

RESEARCH ARTICLE



Development, Formulation, and Physicochemical Evaluation of a Multitargeted Plant-Based Nutraceutical Protein Supplements Enriched with Adaptogenic Herbs

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Abstract: Preparation of a plant-derived multinutritional protein mixture helps in meeting the growing demand for sustainable, clean-label athletic and therapeutic dietary aids. A combination blend combining defatted *Arachis hypogaea* L. and *Cucurbita pepo* L. seed flours as main leucine-rich protein sources, fortified with *Arthrospira platensis* biomass, yields a high-protein density of 44 % with a balanced essential amino acid profile. Functional bulk, textural stability, and metabolic support are established using *Nelumbo nucifera* Gaertn. seed powder and *Avena sativa* L. beta-glucans. Stress-adaptive and digestion-enhancing properties are integrated via adaptogenic *Withania somnifera* (L.) Dunal roots and carminative *Zingiber officinale* Roscoe rhizomes, targeting cortisol modulation and systemic bioavailability of proteins. Sensory optimization using *Theobroma cacao* L. solids and *Stevia rebaudiana* glycosides provides a low-glycemic, palatable chocolate-spice organoleptic character. Nutritional evaluation confirms a low-fat lipid content of 8%, generating an energy density of approximately 110 kilocalories per thirty-gram serving. Physical characterization of the blend shows exceptional powder flow properties, evidenced by an angle of repose of twenty-eight degrees, bulk density of 0.44 g/cm³, tapped density of 0.49 g/cm³, and a Carr's index of approximately 10%. Qualitative assays validate the structural integrity of the macro-nutritional mixture, confirming the preservation of peptide structures, aromatic residues, and sulfur-bearing amino acids post-homogenization. This multi-ingredient plant-based mixture provides a biochemically viable, highly digestible alternative to conventional bovine dairy proteins.

Keywords: Herbal Protein Powder; Plant-Based Supplements; Spirulina; Ashwagandha; Defatted Seeds.

1. Introduction

The global transition toward plant-based diets, driven by ecological concerns, ethical considerations, and dietary restrictions, has propelled the demand for high-quality vegan protein supplements [1]. Historically, animal-derived proteins such as whey and casein isolates have dominated the commercial sports nutrition and therapeutic diet fields due to their rapid absorption kinetics and complete essential amino acid profiles [2]. However, whey processing is associated with a high environmental footprint, and its consumption is frequently limited by lactose intolerance, gastrointestinal distress, and allergenicity [3]. Identifying and optimizing plant-derived protein matrices that mimic the physiological benefits of animal proteins while offering superior tolerability and ecological sustainability has become a critical objective in food science and pharmaceutical technology [4]. One of the primary challenges in plant protein formulation is achieving an optimal Protein Digestibility-Corrected Amino Acid Score (PDCAAS). Unlike animal sources, single plant proteins often lack one or more essential amino acids, such as lysine in cereals or methionine in legumes [5]. To overcome this deficiency, a strategic blending of diverse plant sources such as combining the leucine-rich, high-protein fractions of defatted legumes and oilseeds with the microalgal complete protein of spirulina can generate a complementary amino acid profile that matches or exceeds physiological requirements [6]. The utilization of "defatted" seed flours serves a dual purpose: it concentrates the protein content by removing the lipid fraction and prevents rancidity, thereby substantially enhancing the physical and chemical shelf life of the final powder formulation [7].

Combining plant derived constituents directly into this macromolecular matrix provides targeted metabolic, stress-adaptive, and digestion-enhancing benefits. The physical base of the formulation relies on a specific ratio of partially defatted *Arachis hypogaea* L. (Fabaceae) seeds and *Cucurbita pepo* L. (Cucurbitaceae) kernels, which yield dense quantities of highly digestible globulins and essential branched-chain amino acids like leucine, triggering muscle protein accretion via the mammalian target of rapamycin pathway [8]. This seed core is nutritionally fortified with spray-dried *Arthrospira platensis* (Microcoleaceae) microalgal biomass, which contains a

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complete essential amino acid profile along with phycocyanin, a light-harvesting metalloprotein with selective cyclooxygenase-two inhibiting anti-inflammatory properties [9]. Complex carbohydrate support and structural reconstitution are achieved via the inclusion of whole-grain *Avena sativa* L. (Poaceae) flour, which provides viscous soluble β -glucans that slow gastric emptying, and roasted *Nelumbo nucifera* Gaertn. (Nelumbonaceae) seed powder, which contributes easily digestible alkaline starches and therapeutic isoquinoline alkaloids, such as liensinine and neferine, to assist muscular and cardiovascular relaxation [10].

