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## OPTIMIZATION OF PROTEIN HYDROLYSIS CONDITIONS TO INCREASE THE NUTRITIONAL AND FUNCTIONAL VALUE OF SAUSAGES

**Abstract.** The study included control samples without the enzyme and experimental samples with the addition of 6 ml of the enzyme, hydrolyzed at temperatures of 30°C, 40°C and 50°C for 1, 2, 3 and 4 hours. The results showed that the introduction of the enzyme significantly increases the content of amine nitrogen, especially at a temperature of 40°C and a duration of 3 hours, which was determined as optimal conditions. Under these conditions, the maximum amine nitrogen content of 2.10 mg/g was achieved. The prototypes obtained under optimal conditions demonstrated excellent organoleptic and functional properties. The hydrolysate has good fluidity and uniform texture, which is important for its further use in food products. The introduction of hydrolysate into sausages can lead to a number of improvements, such as an increase in nutritional value by enriching the product with easily digestible amino acids and peptides, an improvement in texture due to moisture binding and an increase in viscosity, as well as an improvement in taste qualities without changing the traditional taste and appearance of the product.

**Keywords:** protein, wool by-products, processing, secondary meat products, enzymatic hydrolysis.



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**Introduction.** The study of amine nitrogen during the hydrolysis of the legs of cattle (cattle) to obtain protein hydrolysate is relevant for several reasons. Firstly, protein hydrolysates are widely used in the food industry, medicine and sports nutrition due to their high nutritional value and easy digestibility [1]. Secondly, the use of cattle legs, which are a byproduct of the meat industry, contributes to the rational use of resources and waste reduction [2]. An important aspect is that protein hydrolysis makes it possible to obtain peptides and amino acids, which can have various functional properties, including antioxidant and antimicrobial activity [3]. In addition, hydrolysis methods can affect the degree of

hydrolysis and the composition of amine nitrogen, which directly affects the quality of the final product [4].

To date, the study of optimal hydrolysis conditions, including the choice of enzymes and reaction conditions, is key to improving the efficiency of protein hydrolysate production [5]. It is also important to note that amine nitrogen is one of the important indicators reflecting the degree of hydrolysis and bioavailability of proteins [6]. In the context of growing demand for high-quality protein products, the development of effective methods for obtaining protein hydrolysates from cattle legs is of great importance. Moreover, such research can contribute to the creation of new products with improved functional properties, which meets current trends in healthy nutrition and sustainable development.

**Materials and methods.** The following were used as the starting material for the study: beef legs with a putty joint (purchased at the specialized butcher shop "Green Market" in Almaty), the enzyme BLT 7 (produced by the National Center of Biotechnology, Astana, Republic of Kazakhstan) and commercial Protease from *Bacillus licheniformis* (P4860, Sigma, Denmark). The PH value was determined using acid-base indicators according to GOST 51478-99. The mass fraction of proteins was determined according to GOST 25011-2017. The organoleptic assessment according to GOST 6658-2016 is based on determining the compliance of organoleptic indicators of product quality with the requirements of regulatory and technical documentation.

**Research results.** Enzymatic hydrolysis was carried out for 1,2,3,4 hours at a temperature of 30°C, 40°C, 50°C. To begin with, we weigh a 500 ml flask on the AMPUT brand electronic scales, then fill it with a sample of the product (crushed tissues of cooked beef legs) in the amount of 33 grams, evenly distributing the sample over the flask. Next, distilled water with a volume of 65 ml is poured, 1% is added with BLT 7 enzyme (in 2 flasks), PS. The fermentation time was determined according to objective indicators. It is mixed with a glass stick. Thus, we get 6 flasks with experimental samples. 2 experimental samples treated with 1% BLT 7 enzyme are placed in a TS-1/80 SPU thermostat preheated to 45°C. 4 experimental samples treated with 1% BLT 7 enzyme, and PS enzyme are placed in a Climo-Shaker ISF1-X brand shaker preheated to 45°C with 150 rpm (rpm). After the expiration of the time of 1, 2, 3, 4 hours of enzymatic hydrolysis, samples are taken into test tubes and centrifugation is carried out to separate particles from the solution at a temperature of 40°C for 30 minutes at a rate of 10.8 rcf using a Centrifuge 5415 R brand device. The determination of the dry matter content in the product under study is carried out by drying at a temperature of 90°C to a constant mass for 30 minutes using a VIBRA brand device (Fig. 1).

