Production of High Tannin Content and Antioxidant Activity Extract from an Unripe Peel of *Musa acuminata* (Cavendish) Using Ultrasound-Assisted Extraction (UAE)

Nur Arnisah Ishak,[a] Nor Asma Ab. Razak,[b,*] Mohd Sabri Pak Dek,[c] and Azhari Samsu Baharuddin [a]

Musa acuminata (Cavendish) unripe peel is a waste product of limited value that is generated in large quantities. Therefore, the conversion of this by-product into a more useful product is necessary. This study aimed to optimize the ultrasound-assisted extraction (UAE) parameters, including extraction temperature, extraction time, preincubation time, and solid to solvent concentration from an unripe banana peel using response surface methodology (RSM). The UAE parameters affected the recovery of yield, total tannin content, and flavonoid content with antioxidant activities. The optimum extraction temperature was 60 °C with an optimum extraction time of 30.0 min. Additional optimum conditions included 25.0 min for the preincubation time and 5.03% solid to solvent concentration. The optimum yield processing parameter of crude extract of unripe peel was 14.9% and the total tannin content was 119.2 mg TAE per g of the sample. Furthermore, the content of flavonoid was 29.0 mg RE per g of the sample and the DPPH and ABTS scavenging activity was 80.8% and 84.7%, respectively. The results from this study can be used for further isolation and purification of tannin from unripe banana peel. Further explorations could lead to the possible application of bio-based polymer in packaging materials.

.

Keywords: Tannin; Ultrasound-assisted extraction; Response surface methodology; Optimization; Musa acuminata; Unripe peel; Antioxidant; Radical scavenging

*Contact information: a: Department of Process and Food Engineering, Faculty of Engineering, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia; b: Laboratory of Molecular Biomedicine, Institute of Bioscience, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia; c: Department of Food Science, Faculty of Food Science and Technology, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia; *Corresponding author*: norasmarazak@upm.edu.my*

INTRODUCTION

The natural phenol polymer consists of various constituents, including tannin and lignin, which are widely present in nature. Plant polyphenols have been extensively studied due to their growing commercial value in nutrition, cosmetics, and the packaging industry. These polyphenols are associated with different biological activities such as antimicrobial, antioxidant, and anti-inflammatory properties (Song *et al*. 2013). They also include flavonoids such as flavonols, flavones, and tannin that are present in edible and non-edible plants such as vegetables and fruits (Manach *et al.* 2004). On many occasions, these polyphenols are used as antioxidants. The advantages of using natural sources in comparison to synthetic antioxidants are limitless. For example, a natural antioxidant candidate is safer than synthetic antioxidants for human consumption. One of the promising

natural antioxidants that can be manipulated in food packaging is tannin, which is obtainable from pine, oak, and chestnut trees. Tannin is the second most abundant phenolic resource in nature after lignin. It has been reported as a potent source in the manufacturing of leather in tanning processing, wine production, and wood adhesives. It is also utilized in the manufacturing of anti-corrosive primers that drive market growth. Functioning as an aromatic building block for green chemistry in bio-based polymers, tannin has the potential to be a renewable aromatic resource in the future. Besides that, tannin-biobased polymers have high antioxidant stability when incorporated in nano-fibrillated cellulose films for active packaging (Missio *et al*. 2019). The recovery of tannin from a plant source is the first step in its application. One of the cheapest sources of tannin is from unripe banana peel. The extract containing tannin from the unripe banana peel is considered as an alternative potential local source for further applications. The banana peel is considered a waste of low value in the annual production of banana (Vu *et al*. 2016).

Banana (*Musa* spp.) is one of the most important commercial crops in tropical areas, especially Malaysia. It is cultivated with 128 million tonnes in 150 countries for annual production (Singh 2011). The peel is the main by-product of the banana, which accounts for approximately 38% of the whole fruit weight (Gonzalez-Montelongo *et al.* 2010). In Malaysia, bananas are grown for domestic and export markets, and it has been listed as one of 15 fruits prioritized for commercial cultivation (Ministry of Agriculture 1998). Moreover, Malaysia is among the highest producer with a total export of about 25 thousand tonnes at the USD price of 0.87 per kg (Food and Agriculture Organization [FOA] 2018). *Musa acuminata* is one of the most abundant cultivars in Malaysia and the most popular for international trade and one of the highly exported cultivars is the Cavendish cultivar. The primary characteristic of this cultivar is that it remains green during the ripening process. Therefore, for export purpose, it must be sprayed with ethylene gas during the de-greening process for it to become yellow upon maturation to enhance its market value. The high-quality grade of Cavendish banana makes it appropriate for export as they possess longer shelf-life compared to other cultivars. During the grading process for export, the fruits categorized low-quality grade based on the criteria of size and color will be rejected at the rate 8 to 20%. Thus, the accumulation of banana by-products, particularly in the banana plantation, can be a potential issue. To overcome this problem, the unripe Cavendish banana peel was chosen for the extraction of tannin and antioxidant for biopolymer application in future.

The banana peel has a higher antioxidant activity than the pulp (Sulaiman *et al.* 2011) with more than 40 individual compounds of phenolics identified. Various factors that affect phenolic composition such as variety, maturity, cultivation condition, and pre-treatment processes. Green banana peel has been reported to help in reducing stools for people with diarrheal problems (Rabbani *et al.* 2001). The astringency of tannin in the unripe fruit is reduced during the ripening process. The changes in astringency are reflected in the changes in tannin's molecular size (Goldstein and Swain 1963). The interaction between polyphenols, salivary proteins, and glycoproteins in the mouth will produce an astringent sensation (Goldstein and Swain 1963). Besides, the banana peel contains flavan-3-ols, which is the largest group of phenolics consisting of monomers, dimers, and polymers (tannin). The compound is associated with potent antioxidant capacity (Rebello *et al.* 2014). However, studies regarding the improvement of tannin extraction efficiency from the unripe banana peel for further utilization are scarce.

Extraction is the process used to separate a bioactive compound from its sources in

the plant matrix, which produces concentrated bioactive compound. Water is considered an effective 'green' solvent in extracting the phenolic compound from the banana peel (Someya *et al.* 2002). However, water itself is not sufficient in maximizing the extraction of the bioactive compound. A nonconventional method such as the ultrasound-assisted extraction (UAE) consumes less time for extraction with high efficiency, and potentially be scaled for industrial use compared to the conventional and microwave-assisted extraction methods (Barrera Vazquez *et al.* 2014). Thus, the UAE technique was proposed in this study to extract tannin with antioxidant activity from the unripe banana peel.

The response surface methodology (RSM) has been applied as a powerful tool for optimal extraction conditions and functional compounds of plant material. It is a collection of mathematical and statistical techniques based on the fit of a polynomial equation to the experimental data and describes well the behavior of a data set (Bezerra *et al*. 2008). It is also useful to evaluate the effects of multiple variables as well as their interactions (Bas and Boyaci 2007). RSM is more effective than an orthogonal test design or the single variable method because it requires a reduced number of experimental trials to evaluate the effect of multiple factors and their interactions, as well as to optimize a process (Lee *et al*. 2000). Central composite design (CCD) is one of the types of an RSM design method that was employed in this study. Several factors affect the efficiency of the UAE process and to understand the interaction of each parameter, use of the CCD was proposed.

Therefore, the objective of this study was to investigate the UAE optimal conditions using water for the recovery of tannin compounds with antioxidant activity. The parameters assessed involved the effects of extraction temperature (X_1), extraction time (X_2), preincubation time (X_3), solid to solvent concentration (X_4), and their interactions towards the model response. An optimal condition process through Response Surface Methodology (RSM) could contribute to the unique understanding of the extraction of tannin with antioxidant activity from unripe banana peel for use in packaging and incorporated as adhesive wood through bio-based polymer technology.

