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Obtaining Ingredients Based on Egg and Plant Protein Mixtures with High Functional and Technological Properties

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Abstract.
Most people source protein from animal products such as eggs, meat, and dairy products. However, they are more expensive than plant sources of protein and their consumption can provoke severe allergic reactions. Enzymatic hydrolysis can reduce the allergenicity of egg white, which has the most balanced amino acid composition. The functional properties of egg white can be improved by mixing it with plant protein hydrolysates. This study aimed to select optimal conditions for obtaining mixtures of egg albumin and globulin hydrolysates with plant protein hydrolysates that would have a balanced amino acid composition, low allergenicity, and high functional and technological properties.
The study objects included chicken eggs (74% moisture, 11% crude protein), flax flour (7% moisture, 36% crude protein), corn flour (12% moisture, 8% crude protein), and oat flour (11% moisture, 11% crude protein). The Lowry method, Anson method, ion exchange chromatography, and the ELISA method were used to determine the amino acid composition, allergenicity, as well as functional and technological properties.
Chymopsin was selected as the most effective enzyme for the proteolysis of egg protein isolates – at 50 U/g for globulin and 25 U/g for albumin. The globulin hydrolysates had lower water-holding, emulsifying, and foaming capacities compared to the non-hydrolyzed globulin isolate. The 60-min globulin hydrolysate had the highest fat-holding capacity. The albumin hydrolysates showed lower water-holding, fat-holding, emulsifying, and foaming capacities compared to the non-hydrolyzed albumin isolate. The 90-min albumin hydrolysate was found to be not allergenic. Its mixtures with oat, corn, and flax flour protein hydrolysates were analyzed to determine the ratio that would improve the mixture's functional and technological properties. A 1:5 ratio of albumin hydrolysate and oat flour hydrolysate had higher fat-holding capacity; a 1:5 ratio of albumin hydrolysate and corn flour hydrolysate had higher water- and fat-holding capacities; and a 1:3 ratio of albumin hydrolysate and flax flour hydrolysate had higher emulsifying and fat-holding capacities.
The study proved that mixtures of egg albumin hydrolysates with plant protein hydrolysates have better functional properties, a balanced amino acid composition, and hypoallergenicity.

Keywords. Egg globulin, egg albumin, allergenicity, plant protein hydrolysates, functional and technological properties, enzymatic hydrolysis, amino acid score

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Получение ингредиентов на основе смеси яичного и растительных белков с высокими функциональными и технологическими свойствами

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Аннотация.
Продукты животного происхождения, такие как яйца, мясо и молочные продукты являются основными источниками белка для большинства людей. Однако они характеризуются более высокой стоимостью, по сравнению с растительными источниками белка, и их потребление может провоцировать тяжелые аллергические реакции. Снижение аллергенности наиболее сбалансированного по аминокислотному составу яичного белка может быть достигнуто путем его ферментативного гидролиза, а для повышения его функциональных свойств может применяться смешение с гидролизатами растительных белков. Цель работы – подобрать оптимальные условия получения смесей гидролизатов яичного альбумина и глобулина с гидролизатами растительных белков, обладающих сбалансированным аминокислотным составом, низкой аллергенностью и высокими функционально-технологическими свойствами.
Объектами исследования послужили куриные яйца, льняная, кукурузная и овсяная мука. Для определения аминокислотного состава, аллергенности и функционально-технологических свойств в работе применяли методы: Лоури, Ансона, ионообменной хроматографии, ИФА.
В ходе исследования подобраны ферментные препараты для протеолиза яичных белков: для глобулина – химопсин (50 ед/г), для альбумина – химопсин (25 ед/г). Гидролизаты глобулина обладали более низкими значениями водоудерживающей, эмульгирующей и пенообразующей способностей в отличие от негидролизованных белков. Самое высокое значение жироудерживающей способности наблюдалось у 60-минутного гидролизата глобулина. У гидролизатов альбумина значения водо- и жироудерживающей, эмульгирующей и пенообразующей способностей оказались ниже, чем у негидролизованного изолята альбумина. У 90-минутного гидролизата яичного альбумина отмечено отсутствие аллергенности.
Подобраны соотношения яичного альбумина и растительного белка в виде смеси: для увеличения жироудерживающей способности – альбуминовый гидролизат + гидролизат овсяной муки в соотношении 1:5, для увеличения водо- и жиро-удерживающей способностей – альбуминовый гидролизат + гидролизат кукурузной муки в соотношении 1:5, для увеличения эмульгирующей и жироудерживающей способностей – альбуминовый гидролизат + гидролизат льняной муки в соотношении 1:3.
Доказана целесообразность получения смесей на основе гидролизатов яичного альбумина с гидролизатами растительного белка, отличающихся улучшенными функционально-технологическими свойствами, сбалансированным аминокислотным составом и гипоаллергенностью.

Ключевые слова. Яичный глобулин, яичный альбумин, аллергенность, гидролизат растительного белка, функционально-технологические свойства, ферментативный гидролиз, аминокислотный скор

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Introduction

Currently, most people source protein from products of animal origin, particularly eggs, meat, and dairy products. However, these products are more expensive than plant sources of protein. Although plant proteins can be used as an alternative source of protein, most of them have an unbalanced amino acid composition, which limits their use. Mixing plant proteins with animal proteins can reduce the cost of a product, make its protein composition more balanced, and improve its functional properties.

Russia is one of the largest producers of eggs in the world, but not dried or liquid egg products. In 2022, the Russian poultry sector processed only about 30% of egg production. Therefore, there is a need to develop the technology for deep processing of egg components to obtain bioactive substances with valuable functional properties [1].

Chicken egg white consists of 86–87% water, in which nutrients and B vitamins are dissolved. Its main component makes up 9.7–11.5% [2].

Egg white consists mainly of 69.7% ovalbumin, 9.5% conalbumin, 6.7% ovoglobulin, 12.7% ovomucoid, 1.9% ovomucin, 3% lysozyme, and 0.05% avidin [3]. Of these proteins, ovalbumin and conalbumin (which is a flavoprotein) have the greatest biological value. Ovoglobulin is responsible for the foaminess of egg white, while ovomucin stabilizes its foam [4].

Due to its balanced amino acid composition, egg white protein can be used as a functional ingredient in a wide range of specialized foods with high biological and nutritional value [5]. However, the consumption of egg white can cause severe allergic reactions [6]. People who are allergic to chicken egg are often sensitive to several proteins [7, 8], most commonly to ovomucoid and ovotransferrin.

To prevent or alleviate allergy symptoms, people have to either give up foods containing chicken egg components or consume hypoallergenic products. The second way seems more promising since hypoallergenic products, which contain modified proteins [9], taste like natural products.

