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OPTIMIZATION OF SPRAY DRYING CONDITION FROM TRAMETES VERSICOLOR MUSHROOM EXTRACT

NGUYEN NGOC TUAN[1], NGUYEN THI NGAN[1], NGUYEN NGOC THUAN[1], NGUYEN TRAN NGOC MAI[1], NGUYEN DUC VUONG[1], DAM SAO MAI[2], NGUYEN TAN THANH[3], TRAN DINH THANG[3]

[1] *Viện Công nghệ sinh học và thực phẩm, Trường Đại học Công nghiệp Thành phố Hồ Chí Minh,*
[2] *Phòng quản lý khoa học và hợp tác quốc tế, Trường Đại học Công nghiệp Thành phố Hồ Chí Minh;*
[3] *Viện Hóa sinh môi trường, Trường Đại học Vinh;*

nguyennngoctuan@iuh.edu.vn

Abstract. In this study**,** the optimal conditions for the spray drying from *Trametes versicolor* (V.C.) extracts were determined using response surface method/technique (RSM). One kilogram of material was extracted with 30L ethanol 65% of 70^{o}C for 7 hours. Solids were filtered off, extract was concentrated to obtain solution containing about 20% total solid using a vacuum evaporator. Study on the effects of three independent variables, namely maltodextrin ratio (% w/v), feed flow rate (ml/min) and inlet drying temperature (^{o}C) on the responses; total phenolic content (TPC) and moisture content (MC). The optimal conditions for spray drying process from *T. versicolor* extracts were found to be maltodextrin ratio of 5%w/v, inlet drying temperature of 160^{o}C and feed flow rate of 26.05ml/min. The experimental values of TPC and MC were 35.76±0.04mg GAE/g, 4.43±0.02%.

Keywords. Moisture content (MC), *Trametes versicolor,* total phenolic content (TPC), response surface methodology (RSM).

1. INTRODUCTION

For a long time, mushrooms have been used not only as traditional foods but also as medicinal species in many countries. So far, mushrooms are still used to treat many major diseases in China, Japan, Korea and several other Asian countries. *Trametes versicolor* (L.:Fr.) Pil has been widely used as a medicinal ingredient in both traditional medicine and modern clinical practice [1]. Studies have shown that in addition to nutritional value, antioxidant and anti-inflammatory activity, immune-enhancing activity, anticancer activity, antiviral effects andantimicrobial [2]. However, in Viennam and other countries there are few studies that analyzed *T. versicolor* phenolic compounds composition, important for their antioxidant properties.

Spray drying is a technique of producing a dry powder from pumpable liquid or slurry using rapid drying with a heated gas media. The physicochemical properties of powders produced by spray drying are very much dependent on significant process variables such as air inlet temperature, maltodextrin ratio, air pressure and feed flow rate. Other less important parameters are ignored in this study due to the limitations of the equipment used (e.g., air outlet temperature, atomizer type and size, air humidity, drying air flow rate). The liquid feed was assumed to be close to water characteristics (e.g., viscosity, solid content, surface tension, solvent volatility).

The objective of the present study was to elucidate the effect of adjuvant in the spray-drying of mushroom extract and to find the processing parameters of spray-drying to create powdered mushroom extract with the highest total phenolic content, the total flavonoid content as well as the best moisture content by applying Response Surface Methodology. As far as the authors know, no spray-drying process of phenolics compounds in *Trametes versicolor* products extracts has been reported.

2. MATERIAL AND METHODS

2.1. Material

The basidiomycetes of *Trametes versicolor* were collected at the Pumat National Park of Nghe An Province, Vietnam in September 2018 and identified by Prof. Dr. Ngo Anh, Department of Biology, Hue University. A voucher specimen (Vinh-TĐT 16092018) was deposited at the herbarium of the Department of Chemistry, Vinh University.

2.2. Methods

2.2.1. Spray drying

Spray drying of *Trametes versicolor* mushroom extract with maltodextrin as an additive (wall material) at different ratios was performed using a Buchi B-290 mini spray dryer. The mushroom extract was then mixed with maltodextrin at a ratio of 5%, 10% and 15% (w/v), inlet temperature used to optimize the model were 140, 150 and 160^{o}C. Feed rates used were 20, 25, 30 ml/min. After spray drying processing, the *Trametes versicolor* mushroom powder thus obtained was stored in small glass jars with screw caps to evaluate their ending product quanlity.

