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# Evaluation of *Canavalia ensiformis* L. beans: Comparative Fatty Acid Profile in Extracts and Analysis of Minerals and Proximate Composition

**Abstract.** This study investigates the proximate composition, mineral content, and fatty acid profile of *Canavalia ensiformis* L. beans extracted with various solvents. Proximate analysis revealed a high protein content (23.16%), moderate fiber levels (7.13%), and a total fat content of 2.35%. Elemental analysis identified potassium (18,696.87 mg/kg) as the most abundant macroelement, followed by calcium (1,886.81 mg/kg) and magnesium (1,500.98 mg/kg), whereas iron (50.43 mg/kg) and zinc (33.28 mg/kg) were the predominant microelements. Gas chromatography analysis of fatty acids showed that monounsaturated fatty acids (MUFA) dominated the lipid profile, particularly in the hexane (58.67%), ethanol (59.57%), and raw bean (61.11%) extracts. Oleic acid ($C_{18:1n9c}$) was the most abundant MUFA, reaching 51.39% in the hexane extract. Saturated fatty acids (SFA) were highest in the hexane extract (21.35%), primarily due to palmitic acid ($C_{16:0}$, 18.77%). Polyunsaturated fatty acids (PUFA) were most prevalent in the hexane extract (16.23%) and bean sample (16.08%), with linoleic acid ($C_{18:2n6c}$) being the major component. The aqueous extract contained notable amounts of $\gamma$-linolenic acid ($C_{18:3n6}$, 1.68%). These findings highlight the nutritional potential of *C. ensiformis* beans and suggest that solvent selection significantly influences lipid composition. The high MUFA and PUFA content, particularly in hexane and ethanol extracts, underscores their possible applications in food and pharmaceutical formulations.
**Key words:** *Canavalia ensiformis* L., GC analysis, proximate analysis, micro- and macroelements, fatty acids.

## Introduction

Jack bean (*Canavalia ensiformis* L.) is a tropical leguminous plant from the *Fabaceae* family, known for its high protein content exceeding 20-40%. The rising protein consumption in developing countries, coupled with the high cost of protein imports, has driven the search for more affordable and widely available alternative protein sources [1]. In recent studies, Jack bean milk has the potential to emerge as a new type of plant-based milk. If developed, it could serve as an alternative for individuals with lactose intolerance, milk allergies, or those following a vegetarian diet [2]. Soaking and fermentation are the most commonly used methods for studying *C. ensiformis*, with good results, alongside germination, boiling, autoclaving, genetic manipulation, and other processing methods [3].

In the studies by Kanetro B. [4], the effect of germination of jack bean seeds on the functional properties of its flour was investigated. It was found that 72-hour germination significantly enhanced protein content, protein solubility, and foaming capacity due to an increased amount of hydrophobic amino acids, indicating its potential use in food products with improved nutritional and functional properties. Sutedja M. and her team [5] investigated flavonol glycosides from jack bean (*C. ensiformis*) for their $\alpha$-glucosidase inhibitory activity, a key target in carbohydrate metabolism. The findings revealed potent inhibitory effects of certain compounds, indicating their potential as antidiabetic agents. Saldarriaga F. and his colleagues [6] conducted a study investigating the effects of pH and different light conditions (red, blue, and their combinations) on callus growth and the production of bioactive compounds in *C.*

 

*ensiformis*. Optimal growth and enhanced production of phenols, carotenoids, and antioxidant activity were achieved at pH 5.5 and under 1/3 red-blue (R-B LED) light conditions. In this context, there is increasing interest in studying *C. ensiformis* L. due to its rich composition of bioactive compounds such as phenols, carotenoids, and proteins such as urease and lectin, may be useful for developing supportive therapies or health-promoting applications.

This study investigated proximate, elements analysis of plant and the effect of different solvents on the extraction of fatty acids from *C. ensiformis* seeds.

**Materials and methods**

The mature *Canavalia ensiformis* L. beans were collected from Karachi, Pakistan. *C. ensiformis* (10 kg) were crushed and powdered using an electronic mill. The powdered beans of *C. ensiformis* were macerated in 80% EtOH three times, and the ethanol extract was evaporated under reduced pressure to yield a brown residue (427 g). Furthermore, the ethanol extract was suspended in distilled water and partitioned sequentially with *n*-hexane (85 g), dichloromethane (DCM) (3.67 g), ethyl acetate (2.66 g), and *n*-butanol (49 g), resulting in five portions.

