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Optimization of solvent-free microwave-assisted extraction of antioxidant compounds from *Lagenaria siceraria* fruit by response surface methodology

Sahar Iftikhar[1], *Muhammad Tariq Qamar*[1], *Alev Yüksel Aydar*[2], *Dildar Ahmed*[1,*]

[1] Department of Chemistry, Forman Christian College (A Chartered University), Lahore, Pakistan
[2] Department of Food Engineering, Manisa Celal Bayar University, Manisa, Turkey

ABSTRACT

Lagenaria siceraria (bottle gourd) fruit is highly celebrated for its nutritional as well as therapeutic properties. The present study was undertaken with the aim of exploring an efficient green solvent-free microwave-assisted extraction method for bioactives from this fruit. Optimization was done according to response surface methodology (RSM), where microwave power (W) and time (s) were independent factors, and percent extraction yield, total phenolic content (TPC), total flavonoid content (TFC), 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity, ferric reducing antioxidant potential (FRAP) and iron chelating activity (ICA) were the responses. TPC, TFC, DPPH radical scavenging activity, FRAP and ICA were highest at 480 W and 60 s. The TPC was 288.9 mg GAE · g^{-1} DW (milligram gallic acid equivalent per gram dry weight), TFC was 214.1 mg RE · g^{-1} (rutin equivalent per gram DW), anti-radical activity was 32.96%, FRAP was 289.7 mg AAE · g^{-1} (mg ascorbic acid equivalents per gram) and ICA was 19.52%. The results of the study thus demonstrate that the solvent-free microwave-assisted extraction method, which utilised an optimum power of 480 W and a time of 60 s, is an effective and green method for extraction of antioxidant compounds from bottle gourd fruit.

Keywords: antioxidant, bottle gourd, green extraction, iron chelation, phenolics

Abbreviations: CCD, central composite design; DoE, design of experiment; DPPH, 1,1-diphenyl-2-picrylhydrazyl; FRAP, ferric reducing antioxidant potential; ICA, iron chelating activity; MAE, microwave-assisted extraction; RSM, response surface methodology; TFC, total flavonoid content; TPC, total phenolic content.

INTRODUCTION

Plants are considered as a backbone in the development of medicines as they are a great source of bioactive compounds. There are several drugs that cannot be synthesised in laboratories. Plant-based bioactive compounds often have complex structures and, therefore, it is difficult to synthesise them. Thus, extracting them from plants would be the only means to obtain them (Cragg and Newman, 2013). As is well known, the extracts obtained from plants are extensively used as folk medicines to treat several diseases in both developed as well as underdeveloped countries (Pan et al., 2014).

Lagenaria siceraria (bottle gourd) is a plant belonging to the family Cucurbitaceae. The family Cucurbitaceae comprises 118 genera and close to 825 species (Bisognin, 2002). This is a medicinal plant whose therapeutic effects are widely recognised and celebrated around the world. Archaeologists have proved that humans have been using it since ancient times, and it is considered as the first plant

*Corresponding author.
e-mail: dildarahmed@fccollege.edu.pk (Dildar Ahmed).

species domesticated on earth (Yetişir et al., 2008). Phytochemical studies have detected the presence of several types of chemical compounds in it, including phenolics, terpenoids, glycosides and saponins (Liaqat et al., 2018; Attar and Ghane, 2019). *L. siceraria* fruit is used to treat numerous diseases, including jaundice, bronchial disorders, cardiac disorders and skin diseases (Decker-Walters et al., 2004; Ahmed et al., 2017). It has also been shown to possess antimicrobial, antioxidant, anti-inflammatory, antibacterial and antihyperlipidemic activities and exhibits strong lipase inhibitory activities (Panchal et al., 2014; Ahmed et al., 2017). Further, it has been found to exert anti-obesity effects (Maqsood et al., 2017) and have antidiabetic properties (Saha et al., 2011; Ahmed et al., 2016).

