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Vegan bean-based product enriched with microgreens: chemical, antioxidant, and sensory evaluation

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Abstract

Beans (*Phaseolus vulgaris* L.) are an excellent raw material for developing vegan and gluten-free products because of their high content of proteins, dietary fiber, and bioactive compounds. This study aimed to evaluate the chemical composition, antioxidant capacity, and sensory properties of a fermented Tetovac (var.) bean-based product enriched with different microgreens and spices. Notable differences in phytochemical profiles were observed depending on the type of microgreens used. The contents of total phenolics and carotenoids (fresh weight basis) were highest in the sample with radish microgreens and saffron (1.178 mg GAE/g and 47.56 µg/g, respectively), whereas the highest total flavonoid content was recorded in the sample with pea microgreens (5.243 mg RE/g). In contrast, soluble sugar contents did not differ significantly among the samples ($P > 0.05$). Phytate content decreased in most samples, compared to the control ($P < 0.05$). Analysis of the protein profile of the sample containing pea microgreens during different storage periods revealed an increase in total protein content, particularly in the fraction of small proteins and peptides. Flavor intensity, sourness, bitterness, and firmness increased gradually during storage. Sensory evaluation revealed that samples containing basil and chive microgreens achieved high overall acceptability and purchase intention, whereas samples with radish microgreens were less acceptable due to pronounced bitterness and sourness.

Keywords: carotenoids; CATA method; *Phaseolus vulgaris* L.; phytates; phytochemical composition; peptides

Introduction

Legumes (family *Fabaceae* or *Leguminosae*) represent one of the most economically important and diverse plant families. There are more than 20,000 legume species worldwide (Sandhu and Chaturvedi, 2025). Many of them, such as lentils, peas, broad beans, chickpeas, soybeans, beans (Lima, a common var.), and peanuts, play an important role in human diet (Nartea *et al.*, 2023; Sparvoli *et al.*, 2021). More than 4,000 bean varieties are cultivated globally (Dimopoulou *et al.*, 2024). Beans (*Phaseolus vulgaris* L.), are among the most widely consumed legumes, with annual production estimated at approximately 28.5 million tons (Food and Agriculture Organization [FAO], 2023), and this production continues to increase.

Beans are a source of high-quality nutrients whose proportion varies between varieties: carbohydrates (50–70%), fat (approximately 2%), and proteins/peptides (18–31% of dry weight) (Alfaro-Diaz *et al*., 2023; Dimopoulou *et al*., 2024; Meenu *et al*., 2023; Uebersax *et al*., 2023). Bean proteins contain most of the essential amino acids required for a balanced human diet (Añazco *et al*., 2023). Globulins are the principal storage proteins, with phaseolin being the most abundant one (30–53% of total proteins), followed by albumins, glutelins, and prolamins (Alfaro-Diaz *et al*., 2023; de Fatima Garcia *et al*., 2021). Other components include micronutrients, such as vitamins and nutritional elements (K, Ca, Mg, P, Cu, Fe, Mn, and Zn) (Dimopoulou *et al*., 2024). In addition, beans are a valuable source of secondary metabolites, among which anthocyanins, flavonols, phenolic acids, saponins, and carotenoids represent the major bioactive phytochemicals. Dietary fiber, proteins, peptides, and bioactive phytochemicals are considered particularly beneficial for human health (Dimopoulou *et al*., 2024; Meenu *et al*., 2023). Consequently, beans are regarded as an excellent source of plant-based proteins in gluten-free, vegetarian, and vegan diets (Sparvoli *et al*., 2021; Wesley *et al*., 2021).

The consumption of beans and bean-based foods have been extensively reviewed for potential health benefits, including antidiabetic, antioxidant, anti-inflammatory, anticancer, and anti-hypercholesterolemic effects as well as possible contributions to gut microbiota diversity and weight regulation (Alfaro-Diaz *et al.*, 2023; Dimopoulou *et al.*, 2024; Tarahi, 2024). Owing to their low allergenic potential, beans are often recommended as hypoallergenic food suitable for individuals with celiac disease and for those adopting gluten-free diets (Giuberti *et al.*, 2015; Sparvoli *et al.*, 2021). The biological potential of beans can be additionally improved by fermentation, thermal, proteolytic, or mechanical processing as well as by incorporating them into functional foods, such as bread, pasta, bars, and other nutritional formulations (Ali *et al.*, 2023; FAO, 2025; Nartea *et al.*, 2023; Sparvoli *et al*., 2021; Wesley *et al*., 2021). Fermentation has been used traditionally for food preservation; however, it can also improve nutritional value, reduce the content of anti-nutrients, and increase the concentration of bioactive compounds with antioxidant and probiotic properties (Dimidi *et al*., 2019). Moreover, an increasing number of consumers are choosing plant-based protein sources over animal-derived proteins, motivated not only by health considerations but also by environmental concerns, particularly emission of greenhouse gases (GHG) (Morgan *et al*., 2024).

Examination of bean proteins has been performed commonly following their isolation and separation. The most frequently applied method involves solubilization under alkaline conditions, followed by precipitation at the isoelectric point of proteins (pH 3.8–4.3). Different methods of protein extraction affect the yield of specific proteins as well as their functional properties (Ferreira *et al*., 2022). Thermal processing induces denaturation and fragmentation of proteins resulting in the formation of short-length peptides that are more easily absorbed in the intestines. Furthermore, high temperatures inactivate proteinase inhibitors and lectins present in beans, which are considered antinutritional factors (Alfaro-Diaz *et al*., 2023), contributing to fragmentation.

Owing to their multifunctional properties, beans serve as an ideal raw material for the development of innovative food products designed to enhance functional nutrition (Ali *et al*., 2023). Microgreens, characterized by a high concentration of bioactive compounds and antioxidants (González and Vargas, 2025), are used in the formulation of various functional products. Some microgreens have already been incorporated into functional foods, such as muffins with wheatgrass powder, cupcakes with beetroot and wheatgrass, cookies with wheatgrass powder, instant soup mix based on wheatgrass, and muffins with wheatgrass and mung-bean microgreen powder. In these applications, microgreens further enriched the products, improving their nutritional profile, functional benefits, and sensory characteristics (Rawat *et al*., 2024). However, only limited studies report functional products based on fermented beans (*P. vulgaris* L. var. Tetovac), particularly those enriched with microgreens.

Therefore, the aim of this study was to evaluate the nutritional and chemical composition, antioxidant and sensory properties, and consumer acceptability of a fermented bean-based product enriched with various types of microgreens. In addition, changes in protein and peptide content and the sensory profile of bean-based products during storage were examined.

Materials and Methods

All ingredients used to prepare products were obtained from local suppliers and distributors. Microgreens were dried in a food dehydrator (FDK24DW; Gorenje, China). Briefly, the samples were prepared using Tetovac white beans (*P. vulgaris* L.), pea protein (10%), potato starch (10%), coconut oil (15%), sunflower lecithin (2%), and salt (1%). For fermentation, yeast *Saccharomyces cerevisiae* (Danija Ltd., Serbia) was added. The samples were coated with dried microgreens (basil, pea, daikon radish, and chives). Red pepper (spicy sweet paprika) and saffron were added to two product samples (of the five different product samples used). The control bean-based sample was prepared without microgreens and/or spices. The composition and codes of product samples are given in Table 1.

Preparation of bean-based product samples

Bean seeds were washed and soaked in cold water for 6 h, after which they were strained and steamed in a food pasteurizer (ECG MHZ 270SD, The Czech Republic) for 120 min at 100°C. After cooking, the beans were crunched with a stick blender, cooled to a temperature of 25°C, and yeast *Saccharomyces cerevisiae* was added (1% relative to the mass of the cooked beans). Incubation lasted for 12 h at a constant temperature (25°C) in the same apparatus that was used for cooking. Spices were added to a part of the mass, which was homogenized and placed in a refrigerator (at 4°C for approximately 8 h) before making round cheese-like samples. Then, the samples were coated with dried microgreens. The appearance of the samples is presented in Figure 1.

Two sets of fermented bean-based product samples were prepared. The first set included samples with different microgreen coatings (Table 1), while the second set comprised samples coated with pea microgreens (BP), stored at 4°C for different periods over 30 days (Table 2).

Chemical analyses

Extraction procedures

For analyzing chemical parameters (total phenolic content [TPC], total flavonoid content [TFC], carotenoids

Table 1. Codes and composition of fermented bean-based product samples.

Sample code	Main component	Microgreens	Spice
BB	Beans^1 (B)	Basil^2 (B)	/
BP	Beans (B)	Pea^3 (P)	/
BR	Beans (B)	Daikon radish^4 (R)	/
BRpC	Beans (B)	Chives^5 (C)	Red pepper^6 (Rp)
BSR	Beans (B)	Daikon (Sango) radish^4 (R)	Saffron^7 (S)
Control	Beans (control)	/	/

Notes: ^{1}*Phaseolus vulgaris* L.; ^{2}*Ocimum basilicum* L.; ^{3}*Pisum sativum* L.; ^{4}*Raphanus sativus* var. *longipinnatus;* ^{5}*Allium schoenoprasum* L.; ^{6}*Capsicum annuum* var. *acuminatum*; and ^{7}*Crocus sativus* L.
BB: beans/basil; BP: beans/pea; BR: beans/radish; BRpC: beans/red pepper/chives; and BSR: beans/saffron/radish.