*Proximate analysis.* The beans of *C. ensiformis* L. were analysed in triplicate for proximate composition: (total ash, dry matter, crude fiber, and fat contents) was conducted using standard methods of the Pharmacopoeia of the Republic of Kazakhstan and total crude protein by the Kjeldahl method [6].

*Micro and macro element analysis* was conducted using a Shimadzu AA 6200 (Japan) dual-beam atomic absorption spectrophotometer with flame and electrothermal atomization. Standard procedures were followed to ensure accuracy and reproducibility.

*The fatty acid composition* of the plant extracts was performed using a «Chromos-1000» (Russia) gas chromatograph equipped with a flame ionization detector (FID). A CP-Sil 88 for FAME 100x0.25x0.36 mm column was used for separation. Chromatographic Conditions: Oven Temperature Program: Initial temperature: 70 °C, hold for 3 min, Ramp: 8 °C/min to 120 °C, hold for 2 min, Ramp: 5 °C/min to 200 °C, hold for 4 min, Ramp: 7 °C/min to 220 °C, hold for 61.5 min, Total run time: 99.61 min. Instrument Temperatures: Column oven: 70 °C; Injector: 250 °C; FID detector: 280 °C. Carrier gas (nitrogen): 1.7 kgf/cm². Auxiliary nitrogen flows: 25 cm³/min and 40 cm³/min. Hydrogen: 20 cm³/min. Air: 200 cm³/min. The sample was dissolved in hexane as a solvent and injection volume of 0.1 μL.

**Results and discussion**

The proximate compositions were determined in triplicate, with all data presented on a dry weight basis and expressed as percentages (%). The moisture and ash content exhibited moderate levels, aligning with the standards outlined in the Pharmacopoeia of the Republic of Kazakhstan and the result is in agreement with the research investigation of Vadivel V. who reported values of 3.8% to 9.2% moisture and for ash values from 3.0% to 5.8% [7-8]. In the present study, the range of protein (23.16%) found lower than some other research [7, 9] (Table 1)

**Table 1** – Proximate analysis of *Canavalia ensiformis* L. beans

| Parameter | Percentage Values (Dry Weight Basis) ± SD |
|---|---|
| Ash | 3.87 ± 0.19 |
| Dry matter | 96.13 ± 4.81 |
| NDF | 7.13 ± 1.06 |
| Crude Protein | 23.16 ± 1.39 |
| Fat | 2.35 ± 0.19 |
| Moisture | 6.51 ± 0.33 |
| Carbohydrate | 64.11 ± 3.21 |
| Energy content, kcal/kJ | 370/1579 |
| Note: *NDF – Neutral Detergent Fiber | |

The results of elemental analysis are given in Table 2. Among the macroelements, potassium (K) was the most abundant (18,696.87 mg/kg), playing a key role in cellular function, osmotic balance, and enzyme activation [10]. Calcium (Ca) (1,886.807 mg/kg) and magnesium (Mg) (1,500.978 mg/kg) contribute to bone health, muscle function, and metabolic processes, while sodium (Na) (395.864 mg/kg) regulates fluid balance and nerve transmission [11-12].

Regarding microelements, iron (Fe) (50.428 mg/kg) is crucial for oxygen transport and energy metabolism [13]. Zinc (Zn) (33.278 mg/kg) plays a vital role in immune function, wound healing, and enzymatic activity [14]. Copper (Cu) (11.273 mg/kg) is essential for redox reactions, connective tissue formation, and nervous system maintenance [15]. Manganese (Mn) (8.602 mg/kg) supports bone development and antioxidant defense, whereas nickel (Ni) (8.929 mg/kg) is involved in enzyme function and microbial metabolism [15].

