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Research Article

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Extraction optimization of total flavonoids and antioxidant activities from *Suaeda glauca* Bge leaves

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ABSTRACT

The total flavonoids from Suaeda glauca Bge leaves were studied using ultrasound-assisted extraction (UAE) in this work, and total flavonoids content (TFC) was measured by UV-spectrophotometric method. The inhibiting effect (DPPH%), reducing power, and inhibiting effect of hydroxyl radical (OH%) of sample extract was also investigated. Three important factors with regard to temperature (30-80°C)，time (10-60 min) and liquid-solid ratio (5-50 ml·g^{-1}) were optimized using RSM for obtaining maximum values of response function. The results showed that the optimal extraction condition was at 45.28 ℃ of extraction temperature, 53.91 min of extraction time, and 30.34 ml·g^{-1} of liquid-solid ratio, respectively. Under those conditions, TFC, DPPH%, reducing power and OH% were found to be 121.0 mg RE·g^{-1}, 94.67%, 740.07 mg AA·g^{-1} DW, and 90.91%, respectively. These results demonstrated that the regression model was obtained, and be capable of accurately estimating the total flavonoids from Suaeda glauca Bge leaves in the field of the deep processing of Suaeda glauca Bge resources, as well as desirable anti-DPPH radical activity, reducing power and anti-hydroxyl radical activity. In addition, high linear correlation between TF and OH% indicated inhibition of hydroxyl radical was attributed to flavonoids compounds.

Keywords: *Suaeda glauca* Bge, Total flavonoids, Reducing power, Response surface methodology, Ultrasound-assisted extraction

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INTROUDUCTION

Natural antioxidants can be more effective to treat chronic renal disease such as cancer, diabetes, aging, heart and degenerative diseases etc[1]. Antioxidant therapy might be useful in preventing or delaying the progression of these diseases, but a major mechanism from oxidative damage is still not clear. The recent studies [2, 3] indicated that free radicals have considerable evidences inducing oxidative damage. Consequently, an extensively study of natural antioxidants have been more frequently undertaken than synthetic antioxidants [4-6].

Suaeda glauca Bge leaves are selected as the object of study, which grows on the seashore or salt flats and belongs to *Suaeda glauca* family. It is a well-known and very important traditional Chinese drug, and often used for treating diarrheas, fevers, bad digestion, etc [7]. Also, It possess many nutrition ingredients, consequently considered as a green food and is edible as a good vegetable for human and animal feed [8]. Although it reveals a potential in the therapy of influenza, Information is scanty in available literatures regarding total flavonoids contents, antiradical capacity and reducing power in *vitro*. The primary aim of this study was to extract total flavonoids and research antiradical capacity and reducing power, and further optimize the extraction process to acquire a good performance process. This is helpful for future deep investigation and development of *Suaeda glauca* Bge in the efficacy of herbal therapies.

EXPERIMENTAL SECTION

Chemicals and plant materials

Aluminium trichloride, potassium ferricyanide, trichloroacetic acid, 1,1-diphenyl-2-picryl hydrazyl (DPPH), ferric chloride, ascorbic acid, sodium salicylate, H_2O_2 were of analytical grade and purchased from Sinopharm Chemical Reagent Co. (Shanghai, China). Rutin was purchased from from Beijing Yingze New Chemical Technology Research Institute Beijing, China).

Preparation and of extract

Suaeda glauca Bge leaves were picked from Dafeng seawall (YanCheng, JiangSu, China) in 2013 March. It was cleaned and dried in a drying oven in the laboratory until a constant weight. It was pulverized by the Joyoung soybean machine (DJ13B-D18D, Shandong, China) into powder, and then sealed in plastic bottles for further analysis after 40 mesh sieving.

According to RSM design (Table 1), *Suaeda glauca* Bge leaves powder was accurately weighed with 4.0 g into the flask, mixed with water of a certain liquid-solid ratio range 5-50 ml·g^{-1}, which was placed into the ultrasonic cleaner for extraction time (10-60 min) at the temperature range 30-80°C, followed by centrifugation and take the supernatant for the quantitative determination of total flavonoids content (TFC) and antioxidant activities after quantitative dilution.