2.2.2. Moisture content (MC)

The moisture content was determined based on AOAC method (AOAC, 2000). Triplicate samples of *Trametes versicolor* mushroom powder (5g) were weighed and dried in oven at 105ºC until its weight is constant.

2.2.3. Total Phenolic Content (TPC)

The TPC was measured according to the method reported by Singleton *et al.* [3] with some modifications. This method is based on measuring color change caused by reagent by phenolates in the presence of sodium carbonate. 1ml of sample was mixed 5ml of Folin – Ciocalteu's solution. After 3 min, 4ml of 7.5% sodium carbonate solution was added to mixture and adjusted to 10 ml with deionized water. The mixture was allowed to stand at room temperature in the dark environment for 60 min. The color change was determined by scanning the wavelength at 765nm (Agilent 8453 UV – Visible Spectrophotometer) since maximum absorbance was obtained. TPC of the *Trametes versicolor* extract was determined as mg gallic acid equivalent using the standard curve prepared at different concentrations of gallic acid (10-60 μg/ml, $y = 0.011x – 0.005$; $R^2 = 0.998$) and reported as mgGAE/g dry weight (DW).

2.2.4. Experimental design

Response Surface Methodology was used to determine the optimum levels of maltodextrin ratio (% w/v), feed flow rate (ml/min) and inlet drying temperature (^{o}C) as drying on two responses namelyTPC and MC in the *Trametes versicolor*extracts spray-drying. These three factors, namely maltodextrin ratio (A), inlet drying temperature (B) and feed flow rate (C) were coded into three levels (-1, 0, +1). The coded independent variables used in the RSM design are shown in table 1. Ranges of maltodextrin ratio, inlet drying temperature and feed flow rate and the central point were selected based on preliminary experimental results [4-10].

3. RESULTS AND DISCUSSION

3.1. Model Fitting

The responses consisting of TPC and MC for Spray drying from *Trametes versicolor* mushroom (V.C) extracts were optimized based on the box-behnken. The box-behnken with three independent variables was used as follows: maltodextrin ratio, inlet drying temperature and feed flow rate. The input range of the selected variables was determined by the preliminary experiments (Table 1). Two dependent variables including total phenolic content and moisture content were determined following spray-drying under optimal conditions. These experimental values were compared with those of the predicted values to check the validity of the model.

Table1: Coded level of independent variables used in the RSM design

Independent variables	Coded symbols	Coded variable levels		
		-1	0	+1
Maltodextrin ratio (%)	A	5	10	15
Inlet drying temperature (^oC)	B	140	150	160
Feed flow rate (ml/min)	C	20	25	30

The effects of the extraction parameters were evaluated using the program Design- Expert®, version 7.0.0. The response variable was fitted be a second- order polynomial model as follows:

$$y = \beta_0 + \sum_{i=1}^{k} \beta_i X_i + \sum_{i=1}^{k} \beta_{ii} X_i^2 + \sum\sum_{i<j} \beta_{ij} X_i X_j$$

Table2: Experimental Design and Response Values

RUN	Maltodextrin ratio A (%w/v)	Inlet drying temperature B (^oC)	Feed flow rate C (ml/min)	TPC Y_1 (mgGAE/g)	MC Y_2 (%)
1	-	-	0	37.69	5.37
2	-	+	0	36.48	4.41
3	+	-	0	29.79	7.16
4	+	+	0	28.08	4.59
5	0	-	-	34.79	6.43
6	0	+	-	32.95	4.54
7	0	-	+	35.04	6.63
8	0	+	+	34.58	4.87
9	-	0	-	36.72	5.72
10	+	0	-	28.42	6.53
11	-	0	+	37.25	5.91
12	+	0	+	29.26	6.83
13	0	0	0	34.12	6.04
14	0	0	0	34.03	6.09
15	0	0	0	34.18	6.01
16	0	0	0	34.21	6.03
17	0	0	0	34.24	6.06

The values of the two evaluation indices for each spray-drying condition were listed in Table 2. The maximal TPC was 37.69 mgGAE/g at maltodextrin ration5%w/v,Inlet drying temperature 140^{o}C and feed flow rate 25mL/min; The minximal MC was 4.41% atmaltodextrin ration 5%w/v, Inlet drying temperature 160^{o}C and feed flow rate 25ml/min. From the multiple linear regression analyses of the 17data entries, empirical second-order polynomial models of TPC and MC scavenging capacity were derived (Table 3).

