Trop J Nat Prod Res, September 2025; 9(9): 4331 - 4335 **ISSN 2616-0684 (Print)**
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Original Research Article

Optimization of Microwave - Assisted Extraction Conditions for Total Saponin Content and Antioxidant Activity of *Launaea sarmentosa* Leaves

Thu M. Tran[1], Huyen T. Nguyen[2], Giang T.T. Dinh[3], Tai T. Tran[3], Quang D. Ho[2], Thang C. Truong[4], Khanh K. Nguyen[5], Kieu P. Nguyen[1], Tuan N. Nguyen[1], Thanh T. Nguyen[2]*

[1]*Institute of Biotechnology and Food Technology, Industrial University of Ho Chi Minh City, Ho Chi Minh City 70000, Vietnam.*
[2]*School of Chemistry, Biology, and Environment, Vinh University, Vinh City, Nghe An Province 43000, Vietnam.*
[3]*Department of Chemistry, Vinh University, Vinh City, Nghe An Province 43000, Vietnam*
[4]*Faculty of Pharmacy, Vinh Medical University, Vinh City, Nghe An Province 43000, Vietnam*
[5]*Faculty of Food Technology, Binh Dương University, Ho Chi Minh City 70000, Vietnam.*

ARTICLE INFO

Article history:
Received 28 April 2025
Revised 03 August 2025
Accepted 05 August 2025
Published online 01 October 2025

ABSTRACT

Launaea sarmentosa is an herbaceous plant rich in saponin, especially in the leaves. The plant has been shown to have antioxidant activity. The present study aimed to optimize the extraction conditions for maximizing both total saponin content (TSC) and antioxidant activity of the plant leaves. To achieve this, Response Surface Methodology (RSM) was applied using a Box-Behnken Design (BBD), which involved three independent variables: microwave power, extraction time, and ethanol concentration. These parameters were systematically optimized to maximize the extraction yield of the desired bioactive compounds. TSC was determined spectrophotometrically using diosgenin as the reference standard. Antioxidant activity was assessed through the DPPH (2,2-diphenyl-1-picrylhydrazyl) free radical scavenging method. The optimal conditions determined through the model were microwave power of 330 W, extraction time of 22 minutes, and ethanol concentration of 64%. Under these conditions, the experimental values of TSC and antioxidant activity were found to be 9.56 ± 0.24 mg DE/g dry weight and 87.62 ± 0.46%, respectively. The findings indicate that microwave-assisted extraction, when optimized using RSM, is an effective approach for enhancing the recovery of bioactive components from *L. sarmentosa*. This study provides a scientific basis for the efficient use of this plant as a natural source of antioxidants. In addition, the results gave valuable insights into the development of plant-based functional ingredients for pharmaceutical and nutraceutical applications. By optimizing the extraction conditions, this work demonstrated the potential of *Launaea sarmentosa* as a promising candidate for use in health-promoting products, paving the way for its broader industrial application.

Keywords: *Launaea sarmentosa*, Antioxidant, Saponin content, Microwave-Assisted extraction, Response surface methodology

Introduction

Launaea sarmentosa (Willd) Schultz-Bip.ex Kuntze is a creeping herbaceous plant in the *Asteraceae* family. It grows on sandy coasts in Malaysia, Sri Lanka, China, India, and Vietnam,[1] particularly on tropical Indian coastlines. Saponin can be found in all parts of the plant, especially the leaves. Studies have shown that compounds from the plant have potential antioxidant properties, making them essential. The herb contains a lot of aromatic compounds, mostly phenols, and their derivatives, which have shown protective effect against plant microbes.[2] *Launaea* holds immense importance owing to its ethnobotanical uses, phytochemical composition, biological effects, and the presence of biologically active compounds, including terpenoids, phenolics, sesquiterpenoids, flavonoids and saponins.[3] The roots and leaves of *Launaea* contains calcium oxalate crystals, tannins, alkaloids, amino acids, carbohydrates, glycosides, and steroids.[4]

*Corresponding author. Email: nguyentanthanh@vinhuni.edu.vn
Tel: +84-989339115

Citation: Rahmad Darmawan, Ira Humairah, Ema Qunianingsih, Ervi A. Munthe, Linda. Effect of *Annona muricata* L. (Soursop) on Blood Glucose Level in a Diabetic Rat Model: A Meta-Analysis. Trop J Nat Prod Res. 2025; 9(9): 4331 – 4335 https://doi.org/10.26538/tjnpr/v9i9.31

