



Ohmic heating extraction of radish (*Raphanus sativus* L.) leaf phenolic extract: Numerical optimization and kinetic modeling

Zandi, M. ^{1*}, Ganjloo, A. ¹, Bimakr, M. ¹, Nasiri, M. ²

1. Associate Professor, Department of Food Science and Engineering, Faculty of Agriculture, University of Zanjan, Zanjan, Iran.

2. Department of Food Science and Engineering, Faculty of Agriculture, University of Zanjan, Zanjan, Iran.

ABSTRACT

This research aimed to model the kinetic of ohmic heating extraction method of radish (*Raphanus sativus* L.) leaf extract in order to understand and optimize the extraction process. The effect of extraction parameters on extraction yield, total phenol content and DPPH scavenging activity was discussed, at a gradient voltage of 10-30 V/cm, temperature of 30-60°C, solvent ration (Ethanol to water) of 30-100% and extraction time of 10-30 min. Different empirical models such as first order, adsorption and sigmoid models presented to predict the kinetics of mass transfer without taking into account the underlying phenomena. Results indicated that the effect of gradient voltage, extraction time and solvent ratio (ethanol to water) on the extraction yield, total phenolic content and DPPH scavenging activity were significant ($p < 0.05$). Although increasing extraction temperature significantly resulted in a lower total phenolic content and DPPH scavenging activity of extract ($p < 0.05$), however, this parameter no significant effect the extraction yield ($p > 0.05$). Under optimum conditions, the experimental extraction yield and total phenol content were close to the predicted values calculated from the quadratic response surface model. Based on kinetics modeling that has been done, it can be said that the sigmoid kinetic model more can represent well the experimental results of radish leaf extract by the ohmic heating method when compared with the first-order and adsorption kinetic models.

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*Corresponding Author E-Mail:
Zandi@znu.ac.ir

1. Introduction

Today, the tendency to use bioactive compounds based on plant sources has increased. Plants contain a wide range of bioactive compounds such as essential oils, extracts, phytochemical compounds and pigments [1]. These compounds are widely used in the food, pharmaceutical and cosmetic industries. Extracts are plant secondary metabolites and functional compounds [2]. With the increasing demand of medicinal products in the world, the manufacturers of medicinal plants have paid attention to the use of the most suitable extraction technologies to produce high-quality extracts [3]. The method chosen to recover bioactive compounds from plants must be such that it preserves the natural components of the plant. On the other hand, using different methods with the same solvent can cause a wide change in the extraction efficiency [4]. The traditional extraction methods are based on placing the plant in a suitable solvent, and stirring or heating is used to increase the speed of the process. Among these methods, soxhlet, soaking and percolation can be mentioned [2, 5]. Although traditional methods are simple and widely used, they are mostly time-consuming, costly and inefficient, and in addition to consuming a lot of energy and solvent, they also have low efficiency and mainly cause thermal damage to the extracted extract; Therefore, researchers seek to use new methods to reduce these disadvantages and limitations. The development of new methods has significant advantages, such as reducing the consumption of organic solvents, improving extraction efficiency, selectivity and minimizing sample destruction compared to traditional methods, and they also cause the loss of undesirable and insoluble components from the extract. Extraction with the help of microwaves and ultrasonic waves are among the most common new methods [4, 6]. The resistance heating process is a method that has been presented in several reports of its use as resistance distillation in the extraction of essential oils. So far, resistance heating has been proposed as an accelerating method to improve the performance of water distillation and steam distillation, which has accelerated the distillation process [1, 7-11]. In the resistance heating process, the food is

placed between two electrodes as an electrical resistance, and when the electric current passes through it and due to the resistance of the material, it causes its volume to heat up in a short period of time. This process enables rapid heating (from a few seconds to a few minutes). Electrical conductivity is the main variable that determines the heat rate of a resistance heating operation. The resistance heating process can reduce thermal damage, loss of nutrients and deposition during the thermal process of suspended and cut-sensitive food particles [12]. Several researchers have investigated the industrial applications of resistance heating, including aseptic processing¹ And sterilization, pasteurization, drying and distillation have been investigated. However, the use of resistance heating for extract extraction is quite new. Wastes have valuable active substances and the increasing population requires the use of wastes and by-products in order to ensure food security. Increasing people's awareness of natural compounds is the concern of many researchers. Radish leaves are one of the by-products of agriculture that have limited consumption fresh as a salad or cooked and combined with other foods as appetizers, soups and snacks.

radish (*Radish sativa*L.) is a plant whose leaves and roots are consumed fresh or cooked all over the world [13]. Radish root has more consumption and most of its leaves are thrown away after harvesting. Radish is a rich source of minerals (potassium and calcium), vitamins (B₆ and C) and fiber, which is also known in South Asia because of its medicinal properties [13-16]. Phytochemical studies show that radish contains polysaccharides, proteins, isoperoxidases, polyphenols, peroxidases, sinapine, flavonoids, phenolic acid, anthocyanins, raphanoosanins and alkaloids. Radish root and leaves have anti-inflammatory, antifungal, antimicrobial, antioxidant and antihemorrhagic effects [13, 17-19]. Research results have proven that radish extract is effective in treating diseases such as cancer, diabetes, liver infections and inflammation [13, 19, 20]. Investigation of the compounds of radish plant shows that free and

¹. Aseptic

bound phenolics, which include vanillic acid, coumaric acid, pyrogallol, and epicatechin, are mostly found in radish roots and leaves [19, 20]. Research in the field of phytochemical and medicinal activities of radish leaves is relatively limited [21]. Although radish leaves have strong antioxidant properties and anticancer and antihypertensive properties, they are usually thrown away. Radish leaf extract can be used in food, pharmaceutical, cosmetic and health products, and these leaves have anti-cancer and anti-hypertensive properties [14, 19]. There are different methods for extracting radish leaves, but resistance heating is one of the new extraction methods.

Various factors such as voltage gradient, type of solvent, polarity and non-polarity of solvent, particle size, ratio of substance to solvent, pH of solvent and temperature are effective on the efficiency and quality of extraction by resistance heating method; Therefore, it is possible to achieve a more efficient extraction and obtain the optimal extraction method by choosing the correct factors affecting the extraction process in terms of quantity and quality [1, 16, 22]. In order to design, analyze, predict and control the extraction process, it is necessary to determine the variables affecting the mass transfer process, including the extraction efficiency. For this purpose, mathematical models can be used to study extraction kinetics. Various experimental kinetic models such as pseudo-first-order model, Peleg, Weibull and film theory have been used in various researches to model the kinetics of extract or essential oil extraction [22-24]. Examining various sources shows that until now no research has been done in the field of extracting extract from radish leaves by resistance heating method; Therefore, the purpose of the present research is primarily to investigate the effect of various factors, including solvent ratio, temperature, time, and voltage gradient, on the efficiency and quality of radish leaf extract extraction using the resistance heating method and comparing it with the standard extraction method; The aim is to optimize the extraction process of radish leaf

extract by resistance heating method and finally to investigate the kinetics of extracting extract from radish leaf with experimental kinetic models and factors affecting it.