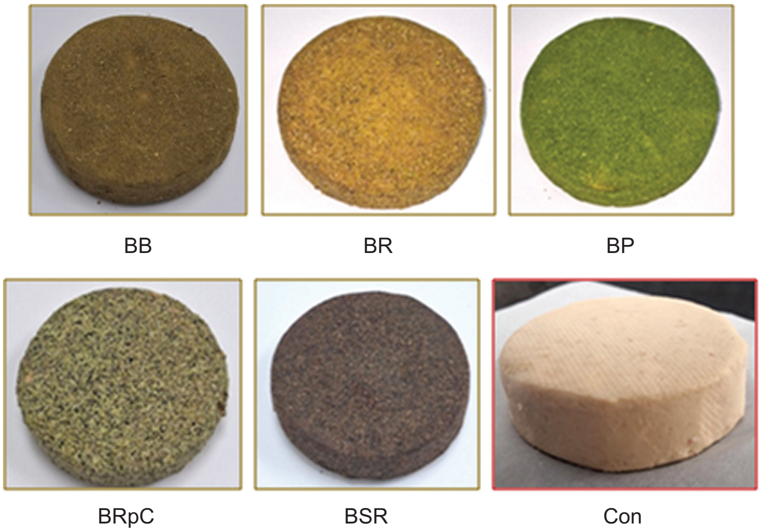

Figure 1. Fermented bean-based product enriched with different microgreens (BB: beans/basil; BP: beans/pea; BR: beans/radish; BRpC: beans/red pepper/chives; BSR: beans/saffron/radish) and a control sample (Con).

Table 2. Codes of fermented bean-based product with pea microgreens (BP) stored for different periods.

Sample code	Sample	Days of storage
BP1	Beans with pea microgreens (BP)	1
BP10		10
BP20		20
BP30		30

(Car), and soluble sugars (SS)) and antioxidant activity, the samples were extracted as follows: 2 g of each sample was extracted with 20 mL of 80% acetone using a mechanical shaker (MX-RD-E; DLAB, Beijing, China) for 3 h at room temperature.

Other analyses were performed as described below.

Proximate composition

The following methods were used to determine the nutritional composition of samples: nitrogen and crude protein (N × 6.25) content was determined according to International Organization for Standardization (ISO, 2013) ISO 20483:2013; total lipid content was measured by following the methods described by Folch *et al.* (1957) and Ljubobratović *et al.* (2021); moisture content was analyzed according to ISO 5534:2004 (ISO, 2004); ash content was determined according to the method described by Benton Jones (2001); and pH was measured as described by Barampama and Simard (1995). Total carbohydrates (excluding crude fiber) were calculated using the following equation (Shimelis and Rakshit, 2005):

Carbohydrates (%) = 100% − (protein [%] + lipids [%] + moisture [%] + ash [%]).

Total phenolic and total flavonoid content

Total phenolic content and TFC were determined using Folin–Ciocalteu's reagent and the aluminum chloride colorimetric assay, respectively (Kostić *et al.* 2021b).

For determining TPC (expressed as milligrams of gallic acid equivalent per gram of fresh sample weight [mg GAE/g FW]), 60 μL of previously prepared sample was mixed with 120 μL of Milli-Q water, 900 μL of Folin–Ciocalteu's reagent, and 900 μL of 7.5% sodium carbonate solution. The reaction mixture was vortexed and incubated in the dark for 90 min at room temperature.

For determining TFC (expressed as milligram of rutin equivalent per gram of fresh weight sample [mg RE/g FW]), 208 μL of the sample was mixed with 950 μL of Milli-Q water and 69 μL of 5% sodium nitrite solution. After 6 min, 125 μL of 10% aluminum chloride solution was added, and the mixture was incubated for 5 min in the dark. Subsequently, 415 μL of 1-M sodium hydroxide and 230 μL of distilled water were added and mixed. The absorbance was measured at 765 nm for TPC and 510 nm for TFC using an *ultraviolet–visible* (UV-VIS) spectrophotometer (UV-1800; Shimadzu USA Manufacturing Inc., USA).

Total carotenoid content (Car)

Total carotenoid content was determined spectrophotometrically according to the method described by Zeb (2015). The absorbance of the extract was measured at 453 nm using a UV-VIS spectrophotometer. Total carotenoid content (expressed as microgram per gram of fresh weight sample [μg/g FW]) was calculated using the following equation:

$$\text{Total carotenoids } (\mu g/g) = (A_{453} \times V \times 10^4) \div (A^{1\%}/_{1cm} \times m),$$

where A_{453} is the absorbance at 453 nm, V is the extract volume (mL), m is the sample mass (g), and $A^{1\%}/_{1cm}$ is the specific extinction coefficient (2592) of carotenoids in acetone (de Carvalho *et al.*, 2012).

Phytate content

Phytate content (Phy) was determined according to the method described by Yimer *et al.* (2023). Briefly, 0.2 g of the sample was extracted with 20 mL of 2.4% HCl using a mechanical shaker (MX-RD-E, DLAB, Beijing, China) for 1 h at room temperature. After extraction, the mixture was centrifuged at 3000 × g for 30 min. Subsequently, an aliquot of the supernatant (3 mL) was reacted with 1 mL of Wade's reagent, composed of 0.03% ferric chloride solution and 0.3% sulfosalicylic acid dissolved in water. The reaction mixture was briefly vortexed for 5 s, and the absorbance was measured at 500 nm using a UV–VIS spectrophotometer. Deionized water was used as a blank. The phytate content (expressed as milligram of sodium phytate per gram of fresh weight sample [mg NaPhy/g FW]) was quantified using a calibration curve prepared with sodium phytate.

Soluble sugars content

Soluble sugars (SS) were measured using standard phenol–sulfuric acid method as described by Dubois *et al.* (1951) with some modifications (Aluta *et al.*, 2023). Briefly, 65 μL of the extract was mixed with 135 μL of distilled water and 200 μL of 5% phenol solution. Samples placed in glass tubes were mixed with 3 mL of concentrated sulfuric acid, and left for for 30 min for color development. The absorbance was measured at

490 nm. Glucose was used as a standard for calibration curve development. The results were expressed as milligram of glucose per gram of fresh weight sample (mg Glc/g FW).

Measurements for pH, TPC, TFC, Car, Phy, and SS were performed in triplicate, and the results were expressed as mean ± standard deviation (SD).

Isolation of proteins/peptides and electrophoretic analysis

Protein isolation from bean-based formulations with pea microgreens was performed by applying the procedure suitable for bean flour (Morales-de Leon *et al.*, 2007). The following samples were tested: dry beans (milled), cooked beans, and fermented formulations stored for up to 30 days. Two samples were analyzed from each formulation: from the central part of the product, and the other from 1mm close to layer, but not in contact with the pea microgreens. The concentration of protein/peptide was measured using bicinchoninic acid (BCA) reagent (Pierce Biotechnology, Rockford, USA).

A portion of 1 g of sample was soaked in 10 mL of NaOH (pH 8.5), vigorously vortexed, and incubated in a rotator at room temperature for 1 h. The suspension was centrifuged at 12,000×*g* for 20 min to remove particles. Supernatant pH was adjusted to 4.0, and the mixture was centrifuged again after 1-h incubation. A precipitate was formed in the case of raw bean flour and cooked beans, which was partially dissolved in NaOH (pH 8.5). The suspension originating from bean-based formulations was turbid and without precipitate. Standard sodium dodecyl sulfate (SDS) electrophoresis was performed under reducing conditions using 12% gel and Coomassie brilliant blue (CBB) or silver staining.

Antioxidant properties

Antioxidant activity of the samples was determined using different assays: total antioxidant capacity (TAC), cupric ion reducing antioxidant capacity (CUPRAC), and 2,2-diphenyl-1-picrylhydrazyl ($DPPH^{\cdot}$) radical scavenging assay.

The TAC was determined using the *in vitro* phosphomolybdenum total antioxidant capacity method described by Prieto *et al.* (1999). An aliquot of 50 µL of the diluted sample was mixed with 3 mL of phosphomolybdenum reagent, incubated at 95°C for 90 min, cooled to room temperature, and the absorbance was measured at 695 nm. The results were expressed as milligram of ascorbic acid equivalent to per gram of fresh weight sample (mg AAE/g FW).

The CUPRAC assay was performed as described by Kilibarda *et al*. (2021). Briefly, 0.5 mL of the extract was mixed with 0.5 mL of 0.01M copper(II) chloride, 0.5 mL of 0.0075M neocuproine solution prepared in absolute ethanol, and 0.5 mL of 1M ammonium acetate buffer solution in a plastic tube. The reaction mixture was incubated at room temperature for 30 min in the dark, and the absorbance was measured at 450 nm. Ascorbic acid was used for constructing calibration curve, and the results were expressed as milligram of ascorbic acid equivalent to per gram of fresh weight sample (mg AAE/g FW).

The free radical scavenging activity of the extracts was evaluated using the $DPPH^{\cdot}$ radical scavenging assay (Kostić *et al*., 2021a). Briefly, 100 µL of the extract was mixed with 100 µL of Milli-Q water and 1,690 µL of DPPH working solution (0.062 g/100 mL) prepared in methanol. The reaction mixture was vortexed and incubated at room temperature for 30 min in the dark, and the absorbance was measured at 517 nm. The obtained results were expressed as the percentage of DPPH radical inhibition and calculated according to the following equation:

$$DPPH^{\cdot} \text{ radical scavenging activity (\%)} = (Ac - As)/Ac \times 100,$$

where Ac is the absorbance of the $DPPH^{\cdot}$ working solution and As is the absorbance of the sample mixed with DPPH working solution. The results were expressed as Trolox equivalent based on fresh sample weight (µmol TE/g FW).

All antioxidant assays were performed in triplicate, and the results were expressed as mean ± SD. Phytochemical composition analyses and antioxidant activity assays of fermented bean-based product with different microgreen coatings (beans/basil [BB], beans/pea [BP], beans/radish [BR], beans/red pepper/chives [BRpC], and beans/saffron/radish [BSR]) were performed after storing samples for 2 weeks at 4°C.

Sensory evaluation

Samples of five fermented bean-based products with different microgreen-coatings (BB, BP, BR, BRpC, and BSR) were subjected to consumer sensory testing, while the product samples coated with only pea microgreens and stored for 30 days (BP1, BP10, BP20 and BP30) were subjected to descriptive sensory evaluation. All sensory tests were conducted in the sensory testing laboratory of the University of Belgrade, Belgrade, Serbia.