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

Microwave-assisted extraction (MAE) offers numerous benefits compared to conventional extraction techniques, including reduced extraction durations, minimized solvent consumption, enhanced extraction efficiency, improved product quality, and reduced expenses. For example, the MAE technique can extract compounds in just a few minutes, while the Soxhlet method takes several hours. Additionally, the cost of MAE equipment is lower than that of supercritical fluid extraction equipment.[5] In MAE, microwave energy generates high temperatures and pressures during extraction, increasing the plant material's water absorption capacity and releasing organic compounds into the solvent. These modifications can increase the extraction efficiency in obtaining target analytes from plant material.[6]

Response Surface Methodology (RSM) is an effective statistical approach used to optimize experimental parameters and investigate critical factors, helping to minimize the number of experiments needed. RSM assists in delineating the impacts of independent variables, both individually and when combined.[7] Verifying the model's predicted values experimentally is crucial to implementing RSM. RSM is an effective tool for optimizing technological processes, offering greater efficiency and cost-effectiveness compared to the traditional one-factor-at-a-time approach. In this research, RSM was used to optimize the conditions for extracting antioxidant compounds and total saponin from the leaves of *Launaea sarmentosa*, which are the main compounds found in the genus *Launaea*.[8]

Materials and Methods

Plant collection and identification

The leaves of *Launaea sarmentosa* were collected from Nghi Loc District in Nghe An Province, Vietnam (18.833°N 105.583°E) in September 2023. The plant material was subsequently identified by the Institute of Ecology and Biological Resources, Vietnam Academy of Science and Technology, where voucher number LSNL2023 was assigned.

Plant extraction

Following drying with a heat pump drier at 40 - 50°C, the leaves were pulverized employing a mechanical grinder. For each extraction, exactly 1.0 g of the powdered sample was mixed with 20 mL of ethanol–water solution at a specified concentration, as defined in the experimental design. The extraction was conducted using a microwave-assisted extraction system equipped with a digital power control unit. Each extraction was performed under controlled conditions following the Box-Behnken design, with microwave power levels ranging from 200 to 400 W, extraction durations from 10 to 30 minutes, and ethanol concentrations from 40% to 80% (v/v). A 100 mL round-bottom flask was used to contain the mixture of sample and solvent, which was then exposed to microwave radiation according to the predefined power conditions. Upon completion of the extraction, the mixture was allowed to cool to ambient temperature and subsequently filtered using Whatman No. 1 filter paper to obtain the extract. The final samples were kept at 4°C prior to analytical testing.

Determination of total saponin content (TSC)

TSC was determined using the combined and modified techniques outlined by Hu *et al*. (2012)[9] and Moyo *et al*. (2013).[10] Exactly 0.2 mL of *L. sarmentosa* leaf extract, 0.35 mL of vanillin solution (8% in ethanol), and 0.8 mL of absolute methanol were accurately transferred into a 15 mL glass test tube. Subsequently, 1.25 mL of sulfuric acid (72%) was added, and the test tube was sealed and shaken thoroughly. The test tube caps were slightly loosened before placing the tubes in a water bath maintained at 60°C for 10 minutes. After heating, the tubes were left to cool at room temperature for 5 minutes. The absorbance of the resulting reaction mixture was recorded at 544 nm using an Agilent 8453 UV-Vis spectrophotometer, with absolute methanol serving as the blank.

A standard curve of Diosgenin ($y = 0.0002x + 0.031$; $R^2 = 0.9962$) was prepared using various concentrations of diosgenin ranging from 100 to 700 mg/mL. The measurement was repeated three times. The total saponin content was determined using the following equation (Equation 1):

$$\mathrm{TSC} = \mathrm{V}\frac{C}{m} \qquad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (1)$$

Where;

C is the concentration determined using the diosgenin standard curve (mg/mL), m is the sample's dry weight (g), V is the volume of extraction solvent (mL).

Total saponin content in the sample was expressed as milligrams of Diosgenin Equivalent (DE) per gram of dry weight (mg DE/g dw).

