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Development and optimisation of plant-based protein–vitamin concentrates from pea, soybean, and oat for potential sports nutrition applications

Massimzhan Velyamov, Zhumatay Urazbayev, Shukhrat Velyamov, Gulnur Srash, Turar Bakytzhan, Dulat Zhalelov, Madina Sadykova, Yelena Solodova

ABSTRACT

The modern sports nutrition industry requires affordable plant-based protein sources with high nutritional potential that can partially replace animal-derived proteins and reduce reliance on imports. Legume grains, as valuable sources of protein and essential amino acids, have considerable potential for developing functional food products. This study aimed to develop and optimise formulations and processing technology for protein–vitamin concentrates (PVCs) derived from plant raw materials-peas, soybeans, and oats-enriched with natural vitamin sources (rosehip and lemon) to expand the availability of affordable sports nutrition products in the Republic of Kazakhstan. The protein base was obtained from regionally adapted cultivars: peas (*Aksary*), soybeans (*Zhansaya*), and oats (*Syrgalym*). The technological process included raw material preparation, aqueous extraction, and enzymatic hydrolysis using pancreatin (3%, pH 8.0–8.2, 43–45 °C), followed by enrichment with lemon extract (10%) and rosehip extract (20%), stabilisation with apple pectin (1%), and drying by either lyophilisation (–50 °C) or convective drying (50 °C). The resulting samples were evaluated for physicochemical, biochemical, sensory, and safety parameters using standardised analytical methods harmonised with internationally recognised ISO and AOAC standards. Optimisation of the formulations increased the content of amino acids, vitamins, and minerals. The obtained PVCs contained 18.79–26.75% protein, L-arginine (1.795–3.631 g/100 g), lysine (1.108–1.222 g/100 g), vitamin C (110.8–125.15 mg/100 g), β-carotene (0.056–0.067 mg/100 g), B-group vitamins, magnesium (119.08–146.57 mg/100 g), and zinc (3.34–3.63 mg/100 g). All samples complied with regulatory requirements for physicochemical, biochemical, sensory, and safety indicators. Convective drying preserved key nutritional and quality parameters at levels comparable to lyophilisation while offering advantages in process efficiency. The developed PVCs are characterised by high protein content and a favourable micronutrient profile and could be further investigated as potential ingredients for the development of functional sports nutrition products. Their implementation could contribute to the development of domestic production and reduced dependence on imported sports nutrition products.

Keywords: protein–vitamin concentrates, enzymatic hydrolysis, plant-based proteins, sports nutrition, functional foods

INTRODUCTION

Modern sports nutrition is an important area of nutritional science that provides athletes with essential macro- and micronutrients to maintain physical performance, accelerate recovery, and support adaptation to intensive physical loads **[1], [2], [3], [4].** Under high training intensity, the demand for proteins, vitamins, amino acids, and antioxidants-key components for muscle tissue regeneration and immune support-significantly increases **[5]**, **[6]**.

Proteins are fundamental components of the athlete's diet, performing both structural and regulatory functions. They supply indispensable amino acids and help restore biochemical homeostasis after physical exertion **[7]**, **[8]**, **[9]**. In line with global trends toward sustainable development and environmental safety, interest in plant-based protein sources as alternatives to animal proteins is steadily increasing **[10]**.

Technologies for obtaining protein ingredients from plant raw materials-primarily legumes (peas, soybeans, beans, lentils) and cereals (oats, wheat, corn)-are of considerable scientific and practical importance, and their application is expected to expand in the coming years **[11]**, **[12]**, **[13]**, **[14]**. The growing consumption of plant-based products is driven both by the expansion of vegetarian and vegan diets and by the development of the functional and specialised food supplement market, including sports nutrition **[15]**.

A key limitation of plant proteins is their relatively low digestibility and, in some cases, an unbalanced amino acid composition. Enzymatic hydrolysis is an effective approach for improving their biological value, enabling the production of readily digestible low-molecular-weight polypeptides, peptides, and amino acids through protein macromolecule cleavage **[16]**, **[17]**.

Pea protein, used alone or in combination with other crops, has been identified as a promising raw material for functional protein ingredients **[16]**, **[18]**, **[19]**, **[20]**, **[21]**. Previous studies have shown that pea-derived hydrolysates possess favourable organoleptic properties, a balanced amino acid profile, and high bioavailability **[22]**, **[23]**, **[24]**. These hydrolysates are enriched with branched-chain amino acids (BCAAs), arginine, and lysine-compounds important for muscle protein synthesis and post-exercise recovery **[25]**, **[26]**, **[27]**. L-arginine has also been associated with metabolic regulation and vascular function, which supports its relevance in sports nutrition formulations **[25]**, **[28]**.

Considering these factors, pea protein can be regarded as a valuable source of amino acids and a promising raw material for the production of protein–vitamin concentrates for sports nutrition **[29]**, **[30]**, **[31]**.

The development of plant-based products is particularly relevant for Kazakhstan due to the high dependence on imported sports nutrition and the growing demand for affordable, environmentally safe, and functionally enriched products. Recent studies (2024–2025) have confirmed the high nutritional and biological value of proteins from regionally adapted pea (*Aksary*), soybean (*Zhansaya*), and oat (*Syrgalym*) cultivars, as well as vitamin-rich sources such as lemon and rosehip fruits **[32]**, **[33]**. These components demonstrate strong potential to develop new formulations of protein–vitamin concentrates and to expand the range of functional sports nutrition products.

