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The effect of high-moisture extrusion on the water holding capacity of pea protein texturized products

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Abstract: *Single factor experiments and Box-Behnken principle were used to optimize the extrusion parameters. The results showed that the changes of the three operating parameters of screw speed, extrusion temperature and moisture have a great impact on the water holding capacity (WHC) of that texturized products. The single factor experiments determined that the best range of screw speed is 130 rpm-170 rpm, the best range of cooking temperature is 160 ℃-180 ℃, and the best range of moisture is 50%-60%. After optimization by response surface analysis, the optimal process parameters were determined to be the screw speed of 149 rpm, the extrusion temperature of 167 ℃, the moisture of 56%, the WHC value of 158.16%.*

Key words: *Pea; Extrusion; Response surface analysis; water holding capacity*

With the increasing demand for meat and the shortage of animal protein, the development of meat substitutes has gradually become a new development trend. At the same time, people's requirements for food health and nutrition have increased, and animal meat has not been selected due to long-term consumption of food-borne diseases[1] . Pea is the second soybean crop in the world. It is rich in plant protein resources. The development of plant protein texturized products using pea protein as a raw material has become a research hotspot in recent years[2].R Alonso[3]et al. found that high-moisture extrusion cooking results in changes in the structure of pea protein, forming a three-dimensional network structure with higher water holding capacity, lower oil holding capacity and solubility. Osen[4]et al. extruded pea protein to form a fibrous product with a muscle-like texture, and found that its product can be a good substitute for meat. The amino acid composition of pea protein is relatively balanced. Among the 8 essential amino acids contained in the human body, all other amino acids except methionine have reached the FAO/WHO recommended standards[5].At present, the use of pea protein is mainly used in feed production, causing a waste of resources[6].Therefore, the research of pea protein texturized products not only enriches the research content of plant protein texturized products, but also becomes an important way to broaden the application field of pea protein and increase its added value[7].

High-moisture extrusion cooking is a mechanical process used to process continuous heating of food ingredients. It is a new food processing technology that integrates various processing units such as raw material conveying, mixing, melting, and extrusion molding[8].Under high temperature and high pressure conditions, a series of operations such as shearing, homogenization, compression, flow alignment, and sterilization can be realized in a short time. The twin-screw high-moisture extrusion cooking has the characteristics of high process integration, high efficiency and low energy consumption, and is used in the production of plant protein texturized products. The product has a rich fiber structure and a texture closer to meat, so it can be eaten directly without rehydration [8].Plant protein texturized products meet the health requirements of modern people and reduce the risk of cardiovascular and cerebrovascular diseases[9].According to the analysis of the extrusion model, the operating parameters of screw speed, cooking temperature, and moisture have a direct impact on plant protein texturized products. According to the analysis of the extrusion model, the operating parameters of screw speed, cooking temperature, and moisture have a direct impact on plant protein texturized products, and the quality of plant protein meat can be controlled by changing the operating parameters[10] .It is of great significance for optimizing the production process and improving the production level of plant tissue protein industrialization.

In this study, pea protein isolate was used as a raw material to make texturized products, and the product quality was changed by controlling the screw speed, cooking temperature, and moisture, and WHC was used as an evaluation index.The indicators for evaluating plant protein texturized products include many indicators such as texture and color. WHC is mostly used to measure food quality. It is an important quality control indicator in the evaluation of texturized products. Increasing WHC can significantly improve the hardness, elasticity, and taste of texturized products.The purpose of this study is to explore the influence of extrusion operation parameters (screw speed, cooking temperature, moisture) on the WHC of pea protein texturized products, so as to optimize the production process and provide a basis for the industrial production of pea protein texturized products.

I. Materials and Methods

1.1 Raw material

Pea protein isolate, purchased from Shandong Jianyuan Biotechnology Co., Ltd., with a content of 75%.

1.2 Equipment

Co-rotating twin-screw extruder, UVTE36-24 type, Changsha Chuangxiang Food Technology Co., Ltd.

1.3 Sample making method

A combination test of screw speed, cooking temperature, and moisture is carried out.Cooking temperature setting: There are five temperature zones in the sleeve, and the main temperature zone is zone *III*. During the test, the other temperature zones, namely, zone *I* 40 °C; zone *II* 120 °C; zone *IV* 145 °C; zone *V* 140 °C remained unchanged during the extrusion process. The feeding speed is 10 kg/h.