EXPERIMENTAL

Materials

The unripe *Musa acuminata* (Cavendish) were collected from Ladang Puchong UPM Serdang approximately 3 months after the plant's flower bloom. The peel of the fruit was removed, washed carefully, and dried by using an oven at 40 °C for 2 days. After the plant materials were dried, the peel portion was ground into powder using a blender and sieve (50 µm size). The sample was packaged, kept in a secure place, and stored in the refrigerator at 4 °C, which prevented contamination during the experimental period.

Chemicals

Folin-Ciocaltue's phenol reagent, sodium carbonate, methanol, ethanol, and acetone were purchased from R & M Chemicals (R & M Marketing, London, UK). The 2,2-diphenyl-1-picryhydrazyl (DPPH) radical, 2,2-azino-bis (3-ethylbenzothiazoline)-6-sulforic acid (ABTS), potassium persulfate, aluminium chloride, tannic acid, rutin, trolox, and gallic acid were purchased from Sigma-Aldrich (St. Louis, MO, USA). All the chemicals used throughout the experiment were analytical grade and stock solutions were prepared using purified deionized water.

Methods

Selection of variables

A preliminary study involving UAE and conventional extraction method were conducted to determine the recovery of tannin and antioxidant content. The effect of different variables such as extraction temperature, extraction time, preincubation time of extraction, solid to solvent concentration, and pulse intensity of the ultrasound are known to affect the extraction yield and polyphenol contents. Therefore, to select the method that yields high total tannin content, a conventional extraction (decoction) procedure was carried out, by 1.0 g of the sample was weighed and mixed with 40 mL of deionized water. It was then placed in of a circulating water bath (Protech, Tech-Lab Manufacturing Sdn. Bhd, Selangor, Malaysia) at 60 °C for 1 h with 960 W, 50 Hz 830-S1. For the UAE, the parameters were the same with the addition of 10% pulse intensity using 770 W, 60 Hz WUC-D10H of an Ultrasonic Cleaner Set (Daihan Scientific Co., Ltd, Seoul, South Korea). The resulting extract was oven dried until a constant weight was reached. The different levels of pulse intensity (low, medium, and high) were examined with a temperature of 30 °C, an extraction time of 30.0 min, and 2.5% of solid to solvent concentration were used. The selection of the best pulse intensity was based on the maximum value of the total tannin content of the water extract.

Determination of yield percentage

The amount of yield percentage in the crude extract for each extraction condition was determined based on the weight difference. A volume of the crude extract was dried to constant weight using an oven drying setting (Memmert, Germany) of 40 °C. The extractable crude extract was calculated according to Eq. 1,

$$\text{Yield percentage} = (\text{the dried extract (g)} \div \text{sample used (g)}) \times 100 \quad (1)$$

Total tannin content analysis

The total tannin content in the extracts was measured in terms of tannic acid equivalents, according to a modified Folin-Ciocaltue's colorimetric method by Haile and Kang (2019). Briefly, an extract sample of 1.0 mL was mixed properly with 7.5 mL of deionized water. Next, 0.5 mL Folin-Ciocalteu's reagent (50% volume per volume) and 1.0 mL saturated Na_2CO_3 (35% mass per volume) was added to this mixture. The mixtures were vortexed and kept in the dark for 30.0 min. The absorbance was measured at 760 nm using a spectrophotometer (Synergy H1, BioTek, Vermont, USA). A standard curve was prepared by using tannic acid. The result was expressed as a tannic acid equivalent in mg per g of the sample.

Total flavonoid content

The total flavonoid content was determined by the aluminium chloride colorimetric as described by Yusri *et al*. (2012) with some modification. Briefly, 100 µL of the sample extract with 1000 ppm was added with 100 µL of 10% (weight per volume) aluminium chloride. This mixture was incubated at room temperature for 10.0 min. The absorbance was measured at 435 nm by using ELISA reader spectrophotometry (Synergy H1, BioTek, Vermont, USA). Quantification of the flavonoid content was done based on the standard curve of rutin prepared in methanol. The results were expressed as mg rutin equivalent per g sample (mg RE/g sample) of the plant extract.

DPPH radical scavenging ability assay

The antioxidant activity was determined using a DPPH assay and was done according to an adapted method Chan *et al.* (2002). A volume of 50 µL extracts and a standard of Trolox solution was mixed with 195 µL of a methanolic solution of DPPH. The mixture was vortexed and incubated at room temperature in the dark for 1 h. The absorbance was measured at 540 nm using an ELISA plate reader spectrophotometer. The Trolox solution was used as the standard. The experiment was performed in triplicate and the percent inhibition was calculated from the control using Eq. 2,

$$\text{Scavenging activity (\%)} = \frac{\text{(Absorbance Control - Absorbance sample)}}{\text{Absorbance control}} \times 100 \quad (2)$$

ABTS radical cation inhibition antioxidant assay

The ABTS radical cation inhibition antioxidant assay was performed according to Vuong *et al*. (2013) with some modification. The 2-azino-bis(3-ethylbenzothiazoline)-6-sulforic acid (ABTS) was prepared by mixing 13.24 mg of potassium persulfate with 76.8 mg of ABTS until it was dissolved. This mixture was kept in the dark for 16 h. This solution was diluted with deionized water until an absorbance of 0.7 ± 0.005 was obtained. A total of 216 µL of this solution was added with 24 µL of the sample and Trolox as the standard. The mixture was kept in the dark for 1 h before taking an absorbance measurement at 735 nm. The experiment was performed in triplicate and the percent inhibition was calculated from the control using Eq. 2.

Experimental design for the ultrasound-assisted extraction (UAE)

The optimization of ultrasound-assisted extraction (UAE) was carried out using a 770 W, 60 Hz WUC-D10H Ultrasonic Cleaner Set (Daihan Scientific Co., Ltd. Seoul, South Korea). The pulse intensity controls the processor, which allows the ultrasonic vibrations to be set at any desired level between the range of 10% to 100%. A sample of 1.0 g of unripe *Musa acuminata* (Cavendish) banana peel powder was mixed with 7.9 to 48.8 mL of deionized water at room temperature for 2.5 min to 32.5 min before the extraction process was performed. The mixture was then kept in the ultrasonic water bath cleaner at various temperatures (15 °C to 75 °C) and time (1.5 min to 39.5 min) with a constant 10% pulse intensity of ultrasound power during the extraction process. The extraction temperature, time, preincubation time treatment and percentage of solid to solvent concentration were based on the experimental design generated by the Design Expert Software Version 6.0.4 (State Ease, Inc.). The extract was filtered using Whatman No.1 filter paper and stored at -4 °C for the next experimental analysis. The optimization experiment was carried out using the RSM for the extraction of tannin content and antioxidant activity in the unripe *Musa acuminata* (Cavendish) peel using water as the solvent. A Central Composite Design (CCD) was used to investigate the effect of four variables consisting of 30 experimental runs where 6 central points were carried out. The variables involved were extraction temperature (X_1; 15 °C to 75 °C), extraction time (X_2; 1.5 min to 39.5 min), preincubation time (X_3; 2.5 min to 32.5 min) and solid to solvent concentration (X_4; 2.05% to 12.65%) with 1.41421 alpha. Each variable coded at its five levels (-2, -1, 0, 1, 2) and were represented by the lower, middle, and higher values. The coded and un-coded levels of independent variables used in the CCD design are listed in Table 1. The experimental data were fitted to the second-order polynomial model to obtain the regression coefficient.