Scientific Hypothesis

The study is based on the hypothesis that the enzymatic hydrolysis of plant proteins (pea, oat, and soybean), combined with enrichment using natural extracts (lemon and rosehip), will produce PVCs with a standardized amino acid profile and enhanced micronutrient content. It is further hypothesized that these concentrates will comply with established safety standards and exhibit favourable sensory characteristics compared to non-hydrolysed mixtures.

Furthermore, it is assumed that the optimisation of processing parameters, specifically the drying method, will ensure the stability and nutritional quality of the PVCs, making them suitable as potential ingredients for sports nutrition formulations.

Objectives

The primary objective of this study was to develop and optimise formulations and processing technology for protein–vitamin concentrates derived from plant raw materials (pea, soybean, and oat) enriched with natural vitamin sources (lemon and rosehip) for potential use in sports nutrition.

To achieve this objective, the following tasks were addressed:

1. To evaluate the feasibility of using locally sourced plant raw materials for the development of functional protein formulations.
2. To optimise key technological parameters, including enzymatic hydrolysis and drying methods (lyophilisation and convective drying).
3. To characterise the physicochemical and biochemical properties of the developed protein–vitamin concentrates.
4. To assess nutritional composition and safety indicators in accordance with regulatory standards.
5. To evaluate sensory properties of the developed products.

MATERIALS AND METHODS

The research was conducted at LLP "Kazakh Research Institute of Processing and Food Industry" of the Ministry of Agriculture of the Republic of Kazakhstan, in the laboratory of "Biotechnology, Quality, and Food Safety"

Samples

Sample description: Regionally adapted plant cultivars were used as the protein base: pea (*Aksary*), oat (*Syrgalym*), and soybean (*Zhansaya*). Vitamin components were represented by commercially available rosehip and lemon fruits characterised by high biological value. All raw materials were obtained in dried, whole form prior to processing.

Sample collection: Plant raw materials were handled in accordance with applicable regulatory requirements. Pea, soybean, and oat grains were stored in dry conditions at ambient temperature (18 ± 2 °C) in hermetically sealed containers. Rosehip and lemon fruits were stored under refrigerated conditions at 4 ± 1 °C until further processing.

Sample preparation: Prior to analysis, raw materials underwent standard technological preparation, including sorting, washing, drying, and milling. The samples were washed with purified water and dried to a residual moisture content of 8–10%. Dried materials were milled to obtain a homogeneous powder with a particle size of approximately 0.8 mm, suitable for subsequent analyses. Aliquots of 10 ± 1 g of the milled material were taken for laboratory investigations (see Figure 1).

Figure 1 Regionally adapted plant raw materials used in the study and their milled forms: a) pea (*Aksary cultivar*); b) soybean (*Zhansaya cultivar*); c) oat (*Syrgalym cultivar*); d) dried rosehip fruits (*commercial cultivar*); e) dried lemon slices (*commercial cultivar*).

Number of samples analysed: The total number of samples (n = 42) was derived from a factorial design: 2 main formulations (F1: Pea+Oat, F2: Pea+Soy) × 2 drying methods (Convective, Lyophilization) × 3 independent experimental batches, with additional samples for intermediate optimization steps. To ensure reliability, each independent experiment was performed in triplicate (experimental replicates), and each laboratory measurement for these samples was conducted in three parallel runs (analytical replicates).

Chemicals

The following reagents and materials were used: pancreatin enzyme preparation (activity 75 USP U/mg; Pancreatin 3X, 3 NF/USP, RM7348, HiMedia, India); apple pectin (Sigma-Aldrich, USA); sodium hydroxide solution (10.0 N, JSC Kaustik, Russia); aqueous lemon extract (10%) and rosehip extract (20%) prepared from pre-dried and milled plant materials (particle size ~0.8 mm); and the LAA-21 calibration kit for amino acid analysis by capillary electrophoresis (Lumex Instruments, Russia).

All reagents were of analytical grade and obtained from certified suppliers. Solutions were prepared using distilled water and stored in accordance with laboratory quality control procedures.

Animals, Plants and Biological Materials

Plant protein sources included regionally adapted cultivars of pea (*Pisum sativum* L., *Aksary*), soybean (*Glycine max* L., *Zhansaya*), and oat (*Avena sativa* L., *Syrgalym*). Vitamin sources were rosehip (Rosa canina L.) and lemon (Citrus limon L.) fruits, obtained from commercial suppliers. All plant materials were provided by the Kazakh Research Institute of Agriculture and Plant Growing LLP, Almaty Region, Republic of Kazakhstan.

Instruments

The following instruments were used in this study: pH meter PB-11 (Sartorius, Germany); bench-top centrifuge SM-12 (Tagler, Russia); rotary evaporator XD-52AA (China); capillary electrophoresis system Kapel-105M (Lumex, Russia); laboratory thermostat TS-1/20 SPU (Russia); laboratory shaker PE-6500 (Russia); refractometer IRF-454 B2M (Russia); drying oven ShS-80-01 SPU (Russia); freeze dryer EV-DF10A (Russia); convective dryer Kitfort KT-1910 (Germany); electric cooking boiler KPEM-100-OM2 (ChAZ, Russia); laboratory water bath series PE-4310–4313 (Russia); spectrophotometers PE-5300VI and PE-5300UV (Russia); analytical balances VK-150.1, VK-300.1, and VK-600 (Russia); and photoelectric photometer KFK-3 (Russia).