The raw materials are discharged from the solid feeder and enter the extruder conveying box through the feed port. Water is pumped into the extruder from the liquid feeder. The mixture is melted, sheared, and extruded in the extruder meshing box. It enters the extrusion mould through the die head and forms from the discharge port. The samples are weighed regularly and stored in the incubator for testing.

1.4 Single factor experimental design

1.4.1 Single factor test of screw speed

Screw speed setting: 100, 125, 150, 175, 200rpm.

Temperature setting: 40 °C in zone *I*; 120 °C in zone *II*; 170 °C in zone *III*; 145 °C in zone *IV*; 140 °C in zone *V*.

Moisture setting: 55%.

1.4.2 Single factor test of cooking temperature

Cooking temperature setting(zone *III*):150, 160, 170, 180, and 190°C.

Screw speed setting: 150 rpm.

Moisture setting: 55%.

1.4.3 Single factor test of cooking moisture

Moisture setting: 45%, 50%, 55%, 60%, 65%.

Temperature setting: 40 °C in zone *I*; 120 °C in zone *II*; 170 °C in zone *III*; 145 °C in zone *IV*; 140 °C in zone *V*.

Screw speed setting: 150 rpm.

1.5 Box-Behnken experimental design

Through the single factor experiment, the level value range of each operating parameter was determined. On this basis, the orthogonal experiment was designed using the Box-Behnken principle to optimize the process. Taking screw speed (A), cooking temperature (B), and moisture (C) as the influencing factors, taking WHC as the response values R_1, and designing a quadratic regression equation with three factors and three levels to simulate Combine influencing factors and indicators to obtain optimal process parameters. The factor level coding is shown in Table 1.

Table 1 Independent variables and test design levels

Level	Factors		
	Screw speed (rpm)	Cooking temperature(°C)	Moisture (%)
-1	130	160	50
0	150	170	55
1	170	180	60

1.6 Determination of water absorption

Take 10 g of the dried sample (calculated as W1 after weighing), rehydrate at 60 °C for 2 h, take it out and place it in a cool place for 5 minutes and then weigh it (calculated as W2 by weight) according to the following formula.

$$WHC = \frac{W_2 - W_1}{W_1} \times 100\ \%$$

II. Results and analysis

2.1 Influence of screw speed on WHC

When the screw speed is below 150rpm, it has a positive effect on the WHC of the extruded product, and when the screw speed is above 150rpm, it has a negative effect on the WHC of the extruded product (the results are shown in Fig. 1). When the screw speed decreased, the material receives insufficient shearing force in the extruder, the hydrogen bond is not fully broken, the product organization is insufficient, and the WHC decreased. When the screw speed increases, the shear force of the material during extrusion increases. Under the combined action of pressure and temperature, the protein denatures, expands, and reorganizes, and the hydrogen bond breaks gradually. The formation of disulfide bonds increases, the peptide chain rearranges and cross-links to form a stable spatial structure, which leads to an increase in WHC. When the screw speed continues to increase, the material flows in the extruder faster, and the short reaction time leads to reduced texturized and lower WHC. In general, the WHC of texturized products is not greatly affected by the screw speed. According to the test results, 130rpm-170rpm is selected as the best screw speed range.

2.2 Influence of cooking temperature on WHC

When the cooking temperature is lower than 170 ℃, the cooking temperature has a positive effect on the water absorption of the extruded product, and when it is higher than 170 ℃, it has a negative effect (The results are shown in Fig. 2). When the temperature is lower than 160 degrees Celsius, the protein is not heated enough and the denaturation is insufficient.The product presents a state of compactness and less pore structure, resulting in a decrease in WHC. As the temperature increases, the degree of melting of the material in the extruder increases, the movement of water molecules intensifies, the pressure increases, and the protein cross-linking and recombination are gradually sufficient. The extruder works stably, the product surface is smooth and uniform. The shape structure is formed and stable, and the WHC is improved.This is consistent with the results of Yang Zhen et al[11]. When the temperature continues to rise,the evaporation rate of water is faster, the movement of water molecules will be more violent and uncontrollable, the formed product will be uneven, and the discharge port will be prone to spraying. According to the test results and comprehensive consideration of various factors, 160℃-180℃ is selected as the best cooking temperature range.

