Formulation of a whey-based beverage supplemented with collagen protein hydrolysate

Mariya Gorbacheva[1*], *Inessa* Zachesova[1], *Konstantin* Esepenok[1], and *Lyudmila* Zemtsova[1]

[1]Moscow State Academy of Veterinary Medicine and Biotechnology - MVA named after K.I. Skryabin, 23, Akademika Skryabin Street, Moscow, 109472, Russia

Abstract. Production of enriched foods is one of the priorities of the Russian policy focused on national health. The use of collagen is of strong interest in many sectors of national economy, with food production being no exception. This is due to a variety of its properties. The article deals with the formulation of the enriched whey-based beverage supplemented with collagen protein hydrolysate. The objects of research were experimental variants of formulas of a whey-based beverage resulting from the manufacture of Adyghe cheese with a mass fraction of fat of 45%. The tested samples were added with, g/100 g: collagen protein hydrolysate from 1.5 to 3.5, and honey from 5 to 7. The choice of input raw components has been justified, and the benefits of addition of collagen in the formulation of the whey-based beverage have been demonstrated. It was found that the sample beverage had the most balanced composition and flavour profile with the following component ratio, g/100 g: cheese whey of 91.23; honey of 6.0; pectin of 0.25; collagen protein hydrolysate of 2.5; preservative of 0.02. All variants of the beverage had low energy values, not exceeding 78.6 kcal (330.12 kJ) per 100 g. Due to low content of fat and casein, the beverage can be classified as a dietary product, and the introduction of collagen hydrolysate as an enrichment agent in its formulation increases the biological value of the beverage. Daily consumption of a daily serving (330 ml) of this beverage has been proven to provide 22.57% of the recommended daily intake of protein.

1 Introduction

Ready-to-drink protein beverages are unique food products that are made with a variety of bioactive ingredients. These beverages can be helpful in meeting your daily nutrient requirements. In addition, they can be consumed on the go, which is important to people who prefer active lifestyles. Thanks to their exceptional properties, nowadays, protein drinks are increasingly being promoted as sport foods. In the Russian market, consumer demand for protein drinks is at the initial stage. This is due to a number of factors. First, the range of this product category is still limited. The market is dominated by energy and sports drinks with high caffeine and sugar content. Second, there is limited public awareness on the benefits of protein-enriched foods. Many people do not realise that protein is crucial to

* Corresponding author: gmv76@bk.ru

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maintaining good health, aiding muscle recovery after exercise and helping to control weight. Protein drinks can be a great source of essential amino acids, especially for those with active or athletic lifestyles. It is worth noting that in recent years there has been an increased interest in healthy food. People tend to choose food that are beneficial to their bodies. This sets the stage for a growing demand for protein drinks that can be tasty and nutritious at the same time.

The papers of the Russian scientists such as A.A. Kochetkova, G.L. Filonova, T.I. Ivanova, V.M. Pozniakovsky, N.V. Nepovinnykh, L.A. Tenkovaskaya, E.I. Cherevach, V.M. Vorobyova, L.V. Antipova, E.A. Smirnova, and others, highlighted the importance of high technical solution in production of enriched and specialized food products.

Recently, there has been a high level of interest in various products containing collagen, especially dietary supplements, enriched food products, including medical, cosmetic and veterinary products [1, 2]. The widespread use of collagen in various economic sectors is driven by its production properties such as gelation, water binding, thickening, emulsion formation, foam formation, texturing, stabilisation, adhesion and bonding, protective colloidal function and film formation [3]. The versatility of collagen makes it indispensable in a number of industries, opening new horizons for research and product innovation. In the Federal State Budgetary Educational Institution of Higher Education Moscow State Academy of Veterinary Medicine and Biotechnology – MVA named after K.I. Skryabin, original technologies for collagen production have been developed at the S.A. Kasparyants Department of Technology and Quality Management of Agricultural Products.

The research helped to find that biological substances obtained from cattle split leather, reindeer hides and cattle tendons have high characteristics. The dry matter content of these samples ranges from 0.8% to 1.8%. Protein levels were 97.9% in the sample obtained from reindeer hide, and 99.1% in protein refined condensates (PRC) from tendons. These data suggest that the obtained biological substances can be used as a high-quality source of protein, suitable for the formulation of supplements, dietary nutrition, and for the production of functional food products [4]. The unquestionable advantage of collagen use in liquid food formulations is its neutral taste and smell, use of its hydrolysed and native forms, the latter additionally acting as a stabiliser. It follows from the above that research and innovative technical solutions in the production of collagen-containing beverages are relevant and timely.