2- Materials and methods

2-1- Materials

radish leaves (*Radish sativa* L.) was purchased from a local market in Zanzan city. 2, 2-diphenyl-1-picrylhydrazyl and sodium carbonate from Sigma Aldridge (USA), Folin-Cyathio reagent and gallic acid from Merck (Germany) and ethanol from Kimia. Alcohol was prepared. Other chemicals with laboratory grade purity were also obtained from Sigma Aldridge (USA).

2-2-Sample preparation

After washing the radish leaves in cold water, the samples were shade dried at a temperature of 25 degrees Celsius until reaching a moisture content of 5% (based on wet weight). The dried radish leaves were ground using a laboratory grinder and then passed through a laboratory sieve with 18 mesh and stored in polyethylene bags impermeable to air and moisture until the experiments were performed.

2-3- extracting the extract

In the resistance heating extraction process, a resistance heating system including a power source, treatment chamber, stainless steel electrodes, voltage and amperage control system, temperature sensor, and safety and protection system were used (Figure 1). The power supply or variable voltage variac could adjust the voltage between 0 and 300 volts with a sensitivity of 1 volt. The treatment chamber or resistance distillation cell was a Pyrex glass flask with a volume of 100 ml, which had two entrances for the electrodes of the device and one entrance for the temperature sensor. The distance between the electrodes was 4 cm. In order to perform the extraction process, 3 grams of radish leaf powder was mixed with 30 ml of solvent (ratio 1:10 (weight/volume)) and before the extraction process, in order to increase the efficiency and extraction speed, it was soaked in the solvent for

1 hour at room temperature. . Next, the mixture was transferred to the resistance heating treatment chamber and depending on the treatment (Table 1) in the voltage gradient (10-30 V/cm), the process temperature (30-60 degrees Celsius), the time of the extract extraction process

(10-30 minutes) and the ratio of ethanol to water (30 to 100 percent (volume/volume)) was given; For this purpose, by placing the electrodes, the extraction process was carried out depending on the treatment in voltage, temperature, solvent ratio and specific time.

Table 1 Design matrix of the response surface methodology.

Solvent ratio (%)	Hour(Min)	Temperature (°C)	Voltage gradient(V/cm)	Run
65	20	30	20	1
44.2	25.9	36.1	14	2
65	20	45	20	3
85.8	14	36.1	25.9	4
65	20	45	10	5
65	20	45	20	6
65	20	45	30	7
30	20	45	20	8
65	20	45	20	9
44.2	14	53.9	25.9	10
85.8	25.9	53.9	14	11
65	20	60	20	12
85.8	25.9	36.1	25.9	13
65	30	45	20	14
65	20	45	20	15
65	10	45	20	16
85.8	14	53.9	14	17
100	20	45	20	18
44.2	25.9	53.9	25.9	19
44.2	14	36.1	14	20
65	20	45	20	21

The control sample was prepared at the same temperature, time and solvent ratio as the optimal sample. Finally, the extract was filtered using filter paper (Whatman, No. 1) and to separate the solvent from the rotary evaporator under vacuum.² It was used at a temperature of 40 degrees Celsius and a speed of 95 rpm. To ensure

the removal of all the solvent, the extract was placed in an oven at a temperature of 40 degrees Celsius until a constant weight was reached. The resulting extract was poured into dark and airtight glass containers and kept in the freezer at -18 ± 1 degrees Celsius until the tests were performed.

². Rotary evaporator



Fig 1 Ohmic heating system.

2-4-Evaluating the quantitative and qualitative characteristics of the extract

To measure the extraction efficiency of the extract (EY, percentage), first its solvent was completely removed in the oven and its mass (m_{ex}) was measured. The amount of extraction efficiency with the help of dry plant weight (gram, m_{dp}) was obtained using equation 1:

$$EX(\%) = \frac{m_{ex}}{m_{dp}} \times 100 \quad (1)$$

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The total phenolic content (TPC) was measured by Folin-Ciocalteu method. This colorimetric method is through the reduction of Folin-Ciocalteu reagent³ It acts by phenolic compounds in an alkaline environment and creates a blue complex. For this purpose, 20 microliters of the extract was mixed with 2 milliliters of ethanol, and then 750 microliters of Folin-Siocaltio reagent (0.1% (vol/vol)) was added to them and it was placed for 5 minutes at room temperature and in a dark environment. Next, 750 microliters of sodium carbonate was added to them and kept in the dark for 90 minutes. Finally, the absorbance was measured at a wavelength of 769 nm using a visible-ultraviolet light spectrometer. In order to draw a standard curve, gallic acid solution was prepared in ethanol (0 to 1000 $\mu\text{g/ml}$) and its total phenol content was measured by the above

method and a gallic acid standard curve was drawn. Total phenol content was expressed as milligrams of gallic acid equivalent per gram of extract weight [11, 23]. The antioxidant activity of radish leaf extract was investigated by measuring the free radical inhibition activity of 2 and 2-diphenyl-1-picrylhydrazyl (DPPH). This is a common and quick method in which the color of the solution changes from purple to yellow by the regeneration of DPPH free radicals. The degree of decolorization shows the power of inhibiting free radicals by the relevant anti-radical compounds and is measured by a visible-ultraviolet light spectrometer at a wavelength of 517 nm. For this purpose, 2 ml of the extract diluted with water-ethanol (final concentration 0.5 mg/ml) was mixed with 2 ml of DPPH solution (0.1 mmol) and the resulting mixture was shaken well and kept in the dark for 30 minutes. Placed at room temperature. Then the absorption of the mixture was measured by a visible-ultraviolet light spectrometer at a wavelength of 517 nm. The inhibition percentage of DPPH free radicals was calculated through equation 2 [11, 23]:

$$DPPH(\%) = \frac{A_c - A_s}{A_c} \times 100 \quad (2)$$

$$DPPH(\%) = \frac{A_c - A_s}{A_c} \times 100$$

That A_c And A_s , It is equal to the absorbance of the control sample and the sample containing the

³. Folin-Ciocalteu

extract, respectively.