Analysis of total flavonoids content (TFC) and antioxidant activities

Determination of total flavonoids

TFC of extracts were determined in triple by the technique described in the paper [9]. Exactly 2 ml of plant extract solution was mixed with 2 ml of aluminium trichloride (2%) in methanol. The mixture was placed at room temperature for 10 min, and the absorbance was measured using a UV-spectrophotometer (SPECORD-50, Germany Jena Co, Ltd.) at 415 nm. The mixture solution without extract was used as the blank. Rutin was used as equivalents (RE) for making the standard curve (Absorbance=0.5716 RE mg/ml-0.0839, R^2=0.9991). TFC in extracts was expressed as mg RE per gram dry weight (DW) of plant. The results were mean values±standard error. TFC in *Suaeda glauca* Bge leaves extracts using rutin as equivalents (RE) was calculated by the following equations:

$$\text{TFC}\ \left(\text{mg RE}\cdot\text{g}^{-1}\right)=\frac{[(y+0.0839)/0.5716]\times V}{\text{mass of sample}}\times 100 \quad (1)$$

V was the volume of solution (ml), *y* was the absorbance of the extracts and mass of sample refer to plant powder studied (g).

Determination of reducing power

The total antioxidant potential of the extracts was researched further by reducing power analysis which is based on a relox reaction. In the reaction system, an ferric ions in excess is easily reduced by antioxidants. The reducing power was investigated according to the method [10]. Briefly, Sample extracts of 70% ethanol was actually taken for 0.5, 1.0, 1.5, 2.0, 2.5, 3.0,3.5 ml into a flask of 10 ml, respectively, then was mixed with phosphate buffer (1ml, 0.2 mol/L, pH 6.6) and 1 ml of potassium ferricyanide (1%). The mixture was incubated for 20 min at 50°C. The mixture was added by 1 ml of trichloroacetic acid (10%), further centrifuged at 1000 rpm for 10 min. The upper layer of solution (1 ml) was diluted with distilled water (2 ml) and Ferric chloride (0.1%) for 0.4 ml. The absorbance of the final solution was measured at 760 nm. Blank was prepared with all the reaction agents without extract. Ascorbic acid (AA) was used as a standard. The reducing power of extracts was expressed as mg AA per gram DW according to the calibration curve: y = 0.0116x, R^2 = 0.9786, where x was the absorbance and y was reducing power mg AAE·g^{-1} DW.

Assay of DPPH radical scavenging activity

The scavenging activity of 1,1-diphenyl-2-picryl hydrazyl (DPPH) was analyzed according to a method [11-12]. The extract were added into a methanolic solution of DPPH radical (0.5 mM), then the mixtures were vigorously shaked and allowed to stand for 30 min at 25°C in the dark. The absorbance at 517 nm from the resulting solution was scanned using a spectrophotometer against a blank sample without DPPH. The scavenging capacity of the DPPH radical was calculated by using the formula below:

$$\text{DPPH}(\%)=\frac{\text{A}_0\text{-A}_\text{s}}{\text{A}_0}\times 100 \quad (2)$$

A_0 is the absorbance of DPPH solution without sample and A_s is the absorbance of the extract studied.

Assay of hydroxyl radicals scavenging activity

The hydroxyl radical scavenging activity was studied according to the previous method [13, 14]. The extract sample was accurately taken to 10 ml flask at the volume of 0.5, 1, 1.5, 2.0, 2.5, 3.0, 3.5 and 4 ml, respectively. Then 1 ml $FeSO_4$ (1.5 mM), 0.3 ml of sodium salicylate (20 mM) were also added into the above extract sample solution. Meanwhile, the mixture was reacted with 0.7 ml of H_2O_2 (6 mM). The reaction solution was incubated for 1 h at 37°C. The absorbance at 562 nm was measured. The capability of scavenging hydroxyl radical of extract samples were calculated by the following equation:

$$\mathrm{OH}(\%) = \frac{\mathrm{A}_0\text{-}\mathrm{A}_s}{\mathrm{A}_0} \times 100 \tag{3}$$

A_0 was the absorbance of the control (without sample) and A_s was the absorbance of the extract samples.

