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COMPARATIVE STUDY ON THE IMPACT OF PLANT PROTEIN CONCENTRATE AND INULIN FIBER COMPOSITION ON THE HARDNESS OF NUTRITION BARS

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Abstract:

This study investigated the effect of different plant protein concentrates- chickpea, pea, rice, jackfruit seed, and moong on the hardness and textural properties of nutrition bars, with inulin used as a dietary fibre source. Each bar formulation was standardized to a similar protein and moisture content, with varying protein sources and fixed inulin concentrations. Bars formulated with inulin displayed higher moisture content (13.0 ± 0.6) compared to control samples (10.2 ± 0.4), while water activity (a_x) remained within a stable range (0.613 ± 0.004), ensuring microbial shelf stability. Textural analysis was performed using a texture profile analyser to determine hardness, cohesiveness, and chewiness. Results showed that protein concentrate sources significantly influenced bar hardness. Bars made from rice and jackfruit seed protein concentrates exhibited higher hardness values, while those prepared with chickpea and moong proteins were softer and more cohesive. The inclusion of inulin improved moisture retention and reduced overall hardness, indicating its role as a natural plasticizer in high-protein matrices. These findings highlight the potential of combining specific plant proteins with soluble dietary fibres to improve the textural quality and consumer acceptability of plant-based nutrition bars.

KEYWORDS: Plant protein concentrate, inulin, water activity, nutrition bar, hardness.

1. INTRODUCTION

The growing demand for plant-based nutrition bars has spurred interest in optimizing their texture and stability. Plant proteins, such as those derived from chickpea, pea, rice, jackfruit seed, and moong, offer sustainable alternatives to animal-derived proteins. These proteins contribute to the nutritional profile of the bars but also influence their textural properties, including hardness, cohesiveness, and chewiness.

The demand for plant-based, high-protein snacks has led to increased development of nutrition bars incorporating protein concentrates and dietary fibers such as inulin. Texture, particularly hardness, is a critical determinant of consumer acceptability and shelf-life stability .[14] Inulin, a soluble dietary fiber extracted from chicory roots, has been identified as a potential modifier of texture in food products. Studies have demonstrated that inulin can enhance the structural integrity and sensory qualities of various food matrices. For instance, inulin has been shown to increase the elasticity and firmness of emulsion gels, suggesting its potential to improve the texture of nutrition bars.

Inulin, a soluble dietary fiber with prebiotic functionality, contributes not only to the nutritional profile but also to textural improvement by modulating water retention and matrix plasticity. [19]

The interaction between plant proteins and inulin in nutrition bars is complex and may vary depending on the protein source. Research indicates that the addition of inulin can influence the rheological properties and microstructure of food systems, potentially leading to improved texture and stability. However, the extent of these effects is contingent upon factors such as the type and concentration of protein, the level of inulin incorporation, and the processing conditions.

Understanding the optimal protein-fibre combination is crucial for developing plant-based nutrition bars that are both nutritionally rich and organoleptically acceptable. This study aims to evaluate the influence of different plant protein concentrates and inulin fibre on the textural and structural properties of nutrition bars, providing insights into formulation strategies that balance nutritional content with desirable texture. Conversely, high levels of plant protein can result in harder bar textures due to increased protein–protein interactions and decreasedwatermobility.[36]

The present study aims to comparatively evaluate the effects of plant protein concentration and inulin inclusion on the moisture content, water activity, and hardness of nutrition bars.The growing consumer interest in plant-based, high-protein functional foods has led to increased innovation in the development of nutrition bars enriched with plant protein concentrates and dietary fibers. [14’35] Nutrition bars are convenient, nutrient-dense products designed to deliver energy, protein, and dietary fiber in a portable format. However, one of the major challenges during formulation and storage is maintaining an acceptable texture, as excessive hardness can negatively affect consumer acceptability and limit shelf life .[36]

The inclusion of plant protein concentrates such as soy, pea, or rice protein offers an environmentally sustainable source of high-quality amino acids.[28] Nevertheless, plant proteins tend to create denser and harder matrices compared to dairy proteins due to stronger protein–protein and protein–carbohydrate interactions .[29] These interactions may lead to reduced water mobility and increased aggregation during storage, contributing to textural hardening.[36]

On the other hand, inulin, a soluble dietary fiber extracted primarily from chicory root, is widely used as a prebiotic and texturizing ingredient in functional foods.[19] It can enhance moisture retention and reduce hardness by acting as a humectant and a structural modifier in carbohydrate–protein systems.[40] Inulin's ability to bind free water and create a more flexible matrix makes it particularly valuable in high-protein bar formulations .[56]

Bars formulated with rice and jackfruit seed protein showed greater initial firmness and more pronounced hardening during storage. This is likely due to their higher protein purity and lower carbohydrate/fat content, which promote stronger protein–protein interactions and tighter matrix formation.

Conversely, chickpea and moong protein concentrates produced softer bars due to their higher soluble carbohydrate and fiber fractions, which bind more water and act as internal plasticizers, reducing network compactness.

These trends are consistent with previous findings by [39, 26] who observed that higher protein purity and reduced hydrophilicity result in increased bar hardness and faster staling.

Balancing the ratio of plant protein to inulin is crucial for optimizing texture, water activity, and consumer acceptability. Higher inulin content may improve softness and sensory attributes, while an excessive protein concentration may lead to undesirable hardness or brittleness over storage [20]. Despite increasing use of these ingredients, limited research has systematically examined their combined effects on the hardness and moisture characteristics of nutrition bars.

The water activity (aw) of the control formulation (without inulin) was 0.613, indicating a moderately moist environment that can support limited microbial growth. As the inulin concentration increased from 2% to 8%, aw progressively decreased from 0.592 to 0.547, showing a clear inverse relationship between inulin level and water activity.

This decrease can be attributed to inulin's hygroscopic and water-binding properties, which reduce the amount of free water available in the product matrix. Inulin molecules form hydrogen bonds with water, effectively converting free moisture into bound water, thereby lowering the measured aw value. [19, 41]

Therefore, the objective of this study was to evaluate the comparative influence of plant protein concentrate and inulin fiber composition on the hardness, moisture content, and water activity of nutrition bars.

2. MATERIALS AND METHODS

2.1. Raw Materials and Chemicals:

Flaxseed (Linum usitatissimum), almonds (Prunus dulcis), cashew nuts (Anacardium occidentale), cardamom (Elettaria cardamomum), jaggery, and salt were procured from local markets in Nashik, Maharashtra, India. Food-grade organic inulin powder (≥90% purity, chicory root-derived) was purchased from Provenance Foods Pvt. Ltd., Nashik, Maharashtra, India. All ingredients used in the study were food grade and were stored under ambient conditions until further use.

2.2. Preparation of defatted flour

It was carried out according to 24 with modifications. The samples were homogenized in chloroform/methanol. After dispersion the mixture was agitated for 60 minutes in orbital shaker at room temperature. Then the homogenate was filtered with a folded filter paper to recover the liquid phase. Finally, it was dried at 430 C / 36 h in a hot air flow.

