

## SAPONIN EXTRACTION FROM INFLORESCENCE OF *Musa balbisiana*

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### ABSTRACT

*Musa balbisiana* is known as a plant rich in nutritional value and has been used in popular medicine for many years. The study aimed to optimize the extraction process to obtain the extract-enriched saponin from the inflorescence of *M. balbisiana* by the response surface method (RSM). In addition, the bioactive components in the extract were screened. The results showed that *M. balbisiana* inflorescence extract contained significant amounts of saponins, polyphenols, and flavonoids. The optimal saponin extraction conditions, such as solvent concentration, temperature, and extraction time, are determined by the response surface method (RSM). The saponin content was 56.84 mg/g<sub>dm</sub> at optimal conditions was 62.41% methanol, the temperature of 65.86 °C, and the extraction time of 84.81 min.

**Keywords:** Extraction, inflorescence, *Musa balbisiana*, saponin.

### 1. INTRODUCTION

*Musa balbisiana* Colla is native to Indo-Malaysia, Asia, and Australasia, belongs to the Musaceae family. This plant is outstanding for its nutritional values and antioxidant capacity due to a variety of nutrients and biological compounds such as polyphenols, carotenoids, protein, fiber, minerals, vitamins, unsaturated fatty acids, and potassium [1]. Parts of the *M. balbisiana*, namely seeds, fruits, and inflorescences, have been used in traditional medicine to treat many disorders related to anti-diabetics, antipyretic, and delirium. Besides banana fruits, banana inflorescence has been used to support many medical disorders, such as lung problems in folk medicine. Some studies have indicated that it contains a broad range of biologically active compounds such as saponins, polyphenols, etc., which are these bioactivities' main components. Sanjay *et al.* (2018) reported that the main chemical compositions in the methanolic extract from *M. balbisiana* inflorescence include alkaloids, phenolics, and flavonoids. This extract had the significant capacity of scavenging free radicals and reducing oxidants in the DPPH and FRAP assays [2]. Tin *et al.* also revealed that the inflorescence of *M. balbisiana* in India also contained a number of triterpenes, which constitute a potential source of high-value compounds for nutraceutical, pharmacological, and food applications [3]. In addition, there is a high demand for elucidating these experiences with scientific evidence for further research and applications on a larger scale. Finding extraction conditions and determining the chemical components is one of the prerequisites for in-depth studies on biological activities and applications.

Saponins are a large group of glycosides, which are naturally occurring organic compounds that have at least one C-3 glycosidic bond between the aglycone and a sugar chain. Hydrolysis of the saponin molecule produces two parts, an aglycone (sapogenin) and a part of a sugar part [4]. According to the structure of the aglycone or of sapogenin, saponins are classified into neutral saponins and saponins. Neutral saponins are steroid derivatives with twisted side chains found in monocotyledonous angiosperms and acid saponins, with triterpenoids being the most common structural type and occurring mainly in dicotyledonous angiosperms. Saponins often have detergent properties and stable foaming properties when in contact with water [5]. Saponins are present in many plant species, both wild and cultivated. Studies have shown that saponins have biological activities such as anti-bacterial, anti-oxidant, anticoagulant, anticancer, and anti-inflammatory [6]. Thus, saponins are attracting the attention

of researchers around the world to study. Among available natural resources, plants are an important one that has gained more concern.

Solvent-assisted extraction is a method used to extract substances from a solution using a suitable solvent. This method usually has better extraction performance than some other methods. The solvent can effectively and quickly separate the substance to be extracted. This method allows the use of a variety of solvents to suit the nature and solubility of the substance to be extracted. This helps to ensure that the substance to be extracted can be efficiently separated and applies to a wide variety of samples and substances to be extracted [7]. This study aimed to establish a foundational framework for further research and practical applications by determining the optimal conditions for saponin extraction using solvents from the inflorescence of *M. balbisiana*, a plant native to Vietnam.

