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Impact of Dehulling on the Nutritional Composition, Bioactive Compounds, and Antioxidant Potential of Moroccan Faba Bean (*Vicia faba* L.) Cultivars

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ABSTRACT

Background: Faba bean (*Vicia faba* L.) is a nutrient rich yet underutilized legume endowed with considerable potential for enhancing food security and plant-based nutritional sustainability. In Morocco, however, the compositional diversity of local varieties and their biochemical response to processing interventions remain insufficiently characterized, thereby constraining the optimal valorization of this crop within sustainable dietary systems.

Aims: This study aimed to evaluate the nutritional composition, phytochemical profile, and antioxidant properties of nine Moroccan faba bean varieties, and to assess the influence of dehulling on their biochemical composition.

Methods and Methods: A comparative cross-sectional analysis was conducted on nine *Vicia faba* L. varieties cultivated in Morocco. Both whole seed (WS) and dehulled seed (DS) fractions were analyzed for proximate composition, mineral content, amino acid and fatty acid profiles, and phytochemical constituents. Antioxidant activities were determined employing three complementary assays: DPPH radical scavenging, ABTS radical cation decolorization, and ferric reducing antioxidant power (FRAP).

Results: Significant varietal and processing effects were observed across all measured parameters. Crude protein content ranged from 20.05% (Alfia 21, WS) to 28.43% (Zina, DS), with dehulling consistently increasing protein concentration across varieties. Lysine and leucine were identified as the most abundant essential amino acids, with particularly elevated levels recorded in the Alfia 17 variety. Fatty acid profiling revealed a predominance of unsaturated fatty acids—most notably linoleic acid (45.31 – 54.42%)—, the proportion of which increased in DS fractions, whilst palmitic acid decreased correspondingly. Among macro-elements, potassium was the most abundant, followed by phosphorus and sulfur; iron and zinc constituted the principal microelements. Dehulling reduced calcium content (by up to 52.9%), whilst increasing phosphorus and sulfur concentrations. Total polyphenol content ranged from 2.86 to 4.26 mg GAE/g DM in WS fraction and declined significantly in DS fractions following seed coat removal. Antioxidant activities were higher in whole seeds across all varieties and assays.

Conclusions: Moroccan *Vicia faba* L. varieties exhibit substantial nutritional diversity, characterized by elevated protein content and favorable unsaturated fatty acid profiles. Dehulling enhances protein concentration and improves the bioavailability of select minerals; however, it concurrently diminishes antioxidant capacity as a consequence of phenolic compound loss associated with seed coat removal. These findings underscore the imperative to optimize processing strategies that reconcile nutrient enrichment with the retention of bioactive compounds, thereby maximizing the nutritional and functional value of faba bean in food system applications.

Keywords *Vicia faba* L.; Nutritional Quality; Phenolic Compounds; Antioxidant Activity; Dehulling; Moroccan Legumes.

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1 INTRODUCTION

In developing countries, rapid population growth and limited economic resources significantly impact public health and national productivity. In this context, food safety and nutritional quality remain among the main challenges of these countries (Oluwole *et al.*, 2023; Food and Agriculture Organization [FAO], 2024). In developing Afro-Asian

countries, the number of nourishing mothers and children suffering from protein-calorie malnutrition (PCM) exceed 170 million requiring the urgent need for new protein sources and valorization of existing ones (Sharma, 2022).

Worldwide, major interest has been reported on legumes for their exceptional nutritional properties, including protein content, macro and micro-nutrients composition, and beneficial effects on human health including the risks of

coronary heart disease, stroke, high blood pressure and diabetes (Thorisdottir et al., 2023). Among grain legumes, faba bean (*Vicia faba* L.) is an annual pulse from the *Fabaceae* family, crops that have been growing for thousands of years in the Mediterranean region, Africa and Asia, and is currently cultivated in more than sixty-six countries (Abou-Khater et al., 2022; Martineau-Côté et al., 2022a). Faba bean exhibits a strong capacity for atmospheric nitrogen fixation in agricultural soils (Zhou et al., 2018) and can be used in soil rotation (Aschi et al., 2017), improving the yield of other crops, such as wheat (Xiao et al., 2018), and greatly reducing the need of synthetic fertilizers, making this legume an ecologically, economically and nutritionally significant crop (Xiao et al., 2021).

From a nutritional perspective, faba beans dry seeds are widely recognized as an exceptional source of dietary protein, typically ranging from 26% to 33% on a dry weight basis (Labba et al., 2021). Characterized by an elevated concentration of essential amino acids—most notably lysine—this legume serves as an optimal nutritional complement to rectify the amino acid profiles of traditional, cereal-based Mediterranean diet (Khayour et al., 2023; Skylas et al., 2019). Moreover, faba beans constitute a valuable source of essential minerals, carbohydrate, dietary fibers and vitamins (Dhull et al., 2022).

Notably, the crop exhibits a superior mineral profile relative to both cereals and alternative leguminous species (Millar et al., 2019). It remains predominantly integrated into global agricultural systems as a non-genetically modified crop (non-GMO) (Calabrò et al., 2014). From a lipid standpoint, faba beans feature a low total fat content dominated by high-quality unsaturated fatty acids, specifically oleic and linoleic acids (Choi et al., 2023). Moreover, epidemiological and immunological insights suggest that the species displays lower allergenicity than other major pulses, including peas, lentils and chickpeas (Smits et al., 2021).

Concurrently, *V. faba* constitutes a significant source of bioactive secondary metabolites—most notably polyphenols, flavonoids, and tannins—widely reported to be associated with various health benefits, including antidiabetic properties (Sathya Prabhu & Devi Rajeswari, 2018), a reduced epidemiological incidence of colorectal malignancies (Aune et al., 2011) and cytoprotection against hydrogen peroxide (H₂O₂)-induced oxidative stress at the cellular level (Siah et al., 2012). The presence of compounds, classified as antinutritional factors (ANFs) that can modulate protein digestibility and mineral bioaccessibility, does not fundamentally compromise the beneficial effect of these bioactive compounds.

Scientific evidences demonstrates that the chemical profile and overall nutritional quality of faba beans are strictly

modulated by an intricate interplay of intrinsic factors—primarily genotypic variations—and extrinsic vectors, including soil properties, edaphic features, and regional agronomic conditions. Furthermore, post-harvest unit operations, such as soaking, thermal extrusion, and mechanical decortication (dehulling), have been reported to efficiently reduce endogenous pools of polyphenols and condensed tannins, thereby elevating nutrient bioavailability and apparent protein concentrations (Alonso et al., 2000; Cormack et al., 2026; Johnson et al., 2020; Valente et al., 2018). For instance, a comparative evaluation across 22 faba bean cultivars revealed that mechanical dehulling provoked a relative increase in protein content ranging from 9.31% to 17.69%, coupled with a simultaneous reduction in condensed tannin levels ranging from 1.70% to 66.99% (Choi et al., 2023).

From a morpho-anatomical standpoint, faba bean genotypes exhibit extensive phenotypic polymorphism regarding seed geometry, pigmentation, and scale. Taxonomically, three primary botanical sub-varieties are differentiated based on seed mass indices: *var. minor* which encompasses small-seeded genotypes weighing less than 0.5 g seed⁻¹; *var. major*, characterized by large-seeded variants exceeding 1 g seed⁻¹ and *var. equina*, which constitutes an intermediate medium-seeded morphotype (Ohm et al., 2024).

Within the agricultural landscape of Morocco, as observed across parallel North African territories, a diverse array of indigenous faba bean cultivars is regularly cultivated but remain underutilized within modern agri-food frameworks. To address this gap, the present investigation was designed to comprehensively characterize the predominant locally grown faba bean varieties through the systematic profiling of their macro- and micronutrient distributions. Concurrently, this work evaluates the precise impact of mechanical dehulling on the raw seed composition, aiming to establish an empirical baseline to facilitate the optimized exploitation, industrial formulation, and targeted technological valorization of this sustainable grain legume.

2 MATERIALS AND METHODS

2.1 Plant Material and Sample Preparation

This investigation evaluated nine distinct cultivars of faba bean (*Vicia faba* L.). Morpho-anatomical classification distinguished five large-seeded genotypes belonging to *var. major* (Aguadulce, Defes; Hiba, Lobab and Karabiga) and four small-seeded genotypes belonging to *var. minor* (Alfia 5, Alfia 17, Alfia 21 and Zina). Certified dry seeds were obtained from the National Institute of Agricultural Research (INRA) in Rabat, Morocco.

Prior to analytical characterization, the seeds were manually cleaned to eliminate extraneous matter and defects. Mechanical decortication (dehulling) was performed to separate the cotyledon from the seed coat. The resulting matrices were subjected to a two-step grinding process: an initial coarse milling to disrupt macro-structural integrity, followed by fine pulverization employing an ultra-centrifugal mill (ZM200, Retsch GmbH, Haan, Germany) equipped with a 0.5 mm aperture sieve to achieve an analytically homogenous flour.

2.2 Nutritional and Compositional Characterization

2.2.1 Proximate Composition Analysis

Proximate chemical constituents were determined in accordance with the standardized protocols established by the Association of Official Analytical Chemists [AOAC] (2019).