Consumer tests

Consumer tests were conducted with 60 students and employees of the University, selected based on their regular consumption of bean-based products. The age and gender distribution of the consumer sample are shown in Supplementary Table S1. A sample of 60 untrained consumers was sufficient for both check-all-that-apply (CATA) and question sensory profiling and sensory acceptability tests in this research (Ares *et al.*, 2014; Hough *et al.*, 2006).

Sensory profiling

The CATA procedure was used to obtain products' sensory profiles from the consumer panel, as described by Buck and Kemp (2018). A list of 21 sensory attributes/descriptors (Supplementary Table S2) was presented to panelists, who indicated which attributes adequately described their sensory experience of the evaluated product by ticking appropriate boxes. The list was generated by descriptive sensory panel during preliminary testing and training sessions. Two expressions related to purchase intention ('willing to buy' and 'not willing to buy') were also included in the attributive list.

Acceptance tests

Overall acceptability, as well as acceptability of flavor and texture of the tested products, was assessed using the 9-point Hedonic scale (1 = dislike extremely, 5 = neither like nor dislike, 9 = like extremely). The 9-point just-about-right (JAR) scale was also used to evaluate the intensity of salty taste (not salty enough/JAR/too salty), sour taste (not sour enough/JAR/too sour), flavor of the beans and flavor of the coating herbs (not enough/JAR/too much), firmness (too soggy/JAR/too firm, brittle), crumbliness (JAR/too crumbly), and hardness (too soft/JAR/too hard).

Descriptive sensory analysis

Descriptive sensory evaluation was conducted by 14 semi-trained staff members experienced in sensory profiling, using a methodology similar to the flesh profiling technique described by Delarue (2014). Several training sessions were held with panelists to prepare a list of sensory attributes for the tests (Supplementary Table S3), following the procedure described by Heymann *et al.* (2014), using samples of the fermented bean-based products with different microgreen coatings prepared specifically for this purpose. All evaluated product samples were served simultaneously in a random order to panelists, who rated these attributes in their own way by directly comparing the samples and using 15-cm intensity line scales. The panelists were instructed to remove coatings from samples after evaluating appearance of the product, before tasting. The test was performed once.

Statistical analysis

Phytochemical data

The phytochemical data were analyzed using ANOVA and Tukey's Honest Significant Difference (HSD) test for multiple pairwise comparisons at 0.05 level of statistical significance. In addition, Principal Component Analysis (PCA) based on correlation matrix with Varimax rotation was applied.

Sensory data

The hedonic data were analyzed using ANOVA and Tukey's HSD test for multiple pairwise comparisons ($\alpha = 0.05$). The mean-drop analysis was conducted following the procedure described by Schraidt (2009). JAR scores were grouped into three categories ('below JAR', 'at JAR', and 'above JAR') as described by Tomic *et al.* (2017), and the mean overall hedonic score was calculated for each category and statistically compared using ANOVA and Tukey's HSD test. The cutoff value, that is, the minimum percentage skew for 'not just right', was set at 20% of the total consumer panel.

To include the overall hedonic ratings in multivariate statistical analysis of the binary CATA data, which consisted of sensory profile and purchase intention data, the overall acceptability was transformed into two variables, 'like' and 'dislike', as described by Tomic *et al.* (2024). Of the total 25 variables (21 sensory descriptive, 2 purchase intentions, and 2 hedonic ratings), seven variables selected with less than 20% of the total number of occurrences for each product sample, which was considered a random selection (Lee *et al.*, 2023), were excluded from statistical analysis. The remaining 18 variables were first subjected to the Cochran's *Q* test, followed by multiple pairwise comparisons using the Sheskin's critical difference procedure to identify those that significantly discriminate among samples ($\alpha = 0.05$). In all, 13 statistically significant variables were subjected to correspondence analysis (CA).

The descriptive sensory data were first standardized across assessors (Romano *et al.*, 2008); then, ANOVA was performed (with 'assessors' as a random factor), followed by Tukey's HSD multiple pairwise comparison test ($\alpha = 0.05$). After excluding four attributes that were not statistically significant, the raw descriptive data of the remaining 14 attributes were subjected to Generalized Procrustes Analysis (GPA), followed by PCA based on correlation matrix applied to the GPA-consensus data.

Results and Discussion

Chemical profile of bean-based samples

In recent decades, increasing attention has been devoted to the development of new functional food products. Such foods serve multiple roles; in addition to meeting primary dietary requirements, they may contribute to both mental and physical health (Rawat *et al.*, 2024). Microgreens are considered novel functional food sources and have gained increasing acceptance and popularity in the market because of their high nutrient density and potential health-promoting properties (Dhaka *et al.*, 2023). Ascorbic acid, α-tocopherol, β-carotene, phenolic antioxidants, carotenoids, anthocyanins, glucosinolates, and sugars are among the major bioactive compounds found in microgreens in substantial amounts (Bhaswant *et al.*, 2023).

Proximate nutritional composition

The values of nitrogen, crude protein, lipid, ash, and moisture content in fermented bean-based products coated with different microgreens are presented in Table 3. Sample BRpC exhibited the highest content of nitrogen, crude protein, lipids, and ash. In contrast, sample BB showed the highest moisture content and the lowest values of nitrogen, crude protein, and lipids. High lipid content may be attributed to both composition of fermented bean-based product and fermentation process itself, as the total lipid content may increase during fermentation (Rohaman, 1990). Beans and other legumes represent valuable sources of dietary fiber and plant-based proteins, and thereby associated with beneficial effects on human health (Meenu *et al.*, 2023; Vasić *et al.*, 2009). Moreover, beans are widely used in various vegan and gluten-free products (Sparvoli *et al.*, 2021; Wesley *et al.*, 2021).

It has been reported that food products formulated with microgreens have enhanced protein, fat, ash, and dietary fiber content (cup-cakes with beetroot and wheatgrass), protein, fat, and ash content (cookies with wheatgrass powder), protein, fat, and crude fiber content (instant soup mix based on wheatgrass), protein, ash, fat, dietary fiber, total phenolics, and flavonoids (muffins with wheatgrass and mung-bean microgreens powder), and phenolic compounds (muffins with wheatgrass powder) (Rawat *et al.*, 2024). Observed differences among examined samples probably reflect the influence of different microgreen coatings and added spices on nutritional composition.

Phytochemical composition

Table 4 presents the results of phytochemical composition of the studied bean-based product samples.

pH values of the samples showed significant differences, compared to the control sample ($P < 0.05$), with the exception of the sample containing daikon radish microgreens (BR). The carotenoid content differed significantly among all samples ($P < 0.05$). The sample enriched with daikon radish and saffron (BSR) exhibited the highest carotenoid content, followed by the sample coated with pea microgreens (BP), while the control sample showed the lowest value. This observation is expected because both saffron spice (Popović-Djordjević *et al.*, 2021) and pea microgreens (Bhaswant *et al.* 2023) are good sources of carotenoids. Major secondary metabolites in saffron stigmas are carotenoids, which are responsible for the color of saffron (Popović-Djordjević *et al.*, 2021). Moreover,

Table 3. Nutritional profile of fermented bean-based product samples with various microgreens.

Sample	Nc (%)	CPcFW (%)	LcFW (%)	TCcFW (%)	Moisture (%)	Ash (%)
BB	2.65	16.55	27.30	30.13	23.05	3.86
BP	2.87	17.92	28.20	31.61	18.51	4.61
BR	2.73	17.08	30.15	33.42	15.68	4.35
BRpC	2.99	18.70	30.25	28.59	18.64	4.69
BSR	2.67	16.66	27.55	35.26	16.66	4.65

Notes: BB: beans/basil microgreens; BP: beans/pea microgreens; BR: beans/radish microgreens; BRpC: beans/red pepper/chives microgreens; BSR: beans/saffron/radish microgreens; Nc: nitrogen (N) content; CPcFW: crude protein content in the sample (fresh weight basis); LcFW: lipid content in the sample (on fresh weight basis); TCcFW: total carbohydrate content in the sample (on fresh weight basis).

Table 4. **Phytochemical composition of fermented bean-based products with different microgreen coatings.**

Sample	pH	TPC (mg GAE/g FW)	TFC (mg RE/g FW)	Car (µg/g FW)	Phy (mg NaPhy/g FW)	SS (mg Glc/g FW)
BB	4.91 ± 0.02^{c}	1.003 ± 0.014^{b}	4.419 ± 0.013^{d}	14.48 ± 0.16^{d}	34.233 ± 0.679^{a,b}	19.719 ± 0.547^{a,b}
BP	4.88 ± 0.01^{b}	0.871 ± 0.066^{a}	5.243 ± 0.025^{e}	18.36 ± 0.36^{e}	33.582 ± 0.198^{a}	17.572 ± 2.214^{a}
BR	4.85 ± 0.01^{a}	1.001 ± 0.014^{b}	2.507 ± 0.175^{a}	9.92 ± 0.12^{c}	33.938 ± 0.043^{a,b}	22.907 ± 1.174^{b}
BRpC	4.91 ± 0.01^{c}	1.010 ± 0.019^{b}	3.693 ± 0.300^{b,c}	4,04 ± 0.08^{b}	34.249 ± 0.014^{a,b}	19.931 ± 1.360^{a,b}
BSR	4.90 ± 0.01^{b,c}	1.178 ± 0.004^{c}	3.800 ± 0.256^{c}	47.56 ± 0.76^{f}	34.729 ± 0.269^{b,c}	19.232 ± 2.167^{a,b}
Con	4.84 ± 0.01^{a}	0.917 ± 0.052^{a,b}	3.188 ± 0.244^{b}	2.00 ± 0.08^{a}	35.438 ± 0.043^{c}	19.219 ± 0.540^{a,b}