Several fast, efficient and environmentally friendly methods of extraction of bioactive compounds have been demonstrated in numerous studies in the modern literature. One of these methods is microwave-assisted extraction (MAE), which involves irradiating the sample with microwaves to efficiently release chemical compounds from a plant biomass (Wang and Weller, 2006; Pinto et al., 2021). Studies have revealed that MAE is more efficient than many conventional methods in terms of yield and time consumption (Doulabi et al., 2020). Solvent-free microwave-assisted extraction (SFME) has been considered as a green technique for extraction of essential oils from herbs. A comparison study between SFME and conventional hydrodistillation showed that SFME is a quicker and more effective and environmentally friendly approach for extraction of oil from herbs (Filly et al., 2014).

The response surface methodology (RSM) is a commonly used mathematical and statistical method for modelling and evaluating a process in which multiple variables affect the response of interest (Aydar, 2018a). It has been successfully applied to determine MAE parameters in several studies (Dahmoune et al., 2015). Numerous solvents have been used for extraction of bioactive substances from this plant; however, no-solvent characterisation of such bioactives is yet to be studied in detail (Atique et al., 2018). In a previous study, we extracted bioactive compounds from the fresh fruit of *L. siceraria* without the use of any added solvent and the results were promising, which motivated us to design the present study (Ahmed et al., 2018). In a more recent study, we found that MAE was an efficient technique for extraction of antioxidant compounds from this plant using ethyl acetate as a solvent (Abbas et al., 2021). The present investigation is intended as a continuation to those studies, and our motivation in carrying it out is to evaluate the effect of microwave irradiation on the solvent-free extraction of bioactive substances from the bottle gourd fruit. Thus, the main purpose of this study was to develop a solvent-free efficient method for extraction of antioxidant compounds from *L. siceraria* assisted by microwave irradiation and to conduct extraction optimization under RSM using central composite design (CCD). Power and time were selected as independent variables (factors), each with three levels, and TPC, TFC and DPPH radical scavenging activity and iron chelating activity (ICA) as responses.

MATERIALS AND METHODS

Chemicals and equipment

The MAE was conducted with a microwave oven (model №. DW-162-HP, Dawlance, Lahore, Pakistan) having various control factors, with a maximum power of 1,200 W. Analytical grade chemical reagents were used for bioactivities. Folin-Ciocalteu reagent and sodium carbonate were obtained from Merck (Darmstadt, Germany). Iron sulphate-7-hydrate, ferrozine, iron chloride-6-hydrat, sodium acetate trihydrate, ascorbic acid and 1,1-diphenyl-2-picrylhydrazyl (DPPH) were purchased from Sigma-Aldrich (St. Louis, USA). 2,4,6-Tris-(2-pyridyl)-s-triazine (TPTZ) was purchased from Alfa-Aesar (Karlruhe, Germany).

Collection and processing of plant material

Fresh pieces of young fruit of *L. siceraria* were obtained from an agriculture farm in Pakpattan, Punjab, Pakistan in July 2019. They were washed and cut into pieces with a kitchen knife. The pieces were ground in a blender (Panasonic, Osaka, Japan). After blending, a paste-like material was used for extraction.

Microwave-assisted extraction and design of experiment

In this study, microwave power and extraction time were selected as independent factors and each factor had three levels based on our previous study (Abbas et al., 2021). The response factors included extraction yield, total phenolic content (TPC), total flavonoid content (TFC), DPPH radical scavenging activity, ferric reducing antioxidant potential (FRAP) and iron chelating activity (ICA).

The design of experiment (DoE) was created with the help of Design Expert software (version 11, Stat-Ease, Inc., Minneapolis, MN, USA). Response surface methodology (RSM) combined with CCD was used for determination of optimal operational conditions. For CCD, a total number of 11 designed runs of experimental conditions including three at centre points (480 W 60 s), four axial points and four corner points were selected. All experiments were carried out in triplicate and mean values were used for analysis. Factors and their coded levels for the CCD design are provided in Table 1. The responses were selected as percent extraction yield, DPPH radical scavenging activity, TPC, TFC and ICA. A total of 11 experiments were conducted to optimise the parameters. The experimental data were analysed to determine the best statistical model to fit (linear, quadratic, cubic or 2FI [two factor interaction]). Constant term, A and B (linear coefficients for power and time,

Table 1. Factors and their levels for the CCD design (α = 1.4142).