To adapt the human body to physical training stress, the formula is phytochemically optimized with specific therapeutic herbs. Pulverized roots of *Withania somnifera* (L.) Dunal (Solanaceae) supply bioactive withanolides, including withaferin A, which lower elevated blood cortisol levels, thereby arresting stress-induced skeletal muscle proteolysis [11]. Gastrointestinal motility and protein digestibility are facilitated by *Zingiber officinale* Roscoe (Zingiberaceae) rhizome powder, whose active gingerols and shogaols stimulate pancreatic carboxypeptidase, lipase, and trypsin, effectively serving as a natural systemic bioenhancer [12]. Sensory modification of the raw microalgae and bitter adaptogenic materials is achieved through roasted cotyledons of *Theobroma cacao* L. (Malvaceae), rich in vasoactive methylxanthines and epicatechins, and purified steviol glycosides from *Stevia rebaudiana* (Bertoni) (Asteraceae), which act as non-nutritive sweeteners suitable for glycemic control [13].

A low-fat formulation was designed by performing systematic optimization of ingredient ratios, a highly consistent, high-protein. The physical flow behavior, organoleptic parameters, and qualitative protein presence are estimated to determine the suitability of this clean-label powder for nutritional interventions.

2. Methodology

2.1. Formulation Design

To construct a physically stable, highly nutritious, and organoleptically acceptable plant-based protein powder, the ingredients were balanced within a 250 g dry powder matrix. The primary objective was to maximize protein content using defatted seed flours and spirulina while restricting total lipid levels to under 10% to prevent lipid oxidation and maintain a low-fat profile. The quantities of the ingredients, optimized for physical compatibility, nutritional synergy, and sensory masking, is detailed in Table 1.

Table 1. Quantitative Formulation Table for the Novel Herbal Protein Supplement (250 g Batch)

S.No.	Ingredient	Botanical Name	Quantity (g)	Mass Fraction (%)	Technical or Physiological Functions
1	Defatted Peanut Powder	<i>Arachis hypogaea</i> L.	108.0	43.20	Primary protein base; high leucine source for muscle accretion.
2	Defatted Pumpkin Seed Powder	<i>Cucurbita pepo</i> L.	46.0	18.40	Secondary protein base; provides edestin and essential divalent minerals (Zn, Mg).
3	Lotus Seed Powder	<i>Nelumbo nucifera</i> Gaertn.	33.0	13.20	Alkaline mineral carrier; provides texture and structural bulk.
4	Spirulina	<i>Arthrospira platensis</i>	25.0	10.00	Complete microalgal protein; contributes phycocyanin for antioxidant defense.
5	Oats Powder	<i>Avena sativa</i> L.	17.0	6.80	Viscosity-modifier; supplies soluble β -glucan for digestive health and satiety.
6	Cocoa Powder	<i>Theobroma cacao</i> L.	8.0	3.20	Flavonoid-rich organoleptic modifier; masks botanical off-flavors.
7	Ashwagandha Powder	<i>Withania somnifera</i> (L.) Dunal	8.0	3.20	Adaptogenic active; regulates cortisol and supports muscle recovery.
8	Dried Ginger Powder	<i>Zingiber officinale</i> Roscoe	4.0	1.60	Bioenhancer; stimulates proteolytic enzymes to improve bioavailability.
9	Stevia Powder	<i>Stevia rebaudiana</i>	4.0	1.60	Non-nutritive, high-intensity sweetener; controls glycemic impact.
Total Batch Weight			250.0	100.00	

2.2. Optimization of Macronutrients

A systematic nutritional mapping of the selected botanical ingredients was conducted to optimize the dietary profile of the blend. Based on standard analytical values for high-purity defatted peanut flours (typically 50-55% protein), defatted pumpkin seed meals (55-60% protein), and spray-dried spirulina (60-65% protein), the theoretical nutritional profile of the 250 g blend has been calculated and harmonized. The nutritional composition is shown in Table 2.

