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# **Evaluation and Optimization of Extraction Conditions for Protein Isolates from Underutilized Ethiopian** *Lupinus spp.*: Chemical Characterization and Yield Assessment

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## **INTRODUCTION & AIM**

Global protein demand continues to rise due to population growth, changing diets, and concerns over the sustainability of animal-based proteins (Karabulut et al., 2024). In response, plant-based proteins are emerging as sustainable, affordable, and nutritionally adequate alternatives in modern food systems (Xiao et al., 2023). Lupine (*Lupinus spp.*) is a promising plant-based protein source with high nutritional value and functional potential for food formulation (Zafeiriou et al., 2021). Despite its adaptability to diverse agro-ecological zones, lupine remains underutilized in Ethiopia. Ethiopia hosts a traditional *Lupinus albus* landrace alongside recently introduced *Lupinus angustifolius* varieties that could enhance local protein self-sufficiency. However, comparative information on their chemical composition, anti-nutritional factors, protein quality, and protein isolate characteristics is lacking. Generating such information is essential for optimizing processing strategies and promoting lupine-based protein ingredients as sustainable alternatives to animal-derived proteins.









Figure 1. List of Ethiopian *Lupinus* genotypes used for protein isolate extraction: one *L* albus (Local) and three *L* angustifolius varieties (Vitabor, Probor, and Sanabor).

#### **METHOD** Lupine Genotypes (Local, Vitabor, Sanabor & Probor) Determination of Determination of Debittering and Defatting Chemical Composition Chemical Composition & Yield Lupin protein extraction (Extraction Lupine protein Flour (g/100g) pH (A) 7-10 and Isoelectric isolate (LPI) % Total alkaloid precipitation pH (B) 4-5 Moisture content Moisture content Ether extract Design-Expert ®, version 13, Stat-Crude protein Ease (Minneapolis USA (CCD) was LPI Yield Ether extract used (total 52 runs) LPI Recovery EAA mg/g, LPI Purity EAAI mg/g, The significance was examined by Total alkaloid Amino acid score ANOVA and the test level was 5%. mg/100g mg/g Numerical optimization was applied.

Figure 2. Experimental Workflow Illustrating Protein Isolate Preparation, Analytical Determination, and Optimization Design for Four Ethiopian *Lupinus* Genotypes.

## **CONCLUSION**

- Ethiopian lupine genotypes showed high protein content (≥40 g/100 g) and a complete EAA profile.
- Under optimized extraction conditions, *Vitabor* protein isolate demonstrated superior amino acid balance, high purity, yield, and minimal alkaloid content.
- This establishes them as a high-quality and environmentally sustainable alternative to animal proteins, particularly relevant for nutrition improvement in low-income regions.
- Local and Probor isolates provide complementary lipid and purity advantages, supporting the development of functional lupine proteins for sustainable, plant-based formulation.

## **RESULTS & DISCUSSION**

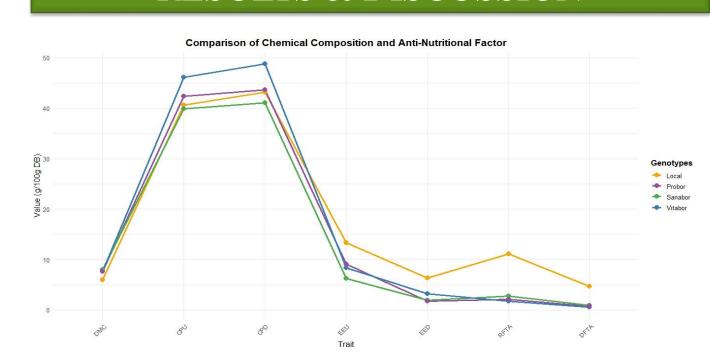


Figure 3. Chemical Composition and Anti-nutritional factor of Lupine Flour

The variables include DMC, the moisture content of defatted flour; CPU, the crude protein from undefatted flour; CPD, the crude protein from defatted flour; EEU, the ether extract content from undefatted flour; EED, the ether extract from defatted flour; RFTA, the total alkaloid from raw flour; and DFTA, the total alkaloid from debittered flour.

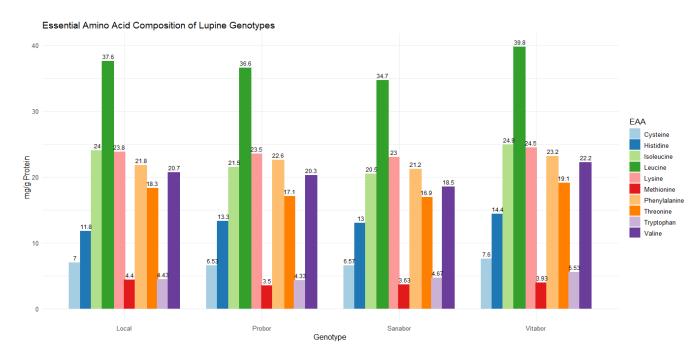


Figure 5. Essential Amino Acid composition of Lupine Flour

Table 1. Optimized conditions for lupine protein isolates of local (L. *albus*) and other varieties of (*L. angustifolius*)

Genotypes	A	В	PIY	PIP	PIR	PIMC	PIEE	TA	Desirability
Local	10.0	5.00	25.8	88.9	53.4	4.75	2.59	16.4	0.88
Vitabor	9.86	4.66	26.4	90.7	50.3	3.58	1.38	0.24	0.64
Sanabor	10.0	4.69	22.0	88.5	47.2	3.18	0.85	0.98	0.75
Probor	9.58	4.64	22.6	90.6	47.9	4.15	0.80	0.71	0.71

PIY, protein isolate yield; PIP, protein isolate purity; PIR, protein isolate recovery; PIMC, protein isolate moisture content; PIEE, protein isolate ether extract; TA, total alkaloid.

### FUTURE WORK / REFERENCES

- These findings support the development of functional LPIs to meet global needs for sustainable nutrition, food innovation, and plant-based dietary solutions.
- Further work will enhance these isolates by investigating their structural, functional, and bioactive properties, enabling broader applications in the food, nutraceutical, and therapeutic sectors.