ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

New Types Of Protein Products Based On Modified Blood Plasma.

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ABSTRACT

This article presents the results of a study of the feasibility of using blood plasma modified by enzymatic hydrolysis as the basis for the production of whipped beverages. For this, the conditions for obtaining blood plasma hydrolyzate were selected, a composition of a dry protein basis was proposed; physicochemical and functional-technological properties were studied, as well as the biological value of the developed dry protein basis. Evaluation of amino acid composition showed the usefulness of the protein of the product and its high biological value. The study of the functional and technological properties of the dry base solution revealed high foaming, multiplicity and stability of the foam. Thus, the developed dry protein base can be recommended for the production of whipped cocktails enriched with high-grade animal protein.

Keywords: protein products, blood plasma, enzymatic hydrolysis, collagenase, animal protein.

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ISSN: 0975-8585

INTRODUCTION

The existing protein deficiency in human diets necessitates an increase in the volume and expansion of the range of production of protein-containing products. The important point is the quality of the protein, which requires a search for unconventional sources and ways of its rational use. One of the most valuable sources of complete animal protein is the blood plasma of slaughter animals. The high biological value of the protein composition of blood plasma causes a high coefficient of its digestibility, and the presence of such functional and technological properties as the ability to gel and foaming, makes it possible to use it in various food products [1, 2, 7, 8].

In the category of functional products, a significant role is given to beverages that can not only meet the body's need for fluids, but also serve as a source of scarce food components, playing the role of a tool for the prevention of nutrition-related diseases. Drinks are an excellent basis for the enrichment of vitamin-mineral complexes, making dietary fiber, saturation with proteins, amino acids and other essential components, becoming their sources for the human body. In this regard, it is of interest to study the methods of deep processing of the liquid fraction of the blood of slaughter animals - plasma, as the basis for the production of biologically valuable beverages [3, 4, 5, 6].

At the same time, one of the limitations of using blood plasma is a short shelf life, which requires the selection of a preservation method that ensures the safety of its nutritional qualities and functional properties.

The use of heat treatment to increase the storage capacity of blood plasma does not give the desired bactericidal effect, since it is not allowed to heat the product above 52-55 °C due to the denaturation of proteins. One of the ways to solve this problem is the directional change in the structure of plasma proteins, which would not undergo degradation during heat treatment in the temperature range of 60-65 ° C. This can be achieved by carrying out hydrolysis to obtain amino acids and peptides, which have a greater thermal stability due to the shorter length of the molecule.

In addition, hydrolysis enhances the biological value of raw materials by increasing the digestibility of the protein substrate.

Objective. Justification of the prospects of using modified blood plasma as a basis for the production of biologically valuable beverages.

Objectives of the study: to select the conditions for obtaining blood plasma hydrolyzate, to propose a composition of a dry protein basis; to study the physico-chemical and technological properties, as well as the biological value of the developed dry product.

MATERIALS AND METHODS

Experimental studies were conducted on the basis of the department of technology of production and processing of agricultural products, as well as in the accredited educational and scientific testing laboratory of the Stavropol State Agrarian University.

As the objects of study were used: blood plasma of cattle; Proteolytic enzyme preparations: megaterin, collagenase, protosubtilin and trypsin; liquid and dry blood plasma hydrolysates; plasma-based foam and dry protein base.

In the course of research, the main indicators were determined by the following methods: protein content - by the Kjeldahl method; carbohydrate content - by Bertrand method; ascorbic acid content - by calculation; dry matter content - by the refractometric method; active acidity - by potentiometric method; solubility index - according to GOST 30305.4-95; amino acid composition - on the AAA-400 amino acid analyzer by standard methods; microbiological indicators - by the standard method; amino acid fast and biological value - by calculation method according to N.N. Lipatov; the multiplicity of foam was established as the ratio of its volume to the volume of the solution that went to the formation of foam; foam stability was fixed by the time of destruction of the foam column. The triple repetition of experiments, mathematical processing was carried out in the program Microsoft Office Excel 2007.

ISSN: 0975-8585

RESULTS AND DISCUSSION

In the production of healthy foods, the most appropriate use of enzymatic hydrolysis, which allows you to initiate profound changes in the structure of proteins. In addition, the use of enzyme technologies will ensure environmental friendliness of technological processes.