## 2. MATERIALS AND METHODS

### 2.1. Materials

The inflorescence of *M. balbisiana* was sourced from Hue Tinh commune, Chau Phu district, An Giang province. Harvesting occurred in late summer, with a 15 cm section left attached at the distal end of the banana bunch following the cessation of fruit formation. The bracts, which ranged in colour from bright red to dark violet, were collected and transported to the laboratory [9]. There, the inflorescences were cleaned to eliminate impurities, dried at 60 °C to gain a moisture content under 10%, ground into a fine powder, and sieved using a 0.3 mm mesh. The resulting powder was stored in a zipper bag for subsequent experimental use.

Chemicals in the study were at analysis grade: oleanolic acid, vanillin, glacial acetic acid, perchloric acid, methanol, ethanol, ethyl acetate and n-butanol, Folin-Ciocalteu reagent, gallic acid (GA) solution, H<sub>2</sub>O<sub>2</sub> solution, quercetin (QE).

### 2.2. Methods

#### 2.2.1. Phytochemical screening

Table 1. Phytochemical screening tests

Constituents	Experiments	Observation
Saponin steroid	5 mL of 0.5 N NaOH solution (pH = 13) + 3 drops of sample	Durable foam appearance
Saponin triterpenoid	5 mL of 0.1 N HCl solution (pH = 1) + 3 drops of sample	Durable foam appearance
Tannin	Reaction with vanillin/ H <sub>2</sub> SO <sub>4</sub>	Positive (a deep red colour)
Flavonoid	Reaction with 10% NaOH solution	Dark yellow appearance
Alkaloid	Reaction with Wagner reagent	Brown precipitate appearance
Steroid	React with Liebermann-Burchard reagent	Green, pink, orange, or red colour (solid colour) appearance
Glycozite	Reaction Keller-Killian	Reddish brown colour between two layers of liquid appearance
Anthocyanin	1 mL the extraction + 5 mL pH = 1	Red color appearance
	1 mL the extraction + 5 mL pH = 4.5	Purple color appearance
Phenolic	50 µL the extraction + 500 µL distilled water + 2-3 drops of FeCl <sub>3</sub> (5%)	Dark blue precipitate appearance
Carotenoid	React with acetone	Clear dark yellow fluid appearance

The extract from *M. balbisiana* inflorescence used for phytochemical screening was prepared as follows. 1 gram of the dried material was soaked sequentially in solvents at a raw material-to-solvent

ratio of 1:20 (w/v) using distilled water, 80% methanol (v/v), and 80% ethanol (v/v) for 24 hours at room temperature with constant shaking. Each mixture was filtered through Whatman No. 1 filter paper. The resulting filtrates were subsequently analyzed to quantify the bioactive compounds present in the extracts. Qualitative phytochemical screening was performed to determine the primary groups of components, including saponins, alkaloids, tannins, terpenoids, carbohydrates, cardiac glycosides, anthraquinone glycosides, flavonoids, and phenols. The analysis involved colour-based reactions following the methodologies described by Sofowora *et al.* (1993) and Tiwari *et al.* (2011), as outlined in Table 1 [9, 10].

### 2.2.2. Saponin extraction

#### a. Saponin extraction

1 gram of raw material (calculated on a dry mass basis) was combined with a solvent at varying concentrations (60%, 70%, 80%, and 90%) and material-to-solvent ratios (1:10, 1:20, 1:30, and 1:40, w/v). The extraction was conducted in a thermostatic bath (Memmert WNB22, Germany) at temperatures of 40 °C, 50 °C, 60 °C, and 70 °C for 60, 90, 120, and 150 min. The resulting mixtures were centrifuged at 5000 rpm for 15 min using a centrifuge (Philips, HR2096/00), followed by filtration through filter paper to obtain a homogeneous extract. The total saponin content was then measured spectrophotometrically at a wavelength of 550 nm.

#### b. Optimization of saponin extraction

The response surface methodology (RSM) was used to evaluate the effects of factors involved in solvent-assisted extraction on saponin content. Three independent variables were considered: solvent concentration ratio ( $X_1$ , v/w), extraction temperature ( $X_2$ , °C), and extraction time ( $X_3$ , min). The central composite design (CCD) was used to construct experiments aimed at developing a quadratic model for the response variables. CCD includes a fractional factorial design with a center point, supplemented with extreme values (both low and high) for each factor to facilitate the estimation of curvature. The dependent variable, or response variable, was the saponin content ( $Y$ , mg/gdm). The experimental levels for the variables are summarized in Table 2.

