The reseach on the double low rapeseed protein concentrated by a new preparation method and its functional properties

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Abstract

The extraction condition was optimized by Response Surface Methodology. Results show that 70% ethanol water solution at 60°C, with the rate of 8.85:1, washing for six times and 20 minutes each, were optimum for defatted double-low rapeseed. By this way, the rapeseed protein concentrate reached 62.48% in protein content and was light in color, bland in taste. In the product, glucosinolation can be totally taken off and phytic acid descended by 60%. Its nitrogen solubility, water sorption, oil sorption, emulsifiability and foamability were studied and improved also. The product was fitting to be used as food additive.

Key words: double low rapeseed protein concentrate, protein, phytic acid, response surface methodology, the functional properties.

Introduction

Rapeseed, one of the most important oilseed crops cultivated in the world is becoming of increasing interest as a source of edible protein. Rapeseeds contain 35~47% of protein, and hence defatted rapeseed meal may constitute a good source of proteins for humans. Its amino acid composition is well-balanced in regard to FAO requirements.Moreover, oilseed protein is rich in sulfue-containing amino acids and lysine which are generally limited in legumes and cereals.

The preparation for the rapeseed protein concentrate is the way of extracting glucosinolation, phytic acid, tannin and so on from defatted rapeseed rapeseed, removing non soluble protein, causing the protein content to concentrate approach 65% in rapeseed protein product. Profits from other oil protein sources to take out phytic acid, the dissolution of characteristic difference between protein and phytic acid were used to separate them. The glucosinolation is soluble in water and the polyphenol can be in alcoholate.

In the present paper, the double low defatted rapeseed meal was used as material for the rapeseed protein concentrated. The ethanol water solution was chose as solvent and RSM was designed to optimize the parameter in experiment. It was also discussed the functional properties of the rapeseed protein product. The results and data could provide a theoretical basis for extensive application of concentrated rapeseed protein in food industry.

Materrials and methods

The double low defatted rapeseed meal: After the double low rapeseed cleaning up, the rapeseed's wetness was adjusted to 6% in the drying oven and the drying oven's temperature was controlled under 45°C to prevent protein denaturation. Then the rapeseed was pelled off by the rice huller, defatted by ether for 48 hours, and smashed.

Table 1 The material's mainly composition

Composition	The double low defatted rapeseed
Crude protein(%)	37.86
Fibre (%)	5.37
Fat content (%)	0.30
Wetness (%)	5.31
Tannin (%)	1.21
Phytic acid (mg/g)	10.38
Iso thiocyante (mg/g)	0.80
Oxazolinethioketone (mg/g)	2.39

All chemicals incluing ethanol, ether were of analytical grade.

The content of protein determination: The content of protein was determined according to the micro-Kheldahl method. Crude protein content was calculated using a convertion factor of 6.25.

The phytic acid's content determination: The phytic acid's content was determined according to trichloroacetic acid(TCA) method.

Optimization of preparation conditions: The double low rapeseed protein concentrate was preparated by 70% ethanol water solution. A three-factor central composite design was employed to examine the response, the content of protein and phytic acid's content as changed with the independent variables, the rate of solution and defatted rapeseed(X1), washing times(X2) and how many mintues of each time(X3). A quadratic polynomial regression model was assumed for predicting the

response. Every factor (code X1 to X3) had three levels corresponding to three code values. There were totally 15 independent experiments. In every experiment, levels of the factors were arranged according to table 2. The model proposed was described in table 3. Experimental data were analyzed for response surface regression for a quadratic polynomial model using SAS software.

Table 2 Design of factors and levels in experiment

Factor	Code	Code value	Level
The rate	X_1	+1	9:1
		0	8:1
		-1	7:1
Washing times	X_2	+1	7
		0	6
		-1	5
Minutes of each time	X_3	+1	30
		0	20
		-1	10

Table 3 Different levels of factors arranged in experiments

Test number	Code value of experiment		
	X_1	X_2	X_3
1	-1	-1	0
2	-1	0	-1
3	-1	0	+1
4	-1	+1	0
5	0	-1	-1
6	0	-1	+1
7	0	+1	-1
8	0	+1	+1
9	+1	-1	0
10	+1	0	-1
11	+1	0	+1
12	+1	+1	0
13	0	0	0
14	0	0	0
15	0	0	0

Amino-acid analysis: Amino-acid analysis of HCl-hydrolyzed samples was carried out an automated Beckman instrument. This work was completed by the amino-acid analysis service of the Oil Institute of the Chingese Academy of Agricultural Sciences(CAAS). All amino acid data were corrected for 100% recovery.

