Caspian Journal of Environmental Sciences
Online ISSN: 1735-3866
Print ISSN: 1735-3033

Development of a functional instant drink using soy isolate and collagen hydrolysate

Dina Dautkanova[1], Assemay Kazhymurat[2]*, Nurlan Dautkanov[1], Zhibek Ussembayeva[1], Saltanat Mussayeva[3]

1. Kazakh Research Institute of Processing and Food Industry, Almaty, Kazakhstan
2. JSC Almaty Technological University, Almaty, Kazakhstan
3. South Kazakhstan University named after M. Auezova, Shymkent, Kazakhstan

* *Corresponding author's email: assemay2006.87@mail.ru*

ABSTRACT

This study presents the development and optimization of a functional instant drink based on soy protein isolate and collagen hydrolysate. The amino acid composition analysis demonstrated the complementarity of the two protein sources: soy isolate is rich in essential amino acids crucial for muscle protein synthesis, while collagen hydrolysate contributes high levels of glycine, proline, and hydroxyproline, important for connective tissue health. A 1:1 protein blend ensured a balanced amino acid profile, enhancing both nutritional and functional properties. Using response surface methodology (RSM) and second-order regression modeling, the optimal formulation was identified to ensure maximum solubility (90.36%) and excellent organoleptic characteristics (9.25/9 points). The statistical analysis confirmed the significance of soy isolate and collagen hydrolysate levels, as well as their interactions with psyllium fiber. Notably, collagen showed a consistently positive impact on solubility and sensory properties, while psyllium negatively influenced both parameters at higher concentrations. The final formulation demonstrated high protein content (87.3%), dry matter content (96.3%), and compliance with microbiological and toxicological safety standards. These results highlight the potential of the developed instant beverage as a functional product suitable for preventive nutrition, sports applications, and the nutraceutical market.

Keywords: Soy protein isolate, Collagen hydrolysat, Instant functional drink.
Article type: Research Article.