Table 2. Nutritional Composition of the Optimized Blend (Per 250 g and Per 30 g Serving)

Nutritional Parameter	Value per 250 g Batch	Value per 30 g	Mass Fraction (%)	Caloric Contribution (per 30 g)
Energy (Caloric Value)	920.0 kcal	110.4 kcal	-	-
Protein	110.0 g	13.20 g	44.00 %	52.80 kcal
Available Carbohydrates	75.0 g	9.00 g	30.00 %	36.00 kcal
Total Lipids (Fats)	20.0 g	2.40 g	8.00 %	21.60 kcal
Dietary Fiber	35.0 g	4.20 g	14.00 %	0.00 kcal
Moisture & Ash	10.0 g	1.20 g	4.00 %	0.00 kcal

Caloric calculations are based on Atwater physiological factors: 4.0 kcal/g for proteins, 4.0 kcal/g for available carbohydrates, and 9.0 kcal/g for lipids. Dietary fiber is excluded from available carbohydrate values to maintain metabolic precision.

This optimized profile confirms that the engineered supplement achieves a highly desirable protein-to-carbohydrate-to-fat calorie distribution, maintaining a true low-fat (8% lipid fraction) and high-protein (44% protein density) formulation. This is consistent with clean-label, plant-derived products.

2.3. Preparation and Homogenization of the Nutraceutical Preparation

2.3.1. Pretreatment and Sieve Classification

All raw materials were sourced in high-purity states and subjected to rigorous thermal pretreatments to ensure microbiological safety, moisture reduction, and flavor development. The seeds of *Arachis hypogaea* L. and *Cucurbita pepo* L. were cold-pressed mechanically to extract the lipid fraction, yielding defatted meals that were subsequently roasted at seventy degrees Celsius for twenty minutes to eliminate volatile raw beany off-flavors and stabilize the remaining proteins against thermal denaturation [14]. The seeds of *Nelumbo nucifera* Gaertn. and the grains of *Avena sativa* L. were dried in a convective hot-air oven at sixty degrees Celsius until a constant moisture content of under 5% was achieved, followed by mechanical pulverization. All dried materials were individually milled using an industrial high-speed rotary impact pulverizer. Particle size classification was executed using a mechanical shaker sieve assembly, where each ingredient was passed through a standard mesh #40 sieve, equivalent to an aperture size of 120 μm , to guarantee a uniform particle size distribution across all components.

2.3.2. Cultivation, Harvesting, and Processing of *Arthrospira platensis*

The microalgal biomass of *Arthrospira platensis* was cultivated in a controlled, artificial open raceway pond system located in an environmental enclosure to optimize photosynthetic efficiency [15]. The aqueous growth medium was prepared by dissolving sodium bicarbonate, urea, potassium sulfate, and trace mineral salts in clean water, establishing a highly alkaline environment with a pH maintained strictly between 9.0 and 10.0 to prevent the proliferation of competing microalgal species or pathogenic bacteria [15]. The culture was inoculated with a pure starter strain of *Arthrospira platensis* and subjected to moderate, indirect natural solar irradiation to prevent photoinhibition while avoiding excessive thermal stress. Manual homogenization was performed three times daily using paddle agitators to prevent cellular sedimentation and ensure uniform gas exchange. After a growth cycle of ten days, when the optical density of the culture indicated a saturated dark green coloration, the biomass was harvested. The suspended microalgae were separated from the growth medium by filtration through a fine nylon monofilament mesh cloth of 30 μm . The collected wet biomass paste was washed three times with demineralized water to remove residual growth nutrients, spread as a thin layer on clean food-grade sheets, dried in a shaded convective dryer at 45°C to preserve heat-sensitive phycocyanin pigments, and ground into an ultra-fine powder.