Table3: Empiric second- order polynomial model of TPC and MC

Response	Model equations	R^2	p-value
Y_1 – TPC	Y_1= 35.35- 0.28A-2.09B + 0.95C - 0.075AB + 0.69AC + 0.093BC + 0.14A^2 – 0.46B^2 +0.17C^2	0.9993	<0.0001
Y_2 – MC	Y_2= 6.29– 0.78A + 0.30B + 1.12C – 0.24AB + 0.065AC + 0.033BC - 0.65A^2 – 0.0006B^2 + 0.87C^2	0.9988	<0.0001

ANOVA results for multiple regression analysis and response surface quadratic model of $Y_{1,}$ Y_2 were evaluated using the corresponding F, p and R^2 values (Table 4). F values of $Y_1$and Y_2 were calculated to be 1041.74; 653.29, both leading to a p-value<0.05, suggesting the both the models were statistically extremely significant. The models' coefficient of determination (R^2) were 0.9993and 0.9988, indicating that more than 99.93% and 99.88% of the response variability were explained, and supporting a good accuracy and ability of the established model within the range limits used. The F-values of Lack of Fit of $Y_{1,}$ Y_2were 3.88; 3.30, respectively, implying that the Lack of Fit was not significant relative to the pure error. This indicated that the accuracy of the polynomial model was adequate.

Table4: Regression coefficients of the predicted second- order polynomial models for the total phenolic compoundsand moisture content.

Source	Y_1 – TPC		Y_2 – MC	
	F- value	p- value	F- value	p- value
Model	1041.74	<0.0001***	653.29	<0.0001***
A	13.40	<0.0081**	866.09	<0.0001***
B	1897.08	<0.0001***	323.65	<0.0001***
C	13.57	0.0078**	155.76	<0.0001***
AB	4.04	0.0844^{NS}	351.10	<0.0001***
AC	30.77	0.0009***	2.29	0.1740^{NS}
BC	1.55	0.2528^{NS}	1.64	0.2413^{NS}
A^2	5.39	0.0532^{NS}	954.21	<0.0001***
B^2	450.62	<0.0001***	0.64	0.4500^{NS}
C^2	0.51	0.4986^{NS}	108.66	<0.0001***
Lack of Fit	3.88	0.1118^{NS}	3.30	0.1396^{NS}
R^2	0.9993		0.9988	

*p< 0.05; **p< 0.01; ***p< 0.001; NS: non-significant.

3.2. Optimization and Model Verification

Based on the empirical second- order polynomial model, the experimental data was analyzed by RSM using the Design- Expert 7.0 software. The X- and Y- axes of the three- dimensional response surfaces represented two factors, for maltodextrin ratio and inlet drying temperature(feed flow rate = 25ml/min), inlet drying temperatureand feed flow rate (maltodextrin ratio = 10%), maltodextrin ratio and feed flow rate (inlet drying temperature = 150 °C). The Z- axes represented one of the four evaluation indices (TPC, MC). Twodimensional response surfaces were constructed as depicted in Fig 1.