Determination of antioxidant activity

The antioxidant activity of *L. sarmentosa* leaf extract was assessed through the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay. The DPPH radical scavenging assay quantifies a compound's or sample's ability to neutralize the stable 1,1-diphenyl-2-picrylhydrazyl radical.[11] The DPPH radical scavenging activity of the extract was conducted as follows: In a test tube, 10 µL of the sample, 0.5 mL of 0.5 mM DPPH solution was combined with 1 mL of ethanol, and 990 µL of 100 mM sodium acetate buffer (pH 5.5). The solution was mixed gently and kept in the dark at room temperature for 5 minutes to allow the reaction to occur. Thereafter, the absorbance of the solution was recorded at 517 nm using a UV-Vis spectrophotometer.

Triplicate assays were conducted for accuracy, with DPPH scavenging activity quantified according to the formula in Equation 2.

$$\text{Radical scavenging activity (\%)} = \left(1 - \frac{\text{Abssample}}{\text{Abscontrol blank}}\right) \times 100 \qquad \ldots\ldots\ldots\ldots\ldots\ldots \quad (2)$$

Where;

Abssample is the absorbance of sample extract, and Abscontrol blank is the absorbance of blank control.

Experimental design

Microwave-assisted extraction optimized the experimental design using Response Surface Methodology (RSM). Box-Behnken Design (BBD) comprising seventeen experimental runs, which included five repetitions at the center point to ensure accuracy. The microwave power (X_1), extraction time (X_2), and ethanol concentration (X_3) were considered as the independent variables, while the total saponin content (TSC) (Y_1) and antioxidant activity (Y_2) served as the dependent variables. The experiments were conducted in replicates, and the response values were calculated as the averages.

Table 1: The coded and uncoded independent variables used in the Box-Behnken Design

Independent variables	Symbols	Coded levels		
		-1	0	+1
Microwave power (W)	X_1	200	300	400
Extraction time (min)	X_2	10	20	30
Ethanol concentration (%)	X_3	40	60	80

Statistical analysis

Data were presented as mean ± standard deviation (SD) of triplicate determination. Differences between means were determined using one-way analysis of variance (ANOVA) using Design Expert (v7.0).

Results and Discussion

Fitting the models

To optimize the recovery of bioactive compounds from the leaves of *L. sarmentosa*, a BBD was applied, targeting the optimization of total saponin content and antioxidant activity. The model enabled an understanding of how different processing variables influenced the outcomes, thereby supporting the identification of ideal extraction parameters. Table 2 outlines the design involving three primary response metrics. The design encompassed 17 experiments, with five replicates at the central point. For this research, model selection was based on higher-order polynomial equations that provided meaningful additional coefficient estimates and ensured no aliasing among the variables. A second-order polynomial model was suggested by the software, showing a good fit across all three independent variables considered in the study.

The following quadratic polynomial equations [Eqs. (3–4)] represent the definitive empirical regression model that depicts the connection between responses and the three variables tested for total saponin content and antioxidant activity:

$$Y_1 = 9.58 + 0.21X_1 + 0.17X_2 + 0.11X_3 - 0.095X_1X_2 - 0.05X_1X_3 - 0.13X_2X_3 - 0.25X_{12} - 0.32X_{22} - 0.16X_{32} \qquad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (3)$$

$$Y_2 = 88.17 + 1.06X_1 + 0.15X_2 + 0.022X_3 + 0.065X_1X_2 + 0.007X_1X_3 - 0.17X_2X_3 - 1.61X_{12} - 0.67X_{22} - 0.67X_{32} \qquad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (4)$$

Where;

Y_1 is total saponin content (TSC), Y_2 is the antioxidant activity, while the independent variables include X_1 (microwave power), X_2 (extraction time), and X_3 (ethanol concentration).

The statistical relevance of the model's coefficients was assessed through analysis of variance (ANOVA), as presented in Table 3.