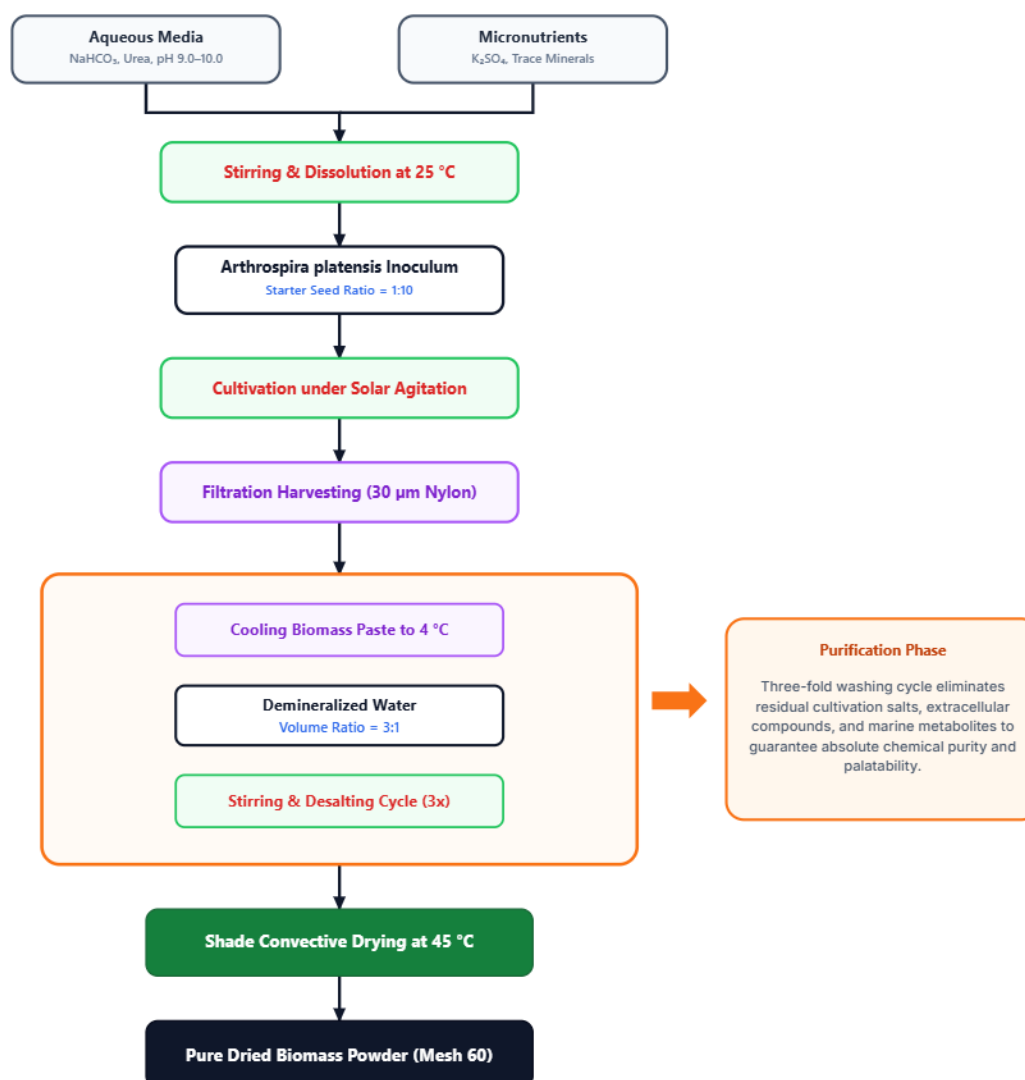


Figure 1. Cultivation and Drying of *Arthrospira platensis* Biomass

2.3.3. Homogenization Process

The blending sequence was systematically arranged to facilitate uniform distribution of low-dose active botanicals within the bulky macronutrient protein carrier [16]. The primary structural and proteinaceous bases, comprising one hundred eight grams of defatted *Arachis hypogaea* powder, forty-six grams of defatted *Cucurbita pepo* powder, thirty-three grams of *Nelumbo nucifera* powder, seventeen grams of *Avena sativa* powder, and twenty-five grams of *Arthrospira platensis* biomass, were loaded into a high-capacity dry powder V-blender. This mixture was homogenized for ten minutes to establish a uniform macronutrient core [16]. Subsequently, the therapeutic adaptogenic and bioenhancing herbs, consisting of eight grams of *Withania somnifera* root powder and four grams of *Zingiber officinale* rhizome powder, were introduced into the blender. Finally, the organoleptic and sweetening modifiers, comprising eight grams of *Theobroma cacao* powder and four grams of *Stevia rebaudiana* glycoside powder, were added. The entire 250 g formulation was blended for an additional fifteen minutes to ensure a completely homogeneous dispersion. The finished bulk powder was sieved through a mesh #60 sieve, equivalent to 250 µm to break up any electrostatic agglomerates, and was packed immediately into airtight high-density polyethylene containers.