5-2-Optimization of the extraction process and comparison with the control sample

A treatment was chosen as the optimal treatment that has the maximum extraction efficiency, total phenol content and inhibition of DPPH free radicals, and for this purpose, the degree of importance and weight of each response was considered the same, and finally, a sample with maximum desirability was selected. Finally, the optimal treatment was compared with the control sample in terms of quantitative and qualitative characteristics.

6-2-Examination of extraction efficiency kinetics

In the present study, in order to investigate the effect of treatment on the kinetics of extraction

efficiency from radish leaves by resistance heating method, first-order, absorption and sigmoid kinetic models (Table 2).

used. The fitting of the kinetic models of the experimental data was done with the help of nonlinear regression and by the data fitting toolbox of MATLAB software (a2019R, USA). In these equations, C_t , C_{eq} , K_A and b , respectively, equal to the extraction efficiency or the total phenolic content of the extracted extract at time t (mg/g), the equilibrium extraction efficiency or saturation or the total phenolic content of the equilibrium or saturated extracted extract when t tends to infinity (mg/g), the rate constant Total extraction (per minute) and constant extraction. t_0 and T_1 It is also equal to the time required to halve the extraction efficiency and the diffusion time constant, which is the slope of the sharp part of the curve.

Table 2 Mathematical models used to evaluate the extraction kinetics.

Reference	Equation	Model
[23]	$C_t = C_{eq} (1 - e^{-K_A \times t})$	First order
[25]	$C_t = C_{eq} + \frac{t}{b + t}$	Adsorption
[26]	$C_t = C_{eq} - \frac{C_{eq}}{1 + \exp\left(\frac{t - t_0}{T_1}\right)}$	Sigmoid

Evaluation and comparison of models with the help of explanatory coefficient (R^2 R^2 R^2)⁴ between the actual value and the predicted value and the root mean square error (RMSE)⁵ Done:

$$R^2 = 1 - \left(\frac{\sum_{i=1}^n (O - P)^2}{\sum_{i=1}^n (O - \bar{P})^2} \right) \quad (3)$$

$$R^2 = 1 - \left(\frac{\sum_{i=1}^n (O - P)^2}{\sum_{i=1}^n (O - \bar{P})^2} \right) \quad (4)$$

$$RMSE = \sqrt{\frac{(O - P)^2}{n - p}}$$

$$RMSE = \sqrt{\frac{(O - P)^2}{n - p}}$$

That O , P , p and n are the experimental data, the predicted data, the number of observations and the number of model variables, respectively.

2-7-Statistical analysis

To optimize the effective factors on extract extraction by resistance heating method, including voltage gradient, temperature, ethanol-

⁴. R-squared

⁵. Root mean squared error

water ratio and time, from the CCD design.⁶ With the help of Design Expert software⁷ used. The selection of factor levels was based on previous researches and numerical optimization was used

to determine the optimal conditions. The graphs were drawn with the help of Excel software (version 2016, USA).

Table 3 ANOVA table showing the extraction variables as linear, quadratic and interaction terms on each response variable

Source	Yield (%)		Total phenolic content (mg GAE/g extract)		DPPH (%)	
	Sum of square	p-value	Sum of square	p-value	Sum of square	p-value
Model	388.35	< 0.0001	1379.60	< 0.0001	394.76	< 0.0001
A-Voltage gradient (V/cm)	49.00	< 0.0001	176.34	< 0.0001	70.54	< 0.0001
B- Extraction temperature	0.0937	0.6698	162.00	< 0.0001	33.05	< 0.0001
C- Extraction time	6.55	0.0095	54.12	0.0019	42.98	< 0.0001
D-Solvent ratio (%)	174.00	< 0.0001	318.40	< 0.0001	22.99	0.0001
AB	58.57	< 0.0001	63.97	0.0012	6.96	0.0033
AC	0.2237	0.5146	6.68	0.1127	1.95	0.0570
AD	0.9462	0.2044	243.39	< 0.0001	41.25	< 0.0001
BC	0.1999	0.5371	0.0055	0.9592	0.0036	0.9181
BD	0.2612	0.4827	0.0630	0.8628	1.17	0.1015
CD	7.94	0.0062	5.93	0.1306	0.4851	0.2602
A ²	6.51	0.0097	0.0179	0.9266	0.9810	0.1275
B ²	3.66	0.0312	173.31	< 0.0001	32.34	< 0.0001
C ²	3.91	0.0276	0.5204	0.6228	34.79	< 0.0001
D ²	58.86	< 0.0001	46.43	0.0027	1.30	0.0878
Residual	2.80		11.62		1.88	
<i>Lack of Fit</i>	1.96	0.0911	6.84	0.1690	1.14	0.1577
Pure Error	0.8454		4.78		0.7481	
Cor Total	391.15		1391.22		396.64	
R²	0.9928		0.9916		0.9953	
CV	2.91		1.13		0.79	

* p -value<0.05 is significant at α <0.05 and *lack of fit* is not significant at p -value>0.05.

3. Results and Discussion

3-1- Extraction efficiency

The results of the analysis of the main and mutual effects of factors including voltage gradient, temperature, time and ethanol-water ratio on the extraction efficiency are shown in Table 3. As can be seen, the quadratic model was able to predict the results of extraction efficiency with a high explanatory coefficient. The significance of the model (<0.05 p) and the non-significance of the misfit error (> 0.05 p) indicates the success of the quadratic model. Also, the coefficient of variation less than 3 indicates (2.91) the high accuracy of the tests. The results of the statistical analysis showed that all the main effects, except the process temperature, all the secondary effects and

the interaction effects between the process time and the ratio of ethanol to water and between the voltage gradient and the temperature of the process on the extraction efficiency of radish leaves were significant (<0.05 p). The final equation for predicting the efficiency of extracting extract from radish leaves by resistance heating method is equation 5:

$$(5) \text{Yeild (\%)} = 22.41 + 2.94A + 0.6928C - 5.55D + 4.20AB + 0.9946CD + 0.6559A^2 - 0.4949B^2 - 0.5113C^2 + 1.98 D^2$$

where A, C and D are respectively equal to the main effects of voltage gradient, process time and ethanol to water ratio and AB, CD, A², B², C² and D² It is also equal to the interaction effect of voltage gradient and process temperature, the interaction effect of process time and ethanol-

⁶. Central Composite Design

⁷. DesignExpert

water ratio, the quadratic effect of voltage gradient, the quadratic effect of temperature, the quadratic effect of time and the quadratic effect of ethanol-water ratio.

The coefficients before each effect indicate its effect on extraction efficiency, and the larger this coefficient is, the greater the sensitivity of the response; Therefore, the main effect of the ratio of ethanol to water has the greatest effect on the extraction efficiency, and the secondary effect of the process temperature has the least effect on the response.