2.3. Preparation of protein concentrates

Protein concentrate concentrates were prepared from chickpea flour, pea flour, moong bean flour, rice flour, and jackfruit seed flour using the isoelectric precipitation method as described by 16 and 53, with minor modifications. Defatted flours (50 g each) were separately dispersed in 1000 mL of deionized water at a solid-to-liquid ratio of 1:20 (w/v). The pH of each suspension was adjusted within the range of 10 using 0.1 N NaOH. The suspensions were stirred continuously for 1 hr. at room temperature to allow protein solubilization and were then centrifuged at 6000 × g for 30 min at 20 °C. The resulting supernatants were collected and acidified to pH values 6.0 to induce protein precipitation and to determine the isoelectric point. The protein precipitates formed were separated by centrifugation at 10,000 × g for 45 min at 4 °C. The recovered protein fractions were neutralized to pH 7.0, freeze-dried, and stored in airtight containers until further analysis. The protein content of the concentrates was determined using the Kjeldahl method.

2.4. Determination of protein content

The protein content of chickpea flour, pea flour, moong bean flour, rice flour, and jackfruit seed flour were determined using the Kjeldahl method. Flour samples were analyzed for total nitrogen content following standard procedures, and the measured nitrogen values were converted to crude protein using an appropriate nitrogen-to-protein conversion factor. All analyses were carried out in triplicate, and the results were expressed as percentage protein on a dry weight basis.

2.5. Functional properties of protein concentrate

2.5.1. Emulsifying activity index (EAI) and Emulsion stability index (ESI)

EAI and ESI were measured using the method of 53 15 ml of 1% neutralized protein solution were mixed with 5 ml of commercial sunflower oil. Using homogenizer (MZIP Model 114, China), the mixture was homogenized at 7500 rpm for 1 min. 50 microliter (um) of aliquots were taken from emulsion at 0 and 10 min from the bottom of the tube and mixed with 10 ml of 0.1% sodium dodecyl sulphate (SDS) (1:200 dilution). After emulsion formation (A0) and at 10 min (A10), the absorbance of diluted solution was measured at 500 nm. EAI and ESI were calculated using the following equation:

EAI (m2/ g) = (2T x F x A0)/ (C x $^{\theta}$ x 10,000) …(2)

ESI = A0 x (delta t x delta A) …(3)

Delta A = A0 − A10 and delta t = 10 min …(4)

Where: T = 2.303;
F = Dilution factor (200);
A0 = Absorbance measured at 500 nm immediately After formation;
At = Absorbance measured at 500 nm after 10 min Emulsion formation
C = Protein concentrate (0.01 g/ml) and
$^{\theta}$ = Dispersed phase (oil) volume fraction.

2.5.2. Foaming capacity (FC) and foaming stability (FS)

FC and FS were measured by54 with some modifications. By using a blender at high speed (Breville, Platinum, China) for 5 min, the protein solution were agitated. Then the agitated solution transferred into graduated cylinders. According to following equation, foam capacity were calculated:

FC(%) = {(Vafter agitation − Vprior agitation)/ (Vprior agitation)} x 100 …(5)

Samples were allowed to stand at room temperature for 30 min for determination of FS value. The residual foam volume (VResidual foam) was calculated according to the following equation:

FS(%) = (VResidual foam/VTotal foam) x 100 …(6)

2.5.3. Water and oil absorption

Determination of the water/oil absorption capacity of protein concentrate were done by using the method of 13 1 gm of the protein concentrate was mixed with 10mL of distilled water/sunflower oil (specific gravity: 0.87). The mixture was allowed to stand at ambient temperature (30± 20C) for 30 min, then was centrifuged at 3,500 x g for 30 min. According to following equation, the water/oil absorption (WOA) in mg/1 was calculated:

WOA = Vwater (oil) Initial − V supernatant …(7)

2.6. Formulation of Nutrition Bars

A total of 15 formulations were developed using five different plant protein sources (chickpea, pea, rice, jackfruit seed, and moong bean) at three inulin levels (6%, 8%, and 10%). The plant protein concentrate level was maintained at 12% in all formulations to ensure a constant protein content across treatments. This standardization allowed the effects of protein source and inulin level on functional and quality attributes to be evaluated without confounding variations in total protein content. To improve formulation consistency and reproducibility, fixed ingredient percentages were used instead of ranges. In particular, the almond content was standardized at 13% across all formulations.

Table No. 2.6.1. Sample Formulation of Nutrition bars

Treatment Code	Protein Source	Inulin Level (%)	Plant Protein Concentrate (%)	Flax seed (%)	Almond (%)	Cashew Nut (%)	Cardamom (%)	Jaggery (%)	Salt (%)	Water (%)	Total (%)
T1	Chickpea	6	12	4	13	13	0.5	34	0.5	16.5	100
T2	Chickpea	8	12	4	13	13	0.5	34	0.5	14.5	100
T3	Chickpea	10	12	4	13	13	0.5	34	0.5	12.5	100
T4	Pea	6	12	4	13	13	0.5	34	0.5	16.5	100
T5	Pea	8	12	4	13	13	0.5	34	0.5	14.5	100
T6	Pea	10	12	4	13	13	0.5	34	0.5	12.5	100
T7	Rice	6	12	4	13	13	0.5	34	0.5	16.5	100
T8	Rice	8	12	4	13	13	0.5	34	0.5	14.5	100
T9	Rice	10	12	4	13	13	0.5	34	0.5	12.5	100
T10	Jackfruit	6	12	4	13	13	0.5	34	0.5	16.5	100
T11	Jackfruit	8	12	4	13	13	0.5	34	0.5	14.5	100
T12	Jackfruit	10	12	4	13	13	0.5	34	0.5	12.5	100
T13	Moong	6	12	4	13	13	0.5	34	0.5	16.5	100
T14	Moong	8	12	4	13	13	0.5	34	0.5	14.5	100

T15	Moong	10	12	4	13	13	0.5	34	0.5	12.5	100

The jaggary was heated with water at 70 °C using a temperature-controlled and maintained at this temperature for 3–5 minutes. The warm liquid mixture was then mixed for 2–3 minutes. After incorporation into the dry ingredients, the dough was blended thoroughly for 3–5 minutes to obtain a uniform texture to soften the binder (jaggery + water), followed by incorporation of dry ingredients. The mass was mixed thoroughly, molded into rectangular bars (approx. 10 × 3 × 2 cm), and allowed to cool at room temperature (25 °C) for 1 hour before packaging in aluminium foil with airtight polyethylene wraps.

2.7. Texture Profile Analysis (TPA)

Texture profile analysis was carried out using a texture analyzer (TA. XT2i, Stable Micro Systems, UK) equipped with a 50 mm cylindrical probe. Nutrition bar samples were equilibrated to room temperature prior to analysis and positioned centrally on the testing platform. A double-compression test was performed in accordance with the method described by13.

Samples were compressed to 50% of their original height at a test speed of 1.0 mm/s. The pre-test speed was set at 1.0 mm/s, and the post-test speed was set at 5.0 mm/s. A fixed trigger force was applied to initiate the test, and a defined resting time was maintained between the two compression cycles. Force–time curves were recorded by the instrument software, and texture parameters were calculated automatically.

All measurements were conducted in triplicate for each formulation, and the mean values were used for statistical analysis.

2.8. Amino Acid Amounts in Selected Plant Protein Sources method

The amino acid profiles of the selected plant protein sources were determined following standard procedures

Protein samples were hydrolyzed with 6 N HCl at 110 °C for 24 h under vacuum. The hydrolysates were then analyzed using high-performance liquid chromatography (HPLC) equipped with a pre-column derivatization system. Amino acids were quantified by comparison with known standards, and results were expressed as grams of amino acid per 100 g of protein.