- **Moisture Content:** Evaluated gravimetrically via thermal dehydration at 105 °C for 24 hours until constant mass was achieved.
- **Crude Ash:** Quantified by the incineration of the organic matrix in a muffle furnace at 550°C overnight.
- **Crude Lipid Fraction:** Extracted via the continuous Soxhlet reflux method utilizing analytical-grade hexane.
- **Crude protein:** Total nitrogen content was determined employing the Kjeldahl method, via a macro-digestion block (DK 20, VELP Scientifica Srl, Usmate, Italy) for sample mineralization and a semi-automated distillation unit (UDK 139, VELP Scientifica) for ammonia liberation. Total protein content was calculated utilizing the specific legume nitrogen-to-protein conversion factor of 5.4 (Mariotti *et al.*, 2008).
- **Total Carbohydrate:** Estimated by difference utilizing the following empirical formula:

$$[100 - (\text{protein} + \text{fat} + \text{ash} + \text{moisture})]$$

2.2.2 LC-MS/MS Profiling of Amino Acids

Amino acid profiles were resolved following a modified acid hydrolysis protocol based on the method described by (Özcan & Şenyuva, 2006). A total of 100 mg of homogenized bean flour was mixed with 8 mL of 6 mol. L⁻¹ HCl and subjected to hydrothermal hydrolysis at 110°C for 24 h within sealed, nitrogen-flushed ampoules. Post-hydrolysis, the cooled matrices were quantitatively diluted to 10 mL with ultra-pure water.

An aliquot of 2 µL was injected into a liquid chromatography-tandem mass spectrometry system (LC-MS/MS, Thermo Fisher Scientific) equipped with a Phenomenex C18 analytical column (250 µm × 4.6 µm × 3 µm), and coupled to a Fortis TSQ triple-quadrupole mass

spectrometer operating in Selected Ion Monitoring (SIM) positive ionization mode.

The binary mobile phase consisted of Eluant A (3% MeOH, 0.2% formic acid, and 0.01% acetic acid v/v/v) and Eluant B (50% MeOH, 0.2% formic acid, and 0.1% acetic acid v/v/v), under an optimized gradient profile: an initial composition of 94% A and 6% B was maintained for 8 minutes, linearly adjusted to 80% A and 20% B over 20 min, held constant for 27 min, restored to initial conditions at 28 min, and equilibrated until the terminal run time of 40 min.

2.2.3 GC Profiling of Fatty Acids

Lipophilic profiles were analyzed following the derivatization of free fatty acids into volatile fatty acid methyl esters (FAME) according to ISO standards (ISO, 2011). Briefly, 500 mg of flour was extracted with 5 mL of analytical-grade hexane under rigorous mechanical agitation. The mixture was saponified by adding 200 µL of a methanolic potassium hydroxide (2N), followed by transesterification via the addition of 1 g of anhydrous sodium hydrogen sulfate. The resulting mixture was vortexed and centrifuged: the upper lipophilic supernatant was collected and passed through a 0.25 µm polytetrafluoroethylene (PTFE) syringe filter (Bahorun *et al.*, 1996).

Chromatographic separation of the FAME aliquots was executed on a gas chromatograph equipped with a flame ionization detector (GC-FID, Clarus 580, Perkin-Elmer) employing a highly polar capillary column (60 m × 0.25 mm internal diameter × 0.25 µm). The initial oven temperature was established at 85 °C for 4 min, ramped to 175°C at a rate of 15 °C.min⁻¹ (held for 15 min), increased to 220 °C at 25 °C.min⁻¹ (held for 25 min), and reached a final ceiling of 240°C at a rate of 4 °C.min⁻¹ (held for 10 min). The injector and detector manifolds were maintained at 220 °C and 140°C, respectively. Ultra-pure nitrogen served as the carrier gas at a constant volumetric flow rate of 40 mL.min⁻¹.

2.2.4 ICP-OES Minerals Determination

The quantitation of eleven mineral elements (Na, K, Mg, Ca, P, S, Mn, Zn, Cu, B and Fe) was achieved via wet closed-vessel hydrogen peroxide-nitric acid (1:3 ratio) digestion adapting the protocols outlined by Amarakoon *et al.* (2012) and Thavarajah *et al.* (2009). After digestion, mineral concentrations were determined through an Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES, iCAP PRO Series, Thermo Fisher Scientific). Trace elements (Mn, Zn, Cu, B and Fe) were monitored via an axial plasma configuration to optimize sensitivity, whereas macrominerals (Na, K, Mg, Ca, P and S) were resolved via radial plasma view to accommodate higher linear dynamic concentrations.

2.3 Phytochemical Profiling and Antioxidant Capacity

Due to the photosensitive nature of polyphenolic secondary metabolites, all extraction and analytical protocols were executed under attenuated light conditions. Spectrophotometric measurements were performed through a double-beam UV-Vis spectrophotometer (GENESYS 10S, Thermo Fisher Scientific).

2.3.1 Hydro-Acetic Extraction Protocol

A 1 g sample of flour was mixed with 40 mL of an aqueous acetone solvent system (50:50, v/v). Solid-liquid extraction was carried out over a 4-h window within a thermostatically controlled water bath set at 40 °C under continuous reciprocal shaking. The obtained extracts were centrifuged at 3600 rpm for 15 min, and the resulting clear supernatant was recovered and filtered through Whatman No.1 filter paper. The liquid extracts were cryopreserved at -20°C until analysis (Turkmen *et al.*, 2006).

2.3.2 Total Polyphenolic Contents (TPC)

Total polyphenolic content was determined via the Folin–Ciocalteu colorimetric assay as outlined by Singleton & Rossi (1965). Absorbance was measured at 760 nm, and results were expressed as mg gallic acid equivalents per gram of dry matter (mg GAE.g⁻¹ DM).

2.3.3 Total Flavonoid Content (TFC)

Total flavonoid content was determined employing the aluminum chloride colorimetric method as described by Bahorun *et al.* (1996). Absorbance was determined at 430 nm, and results were expressed as milligrams of quercetin equivalents per gram of dry matter (mg QE.g⁻¹ DM).

2.3.4 Condensed Tannin Fraction

Condensed tannin content was determined through the vanillin assay in acidic medium in accordance with the method of Julkunen-Tiitto (1985). Absorbance was measured at 500 nm, and results were expressed as mg catechin equivalents per gram of dry matter (mg CE.g⁻¹ DM).

2.3.5 Antioxidant Activities

The radical quenching and reducing capacities of the extracts were validated using three independent colorimetric mechanisms as described by Desta *et al.* (2022):

- DPPH• Radical Scavenging Activity: Assessed by tracking the radical reduction of 2,2-diphenyl-1-picrylhydrazyl at 517 nm. Results were computed against an ascorbic acid reference calibration ($R^2 = 0.9996$) and are expressed as milligrams of ascorbic acid equivalents per gram of dry seed weight (mg AAE.g⁻¹)

- ABTS•+ Radical Scavenging Activity: Determined by measuring the discoloration of the 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) radical cation at 734 nm. The kinetic responses were evaluated against a Trolox calibration curve ($R^2 = 0.9997$) and are reported as milligrams of Trolox equivalents per gram of dry seed weight (mg TE.g⁻¹)
- Ferric Reducing Antioxidant Power (FRAP): measured at 593 nm. The reducing capacity was calibrated employing ascorbic acid ($R^2 = 0.9990$) and reported as mg AAE.g⁻¹.

2.4 Statistical Modeling and Analysis

All experimental data were analyzed using the Generalized Linear Model (GLM) procedure within SAS statistical software (version 9.1.3, SAS Institute, Cary, NC, USA). The model included Variety and State (Whole vs. Dehulled) as fixed factors, with their interaction. Post-hoc comparisons among varieties were performed using Duncan's multiple range test (DMRT) at a strict statistical significance threshold of $p < 0.05$.

3 RESULTS

3.1 Proximate Composition

Moisture Dynamics

The proximate composition profiles across the selected faba bean cultivars are presented in Table 1. The residual moisture fractions were significantly influenced ($p < 0.05$) by the individual genotypic background and the mechanical decortication process. Within the whole seeds (WS) matrices, moisture content varied from 8.41% in the Alfia5 cultivar to 9.98% in the Defes cultivar.

The dehulled seed (DS) matrices displayed a distinct moisture range, shifting from a minimum threshold of 8.89% in Lobab to a maximum of 10.33% in Alfia5.

Total Inorganic Matter (Crude Ash)

The total crude ash fraction, representing the baseline inorganic mineral pool, exhibited significant cultivar-dependent variation. Values ranged from 3.06 to 3.68% within the WS matrices, and from 3.11 to 3.79% within the corresponding DS flours.

The highest absolute mineral concentrations were observed in the Hiba cultivar for the whole seeds and Alfia 5 cultivar for the dehulled seeds. Statistical evaluation revealed that mechanical dehulling did not exert a significant main effect on total inorganic ash levels.