The advantages of enzymatic hydrolysis should also include the softness of flow conditions with obtaining a mixture of hydrolysis products with the lowest degree of racemization of amino acids.

In studies, the optima of the action of proteolytic enzymes at different pH and temperature values were studied to establish the parameters of their greatest activity.

Sodium caseinate at pH = 7 was used as a substrate for determining the temperature optimum (Fig. 1).

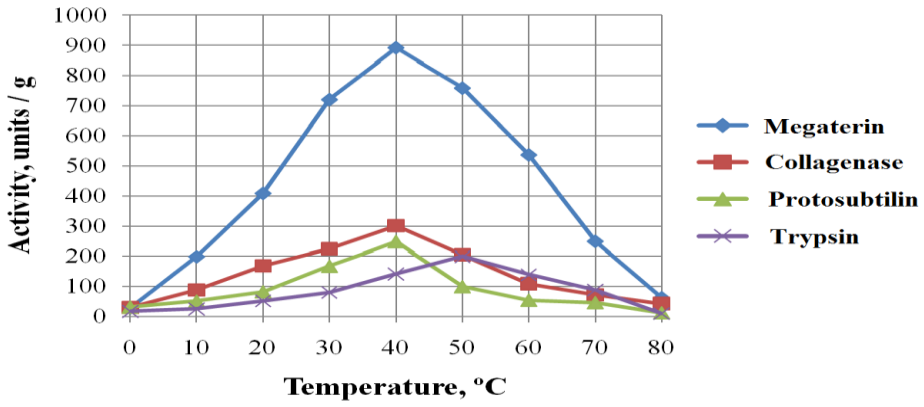

Figure 1: Effect of temperature on the activity of enzyme preparations

It is established that megaterine, Protosubtilin and Collagenase show maximum activity at temperatures of 37-40°c. For Trypsin optimal action is defined in the temperature range 48-52 ° C.

The results of studying the influence of active acidity of the environment on the proteolytic activity of enzymes are presented in Figure 2.

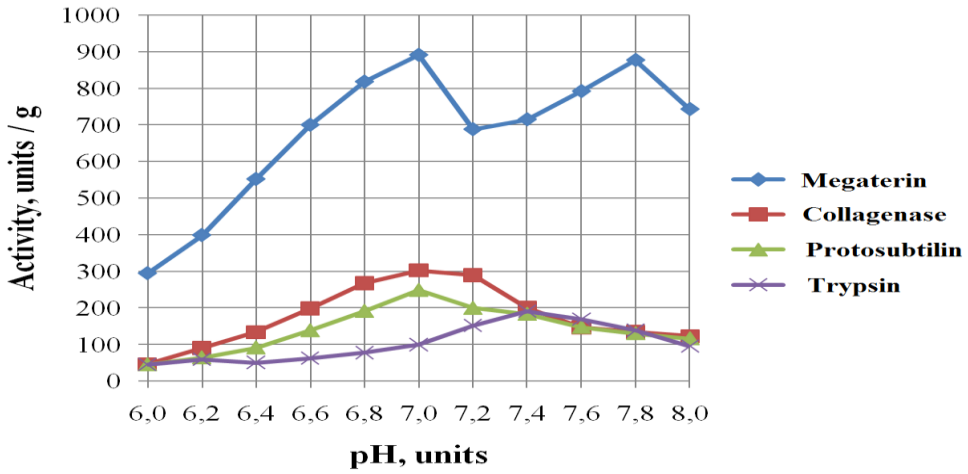

Figure 2: Effect of pH on the activity of enzyme preparations (t = 37 ° C)

The maximum activity of protosubtilin and collagenase is achieved at pH 6.8 - 7.2, for trypsin at t = 37 °C the optimum pH value is 7.4 units, and the maximum activity is reached at pH = 8 (t = 50 °C) . For megaterin,

ISSN: 0975-8585

two peak activity values (pH = 7.0; pH = 7.8) were noted, which can be attributed to the heterogeneous nature of this enzyme preparation.

Further studies were aimed at studying the effectiveness of the use of these enzyme preparations for the hydrolysis of plasma proteins. On the basis of preliminary experimental studies, it was found that for successful hydrolysis it is necessary to ensure that the ratio of protein to substrate in the system is 100: 1, as a result of which the blood plasma (substrate) must be diluted with 1: 1 water. The results of studying the effect of enzyme dosages on the efficiency of hydrolysis are presented in Figure 3.