INTRODUCTION

The modern food industry is witnessing a surge in the development of functional foods, with beverages leading the way due to their convenience and ease of consumption. Functional drinks, designed to offer health benefits beyond basic nutrition, are gaining traction as consumers seek proactive approaches to wellness (Puiggròs *et al.* 2017). These beverages often incorporate bioactive ingredients like phenolic compounds, vitamins, minerals, amino acids, unsaturated fatty acids, and peptides derived from probiotic microbes, plants, or animals. The formulation of functional drinks utilizes diverse natural raw materials, reflecting a growing interest in leveraging traditional knowledge and biodiversity to create health-promoting products (Gupta *et al.* 2023). Functional beverages must contain adequate amounts of functional components, generally between 10 and 50% of the recommended daily intake, to be categorized as such. The global market for functional beverages is expanding rapidly, driven by increasing consumer awareness of the link between diet and health, with projections indicating continued growth in the coming years (Sharma *et al.* 2021) . This heightened interest underscores the importance of innovation in the food sector, particularly in research and development, to meet evolving consumer needs and preferences. The exploration of novel protein sources, such as soy isolate and collagen hydrolysate, holds significant promise for creating functional instant drinks with enhanced nutritional profiles and targeted health benefits (Gupta *et al*. 2023). Soy isolate, derived from soybeans, is a high-quality plant-based protein source containing all essential amino acids, making it a valuable ingredient for enhancing the nutritional value of food products. Soy protein has been extensively studied for its potential health benefits, including reducing cholesterol levels and improving bone health, which positions it as a compelling ingredient for functional foods. Additionally, soy protein exhibits excellent functional properties, such as emulsifying and gelling capabilities, making it suitable for various food applications (Gupta *et al*. 2023; Intrasook *et al*. 2024). Collagen hydrolysate, on the other hand, is derived from collagen, the most abundant protein in the human body, and is rich in amino acids like glycine, proline, and hydroxyproline. These amino acids are crucial for maintaining the structural integrity of skin, joints, and bones, thus offering potential benefits for joint health, skin elasticity, and bone density. Collagen hydrolysate is produced through enzymatic hydrolysis, breaking down collagen into smaller peptides that are easily digestible and bioavailable, increasing its utility in functional beverages. The combination of soy isolate and collagen hydrolysate in a functional instant drink can offer a synergistic effect, providing a complete protein source with a range of amino acids and potential health benefits (Intrasook *et al.* 2024). When formulating an instant drink, it is essential to consider factors such as solubility, dispersibility, taste, and stability to ensure a palatable and effective product. Soy isolate and collagen hydrolysate have different solubility properties, and the formulation process should optimize their interaction to prevent sedimentation or aggregation in the final product. Using encapsulation technologies, such as protein-based nanostructures, can protect bioactive molecules and comply with the requirements for food industry applications (Gerliani *et al*. 2019). Furthermore, the taste of soy isolate can be a concern for some consumers, and masking agents or flavor enhancers may be necessary to improve the overall sensory experience. The addition of whey powder in plant-based beverages containing soy extract can influence the particle size and stability of the proteins, affecting the beverage's overall characteristics. Moreover, the stability of the protein hydrolysates during storage and distribution must be addressed to maintain their bioactivity and prevent degradation (Nonglait *et al.* 2022). The optimization of these factors can lead to a functional instant drink that is not only nutritious but also enjoyable to consume. Soy beverages can serve as an alternative to cow's milk, but a major limiting factor in their use is the presence of considerable amounts of non-digestible oligosaccharides and the unpleasant odor and taste caused by lipoxygenase, an enzyme from soybean grain. Protein ingredients can be obtained from plant raw materials through different technological processes such as precipitation, fractionation, and extraction, which seems to be valuable for the improvement of nutritional profile of plant-based milk alternatives (Liu *et al.* 2023). Plant-based proteins have drawn considerable interest lately due to their health benefits, sustainable origins, and affordable costs compared to those derived from animals. In order to secure food supplies for the world's expanding population while also addressing the concerns about climate change, it may become necessary to move toward a more plant-based diet. The need for alternative protein sources to meet global protein needs has been brought on by the expansion of the world's population and the growing scarcity of resources (Vogelsang-O'Dwyer *et al.* 2021). Due to their superior nutritional qualities, widespread availability, and well-established functionality in food systems, plant-based proteins have been widely used in the food industry. Plant proteins differ from animal proteins in terms of structure, composition, and food functionality. This review examines the possibility of creating a new generation of semi-purified, plant-derived ingredients with enhanced sustainability and health benefits, as well as how protein can be extracted from plant materials to create protein-rich ingredients for plant-based foods. The increasing demand for plant-based protein is fueled by consumer dietary changes, health claims, and the sustainability of plant-based protein production compared to animal protein (Akharume *et al.* 2021). Plant-based products are becoming more and more popular as consumers look for sustainable and ethical food choices. Plant-based options are viewed as ecologically beneficial because they use fewer natural resources and have a smaller carbon footprint than meat-based products. A wide variety of plant proteins are used to develop meat and dairy substitutes; however, their characteristics differ from those of animal proteins and are not adequately described (da Silva *et al.* 2024). The food industry is increasingly interested in plant-based milk substitutes as more consumers look to cut back on their intake of animal products like cow's milk for moral, environmental, and health reasons. Using plant proteins in place of animal proteins presents a number of opportunities and challenges in the food industry. Due to sustainability issues, health concerns related to animal fat intake, lactose intolerance, and allergies to milk proteins, plant-based alternatives to dairy milk are gaining market share relative to cow's milk in the United States, Europe, Australia, and New Zealand, and are forecasted to grow at a rate of 8-15% (Short *et al.* 2021). Innovation in the food sector has been sparked by the growing consumer demand for plant-based substitutes for conventional animal products, such as cheese. Using dryfractionated pea protein and inulin-olive oil emulsion may improve the formulation of plant-based cheese on a wider scale, as the ingredients and product ideas that have been developed may be used to do so. Plant-based diets are becoming increasingly popular because they are thought to be healthier for both people and the environment than diets that include meat (Mefleh *et al.* 2022). Due to the growing trend of vegetarianism, consumers' increased awareness of the effects of their food choices on the environment and health, and the limited use of dairy products in some regions, there is an increasing demand for plant-based products. The market for plant-based foods is expanding, which presents opportunities for innovation and the creation of novel products that cater to a wide range of consumer preferences and dietary requirements. It is also believed that the importance of meat alternatives will continue to increase because of concerns on limited sustainability of the traditional meat production system. Meat analogues can be manufactured utilizing protein ingredients such as soy concentrate, gluten, soy isolate, pea concentrate, and canola concentrate (Lee *et al*. 2020). Due to ethical and environmental concerns associated with meat consumption, the development of meat substitutes has grown rapidly. Using pulses to produce plant-based cheese alternatives seems promising because of their nutritional value, which opens up opportunities for product development in this area (Lu *et al*. 2025). It is very important to explore the functionality of plant-based proteins, as well as how processing techniques affect their functional properties. Soy protein isolate is an excellent source of protein for meat alternatives and has a number of health benefits. It has been demonstrated to lower cholesterol. Soy protein isolate can be utilized in the creation of meat substitutes as well as other food items. Collagen hydrolysate is a popular ingredient in functional foods and supplements because of its potential health benefits, especially for joint health and skin elasticity (Lee *et al*. 2022). There has been an increased research and development in the field of alternative food products, such as meat analogues, as a result of the rising interest in food products that do not contain animal-based ingredients. Examining the nutritional profiles, environmental effects, and consumer acceptability of plant-based meat substitutes requires an interdisciplinary approach. It has been determined that the environmental effect and resource demand are minimal for plant-based meat substitutes in general (Andreani *et al*. 2023). They typically have a smaller carbon footprint and require less land and water than conventional meat production. Innovations in processing technologies, ingredient sourcing, and formulation strategies are essential for improving the texture, taste, and nutritional value of meat substitutes. Texture, taste, and nutritional content are the main obstacles in the development of plant protein-based meat alternatives (Zhang *et al.* 2021). Soy-based products have been used in the meat processing industry for over 60 years because they are plant-based ingredients that have been chosen for certain jobs in meat products and have made ingredients available through a wide range of processes to change or optimize their functionalities (Zhang *et al.* 2021) . Proteins are a fundamental component of meat analogues, and their functionalities have been thoroughly researched to mimic the look and feel of meat. Almost every plant protein can be utilized as a raw material for meat analogues, but their suitability is determined by a number of crucial factors. These factors include the accessibility of a steady supply, cost, flavour, and functional qualities. The functionality of plant proteins is influenced by a number of factors, including cultivars, genotypes, extraction and drying methods, protein level, and preparation methods (Tang *et al*. 2024). Alternative proteins, such as those derived from plants, fermentation, or cultured from animal cells, utilize a considerably smaller amount of land and water than traditional meat production. These novel proteins have a number of technological and functional qualities that can be used to create a variety of protein systems on different scales, which can be customized for a specific application in innovative food products. It is important to remember that novel protein sources must establish new value chains, and issues such as production costs, food safety, scalability, and consumer acceptance must be taken into consideration (Carey *et al.* 2023).

MATERIALS AND METHODS

Recipe development. A series of soluble drink formulations with various ratios of soy isolate and collagen hydrolysate have been created. The protein concentration was optimized to improve the functional properties and nutritional value of the final product, and preliminary tests were conducted to determine the ideal protein ratio for taste, texture, and solubility.