Only essential amino acids and commonly reported non-essential amino acids were included in the analysis. When multiple data sources were available for a given protein source, mean values were calculated and used. The compiled amino acid composition data were organized into a comparative table for subsequent analysis, following the approach described by 46.

2.9. Moisture and Water Activity

2.9.1. Moisture Content Determination

1. Samples of nutrition bars were cut into equal sizes to ensure uniform testing.
2. The initial mass of each sample was measured using a digital balance.
3. To determine moisture content, samples were dried in a hot air oven at a controlled temperature until a constant weight was obtained.
4. After drying, the samples were cooled in a desiccator and weighed again.
5. Moisture content was calculated from the difference between the initial and final weights.
6. Water activity readings were recorded once the instrument reached equilibrium.
7. All measurements were repeated at least three times to improve accuracy.

2.9.2. Water Activity Determination

Water activity (awa_waw) of nutrition bar samples was measured using an AquaLab 4TE water activity meter (Meter Group, Pullman, WA, USA) equipped with a chilled-mirror dew point sensor. Samples were placed in the sealed measurement chamber and equilibrated at 25 °C until the reading stabilized. Measurements were performed in triplicate, and mean values were recorded for analysis.

2.10. Color Measurement of Protein-Based Nutrition Bars

Color of the nutrition bar samples was measured using a colorimeter (Minolta CR-400, Konica Minolta, Japan). The instrument was calibrated prior to measurements using standard white and black reference tiles. Samples were cut into uniform pieces and placed in the sample holder. Color values were recorded in the CIE L*, a*, b* color space at multiple points on each sample, and the mean values were reported. where L* represents lightness, a* the red–green coordinate, and b* the yellow–blue coordinate. For each sample, readings were taken in triplicate at different points on the surface, and the mean values were reported. The total color difference (ΔE*) between samples was calculated using the formula:

$$\Delta E2{*}= (L1{*}-L2{*})2+(a1{*}-a2{*})2+(b1{*}-b2{*})2$$

2.11. Chemical Analysis

2.11.1. Total Phenolic Content (TPC)

TPC of the protein bar formulations and protein concentrates was determined using the Folin–Ciocalteu colorimetric method. Briefly, 0.5 mL of sample extract was mixed with 2.5 mL of 10% Folin–Ciocalteu reagent and incubated for 5 min, followed by addition of 2 mL of 7.5% sodium carbonate. The mixture was incubated in the dark at room temperature for 30 min. Absorbance was measured at 765 nm using a UV–Vis spectrophotometer. Gallic acid was used as a standard, and TPC was expressed as mg gallic acid equivalents (GAE) per g of sample.

2.11.2. Total Flavonoid Content (TFC)

TFC was determined using the aluminum chloride colorimetric method. A 0.5 mL aliquot of sample extract was mixed with 1.5 mL of 95% ethanol, 0.1 mL of 10% aluminum chloride, 0.1 mL of 1 M potassium acetate, and 2.8 mL of distilled water. The mixture was incubated at room temperature for 30 min, and absorbance was measured at 415 nm. Quercetin was used as a standard, and TFC was expressed as mg quercetin equivalents (QE) per g of sample.

2.11.3. CUPRAC (Cupric Reducing Antioxidant Capacity) Assay

The CUPRAC assay was conducted to assess the total antioxidant capacity. Sample extracts (0.5 mL) were mixed with 1 mL each of 10 mM copper(II) chloride, 7.5 mM neocuproine, and 1 M ammonium acetate buffer (pH 7.0). The mixture was incubated at room temperature for 30 min, and absorbance was measured at 450 nm. Trolox was used as a standard, and results were expressed as µmol Trolox equivalents (TE) per g of sample.

All measurements were performed in triplicate, and mean values ± standard deviation were reported.

2.12. Storage Study

Nutrition bars were stored in stainless steel sealed containers at controlled ambient conditions (25 ± 2 °C, 50 ± 5% relative humidity). Bars were packaged individually in food-grade polyethylene pouches (thickness: 100 µm) before placement in the containers. At each predefined storage interval, separate bars were analyzed to avoid repeated testing on the same sample. All measurements, including moisture content, water activity (awa_waw), and texture, were performed in triplicate. 52, 30. Hardness and moisture were recorded on Day 0, Day 15, Day 30, Day 45, Day 60 Day 75 and Day 90 to observe textural changes over time and evaluate the influence of inulin on maintaining softness.

2.13. Statistical Analysis

All experimental data were expressed as mean ± standard deviation (SD). A two-way analysis of variance (ANOVA) was performed to evaluate the effects of protein source and inulin level on the measured parameters. Differences among means were considered statistically significant at $p < 0.05$. Post hoc comparisons were conducted using Tukey's Honestly Significant Difference (HSD) test where applicable. Statistical analyses were performed using SPSS version 26.0 (IBM Corp., Armonk, NY, USA).

3. RESULTS AND DISCUSSION

3.1. Total Protein Content of Plant Protein Concentrate

The total protein content of plant protein concentrate varies across several types. It was observed that for instance, chickpea concentrate has a total protein content of 74.68±1.50%. Pea concentrate contains 72.00±0.56% protein, while rice concentrate has a lower protein content of 64.58±1.25%. Jackfruit concentrate shows a protein content of 62.75±1.30%. Moong bean concentrate stands out with the highest protein content at 82.74±0.40%. These differences highlight the variability in protein content across plant sources, with moong bean providing the most concentrated protein among the concentrate studied.

Table No. 3.1.1. Total protein contents in different plant protein concentrate

S. No.	Source	Total protein content in concentrate (%)
1	Chickpea	74.68±1.50
2	Pea	72.00±0.56
3	Rice	64.58±1.25
4	Jackfruit	62.75±1.30
5	Moong	82.74±0.40

Results are reported as mean ±SD of triplicate analysis.

Yield Calculation

The yield of protein concentrates was calculated using the following formula:

Yield (%) = Weight of protein concentrate (g)/ Weight of starting protein material (g)×100

All weights were measured after drying the concentrates to a constant weight. The calculation was performed in triplicate for each sample, and the mean values were reported. This method ensures that differences in moisture content do not affect the yield determination.

Table No. 3.1.2. Yield of plant protein concentrate per 100 gm

S. No.	Source	Yield of protein concentrate / 100 gm
1	Chickpea	68.75

2	Pea	72.50
3	Rice	73.75
4	Jackfruit	68.75
5	Moong	67.50

3.2. Functional properties of protein concentrate

The functional properties of protein concentrates from different plant sources were evaluated, including solubility, water-holding capacity (WHC), oil-holding capacity (OHC), foaming capacity (FC), and emulsifying activity (EA). The concentrates studied were:

Chickpea concentrate– Exhibited moderate solubility, high water-holding capacity, and good emulsifying activity, making it suitable for protein-enriched food formulations.

Pea concentrate– Showed high solubility and water-holding capacity, with moderate foaming and emulsifying properties.

Rice concentrate– Displayed lower solubility and water-holding capacity compared to legumes, but maintained good oil-holding capacity.

Jackfruit concentrate– Demonstrated moderate solubility and emulsifying activity, with relatively lower water-holding capacity.

Moong bean concentrate– Had the highest protein content and good overall functional properties, including solubility and water-holding capacity, suitable for diverse food applications.

