	%Yield, Predicted
0	1.5	120	20	1.625935	1.108974	8.9360695537
−−0	1	60	20	16.33208	1.372915	20.304987626
0++	1.5	180	30	28.65358	0.8004	32.696897338
0	1.5	120	20	7.729351	1.719523	8.9360695537
+0+	2	120	30	30.1564	2.314596	30.085995228
0	2	180	20	7.6226	3.020772	3.6496907623
+−0	2	60	20	31.10671	4.249723	26.182586257
0+−	1.5	180	10	2.388346	0.200558	2.606182537
0	1.5	120	20	17.45292	6.408782	8.9360695537
−+0	1	180	20	11.64339	5.032145	16.567506693
+0−	2	120	10	7.100751	1.801332	16.06818871
−0−	1	120	10	1.384608	0.485546	1.455015999
−0+	1	120	30	60.70682	6.176799	51.739385239
0−−	1.5	60	10	17.72431	8.731419	13.680997672
0−+	1.5	60	30	37.68556	6.30836	42.680093555

Table 2. Generated Data Points for the Response Surface Plot

Term	Estimate	Std Error	T Ratio	Prob>|t|
Intercept	8.9360696	5.482823	1.63	0.1641
pH(1,2)[1]	-1.760054	3.35753	-0.52	0.6225
Irradiation time(60,180)[1]	-6.567594	3.35753	-1.96	0.1078
S/L ratio (10,30)[1]	16.075544	3.35753	4.79	0.0049
pH*Irradiation time[2]	-4.698854	4.748264	-0.99	0.3678
pH*S/L Ratio[2]	-9.066641	4.748264	-1.91	0.1145
Irradiation time*S/L ratio[2]	1.575996	4.748264	0.33	0.7534
pH*pH[2]	5.482159	4.94215	1.11	0.3178
Irradiation time*Irradiation time[2]	2.2579642	4.94215	0.46	0.6669
S/L ratio*S/L ratio[2]	10.418918	4.94215	2.11	0.0888

Parameters were estimated at significance level $\alpha = 0.05$

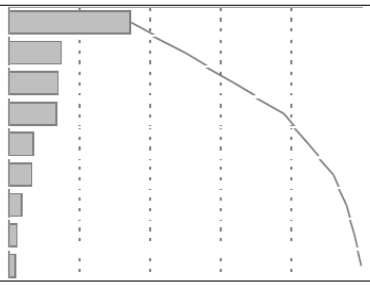
[1]First-order response surface model; [2]Second-order response surface model

The contour and response surface plots in Figure 1 represent the effects of each process parameter on pectin yield. In the contour plots (A-C), the areas with the highest pectin yields are colored red. Based on (A), the maximum yield is between 25 to 30 mL/g S/L ratio and pH 1 to 1.6. Based on (B), the maximum yield can be obtained with a S/L ratio of about 25 to 30 mL/g and an irradiation time of about 60 to 125 s. Based on (C), the maximum yield is at 60 to 125 s irradiation time and pH 1 to 2. On the other hand, the curved surface in green color of the 3D response surface plots corresponds to the statistically significant quadratic terms of the model. As seen in the figures (D-F), the curvature is going in the direction of the S/L ratio relative to the yield. This implies that the S/L ratio had the most significant impact on pectin yield. All the other interactions had a non-significant effect.

To better visualize the observations, a Pareto plot was constructed using the converted estimates from the response surface plots, as shown in Table 3. Each process parameter was ranked according to its significance to pectin yield - S/L ratio ranked first, followed by the irradiation period, and lastly is the pH. To obtain the optimum extraction conditions, a prediction expression (Eq. 1) was generated from the actual yield data. The predictive ability of the expression was evaluated using linear regression (Figure 2, Table 4), residual fit (Figure 3), ANOVA (Table 5), and lack of fit (Table 6). The predictive ability of the model was assessed to determine how well the model fits the data and whether it can be used to predict the optimum extraction conditions (Table 7).