To reduce the allergenicity of egg whites, they are exposed to thermal treatment, enzymatic hydrolysis, radiation, ultrasound, and other treatments [10, 11].

Modern food industry seeks to obtain functional food ingredients, including enzymatic hydrolysates of protein isolates [12]. Their advantages over egg white protein are higher solubility in water and better digestibility and absorption in the gastrointestinal tract. In addition, proteolysis leads to the formation of a wide range of bioactive peptides [13].

The most important functional and technological properties of food ingredients are their water- and fat-holding capacities, as well as emulsifying and foaming capacities [14]. Although egg white hydrolysates do not have high functional properties, their advantages over non-hydrolyzed egg white include lower allergenicity and a balanced amino acid composition. Their functional properties can be improved by using plant protein isolates and hydrolysates, for example, obtained from oat, corn or flax flours.

Oat flour has a balanced amino acid composition, which is similar to that of muscle protein. On average, oat flour consists of 12% protein containing albumins, globulins, prolamins, and glutelins. Their content (≈ 40–45%) can vary depending on the method of extraction [15]. Oat flour protein has a high water-holding capacity.

Corn flour protein is low in tryptophan, methionine, and lysine, but it is fairly rich in leucine [16]. This plant protein contains 40.3% glutelins, 29.9% prolamins, 15.5% scleroproteins, 9.6% albumins, and 4.7 % globulins [17]. Due to its good water-holding properties [18], corn flour protein, when mixed with egg white, can increase the water-holding capacity of chicken egg protein isolate.

Flax flour contains 21–26% of protein substances, namely globulins (up to 95%), glutelins (3%), albumins (1%), and prolamins (1%) [19]. Flax flour proteins have a high emulsifying capacity [20], which can improve this indicator in mixtures with egg white hydrolysates.

However, plant proteins can cause allergies. Therefore, they are exposed to enzymatic hydrolysis in order to produce allergen-free hydrolysates [21].

In this study, we aimed to select optimal conditions for obtaining mixtures of egg albumin and globulin hydrolysates with plant protein hydrolysates that have a balanced amino acid composition, low allergenicity, and high functional and technological properties.

Study objects and methods

We used the following study objects: chicken eggs according to State Standard 31654-2012 (74% moisture, 11% crude protein); flax flour (7% moisture, 36% crude protein) (Kompas Zdorovya, Russia); corn flour (12% moisture, 8% crude protein) (Garnets, Russia); oat flour (11% moisture, 11% crude protein) (Garnets, Russia); hydrolysates of egg globulin, egg albumin, oat protein, corn protein, and flax protein; and mixtures of hydrolysates: egg albumin + oat protein (5:1, 3:1, 2:1, 1:1, 1:2, 1:3, and 1:5), egg albumin + corn protein (5:1, 3:1, 2:1, 1:1, 1:2, 1:3, and 1:5), and egg albumin + flax protein (5:1, 3:1, 2:1, 1:1, 1:2, 1:3, and 1:5).

The enzymatic protein hydrolysates were obtained from enzyme preparations with specific proteolytic activity measured by the Anson method (State Standard 20264.2-88), namely chymopsin (2000 U/g protein; Samson-Med, Russia), pancreatin (177 U/g protein; Biosintez, Russia), and beef pepsin (7500 U/g protein; Moscow Rennet Plant, Russia).

Enzymatic hydrolysis of proteins. Proteins (40 g/L) were enzymatically hydrolyzed for 2 h, with enzyme activity of 50 U/g substrate, under optimal temperature and pH conditions. The enzymatic reaction was stopped and non-hydrolyzed protein was separated by precipitation with 50% trichloroacetic acid and subsequent centrifugation at 6000 rpm for 15 min. Then, the concentration of hydrolysis products in the supernatant was measured using the modified Lowry method. The degree of protein hydrolysis was determined as a ratio between the concentration of the protein's low-molecular fraction in the hydrolysate and the initial protein concentration [22].

Selection of enzyme preparation. Three enzyme preparations were studied, namely pepsin, pancreatin, and chymopsin. Hydrolysis was carried out for 2 h at a

substrate concentration of 40 g/L, proteolytic activity of 25–75 U/g substrate, 40°C, and pH 7.6–8.2. The hydrolysates were analyzed for the content of low-molecular fraction of protein by the modified Lowry method. The degree of protein hydrolysis was determined as described above.

Egg globulin preparation. After separating the yolk, a 4-fold volume of distilled water was added to the egg white. The mixture was kept for 30 min at room temperature. The sediment was separated by centrifugation at 6000 rpm, 20°C for 15 min and dried in air. The resulting fraction of egg globulin contained 83% of the main component.

Egg albumin preparation. The supernatant obtained after isolating the egg globulin was used to isolate the albumin fraction. For this, a 3-fold volume of 96% ethyl alcohol was added to the supernatant, and the albumin fraction was precipitated at room temperature for 1 h. The sediment was separated by centrifugation at 6500 rpm, 20°C for 10 min and dried in air. The resulting fraction of egg albumin contained 88% of the main component.

Oat flour protein hydrolysis. Prior to protein isolation, oat flour was separated from starch by washing three times with water (3:20) for 20 min at room temperature and centrifuging at 5200 rpm for 15 min. Then, the flour was diluted 10 times with distilled water at pH adjusted to 7.0 and thermostatted for 1.5 h at 50°C. The solid phase was separated by centrifugation at 5500 rpm for 15 min. Protein was precipitated from the extract at the isoelectric point at pH 3. The protein precipitate, which was mainly a globulin fraction containing 82% of the main component, was separated by filtration and dried in air at room temperature. To obtain a protein hydrolysate, a 1% suspension of the protein was hydrolyzed with pancreatin (5 U/mL) for 60 min at 40°C with constant stirring. The hydrolysate was dried in air at room temperature.

Corn flour protein hydrolysis. Prior to protein extraction, starch was removed from corn flour as described above for oat flour. Protein was extracted with a 0.1 N sodium hydroxide solution from a 10% suspension with constant stirring at room temperature for 30 min. The supernatant was separated by centrifugation at 6000 rpm for 15 min. Protein was precipitated from the supernatant at pH 6.0, after which the precipitate was separated by centrifugation at 6000 rpm for 15 min and dried in air at room temperature. A 2% solution was prepared from the protein precipitate, which was mainly a globulin fraction containing 85% of the main component. The pH was adjusted to 8.2, and the enzyme preparation trypsin was added until an activity of 0.35 U/mL was achieved. Hydrolysis was carried out at 40 °C for 60 min with constant stirring. The hydrolysate was dried in air at room temperature.