In order to evaluate the ANOVA results of the regression and response

surface models describing Y_1 and Y_2, the associated p-values and R^2 values were examined. The calculated F values for Y_1 and Y_2 were 29.33 and 1049.97, respectively, both yielding p-values <0.05. This suggests that both models exhibited statistical significance. The R^2 values obtained for the models was 0.9742 and 0.9993, signifying that over 97.42% and 99.93% of the response variation was successfully modeled. This showcases the exceptional accuracy and proficiency of the established model within the defined range boundaries. Lack-of-fit test statistics (F-values) for Y_1 and Y_2 were 3.13 and 1.07, respectively indicating that the lack-of-fit value was not statistically significant relative to the residual error, thereby confirming the adequacy of the polynomial model.

Table 2: The experimental results from the Box-Behnken Design

RUN	X_1 (W)	X_2 (min)	X_3 (%)	TSC Y_1 (mg DE/g dw)	DPPH RSA Y_2 (%)
1	300	20	60	9.64	88.09
2	200	20	40	8.88	84.81
3	300	30	40	9.23	87.11
4	400	10	60	9.18	86.73
5	300	10	40	8.65	86.53
6	200	10	60	8.49	84.71
7	300	20	60	9.52	88.23
8	200	20	80	9.09	84.88
9	400	30	60	9.34	87.21
10	300	20	60	9.53	88.19
11	300	10	80	9.23	86.91
12	300	20	60	9.57	88.17
13	300	20	60	9.64	88.17
14	300	30	80	9.28	86.80
15	400	20	40	9.34	86.92
16	400	20	80	9.35	86.96
17	200	30	60	9.03	84.93

TSC: Total Saponin Content, RSA: Radical Scavenging Activity

Response surface analysis

The extraction conditions that yielded the highest total saponin content (TSC) and antioxidant activity (DPPH radical scavenging activity) were affected by extraction time, microwave power, and ethanol concentration.

Response surface analysis of TSC

Figure 1 showcases response surface plots that illustrate the impact of independent variables and their interactions on the yields of TSC during *L. sarmentosa* leaf extraction. As depicted in Figure 1 and detailed in Table 3, it is evident that all three factors, namely ethanol concentration, microwave power and extraction time, exhibit statistically significant quadratic effects ($p < 0.0001$). Figure 1a shows the response surface plot illustrating the relationship between microwave power and extraction time, showcasing their combined impact on TSC, while holding ethanol concentration constant at 60%. The TSC yields showed an increase as the microwave power was raised from 200 to 350 W. Though, from 350 to 400 W, there was a slight tendency for TSC to decrease, although not significantly. These results corroborate the findings previously reported by Akbari *et al.* (2019).[12] The maximum amount of total saponin can be achieved at a microwave power of 300-400 W.

Table 3: Analysis of variance **(**ANOVA) for the response surface of the model

Source	Y_1 – TSC			Y_2 – DPPH RSA		
	Mean Square	F-value	p-value	Mean Square	F-value	p-value
Model	0.19	29.33	< 0.0001^S	2.81	1049.97	< 0.0001^S
X_1	0.37	57.75	0.0001^S	9.01	3368.23	< 0.0001^S
X_2	0.22	34.53	0.0006^S	0.17	63.97	< 0.0001^S
X_3	0.09	14.10	0.0071^S	4.050E-003	1.51	0.2583^{NS}
X_1X_2	0.03	5.64	0.0493^S	0.017	6.32	0.0402^S
X_1X_3	0.01	1.56	0.2516^{NS}	2.250E-004	0.084	0.7802^{NS}
X_2X_3	0.07	10.97	0.0129^S	0.12	44.50	0.0003^S
X_1^2	0.27	41.51	0.0004^S	10.91	4080.04	< 0.0001^S
X_2^2	0.43	66.81	< 0.0001^S	1.86	696.07	< 0.0001^S
X_3^2	0.11	17.63	0.0040^S	1.88	701.32	< 0.0001^S
Lack of Fit	0.01	3.13	0.1498^{NS}	2.775E-003	1.07	0.4566^{NS}
R^2	0.9742			0.9993		
C.V%	0.87			0.06		

TSC: Total Saponin Content, RSA: Radical Scavenging Activity, S: significant ($p < 0.05$); NS: non-significant

The TSC exhibited an upward trend as the extraction time extended between 10 and 25 minutes. Though, as the extraction time continued to increase beyond 25 minutes, the TSC declined. This result aligns with the findings reported by Xu *et al*. (2012).[13] In Figure 1b, the response surface illustrates the impact of varying microwave power and ethanol concentration on TSC at an extraction time of 20 minutes. The TSC yields exhibited an upward trend as the ethanol concentration was raised from 40% to 70%. However, once the ethanol concentration surpassed 70% and reached 80%, a decrease in TSC yields was observed. The findings of this study are consistent with those reported by Amid *et al*. (2013).[14] The highest saponin content in *L. sarmentosa* leaf extract can be obtained when the ethanol concentration ranges between 60% and 80%.