Table 1. Proximate Composition of the Nine Faba Bean Varieties Flour (g.100g⁻¹ Dry Matter)

Varieties	Moisture		Ash		Crude fat		Protein		Carbohydrate	
	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS
Zina	9.23 ± 0.00 ^d	9.88 ± 0.12 ^b	3.33 ± 0.03 ^c	3.21 ± 0.02 ^f	1.84 ± 0.05 ^a	1.96 ± 0.01 ^a	25.46 ± 0.08 ^a	28.43 ± 0.24 ^a	60.14 ± 0.15 ^b	56.51 ± 0.36 ^g
Lobab	9.89 ± 0.00 ^a	8.89 ± 0.09 ^f	3.21 ± 0.03 ^d	3.35 ± 0.01 ^d	1.40 ± 0.04 ^e	1.47 ± 0.05 ^{ef}	21.55 ± 0.24 ^g	27.58 ± 0.10 ^c	63.95 ± 0.30 ^b	58.70 ± 0.23 ^c
Hiba	8.57 ± 0.03 ^f	10.21 ± 0.23 ^a	3.68 ± 0.02 ^a	3.57 ± 0.01 ^b	1.46 ± 0.03 ^e	1.53 ± 0.06 ^e	25.13 ± 0.09 ^b	27.69 ± 0.08 ^c	61.16 ± 0.17 ^f	57.00 ± 0.30 ^{ef}
Alfia5	8.41 ± 0.03 ^f	10.33 ± 0.12 ^a	3.64 ± 0.02 ^b	3.79 ± 0.02 ^a	1.27 ± 0.03 ^f	1.43 ± 0.09 ^f	23.73 ± 0.21 ^d	27.77 ± 0.12 ^b	62.95 ± 0.24 ^c	56.68 ± 0.12 ^g
Alfia17	8.86 ± 0.03 ^e	9.28 ± 0.03 ^d	3.06 ± 0.01 ^e	3.26 ± 0.02 ^e	1.72 ± 0.06 ^b	1.83 ± 0.03 ^{bc}	22.48 ± 0.25 ^f	27.77 ± 0.12 ^b	63.88 ± 0.31 ^b	57.86 ± 0.19 ^d
Alfia21	8.52 ± 0.05 ^f	9.65 ± 0.04 ^c	3.17 ± 0.02 ^d	3.34 ± 0.02 ^d	1.57 ± 0.04 ^d	1.66 ± 0.03 ^d	20.05 ± 0.19 ^h	22.09 ± 0.17 ^e	66.68 ± 0.18 ^a	63.26 ± 0.20 ^a
Defes	9.98 ± 0.12 ^a	9.36 ± 0.12 ^d	3.34 ± 0.02 ^c	3.50 ± 0.01 ^c	1.75 ± 0.03 ^b	1.86 ± 0.04 ^b	24.23 ± 0.10 ^c	27.97 ± 0.09 ^b	60.70 ± 0.20 ^g	57.31 ± 0.26 ^e
Aguadulce	9.59 ± 0.20 ^b	9.58 ± 0.19 ^c	3.65 ± 0.02 ^{ab}	3.23 ± 0.02 ^{ef}	1.66 ± 0.05 ^c	1.76 ± 0.03 ^c	23.38 ± 0.15 ^e	26.79 ± 0.10 ^d	61.72 ± 0.38 ^e	58.63 ± 0.29 ^c
Karabiga	9.42 ± 0.13 ^c	9.11 ± 0.03 ^e	3.18 ± 0.02 ^d	3.11 ± 0.00 ^g	1.18 ± 0.04 ^g	1.29 ± 0.04 ^g	23.96 ± 0.21 ^{cd}	26.97 ± 0.21 ^d	62.25 ± 0.19 ^d	59.52 ± 0.17 ^b
Dehulling effect	***		-		***		***		***	

Note: Data presented as means of triplicates ± standard deviation. The values within the same column with the same letter are not significantly different ($p > 0.05$)^a. Asterisks indicate significance at * $p \leq 0.05$, ** $p \leq 0.01$, or *** $p \leq 0.001$ -no significance

Crude Lipid Dynamics

Although the baseline crude fat content of *Vicia faba* seeds is minor, it varied significantly ($p < 0.05$) among the different cultivars in both WS and DS. The Zina cultivar exhibited the highest lipid accumulation ($1.84 \text{ g} \cdot 100^{-1} \text{ g DM}$), whereas Karabiga possessed the lowest concentration ($1.18 \text{ g} \cdot 100^{-1} \text{ g DM}$). Dehulling increased crude fat across all cultivars from 4.79 to 12.60%. The Variety $\times$ State interaction was not significant for fat as displayed in Table S1.

Crude Protein Alterations

Protein content was highly sensitive to both genotypic differences and processing states ($p < 0.05$). Within the WS configuration, baseline protein levels ranged from 20.05% in the Alfia 21 cultivar to a maximum of 25.46% in the Zina cultivar. Dehulling induced a significant relative enrichment in protein across all genotypes. In the DS flours, total protein levels increased to a range between 22.09% (Alfia 21) and 28.43% (Zina). This represents a relative protein increase of 10.17 to 27.98% post-decortication.

Total Carbohydrate Profiles

Total carbohydrate content varied significantly among the evaluated cultivars, ranging from 60.14% to 66.68% in the WS forms, and decreasing to a range of 56.52% to 63.26% within the DS matrices. The lowest carbohydrate concentrations were observed in the Zina cultivar, matching its high protein profile, while the highest values were found in the Alfia 21 cultivar. Mechanical decortication (dehulling) exerted a significant reducing effect on total carbohydrates, driving net decreases ranging from 4.38% to 9.96% across the cultivars.

3.2 Amino Acids (AA) Composition and Proteomic Quality

The detailed profiles of indispensable (essential) and non-indispensable for both the WS and DS forms are presented in Table 2. Among the indispensable amino acids (IAAs), lysine and leucine predominated: lysine ranged from $72.51 \text{ mg} \cdot \text{g}^{-1}$ protein in Alfia 5 to $82.73 \text{ mg} \cdot \text{g}^{-1}$ in Alfia 17. Leucine concentrations exhibited a narrow range, from 69.71 mg/g protein (Hiba) to $72.36 \text{ mg} \cdot \text{g}^{-1}$ protein (Alfia 17). Conversely, isoleucine was identified as the limiting IAA across all samples, displaying the lowest relative abundance ($22.03\text{--}34.89 \text{ mg} \cdot \text{g}^{-1}$ protein).

Among the non-indispensable amino acids (NIAAs), aspartic acid, asparagine, arginine, and glutamic acid were highly abundant ($95.16\text{--}117.48 \text{ mg} \cdot \text{g}^{-1}$; $100.33\text{--}104.46 \text{ mg} \cdot \text{g}^{-1}$; $88.37\text{--}120.61 \text{ mg} \cdot \text{g}^{-1}$; and $138.05\text{--}148.60 \text{ mg} \cdot \text{g}^{-1}$ protein, respectively). Cysteine was the least abundant NIAA ($12.08\text{--}15.85 \text{ mg} \cdot \text{g}^{-1}$ protein). Tryptophan and methionine

were not detected in the extracts, likely due to degradation during acid-catalyzed HCl hydrolysis.

3.3 Lipophilic Profiling and Fatty Acid Alterations

The FA composition of the studied faba bean cultivars, in is detailed in Table 3. The dehulling process significantly affected the content of most FAs, inducing a decrease in saturated fatty acids (SFAs). Palmitic acid (C16:0), was significantly ($p < 0.001$) affected by both processing and variety, ranged from 15.51 to 19.19% in WS and dropped to 14.69–18.66% in DS, with reduction rates spanning 1.53% to -17.35% depending on the variety. Stearic acid (C18:0) was similarly affected by both factors ($p < 0.01$ and $p < 0.001$, respectively), decreasing from 1.62–2.87% in WS to 1.38–2.67% in DS. In most varieties, the reduction ranged from 8.64% to 40.00%, with the exception of Defes and Alfia17, which exhibited increases of 13.62% and 19.39%, respectively. As a result, total SFA decreased significantly ($p < 0.001$) from 17.92–22.18% in WS to 16.63–20.70% in DS, with reduction rates between -1.11% and -14.99% . The Cultivar $\times$ Processing State interaction did not significantly influence SFA dynamics as displayed in Supplementary Data Table S1.

Conversely, unsaturated fatty acids (UFAs) increased post-dehulling, rising from 78.31%–82.29% in WS to 79.65%–84.01% in DS (a relative increase of $+0.28\%$ to $+4.26\%$). Oleic acid (C18:1), was significantly influenced by cultivar ($p < 0.001$) but was unaffected by processing ($p > 0.05$), maintaining stable levels across both states ($23.00\%\text{--}31.59\%$ in WS vs. $22.38\%\text{--}31.36\%$ in DS). Linoleic acid (C18:2), the predominant polyunsaturated fatty acid (PUFA) in faba beans, was highly sensitive to both processing ($p < 0.001$) and cultivar ($p < 0.001$). Dehulling significantly increased linoleic acid levels, which rose from 45.31–54.42% (WS) to 49.56–58.19% (DS), reflecting a relative increase of $+1.91\%$ to $+12.59\%$ (with the sole exception of Alfia17 (-1.95%)). A significant Cultivar $\times$ Processing State interaction ($p < 0.05$) indicate that the linoleic acid response to dehulling is genotype-dependent. Linolenic acid (C18:3) displayed the most variable response to dehulling, being significantly influenced by processing ($p < 0.01$), cultivar ($p < 0.001$), and their interaction ($p < 0.001$). Its values ranged from 0.27–3.01% (WS) to 0.26–3.41% (DS), reflecting highly contrasting genotypic responses that ranged from sharp decreases (-88.55% in Hiba) to substantial increases ($+1114.81\%$ in Alfia 17). Finally, docosapentaenoic acid DPA levels were significantly reduced by dehulling ($p < 0.001$) and varied considerably across cultivars ($p < 0.001$), declining from 0.53–3.62% (WS) to 0.04–1.52% (DS), represents a net reduction of -18.87% to -96.12% .