Optimization ultrasonic-assisted extraction by response surface method (RSM)

To further study the interaction between the variables, we optimized the experimental conditions by Box-Behnken design (BBD) of RSM using Design-Expert 8.0.7.1 [15, 16]. BBD with three factors was applied to set the three variables containing temperature for 30-80°C, ultrasonic time for 10-60 min, and liquid–solid ratio for 5-50 $ml \cdot g^{-1}$. The factors and levels were shown in Table 1. The actual result was obtained by experimental operation at each team condition by UAE. The data was analyzed by RSM, then the fitting model was established, and the optimal conditions were got. The validate test was carried out in triple to determine the repeatability of the model. The three samples of the same weight (0.5 g) were performed under the resulting optimal conditions.

Statistical analysis

RSM was used to analyze the effect of three factors including extraction temperature (X_1); extraction time (X_2) and solid-liquid ratio (X_3) on the response functions of TF (Y_1), DPPH% (Y_2), reducing power (Y_3) and OH% (Y_4) of the extract. A Box-Behnken Design (BBD) from RSM was employed to design the experimental arrangement. The software package of Design-Expert 8.0.7.1 was applied to design variables with actual and coded levels along with response variables. The experimental design included twelve factorial points, and five repeated points. These values were calculated according to regression model related to the three variables in a second-order polynomial equation.
Statistical significance of coefficients in the regression equations was examined and analysis of variance (ANOVA) was carried. A general function formula is used below:

$$Y = b_0 + \sum_{i=1}^{n} b_i X_i + \sum_{i<j}^{n} b_{ij} X_i X_j + \sum_{i=i}^{n} b_{ii} X_i^2 \tag{4}$$

Y is the response function, b_0, b_i, b_{ij}, b_{ii} are regression coefficients of the constant term, linear coefficient, the cross coefficient and quadratic coefficient, respectively. According to equation (4), four functions of this study were expressed as equation (5):

$$\begin{aligned} Y_{\mathrm{k}} &= b_0 + b_1 X_1 + b_2 X_2 + b_3 X_3 + b_{12} X_1 X_2 + b_{23} X_2 X_3 + b_{13} X_1 X_3 \\ &\quad + b_{11} X_1^2 + b_{22} X_2^2 + b_{33} X_3^2 \end{aligned} \tag{5}$$

Lack of fit, correlation coefficients R^2 and adjusted-R^2 were also analyzed. When R^2 is up to 0.80, it was considered to be a good fitness of a response model. The initial models can be adjusted by removing some non-significant terms (quadratic terms or interaction terms) so that the corresponding variables were to be more significant ($p < 0.05$). So the final model was determined. All tests were performed in triplicate.

RESULTS AND DISCUSSION

Effect of extraction parameters on TF

The experimental design was presented in Table 1. According to 17-run experimental parameter, the results were also showed in Table 1. Figure 1a, b and c showed the response surface of the effect of extraction conditions of temperature, time and liquid-solid ratio on TFC by UAE technique. Table 2 showed analysis of the variance of regression coefficients of each response variable in the regression model. Among the influence factors, liquid-solid ratio revealed the significant ($p<0.05$) linear and quadratic effects. Higher liquid-solid ratio could have higher solubility of active compounds including flavonoids compounds and other antioxidants to the solvents. With the

increase of temperature, the target compound solubility increased because of the increase of mass transfer and solvent diffusion rate. The extraction time also had a significant (p<0.05) effect at linear level. Meanwhile, the increased extraction time enhanced the exchange of solvent penetration and active compositions. The highest TFC (124.92 mg RE·g^{-1}) was optimized under the extraction parameters of 45.28°C, extraction time for 53.91min and 30.34 ml·g^{-1} of liquid-solid ratio by RSM.