Laboratory Methods

The quality of plant raw materials (pea, soybean, oat, rosehip, and lemon) was evaluated using standardised analytical methods. Total protein content was determined according to GOST 13496.4-2019 **[34]** and GOST R 51417-99 **[35]**, while fat content was measured according to GOST 13496.15-2016 **[36]**. Carbohydrate content was determined using GOST 8756.13-87 **[37]**, and moisture and ash contents were measured according to GOST 13496.3-92 **[38]** and GOST R 51418-99 **[39]**, respectively. Energy value was calculated using the Skurikhin method **[40]**.

Macro- and microelements (Mg, Ca, Fe, Zn) were analysed according to GOST 32343-2013 **[41]**, with zinc additionally determined using GOST 30692-2000 **[42]**. Soluble solids and titratable acidity were determined according to GOST R 51433-99 **[43]** and GOST 25555.0-82 **[44]**, respectively. Fibre and starch contents were determined according to GOST 13496.2-91 **[45]** and GOST 26176-2019 **[46]**.

β-Carotene (vitamin A) content was determined according to GOST 13496.17-2019 **[47]**, and B-group vitamins and vitamin C were analysed according to GOST 31483-2012 **[48]**. Water-soluble antioxidant levels were determined according to GOST R 54037-2010 **[49]**, and nitrate content was measured according to GOST 13496.19-2015 **[50]**.

Toxic elements were determined as follows: Pb and Cd according to GOST 30692-2000 **[51]**, As according to GOST 26930-86 **[52]**, and Hg according to GOST 26927-86 **[53]**. Mycotoxins (aflatoxin B1) were analysed according to GOST 30711-2001 **[54]**, and organochlorine pesticides (HCH isomers and DDT with metabolites) were determined according to ST RK 2011-2010 **[55]**.

The analytical procedures for protein (Kjeldahl method), ash, and moisture content were conducted in accordance with AOAC Official Methods 2001.11 and ISO 20483:2013. The comparative analysis demonstrated a deviation of less than 1.5% from standardized reference materials, validating the equivalence of the laboratory protocols used.

All analyses were performed in triplicate using validated methodologies and calibrated laboratory equipment.

Sensory evaluation was conducted in accordance with ISO 8586 standards. Evaluation was performed at 22 ± 1 °C using a five-point hedonic scale. Data were analyzed using the Friedman test to determine significant differences between formulation scores.

Compliance with International Standards. Although some analytical procedures were originally conducted according to GOST standards, the applied methodologies are equivalent to internationally recognised ISO and AOAC methods. Where applicable, ISO-equivalent analytical principles were followed, ensuring compatibility with international food analysis practices and reproducibility across laboratories.

Description of the Experiment

Study flow: The study involved the stepwise development of a technology for producing PVCs from locally sourced plant raw materials: pea (*Aksary*), soybean (*Zhansaya*), and oat (*Syrgalym*), enriched with aqueous extracts of lemon and rosehip. Raw materials were washed and dried at 60 ± 1 °C to a residual moisture content of approximately 10%, then milled to a particle size of ~0.8 mm. Two formulations were prepared: formulation 1 (20% pea + 5% oat) and formulation 2 (20% pea + 5% soybean). Each mixture was subjected to aqueous extraction at 43 ± 1 °C for 30 min, followed by enzymatic hydrolysis with pancreatin (3%) at pH 8.0–8.2 and 43–45 °C for 3–4 h. After hydrolysis, apple pectin (1% of the hydrolysate mass) was added as a stabiliser. The mixture was homogenised, and the enzyme was thermally inactivated, followed by pasteurisation at 85 ± 2 °C for 30 min. The optimisation of the PVCs formulations and processing parameters was based on pre-defined criteria: ensuring a balanced essential amino acid profile and preserving maximum Vitamin C retention. The hydrolysates were enriched with aqueous vitamin extracts of lemon and rosehip (5% each, v/v), previously prepared by extraction at 60 ± 5 °C for 60 min (10% and 20% extracts, respectively). The mixtures were homogenised prior to drying. Drying was performed using two methods: freeze-drying at −50 °C for 24 ± 2 h and convective drying at 50 °C for 24 ± 2 h. The dried concentrates were milled, packaged, and stored at 10 ± 2 °C until further analysis. Freeze-drying was utilized as a reference (control) method to establish the maximum possible nutrient retention. Subsequent detailed analyses focused primarily on convective drying due to its greater feasibility for industrial-

scale production and cost-effectiveness in the Republic of Kazakhstan, provided that quality parameters remained comparable to the lyophilised samples.

Quality Assurance

Number of repeated analyses: Each analysis was performed in triplicate.

Number of experiment replication: Each experiment was conducted three times.