Table 1. Independent Variables and Their Levels Used in the RSM Design

Symbols	Factors	Coded Levels				
		(-2)	(-1)	(0)	(+1)	(+2)
X_1	Extraction temperature (°C)	15.0	30.0	45.0	60.0	75.0
X_2	Extraction time (min)	1.5	11.0	20.5	30.0	39.5
X_3	Preincubation time (min)	2.5	10.0	17.5	25.0	32.5
X_4	Solid to solvent concentration (%)	2.05	4.70	7.35	10.00	12.65

Statistical analysis

The optimal conditions for extraction were determined using the RSM and performed using the Design Expert Version 6.0.4 program. The effects of four independent variables at five levels of dependent variables were investigated using CCD. The least-squares regression was used to fit the data to a quadratic model, which was calculated using Eq. 3,

$$Y = \beta_0 + \Sigma\beta_i X_i + \Sigma\beta_{ii} X_i^2 + \Sigma\Sigma\beta_{ij} X_i X_j \quad (3)$$

where Y is the predicted response, β_0 is a constant, β_i is the linear coefficient, β_{ii} is the quadratic coefficient, β_{ij} is the interaction coefficient of variables i and j, and Xi and Xj are independent variables.

Three-dimensional response surface plots were generated by keeping one response variable at its optimal level and plotting it against two factors (independent variables). The coded values of the experimental factors and factor levels used in the response surface analysis are given in Table 1. The complete design using aqueous extraction consisted of 30 experimental runs, including six replications of the center point and the corresponding Y1 to Y5 values. The optimum values of the selected variable were obtained by solving the regression equation and analyzed using the responses surface contour plots. The verification of the validity and adequacy of the predictive extraction model was realized in these optimum conditions of extraction temperature, extraction time, preincubation time, and solid to solvent concentration.

The individual linear, quadratic, and interaction regression coefficient were determined by an analysis of variance (ANOVA) using the Design Expert Version 6.0.4 (State Ease, Inc., Minnesota, United States). The coefficient of determination (R^2) and ANOVA were used for the fitness of the polynomial equation with the responses. The significance of all the terms of the polynomial equation was analyzed statistically by computing the F value at $p < 0.05$.

RESULTS AND DISCUSSION

Preliminary Experiments

The results of the preliminary experiments, showed that the percentage of yield and tannin content of crude extract with UAE are higher than the conventional method of extraction (Table 2). The improvement of tannin content in the crude extract of UAE might be due to the cavitation effects caused by the application of the ultrasound. The amplitude of an ultrasound wave travels through a mass medium and causes compression of water molecules, ultimately resulting in localized changes in the density and modulus. The abrupt

decrease in pressure at the edges of the sawtooth-shaped ultrasonic wave in the negative pressure cycle generates small bubbles. These bubbles collapse in a positive pressure cycle and produce turbulent flow conditions, which are associated with pressures and temperatures (Liu *et al*. 2014).

Table 2. Comparison of Extraction Methodology of Conventional and Ultrasound-Assisted Extraction

Method of Extraction	Yield (%)	Total Tannin Content (mg TAE/g sample)
Conventional extraction	15.60 ± 0.85	34.95 ± 1.91
Ultrasound-assisted extraction	16.92 ± 1.42	53.65 ± 7.64
*Note: Data are expressed as mean ± standard deviation		

In the present study, the level of pulse intensity (low, medium, or high) with different ultrasonic power showed no visible effect on the tannin content extracted from the unripe banana peel. The tannin content was 40.5 mg TAE per g of the sample when compared with the medium and high application of the pulse intensity, which were 31.7 mg TAE per g of the sample and 32.2 mg TAE per g of the sample, respectively. The results were obtained at the extraction temperature of 30 °C, extraction time of 5.0 min, and 2.5% of solid to solvent concentration. However, a low pulse intensity (10%) of ultrasonic power showed a slightly higher tannin content compared to the 50% and 100% power level. This finding was different from the study by He *et al*. (2016), which found that the yield of anthocyanin, ascorbic acid, and tannin from myrobalan nut increases with ultrasonic power. The contradicting observation may be due to the presence of hard cell walls of the myrobalan nut, which are not so permeable compared to the banana peel.

The application of low pulse intensity is beneficial in the industrial scale as it consumes little energy when used on as a proven by Pradal *et al*. (2016), who showed that lower energy consumption was observed when 50 W of ultrasound power was applied instead of 75 and 100 W. The total tannin content was higher when a low-intensity pulse (10%) was applied in the extraction process. Thus, the low pulse intensity was applied in all extraction processes using UAE in the present study.

To understand the effect of extraction temperature, time, preincubation time, and the solid to solvent concentration on the tannin content, flavonoid content, and also antioxidant activity, the variables were optimized using RSM. All variables were fixed at five levels for extraction temperature, extraction time, preincubation time of extraction, and concentration of solid to solvent as shown in Table 1. The results for all responses are displayed in Table 3. The pulse intensity of the UAE was kept constant at 10%.

Optimization of Ultrasound-Assisted Extraction (UAE)

The second-order polynomial equation was used in optimizing the extraction process. The model showed a highly significant effect on the percentage of yield, total tannin content, total flavonoid content, DPPH scavenging activity, and the ABTS scavenging activity data, as shown in Table 4. The fitness and adequacy of the model were judged by the coefficient of determination (R^2) and the significance of the lack-of-fit. The R^2 is defined as the ratio of explained variation to the total variation. The better the empirical model fits the actual data, the closer the R^2 values are to unity.