Table 1 Experimental design and results of TPC, DPPH%, reducing power and OH% of the *Suaeda glauca* Bge Leaves extract by UAE technique

No.	X_1 /℃	X_2 / min	X_3 /(ml·g^{-1})	Y_1/(mg RE·g^{-1})		Y_2/%		Y_3/ (mg AA·g^{-1} DW)		Y_4/%	
				Actual*	predicted	Actual*	predicted	Actual*	predicted	Actual*	predicted
1	30	10	27.5	108.4±0.11	10.72	90.21±0.04	90.23	580.31±0.01	579.42	84.64±0.02	84.37
2	80	10	27.5	103.9±0.09	10.27	92.96±0.02	93.10	686.55±0.03	683.50	86.56±0.03	86.02
3	30	60	27.5	114.9±0.06	11.61	95.33±0.01	95.19	749.78±0.02	752.83	92.44±0.02	92.98
4	80	60	27.5	123.5±0.03	12.47	95.08±0.02	90.06	723.16±0.04	724.05	90.73±0.01	91.00
5	30	35	5	75.3±0.42	7.53	87.94±0.02	88.22	563.75±0.02	571.11	79.41±0.04	80.12
6	80	35	5	75.7±0.35	7.57	87.57±0.05	87.72	621.19±0.03	630.71	79.42±0.03	80.40
7	30	35	50	83.5±0.04	8.35	91.84±0.03	91.69	689.89±0.01	680.37	85.67±0.03	84.69
8	80	35	50	87.4±0.03	8.74	90.22±0.01	89.94	703.44±0.02	696.08	84.79±0.01	84.08
9	55	10	5	62.0±0.25	6.32	90.45±0.02	90.16	492.06±0.04	485.59	72.58±0.02	72.14
10	55	60	5	79.1±0.20	7.79	90.63±0.04	90.50	695.51±0.01	685.10	81.73±0.02	80.49
11	55	10	50	71.2±0.14	7.24	92.25±0..2	92.38	655.03±0.03	665.44	76.58±0.02	77.83
12	55	60	50	89.9±0.07	8.87	93.67±0.02	93.96	673.42±0.03	679.89	82.61±0.03	83.05
13	55	35	27.5	120.2±0.14	11.98	95.51±0.04	95.06	740.91±0.02	731.42	92.79±0.03	91.06
14	55	35	27.5	121.7±0.16	11.98	95.37±0.01	95.06	729.68±0.01	731.42	91.73±0.04	91.06
15	55	35	27.5	117.8±0.21	11.98	94.99±0.01	95.06	733.85±0.03	731.42	91.43±0.04	91.06
16	55	35	27.5	119.1±0.17	11.98	94.14±0.03	95.06	716.79±0.03	731.42	87.76±0.01	91.06
17	55	35	27.5	120.4±0.15	11.98	95.29±0.03	95.06	725.89±0.03	731.42	91.57±0.01	91.06

** Data was expressed as mean ±S.D. (n=3) .*

X_1 (Temperature, ℃), X_2 (Extraction time, min), X_3 (Liquid-solid ratio, ml·g^{-1}), Y_1 (TFC, (mg RE·g^{-1})), Y_2 (DPPH, %), Y_3 (Reducing power, mg AA·g^{-1} DW), Y_4 (OH, %).

Table 2 Analysis of the variance of regression coefficients of each response variable in the regression model