Flax flour protein hydrolysis. Protein extract was obtained from flax flour with a 0.5 M sodium bicarbonate solution (flour to solution ratio of 1:8) at 25°C for 30 min with constant stirring. The resulting suspension was separated by centrifugation at 6500 rpm for 15 min. Then, 2 volumes of ethyl alcohol were added to precipitate protein from the flax extract at a pH of 4.5–5.0 at a reduced temperature (2–8°C) for 4 h. The precipitate was separated by centrifugation at 2000 rpm for 15 min and dried at room temperature. To obtain a hydrolysate, a 4% suspension of the flax protein globulin fraction containing 84% of the main component was hydrolyzed with pancreatin (5 U/mL) at pH 7.6, 40°C for 30 min with constant stirring. The resulting hydrolysate was dried in air at room temperature.

The effect of enzymatic hydrolysis time on the functional properties of hydrolysates. Hydrolysates based on egg globulin and albumin, as well as oat, corn, and flax globulins, were prepared at different enzymolysis times. For this, 1 g of a hydrolysate was mixed with water (1:25) and an enzyme preparation was added until its proteolytic activity in the solution reached 40 U/g protein. Hydrolysis was carried out at the optimum temperature and pH for 15, 30, 60, 90, and 120 min. Heating protein hydrolysates may produce an unpleasant odor due to side chemical reactions such as the Maillard and Strecker reactions [23]. Therefore, the enzymes were inactivated by cooling to –10°C. The hydrolysates were dried at 50°C prior to the analysis of their functional properties.

Protein content. The content of protein in the hydrolysates was quantified by a modified Lowry method (high-molecular and low-molecular fractions determined separately), with a preliminary precipitation of the high-molecular fraction with 50% trichloroacetic acid.

Total proteolytic activity. The total proteolytic activity was measured by a modified Anson method. A unit of proteolytic activity was defined as an enzyme's ability to convert sodium caseinate into a form unprecipitable with trichloroacetic acid in an amount corresponding to 1 μmol of tyrosine in one min at 30°C.

Allergenicity of egg albumin and globulin hydrolysates. The allergenicity of egg albumin and globulin hydrolysates was determined by an enzyme immunoassay according to the ALINORM 08/31/26 Standard for food products (Methodological Guidelines 4.1.2880-11 4.1). The assay took into account the specific interaction between the allergenic protein in the sample and the antibodies to it contained in the test solution (Siemens, Germany). For the assay, we placed 100 μL of a 1% solution of the test sample into a well of the plate and added 100 μL of the test conjugate solution containing antibodies to immunoglobulin E. This caused an "antibody-antigen" complex to form and precipitate. The resulting analytical signal, which depended on the interaction between the antibody-antigen complex and the conjugate on the surface of the plate wells, was measured in terms of optical density at 450 nm. Then, this value was recalculated for egg white (an optical density value of one corresponding to 50 μg/L (1 kU/L) of immunoglobulin E

in the sample. Allergenicity was considered low if the immunoglobulin E content was at least 35 μg/L and moderate if it was under 150 μg/L.

Amino acid composition. The amino acid composition of the hydrolysate samples and their mixtures was determined by ion exchange chromatography on an ARACUS amino acid analyzer (membraPure GmbH, Bodenheim, Germany) equipped with a C18 column and a refractive index detector. High-performance liquid chromatography was supplemented with mass spectrometry, with electrospray ionization for amino acid separation followed by a ninhydrin reaction and photometric detection [24].

Fat-holding capacity. Glass centrifuge tubes were filled with 0.5 g of the test sample and then 0.125 to 0.625 mL of vegetable oil was added at 0.125 mL intervals. The contents of the tubes were mixed for 10 min, after which the samples were stirred for 15 min, cooled to room temperature, and centrifuged at 1500 rpm for 15 min. The fat-holding capacity was determined as the maximum amount of added oil which did not cause the oil phase to separate during the test (expressed in terms of 1 g of preparation) [25].

Water-holding capacity. Glass centrifuge tubes were filled with 0.5 g of the test sample and 1.5 to 2.5 mL of water was added at 0.25 mL intervals. The test continued as described above for the fat-holding capacity. The water-holding capacity was determined as the maximum amount of added water which did not cause the aqueous phase to separate during the test (expressed in terms of 1 g of preparation) [24].

Emulsifying capacity. Glass centrifuge tubes were filled with 1 g of the test sample and then mixed with 5 mL of water and 5 mL of oil. The contents of the tubes were stirred for 10 min, followed by the procedure described above. The emulsifying capacity was determined as a percentage ratio between the aqueous and oil phases separated from the emulsion [26].

Foaming capacity. Conical flasks (50 mL) were filled with 0.25 g of the test sample and 25 mL of water was added. The resulting solution was shaken with a shaker for 30 s. It was then poured into a measuring cylinder to measure the height of the foam column [27].

Results and discussion

Allergenicity is a significant disadvantage of egg protein isolates, which limits their use in the food industry. This problem can be solved by modifying the protein by hydrolysis. The resulting hydrolysates not only have lower allergenicity, but also better functional and technological properties.

Enzymatic hydrolysis is the best method of protein hydrolysis since it occurs under milder conditions compared to acid or alkaline hydrolysis. However, its use is constrained by the high cost of enzyme preparations.

The type of enzyme and hydrolysis time affect the functional and technological properties of protein hydrolysates, as well as their allergenicity. First, to determine the most effective enzyme, we hydrolyzed isolates of egg white globulin and albumin fractions with pancreatin (177 u/g), pepsin (1070 u/g), and chymopsin (2000 u/g) (Table 1).

As can be seen, chymopsin with an activity of 2.0 U/mL for globulin and 1.0 U/mL for albumin ensured the highest degree of hydrolysis and enzyme efficiency. A further increase in its activity did not lead to a higher degree of hydrolysis.

Next, we determined the hydrolysis time that would decrease the allergenicity of protein isolates and improve their functional properties. For this, egg albumin and globulin isolates were hydrolyzed with chymopsin at a selected dosage for 10, 20, 30, 60, 90, and 120 min. The resulting hydrolysates were dried in air at room temperature, after which we determined their water-holding, fat-holding, emulsifying, and foaming capacities (Fig. 1).

We found that the hydrolysates obtained at different enzymolysis times had significantly different functional and technological properties compared to the original proteins.

The water-holding capacity of all egg globulin hydrolysates was significantly lower than that of the original isolate (max. 4.24 g protein/g water). The fat-holding capacity of egg globulin was high before hydrolysis and increased slightly from 0.52 to 0.60 g protein/g oil after 60 min of hydrolysis. The emulsifying and foaming capacity values were the highest in the original globulin isolate.

As for egg albumin, the highest water-holding and fat-holding values were observed in the non-hydrolyzed isolate (4.55 g protein/g water and 0.88 g protein/g oil, respectively). The emulsifying and foaming values were also higher in the non-hydrolyzed isolate.