The objective of the paper is to formulate the enriched whey-based beverage supplemented with collagen protein hydrolysate.

2 Materials and methods

The objects of research were experimental variants of formulas of a whey-based beverage with the addition of collagen protein hydrolysate and honey. The control sample was a beverage without the added collagen protein hydrolysate.

Cheese whey, a milk processing by-product resulting from the manufacture of Adyghe cheese with a mass fraction of fat of 45%, was used as a main ingredient (Table 1). The main raw material for Adyghe cheese was whole cow's milk with mass fat content of 4.5%.

Table 1. Physical and chemical parameters of cheese whey.

Parameter name	Parameter value
Mass fraction of dry matter, %, including:	6.15±0.10
protein	0.63±0.01
fat	0.22±0.01
carbohydrates	5.3
lactose	4.94±0.02
Titrable acidity, °T	13.0±0.42
Density, kg/m^3	1.021.0±0.5

In order to improve the taste and flavour, as well as the storage quality of the beverage, natural flower honey, apple pectin, potassium sorbate have been added to its formulation. To enrich the product, preference was given to the hydrolysed form of collagen from cattle split leather trimmings without flavourings, dyes and artificial additives with the following characteristics: mass fraction of protein of not less than 96.0%, pH of 4.5, density of not less than 1,004.0 kg/m^3, and TVC of not more than 10 CFU/cm^3.

In view of the fact that the properties of the finished product directly depend on the selected raw material components, their quality was previously analysed according to GOST R 53438-2009, GOST 33957-2016, GOST 33692-2015, GOST R 54644-2011, GOST 19792-2017. It was determined that whey, honey and collagen protein hydrolysate met the requirements of standards in terms of organoleptic properties.

The prepared beverages have been tested for organoleptic, physical and chemical quality parameters in accordance with GOST 33957-2016. Mass fraction of dry matter was determined according to GOST R 54668-2011, and mass fraction of total protein was determined according to R 4.1.1672-03. Amino acid contained in the beverage have been determined by liquid chromatography according to GOST 34230-2017 using a liquid chromatograph, which comprised FLD detector No. DEAE303107, DAD detector No. DEAEK06520, auto sampler No. DEAEQ31879, column oven No. DEAEM05719, model 1260 Infinity, II LC.

The flowchart of preparation of the whey-based enriched beverage is given in Figure 1.