Table 3. Pareto Plot of the Transformed Estimates from Response Surface Plot

Term	Orthog Estimate
S/L Ratio (10,30)	11.73992
S/L Ratio*S/L Ratio	5.16923
Irradiation Time (60,180)	-4.79629
pH*S/L Ratio	-4.68199
pH*Irradiation Time	-2.42648
pH*pH	2.28324
pH (1,2)	-1.28536
Irradiation Time*S/L Ratio	0.81384
Irradiation Time*Irradiation Time	0.72478



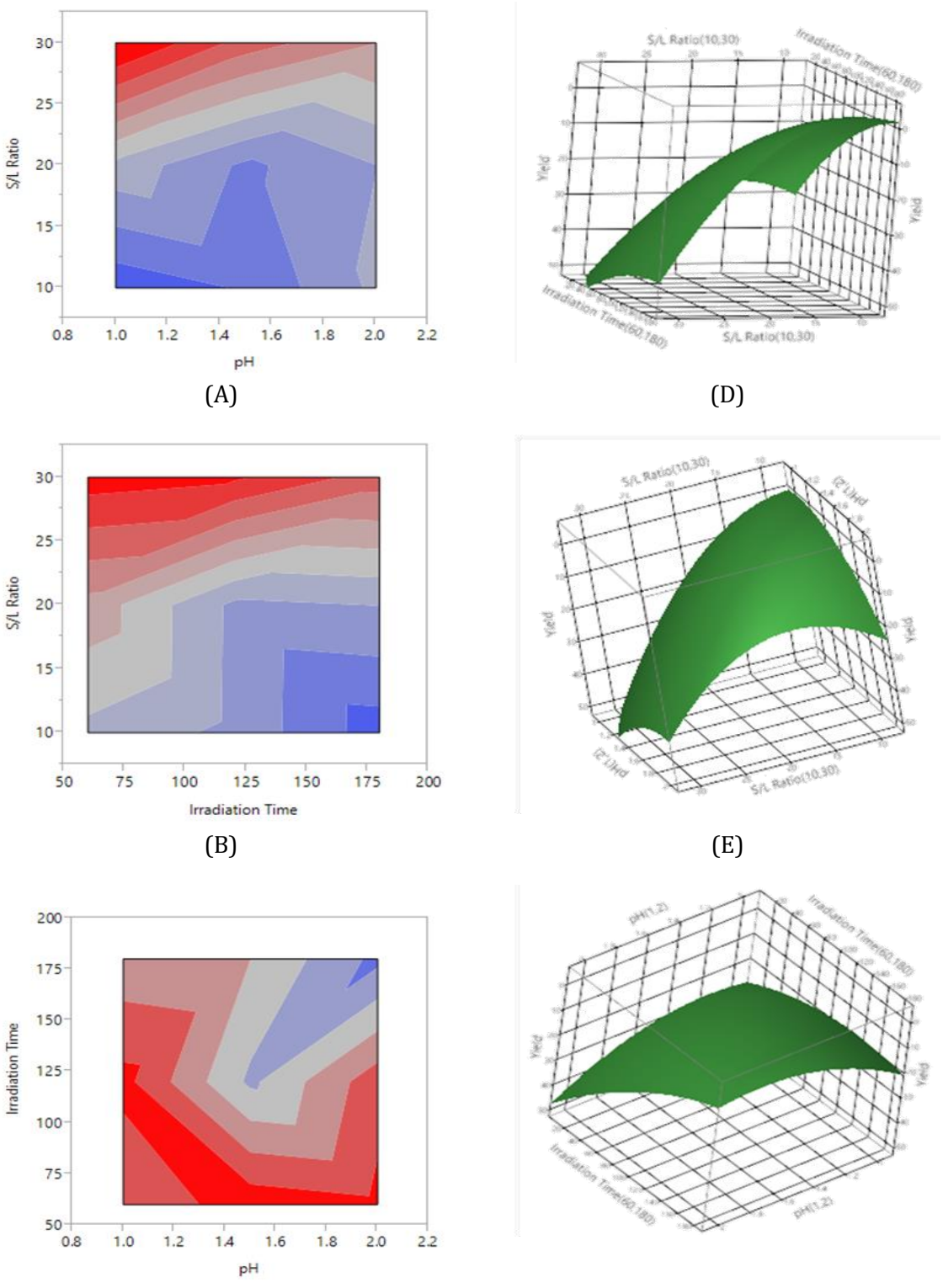


Figure 1. 2D Contour (A-C) and 3D Response surface (D-E) plots of the microwave-assisted extraction of pectin from dalanghita peels: (A,D) S/L ratio vs irradiation time, (B,E) S/L ratio vs pH, and (C,F) irradiation time vs pH.