Reference materials: To ensure analytical accuracy and reproducibility, certified reference materials and secondary standards were used where applicable. The Kjeldahl method for total nitrogen determination was validated using certified protein standards **[34]**. Amino nitrogen content was determined according to GOST 29311-92 **[56]**, and sterility testing was performed according to GOST ISO 11737-2-2011 **[57]**. Amino acid analysis was performed using a Kapel-105M capillary electrophoresis system (Lumex, Russia) calibrated with the LAA-21 amino acid standard kit (Sigma-Aldrich, USA). All reagents were of analytical grade.

Calibration: All instruments were calibrated according to the manufacturers' specifications prior to analysis. The pH meter (PB-11, Sartorius) was calibrated using standard buffer solutions. The capillary electrophoresis system was calibrated using certified amino acid standards to ensure analytical accuracy.

Laboratory accreditation: Experimental work, including sample preparation and analysis, was performed at the Kazakh Research Institute of Processing and Food Industry LLP and in the accredited testing laboratory of Almaty Technological University JSC. Both laboratories operate in compliance with ISO/IEC 17025 standards.

Data Access

Primary data supporting the findings of this study are available from the corresponding author upon reasonable request, subject to institutional regulations.

Statistical Analysis

All experiments were conducted in triplicate, and results are presented as mean ± standard deviation. Statistical analysis was performed using IBM SPSS Statistics 26.0 (IBM Corp., USA). Data normality was assessed using the Shapiro–Wilk test. Differences between experimental groups were evaluated using one-way analysis of variance (ANOVA) followed by Tukey's post hoc test. Statistical significance was accepted at $p < 0.05$ **[58]**. Variance homogeneity was verified using Levene's test.

Reporting and transparency statement

The authors confirm the transparency of the methodology and the reliability of the presented results:

Study design: This research is an experimental laboratory study focused on the development and optimization of production technologies for PVCs.

Randomization and blinding: Due to the specific technical nature of food biotechnology processes, randomization and blinding (masking) methods were not applied.

Sample size determination: The sample size and the total number of analyzed samples ($n = 42$) were determined based on preliminary physicochemical and organoleptic assessments, as well as results from previous research stages.

Replication: To ensure accuracy and reproducibility, each technological experiment was conducted in triplicate, and all laboratory analyses were performed in three parallel measurements.

Data exclusion: All data obtained during the experiments were included in the analysis; no samples or results were excluded.

Data availability: The raw data supporting the findings of this study are available from the corresponding author upon reasonable request.

Compliance with standards: Experimental work was carried out in the accredited laboratories of the Kazakh Research Institute of Processing and Food Industry and Almaty Technological University, operating in accordance with ISO/IEC 17025 standards. Analytical methods were harmonized with international ISO and AOAC standards.

RESULTS AND DISCUSSION

Based on previous studies on the development of PVCs from plant raw materials for sports nutrition, four experimental formulations were initially proposed. However, preliminary physicochemical and sensory evaluations identified two formulations with superior nutritional and organoleptic characteristics, which were selected for further investigation.

In the present study, additional optimisation of the technological regime was performed to improve the amino acid profile of the resulting PVCs . The modification strategy focused on complementing pea protein with soybean and oat components in order to enhance the content of essential amino acids and improve overall protein quality.

The obtained results are consistent with previously reported studies on plant protein hydrolysates, which have been reported to be associated with improved digestibility and enhanced amino acid bioavailability; however, these effects were not directly assessed in the present study **[9]**, **[18]**, **[22]**. Similar increases in free amino acids

and peptides after hydrolysis of legume proteins have been reported in recent studies using pancreatin and other proteolytic enzymes, supporting the effectiveness of the applied approach.

The amino acid composition observed in this study, particularly the elevated levels of arginine and lysine, aligns with findings reported for pea- and soybean-based protein ingredients used in sports nutrition formulations **[20]**, **[25]**. These amino acids are known to contribute to protein metabolism and post-exercise recovery, confirming the relevance of the developed concentrates for specialised nutrition.

The comparison of drying methods demonstrated that convective drying preserved key nutritional characteristics at levels comparable to freeze-drying. Similar observations have been reported in recent studies evaluating the effects of drying techniques on protein powders and bioactive compounds, where moderate-temperature convective drying provided acceptable retention of nutritional quality while improving technological feasibility **[19]**.

The regionally adapted pea cultivar *Aksary* is characterised by a relatively low methionine content (0.290 ± 0.023 g/100 g). In contrast, literature data indicate higher methionine levels in soybean (0.547 ± 0.030 g/100 g) and oat grains (0.350 ± 0.035 g/100 g) **[59]**, **[60]**, **[61]**. **[62]**. These differences support the rationale for combining legumes and cereals to achieve a more balanced amino acid composition, which is consistent with previously reported protein complementation strategies in plant-based nutrition.

Based on this approach, *Aksary* pea was used as the primary protein source, while soybean and oat grains recommended for cultivation in Kazakhstan were incorporated as complementary components. This formulation strategy aimed to enhance the biological value of the developed concentrates while maintaining compatibility with locally available raw materials. Natural extracts of lemon and rosehip were included as sources of vitamins and bioactive compounds.

To identify the most suitable soybean and oat cultivars, a comparative analysis of total protein content was performed. The results are presented in Table 1.

Table 1 Total protein content in selected regionally adapted soybean and oat cultivars.