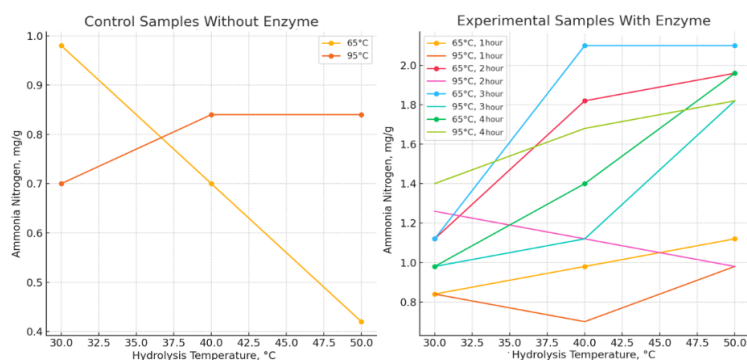


Fig. 1. Investigation of amine nitrogen in the obtained hydrolysates

To assess the depth of hydrolysis, the method of formal Serensen titration was used, which allows us to determine the content of amine nitrogen in samples. The experiment was carried out at different temperatures and duration of hydrolysis, both with and without the addition of the enzyme. For control samples hydrolyzed at a temperature of 65°C, a decrease in the content of amine nitrogen is observed with an increase in the hydrolysis temperature. At 95°C, the amine nitrogen content remains stable at temperatures of 40°C and 50°C. The addition of the enzyme significantly increases the content of amine nitrogen. At 65°C, the maximum content of amine nitrogen (2.10 mg/g) is achieved at 3 hours of hydrolysis and temperatures of 40°C and 50°C. At 95°C, the maximum content of amine nitrogen (1.82 mg/g) is achieved at 3 hours of hydrolysis and a temperature of 50°C. With longer periods of hydrolysis (3 hours), the maximum content of amine nitrogen is achieved.

Organoleptic studies have shown positive consumer properties of the resulting protein hydrolysate. Volumetric hygroscopic powder of homogeneous mass, pleasant light color, with a slight noticeable smell of raw materials. Based on the conducted research, a technological scheme for the production of protein hydrolysate from wool by-products is proposed (Fig. 2).

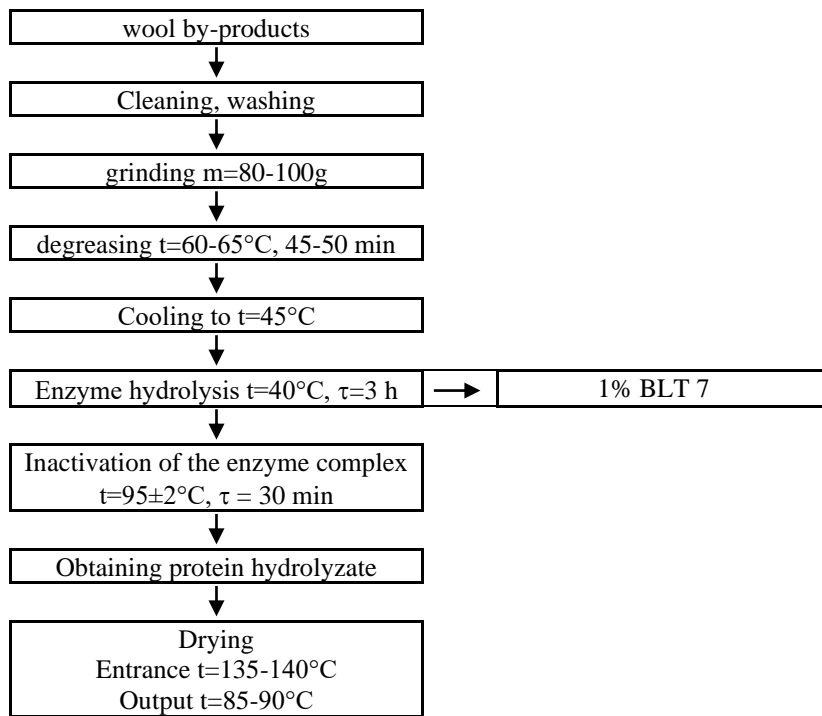


Fig. 2. Technology for the production of protein hydrolysate from wool by-products