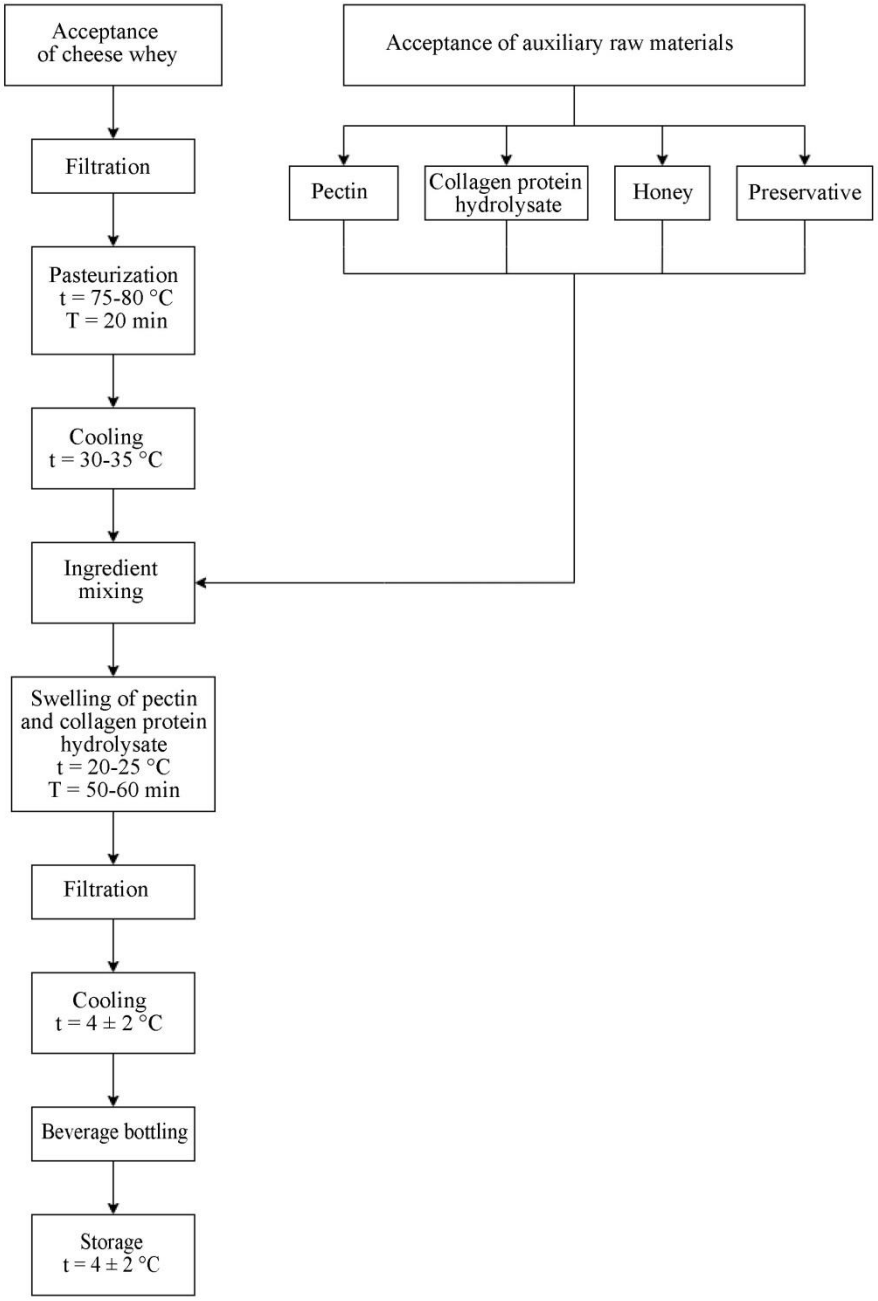

Fig. 1. Flowchart of preparation of whey-based enriched beverage.

Figure 1 shows that the production of the enriched beverage consists of the following basic stages: acceptance and control of raw materials quality, filtration, pasteurization, ingredient mixing, swelling of pectin and collagen protein hydrolysate, filtration, cooling, beverage bottling and storage.

3 Results

It follows from SanPin (sanitary regulations and standards) 2.3.2.2804-10 “Additions and amendments No. 22 to SanPin 2.3.2.1078-01 “Hygienic requirements of safety and nutritional value of food products”, that the beverage can be classified as an enriched product, provided that at the normal rate of its consumption (daily portion), it satisfies 15 to 50% of the physiological need for the corresponding micronutrient. It is recommended to enrich it with the substances whose deficiency is widespread and proven [5]. Three formulations of the beverage samples and one control sample have been developed to select the ingredient ratio ensuring high biological value and rich appetizing flavour of the liquid product (Table 2). It should be noted that collagen concentration ranged from 1.5 to 3.5 g/100 g.

Table 2. Formulation of whey-based enriched beverage, kg/100 kg.

Ingredient name	Sample 1	Sample 2	Sample 3	Sample 4 (control)
Cheese whey	93.23	91.23	89.23	93.73
Honey	5.0	6.0	7.0	6.0
Pectin	0.25	0.25	0.25	0.25
Collagen protein hydrolysate	1.5	2.5	3.5	-
Preservative (potassium sorbate)	0.02	0.02	0.02	0.02

It is important to emphasise the outstanding properties of whey, which is a unique product with a high nutritional value due to the significant content of organic and salt substances. Lactose is the main component of the milk whey and makes up about 70% of the dry matter, containing valuable carbohydrates [6]. The results of the information search show that it contains up to 30 macro- and microelements and the following vitamins, μg/kg: β-carotene - 13; A - 22; E - 227; B1 - 315; B2 - 1389, B_6 - 524; choline - 160000, PP - 140, C - 500. Whey is, among other things, rich in protein (6 to 10 g/l), which makes it widely used for nutritional, biological and functional applications [6, 7].

Honey is an exceptional product that contains not only carbohydrates, but also many other beneficial components. It is important to note that carbohydrates in honey are represented mainly by monosaccharides - glucose and fructose that contain about 95-99% of the dry matter. The percentage ratio of monosaccharides is 32-36% and 36-39%, respectively, which ultimately determines the main properties of honey: its sweetness, nutritional value, crystallizability, hygroscopicity, etc. In addition to monosaccharides, honey contains organic acids such as gluconic acid, which is critical in maintaining the acid-base balance of the body. The minerals in honey are represented by potassium, calcium, magnesium, iron and other elements essential for the normal vital activity. B vitamins and vitamin C help strengthen the immune system and improve overall health (*L.A. Tenkovskaya, 2015*).