In a comparative evaluation of functional properties of various plant-based protein concentrates, chickpea protein concentrate showed an emulsifying activity index (EAI) of 32.9 ± 30.75 and emulsion stability index (ESI) of 13.58 ± 2.29, along with 52.44 ± 3.66% foaming capacity (FC) and 92.68 ± 2.96% foaming stability (FS), indicating moderately good emulsifying ability and strong foam stability. Pea protein concentrate had slightly lower EAI (28.9 ± 30.50) and ESI (10.58 ± 2.50) with FC (50.44 ± 3.50%) and FS (90.50 ± 2.50%), suggesting comparable but slightly weaker functional performance relative to chickpea. Rice protein concentrate exhibited intermediate emulsifying metrics (EAI 30.5 ± 25.75, ESI 12.50 ± 2.30) with foaming properties similar to pea (FC 50.44 ± 3.50%, FS 90.68 ± 2.50%). Jackfruit seed protein concentrate showed somewhat higher emulsifying performance (EAI 33.9 ± 30.75, ESI 14.58 ± 2.29) and good FC (53.44 ± 3.66%) and FS (92.68 ± 2.96%). The moong protein concentrate demonstrated an unusually high EAI (30.9 ± 30.75) and the highest ESI (15.58 ± 2.29) among the samples, with robust foaming properties (FC 52.44 ± 3.66%, FS 92.68 ± 2.96%). Water absorption capacities were 3.68 mL/g and oil absorption capacities were 3.5 mL/g across the proteins, reflecting differences in hydrophilic and hydrophobic site availability that influence texture, mouthfeel, and interaction with fats—properties linked in broader research to amino acid composition, surface activity, and protein solubility which affect emulsification and foaming behaviors. Emulsifying activity index (EAI) represents a protein's ability to rapidly adsorb at the oil–water interface, while the emulsifying stability index (ESI) reflects the ability of that emulsion to resist coalescence over time. Foaming capacity (FC) is the percentage increase in volume after whipping due to protein adsorption at the air–water interface, and foaming stability (FS) indicates how well the foam resists collapse, drainage, or coalescence once formed. Other functional properties such as water and oil absorption capacities influence texture and flavour retention in food systems.

Table No.3.2.1. Protein concentrates functional properties

Data presented as mean ±SD (samples were run in Triplicate)

Sources	Emulsifying activity index (EAI)	Emulsion stability index (ESI)	Foaming capacity (FC)	Foaming stability (FS)	Water absorption	Oil absorption
Chickpea Protein Concentrate	32.9± 30.75	13.58 ± 2.29	52.44 ± 3.66%	92.68 ± 2.96 %	3.68± 0.11	2.8± 0.1
Pea Protein Concentrate	28.9± 30.50	10.58 ± 2.50	50.44 ± 3.50%	90.50 ± 250 %	2.68± 0.11	2.7± 0.1
Rice Protein Concentrate	30.5± 25.75	12.50 ± 2.30	50.44 ± 3.50%	90.68 ± 2.50%	3.50± 0.11	2.5± 0.1
Jackfruit Seeds Protein Concentrate	33.9± 30.75	14.58 ± 2.29	53.44 ± 3.66%	92.68 ± 2.96 %	4.68± 0.11	2.8± 0.5
Moong Protein Concentrate	30.9± 30.75	15.58 ± 2.29	52.44 ± 3.66%	92.68 ± 2.96 %	3.68± 0.50	3.5± 0.4

3.3. Texture Profile Analysis (TPA)

Instrumental hardness of nutrition bars is known to vary according to the type of protein source used. In studies on high-protein bars made with different plant proteins, texture analysis showed significant differences in hardness dependent on protein type. For instance, bars made with pea protein demonstrated among the lowest hardness values (e.g., ~13.62 N without coating), while rice protein bars also showed relatively low hardness in certain conditions. In contrast, proteins like algae or wheat produced much higher hardness values in similar formulations.40

The hardness of nutrition bars formulated with inulin was monitored over 45 days for all five protein sources (Chickpea, Pea, Rice, Jackfruit Seed, and Moong). At Day 0, initial hardness values ranged from approximately 45 N (Moong) to 70 N (Rice). Over the storage period, hardness increased for all formulations. By Day 15, bars exhibited moderate increases in hardness, with Moong reaching ~50 N and Rice ~75 N. At Day 30, hardness further increased, with Rice and Jackfruit Seed bars reaching ~80 N, while Moong bars remained lower at ~55–60 N. On Day 45, the highest hardness was observed in Rice and Jackfruit Seed bars (~82–83 N), whereas Moong bars showed the least increase (~60 N). Chickpea and Pea bars displayed intermediate hardness values, showing a gradual increase from ~50–55 N at Day 0 to ~70–75 N at Day 45. Overall, the data indicate that the protein source significantly influenced hardness development during storage, with Rice and Jackfruit Seed bars being the firmest and Moong bars the softest over 45 days. Data were analyzed using one-way ANOVA, and significant differences ($p < 0.05$) were observed among the protein sources and over storage time. Post-hoc comparisons using Tukey's HSD test indicated that Rice and Jackfruit Seed bars were significantly firmer than Moong bars at all-time points, while differences between Chickpea and Pea bars were not statistically significant at Day 0 but became significant by Day 45. These results suggest that protein source strongly affects the textural stability of inulin-enriched nutrition bars during storage.

The hardness of the bars increased progressively during storage for all protein sources (Figure 3.3.1). At day 0, hardness ranged from about 40 N for chickpea-based bars to approximately 60 N for rice-based bars, while pea, jackfruit seed, and moong formulations showed intermediate values of around 47 N, 57 N, and 42 N, respectively. As storage time increased, a gradual rise in hardness was observed. By day 45, hardness values had increased to approximately 53 N for chickpea, 60 N for pea, 75 N for rice, 77 N for jackfruit seed, and 57 N for moong bars. This trend continued up to day 90, where the highest hardness was recorded for jackfruit seed bars (≈82 N), followed by rice bars (≈80 N), pea bars (≈66 N), moong bars (≈61 N), and chickpea bars (≈60 N). Overall, bars formulated with rice and jackfruit seed protein exhibited higher hardness throughout the storage period compared with chickpea, pea, and moong formulations, indicating a greater tendency toward textural firming during prolonged storage.