Factors	Symbols	Levels				
		−α	−1	0	1	+α
Microwave power (W)	A	330	360	480	600	630
Time (s)	B	18	30	60	90	102

CCD, central composite design.

respectively), AB (interactive term coefficients), and A^2 and B^2 quadratic term coefficients were used to represent the model's coefficients. The correlation coefficient (R^2), adjusted determination coefficient ($R^2_{adj.}$) and adequate precision were used to assess the model's adequacy; the model was regarded as adequate when its model *p*-value ≤ 0.05, R^2 was highest, root mean square error (RMSE) was lowest and adequate precision was <4. The goal of optimising multiple responses was to achieve maximum desirability. Various three-dimensional response surface plots were generated for each response using RSM.

Extraction procedure

The extraction was performed by using a funnel-over-funnel method. For this, 20 g *L. siceraria* fresh fruit ground material was weighed and transferred onto the filter paper placed in a glass funnel. The fruit paste was covered with another funnel. This setup was placed in a microwave oven and heated according to the run of the DoE. Followed by this, the filter paper was pressed to squeeze the extract out of it. The water present in the extract was evaporated in a rotary evaporator and the extract was removed from the rotary flask using ethanol (5 mL). Dried plant extract was obtained after evaporation of ethanol at room temperature. The percent extraction yield was calculated based on weight of ground fruit material used for extraction.

Total phenolic content

The TPCs of the *L. siceraria* extracts obtained from MAE were estimated by a reported method that uses Folin-Ciocalteu reagent (Ahmed et al., 2016). Reagents were prepared as per the reported methods. A measured amount (10 mg) of dried plant extract was dissolved in 10 mL distilled water to make the plant extract solution to attain a concentration of 1 mg · mL^{-1}. In a clean test tube, 60 µL (0.06 mL) plant extract sample was diluted by adding 4.7 mL distilled water. Then, 300 µL FC reagent was put into it. The mixture was incubated for 8 min. After 8 min, 900 µL of 20% sodium carbonate was added to it. This solution was kept at 40 °C for 30 min. After the incubation, absorption was measured at 765 nm wavelength using UV-Visible spectrophotometer (UVD-3200, Labomed Inc., Los Angeles, California, US) The TPC was calculated as milligrams gallic acid equivalents per gram of the dried extract. The blank solution contained everything except the plant extract. Instead of the plant extract, the same volume of distilled water was put into it.

Total flavonoid content

The TFC of the *L. siceraria* extract was measured based on a reported procedure (Ahmed et al., 2016). Reagents were prepared as per the reported procedure. The measured amount (10 mg) plant extract solution was solvated with 10 mL distilled water. In a clean test tube, 0.3 mL plant extract was placed. It was diluted by adding 30% aqueous methanol. Then, 150 µL of 0.5 M sodium nitrite ($NaNO_2$) followed by 150 µL of 0.3 M aluminium chloride ($AlCl_3$) were added. After an interval of 5 min, 1 mL of 1 M NaOH was added to this mixture. The absorption was measured at 506 nm using UV-Visible spectrophotometer. The TFC was quantified as milligram rutin equivalents per gram of the dried extract. The blank solution contained everything except the plant extract. Instead of the plant extract, the same volume of distilled water was added.