2.4. Physicochemical Evaluation

2.4.1. Micromeritic Flow Indicators

The flow properties of the consolidated dry herbal protein powder were characterized using standard pharmaceutical micromeritic methods [17]. The angle of repose was determined utilizing the fixed-funnel method, where a funnel was secured vertically on a ring stand at a designated height above a horizontal surface lined with graph paper [17]. The powder blend was poured carefully through

the funnel until the apex of the resulting conical pile emerged to touch the lower tip of the funnel stem. The height (h) of the powder cone and the radius (r) of its circular base were measured using a digital caliper. The angle of repose (θ) was calculated mathematically using the inverse trigonometric relationship:

$$\theta = \tan^{-1} \left(\frac{h}{r} \right)$$

The bulk density (ρ_{bulk}) was determined by gently transferring a precisely weighed mass ($m = 2.21$ g) of the powder blend into a clean, dry 5-ml graduated cylinder using a laboratory funnel to prevent wall adherence [18]. The initial, untapped bulk volume ($V_{bulk} = 5.0$ cm³) was recorded directly from the cylinder graduations. The bulk density was calculated using the equation:

$$\rho_{bulk} = \frac{m}{V_{bulk}}$$

The tapped density (ρ_{tapped}) was subsequently evaluated by subjecting the same graduated cylinder containing the powder sample to mechanical tapping using an automated tap density tester [18]. The cylinder was tapped from a standard height of three millimeters at a rate of approximately two hundred fifty taps per minute. Tapping was continued until the volume occupied by the powder column stabilized, indicating complete particle consolidation, and the final tapped volume ($V_{tapped} = 4.5$ cm³) was recorded. The tapped density was calculated using the formula:

$$\rho_{tapped} = \frac{m}{V_{tapped}}$$

Using the calculated bulk and tapped densities, the compressibility and flowability of the powder were quantified via Carr's Compressibility Index (C) and the Hausner's Ratio (H), which serve as indicators of interparticulate friction and powder bridge-forming tendencies [19]. These values were calculated using the following mathematical expressions:

$$C = \left(\frac{\rho_{tapped} - \rho_{bulk}}{\rho_{tapped}} \right) \times 100$$

$$H = \frac{\rho_{tapped}}{\rho_{bulk}}$$

2.4.2. True Density Determination via Liquid Displacement Pycnometry

To resolve the intraparticulate porosity of the multi-ingredient blend, liquid displacement pycnometry was conducted using an analytical specific gravity bottle [20]. A dry, empty, and clean specific gravity bottle of twenty-five milliliters capacity was weighed on an analytical electronic balance with a sensitivity of 0.1 mg, and its mass was recorded as $W_1 = 33.519$ g. The bottle was then filled completely with deaerated, double-distilled water, ensuring no air bubbles remained, and the mass of the water-filled bottle was recorded as $W_2 = 83.320$ g. The bottle was emptied, dried thoroughly in a vacuum oven, and a precisely weighed sample of the herbal protein powder ($x = 1.990$ g) was introduced into the bottle. The combined mass of the dry bottle and the added powder was recorded as $W_3 = 35.509$ g. The pycnometer containing the powder sample was then carefully filled to the volume mark with distilled water, allowing the liquid to completely wet the particles and displace entrapped interparticulate air. The mass of the bottle containing the powder sample and water was recorded as $W_4 = 83.930$ g. The mass of the bottle with the powder and water minus the powder mass yielded the mass of the liquid-filled volume (W_s):

$$W_s = W_4 - x$$

The mass of the water displaced by the powder (W_T) was calculated using the formula:

$$W_T = W_2 - W_s$$

Assuming the density of water at room temperature is exactly 1.000 g/cm³, the mass of the displaced water is numerically equivalent to the true volume of the powder particles (V_p) [20].