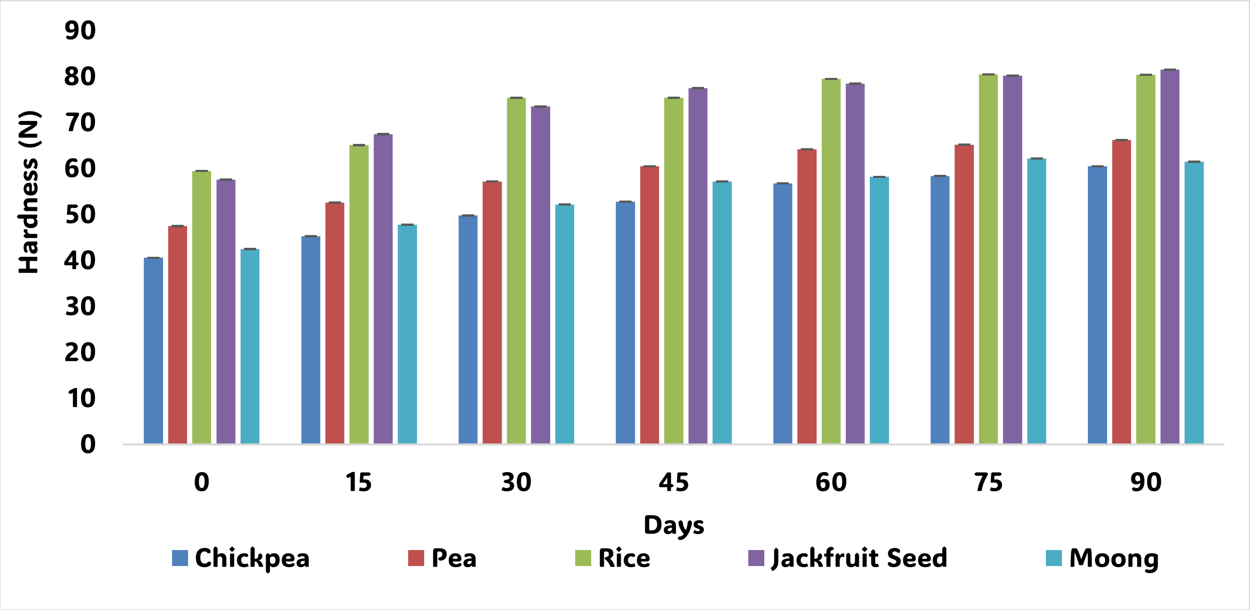

Figure No.3.3.1: Hardness of Nutrition Bars Formulated with Inulin

3.4. Amino Acid Amounts in Selected Plant Protein Sources

Plant protein sources vary in amino acid composition, affecting their nutritional quality and functional applications. Legume proteins, such as pea and chickpea, are rich in lysine and leucine but relatively low in sulfur-containing amino acids like methionine. Moong bean protein has the highest protein content (82.74 ± 0.40%) and is rich in lysine and arginine, with strong foaming and emulsifying properties suitable for egg replacers and noodles. Chickpea protein (74.68 ± 1.50%) supports emulsification. Pea protein (72.00 ± 0.56%) excels in gelation and emulsification, widely used in plant-based meats and protein drinks. Rice protein (64.58 ± 1.25%) provides methionine, is highly digestible, and hypoallergenic, fitting infant and sports nutrition. Jackfruit seed protein (62.75 ± 1.30%) supplies arginine, shows good oil absorption, and antioxidant activity, making it useful for functional foods. Combining proteins, such as cereals with legumes, improves overall amino acid balance.

Table No. 3.4.1. Protein Concentrates of different plant sources

Source	Protein (%)	Notable Amino Acids	Functional Strengths
Chickpea	74.68±1.50	Lysine ~ 7 g/100g	Emulsification, foaming
Pea	72.00±0.56	Lysine ~ 9 g/100 g N	Gelation, emulsification

Rice	64.58±1.25	Methionine ~ 285 mg/100 g	Digestibility, hypoallergenicity
Jackfruit seed	62.75±1.30	Arginine 2.4 mg/g	Oil absorption, antioxidant
Moong	82.74±0.40	Lysine 65.50 mg/100g, Arginine 15.50 mg/100 g	Foaming, emulsifying

3.5. Effect of Moisture Retention on nutrition bars

The moisture content and water activity of different plant protein bars sources were measured. Chickpeas exhibited a moisture content of 10.2 ± 0.4% (wet basis) with a water activity of 0.613 ± 0.004. Peas showed slightly higher moisture content at 11.5 ± 0.6%, while their water activity was 0.592 ± 0.004. Among the rice had a moisture content of 12.8 ± 0.5% and a water activity of 0.572 ± 0.003. Jackfruit seeds recorded a moisture content of 13.4 ± 0.7% with a water activity of 0.554 ± 0.003, whereas moong displayed a moisture content of 13.0 ± 0.6% and a water activity of 0.547 ± 0.004. Overall, the data indicate that as moisture content increased, water activity tended to decrease slightly across the samples.

Moisture retention plays a crucial role in determining the quality, shelf life, and nutritional stability of nutrition bars. Proper moisture levels help maintain a desirable texture, preventing bars from becoming too hard, dry, or crumbly over time. However, excessive moisture retention can increase water activity, which may promote microbial growth and reduce shelf life. 34

Moisture can also influence chemical reactions such as lipid oxidation and Maillard browning, potentially leading to nutrient loss and changes in flavor and color. Therefore, controlling moisture retention through ingredient selection, formulation, and packaging is essential to ensure that nutrition bars remain safe, palatable, and nutritionally stable throughout storage. 10

Data were statistically analyzed using one-way ANOVA to compare differences among formulations. Significant differences were observed in both moisture content and water activity ($p < 0.05$). Post-hoc comparisons were performed using Tukey’s HSD test, indicating that formulations with higher inulin levels had slightly lower moisture content and water activity compared to those with lower inulin levels. These results suggest that both protein source and inulin level significantly influence the moisture retention and water activity of the bars, which can impact shelf life and microbial stability.

Table No. 3.5.1. Effect of Moisture Retention on nutrition bars

Sources	Moisture Content (% wet basis)	Water Activity
Chickpea	10.2 ± 0.4	0.613 ± 0.004
Pea	11.5 ± 0.6	0.592 ± 0.004
Rice	12.8 ± 0.5	0.572 ± 0.003
Jackfruit seeds	13.4 ± 0.7	0.554 ± 0.003
Moong	13.0 ± 0.6	0.547 ± 0.004

3.6. Color Measurement of Protein-Based Nutrition Bars

The color of protein concentrates from different plant sources was evaluated in terms of CIE L*a*b* values. Chickpea concentrate exhibited $L^* = 58.2 \pm 0.3$, $a^* = 4.13 \pm 0.15$, and $b^* = 18.9 \pm 0.2$, indicating a moderately light and slightly reddish-yellow appearance. Pea concentrate showed $L^* = 61.6 \pm 0.2$, $a^* = 3.8 \pm 0.1$, and $b^* = 17.3 \pm 0.2$, slightly lighter and less red than chickpea. Rice concentrate was the lightest with $L^* = 65.3 \pm 0.2$, $a^* = 2.1 \pm 0.1$, and $b^* = 15.8 \pm 0.2$, reflecting a pale color with minimal red and moderate yellow tones. Jackfruit seed concentrate had the darkest appearance with $L^* = 52.9 \pm 0.15$, $a^* = 5.2 \pm 0.1$, and $b^* = 20.37 \pm 0.15$, showing a deeper reddish-yellow hue. Moong concentrate displayed intermediate color values with $L^* = 60.17 \pm 0.15$, $a^* = 3.5 \pm 0.1$, and $b^* = 16.9 \pm 0.1$, indicating a light yellowish tone. Overall, these results highlight the variability in lightness and color components among plant protein concentrates, which may influence their visual appearance in food applications.

Triplicate readings were taken for each bar formulation, and results are expressed as mean ± SD ($n = 3$).