Manufacturing process. The developed ingredients for the recipe were carefully measured and combined in a controlled environment. To ensure consistency and uniformity, the powdered components were combined using a high-speed mixer. To guarantee the stability and long shelf life of the soluble drink powder, the mixture was dried using a low-temperature method. After drying, the powder was sieved to achieve a uniform particle size, which improved its flowability and solubility upon reconstitution. To prevent moisture ingress and maintain product quality, the finished powder was packaged in airtight sachets with a moisture-proof barrier.

Quality and safety control. The content of heavy metals and mineral substances was determined by atomic absorption spectroscopy (AAS) on the "Kvant-Z.EA-T" spectrometer with electrical atomization (Delgado-Andrade et al. 2003).

The pesticide content in finished products was determined using the stationary analytical gas chromatograph "Kristallux-4000M" (Russia) with an electron capture detector and the software "NetChrom" (Russia; Capita *et al.* 2004). When studying the microflora of the instant beverage, classical methods of microbiological analysis were used: methods for sampling and preparing samples for microbiological analyses (Wagner *et al.* 2019).

To determine the amino acid composition of the protein components, a chromatographic method was used. The protein content in the powder was determined using the Kjeldahl method (Jarvas *et al.* 2020).

Organoleptic evaluation. Methods of determining product quality based on the perception of its appearance, color, smell, taste, consistency, and other sensory characteristics were performed. The evaluation was conducted under standardized conditions using a 10-point scale according to GOST R 56784-2015 – Instant beverages. General technical.

Process optimization. To optimize the formulation of the instant functional drink, the response surface methodology was used (Myers *et al.* 2016).

Modelling and processing of experimental data were carried out using the Statistica 10 package (StatSoft, Inc.).

RESULTS

Results of optimizing the recipe composition of an instant functional drink based on quality indicators and functional properties are as follows. At the first stage of the research, in accordance with the set tasks, work was carried out to optimize the recipe composition of the drink with collagen hydrolysate and soy isolate with the addition of psyllium fibre. To describe the dependence of solubility (Y_1) and organoleptic assessment (Y_2) on the concentrations of three key components — soy isolate (A), collagen hydrolysate (B), and psyllium (C) — second-order regression models were constructed. Such models allow for the consideration of not only the direct influence of each factor but also their quadratic effects and interactions with each other.

Solubility model (Y_1):

$$Y_1 = 94.59 - 2.20A + 0.43B - 0.58C - 0.008A^2 + 0.25B^2 - 0.56C^2 + 0.019AB + 0.78AC - 0.83BC \quad (1)$$

Model organoleptic (Y_2):

$$Y_2 = -0.59 + 3.07A + 2.33B + 0.03C - 0.43A^2 - 0.41B^2 - 0.34C^2 + 0.074AB + 0.29AC - 0.088BC. \quad (2)$$

Each equation is a second-degree polynomial that includes:

- linear terms (A, B, C), reflecting the main influence of the factor;
- quadratic terms (A^2, B^2, C^2), reflecting the nonlinearity of the dependence;
- pairwise interactions (AB, AC, BC), reflecting synergistic or antagonistic effects when two factors are present simultaneously.

The model's form allows not only predicting response values under various factor combinations but also optimizing the product composition based on selected quality criteria. As a result of the modelling, the following regression equations for solubility and organoleptics were obtained:

$$Y_1 = 94.59 - 2.20A + 0.43B - 0.58C - 0.008A^2 + 0.25B^2 - 0.56C^2 + 0.019AB + 0.78AC - 0.83BC$$

$$Y_2 = -0.59 + 3.07A + 2.33B + 0.03C - 0.43A^2 - 0.41B^2 - 0.34C^2 + 0.074AB + 0.29AC - 0.088BC$$

The greatest influence on solubility is exerted by the negative coefficient at A (soy isolate), while the positive coefficients at A and B affect the organoleptic evaluation. Psyllium, on the other hand, has a reducing effect, especially in combination with collagen. The experimental design matrix and response functions are presented in Table 1. To check the significance of the regression coefficients (1) and (2), Pareto charts were constructed, as shown in Fig. 1 (L – linear effect, K – quadratic effect). The specified Pareto chart (Fig. 1) presents the standardized coefficients, which are sorted by absolute values. The diagram reflects the relative statistical significance of the regression coefficients in the response surface model developed for optimizing the formulation of an instant functional beverage. The vertical bars show the absolute values of the t-statistics (|t|), calculated as the ratio of the regression coefficient to the corresponding *p*-value. This allows for a quantitative assessment of each factor's contribution to the model's predictive capability.

Table 1. Experiment planning matrix and response functions.

A (Soy Isolate; g/100g)	B (Collagen; g/100g)	C (Psyllium; g/100g)	Y_1 (Solubility; %)	Y_2 (Organoleptics), points
4.0	2.0	1.0	88.4	9.5
4.0	2.0	3.0	86.2	8.6
4.0	4.0	1.0	93.4	9.8
4.0	4.0	3.0	83.6	8.5
6.0	2.0	1.0	86.3	7.2
6.0	2.0	3.0	82.9	7.4
6.0	4.0	1.0	86.9	7.7
6.0	4.0	3.0	84.1	7.5
3.318	3.0	2.0	86.2	8.2
6.682	3.0	2.0	86.3	8.5
5.0	1.318	2.0	86.8	8.4
5.0	4.682	2.0	87.2	8.4
5.0	3.0	0.318	85.6	8.6
5.0	3.0	3.682	84.2	8.7
5.0	3.0	2.0	86.7	9.3
5.0	2.0	2.0	87.2	9.3
4.5	3.0	1.5	86.4	9.5
6.0	3.5	2.5	87.1	9.0

The model takes into account linear factors (A – soy isolate, B – collagen hydrolysate, C – psyllium), their quadratic terms (A^2, B^2, C^2), as well as interactions (AB, AC, BC). The columns are sorted in descending order of significance, so the most influential variables are on the left. On the secondary Y-axis, the cumulative curve (red line) is shown, demonstrating the accumulated percentage contribution of factors to the overall significance of the model. The dashed horizontal line at the 80% level serves as a visual boundary based on the Pareto principle (80/20 rule), allowing the identification of key variables.