Table 2. Amino acid compositions of the nine whole and dehulled faba bean varieties, expressed as mg of amino acid per g of protein

Varieties	Zina		Lobab		Hiba		Alfia5		Alfia17		Alfia21		Defes		Aguadulce		Karabiga		mg/kg per day ^A	protein ^B mg/g	
	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS			
Essential amino acids																					
Histidine	33.28	36.55	37.20	36.31	25.37	27.72	39.66	38.23	35.61	37.26	35.74	36.17	37.47	36.17	30.55	35.49	35.47	38.97	10	15	
Isoleucine	25.88	33.21	33.86	22.98	22.03	34.38	26.32	34.89	22.27	33.92	22.40	32.83	24.13	22.21	22.21	32.15	22.13	25.63	20	30	
Leucine	70.23	70.81	71.41	69.21	69.71	70.50	72.07	72.29	72.36	68.55	71.46	68.67	70.45	70.27	69.88	69.83	69.81	73.04	39	59	
Lysine	79.85	81.06	81.71	80.83	79.89	72.32	72.51	74.24	82.73	80.13	81.77	80.26	80.69	81.97	80.07	80.01	79.99	73.55	30	45	
Phenylalanine	40.55	42.82	43.41	42.61	41.75	43.89	44.07	45.65	44.35	41.97	43.47	42.08	42.48	43.66	41.91	41.86	41.84	45.02	25 ^c	38 ^c	
Threonine	38.32	42.85	43.44	39.45	41.78	43.92	44.10	45.68	44.38	42.00	43.50	42.11	42.51	43.69	41.94	41.89	42.49	45.05	15	23	
Valine	34.79	37.47	37.97	37.29	36.56	38.37	38.52	39.86	38.76	36.75	38.02	36.85	37.18	38.18	36.70	36.65	36.64	39.33	26	10	
Non-essential amino acids																					
Aspartic acid	102.95	108.48	109.13	108.25	99.30	109.65	109.85	99.59	95.16	107.54	109.19	107.67	108.10	111.40	117.48	107.42	98.40	100.90	-	-	
Asparagine	100.33	102.78	103.43	102.55	101.60	103.95	104.15	105.89	104.46	101.84	103.49	101.97	102.40	103.70	101.78	101.72	101.70	105.20	-	-	
Arginine	98.21	113.69	114.34	113.46	98.51	92.86	115.06	116.80	88.37	100.75	92.40	97.88	99.31	114.61	112.69	111.63	120.61	110.11	-	-	
Alanine	52.18	65.66	62.31	65.43	64.48	56.83	67.03	58.77	67.34	64.72	46.37	64.85	55.28	66.58	64.66	64.60	54.58	58.08	-	-	
Cysteine	12.08	13.48	14.13	13.25	12.30	14.65	15.85	16.59	14.16	12.54	14.19	12.67	13.10	15.40	12.48	12.42	12.40	15.90	-	-	
Glutamic acid	143.2	136.68	147.33	146.45	145.50	147.85	138.05	149.79	140.36	155.74	147.39	142.87	145.30	147.60	145.68	138.62	148.60	149.10	-	-	
Glycine	32.77	36.31	36.96	36.08	35.13	37.48	37.68	39.42	37.99	35.37	37.02	35.50	35.93	37.23	35.31	35.25	35.23	38.73	-	-	
Serine	41.3	54.48	52.06	48.58	52.71	51.83	50.88	53.23	53.43	55.17	53.74	51.12	52.77	51.25	51.68	52.98	51.06	51.00	-	-	
Taurine	25.149	32.239	31.029	24.289	31.35	30.914	30.439	31.614	31.71	32.58	31.87	30.56	31.384	30.624	30.84	31.489	30.53	30.50	-	-	
Proline	38.92	38.12	41.70	35.48	45.28	50.06	49.86	48.64	53.44	47.22	57.02	45.80	61.60	44.38	62.18	30.96	60.76	34.54	-	-	

Note: A: Amino acid recommendations, mg/kg¹ body mass per day for adults (WHO/ FAO/UNU, 2007). B: Amino acid recommendations, mg/kg¹ protein consumed for adults (WHO/ FAO/UNU, 2007). C: Tyrosine and Phenylalanine

Table 3. Fatty Acids (FA) Profile of the Nine Faba Bean Varieties, Expressed as gram of FA per 100g of Total Amount

Varieties	Palmitic acid			Stearic acid			Oleic acid			Linoleic acid			DPA ^A			TSPA ^B			TUPA ^C		
	WS	DS		WS	DS		WS	DS		WS	DS		WS	DS		WS	DS		WS	DS	
Zina	17.56 ± 0.64 ^{ab}	16.23 ± 0.48 ^{ab}	2.37 ± 0.05 ^a	1.87 ± 0.35 ^{ab}	27.64 ± 0.06 ^{ab}	28.06 ± 0.07 ^a	45.31 ± 1.15 ^a	50.33 ± 0.27 ^a	2.48 ± 0.02 ^{ab}	2.56 ± 0.19 ^{ab}	2.48 ± 0.02 ^{ab}	2.56 ± 0.19 ^{ab}	3.62 ± 0.27 ^a	1.52 ± 0.25 ^a	20.00 ± 0.99 ^c	18.11 ± 0.97 ^{abcde}	20.00 ± 0.99 ^c	18.11 ± 0.97 ^{abcde}	79.11 ± 0.54 ^c	82.48 ± 0.95 ^{ab}	
	17.60 ± 0.31 ^{abc}	16.69 ± 0.14 ^a	2.56 ± 0.27 ^{ab}	1.65 ± 0.23 ^a	24.93 ± 0.88 ^b	25.85 ± 0.71 ^{ac}	50.49 ± 0.50 ^{bc}	53.25 ± 0.79 ^a	3.01 ± 0.37 ^b	2.75 ± 0.72 ^{ab}	3.01 ± 0.37 ^b	2.75 ± 0.72 ^{ab}	1.90 ± 1.27 ^b	0.88 ± 0.28 ^b	19.38 ± 1.18 ^c	18.13 ± 0.34 ^{abcde}	19.38 ± 1.18 ^c	18.13 ± 0.34 ^{abcde}	79.15 ± 2.07 ^c	81.81 ± 0.71 ^{bc}	
Hiba	16.78 ± 0.38 ^{de}	14.69 ± 0.45 ^c	1.62 ± 0.02 ^c	1.48 ± 0.17 ^b	27.08 ± 0.67 ^a	28.42 ± 0.18 ^b	53.82 ± 2.14 ^{cd}	54.85 ± 0.53 ^c	2.27 ± 0.67 ^{abc}	2.27 ± 0.11 ^c	2.27 ± 0.67 ^{abc}	2.27 ± 0.11 ^c	0.65 ± 0.10 ^c	0.06 ± 0.02 ^d	17.92 ± 0.73 ^d	16.63 ± 0.07 ^e	17.92 ± 0.73 ^d	16.63 ± 0.07 ^e	82.09 ± 1.26 ^{ab}	84.01 ± 0.82 ^a	
	19.19 ± 0.87 ^d	15.86 ± 0.39 ^{abc}	2.30 ± 0.29 ^a	1.38 ± 0.38 ^a	24.50 ± 0.37 ^b	23.76 ± 1.06 ^{cd}	52.57 ± 0.28 ^{cd}	58.19 ± 0.18 ^d	1.96 ± 0.30 ^c	0.32 ± 0.06 ^c	1.96 ± 0.30 ^c	0.32 ± 0.06 ^c	1.05 ± 0.02 ^c	0.12 ± 0.01 ^d	20.75 ± 0.16 ^{bc}	17.64 ± 1.10 ^{de}	20.75 ± 0.16 ^{bc}	17.64 ± 1.10 ^{de}	78.79 ± 0.43 ^c	80.77 ± 1.01 ^{bcd}	
Alfia17	19.00 ± 0.25 ^d	18.18 ± 0.40 ^d	1.65 ± 0.11 ^c	1.97 ± 0.37 ^{ab}	24.25 ± 0.14 ^{bc}	24.25 ± 0.57 ^{cd}	54.42 ± 0.45 ^d	53.36 ± 0.27 ^{bc}	0.34 ± 0.05 ^d	2.98 ± 0.29 ^{ab}	0.34 ± 0.05 ^d	2.98 ± 0.29 ^{ab}	0.53 ± 0.05 ^c	0.43 ± 0.12 ^c	20.21 ± 0.77 ^c	19.56 ± 0.69 ^{ab}	20.21 ± 0.77 ^c	19.56 ± 0.69 ^{ab}	79.77 ± 0.68 ^{bc}	80.12 ± 0.78 ^{cd}	
	17.36 ± 0.15 ^d	15.85 ± 0.18 ^{abc}	2.65 ± 0.15 ^b	2.26 ± 0.28 ^{ab}	28.22 ± 0.15 ^a	27.42 ± 0.11 ^{ab}	47.28 ± 0.16 ^{ab}	53.23 ± 0.17 ^b	2.84 ± 0.08 ^{ab}	3.41 ± 0.16 ^a	2.84 ± 0.08 ^{ab}	3.41 ± 0.16 ^a	1.82 ± 0.21 ^b	0.11 ± 0.04 ^d	20.47 ± 0.19 ^c	18.45 ± 0.22 ^{bcd}	20.47 ± 0.19 ^c	18.45 ± 0.22 ^{bcd}	80.59 ± 0.42 ^{abc}	81.41 ± 0.45 ^{bcd}	
Defes	15.51 ± 0.09 ^e	15.04 ± 0.51 ^{bc}	2.35 ± 0.19 ^a	2.67 ± 0.18 ^b	31.59 ± 0.33 ^d	31.36 ± 0.09 ^e	47.57 ± 0.03 ^{ab}	49.56 ± 0.10 ^a	2.58 ± 0.19 ^{abc}	3.05 ± 0.46 ^{ab}	2.58 ± 0.19 ^{abc}	3.05 ± 0.46 ^{ab}	1.03 ± 0.06 ^c	0.04 ± 0.01 ^d	18.03 ± 0.25 ^d	17.83 ± 1.09 ^{abcde}	18.03 ± 0.25 ^d	17.83 ± 1.09 ^{abcde}	82.29 ± 0.42 ^a	82.52 ± 1.46 ^{ab}	
	18.48 ± 0.27 ^{cd}	15.93 ± 0.59 ^{abc}	2.87 ± 0.08 ^b	2.22 ± 0.53 ^{ab}	25.17 ± 0.46 ^{ab}	26.67 ± 0.46 ^{ab}	50.26 ± 0.89 ^{bc}	52.75 ± 0.08 ^b	0.27 ± 0.04 ^d	3.28 ± 0.24 ^{ab}	0.27 ± 0.04 ^d	3.28 ± 0.24 ^{ab}	1.85 ± 0.16 ^b	0.20 ± 0.02 ^{cd}	21.83 ± 0.78 ^{ab}	19.45 ± 0.08 ^{abc}	21.83 ± 0.78 ^{ab}	19.45 ± 0.08 ^{abc}	78.31 ± 1.61 ^c	81.57 ± 1.25 ^{bcd}	
Karabiga	18.95 ± 0.68 ^{cd}	18.66 ± 0.65 ^d	2.72 ± 0.14 ^{ab}	1.91 ± 0.01 ^c	23.00 ± 0.01 ^c	22.38 ± 0.68 ^d	50.53 ± 0.20 ^{bc}	53.83 ± 0.47 ^{bc}	2.20 ± 0.17 ^{bc}	2.46 ± 0.25 ^b	2.20 ± 0.17 ^{bc}	2.46 ± 0.25 ^b	1.71 ± 0.21 ^b	0.42 ± 0.04 ^c	22.18 ± 0.76 ^a	20.70 ± 0.91 ^a	22.18 ± 0.76 ^a	20.70 ± 0.91 ^a	78.61 ± 1.08 ^c	79.65 ± 0.96 ^d	