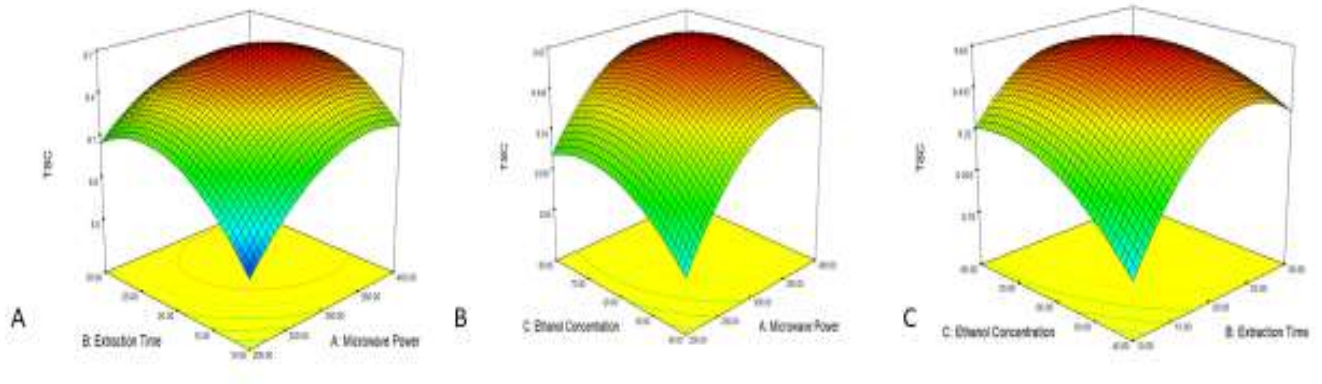

Figure 1: The response surface plot of total saponin content (TSC)

Response surface analysis of antioxidant activity

Figure 2 displays the surface plots for the antioxidant activity of *L. sarmentosa* leaves extract. In Figure 2a, the 3D plot depicts the response surface for microwave power (X_1) and extraction time (X_2), while keeping the ethanol concentration fixed at 60%. The response surface plot suggested that microwave power had a relatively minor effect, whereas extraction time had a more significant impact on the antioxidant activity. It can be observed that antioxidant activity increased as microwave power increased from 200 to 330 W, but declined when the power was further increased from 330 to 400 W. This trend is consistent with the findings reported by Thanh *et al*. (2023).[15] The maximum antioxidant activity was achieved at a microwave power of 300 - 360 W. As the extraction time increased from 10 to 20 minutes, antioxidant activity improved; however, extending the time to 30 minutes led to a reduction.

In Figure 2b, the 3D surface plots illustrate the interplay between ethanol concentration (X_3) and microwave power (X_1) at an extraction time of 20 minutes. The 3D surface plots in Figure 2b are similar to that of Figure 2a. The antioxidant activity increased as the ethanol concentration increased from 40% to 60%. However, beyond this point, when the ethanol concentration was increased from 60% to 80%, the antioxidant activity decreased.

Figure 2c illustrates the interaction between ethanol concentration (X_3) and extraction time (X_2), with microwave power maintained at 300 W. Increasing the ethanol concentration promoted cell membrane disruption, thereby improving solvent penetration into the solid matrix. The antioxidant activity increased as the extraction time increased from 10 to 23 minutes, followed by a decline between 23 and 30 minutes. As the ethanol concentration increased from 40% to 65%, there was an increase in the antioxidant activity. Following the continuous increase in ethanol concentration from 65% to 80%, the antioxidant activity exhibited a decrease. These findings align with those reported in a study conducted by Chi *et al*. (2022).[16]