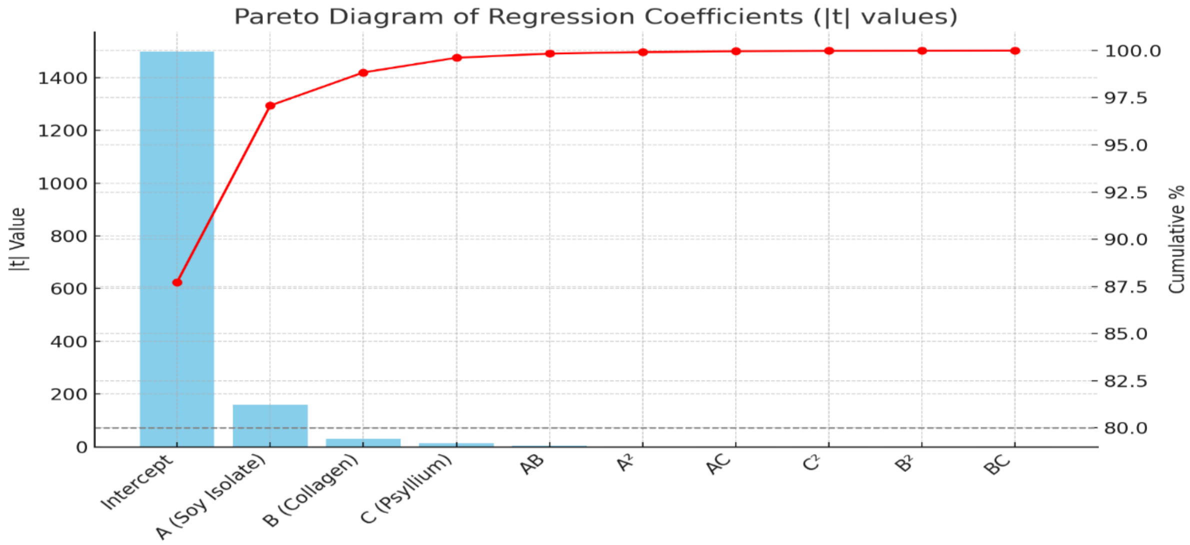

Fig. 1. Pareto chart of the significance of standardized regression coefficients (|t|-values).

The analysis showed that the highest |t| values are observed for factors A (soy isolate), B (collagen hydrolysate), and C (psyllium), indicating their decisive influence on the target response (e.g., solubility or organoleptic properties). Interaction terms and quadratic terms turned out to be less significant—especially BC, B^2, and C^2—which may indicate the possibility of excluding them from the model without losing prediction accuracy. This visualization allows for the objective identification of the most significant factors and their use in simplifying the model and enhancing its interpretability. The adequacy of the developed models was tested using the method of analysis of variance, and its results are presented in Table 2. The analysis of variance was conducted to assess the significance of the influence of each factor and their interactions on the response — in this case, the solubility or organoleptic evaluation of the functional beverage. As shown in the Table, factor B (collagen hydrolysate) has the greatest influence on the response ($F = 6.23$, $p = 0.0547$), being on the verge of statistical significance ($p < 0.05$). Moderate influence is also exerted by the quadratic and interaction terms, such as A^2, AB, and BC, with *p*-

values ranging from 0.09 to 0.11. Factors A (soy isolate) and C (psyllium), as well as their interactions AC and C^2, did not have a significant impact on the response ($p > 0.05$), which may indicate the possibility of excluding these terms in further model simplification. The remaining variance is explained by the residual error, corresponding to 5 degrees of freedom, which indicates the adequacy of the model for describing the experimental data at this sample size.

Table 2. Analysis of variance.

Factor	**Sum of Squares (SS)**	**Degrees of Freedom (df)**	**F-value**	***p*-value**
A	0.010276	1.0	0.029914	0.869469
B	2.141546	1.0	6.234135	0.054702
C	0.074922	1.0	0.218101	0.660134
A2	1.510051	1.0	4.395826	0.090139
B2	1.196169	1.0	3.482100	0.121028
C2	0.117477	1.0	0.341981	0.584081
AB	1.407680	1.0	4.097819	0.098826
AC	0.130972	1.0	0.381266	0.563959
BC	1.286619	1.0	3.745407	0.110735
Residual	1.717596	5.0	-	-

Recipe optimization

Through numerical optimization with equal significance of Y_1 and Y_2, the maximum point was determined: A = 4.0 g/100 g, B = 4.0 g/100 g, and C = 1.0 g/100 g. At these values, the predicted solubility responses were 90.36%, and the organoleptic evaluation was 9.25 points. Thus, this formulation ensures technological stability, a high degree of solubility, and excellent sensory acceptability. For visual analysis of the influence of formulation factors on responses, 3D response surfaces were constructed, reflecting the behavior of solubility and organoleptics depending on the variation of two factors with a fixed third factor (Figs. 2 - 3). The most indicative were the graphs with a fixed psyllium content (2.0 g/100 g), as it showed the greatest negative impact on both responses when exceeding a certain threshold.