The hydrolysates with lower functional and technological properties compared to non-hydrolyzed proteins should not be used to prepare functional food products.

Table 1. Determination of enzyme type and dosage for effective hydrolysis of egg albumin and globulin

Таблица 1. Определение типа ферментного препарата и его дозировки для эффективного гидролиза яичного альбумина и яичного глобулина

Enzyme	Enzyme activity, U/mL	Hydrolysis degree, %	
		Globulin	Albumin
Pancreatin	1.0	31 ± 2	31 ± 2
	2.0	35 ± 2	29 ± 2
	3.0	36 ± 2	30 ± 2
Pepsin	1.0	30 ± 2	27 ± 2
	2.0	49 ± 3	57 ± 3
	3.0	50 ± 3	58 ± 3
Chymopsin	1.0	68 ± 3	74 ± 4
	2.0	71 ± 4	70 ± 4
	3.0	70 ± 4	71 ± 4

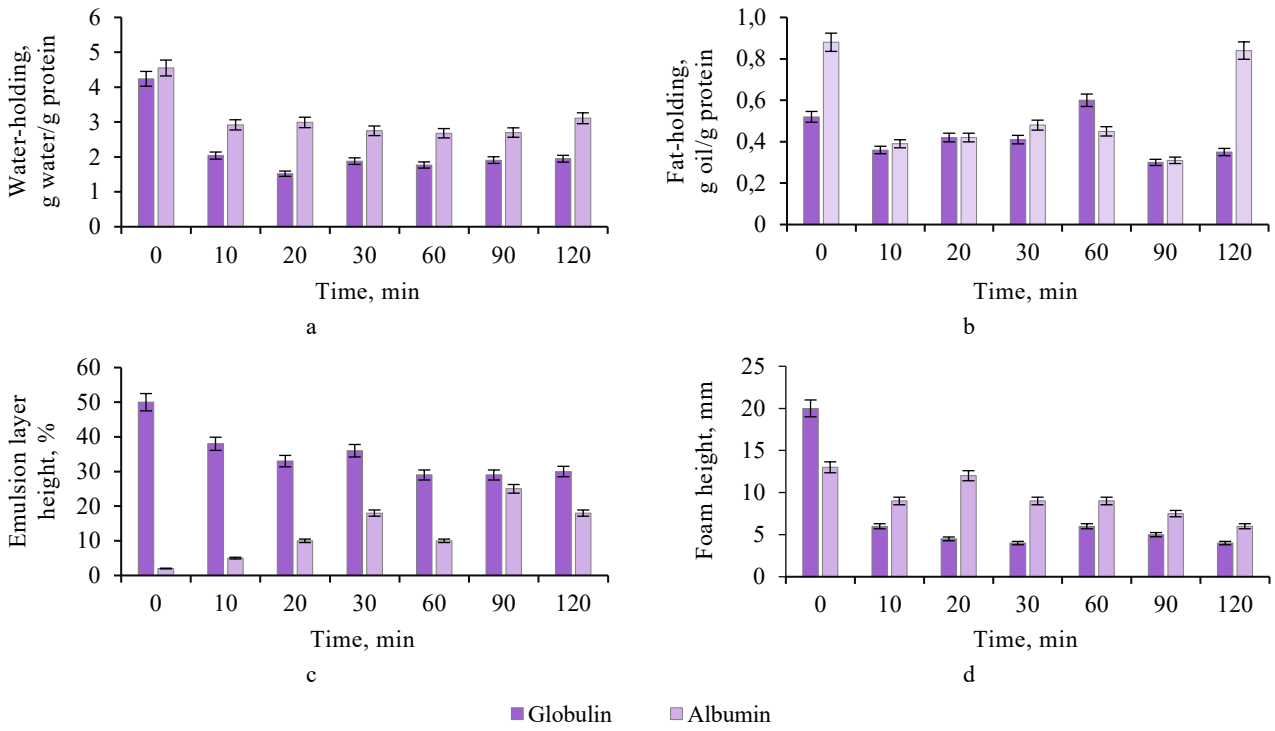

Figure 1. Effect of enzymatic hydrolysis time on the functional properties of egg albumin and globulin hydrolysates: a – water-holding capacity, b – fat-holding capacity, c – emulsifying capacity, and d – foaming capacity

Рисунок 1. Влияние времени ферментативного гидролиза яичного альбумина и яичного глобулина на функциональные свойства гидролизатов: a – водоудерживающая способность, b – жироудерживающая способность, c – эмульгирующая способность, d – пенообразующая способность

Table 2. Allergenicity of egg globulin and albumin and their hydrolysates

Таблица 2. Исследование аллергенности яичного глобулина и яичного альбумина и их гидролизатов

Study object	Allergenicity, kU/L	Allergen-specific IgE antibody level
Egg globulin	0.55 ± 0.03	Low
Egg albumin	15.30 ± 0.80	High
Egg albumin hydrolysate, 10 min	4.50 ± 0.20	High
Egg albumin hydrolysate, 20 min	2.40 ± 0.10	Medium
Egg albumin hydrolysate, 30 min	2.80 ± 0.10	Medium
Egg albumin hydrolysate, 60 min	2.40 ± 0.10	Medium
Egg albumin hydrolysate, 90 min	0.35 ± 0.02	Not detected
Egg albumin hydrolysate, 120 min	0.32 ± 0.02	Not detected

According to literature, egg white is highly allergenic. Therefore, we tested egg albumin and globulin isolates and their hydrolysates for allergenicity using the enzyme immunoassay method described above (Table 2).

As can be seen in Table 2, allergenicity was low in the globulin isolate and high in the albumin isolate. However, the longer egg albumin was hydrolyzed, the lower was its allergenicity. Since no allergenicity was detected after a 90 min enzymatic hydrolysis, this egg albumin hydrolysate can be used to produce hypoallergenic food products. In contrast, egg globulin hydrolysates cannot be recommended for functional nutrition as their functional properties are much lower compared to those of egg globulin isolates. Therefore, only egg albumin hydrolysates were exposed to further analyses.

Although the 90-min egg albumin hydrolysate was not allergenic, its functional properties needed improving. For this, we mixed it with oat, corn, and flax flour globulins and their hydrolysates in different ratios.

Previous studies [28, 29] selected optimal conditions for obtaining hydrolysates of oat, corn, and flax flour globulins with high water-holding and emulsifying capacities.