No.	Plant species	Cultivar	Total protein content, % (mean ± SD)
1	Soybean	*Aisaule*	36.29 ± 0.10^{a}
2	Soybean	*Zhansaya*	38.25 ± 0.29^{b}
3	Oat	*Alaman*	16.16 ± 0.04^{c}
4	Oat	*Syrgalym*	16.65 ± 0.08^{d}

Note: Values are expressed as mean ± standard deviation (n = 3). Different superscript letters (a, b, c, d) within the column indicate statistically significant differences between the cultivars ($p < 0.05$) according to Tukey's HSD test.

The analysis of the raw materials revealed significant variations in protein content among the studied cultivars (Table 1). Among the soybean samples, the *Zhansaya* cultivar exhibited a significantly higher protein concentration (38.25 ± 0.29%) compared to the *Aisaule* cultivar (36.29 ± 0.10%) ($p < 0.05$). Similarly, for the oat cultivars, a statistically significant difference was observed: *Syrgalym* contained 16.65 ± 0.08% of total protein, surpassing the *Alaman* cultivar (16.16 ± 0.04%) ($p < 0.05$). These results, confirmed by ANOVA and Tukey's HSD test, justify the selection of *Zhansaya* and *Syrgalym* as the primary components for the developed PVCs.

For formulation optimisation and cost reduction, the proportion of complementary components was standardised at 5% of oat flour (*Syrgalym*) for formulation 1 and 5% of soybean flour (*Zhansaya*) for formulation 2. Pea flour (*Aksary cultivar*) served as the primary component in both formulations (20%). Each formulation additionally contained lemon extract (10%) and rosehip extract (20%) at 5% of the hydrolysate volume, while apple pectin (1% w/w of the hydrolysate) was used as a stabiliser **[63]**.

Based on the optimised parameters, pilot-scale production of PVCs was carried out under semi-industrial conditions. Each formulation was processed in batches of 25 ± 1 L using an automated cooking boiler (KPEM-100-OM2, 100 L capacity) with controlled temperature and continuous mixing. Freeze-drying was performed using an EV-DF10A unit at −50 °C for 18–20 h under a residual pressure of 1 ± 0.5 Pa, while convective drying was carried out in a Kitfort KT-1910 dehydrator at 50 °C for 18–20 h.

During pilot-scale processing, 800 ± 10 mL of each batch were subjected to freeze-drying, while the remaining volume (approximately 24.2 ± 0.1 L) was processed by convective drying. The yield of dry product after freeze-drying was 198 ± 1 g for formulation 1 and 201 ± 1 g for formulation 2, corresponding to approximately 25% recovery. Convective drying produced 5.8 ± 0.1 kg and 5.9 ± 0.1 kg of dry product for formulations 1 and 2, respectively (approximately 24% yield).

The obtained dried PVCs samples were subsequently analysed for their key physicochemical and biochemical characteristics. Detailed results are presented in Table 2.

Table 2 Biochemical and physicochemical parameters of dried protein–vitamin concentrate samples based on pea, soybean, and oat, enriched with lemon and rosehip extracts after drying.

Parameters	Formula 1 (pea+oat) Freeze drying	Formula 1 (pea+oat) Convective drying	Formula 2 (pea+soy) Convective drying
Protein content, %	26.75 ± 0.29^{a}	24.62 ± 0.20^{b}	18.79 ± 0.16^{c}
Fat content, %	n.d.	0.97 ± 0.005^{a}	2.66 ± 0.01^{b}
Carbohydrate content, %	n.d.	62.50 ± 0.75^{a}	53.75 ± 0.63^{b}
Moisture content, %	n.d.	6.64 ± 0.05^{a}	5.07 ± 0.02^{b}
Ash content, %	n.d.	2.72 ± 0.01^{a}	2.45 ± 0.01^{b}
Dry matter content, %	n.d.	93.35 ± 1.01^{a}	94.92 ± 1.05^{b}
Fiber content, %	n.d.	14.83 ± 0.11^{a}	16.41 ± 0.13^{b}
Starch content, %	n.d.	46.44 ± 0.51^{a}	34.30 ± 0.40^{b}
Energy value, kcal	n.d.	341.58^{a}	300.66^{b}
Titratable acidity, °T	n.d.	29^{a}	30.5^{b}
Nitrate content, mg/kg	n.d.	66.09^{a}	82.53^{b}
Water-soluble antioxidants, mg/g	n.d.	4.70 ± 0.0710	1.74 ± 0.0382^{b}
β-carotene content, mg/100 g	0.061 ± 0.0008^{a}	0.056 ± 0.0006^{a}	0.067 ± 0.001^{b}
B vitamins content, mg/100 g			
B1	0.801 ± 0.160^{a}	0.784 ± 0.157^{a}	0.820 ± 0.164^{b}
B2	0.169 ± 0.071^{a}	0.156 ± 0.065^{a}	0.191 ± 0.080^{b}
B3	1.98 ± 0.356^{a}	1.89 ± 0.340^{a}	1.95 ± 0.351^{b}
B5	1.66 ± 0.33^{a}	1.54 ± 0.31^{a}	1.63 ± 0.326^{b}
B6	0.24 ± 0.048^{a}	0.18 ± 0.036^{a}	0.27 ± 0.054^{b}
Bc	0.069 ± 0.014^{a}	0.062 ± 0.012^{a}	0.070 ± 0.014^{b}
Vitamin C, mg/100 g	125.15 ± 15.10^{a}	112.25 ± 12.80^{a}	110.80 ± 11.75^{a}
Magnesium, mg/100 g	-	119.08 ± 1.31^{a}	146.57 ± 1.07^{b}
Zinc, mg/100 g	-	3.34 ± 0.03^{a}	3.63 ± 0.05^{b}
Amino nitrogen, mg/g	19.46 ± 0.15^{a}	18.76 ± 0.22^{b}	18.2 ± 0.11^{c}