Notes: BB: beans/basil microgreens; BP: beans/pea microgreens; BR: beans/radish microgreens; BRpC: beans/red pepper/chives microgreens; BSR: beans/saffron/radish microgreens; TPC: total phenolic content (milligram of gallic acid per gram of fresh sample weight); TFC: total flavonoid content (milligram of rutin per gram of fresh weight sample); Car: carotenoids content (microgram per gram of fresh weight sample); Phy: phytate content (milligram of sodium phytate per gram of fresh weight sample); SS: soluble sugars content (milligram of glucose per gram of fresh weight sample).
Values are expressed as mean ± SD (n = 3).
Different superscript alphabets within the same column indicate statistically significant difference according to Tukey's test ($P < 0.05$).

the highest TPC value was also observed in BSR sample ($P < 0.05$), confirming that saffron is rich in phenolic compounds (Popović-Djordjević *et al.*, 2021). Literature data suggest considerably lower TPC values in raw and cooked beans (Akillioglu and Karakaya, 2010). In contrast, phytate content was significantly lower ($P < 0.05$) in all samples, except for BSR. Degradation of phytates during fermentation is favorable because lower phytate levels improve nutrient utilization and product's functional properties (Liu *et al.*, 2024). Among the examined samples, the product enriched with radish microgreens showed the lowest TFC. Previous studies have reported very low or undetectable flavonoid content in raw and cooked beans (Akillioglu and Karakaya, 2010; Lin *et al.*, 2008). In contrast, samples enriched with microgreens, especially BP (pea microgreens), showed higher TFC, indicating that microgreens were the main contributors to the flavonoid content of the examined products. Overall, the obtained results demonstrated clear differences in the levels of bioactive compounds depending on the type of microgreens used. Soluble sugar contents did not differ significantly from the control ($P > 0.05$). These findings suggest that microgreens can further enrich bean-based products, potentially improving their nutritional profile and functional properties. Previous research has shown that vegetable microgreens are naturally rich in bioactive compounds and exhibit high antioxidant activity, supporting their use in enhancing the functional quality of food products (Stajčić *et al.*, 2024).

Antioxidant properties

Saffron spice significantly contributed to the highest carotenoid and TPC) observed in the BSR sample. The sample containing red pepper/chive microgreens (BRpC) exhibited the highest antioxidant activity in TAC and CUPRAC assays, while the sample with pea microgreens (BP) showed highest activity in the CUPRAC assay. In addition, the samples containing saffron and daikon radish (BRS) and basil microgreens (BB) demonstrated the highest DPPH$^{\cdot}$s cavenging activity (Figure 2). Previous studies have shown that fermentation of soybean, chickpea, and beans results in higher levels of phenols and flavonoids, thereby enhancing antioxidant potential (Liu *et al.*, 2022; Saharan *et al.*, 2020). Moreover, microgreens are reported to increase the antioxidant content (Rawat *et al.*, 2024), which is consistent with the results obtained for the bean-based products analyzed in this study.

The phytochemical composition and antioxidant activity data were further analyzed using PCA to observe this set of variables simultaneously. The results are shown in Figure 3. The first four principal components (PCs) were retained to describe bean-based products, accounting for 86.3% of total variability in the original dataset. PC1 was mainly characterized by pH, TAC, and DPPH$^{\cdot}$ PC2 was characterized by Car; and TPC and PC3 showed strong correlations with TFC and SS, while PC4 distinguished the products primarily according to differences in CUPRAC (strong correlations were defined by loading values ≥0.72). Based on the position of the control sample (Con), it is evident that the addition of microgreen coatings and spices contributed to higher values for most of the evaluated ochemical parameters. Notably, each extracted PC differentiated bean-based products in a distinct manner, indicating that no clear pattern was found in the differentiation between samples based solely on phytochemical composition. The samples coated with radish microgreens (BR) and pea microgreens (BP)

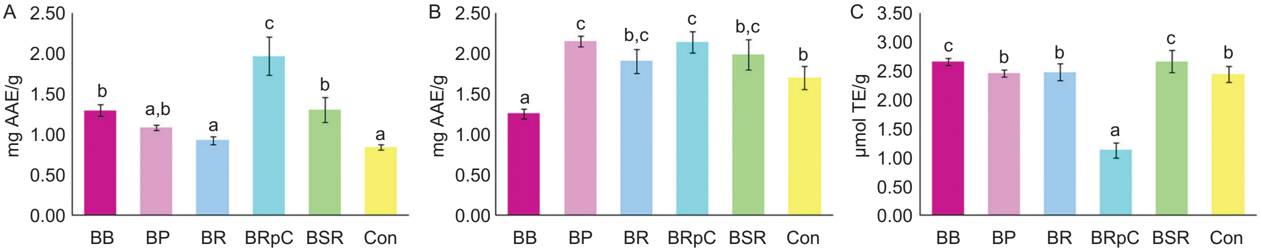

Figure 2. (A) Total antioxidant capacity (TAC: milligrams of ascorbic acid equivalent to per gram of fresh weight sample); (B) cupric reducing antioxidant capacity (CUPRAC: milligrams of ascorbic acid equivalent to per gram of fresh weight sample); and (C) DPPH·scavenging activity (DPPH:·Trolox equivalent to per gram of fresh weight sample) of vegan bean-based product. BB: beans/basil microgreens; BP: beans/pea microgreens; BR: beans/radish microgreens; BRpC: beans/red pepper/chives microgreens; BSR: beans/saffron/radish microgreens. The values are represented as mean ± SD (n = 3). Different superscript alphabets indicate significant differences according to Tukey's test (*P* < 0.05).

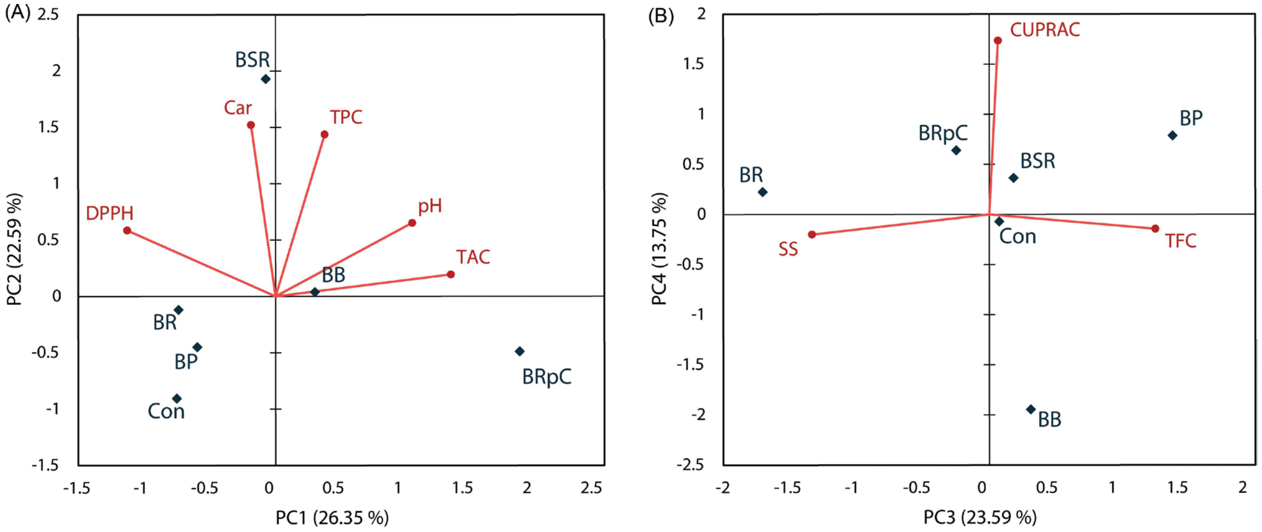

Figure 3. Results of Principal Component Analysis (PCA) with varimax rotation applied to the phytochemical composition and antioxidant activity data of the fermented bean-based products with different microgreen coatings, stored for 2 weeks at 4°C. BB: beans/basil microgreens; BP: beans/pea microgreens; BR: beans/radish microgreens; BRpC: beans/red pepper/chives microgreens; BSR: beans/saffron/radish microgreens.

showed similar values of DPPH·, pH, and TAC, but differed greatly in SS (significantly higher in BR) and TFC (significantly higher in BP) content. The phytochemical profiles of the samples with radish microgreen coating (BR and BSR) were clearly influenced by the presence of saffron in the BSR sample, which exhibited higher levels of Car, TPC, and TFC, whereas the BR sample showed higher DPPH· and SS content. BRpC samples with chive microgreen coating and red pepper were characterized by higher values of pH, TAC, and CUPRAC.

Analysis of proteins/peptides

pH value, and nitrogen and crude protein content were measured in BP samples during storage prior to determining the concentration of proteins and peptides (Table 5). The nitrogen and crude protein content increased from day 1 to day 20, followed by a slight decrease after 30 days of storage. Experimental studies have shown that protein content in the substrate may increase during fermentation because of microbial activity involving yeast and bacteria. Agricultural materials with considerable carbohydrate content, such as beans, represent suitable substrates for the growth of microorganisms in terms of both cell number and total amount of protein (Thakaew *et al.*, 2024). pH value remained relatively stable during the first 20 days but increased slightly day 30. The breakdown of amino acids releases ammonia, which causes pH to stabilize or even slightly rise in the final stages of fermentation. The obtained results were consistent with previously reported findings (Jung *et al.*, 2016; Onwurafor *et al.*, 2014).

The highest concentration of proteins and peptides was measured in the extract of raw bean. Changes occurring during processing and fermentation influenced protein structure and stability (Table 6). For the purpose of this study, proteins and peptides were analyzed without considering the yield. The protein and peptide content in extracts increased during storage, confirming previous results (Table 5).

All analyzed samples were subjected to electrophoresis and CBB staining. Proteins extracted from raw and cooked beans retained their native structure (Figure 4A). BP samples were the suspensions whose turbidity decreased during maturation and storage, gradually forming small particles (Figure 4B). The central part and region close to pea microgreens (BP) within the same bean-based product showed similar appearance and identical protein/peptide concentration (Figure 4C). To determine the approximate molecular masses of separated proteins and peptides, electrophoresis was repeated using only fractions from the central part of BP formulations, and the bands were visualized by silver staining (Figure 4D). The most abundant molecular species exhibited a mass close to 17 kDa marker, while proteins with higher molecular masses were visible faintly only at approximately 50 kDa and 30 kDa.