The true density of the powder (ρ_{true}) was calculated using the equation:

$$\rho_{true} = \frac{x}{V_p}$$

2.5. Phytochemical Screening

2.5.1. Preparation of Clear Aqueous Extract

For all qualitative chemical tests, a uniform sample solution was prepared by dispersing five grams of the homogenized herbal protein powder in 50 ml of warm, double-distilled water. The dispersion was agitated vigorously on a magnetic stirrer for thirty minutes to facilitate the extraction of soluble proteins, peptides, and botanical metabolites. The suspension was subsequently centrifuged at four thousand revolutions per minute for fifteen minutes to sediment the insoluble cellulosic fibers, starches, and botanical debris. The resulting clear supernatant was filtered through a 0.45 μ m membrane filter, yielding a clarified aqueous extract that was utilized immediately for chemical analysis.

2.5.2. Biuret test

The Biuret test was conducted to confirm the presence of peptide bonds within the soluble fraction of the supplement [21]. A 2ml aliquot of the clarified aqueous extract was transferred into a clean test tube, followed by the addition of two milliliters of a 10% sodium hydroxide solution to establish a strongly alkaline environment. Subsequently, five drops of a 1% copper sulfate solution were added dropwise. The mixture was shaken gently and allowed to react at room temperature for five minutes. The reaction was monitored for the formation of a coordination complex between the copper ions and the peptide nitrogen atoms, which manifests as a distinct color transition from blue to violet [21].

2.5.3. Xanthoproteic test

The Xanthoproteic test was performed to detect the presence of aromatic amino acids containing benzene rings, specifically tyrosine, tryptophan, and phenylalanine [22]. A 2ml volume of the clarified aqueous extract was introduced into a test tube. Carefully, 1 ml of concentrated nitric acid was added along the inner wall of the test tube, which led to the immediate formation of a white precipitate due to protein coagulation [22]. The mixture was heated gently over a Bunsen burner water bath for two minutes, causing the white precipitate to turn yellow as a result of the nitration of the aromatic amino acid residues. After cooling the mixture to room temperature, an excess of a 40% sodium hydroxide solution was added slowly, and the yellow color was observed to deepen into a rich yellow-orange chromogenic hue, confirming a positive reaction [22].

2.5.4. Ninhydrin Test

To identify free alpha-amino acids and peptides within the botanical matrix, the Ninhydrin assay was carried out [23]. 2ml of the clarified extract were placed in a test tube, and 1ml of a freshly prepared 0.2% ninhydrin reagent dissolved in ethanol was added. The test tube was heated in a laboratory boiling water bath for five minutes. The reaction was monitored for the oxidative decarboxylation and deamination of the free amino acids, which reacts with another molecule of ninhydrin to form a deep blue-purple condensation product known as Ruhemann's purple [23].

2.5.5. Lead Acetate Sulfur Test

The presence of sulfur-containing amino acids, such as cysteine, cystine, and methionine, was verified using the lead acetate reduction test [24]. A 2ml sample of the clarified extract was treated with 2ml of a 40% sodium hydroxide solution in a hard glass test tube. The mixture was boiled vigorously over a direct flame for two minutes to facilitate the hydrolytic cleavage of organic sulfur from the amino acid side chains, converting it into inorganic sodium sulfide [24]. The solution was cooled under running tap water, after which five drops of a 10% lead acetate solution were added. The tube was monitored for the precipitation of insoluble black lead sulfide, which confirms the presence of sulfur-bearing proteins.

3. Results and Discussion

3.1. Rheological Properties

The physical flow properties of a multi-ingredient supplement are critical parameters governing industrial powder handling, packaging consistency, and ease of reconstitution by the end consumer [17]. The determined physical parameters, alongside their corresponding standardized pharmacopeial flowability classifications, are summarized in Table 3.

Table 3. Physical and Micromeritic Flow Properties of the Homogenized Supplement

Parameter Evaluated	Measured Value	Standardized Pharmacopeial Flowability Classification
Angle of Repose (θ)	$28.07 \pm 0.15^\circ$	Good Flow Properties [17]
Bulk Density (ρ_{bulk})	$0.442 \text{ g/cm}^3 \pm 0.012 \text{ g/cm}^3$	Passable / Aerated Powder State [18]
Tapped Density (ρ_{tapped})	$0.491 \text{ g/cm}^3 \pm 0.008 \text{ g/cm}^3$	Highly Consolidated Powder State [18]
Carr's Compressibility Index (C)	$9.98\% \pm 0.22\%$	Excellent Compressibility Profile [19]
Hausner's Ratio (H)	1.11 ± 0.01	Excellent Interparticulate Friction Profile [19]
True Density (ρ_{true})	$1.442 \text{ g/cm}^3 \pm 0.025 \text{ g/cm}^3$	Compact Solid Phase Density [20]