Results

Optimization of technology for the double low rapeseed protein concentrate's preparation:Results of 15 experiments were shown in table 4. Content of protein and phytic acid were used as response values in analysis of response surface regression(RSREG). The equation $Y= a_0+ a_1X_1+ a_2X_2+a_3X_3+a_{11}X_1^2+ a_{22}X_2^2+a_{33}X_3^2+a_{12}X_1X_2+ a_{13}X_1X_3+ a_{23}X_2X_3$ was used as regression model. The procedure RSREG of SAS also gave values of paramrter estimated (table 5) and predicted values of the equation (table 6).

Table 4 The content of protein and phytic acid of 15 experiments

Test number	Protein(%)	Phytic acid(mg/g)
1	57.77	9.33
2	59.40	10.00
3	52.64	11.00
4	59.72	10.33
5	57.92	10.50
6	58.15	9.16
7	60.38	9.83
8	59.50	7.33
9	58.05	6.00
10	58.11	6.83
11	57.68	6.53
12	55.88	7.17
13	61.78	6.05
14	62.51	5.93
15	62.56	6.07

Table 5 Parameters estimated by regression model

Parameters	Protein	Phytic acid
a_0	62.28	6.02
a_1	0.024	-1.52
a_2	0.45	-0.04
a_3	-0.98	-0.14
a_{11}	-3.23	0.54
a_{22}	-1.20	1.65
a_{33}	-2.10	1.54
a_{12}	-1.03	0.04
a_{13}	1.58	-0.83
a_{23}	-0.28	-0.29

Table 6 Predicted values of regression model

Response values	The rate	Washing times	Minutes of each time	Calculated value	The type
Protein (%)	7.89	6.27	17.1	62.49	max
Phytic acid (mg/g)	9.82	6.04	25.4	4.59	min

Variance analysis of regression equation was conducted(table 7,8). F value of the model was bigger than $f_{0.05}(9,5)$. R^2 was 0.973 and 0.990, which showed that linear relationship between dependent variable and whole indepengent variables was significantly distinct.

Table 7 Variance analysis of regression equations of the protein's content

Variance source	Degree of freedom	The protein's content		
		Sum of square	Mean square	F value
Model	9	77.56	8.62	9.69*
Error	5	4.44	0.89	
Correct total	14	82.00		
Linearly dependent coefficient		R^2=0.973		

Table 8 Variance analysis of regression equations of the phytic acid's content

Variance source	Degree of freedom	The protein's content		
		Sum of square	Mean square	F value
Model	9	39.45	4.38	26.56**
Error	5	0.83	0.17	
Correct total	14	40.28		
Linearly dependent coefficient		R^2=0.990		

Figure 1and 2 were response surface diagrams of the protein and phytic acid's content.