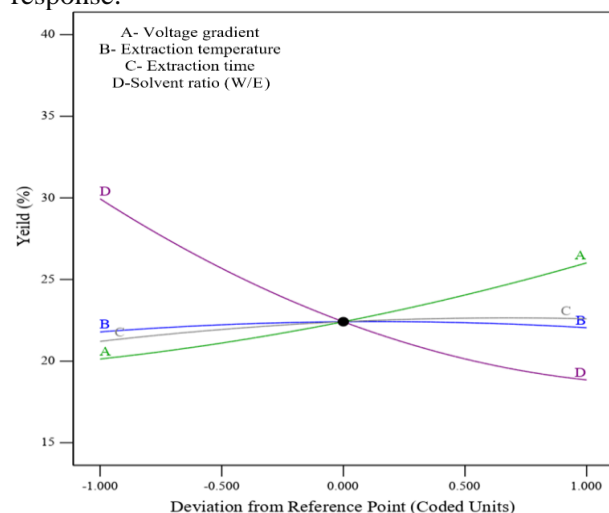


Fig 2 Perturbation plot for extraction yield.

After the ratio of ethanol to water, the interaction effect of voltage gradient and process temperature has the greatest effect on extraction efficiency. The positive and negative signs before each effect indicate the increasing and decreasing effects of that effect on the response, respectively. As can be seen in the saturation graph (Figure 2), the highest slope of the graph among the main effects is related to the graph of the ratio of ethanol to water, and then the slope of the graph of the main effect of voltage gradient is higher, which indicates its greater effect on the changes in extraction efficiency. The negative sign before the coefficient of the main effect of the ratio of ethanol to water in equation 5 is also observed as the reverse trend of the graph of this main effect compared to other main effects in the saturation graph. The significance of the quadratic effect of all factors on the extraction

efficiency is clearly evident in the saturation diagram, so that the shape of all the main effects diagrams is curved.

One of the goals in the extraction process is to increase the extraction efficiency of the extract; The results showed that depending on the type of treatment, the extraction efficiency of radish leaves with resistance heating method varied between 19-37.7%. The extraction efficiency of radish leaf extract depends on various factors such as the type and growth conditions and time of harvesting radish leaves, extraction method, extraction temperature, type of solvent, etc. Many researches in the field of extract extraction from

Radish root is done; However, there are limited sources in extracting extract from radish leaves. In research⁸ and colleagues (2018) reported the extraction efficiency of radish leaf extract with supercritical fluid method (carbon dioxide) in 7-33% depending on the type of treatment [27]. Quercetin extraction efficiency⁹ of radish leaves with water solvent and with the help of soaking, soxhlet, digestion and extraction with the help of ultrasound was less than 0.4%. While with ethanol solvent, the efficiency in the methods of soaking, Soxhlet, digestion and extraction with the help of ultrasound was 1.2, 0.45, 2.25 and 12%, respectively [28], which shows the great effect of the extraction method and the type of solvent on the efficiency. It has extraction. The three-dimensional diagrams of the main and mutual effects of the studied factors on the extract efficiency of radish leaves are shown in Figure 3. The increase in extract extraction efficiency by increasing the voltage gradient may be attributed to the non-thermal effect of the resistance heating process. In the resistance heating process, the passage of electric current along with the generation of heat due to the internal resistance of the mixture, by passing through the glands containing the extract, causes them to tear, and therefore facilitates the extraction of the effective substance from them, and as a result, increases the extraction efficiency [11, 23]. Therefore, as the voltage gradient increases, the non-thermal effect of the resistance heating method also increases, and as a result, the extraction efficiency

⁸.Goyeneche

⁹.Quercetin

increases. So far, no research has been done in the field of extraction of extract by resistance heating method, but researches on extraction of essential oil by resistance distillation method have also achieved similar results [1, 8-11]. Although the main effect of temperature did not have a significant effect on the extraction efficiency ($>$

0.05). p) but its interaction with the voltage gradient had a great effect on the extraction speed of the extract, which seems to be due to its effect on reducing the viscosity of the liquid phase and accelerating the extraction of the extract from the damaged glands.

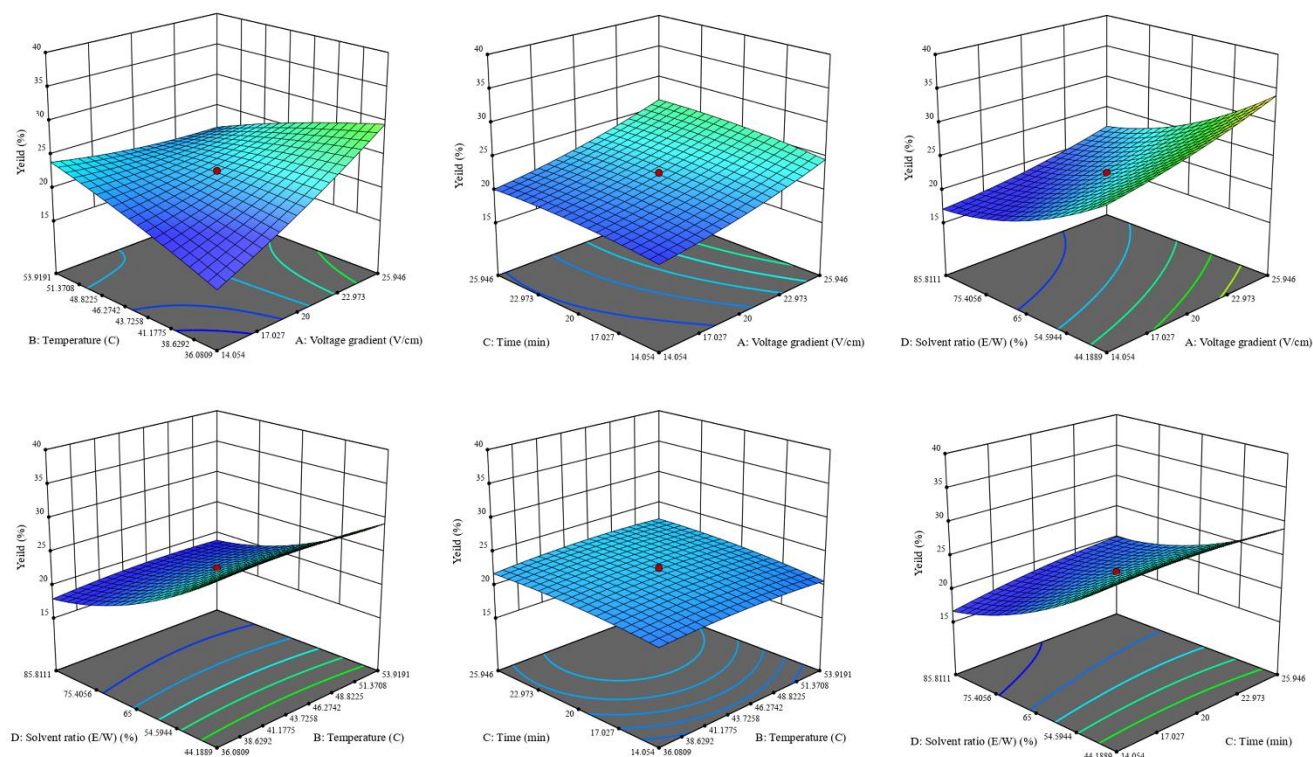


Fig 3 Changes in extraction yield of radish leaf extract vs. independent variables.