2.3 Influence of moisture on WHC

From the test results, there is an overall positive effect between the WHC and moisture of the sample（The results are shown in Fig. 3）. When the moisture content is 45%, Material melting effect is not obvious, the extruder discharges slowly, and it is easy to block the mold. The accumulation of materials in the extruder has an obvious effect, the pressure rises, the product is dry and hard, and the structure is too dense, which makes it difficult to form a pore structure. With the increase of water content, the molten state of the material becomes more sufficient, there is enough water for protein denaturation, reorganization, and cross-linking, the state of the extruder tends to be stable, the degree of texturized of the product is improved, and the product is uniform and shiny. The internal pore structure increases with the increase of moisture, and the WHC increases. When the moisture is 65%, the ratio of the material to the moisture is unbalanced, resulting in the product becoming loose and broken, it cannot be selected as an ideal product. According to the test results, choose 50%-60% as the best moisture range.

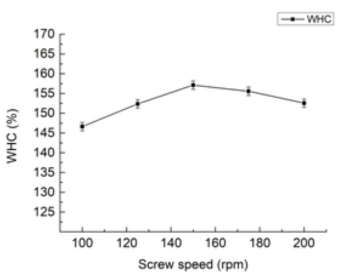

Fig. 1 Influence of screw speed on WHC

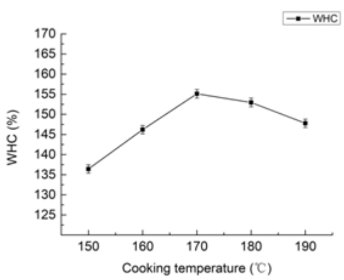

Fig. 2 Influence of cooking temperature on WHC

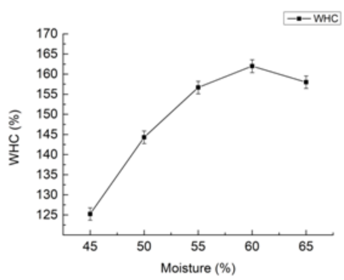

Fig. 3 Influence of moisture on WHC

2.4 Response surface method to optimize the preparation process

2.4.1 Experimental design and results

The Box-Behnken experiments design and result are shown in Table 2:：

Table 2 Box-Behnken experiments design and result

Number	x_1(rpm)	x_2(℃)	x_3(%)	WHC(%)	Number	x_1(rpm)	x_2(℃)	x_3(%)	WHC(%)
1	1	0	-1	143.44	10	-1	0	-1	145.46
2	-1	0	1	153.85	11	-1	1	0	154.43
3	0	0	0	157.02	12	1	-1	0	151.24
4	0	1	-1	143.84	13	0	-1	1	155.20
5	0	0	0	158.60	14	1	0	1	153.42
6	0	0	0	156.48	15	-1	-1	0	150.62
7	0	-1	-1	143.03	16	0	0	0	157.29
8	0	1	1	140.26	17	0	0	0	156.62
9	1	1	0	154.69					

Taking screw speed (x_1), cooking temperature (x_2), and moisture (x_3) as independent variables, taking WHC (R_1) as the response value, data analysis is carried out through the software Design-Expert, and the model is established as follows:

$$R_1 = 157.60 - 0.20x_1 - 0.86x_2 + 3.37x_3 - 0.090x_1x_2 + 0.40x_1x_3 - 3.94x_2x_3 - 0.70x_1^2 - 4.16x_2^2 - 7.86x_3^2$$

The variance analysis is shown in Table 3.

Table 3 Variance analysis

Source	Sum of Squares	df	Mean Square	F	Prob > F	Significant
Model	516.11	9	57.35	5.45	0.0180	*
A	0.31	1	0.31	0.029	0.8690	
B	5.90	1	5.90	0.56	0.4785	
C	90.86	1	90.86	8.63	0.0218	*
AB	0.032	1	0.032	0.003	0.9573	
AC	0.63	1	0.63	0.060	0.8135	
BC	62.02	1	62.02	5.89	0.0456	*
A2	2.05	1	2.05	0.20	0.6720	
B2	72.81	1	72.81	6.92	0.0339	*
C2	260.19	1	260.19	24.71	0.0016	*
Residual	73.70	7	10.53			
Lack of Fit	70.53	3	23.51	29.69	0.0034	*
Pure Error	3.17	4	0.79			
Cor Total	589.81	16				

"Prob > F" < 0.05 is significant, which is indicated by "*" .