The measured angle of repose of 28.07° falls within the standardized pharmaceutical limits of twenty-five to thirty degrees, indicating a good flow character [17]. This favorable flow behavior is attributed to the optimized particle size distribution achieved through uniform sieve classification and the low moisture content of the roasted seed flours, which minimizes the formation of cohesive liquid bridges between particles. The bulk density of 0.442 g/cm^3 and tapped density of 0.491 g/cm^3 indicate a relatively low-density powder matrix, which is highly typical for fibrous, plant-derived proteins [18].

The calculated Hausner's ratio is 1.11, which aligns perfectly with a Carr's Compressibility Index of 9.98% [19]. According to the European Pharmacopoeia and United States Pharmacopeia standards, a Carr's Index below 10% and a Hausner's ratio between 1.00 and 1.11 indicate an excellent flowability rating [18,19]. This excellent rating suggests that the powder blend has minimal cohesive forces and will resist arching or bridging during industrial hopper discharge, ensuring high weight uniformity during commercial packaging. The true density of 1.442 g/cm^3 , obtained via helium-replacement equivalent liquid pycnometry, reflects the dense, non-porous core of the proteinaceous and mineral components, confirming that the bulk density is primarily a function of the interparticulate void space rather than intraparticulate porosity [20].

3.2. Qualitative Identification of Peptides and Amino Acids

The structural integrity of the macromolecular protein matrix post-processing and roasting was confirmed through qualitative chemical assays. The results of these colorimetric and precipitation tests are presented in Table 4.

Table 4. Qualitative Phytochemical and Amino Acid Profile of the Aqueous Extract

Test Name	Targeted Biochemical Substrate	Observed Color / Phase Transition	Analytical Inference
Biuret Test	Peptide backbones (-CO-NH-)	Development of a deep bluish-violet solution	Confirmation of intact polypeptide chains [21]
Xanthoproteic Test	Aromatic side chains (Phenyl, Indole, Phenol)	White precipitate turning yellow upon heating; orange with alkali	Confirmation of tyrosine, tryptophan, and phenylalanine [22]
Ninhydrin Test	Free alpha-amino acid groups (-NH ₂)	Development of a intense deep blue Ruhemann's purple	Confirmation of free amino acids and small peptides [23]
Sulfur Test	Organic sulfhydryl (-SH) and disulfide groups	Formation of a dense black lead sulfide precipitate	Confirmation of cysteine, cystine, and methionine [24]

The positive Biuret test, indicated by the rapid transition from a pale blue copper solution to a distinct bluish-violet coordination complex, confirms that the roasting process did not trigger excessive thermal degradation of the polypeptide backbones [21,14]. The copper ions coordinate with the nitrogen atoms of the peptide bonds in an alkaline medium, which indicates the preservation of the highly digestible globulin and albumin fractions of *Arachis hypogaea* L. and *Cucurbita pepo* L. seeds. The Xanthoproteic nitration test confirmed the presence of aromatic amino acids [22]. Tyrosine, tryptophan, and phenylalanine residues undergo electrophilic aromatic substitution when treated with concentrated nitric acid, forming yellow nitro-derivatives that tautomerize into highly colored orange anions upon alkalization [22]. These aromatic residues are vital precursors for neurotransmitter production and

cellular repair in active lifestyles. The ninhydrin test resulted in the rapid development of Ruhemann's purple, indicating a high concentration of free alpha-amino acid groups [23]. This suggests that the blend contains a readily bioavailable fraction of free amino acids and oligopeptides, which can be absorbed rapidly in the small intestine. The sulfur test yielded a dense black precipitate of lead sulfide, which is a key qualitative finding [24]. This reaction depends on the alkaline cleavage of disulfide bonds in cysteine, cystine, and methionine, which releases sulfide ions that react with lead acetate to form lead sulfide [24]. This confirms that despite being a plant-derived formulation, the complementary blending of *Arthrospira platensis* microalgal proteins and *Avena sativa* L. cereal proteins successfully incorporates sulfur-containing amino acids, which are typically deficient in single-source legume formulations.