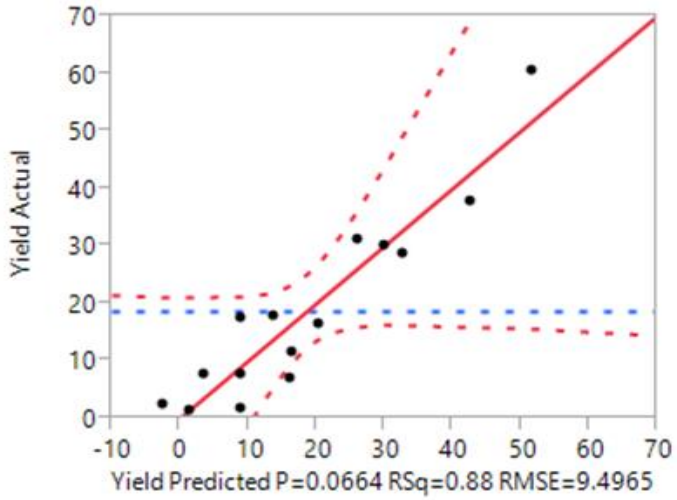


Figure 2. Model fit. Actual % yield of pectin vs the calculated % yield from the prediction expression.

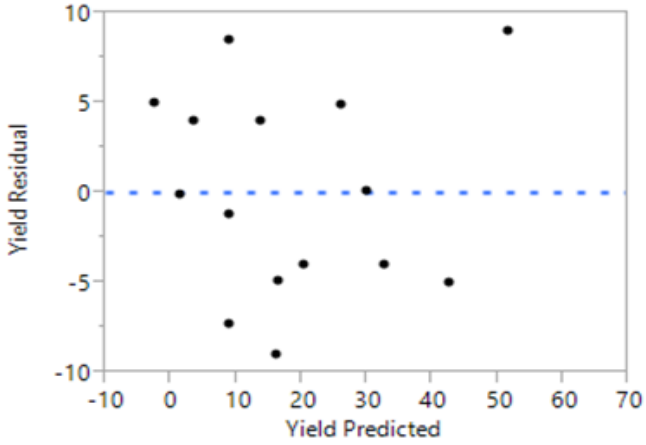


Figure 3. Residual fit. Residual of the actual % yield of pectin vs the predicted yield.

Table 4. Linear Regression Parameters of the Model Fit.

Parameter	Value
R^2	0.881403
R^2 Adj	0.667928
Root Mean Square Error	9.496528
Mean of Response	18.62089
Observations (or Sum Wgts)	15

Table 5. Analysis of Variance (ANOVA) of the Model Fit (α = 0.05).

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	9	3351.1931	372.355	4.1288
Error	5	450.9202	90.184	**Prob > F**
C. Total	14	3802.1134		0.0664

Table 6. Lack of Fit (α = 0.05).

Source	DF	Sum of Squares	Mean Square	F Ratio
Lack Of Fit	3	323.48920	107.830	1.6924
Pure Error	2	127.43101	63.716	**Prob > F**
Total Error	5	450.92022		0.3924
				Max RSq
				0.9665

Table 7. Replicability of Model

pH	Irradiation Time, s	S/L ratio	Predicted % Yield	Actual % Yield	% Error
1.5	120	30	35.43053	31.74741±0.054867	10.39533

Figure 2 shows the actual %yield versus the %predicted yield plot of the model, with an R^2 of 0.8814 (Table 4). The resulting R^2 is close enough to 1, implying that the model has fairly good predictive power and has good linearity. Ideally, the R^2 value should be no less than 0.8 to fit a regression model. Furthermore, the model has a prob>F value (0.0664) greater than α = 0.05 in the ANOVA test (Table 5), indicating that there is no statistically significant difference between the actual and predicted yields.

In addition to the R^2, a residual plot was also created. As shown in Figure 3, the points are in random order. This means that the model fit is indeed linear. R^2 is only one indicator of how well the model fits the data. Even if a model has a high R^2, this does not guarantee better predictive ability. A residual plot is necessary to verify that the model is adequate and meets the assumptions of the analysis (Minitab, n.d.).

In the Lack of Fit test (Table 6), the prob>F value of 0.3924 is greater than α = 0.05. Thus, the null hypothesis that the model error mean square is equal to the true error was accepted. In optimization studies, a non-significant p-value indicates that the model fits the data well. Overall, the results revealed that process parameters used in the study have a significant impact on pectin yield. The optimum conditions based on the model are pH 1.5, irradiation time of 120 s, and S/L ratio of 30. Under these conditions, the expected yield is 35.43%. Through a validation experiment, it was calculated that the model has a 10.40% error (Table 7).