DPPH radical scavenging activity

Using the method reported in Ahmed et al. (2017), the DPPH assay was applied to quantify the antioxidant activity of *L. siceraria* extract obtained from MAE. Reagents were prepared as per the reported procedure. The concentration of the plant extract solution in distilled water was 1 mg · mL^{-1}. In a test tube, 4 mL DPPH working solution was mixed with 0.4 mL sample. The mixture was incubated at 37 °C for 30 min. Later, it was placed in a spectrophotometer and absorbance was noted at 517 nm. The control contained everything except the plant extract. Ascorbic acid was used as positive control and the antioxidant activity was expressed as percentage of inhibition, in accordance with the following formula:

$$\%\text{Activity} = [(\text{Ac} - \text{As})/\text{Ac}] \times 100$$

where Ac and As are the absorbance of the control and sample, respectively.

Ferric reducing antioxidant potential assay

This assay was conducted as per the protocol of Benzie et al. (1999). Ten milligrams of dried plant extraction was diluted with 10 mL of distilled water. In a test tube, 2.85 mL FRAP reagent was mixed with 150 µL plant extract solution. The mixture was incubated in the dark for 30 min. Absorbance was measured at 593 nm using UV-Visible spectrophotometer. Ascorbic acid was used as a standard and the FRAP was expressed as milligram ascorbic acid equivalent per gram DW.

Iron (metal) chelating activity

The iron(II) chelating activity of *L. siceraria* solvent-free extract was measured using a method reported by Ebrahimzadeh et al. (2008). The concentration of the plant extract solution in distilled water was 1 mg · mL^{-1}. In a test tube, 50 µL of $FeCl_2$ was placed, followed by the addition of 1 mL of plant extract. Then, 2 mL water was added. Thereafter, 0.2 mL of ferrozine was added to initiate the reaction. The solution was mixed well. After 10 min, the absorption was measured by using

UV-Visible spectrophotometer at 562 nm. The control contained everything except the plant extract. Instead of the plant extract, the same volume of EDTA was added. The ICA was expressed as a percentage, in terms of the following formula:

$$\% \text{ ICA} = [(Ac - As)/Ac] \times 100$$

where Ac and As are the absorbance of the control and sample, respectively.

Data analysis

Statistical analysis was carried out through Design Expert software (version 11) and the model fitness was evaluated by evaluating the coefficient of determination (R^2), lack-of-fitness model, ANOVA for linear model and F-value for each response.

RESULTS AND DISCUSSION

Percent yield of extraction

The results of percent yield obtained from *L. siceraria* are shown in Table 2. The highest yield of 4.02% was obtained at 600 W power and 90 s time. The lowest yield was observed at a power of 330 W and time of 60 s. The ANOVA results revealed that microwave power had a significant effect on yield ($p < 0.05$), whereas time had no effect on yield ($p > 0.05$). The yield increased with the increase in power and did not change significantly over time. An analysis of the effect of the independent factors on extraction yield showed a significant variation in percent yield resultant to varying the time and power during the extractions, and the model was significant with an R^2 value of 0.9935. The *p*-value of the model being <0.05 and the *p*-value of lack of fit being >0.05 also confirmed that the model is significant. The extraction yield was significantly affected by the linear effect of microwave time (B), power (A) and the quadratic effect of power (A^2). The response surface plot (Figure 1A) demonstrates that the extraction yield increased slightly over time, from 30 s to 90 s. This could be because microwaves increase the internal pressure of solid media and improve extraction; as a result, phenolic and antioxidant compounds can be leached in less time using microwaves than what would be possible with the use of traditional extraction methods (Aydar, 2018b). However, an increase in yield was observed while increasing power from 360 W to 600 W and a similar trend was further confirmed in the respective contour lines. The following coded and actual equation for the response of percent yield was established. These equations provide us with information concerning the response of every factor at each level.

$$\% \text{ Yield} = +1.50 + 1.34A + 0.1471B + 0.0975AB + 0.8723A^2 + 0.1014B^2$$

The regression equation for percent yield while considering only significant terms is written as:

$$\% \text{ Yield} = +1.49644 + 1.33722A + 0.872308A^2$$

Table 2. Solvent-free microwave-assisted extraction of bioactive compounds form *Lagenaria siceraria* fresh fruit: Independent and response factors along with the predicted values.