Note: Values are presented as mean ± SD (n = 3). Different superscript letters (a, b, c) within a row indicate statistically significant differences ($p < 0.05$) determined by Tukey's HSD test. "n.d." - not determined (parameters for freeze-dried samples were analyzed focusing on the preservation of labile bioactive compounds).

Comparative analysis of drying methods for Formula 1 showed that freeze-drying significantly preserved more protein (26.75%) compared to convective drying (24.62%) ($p < 0.05$). However, for vitamin C and B-group vitamins, the differences between drying methods were not statistically significant ($p > 0.05$), which indicates the efficiency of the chosen convective temperature (50 °C).

Formula 1 (pea+oat) showed a significantly higher antioxidant activity (4.70 mg/g) compared to Formula 2 (pea+soy) (1.74 mg/g) ($p < 0.05$), likely due to the synergistic effect of oat polyphenols and rosehip extract.

Based on the data presented in Table 2, both freeze-drying and convective drying preserved the nutritional and biochemical quality of the developed concentrates. Minor variations in protein, vitamin C, β-carotene, and amino nitrogen contents were observed between the drying methods; however, these differences remained within a comparable range and did not substantially affect the overall nutritional profile.

From a technological perspective, convective drying demonstrated comparable compositional characteristics while offering advantages in process simplicity and scalability. Therefore, convectively dried samples were selected for further investigation.

The analytical results obtained for the pilot-scale batches indicate that the developed PVCs possess balanced biochemical and physicochemical properties, including protein, lipid, carbohydrate, antioxidant, vitamin, and mineral components, as well as favorable fiber content and energy value. These characteristics are consistent with regulatory and literature-reported criteria for functional sports nutrition products **[64]**, **[65]**.

The developed concentrates also demonstrated a diverse amino acid composition (Table 3), confirming their potential as plant-based protein ingredients for specialised nutrition applications.

Table 3 Essential and sports-relevant amino acids (g/100 g product, mean ± SD).

No.	Amino acid	Formula 1 (pea+oat) (g/100 g)	Formula 2 (pea+soy) (g/100 g)	FAO/WHO requirement (mg/kg/day)
1	Lysine	1.108 ± 0.377 [a]	1.222 ± 0.415 [b]	30
2	Phenylalanine	1.785 ± 0.535 [a]	1.145 ± 0.344 [b]	25 (with tyrosine)
3	Tyrosine	3.015 ± 0.905 [a]	2.062 ± 0.619 [b]	25 (total)
4	Histidine	0.517 ± 0.258 [a]	0.126 ± 0.063 [b]	10
5	Leucine	2.277 ± 0.592 [a]	1.909 ± 0.496 [b]	39
6	Isoleucine	(included)	(included)	20
7	Valine	2.092 ± 0.837 [a]	1.145 ± 0.458 [b]	26
8	Methionine	0.738 ± 0.251 [a]	0.107 ± 0.036 [b]	15 (with cysteine)
9	Threonine	2.277 ± 0.911 [a]	1.985 ± 0.794 [b]	15
10	Arginine	3.631 ± 1.452 [a]	1.795 ± 0.718 [b]	~50–60 (conditionally essential)

Note: Values are expressed as mean ± SD (n = 3). Different superscript letters (a, b) within a row indicate statistically significant differences between Formula 1 and Formula 2 ($p < 0.05$) according to Tukey's HSD test. * Reference pattern for adults (FAO/WHO/UNU, 2007). ** Total for Phenylalanine + Tyrosine. *** Total for Methionine + Cysteine.

The analysis of the amino acid composition of the developed formulations demonstrates their compliance with current requirements for protein ingredients in sports nutrition in terms of biological value and functional relevance.

According to the recommendations of the Food and Agriculture Organization and the World Health Organization, both formulations contain key essential amino acids, including lysine, threonine, valine, leucine, isoleucine, phenylalanine (in combination with tyrosine), and histidine. This indicates a well-balanced amino acid profile approaching the reference pattern and confirms their suitability as sources of complete plant-based protein.

In sports nutrition, particular importance is attributed to the content of branched-chain amino acids (BCAAs: leucine, isoleucine, and valine), which play a central role in regulating muscle protein synthesis via activation of the mTOR signaling pathway. Formulation 1 exhibited higher levels of these amino acids, indicating its advantage in terms of post-exercise recovery and maintenance of muscle mass.

The analysis of amino acid composition further revealed elevated levels of L-arginine (1.795–3.631 g/100 g) and lysine (1.108–1.222 g/100 g) in the developed PVCs. These amino acids are widely recognised as important components of plant protein formulations and have been associated with protein metabolism and post-exercise recovery in the scientific literature **[66]**. L-Arginine and lysine are known to participate in metabolic pathways related to protein synthesis and physiological regulation, including vascular and immune functions **[67]**, **[68]**. Their presence, together with a balanced essential amino acid profile, reinforces the functional relevance of the developed concentrates for sports nutrition applications.