It can be observed that when the S/L ratio is less than 30, it leads to low pectin yields. Greater S/L ratio promotes swelling of the dalanghita peel solids. Because the surface area of each particle is increased, there is more contact between the microwaves and the material. The microwaves cause the material's cell walls to rupture, releasing pectin into the surrounding medium. There is a high risk of saturating the solvent at S/L ratios lower than 30 mL/g. When the solvent is saturated, the remaining pectin in the sample is less likely to be released into the solvent, lowering the percent yield. When the solvent becomes supersaturated, pectin precipitates out of the solution. This precipitated pectin will thicken with the residues and will not be transferred to the filtrate during filtration. This will also decrease the observed yield.

Furthermore, longer irradiation time and low pH were found to be favorable for a high pectin yield. The optimum irradiation time is set at 120 s. There is a high likelihood that the solids are underexposed to microwaves at irradiation times less than 120 s. Less exposure would decrease the yield since fewer cell walls are ruptured. On the other hand, overexposure to microwaves at irradiation time > 120 s would most likely cause degradation of pectin. At pH < 1.5, pectin should be soluble enough due to the acidic nature of its repeating units. However, at very low pH, there

is the risk of pectin hydrolysis, which would negatively affect experiment yield. At pH > 1.5, pectins are more likely to aggregate and precipitate out of the solution even before filtration.

Characterization of Extracted Pectin from Dalanghita Peels. Pectin is a complex polysaccharide consisting mainly of straight-chain α-D-galacturonic acid residues linked by (1→4) glycosidic bonds. Its composition and structure depend highly on the source and the conditions applied during extraction (Belkheiri et al., 2021). These countless possibilities make pectin research a promising area for further studies.

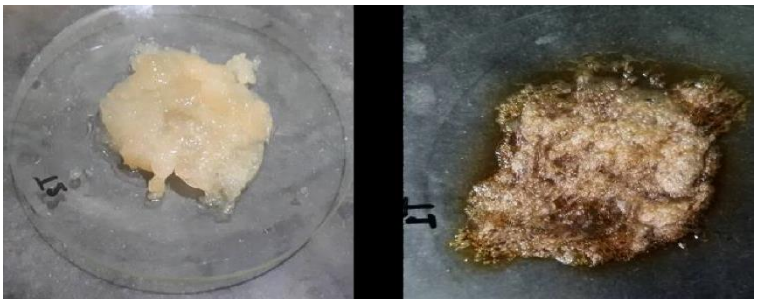


Figure 4. Physical appearance of wet dalanghita pectin (left) and dried dalanghita pectin (right).

Table 8. Physicochemical Properties of Pectin Extracted from Dalanghita Peels

Property	**Observation**
Color	Light yellow (wet); Dark brown (dry)
pH	2.67
Solubility	Soluble: HCl, NaOH, hot water Partially soluble: room temp water, DMSO Insoluble: ethanol, cold water
Chemical composition:	
Equivalent weight (EW)	122.95 ± 8.36
Methoxyl content (MeO), %	4.94 ± 0.68
Anhydrouronic acid (AUA), %	171.67 ± 9.62
Degree of esterification (DE), %	16.37 ± 2.37

The physicochemical properties of pectin from dalanghita peels are summarized in Table 8. The extracted pectin was light yellow in color when wet, and dark brown in color when completely oven-dried (Figure 4). It was soluble in acid, base, and hot water. It was partially soluble in room temperature water and DMSO, and was completely insoluble in ethanol and cold water. The resulting solutions were observed to have thickened upon the dissolution of pectin. The pH of the pectin is 2.67.

The methoxyl content of the pectin extracted from dalanghita peels was calculated to be 4.94%, while the degree of esterification was 16.37%. Generally, pectins are classified according to their methoxyl content and degree of esterification (DE). Pectins with a DE greater than 50% are called high ester pectins (or high methoxyl pectins), while those with a DE of less than 50% are called low ester pectins (or low methoxyl pectins). DE is defined as the percentage of esterified carboxyl groups in the pectin structure, while methoxyl content is defined as the mol quantity of methyl alcohol in 100 mL of galacturonic acid. Both parameters are used to describe pectin's gelling properties. The lower their values, the less capable they are of forming gels. DE and methoxyl content can be calculated by determining the equivalent weight of pectin using standard titration with sodium hydroxide. Equivalent weight is the total free galacturonic acid (not esterified) in the structure of pectin (May 1990; Ismail et al. 2012).