Run order	Power (W)	Time (s)	Extraction yield (%)		TPC (mg GAE · g^{-1})		TFC (mg RE · g^{-1})		FRAP (mg AAE · g^{-1})		DPPH radical scavenging (%)		ICA (%)	
			Actual	Predicted	Actual	Predicted	Actual	Predicted	Actual	Predicted	Actual	Predicted	Actual	Predicted
1	630.00	60	3.77	3.71	170.79	189.03	129.55	108.59	237.57	242.85	27.75	28.54	16.65	17.12
2	480.00	60	1.50	1.50	288.88	283.71	213.40	216.97	281.29	283.81	32.12	32.39	19.27	19.43
3	480.00	102	1.87	1.91	218.46	233.30	148.70	159.04	183.16	172.17	22.31	21.47	13.38	12.88
4	480.00	60	1.50	1.50	286.83	283.71	214.10	216.97	289.67	283.81	32.96	32.39	19.27	19.43
5	360.00	30	1.23	1.08	193.16	194.96	96.06	110.74	261.57	244.61	29.15	28.18	17.49	16.91
6	360.00	90	1.30	1.18	250.79	241.13	143.91	143.97	153.97	159.18	20.39	21.04	12.23	12.62
7	600.00	30	3.56	3.56	200.81	196.99	80.13	103.01	207.03	192.65	25.70	24.49	15.42	14.69
8	330.00	60	1.00	1.19	202.72	208.31	115.86	112.20	214.60	222.96	26.46	26.70	15.87	16.02
9	600.00	90	4.02	4.05	151.22	135.93	61.15	59.80	198.88	206.67	23.88	24.28	14.33	14.57
10	480.00	18	1.41	1.49	246.07	244.68	191.36	167.20	203.56	224.13	25.35	26.81	15.21	16.08
11	480.00	60	1.50	1.50	285.72	283.71	211.64	216.97	285.34	283.81	32.39	32.39	19.52	19.43

DPPH, 1,1-diphenyl-2-picrylhydrazyl; FRAP, ferric reducing antioxidant potential; GAE, gallic acid equivalent; ICA, iron chelating activity; TFC, total flavonoid content; TPC, total phenolic content.

Total phenolic content

The results of TPC are shown in Table 2. The maximum content obtained was 288.88 ± 0.016 mg GAE · g^{-1} at 480 W and 60 s, and the least was 151.22 ± 0.055 mg GAE · g^{-1} extract at 600 W and 90 s. Increasing microwave power can improve extraction efficiency by boosting molecular interactions between the electromagnetic field and the sample. Longer exposure to a higher microwave power, on the other hand, may destroy some phenolic compounds (Ismail-Suhaimy et al., 2021). In the present study, lower TPC was achieved corresponding to microwave power >480 W and time >60 s. It appears that a lower power is not efficient for extraction of phenolics while a higher power may result in the degradation of these substances (Lovrić et al., 2017; Abbas et al., 2021). Since the *p*-value of lack of fit was significant ($p < 0.05$), there is no model well-fitted to TPC. As a result, the discrepancy between predicted and actual values in TPC was slightly larger than those in other well-fitting responses such as yield, DPPH radical scavenging activity and ICA. The microwave irradiation had an enormous effect on the no-solvent extraction of phenolics from the fruit of *L. siceraria*, as can be surmised from the fact that the results of the present study are highly enhanced in comparison with those of a previous study carried out without the aid of microwaves (Ahmed et al., 2018). The no-solvent MAE in the present study also showed higher TPC as compared to the heat-dried MAE discussed in the study of Abbas et al. (2021), in which ethyl acetate was used as a solvent.