Also, the non-linear effect of temperature on the extraction efficiency was effective, which is probably related to the relationship between temperature and the viscosity of the liquid phase. The results showed that increasing the extraction time increases the extraction efficiency, which is due to the fact that the time available for the extract increases and more extract is extracted; Similar results of the effect of time on the extraction of extract from radish leaves with methods such as soaking, Soxhlet, digestion, extraction with the help of ultrasound and supercritical fluid have been reported by other researchers [27, 28]. The main effect of the ratio of ethanol to water has the greatest effect on the extraction efficiency, so that with the increase in the polarity of the solvent (increasing the share of ethanol), the extraction efficiency decreases

drastically, which indicates the greater contribution of polar compounds in radish leaf extract. Finally, it was found that both linear and non-linear effects of voltage gradient, process time and ethanol-water ratio were effective on the extraction efficiency of radish leaf extract.

2-3- The total phenol content of the extract

The results of statistical analysis (Table 3) showed that the quadratic model was significant on the results of total phenol content ($<0.05p$); Also, the non-significant model misfit error (>0.05). p) and the high explanation coefficient and low variation coefficient also indicate the correct performance of the quadratic model. As can be seen in Table 3, the effect of all the main effects of voltage gradient, process time and temperature, and the ratio of ethanol to water on

total phenol content was significant (<0.05). p). Among the mutual effects, only the mutual effects between the voltage gradient and the process temperature, as well as between the voltage gradient and the ratio of ethanol to water, were significant (<0.05). p). Finally, among the quadratic effects, the quadratic effect of process temperature and ethanol-water ratio on total phenol content was significant (<0.05). p). The equation created for predicting the total phenol content of the extract is as equation 6:

(6)

$$\text{TPC} = 124 + 5.58A - 5.35B + 1.99C + 7.50D - 4.39AB + 8.57AD - 3.341B^2 + 1.76D^2$$

where A, B, C and D are respectively equal to the main effects of voltage gradient, process temperature, process time and ethanol to water ratio and AB, AD, B^2 and D^2 It is also equal to the interaction effect of voltage gradient and process temperature, the interaction effect of voltage gradient and ethanol-water ratio, the quadratic effect of temperature and the quadratic effect of ethanol-water ratio. According to the coefficients, the content of total phenol is most sensitive to changes in the interaction effect of voltage gradient and ethanol-water ratio, and among the main effects, the main effect of ethanol-water ratio and the main effect of process temperature have the most increasing and decreasing effects on total phenol content. . The quadratic effect of temperature also decreased the content of total phenol.

The sensitivity of total phenol content to the main effects of voltage gradient, process temperature, process time and ethanol-to-water ratio is shown in the saturation diagram (Figure 4).

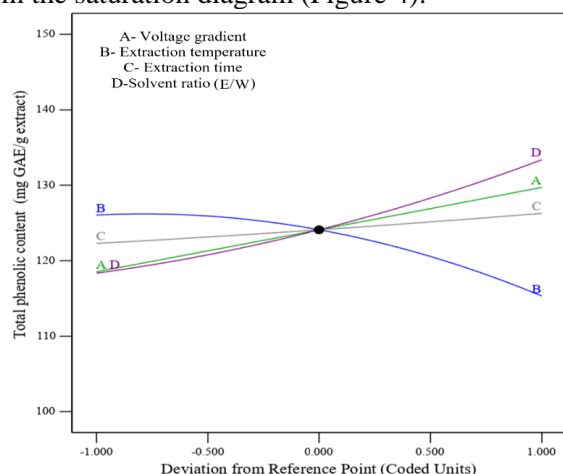


Fig 4 Perturbation plot for total phenolic content.

As can be seen in the saturation diagram, changes in the process temperature have the greatest effect on the total phenol content, which is a weakening effect on the response. In the diagram, the nonlinear effects of the process temperature and the ratio of ethanol to water are clearly visible. The results of total phenol content for different treatments of resistance heating process varied between 106.1-142.3 mg of gallic acid equivalent per gram of extract. In a research, the amount of phenolic content of radish leaf extract extracted by supercritical fluid method was reported between 9.97-13.61 mg of gallic acid equivalent per gram of dry plant, which is consistent with the results of this research according to the extraction efficiency of the extract [27].

Also, Khan et al. (2022) also reported the total phenolic content of radish leaf extract as 125.3 mg equivalent of gallic acid per gram of extract [29].

The main and reciprocal effects of variables affecting total phenol content in resistance heating process are shown in Figure 5. As can be seen in the figure, with the increase of the voltage gradient, the amount of total phenol content increases, which is probably due to the increase of the non-thermal effect of the resistance heating process, which causes less thermal damage to the extract. On the other hand, phenolic compounds in plant cell walls create strong bonds with lignin, which makes their extraction more complicated. Dissolving these strong bonds of phenolic compounds is difficult and only methods such as resistance heating can destroy plant cell walls and facilitate the release of cellular secondary metabolites [28]. Therefore, by increasing the voltage gradient, the separation of phenolic compounds is facilitated. Of course, it should be noted that increasing the voltage gradient too much can cause damage to phenolic compounds. So far, no research has been done in the field of extraction of extract by resistance heating method, but similar results of extracting essential oil by resistance distillation method have been reported [1, 7, 11]. The mutual effect of voltage and temperature gradient caused a significant decrease in total phenol content (<0.05). p) which is probably due to the intensification of the thermal effect of the resistance heating method with increasing temperature. On the other hand, the mutual effect of the voltage gradient and the

ratio of ethanol to water also caused an increase in the total phenol content of the extract, which is probably because the non-thermal effect of this method intensifies with the increase of the voltage gradient and causes more damage to the glands containing the extract and at the same time More phenolic compounds are removed due to their greater solubility in ethanol. As it is clear

from the results, increasing the temperature significantly decreased the content of total phenol ($<0.05.p$).