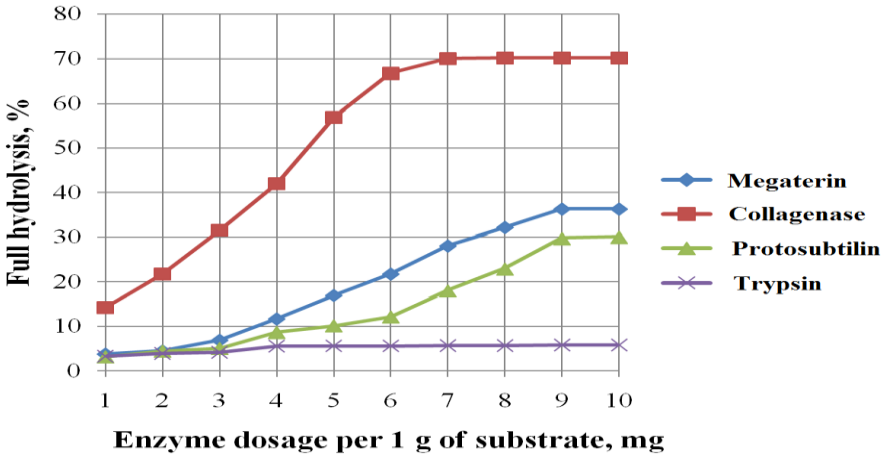

Figure 3: Dynamics of hydrolysis of plasma proteins by various enzymes

It was established that the introduction of the collagenase preparation into the system in quantities of more than 7×10^{-3} g of the enzyme per 1 g of the substrate is impractical because it does not lead to a further increase in the degree of hydrolysis of proteins. For megaterin, the maximum value of the complete hydrolysis is achieved at a concentration of 3.8×10^{-3} g, protosubtilin - 3.0×10^{-3} g, which is slightly lower than for collagenase.

Evaluation of hydrolytic indicators allowed to establish that the drug collagenase provides the most complete course of hydrolysis, forming 2.9 times more free amino acids in comparison with megaterin, including 1.7 times more essential.

For further research, enzyme preparations that showed the greatest efficacy — collagenase and megaterin — were used.

The results of studying the effect of the duration of hydrolysis on its completeness are presented in Figure 4.

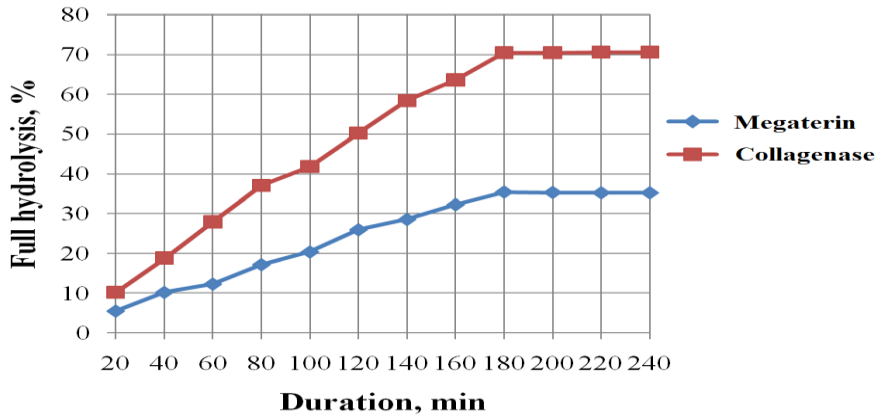

Figure 4: Effect of the duration of hydrolysis on its completeness

It was established that the hydrolysis proceeds most intensively for 2.8–3.2 hours, the further course of the process was characterized by a sharp decrease in the accumulation of hydrolysis products. Evaluation of the obtained data showed that the drug collagenase provides a significantly greater completeness of hydrolysis.

Applying regression analysis using multivariate planning to process the obtained experimental data, optimized parameters for the hydrolysis of blood plasma proteins by the collagenase preparation were obtained (Table 1).