Total flavonoid content

The results of TFC (milligram rutin equivalent per gram extract) obtained from *L. siceraria* fruit are shown in Table 2. The maximum content obtained was 214.10 ± 0.318 mg RE · g^{-1} extract and the least was 61.15 ± 0.017 mg RE · g^{-1}. Similar to the TPC, the differences between predicted and observed values of TFC were higher since its model was inadequate. Moreover, the optimum power and time for TFC were also, respectively, 480 W and 60 s, which is another similarity with TPC. The trend might be due to the same reasons explained in the case of TPC. Heating the biomass to extract bioactive compounds from it is important as it assists their release from the biomatrix. However, higher heating conditions may break up the flavonoids or other polyphenolic biomolecules. Thus, it is desirable to explore the optimum heating conditions required for extraction of these compounds without damaging them (Routray and Orsat, 2021).

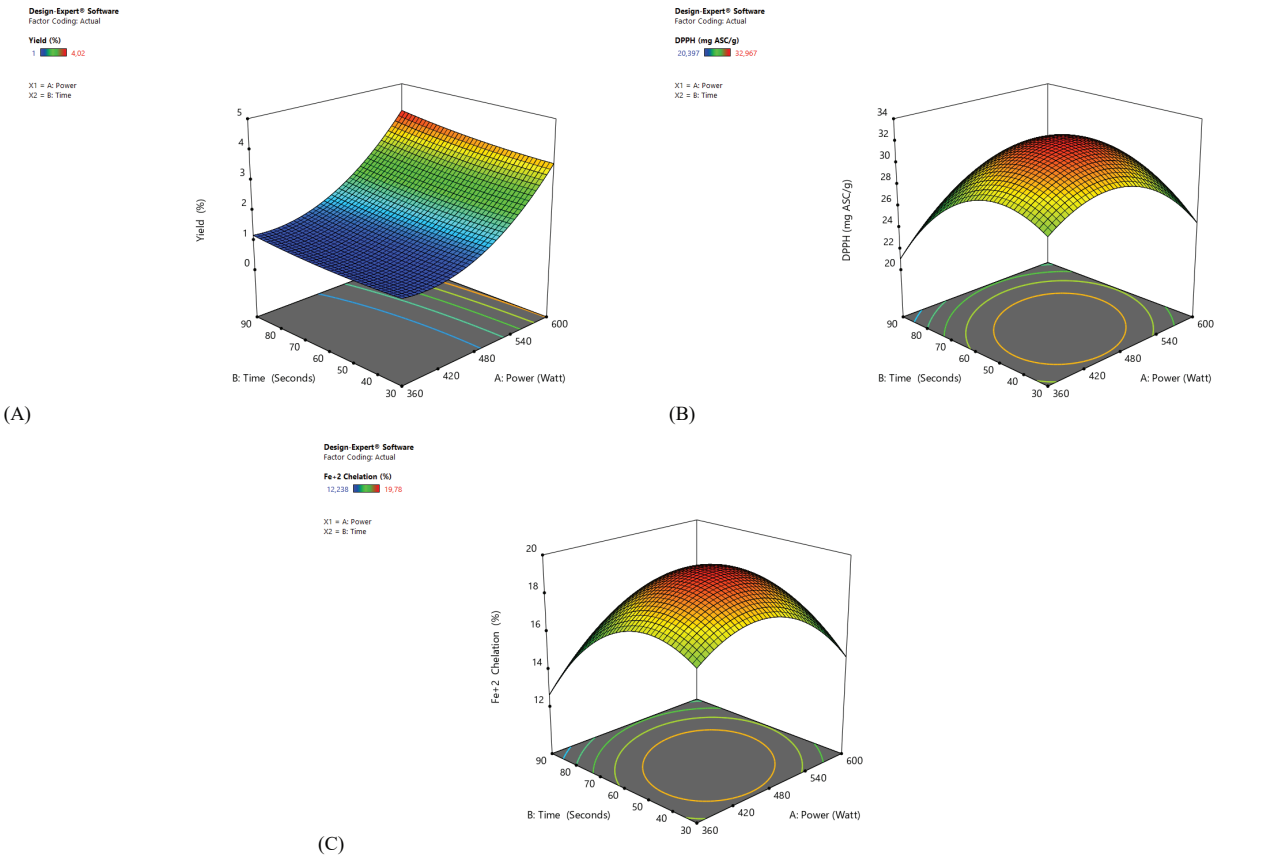

Figure 1. 3D surface plots for yield (A), DPPH (B) and ICA (C). DPPH, 1,1-diphenyl-2-picrylhydrazyl; ICA, iron chelating activity.