**Table 2** – Mineral composition of *Canavalia ensiformis* L. beans

| Minerals | Concentration, mg/kg |
|---|---|
| Zn | 33.278 |
| Cu | 11.273 |
| Pb | 7.765 |
| Cd | 0.978 |
| Fe | 50.428 |
| Ni | 8.929 |
| Mn | 8.602 |
| Ca | 1886.807 |
| Mg | 1500.978 |
| K | 18696.87 |
| Na | 395.864 |

The fatty acid profile of *C. ensiformis* beans (Table 3) and its various solvent extracts revealed a diverse range of saturated, monounsaturated, and polyunsaturated fatty acids, highlighting their potential nutritional and functional significance. Saturated fatty acids were present in varying concentrations across different extracts. The total SFA content was highest in the hexane extract (21.346%) and the bean sample (18.629%), primarily due to the presence of palmitic acid ($C_{16:0}$) (18.768% and 13.180%, respectively) and stearic acid ($C_{18:0}$). These fatty acids are known to contribute to membrane stability and are common in plant-based lipid sources [15]. The aqueous extract also showed a relatively high SFA content (10.916%), with notable amounts of stearic acid ($C_{18:0}$) (4.842%) and palmitic acid ($C_{16:0}$) (4.302%). However, shorter-chain saturated fatty acids such as butyric acid ($C_{4:0}$) and capric acid ($C_{10:0}$) were either absent or found in trace amounts.

Monounsaturated fatty acids (MUFA) constituted the dominant fraction in most extracts, particularly in the hexane (58.667%), 70% ethanol (59.565%), and raw bean (61.106%) samples. Oleic acid ($C_{18:1n9c}$) was the most abundant MUFA, accounting for 50.102% in the bean sample and 51.389% in the hexane extract, reinforcing the potential health benefits of these extracts in terms of cardiovascular health and anti-inflammatory properties [16-20]. The ethanol extract exhibited a significant proportion of myristoleic acid ($C_{14:1}$) (16.349%). Additionally, eicosenoic acid ($C_{20:1}$) was notable in the bean sample (9.296%) and hexane extract (6.121%), which could contribute to lipid metabolism regulation [21].

Polyunsaturated fatty acids (PUFA), particularly linoleic acid ($C_{18:2n6c}$) and linolenic acid ($C_{18:3n3}$), were found in notable amounts. The highest PUFA content was recorded in the hexane extract (16.229%) and bean sample (16.078%), with linoleic acid contributing significantly to these values. Linoleic acid is an essential fatty acid involved in cell membrane function and inflammatory response regulation. The aqueous extract contained γ-linolenic acid ($C_{18:3n6}$) (1.675%), which has been associated with anti-inflammatory and neuroprotective effects [22-25]. However, the DCM extract exhibited the lowest PUFA content (0.410%), suggesting its limited ability to extract these essential fatty acids.

**Table 3** – Fatty acid profile of *C. ensiformis* beans extracted with various solvent

| RT | Fatty acid | Concentration, % | | | | | |
|---|---|---|---|---|---|---|---|
| | | *Canavalia ensiformis* L. beans | 70 % EtOH extract | Hexane Extract | DCM extract | Butanol extract | Aqueous extract |
| 16.47 | $C_{4:0}$ Butyric acid | NF | NF | NF | NF | 0.267 | NF |
| 29.48 | $C_{10:0}$ Capric acid | 0.020 | NF | NF | NF | NF | 0.825 |
| 34.38 | $C_{12:0}$ Lauric acid | 0.045 | NF | NF | NF | NF | 0.135 |
| 39.1 | $C_{14:0}$ Myristic acid | 0.346 | NF | 0.316 | 0.008 | 0.209 | 0.584 |
| 41.18 | $C_{14:1}$ Myristoleic acid | 0.200 | 16.349 | 0.276 | 0.007 | 0.408 | 0.108 |
| 43.6 | $C_{15:1}$ Pentadecanoic acid | NF | NF | NF | NF | 0.005 | NF |
| 43.85 | $C_{16:0}$ Palmitic acid | 13.180 | 9.242 | 18.768 | 0.498 | 3.739 | 4.302 |
| 45.87 | $C_{16:1}$ Palmitoleic acid | 0.870 | 1.638 | 0.664 | 0.019 | 2.023 | 0.298 |
| 46.28 | $C_{17:0}$ Margaric acid | 0.151 | NF | 0.170 | NF | NF | NF |
| 48.31 | $C_{17:1}$ Margaroleic acid | 0.331 | 3.870 | 0.218 | NF | NF | NF |