During the hydrolysis of meat and bone raw materials, one of the main criteria is fat content, with a fat content of 15-20%, the process of obtaining protein hydrolysates becomes more complicated. One of the most common ways to degrease bones is heat treatment. Heating the raw materials denatures the proteins, making it easier to extract fat from the bones. During experimental studies, the shins of horses, cows, and sheep are stored at a temperature of 0-6°C and, no later than 8 hours after cleaning from bones, they are handed over for degreasing. If

necessary, these secondary products can be stored at a temperature of  $-18^{\circ}\text{C}$  for no more than two months. Taking into account the peculiarities of the preparation of raw materials, the technological process was processed and a technological scheme for obtaining protein hydrolysate was proposed.

Samples of cow, horse and sheep paws were selected for experimental studies. As the optimal criteria for choosing an enzyme preparation for the purpose of shin hydrolysis, the pH value was determined to reflect the activity (pH 7-9) corresponding to the range of raw solutions containing collagen. According to the instructions for use of the enzyme preparation *Bacillus licheniformis*, the home proteolytic enzyme BLT 7 (*Bacillus licheniformis*) and the commercial protease *Bacillus licheniformis* have an optimal pH value of 7.5. the temperature optimum for both enzyme preparations is  $45^{\circ}\text{C}$ . To study the effect of enzyme preparations on connective tissue raw materials, beef, horse and sheep shins processed using traditional processing technology of meat and bone by-products used in meat processing enterprises were used. The shins are cut with a band saw into discs 15-20 mm wide and weighing 80-100 g. Since the reaction of enzymatic hydrolysis of proteins in an aqueous medium occurs on the basis of literature data, in this dissertation work it was decided to degrease meat bone raw materials in a wet way. 200 ml of distilled water was added to 100 g of shin and the suspension was heated at a temperature of  $60-65^{\circ}\text{C}$  for 40-45 minutes. Waste oil was isolated. The fat-free bones were separated, and the meat part was sent to a fermentation container.

These results show the same effect as the commercial PS enzyme and the BLT 7 enzyme extract. Examination of control and experimental samples (determination of pH). Studies have shown that a processed enzyme extract of 1% BLT7 and commercial PS were achieved for protein hydrolysis after 3 hours in all samples. When the hydrolysis time was increased by more than 3 hours, the samples had a foreign odor. Thus, as a result of a series of experiments, 1% BLT 7 was selected as an enzyme extract for the production of protein hydrolysate. After completion of the hydrolysis process, the substrate is heated to a temperature of  $95 \pm 2^{\circ}\text{C}$  for 30 minutes to inactivate the enzymes and thermocoagulate the residual protein. The resulting protein hydrolysate is filtered and sent to dry. The drying of the hydrolysate was carried out on a spray dryer Spray Dryer NSP-1500. Mode: drying temperature is  $135-140^{\circ}\text{C}$  at the inlet and  $85-90^{\circ}\text{C}$  at the outlet. The results of the organoleptic evaluation of protein hydrolysate are presented in Table 1.

Table 1

## Results of organoleptic evaluation of protein hydrolysate

Name of indicators	Description
Appearance	Dry product with hygroscopic uniform powder-like consistency
Colour	Light blond color
Smell	The smell of the resulting raw materials is characteristic, not pronounced

Thus, the effectiveness of using 1% BLT 7 as an enzyme extract preparation is justified, which ensures the hydrolysis of beef leg proteins. The technological scheme for obtaining protein hydrolysate from wool by-products has been improved and proposed.

**Discussion of scientific results.** The resulting hydrolysate has good flowability and uniform texture, which is important for its further use in food

products. Hygroscopicity indicates the ability of the product to absorb moisture from the environment, which may require additional packaging to prevent clumping. The light color of the hydrolysate indicates minimal changes in proteins and the absence of significant thermal effects that could lead to darkening of the product. This is a positive indicator, since the light color facilitates the use of hydrolysate in various food products without changing their appearance. The unobtrusive smell is a positive quality, as it allows the use of hydrolysate in a wide range of food products without the risk of changing their flavor. This is especially important for products where preserving the original taste and smell is critical.