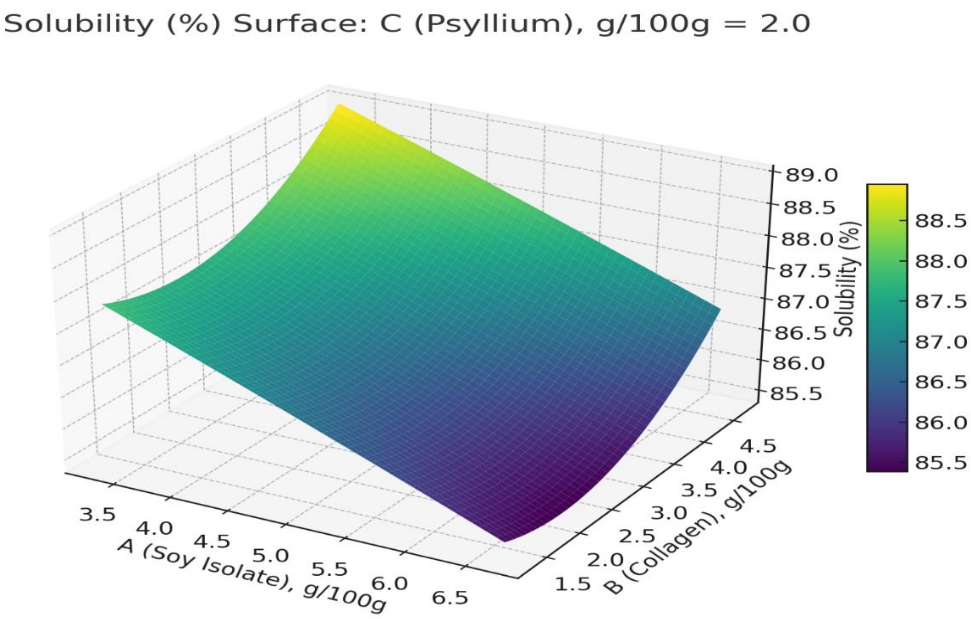

Fig. 2. Response surface of solubility (Y_1) as a function of soy isolate content (A) and collagen hydrolysate (B) at a constant level of psyllium (C = 2.0 g/100 g).

The graph presents a three-dimensional response surface illustrating the change in the solubility percentage of the functional instant drink depending on the variation of two factors: the content of soy isolate (A, g/100 g) and collagen hydrolysate (B, g/100 g), with a constant psyllium concentration of 2.0 g/100 g. The X-axis reflects the concentration of soy isolate in the range of 3.5 to 6.5 g/100 g, the Y-axis shows the collagen content from 1.5 to 4.5 g/100 g, and the Z-axis represents the response value, i.e., the solubility (%) of the product. The color scale on the right complements the visualization, showing solubility gradations from 85.5% to 89.0%. The analysis of the graph shows that the highest solubility values are achieved with an increased content of collagen hydrolysate, especially in combination with moderate levels of soy protein (~4.0–4.5 g/100 g). In contrast, higher concentrations of soy isolate (closer to 6.5 g/100 g) combined with low collagen content lead to a decrease in solubility. Thus, the graph demonstrates a synergistic interaction between the two protein components, where collagen enhances solubility, while an excess of soy protein may have the opposite effect. This data is crucial when selecting the optimal protein ratio in the product formulation.

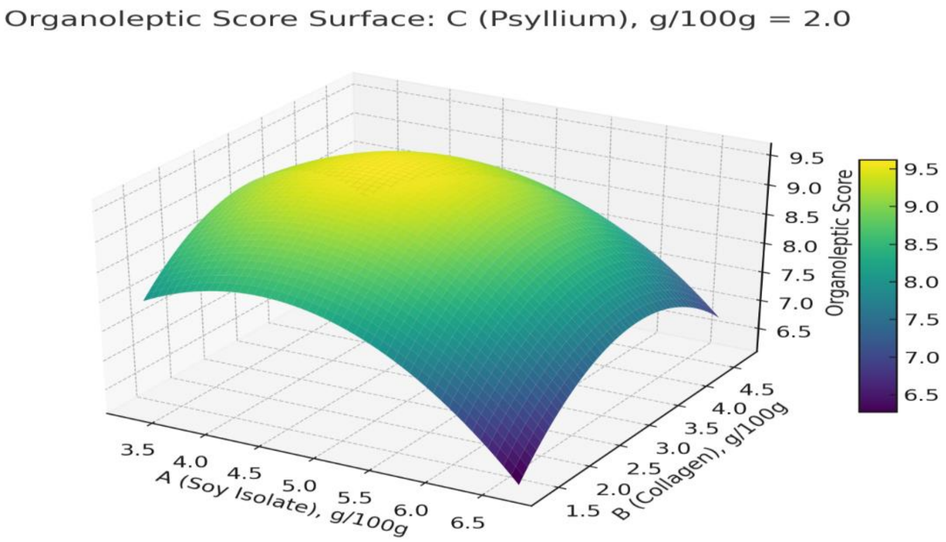

Fig. 3. Response surface of organoleptic evaluation depending on the content of soy isolate (A) and collagen hydrolysate (B) at a fixed level of psyllium (C = 2.0 g/100 g).