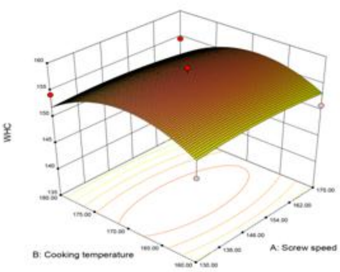

Fig. 4 The interaction of screw speed and cooking temperature

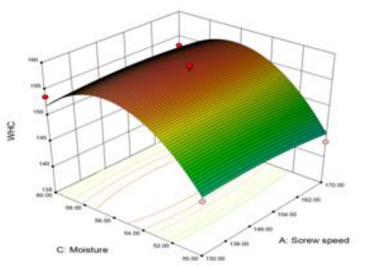

Fig. 5 The interaction between screw speed and moisture temperature

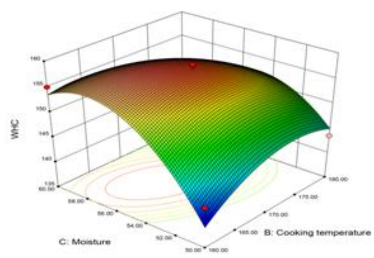

Fig. 6 The interaction between cooking temperature and moisture

2.4.2 WHC response surface analysis

The regression model between the independent variable and the dependent variable is significant ($p < 0.05$), and the lack of fit item is not significant ($p > 0.05$) (Table 5). The analysis of variance showed that the R^2 value was 0.8750 and the Adjusted R^2 value was 0.7144, indicating that the regression model is credible. Response surface analysis shows the overall trend of WHC changes is to increase firstly and then decrease. When the screw speed is reduced, the material receives insufficient shearing force, resulting in a low degree of product texturization. The increase in the screw speed and the too short residence time of the material in the shearing module of the extruder make it difficult to form the product structure and reduce WHC. Only the screw speed is suitable, the product has the best WHC (Fig. 4). When the screw speed is constant, the WHC rises first and then decreases with the increase of the cooking temperature. Lower temperature affects protein denaturation. At higher temperatures, water evaporates quickly, and the product integrates into a scorched paste with lower WHC. The water absorption of the product will change greatly with the change of moisture, no matter which value the screw speed is fixed to. Showing a trend of first increasing and then decreasing. The WHC of 60% moisture samples is higher than the WHC of 50% moisture samples (Fig. 5). The interaction between cooking temperature and moisture has a significant effect on the water absorption of the product. When the water is fixed, the water absorption of the product first rises and then decreases with the increase of the cooking temperature. When the cooking temperature is fixed, the water absorption of the product first increases and then decreases as the moisture increases. Overall, moisture has a positive effect on the WHC of extruded products (Fig. 6). This is consistent with the results of Kaur Maninder [12] et al.It can be seen from the quadratic regression equation that the interaction between cooking temperature and moisture is a significant factor affecting the WHC.

Through the F test (Table 3), it is found that the influence of the operating parameters on the index is: C > B > A, that is, moisture > cooking temperature > screw speed. The relationship between cooking temperature and screw speed is consistent with the research results of Yang Yong et al.The best parameters obtained through response surface analysis are screw speed: 149.04 rpm; cooking temperature: 167.70 ℃; moisture: 56.36%. The optimal value of WHC response value R_1 is 158.16%. Considering the actual test factors and the operating accuracy of the extruder, the process parameters are set as screw speed: 149 rpm; cooking temperature: 167 ℃; moisture: 56%. Under this process condition, verified by three parallel experiments, the average product WHC is 158.15%, the predicted value of the quadratic regression equation is consistent with the actual value, and the process parameters have been optimized.

III. Conclusion

The operating parameters of screw speed, cooking temperature, and moisture will have a great impact on the WHC. Through single factor tests, the optimal range of screw speed is determined to be 130 rpm-170 rpm. The optimum range of temperature is 160 ℃-180 ℃, and the optimum range of moisture is 50%-60%. After response surface optimization and comprehensive consideration, it is determined that the best process parameters are screw speed: 149 rpm; cooking temperature;167 ℃;moisture: 56%, and the optimal water absorption value is 158.16%.

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