Table 1: The parameters of the enzymatic hydrolyzate of plasma proteins

Process parameters	Value
Mass fraction of enzyme, %	0,35
Active acidity, units	6,8-7,2
Temperature, °C	35-40
Duration, h	3,0-3,5
Concentration of protein in the substrate,%	3,5

The resulting hydrolyzate was further subjected to pasteurization at a temperature of 56 - 58 °C, with a concomitant decrease in pH to 3.5-4.5 with the help of various additives. This avoided the denaturation of non-hydrolyzed proteins, precipitation, and also caused the inactivation of the enzyme.

For convenience of further use and increase in storage capacity, the resulting hydrolyzate was dried in a spray dryer, which prevented the denaturation of proteins and ensure the high solubility of the final product in water.

It was decided to use dry hydrolyzate to develop a dry basis for the preparation of cocktails. In addition to the dry hydrolyzate, fructose, as an alternative to beet sugar, and ascorbic acid were added to the dry base for cocktails (Table 2).

Table 2: Dry cocktail base formulation

Component	Consumption of raw materials, kg / 100 kg of product
Dry hydrolyzed blood plasma	56,8
Vitamin C	3,4
Fructose	39,8

The results of the study of physico-chemical, organoleptic indicators and amino acid composition of the proposed dry basis for cocktails are presented in tables 3 and 4.

Table 3: The results of the study of physico-chemical and organoleptic characteristics of the dry basis for cocktails

Indicators	Value
Dry matter content,%	86,7
Solubility,%	96,0
Bulk mass, kg / m^3	310,4
Titratable acidity, °T	20,0
Protein content,%	46,8
Carbohydrate content,%	37,1

Ascorbic acid content, mg / 100 g	1900,0
Taste and smell	Sweet-sour, pleasant aroma and taste of protein present
Colour	Light cream, homogeneous throughout the product
Structure	Powdered, lumps easily destroyed

Table 4: Results of the study of the amino acid composition of the dry base for cocktails

Amino acids	Amino acid content, g / 100 g of product	Amino acid score, %	Utilization factor
Threonine	2,36	59,0	0,571
Methionine	1,18	33,71	1,000
Lysine	4,0	72,73	0,464
Leucine	5,19	74,14	0,455
Phenylalanine	4,2	70,0	0,482
Isoleucine	1,6	40,0	0,843
Valin	1,84	36,8	0,916
Tryptophan	0,7	70,0	0,482

By calculation, it was established that the coefficient of differences in the amino acid protein content of the product is 23.33%, its biological value is 76.67%, and the ratio of comparable redundancy is 0.2650.

An important condition for the possibility of using the developed dry basis for the production of whipped cocktails is the presence of such technological properties as good foaming properties, multiplicity and durability of the foam. The control sample was a solution of dry blood plasma. The mass fraction of protein in the control and experimental samples was 5%, the temperature of the samples was in the range of 4-6 ° C. The research results are presented in table 5.

Table 5: Results of the comparison of the functional properties of dry blood plasma and the developed protein basis

Component	Foaming capacity, cm^3 per time interval, with					Frequency ratio of foam for a time interval, with				
	30	60	120	180	240	30	60	120	180	240
Solution of dry blood plasma	171,0	199,0	248,0	226,0	294,0	1,4	1,7	2,1	2,7	2,9
Dry base solution	197,0	219,0	272,0	295,0	318,0	1,9	2,0	2,7	3,0	3,2

The dry base solution for whipped cocktails was characterized by the best values of foam formation, the multiplicity and stability of the foam, slightly exceeding the control sample based on dry blood plasma. Perhaps this is due to the presence of fructose and ascorbic acid in the composition of the dry protein basis. The stability of the foam solution of dry blood plasma was about 40 minutes, while the dry protein base retained the resulting volume for up to 1.5 hours.

The study of storage ability revealed the absence of pathogenic microflora in a dry product with a shelf life of up to 12 weeks at a temperature not higher than 4 ° C. The indicator total microbial countremained within 2.4×10^3 CFU in 1 g of the product.

ISSN: 0975-8585

CONCLUSION

Conducted studies allowed to establish the optimal parameters of the enzymatic hydrolysis of the blood plasma of slaughter animals by the proteolytic preparation collagenase. Dry protein base on the basis of blood plasma hydrolyzate is characterized by high quality indicators and can be recommended as a basis for the production of whipped cocktails enriched with high-grade animal protein.

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