The key properties of collagen, such as non-toxicity, complete absorption in the body and stimulation of reparative processes, constitute favourable background for this natural biopolymer to be used in the studied whey-based beverage.

The organoleptic test of the enriched beverage showed that the samples were of high quality and obtained high scores (Figure 2). Sample 2 obtained the highest score of 4.9, outperforming Sample 1 and Sample 4 by 11.36% and Sample 3 by 14.49%, respectively.

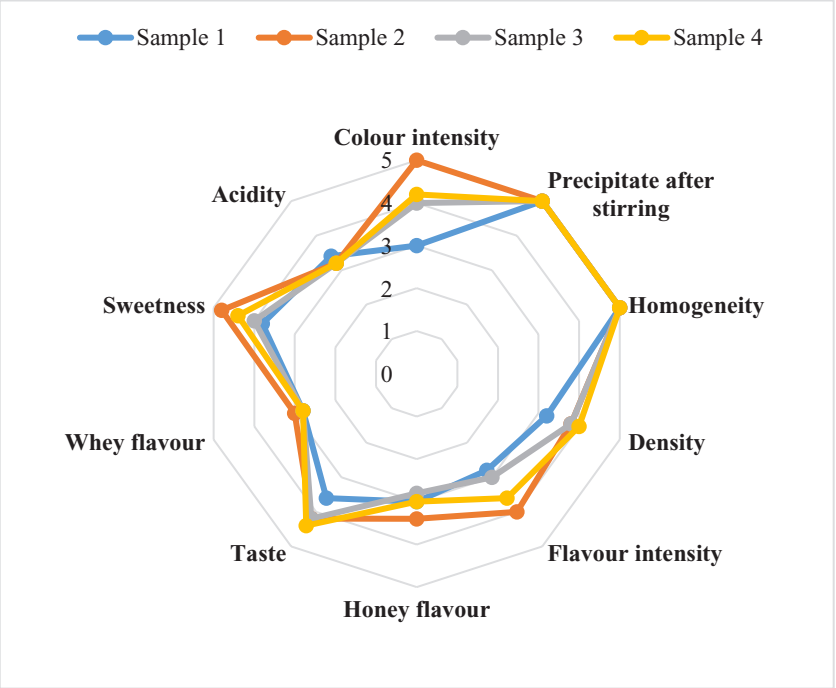

Fig. 2. Organoleptic properties of the whey-based enriched beverage.

Significant difference in the evaluation of organoleptic properties was observed for colour intensity, density and taste of the product. The control Sample 4 had the lowest score of 3.8 for these parameters. The density of Sample 2 and Sample 3 was scored 4.8 points by the commission. Interestingly, Sample 2 surpassed the other samples in terms of flavour and whey flavour. The same applies to acidity and sweetness (Figure 2). Therefore, tasting evaluation of quality established that Sample 2, supplemented with collagen protein hydrolysate, have the high organoleptic properties. Importantly, the addition of collagen protein hydrolysate as an ingredient to the beverage does not adversely affect its flavour and taste.

Samples of the beverage were subjected to physical and chemical tests that are summarized in Table 3.

Table 3. Physical and chemical quality parameters of the whey-based enriched beverage.

Parameter	Sample 1	Sample 2	Sample 3	Sample 4 (control)
Mass fraction of dry matter, %, including:	14.4±0.3	17.1±0.3	19.3±0.3	13.9±0.3
protein	2.28±0.11	3.42±0.14	4.39±0.17	0.97±0.07
fat	0.19±0.14	0.28±0.14	0.28±0.14	0.22±0.14
carbohydrates	11.93	13.40	14.63	12.71
Density, kg/m^3	1,054.0±1.0	1,056.0±1.0	1,069.0±1.0	1,050.0±1.0
Energy, kcal/KJ	58.55/ 245.91	69.8/ 293.16	78.6/ 330.12	64.38/ 270.40

It was found that Sample 3 had a dry matter concentration of 19.3%, which was 34.03%, 12.87% and 38.85% more as compared to the rest of the samples (Table 3). Also, Sample 3 was found to have a higher protein content of 4.39%, which can be explained by

the higher concentration of collagen protein hydrolysate in the beverage formulation as opposed to the other samples. No significant difference in the values of mass fraction of fat in the tested samples was found, as it did not exceed 0.28±0.14%. The amount of carbohydrates in the beverage samples directly depended on the amount of added honey; therefore, Sample 3 had the highest amount of carbohydrates, i.e. 14.63%, as its honey content was 7 kg/100 kg as per the formulation. Notably, the selected combination of collagen and honey influenced the density of Sample 3, which was 1,069 kg/m³. On the contrary, the lowest value of density was detected in control Sample 4. It was found that all variants of the beverage had low energy values, not exceeding 78.6 kcal (330.12 kJ) per 100 g. Due to low content of fat and casein, the whey-based beverage can be classified as a dietary product, and the addition of collagen hydrolysate in its formulation increases the biological value of the beverage.