According to the literature, bean proteins have masses ranging between 21 kDa and 86 kDa (Morales-de Leon *et al.*, 2007). The major proteins include phaseolin (43–45 kDa), a storage protein, and albumins (27 kDa), which include enzymes, enzyme inhibitors, and lectins (Mundi and Aluko, 2012; Tarahi, 2024). These proteins were also dominant in the extracts obtained from raw and cooked beans in the present study. Bean processing methods, such as cooking, can inactivate proteinase inhibitors, thereby creating conditions favorable for protein proteolysis. In addition, microbial fermentation promotes peptide formation (de Fatima Garcia *et al.*, 2021), as observed in the examined samples during prolonged fermentation. The vacuole of *S. cerevisiae* contains several proteases, such as proteinase A and B, carboxypeptidase X and S, aminopeptidase I and Y, dipeptidyl aminopeptidase B, etc. (Hecht *et al.*, 2014). They are active at distinct amino acid sites, enabling efficient proteolysis of different proteins. According to our results, very little intact protein

Table 5. Changes in pH value and crude protein content of fermented bean-based formulation with pea microgreen (BP) during storage period.

Sample	Storage period (days)	pH	Nc (%)	CPcFW (%)
BP1	1	4.23	1.36	8.52
BP10	10	4.32	1.84	11.50
BP20	20	4.32	2.14	13.38
BP30	30	4.60	2.05	12.82

Notes: Nc: nitrogen (N) content; CPcFW: crude protein content in samples (fresh weight basis).

Table 6. Proteins and peptides in bean extracts.

Sample	Raw bean	Cooked bean	BP1	BP10	BP20	BP30
Proteins/peptide concentration (mg/mL)	9.74	1.00	0.60	0.54	0.95	1.81
Total extracted proteins/peptides (mg)	87.66	9.00	5.40	4.86	8.55	16.29

Note: BP: beans/pea microgreens.

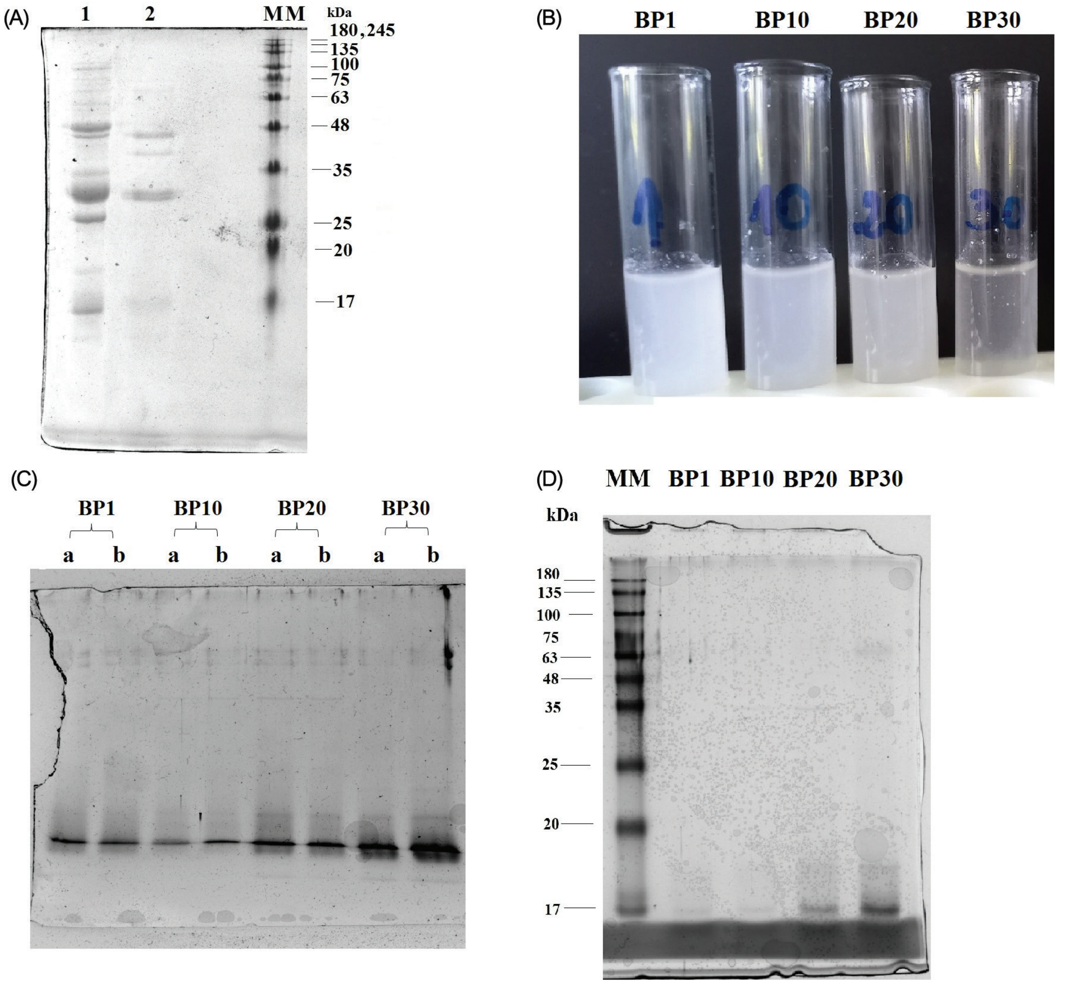

Figure 4. (A) Electrophoresis of extracted proteins/peptides from (1) raw and (2) cooked beans, and molecular mass markers (MM); (B) appearance of protein/peptide extracts; (C) electrophoresis of extracted proteins/peptides—comparison of sampling sites (a: samples close to pea microgreen, b: samples from the central part of the formulation); (D) electrophoresis of extracted proteins/peptides—comparison of samples at different storage periods: 1 day (BP1), 10 days (BP10), 20 days (BP20), and 30 days (BP30).

remained in the extract after fermentation. Montoya *et al.* (2008) examined the susceptibility of 43 phaseolin types to proteolysis and reported that heat treatment significantly increased proteolysis, whereas amino acid composition had a minor influence only. Results from *in vitro* proteolysis experiments using pepsin and pancreatin demonstrated some differences among cultivars; however, heating for 6 h increased the overall proteolysis to nearly 100% in certain types of phaseolins. This effect was attributed to structural changes characterized by a decrease in α-helixes and an increase in random structures. When the results for protein/peptide concentration were compared to moisture content, the increase in concentration was much more pronounced than the increase in dry mass or pH. This finding supports the conclusion that the increased concentration of small protein and peptide, as well as their solubilization, is a consequence of protein fragmentation during sample storage and aging.

Small bean proteins and peptides exert antioxidant and inhibitory activity against a number of enzymes (de Fatima Garcia *et al.*, 2021; Hu *et al.*, 2023). Two main mechanisms, identified using bioinformatics-based approaches, are proposed to explain enzyme inhibition: 'alteration of enzyme conformation' and 'competition with substrates' (Ngoh and Gan, 2018). Although gastrointestinal digestion enables proteolytic cleavage of proteins into small peptides, foods containing pre-fragmented proteins may contribute to faster, easier, and more efficient nutrient absorption. Fermentation processes based on microbial enzymes are used widely to transform various substrates, including grains and vegetables (Bonsu *et al.*, 2025; Peres Fabbri *et al.*, 2024). Peptides derived from fermented rice, oat, quinoa, soy, and red beans are shown to exert desirable nutritional and health effects. Numerous plant fermentation methods exist, and choice of a particular method depends on the nature of the substrate, its state (liquid or solid), the strain (single or mixed, fungi or bacteria), and other physicochemical variables that characterize the technological process (Zhao *et al.*, 2024).

The antioxidant activity of bean-derived peptides is documented extensively, including those obtained from bean husks (Shchypanskyi *et al.*, 2025). Bean peptides obtained after enzymatic hydrolysis with pepsin are reported to exhibit significantly stronger antioxidant activity than protein isolates, and even greater activity than ascorbic acid, which is commonly used as a reference substance for antioxidant evaluation (Sarker *et al.*, 2020). Although proteins interact with reactive oxygen and nitrogen species and scavenge free radicals, peptides often exhibit a higher potential to neutralize pro-oxidant species. This effect includes the ability to chelate metal ions that initiate the Fenton reaction. Furthermore, peptides, and specifically those containing sulfur, aromatic, and imidazole-containing structures, are often more effective antioxidants than amino acids (Davies, 2005; Ohashi *et al.*, 2015).

Sensory evaluation

Consumer tests

The results of consumer sensory evaluation of fermented bean-based products with different microgreen coatings, obtained using the CATA method, are shown in Table 7 and Figure 5. The overall hedonic ratings included in this statistical analysis (as described in the Materials and Methods section) provided additional information on acceptability in terms of the proportion of participants who reported that they actually 'liked' or 'disliked' the products. Seven sensory attributes (out of 21 attributes) were excluded from the statistical analysis because they were selected in less than 20% of the total responses for each product sample (Supplementary Table S2).

Table 7. Proportions of perceptual, hedonic, and purchase intention responses for fermented bean-based product with different microgreen coatings.