Source	Df	TFC		DPPH%		Reducing power		OH%	
		Sum of squares	p-value	Sum of squares	p-value	Sum of squares	p-value	Sum of squares	p-value
Model	9	7220.4758	< 0.0001[a]	110.5382	< 0.0001[a]	82895.4832	<0.0001[a]	568.1755	0.0003[a]
X_1	1	8.8200	0.1253[b]	2.5200	0.0137[a]	2835.4215	0.0052[a]	0.0544	0.8987[b]
X_2	1	478.9513	< 0.0001[a]	1.8432	0.0267[a]	22889.4408	<0.0001[a]	92.1403	0.0010[a]
X_3	1	199.0013	< 0.0001[a]	16.2165	< 0.0001[a]	15248.6916	<0.0001[a]	34.0725	0.0131[a]
X_1X_2	1	42.9025	0.0064[a]	16.0000	< 0.0001[a]	4412.9449	0.0016[a]	3.2942	0.3385[b]
X_1X_3	1	3.0625	0.3392[b]	0.3906	0.2392[b]	481.5830	0.1437[b]	0.1980	0.8084[b]
X_2X_3	1	0.6400	0.6534[b]	0.3844	0.2426[b]	8561.8009	0.0002[a]	2.4336	0.4066[b]
X_1X_1	1	5.2817	0.2200	29.2624	< 0.0001[a]	1005.8114	0.0489[a]	2.3182	0.4174[b]
X_2X_2	1	153.8612	0.0002[a]	0.3272	0.2775[b]	4051.0814	0.0020[a]	43.2641	0.0074[a]
X_3X_3	1	6158.6527	< 0.0001[a]	38.6883	< 0.0001[a]	21465.5457	<0.0001[a]	378.0425	<0.0001[a]
Residual	7	20.3795		1.6520		1243.4671		21.8578	
Lack of fit	3	11.7675	0.2831[b]	0.4492	0.7033[b]	610.0568	0.3938[b]	7.1291	0.6255[b]
Pure error	4	8.6120		1.2028		633.4103		14.7287	
Cor total	16	7240.8553		112.1902		84138.9504		590.0333	
R^2			0.9972	0.9853		0.9852		0.9630	
Adj R^2			0.9936	0.9663		0.9662		0.9153	
Pred R^2			0.9721	0.9192		0.8722		0.7677	

[a] 5% significant level; [b] Not significant relative to the pure error.

Effect of extraction parameters on DPPH%

The Figure 1d, e and f indicated the influence of temperature (30-80°C), time (10-60 min) and liquid-solid ratio (5-50 ml·g^{-1}) on DPPH% of *Suaeda glauca* Bge leaves extracted by UAE. The liquid-solid ratio presented remarkably significant (p<0.05) linear and quadratic effects on DPPH%. By increasing the liquid-solid ratio, the target components of inhibiting DPPH radical have full dissolution related to increasing concentration gradient which is driving force in consistent with mass transfer principles. At 30.34 ml·g^{-1} of liquid-solid ratio, DPPH% reached to the maximum value 95.99% optimized by RSM. The temperature showed rapid increase at the beginning of the extraction corresponding with the dissolution of soluble components. Then the slow decrease of DPPH% was observed related to degradation of active ingredients because of higher temperature. The optimal temperature was obtained for 45.28℃, in a case, giving the equilibrium concentration of the target compositions in the extract. The time effect was found different tendency to temperature and liquid-solid ratio. It always revealed steady increase of DPPH% from 10 min to 60 min. However, the longer time for higher DPPH% was obviously impractical performance from the economical point of view.

Effect of extraction parameters on reducing power

The reducing power may serve as a significant indicator of potential antioxidant activity because of a direct correlation between reducing power and antioxidant activities of extracts. Three factors had basically a significant effect on reducing power activity of the extract from *Suaeda glauca* Bge leaves. But liquid-solid ratio revealed strongest effect. As shown in Figure 1h and i, an extreme value was found at liquid-solid ratio for 30.34 $ml \cdot g^{-1}$ at the extraction temperature for 45.28°C, extraction time for 53.91min after optimization by RSM. The temperature and time seemed to presented the same tendency for reducing power activity from the extract, which was the increase of reducing power with temperature and time (see Figure 1g), while extraction time also showed a significant (p<0.05) linear and quadratic effect on reducing power.