The graph presents a three-dimensional response surface characterizing the change in organoleptic evaluation (on a 9-point scale) of the functional instant drink depending on the variation in the content of soy isolate (X-axis, from 3.5 to 6.5 g/100 g) and collagen hydrolysate (Y-axis, from 1.5 to 4.5 g/100 g), with a constant level of psyllium — 2.0 g/100 g. The Z-axis reflects the values of the organoleptic assessment, ranging from 6.3 to 9.5 points. The color scale visualizes changes in the indicator: from purple (low values) to yellow (maximum scores). Surface analysis shows that the highest organoleptic scores are achieved with a balanced content of both proteins: around 4.5 g/100 g soy isolate and 3.0–3.5 g/100 g collagen hydrolysate. By excessive increase of one of the components (especially A > 6.0 g/100 g or B > 4.5 g/100 g), the organoleptic characteristics deteriorate, likely due to excessive viscosity, foam, or a specific aftertaste. This graph confirms the presence of an optimal range of protein ratios that provide the best sensory properties of the product, which is an important criterion in developing a commercially successful formulation. Thus, the graphs confirm the conclusions obtained from the regression equations and allow for the visual identification of the recipe's optimal area. Response surfaces were constructed for visual analysis. The graphs showed the optimal zone near the minimum values of psyllium and maximum values of collagen, with a moderate level of soy protein. This is consistent with the regression analysis. Results of the study on the chemical composition and quality indicators of an instant drink with collagen hydrolysate and soy isolate are provided. The chemical composition and main quality indicators of the instant drink with collagen hydrolysate and soy isolate obtained based on optimal ratio of recipe components, presented in Table 3. For this, experimental samples of the new instant drink were developed under laboratory conditions. The developed samples of the instant drink were packaged in sachet bags, and their chemical composition, safety indicators, and microbiological indicators were determined.

Table 3. Physico-chemical indicators of instant drink

Indicators	**Control instant drink sample**	**Instant drink with collagen hydrolysate and soy isolate**
Mass fraction of moisture (%)	5.70 ± 0.06	5.6 ± 0.0 5
Mass fraction of protein (%)	86 ± 1.0	87.30 ± 1.06
Moisture binding capacity (%)	700	800
Ash content (%)	4, 73 ± 0.05	4. 85 ± 0.05
Mass fraction of dry matter (%)	95.30 ± 1.30	9 6 .30 ± 1.30

The instant drink with collagen hydrolysate and soy isolate has improved physicochemical properties compared to the control sample: it contains a higher protein content of 87.3%, more dry matter at 96.3%, and ash at 4.85%, with a slightly lower moisture content of 5.6%. Additionally, the water-holding capacity increased from 700 to 800, indicating an improvement in functional properties.

Table 4. Microbiological indicators.

Microbiological indicators	Instant drink sample	
	Control	**Instant drink with collagen hydrolysate and soy isolate**
Mesophilic Aerobic and Facultative Anaerobic Microorganisms (CFU g^{-1})	17×10^{2}	
Molds (CFU g^{-1})	Not found	Not found
Coliform Bacteria (Coliforms) per g	Not found	Not found
Yeasts (CFU g^{-1})	Not found	Not found

Table 5. Safety Indicators of instant beverage.

Name of the indicator	Instant drink sample	
	Control	**Instant drink with collagen hydrolysate and soy isolate**
Toxic elements (mg kg^{-1})		
Lead	Not found	Not found
Arsenic	Not found	Not found
Cadmium	Not found	Not found
Mercury	Not found	Not found
Pesticides (mg kg^{-1})		
hexachlorocyclohexane (a-, p-, y-isomers)	Not found	Not found
DDT and its metabolites	Not found	Not found
Radionuclides (Bq kg^{-1})		
potassium - 40	Not found	Not found

All safety indicators of the instant drink with collagen hydrolysate and soy isolate are within acceptable norms: toxic elements (lead, arsenic, cadmium, mercury), pesticides (hexachlorocyclohexane, DDT and its metabolites), and radionuclides (potassium-40) were not detected, indicating its complete safety for consumption. Table 5 demonstrates the amino acid composition of soy isolate, collagen hydrolysate, and their mixture in a 1:1 mass ratio. Significant differences in the amino acid profile reflect the functional orientation of each component. The collagen hydrolysate contains an extremely high amount of glycine (22.0 g/100 g protein), proline (13.5 g/100 g), and hydroxyproline (12.0 g/100 g), while in the soy isolate, these values are 1.5, 1.9, and 0 g/100 g, respectively. These amino acids play a key role in supporting the structure of connective tissue and collagen synthesis. Soy isolate, on the other hand, is a source of essential amino acids: leucine (8.0 g), isoleucine (5.0 g), lysine (6.4 g), methionine (1.3 g), phenylalanine (5.2 g), which significantly exceed the values of collagen hydrolysate (leucine — 2.9 g, lysine — 3.5 g, methionine — 0.6 g, etc.). These amino acids are particularly important for muscle protein synthesis and overall protein balance in the body. The combination of soy protein and collagen in a 1:1 ratio allows for a balanced amino acid profile, increasing the content of both structural and essential amino acids. The mixture shows an increase in glycine to11.75 g/100 g, proline to 7.7 g, leucine to 5.45 g, as well as maintaining high levels of glutamic acid (14.1 g) and arginine (7.45 g).

DISCUSSION

The market for functional food products is expanding as consumers become more health-conscious. Soluble drinks that are both healthy and easy to prepare are becoming increasingly popular. The development of a functional soluble drink combining soy isolate and collagen hydrolysate aligns with this trend (Gupta *et al*. 2023). The research results showed that the application of the response surface methodology (RSM) using second-order regression modelling allows for the effective optimization of the formulation of the functional instant drink. The constructed regression equations for solubility (Y_1) and organoleptic evaluation (Y_2) demonstrate high explanatory power and statistical significance of the key factors. The greatest influence on solubility was exerted by the negative linear effect of soy isolate, which is likely due to its ability to aggregate and form less soluble protein complexes at high concentrations. In contrast, the collagen hydrolysate showed a positive effect on both responses, which can be attributed to its high solubility and favourable impact on the product's consistency. Psyllium, as a source of insoluble fibre, reduced both solubility and organoleptic qualities at concentrations above 2 g/100 g.

Table 5. Comparative analysis of amino acid indicators.