Note: Data presented like means of triplicates ± standard deviation. The values within the same column with the same letter are not significantly different ($p > 0.05$). A: Docosapentaenoic Acid, B: Total Saturated Fatty Acids, C: Total unsaturated Fatty Acids

3.4 Mineral Profile and Elemental Flux

The quantitative distribution of macro-elements (Na, K, Mg, Ca, P, S) and micro-elements (Mn, Zn, Cu, B, Fe) within the whole seed (WS) and dehulled seed (DS) matrices across the nine faba bean cultivars is compiled in Table 4 and Tables 5, respectively.

Potassium (K) emerged as the predominant macro-mineral across all investigated genotypes, ranging from 931.16 mg.100 g⁻¹ DM in the Karabiga cultivar to 1166.52 mg.100 g⁻¹ DM in the Hiba cultivar. This was followed by phosphorus (P), which varied from 426.00 mg.100 g⁻¹ DM (Zina) to 620.10 mg.100 g⁻¹ DM (Defes). Calcium (Ca), magnesium (Mg), and sulfur (S) occurred at moderate concentrations ($\approx 94 - 200$ mg.100 g⁻¹ DM), whereas Sodium (Na) represented the minor macro-elemental fraction (35.52 mg.100 g⁻¹ DM (Hiba) and 81.94 mg.100 g⁻¹ DM (Alfia 5).

Among the trace elements, iron (Fe) and zinc (Zn) predominated. Fe ranged from 5.67 mg.100 g⁻¹ DM (Alfia 5) to 8.63 mg.100 g⁻¹ DM (Alfia 21), while Zn ranged from 2.48 mg.100 g⁻¹ DM (Alfia 17) to 7.46 mg.100 g⁻¹ DM (Hiba).

Dehulling markedly reduced Ca levels (-8.81% to -52.90%) and increased P levels (+7.46% to +20.50%) and S concentrations (+7.80% to +16.16%). With the singular exception of Zn, most minerals were significantly affected by dehulling ($p < 0.05$). Significant varietal effects were observed across all minerals ($p < 0.05$).

3.5 Total Polyphenols Content (TPC)

The phenolic profiles of the studied faba bean cultivars, analyzed in both whole and dehulled forms, are presented in Table 6. Dehulling significantly reduced the TPC across all studied varieties. TPC values decreased in all nine varieties following dehulling, with values declining from 2.86 – 4.26 mg GAE.g⁻¹ DW in whole seeds to 2.13 – 3.88 mg GAE.g⁻¹ DW in dehulled seeds. The rate of decrease ranged from -8.31% (Hiba) to -36.77% (Alfia 17), indicating that Hiba was the least affected by dehulling, while Alfia 17 suffered the greatest proportional loss.

3.6 Condensed Tannins content (CTC)

As for TPC, condensed tannin content (CTC) was reduced in all varieties after dehulling, dropping from 2.38 – 2.88 mg CE.g⁻¹ DW (WS) to 1.52 – 2.42 mg CE.g⁻¹ DW (DS), with reduction percentages ranging from -13.26% in Aguadulce to -43.91% in Lobab, confirming that tannins are substantially concentrated in the hull fraction.

3.7 Total Flavonoids Content (TFC)

The total flavonoid content (TFC) exhibited the most variable response to dehulling, with values ranging from 0.20

– 0.62 mg QE.g⁻¹ DW (WS) to 0.03–0.35 mg QE.g⁻¹ DW (DS). Reduction rates spanned from -85.0% in Karabiga to -21.2% in Aguadulce, with the notable exception of Alfia21, which exhibited a slight increase of +12.9% following dehulling (from 0.31 to 0.35 mg QE.g⁻¹ DW), suggesting a possible redistribution or differential localization of flavonoids between the hull and cotyledon fractions in this particular variety. Overall, these results confirm that the hull constitutes a major reservoir of phenolic compounds, and its removal through dehulling leads to a significant reduction in the antioxidant related phytochemical content of the seeds, with the extent of this reduction being variety-dependent.

3.8 Antioxidant Activities

The multi-mechanism antioxidant capacities of the whole and dehulled faba bean flours, resolved via the DPPH•, ABTS•+, and FRAP configurations, are compiled in Table 6. Dehulling significantly reduced the antioxidant activity of seeds across all varieties and for all three assessed antioxidant properties ($p < 0.05$). DPPH radical scavenging activity declined from a WS range of 3.17 – 4.59 mg AAE.g⁻¹ DW to a DS range of 1.71 – 3.85 mg AAE.g⁻¹ DW following decortication. The percentage reduction ranged from -13.1% in the Alfia 5 cultivar and a maximal collapse of -52.4% in both Lobab and Aguadulce cultivars.

ABTS radical cation scavenging activity displayed a comparatively more moderate reduction following dehulling, with values decreasing from 4.04 – 5.13 mg TE.g⁻¹ DW (WS) to 3.68 – 4.39 mg TE.g⁻¹ DW (DS). The reduction rates ranged from -2.5% in Karabiga, which retained nearly all of its ABTS activity after decortication, to -25.5% in Lobab, suggesting that the hull contributes differentially to DPPH and ABTS scavenging capacities, and that hydrophilic antioxidants measured by ABTS may be more evenly distributed across seed fractions.

The ferric reducing antioxidant power (FRAP) was the most consistently and severely attenuated functional parameter across all samples. Net reducing capacity collapsed from 1.51 – 2.40 mg AAE.g⁻¹ DW (WS) to 0.88–1.46 mg AAE.g⁻¹ DW (DS). Reduction rates ranged from -22.1% in Hiba to -55.0% in Lobab, with majority of the evaluated cultivars exhibiting net losses exceeding 35%.

In summary, the relative loss of antioxidant performance triggered by mechanical decortication was consistently most severe within the Lobab cultivar across all three independent analytical assays. Conversely, the Alfia 5 and Karabiga exhibited the highest DPPH• and ABTS•+ scavenging activities. These findings highlight that decortication results in a substantial and assay dependent reduction in antioxidant potential, the extent of which is markedly modulated by the genetic background of the variety.