Attributes	Samples				
	BB	BP	BR	BRpC	BSR
Spring onion/garlic (flavor)	0.150^{a}	0.033^{a}	0,100^{a}	0.800^{b}	0.083^{a}
Beans (flavor)	0.667^{a}	0.633^{a}	0.170^{a}	0.583^{a}	0.567^{a}
Sweet pepper (flavor)	0.100^{a}	0.083^{a}	0.067^{a}	0.617^{b}	0.167^{a}
Radish (flavor)	0.017^{a}	0.050^{a}	0.583^{b}	0.050^{a}	0.383^{b}
Peas (flavor)	0.083^{a}	0.483^{b}	0.167^{a}	0.183^{a}	0.167^{a}
Basil (flavor)	0.650^{b}	0.167^{a}	0.067^{a}	0.100^{a}	0.033^{a}
Spices (flavor)	0.767^{b}	0.517^{a}	0.567^{a,b}	0.700^{a,b}	0.600^{a,b}
Leather, new shoes (flavor)	0.050^{a}	0.050^{a}	0.117^{a}	0.100^{a}	0.683^{b}
Salty	0.433^{a}	0.400^{a}	0.400^{a}	0.367^{a}	0.283^{a}
Sour	0.683^{a}	0.617^{a}	0.700^{a}	0.517^{a}	0.617^{a}
Bitter	0.183^{a,b}	0.183^{a,b}	0.283^{b}	0.067^{a}	0.317^{b}
Spreadable	0.350^{a}	0.267^{a}	0.333^{a}	0.400^{a}	0.400^{a}
Soggy	0.200^{a,b}	0.200^{a,b}	0.250^{a,b}	0.150^{a}	0.317^{b}
Sandy	0.217^{a}	0.233^{a}	0.183^{a}	0.200^{a}	0.200^{a}
Liked it	0.567^{b,c}	0.417^{a,b}	0.367^{a,b}	0.717^{c}	0.267^{a}
Disliked it	0,283^{a,b}	0.367$^{a–c}$	0.450^{b,c}	0.183^{a}	0.533^{c}
Willing to buy	0.550^{b,c}	0.467^{b,c}	0.333^{a,b}	0.650^{c}	0.233^{a}
Not willing to buy	0.450^{a,b}	0.533^{a,b}	0.667^{b,c}	0.350^{a}	0.767^{c}

Notes: The product was stored for 2 weeks at 4°C, and was evaluated by a panel of 60 consumers using the check-all-that-apply (CATA) procedure.
BB: beans/basil microgreens; BP: beans/pea microgreens; BR: beans/radish microgreens; BRpC: beans/red pepper/chives microgreens; BSR: beans/saffron/radish microgreens.
Different superscript alphabets within the same column, indicate statistically significant differences according to Tukey's test ($P < 0.05$).

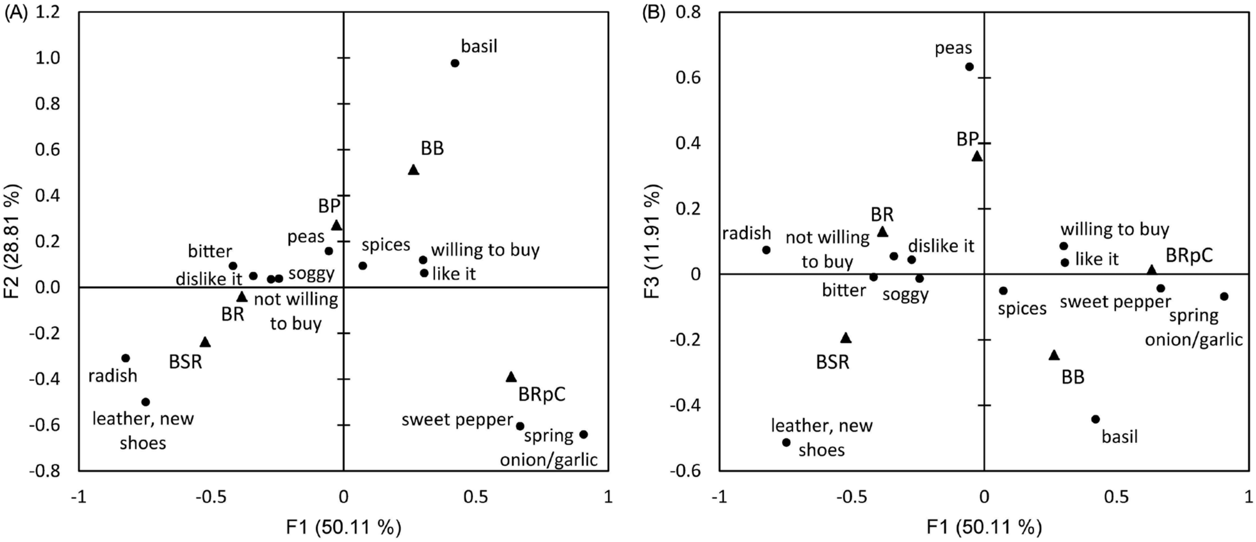

Figure 5. Perceptual, hedonic, and purchase intention responses to the fermented bean-based product with different microgreen coatings, stored for 2 weeks at 4°C. BB: beans/basil microgreens; BP: beans/pea microgreens; BR: beans/radish microgreens; BRpC: beans/red pepper/chive microgreens; BSR: beans/saffron/radish microgreens. Results of the matching analysis were applied to the binary CATA data (N = 60).

The remaining attributes were first tested for statistical significance, and those showing significant differences ($P < 0.05$) were subjected to dimension-reduction analysis. According to a scree plot (where the third point indicated the point at which eigenvalues stopped decreasing rapidly), the first three factors were retained, explaining 90.8% of the total variance in raw contingency data (Figure 5).

From the results, participants were able to identify main flavor components originating from the key ingredients of products. The sample containing red bell pepper and coated with chive microgreens (BRpC) was characterized by spring onion/garlic flavor notes (80% of participants) derived from chives, as well as by a sweet bell pepper flavor (62% of participants) (Table 7). The radish flavor was perceived by 58% and 38% of the participants in the samples coated with radish microgreens (BR and BSR, respectively; no significant difference at 0.05). Interestingly, the sample containing saffron (BSR) was associated with the smell of new leather shoes, as 68% of participants checked the descriptor 'leather/new shoes' when evaluating this sample. In the complex matrix of BSR formulation, saffron with its key aroma and bitter-tasting compounds, safranal and picrocrocin, may be a major contributor of specific sensory attributes associated with this product. Moreover, the presence of enzymes and the fermentative activity of yeast species may also influence the overall flavor and aroma of a BSR product (Giuffrè and Giuffrè, 2024; Popović-Djordjević *et al.*, 2021). The basil aroma was perceived by 65% of participants in the sample coated with basil microgreens (BB), whereas the pea aroma was primarily detected in the product coated with pea microgreens (BP, 48%). In all cases, the proportion of participants who selected the corresponding flavor descriptor for the other evaluated products was low (<20%). None of the samples differed significantly in terms of sourness and saltiness, whereas the products coated with radish microgreens (BR and BSR) were perceived as significantly more bitter ($P < 0.05$) than the others. Although no significant difference ($P > 0.05$) in sourness was observed among the samples, this sensory attribute was recognized by a large number of participants for each sample (52–70%), indicating that sourness was a prominent characteristic of the products.

Observing hedonic responses and purchase intention, a clear separation was observed between the samples containing radish microgreens (BR and BSR) and those containing chive (BRpC) or basil microgreens (BB). Approximately 50% of participants disliked the former two products, and 67–77% indicated that they would not be willing to purchase them (no statistically significant differences were observed between BR and BSR for these variables at $\alpha = 0.05$). In contrast, more than 70% of participants liked the product with red bell pepper and chive microgreens (BRpC), and 65% expressed willingness to purchase it. For BB sample, approximately 55% of participants expressed both liking and purchase intention, with no statistically significant differences between these two variables. The results regarding the acceptability of bean-based formulations, presented in Table 7 and Figure 5, include hedonic data converted into binary form to indicate the actual proportions of participants who 'liked' or 'disliked' the products.

Table 8 shows mean hedonic ratings of the evaluated products for the entire consumer panel. The overall acceptability scores for the samples containing radish microgreens (4.6 ± 2.4 and 4.2 ± 2.7 for BR and BSR, respectively) were within the 'neither like nor dislike' range (values between 4 and 6 on the 9-point hedonic scale) and were significantly lower than the scores for the samples containing chive (BRpC, 6.3 ± 2.3) and basil (BB, 5.6 ± 2.3) microgreens ($P < 0.05$). These values indicate that the consumer panel preferred the latter two products. The reasons for this lower acceptability could be the flavor of these bean-based products, as no statistically significant differences ($P > 0.05$) were observed among the samples in terms of texture acceptability. A similar trend was observed for flavor ratings: the mean hedonic scores for flavor were significantly lower ($P < 0.05$) for the radish microgreen-coated samples than for the samples coated with chive or basil microgreens and were close to the borderline between the 'dislike' and 'neither like nor dislike' options (approximately 4.0). In contrast, the flavor hedonic scores for BRpC and BB were around 6.0, corresponding to the borderline between the 'neither like nor dislike' and 'like' options. Negative flavor notes that may have influenced product acceptance include bitterness (in combination with sourness) and an aroma described as leather or new leather shoes. The products containing radish microgreens were perceived as significantly more bitter by the consumer panel, compared to BRpC and BB, while the product containing saffron exhibited the 'leather/new shoes' aroma note, which was considered atypical for food products. The identification of atypical and polarizing descriptors, such as 'leather' or 'new shoes', suggests a potential limitation in the flavor profile of saffron-enriched formulation, indicating that chemical interactions between saffron volatiles and fermented bean matrix produce off-flavors that severely compromise consumer hedonic perception. Additionally, significantly higher bitterness scores observed for radish-coated samples suggest that the concentration of glucosinolate-derived compounds (Bell *et al.*, 2018; Zhong *et al.*, 2023) may have exceeded the threshold of sensory tolerance. This flavor imbalance probably contributed to reduced consumer acceptability and purchase intention.

Further insights into the lower acceptance and potential strategies for product optimization can be derived from the results of the mean drop analysis presented in Figure 6. This mathematical tool combines hedonic data with information on the appropriateness of the intensity of selected sensory attributes obtained using just-about-right scales. A point in the plot showing a large proportion of consumers (≥20% in this case) together with a significantly larger mean drop value for a particular attribute indicates that the attribute deviates from its optimal value and therefore represents a potential direction for product modification (Lawless and Heymann, 2010). In general, the evaluated products were perceived by a considerable proportion of participants (≥20% and up to 53%) as 'too sour' (all samples), with 'too much herbal flavor' (BRpC, BB, and BR), and as having 'not enough bean flavor', which consumers probably expected (BB, BP, BR, and BSR). However, only four consumer groups appeared in the plot with a significantly larger mean drop value ($P < 0.05$): two groups perceived the samples BB and BSR as 'too sour', while two perceived the samples BP and BR as having

Table 8. Acceptability ratings for fermented bean-based product with different microgreen coatings on a 9-point hedonic scale.