Amino acid	Soy isolate (g/100g protein)	Collagen hydrolysate (g/100g protein)	Mixture 1:1 (g/100g protein)
Glycine	1.5	22.0	11.75
Proline	1.9	13.5	7.7
Hydroxyproline	0.0	12.0	6.0
Alanine	4.5	8.9	6.7
Glutamic acid	18.2	10.0	14.1
Aspartic acid	11.6	6.5	9.05
Serene	5.1	3.2	4.15
Threonine	4.2	2.1	3.15
Arginine	7.4	7.5	7.45
Cysteine	1.4	0.0	0.7
Valin	5.0	2.4	3.7
Isoleucine	5.0	1.5	3.25
Leucine	8.0	2.9	5.45
Phenylalanine	5.2	2.0	3.6
Tyrosine	3.9	0.5	2.2
Histidine	2.6	1.0	1.8
Methionine	1.3	0.6	0.95
Lysine	6.4	3.5	4.95

The analysis of Pareto charts and the results of the dispersion analysis confirmed the significance of linear factors A and B, as well as the partial significance of their quadratic and interacting effects. The interaction results of AC and BC are particularly interesting, where synergy or antagonism is observed in the simultaneous presence of fibre and proteins. The numerical optimization conducted allowed for the determination of the formulation combination (A = 4.0 g/100 g, B = 4.0 g/100 g, C = 1.0 g/100 g), ensuring a high level of solubility (90.36%) and sensory perception (9.25 points on a 9-point scale). The conducted chemical and microbiological analyses confirmed the safety, stability, and nutritional value of the obtained beverage. In particular, the protein content reached 87.3%, the dry matter content was 96.3%, and the microbiological indicators and toxic element content met safety requirements. A comparative analysis of the amino acid composition showed that the combination of soy isolate and collagen hydrolysate provides a balanced amino acid profile. Collagen contributes glycine, proline, and hydroxyproline, which are necessary for the synthesis of connective tissue, while the soy isolate compensates for the deficiency of essential amino acids—leucine, isoleucine, methionine, and lysine. Thus, the joint use of the two proteins allows for both nutritional completeness and functional characteristics. Using RSM and regression analysis methods allowed for the identification of significant factors and the construction of reliable response models. The development of a functional instant drink using soy isolate and collagen hydrolysate involves leveraging the nutritional and functional properties of both components to create a health-promoting beverage. Soy protein isolate (SPI) is known for its high protein content and essential amino acids, while collagen hydrolysate offers benefits for bone, cartilage, and skin health. Combining these ingredients can result in a beverage that supports overall health and wellness. The following sections detail the key aspects of developing such a drink (Zarubin *et al.* 2024). While the combination of soy isolate and collagen hydrolysate offers numerous health benefits, challenges such as flavour masking and maintaining beverage stability must be addressed. Using flavour enhancers and stabilizers can improve consumer acceptance and product quality. Additionally, leveraging sustainable sources for collagen hydrolysate not only enhances the nutritional profile, but also contributes to environmental sustainability by utilizing fish processing waste (Kuprina *et al*. 2020). The improvement of the amino acid composition of the mixture of soy isolate and collagen hydrolysate is due to their complementarity. The collagen hydrolysate significantly enriches the mixture with structural amino acids-glycine, proline, and hydroxyproline—important for skin and connective tissue health (Li & Wu 2018). At the same time, the soy isolate compensates for the deficiency of essential amino acids such as leucine, isoleucine, lysine, and methionine, which are necessary for muscle protein synthesis. The combination of these two proteins allows for a balanced amino acid profile. This enhances the biological value and digestibility of the protein in the body. In particular, the addition of soy protein improves the content of sulphur-containing amino acids, compensating for their deficiency in collagen. Thus, the resulting mixture represents a functionally complete protein component. The combination of plant and animal proteins provided a balanced amino acid profile, making the drink promising for use in nutraceuticals, sports, and functional nutrition.

CONCLUSIONS

During the study, an optimal recipe for an instant functional drink was developed. The resulting product demonstrated high levels of solubility, organoleptic properties, and protein value, as well as compliance with safety requirements.

ACKNOWLEDGMENTS

The article was prepared under the scientific project, registration number № 0124PK00891, IRN AP22686505, titled "Development of technology for instant functional drinks." (contract № 176-ZhG5 24-26 dated June 20, 2024), granted by the Science Committee of the Ministry of Science and Higher Education of the Republic of Kazakhstan.

REFERENCES

Akharume, FU, Aluko, RE & Adedeji, AA 2021, Modification of plant proteins for improved functionality: A review. Comprehensive Reviews in Food Science and Food Safety, 20: 198-224.

Andreani, G, Sogari, G, Marti, A, Froldi, F, Dagevos, H & Martini, D 2023, Plant-based meat alternatives: Technological, nutritional, environmental, market, and social challenges and opportunities. Nutrients, 15: 452.

Capita, R, Prieto, M & Alonso-Calleja, C 2004, Sampling methods for microbiological analysis of red meat and poultry carcasses. Journal of Food Protection, 67: 1303–1308.

Carey, CN, Paquette, M, Sahye-Pudaruth, S, Dadvar, A, Dinh, D, Khodabandehlou, K, Liang, F, Mishra, E, Sidhu, M & Brown, R 2023, The environmental sustainability of plant-based dietary patterns: A scoping review. The Journal of Nutrition, 53: 857-869.

Delgado-Andrade, C, Navarro, M, Lopez, H & López, MC 2003, Determination of total arsenic levels by hydride generation atomic absorption spectrometry in foods from south-east Spain: Estimation of daily dietary intake. Food Additives and Contaminants, 20: 923-932.

Gerliani, N, Hammami, R & Aider, M 2019, Production of functional beverage by using protein-carbohydrate extract obtained from soybean meal by electro-activation. LWT, 113: 108259.