Table 4. Major Elements Composition of Faba Bean Varieties (mg.100g⁻¹)

Varieties	Na		K		Ca		Mg		P		S	
	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS
Zina	44.87±0.95 ^f	33.98±0.37 ^b	1069.37±1.87 ^c	1068.44±4.44 ^f	117.97±1.410 ^d	60.07±2.26 ^d	94.19±1.45 ^f	80.48±1.49 ^b	425.97±1.38 ^f	474.33±2.47 ^f	159.72±1.73 ^f	177.19±0.84 ^f
Lobab	43.80±0.27 ^e	35.50±0.68 ^e	1003.48±4.04 ^e	1111.94±4.38 ^f	133.35±7.14b ^d	77.20±2.29 ^c	98.16±1.48 ^e	86.29±1.08 ^e	494.46±0.88 ^b	560.80±1.29 ^b	159.54±0.51 ^e	180.03±0.26 ^e
Hiba	35.52±0.58 ^b	35.50±0.86 ^c	1166.16±2.63 ^a	1220.24±8.14 ^b	153.38±0.39 ^f	100.91±1.62 ^b	104.30±0.55 ^d	92.40±0.73 ^f	537.00±1.46 ^f	647.10±2.04 ^e	179.68±0.65 ^e	196.26±1.07 ^e
Alfia5	81.93±0.13 ^a	65.69±0.61 ^b	1133.52±10.98 ^f	1233.84±3.64 ^f	137.62±11.56 ^{abc}	81.06±9.77 ^c	137.36±1.54 ^f	123.74±0.57 ^e	607.39±1.64 ^f	719.35±0.75 ^a	199.22±1.07 ^e	231.46±0.49 ^e
Alfia17	70.01±0.16 ^b	54.22±0.36 ^c	1013.94±2.67 ^d	1021.14±0.99 ^b	113.46±0.59 ^f	103.48±8.35 ^b	112.48±1.51 ^c	92.53±1.77 ^f	597.56±1.57 ^e	642.17±1.88 ^d	178.24±0.79 ^e	192.09±0.12 ^d
Alfia21	57.84±0.52 ^d	43.12±0.31 ^d	977.06±4.27 ^f	1055.44±1.60 ^f	145.55±10.13 ^{ab}	83.78±5.35 ^c	110.46±1.35 ^c	94.81±0.37 ^e	566.74±0.97 ^f	637.52±0.38 ^e	159.17±0.71 ^e	173.44±0.49 ^e
Defes	62.59±0.33 ^c	79.23±1.19 ^a	1012.35±6.89 ^{de}	1161.11±1.45 ^e	145.98±0.84 ^{ab}	117.18±0.46 ^e	105.49±0.81 ^d	111.96±0.29 ^b	620.09±0.84 ^f	695.61±2.51 ^b	172.73±0.24 ^d	196.55±0.15 ^e
Aguadulce	43.35±0.39 ^e	37.70±0.55 ^f	1166.52±3.52 ^e	1134.64±2.54 ^d	127.17±7.74 ^{de}	59.89±0.15 ^d	121.44±0.98 ^b	102.76±0.35 ^e	584.47±0.91 ^d	630.95±0.89 ^f	156.13±0.75 ^f	173.08±0.33 ^e
Karabiga	50.62±0.59 ^f	41.03±0.59 ^f	931.15±0.85 ^f	1018.34±3.16 ^b	114.76±0.97 ^f	60.28±0.22 ^d	105.81±0.45 ^d	97.24±0.85 ^d	516.39±1.72 ^e	595.57±1.85 ^e	186.29±0.36 ^f	209.28±0.93 ^e

Note: Data presented like means of triplicates ± standard deviation. The values within the same column with the same letter are not significantly different ($p > 0.05$)

Table 5. Minor Elements Composition of Faba Bean Varieties (mg.100g⁻¹)

Varieties	Zn		Mn		Fe		Cu		B	
	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS
Zina	4.99 ± 0.23 ^b	5.39 ± 0.41 ^{ab}	0.89 ± 0.01 ^c	0.8 ± 0.01 ^c	6.95 ± 0.11 ^{bc}	6.58 ± 0.06 ^d	1.23 ± 0.06 ^{ab}	1.29 ± 0.04 ^f	0.51 ± 0.02 ^{bc}	0.29 ± 0.01 ^{bc}
Lobab	3.73 ± 0.70 ^{abd}	3.57 ± 0.30 ^{bd}	1.46 ± 0.01 ^a	1.26 ± 0.00 ^{ab}	6.79 ± 0.08 ^c	7.98 ± 0.04 ^{ab}	0.86 ± 0.03 ^c	1.02 ± 0.05 ^b	0.56 ± 0.03 ^{bc}	0.40 ± 0.00 ^{bc}
Hiba	7.46 ± 0.06 ^c	4.96 ± 0.08 ^{abc}	1.09 ± 0.02 ^c	1.02 ± 0.00 ^{bc}	7.36 ± 0.09 ^{ab}	7.67 ± 0.02 ^c	1.27 ± 0.04 ^{ab}	1.35 ± 0.03 ^b	0.81 ± 0.02 ^{abc}	0.55 ± 0.01 ^{abc}
Alfia5	4.75 ± 0.03 ^{bc}	6.25 ± 0.05 ^a	1.32 ± 0.01 ^{ab}	1.42 ± 0.00 ^a	5.67 ± 0.02 ^c	6.75 ± 0.05 ^{cd}	1.36 ± 0.05 ^a	1.73 ± 0.02 ^a	0.40 ± 0.06 ^{bc}	0.02 ± 0.01 ^c
Alfia17	2.48 ± 0.07 ^d	2.77 ± 0.06 ^d	1.21 ± 0.01 ^{ab}	1.00 ± 0.01 ^{bc}	7.40 ± 0.09 ^{ab}	7.66 ± 0.07 ^c	1.02 ± 0.07 ^{bc}	1.04 ± 0.02 ^b	1.48 ± 0.27 ^a	0.98 ± 0.05 ^c
Alfia21	3.34 ± 0.03 ^{bd}	3.31 ± 0.30 ^{cd}	1.23 ± 0.03 ^{ab}	1.17 ± 0.10 ^{ab}	8.62 ± 0.30 ^a	8.48 ± 0.52 ^{ab}	0.99 ± 0.05 ^{bc}	1.07 ± 0.03 ^b	1.02 ± 0.07 ^{ab}	0.66 ± 0.04 ^{ab}
Defes	2.77 ± 0.14 ^{bd}	4.31 ± 0.34 ^{ab}	1.13 ± 0.00 ^{bc}	1.14 ± 0.01 ^{abc}	6.93 ± 0.07 ^{bc}	8.11 ± 0.14 ^{ab}	1.17 ± 0.07 ^{ab}	1.34 ± 0.02 ^b	0.20 ± 0.01 ^c	0.06 ± 0.02 ^c
Aguadulce	3.52 ± 0.56 ^{bd}	3.09 ± 0.14 ^{cd}	1.11 ± 0.01 ^{bc}	0.92 ± 0.01 ^{bc}	7.86 ± 0.14 ^{ab}	7.75 ± 0.16 ^{bc}	1.21 ± 0.10 ^{ab}	1.13 ± 0.07 ^b	0.99 ± 0.05 ^{ab}	0.57 ± 0.06 ^{abc}
Karabiga	2.72 ± 0.13 ^{cd}	3.65 ± 0.50 ^{bd}	1.05 ± 0.00 ^{bc}	1.02 ± 0.00 ^{bc}	7.49 ± 0.02 ^{ab}	8.99 ± 0.07 ^a	1.02 ± 0.03 ^{bc}	1.23 ± 0.04 ^b	0.69 ± 0.02 ^{bc}	0.49 ± 0.00 ^{bc}

Note: Data presented like means of triplicates ± standard deviation. The values within the same column with the same letter are not significantly different ($p > 0.05$)

Table 6. Polyphenolic Composition and Radical Scavenging Activity of Faba Bean Cultivars