This is due to thermal damage to phenolic compounds at high temperature. Similar results were obtained in extracting radish leaf extract with supercritical fluid method [27].

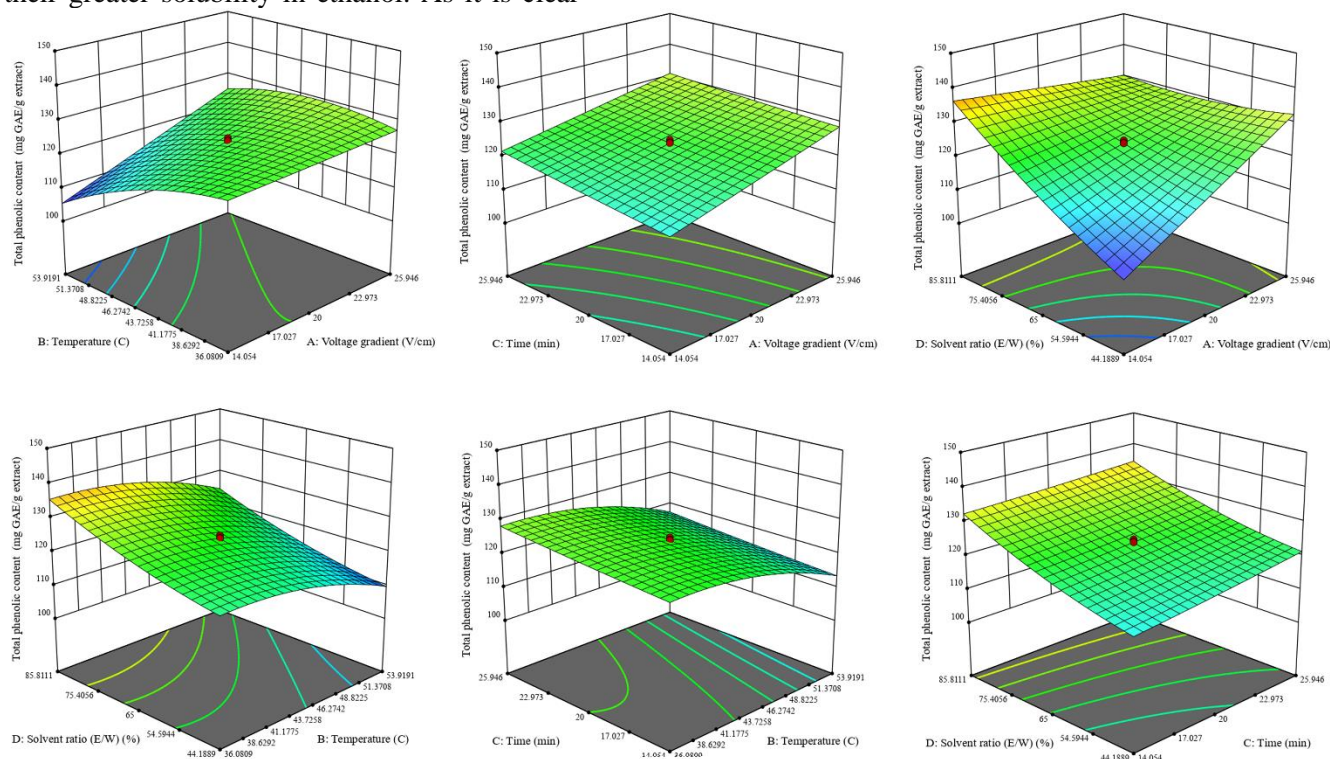


Fig 5 Change in total phenolic content of radish leaf extract vs. independent variables.

Increasing the extraction time had a small effect on increasing the total phenol content, which may be because the necessary time is provided for the extraction of phenolic compounds. On the other hand, with an increase in the ratio of ethanol to water, the extraction rate of phenolic compounds increased, which indicates the greater solubility of these compounds in ethanol. Similar results were obtained by Sharifi et al. (2016) in extracting corsine as a phenolic compound from radish leaves with different methods and solvents [28]. Finally, process temperature and ethanol-water ratio had non-linear effects on total phenol content.

3-3- Antioxidant activity

As can be seen in Table 3, the quadratic model with the explanation coefficient equal to 0.9953 and the coefficient of variation equal to 0.79 was significant in the analysis of the measurement

data of DPPH free radical scavenging activity ($<0.05p$) and since the misfit error was not significant ($> 0.05.p$) as a result, this model can be used to analyze the results. The results showed that all the main effects of voltage gradient, temperature and process time and ethanol-to-water ratio, as well as the mutual effects between voltage gradient and temperature and between voltage gradient and ethanol-to-water ratio and second-order effects of temperature and process time on DPPH free radical scavenging activity are significant. became ($<0.05p$). The final equation for predicting DPPH free radical scavenging activity is presented in Equation 7:

$$(7) \text{ DPPH scavenging activity} = 71.59 + 3.53A - 2.42B + 1.77C + 2.02D - 1.45AB + 3.53AD - 1.47B^2 + 1.53C^2$$

where A, B, C and D are respectively equal to the main effects of voltage gradient, process

temperature, process time and ethanol to water ratio and AB, AD, B² and C². It is also equal to the interaction effect of voltage gradient and process temperature, the interaction effect of voltage gradient and ethanol to water ratio, the quadratic effect of process temperature and the quadratic effect of process time. As can be seen in the above relationship, the main effect of voltage gradient and the interaction effect of voltage gradient and ethanol-water ratio had the greatest effect on the change in DPPH free radical inhibition activity. By examining the saturation diagram (Figure 6), it is clear that the most sensitive response after the main effect of voltage gradient is to the main effect of temperature, although the relationship between DPPH free radical inhibition activity and temperature effect is inverse. The curvature of the graphs of the main effects of time and process temperature also indicate their second-order effect on the response. DPPH free radical inhibition activity of radish leaf extract obtained from different resistance heating treatments varied between 63.45-63.43%.

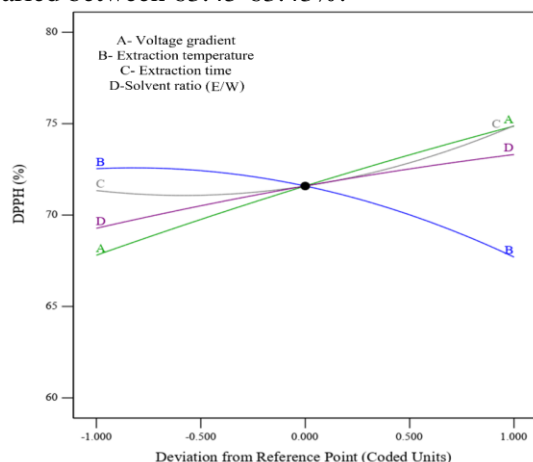


Fig 6 Perturbation plot for DPPH scavenging activity.