Gupta, A, Sanwal, N, Bareen, MA, Barua, S, Sharma, N, Joshua Olatunji, O, Prakash Nirmal, N & Sahu, JK 2023, Trends in functional beverages: Functional ingredients, processing technologies, stability, health benefits, and consumer perspective. Food Research International (Ottawa, Ont.), 170: 113046.

Gupta, A, Sanwal, N, Bareen, MA, Barua, S, Sharma, N, Olatunji, OJ, Nirmal, NP & Sahu, JK 2023, Trends in functional beverages: Functional ingredients, processing technologies, stability, health benefits, and consumer perspective. Food Research International, vol. 170: 113046.

Intrasook, J, Tsusaka, TW & Anal, AK 2024, Trends and current food safety regulations and policies for functional foods and beverages containing botanicals. Journal of Food and Drug Analysis, 32: 112-139.

Jarvas, G, Guttman, A, Miękus, N, Bączek, T, Jeong, S, Chung, DS, Pätoprstý, V, Masár, M, Hutta, M & Datinská, V 2020, Practical sample pretreatment techniques coupled with capillary electrophoresis for real samples in complex matrices. TrAC Trends in Analytical Chemistry, 121: 115702.

Kuprina, E, Yakkola, A, Kopylov, A, Zashikhin, M & Kuznetsova, A 2020, Development of functional product enriched with collagen hydrolysate from fish processing waste. E3S Web of Conferences, 6026.

Lee, HJ, Yong, HI, Kim, M, Choi, Y-S & Jo, C 2020, Status of meat alternatives and their potential role in the future meat market—A review. Asian-Australasian Journal of Animal Sciences, 33(10):1533.

Lee, J-S, Choi, I & Han, J 2022, Construction of rice protein-based meat analogues by extruding process: Effect of substitution of soy protein with rice protein on dynamic energy, appearance, physicochemical, and textural properties of meat analogues. Food Research International, 161: 111840.

Li, P & Wu, G 2018, Roles of dietary glycine, proline, and hydroxyproline in collagen synthesis and animal growth. Amino acids, 50: 29-38.

Liu, H, Xu, X, Cui, H, Xu, J, Yuan, Z, Liu, J, Li, C, Li, J & Zhu, D 2023, Plant-based fermented beverages and

key emerging processing technologies. Food Reviews International, 39: 5844-5863.
Lu, D, Roy, D, Acevedo-Fani, A, Singh, H & Ye, A 2025, Investigation of various plant protein ingredients for processed cheese analogues: physical properties and microstructure compared with milk proteins. International Journal of Food Science and Technology, 60: 018.
Mefleh, M, Pasqualone, A, Caponio, F, De Angelis, D, Natrella, G, Summo, C & Faccia, M 2022, Spreadable plant-based cheese analogue with dry-fractioned pea protein and inulin–olive oil emulsion-filled gel. Journal of the Science of Food and Agriculture, 102(12): 5478-5487.
Myers, RH, Montgomery, DC & Anderson-Cook, CM 2016, Response Surface Methodology: Process and Product Optimization Using Designed Experiments, John Wiley & Sons.
Nonglait, DL, Chukkan, SM, Arya, SS, Bhat, MS & Waghmare, R 2022, Emerging non-thermal technologies for enhanced quality and safety of fruit juices. International Journal of Food Science and Technology, 57: 6368–6377.
Puiggròs, F, Muguerza, B, Arola-Arnal, A, Aragonès, G, Suárez-Garcia, S, Bladé, C, Arola, L & Suárez, M 2017, Functional beverages. Innovative Technologies in Beverage Processing, pp. 275-296.
Sharma, Shagun, Singh, A, Sharma, Swati, Kant, A, Sevda, S, Taherzadeh, MJ & Garlapati, VK 2021, Functional foods as a formulation ingredient in beverages: technological advancements and constraints. Bioengineered, 12: 11055-11075.
Short, EC, Kinchla, AJ & Nolden, AA 2021, Plant-based cheeses: A systematic review of sensory evaluation studies and strategies to increase consumer acceptance. Foods, 10: 725.
da Silva, VT, Mateus, N, de Freitas, V & Fernandes, A 2024, Plant-Based Meat Analogues: Exploring Proteins, Fibers and Polyphenolic Compounds as Functional Ingredients for Future Food Solutions. Foods,13(14): 1-33.
Tang, J, Yao, D, Xia, S, Cheong, L & Tu, M 2024, Recent progress in plant-based proteins: From extraction and modification methods to applications in the food industry. Food Chemistry: X, 23: 101540.
Vogelsang-O'Dwyer, M, Zannini, E & Arendt, EK 2021, Production of pulse protein ingredients and their application in plant-based milk alternatives. Trends in Food Science & Technology, 110: 364-374.
Wagner, AO, Markt, R, Mutschlechner, M, Lackner, N, Prem, EM, Praeg, N & Illmer, P 2019, Medium preparation for the cultivation of microorganisms under strictly anaerobic/anoxic conditions. Journal of Visualized Experiments: JoVE, 150: 10-3791.
Zarubin, NY, Kharenko, EN, Bredikhina, OV, Lavrukhina, EV, Rysakova, KS, Novikov, VY, Leonov, GE, Vakhrushev, IV, Zolotarev, KV & Mikhailov, AN 2024, An isotonic drink containing pacific cod (Gadus macrocephalus) processing waste collagen hydrolysate for bone and cartilage health. Marine Drugs, 22: 202.
Zhang, T, Dou, W, Zhang, X, Zhao, Y, Zhang, Y, Jiang, L & Sui, X 2021, The development history and recent updates on soy protein-based meat alternatives. Trends in Food Science & Technology, 109: 702–710.