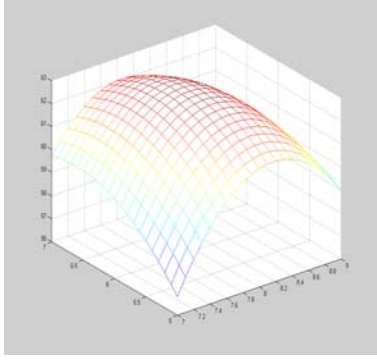

Figure 1 (1) :X-washing times Y-the rate Z-protein minutes of each time=17.1min

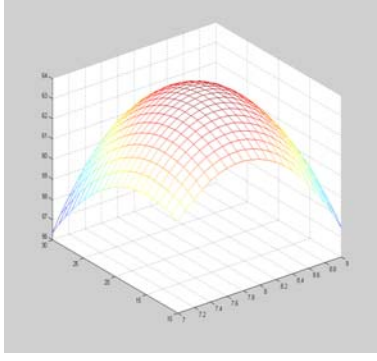

Figure 1 (2) :X- minutes of each time Y- the rate Z- protein washing times =6.27

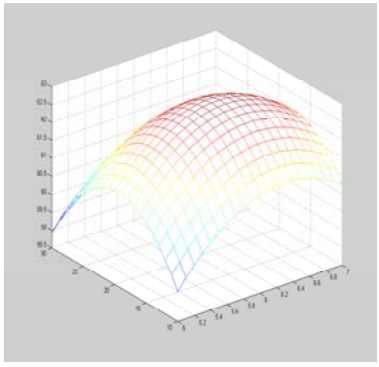

Figure 1(3): X- minutes of each time Y- washing times Z- protein the rate =7.89

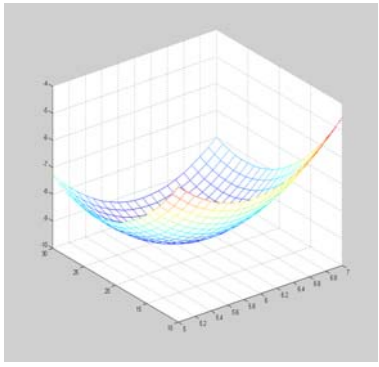

Figure 2(1):X- minutes of each time Y- washing times Z-phytic acid the rate =9.82

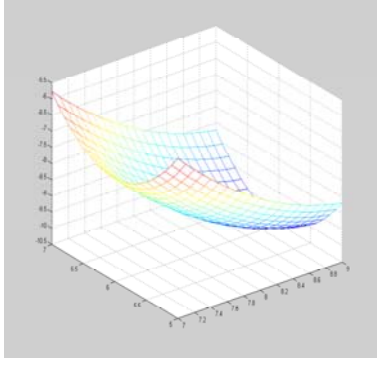

Figure 2 (2) :X- washing times Y- the rate Z- phytic acid minutes of each time =25.4min

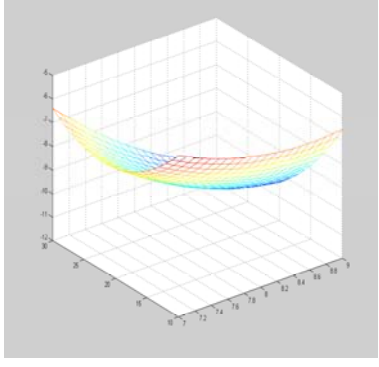

Figure 2 (3) :X- minutes of each time Y- the rate Z- phytic acid washing times =6.04

Discussion

Considering the interaction of all the variables, the optimum conditions for the preparation of double low rapeseed peotein concentrate can be calculated by the assumed equation as follows:70% ethanol water solution at 60°C, with the rate of 8.85:1, washing for six times and 20 minutes each.

By this way, the double low rapeseed protein concentrate reached 62.48% in protein content and was light in color, bland in taste. In the product, glucosinolation can be totally taken off and phytic acid descended by 60%. The 61% of the material can be gained.

The functional properties of double low rapeseed protein concentrate:The functional properties incluing solubility, water sorption, oil sorption, emulsifiability, foamability and so on can have the influence on the physics or chemical property to food quality. In recent years, protein products in food application are considered about not only its nutrition but also the physico-chemical properties. Therefore,the resrch on the functional properties of the double low rapeseed protein concentrate is extremely essential. The experiments were compared the functional properties of the double low rapeseed protein concentrate(RPC) and the soybean protein concentrate(SPC).(Table9,Figure3,4,5,6)

Table 9 The solubility of the RPC and SPC(pH=7.0)

Product	Soluble protein(%)	Nitrogen solubility(NSI,%)
RPC	2.12	3.37
SPC	3.47	5.10

Results showed the RPC's nitrogen solubility was lower for the protein denaturation, the RPC's water sorption and oil sorption were higher than SPC. But the RPC's emulsifiabilityand foamability were not as good as the SPC.