The experiment design was carried out with 20 experiments (8 experiments at the edge, six experiments at the centre, and six experiments on the swingarm). The linear regression equation with quadratic form was determined by JMP 10 software:

$$Y(\%) = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + b_{11}X_{12} + b_{22}X_{22} + b_{33}X_{32} + b_{12}X_1X_2 + b_{13}X_1X_3 + b_{23}X_2X_3$$

In which,  $b_0$ ,  $b_1$ ,  $b_2$ ,  $b_3$ ,  $b_{11}$ ,  $b_{22}$ ,  $b_{33}$ ,  $b_{12}$ ,  $b_{13}$ ,  $b_{23}$  were the coefficients of the variable  $X_1$ ,  $X_2$ ,  $X_3$ ,  $X_{11}$ ,  $X_{22}$ ,  $X_{33}$ ,  $X_1X_2$ ,  $X_1X_3$ ,  $X_2X_3$  respectively.

The data obtained from the experimental results were analyzed using the JMP software version 10.0.

Table 2. Experimental levels for optimization

Variables	Coded variables	Unit	Levels				
			-1.68 (- $\alpha$ )	-1	0	1	+1.68 (+ $\alpha$ )
Solvent concentration	$X_1$	%	43.2	50	60	70	76.8
Temperature	$X_2$	°C	43.2	50	60	70	76.8
Time	$X_3$	min	58.2	65	75	85	91.8

## 2.3. Analysis methods

### 2.3.1. Total saponins content determination

The spectrophotometric method is based on the Rosenthaler reaction between the sample solution and the reagent vanillin/glacial acetic acid, perchloric acid, measuring the absorbance at 550 nm [12].

### 2.3.2. Total flavonoids content determination

The total flavonoid content was measured following the method described by Chang *et al.* (2002), which involved constructing a calibration curve using quercetin (QE) as the standard. The absorbance was recorded at 765 nm.

### 2.3.3. Total polyphenol content determination

The polyphenol content (TPC) was determined using the Folin-Ciocalteu method according to the description of Feduraev *et al.* (2019) [14].

### 2.3.4. Carotenoid content determination

The carotenoid content was determined as the description by Kotíková *et al.* (2011) [15]. The formula was followed.

$$C_a = 17.75 * A_{662} - 2.35 * A_{645}$$

$$C_b = 18.61 * A_{645} - 3.96 * A_{662}$$

$$C_{x+c} = (1000 * A_{470} - 2.27 * C_a - 81.4 * C_b) / 227$$

$$X = \frac{C_x + c * V * F}{m * (100 - w) / 100}$$

In which: C<sub>a</sub>: chlorophyll a content (µg/mL); C<sub>b</sub>: chlorophyll b content (µg/mL); C<sub>x+c</sub>: carotenoid content (µg/mL); V: volume of extract (mL); F: dilution factor; m: sample mass (g); w: moisture content of raw materials (%).

### 2.3.5. Tannin content determination

The tannin content was evaluated with potassium iodate test, following the protocol of Rhazi Naima *et al.* (2015). A tannic acid solution was used for calibration curve [16].

### 2.3.6. Alkaloid content determination

The total alkaloid content was determined as the description by Chia Hau Lee *et al.* (2021) with atropine used as a standard [17].

## 2.4. Data analysis

The experiments were conducted in triplicate. Experimental data were analyzed using Microsoft Excel 2016, while the ANOVA statistical analysis was performed with SPSS 20 software. Optimization and data evaluation were carried out using JMP 10 software.

## 3. RESULTS AND DISCUSSION

### 3.1. Phytochemical screening

The obtained ethanolic, methanolic, and aqueous extracts were determined to identify the main components such as saponins, polyphenols, tannins, flavonoids, alkaloids, steroids, glycosides, anthocyanins, and carotenoids were identified as described in Section 2.3.1. The results are presented in Table 3.