Figure 2 shows the values of water-holding, fat-holding, emulsifying, and foaming capacities for mixtures of the 90-min enzymatic lysate of egg albumin with oat,

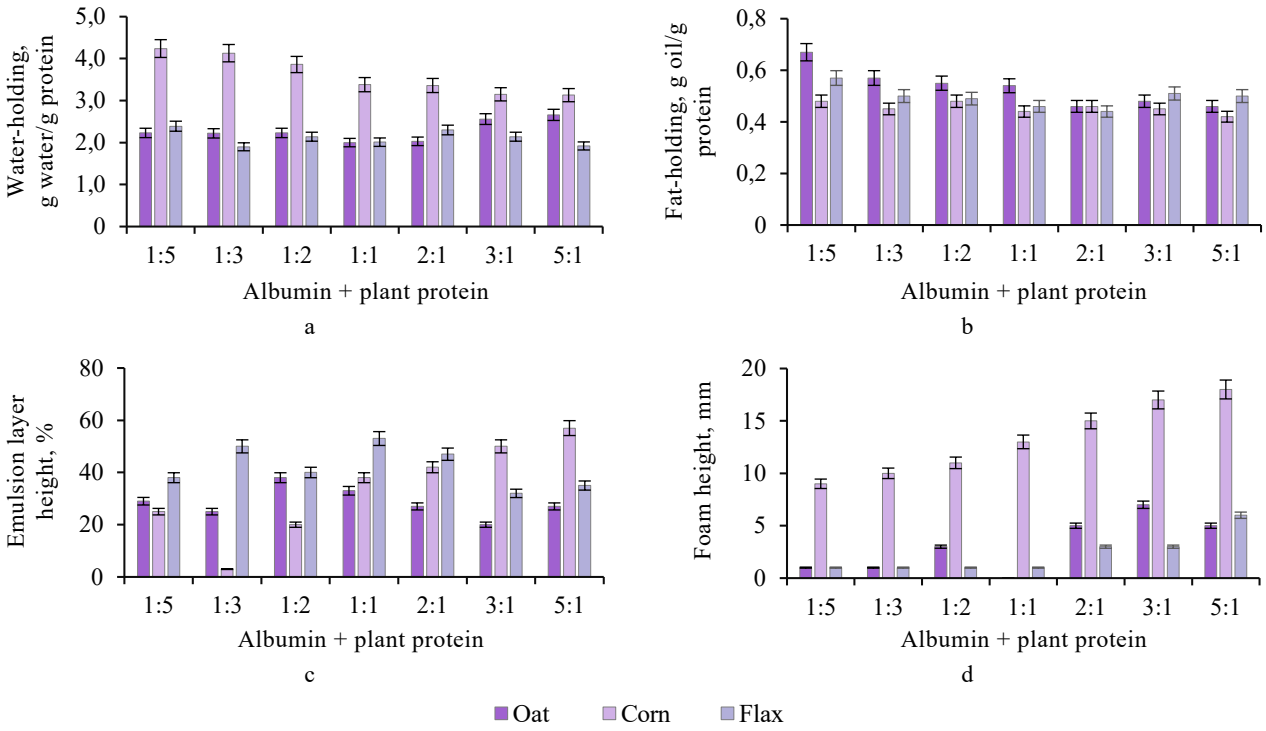

Figure 2. Effect of egg albumin hydrolysate + plant protein hydrolysate ratios on the functional and technological properties of mixed hydrolysates: a – water-holding capacity, b – fat-holding capacity, c – emulsifying capacity, and d – foaming capacity

Рисунок 2. Влияние массового соотношения гидролизат яичного альбумина: гидролизат растительного белка на функционально-технологические свойства смешанных гидролизатов: a – водоудерживающая способность, b – жироудерживающая способность, c – эмульгирующая способность, d – пенообразующая способность

corn, and flax flour hydrolysates with the best functional and technological properties.

The highest water-holding capacity was found in the mixtures of albumin and corn flour globulin hydrolysates, especially in a 1:5 ratio (4.24 g water/g protein). The mixtures of albumin hydrolysates with oat and flax flour globulin hydrolysates had significantly lower water-holding capacity (2.66 g water/g protein in a 5:1 ratio and 2.39 g water/g protein in a 1:5 ratio, respectively). Therefore, a 1:5 ratio of egg albumin hydrolysate and corn protein hydrolysate can be recommended for use to increase the water-holding capacity of food products.

The fat-holding capacity values were found to be in the same range for all the mixtures. The highest values were observed in the mixtures of albumin + oat protein (1:5; 0.67 g oil/g protein), albumin + corn protein (1:2 and 1:5; 0.48 g oil/g protein), and albumin + flax protein (1:5; 0.57 g oil/g protein). Thus, a 1:5 ratio of egg albumin hydrolysate and oat flour protein hydrolysate can be recommended to increase the fat-holding capacity of food products.

The highest emulsifying capacity values were found in a 1:2 ratio of egg albumin and oat globulin (38%), a 5:1 ratio of egg albumin and corn globulin (57%), and a 1:1 ratio of albumin and flax protein (53%). Therefore, the last two mixtures can be used to increase the emulsifying capacity of food products.

The foaming capacity values were lower in all the mixtures of albumin with plant proteins. The greatest foam height was observed in a 3:1 ratio of egg albumin and oat globulin, a 5:1 ratio of egg albumin and corn globulin, and a 5:1 ratio of egg albumin and flax globulin. Since the maximum foaming capacity was demonstrated by the mixture with flax protein, it can be used to increase the foaming capacity of food products.

Thus, our results showed that the water- and fat-holding capacities can be significantly increased by adding corn globulin to the egg albumin hydrolysate in a ratio of 1:5. However, the emulsifying capacity of this mixture was similar to that of the egg albumin hydrolysate, while its foaming capacity decreased about 3 times. Adding oat globulin to the egg albumin hydrolysate in a ratio of 1:5 more than doubled the fat-holding capacity, with the water-holding and emulsifying capacities remaining unchanged and the foaming capacity decreasing to its minimum. The emulsifying capacity can be improved by adding flax globulin to the egg albumin hydrolysate in a ratio of 1:3. The mixture's fat-holding capacity was also higher, while the water-holding and foaming capacities were lower compared to those of the egg white hydrolysate.

One of the significant disadvantages of plant proteins is their low content of some amino acids. In particular, oat protein is low in glycine, cysteine, tryptophan, and arginine. Corn protein is low in glycine, arginine, tryptophan, methionine, cysteine, and histidine. Flax protein has a reduced content of cysteine, methionine, proline, and histidine. At the same time, egg proteins have a balanced amino acid composition. Therefore, we determined the amino acid composition of the obtained mixtures of egg albumin with plant globulins in order to select a sample with a balanced amino acid composition in addition to high functional properties (Table 3).

Table 4 shows amino acid scores for the selected mixtures. As can be seen, the mixtures of egg albumin hydrolysate and plant globulin hydrolysates had higher scores for almost all essential amino acids.