The lysine content, commonly a limiting amino acid in cereal-based products, was adequate in both formulations, reflecting the effective complementarity of plant raw materials and contributing to improved protein quality and bioavailability. At the same time, the content of sulfur-containing amino acids, particularly methionine, remains relatively low, which is typical for plant-based protein systems and may be considered a limiting factor affecting the overall amino acid score.

Comparative analysis indicates that formulation 1 has a more pronounced functional potential for sports nutrition applications, owing to its higher BCAAs and arginine content, whereas formulation 2 exhibits a comparable but less pronounced profile.

The developed PVCs therefore represent a viable dietary source for supplementing protein intake in physically active individuals. In practical applications, these concentrates can be consumed in reconstituted forms, such as dispersions in water or juice. The appropriate intake level should be determined individually, taking into account physiological needs, training intensity, and total daily macronutrient requirements, rather than adhering to a fixed dosage **[69]**.

Overall, the obtained results confirm that the developed concentrates can be considered promising plant-based ingredients for specialised sports nutrition, with further optimisation recommended to improve limiting amino acid balance and overall protein quality.

At the next stage, the nutritional value and safety parameters of the pilot-scale batches produced using both formulations were evaluated. The results are presented in Table 4.

Table 4 Safety indicators of convectively dried PVCs developed from plant raw materials.

N	Parameters	Regulatory permissible limits	Actual results obtained	
			Formulation 1 (pea+oat) convective drying	Formulation 2 (pea+soybean) convective drying
1	Lead, mg/kg	1.0	0.0085±0.0006	0.0093±0.0010
2	Cadmium, mg/kg	0.2	0.0006±0.00002	Not detected
3	Arsenic, mg/kg	1.0	0.0010±0.0002	0.0008±0.0001
4	Mercury, mg/kg	0.03	Not detected	Not detected
5	Potassium-40, Bq/kg	60	56±2	60±3
6	Cesium-137, Bq/kg	60	Not detected	Not detected
7	Pesticides:			
	HCH (α, β, γ -isomers), mg/kg	0.5	Not detected	Not detected
	DDT and its metabolites, mg/kg	0.05	Not detected	Not detected
8	Aflatoxin B1, mg/kg	0.005	Not detected	Not detected
9	MAFAnM, CFU/g (cm^3) ≤	$5x10^4$	$9x10^3$	$6x10^3$
10	Coliforms, in 1.0 g of product	Not permitted	Not detected	Not detected
11	E. coli, in 0.1 g	Not permitted	Not detected	Not detected
12	Pathogens incl. Salmonella, in 25g	Not permitted	Not detected	Not detected
13	Mould, CFU/g	10	1.0	1.0
14	Yeasts, CFU/g	10	Not detected	Not detected

Note: ± standard deviation.

As shown in Table 4, the developed PVCs demonstrated a favourable safety profile while maintaining balanced nutritional characteristics. Protein content ranged from 18.79 ± 0.16% to 24.62 ± 0.20%, amino nitrogen from 18.2 ± 0.11 mg/g to 18.76 ± 0.22 mg/g, fat from 0.97 ± 0.01% to 2.66 ± 0.01%, and carbohydrates from 53.75 ± 0.63% to 62.50 ± 0.75%. The calculated energy value ranged from 300.66 to 341.58 kcal, indicating a nutritionally dense product.

Safety evaluation showed that the analysed samples complied with relevant regulatory limits for heavy metals (Pb, Cd, As, Hg), pesticide residues, aflatoxin B1, and microbiological indicators. These results suggest that the developed concentrates meet general safety criteria for plant-based food products.

Sensory evaluation of pilot-scale batches was conducted using a five-point hedonic scale assessing appearance, texture, taste, colour, and aroma. The evaluation was performed by a trained panel (n = 12). Results are presented as mean ± standard deviation. Formulation 1 (pea + oat) received a score of 4.1 ± 0.3, while formulation 2 (pea + soybean) received 4.3 ± 0.2. Statistical analysis using the Friedman test indicated that the differences between formulations were not statistically significant ($p > 0.05$).

Overall, both formulations demonstrated favourable sensory acceptance and stable physicochemical characteristics. The combination of balanced composition and acceptable sensory properties supports the potential applicability of the developed protein–vitamin concentrates as plant-based ingredients for specialised nutrition.

The photograph of the final product is presented in Figure 4.

Figure 4 Final product of developed PVCs.

LIMITATIONS

While this study provides significant insights into the optimization of plant-based PVCs, several limitations should be considered when interpreting the results.

Absence of Bioavailability Data: A primary limitation is that this research did not involve direct in vivo or in vitro measurements of protein digestibility, bioavailability, or amino acid utilization. Although the nutritional potential was estimated based on biochemical profiles and the degree of enzymatic hydrolysis, the actual metabolic absorption and physiological efficiency of these concentrates remain to be empirically validated.

Scale of Production: The experimental work was conducted at a laboratory and pilot scale using specific regionally adapted cultivars. Consequently, the results regarding yield, consistency, and chemical stability may differ when transitioned to large-scale industrial production.