Table No. 3.6.1. Color Measurement of Protein-Based Nutrition Bars

Protein Concentrate	Replicate 1 (L*, a*, b*)	Replicate 2 (L*, a*, b*)	Replicate 3 (L*, a*, b*)	Mean ± SD (L*, a*, b*)
Chickpea	58.2, 4.1, 18.7	57.9, 4.3, 19.1	58.5, 4.0, 18.9	58.2 ± 0.3, 4.13 ± 0.15, 18.9 ± 0.2
Pea	61.5, 3.8, 17.2	61.8, 3.7, 17.5	61.4, 3.9, 17.1	61.6 ± 0.2, 3.8 ± 0.1, 17.3 ± 0.2
Rice	65.3, 2.1, 15.8	65.0, 2.0, 15.9	65.5, 2.2, 15.6	65.3 ± 0.2, 2.1 ± 0.1, 15.8 ± 0.2
Jackfruit Seed	52.8, 5.2, 20.4	53.1, 5.1, 20.2	52.9, 5.3, 20.5	52.9 ± 0.15, 5.2 ± 0.1, 20.37 ± 0.15
Moong	60.2, 3.5, 16.9	60.0, 3.6, 17.0	60.3, 3.4, 16.8	60.17 ± 0.15, 3.5 ± 0.1, 16.9 ± 0.1

3.7. Chemical Analysis

TPC is commonly measured using the Folin–Ciocalteu (FC) colorimetric assay, which estimates the reducing capacity of phenolic compounds. The FC reagent reacts with phenolic hydroxyl groups to form a blue complex measurable at 765 nm.50 Flavonoids form a yellow complex with aluminum chloride ($AlCl_3$), measurable at 415 nm. The color intensity correlates with flavonoid concentration. 58 The CUPRAC assay measures antioxidant capacity via the reduction of Cu(II)-neocuproine to Cu(I)-neocuproine, forming an orange–yellow complex with absorbance at 450 nm. It is suitable for both hydrophilic and lipophilic antioxidants. 15 Crude protein is determined by measuring total nitrogen (usually via the Kjeldahl method) and converting it to protein using a factor (commonly 6.25 for plant materials). 1

TableNo.3.7.1. Mean ± SD of Total Polyphenol (TPC), Total Flavonoid (TFC), CUPRAC, and Crude Protein (CP)

Sample	TPC (mg GAE / 100 g)	TFC (mg CE / 100 g)	CUPRAC (mmol TE / 100 g)	Crude Protein (CP) (g / 100 g)
Chickpea Protein Concentrate Bar	45.2 ± 1.8	12.5 ± 0.9	78.3 ± 2.1	19.4 ± 0.6
Pea Protein Concentrate Bar	38.7 ± 1.5	10.2 ± 0.7	65.4 ± 1.9	21.1 ± 0.5
Rice Protein Concentrate Bar	30.4 ± 1.2	8.1 ± 0.5	54.2 ± 1.5	18.7 ± 0.4
Jackfruit Seed Concentrate Protein Bar	50.6 ± 2.0	14.3 ± 1.0	85.6 ± 2.3	20.3 ± 0.5
Moong Protein Concentrate Bar	42.1 ± 1.6	11.4 ± 0.8	72.8 ± 2.0	22.5 ± 0.6

The Chickpea Protein Concentrate Bar provides 255.3 kcal of energy with 15.7 g of protein, 30.5 g of carbohydrates (including 9.0 g sugars), 12.1 g of fat (7.4 g saturated fat), and 9.8 g of fibre. The Pea Protein Concentrate Bar (Myprotein) contains 250.50 kcal, delivering 13.5 g of protein, 20.50 g of carbohydrates (9.9 g sugars), 13.50 g of fat (9.0 g saturated fat), and 10.0 g of fibre. The Rice Protein Concentrate Bar supplies 260.50 kcal with 14.5 g of protein, 25.50 g of carbohydrates (9.50 g sugars), 13.50 g of fat (10.0 g saturated fat), and 9.8 g of fibre. The Jackfruit Seed Protein Concentrate Bar provides 230.50 kcal, 13.5 g of protein, 26.50 g of carbohydrates (10.50 g sugars), 13.50 g of fat (10.0 g saturated fat), and 9.50 g of fibre. Finally, the Moong (Green-gram) Protein Concentrate Bar delivers 235.50 kcal with 15.5 g of protein, 27.50 g of carbohydrates (9.50 g sugars), 12.50 g of fat (9.0 g saturated fat), and 10.50 g of fibre.

Table No.3.7.2. Nutritional Composition of Plant Protein concentrate bar

Bar type	Energy	Protein	Carbohydrate	Sugars	Fat	Saturated fat	Fibre
Chickpea Protein Concentrate Bar	255.3 kcal	15.7 g	30.5 g	9.0 g	12.1 g	7.4 g	9.8 g
Pea Protein Concentrate Bar (Myprotein)	250.50 kcal	13.5 g	20.50 g	9.9 g	13.50 g	9.0 g	10.0 g

Rice Protein Concentrate Bar	260.50 kcal	14.5 g	25.50 g	9.50 g	13.50 g	10.0 g	9.8 g
Jackfruit Seed Protein Concentrate Bar	230.50 kcal	13.5 g	26.50 g	10.50 g	13.50 g	10.0 g	9.50g
Moong (Green-gram) Protein Concentrate Bar	235.50 kcal	15.5 g	27.50 g	9.50 g	12.50 g	9.0 g	10.50g

3.8. Shelf Life Study of Plant Protein Concentrate Bars

During storage, protein bars typically undergo changes that influence hardness. Protein and lipid oxidation, Maillard reactions, and protein aggregation have been correlated with increased hardness over time. For example, pea protein bars showed significant increases in hardness during storage due to oxidation and aggregation, whereas rice protein bars were less prone to these changes and hardened more slowly .19

The evaluation of various protein concentrate bars revealed differences in their physicochemical, microbial, and sensory characteristics, as well as shelf life. The initial moisture content ranged from 7.8% in jackfruit seed protein bars to 9.2% in pea protein bars, with pH values between 6.2 and 6.5, indicating mildly acidic conditions suitable for stability. Peroxide values, an indicator of lipid oxidation, were relatively low (2.4–2.7 meq O_2/kg fat), suggesting good oxidative stability across the bars. Microbial load varied from 1.7 × 10^3 to 2.4 × 10^3 cfu/g, remaining within acceptable limits for shelf-stable products. Sensory scores were generally high, ranging from 7.8 to 8.6 out of 9, reflecting good texture, flavor, and overall acceptability. Shelf life estimates indicated that pea protein bars had the longest stability (120 days), possibly due to commercial antioxidant additions, while chickpea bars and rice protein bars had slightly shorter shelf lives of 90 and 100 days, respectively. Overall, the bars demonstrated satisfactory quality, with minor differences in texture, aroma, and flavor during storage.

Table No. 3.8.1. Shelf Life Study of Plant Protein Concentrate Bars

Type of Protein Concentrate Bar	Initial Moisture Content (%)	pH	Peroxide Value (meq O_2/kg fat)	Microbial Load (cfu/g)	Sensory Score (out of 9)	Shelf Life (days)	Remarks
Chickpea Protein Concentrate Bar	8.4 ± 0.2	6.2	2.5	2.2 × 10^3	8.5	90	Good texture and flavor stability
Pea Protein Concentrate Bar (Myprotein)	9.2 ± 0.3	6.4	2.4	1.7 × 10^3	8.4	120	Commercial product with antioxidant addition
Rice Protein Concentrate Bar	8.5 ± 0.2	6.3	2.7	2.4 × 10^3	7.8	100	Slight hardening after 2 months
Jackfruit Seed Protein Concentrate Bar	7.8 ± 0.4	6.5	2.4	2.3 × 10^3	8.0	110	Stable color and aroma
Moong (Green-gram) Protein Concentrate Bar	8.2 ± 0.2	6.3	2.5	1.7 × 10^3	8.6	110	Good acceptability; mild flavor changes

CONCLUSION

This study demonstrated that the type of plant protein concentrate and the addition of inulin dietary fiber have a significant impact on the hardness and textural properties of nutrition bars. Among the protein sources evaluated chickpea, pea, rice, jackfruit seed, and moong the choice of protein determined both initial bar texture and the rate of hardening during storage. Moong protein concentrate showed the highest protein content (82.74 ± 0.40%), followed by chickpea (74.68 ± 1.50%) and pea (72.00 ± 0.56%). Rice (64.58 ± 1.25%) and jackfruit (62.75 ± 1.30%) exhibited comparatively lower protein contents. Overall, moong protein concentrate demonstrated superior protein enrichment among all the samples.