Table 3. Chemical compositions of ethanolic, methanolic, and aqueous extracts

No.	Constituents	Methanolic extract	Ethanolic extract	Aqueous extract	No.	Constituents	Methanolic extract	Ethanolic extract	Aqueous extract
1	Saponins	+++	+++	++	6	Steroids	-	-	-
2	Polyphenols	+++	+++	++	7	Glycosides	-	-	-
3	Tannins	+	+	+	8	Anthocyanins	-	-	-
4	Flavonoids	+++	+++	++	9	Carotenoids	+	+	+
5	Alkaloids	+	+	+	+: present moderately; ++: present strongly; +++: present very strongly; -: signifies absence.				

Table 3 shows that glycosides, anthocyanins, and steroids were not detected in any of the three extracts. There were identified clearly with saponins, polyphenols, flavonoids and less with tannins, alkaloids and carotenoids, each exhibiting distinct characteristics, which were reflected in the observed colour intensity. The content of these compounds was subsequently determined, and the results are presented in Table 4.

Table 4. Content of bioactive components in *M. balbisiana* inflorescence

Contents	Methanolic extract	Ethanolic extract	Aqueous extract
Saponin (mg/g <sub>dm</sub> )	46.98 ± 1.17 <sup>a</sup>	42.97 ± 1.45 <sup>b</sup>	30.26 ± 1.06 <sup>c</sup>
Polyphenol (mgGAE/g <sub>dm</sub> )	35.17 ± 1.54 <sup>a</sup>	31.81 ± 2.00 <sup>b</sup>	22.68 ± 0.97 <sup>c</sup>
Flavonoid (mgQE/g <sub>dm</sub> )	24.98 ± 1.47 <sup>a</sup>	20.42 ± 1.66 <sup>b</sup>	16.96 ± 1.41 <sup>c</sup>
Carotenoid (mg/g <sub>dm</sub> )	3.45 ± 0.05 <sup>a</sup>	2.27 ± 0.01 <sup>a</sup>	1.83 ± 0.06 <sup>b</sup>
Tannin (mg/g <sub>dm</sub> )	5.17 ± 0.02 <sup>a</sup>	0.99 ± 0.13 <sup>b</sup>	1.08 ± 0.04 <sup>b</sup>
Alkaloid (mgAE/g <sub>dm</sub> )	1.46 ± 0.05 <sup>b</sup>	3.63 ± 0.15 <sup>a</sup>	3.86 ± 0.13 <sup>a</sup>

Note: In the same rows, different letters represent a statistically significant difference at  $p < 0.05$ .

Total saponin content was accounted for as the highest content in the inflorescence of *M. balbisiana* extracts, followed by polyphenols and flavonoids.

### 3.2. Saponin extraction

#### 3.2.1. Effects of solvent on the extraction of saponins

The above experimental results show that the TSC content in the extracted material with methanol solvent is higher than that of ethanol and distilled water. Therefore, methanol was selected as the investigation solvent for the next experiments. The solubility of bioactive substances such as saponins in plant materials varies depending on the type of solvent. Thus, the concentration and volume of the solvent affected the extracted saponin content. The effects of solvent concentration and material/solvent ratio on obtained saponins content were presented in Fig. 1 and Fig. 2.

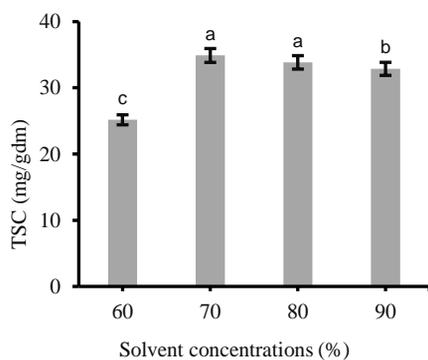


Fig.1. Effects of solvent concentration on saponins extraction

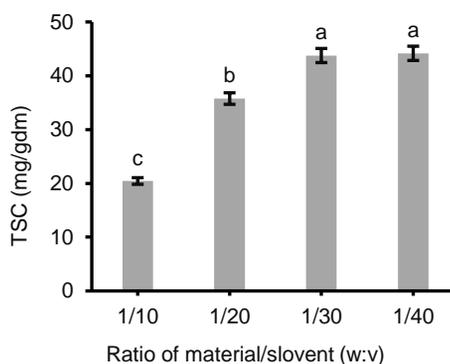


Fig. 2. Effects of material/solvent ratio on saponins extraction

Note: Different letters on the bar reveal a statistically significant difference ( $p < 0.05$ ).