Table 3. Central Composite Design Setting in the Coded Forms

Run	(X_1)	(X_2)	(X_3)	(X_4)	(Y1)	(Y2)	(Y3)	(Y4)	(Y5)
1	+1	+1	-1	+1	7.25 ± 1.24	143.36 ± 6.57	37.00 ± 4.04	85.89 ± 0.74	85.65 ± 2.50
2	-1	+1	+1	+1	11.13 ± 2.57	51.93 ± 8.59	8.07 ± 4.04	54.02 ± 1.83	41.39 ± 1.09
3	+1	+1	+1	-1	12. 50 ± 0.60	113.00 ± 1.24	30.79 ± 1.01	83.01 ± 1.15	85.65 ± 3.12
4	-1	+1	-1	+1	12.84 ± 0.74	66.21 ± 3.54	12.71 ± 1.52	57.02 ± 0.47	49.44 ± 0.32
5	-1	+1	+1	+1	7.70 ± 0.74	114.79 ± 7.19	21.29 ± 1.52	79.67 ± 1.15	81.40 ± 1.10
6	-1	-1	+1	-1	10.54 ± 0.74	89.43 ± 0.50	15.07 ± 0.71	66.41 ± 1.76	86.64 ± 3.47
7	+1	-1	-1	-1	15.55 ± 0.12	68.36 ± 3.54	15.07 ± 0.71	38.77 ± 0.95	50.88 ± 2.34
8	0	0	0	0	9.92 ± 0.57	65.38 ± 4.31	23.07 ± 0.50	70.00 ± 8.32	54.19 ± 1.72
9	+1	+1	-1	-1	11.67 ± 0.20	75.50 ± 4.55	36.74 ± 2.97	89.62 ± 1.15	68.54 ± 1.72
10	-1	+1	-1	-1	15.75 ± 0.36	39.43 ± 1.01	18.43 ± 3.54	43.01 ± 0.07	33.44 ± 3.90
11	0	0	0	0	9.40 ± 1.88	64.43 ± 5.05	21.50 ± 2.02	60.00 ± 1.83	53.53 ± 2.97
12	-1	-1	-1	+1	10.12 ± 2.92	61.57 ± 5.00	10.45 ± 2.30	85.43 ± 0.57	74.61 ± 3.15
13	+1	+1	+1	-1	15.47 ± 0.84	79.07 ± 0.51	16.64 ± 3.98	70.53 ± 1.89	64.59 ± 2.84
14	+1	-1	+1	-1	15.73 ± 2.29	56.93 ± 6.57	22.36 ± 3.03	71.66 ± 7.94	49.33 ± 0.79
15	+1	-1	+1	+1	11.66 ± 0.64	33.36 ± 9.56	11.50 ± 2.02	39.47 ± 0.50	27.15 ± 6.24
16	+1	-1	-1	0	16.07 ± 0.48	82.29 ± 3.03	19.74 ± 3.93	51.87 ± 0.95	80.35 ± 4.68
17	-1	-1	+1	+1	16.07 ± 0.94	42.64 ± 0.51	20.07 ± 3.98	52.39 ± 0.07	38.85 ± 0.50
18	-1	-1	-1	+1	12.51 ± 0.35	53.00 ± 4.04	20.55 ± 3.70	84.63 ± 0.50	41.39 ± 0.16
19	0	0	0	-1	7.14 ± 0.74	48.40 ± 0.61	24.26 ± 0.82	64.50 ± 1.49	51.00 ± 3.02
20	0	0	0	-1	7.31 ± 0.51	62.28 ± 1.52	24.74 ± 2.06	53.78 ± 0.50	65.43 ± 0.78
21	0	0	0	-2	17.82 ± 0.00	61.21 ± 16.16	42.00 ± 12.62	67.18 ± 0.50	63.14 ± 8.98
22	0	0	0	0	7.88 ± 0.51	63.00 ± 12.02	24.86 ± 0.51	84.63 ± 0.50	58.61 ± 5.46
23	0	0	0	0	7.88 ± 0.31	63.80 ± 2.02	20.45 ± 3.67	64.50 ± 1.49	66.56 ± 0.78
24	0	0	0	0	10.08 ± 0.23	46.56 ± 1.03	16.29 ± 3.54	53.79 ± 0.50	45.81 ± 2.65
25	0	0	+2	0	12.60 ± 0.17	73.36 ± 1.52	20.57 ± 0.51	67.17 ± 0.50	70.86 ± 2.19
26	0	+2	0	0	11.10 ± 0.42	99.07 ± 0.51	35.21 ± 6.06	84.43 ± 0.66	63.58 ± 4.06
27	0	0	-2	0	12.00 ± 0.68	75.87 ± 1.52	24.50 ± 1.43	56.84 ± 2.17	36.30 ± 1.36
28	0	0	0	+2	6.78 ± 1.67	55.50 ± 8.59	29.35 ± 3.25	26.20 ± 0.76	39.85 ± 0.79
29	0	-2	0	0	11.12 ± 0.91	74.07 ± 2.52	22.36 ± 3.11	65.74 ± 2.57	38.83 ± 3.35
30	+2	0	0	0	11.49 ± 0.12	95.05 ± 3.11	15.93 ± 4.34	85.60 ± 0.34	56.95 ± 1.58

*Note: Y1 = Percentage of yield, Y2 = Total tannin content (mg TAE/g sample), Y3 = Total flavonoid content (mg RE/g sample), Y4 = DPPH scavenging activity (%), Y5 = ABTS scavenging activity (%) and ± standard deviation (*n*=3)

The parameters of regression equations obtained by the fitting of yield, total tannin content, total flavonoid content, DPPH scavenging activity, ABTS scavenging activity, and the R^2 value are 0.7493, 0.8662, 0,8720, 0.8578, and 0.7237, respectively.

The regression coefficients for dependent variables were obtained by multiplying the linear regressions, as shown in Table 4. The extraction temperature (X_1) has a positive linear effect and is significant ($p < 0.05$) to all responses except for the yield, which has a significant negative effect ($p < 0.05$). The result is similar to the study by Wu *et al*. (2014), who reported that the extraction temperature also significantly affects the total tannin content. Even though the preincubation time of extraction (X_3) does not significantly affect any of the responses ($p > 0.05$), the interaction effects of the preincubation time with other variables were significant ($p < 0.05$). For instance, there are significant interactive effects of the preincubation time and solid to solvent concentration (X_{34}) on total tannin content, DPPH, and ABTS scavenging activities ($p < 0.05$). The interaction parameter between X_{13} and X_{23} also had a significant effect on the selected response variable, as shown in Fig 1. The nonsignificant lack of fit value from the ANOVA analysis indicated the fitness of the

model, which adequately fit the experimental data ($p < 0.05$) for all response variables. Response surface 3D graphs were generated for each response, which showed the interaction between independent variables on the responses.

Effect of Extraction Variables on Yield Percentage

In the present study, the effect of extraction variables on yield recovery from the unripe banana peel was studied. The effects of the studied variables on the yield were analyzed and shown in Table 4.

Table 4. Regression Coefficients (β), Coefficient of Determination (R^2), and the F-test Value of the Predicted Second Order Polynomial Models for Antioxidant Compounds and Activities

Regression Coefficients (β)					
Intercept	Y1	Y2	Y3	Y4	Y5
X_0	8.12 **	63.13 ***	24.39 ***	72.35 ***	56.61 *
Linear					
X_1	-1.12 **	14.70 ***	2.00 *	9.35 ***	10.12 **
X_2	-0.49	10.25 **	3.02 ***	4.71 *	4.60
X_3	0.098	-0.57	-1.36	-0.76	2.49
X_4	-1.83 ***	-2.02	-2.44 **	-3.71 *	-5.26 *
Quadratic					
X_1^1	0.78 *	2.50	-2.82 ***	-0.17	-0.37
X_2^2	0.86	6.20 *	0.41	1.69	-0.41
X_3^3	1.16 **	3.21	-1.21	1.18	0.18
X_4^4	1.16 **	-0.61	2.07 **	-8.02 ***	-0.34
Interactions					
X_{12}	-0.16	10.28 *	5.70 ***	3.77	2.76
X_{13}	-0.15	-0.99	-1.60	-7.87 **	-2.96
X_{14}	-012	1.44	-0.69	-1.03	-1.57
X_{23}	-0.16	4.81	-1.95 *	2.22	5.08
X_{24}	-0.69	10.99 *	-0.87	0.92	5.68
X_{34}	0.41	-9.65 *	-0.92	-6.21 *	-7.21 *
R^2	0.7493	0.8662	0.8720	0.8578	0.7237
F value (model)	6.31 **	11.65 ***	12.27 ***	10.86 ***	4.72 *
F value (Lack of fit)	2.65	3.13	3.79	0.37	3.82

*Note: X_1 = Extraction temperature (°C), X_2 = Extraction time (min), X_3 = Preincubation time (min), X_4 = Solid to solvent concentration (%), *Y1* = Yield (%), *Y2* = Total Tannin content (mg TAE/g sample), *Y3* = Total flavonoid content (mg RE/g sample), *Y4* = DPPH Scavenging activity (%), *Y5* = ABTS Scavenging activity (%), and R^2 = Coefficient of determination; the level of significance is *$p < 0.05$, **$p < 0.01$, and ***$p < 0.001$