Protein hydrolysate obtained under optimal conditions (40°C, 3 hours, 2.10 mg/g of amine nitrogen) has high functional and organoleptic properties, which makes it an excellent additive to sausage products. The introduction of hydrolysate into the sausage formulation can lead to the following improvements, such as: increased nutritional value: hydrolysate enriches the product with easily digestible amino acids and peptides, improving its biological value; texture improvement: hydrolysate can help improve the texture of sausages by binding moisture and increasing viscosity; improved taste qualities: the unobtrusive smell and light color of the hydrolysate allow it to be used in sausages without changing their traditional taste and appearance.

**Conclusion.** The study showed that the introduction of the enzyme significantly increases the efficiency of protein hydrolysis. The optimal conditions for the hydrolysis of protein hydrolysate from the legs with a putty joint of cattle were determined as a temperature of 40°C and a duration of 3 hours, at which the maximum amine nitrogen content of 2.10 mg/g is achieved. This hydrolysate can be effectively used in sausage products to improve its nutritional and functional properties.

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#### **ШҰЖЫҚТАРДЫҢ ТАҒАМДЫҚ ЖӘНЕ ФУНКЦИОНАЛДЫҚ ҚҰНДЫЛЫҒЫН АРТТЫРУ ҮШІН АҚУЫЗ ГИДРОЛИЗИНІҢ ШАРТТАРЫН ОҢТАЙЛАНДЫРУ**

**Аңдатпа.** Зерттеуге 1, 2, 3 және 4 сағат ішінде 30°C, 40°C және 50°C температурада гидролизденетін 6 мл фермент қосылған ферментсіз бақылау үлгілері мен прототиптер кірді. Нәтижелер ферментті еңгізу амин азотының құрамын айтарлықтай арттыратынын көрсетті, әсіресе 40°C температурада және ұзақтығы 3 сағат – оңтайлы жағдайлар ретінде анықталды. Бұл жағдайда амин азотының максималды мөлшері 2,10 мг/г құрайды. Оңтайлы жағдайларда алынған прототиптер керемет органолептикалық және функционалдық қасиеттерді көрсетті. Гидролизат жақсы сұйықтыққа және біркелкі құрылымға ие, бұл оны тамақ өнімдерінде одан әрі пайдалану үшін маңызды. Шұжықтарға гидролизатты енгізу өнімді оңай сіңетін аминқышқылдары мен пептидтермен байыту арқылы тағамдық құндылығын арттыру, ылғалды байланыстыру және тұтқырлықты арттыру арқылы құрылымды жақсарту және өнімнің дәстүрлі дәмі мен сыртқы түрін өзгертпестен дәмді жақсарту сияқты бірқатар жақсартуларға әкелуі мүмкін.

**Тірек сөздер:** ақуыз, жүнді субөнімдер, өңдеу, қайталама ет өнімдері, ферментативті гидролиз.

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#### **ОПТИМИЗАЦИЯ УСЛОВИЙ ГИДРОЛИЗА БЕЛКОВ ДЛЯ ПОВЫШЕНИЯ ПИТАТЕЛЬНОЙ И ФУНКЦИОНАЛЬНОЙ ЦЕННОСТИ КОЛБАСНЫХ ИЗДЕЛИЙ**

**Аннотация.** Исследование включало контрольные образцы без фермента и опытные образцы с добавлением 6 мл фермента, гидролизующиеся при температурах 30°C, 40°C и 50°C в течение 1, 2, 3 и 4 часов. Результаты показали, что введение фермента значительно повышает содержание аминного азота, особенно при температуре 40°C и продолжительности 3 часа, что было определено как оптимальные условия. При этих условиях достигнуто максимальное содержание аминного азота 2,10 мг/г. Опытные образцы, полученные при оптимальных условиях, продемонстрировали отличные органолептические и функциональные свойства. Гидролизат обладает хорошей текучестью и однородной текстурой, что важно для его дальнейшего использования в пищевых продуктах. Введение гидролизата в колбасные изделия может привести к ряду улучшений, таких как повышение питательной ценности за счет обогащения продукта легко усваиваемыми аминокислотами и пептидами, улучшение текстуры благодаря связыванию влаги и увеличению вязкости, а также улучшение вкусовых качеств без изменения традиционного вкуса и внешнего вида продукта.

**Ключевые слова:** протеин, шерстные субпродукты, переработка, вторичные мясные продукты, ферментативный гидролиз.