*Continuation of the table*

| RT | Fatty acid | Concentration, % | | | | | |
|---|---|---|---|---|---|---|---|
| | | *Canavalia ensiformis* L. beans | 70 % EtOH extract | Hexane Extract | DCM extract | Butanol extract | Aqueous extract |
| 48.87 | $C_{18:0}$ Stearic acid | 1.436 | NF | 1.285 | 0.078 | 3.045 | 4.842 |
| 50.19 | $C_{18:1}$(*trans*-9) Elaidic acid | 0.247 | 1.169 | NF | NF | 12.786 | 4.479 |
| 50.81 | $C_{18:1n9c}$ Oleic acid | 50.102 | 24.930 | 51.389 | 1.286 | 17.684 | 4.152 |
| 52.34 | $C_{18:2n6t}$ *trans*-Linoleic acid | NF | NF | NF | NF | 0.353 | 0.156 |
| 53.85 | $C_{18:2n6c}$ Linoleic acid | 14.696 | 9.518 | 15.378 | 0.399 | 3.544 | NF |
| 54.64 | $C_{18:3n3}$ Linolenic acid | 0.547 | NF | 0.598 | 0.010 | 0.286 | 0.758 |
| 56.38 | $C_{18:3n6}$ γ-Linolenic acid | 0.133 | 1.988 | 0.029 | NF | 1.675 | NF |
| 57.01 | $C_{20:0}$ Arachidic acid | 1.628 | NF | 0.451 | 0.006 | 0.352 | 0.228 |
| 57.6 | $C_{20:1}$ Eicosenoic acid | 9.296 | 2.367 | 6.121 | 0.142 | 0.418 | NF |
| 56.38 | $C_{20:3n6}$ Homo-γ-Linolenic acid | 0.509 | NF | 0.091 | NF | NF | 0.102 |
| 65.09 | $C_{20:3n3}$ Eicosatrienoic acid | 0.192 | 0.344 | 0.133 | NF | 0.337 | 0.254 |
| 60.9 | $C_{21:0}$ Heneicosic acid | 0.070 | NF | 0.027 | NF | NF | NF |
| 66.09 | $C_{22:0}$ Behenic acid | 0.185 | NF | 0.059 | NF | NF | NF |
| 72 | $C_{23:0}$ Tricosanoic acid | 1.568 | NF | 0.270 | 0.005 | NF | NF |
| 76.28 | $C_{24:1}$ Nervonic acid | 0.061 | NF | NF | NF | NF | NF |
| Saturated acids | | 18.629 | 9.242 | 21.346 | 0.595 | 7.611 | 10.916 |
| Monounsaturated acids | | 61.106 | 59.565 | 58.667 | 1.458 | 33.267 | 8.946 |
| Polyunsaturated acids | | 16.078 | 11.849 | 16.229 | 0.410 | 6.196 | 2.587 |
| Note: *NF – not found | | | | | | | |

## Conclusion

The results of this study provide valuable insights into the nutritional composition and fatty acid profile of *Canavalia ensiformis* L. beans. The beans were found to have a high protein content (23.16%) and moderate levels of fiber (7.13%) and fat (2.35%), making them a potential source of plant-based protein. Mineral analysis showed that potassium (18,696.87 mg/kg) was the most abundant macroelement, followed by calcium (1,886.81 mg/kg) and magnesium (1,500.98 mg/kg), indicating their potential contribution to essential dietary minerals.

Fatty acid analysis revealed that monounsaturated fatty acids (MUFA) were the predominant lipid fraction, with oleic acid ($C_{18:1n9c}$) reaching 51.39% in the hexane extract. Saturated fatty acids were highest in the hexane extract (21.35%), while polyunsaturated fatty acids, particularly linoleic acid ($C_{18:2n6c}$), were most abundant in the hexane (16.23%) and raw bean (16.08%) samples. The choice of extraction solvent significantly influenced the fatty acid composition, with hexane and ethanol yielding extracts rich in nutritionally beneficial lipids.

These findings highlight the nutritional potential of *C. ensiformis* beans and their possible applications in food and pharmaceutical industries. Further research is needed to explore the bioavailability of their nutrients, the effects of processing on lipid composition, and potential functional applications of the extracted compounds.

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## Conflict of interest

All authors are aware of the article's content and declare no conflict of interest.

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