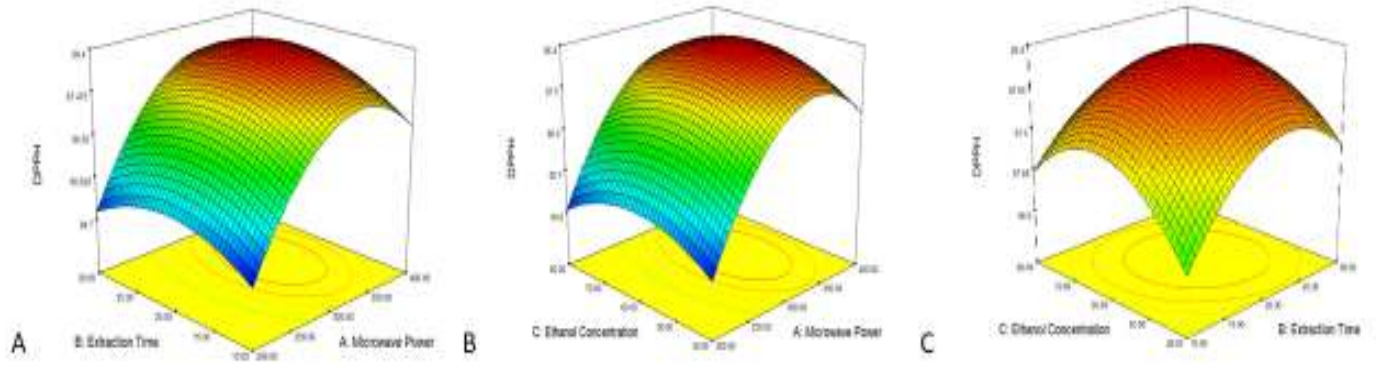

Figure 2: The response surface plot of antioxidant (DPPH radical scavenging) activity

Optimization and model verification

By applying simultaneous optimization via the desirability function method, the optimal conditions for extracting *L. sarmentosa* leaf compounds were determined as a microwave power of 330 W, an ethanol concentration of 64% and an extraction time of 22 minutes. These conditions offer the optimal combination for achieving the highest TSC and antioxidant activity. The experimental results yielded extraction yields of 9.56 ± 0.24 mg DE/g dw for total saponin content and 87.62 ± 0.46% for DPPH radical scavenging activity. There was a high level of consistency between the experimental data and the predicted outcomes (TSC = 9.6432 mg DE/g dw, DPPH radical scavenging activity = 88.3249%) generated from the corresponding regression models with CV values ranging between 0.79% and 0.83%.

Conclusion

The successful use of the response surface methodology facilitated the determination of optimal microwave-assisted extraction conditions for maximizing the yield of total saponin content and antioxidant activity of *L. sarmentosa* leaves. Under the optimized conditions, which included a microwave power of 330 W, an extraction time of 22 minutes, and an ethanol concentration of 64%, the experimental values for total saponin content and antioxidant activity were measured at 9.56 ± 0.24 mg DE/g dw and 87.62 ± 0.46%, respectively.

Conflict of Interest

The author`s declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgements

The authors gratefully acknowledge the support provided by the Institute of Biotechnology and Food Technology, Industrial University of Ho Chi Minh City, for access to testing instruments.