The extraction time (X_2) and preincubation time (X_3) of the UAE did not significantly affect the yield percentage of the crude extract. However, the extraction temperature (X_1) and the solid to solvent concentration (X_4) showed a significant, linear effect on the yield percentage ($p < 0.05$). The yield percentage of the crude extract of the unripe banana peel was profoundly affected ($p < 0.001$) by the percentage of the solid to solvent concentration. The quadratic effect of X_1^1, X_3^3, and X_4^4 also gave a significantly positive effect on the yield

percentage. For modelling the extraction process, a reduced quadratic model was used according to the second-order polynomial formula in Eq. 4,

$$Y_1 = 8.12 - 1.12X_1 - 0.49X_2 + 0.098X_3 - 1.83X_4 + 0.78X_1^1 + 0.86X_2^2 + 1.16X_3^3 + 1.1X_4^4 + 0.4X_{34} \quad (4)$$

As X_4 decreased and became more diluted in the solution, the yield percentage significantly increased ($p < 0.05$). Whereas, the percentage of yield increased when the solid to solvent concentration was 4.7% and decreased when the solution of solvent and solid was further increased to 10%. This was due to the mass transfer principle because the driving force of a mass transfer is the concentration gradient between the solid and liquid matter, which are more significant when applying a low sample to solvent ratio. The low solid to solvent concentration will increase the usage of solvent during the extraction process. It was observed that the percentage of yield increased with the decreasing temperature and solid to solvent concentration (%).

Effect of Extraction Variables on the Total Tannin Content

The impact of four extraction parameters on the total tannin content from the unripe banana peel is presented in Table 4. Statistical analysis showed that only the extraction temperature (X_1) and extraction time (X_2) had a significant impact on the total tannin content ($p < 0.05$). By evaluating the basis of the regression coefficient, the extraction temperature (X_1) had the most significant impact on the response among other factors. The increased extraction temperature (X_1) led to an increment in the total tannin content as it gave a positive, linear, significant effect ($p < 0.05$). The linear effect (X_2) and quadratic effect (X_2^2) of the extraction time and the interaction effect of X_{12}, X_{24}, and X_{34} showed a significant positive effect ($p < 0.05$), except for the interaction of X_{34} ($p > 0.05$) on the total tannin content (Table 4). The nonsignificant value of the lack of fit ($R^2 = 0.8662$) reflected a good prediction model of variable spatial influence to the response. The variation may be due to the regression coefficients being calculated and fitted to a second-order polynomial as follows in Eq. 5,

$$Y_2 = 63.13 + 14.7X_1 + 10.25X_2 - 0.57X_3 - 2.02X_4 + 6.2X_2^2 + 3.21X_3^3 + 10.28X_{12} + 4.81X_{23} + 10.99X_{24} - 9.65X_{34} \quad (5)$$

With the increased temperature and time of the extraction process, the total tannin content significantly increased ($p < 0.05$), as shown in Fig 1(a). At a lower extraction time (11.0 to 20.5 min) and temperature (30.0 °C to 37.5 °C), no interaction effect was observed. However, as the extraction time and extraction temperature increased, their interaction enhanced the total tannin content of the crude extract. The ultrasound is effective even in the later stage of the extraction process through enhance internal diffusivity, even though the effects were smaller. Based on the regression coefficient value, the linear effect concentration of solid to solvent does not significantly affect the total tannin content ($p > 0.05$), but their interaction with extraction time and preincubation time (X_{24} and X_{34}) is significant ($p < 0.05$) as shown in Fig. 1(b through c). An increase in the percentage of the solid to solvent concentration and extraction time increased the total tannin content. A low percentage in the concentration of solid to solvent during a low extraction time (11.0 min) increased the total tannin content. This is caused by the driving force of the concentration gradient between the solid and solvent being greater. The extraction time of 30.0 min was found to be the optimum operating time for the ultrasound-assisted extraction. As the

temperature increased, it helped to increase the solubility and diffusivity of the solid in the solvent solution (Eikani *et al*. 2012). The total tannin content was significantly affected ($p < 0.05$) at the extraction time of 11.0 min and temperature from 30 °C to 60 °C. The mechanical effect of the ultrasound contributes to the extraction process when used at a constant range of 10% nominal power. The contact surface area between the solid and liquid matter was increased by the mechanical effect, which caused the penetration of the solvent into the sample powder. The ultrasonic power generated over a very short time caused high temperature and pressure inside the formed bubble and thus, creating a violent shock wave. Moreover, the high-speed jet, which enhanced the penetration of the solvent into the cell tissue, helped accelerate the release process of the intracellular product into the water by disrupting it (Zhang *et al*. 2008). The presence of the violent shock wave and high-speed jet might cause the molecules to mix better, which enhanced the mass transfer rate in previous studies. A previous study showed a different result as the extraction time was not significantly affected by the condensed tannin content (Vu *et al*. 2016). This variation may be due to different solvents used in the experiment with different polarities of the solvent.

Figure 1 (b) shows the interaction effect of the solid to solvent concentration with time might be due to the increase in extraction temperature with the solid to solvent concentration. This causes the solubility to increase, as there is an improvement in the mass transfer for penetration of the solvent into the plant matrix (Al-Farsi *et al*. 2008). Cacace and Mazza (2003) showed that the solvent to solid concentration allows for the diffusion coefficient of the samples. Moreover, the interaction between the preincubation time and the solid to solvent concentration were observed to have a significant effect on the total tannin content ($p < 0.05$). The unripe Cavendish banana peel was significantly affected by the extraction temperature and time ($p < 0.05$), while the interaction of X_{12}, X_{24}, and X_{34} also significantly affected the experiment ($p < 0.05$).

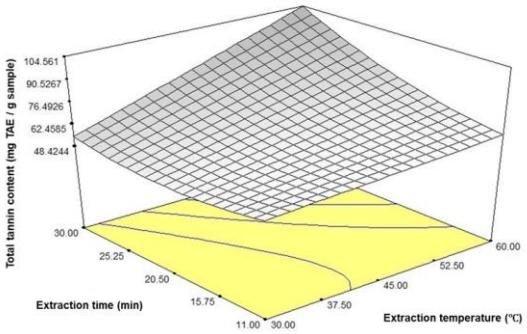
Effect of Extraction Variables on the Total Flavonoid Content

The effect of linear extraction temperature (X_1), extraction time (X_2), and solid to solvent concentration (X_4) showed a significant effect ($p < 0.05$) on the total flavonoid content (Table 4). Similarly, the interaction effect between the extraction temperature with time and the extraction time with preincubation time also showed a significant effect ($p < 0.05$) on the total flavonoid content (Table 4). Among these variables, the total flavonoid content depended more on X_{12} followed by X_2, X_4, X_1, and X_{23}, which are associated with higher regression coefficient values. The nonsignificant value for the lack of fit showed that the model was fitted with a good prediction ($R^2 = 0.8720$). The quadratic effects of the extraction temperature and percentage of solid to solvent concentration have significant effects on the flavonoid content, similar to the linear effects. RSM models developed to predict the flavonoid content of the sample were found to be reliable. These models can be fitted into the following second-order polynomial formulas of Eq. 6,

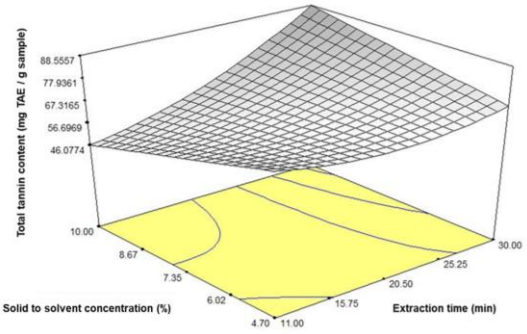
$$Y3 = 24.39 + 2X_1 + 3.02X_2 - 1.36X3 - 2.44X_4 - 2.82X_1^1 - 1.21X_3^3 + 2.07X_4^4 + 5.7X_{12} - 1.6X_{13} - 1.95X_{23} \quad (6)$$

Two stages existed in the extraction process. The first stage can be characterized by a rapid profile, which involves the penetration of the solvent into the cellular structure and the dissolution of soluble constituents into the solvent.

