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The Effect of Extrusion Puffing Process on the Gelatinization Degree of Starch in Carp Feed and Intestinal Glucose Absorption Was Investigated

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ABSTRACT
In the process of aquaculture feed production, the processing technology plays a crucial role, and different processes will directly affect the characteristics of the feed which will affect the absorption and development of fish and so on. In the application of extrusion pelleting process, through the combined action of high temperature, high pressure and shear force, it can effectively improve the degree of starch gelatinization of feed and the absorption of intestinal glucose, and it also has the effect of sterilization, that the feed has the characteristics of safety and stability. Based on this, this paper starts with the principle of extrusion pelleting process and analyzes it through experimental design, explores the influence of this technology on the degree of starch gelatinization and intestinal glucose absorption of carp feed.

KEYWORDS
Extrusion-puffing Process; Carp feed; Starch gelatinization degree; Intestinal glucose absorption degree

1 The principle of extrusion puffed technology

Extrusion puffed technology belongs to a kind of food processing technology. Through the physical pressure the original material, the original material is made to undergo chemical changes. In the process of extrusion puffy, the high-speed rotating screw in the extruder is used to, friction and extrusion the original material. After the original material is treated with strong physical work, it produces a lot of heat and the intermolecular force inside it also changes After the thermal energy is produced by mechanical action, the external steam heating, electric heating and other devices of the extruder will continue to heat the original material, so that the temperature the original material quickly rises. Based on the synergistic effect of high temperature and high shear force, the moisture contained in the original material is quickly vaporized, but because mechanical equipment is a closed device, the steam can not be discharged normally, so the pressure of the environment where the original material is located will increase, when the temperature reaches 150-200 ℃, the pressure can reach more than 1 MPa. At this time, the original material is in a molten state, the molecular structure loose and it has fluidity.

When the raw material is extruded from the mold, the original high pressure state will quickly drop to the normal pressure state, and the volume can expand about 2000 times under the action of thermal steam expansion. During the extremely expansion process, a large impact force will be generated, which will cause the internal structure of raw material to be destroyed, and then a loose and porous structure will be formed. During the expansion of the raw material, the raw starch in the grain will quickly convert into starch, the hydrogen bond in the starch molecule will break, and the original crystal structure will be destroyed after the destruction, and the digestibility of the feed will be greatly. At the same time, the protein in the raw material has undergone denaturation, and the molecules have been rearranged, and some of the fat contained has been oxid and decomposed, greatly improving the nutritional value of the feed, and the taste is better.

2 Experimental design of extrusion puffing process

In the process of studying the effect of extrusion puffing process on the gelatinization degree of starch carp feed and the intestinal glucose absorption, the extrusion puffing process experiment was designed, and the experiment materials, scheme and so on are as follows.

2.1 Materials selection for experiments

The experimental carps were purchased from local qualified aquaculture households, and healthy carps with strong physique lively and consistent weight were selected, with an average body weight of 50.00 ± 5.00g. The main raw materials of the basic feed were meal, soybean meal, corn starch, wheat bran, fish oil, calcium dihydrogen phosphate,

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premixed complex vitamin, premixed complex mineral, etc. and all kinds of raw materials were fresh, non-mouldy, non-impurities, qualified raw materials that meet the feed hygiene standard and quality standard. The extrusioning equipment used was a twin-screw extrusion puffing machine, which has the characteristics of high production efficiency, stable operation, easy adjustment of various important indicators such as temperature pressure, screw speed, etc. during the extrusion process to meet the requirements of feed processing preparation under different experimental conditions. Temperature range room temperature ~ 200 ℃ accuracy ± 1 ℃, screw speed 50 ~ 500r/min.

Other experimental instruments and reagents: analytical balance with an accuracy of 0.0001g to weigh feed raw materials and experimental samples;-speed crusher to crush feed raw materials to achieve the corresponding particle size to make mixing uniform; electric heating constant temperature drying oven to dry samples and determine moisture content; muffle to determine ash content; Kjeldahl nitrogen determinator to determine crude protein content of feed and fish body; Soxhlet extractor to determine crude fat content; enzyme meter to determine amylase gelatinization degree, corresponding enzyme activity; centrifuge to separate each component of the sample; anhydrous ether, concentrated sulfuric acid, sulfate, potassium sulfate, sodium hydroxide, hydrochloric acid, amylopectinase, 3, 5-dinitrosalicylic acid other chemical reagents are all of analytical purity.