Fig. 1 indicated that the saponin content tended to increase gradually from the concentration of 60% to 70% and reached the highest point at 70% with  $34.89 \pm 1.47$  (mg/g<sub>dm</sub>). The figure was  $32.87 \pm 2.59$  mg/g<sub>dm</sub> at 90%. Higher solvent concentration would result in a stronger penetration rate into the cell and enhance the extraction performance. However, high solvent concentration with high polarity would not result in higher target compound content due to much-accompanied impurities. In the case of the low solvent concentration, the low polarity can reduce the ability to dissolve and separate the substances from the material into the solvent, reducing the extraction efficiency. In this experiment, 70% was selected for the following experiments. Additionally, the saponin content showed a significant increase, rising from  $20.44 \pm 2.25$  mg/g<sub>dm</sub> to  $43.76 \pm 1.08$  mg/g<sub>dm</sub> as the raw material/solvent ratio

changed from 1:10 to 1:30 (w:v). However, the saponin content slightly increased from  $44.17 \pm 1.19$  mg/g<sub>dm</sub> (1:40 w/v) without different significance with the ratio of 1/30 (w/v). Cui *et al.* (2019) also found that the extraction yield of saponin compounds significantly increased with a higher solvent volume [16]. In this study, the saponin extraction efficiency reached its maximum at a material-to-solvent ratio of 1:30 (w:v) and did not increase further with higher solvent volumes. In fact, the solvent used in the extraction process needs a sufficient amount to penetrate the raw materials, pulling the dissolved components into the extract. The lower the solvent volume used, the lower the saponin content obtained in the corresponding extract. However, the amount of the solvent should be high enough to help the solvent penetrate the plant cell wall of the material; along with the formation of a concentration gradient from the inside of the cell to the environment, the solvent pulls the soluble components of the raw material. Therefore, from the viewpoint of the economy, a material: solvent ratio of 1:30 (w:v) is sufficient to extract the target saponin compounds.

### 3.2.2. Effects of temperature and time on saponin extraction

Two factors of temperature and time extraction also have a significant impact on extraction efficiency [19]. In this study, the temperature from 40 °C to 70 °C and time extraction from 60 min to 150 min were investigated. The results are presented in Fig. 3 and Fig. 4.

Saponin content increased significantly from 40 °C to 60 °C and tended to decrease at higher temperatures. This could be because a low extraction temperature slows down the mass transfer process, but at a high extraction temperature, the solvent would evaporate, and part of the saponin would be oxidized and reduced in content. Saponins are less durable compounds, so they can be decomposed if the extraction temperature is too high enough [20]. The similarity tendency was observed in the study of Menghao Du *et al.* (2014); the saponins content extracted from the Soup tree (*Sapindus mukorossi* Gaertn) at 95% EtOH, 60 °C was higher than the temperature range of 20 °C - 50 °C. There was no significant difference in saponin content at 60 °C and 70 °C. Thus, 60°C was the chosen temperature in this study. On the other hand, the saponin content increased sharply from  $43.37 \pm 1.60$  mg/g<sub>dm</sub> (60 min) to  $51.82 \pm 2.57$  mg/g<sub>dm</sub> (90 min) and reached the peak at 120 min ( $53.02 \pm 2.72$  mg/g<sub>dm</sub>) before decreasing to  $47.03 \pm 1.31$  mg/g<sub>dm</sub> (150 min) (Fig. 4). In fact, the short time would not be enough for the solvent to penetrate the cells to extract saponin into the plant cells, which results in low extraction efficiency. In contrast, prolonged extraction time increased the diffusion of the target compounds into the solvent and enhanced the yield extract [14]. The results indicated that there was no remarkable difference in saponin content for the survey time at 90 min and 120 min. Therefore, 90 min was selected for further experiments. Gyu *et al.* (2020) found suitable conditions to extract saponins, flavonoids, and polyphenols from *Chenopodium quinoa* Willd using methanol as extraction solvent at 60 °C for 90 min to obtain a significant saponin content (13.39 g/100g<sub>dm</sub>) [21].