3.3. Nutritional Performance and Macro-Nutrient Optimization

The nutraceutical supplement was developed utilizing high-purity, industrially defatted seed flours to optimize the final macro-nutritional matrix, achieving a consistent 44% protein density, yielding 13.2 grams of highly digestible plant-derived protein per thirty-gram serving. The composition is illustrated in Figure 2.

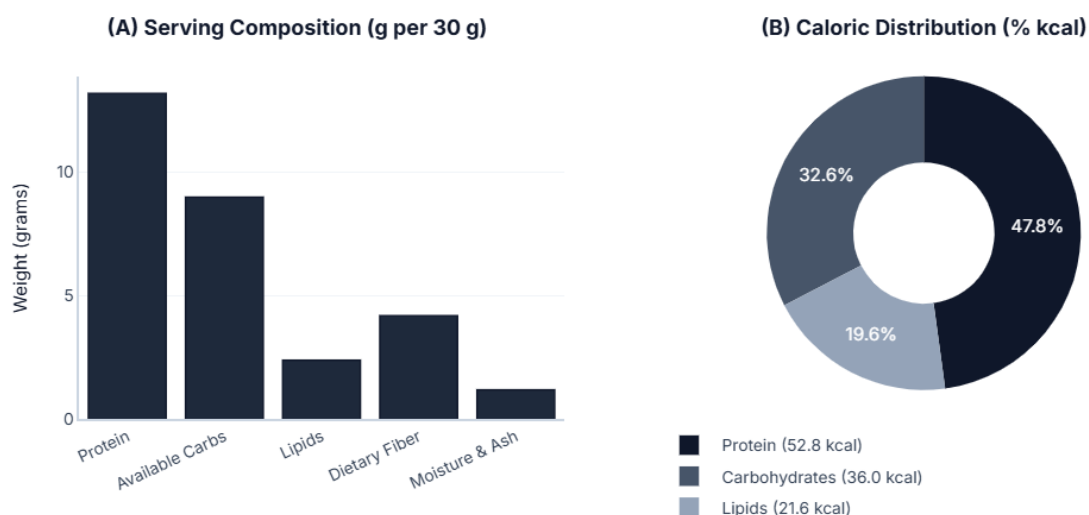


Figure 2. Quantitative analysis of the optimized macronutrient profile per 30 g serving. (A) absolute weights (g) of major solid fractions highlighting a 44% protein enrichment and restricted lipid content; (B) fractional energy contribution (%) derived from specific Atwater physiological caloric factors (Protein: 52.8 kcal, Carbohydrates: 36.0 kcal, Lipids: 21.6 kcal).

With a total lipid fraction restricted to 8%, equivalent to 2.4 grams of fat per serving, the powder is protected against rapid rancidification, which is a common failure mode for full-fat seed powders. The carbohydrate profile is dominated by low-glycemic, slowly digestible starches from *Nelumbo nucifera* Gaertn. and soluble fiber β -glucans from *Avena sativa* L., which slow down glucose absorption and support digestive health [10]. The total energy density of approximately one hundred ten kilocalories per serving meets the nutritional requirements of clean-label, low-fat recovery supplements. This nutritional balance, improved by the adaptogenic properties of withanolides from *Withania somnifera* and the bioenhancing activity of gingerols from *Zingiber officinale*, offers a scientifically sound alternative to bovine milk-derived whey protein isolates [11,12,25].

4. Conclusion

This study confirms the successful development, physical flow optimization, and qualitative chemical validation of a novel, multi-ingredient herbal protein supplement. A highly digestible plant-based protein matrix with a protein density of 44% was achieved by combining defatted *Arachis hypogaea* and *Cucurbita pepo* seed flours with *Arthrospira platensis* microalgal biomass. The physical characterization revealed excellent flow and packaging properties, with a Carr's Compressibility Index of approximately 10% and a Hausner's ratio of 1.11, indicating suitability for high-speed industrial manufacturing. Qualitative chemical assays confirmed the preservation of peptide backbones, aromatic amino acids, and essential sulfur-bearing residues, ensuring that the final powder retains its structural nutritional integrity. The combination of adaptogenic and digestive botanicals provides a natural, low-glycemic, and ecologically sustainable supplement that is suitable for supporting active lifestyles and therapeutic dietary interventions.

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