The mixture of egg albumin hydrolysate with oat globulin hydrolysate had scores below 100 for lysine (87), threonine (93), valine (94), as well as methionine and cystine (71). The mixture of egg albumin hydrolysate with corn globulin hydrolysate had the following limiting amino acids: lysine (62), isoleucine (80), threonine (90), methionine and cystine (71), and valine (79). In the mixture of egg albumin hydrolysate with flax globulin hydrolysate, the limiting amino acids were valine (86) and methionine + cystine (51).

Of the three mixtures under study, a 1:3 ratio of egg albumin hydrolysate and flax globulin hydrolysate

Table 3. Amino acid composition of hydrolysates of egg albumin; oat, corn, and flax flour globulins; and their mixtures

Таблица 3. Аминокислотный состав гидролизатов яичного альбумина, овсяного, кукурузного, льняного глобулинов муки и их смесей

Amino acid, %	Hydrolysate of						
	egg albumin, 90 min	oat globulin, 60 min	corn globulin, 30 min	flax globulin, 30 min	egg albumin, 90 min +		
					egg albumin + oat globulin, 1:5	corn globulin, 1:5	flax globulin, 1:3
Gly	3.20 ± 0.20	0.63 ± 0.03	0.41 ± 0.02	14.00 ± 0.70	1.00 ± 0.10	0.93 ± 0.05	11.00 ± 1.00
Ala	7.20 ± 0.40	6.80 ± 0.30	6.40 ± 0.30	6.30 ± 0.30	6.90 ± 0.30	6.50 ± 0.30	6.50 ± 0.30
Val	1.70 ± 0.10	5.30 ± 0.30	4.40 ± 0.20	5.20 ± 0.30	4.70 ± 0.20	4.00 ± 0.20	4.30 ± 0.20
Ile	5.50 ± 0.30	3.90 ± 0.20	2.70 ± 0.10	4.70 ± 0.20	4.20 ± 0.20	3.20 ± 0.20	4.90 ± 0.20
Leu	9.90 ± 0.50	7.40 ± 0.40	12.00 ±1.00	7.00 ± 0.40	12.00 ± 1.00	12.00 ± 1.00	7.70 ± 0.40
Pro	4.50 ± 0.20	4.30 ± 0.20	5.90 ± 0.30	1.90 ± 0.10	4.30 ± 0.20	5.70 ± 0.30	2.60 ± 0.10
Ser	7.40 ± 0.40	6.00 ± 0.30	4.40 ± 0.20	7.50 ± 0.40	6.20 ± 0.30	7.50 ± 0.40	7.50 ± 0.40
Thr	5.80 ± 0.30	3.30 ± 0.20	3.20 ± 0.20	3.60 ± 0.20	3.70 ± 0.20	3.60 ± 0.20	4.20 ± 0.20
Cys	1.00 ± 0.10	1.10 ± 0.10	1.80 ± 0.10	0.90 ± 0.10	1.10 ± 0.10	1.70 ± 0.10	0.92 ± 0.10
Met	3.50 ± 0.20	2.60 ± 0.10	1.90 ± 0.10	1.40 ± 0.10	2.80 ± 0.10	2.20 ± 0.10	1.90 ± 0.10
Asp	6.30 ± 0.30	9.10 ± 0.50	9.20 ± 0.50	3.80 ± 0.20	8.60 ± 0.40	8.70 ± 0.40	4.40 ± 0.20
Glu	17.00 ± 0.90	17.00 ± 0.90	18.00 ± 1.00	9.50 ± 0.50	17.00 ± 1.00	18.00 ± 1.00	11.00 ± 1.00
Lys	7.90 ± 0.40	4.20 ± 0.20	2.50 ± 0.10	5.80 ± 0.30	4.80 ± 0.20	3.40 ± 0.20	6.30 ± 0.30
Arg	5.70 ± 0.30	0.64 ± 0.03	0.44 ± 0.02	4.20 ± 0.20	1.50 ± 0.10	1.30 ± 0.10	4.60 ± 0.20
His	2.40 ± 0.10	0.32 ± 0.02	0.36 ± 0.02	1.00 ± 0.10	0.73 ± 0.04	1.20 ± 0.10	1.40 ± 0.10
Phe	6.30 ± 0.30	2.80 ± 0.10	4.90 ± 0.20	5.10 ± 0.30	3.40 ± 0.20	5.10 ± 0.30	5.40 ± 0.30
Trp	5.70 ± 0.30	1.30 ± 0.07	0.61 ± 0.03	3.50 ± 0.20	2.00 ± 0.10	3.90 ± 0.20	4.10 ± 0.20
Tyr	2.80 ± 0.10	6.40 ± 0.30	3.80 ± 0.20	3.50 ± 0.20	5.80 ± 0.30	3.40 ± 0.20	3.30 ± 0.20

Table 4. Amino acid scores, %, for mixtures of egg albumin hydrolysates with plant globulin hydrolysates

Таблица 4. Аминокислотный скор, %, смесей гидролизатов яичного альбумина с гидролизатами растительных глобулинов

Amino acid	Hydrolysate of						
	egg albumin, 90 min	oat globulin, 60 min	corn globulin, 30 min	flax globulin, 30 min	albumin, 90 min +		
					oat globulin, 1:5	corn globulin, 1:5	flax globulin, 1:3
Lys	144 ± 7	76 ± 4	45 ± 2	105 ± 5	87 ± 4	62 ± 3	115 ± 6
Met+Cys	129 ± 6	67 ± 3	67 ± 3	42 ± 2	71 ± 4	71 ± 4	51 ± 3
Ile	138 ± 7	99 ± 5	68 ± 3	118 ± 6	105 ± 5	80 ± 4	123 ± 6
Leu	141 ± 7	106 ± 5	170 ± 9	100 ± 5	174 ± 9	166 ± 8	110 ± 6
Thr	145 ± 7	83 ± 4	80 ± 4	90 ± 5	93 ± 5	90 ± 5	105 ± 5
Phe+Tyr	152 ± 8	153 ± 8	145 ± 7	156 ± 8	153 ±8	142 ± 7	145 ± 7
Trp	570 ± 29	130 ± 7	60 ± 3	350 ± 18	200 ± 10	390 ± 20	410 ± 21
Val	34 ± 2	106 ± 5	88 ± 4	104 ± 5	94 ± 5	79 ± 4	86 ± 4

had the best amino acid composition, with the highest scores and fewer limiting amino acids.

Conclusion

Chymopsin was selected as the most effective enzyme for the proteolysis of egg protein isolates – at 50 U/g for globulin and 25 U/g for albumin.

Egg globulin and albumin isolates were hydrolyzed at different times with the selected enzymes to study the hydrolysates' functional and technological properties. All globulin hydrolysates had lower water-holding, emulsifying, and foaming capacities compared to the non-hydrolyzed globulin isolate. The 60-min globulin hydrolysate had the highest fat-holding capacity. All albumin hydrolysates showed lower water-holding, fat-holding, emulsifying, and foaming capacities compared to the non-hydrolyzed albumin isolate. The hydrolysis reduced the allergenicity of egg albumin and its 90-min hydrolysate was found to be not allergenic.