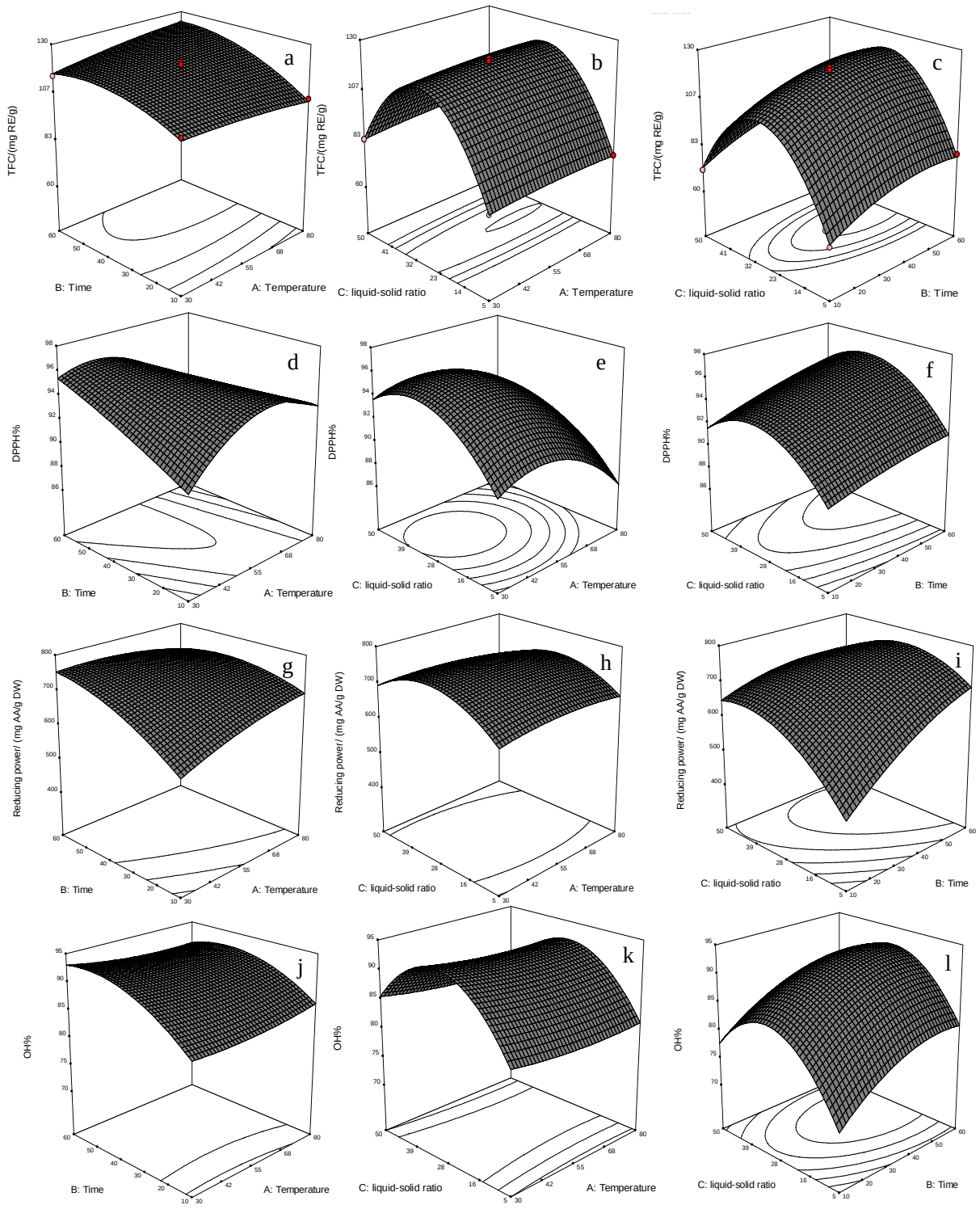
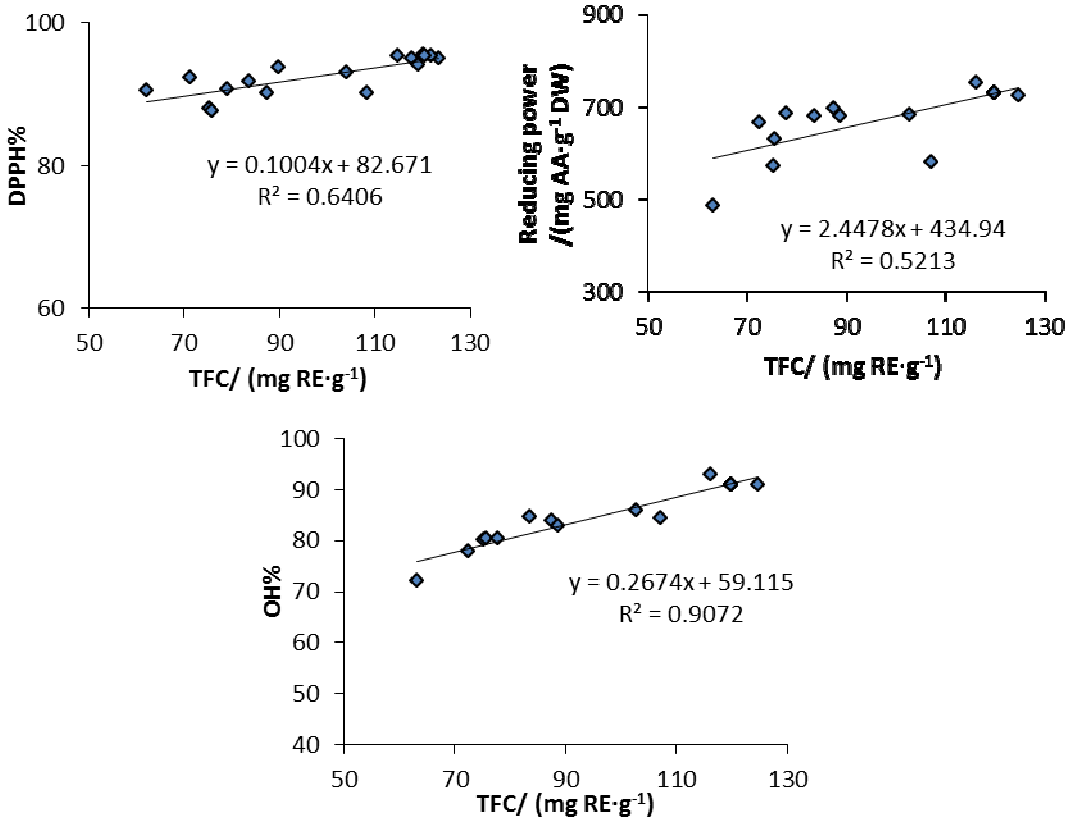