In a research, Guinche et al. (2018) reported the DPPH free radical scavenging activity of radish leaf extract extracted by supercritical fluid method between 0.274-0.403% based on dry plant, which according to the extraction efficiency of the extract is consistent with the results of this research. has [27].

The three-dimensional diagram of changes in DPPH free radical scavenging activity of radish leaf extract, which shows the main and reciprocal effects, is shown in Figure 7. As can be seen in the figure, with the increase of the voltage gradient due to the increase of non-thermal effects, less thermal damage is caused to the extract, and on the other hand, the increase of the extraction time allows the extraction of more antioxidant compounds. Also, the increase in DPPH free radical inhibition activity with an increase in the ratio of ethanol to water indicates greater solubility of antioxidant compounds in ethanol. As expected, the increase in temperature caused a decrease in DPPH free radical inhibition activity. Similar results of the effect of temperature on DPPH free radical inhibition activity of the extract were obtained in the extraction of radish leaf extract by supercritical fluid method [27].

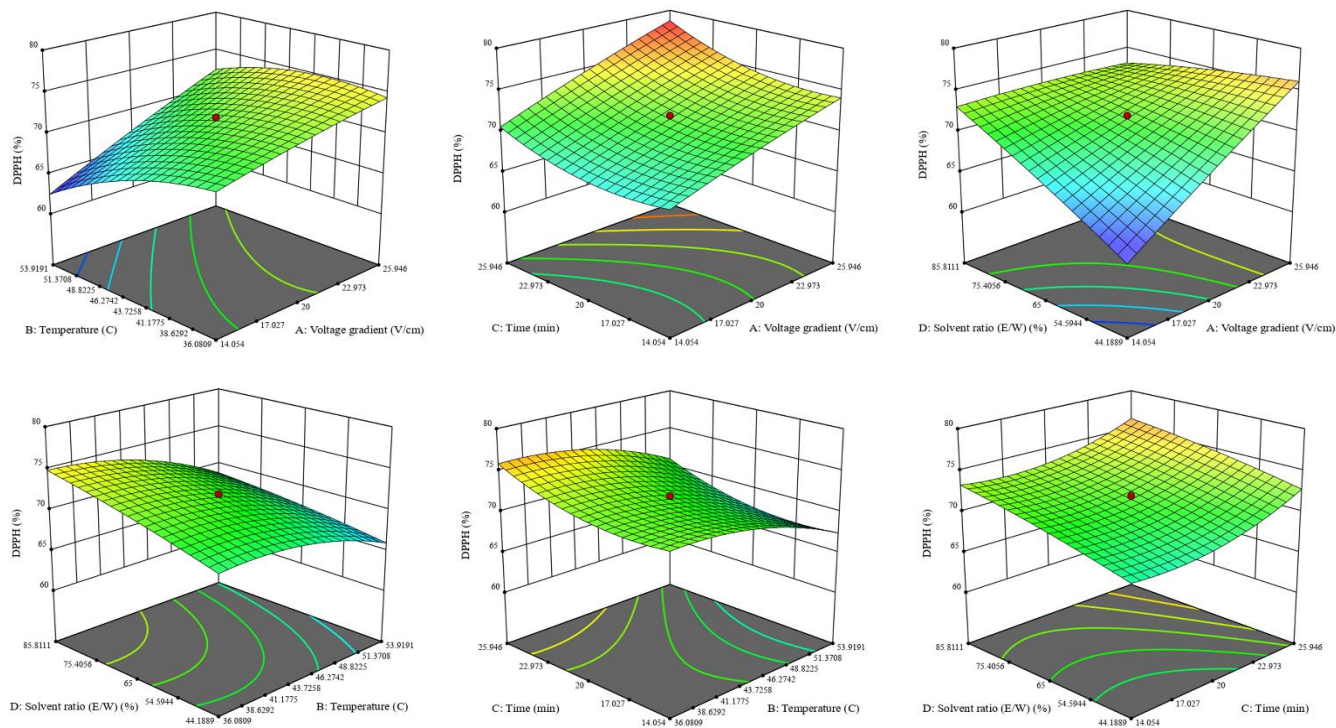


Fig 7 Change in DPPH scavenging activity of radish leaf extract vs. independent variables.

4-3- Optimizing the process and comparing it with the soaking method

In order to obtain the optimal treatment, numerical optimization was carried out with the aim of finding a process with the maximum extract extraction efficiency, maximum total phenol content and DPPH free radical inhibition activity of the extract. The results of the optimization determined a process with a voltage gradient equal to 25.9 V/cm, a temperature of 37

degrees Celsius, a process time of 26 minutes, and a ratio of ethanol to water of 44.2% as the optimal process. The prediction results for the optimal treatment with the experimental results are shown in Table 4. As can be seen in the table, there is no significant difference between the predicted and experimental results for the optimal resistance heating treatment ($> 0.05p$) which indicates the accurate prediction of the model. This is significant with a high explanation coefficient (<0.05). p quadratic model and non-significance ($0.05>p$) misfit error was predicted.

Table 4 Multi-response numerical prediction results.

	Predicted values	Experimental values
Extraction yield (% w/w)	36.80	37.18±0.59
Total phenolic content (mg GAE/g extract)	132.69	130.95±2.13
DPPH (%)	79.29	80.32±1.21

5-3- Kinetic modeling of extraction efficiency and total phenol content

As mentioned earlier, kinetic modeling was done with three first-order models, absorption and sigmoid. The results showed that all three models were able to model the changes of both extraction efficiency and total phenol content with an explanation coefficient of 0.93 (Table 4). So that the explanation coefficient for The fitting curves of extraction efficiency and

total phenol content were 0.9943 and 0.9986, respectively. The sigmoid model is used to describe it when the kinetics of essential oil extraction does not follow the exponential pattern [23]. In the sigmoid model t_0 It is equivalent to the time when the response rate (extraction efficiency or total phenol content) has reached 50% and T_1 It is also equal to the slope of the beginning of the curve (its sharp part) and in fact it is the same as the propagation time constant. Whatever t_0 Smaller and T_1 The larger it is, the

faster the response changes.

Table 5 Kinetic parameters and statistical values of first-order, adsorption and sigmoid models.