The non-allergenic 90-min egg albumin hydrolysate was mixed with oat, corn, and flax flour protein hydrolysates to determine the ratio that would improve the mixture's functional and technological properties. According to the results, a 1:5 ratio of albumin hydrolysate and oat flour hydrolysate had higher fat-holding capacity; a 1:5 ratio of albumin hydrolysate and corn flour hydrolysate had higher water- and fat-holding capacities; and a 1:3 ratio of albumin hydrolysate and flax flour hydrolysate had higher emulsifying and fat-holding capacities.

The scores for most essential amino acids in the selected mixtures exceeded 100%.

Contribution

D.V. Prikhodko conducted the experiments, processed the data, and wrote the manuscript. P.A. Karpushina conducted the experiments, processed the data, and formulated intermediate conclusions. A.A. Krasnoshtanova edited and corrected the final version of the manuscript and formulated the conclusions.

Conflict of interest

The authors declare no conflict of interest.

Критерии авторства

Приходько Д. В. – проведение экспериментов, обработка данных, написание рукописи. Карпухина П. А. – проведение экспериментов, обработка данных, формулировка и составление промежуточных выводов. Красноштанова А. А. – редактирование и корректировка рукописи, формулировка выводов.

Конфликт интересов

Авторы заявляют об отсутствии конфликта интересов.

References / Список литературы

1. Krasnoshtanova AA, Yudina AN. Egg components like a basis for development of scientific foundation of technology for products of lipid and protein origin with high level value. Butlerov Communication. 2022;70(6):109–123. (In Russ.) [Красноштанова А. А., Юдина А. Н. Роль яичных компонентов в разработке научных основ технологии получения продуктов липидной и белковой природы с высокой добавленной стоимостью. Бутлеровские сообщения. 2022. Т. 70. № 6. С. 109–123.] https://elibrary.ru/QHQIPB

2. Bizanov G. IgY extraction and purification from chicken egg yolk. Journal of the Hellenic Veterinary Medical Society. 2018;68(3):265–272. https://doi.org/10.12681/jhvms.15466

3. Li Z, Huang X, Tang Q, Ma M, Jin Y, *et al.* Functional properties and extraction techniques of chicken egg white proteins. Foods. 2022;11(16):2434. https://doi.org/10.3390/foods11162434

4. Abeyrathne E, Huang X, Ahn DU. Antioxidant, angiotensin-converting enzyme inhibitory activity and other functional properties of egg white proteins and their derived peptides – A review. Poultry Science. 2018;97(4):1462–1468. https://doi.org/10.3382/ps/pex399

5. Sidorova YuS, Mazo VK, Zorin SN, Stefanova IL. The evaluation of biological value and immunochemical characteristics of the coagulated chicken egg white. Problems of Nutrition. 2018;87(1):44–50. (In Russ.) [Сидорова Ю. С., Мазо В. К., Зорин С. Н., Стефанова И. Л. Оценка биологической ценности и антигенности коагулированного белка куриного яйца. Вопросы питания. 2018. Т. 87. № 1. С. 44–50.] https://doi.org/10.24411/0042-8833-2018-10005

6. de Pilli T. Development of a vegetable oil and egg proteins edible film to replace preservatives and primary packaging of sweet baked goods. Food Control. 2020;114:107273. https://doi.org/10.1016/j.foodcont.2020.107273

7. Shukla P, Chopada K, Sakure A, Hati S. Current trends and applications of food-derived antihypertensive peptides for the management of cardiovascular disease. Protein & Peptide Letters. 2022;29(5):408–428. https://dx.doi.org/10.2174/0929866529666220106100225

8. Réhault-Godbert S, Guyot N, Nys Y. The golden egg: Nutritional value, bioactivities, and emerging benefits for human health. Nutrients. 2019;11(3):684. https://doi.org/10.3390/nu11030684

9. Gromov DA, Borisova AV, Bakharev VV. Food allergens and methods for producing hypoallergenic foods. Food Processing: Techniques and Technology. 2021;51(2):232–247. (In Russ.) [Громов Д. А., Борисова А. В., Бахарев В. В. Пищевые аллергены и способы получения гипоаллергенных пищевых продуктов. Техника и технология пищевых производств. 2021. Т. 51. № 2. С. 232–247.] https://doi.org/10.21603/2074-9414-2021-2-232-247

10. Lesmes U. Quantifying digestion products: Physicochemical aspects. In: Gouseti O, Bornhorst G, Bakalis S, Mackie A, editors. Interdisciplinary Approaches to Food Digestion. Cham: Springer; 2019. pp. 231–253 https://doi.org/10.1007/978-3-030-03901-1_11

11. Mahler V, Goodman RE. Definition and design of hypoallergenic foods. In: Kleine-Tebbe J, Jakob T, editors. Molecular allergy diagnostics: Innovation for a better patient management. Cham: Springer; 2017. pp. 487–511. https://doi.org/10.1007/978-3-319-42499-6_27

12. Monaci L, Pilolli R, de Angelis E, Crespo JF, Novak N, *et al.* Chapter three – Food allergens: Classification, molecular properties, characterization, and detection in food sources. Advances in Food and Nutrition Research. 2020;93:113–146. https://doi.org/10.1016/bs.afnr.2020.03.001

13. Milentyeva IS, Davydenko NI, Rasshchepkin AN. Casein proteolysis in bioactive peptide production: Optimal operating parameters. Food Processing: Techniques and Technology. 2020;50(4):726–735. (In Russ.) [Милентьева И. С., Давыденко Н. И., Расщепкин А. Н. Подбор рабочих параметров для проведения направленного протеолиза казеина с целью получения биопептидов. Техника и технология пищевых производств. 2020. Т. 50. № 4. С 726–735.] https://doi.org/10.21603/2074-9414-2020-4-726-735

14. Agarkova EYu, Kruchinin AG, Ryazantseva KA, Sherstneva NE. Functional property enhancement of milk-based proteins using enzymatic hydrolysis. Milk Processing. 2020;(2):16–19. (In Russ.) [Агаркова Е. Ю., Кручинин А. Г., Рязанцева К. А., Шерстнева Н. Е. Повышение функциональных свойств белков молочной сыворотки путем ферментативного гидролиза. Переработка молока. 2020. № 2. С. 16–19.] https://doi.org/10.33465/2222-5455-2020-02-16-18