The no-solvent MAE in the present study also showed higher TFC as compared to the heat-dried MAE for which ethyl acetate was used as a solvent (Abbas et al., 2021).

DPPH radical scavenging activity

The percentage anti-radical or DPPH radical scavenging activity values of *L. siceraria* solvent-free extract obtained using MAE technique are displayed in Table 2. The highest anti-radical activity was 32.967 ± 0.042% and the least was 20.397 ± 0.021%. Similar to TPC and TFC, the optimum power and time for DPPH radical scavenging activity were also 480 W and 60 s, respectively. The observation may be explained based on the fact that although antioxidant activities may be due to substances other than polyphenols, the latter contribute considerably to these activities (Kainama et al., 2020). Table 3 illustrates that the DPPH radical scavenging activity was significantly influenced by the linear effect of microwave time (B), interactive effect of microwave power and microwave time (AB), quadratic effect of microwave power (A^2) and time (B^2). DPPH radical scavenging activity of *L. siceraria* was decreased as microwave time increased (Figure 1B). The lower DPPH radical scavenging activity of the *L. siceraria* extracts obtained when the microwave time was >60 s might be due to the degradation of some antioxidant compounds.

Table 3. Regression coefficients for different responses.

Coefficients	Extraction yield	DPPH	ICA
Model	Quadratic	Quadratic	Quadratic
Intercept	1.49644	32.3854	19.431
A	1.33722**	−0.11279	−0.06789
B	0.147079	−1.83707*	−1.10224*
AB	0.0975	1.73625*	1.04175*
A^2	0.872308**	−3.73041*	−2.23822*
B^2	0.10136	−4.15794**	−2.49495**
Lack-of-fit *p*-value	0.051	0.0752	0.0753
Model *p*-value	0.0001	0.0006	0.0016
R^2	0.9935	0.9610	0.9610
$R^2_{adj.}$	0.9870	0.9220	0.9220
RMSE	0.0001	0.4190	0.2515
PRESS	2.11	46.94	16.90
CV (%)	11.95	4.06	4.06
Adequate precision	16.4340	14.2238	14.2236
F-value	41.29	28.15	28.15
BIC	6.09	39.09	27.86
AICc	24.70	57.71	46.47

*Significant (<0.05).
**Significant (<0.001).
DPPH, 1,1-diphenyl-2-picrylhydrazyl; ICA, iron chelating activity; RMSE, root mean square error; PRESS, predicted residual error sum of squares; CV, coefficient of variation; BIC, Bayesian information criterion; AICc, Akaike information criterion.

The regression equation for DPPH radical scavenging activity while considering the significant terms is given below:

$$\text{DPPH (\%)} = 32.3854 - 1.83707B + 1.73625AB - 3.73041A^2 - 4.15794B^2$$

Ferric reducing antioxidant potential

The FRAP outcomes of *L. siceraria* solvent-free extracts obtained through MAE are given in Table 2. The maximum FRAP value obtained was 289.67 ± 0.042 mg AAE · g^{-1} at 480 W and 60 s, while the minimum was 153.97 ± 0.021 mg AAE · g^{-1} at 360 W and 90 s. Even though the *p*-value of the model was significant with $p < 0.05$, the *p*-value of lack of fit was <0.05. Thus, no model was found to fit to FRAP results similar to TPC and TFC.