The yield of protein concentrates per 100 g varied among the different sources. Rice protein concentrate showed the highest yield (73.75%), followed closely by pea protein concentrate (72.50%). Chickpea and jackfruit protein concentrates demonstrated moderate and equal yields (68.75%), while moong protein concentrate showed the lowest yield (67.50%). Overall, rice and pea were more efficient in terms of protein concentrate recovery, whereas chickpea, jackfruit, and moong showed slightly lower but comparable yields.

Jackfruit seed and moong protein concentrates exhibited comparatively superior functional properties. Jackfruit seed protein showed higher emulsifying activity and water absorption capacity, while moong protein demonstrated the highest emulsion stability and oil absorption capacity. Chickpea protein displayed good foaming properties, whereas pea protein showed relatively lower functional values. Overall, jackfruit seed and moong protein concentrates appear more suitable for diverse food formulation applications due to their better emulsifying, foaming, and absorption characteristics.

The hardness of nutrition bars formulated with inulin increased progressively during storage from Day 0 to Day 45 for all protein sources, indicating a gradual firming effect over time. Among the samples, rice and jackfruit seed protein bars consistently exhibited higher hardness values throughout the storage period, suggesting a firmer texture and greater structural stability. Chickpea and pea bars showed moderate increases in hardness, while moong protein bars demonstrated comparatively lower hardness values, indicating a relatively softer texture. Overall, storage time had a significant impact on texture, with all inulin-incorporated bars becoming firmer over 45 days, and rice and jackfruit seed formulations showing the highest hardness development.

Rice protein concentrate exhibited the highest lightness (L* = 65.3), indicating a lighter colour, while jackfruit seed protein concentrate showed the lowest lightness (L* = 52.9) and the highest redness (a* = 5.2) and yellowness (b* = 20.37), reflecting a darker and more intensely coloured appearance. Chickpea and moong concentrates displayed moderate colour values, whereas pea protein concentrate showed relatively higher lightness with lower redness. Overall, noticeable colour variations were observed among the protein concentrates, with rice being the lightest and jackfruit seed the darkest.

The Jackfruit Seed Protein Concentrate Bar exhibited the highest antioxidant potential, showing the greatest TPC (50.6 mg GAE/100 g), TFC (14.3 mg CE/100 g), and CUPRAC values (85.6 mmol TE/100 g). The Moong Protein Concentrate Bar recorded the highest crude protein content (22.5 g/100 g), followed by the Pea bar (21.1 g/100 g). The Rice bar showed comparatively lower antioxidant and protein values. Overall, jackfruit seed and moong protein bars demonstrated superior functional and nutritional properties among the samples.

All formulated protein concentrate bars provided comparable energy (230–261 kcal) and balanced macronutrient profiles. The Chickpea and Moong bars delivered the highest protein content (15.7 g and 15.5 g, respectively), while the Rice bar provided the highest energy (260.50 kcal). The Pea and Moong bars showed relatively higher fibre content (10.0 g and 10.50 g), and fat levels were similar across all variants. Overall, Chickpea and Moong protein bars demonstrated slightly better nutritional profiles in terms of protein and fibre content, making them promising options for high-protein snack formulations.

The quality and shelf stability of protein concentrate bars from different plant sources were evaluated. Initial moisture content ranged from 7.8 ± 0.4% in jackfruit seed bars to 9.2 ± 0.3% in pea protein bars, while pH values were slightly acidic, between 6.2 and 6.5. Peroxide values were low (2.4–2.7 meq O_2/kg fat), indicating minimal lipid oxidation at the start of storage. Microbial loads were within safe limits, ranging from 1.7×10^3 to 2.4×10^3 cfu/g. Sensory evaluation revealed high acceptability across all bars, with scores from 7.8 to 8.6 out of 9; moong protein bars scored the highest, while rice bars scored slightly lower due to minor hardening after two months. Shelf life varied from 90 days for chickpea bars to 120 days for pea protein bars, which included antioxidant addition. Overall, chickpea and moong protein bars demonstrated good texture and flavor stability, jackfruit bars maintained stable color and aroma, and rice bars exhibited slight textural changes over time.

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REFERENCES

1. AOAC International. Official Methods of Analysis of AOAC International. 21st ed. AOAC International; 2019. Method 979.09.
2. Aluko RE. Functional properties of plant proteins. J Food Sci. 2004;69(3):R93-R104.
3. AOAC International. Official Methods of Analysis of AOAC International. 21st ed. AOAC International; 2019. Method 978.18.
4. Ainsworth EA, Gillespie KM. Estimation of total phenolic content and other oxidation substrates by Folin–Ciocalteu reagent. Nat Protoc. 2007;2(4):875-877.
5. Aprodu I, Banu I. Effect of inulin addition on dough rheology and bread quality. Food Sci Technol Int. 2015;21(8):621-630.
6. Arya SS, Salve AR. Influence of inulin on functional and textural properties of baked snack bars. J Food Process Preserv. 2019;43(5):e13936.
7. AOAC International. Official Methods of Analysis of AOAC International. 21st ed. AOAC International; 2019. Method 970.53.