A

B

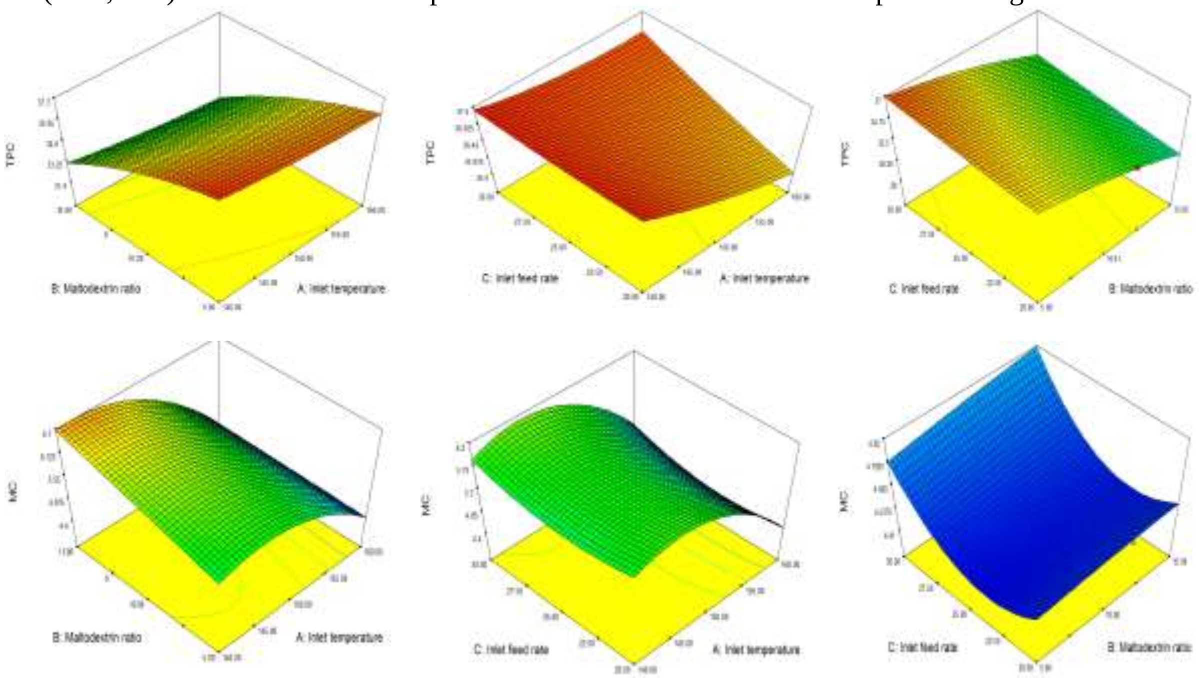

Fig 1: Response surface of TPC and MC. (A) TPC, (B) MC .

The optimal values of the independent variables were obtained by solving second - order regression equations using a numerical optimization method. Experimental data suggested the existence of optimization of total phenolic compound and moisture content occurred with feed flow rate 26.05ml/min, Mmaltodextrin ratio 5%at 160^{o}C.

Table 5: Optimum conditions, predicted and experimental values of responses on spray drying of *Trametes versicolor*extract[a].

Independent variables			Dependent variables (Response)	Optimum value	
A	B	C		Experimental[b]	Predicted
5	160	26.05	Y_1	35.76±0.04	36.3266
			Y_2	4.43±0.02	4.4817

[a]A, Maltodextrin ratio (%); B, Inlet drying temperature; C, Feed flow rate(ml/min); Y_1, TPC (mgGAE/g); Y_2, MC (%). [b]Mean ± standard deviation (SD) of four determinations (n= 4) from two crude extract replications.

4. CONCLUSION

This study showed that the optimal conditions for spray drying from *Trametes versicolor* mushroom (V.C) extracts. The optimal conditions for process spray drying from *T. versicolor* extracts were found to be maltodextrin ratio of 5%w/v, inlet drying temperature of 160^{o}C and feed flow rate of 26.05ml/min. The experimental values of Total Phenolic Content (TPC) and Moisture Content (MC) were 35.76±0.04mg GAE/g, 4.43±0.02%.

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TỐI ƯU HÓA ĐIỀU KIỆN SẤY PHUN ĐỐI VỚI DỊCH CHIẾT NẤM VÂN CHI (*TRAMETES VERSICOLOR*)

Tóm tắt. Trong nghiên cứu này, chúng tôi tiến hành tối ưu hóa các điều kiện sấy phun đối với dịch chiết nấm vân chi (*Trametes versicolor*), sử dụng phương pháp đáp ứng bề mặt (RSM). Nấm vân chi (1kg) được ngâm chiết với ethanol 65% (30L) ở 70^0C trong thời gian 7 giờ. Sau đó, dịch chiết được lọc bỏ phần rắn, loại bỏ dung môi dưới áp suất thấp đạt độ ẩm 20%. . Chúng tôi tiến hành nghiên cứu ảnh hưởng của ba yếu tố độc lập, tỉ lệ maltodextrin (% kl/v), tốc độ dòng (ml/phút) và nhiệt độ sấy đầu vào (^{0}C) nhằm khảo sát tổng hàm lượng phenolic (TPC) và độ ẩm (MC). Điều kiện tối ưu để sấy phun dịch chiết nấm vân chi được xác định là tỷ lệ maltodextrin 5% kl/v, nhiệt độ sấy đầu vào 160^oC và tốc độ dòng cấp liệu 26,05ml / phút. Giá trị thực nghiệm của TPC và MC là 35,76 ± 0,04mg GAE / g, 4,43 ± 0,02%.

Từ khóa.Độ ẩm (MC), *Trametes versicolor*, tổng hàm lượng phenolic (TPC), phương pháp đáp ứng bề mặt (RSM)

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