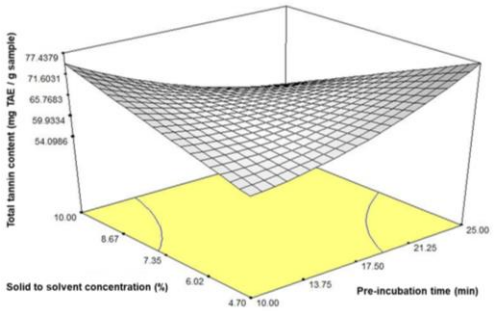
(a)


Total tannin content (mg TAE / g sample)
104.561
90.5267
76.4926
62.4585
48.4244
Extraction time (min)
30.00
25.25
20.50
15.75
11.00
Extraction temperature (°C)
30.00
37.50
45.00
52.50
60.00

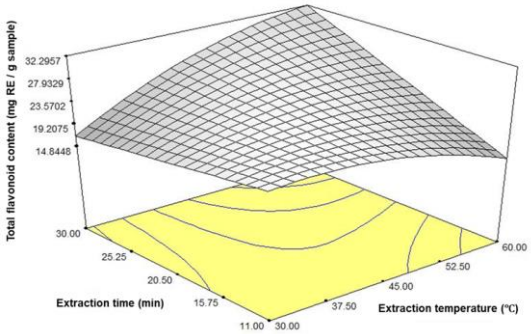
(b)


Total tannin content (mg TAE / g sample)
88.5557
77.9361
67.3165
56.6969
46.0774
Solid to solvent concentration (%)
10.00
8.67
7.35
6.02
4.70
Extraction time (min)
11.00
15.75
20.50
25.25
30.00

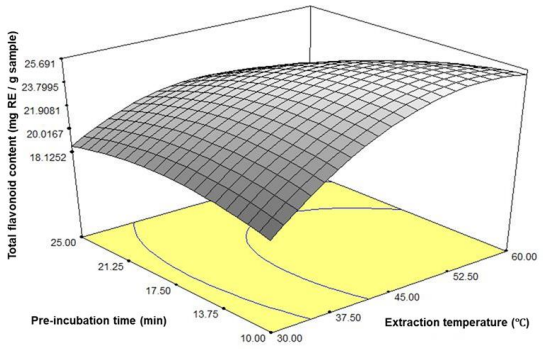
(c)


Total tannin content (mg TAE / g sample)
77.4379
71.6031
65.7683
59.9334
54.0986
Solid to solvent concentration (%)
10.00
8.67
7.35
6.02
4.70
Pre-incubation time (min)
10.00
13.75
17.50
21.25
25.00

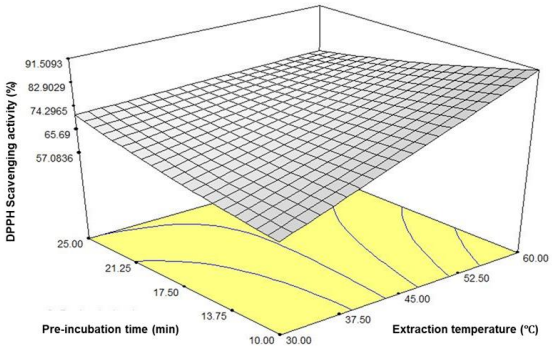
(d)


Total flavonoid content (mg RE / g sample)
32.2957
27.9329
23.5702
19.2075
14.8448
Extraction time (min)
30.00
25.25
20.50
15.75
11.00
Extraction temperature (°C)
30.00
37.50
45.00
52.50
60.00

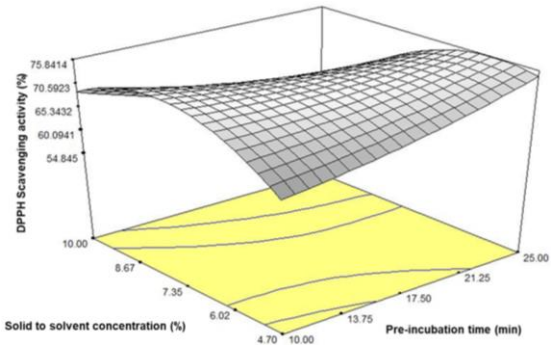
(e)


Total flavonoid content (mg RE / g sample)
25.691
23.7995
21.9081
20.0167
18.1252
Pre-incubation time (min)
25.00
21.25
17.50
13.75
10.00
Extraction temperature (°C)
30.00
37.50
45.00
52.50
60.00

(f)


DPPH Scavenging activity (%)
91.5093
82.9029
74.2965
65.69
57.0836
Pre-incubation time (min)
25.00
21.25
17.50
13.75
10.00
Extraction temperature (°C)
30.00
37.50
45.00
52.50
60.00

(g)


DPPH Scavenging activity (%)
75.8414
70.5923
65.3432
60.0941
54.845
Solid to solvent concentration (%)
10.00
8.67
7.35
6.02
4.70
Pre-incubation time (min)
10.00
13.75
17.50
21.25
25.00

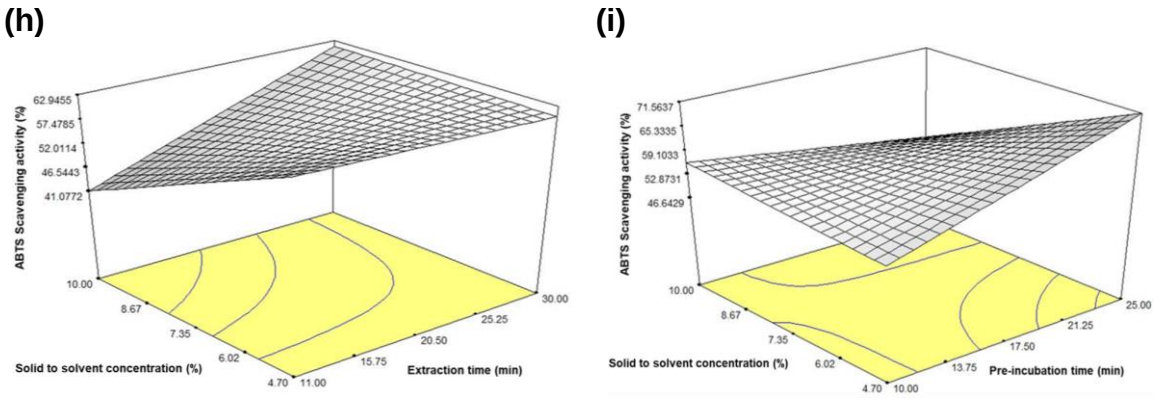


Fig. 1. The interaction effect of extraction variables on (a through c) the total tannin content, (d through e) total flavonoid content, (f through g) DPPH Scavenging activity, and (h through i) ABTS scavenging activity

Next, external diffusion from the porous structure of the residual solids of the sample occurs and transferred into the bulk solution. In the present study, the total flavonoid content was significantly affected ($p < 0.001$) in a time-dependent manner extracted at 30.0 min. This observation attributed to the fact that UAE will generate an ultrasound wave that could disrupt the cell wall of the solid matter, causing the contact area between the solvent and material to be increased. The interaction effect of the extraction temperature and time (X_{12}) showed a significant, positive effect ($p < 0.001$) on the flavonoid content Fig. 1(d). As the temperature of the extraction increased to 45 °C with an increased time of extraction, the total flavonoid content increased gradually. Further increase in temperature to 60 °C and the extraction time, resulted in a rise of flavonoid content, as shown in Fig. 2(d). In contrast, a previous study by Vu *et al*. (2016) reported that the extraction temperature and the percentage of solid to solvent concentration did not affect the extraction of crude extract using the same approach.