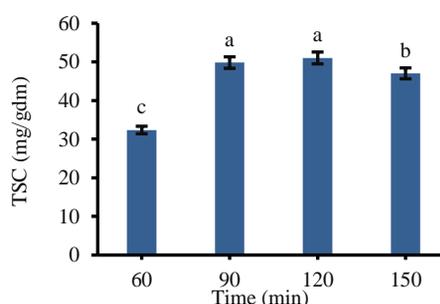
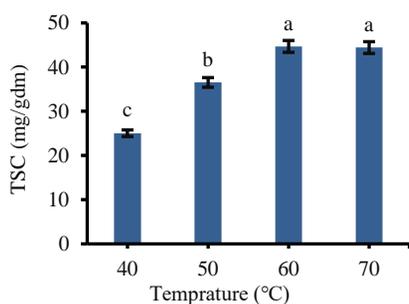


Fig. 3. Effects of temperature on saponin extraction Fig. 4. Effects of time on saponins extraction

Note: Different letters on the bar reveal a statistically significant difference ( $p < 0.05$ ).

### 3.3. Optimizing conditions for extracting saponins

In determining the optimal conditions for saponin extraction from *M. balbisiana* inflorescence, it is essential to consider the factors influencing the extraction process, including solvent concentration, temperature, and extraction time. By examining the interactions between these factors, the optimal conditions can be identified. The results of the saponin content obtained through the CCD in the RSM,

with three factors—solvent concentration ( $X_1$ ), extraction temperature ( $X_2$ ), and extraction time ( $X_3$ ) are presented in Table 5.

Table 5. CCD design arrangement and responses

No.	Coded variables			Saponin (mg/g <sub>dm</sub> )	No.	Coded variables			Saponin (mg/g <sub>dm</sub> )
	$X_1$	$X_2$	$X_3$			$X_1$	$X_2$	$X_3$	
1	-1	-1	-1	45.62	11	0	-1.68	0	46.13
2	-1	-1	1	49.03	12	0	1.68	0	52.33
3	-1	1	-1	49.25	13	0	0	-1.68	49.85
4	-1	1	1	55.36	14	0	0	1.68	52.92
5	1	-1	-1	49.79	15	0	0	0	56.48
6	1	-1	1	51.75	16	0	0	0	55.39
7	1	1	-1	47.08	17	0	0	0	56.55
8	1	1	1	55.34	18	0	0	0	54.13
9	-1.68	0	0	45.35	19	0	0	0	55.06
10	1.68	0	0	54.36	20	0	0	0	54.36

Table 6. Results of significance analysis of coefficients of the regression equation

Coefficient	Coeff. SC	Std. Err.	P-value	Coefficient	Coeff. SC	Std. Err.	P-value
$b_0$	55.31	0.72	<.0001	$b_{13}$	0.09	0.62	0.8913
$b_1$	1.45	0.48	0.0124	$b_{23}$	1.13	0.62	0.1017
$b_2$	1.56	0.48	0.0086	$b_{11}$	-1.82	0.47	0.0029
$b_3$	1.82	0.48	0.0034	$b_{22}$	-2.04	0.47	0.0014
$b_{12}$	-1.14	0.62	0.0991	$b_{33}$	-1.28	0.47	0.0207
$R^2 = 0.8843$				$R^2_{adj} = 0.7801$			