Figure 1 Response surface for the effect of temperature, time and liquid-solid ratio on TFC, DPPH%, reducing power and OH%

Figure 2 The correlation of between TFC and DPPH% (Reducing power, OH%)



Effect of extraction parameters on OH%

As the most reactive free radicals, hydroxyl radicals can react with almost all the bio-macromolecules, resulting in damage to the adjacent biomolecules in living systems. Thus, inhibition and elimination of it is important. As we all know, the liquid–solid ratio enhances the extraction efficiency of active components by exposing more surface area of the solid sample to the solvent. As seen in Figure. 1k and l, OH% had gradually increased to 30.34 ml·g^{-1} as the increase of liquid-solid ratio, then achieved to highest value (92.25%) when the solid sample was completely immersed and anti-hydroxyl radical component was fully extracted in the case of reaching the biggest contact area. However, when the liquid-solid ratio was higher than 30.34 ml·g^{-1}, OH% was decreased. This might be the reason that dilution of the active compositions due to superfluously high ratio led to lower OH% for lower concentration. In spite of this, the high correlation coefficients (r^2=0.9072) was found between TFC and OH% (see Figure 2), followed by the relationship between TFC and DPPH% (r^2=0.6406), TFC and reducing power (r^2=0.5213). This implied that the task of removing hydroxyl radical was mainly achieved by flavonoids compounds in this study.

Regression model

The independent factors including variables extraction temperature, extraction time and liquid-solid ratio were optimized as variables of TFC, DPPH%, reducing power and OH%. The regression model involving linear, quadratic and interaction terms was built up by RSM. The statistical significance of the terms, regression analysis and ANOVA were carried out. The result in Table 1 was the predicted values and actual value of four response functions. They should be in accordance with a polynomial model obtained in data processing using RSM software. The ANOVA results from Table 2 indicated the four regression models were highly significant (p<0.01). Each predicted response function could be obtained by the following polynomial equations（6-9）:

$$Y_1 = 39.3255 + 0.01294X_1 + 0.67878X_2 + 4.26623X_3 + 0.00524X_1X_2 + 0.00071X_2X_3 + 0.00156X_1X_3 - 0.00179X_1^2 - 0.00967X_2^2 - 0.07555X_3^2 \quad (6)$$