	Extraction yield	Total phenolic content
First-order model		
K_A	0.2011	0.2147
C_{eq}	23.11	114.07
R^2	0.9847	0.9830
MSE	2.8682	45.49
Adsorption model		
b	2.2964	2.1510
C_{eq}	23.11	114.07
R^2	0.9470	0.9348
RMS		
E	9.7732	169.90
Sigmoid model		
t_0	3.1549	3.9427
T_1	4.3919	5.3743
R^2	0.9943	0.9986
RMS		
E	1.0750	2.7359

The graph of the experimental data of extraction efficiency and total phenol content compared to the predicted data by the first-order, absorption and sigmoidal kinetic models is shown in Figure 8. As can be seen, the sigmoid model was able to fit the changes of extraction efficiency and total phenol content more accurately than other models, and therefore its output is very close to the experimental data.

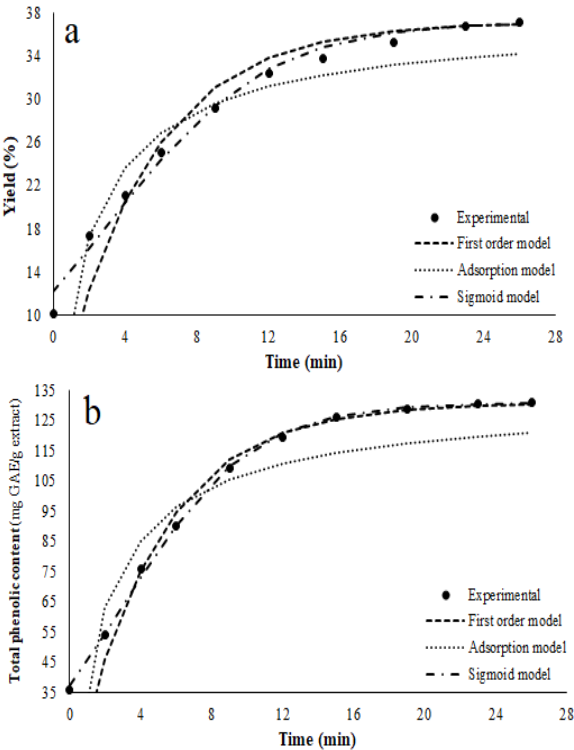


Fig 8 Comparison of the First order, Adsorption and Sigmoid models with the experimental data.

4- General conclusion

In this research, with the aim of optimization, the effects of voltage gradient, process temperature, process time and ethanol-water ratio in resistance

heating method were investigated. The results showed that the quadratic model was able to predict all the answers with an explanation coefficient above 0.99. The low coefficient of variation is significant (<0.05). p) model and non-significance ($> 0.05p$) the misfit error also indicates the correct performance of the model. The results showed that all the main effects, except for the process temperature, all secondary effects and interaction effects between the process time and the ratio of ethanol to water and between the voltage gradient and the process temperature on the extraction efficiency of radish leaves were significant (<0.05). p). Of course, the relationship between the extraction efficiency and the ratio of ethanol to water was inverse. Also, the effect of all main effects and mutual effects between voltage gradient and process temperature, as well as between voltage gradient and ethanol-water ratio on total phenol content and DPPH free radical inhibition activity were significant (<0.05). p). Finally, among the quadratic effects, the quadratic effect of process temperature and ethanol-water ratio on total phenol content and the quadratic effects of temperature and process time on DPPH free radical inhibition activity were significant (<0.05). p). In both responses of total phenol content and DPPH free radical scavenging activity, the relationship between the main effect of temperature and the response was inverse. The results of the optimization determined a process with a voltage gradient equal to 25.9 V/cm, a temperature of 37 degrees Celsius, a process time of 26 minutes, and a ratio of ethanol to water of 44.2% as the optimal process. Among the first-order, absorption, and sigmoidal kinetic models, the sigmoidal model had a more successful performance in modeling the kinetics of responses; So that the coefficient of explanation for fitting the curves of extraction efficiency and total phenol content was equal to 0.9943 and 0.9986 respectively.

5- Appreciation and thanks

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استخراج عصاره فنولی از برگ تربچه (*Raphanus sativus* L.) به روش حرارت‌دهی مقاومتی:

بهینه‌سازی عددی و مدل‌سازی سینتیکی

محسن زندی^{۱*}، علی گنجلو^۱، ماندانا بی‌مکر^۱، مینا نصیری^۲

۱- دانشیار، گروه علوم و مهندسی صنایع غذایی، دانشکده کشاورزی، دانشگاه زنجان، زنجان، ایران.

۲- گروه علوم و مهندسی صنایع غذایی، دانشکده کشاورزی، دانشگاه زنجان، زنجان، ایران.

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* مسئول مکاتبات:

Zandi@znu.ac.ir

این پژوهش با هدف بهینه‌سازی عددی و مدل‌سازی سینتیکی استخراج عصاره فنولی از برگ‌های تربچه (*Raphanus sativus* L.) به روش حرارت‌دهی مقاومتی به منظور درک بهتر و بهینه‌سازی فرآیند استخراج انجام شد. اثر متغیرهای استخراج بر راندمان استخراج، محتوای فنول کل و فعالیت مهار رادیکال‌های آزاد DPPH، در ولتاژ گرادیان برابر ۳۰-۱۰ ولت بر سانتی‌متر، دمای ۳۰-۶۰ درجه سلسیوس، نسبت حلال (اتانول به آب) ۱۰۰-۳۰٪ و زمان استخراج ۱۰-۳۰ دقیقه مورد بررسی قرار گرفت. مدل‌های تجربی مختلف مانند مدل‌های مرتبه اول، جذب و مدل‌های سیگموئید برای پیش‌بینی سینتیک انتقال جرم بدون در نظر گرفتن پدیده‌های زمینه‌ای بررسی شدند. نتایج نشان داد که اثر گرادیان ولتاژ، زمان استخراج و نسبت حلال (اتانول به آب) بر راندمان استخراج، محتوای فنول کل و فعالیت مهار DPPH معنی‌دار بود ($p < 0.05$). اگرچه افزایش دمای استخراج به‌طور معنی‌داری منجر به کاهش محتوای فنول کل و فعالیت مهار رادیکال‌های آزاد DPPH شد ($p < 0.05$)، اما این متغیر تأثیر معنی‌داری بر عملکرد استخراج نداشت ($p > 0.05$). در شرایط بهینه، راندمان استخراج و محتوای فنول کل تجربی نزدیک به که مدل سینتیکی سیگموئیدی در مقایسه با مدل‌های سینتیکی مرتبه اول و جذبی، بیشتر می‌تواند نتایج تجربی عصاره برگ تربچه را با روش حرارت‌دهی مقاومتی مدل نمایند.