2.2 Preparation of feed

On the basis of the basic feed formula, four different gelatinization degrees of experimental feeds were prepared by adjusting the extrusionelleting process parameters. The specific process parameters are shown in the following Table 1.

Table 1 Process parameter setting

Experimental group	Temperature (℃)	screw speed(r/min)	Moisture content(%)
Group A	120	150	18
Group B	135	200	20
Group C	150	250	22
Group D	165	300	24

2.3 Experimental plan

The gelatinization degree of each feed was tested by enzymatic hydrolysis method. The raw materials of each formula were weighed and mixed evenly. The raw materials were crushed by high-speed universal crusher (particle size 0.30 mm). After extrusion pelleting, material could be evenly heated and subjected to shear force. The crushed materials were sent into the feed hopper of the double-screw extrusion pelletizer, and then extr pelletizing was carried out according to the set process parameters. The extrusion pelletized feed was cut into particles of appropriate length by a cutting knife device. After natural cooling drying, the moisture content of the feed was reduced to 10% ~ 12%, and the feed was packed.

A total of 360 healthy carps were selected and randomly divided into four groups, namely A, B, C, and D. The groups of carps were then placed in four indoor breeding ponds of the same size, equipped with circulating water systems and aeration devices, to provide fresh water quality and sufficient dissolved for this experiment. In addition, the water temperature of the indoor breeding ponds was kept constant at 25 ± 2℃, with a pH of 7.08.0 and ammonia nitrogen content of <0.05 mg/L. On this basis, an 8-week experiment was carried out. During the experiment, was fed at fixed times every day at 9:00 and 16:00, with a feed input of 3%-5% of the carp's weight. The amount of feed was adjusted stage by stage according to the feeding situation and growth situation of the carp, so as to ensure the scientificity of the carp farming feed input.

At the end of this experiment, 5 individuals of the carp were randomly selected from each of the four groups, and then the intestinal tissue of thep was immediately removed, and the tissue was equally divided into 2 parts, one part was used for the determination of enzyme activity related to intestinal glucose absorption, and the part was used for the determination of the gene and protein expression levels of the intestinal glucose transporter (SGLT1 and GLUT2), and the average value of five data in each group was taken as the final determination result.

3 The effect of extrusion puffing process on the gelatinization degree of starch in carp feed and intestinal glucose absorption; result analysis

3.1 The effects of extrusion expansion technology on the starch gelatinization degree of carp feed

The measurement results of the starch gelatinization degree of carp feed under different extrusion expansion process conditions are shown in Table 2.

Table 2 Measurement results of the gelatinization degree of starch in carp feed under different extrusion puffing process conditions

Experimental group	Temperature (℃)	screw speed(r/min)	Moisture content(%)	Starch gelatinization degree(%)
Group A	120	150	18	62.35±2.13
Group B	135	200	20	70.45±2.56
Group C	150	250	22	78.67±3.02
Group D	165	300	24	87.56±3.54

As can be seen from the data results in Table 1, with the increase in extrusion temperature, the gelatinization degree of feed starch has been significantly improved. The starch gelatinization degree has increased to 87.56% from 120°C to 165° C, and it has risen to 83.21% when the rotational speed has increased from 150 r/min to 300 r/min.Similarly, the moisture content also affects the gelatinization degree of starch. When the moisture content has increased to 24%, the gelatinization degree has risen from 68.24% at 18% moisture content to 85.67%. Moisture is one of the essential factors affecting starch gelatinization. Sufficient moisture is conducive to the full absorption of water by starch granules, enabling the starch granules to fully expand and absorb water, which is beneficial to the development of gelatinization.