Iron (metal) chelating activity

The outcomes of iron metal chelating activity of *L. siceraria* solvent-free extract obtained through MAE are displayed in Table 2. The highest and the lowest metal chelating activities of the extracts were 19.520 ± 0.025% and 12.238 ± 0.012%, respectively. Similar to other bioactivities, the optimum power and time for ICA were 480 W and 60 s, respectively. The influence of independent factors on ICA is exhibited in Table 3. The *p*-value of the model, 0.0016, and the *p*-value of lack of fit, 0.0753, show that the model is significant. The *p*-value <0.05 shows that the model terms are significant. The data displayed in Table 3 demonstrate that the metal chelating activity was significantly influenced by the linear effect of microwave time (B), interactive effect of microwave power and microwave time (AB), quadratic effect of microwave power (A^2) and time (B^2). Figure 1C confirms that a greater content of ICA can be obtained by maintaining the extraction power and time at around 480 W and 60 s, respectively. The regression equation for ICA without considering the non-significant term is given below:

$$\text{ICA (\%)} = 19.431 - 1.10224B + 1.04175AB - 2.23822A^2 - 2.49495B^2$$

Validation of models

Experiments were carried out to validate regression models under optimum conditions, which were 480 W and 60 s for microwave power and time, respectively. Table 4 shows the predicted and observed means for responses in optimum extraction conditions. For yield, DPPH radical scavenging activity and ICA, the error rates between predicted and observed values were −0.74%, 1.75% and 0.46%, respectively. Since no well-fitting model for TPC, TFC and FRAP activity was observed, optimization and validation were not performed on these responses. The notable finding, however, is that 480 W power and 60 s time were the most efficient for all the bioactivities

Table 4. Predicted and observed means of responses in optimum extraction conditions.

Optimum conditions		Responses	Predicted mean	Observed mean	Error rate (%)
Power (W)	Time (s)	Extraction yield (%)	4.05	4.02	−0.74
480	60	DPPH	32.39	32.96	1.75
		ICA	19.43	19.52	0.46

DPPH, 1,1-diphenyl-2-picrylhydrazyl; ICA, iron chelating activity.

determined in this study. These factors can fairly be recommended for a large-scale extraction of bioactive substances from *L. siceraria* fruit using microwave-assisted no-solvent extraction process. As noted above, ANOVA results for TPC, TFC and FRAP activity revealed that they were not well-fitted to any model since lack-of-fit *p*-values were <0.05 and owing to low lack-of-fit F-values. This indicated that the models could not be used to predict the response. In modelling, for a model to be significant, the model *p*-value should be <0.05 and lack-of-fit *p*-value should be >0.05.

The method proposed in the current study is green as it uses no solvent for extraction and microwave irradiation is required only for a short period of time. The same reasons also make the method fast as well as cost effective. Keeping in view the biological benefits of bottle gourd fruit, the proposed strategy can effectively be used for extraction of antioxidant biomolecules from it in a green, safe, fast and cost-effective way (Amin et al., 2022).

CONCLUSIONS

The aim of this research was to explore a viable green method for extraction of chemical compounds from *L. siceraria* using a microwave-assisted solvent-free extraction. An optimum extraction of bioactive compounds including polyphenolics and antioxidants occurred at 480 W and 60 s. In addition, the time economy was one of the most prominent features of MAE technique. The extraction of bioactive compounds without the use of any added organic solvent makes the method further environmentally friendly. The models were able to produce good predictions for extraction yield, DPPH radical scavenging activity and metal chelation activity with low error rates (<2%), as discussed in the evaluation section. In the future, the method can be further developed into an efficient process for the extraction of chemical compounds on a large scale.

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AUTHOR CONTRIBUTIONS

D.A. designed the research and supervised it, and contributed in manuscript writing. S.I. suggested the methodology, performed the literature review and wrote the manuscript. M.T.Q. carried out data interpretation and statistical analysis. A.Y.A. carried out optimization analysis and helped in data interpretation. All the authors approved the final version.

CONFLICT OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have exerted an influence on the study reported in this paper.

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