$$Y_2 = 69.57881 + 0.56881X_1 + 0.21126X_2 + 0.40387X_3 - 0.00320X_1X_2 + 5.51111X_2X_3 - 0.00056X_1X_3 - 0.00422X_1^2 - 4.46X_2^2 - 0.00599X_3^2 \quad (7)$$

$$Y_3 = 108.52699 + 5.86974X_1 + 10.79841X_2 + 13.64909X_3 - 0.05314X_1X_2 - 0.08225X_2X_3 - 0.01951X_1X_3 - 0.02473X_1^2 - 0.04963X_2^2 - 0.14104X_3^2 \quad (8)$$

$$Y_4 = 62.38959 - 0.07219X_1 + 0.61276X_2 + 1.19145X_3 - 0.00145X_1X_2 - 0.00139X_2X_3 - 0.00041X_1X_3 + 0.00118X_1^2 - 0.00513X_2^2 - 0.01870X_3^2 \quad (9)$$

In Eq. (6), the correlation coefficients R^2 was 0.9972, implying that the sample variation of 99.72% for TFC was attributable to the three independent variables. The adjusted correlation coefficient ($AdjR^2$=0.9936) was used for analyzing the fitting correlation of the regression equation. The higher it is the better fitting between the actual and predicted values. The R^2 and $AdjR^2$ for Eq. (7) was 0.9853, 0.9663, respectively, which showed that there was 98.53% of total variation from three variables in this test. The R^2 and $AdjR^2$ for Eq. (8-9) were, 0.9852, 0.9630 and 0.9662, 0.9153, respectively. The value of R^2 is higher than 0.9, so these models were goodness-of-fit and adequacy, and capable of elucidating the relationships between three factors. Meanwhile, each of lack of fit was also not significant (p>0.05). All these good information further strengthened the reliability of the models.

Optimization conditions and Verification experiment

The optimization procedures were performed for predicting the optimal level of three factors to obtain maximum values of TFC, DPPH%, reducing power and OH%. The final optimal condition was extraction temperature for 45.28°C, extraction time for 53.91min, and liquid-solid ratio for 30.34 ml·g^{-1}. In the case, the predicted values of the models were TFC for 124.92 mg RE·g^{-1}, DPPH% for 95.99, reducing power for 755.71 mg AA·g^{-1} DW, and OH% for 92.25, respectively. Considering the practice operability, the optimal condition can be modified as follows: extraction temperature for 45°C, extraction time for 55 min, and liquid-solid ratio for 30 ml·g^{-1}. Under the modified condition, the verification experiments were carried out, providing the highest 121.0 mg RE·g^{-1} of TFC, DPPH% for 94.67, reducing power for 740.07 mg AA·g^{-1} DW, and OH% for 90.91, respectively, which have RSD for 5.44, 1.18, 2.29, and 1.47%, respectively. The result indicated the model is satisfactory and accurate in predicting TFC, DPPH%, reducing power, and OH% of the extract from *Suaeda glauca* Bge leaves.

CONCLUSION

In this study, ultrasound-assisted extraction technique was used to extract TF from *Suaeda glauca* Bge leaves and antioxidant activities including DPPH%, reducing power and OH% was investigated. Three important factors with regard to temperature (30-80°C)，time (10-60 min) and liquid-solid ratio (5-50 ml·g^{-1}) were optimized using RSM for obtaining maximum values of response function. The results demonstrated that liquid-solid ratio had highly significant effect on the four response functions followed by temperature and time. The optimal predicted TFC (124.92 mg RE·g^{-1}), DPPH% (95.99), reducing power (755.71 mg AA·g^{-1} DW) and OH% (92.25) were obtained at 45.28°C of extraction temperature, 53.91min of extraction time, and 30.34 ml·g^{-1} of liquid-solid ratio. Under optimal conditions modified, the experimental data of four response functions had good correlation with predicted value. The antioxidant activities of the extract were also evaluated in vitro systems. The results showed that the significant inhibitory effects on DPPH and OH radical of the sample extract were observed. It also exhibited strong reducing power activity. These results suggest that TF from *Suaeda glauca* Bge leaves seems to be suitable natural antioxidant, as well as UAE technique appears to have the advantage of energy-saving, less time-consuming, clean and green for the extraction of antioxidant compounds from plant materials.

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