Varieties	TPC (mg GAE/g DW)		CTC (mg CE/g DW)		TFC (mg QE/g DW)		DPPH• (mg AAE/g DW)		ABTS•+ (mg TE/g DW)		FRAP (mg AAE/g DW)	
	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS	WS	DS
Zina	3.96 ± 0.06 ^{ab}	3.16 ± 0.17 ^{bc}	2.75 ± 0.20 ^{ab}	1.70 ± 0.05 ^b	0.36 ± 0.15 ^b	0.24 ± 0.16 ^b	4.41 ± 0.04 ^a	3.41 ± 0.28 ^{ab}	4.44 ± 0.02 ^{ab}	3.96 ± 0.01 ^{bcd}	1.61 ± 0.01 ^d	1.02 ± 0.01 ^{bc}
Lobab	3.35 ± 0.03 ^{bc}	2.66 ± 0.48 ^{bc}	2.71 ± 0.09 ^{ab}	1.52 ± 0.37 ^a	0.62 ± 0.02 ^a	0.12 ± 0.08 ^{ab}	4.41 ± 0.05 ^a	2.10 ± 0.01 ^{bc}	4.94 ± 0.03 ^a	3.68 ± 0.10 ^d	2.40 ± 0.03 ^a	1.08 ± 0.01 ^{bc}
Hiba	3.73 ± 0.16 ^{bc}	3.42 ± 0.07 ^{ab}	2.88 ± 0.12 ^a	2.15 ± 0.03 ^{ab}	0.22 ± 0.06 ^b	0.15 ± 0.04 ^{ab}	4.30 ± 0.07 ^a	3.08 ± 0.10 ^{bc}	5.13 ± 0.05 ^a	4.21 ± 0.05 ^{bc}	1.72 ± 0.03 ^{cd}	1.34 ± 0.06 ^{ab}
Alfia5	4.26 ± 0.01 ^a	3.88 ± 0.01 ^a	2.42 ± 0.23 ^b	1.62 ± 0.06 ^b	0.35 ± 0.10 ^b	0.17 ± 0.01 ^{ab}	4.43 ± 0.04 ^a	3.85 ± 0.07 ^a	4.18 ± 0.04 ^b	3.87 ± 0.01 ^{cd}	1.83 ± 0.07 ^{abcd}	1.17 ± 0.01 ^{abc}
Alfia 17	3.59 ± 0.00 ^{bc}	2.27 ± 0.27 ^{bc}	2.38 ± 0.19 ^b	1.75 ± 0.04 ^{ab}	0.26 ± 0.04 ^b	0.17 ± 0.01 ^{ab}	4.34 ± 0.03 ^a	2.36 ± 0.02 ^{bc}	4.67 ± 0.17 ^{ab}	4.32 ± 0.08 ^{ab}	1.96 ± 0.02 ^{abcd}	1.25 ± 0.04 ^{ab}
Alfia21	4.10 ± 0.09 ^a	2.97 ± 0.00 ^{bc}	2.61 ± 0.21 ^{ab}	1.98 ± 0.16 ^b	0.31 ± 0.05 ^b	0.35 ± 0.05 ^a	4.30 ± 0.00 ^a	2.49 ± 0.11 ^{bc}	4.68 ± 0.02 ^{ab}	4.39 ± 0.00 ^a	2.26 ± 0.03 ^{ab}	1.46 ± 0.04 ^a
Defes	3.09 ± 0.00 ^{bc}	2.51 ± 0.01 ^{bc}	2.82 ± 0.09 ^a	2.38 ± 0.30 ^a	0.23 ± 0.08 ^b	0.15 ± 0.06 ^{ab}	4.59 ± 0.08 ^a	2.84 ± 0.22 ^{bc}	4.52 ± 0.03 ^{ab}	3.87 ± 0.03 ^{cd}	1.59 ± 0.06 ^{cd}	1.01 ± 0.01 ^{bc}
Agnadulce	3.64 ± 0.05 ^{bc}	2.73 ± 0.05 ^{bc}	2.79 ± 0.05 ^a	2.42 ± 0.05 ^a	0.33 ± 0.01 ^b	0.26 ± 0.12 ^{ab}	4.43 ± 0.01 ^a	2.11 ± 0.05 ^{bc}	5.10 ± 0.03 ^a	4.07 ± 0.02 ^{abcd}	2.20 ± 0.09 ^{abc}	1.30 ± 0.03 ^{ab}
Karabigua	2.86 ± 0.03 ^c	2.13 ± 0.07 ^c	2.65 ± 0.18 ^{ab}	1.81 ± 0.20 ^{ab}	0.20 ± 0.00 ^b	0.03 ± 0.01 ^b	3.17 ± 0.01 ^b	1.71 ± 0.04 ^c	4.04 ± 0.03 ^b	3.94 ± 0.03 ^{cd}	1.51 ± 0.01 ^d	0.88 ± 0.01 ^c

Note: Data presented as means of triplicates ± standard deviation. The values within the same column with the same letter are not significantly different ($p > 0.05$)

4 DISCUSSION

4.1 Proximate Composition

The variable response of moisture content to dehulling across varieties reflects a complex relationship between seed coat morphology and the hygroscopic behavior of the cotyledon. The seed coat acts as a selective moisture barrier. Its removal directly exposes the cotyledon tissue to ambient humidity, which can result in increased moisture uptake in highly hygroscopic genotypes (Upreti *et al.*, 2024). This mechanism explains the significant moisture increases observed in cultivars such as Hiba (from 8.57 to 10.21%) and Alfia 5 (from 8.41 to 10.33%). Conversely, the post-dehulling moisture declines seen in Lobab (9.89 to 8.89%) and Defes (9.98 to 9.36%) may be attributable to differences in the moisture content of the hull fraction itself relative to the cotyledon in those genotypes, or to differences in the post dehulling equilibration conditions.

The ash values (3.06–3.68% in WS; 3.11–3.79% in DS) are consistent with established literature for *Vicia faba*, which typically fall between 2.5 and 4.0% (Crépon *et al.*, 2010; Multari *et al.*, 2015). The absence of a consistent directional dehulling effect on ash content indicates that minerals are not distributed uniformly between the hull and cotyledon fractions across these nine genotypes. In certain legume species, the seed coat concentrates specific structural minerals, particularly calcium and magnesium (Sharan *et al.*, 2021), the removal of which lowers total ash. However, the cotyledon is simultaneously the primary repository for phosphorus, potassium, and iron (Montanha *et al.*, 2022). When the non-mineralized hull mass is discarded, these cotyledon-bound elements undergo a relative mathematical concentration on a dry matter basis. The highly variety-dependent ash response highlights a strong genotypic control over spatial mineral partitioning during seed development.

Dehulling induced a modest but consistent upward in crude fat (1.18–1.84 g.100g⁻¹ DM), which falls within the historical ranges of 1.2–1.9% reported by Alghamdi (2009) and 0.79–2.07% reported by Choi *et al.* (2023). The lack of a significant Variety × State interaction for fat content indicates that the effect of processing on this trait is stable across genotypes.

The broad range of total protein content (20.05–28.43% across all samples) underscores both the intrinsic genetic diversity and the environmental plasticity of these cultivars. This profile closely matches the 22.7–28.3% protein range reported for Swedish faba beans (Labba *et al.*, 2021), sits within the 19.83–36.61% range found across 22 South Korean cultivars (Choi *et al.*, 2023), and falls slightly below the upper limits documented in Saudi Arabian (27.5–34.3%) and Mexican (26.59–35.17%) genotypes (Alghamdi,

2009; Corzo-Ríos *et al.*, 2022). These geographical shifts are typical of the strong interactive influences of macroclimate, soil composition, and agronomy on legume storage protein deposition (Crépon *et al.*, 2010).

Crucially, the significant surge in protein concentration following decortication is not merely a mathematical artifact of removing the hull weight. It represents a functional enrichment of the flour in highly digestible storage globulins—specifically legumin (11S) and vicilin (7S)—which are compartmentalized inside the protein bodies of the cotyledon parenchyma (Tiruneh *et al.*, 2025; Warsame *et al.*, 2022). These proteins exhibit superior solubility, digestibility, and techno-functional properties compared to the structural and cell-wall-bound proteins of the seed coat, whose bioavailability is frequently compromised by the formation of insoluble phenolic-protein complexes (Alonso *et al.*, 2000). Dehulling thus optimizes both gross protein yield and net nutritional quality.

The protein gain varied widely by genotype, peaking at +27.98% in Lobab while reaching only +10.17% in Alfia 21. Cultivar Alfia 21 exhibited both the lowest baseline protein content and the lowest post-processing gain, revealing a highly protein-dilute cotyledon fraction that is instead compensated by elevated carbohydrate reserves. This prominent trade-off aligns with the well-established negative correlation between protein and carbohydrate synthesis in grain legumes, governed by competitive assimilate partitioning during seed filling (Choi *et al.*, 2023).

The significant and consistent reduction in carbohydrate content post-dehulling across all varieties is primarily attributable to the removal of the seed coat, a matrix composed primarily of structural non-starch polysaccharides, notably cellulose, hemicellulose, and pectin (Saldanha do Carmo *et al.*, 2022). These insoluble dietary fibers dominate the hull dry matter but are virtually absent within the starch- and protein-dense cotyledon cells. This systematic drop mirrors trends recorded in dehulled mung beans, chickpeas, and common beans (Mubarak, 2005; Wang *et al.*, 2010). This carbohydrate reduction yields a classic nutritional trade-off: it reduces the total dietary fiber intake, yet it simultaneously optimizes the starch-to-fiber ratio, rendering the resulting flour ideal for applications demanding exceptional digestible energy, smooth texturization, and low grittiness (Boye *et al.*, 2010).

4.2 Amino Acid Dynamics and Bioavailability

The predominance of lysine and leucine among the IAAs across all nine cultivars and both hull conditions matches the benchmarks established by Labba *et al.* (2021). This specific profile highlights the exceptional capacity of these faba bean flours to correct the native lysine deficiencies characteristic of cereal-based Mediterranean dietary staples (Millar *et al.*,

2019; Rahate *et al.*, 2021). The effect of dehulling on IAA concentrations was not uniform across varieties, suggesting that the distribution of amino acids between the hull and cotyledon fractions differs by variety. The consistent increase in histidine following dehulling in most varieties' points to a preferential concentration of this amino acid in the cotyledon. The divergent response of isoleucine increasing in certain varieties and decreasing in others may reflect differences in seed morphology, hull thickness, or protein body distribution among varieties, and warrants further investigation. The relative stability of lysine across dehulling treatments in most varieties is nutritionally favorable, as it confirms that this key limiting amino acid is largely retained in the cotyledon fraction regardless of variety.

The high concentrations of glutamic acid, aspartic acid, and arginine among non-IAAs enhance the functional and sensory value of faba bean protein, as these amino acids contribute to umami perception, nitrogen metabolism, and buffering capacity in food systems (Rahate *et al.*, 2021). The marked increase in arginine following dehulling in several varieties suggests its preferential localization in the cotyledon, which has implications for the use of dehulled faba bean flour in functional food formulations where arginine bioavailability is desirable. The notable reduction in proline observed post-dehulling in several varieties, particularly within Defes, Karabiga, and Aguadulce, suggests that this amino acid is concentrated within the hull fraction, though the trend remains highly genotype-dependent.