According to Joglekar and May (1987), for the model to fit, the  $R^2$  value must be at least 0.8, and  $R^2_{adj}$  parameter greater than four is required [22]. The values of  $R^2 = 0.8843$ ,  $R^2_{adj} = 0.7801$ . The correlation coefficient  $R^2$  reached 0.8843, indicating that the model explained 88.43% of the data variability. In addition, the level of fit of the model was also evaluated by the F value of Lack of fit. The P value was used to test the significance of each regression coefficient. Specifically, the factors with P value < 0.05 were considered to influence the response. Of the nine regression coefficients (except  $b_0$ ), three regression coefficients were not significant with confidence  $P > 0.05$ , namely  $b_{12}$ ,  $b_{13}$ , and  $b_{23}$ , which proved the interaction between  $X_1X_2$ ,  $X_1X_3$ , and  $X_2X_3$  had no significant effect on the response. For negative values, the regression coefficients  $b_{11}$ ,  $b_{22}$ , and  $b_{33}$  were the factors that had a negative impact on the response, reducing the obtained saponin content. Regression coefficient  $b_3$  had the largest positive value, showing that  $X_3$  (extraction time) has a large positive impact on the objective function. Table 6 also showed the influence of solvent concentration ( $X_1$ ), extraction temperature ( $X_2$ ), and extraction time ( $X_3$ ) on the obtained saponin content, influencing factors according to the second-order polynomial equation. After analyzing ANOVA using JMP software, the following equation was shown:

$$Y = 55.31 + 1.45X_1 + 1.56X_2 + 1.82X_3 - 1.82X_{12} - 2.04X_{22} - 1.28X_{32}$$

ANOVA analysis results indicated that the maximum saponin content was 56.84 mg/kg<sub>dm</sub> at extracted conditions: methanol 62.41%, 65.86 °C, and extraction time 84.81 min. The response surface model showed the influence of the investigated factors on the saponin content, and the model predicted the optimal conditions (Fig. 5 and Fig. 6). The extraction temperature increase would lead to the increased kinetics of substances, causing substances to move at a faster rate than solvent dissolution. The longer extraction time facilitates the saponins' extraction process, leading to an increase in saponin content. However, high temperature and longer extraction time also lead to the loss of solvent; the solute concentration would reach equilibrium, so the content of saponin did not increase but tended to decrease because saponin was easily decomposed with conditions of high temperature for a long time.

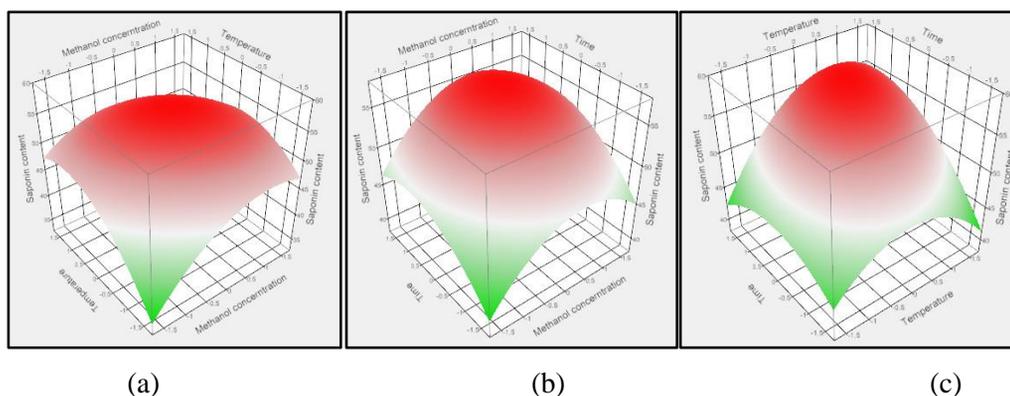


Fig. 5. Response surface model (a, b, c) showing the influence of factors (solvent concentration  $X_1$ ; extraction temperature,  $X_2$ ; extraction time,  $X_3$ ) on saponin content