Thus, the most efficient extraction time in achieving maximum flavonoid content is 30.0 min. The interaction between the extraction temperature and time was crucial for determining the flavonoid content of the crude extract.

Effect of UAE Extraction Variables on Antioxidant Activity

The parameters that showed a significant, linear effect toward DPPH scavenging activity were extraction temperature (X_1), extraction time (X_2), and solid to solvent concentration (X_3) ($p < 0.05$). In contrast, the preincubation time of extraction (X_3) had no significant effect on the ABTS scavenging activity ($p > 0.05$). However, the interactive effect of the preincubation time and solid to solvent concentration (X_{34}) significantly affected ($p < 0.05$) both antioxidant activities based on the regression of the coefficient value (Table 4). Figure 1 (f through i) illustrated the three-dimensional response surface plots by representing the response in function of the two factors while keeping the others at constant middle values to show selected parameters on antioxidant activity. The DPPH scavenging activity increased with the increased temperature and the preincubation time of extraction (X_{13}), as shown in Fig. 1(h). The extraction temperature (X_1) and solid to solvent concentration (X_4) significantly affected the scavenging activity of ABTS ($p < 0.05$). The predicted model for the extraction of the high antioxidant activity from the unripe banana peels were calculated using Eq. 7 and Eq. 8,

$$Y4 = 72.35 + 9.35X_1 + 4.71X_2 - 0.76X_3 - 3.71X_4 + 1.18X_3^3 - 8.02X_4^4 + 3.77X_{12} - 7.87X_{13} + 2.22X_{23} - 6.21X_{34} \quad (7)$$

$$Y5 = 56.61 + 10.12X_1 + 4.6X_2 + 2.49X_3 - 5.26X_4 + 2.76X_{12} - 2.96X_{13} - 1.57X_{14} + 5.08X_{23} + 5.68X_{24} - 7.21X_{34} \quad (8)$$

Since the unripe banana peel contains various chemical compounds with different polarity and biological activities, different assays were needed to measure the antioxidant activity. Since antioxidant activity differed in terms of the different mechanisms, the various assays utilized have their advantages and limitations. Thus, using a single antioxidant assay is not enough to understand the antioxidant potential of a plant extract. In this study, the capacity of the studied extract to scavenge radicals 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2-azino-bis(3-ethylbenzothiazoline)-6-sulforic acid (ABTS) were evaluated. The principle behind DPPH free radical assay is that the antioxidants react with the stable DPPH radical and convert it into 1,1-diphenyl-2-picry l hydrazine. Meanwhile, regarding temperature, the DPPH scavenging activity of the antioxidant increased with an increasing extraction temperature of up to 60 °C (Fig. 1[g]). The high temperature, may cause certain antioxidants to mobilize, which was already mobilized at lower temperatures (Wettasinghe and Shahidi 1999). A 60 °C extraction temperature, 30.0 min extraction time, 10.0 min preincubation time, and 10% solid to solvent concentration yielded the best performance of DPPH and ABTS scavenging activity.

Optimization of the Ultrasonic-Assisted Extraction Condition

The optimal condition of UAE was determined by maximizing the desired responses using the Design Expert Software Version 6.0.4. The experimental values were compared with the predicted values based on the coefficient of variance (CV%) to determine the validity of the model. The UAE optimal conditions were determined according to the highest yield percentage, total tannin content, total flavonoid content, and the DPPH and ABTS scavenging activity. The optimum extraction conditions were obtained at an extraction temperature (X_1) of 60 °C, an extraction time (X_2) of 30.0 min, a preincubation time (X_3) of 25.0 min, and a solid to solvent concentration (X_4) of 5.03%. All measurements under the optimized conditions were conducted in triplicate and tabulated in Table 5. The experimental values were not significantly different with the predicted values under the optimal conditions. These results indicated that both experimental and predicted values were in agreement and reliable for the extraction process.

Table 5. Experimental Data of the Validation of Predicted Values at Optimal Extraction Conditions

Optimized Ultrasound-Assisted Extraction (UAE)			
Dependent variables	Predicted value	Experimental value	% Differences (CV)
Percentage of yield	13.80	14.88 ± 1.12	5.33
Total tannin content (mg TAE/g sample)	112.63	119.17 ± 1.79	3.99
Total flavonoid content (mg RE/g sample)	30.10	29.00 ± 2.53	2.63
DPPH scavenging activity (%)	87.34	80.82 ± 5.21	5.48
ABTS scavenging activity (%)	86.31	84.70 ± 3.77	1.33
*Note: CV= Coefficient of variation and ± standard deviation (n=3)			

CONCLUSIONS

1. *Musa acuminata* (Cavendish) unripe peel is a low cost and easy availability source of valuable bioactive compounds. The optimal condition extraction process of tannin content with antioxidant activities from unripe *Musa acuminata* (Cavendish) peel was successfully obtained using the response surface methodology (RSM) through UAE.
2. The optimised conditions of tannin and antioxidant activities were 60 °C extraction temperature, 30.0 min extraction time, 25.0 min preincubation time, and 5.03% solid to solvent concentration. The optimized condition was fitted with experimental values through validation. It is concluded that the extraction temperature (X_1) and extraction time (X_2) are highly dependent on the maximum total tannin content.
3. The extract containing tannin from the unripe banana peel is considered as an alternative potential local source and thus, these optimal condition process could contribute to the possibility of tannin and antioxidant for further applications. For example, the packaging products and incorporated as adhesive wood through bio-based polymer technology.

ACKNOWLEDGMENTS

The authors are grateful for the support and facilities of the Institute of Bioscience, Faculty of Engineering and Faculty of Food Science and Technology from the University of Putra Malaysia and the sample obtained from Ladang Puchong, UPM by Agrofresh International Sdn Bhd.

REFERENCES CITED

Al-Farsi, M. A., and Lee, C. Y. (2008). "Optimization of phenolics and dietary fibre extraction from date seeds," *Food Chemistry* 108(3), 977-985. DOI: 10.1016/j.foodchem.2007.12.009

Barrera Vazquez, M. F., Comini, L. R., and Martini, R. E. (2014). "Comparisons between conventional, ultrasound-assisted and microwave-assisted methods for extraction of anthraquinones from *Heterophyllaea pustulata* Hook f. (Rubiaceae)," *Ultrasonics Sonochemistry* 21, 478-484. DOI: 10.1016/j.ultsonch.2013.08.023

Baş, D., and Boyacı, İ. H. (2007). "Modeling and optimization I: Usability of response surface methodology," *Journal of Food Engineering* 78(3), 836-845. DOI: 10.1016/j.jfoodeng.2005.11.024

Bezerra, M. A., Santelli, R. E., Oliveira, E. P., Villar, L. S., and Escaleira, L. A. (2008). "Response surface methodology (RSM) as a tool for optimization in analytical chemistry," *Talanta* 76(5), 965-977. DOI: 10.1016/j.talanta.2008.05.019

Chan, K.W., Khong, N. M., Iqbal, S., Umar, I. M., and Ismail, M. (2002). "Antioxidant property enhancement of sweet potato flour under simulated gastrointestinal pH," *International Journal of Molecular Sciences* 13(7), 8987-8997. DOI: 10.3390/ijms13078987

Cacace, J. E., and Mazza, G. (2003). "Mass transfer process during extraction of phenolic

compounds from milled berries," *Journal of Food Engineering* 59(4), 379-389. DOI: 10.1016/S0260-8774(02)00497-1

Eikani, M. H., Golmohammad, F., and Homami, S. S. (2012). "Extraction of pomegranate (*Punica granatum* L.) seed oil using superheated hexane," *Food and Bioproduct Processing* 90, 32-36. DOI: 10.1016/j.fbp.2011.01.002

Food and Agriculture Organization of the United Nations (FAO) (2018). "Food and agricultural data (http://www.fao.org/faostat/en/#home), Accessed Day Month 2017.