Of particular interest, the absence of methionine and tryptophan in all hydrolysates is consistent with the well documented limitations of acid hydrolysis, which causes the degradation of sulfur containing and indole containing amino acids respectively (Labba *et al.*, 2021). Regardless, these findings reaffirm the importance of combining faba beans with sulfur amino acid-rich sources when utilized as a primary dietary protein.

Taken together, the results indicate that dehulling exerts a variety dependent effect on the amino acid composition of faba beans, generally concentrating cotyledon bound amino acids while partially redistributing others. These differences have practical implications for variety selection in food processing contexts where specific amino acid targets are prioritized.

4.3 Fatty Acid Architecture

The fatty acid profiles, characterized by the predominance of linoleic (C18:2, n-6) and oleic (C18:1, n-9) acids, match the classic lipid fingerprints of grain legumes documented by Pastor-Cavada *et al.* (2009) and Yoshida *et al.* (2009). The relatively high unsaturation degree underscores the favorable nutritional quality of faba bean lipids.

The significant drop in palmitic acid (C16:0) paired with the simultaneous accumulation of linoleic acid post-dehulling reveals a tissue-specific distribution of fatty acids within the seed. Palmitic acid serves as a fundamental metabolic substrate for suberin biosynthesis within the outer seed coat cells (Franke & Schreiber, 2007). Its sharp decline following dehulling validates this anatomical partitioning. In contrast, the enrichment of linoleic acid in dehulled seeds suggests its primary synthesis in cotyledonary, consistent with their metabolic role.

The detection of docosapentaenoic acid (DPA; C22:5, n-3), though in minor amounts, is noteworthy, as it confirms earlier reports of minor long-chain omega-3 fatty acids occur natively within wild and naturalized legume matrices (Howe et al., 2007; Pastor-Cavada et al. 2009; Yoshida et al. 2009). Interestingly, the general increase in total unsaturated fatty acids (TUFA) seen here after hull removal contrasts with the findings of Choi et al. (2023), who reported no significant dehulling effect on TUFA or TSFA. This discrepancy is likely driven by unique lipid partitioning behaviors unique to these specific Moroccan genotypes.

4.4 Mineral Flux and Genotypic Translocation

The mineral configurations closely track reference values for high-protein pulses (Khazaei & Vandenberg, 2020; Mattila et al., 2018). Potassium and phosphorus represented the predominant elemental fractions, confirming the crop's significant contribution to dietary electrolyte balance and energy metabolism.

The marked reduction in calcium content following dehulling (−8.81% to −52.90%) corroborates its localization within the seed coat (Blair et al., 2013; Moraghan et al., 2006). In contrast, the increase in phosphorus and sulfur concentrations following dehulling (+7.46% to +20.50% and +7.80% to +16.16%, respectively) likely reflects their association with metabolically active cotyledons.

The highly significant varietal effect ($p < 0.05$) across nearly all mineral elements highlights a powerful genotypic influence over mineral accumulation and translocation dynamics. The observed levels of iron (5.67–8.63 mg.100g⁻¹) and Zn (2.48 – 7.46 mg.100g⁻¹) are of immense nutritional relevance given the global concern over iron and zinc deficiencies.

4.5 Phytochemical and Antinutritional Profiles

The significant decline in TPC and condensed tannins following dehulling is consistent with the well-established role of the seed coat as the primary reservoir of phenolic compounds in legumes (Çalışkantürk Karataş et al., 2017). Legume hulls typically concentrate these secondary metabolites to serve as a biochemical shield against field

pathogens and environmental oxidative stress (Ferreira et al., 2022). Consequently, the mechanical decortication successfully strips away this anti-nutritional barrier.

The non-significant effect of dehulling on total flavonoid content, however, suggests that flavonoids in faba bean are not exclusively or predominantly localized in the seed coat, but are also distributed throughout the cotyledon tissue. This finding is consistent with reports in further legume species where flavonoids particularly isoflavones and flavonols are present in appreciable quantities in the cotyledon fraction and are therefore largely retained after hull removal (Okafor et al., 2022). This differential distribution of phenolic subclasses within the seed architecture has crucial implications for processing: while dehulling effectively reduces condensed tannins and total polyphenols, it does not significantly compromise the flavonoid pool, which may be particularly relevant for food applications targeting the health promoting properties associated with this subclass, including anti-inflammatory, antioxidant, and cardioprotective effects (Liu et al., 2024).

The TPC values obtained in whole seeds (2.86 – 4.26 mg GAE.g⁻¹ DM) are slightly lower than those reported by Chaieb et al. (2011), a discrepancy that could be attributed to differences in extraction solvents, solvent to sample ratios, and variety specific phenolic profiles. Such methodological variability in polyphenol quantification has been widely acknowledged in the legume literature (Xu et al., 2007). The condensed tannin concentrations recorded in WS (2.38 – 2.88 mg CE.g⁻¹ DM) fall within the broad range reported by Oomah et al. (2011) (0.50 – 5.42 mg CE.g⁻¹ DM) and corroborate the observation by Sharan et al. (2021) that condensed tannins represent 72–82% of total seed coat phenolics in faba beans.

From a nutritional standpoint, the reduction of condensed tannins through dehulling carries a dual significance: while tannins are recognized antinutritional factors that form complexes with dietary proteins and reduce their digestibility and bioavailability (Cormack et al., 2026), additionally they exert beneficial biological effects including antioxidant, antimicrobial, and anti-inflammatory activities (Cosme et al., 2025). Dehulling therefore represents a tradeoff between improved protein digestibility and a partial reduction in bioactive compound content one that is nuanced by the fact that flavonoids, an important bioactive subclass, are not significantly affected by the process.

4.6 In Vitro Antioxidant Dynamics

The consistent reduction in antioxidant activity as measured by DPPH•, ABTS•+, and FRAP assays across all cultivars post-dehulling directly reflects the loss of hull associated polyphenols and condensed tannins (Martineau-

Côté et al., 2022b; Multescu et al., 2024). These compounds neutralize free radicals via hydrogen-atom transfer or single-electron donation; thus, removing the hull systematically reduces the total radical-quenching capacity of the resulting flours (Hernández-Ruiz et al., 2025).

The partial preservation of antioxidant activity in dehulled flours across all varieties further proves that cotyledon-bound phenolic elements including the flavonoid fraction, continue to supply meaningful protective activity, as documented in lentils and chickpeas (Dueñas et al., 2006).

The significant Variety × State interaction ($p < 0.05$) across these functional assays confirms that genotypic variations in hull phenolic composition influence the magnitude of bioactive compound loss upon dehulling. This diversity is regulated by allelic variations at loci governing the core steps of the tannin and flavonoid biosynthetic pathways (Gutierrez & Torres, 2019).

While these findings differ slightly from the profiles noted by Choi et al. (2023), potentially due to differences in variety selection, growing conditions, or processing methods, they firmly reinforce the dual nature of faba bean processing. Dehulling is highly effective at reducing antinutritional tannins, yet it simultaneously depletes the raw antioxidant potential of the flour.

For commercial food applications where both exceptional protein bioavailability and high antioxidant activity are required simultaneously, alternative processing avenues should be explored. These include partial decortication regimes, the technical valorization of the discarded hull as a standalone functional fiber ingredient, or the controlled fortification of dehulled flours using targeted hull phenolic extracts (Ozkan et al., 2025)

5 CONCLUSIONS

This study demonstrates that mechanical dehulling significantly affects the proximate profiles, bioactive compound content, and antioxidant activity of nine Moroccan faba bean varieties, with all changes heavily modulated by the genetic background of the cultivar.

- **Nutritional Optimization:** Dehulling consistently increased total crude protein content across all investigated genotypes, reduced carbohydrate content, and shifted lipid profiles toward a highly favorable unsaturated fatty acid distribution.
- **Mineral and Micronutrient Flux:** Ash and moisture responses were highly variety-dependent. Dehulling provoked severe losses in calcium due to its structural localization within the seed coat, but successfully concentrated phosphorus and sulfur reserves within the cotyledon fraction. The iron and zinc concentrations

observed highlight a strong potential for targeted breeding and nutritional biofortification.

- **Phytochemical Trade-offs:** Total polyphenols and condensed tannins dropped sharply across all varieties, verifying their role as primary physical and chemical shields within the seed coat. Total flavonoids displayed immense phenotypic diversity; while heavily depleted by dehulling in most cultivars, they were uniquely enriched within the dehulled cotyledon of the Alfia 21 cultivar.
- **Antioxidant Capacity:** Net antioxidant performance (DPPH•, ABTS•+, and FRAP) was consistently superior in whole seeds, directly reflecting the removal of highly active hull-bound phenolics.

In conclusion, while mechanical dehulling serves as an efficient processing strategy to improve macronutrient purity, protein bioavailability, and lipid profiles, it simultaneously causes a significant loss of bioactive phytochemicals and radical-scavenging potential. These findings provide a valuable, comprehensive baseline for selecting and utilizing specific Moroccan faba bean varieties to develop optimized functional foods, plant-based protein isolates, and highly bioavailable nutritional formulations.

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