Samples	Hedonic attributes		
	Overall acceptability	Flavor acceptability	Texture acceptability
BB	5.6 ± 2.3^{b,c}	5.4 ± 2.5^{b}	6.5 ± 2.1^{a}
BP	5.0 ± 2.5^{a,b}	4.9 ± 2.3$^{a.b}$	5.9 ± 2.2^{a}
BR	4.6 ± 2.4^{a}	4.1 ± 2.5^{a}	5.5 ± 2.3^{a}
BRpC	6.3 ± 2.3^{c}	6.1 ± 2.5^{b}	6.3 ± 2.0^{a}
BSR	4.2 ± 2.7^{a}	3.8 ± 2.0^{a}	5.8 ± 2.3^{a}

Notes: The product was stored for 2 weeks at 4°C.
BB: beans/basil microgreens; BP: beans/pea microgreens; BR: beans/radish microgreens; BRpC: beans/red pepper/chives microgreens;
BSR: beans/saffron/radish microgreens.
Values are mean ± SD (N = 60).
Values marked with the same superscript alphabets within the same column are not statistically different (α = 0.05).

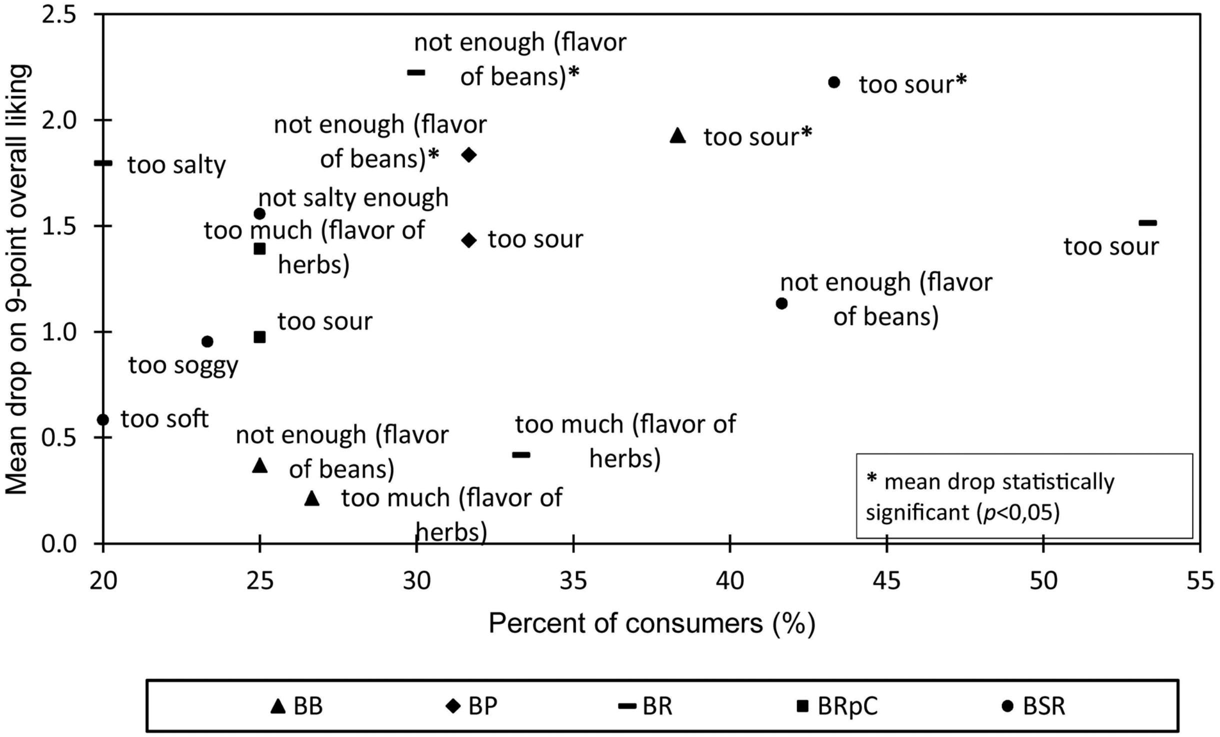

Figure 6. Mean drop analysis of the acceptability data for fermented bean-based product with different microgreen coatings stored for 2 weeks at 4°C (N = 60). BB: beans/basil microgreens; BP: beans/pea microgreens; BR: beans/radish microgreens; BRpC: beans/red pepper/chive microgreens; BSR: beans/saffron/radish microgreens.

'not enough bean flavor'. Considering that sourness was identified as a prominent characteristic of the evaluated products and that no significant differences in sourness were observed among the samples (although the CATA method cannot be considered a sensitive tool for sensory discrimination), the results of the mean drop analysis suggest that the main direction for optimizing these fermented bean-based products is to reduce sourness. In other words, this implies that the fermentation process should be controlled more tightly. Potential approaches may include regulating inoculation and inoculum potential/concentration, adjusting fermentation and storage temperatures, controlling redox potential, adding humectants, such as salt, or managing the presence of off-gases, such as CO_2.

Descriptive sensory evaluation

Descriptive sensory evaluation was conducted to investigate changes in the sensory profile of fermented bean-based product during 1 month of refrigerated storage at 4°C. Pea microgreens were selected for this purpose as the most 'neutral' coating option, according to the results of consumer sensory tests. The product was evaluated after 1, 10, 20, and 30 days of storage. The descriptive data for each attribute were first tested for statistical significance at $\alpha = 0.05$ (Supplementary Table S4). Of the 20 sensory attributes evaluated, six were excluded from further analysis as they were not statistically significant (Supplementary Table S3). The remaining attributes were subjected to dimension-reduction analysis as described previously, and the results are shown in Figure 7 and Supplementary Table S5. According to a scree plot criterion, the first two principal components (PC1 and PC2) were retained to describe product samples in a new two-dimensional (2D) PC space. However, all sensory attributes showed strong correlations with PC1 (loading values ≥ 0.72; Elmore *et al.*, 1999), and weaker correlations with PC2 (Supplementary Table S5), suggesting that differences among samples could be explained primarily by PC1 values.

The evaluated color dimensions (for both coating and product cross section), the overall flavor intensity, and firmness increased gradually ($P < 0.05$) across the sampling time-points (Supplementary Table S4). From the 1st to the 30th day of storage, the color of pea microgreen layer changed gradually from yellow to green, while color of the cross section shifted from creamy to brown. In addition to the accumulation of microbial metabolites and changes in substrate content and conditions during

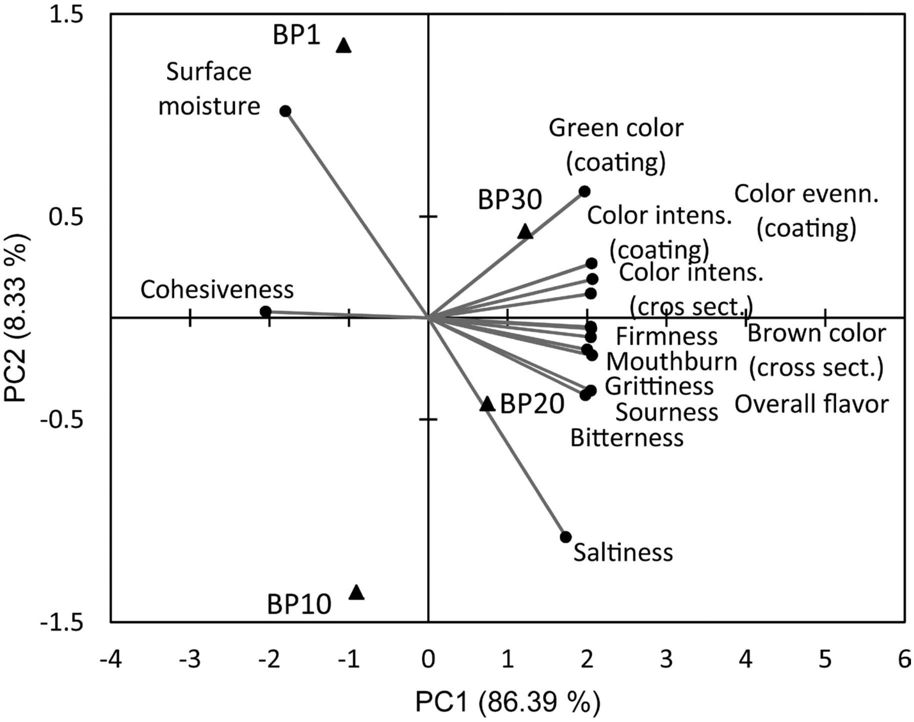

Figure 7. Results of principal component analysis (PCA) applied to the consensus data matrix obtained by applying generalized procrustes analysis (GPA) to descriptive sensory data for the fermented bean-based formulation coated with pea microgreens (BP) stored at 4°C for 1 day (BP1), 10 days (BP10), 20 days (BP20), and 30 days (BP30).

storage, which can suppress cell growth and activity, the fermentation process was partly controlled by storage temperature only (up to 4°C) and continued until the final day of sampling. These changes were probably a major factor contributing to increase in the overall flavor intensity. Sourness, bitterness, and mouthburn sensations were significantly more pronounced in the product after 20 and 30 days of storage than in the samples stored for up to 10 days (Supplementary Table S4), suggesting that these changes were influenced by fermentation and product aging.