8. Apak R, Güçlü K, Özyürek M, Karademir SE. Novel total antioxidant capacity index using cupric ion reducing capability (CUPRAC method). J Agric Food Chem. 2004;52(26):7970-7981.
9. Apak R, Güçlü K, Özyürek M, Karademir SE. The CUPRAC and related antioxidant capacity assays: review. Microchim Acta. 2008;160(4):413-419.
10. Barrett AH, Lloyd BJ. Advanced preservation methods and nutrient retention in food products. J Food Sci Nutr. 2012;63(4):321-330.
11. Banach JC, Clark S, Lamsal BP. Hardening of high-protein nutrition bars: A review. Food Hydrocoll. 2018.
12. Boye J, Zare F, Pletch A. Pulse proteins: Processing, characterization, functional properties and applications in food and feed. Food Res Int. 2010;43(2):414-431.
13. Bourne MC. Texture profile analysis. Food Technol. 1978;32(7):62-66.
14. Crowley SV, Gazi I, Kelly AL, Huppertz T, O'Mahony JA. Influence of protein ingredients on the textural and microstructural properties of nutrition bars. Food Hydrocoll. 2019;87:849-857.
15. Chang CC, Yang MH, Wen HM, Chern JC. Estimation of total flavonoid content in propolis by two complementary colorimetric methods. J Food Drug Anal. 2002;10(3):178-182.
16. Chang YW, Alli I, Konishi Y, Ziomek E. Characterization of protein fractions from chickpea (Cicer arietinum L.) and oat (Avena sativa L.) seeds using proteomic techniques. Food Res Int. 2011;44(9):3094-3104. doi:10.1016/j.foodres.2011.08.001.
17. Chandra S, Samsher. Assessment of functional properties of different flours. Afr J Agric Res. 2013;8:4849-4852.
18. Duncan DB. Multiple range and multiple "F" test. Biometrics. 1955;11:1-42.
19. DePalma G, Smith J, Brown R, et al. Nutritional and physicochemical properties of plant-based protein bars. Foods. 2024;13(2):123-130.
20. Franck A. Technological functionality of inulin and oligofructose. Br J Nutr. 2002; 87(S2):S287-S291.
21. Fang Y, et al. Influence of inulin and fiber addition on the water activity and stability of high-protein foods. J Food Eng. 2018;237:33-41.
22. Francis FJ. Colorimetry of foods. J Food Sci. 2010;50(1):36-43.
23. Friede J, Zavareze E. Protein extraction and isolation techniques in food applications. Food Bioprocess Technol. 2016;9:138-150.
24. Friedman HH, Whitney JE, Szczesniak AS. The texturometer—A new instrument for objective texture measurement. J Food Sci. 1963;28(4):390-396.
25. Folch J, Lees M, Stanley GHS. A simple method for the isolation and purification of total lipids from animal tissues. J Biol Chem. 1957;226:497-509.
26. Gaur VK, et al. Nutritional composition and amino acids of field pea and chickpea. ACS Food Sci Technol. 2018.
27. Hassan A, McMahon DJ. Hardening and microstructure of high-protein nutrition bars made using whey protein concentrate or milk protein concentrate. J Food Sci. 2016.
28. Hettiarachchy NS, Kalapathy U. Isolation and characterization of rice protein concentrate. J Food Sci. 1997;62(5):1116-1119.
29. Jeske S, Zannini E, Arendt EK. Past, present and future: The strength of plant-based dairy substitutes. Food Res Int. 2019;110:42-51. doi:10.1016/j.foodres.2018.03.014.
30. Joyner HS, Smith DM, Wang H. Understanding textural changes in protein bars during storage. J Food Sci. 2018;83(8):1953-1962. doi:10.1111/1750-3841.14285.
31. Jatav S, Soni R, Dunkwal V. Storage stability of millet-based nutri-bars. 2025.
32. Kinsella JE. Functional properties of soy proteins. J Am Oil Chem Soc. 1979;56:242-258.
33. Kowieska A, Gawlik-Dziki U, Dziki D. The effect of protein source on physicochemical and nutritional properties of high-protein bars. Foods. 2020;9(10):1467.
34. Kumar A, Mohanty V, Yashaswini P. Development of high-protein nutrition bar enriched with Spirulina platensis for undernourished children. Curr Res Nutr Food Sci. 2018;6(3):728-735.
35. Labuza TP, Altunakar B. Water activity prediction and moisture migration in multicomponent foods. J Food Eng. 2007;78(1):266-273.
36. Liu F, Ma C, Gao Y, McClements DJ, Ng TB. Structural and functional properties of plant proteins in food systems. Trends Food Sci Technol. 2020;99:1-11. doi:10.1016/j.tifs.2020.02.003.
37. McMahon DJ, Adams SL, McManus WR. Hardness and texture of high-protein nutrition bars: Role of water activity and matrix composition. J Food Sci. 2016;81(5):E1132-E1139.
38. Meter Group. AquaLab 4TE Water Activity Meter – Specifications. Meter Group Inc.
39. Mariotti F, Tomé D. The protein assessment methods: From Kjeldahl to Dumas and beyond. Food Chem. 2019;301:125258.
40. McMahon DJ, Adams SL, McManus WR. Hardening of high-protein nutrition bars and sugar/polyol-protein phase separation. J Food Sci. 2009;74(9):E312-E321.
41. Małecki J, Tomasevic I, Djekic I, Sołowiej BG. The effect of protein source on the physicochemical, nutritional properties and microstructure of high-protein bars intended for physically active people. Foods. 2020;9(10). doi:10.3390/foods9101460.Niness KR. Inulin and oligofructose: What are they? J Nutr. 1999;129(7):1402S-1406S.
42. Nath A, Chattopadhyay PK, Majumdar GC. Effect of inulin on physicochemical, textural, and sensory properties of fiber-enriched protein bars. LWT Food Sci Technol. 2016;73:102-110.

43. Pathare PB, Opara UL, Al-Said FAJ. Colour measurement and analysis in fresh and processed foods: A review. Food Bioprocess Technol. 2013;6:36-60.
44. Pearce KN, Kinsella JE. Emulsifying properties of proteins—Evaluation of a turbidimetric technique. J Agric Food Chem. 1978;26(3):716-723.
45. Raju PS, Bawa AS. Moisture migration and its control in baked products. Crit Rev Food Sci Nutr. 2006;46(6):469-481.
46. Rahmati NF, et al. Effect of dietary fibers on physicochemical and microbial stability of nutrition bars. Food Res Int. 2020;137:109619.
47. Rutherfurd SM, Moughan PJ. Available versus digestible dietary amino acids. Br J Nutr. 2012;108(S2):S298-S305.
48. Srebernich SM, Silva GM, Salvucci RC, Ruffi CRG. Physico-chemical, sensory and nutritional characteristics of cereal bars with addition of acacia gum, inulin and sorbitol. Cienc Tecnol Aliment. 2016;36(3):555-562.
49. Sołowiej E, et al. Addition of inulin to protein bars increases dietary fiber content and improves texture and mouthfeel. Int J Food Sci Nutr. 2021.
50. Sharma M, Shankar P. Organoleptic quality and shelf-life evaluation of nutraceutical sports bars. 2025.
51. Singleton VL, Orthofer R, Lamuela-Raventós RM. Analysis of total phenols and other oxidation substrates by means of Folin–Ciocalteu reagent. Methods Enzymol. 1999;299:152-178.
52. Szczesniak AS. Texture is a sensory property. Food Qual Prefer. 2002; 13(4-5):215-225.
53. Souiy Z, et al. Storage stability of cereal bars under controlled conditions. 2022.
54. El-Sohaimy SA, Sitohy MZ, El-Masry RA. Isolation and partial characterization of chickpea, lupine and lentil seed proteins. World J Agric Sci. 2007;3: 123-129.
55. Tsutsui T. Functional properties of heat-treated egg-yolk low-density lipoprotein. J Food Sci. 1988; 53(4):1103-1106.
56. You XY, Ding Y, Bu QY, Wang QH, Zhao GP. Nutritional, textural, and sensory attributes of protein bars formulated with mycoproteins. Foods. 2024; 13(5):671.
57. Wang Y, Zhang C, Chen L, Zhang H. Effect of inulin on texture and water distribution in high-protein food systems. LWT Food Sci Technol. 2022; 155:112936.
58. Xu X, et al. Effect of protein source on physicochemical and microstructural properties of high-protein bars. Foods. 2021;9(10):1467.
59. Zhishen J, Mengcheng T, Jianming W. Determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. Food Chem. 1999; 64(4):555-559.
60. Zhao X, et al. Color and texture changes in protein-enriched snack bars during storage. J Food Sci Technol. 2023; 60(5):1821-1832. Doi: 10.1007/s13197-023-05721-3.