Goldstein, N. J. L., and Swain, T. (1963). "Changes in tannins in ripening fruits," *Phytochemistry* 2(4), 371-383. DOI: 10.1016/s0031-9422(00)84860-8

González-Montelongo, R., Lobo, M. G., and González, M. (2010). "The effect of extraction temperature, time and number of steps on the antioxidant capacity of methanolic banana peel extracts," *Separation and Purification Technology* 71(3), 347-355. DOI: 10.1016/j.seppur.2009.12.022

Haile, M., and Kang, W. (2019). "Antioxidant activity, total polyphenol, flavonoid and tannin contents of fermented green coffee beans with selected yeasts," *Fermentation* 5(1), 29. DOI: 10.3390/fermentation5010029

He, B., Zhang, L. L., Yue, X. Y., Liang, J., Jiang, J., Gao, X. L., and Yue, P. X. (2016). "Optimization of ultrasound-assisted extraction of phenolic compounds and anthocyanins from blueberry (*Vaccinium ashei*) wine pomace," *Food Chemistry 204*, 70-76. DOI: 10.1016/j.foodchem.2016.02.094

Lee, J., Ye, L., Landen, W. O., and Eitenmiller, R. R. (2000). "Optimization of an extraction procedure for the quantification of vitamin e in tomato and broccoli using response surface methodology," *Journal of Food Composition and Analysis* 13(1), 45-57. DOI: 10.1006/jfca.1999.0845.

Liu, Y., Gong, G., Zhang, J., Jia, S., Li, F., Wang, Y., and Wu, S. (2014). "Response surface optimization of ultrasound-assisted enzymatic extraction polysaccharides from *Lycium barbarum*," *Carbohydrate Polymers* 110, 278-284. DOI: 10.1016/j.carbpol.2014.03.040

Manach, C., Scalbert, A., Morand, C., Rémésy, C., and Jiménez, L. (2004). "Polyphenols: Food sources and bioavailability," *The American Journal of Clinical Nutrition* 79(5), 727-747. DOI:10.1111/j.1365-2621.2009.02077

Ministry of Agriculture. (1998b). *Third National Agricultural Policy (1998-2010): Executive Summary*, Kuala Lumpur, Ministry of Agriculture.

Missio, A., L., Gatto, D., A., and Tondi, G. (2019). "Exploring tannin extracts: Introduction to new bio-based materials," *Brazilian Journal of Wood Science* 10(1), 88-102. DOI: 10.12953/2177-6830/rcm.v10n1p88-102

Pradal, D., Vauchel, P., Decossin, S., Dhulster, P., and Dimitrov, K. (2016). "Kinetics of ultrasound-assisted extraction of antioxidant polyphenols from food by-products: Extraction and energy consumption optimization," *Ultrasonics Sonochemistry* 32, 137-146. DOI: 10.1016/j.ultsonch.2016.03.001

Rabbani, G. H., Teka, T., Zaman, B., Majid, N., Khatun, M., and Fuchs, G. J. (2001). "Clinical studies in persistent diarrhea: Dietary management with green banana or pectin in Bangladeshi children," *Gastroenterology* 121(3), 554-560. DOI: 10.1053/gast.2001.27178

Rebello, L. P. G., Ramos, A. M., Pertuzatti, P. B., Barcia, M. T., Castillo-Muñoz, N., and Hermosín-Gutiérrez, I. (2014). "Flour of banana (Musa AAA) peel as a source of antioxidant phenolic compounds," *Food Research International* 55, 397-403. DOI:

10.1016/j.foodres.2013.11.039

Song, F., Lia, H., Sun, J., and Wang, S. (2013). "Protective effects of hydroxycinnamic acid and cinnamic aldehyde on isoproterenol-induced acute myocardial ischemia in rats," *Journal of Ethnopharmacology* 150(1), 125-130. DOI: 10.1016/j.jep.2013.08.019

Singh, H. P. (2011). "Harnessing the potential of banana and plantain in Asia and the Pacific for inclusive growth," in: *International Society for Horticultural Science (ISHS*), Leuven, Belgium, pp. 495-506.

Someya, S., Yoshiki, Y., and Okubo, K. (2002). "Antioxidant compounds from bananas (Musa cavendish)," *Food Chemistry* 79(3), 351-354. DOI: 10.1016/s03088146(02)00186-3

Sulaiman, S. F., Yusoff, N. A. M., Eldeen, I. M., Seow, E. M., Sajak, A. A. B., Supriatno, and Ooi, K. L. (2011). "Correlation between total phenolic and mineral contents with antioxidant activity of eight Malaysian bananas (*Musa sp*.)," *Journal of Food Composition and Analysis* 24(1), 1-10. DOI: 10.1016/j.jfca.2010.04.005

Vu, H. T., Scarlett, C. J., and Vuong, Q. V. (2016). "Optimization of ultrasound-assisted extraction conditions for recovery of phenolic compounds and antioxidant capacity from banana (Musa cavendish) peel," *Journal of Food Processing and Preservation* 41(5), 1-14. DOI: 10.1111/jfpp.13148

Vuong, Q. V., Hiruna, S., Roach, P. D., Bowyer, M. C., Phillips, P. A., and Scarletta, C. J. (2013). "Effect of extraction conditions on total phenolic compounds and antioxidant activities of Carica papaya leaf aqueous extracts," *Journal Herbal Medicine* 3(3), 104-111. DOI: 10.1016/j.hermed.2013.04.004

Wettasinghe, M., and Shahidi, F. (1999). "Antioxidant and free radical-scavenging properties of ethanolic extracts of defatted borage (*Borago officinalis* L.) seeds," *Food chemistry* 67(4), 399-414. DOI: 10.1016/S0308-8146(99)00137-5Get.

Wu, H., Zhu, J., Diao, W., and Wang, C. (2014). "Ultrasound-assisted enzymatic extraction and antioxidant activity of polysaccharides from pumpkin (*Cucurbita moschata*)," *Carbohydrate Polymers* 113, 314-324. DOI: 10.1016/j.carbpol.2014.07.025

Yusri, N., Chan, K., Iqbal, S., and Ismail, M. (2012). "Phenolic content and antioxidant activity of *Hibiscus cannabinus* L. seed extracts after sequential solvent extraction," *Molecules* 17(11), 12612-12621. DOI: 10.3390/molecules171112612

Zhang, Z. S., Wang, L. J., Li, D., Jiao, S. S., Chen, X. D., and Mao, Z. H. (2008). "Ultrasound-assisted extraction of oil from flaxseed," *Separation and Purification Technology* 62(1), 192-198. DOI: 10.1016/j.seppur.2008.01.014

Article submitted: November 19, 2019; Peer review completed: December 21, 2019; Revised version received: January 20, 2019; Accepted: January 21, 2020; Published: January 28, 2020.
DOI: 10.15376/biores.15.1.1877-1893