According to USP specifications, pharmaceutical-grade pectin has a methoxyl content of not less than 6.7% (Gragasin et al., 2012). A methoxyl content lower than the specified value is only suitable for food. Based on the results, dalanghita could be a good source of food-grade pectin. Low-ester pectins are widely used in the food industry, especially in low-calorie foods, because of their low sugar content and good stability.

The % anhydrouronic acid (AUA), on the other hand, is a good indication of the purity of extracted pectin. It is suggested that the % AUA should not be less than 65% (Ismail et al. 2012). In this case, the extracted pectin had a % AUA of 171.67, which means that the extracted pectin is very pure.

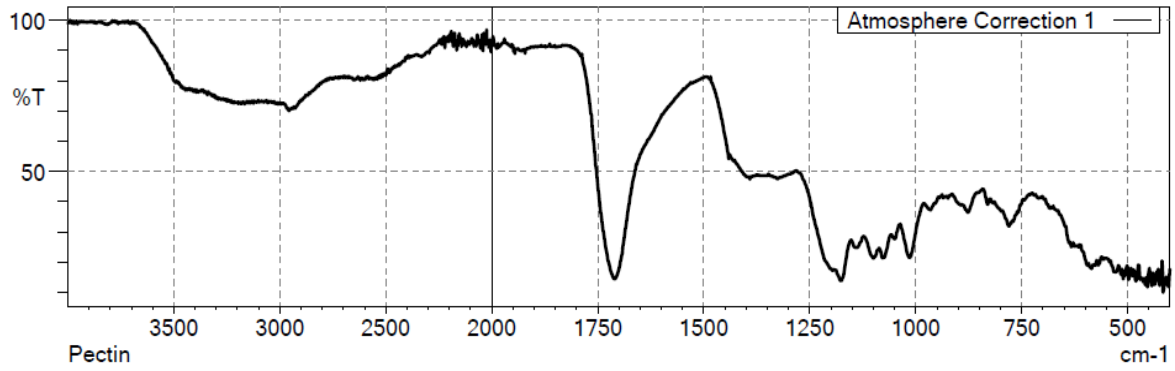


Figure 4. IR spectrum of the pectin extracted from dalanghita peels.

Table 9. Peak Assignments for the IR Spectra of Pectin.

Frequency (cm^{-1})	Type of Vibration
3,371.79	OH stretch, intermolecular H-bridge between the OH groups
2,948.72	CH stretch
1,673.08	C=O stretch
1,397.43	CH_2 scissors vibration
1,333.33	CH deformation
1,179.49	C-O-C antisymmetric stretch
1,019.23	C-O stretch
871.79	C-anomeric group stretch; C_1-H deformation; ring stretch
782.05	CH_2 rocking

FTIR analysis of the extracted pectin from dalanghita peels show prominent peaks at 3,371.79 cm^{-1} (OH stretch), 1,179.49 cm^{-1} (C-O-C antisymmetric stretch), 1,019.23 cm^{-1} (C-O stretch), and 871.79 cm^{-1} (C-anomeric group stretch), indicating the presence of carbohydrates in the structure. The sharp peak at 1,673.08 cm^{-1} (C=O stretch), along with the OH stretch at 3,371.79 cm^{-1} is indicative of the presence of a carboxylic acid in the structure. This can be from the D-galacturonic acid units. The peaks at 2,948.72 cm^{-1} (CH stretch), 1,397.43 cm^{-1} (CH_2 scissors vibration), 1,333.33 cm^{-1} (CH deformation), and 782.05 cm^{-1} (CH_2 rocking) are indicative of the presence of methoxylated D-galacturonic acid units in the extracted pectin.

According to Ismail et al. (2012), the major functional groups in pectin are usually in the region between 1,000 and 2,000 cm^{-1} of the FTIR spectra. An increase in DE values will also increase the intensities and band areas of the esterified carboxyl groups. This could be used to compare the different types of pectin (Ismail et al. 2012).

CONCLUSIONS

The Box-Behnken response surface design was used to develop an optimized microwave-assisted extraction method for pectin from dalanghita (*Citrus reticulata* Blanco) peels. Three process variables were examined: pH, irradiation time, and solid/liquid ratio. From the statistical model (R^2 = 0.8814), the optimum conditions were determined to be at pH 1.5, irradiation time of 120 s, and S/L ratio of 30 mL/g, with a pectin yield of 35.43% (10.40% error). The extracted pectin is a low ester pectin, with a MeO content of 4.94% and 16.37% DE. Low ester pectins are good additives for low-sugar food products. The pectin is also found to be pure, with a % AUA of 171.67.

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