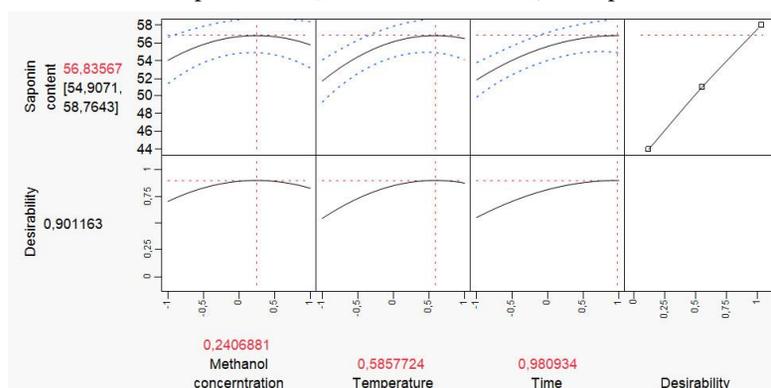


Fig. 6. Saponin content prediction model

Three experiments were done under the obtained optimal conditions, and the results from the predicted and actual experiments were compared. The experimental saponin content was  $55.16 \pm 0.25$  mg/g<sub>dm</sub>, which showed a 2.96% difference (<5%) compared to the predicted saponin content of 56.84 mg/g<sub>dm</sub>, based on the regression equation. This indicates that the experimental saponin content closely matches the value predicted by the second-order regression model, confirming the model's reliability and practical applicability. In a similar study, Do Thi Mai Trinh *et al.* (2022) optimized the extraction conditions for triterpenoid saponins from *Camellia oleifera* seeds using the RSM. The optimal conditions, based on the CCD model, resulted in a maximum triterpenoid saponin content of 5.45% at a solvent-to-material ratio of 19:1 (v/w), with 80% ethanol concentration and a 77-hour extraction time [23]. Similarly, in the work by Zhang *et al.* (2018), the optimization of extraction factors for phytochemical components and antioxidant activity from *Asparagus officinalis* root extracts led to the following optimal parameters: a temperature of 51 °C, 73.02 min of extraction, 75.23% ethanol, and a material-to-solvent ratio of 1:50 (w:v), resulting in a total phenolic content ranging from 14.7 to 35.2 mgGAE/g and total saponin content ranging from 9.2 to 17.1 mgRE/g<sub>dm</sub> [19]. These findings confirm that the optimization results align well with the experimental outcomes.

#### 4. CONCLUSION

The study found the dominant components in the extract from *M. balbisiana* inflorescence. The obtained extract was enriched saponins. It also contained polyphenols, flavonoids, tannins, alkaloids, and carotenoids. In the single-factor investigation for saponin extraction, the selected conditions were 70% methanol, the material/solvent ratio 1/30 w/v, and 60 °C in 90 min. The optimal conditions resulted in a saponin content of 56.84 mg/g<sub>dm</sub>. This is basic information for further studies relating to isolation and biological activities evaluation of the obtained extract for practical applications.

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## TÓM TẮT

### NGHIÊN CỨU TRÍCH LY SAPONIN TỪ HOA CỦA CÂY CHUỐI HỘT *Musa balbisiana*

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*Musa balbisiana* được biết đến là loại cây giàu hoạt chất và được sử dụng phổ biến trong y học dân gian. Nghiên cứu nhằm mục đích tối ưu hóa quá trình chiết xuất để thu được saponin giàu dịch chiết từ hoa *M. balbisiana* bằng phương pháp bề mặt đáp ứng (RSM). Ngoài ra, các thành phần hoạt tính sinh học trong dịch chiết đã được định tính và định lượng. Kết quả cho thấy chiết xuất hoa *M. balbisiana* chứa một lượng đáng kể saponin, polyphenol và flavonoid. Các điều kiện chiết saponin tối ưu như nồng độ dung môi, nhiệt độ và thời gian chiết, được xác định bằng phương pháp bề mặt đáp ứng (RSM). Hàm lượng saponin là 56,84 mg/g<sub>CK</sub> ở điều kiện tối ưu là methanol 62,41%, nhiệt độ 65,86 °C và thời gian chiết là 84,81 phút.

*Từ khóa:* Hoa chuối, *Musa balbisiana*, saponin, trích ly.