The results of this study show that, firstly, the extrusion expansion process parameters such as temperature, screw rotational speed, and moisture content all have a direct impact on the starch gelatinization degree of carp feed. Under the influence of the temperature factor, with the increase in extrusion temperature, the starch gelatinization degree is significantly improved. This is because the temperature provides the energy required for the thermal motion of starch molecules to be enhanced, thus breaking the hydrogen bonds between starch molecules and causing the starch granules to absorb water and expand, resulting in starch gelatinization.Secondly, the increase in screw rotational speed can also promote the improvement of the starch gelatinization degree. The increase in screw rotational speed leads to an increase in the shearing force of the materials in the extruder, which is conducive to the destruction of the structure of starch granules, allowing them to come into contact with water and gelatinize.Thirdly, moisture content is one of the important factors affecting starch gelatinization. Sufficient moisture can ensure that starch granules fully absorb water and expand, making the starch gelatinization reaction more complete. In this study, when the moisture content was increased from 18% to 24%, the starch gelatinization degree increased significantly.

3.2 The influence of extrusion-puffing process on glucose absorption in the intestines of carp

The results of the determination of the expression level of intestinal glucose transporters and glucose absorption rate in carp fed with different feed are shown in Table 3.

Table 3 Results of the study on the glucose absorption in the intestines of carp by the extrusion puffing process

Experimental group	SGLT1 Relative gene expression level	GLUT2 Relative gene expression level	SGLT1 Relative protein expression levels	GLUT2 Relative protein expression levels	Glucose absorption rate(μmol/g・h)
Group A	1.00±0.05a	1.00±0.06a	1.00±0.08a	1.00±0.09a	25.67±2.13a
Group B	1.54±0.10b	1.78±0.12b	1.45±0.12b	1.67±0.15b	32.45±2.56b
Group C	1.98±0.15c	2.34±0.18c	1.89±0.15c	2.23±0.18c	38.67±3.02c
Group D	2.56±0.20d	3.12±0.25d	2.45±0.20d	3.01±0.25d	45.78±3.54d

It can be seen from Table 3 that as the degree of gelatinization of feed starch increased, the relative expression levels of sodium-gluc cotransporter (SGLT1) and glucose transporter (GLUT2) genes and proteins in the intestines of carp showed an upward trend. The relative level of SGLT1 gene in group D with 87.56% gelatinization degree was 2.56, which was significantly higher than that in A (1.00), B (1.54), and C (1.98) ($P < 0.05$); the relative expression of GLUT2 gene was 3.12, which was also significantly higher than that in the other three groups ($P < 0.05$); In terms of expression, the gray value of SGLT1 and GLUT2 bands in group D was significantly higher than that in the other groups, and the relative expression levels were 245 and 3.01, respectively, after analysis by ImageJ software, and their change trend was the same as that of the relative expression level of genes.

Overall, the changes in the rate of glucose absorption were consistent with the trend in the expression of transport proteins. The rate of glucose absorption in D was the highest, reaching 45.78 μmol/g・h, which was significantly higher than that of the other three groups ($P < 005$), and the lowest in group A, which was only 25.67 μmol/g・h. The extrusion puffing treatment changed the gelization degree of starch and had a significant effect on the absorption of glucose in the carp intestine. With the increase of the gelatinization degree, the expression levels ofGLT1 and GLUT2 genes and proteins in the carp intestine increased significantly, and the rate of glucose absorption also increased significantly. This indicated that feed with highatinization degree could improve the active transport and facilitated diffusion capacity of intestine for glucose by promoting the expression of transport proteins. Due to the easy digestion of starch in high gelization feed into glucose, resulting in an increase in intestinal glucose levels, this might be an important inducement for the expression of transport protein genes. The results of the study showed the rise in intestinal glucose levels could activate the related

signaling pathways, induce the transcription and translation of the SGLT1 and GLUT2 genes, and then induce the expression synthesis of transport proteins. High gelatinization feed may also have an impact on the morphology and function of the intestine, such as increasing the length and surface area of intestinali, increasing the activity of intestinal digestive enzymes, and further promoting the absorption of glucose.

4 Conclusion

In summary, the results of this study have important guiding significance for optimizing the feed formula for carp, guiding production practice, and improving the of feed processing technology. In the process of feed processing, the process parameters of extrusion pelleting can be regulated to increase the gelatinization degree of feed starch, thus improve the digestibility and nutritional value of feed, promote the growth of carp, reduce the feed coefficient, and improve the breeding benefit. The use of this process not only to reduce feed waste and reduce breeding costs, but also reduces the pollution caused by feed residues in the aquatic environment, so that the carp breeding industry can take a green sustainable development path.

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