Based on findings of studies of organoleptic characteristics of the whey-based beverage, and its physical and chemical properties, it was established that Sample 2 was the most consumer appealing. Therefore, the amino acid composition was determined in Sample 2 and Sample 4 (control) (Table 4).

Table 4. Amino acids contained the whey-based enriched beverage, mg/100 g.

Parameter	Sample 2	Sample 4 (control)
Lysine*	238.26±31.31	190.57±27.18
Methionine*	< 10	< 10*
Cysteine	87.12±11.23	77.17±13.09
Alanine	288.935±36.12	167.25±22.20
Arginine	351.98±40.29	311.98±41.95
Asparagic acid	292.55±33.11	278.87±32.36
Valine*	161.93±24.63	125.72±21.63
Histidine	22.52±2.19	65.80±20.50
Glycine	1,149.8±96.88	1,091.04±92.24
Glutamic acid	761.50±52.16	591.45±54.86
Isoleucine*	608.38±46.21	< 10*
Leucine*	88.7±14.14	67.12±17.87
Proline	133.49±17.31	< 10*
Serine	176.57±20.23	156.69±18.69
Tyrosine	< 10	10.89±1.39
Threonine*	120.9±10.98	111.50±11.46
Tryptophan*	2,382.09±107.50	1,697.48±85.26
Phenylalanine*	222.20±25.68	198.96±27.74

Note: *essential amino acids

According to the data presented in Table 4, it was found that the whey-based beverage contains all essential amino acids, which accounted for at least 85% of the total amount of amino acids in Sample 2. This is beneficial to the consumer, as the essential amino acids are well known for maintaining normal protein metabolism in the body, synthesis of antibodies, hormones and enzymes, etc. We must highlight the high content (87.12 mg/100 g of product) of cysteine, sulphur-containing amino acid that is synthesised from methionine and contributes to the formation of insulin and immunoglobulins. Of note is the high amount of proline found in the beverage sample with the use of collagen protein hydrolysis. The same applies to other amino acids, the proportion of which prevailed in Sample 2.

The amino acid score was calculated to determine the biological value of proteins. The results are shown in Table 5.

Table 5. Biological value of protein contained in the whey-based enriched beverage, mg/100 g.

Amino acid	Amino acid score	
	of Sample 2	of Sample 4
Isoylecin	4.45	< 10.0
Leucine	0.37	0.28
Lysine	1.27	1.01
Methionine+cystine	0.73	0.65
Phenylalanine+tyrosine	1.08	1.02
Tryptophan	69.65	49.63
Threonine	0.88	0.82
Valine	0.95	0.74

The data in Table 5 show that leucine is the major limiting amino acid. And Sample 2 has rather high value of amino acid score, on the average on 46.6% superior to Sample 4. Whereby, Sample 2 is more balanced in terms of amino acid composition of protein compared to Sample 4 and has a high biological value.

These studies allowed adjusting the final formulation of the whey-based enriched drink (Table 6).

Table 6. Formulation of the whey-based enriched drink.

Ingredients	Ratio kg/100 kg
Cheese whey	91.23
Honey	6.0
Pectin	0.25
Collagen hydrolysate	2.5
Preservative (potassium sorbate)	0.02

The ingredient ratios given in Table 6 have been shown to be preferential for the preparation and use of the enriched whey-based beverage. It is also important that the collagen protein hydrolysate added to the formulation will promote regeneration of mucous membranes of the gastrointestinal tract and boost the immune system.

4 Conclusion

Based on the obtained data it can be summarised that the amount of total protein and essential amino acids contained in the beverage as per the developed formulation enables it to be classified as enriched. Thus, consumption of 330 ml of the proposed enriched drink based on whey provides protein intake of 22.57% of the recommended daily allowance. The production of the collagen-enriched drink will expand the range of liquid dairy products.

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