The texture of the product was also affected. During storage, the product became significantly firmer and less cohesive, with an increased number of small particles perceptible between the tongue and palate during consumption (grittiness). The surface moisture of the product cross section was significantly more pronounced at the beginning of storage, compared to the 10–30-day old product. This increase in bitterness and grittiness, coupled with the loss of cohesiveness, indicates potential over-fermentation or undesirable protein degradation, which may negatively impact consumer acceptability and limit product’s commercial shelf life.

Conclusions

The nutritional potential of the vegan bean-based product prepared and examined in this study was clearly documented with respect to biomolecule content, antioxidant activity, and sensory characteristics. Depending on the presence and absence of spices, the samples with various microgreen coatings exhibited certain differences in their nutritional and chemical characteristics. PCA indicated that the microgreens and spices contributed to an enhanced phytochemical composition, compared to the control sample. The phytate content was significantly lower in all samples except for the formulation containing saffron and daikon radish microgreens. Conversely, saffron spice significantly increased the carotenoid and total phenolic contents. Samples BB, BP, BRpC, and BRS showed improved antioxidant activity, compared to the control. Differences in sensory characteristics were the most pronounced variations. Samples enriched with red pepper, chive, and basil microgreens showed higher overall acceptability and purchase intention, whereas samples enriched with radish microgreens were perceived as bitterer and less pleasant in flavor. Sourness was identified as a key attribute negatively affecting the overall liking, suggesting that improved control of the fermentation process could enhance product acceptability. These findings highlight that microgreen types strongly influence flavor perception and consumer acceptance. Storage period also affected product characteristics, with changes occurring in opposing directions. Longer storage periods increased the content of proteins and particularly those with lower mass. However, sensory properties tended to deteriorate during prolonged storage. The results therefore suggest the existence of an optimal storage period, which enables a compromise between nutritional and sensory characteristics. Overall, the findings suggest that fermentation and addition of microgreens enhance the nutritional and functional value of bean-based products.

However, the findings were based on the study of only one bean variety, and several types of microgreens; this must be expanded to other varieties of both beans and microgreens. Further evaluation of such products may support the development of innovative plant-based foods of particular interest to people following specialized dietary regimes.

From the perspective of industrial applicability and product positioning**, t**he results indicate that fermented bean-based formulations represent a promising matrix for the development of nutritionally enhanced plant-based fermented products. Beans (*P. vulgaris* L.) provide a sustainable and protein-rich raw material, while fermentation improves protein digestibility and reduces antinutritional factors, such as phytates. Incorporation of microgreens further enriches the product with bioactive compounds while modulating sensory characteristics. This formulation flexibility enables targeted optimization of both nutritional and organoleptic properties. Considering the rapid growth of plant-based dairy alternatives, such products could contribute to the diversification of fermented vegan foods, particularly within the clean-label, functional, and gluten-free market segments. In addition, the relatively simple technological process and the availability of raw materials support the potential for industrial scalability and further development of differentiated formulations.

Ethical Statement

The participation of students and academic staff as sensory assessors in the study conducted was in accordance with the Code of Professional Ethics of the University of Belgrade (Senate of the University of Belgrade, 2016). Owing to the nature of the research, ethical approval by the Ethics Committee of the Faculty of Agriculture, University of Belgrade, was not required. All participants in the sensory evaluation were informed about the study, and they gave consent prior to participation. All necessary protocols to protect the rights and privacy of sensory panelists were followed.

Data Availability Statement

All data supporting the findings of the study are included in the article and the Supplementary Material. Further inquiries can be directed to the corresponding author.

Mandatory Disclosure on Use of Artificial Intelligence

The authors declared that no AI-assisted tools were used in the preparation of this manuscript.

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Conflicts of Interest

The authors declared no conflict of interest.

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Supplementary

Table S1. Demographic data of the consumer sample.

Gender	Age (years)				Total
	≤30	31–45	46–55	>55	
Male	12 (20.0%)	6 (10.0%)	3 (5.0%)	1 (1.7%)	22 (36.7%)
Female	21 (35.0%)	6 (10.0%)	4 (6.7%)	7 (11.7%)	38 (63.3%)
Total	**33** (55.0%)	**12** (20.0%)	**7** (11.7%)	**8** (13.3%)	**60** (100.0%)

Note: Frequencies are in their absolute and relative (%) values.

Table S2. List of sensory descriptors used in the study for (consumer) sensory profiling of fermented bean-based formulations enriched with different microgreens.

	Attributes
1	Spring onion/garlic (flavor)
2	Beans (flavor)
3	Ground sweet pepper (flavor)
4	Radish (flavor)
5	Peas (flavor)
6	Basil
7	Spices
8	Leather, new shoes (flavor)
9	Salty
10	Sour
11	Bitter
12	Sweet[1]
13	Mouthburn[1]
14	Firm[1]
15	Spreadable
16	Dry[1]
17	Crumbly[1]
18	Soggy
19	Hard[1]
20	Sandy
21	Sticky (tooth packing)[1]

Note: [1]Excluded from further dimension reduction analysis.

Table S3. List of sensory attributes used in the descriptive sensory analysis of fermented bean-based product with different microgreen coatings.

Sensory attribute	Definition
Appearance	
Coating	
Color description (hue)	The actual color name or hue, such as red, blue, and so on (yellow–green).
Color intensity	The intensity or strength of color from light to dark.
Color evenness	The evenness of distribution of color, not blotchy (uneven/blotchy–even).
Cross section	
Color description (hue)	The actual color name or hue, such as red, blue, and so on (cream–brown).
Color intensity	The intensity or strength of color from light to dark.
Color evenness[1]	The evenness of distribution of color, not blotchy (uneven/blotchy–even).
Texture	
First contact with lips (cross section)	
Stickiness[1] (contact with lips)	Stickiness of the surface of the sample when touching/tapping with lips. (not sticky–very sticky).
Surface moisture	The amount of wetness on surface (the feeling of wetness on the surface of cross section) (dry–wet).
Flavor	
Overall flavor intensity	The intensity of the overall flavor of the product. (none–intensive).
Sourness	The taste stimulated by acids. (none–intensive).
Saltiness	The taste stimulated by salts, such as sodium chloride etc. (none–intensive).
Bitterness	The taste stimulated by substances, such as quinine, caffeine, etc. (none–intensive).
Cooked bean flavor[1]	The intensity of the flavor of cooked and mashed beans (none–intensive).
Mouthburn	Burning sensation in the mouth caused by depletion of water in the oral mucous caused by high concentration of some ingredients, such as salt, acid, sucrose, or alcohol (none–intensive).
Texture	
First compression between tongue and palate	
Firmness	The force required to compress sample between tongue and palate (soft/semisolid–firm).
Cohesiveness (first compression)	Amount sample deforms/strings rather than shears/cuts when compressed between tongue and palate (shears/short–deforms/cohesive).
Manipulation/breakdown	
Cohesiveness of mass[1]	Degree to which the sample holds together in a mass (none–tight mass).
Graininess[1]	Amount of individual particles in the mass (none–many particles).
Grittiness	Amount of small, hard particles between tongue and palate during manipulation (none–high).
Stickiness of mass[1]	Amount of mass that adheres to oral surfaces (none–very sticky).

Note: [1]Excluded from further dimension reduction analysis.

Table S4. **Results of Tukey's HSD multiple pairwise comparison test performed after applying ANOVA on the standardized descriptive data of evaluated fermented bean-based product coated with pea microgreen stored at 4°C for a period of 1, 10, 20, and 30 days (BP1, BP10, BP20, and BP30, respectively).**

Color description (coating)				Color intensity (coating)			Color evenness (coating)		
Subset				Subset			Subset		
1	2	3	4	1	2	3	1	2	3
BP10				BP1			BP10		
	BP1			BP10			BP1		
		BP20			BP20			BP20	
			BP30			BP30			BP30
More yellow		More green							

Color description (cross section)				Color intensity (cross section)			Surface moisture		
Subset				Subset			Subset		
1	2	3		1	2	3	1	2	
BP1				BP1			BP30		
BP10				BP10			BP20		
	BP20				BP20		BP10		
		BP30				BP30		BP1	
More cream		More brown							

Overall flavor intensity				Sourness			Saltiness		
Subset				Subset			Subset		
1	2	3	4	1	2		1	2	
BP1				BP1			BP1		
	BP10			BP10			BP10	BP10	
		BP20			BP20			BP30	
			BP30		BP30			BP20	

Bitterness				Mouthburn			Cohesiveness (first compression)		
Subset				Subset			Subset		
1	2			1	2		1	2	
BP1				BP1			BP20		
BP10				BP10			BP30		
	BP20				BP20			BP10	
	BP30				BP30			BP1	

Firmness				Grittiness	
Subset				Subset	
1	2	3	4	1	2
BP1				BP1	
	BP10			BP10	
		BP20			BP30
			BP30		BP20

Note: Homogeneous subsets are formed at a statistically significance level of 0.05. The intensity of the attributes increases going from subset 1 to subset *n*.

Table S5. Attribute loadings for the first two extracted principal components (PC1 and PC2) by applying Principal Component Analysis (PCA) to the consensus data matrix obtained by applying Generalized Procrust Analysis to the raw descriptive data of the fermented bean-based product with different microgreen coatings, stored at 4°C for a period of 1, 10, 20 and 30 days.

Sensory attributes	PC1	PC2
Appearance		
Coating		
Color description (hue)	**0.948**	0.299
Color intensity	**0.996**	0.092
Color evenness	**0.991**	0.129
Cross section		
Color description (hue)	**0.987**	–0.026
Color intensity	**0.984**	0.057
Texture		
Contact with lips (cross section)		
Surface moisture	**–0.861**	0.489
Flavor		
Overall flavor intensity	**0.991**	–0.089
Sourness	**0.985**	–0.173
Saltiness	**0.834**	–0.519
Bitterness	**0.952**	–0.183
Mouthburn	**0.985**	–0.046
Texture		
Compression between tongue and palate		
Firmness	**0.985**	–0.021
Cohesiveness (First compression)	**–0.982**	0.014
Manipulation/breakdown		
Grittiness	**0.962**	–0.075

Note: Loadings with absolute values >0.72 are in bold.