Lack of Performance Testing: The product is framed for sports nutrition applications; however, no functional performance-related properties or clinical trials involving athletes were conducted. Further research is required to evaluate the impact of these concentrates on muscle recovery, strength, and overall athletic performance in a controlled physiological environment.

Geographic Scope: The study focused on specific cultivars adapted to the climate of Kazakhstan. While these results are highly relevant for regional food security and import substitution, the findings may not be directly generalizable to plant varieties grown under different agro-climatic conditions.

CONCLUSIONS

This study developed and optimised formulations and processing technology for plant-based PVCs derived from pea, soybean, and oat raw materials enriched with natural vitamin sources (rosehip and lemon extracts). The developed concentrates contained 18.79–26.75% protein and demonstrated a balanced macronutrient composition with energy values ranging from 300.66 to 341.58 kcal. The products were characterised by a favourable micronutrient profile, including vitamin C (110.8–125.15 mg/100 g), β-carotene (0.056–0.067 mg/100 g), and essential minerals such as magnesium (119.08–146.57 mg/100 g) and zinc (3.34–3.63 mg/100 g). The amino acid profile demonstrated elevated levels of L-arginine (1.795–3.631 g/100 g) and lysine (1.108–1.222 g/100 g), supporting the compositional and nutritional potential of the developed concentrates. Convective drying at 50 °C preserved key nutritional parameters at levels comparable to freeze-drying while offering advantages in technological simplicity and scalability. Safety assessments confirmed compliance with regulatory limits for heavy metals, pesticides, mycotoxins, and microbiological indicators. Sensory evaluation showed favourable acceptance, with mean scores of 4.1 and 4.3 for the pea–oat and pea–soybean formulations, respectively. These findings indicate that the developed PVCs represent promising plant-based ingredients for specialised and sports nutrition applications. Future studies should include clinical validation, assessment of protein digestibility and bioavailability, storage stability analysis, and evaluation of functional performance outcomes.

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Contact Address:
Massimzhan Velyamov
Affiliation: LLP Kazakh Research Institute of Processing and Food Industry (Almaty branch),
Serkebaev Ave., 62, 050060, Almaty, Kazakhstan,
Tel.: + 7 701 381 88 81
E-mail: vmasim58@mail.ru
ORCID: https://orcid.org/0000-0002-9248-5951
Author contribution: data curation, project administration, conceptualisation, methodology.

Zhumatay Urazbayev
Affiliation: LLP Kazakh Research Institute of Processing and Food Industry (Astana head institute),
st. Akzhol, 26,010000, Astana, Kazakhstan,
Tel.: +7 701 544 3766
E-mail: zhz1964@mail.ru
ORCID: https://orcid.org/0000-0003-1898-0564
Author contribution: funding acquisition, formal analysis.

Shukhrat Velyamov
Affiliation: LLP Kazakh Research Institute of Processing and Food Industry (Almaty branch),
Serkebaev Ave., 62, 050060, Almaty, Kazakhstan,
Tel.: +7 707 210 51 11
E-mail: v_shukhrat@mail.ru
ORCID: https://orcid.org/0000-0002-5997-5182
Author contribution: data curation, writing - original draft, writing - review & editing.

Gulnur Srash
Affiliation: LLP Kazakh Research Institute of Processing and Food Industry (Almaty branch), Serkebaev Ave., 62, 050060, Almaty, Kazakhstan,
Tel.: +7 778 3723848
E-mail: srash.gulnur.99@mail.ru
ORCID: https://orcid.org/0009-0001-2730-4763
Author contribution: writing - original draft, visualisation.

Turar Bakytzhan
Affiliation: Kazakh national agrarian research university, Faculty of Engineering and Technology, Department of "Technology of Food Production and Food Safety", Abay str, 8, 050010, Almaty, Kazakhstan,
Tel.: +7 775 996 6070
E-mail: nurtuganuly.t@mail.ru
ORCID: https://orcid.org/0009-0002-9720-6189
Author contribution: writing - original draft, visualisation.

Dulat Zhalelov
Affiliation: Kazakh national agrarian research university, Faculty of Engineering and Technology, Department of "Technology of Food Production and Food Safety", Abay str, 8, 050010, Almaty, Kazakhstan,
Tel: +7777 188 3689
E-mail: zhalelov.dulat@kaznaru.edu.kz
ORCID: https://orcid.org/0000-0002-9688-2639

Author contribution: formal analysis, investigation, resources, data curation.

Madina Sadykova
Affiliation: LLP Kazakh Research Institute of Processing and Food Industry (Almaty branch),
Serkebaev Ave., 62, 050060, Almaty, Kazakhstan,
Tel.: + 7 747 438 48 49
E-mail: madina.sadukova0209@mail.ru
ORCID: https://orcid.org/0009-0008-9911-6973
Author contribution: data collection, analysis of scientific literature

Yelena Solodova
Affiliation: LLP International Engineering and Technological University, Al-Farabi avenue, 93G/5, 050060, Almaty, Kazakhstan,
Tel.: + 7 777 962 84 58
E-mail: elenaneft3@gmail.com
ORCID: https://orcid.org/0000-0003-0136-4220
Author contribution: data curation, methodology

Corresponding author: **Turar Bakytzhan**