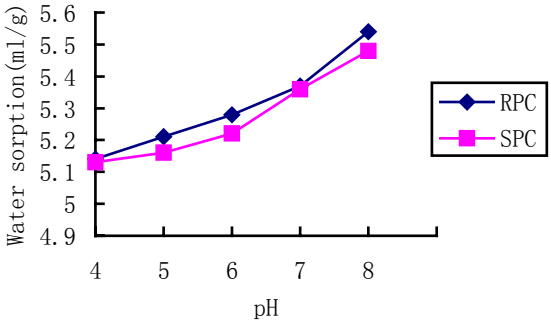

Figure 3 The water sorption of protein products

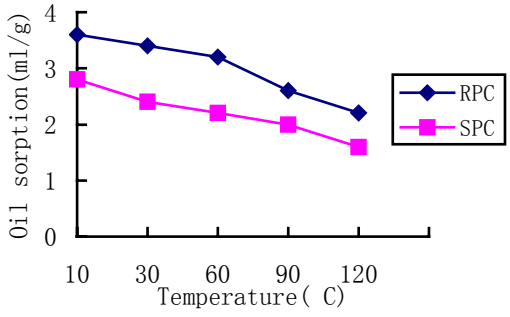

Figure 4 The oil sorption of protein products

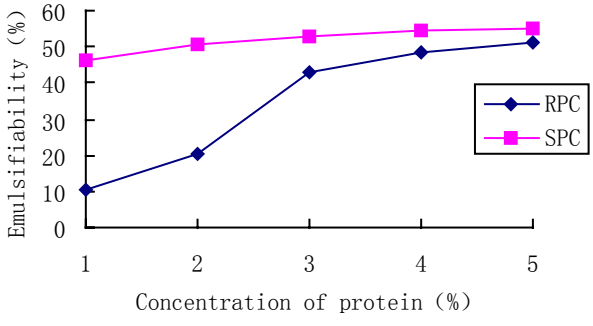

Figure 5 The emulsifiability of protein products

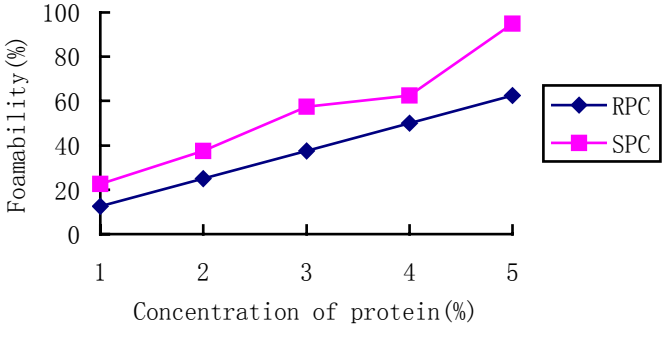

Figure 6 The foamability of protein products

Conclusions

The double low rapeseed protein concentrate was prepared by 70% ethanol water solution. The optimum conditions were established by RSM. These parameters included temperature: 60°C, with the rate of 8.85:1, washing for six times

and 20 minutes each. By this way, the rapeseed protein concentrate reached 62.48% in protein content and was light in color, bland in taste. In the product, glucosinolation can be totally taken off and phytic acid descended by 60%. The 61% of the material can be gained.

Table 10 The product's mainly composition

Composition	The double low rapeseed protein concentrate
Wetness(%)	7.24
Crude protein (%)	62.48
Fat content (%)	0.28
Fibre (%)	6.73
Ash content (%)	4.08
Tannin (%)	0.130
Phytic acid (mg/g)	4.62
Glucosinolation (mg/g)	not detected

Table 11 The product's composition of amino-acid

Amino-acid	Content(g/100g)	Amino-acid	Content(g/100g)
Aspartic acid	8.87	Methionine	1.55
Threonine	4.43	Isoleucine	4.06
Serine	4.13	Leucine	5.32
Glutamic acid	19.74	Tydroxyproline	2.90
Glycine	4.21	Phenylalanine	3.99
Alanine	4.39	Histidine	1.86
Valine	4.35	Lysine	1.83
		Arginine	2.40

The research on the functional properties of double low rapeseed protein concentrate showed the RPC's nitrogen solubility was lower for the protein denaturation, the RPC's water sorption and oil sorption were higher than SPC. But the RPC's emulsifiabilityand foamability were not as good as the SPC.

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