15. Feofilaktova OV, Ponomarev AS. Technological property research of the alternative flour types while food producing in catering in catering. Food industry. 2019;4(2):28–34. (In Russ.) [Феофилактова О. В., Пономарев А. С. Исследование технологических свойств нетрадиционных видов муки при производстве продукции предприятий общественного питания. Индустрия питания. 2019. Т. 4. № 2. С. 28–34.] https://elibrary.ru/VXREVG

16. Ananskikh VV, Shleina LD. About a possibility of receiving maltodextrins from cornmeal. Storage and Processing of Farm Products. 2017;(11):9–13. [Анапских В. В., Шлеина Л. Д. О возможности получения мальтодекстринов из кукурузной муки. Хранение и переработка сельхозсырья. 2017. № 11. С. 9–13.] https://elibrary.ru/YMTKQA

17. Renzyaeva TV, Tuboltseva AS, Renzyaev AO. Various flours in pastry production technology. Food Processing: Techniques and Technology. 2022;52(2):407–416. (In Russ.) [Рензяева Т. В., Тубольцева А. С., Рензяев А. О. Мука различных видов в технологии мучных кондитерских изделий. Техника и технология пищевых производств. 2022. Т. 52. № 2. С. 407–416.] https://doi.org/10.21603/2074-9414-2022-2-2373

18. Mazhulina IV, Tertychnaya TN, Andrianov EA. Formulating a functional cake with hawthorn and flax products. Khleboprodukty. 2018;(5):45–47. (In Russ.) [Мажулина И. В., Тертычная Т. Н., Андрианов Е. А. Разработка рецептуры кекса функционального назначения с продуктами переработки боярышника и льна. Хлебопродукты. 2018. № 5. С. 45–47.] https://elibrary.ru/LBAWMX

19. Varivoda AA. Utilization of oil and fat industry by-products as functional ingredients for the bakery industry. Polzunovskiy vestnik. 2019;(3):3–7. (In Russ.) [Варивода А. А. Использование побочной продукции масложирового производства в качестве функциональных ингредиентов для хлебопекарной отрасли. Ползуновский вестник. 2019. № 3. С. 3–7.] https://elibrary.ru/PMZGPC

20. Merenkova SP, Semizdralova VV, Paymulina AV. Analysis of the influence of linseed meal on structural-and-mechanical properties of meat products. Bulletin of South Ural State University, Series: Food and Biotechnology. 2018;6(4): 42–51. (In Russ.) [Меренкова С. П., Семиздралова В. В., Паймулина А. В. Анализ влияния льняной муки на структурно-механические свойства мясных продуктов. Вестник Южно-Уральского государственного университета. Серия: Пищевые и биотехнологии. 2018. Т. 6. № 4. 42–51.] https://doi.org/10.14529/food180406

21. Krasnoshtanova AA, Shul'ts LV. Preparation and evaluation of the functional properties of protein isolates and hydrolysates from plant raw materials. Chemistry of plant raw material. 2022;(4):299–309. (In Russ.) [Красноштанова А. А., Шульц Л. В. Получение и оценка функциональных свойств белковых изолятов и гидролизатов из растительного сырья. Химия растительного сырья. 2022. № 4. С. 299–309.] https://doi.org/10.14258/jcprm.20220410952

22. Prikhodko DV, Krasnoshtanova AA. Study of the functional properties of wheat gluten fractions. Scientific achievements of the third millennium. SPC LJournal. New York, 2021:161–165. (In Russ.) [Приходько Д. В., Красноштанова А. А. Исследование функциональных свойств фракций пшеничного глютена. Scientific achievements of the third millennium. SPC LJournal. New York, 2021. С. 161–165.] https://doi.org/10.18411/scienceconf-03-2021-30

23. Linhart B, Freidl R, Elisyutina O, Karaulov A, *et al.* Molecular approaches for diagnosis, therapy and prevention of cow's milk allergy. Nutrients. 2019;11(7):1492. https://doi.org/10.3390/nu11071492

24. Babich OO, Razumnikova IS, Poletaev AU, Morozova AI. Keratin containing waste processing and manufacture of albuminous hydrolysates for food and fodder purposes. Food Processing: Techniques and Technology. 2011;(2):7–11. (In Russ.) [Бабич О. О., Разумникова И. С., Полетаев А. Ю., Морозова А. И. Переработка вторичного кератинсодержащего сырья и получение белковых гидролизатов на пищевые и кормовые цели. Техника и технология пищевых производств. 2011. № 2. С. 7–11.] https://elibrary.ru/NYGVEB

25. Kumar D, Chatli MK, Singh R, Mehta N, Kumar P. Antioxidant and antimicrobial activity of camel milk casein hydrolysates and its fractions. Small Ruminant Research. 2016;139:20–25. https://doi.org/10.1016/j.smallrumres.2016.05.002

26. Berdutina AV, Gromov AS. Methodology for determining the emulsion properties of protein preparations. Food Industry. 2009;(9):35–37. (In Russ.) [Бердутина А. В., Громов А. С. Методика определения эмульсионных свойств белковых препаратов. Пищевая промышленность. 2009. № 9. 35–37.] https://elibrary.ru/KWXCQV

27. Popov VN, Plotnikova IV, Magomedov GO, Magomedov MG, Polyakova LE, *et al.* Comprehensive evaluation of foam-forming properties of a serum of serum proteins for producing special purpose products. Food Industry. 2020;(8):42–47. (In Russ.) [Попов В. Н., Плотникова И. В., Магомедов Г. О., Магомедов М. Г., Полякова Л. Е. и др. Комплексная оценка пенообразующих свойств концентрата сывороточных белков для получения продукции специального назначения. Пищевая промышленность. 2020. № 8. С. 42–47.] https://elibrary.ru/LBZLPV

28. Bogdanova LS, Prikhodko DV, Krasnoshtanova AA. Obtaining protein fractions from corn flour and protein-polysaccharide complexes based on them. Butlerov Communication. 2023;73(3):95–103. (In Russ.) [Богданова Л. С., Приходько Д. В., Красноштанова А. А. Получение белковых фракций из кукурузной муки и белково-полисахаридных гелей на их основе. Бутлеровские сообщения. 2023. Т. 73. № 3. С. 95–103.] https://elibrary.ru/WEAAJS

29. Zarubin NYu, Frolova YV, Bredikhina OV. Development of a multifunctional complex based on the raw materials of animal and vegetable origin for use in semifinished fish technology. Proceedings of Universities. Applied Chemistry and Biotechnology. 2017;7(1):119–126. (In Russ.) [Зарубин Н. Ю., Фролова Ю. В., Бредихина О. В. Разработка многофункционального комплекса на основе сырья животного и растительного происхождения для использования в технологии рыбных полуфабрикатов. Известия вузов. Прикладная химия и биотехнология. 2017. Т. 7. № 1. 119–126.] https://elibrary.ru/YINZJZ