References

1. Moghal MMR, Millat MS, Hussain MS, Islam MR. Thrombolytic and Membrane Stabilizing Activities of *Launaea sarmentosa*. Int J Pharmacogn. 2016; 3:354-358. doi: 10.13040/IJPSR.0975-8232.IJP.3(8).354-58.
2. Millat MS, Islam S, Hussain SM, Moghal MR, Islam T. Anti-Bacterial Profiling of *Launaea sarmentosa* (Willd.) and *Bruguiera cylindrical* (L.): Two Distinct Ethno Medicinal Plants of Bangladesh. Eur J Exp Biol. 2017; 7:1-6. doi: 10.21767/2248-9215.100006.
3. Abdelkrim C, Mebarka B, Nasser B, Houria D. Phytochemical and Biological Studies on *Launaea* Cass. Genus (Asteraceae) From Algerian Sahara. Curr Top Phytochem. 2012; 11:67-80.
4. Yusriyya S, Harischa CR, Vinay JS, Rabinarayan A. Pharmacognostical Evaluation of *Launaea sarmentosa* (Willd.) Schultz-Bip. ex Kuntze Root. An Int Q J Res Ayurveda. 2013; 34:90-94. doi: 10.4103/0974-8520.115439.
5. Hao JY, Han W, Huang SD, Xue BY, Deng X. Microwave-Assisted Extraction of Artemisinin from *Artemisia annua* L. Sep Purif Technol. 2002; 28:191–196. doi: 10.1016/S1383-5866(02)00043-6.
6. Kratchanova M, Panchev I, Pavlova E. The Effect of Microwave Heating of Fresh Orange Peels on The Fruit Tissue and Quality of Extracted Pectin. Carbohydr Polym. 2004; 56:181–185. doi: 10.1016/j.carbpol.2004.01.009.
7. Bas D and Boyaci IH. Modeling and Optimization I: Usability of Response Surface Methodology. J Food Eng. 2007; 788:836–845. doi: 10.1016/j.jfoodeng.2005.11.024.
8. Them LT, Tuan NT, Trinh PTN, Tho LTC, Vi LNT, Hung QT, Dung NPT, Thuy NV, Dung LT. Saponin, Polyphenol, Flavonoid Content and α-Glucosidase Inhibitory Activity, Antioxidant Potential of *Launaea sarmentosa* Leaves Grown in Ben Tre Province, Vietnam. IOP Conf Ser Mater Sci Eng. 2019; 542(1):012036. doi: 10.1088/1757-899X/542/1/012036.
9. Hu T, Guo YY, Zhou QF, Zhong XK, Zhu L, Piao JH, Chen J, Jiang JG. Optimization of Ultrasonic-Assisted Extraction of Total Saponins From *Eclipta prostrasta* L. Using Response Surface Methodology. J Food Sci. 2012; 77:975-982. doi: 10.1111/j.1750-3841.2012.02869.x.
10. Moyo M, Amoo SO, Ncube B, Ndhlala AR, Finnie JF, Van Staden J. Phytochemical and Antioxidant Properties of Unconventional Leafy Vegetables Consumed in Southern Africa. S Afr J Bot. 2013; 84:65-71. doi: 10.1016/j.sajb.2012.09.010.
11. Rajha HN, El Darra N, Hobaika Z, Boussetta N, Vorobiev E, Maroun RG, Louka N. Extraction of Total Phenolic Compound, Favonoids, Anthocyanins, and Tannins from Grape Byproducts by Response Surface Methodology. Influence of Solid-Liquid Ratio, Particle Size, Time, Temperature and Solvent Mixtures on the Optimization Process. Food Nutr Sci. 2014; 5:397–409. doi: 10.4236/fns.2014.54048.
12. Akbari S, Abdurahman NH, Yunus RM. Optimization of Saponins, Phenolics, and Antioxidants Extracted from Fenugreek Seeds Using Microwave-Assisted Extraction and Response Surface Methodology as An Optimizing Tool. C R Chim. 2019; 22:714-727. doi: 10.1016/j.crci.2019.07.007.
13. Xu HJ, Shi XW, Ji X, Du YF, Zhu H, Zhang LT. A Rapid Method for Simultaneous Determination of Triterpenoid Saponins In *Pulsatilla turczaninovii* Using Microwave-Assisted Extraction and High-Performance Liquid Chromatography–Tandem Mass Spectrometry. Food Chem. 2012; 135:251-258. doi: 10.1016/j.foodchem.2012.04.081.
14. Amid BT and Mirhosseini H. Effect of Different Purification Techniques on The Characteristics of Heteropolysaccharide-Protein Biopolymer from Durian (*Durio zibethinus*) Seed. Molecules. 2012; 17:10875-10892. doi: 10.3390/molecules170910875.
15. Thanh NT. Optimization of Microwave-Assisted Extraction of Antioxidant Compounds from Roots of *Polygonum multiflorum* Thunb. At Vietnam Using Response Surface Methodology. Malays J Chem. 2023; 25:21-29.
16. Chi TP, Trang BT, Trinh NTN, Tho LTC, Thanh NT, Thang TD, Hang HTL, Quang LD, Tuan NN. Optimization of Ultrasonic-Assisted Extraction of Antioxidant Compounds in Black Shallots (*Allium ascalonicum*) From Vietnam Using Response Surface Methodology. Malays J Chem. 2022; 24:26-35.